BIOINFORMATICS AND BIOMEDICAL RESEARCH JOURNAL
2021, Vol. 4, No. 1, 32 – 38
http://dx.doi.org/10.11594/bbrj.04.01.05

Research Article

Antimicrobial Activity of *Stenochlaena palustris* and *Sauropus androgynus* in *Staphylococcus aureus*, *Escherichia coli* and *Candidia albicans*

Lia Yulia Budiarti[1]*, Isnaini[2], Puteri Dayana[3], Norma Sari[3], and Nur Almira R. S.[3]

[1]Departement of Microbiology, Medical Faculty, Lambung Mangkurat University of Banjarmasin, Indonesia
[2]Departement of Pharmacology and Threapy of Mecial Education of Medical Faculty of Lambung Mangkurat University of Banjarmasin, Indonesia
[3]Student of Medical Education of Medical Faculty Lambung Mangkurat University of Banjarmasin, Indonesia

Article history:
Submission June 2021
Revised June 2021
Accepted June 2021

**Corresponding author:*
E-mail:
lybudiarti@gmail.com

ABSTRACT

Stenochlaena palustris and *Sauropus androgynus* are known to contains antimicrobial substances such as flavonoids, saponins and tannins compounds.The purpose of this study was to analyzes the antimicrobial activity of young and old leaf infusions of *S. palustris* and *S. androgynus* leaves against *Staphylococcus aureus, Escherichia coli* and *Candida albicans.* Analyze the antibacterial activity of a single preparations with a combination preparation of *S.palustris* (SP) and *S.androgynus* (SA) leaves infusion against *S.aureus, E.coli* and *C.albicans*. Leaves of *S.palustris* young part (SP1) taken 0-10 cm from shoots and old parts (SP2) 11-20 cm from shoots, while leaves of *S.androgynus* young part (SA1) leaves number 1 - 10 from the top and the old part (SA2) leaves number 11-20 from the top. The results showed that a single infusion of SP1 75% and SP2 75%, SA1 90% and SA2 90%, and a combination of SP1 75% and SA1 75%, SP2 75% and SA2 75% have the same activity as ampicillin in *S.aureus*. Single infusion of SP1 90% and SP2 90%, SA1 90% and SA2 90%, combination of SP1 75% and SA1 80% and the combination of SP2 80% and SA2 60% have the same activity as ciprofloxacin in *E. coli*. Single infusion of SP1 90% and SP2 90%, and a combination of SP1 80% and SA1 80%, SP2 80% and SA2 80% have the same activity as ketoconazole in *C.albicans*. The difference in activity due to differences in leaf parts used only occurred in *E. coli*, whereas in *S.aureus* and *C.albicans* ($p < 0.05$).

Keywords: *Stenochlaena palustris, Sauropus androgynus, Staphylococcus aureus, Escherichia coli, Candida albicans*.

How to cite:
Budiarti, L. Y., Isnaini, Dayana, P., Sari, N. & Almira, Nur R. S. (2021). Antimicrobal Activity of *Stanochlaena palustris* and *Sauropus androgynus* in *Staphyloccus aureus, Esherichia coli* and *Candida albicans*. *Bioinformatics and Biomedical Research Journal*. *4* (1), 32 – 38. doi: 10.11594/bbrj.04.01.05

Introduction

Stenochlaena palustris (Burm.f) Bedd. and Sauropus androgynus) (L.) Merr are plants that is widely found in South Kalimantan, Indonesia. These plants are used as a vegetable. Leaves of *Stenochlaena palustris* and *Sauropus androgynus* leaves can be used as antimicrobial drugs. (6), (10), (13). The antimicorbial substances contained in the leaves of *S. palustris* which can inhibit bacterial growth are flavonoids and alkaloids, while the antimicrobial substances of the leaves of *S. androgynus (*are flavonoids, saponins and tannins (10), (17).

People also often use these two medicinal ingredients. Medicinal preparations are made by mixing two or more plants in the form of stew preparations, water extracts or infusions. The combination of two or more plants tends to cause drug interactions, because in plants it self contains a lot of active ingredients. Interaction of active ingredients contained in plants can cause synergistic effects or antagonistic effects. The beneficial effect is the synergistic one (16).

