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by Tita Juwitaningsih

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T Juwitaningsih[1]*, S A Sari[1], I S Jahro[1], S Silaban[1], M Simorangkir[1]

Department of Chemistry, Universitas Negeri Medan, Medan-20221, Indonesia

*Corresponding author's email: juwitaningsih@gmail.com

Abstract. The Sayat-sayat(*Leersiahexandra*Sw) plant is a plant used in traditional medicine by the Batak Karo people in North Sumatra. This study aims to examine the phytochemical content, antibacterial activity and cytotoxic test of acetone extract of *L.hexandraSw* seeds. Phytochemical tests include tests for alkaloids, flavonoids, saponins, tannins and steroids. Antibacterial tests were carried out against Propionibacterium acnesATCC (27853), *Escherichia coli* ATCC 25922, *Bacillus cereus* ATCC 1178, *Salmonella enterica* ATCC 14028 with the paper disc diffusion method M02-A11 (CLSI) and continued with the determination of MIC and MBC using the microdilution method M07-A9 (CLSI). The toxicity test used the Brine Shrimp Lethality Toxicity (BSLT) method using Artemia Salina Leach shrimp larvae. The results of phytochemical screening showed that the acetone extract of the *L.hexandraSw* seeds contained alkaloids, flavonoids and terpenoids. The results of the antibacterial activity test showed the best activity against *B. cereus* ATCC 1178 and *S. enterica* ATCC 14028 with an inhibition zone in the range of 8.4 -8.6mm, with MIC and MBC values of 625 µg/mL. The results of the toxicity test are toxic and potentially bioactive.

Keywords: *Leersiahexandra*Sw, phytochemicals, toxicity, antibacterial

1. Introduction

Indonesia has wealth of biodiversity, one of them can be used as traditional medicine. One of biodiversity that has long been used by the Karonese in Sumatera as traditional medicine is the Sayat-sayat plant. These herbs are used to treat toothaches [1,2].In the traditional Senegalese pharmacopoeia, *L.hexandra* is used for the treatment of hemoptysis in patients who experience frequent coughs. In Cameroun, an aqueous extract of *L. hexandra* leaves and stems is traditionally used for the treatment of hypertension [3].

*L. hexandra*Sw is a kind of weed that grows wild in dry, watery, humid and cold areas. Therefore, most research has been carried out on *L.HexandraSw* plants related to the ability of these plants to absorb metals through the roots, such as detoxification in wetlands from harmful water pollutants [Cr (VI)][4]. Use of *L. hexandraSw* for soil phytomediation on soils contaminated with fresh and weathered oil[5], *L. hexandraSw*as phytoremediation of soil and water contaminated by Cr [4,6]. Based on the results of research on existing publications, there is no information / research on *L. hexandra*Sw that supports its use as a traditional medicine. Therefore, this paper reports on the phytochemical screening, antibacterial test and toxicity test of *L.hexandraSw* seeds originating from North Sumatera.

2. Materials and Methods

2.1. Plants extract preparation

The sample studied was *L. hexandraSw* seeds obtained from the herbal medicine shop "Sempurna" Sambu Medan. The dry seeds of *L.hexandraSw* are then mashed (200 grams). After that it was macerated with acetone for 3 x 24 hours. The results of maceration are concentrated by evaporation using a rotary evaporator (Heidolph) until concentrated extract is obtained.

2.2. Antibacterial activity test

Based on the standard method that recommended by Clinical and Laboratory Standards Institute [7]. Antibacterial activity test carried out on pathogen bacterial Propionibacterium acnes ATCC (27853), *Escherichia coli* ATCC 25922, *Bacillus cereus* ATCC 1178, *Salmonella enterica* ATCC 14028.Solution test was prepared at concentration 1% in 10% DMSO which is equivalent to 10,000 µg/mL and a standard solution of 500 µg/mL of chloramphenicol antibiotic. Determination of the Inhibition Zone using the paper disc diffusion method (CLSI-M02-A11) and the determination of the Minimum Inhibitory Concentration (MIC) by the micro dilution method (CLSI-M07-A9). The determination of the minimum bactericidal concentration (MBC) was carried out by growing 10 μL of sample from each hole of the micro plate on the surface of the media so that the MHA MHA[8].

