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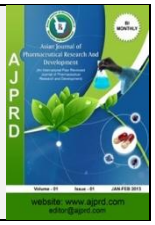

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Research Article

Antibacterial Activity of Ethanolic Extract of Kitolod (*Hippobromalongiflora*) Leaf Against *Staphylococcus aureus* and *Salmonella typhi*

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A B S T R A C T

Objective: The purpose of this study was to determine at what concentration of ethanol extract of kitolod leaves is active against Staphylococcus *aureus* and *Salmonella typhi*.

Methods: Ethanolic extract of Kitolod leaves was tested for phytochemical screening by using standard protocol. Antibacterial testing was using the diffusion disc method to measure the inhibition zone against the *Staphylococcus aureus* and *Salmonella typhi* with various concentration of Kitolod leaves extract (6.25%, 12.5%, 25%, 50%, and 75%).

Results: Phytochemical screening showed that ethanolic extract of Kitolod leaves contain alkaloids, flavonoids and saponins. The antibacterial inhibition of ethanol extract of kitolod leaves against Staphylococcus aureus and Salmonella typhi bacteria at a concentration of 75% had a diameter of 11.3 mm and 12.16 mm with a strong category.

Conclusions: Kitolod leaf could be use as a novel antibacterial agent.

Keywords: Extract, Antibacterial, Kitolod, *Hippobromalongiflora*,*S. aureus, S. typhi*

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INTRODUCTION

Medicinal plants have been known to cure various diseases since ancient times. This is due to the potential of chemical compounds found in these plants that can be used to synthesize new conventional medicines, one of which is the plant kitolod *Hippobroma longiflora* (L.) G. Don[1] . Kitolod plants are used to treat toothache, asthma, bronchitis, laryngitis, cataracts, wound medication, and cancer drugs[2].Ethanol extract of kitolod leaf contain secondary metabolite compounds namely alkaloids, flavonoids, polyphenols, monoterpenoids, sesquiterpenoids, quinins and saponins[3]. According to Ibrahaim explains that the content of secondary metabolites in plants Chitolod has activities such as antioxidants, cytotoxics, anticancer, anti-inflammatory and antimicrobial[4].*Staphylococcus aureus* is one of the important pathogenic bacteria associated with toxin virulence, invasive, and the body's resistance to antibiotics. *Staphylococcus aureus* infection is the main cause of skin, soft tissue, respiratory, bone, joint and endovascular disorders[5]. *Salmonella typhi* is a pathogen that causes typhoid (enteric) fever, which is a systemic infection in humans caused by *Salmonella* serotypes, including *Salmonella serotype Typhi*[6].This research is expected to provide information on secondary metabolite compounds found in chitolod leaves and the antibacterial activity of ethanol extracts of kitolod leaves against *Staphylococcus aureus* and *Salmonella typhi.*

MATERIAL AND METHODS

Plant preparation

Fresh kitolod leaves was collected from local area of from Bakal Julu Village (Dairi Regency North Sumatra Medan, Indonesia), and authenticated by Indonesian Institute of Sciences: Research Center ForBiology (No. 2347/MEDA/2019). Voucher specimen was deposited in the Pharmacognosy Laboratory, Sekolah Tinggi IlmuKesehatan Senior Medan.

Extraction of kitolod leaves

Kitolod leaves simplicia powder was extracted using maceration method with 96% ethanol solvent. Maceration is done by soaking the simplicia of kitolod leaves for 3 days with occasional stirring. The procedure is repeated

until the color is clear. The results of maceration (maserate) were evaporated using a rotary evaporator to obtaincrude extract of Kitolod leaves.

Phytochemical screening of various lotus leaf extract

The crude extract of kitolod leaves was screening by using the standard protocol to know the presence of phytochemical compounds[7].

Antibacterial test

Preparation of antibacterial test will begin with sterilizing the tools and materials to be used, rejuvenation of bacteria, making media, making bacterial suspense, making kitolod leaf extract test solutions and making comparative standard solutions.The determination of the antibacterial activity was carried out with sterilized NA media inserted into 20 mL sterile petri dishes each and allowed to condense at room temperature. The media was dropped with 0.1 mL of bacterial suspension test and flattened using an L bar until smooth and dry. Sterile disk paper with a diameter of 6 mm was dropped with ethanol extract 96% of kitolod leaves as much as 10 µL with each concentration of 6.25%, 12.5%, 25%, 50%, and 75% and then placed on the media so that the solid that had been dripped with a test bacterial suspension, DMSO 10% as a negative control, and chloramphenicol as a positive control. Then incubated at 37°C for 24 hours and after incubation the clear zone was measured using calipers, three replications were performed.

RESULT AND DISCUSSION

Phytochemical Screening

Phytochemical screening results of kitolod leaves ethanol extract showed different phytochemical compounds (Table 1).