The study aimed to test the antimicrobial activity of a single preparation and a combination of infusion of *Stenochlaena palustris* leaves and *Sauropus androgynus* leaves against *staphylococcus aureus*, *Escherichia coli* and *Candida albicans* yeast. It is expected that from the results of this study obtained concentrations that provide optimum bland activity from a single preparation and a combination of infusion of *S. palustris* leaves and *S. androgynus* leaves in inhibiting the growth of *S. aureus, E. coli* and *C. albicans.*

Antibacterial activity tests are carried out by diffusion methods and the observed parameter is the quantity of the bland zone of all treatments tested against *S. aureus*, *E. coli* and *C. albicans.* This research has passed the ethical test with the certificate of the ethics commission of the Faculty of Medicine, Lambung Mangkurat University number: 840, 758-759 /KEPK-FK UNLAM/EC/ VIII / 2018.

Materials and Methods

This true experimental research method uses a posttest control group design. This in vitro study was conducted at the Microbiology Laboratory of the Faculty of Medicine, Lambung Mangkurat University, in August-October 2018. The determination test of the future *Stenochlaena palustris (Burm.f) Bedd* and *Sauropus androgynus* (L.) Merr. leaves was conducted at the Biology Laboratory of the Faculty of Mathematics and Natural Sciences, Lambung Mangkurat University.

Plant Materials

The plant materials used in this study were young leaves (DM) and old leaves (DT) from *Stenochlaena palustris (Burm.f) Bedd* and *Sauropus androgynus* (L.) Merr. Young leaves of *Stenochlaena palustris (Burm.f) Bedd* (SP1), were leaves that are at 0-10 cm from the end of the stem and old leaves of *Stenochlaena palustris (Burm.f) Bedd* (SP2) were leaves that are at 10 cm - 20 cm from the end of the stem. The young leaves of *Sauropus androgynus (L.) Merr* (SA1) were leaves no. 1-10 that counted from the end of the stem and the old leaves *Sauropus androgynus (L.) Merr* (SA2) were leaves no. 11-20 that counted from the end of the stem. The leaves collected were dried in an oven at 60℃ and grinded with a blender into fine powder.

Isolates

Isolates of *S. aureus* ATCC 25923, *E,coli* ATCC 25922, and *C. albicans* ATCC 10231 that cultured in the Microbiology Laboratory of the Faculty of Medicine, University of Lambung Mangkurat were used as research material. Positive control using ampicillin for *S. aureus*, ciprofloxacin for *E. coli* and ketoconazole for *C. albicans*.

Research Procedure

Preparation of Plant Infusion

Simplicia was put into an infusion pot containing aquadest according to the expected concentration. The pot is heated for 15 minutes at 90℃. Then, the infusion water is filtered (3).

Preparation of Plant Infusion Concentration

Preliminary test results obtained the Minimum Inhibitory Concentration (MIC) from the infusion preparations of both test parks against *S.aureus* was 45%, 50% against *E.coli* and 60% against *C. albicans*. The infusion concentration tested on *S. aureus* were 45%, 60%, 75% and

90%; in *E. coli* were 50%, 60%, 70%, 80% and 90%, while in *C. albicans* were 60%, 70%, 80% and 90%.

Preparation of microbial isolate suspension

In each culture of pure microbial isolates (*S. aureus, E.coli, C. albicans*), one scratch of the isolate was taken using the inoculating loop and inserted into a test tube containing 10 ml of physiological salt solution 0.85% (NaCl). The isolates was incubated for 24 hours. The results of the bacteria growth in the form of turbidity were standardized (compared) to the solution turbidity series, Mac Farland Standard 0,5 (equivalent to 0.5×10^8 CFU/ml of bacteria), which seen using white paper as a background. If the bacterial suspension is less turbid, more colony was added, whereas if it was more turbid, 0.85% physiological NaCl was added to the standard of turbidity.

Antimicrobial Test

Mueller Hinton agar (MHA) medium was used for the antibacterial test and Sabouraud dextrose agar (SDA) medium was used for the antifungal test. First, MHA, SDA and suspension of bacteria or fungi isolates whose turbidity had been standardized were prepared. Sterile cotton swabs were dipped into the suspension of bacteria or fungi, wait a moment so that the liquid can absorbed into the sterile cotton swabs. Then, lift the cotton swab and squeeze it by rotating it on the inner wall of the tubes. Spread the cotton swab on the surface of MHA or SDA medium until the entire surface is covered with the suspension. The spreading was done by performing 90^o rotation on the 1st spread to the 2nd spread and 45^o rotation on the 2nd spread to the 3rd spread, then the petri dish is rotated up to 45^o. Put the MHA or SDA medium which has been covered with the bacteria or fungi suspension on the table for 15 minutes to allow the bacterial or fungi suspension to diffuse into the agar.