2.3. Cytotoxic

The cytotoxic test was carried out using the Brine Shrimp Lethality Test (BSLT) method referring to the research of Puspitasari etal. [9]. In summary: Artemia Salina Linch larvae grown from eggs were contacted with samples of various concentrations (1000, 500, 250, 100 and 10 ppm), then left to stand for 24 hours. Then counted the number of shrimp larvae that died from each concentration. The toxicity test for each concentration was repeated 3 times. The toxicity effect was analyzed from the observed percent mortality.

$$\%\ Mortality = \frac{the\ number\ of\ dead\ larvae}{the\ number\ of\ test\ larvae} x\ 100\%$$

If there are dead larvae in control, the percentage of mortality is determined by this formula:

$$\%\ \text{Mortality} = \frac{test-control}{control} \text{x } 100\%$$

The mortality of *A. salina* larvae was obtained through the probity table and regressed linearly.

$$Y = a + bX$$

Information

Y = probity value
a = regression concentration
b = Slope of the regression
X = Logarithm of 10 test concentration

2.4. Phytochemical screening

Phytochemical screening is intended to identify the flavonoids, terpenoids, saponnins, tannins steroids contained in *L. Hexandra Sw*. Phytochemical screening is done by referring to the method that has been done in previous research[10].

3. Results and Discussion

3.1. Antibacterial test

The paper disc diffusion test is a preliminary test to see the inhibition of the sample against pathogenic bacteria. Antibacterial activity was measured from the clear zone diameter (mm). The acetone extract of *L. Hexandra* Sw seeds showed activity against gram-positive bacteria as summarized in Table 1.

Table 1. Data of average value of inhibition zone diameter of acetone extract of *L. Hexandra* Sw

	Average value of the inhibition zone(mm)			
	Gram -		Gram +	
	E. coli	*S. enterica*	*P. acne*	*B.cereus*
Chloramphenicol	22.4	30.7	22.2	25.9
Aceton*L. Hexandra Sw*seeds extract	0	8.6	7.5	8.4

the zone diameter is categorized as weak if it is smaller or equal to 5 mm, moderate category if the inhibition zone is 5-10 mm, strong if the inhibition zone is 10 -19 mm and is said to be very strong if the inhibition zone is greater than or equal to 20 mm [11]. Based on these criteria, the acetone extract of *L.Hexandra Sw* seeds showed moderate category against *S. enterica* ATCC 14028, *P. acne* ATCC (27853) and *B. cereus* ATCC 1178.

When compared with the standard chloramphenicol, the percentage effectiveness of the inhibition zone diameter of *L. hexandra Sw* acetone extract against S. *enterica bacteria* was 28%, the effectiveness against *P.acne bacteria* was 33%, and against B. *cereus bacteria* was 32%.Determination of MIC value to determine the minimum inhibitory concentration against bacteria. The MIC value of the seed extract of *L.Hexandra Sw* is as shown in Table 2.

Table 2. Data value of MIC acetone *L. Hexandra Sw*seeds extract

	MIC (µg/mL)			
	Bacterial			
	Pa	Ec	Se	Bc
Chloramphenicol	0.49	0.97	0.48	0.48
Acetone *L.HexandraSw* seeds extract	5000	ND	625	625

Note ND: Is not done

The activity of an extract is categorized as active when the MIC value is below 100 µg / mL, moderate if the MIC value is around 100 <MIC <625 µg / mL and weak if the MIC value is> 625 µg / mL [12]. Thus, the acetone extract of *L.HexandraSw* seeds inhibited the "medium" category of bacteria *S. enterica* and *B. cereus*, and the "low" category of *P. acne*.Determination of MBC was intended to see whether the acetone extract of *L. Hexandra*Sw seeds had bacteriostaticor bactericidal properties. The MBC value data is summarized inTable 3.

Table 3. Data value of MBC acetone *L. Hexandra Sw*seeds extract

	MIC (µg/mL)			
	Bacterial			
	Pa	Ec	Se	Bc
Chloramphenicol	31.5	31.3	7.8	1.95
Acetone *L.HexandraSw* seeds extract	>5000	ND	625	625

Based on the data in Table 3, the acetone extract of *L.hexandra Sw* seeds is bactericidal against *S.enterica* and *B.cereus* with a concentration of 625 µg / mL and to kill *P.acne* requires a concentration greater than 5000 µg / mL.