Table 1. Phytochemical screening of Kitolod leaves ethanolic extract

No	Screening	Reagent	Ethanolic Extract
1	Alkaloids	Mayer	Positive
		Dragenddroff	Positive
		Bouchardat	Positive
2	Flavonoids	Mg+HCl+Amyl Alcohol	Positive
3	Saponins	Foaming Test	Positive
4	Tannins	$FeCl_3$1%	Negative
5	Triterpenoid/Steroids	Liebermann Bouchard	Negative

Antibacterial test

Antibacterial activity testing of ethanol extract of kitolod leaves was carried out using the disk diffusion method, namely the determination of bacterial sensitivity with a particular substance that may have antibacterial activity using disc paper. Antibacterial testing was carried out with various concentrations of 6.25%, 12.5%, 25%, 50%, and 75%, chloramphenicol and DMSO. The results of testing the antibacterial activity of ethanol extract of chitolod leaves against *Staphylococcus aureus* and *Salmonella typhi* can be seen in Table 2 and Figure 1 below.

Table 2:. Antibacterial result of Kitolod leaves ethanolic extract

Concentration	Zone of inhibition (mm)	
	Staphylococcus aureus	*Salmonella typhi*
75%	11.3	12.6
50%	10	8.9
25%	8.83	7.6
12.5%	7.06	7.83
6,25%	0	7.66
Chloramphenicol	28.66	30.5
DMSO	0	0

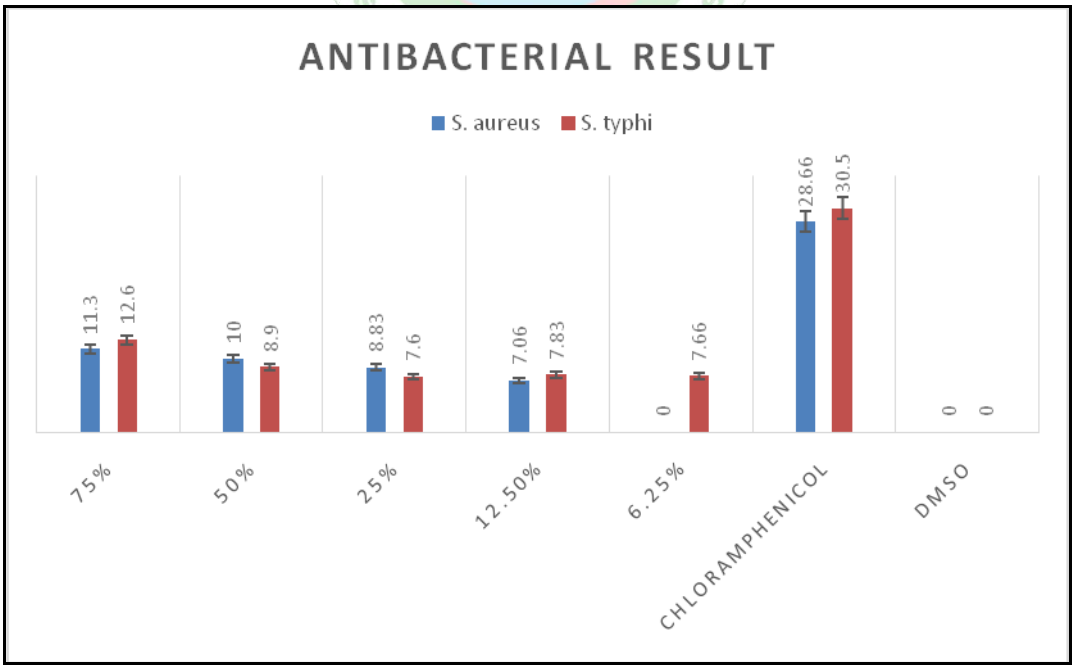

Figure 1: Antibacterial result of Kitolod leaves ethanolic extract

Based on Table 2 and Figure 1 it can be seen that 75% concentration of ethanol extract of kitolod leaves is effective in inhibiting the growth of *Staphylococcus aureus* and *Salmonella typhi* with inhibition zone diameters of 11.3 mm and 12.6 mm, which are classified as strong criteria. Whereas the concentration of 6.25% has the ability to inhibit the growth of *Salmonella typhi* bacteria compared to Staphylococcus aureus. According to Davis and Stout, explain that the criteria for antibacterial inhibition consist

of ≥ 20 mm is very strong, 10-20 mm is strong, 5-10 mm is moderate and ≤ 5 mm is weak[8].

In positive controls using standard chloramphenicol with a concentration of 30 mg / ml. Chloramphenicol works by inhibiting protein synthesis in bacterial cells by reversibly binding to the 50 s ribosome subunit[9]. The negative control used was DMSO with a concentration of 10%. In this study negative control DMSO has no clear zone, so DMSO can be said to not be able to inhibit bacterial growth.

alkaloids can disrupt the constituent components of peptidoglycan on bacterial cells so that the cell wall layers are not formed intact and cause cell death. Another mechanism of antibacterial alkaloids is that the alkaloid component is known as a DNA accelerator and inhibits bacterial cell topoisomerase enzymes[11].