Soak the Paper discs in single preparation and combination of *Stenochlaena palustris* (Burm.f) Bedd leaves infusion and *Sauropus androgynus (L.) Merr* leaves infusion for 15 minutes (until saturated or until paper discs are unable to absorb natural antimicrobial liquids). The soaked paper discs were then placed on an aseptic MHA or SDA medium and incubated for 24 hours at 37^oC.

The result was known by measuring the inhibitory zone diameter using calliper, the diameter of the inhibitory zone was measured from one edge to another through the centre of the paper disc and the value of the inhibitory zone was measured in millimetres.

Results and Discussion

Phytochemical screening tests by researchers previously found the presence of secondary compounds in *Stenochlaena palustris* and *Sauropus androgynus* leaf extracts that had antimicrobial activity. In the presence of relatively similar active contains compounds in *S. palustris* and *S. androgynus* leaves extract, then the incorporation of leaf extracts from these two plants can increase their effectiveness as antimicrobials.

Phytochemical analyses identified high content of fatty acids, flavonoids, and polyphenols as the major bioactive components in *S. androgynus* (19), (7), (8). Phytochemical screening results showed that ethanol extract of 90% *S. androgynus* leaves contains compounds of alkaloids, triterpenoids, saponins, tannins, glycosides polyphenols, and flavonoids (13), (15).

The presence of several secondary metabolites such as alkaloids, flavonoids, phenols, saponins, terpenoids, glycosides and tannins were detected in fractions obtained from *S. palustris* both extracts (14), (2), (4), (11). Flavonoids and phenolic compounds are secondary metabolites with a variety of biological activities, and widely found in plants (14), (11). The consumption of fruits and vegetables that contain high levels of phenolic compounds is often recommended because phenolic compounds represent the most efficient natural antioxidants, and play important roles in the prevention of cardiovascular disease, ageing, and the effective scavenging of oxygen free radicals (14), (2), (4), (11).

Flavonoids are the dominant antibacterial compounds in the leaves *Stenochlaena palustris* and *Sauropus androgynus* leaves. Flavonoid

content in later *Stenochlaena palustris* leaves amounted to 166.1779 ± 4.1420 mg QE /g and in *S. androgynu* leaves by 148.94 ± 0.05 mg QE/g (13). Flavonoids inhibited prostaglandin synthase, lipoxygenase, and cyclooxygenase enzymes, thus inhibiting the occurrence of inflammatory processes and damaging cell membranes so that cell permeability changes which can result in inhibition of cell growth (4), (11), (12). Flavonoids acted as antifungals by denaturing cell proteins and shrinking cell walls so that they can lyse fungal cell walls.

Alkaloids' mechanism as antifungal by inhibiting topoisomerase enzymes causing cell membrane leakage (5), (18), (9). The mechanism of action of alkaloids in the extract of the leaves of the future *S. palustris* and *S. androgynus* leaves is together to work as an antimicrobial (12), (18), (1). It is associated with affecting cell division by inhibiting the activity of dihydroolat reductase, thereby inhibiting the synthesis of nucleic acids (5), (18), (9), (1). Alkaloids inhibited and interfered the constituents of bacterial cell wall peptidoglycan because nitrogen in alkaloid base groups could bind to amino acids in the bacterial cell wall so that the layer of the bacterial cell wall was not formed and lead to cell death (12), (9), (1).

Saponins were a glycoside found in plants and possesed an antibacterial effect. Saponins had activities as an antimicrobial by involving the formation of sterol complexes on the plasma membrane thereby destroying semi-permeability of cell and causes cell death (1). Saponins were bioactive compounds that had antifungal activity involving the formation of sterol complexes on the plasma membrane thereby destroying semi-permeability of cells and causing cell death (5), (1).

The tannin inhibited the reverse transcriptase and DNA topoisomerase enzymes so that bacterial or fungal cells could not formed. Tannins are toxic and can affect changes in cell membrane permeability and reduce the volume of bacterial or fungus cells (2), (5), (9).

