Phytochemical Characterization of Bioactive Compounds on Ethanol Extracts Fermentation Products of Isolate BJDTA21 Endophytic Bacterial Andalas

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Abstract. The West Sumatra, Indonesia, mascot plant is called Andalas (*Morus macroura* Miq.). This plant belongs to the Moraceae family, which has a long history of being used medicinally. The goal of this study was to evaluate the Andalas ethanol extract's potential to prevent the growth of pathogenic microorganisms. The Andalas stems and roots were subjected to the ethanol extraction process. A completely randomized design (CRD) was utilized to determine the ideal concentration of ethanol extracts of Andalas roots and stems for suppressing test microorganisms. In this investigation, there were four different extract concentrations used as treatments: 3.12 percent, 6.25 percent, 12.50 percent, and 25 percent. The paper disc diffusion method was used to conduct antimicrobial activity testing on the test microorganisms (*Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans*). The outcomes revealed that the. The extract's potential mechanisms for exerting antibacterial activity against both of the studied microorganisms include flavonoids, alkaloids, saponins, terpenoids, and tannins.

Keywords: Phytochemical · Endophytic Bacteria · Andalas · Bioactive Compounds

1 Introduction

One of the factors contributing to the rise in infections-related fatalities is bacterial resistance. Doxycycline and tetracycline resistance was common in Staphylococcus aureus isolates from hospital patients (72 % and 69 %). Additionally, penicillin-resistant Staphylococcus is discovered in hospitals. And bacteria isolated from the community [1, 2]. A Exploration of novel antibacterial active chemicals is required.. Andalas (*Morus macroura*) is One of the alternatives for an active compound source. Andalas has several active Antiviral, antioxidant, antifungal, and antibacterial properties are possessed by chemical substances [3, 4].

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The lack of research on the active chemicals in these plants was caused by the Andalas plants' development problems and their rarity. Utilizing endophytic microorganisms that are dependent on andalas plant active chemicals is one of the research methods for these substances. The lack of research on the active chemicals in these plants was caused by the Andalas plants' development problems and their rarity. Utilizing endophytic microorganisms that are dependent on andalas plant active chemicals is one of the research methods for these substances.

It is necessary to do research on the antibacterial activity of Andaleh compounds to provide comparative data that may support the findings of the study conducted by [5–7]. Theoretically, endophytic microorganisms will produce consistently active substances in response to their host. Endophytic bacteria produce secondary metabolites that are consistent with their host, even in quite high levels, according to [8]. Nisa's research, however, yields entirely different findings [9]. When compared to secondary substance extracts of endophytic fungi, the repressive zone of Chromolaena odorata leaf extract displayed better antibacterial activity against Enterobacteria dysenteriae, according to the results of the antibacterial activity study.Because of this, phytochemical characterization of bioactive compounds on ethanol extracts fermentation products of Isolate BJDTA21 endophytic bacterial Andala has been carried out.

2 Method and Materials

2.1 Bacteria Strains

The Gram positive and Gram negative conventional bacterial strains, including Staphylococcus aureus (ATCC.25923) and Escherichia coli, were employed in this study (ATCC.EC 25922). They came from the University of North Sumatra's Medical Faculty's microbiological labs.

2.2 Extracts from Fermentasi Products

The andalas plants were gathered in the andalas village in the Indonesian province of West Sumatra's Tanah Datar regency. They were recognized by the authors using local information from the community and the M. macraura determination key. Roots and stems from the plant are collected. Plants were dried in an oven (set to 45°C) until their weight remained constant. Each dried plant component was pulverized into 300 g of dry powder, which was then macerated for three times 24 hours. Following the maceration procedure, filter paper is used to remove the Andaleh tissue waste from the ethanol solution. A vacuum rotary evaporator is subsequently used to remove the filter results. The extraction procedure is completed.

2.3 Test for Phytochemicals

The With a small modification, Tapwal et al. (2016)'s phytochemical experiment was performed, allowing for the detection of some key ingredients (phenolic, flavonoid, tannin, steroid, terpenoid, and alkaloid). A phytochemical analysis was performed on all of

the soluble ethyl acetate that was recovered from the extraction (already reported) at a concentration of 1000 g/L. In general, the ferric chloride test was used to determine phenolic and tannin, magnesium and hydrochloric acid were added to determine flavonoid, acetate anhydride and sulfuric acid were added to monitor the steroid, sulfuric acid was added to determine terpenoid, and Mayer's and Wagner's reagents were used to monitor alkaloid.

