JTBB
Journal of Tropical Biodiversity and Biotechnology

Journal of Tropical Biodiversity and Biotechnology

online http://jtbb.or.id

ISSN 2540-9581 (online)

Journal of Tropical Biodiversity and Biotechnology

Journal of Tropical Biodiversity and Biotechnology (JTBB) is an open access, peer-reviewed journal dedicated to the publication of novel research concerned with the advancement of tropical biology studies in the Southeast Asia Region.

JTBB publishes the following subjects:

- Descriptive and analytical studies of genetic, species, and community diversity
- Ecological and conservation studies
- The utilisation of tropical organisms in biotechnology
- Novel methods in studying biodiversity
- Etnobotany and etnozoology
- Bioinformatics

Editorial Team

Reviewer List

Name	Institution
Assoc. Prof. Dr. Wong Wey Lim	Department of Biological Science, Faculty of Science, Universiti Tunku Abdul Rahman, Malaysia
Dadan Supardan, S.Si., M. Biotech.	State Islamic University of Mataram, Indonesia
Dr. Arief Darmawan	Faculty of Agriculture, University of Lampung, Indonesia
Dr. Dini Wahyu Kartika Sari, S.Pi., M.Si.	Department of Fisheries, Universitas Gadjah Mada, Indonesia
Dr. E. Charles Morris	Western Sydney University: Penrith, AU
Dr. Endah Retnaningrum, M. Eng	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Dr. Farid Asif Shaheen	Department of Entomology, PMAS-Arid Agriculture University Rawalpinidi, Pakistan
Dr. Ir. Wartika Rosa Farida	Research Center for Biology, Indonesian Institute of Sciences (LIPI), Indonesia
Dr. Mahfut, M.Sc.	Department of Biology, University Lampung, Indonesia
Dr. Maryani, M.Sc.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Dr. Ni Kadek Dita Cahyani	Indonesian Biodiversity Research Center
Dr. Phoon Lee Quen	Department of Allied Health Sciences, Faculty of Science, Universiti Tunku Abdul Rahman, Malaysia
Dr. rer. nat. Andhika Puspito Nugroho, S.Si., M.Si.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Dr. Susanti Withaningsih, M.Si.	Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Padjadjaran, Indonesia

	Indonesia
Dr. Tri Muji Ermayanti	Research Center for Biotechnology, Indonesian Institute of Sciences (LIPI), Indonesia
Dr. Windi Mose	Department of Biology, University of Pattimura, Indonesia
Dra. Kusmiati, M.Si.	Research Center For Biotechnology, Indonesian Institute of Sciences (LIPI), Indonesia
Dra. Tuty Arisuryanti, M.Sc., Ph.D.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Drs. Tri Joko, M.Si.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Husna Nugrahapraja, Ph.D.	Genetics and Molecular Biotechnology, School of Life Sciences & Technology Institut Teknologi Bandung, Indonesia
Imroatushshoolikhah, S.Si, M.Sc.	Research Center for Limnology, Indonesian Institute of Sciences (LIPI), Indonesia
Pranatasari Dyah Susanti, SP, MS	Watershed Management Technology Center (WMTC), Kartasura, Indonesia
Prof. Dr. Drs. Wibowo Mangunwardoyo M.Sc.	Department of Biology, FMIPA, Universitas Indonesia, Indonesia
Prof. Dr. Endang Semiarti, M.S., M.Sc.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Prof. Dr. Jonathan A. Anticamara	Institute of Biology, College of Science, University of the Philippines, Philippines
Prof. Dr. L. Hartanto Nugroho, M.Agr.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Prof. Dr. Purnomo, M.S.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Reni Ambarwati, S.Si., M.Sc.	Department of Biology, Faculty of Mathematics and Natural Sciences, State University of Surabaya, Indonesia
Rina Sri Kasiamdari, Ph.D.	Faculty of Biology, Universitas Gadjah Mada, Indonesia
Siti Nurfadilah, M.Sc.	Purwodadi Botanical Garden, Indonesian Institute of Sciences (LIPI), Indonesia

Journal of Tropical Biodiversity and Biotechnology
Volume 06, Issue 01 (2021): jtbb58528
DOI: 10.22146/jtbb.58528

Short Communication

Antibacterial Activity of *Kecombrang* Flower (*Etlingera elatior* (Jack) R.M. Sm) Extract against *Staphylococcus epidermidis* and *Propionibacterium acnes*

Vilya Syafriana[1]*, Renita Noviani Purba[1], Yayah Siti Djuhariah[1]
1) National Institute of Science and Technology (ISTN), Jl. Moh. Kahfi II, Srengseng Sawah, Jagakarsa, Jakarta Selatan, 12640
* Corresponding author, email: v.syafriana@istn.ac.id

Submitted: 12 August 2020; **Accepted:** 27 December 2020; **Published online:** 23 January 2021

ABSTRACT
This study aimed to determine the antibacterial activity from the ethanol extract of *Kecombrang* flower (*Etlingera elatior* (Jack) R.M. Smith) against *Staphylococcus epidermidis* and *Propionibacterium acnes*. The extract was made by the maceration method with 70% ethanol as a solvent. Antibacterial activity test was carried out by the disk diffusion method with a concentration of 10%, 20%, 40%, and 80%. Meanwhile, the Minimum Inhibitory Concentration (MIC) was done at concentrations of 10%, 8%, 6%, 4%, and 2%. The results showed that the *Kecombrang* flower (*Etlingera elatior* (Jack) R.M.Smith) extract had antibacterial activity against *S. epidermidis* and *P. acnes*. The MIC for *S. epidermidis* is at a concentration of 4%, while in *P.acnes* cannot determine yet.

