Plagiarism Checker X Originality Report

Similarity Found: 7%

Date: Friday, November 6, 2020
Statistics: 112 words Plagiarized / 1572 Total words
Remarks: Low Plagiarism Detected - Your Document needs Optional Improvement.

-----------------------------------------------
INTRODUCTION Antibiotic resistance is one of the biggest threats to global health. It is rising to dangerously high levels in all parts of the world (Aslam et al., 2018). Methicillin-resistance Staphylococcus aureus (MRSA) is a common cause of severe nosocomial infections (Choo & Hambers, 2016). It has developed resistance to numerous antibiotics caused by the misuse and overuse of antibiotics. The MRSA can hydrolyze almost any type of lactams, and its strains spread quickly, leading to a high mortality rate (Hu et al., 2019).

World Health Organization (WHO)'s first global report on antibiotic resistance reveals that more than one-quarter of S. aureus infections in the south-east Asia region are reported to be MRSA, which is home to a quarter of the world's population (Prestinaci et al., 2015). Therefore, alternatives antibacterial against MRSA infections is still sought-after investigation. Many antibacterial drugs were firstly isolated from natural sources (Rossiter et al., 2017). Many studies revealed that medicinal plants provide antibacterial compounds from its secondary metabolites (Gorlenko et al., 2020; Othman et al., 2019; Voravuthikunchai & Kitpipit, 2005). Indonesia is a rich archipelago with an abundance of natural plants to explore. One of them is bandotan (Ageratum conyzoides L.) plants, which grow in many Indonesian regions and are classified as tropical weeds (Kotta et al., 2020; Atisha & Mita, 2018). It is easy to find the weeds because it thrives in any garden and agricultural soils. It is also ubiquitous in disturbed sites and degraded areas (Marks & Nwachuku, 1986). The weeds are also noxiously regarded as harmful for crops, but on the contrary, A. conyzoides have been known since ancient times for their therapeutic benefits (Garg et al., 2015). The weed is traditionally used to treat new wounds, bleeding wounds, ulcers, eczema, bacterial infection diseases, arthrosis, headaches, pneumonia, analgesic, antispasmodic, anti-inflammatory, leprosy, and other skin diseases (Kamboj & Saluja, 2008; Achmad et al., 2020). Recent studies have shown that bandotan leaves have antibacterial activity against Staphylococcus aureus, Porphyromonas gingivalis, Streptococcus mutans, and Escherichia coli (Mentari et al., 2020; Achmad et al., 2020; Sugara et al., 2016).

Many research shows that A. conyzoides have various health benefits. However, there is still limited research on A. conyzoides leaves antibacterial activity specifically towards MRSA. Therefore, this study focused on assessing the antibacterial activity of A. conyzoides leaves extract towards MRSA. MATERIALS AND METHODS Plant material Ageratum conyzoides plants were collected from the rice field at Limbangan, Garut, West Java, Indonesia. The plants were authenticated and determined at the Herbarium
Unit, Department of Biology, Universitas Padjadjaran, Indonesia. Extract preparation

Ageratum conyzoides leaves were washed and dried for ten days at 40°C.

Dried leaves were grinded into powder and evaluated by the distillation method to
analyze the moisture content. Extraction was performed by the maceration method. As
much as 250 g of A. conyzoides leaves were extracted by ethanol 96% at room
temperature. The solvent was replaced three times with fresh solvent every 24 hours.
After filtration of total extracts, A. conyzoides leaves extract were evaporated by rotary
evaporator until it dry and were weighed to determine the yield. Phytochemical
screening The extract was subjected to various phytochemical screening to identify its
chemical constituents, including alkaloids, flavonoids, saponins, tannins, quinones,
steroids, or triterpenoids.

The procedures for detecting those secondary metabolites are referred to Materia
Medika Indonesia volume IV (1980). Bacterial culture Methicillin-resistance
Staphylococcus aureus ATCC 43300 isolates were obtained from the Department of
Pharmacy, Universitas Padjadjaran, Indonesia. The bacteria were maintained on Nutrient
Agar (NA) slope and then subcultured on NA at 37°C for 18-24 hours. Antibacterial
activity Antibacterial activity of A. conyzoides leaves extract was evaluated by disc
diffusion method.

Extract was diluted on dimethyl sulfoxide (DMSO) solution to yield 5%, 7.5%, 10% and
12.5% concentration. Bacteria inoculum was introduced onto the sterile NA plates' surface using a sterile loop and spread over the media for even distribution. The plates were divided into five sections: four sections for extract and one section for gentamicin. Blank sterile paper discs were placed on the NA surface and impregnated with 15 mL of the extracts. The plates were incubated at 37°C for 18-24 hours. The antibacterial activity was expressed as clear inhibition zones produced by the extracts. The test was repeated three times. The minimum inhibitory concentration (MIC) was investigated using the agar dilution method. As much as 1 mL of A.

conyzoides leaves extracts with various concentration (1%, 2%, 3%, 4%, and 5%) were
added into 10 mL NA. Bacterial suspensions were inoculated onto each plate with a
sterile loop, and the presence or absence of bacteria growth is recorded after suitable
incubation. Incubation lasted for 18-24 hours at 37°C. The MIC was determined as the
lowest concentration of A. conyzoides leaves extracts, which completely inhibited
bacterial growth. Furthermore, the MIC was converted by regression linear equation into
its antibiotic dose.