The selection of extraction solvents, generally using the principle of like dissolves like. The solvent used in this study was distilled water which has polar characteristics so that only polar or semi-polar secondary compounds can dissolve in water solvents. Polar and semi-polar secondary compounds that dissolve in water solvents such as flavonoids, phenolics, saponins, and tannins (14), (12), (9).

Stated that Previous research has stated that *Stenochlaena palustris* (SP) 10% - 30% ethanol extract could inhibit the growth of *S. aureus* and *Escherichia coli* (6), (13). Research has stated that *Sauropus androgynus (*SA) 20% ethanol extract could inhibit *S. aureus* growth (10), (13). In this study, the concentration of the inhibitory zone above 15 mm began for 45% for all single preparations. The largest combination of inhibitory zones was 29.54 mm in the combination of SP2 90% and SA2 90%. The largest inhibitory zone was larger than ampicillin which only inhibited 26.86 mm. The infusion of old leaves had larger inhibitory zone compared to young leaves both on a single preparation and in combination (Figure 1).

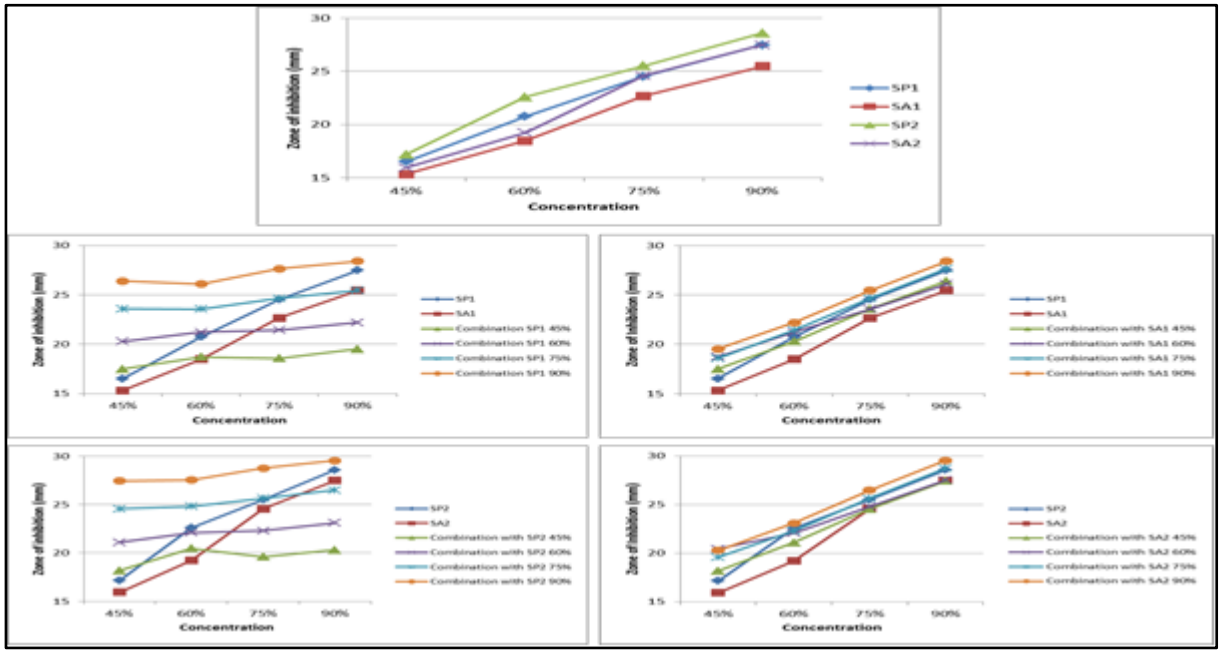

Figure 1. Inhibitory Zone of administration a single infusion and combination of Stenochlaena palustris (SP) and Sauropus androgynus (SA) leavest against Staphylococcus aureus