Keywords: antibacterial, ethanol extract, *Kecombrang* flower, *Propionibacterium acnes, Staphylococcus epidermidis*

Staphylococcus epidermidis and *Propionibacterium acnes* are known as commensals bacteria in human skin which can change into opportunistic (Nakase et al. 2014; Chessa et al. 2015). *Staphylococcus epidermidis* colonizes various areas of the skin, while *P. acnes* resides mainly in the pilosebaceous skin follicles. This microbial interplay, for instance, mediated through molecules involved in intercellular competition or communication, may have an impact on the fine balance of the skin ecosystem. A disturbed balance (dysbiosis) can impact skin health and might initiate or support the events that lead to skin disorders. One of such disorders is acne vulgaris, multifactorial disease of pilosebaceous units of the skin that affects adolescents (Christensen et al. 2016).

Propionibacterium acnes can be related to the initial stage of acne because it causes an increase in the lipogenesis originated in sebaceous glands. It induces inflammation and pustules on the skin (Neves et al. 2015; Blaskovich et al. 2019). Meanwhile, *S. epidermidis* also can be opportunistic when it enters the bloodstream (Nakase et al. 2014; Tabri 2019).

Acne treatment in skin clinics usually uses antibiotics that can overcome inflammation and kill bacteria such as tetracycline, erythromycin, doxycycline, and clindamycin (Nakatsuji et al. 2009; Doğan et al. 2017). However, these drugs have side effects such as irritation and allergy, while

long-term use of antibiotics can cause resistance, organ damage, and immune -hypersensitivity (Adawiyah et al. 2010; Tan et al. 2018; Dikicier 2019). These problems have led many researchers to discover and develop new sources for antimicrobial agents from natural products, e.g. medicinal plants (Abdallah 2011). Sadeek & Abdallah (2019) stated that some phytochemical compounds extracted from medicinal plants showed effective antibacterial potential against multi-drug-resistant pathogens and these compounds could be exploited as antibacterial drugs.

Indonesia is known as one of the countries that have many medicinal plants. One of them is *Kecombrang* (*Etlingera elatior* (Jack) R.M.Smith). *Kecombrang* is a spice plant that belongs to the Zingiberaceae Family and has been used in making medicine as well as flavour enhancers. This plant contains secondary metabolites such as phenols, flavonoids, glycosides, saponins, tannins, steroids, and terpenoids (Silalahi 2017; Juwita et al. 2018; Effendi et al. 2019). Those compounds are known as potential sources for antibacterial agents (Abdallah 2011; Sadeek & Abdallah 2019). Based on Farida & Maruzy (2016) report, *Kecombrang* flower has more antibacterial compounds compare to its rhizome, leaves, or fruit. *Kecombrang* fruit contains flavonoids only. *Kecombrang* leaves contain saponin and flavonoids. The rhizome of *Kecombrang* contains saponin, tannin, sterol, and terpenoid. Meanwhile, *Kecombrang* flower contains flavonoid, saponin, tannin, and terpenoid.

Some studies reported that *Kecombrang* flower extract has an antibacterial activity to some bacteria. Mackeen et al. (1997) reported that ethanol extract of *Kecombrang* flower can inhibit the growth of *Pseudomonas aeruginosa, Escherichia coli, Bacillus megaterium*, and *Cryptococcus neoformans*. Wijekoon et al. (2013) reported that *Kecombrang* flower extract with various solvents (water, 50% ethanol, and 96% ethanol) can inhibit the growth of *Bacillus cereus, Bacillus subtilis, Staphylococcus aureus, Listeria monocytogenes*, and *Klebsiella pneumoniae*. Another research by Ghasemzadeh et al. (2015), showed that the ethanol and water extract of *Kecombrang* flower can inhibit *S. aureus, B. subtilis, E. coli, Salmonella* sp., *Micrococcus* sp., and *Proteus mirabilis*. Naufalin & Rukmini (2018) reported that the ethanol extract of *Kecombrang* flower has better antibacterial activity thBased on that background, this study was done to determine the antibacterial activity from the ethanol extract of *Kecombrang* flower against *S. epidermidis* and *P. acnes*. Since *Kecombrang* flower is high in containing polyphenol compounds, so it is best to use ethanolic solvents (Farida & Maruzy 2016). Ethanol can attract more polyphenol compounds than others (Tiwari et al. 2011). The outcome of the study is expected to show that ethanol extract of *Kecombrang* flower can be used as an alternative for acne treatment. This study was done by several steps, namely, plant source and preparation, extraction, phytochemical screening, disk diffusion test, and dilution method to determine Minimum Inhibitory Concentration (MIC).