2. Toxicity test

Toxicity test is a test to detect the toxic effect of a substance on a biological system and to obtain typical dose-response data from the test preparation. There is a positive correlation between the BSLT method and the cytotoxic test on cancer cell culture, so the method is often used as a preliminary test

to determine whether a compound has potential or not as an anticancer [13]. The sample toxicity test was determined based on the value of the LC_{50} which can kill *A. salina* by up to 50%. Furthermore, statistical calculations are carried out using probit analysis (probability). The results of the cytotoxy test on *A.salina* Leach are summarized inTable 4.

Table 4.BSLT (Brine shrimp lethality test) of aceto extracts *L. hexandraSw*Seeds

Treatment	Number of Mortality of Artemia Salina Leach			
	Concentration (ppm)			
	10	100	500	1000
1	6	8	10	10
2	7	8	9	10
3	8	9	10	10
Number of Mortality	21	25	29	30
Average	7	8.3	9.6	10
% Mortality	70%	83%	96%	100%

Based on the data in Table 4, this study shows that the higher the concentration of the test solution, the more *A.salina* larvae will die. Determination of the LC_{50} value using the probit method which is summarized in Table 5.

Table5. Death of larvae on acetone extracts *L. hexandraSw*Seeds

Concentration	Concentration log(x)	% Mortality	Probity Value
1000	3	100%	7.33
500	2.69	96%	6.75
100	2	83%	5.92
10	1	70%	5.52

From the data in Table 5, it is obtained the straight line equation y = 0.8744x + 4.4787 as shown in Figure 1.

Figure 1. LC_{50} of the acetone seed extract

Based on the regression equation, the value of LD_{50} = 3.9445 ppm is obtained. Determination of bioactive potential is carried out by comparing the LC_{50} value of a sample extract with the following criteria: $LC_{50} \leq 30$ ppm very toxic, 31 ppm $\leq LC_{50} \leq$ 1000 toxic, LC50> 1000 ppm is not toxic. Thus,

the acetone extract of *L. Hexandra*Sw seeds is categorized as very toxic and has the potential as an anticancer.

3.3. Phytochemical screening

Phytochemical screening is one way to determine the active ingredients which are secondary metabolites contained in plant samples. The results of phytochemical screening on *L. Hexandra* Sw seeds are shown in Table 6.

Based on the results of phytochemical screening, the acetone extract of *L.Hexandra Sw* seeds contained secondary metabolites of the alkaloids, flavonoids and terpenoids and tannins. Thus the antibacterial activity is caused by the presence of these secondary metabolites. Alkaloids can interfere with the peptidoglycan components of bacterial cells so that the cell wall layer is not formed completely and causes cell death [14]. Flavonoids cause damage to cell wall permeability [15]. Terpenoids cause cell membrane damage [16,17]. Whereas tannins cause cells to become lysed, besides that they have the ability to activate bacterial enzymes and disrupt the passage of proteins in the inner layers of cells [18]. Likewise, the death of larvae is related to the function of active compounds that can inhibit larvae. The way these compounds work is by acting as stomach poisoning, therefore when the secondary metabolites enter the body of the larvae, it will disturb the digestive tract and inhibit the taste receptors in the mouth area of the larvae [19].

Tabel 6. Phytochemical screening result of *L. hexandra*Sw Seeds

Phytochemical Test	Test Result
Alkaloids	+
Flavanoids	+
Terpenoids	+
Steroids	-
Saponnins	-
Tannins	+

Information:
(+): There is a chemical content
(-): There is no chemical content

4. Conclusion

The acetone extract of the seeds contains secondary metabolites of alkaloids, flavonoids, terpenoids, and tannins. The best activity was shown against *B.cereus* ATCC 1178 and *S.enterica* ATCC 14028 with the inhibition zone in the range of 8.4 -8.6, mm, with MIC and MBC values of 625 μg/mL. The results of the toxicity test were very toxic with an LD_{50} value of 3.9445 ppm.

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