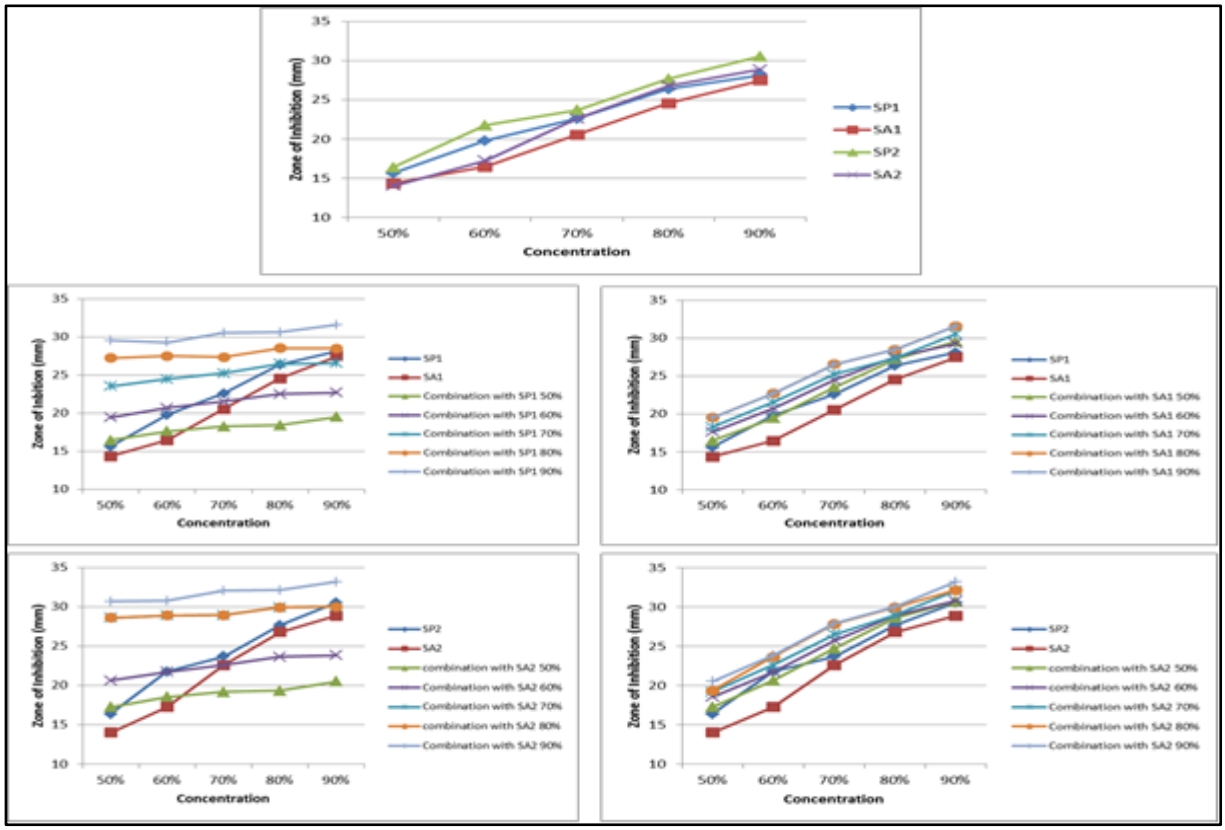

Figure 2. Inhibitory Zone of administration a single infusion and combination of Stenochlaena palustris (SP) and Sauropus androgynus (SA) leavest against Escherichia coli