The *Kecombrang* flower used in this research was obtained from *Kecombrang* plantation in Lubuk Begalung, Padang City, West Sumatra. About eight kg of the flowers was washed with clean water and cut into small pieces, then placed in a container and spread evenly for the drying process. The flowers were dried in the oven with a temperature of 40-50 ºC for 3 x 24 hours. The purpose of drying is to get the simplicia of flower that is not easily damaged and not overgrown with fungus in long-term storage (Sa'adah & Nurhasnawati 2015). The oven was chosen because it can keep at a controlled temperature and gave faster drying (Singh & Laishram 2010). The drying flower then mashed up into a homogenous powder to expand the contact between the solvent and the simplicial. This texture can speed up the extraction process because it was enlarging the contact between the powder

and the solvent (Depkes RI 1989).

The extraction of phytochemicals from *Kecombrang* was done using maceration method. Maceration was chosen because it is a very simple method and could be used for the extraction of thermolabile compounds (Zhang et al. 2018). *Kecombrang* flower powder as much as 500 g were macerated using 70% ethanol as a solvent with a concentration of 1:10. Ethanol was used because it can attract more polyphenol compounds than others (Tiwari et al. 2011). *Kecombrang* flower is high in containing polyphenol compounds, so it is best to use ethanolic solvents (Farida & Maruzy 2016). 70% ethanol is known more polar than pure ethanol because adding water to pure ethanol up to 30% could increase the polarity of ethanol. According to Velavan (2015), the higher concentrations of bioactive flavonoid compounds were detected in 70% ethanol due to their higher polarity than pure ethanol. Moreover, the higher the solvent polarity, the yield obtained will also increase (Noviyanty et al. 2019). The maceration was done 1 x 24 hours with occasional stirring. Remaceration was done for 2 x 24 hours using the same solvent. The filtrate from maceration then evaporated using rotary evaporator into a thick extract (Depkes RI 1995).
an the ethyl acetate's against *B. cereus* and *E. coli.*

Based on that background, this study was done to determine the antibacterial activity from the ethanol extract of *Kecombrang* flower against *S. epidermidis* and *P. acnes*. Since *Kecombrang* flower is high in containing polyphenol compounds, so it is best to use ethanolic solvents (Farida & Maruzy 2016). Ethanol can attract more polyphenol compounds than others (Tiwari et al. 2011). The outcome of the study is expected to show that ethanol extract of *Kecombrang* flower can be used as an alternative for acne treatment. This study was done by several steps, namely, plant source and preparation, extraction, phytochemical screening, disk diffusion test, and dilution method to determine Minimum Inhibitory Concentration (MIC).

The *Kecombrang* flower used in this research was obtained from *Kecombrang* plantation in Lubuk Begalung, Padang City, West Sumatra. About eight kg of the flowers was washed with clean water and cut into small pieces, then placed in a container and spread evenly for the drying process. The flowers were dried in the oven with a temperature of 40-50 ºC for 3 x 24 hours. The purpose of drying is to get the simplicia of flower that is not easily damaged and not overgrown with fungus in long-term storage (Sa'adah & Nurhasnawati 2015). The oven was chosen because it can keep at a controlled temperature and gave faster drying (Singh & Laishram 2010). The drying flower then mashed up into a homogenous powder to expand the contact between the solvent and the simplicial. This texture can speed up the extraction process because it was enlarging the contact between the powder and the solvent (Depkes RI 1989).

The extraction of phytochemicals from *Kecombrang* was done using maceration method. Maceration was chosen because it is a very simple method and could be used for the extraction of thermolabile compounds (Zhang et al. 2018). *Kecombrang* flower powder as much as 500 g were macerated using 70% ethanol as a solvent with a concentration of 1:10. Ethanol was used because it can attract more polyphenol compounds than others (Tiwari et al. 2011). *Kecombrang* flower is high in containing polyphenol compounds, so it is best to use ethanolic solvents (Farida & Maruzy 2016). 70% ethanol is known more polar than pure ethanol because adding water to pure ethanol up to 30% could increase the polarity of ethanol. According to Velavan (2015), the higher concentrations of bioactive flavonoid compounds were detected in 70% ethanol due to their higher polarity than pure ethanol. Moreover, the higher the solvent polarity, the yield obtained will also increase (Noviyanty et al. 2019). The maceration was done 1 x 24

hours with occasional stirring. Remaceration was done for 2 x 24 hours using the same solvent. The filtrate from maceration then evaporated using rotary evaporator into a thick extract (Depkes RI 1995).

The yield extract in this research was about 17.6%. The yield of an extract shows the number of active compounds extracted from the sample. The higher the yield extract, the active compounds contained in the extract also bigger (Harborne 1984; Hasnaeni et al. 2019). The calculation of the yield extract showed in Table 1.

Table 1. Calculation Yield of *Kecombrang* Flower Powder and Extract.

Sample	Flower powder (g)	Extract (g)	Yield (%)
Kecombrang flower	500	88	17.60

The extracted phytochemical was then screened for phytochemical contents based on Materia Medika Indonesia (Depkes RI 1989) and Pandey & Tripathi (2014). Screening includes testing for alkaloids, flavonoids, tannins, saponins, steroids, and triterpenoids. The results of the phytochemical screening showed that *Kecombrang* flower extract contains flavonoid, saponin, and tannin. While for alkaloid and steroid/triterpenoid, it shows negative results. These results following with Silalahi (2017) and Juwita et al. (2018), which stated that *Kecombrang* contains many secondary metabolites from terpenoids and phenolic groups, whereas alkaloid groups have not been reported. The phenolic compounds found in *Kecombrang* are flavonoid, saponin, tannin, and polyphenol. The result of the phytochemical screening showed in Table 2.