Flavonoids provide bacteriolytic effects, inhibit protein synthesis, DNA synthesis, RNA and damage cell membrane permeability[12]. According to Wu et al, flavonoids have antibacterial activity because of the ability of flavonoids to interact with cell membranes and affect cell membrane bioactivity and it has been reported that flavonoids are able to reduce the fluidity of bacterial cell membranes that is directly related to damage to cytoplasmic membranes or indirect damage through autolysis / weakening of the cell wall and consequently osmotic lysis[13].

Ethanol extract 96% of active chitolod leaves as antibacterial due to chemical components contained in the extract. Based on phytochemical screening, ethanol extract of kitolod leaves contain alkaloid, flavonoid, and saponin compounds.The mechanism of action of alkaloids as an antibacterial is by inhibiting the synthesis of nucleic acids, because it can inhibit the enzymes dihydrofolate reductase and topoisimerase I[10]. According to Karou et al.,

The mechanism of action of saponin as an antibacterial causes lysis of the bacterial cell wall and leakage of AKP (Alkaline Phosphate), an increase in saponin concentration causes the protein to dissolve, causing intercellular compounds to diffuse through the outer membrane and cell wall. This causes the cytoplasm to leak out of the cell resulting in cell death[14].

CONCLUSIONS

The ethanol extract of Kitolod leaves has a group of secondary metabolites including alkaloids, flavonoids, and Saponins. The ethanol extract of Kitolod leaves has effective antibacterial activity against Staphylococcus aureus and Salmonella typhi bacteria with a concentration of 75% with a strong category.

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REFERENCES

1. Baibado JT, Hon YC. Biological Functions of The Metabolites From*Euphorbia hirta* L. International Journal of Pharmaceutical and Biological Science Archive 2015; 3(1): 27–9.
2. Utami P. BukuPintarTanamanObat. Jakarta: Penerbit PT. AgromediaPustaka; 2008. p. 147-8.
3. Aqila FN, Sri Agung FK, Yopi I. Antituberculosis Activity Test of Kitolod Leaf Ethanol Extract (*Laurentia longiflora* (L). Paterm). International Journal of Scientific Engineering and Applied Science 2017;7(7).
4. Ibrahim S., Darma RR and Afrizal. Isolation and Elucidation Structure of Triterpenoids from Hippobroma longiflora Leaf Extract and Tested of Antibacterial Activity. Journal of Chemical nd Pharmaceutical Research 2017; 9(11):205–8.
5. Tong SYC, Davis JS, Eichenberger E, Holland TL, Jr VGF. *Staphylococcus aureus* Infections: Epidemiology, Pathophysiology. Clinical Microbiology Reviews 2015; 28(3):602-31.
6. Santos Rl., Shuping Z., Renee MT., Robert A.K., L. Garry A., Andreas J.B. Animal Models of Salmonella Infections: Enteritis Versus Typhoid Fever. Microbes and Infection 2001; 3:1335-44.
7. Sinaga SM, Haro G, Sudarmi S, Iksen I. Phytochemical Screening and Antihyperglycemic Activity of Ethanolic Extract of Coriandrum sativum L. Leaf 2019; 12(4):1992-6.
8. Davis WW, Stout TR. Disc Plate Method of Microbiological Antibiotic Assay: Applied and Enviromental Microbiology 2009; 22 (4):666-70.
9. Kasten, MJ. Clindamycin, Metronidazole, and Chloramphenicol. Mayo ClinicProceedings.1999; 74(8):823-833.
10. Cushine TPT, Benjamart C, LambAJ. Alkaloids: An Overview of Their Antibacterial,Antibiotic – Enhancing and Antivirulence Activities. International Journal of Antimicrobial Agents2014; 44:377–86.
11. Karou D, Savadogo A, CaniniA,Yameogo S, Montesano C, Simpore J, Colizzi V, Traore AS. Antibacterial activity of alkaloids from *Sidaacuta* 2005; 4(12):1452-7.
12. Dzoyem JP, Hiroshi H, Barthelemy N, Bonaventure TN, Kazuhisa S. Antimicrobial Action Mechanism of Flavonoids From Dorstenia Spesies. Drug Discoveries & Therapeutic 2013; 7(2):66–72.
13. Wu T, Mengying H, Xixi Z, Ying Z, Tianfu Q, Siyi P, Xiaoyun X. A StructureActivity Relationship Study of Flavonoids As Inhibitors of E. coli By Membrane Interaction Effect. Biochimica et Biophysica Acta2013;2751–6.
14. Khan MI, Abdulatef A, Jin HS, Jun SB, Min YK,Jong DK. Green Tea Seed Isolated Saponins Exerts Antibacterial Effects Against Various Strains of Gram Positive and Gram Negative Bacteria A Comprehensive Study In Vitro and In Vivo.Evidence-Based Complementary and Alternative Medicine2018;1–12.