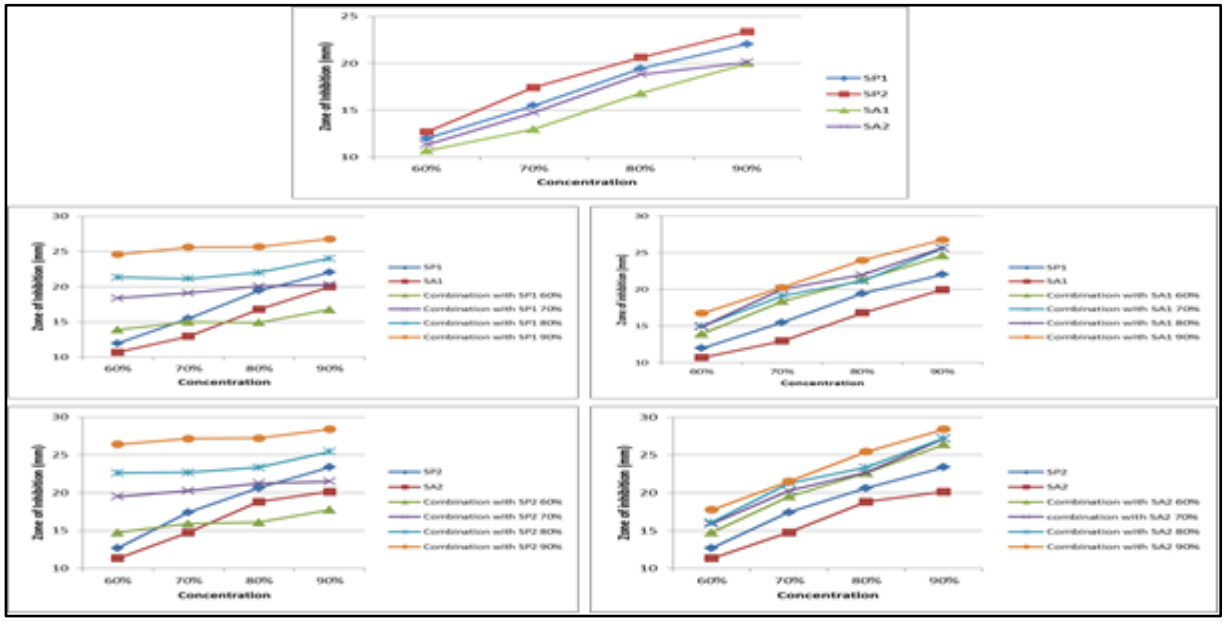

Figure 3. Inhibitory Zone of administration a single infusion and combination of Stenochlaena palustris (SP) and Sauropus androgynus (SA) leavest against Candida albicans

The inhibitory effect of treatment on *E. coli* bacteria, SA1 and SA2 treatment concentrations of 50% begin to form a 14 mm bland zone, while in the administration of SP1 and SP2 provide a larger inhibitory zone. The combination of SP2 90% and SA2 90% forms a inhibitory zone of 33.17 mm. This inhibitory zone was larger than ciprofloxacin inhibitory zone, which was 29.66 mm (Figure 2).

The inhibitory effect of treatment on *C. albincans*, it was seen that the concentration of 60 % of SA1 and SA2 had begun to form 9 mm inhibitory zone, whereas the administration of SP1 and SP2 had larger inhibitory zone. In the combination preparation, the largest inhibitory zone was 28.38 mm in the combination of SP2 90% and SA2 90%. This inhibitory zone was larger than ketokenazole which was 24.12 mm (Figure 3).

The older leaves of *Stenochlaena palustris (Burm.f) Bedd*. had a larger inhibitory zone. This result was corresponded with the research conducted by Rashid et al (2014). But, *Sauropus androgynus (L.) Merr* leaves had the opposite result, the older leaves produced smaller inhibitory zone. There was no known difference in the content of the leaves that are old and young in the two plants, so it is necessary to do further research on the content of what influences the difference in activity in it.

Conclusion

Leaves of *Stenochlaena palustris (Burm.f) Bedd*. and *Sauropus androgynus (L.) Merr* had antibacterial activity. Antibacterial activity of the leaves of *Stenochlaena palustris (Burm.f) Bedd*. Was better than the leaves of Sauropus androgynus (L.) Merr. The old leaves of *Stenochlaena palustris (Burm.f) Bedd*. had better antimicrobial activity compared to young leaves. While the old leaves of *Sauropus androgynus (L.) Merr* had less antimicrobial activity compared to young leaves. The combination of *Stenochlaena palustris (Burm.f) leaves Bedd*. and *Sauropus androgynus (L.) Merr*, both old and young, had better activities compared to single preparation.

References

1. Ali MA, Lafta AH. Jabar SK. Antibacterial Activity of Alkaloidal Compound Isolated from Leaves of *Catharanthus roseaus* (L.) Against Multidrug Resistant Strains. Academic Journal of Research in Pharmaceutical Biotechnology. 2014; 5(2): 21-23.
2. Arullappan S, Sawai S, Chee L, Mahandan M, Shanmugavelan R. Phytochemical Screening and

Evaluation of Cytotoxic Effect and Antioxydant Activity of Fractions Isolated from *Stenochlaena palustris* (Burm.f) Bedd Leaves. *Indian Journal of Pharmaceutical Education and Research*. 2017; 51(45): 735-740