Table 2. Phytochemical Screening of *Kecombrang* Flower Extract.

Metabolites	Phytochemical screening	
	Powder	extract
Alkaloid	-	-
Flavonoid	+	+
Saponin	+	+
Tannin	+	+
Steroid/ Triterpenoid	-	-

(+) : contain tested metabolite; (-) : do not contain tested metabolites

The presence of flavonoids, saponin, and tannin in this research indicates that *Kecombrang* flower extract has the potential as an antimicrobial. As stated by Juwita et al. (2018) that the high potential antimicrobial activity possessed by *Kecombrang* is due to the presence of flavonoid, phenolic, and terpenoid contents.

The antibacterial activity test was carried out by the disk diffusion method on Mueller Hinton Agar (MHA). The MHA was poured into a petri dish and allowed to stand until the media hardens. After the media has hardened, the suspension of bacteria was pipetted about 1 ml onto a petri dish and spread evenly. The Bacterial suspension (9 x 10^7 CFU/ml) was suspended in NaCl 0.9%. After the bacterial suspension dried, sterile disk paper was inserted into a petri dish and 20 µl of the extract was dropped. The concentration of the extract was using 10%, 20%, 40%, and 80% based on

MacKeen et al. (1997) and Kusumawati et al. (2015) with modification. They tested the extract of *Kecombrang* flower and *Kecombrang* leaf (respectively) in concentrations of 20%, 40%, and 80% which showed positive results to the tested bacteria. The tested disks were then incubated for 24 hours at 37°C. The diameter of the inhibition zone was measured using a calliper. The clear zone that appears around the disk was measured as the inhibition zone (IZ) (Hudzicki 2016). The inhibition zones were analyzed statistically by Two-Way Anova and Least Significance Different (LSD) using MS Excel 2010 to see the significant effect of the differences concentrations.

The positive control used in this research is two antibiotics, named ciprofloxacin for *S. epidermidis* and clindamycin for *P. acnes*. Antibiotics are known as a substance that can inhibit the growth of bacteria (Pratiwi 2008). Ciprofloxacin is known as a broad-spectrum antibiotic from quinolone group. Quinolones are known can inhibit or disrupt the nucleic acid synthesis of the bacterial cell (Hogg 2005; Vidyavathi & Srividya 2018). Meanwhile, clindamycin is known as an antibiotic that can inhibit the protein synthesis of the bacteria. Clindamycin usually prescribes for acne treatment (Smieja 1998; Walsh et al. 2016). The use of different antibiotics in this research was due to in preliminary study ciprofloxacin gave a very wide inhibition zone in the dish containing *P. acnes*, so it can create ambiguous results. The negative control used in this test is DMSO 10% (v/v). DMSO is a surfactant that can dissolve polar and nonpolar materials. It also showed no antibacterial activity (de Brito et al. 2017). The measurements of the inhibition zone showed in Figure 1 and Table 3.

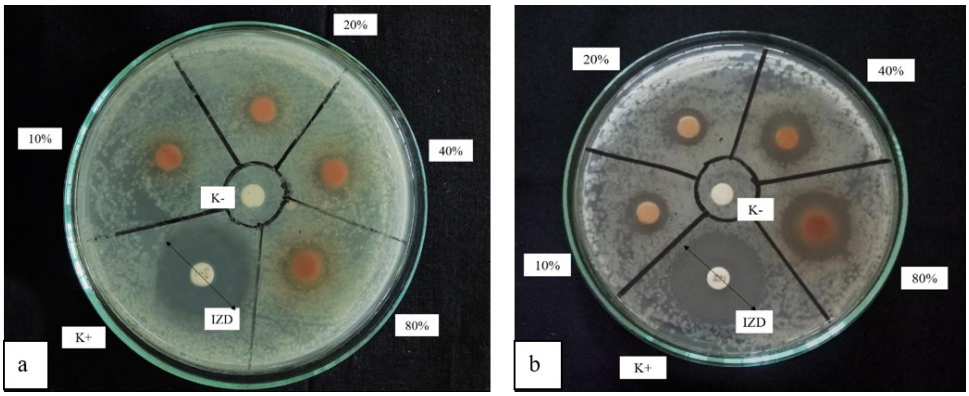

Figure 1. Inhibition zones of ethanol extract of *Kecombrang* flower (*Etlingera elatior* (Jack) R.M.Smith against (a) *S. epidermidis* and (b) *P. acnes*.; IZD: Diameter of Inhibition Zone; K+: positive control; K-: negative control; 10%, 20%, 40%, and 80% indicate the percentage test concentration of the ethanol extract of *Kecombrang* flower.

Table 3. Inhibition Zone (IZ) of *Kecombrang* Flower Extract against *Staphylococcus epidermidis* and *Propionibacterium acnes*.

Bacteria	Inhibition Zone (IZ) (mm)					
	10%	20%	40%	80%	Positive control	Negative control
Staphylococcus epidermidis	10.62±0.06^{b}	11.41±0.06^{b}	12.44±0.12^{b}	14.41± 0.02^{c}	28.67*± 0.17^{d}	-^{a}
Propionibacterium acnes	11.25± 0.05^{b}	11.46± 0.02^{b}	14.51±0.06^{b}	19.37± 0.11^{c}	26.31$^{\#}$±0.06^{d}	-^{a}

10%, 20%, 40%, and 80%: concentration of *Kecombrang* flower extract; *: Ciprofloxacin 5 µg; #: Clindamycin 10 µg; Negative control: DMSO 10%; -: no inhibition zone; inhibition values are expressed as mean ± standard deviation. Different superscript letters show the significant differences ($p < 0,05$), and the same letters show no significant differences in the row.