2.4 Analysis

The mean and standard deviation were used to present all results. Results were further examined using SPSS (version 15) for Windows as necessary.

3 Achievement and Discussion

The extraction of the Andaleh root and stem with ethanol yielded thick extracts weighing 7.92 g and 10.2 g, respectively. The antibacterial activity of the extract revealed that bacteria could not grow at any concentration, according to the data. Figure 1 shows examples of inhibited zones that have formed.

The lowest concentration that has the same inhibition zone as or is greater than the control of ampicilli is the Andaleh ethanol extract concentration that will most effectively stop S. aureus from growing. Table 1 displays the findings of the investigation into the actions of Andaleh ethanol extract in preventing *S. aureus* growth.

Based on statistical analysis (= 0.05), it was determined that the concentration of the stem and root Andaleh ethanol extract had a significant impact on the amount of S. aureus growth inhibition. According to DMRT tests (= 0.05), the concentration of 25 percent and 12.5 percent for the stems and roots of Andaleh ethanol extract, respectively, was the best concentration for preventing the growth of S. aureus.

An antibiotic activity test on *E. coli* bacteria was performed to determine the extract's capacity to prevent the growth of gram-negative bacteria. Table 2 displays the results of the efficiency of stem and root Andaleh ethanol extracts against *E. coli*.

Table 2 Statistics indicate that neither of the two extracts tested could stop C. albicans from growing. The maximum percentage of tests that were run in this experiment was 25%.

An earlier investigation extracted endophytic bacteria from the stem, leaf, and root of Andaleh and evaluated their root tissue in producing anti-microbial compounds. The

Fig. 1. Extract Result

Table 1. *S. aureus* specific antimicrobial activity of stem and root Andaleh ethanol extracts

No	Concentration (%)	Dimensions of the inhibition zone (cm)	
		Stem	**Root**
1	Ampicillin	1,176 [a]	1.176[b]
2	3,125%	0,800 [c]	0.842[d]
3	6,25%	0,827 [c]	0.977[c]
4	12,5%	0,945 [b]	1.19[b]
5	25%	1,149 [a]	1.377[a]

Values are presented as Mean (n=3); One-Way ANOVA analysis was completed before the DMRT test.

Table 2. Phytochemical test results of Andalas endophytic bacterial fermentation products.

Isolat bakteri endofit	Senyawa aktif	Keterangan
BJTA 21	Safonin	+
	Flavonoid	+
	Tanin	−
	Terpenoid	−
	Fenolik	+
ATB 106	Safonin	+
	Flavonoid	+
	Tanin	−
	Terpenoid	−
	Fenolik	−
JDT 1B	Safonin	+
	Flavonoid	+
	Tanin	−
	Terpenoid	−
	Fenolik	+
ATBA41	Safonin	+
	Flavonoid	+
	Tanin	−
	Terpenoid	−
	Fenolik	−

study found that endophytic bacteria isolated from different tissues had different antimicrobial activity. Endophytic bacteria that [5, 7] isolated from the stem dan leaf (respectivly) have the ability to inhibit *S. aureus* better than other test microbes. In contrast, research conducted by [6], shows that endophytic bacteria isolated from roots can inhibit *C. albicans* better.

Although the research was done by [10] showed the endophytic bacteria can produce secondary metabolites that are the same as their hosts, even in relatively high amounts. Contrariwise, Nisa's research [9] shows that the ability of endophytic bacteria to produce antimicrobial compounds is lower than that of plant hosts. This difference in results can be caused by several factors, one of them is the use of solvents to extract active compounds which are not appropriate. In this research, the solvent used was ethanol, which is polar. According to [11], the type of solvent was a factor that influences the concentration and type of compound to be extracted. The polarity of the solvent is an important thing that influences the antimicrobial activity. Therefore, further experiments need to be carried out using non-polar solvents.

4 Conclusion

S. aureus and *E. coli* growth was inhibited most effectively by Andaleh root ethanol extract at 12.5 % and 3.125 %, respectively. The The presence of flavonoids, alkaloids, saponins, terpenoids, and tannins in the extract may be the cause of its antibacterial action against the two bacteria that were tested.

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