3. Badan Pengawas Obat dan Makanan (BPOM) Republik Indonesia. 2010. Acuan sediaan herbal: I. (5). Jakarta
4. Chai T, Kwek M, Ong H, Wong F. Water Fraction of Edible Medicinal Fern *Stenochlaena palustris* is a Potent Alfa Gucosidase inhibitor with Concurrent Antioxidant Activity. *Journal of Food Chemistry*. 2015; 7(2): 1-2.
5. Cikita I, Hasibuan IH, Hasibuan R. Pemanfaatan flavonoid ekstrak daun katuk *(Sauropus androgynus (L)Merr)* sebagai antioksidan pada minyak kelapa. Jurnal Teknik Kimia Universitas Sumatera Utara. 2016;5(1):45-53.
6. Erwin, Anggreini D, Suryani. Chemical analysis and and antibacterial activity of the ethanolic extract of *Stenochlaena palustris.* Der Pharmacia Lettre. 2016; 8(1): 233-6.
7. Laveena KB, Chandra M. Evaluation Of Bioactive Compounds, Antioxidant, And Antibacterial Properties Of Medicinal Plants Sauropus Androgynus L. And Erythrina Variegata L. *Asian Journal of Pharmaceutical and Clinical Research*. 2018; 11(12): 313-317.
8. Majid TS, Muchtaridi. Aktivitas Farmakologi Ekstrak Daun Katuk (*Sauropus androgynus* (L.) Merr). *Farmaka*. 2018; 16(2): 398-405.
9. Malladi S, Ratnakaram VN, Babu KS, Pullaiah T. Evaluation of In Vitro Antibacterial Activity of *Carraluma lasiantha* for Scientific Validation of Indian Traditional Medicine. Cogent Chemistry. 2017; 3(1): 11-16.
10. Mukhriani, Tahar N, Astha ASW. Uji aktivitas antibakteri hasil fraksinasi dari ekstrak metanol daun katuk (*Sauropus androgynous*) terhadap beberapa bakteri patogen. JF FIK UINAM. 2014; 2(1): 12-7.
11. Ndanusa, A. H., Cicuzza, D., Siddique, M. M. 2020. Analysis of the phytochemical contents and anti-oxidative properties of *Stenochlaena palustris.* International Food Research Journal 27(5): 798 – 804.
12. Othman L, Sleiman A, Abdel-Massih M. Antimicrobial Activity of Polyphenols and Alkaloids in Middle Eastern Plants. *Frontiers in Microbiology.* 2019; 10.911.
13. Ramadheni P, Mukhtar H, Prahmono D. Uji aktivitas antibakteri ekstrak etanol daun katuk (Sauropus androgynus (L.) merr) terhadap bakteri *Staphylococcus aureus* dan Eschericia coli dengan metode difusi agar. Indonesia National Research Pharmaceutical Journal. 2017; 2(2): 34-44.
14. Rostinawati T, Shendi S, Maulida F. Aktivitas Antibakteri Ekstrak Etanol Daun Kelakai (*Stenochlaena palustris* (Burm.F) Bedd) Terhadap *Salmonella typhi* dan *Staphylococcus aureus* Dengan Metode Difusi Agar CLSI M02- A11. *Jurnal Farmasi Kesehatan*. 2017; 3(1):1-5
15. Susanti, N.M.P., Budiman, I.N.A., Warditiani, N.K. Skrining Fitokimia Ekstrak Etanol 90% Daun Katuk (*Sauropus androgynus* (L.) Merr.) 2014. Ojs.unud.ac.id. 1: 83-86
16. Syahrir NH, Afendi FM, Susetyo B. Efek sinergis bahan aktif tanaman obat berbasiskan jejaring dengan protein target. Jurnal Jamu Indonesia. 2016; 1(1): 35-46.
17. Tahir B, Saleh C, Pasaribu SP. Uji fitokimia, toksisitas, dan aktivitas antioksidan alami daun tumbuhan kelakai (*Stenochlaena palustris*) dengan metode DPPH [Prosiding Seminar Nasional Kimia 2013]. Samarinda: Universitas Mulawarman. 2013: 141-46.
18. Xie Y, Yang W, Tang F, Chen X. Antibacterial Activities of Flavonoids: Structure- Activity Relationship and Mechanism. *Current Medicinal Chemistry*. 2015; 22: 133-149.
19. Zanga, B., J. Chenga, C.Zhanga, Y. Baib, W.Liub, W.Lic, K. Koikec, T.Akihisaad, F. Fengae, J. Zhangae, *Sauropus androgynus* L. Merr A phytochemical, pharmacogical and Toxocogical review. Journal of Ethnopharmacology. Abstract. Elsevier; 2020; 257: 11277