The data in Table 3 showed that the ethanol extract of *Kecombrang* flower has the activity to inhibit the growth of *S. epidermidis* and *P. acnes*. The results showed that the inhibition zone became larger in line with the greater concentration of the extract. The Inhibition zone at a concentration of 80% showed significant inhibition against those two bacteria compared to other concentrations. The difference in diameter of inhibition zones at each concentration possibly was due to differences in the magnitude of active substances contained in the concentration. The active compounds in higher concentrations are more than the opposite (Lingga et al. 2016). Besides that, the size of the inhibition zone was also influenced by the level of sensitivity of the organism, the culture medium, the incubation conditions, and the diffusion rate of the antibacterial compound (Fitriah et al. 2017). Bacteria have different sensitivity against antibacterial agents. Usually, Gram-positive bacteria are more sensitive to antibacterial agents compared to Gram-negative bacteria. The Gram-negative bacteria have an outer membrane in their cell wall that is not easily penetrated or damaged by certain antibacterial compounds (Breijyeh et al. 2020). The culture medium can influence the inhibition zone because different nutrient contents in media are affecting the bacterial growth. The Composition of culture media exercises a key effect on the susceptibilities of microorganisms, such as free from inhibitor content or the magnesium levels (Daoudi et al. 2020). Besides the nutrient contents in media, agar thickness and inoculum volume can influence the diffusion rate of the antibacterial agent. The higher the thickness of agar layer, the smaller the zone diameter. Similarly, high inoculum concentration shows hazy growth on the media, whereas a low concentration of inoculum shows light and immeasurable zone (Dafale et al. 2016). The particle volume also can influence the diffusion rate. Small particles will diffuse faster than large ones. Other factor that influences the inhibition zone is the incubation conditions, such as pH and temperature. Higher temperatures will increase the diffusion process. The pH of a solvent must be adjusted to neutrality (6.0 – 8.0) or dissolved in buffer solutions because the bacterial growth might be inhibited in too acid or too alkaline media (Valgas et al. 2007). Antibacterial agents have four mechanisms to inhibit the growth of bacteria. First, by inhibiting the growth of cell walls. Secondly, changing the cell membrane permeability; then inhibits the protein synthesis; and the nucleic acids (Hogg 2005). According to Fitriah et al. (2017), each group of compounds can have different effects in inhibiting bacterial growth. The difference in the activity that occurs is caused by secondary metabolites contained having synergistic energy that is different depending on the nature and morphology of bacteria.

The *Kecombrang* flower extract in this research contains flavonoid, tannin, and saponin, which has different mechanisms to inhibit the growth of bacteria. According to Juwita et al. (2018), flavonoid compounds in *Kecombrang* (*Etlingera elatior* (Jack) R.M.Smith) have antibacterial activity by targeting of membrane cell wall due to its capability to composite with extracellular and soluble proteins. This mechanism is similar to saponin, which also attacks the bacterial cell membrane. Saponin can dissolve lipids in bacterial cell membranes (lipoprotein), which causes the bacterial cell to become lysis and death (Syafriana et al. 2019). Meanwhile, tannins act by disturbed the DNA gyrase, which is an enzyme that plays a role in DNA replication (Khameneh et al. 2019). However, we cannot determine yet which compounds have a significant effect in inhibiting bacterial growth because this research has only qualitative phytochemical data, not a quantitative one. So, we cannot find out whether these compounds are equal in quantity or one is higher than another.

The data in Table 3 also showed us that a minimum concentration (10%) can inhibit the two bacteria. Because of that, the antibacterial test

continued to the Minimum Inhibitory Concentration (MIC) value test. MIC value is the lowest concentration value that can inhibit bacterial growth (Dafale et al. 2016). The test was carried out using the solid dilution method which is observing the growth of the bacteria at the lowest concentration of IZ results onto agar media. The concentration used were 10%, 8%, 6%, 4%, and 2%. The liquid medium was mixed with 1 ml of bacterial suspension and 1 ml of predetermined extract concentration. The mixture of cell suspension, media, and extract was then homogenized and incubated for 24 hours at 37^0C. The results of the incubation were then observed for the presence or absence of bacterial colony growth on the media. If there is bacterial growth, the extract cannot inhibit bacterial growth. However, if the media remains clear, it shows that the extract could inhibit bacterial growth (Pratiwi 2008; Hudzicki 2016). The data of MIC was showed in Table 4.

Table 4. Minimum Inhibitory Concentration (MIC) Test of *Kecombrang* Flower Extract Against *Staphylococcus epidermidis* and *Propionibacterium acnes*.