RESULTS AND DISCUSSION This study is experimental laboratory research. The moisture
content of A. conyzoides L. leaves was 5%, which less than 10%. The yields extract obtained with maceration was 17.16 g from 250 g simplicia. The phytochemical screening showed that A. conyzoides leaves contained various secondary metabolites, such as alkaloids, flavonoids, saponins, tannins, steroids, or triterpenoids as shown in Table I. These results agree with the previous study of phytochemical screening of A. conyzoides leaves. It shows alkaloids, saponins, flavonoids, polyphenols, tannins, glycosides, resins, phenols, and essential oils (Achmad et al., 2020; Chew et al., 2018; Amadi et al., 2012). Plants synthesized the secondary metabolites to protect them from predators such as herbivores, insects, and microorganisms. It could kill or inhibit microorganism growth via different mechanisms (Chew et al., 2018). Table I. Phytochemical screening of A. conyzoides leaves

<table>
<thead>
<tr>
<th>Phytochemical</th>
<th>Simplicia</th>
<th>Extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Quinones</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Steroids/Triterpenoids</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

(+): presence; (-): absence of phytochemicals The results of the antibacterial activity were revealed using the disc diffusion method. This is a standard qualitative assay to evaluate the antimicrobial activity of extracts or phytochemicals.

Ageratum conyzoides leaves ethanolic extracts have demonstrated antibacterial activity against MRSA isolate. From Figure 1, it can be seen that the higher extract concentration produced a more expansive inhibition zone. The inhibition zone at 5% concentration of A. conyzoides leaves extract had the lowest average inhibition zone of 15.47 mm, while 12.5% had the largest average inhibition zone of 25.1 mm. Ageratum conyzoides leaves extract with 12.5% concentration, giving almost similar results compared to gentamicin, which was used as a comparative antibiotic. This activity may be attributed to the rich tannins and flavonoid contents of A. conyzoides leaves.

Flavonoids and tannins have been reported to possess antimicrobial activity due to their ability to complex with the bacteria cell wall and inactivate enzymes, microbial adhesion, and cell envelops proteins (Cowan, 1999). These results confirmed the evidence in previous studies that reported that the extract of A. conyzoides has potential antibacterial activity against S. aureus (Garg et al., 2015; Sugara et al., 2016). Figure 1. Antibacterial activity of A. conyzoides leaves extract against MRSA The quantitative analysis using the agar dilution method showed that at a concentration of 1%, 2%, and 3% of A. conyzoides leaves extract still observed MRSA growth; meanwhile, the absence of MRSA growth can be seen at a concentration of 4% and 5% as shown in Table II. It was indicated that extract of A. conyzoides leaves active exhibiting the highest potency with MIC of 4%. Previously, the plant has been reported to have good antibacterial activity.
towards S. aureus with the MIC value of 2% (Budiman & Aulifa, 2020). According to research conducted by Astuti (2015), ethanolic extract of A. conyzoides leaves had the MIC value of 12.5 mg/mL against S. aureus. The MRSA was more challenging to treat than most S. aureus because it is resistant to some commonly used antibiotics.

Therefore, the MIC value for MRSA was higher than S. aureus. Table II. Minimum inhibitory concentration of A. conyzoides leaves extract Extract concentration (%) Bacterial growth _1_+ _2_+ _3_+ _4_- _5_- (_+): growth of bacteria; (-): no growth of bacteria The MIC value of A. conyzoides leaves extracts then converted into equivalency of antibiotic dose. The concentration of 4% A. conyzoides leaves extract concentration was analyzed to determine its antibiotic dosage equivalency using the agar diffusion method as shown in Figure 2. Gentamicin was used as a reference standard antibiotic. Gentamicin is an aminoglycoside that inhibits bacterial protein synthesis by binding to its ribosomes (Krause et al., 2016). The standard dose of gentamicin is 3-6 mg/kg/day divided every eight hours to treat MRSA prosthetic valve endocarditis. Gentamicin is regularly added with rifampin for the first two weeks of treatment (Galar et al., 2019). / Figure 2. Inhibition zone of A. conyzoides leaves extract compared with gentamicin Furthermore, the data was calculated by linear regression, as seen in Figure 3. The obtained equation concluded that 4% of A. conyzoides leaves extract was equivalent to 4.46 x 10^-6 g of gentamicin. This concentration could be considered when designing the next potential drug to treat nosocomial infection caused by MRSA. / Figure 3. Linear regression graphic and equation of gentamicin inhibition zone CONCLUSION Ethanolic extracts of A. conyzoides leaves have potential antibacterial activity against MRSA. Further identification of the active constituents is needed to evaluate its efficacy and safety against MRSA.

INTERNET SOURCES:
----------------------------------------------------------------------------------------
1% - https://link.springer.com/article/10.1007/s41745-020-00187-0
<1% - https://infectologiaemgeral.files.wordpress.com/2017/01/28032484.pdf
<1% - http://www.readbag.com/blackwellpublishing-eccmid16-pdfs-clm-1432