Extract Concentrations	Bacteria	
	Staphylococcus epidermidis	*Propionibacterium acnes*
10 %	-	-
8 %	-	-
6%	-	-
4 %	-	-
2 %	+	-

-: no growth; +: growth

Data in Table 4 showed that MIC in *S. epidermidis* is at a concentration of 4% because, in a concentration of 2%, the bacteria showed growth. Meanwhile, the MIC value of *P. acnes* cannot be determined yet. It's due to at the lowest concentration of the test (2%), the *P. acnes* still showed no growth. The data in Tables 3 and 4 showed that *P. acnes* was more sensitive than *S. epidermidis* against the extract. This data aligned with Nishijima et al. (2000), which reported that *S. epidermidis* indeed more resistant than *P. acnes* when testing against several antibiotics. Based on that data, the tested concentration to *P. acnes* should be below 2% until it shows a MIC value.

In conclusion, the ethanol extract of *Kecombrang* flower (*Etlingera elatior* (Jack) R.M.Smith) can inhibit the growth of *S. epidermidis* and *P. acnes*. This data showed a potency of *Kecombrang* flower extract as an antibacterial agent against *S. epidermidis* and *P. acnes* that causes acne vulgaris. This research is a preliminary study, so further research is needed to ensure a more valid antibacterial activity, such as determining levels of flavonoid, tannin, and saponin. Besides that, it is also necessary to study the mechanism of inhibition or cell damage caused by the secondary metabolites to prove that the mechanisms mentioned above are suitable.

AUTHORS CONTRIBUTION

VS and RNP designed the study and carried out the laboratory work. VS, YSD, and RNP analysed the data and write the manuscript.

ACKNOWLEDGMENTS

This research was supported by the Institution Research Fund 2019, Board of Research and Community Service, National Institute of Science and Technology.

CONFLICT OF INTEREST

The authors declare there is no conflict of interest.

REFERENCES

Abdallah, E. M., 2011. Plants: An alternative source for antimicrobials. *Journal of Applied Pharmaceutical Science*, 6, pp. 16–20.

Adawiyah, J., Priya, G. G., & Roshidah, B., 2010. Oral antibiotics in acne vulgaris: therapeutic response over 5 years. *Malaysian Family Physician*. Available at: http://www.e-mfp.org/.

Blaskovich, M. A. T. et al., 2019. In vitro Antimicrobial Activity of Acne Drugs Against Skin-Associated Bacteria. *Scientific Reports*, 9(1). doi: 10.1038/s41598-019-50746-4.

Breijyeh, Z., Jubeh, B., & Karaman, R., 2020. Resistance of Gram-negative bacteria to current antibacterial agents and approaches to resolve it. *Molecules*, 25(6), pp. 1-23. doi: 10.3390/molecules25061340.

Chessa, D., Ganau, G., & Mazzarello, V., 2015. An overview of *Staphylococcus epidermidis* and *Staphylococcus aureus* with a focus on developing countries. *Journal of Infection in Developing Countries*, 9(6), pp. 547–550, doi: 10.3855/jidc.6923.

Christensen, G. J. M. et al., 2016. Antagonism between *Staphylococcus epidermidis* and *Propionibacterium acnes* and its genomic basis. *BMC Genomics*. BioMed Central Ltd., 17(1). doi: 10.1186/s12864-016-2489-5.

Dafale, N. A., Semwal, U. P., Rajput, R. K., & Singh, G. N., 2016. Selection of appropriate analytical tools to determine the potency and bioactivity of antibiotics and antibiotic resistance. *Journal of Pharmaceutical Analysis*, 6(4), pp. 207–213. doi: 10.1016/j.jpha.2016.05.006.

Daoudi, Y. et al., 2020. Influence of culture media on the Kirby-Bauer method for testing antibacterial activity against Pseudomonas aeruginosa (HO80). *International Journal of Natural Resources*, 1(1), pp. 21-28.

De Brito, R.C., da Silva, G.N, Farias, T.C., Ferreira, P.B., & Ferreira S.B., 2017. Standardization of the safety level of the use of DMSO in viability assays in bacterial cells, paper presented at the *Mol2Net International Conference on Multidisciplinary Sciences*, Biscay, November 2017.

Departemen Kesehatan Republik Indonesia., 1989. *Materia Medika Indonesia Jilid V [The Indonesian Materia Medica 5th Volume]*. Direktorat Pengawasan Obat dan Makanan. Jakarta.

Departemen Kesehatan Republik Indonesia., 1995. *Farmakope Indonesia Edisi IV [The Indonesian Pharmacopeia 4th Edition]*. Departemen Kesehatan Republik Indonesia, Jakarta.

Dikicier, B.S., 2019. Topical treatment of acne vulgaris: Efficiency, side effects, and adherence rate. *Journal of International Medical Research*, pp. 1-6.

Doğan, B., Bektöre, B., Karabacak, E., & Özyurt, M., 2017. Resistance status of antibiotics in Gram-positive bacteria isolated from acne lesions in Istanbul, *Turkderm Deri Hastaliklari ve Frengi Arsivi. Istanbul Assoc. of Dermatology and Venerology,* 51(2), pp. 32–36, doi: 10.4274/turkderm.23169.

Effendi, K. et al., 2019. Analysis of bioactive components and phytochemical of powders stem and leaves of *Kecombrang* (*Etlingera elatior*), in *IOP Conference Series: Earth and Environmental Science*. Institute of Physics Publishing. doi: 10.1088/1755-1315/406/1/012003.

Farida, S., & Maruzy, A., 2016. *Kecombrang* (*Etlingera elatior*): Sebuah tinjauan penggunaan secara tradisional, fitokimia dan aktivitas farmakologinya [Torch Ginger: A review of its tradtional uses, phytochemistry and pharmacology]. *Jurnal Tumbuhan Obat Indonesia [The Journal of Indonesian Medicinal Plant]*, 9(1), pp. 19-28, doi: 10.22435/toi.v9i1.6389.19-28.

Fitriah, Mappiratu, & Prismawiranti, 2017. Uji aktivitas antibakteri ekstrak daun tanaman Johar (*Cassia siamea* Lamk.) dari beberapa tingkat kepolaran pelarut [Antibacterial activity test of Johar plant leaf extract (*Cassia siamea* Lamk.) using several levels of solvent polarity]. *Kovalen,* 3 (3), pp. 242-251.

Ghasemzadeh, A. et al., 2015. Secondary metabolites constituents and antioxidant, anticancer and antibacterial activities of *Etlingera elatior* (Jack) R.M.Sm grown in different locations of Malaysia. *BMC Complementary and Alternative Medicine*. BioMed Central Ltd., 15(1). doi: 10.1186/s12906-015-0838-6.

Harborne, J.B., 1984. *Phytochemical methods: A guide to of modern techniques of plant analysis, 2nd ed.,* Chapman Hall, London.

Hasnaeni, Wisdawati, & Usman, S., 2019. Pengaruh metode ekstraksi terhadap rendemen dan kadar fenolik ekstrak tanaman Kayu Beta-Beta (*Lunasia amara* Blanco) [The effect of extraction method on yield value and phenolic content of Beta-Beta (*Lunasia amara* Blanco) bark extract. *Jurnal Farmasi Galenika [Galenika Journal of Pharmacy]*, 5(2), pp. 175-182.

Hogg. S., 2005. *Essential Microbiology*. John Wiley & Sons Ltd., West Sussex, England.

Hudzicki, J., 2016. *Kirby-Bauer Disk Diffusion Susceptibility Test Protocol*, American Society for Microbiology, viewed 29 August 2018, from https://asm.org/getattachment/2594ce26-bd44-47f6-8287-0657aa9185ad/Kirby-Bauer-Disk-Diffusion-Susceptibility-Test-Protocol-pdf.pdf.

Juwita, T., Puspitasari, I. M., & Levita, J., 2018. Torch ginger (*Etlingera elatior*): A review on its botanical aspects, phytoconstituents and pharmacological activities. *Pakistan Journal of Biological Sciences*, 21(4), pp. 151–165, doi: 10.3923/PJBS.2018.151.165.

Khameneh, B., Iranshahy, M., Soheili, V., & Bazzaz, B. S. F., 2019. Review on plant antimicrobials: A mechanistic viewpoint. *Antimicrobial Resistance and Infection Control.*, 8(118), pp. 1-28, doi: 10.1186/s13756-019-0559-6.

Kusumawati, E. et al., 2015. Uji aktivitas antibakteri ekstrak etanol daun *Kecombrang Etlingera elatior* (Jack) R.M.Sm *terhadap Salmonella typhi* [Antibacterial activity test of ethanol extract of *Kecombrang* leaf *Etlingera elatior* (Jack) R.M.Sm against *Salmonella typhi*], *Jurnal Ilmiah Manuntung*. 1 (1), pp. 1-7.

Lingga, A. R., Pato, U., & Rossy, E., 2016. Uji antibakteri ekstrak batang *Kecombrang (Nicolaia speciosa* Horan*)* terhadap *Staphylococcus aureus* dan *Escherichia coli* [Antibacterial test of *Kecombrang* (*Nicolaia speciosa* Horan) stem extract against *Staphylococcus aureus* and *Escherichia coli*]. *JOM Faperta*, 3(1), pp. 1-15.

Mackeen, M. M. et al*.,* 1997. Antimicrobial and cytotoxic properties of some Malaysian traditional vegetables (ULAM). *Pharmaceutical Biology*. Swets en Zeitlinger B.V., 35(3), pp. 174–178. doi: 10.1076/ phbi.35.3.174.13294.

Nakase, K., Nakaminami, H., Takenaka, Y., Hayashi, N., Kawashima,M., & Noguchi, N., 2014. Relationship between the severity of acne vulgaris and antimicrobial resistance of bacteria isolated from acne lesions in a hospital in Japan. *Journal of Medical Microbiology*, 63(Part 5), pp. 721–728, doi: 10.1099/jmm.0.067611-0.

Nakatsuji, T. et al., 2009.Antimicrobial property of lauric acid against *Propionibacterium acnes*: Its therapeutic potential for inflammatory acne vulgaris. *Journal of Investigative Dermatology*, 129(10), pp. 2480–2488. doi: 10.1038/jid.2009.93.

Naufalin, R. & Rukmini, H. S., 2018.,Antibacterial activity of *Kecombrang* flower extract (*Nicolaia speciosa*) microencapsulation with food additive materials formulation, in *IOP Conference Series: Earth and Environmental Science*. Institute of Physics Publishing. doi: 10.1088/1755-1315/102/1/012035.

Neves, J.R., Francesconi, F., Costa, A. Ribeiro, B.M., Follador, I., & Almeida, L.M.C., 2015. *Propionibacterium acnes* and bacterial resistance. *Surg Cosmet Dermatol*, 7(3 Suppl 1): S27-38.

Nishijima, S., Kurokawa, I., Katoh, N., & Watanabe, K., 2000. The bacteriology of acne vulgaris and antimicrobial susceptibility of *Propionibacterium acnes* and *Staphylococcus epidermidis* isolated from acne lesions. *The Journal of Dermatology,* 27, pp. 318-323.

Noviyanty, A., Salingkat,C.A., Syamsiar, 2019. Pengaruh jenis pelarut terhadap ekstraksi dari kulit buah naga merah (*Hylocereus polyrhizus*) [The effect of solvent type to the quality of red dragon fruit peel (*Hylocereus polyrhizus*) extracts]. *Kovalen*, 5(3), pp. 271-279.

Pandey, A., & Tripathi, S., 2014. Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. *Journal of Pharmacognosy and Phytochemistry*, 2(5), pp. 115-119.

Pratiwi, S. T., 2008. *Mikrobiologi Farmasi [Pharmaceutical Microbiology]*. Penerbit Erlangga, Jakarta.

Sa`adah, H., & Nurhasnawati, H., 2015. Perbandingan pelarut etanol dan air pada pembuatan ekstrak umbi bawang Tiwai (*Eleutherine americana* Merr.) menggunakan metode maserasi [Comparison of ethanol and water solvents in making Tiwai bulb extract (*Eleutherine americana* Merr.) using the maceration method]. *Jurnal Ilmiah Manuntung*, 1(2), pp. 149–153.

Sadeek, A. M., & Abdallah, E. M., 2019. Phytochemical Compounds as Antibacterial Agents: A Mini Review. *Saudi Arabia Glob J Pharmaceu Sci*, 53(4). doi: 10.19080/GJPPS.2019.07.555720.

Silalahi, M., 2017. Senyawa metabolit sekunder pada *Etlingera elatior* (Jack) R. M. Smith [Secondary metabolite compounds in *Etlingera elatior* (Jack) R. M. Smith], paper presented at *Seminar Nasional Pendidikan Biologi dan Saintek [National Seminar on Biological Education and Sciencetech]*, Surakarta, May 20th 2017.

Singh, A., & Laishram, N., 2010. Drying of flowers and other ornamental plant parts in India. *Floriculture and Ornamental Biotechnology*, 4(1), pp. 72-78.

Smieja, M., 1998. Current indications for the use of clindamycin: A critical review. *Can J Infect Dis*, *9*(1), pp. 22–28.

Syafriana, V., Ningsih, W., & Wahidin., 2019. Uji aktivitas antibakteri metanol daun hijau tanaman pucuk merah (*Syzygium myrtifolium* Walp.) terhadap *Pseudomonas aeruginosa* dan *Staphylococcus epidermidis* [Antibacterial activity test from green leaves methanol extract of red shoot plant (*Syzygium myrtifolium* Walp.) against *Pseudomonas aeruginosa* and *Staphylococcus epidermidis*], paper presented at *Seminar Nasional Pemanfaatan Bahan Alam sebagai Obat, Kosmetik dan Pangan Fungsional [National Seminar on Utiklization of Natural Ingredients as Medicine, Cosmetics, and Functional Food]*, Jakarta, June 29Th 2019. http://repository.istn.ac.id/id/eprint/859

Tabri, F., 2019. The association between *Staphylococcus epidermidis* and palmitic acid level in patients with acne vulgaris. *Surgical and Cosmetic Dermatology. Sociedade Brasileira de Dermatologia* 1, 11(2), pp. 105–109. doi: 10.5935/scd1984-8773.20191121382.

Tan, A. U., Schlosser, B. J., & Paller, A. S., 2018. A review of diagnosis and treatment of acne in adult female patients. *International Journal of Women's Dermatology*, 4, pp. 56–71.

Tiwari, P., Kumar, B., Kaur, M., Kaur, G., & Kaur, H., 2011. Phytochemical screening and Extraction: A Review. *Internationale Pharmaceutica Scientia,* 1(1), pp. 98-106.

Valgas, C. et al., 2007. Screening methods to determine antibacterial activity of natural products. *Brazilian Journal of Microbiology*, 38(2), pp. 369–380. doi: 10.1590/S1517-83822007000200034.

Velavan, S., 2015. Phytochemical techniques: A review. *World Journal of Science and Research*, 1(2), pp. 80-91.

Vidyavathi, M., & Srividya, G., 2018. A review on ciprofloxacin: Dosage form perspective, *International Journal of Applied Pharmaceutics*, *10*(4), 6–10. https://doi.org/10.22159/ijap.2018v10i4.25315

Walsh, T. R., Efthimiou, J., & Dréno, B., 2016. Systematic review of antibiotic resistance in acne: An increasing topical and oral threat. *The Lancet Infectious Diseases*, *16*(3), e23–e33. https://doi.org/10.1016/S1473-3099(15)00527-7

Wijekoon, M. M. J. O. et al., 2013. Chemical composition and antimicrobial activity of essential oil and solvent extracts of torch ginger inflorescence (*Etlingera elatior* Jack.). *International Journal of Food Properties*, 16(6), pp. 1200–1210. doi: 10.1080/10942912.2011.579674.

Zhang, Q. W., Lin, L. G., & Ye, W. C., 2018. Techniques for extraction and isolation of natural products: A comprehensive review. *Chinese Medicine (United Kingdom)*, 13(20), pp. 1-26, doi: 10.1186/s13020-018-0177-x.