ANTIBACTERIAL ACTIVITY TEST OF BAY LEAF EXTRACTS (SYZYGIUM POLYANTHUM (WIGHT) WALP.) AGAINST STAPHYLOCOCCUS AUREUS AND ESCHERICHIA COLI: SYSTEMATIC LITERATURE REVIEW

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ABSTRACT

Antibiotic resistance is a global health problem that has severe implications for morbidity, mortality, and health costs. Based on the Antimicrobial Resistant in Indonesia (AMRIN-Study), it was reported that there were bacteria that were resistant to antibiotics, including Escherichia coli, which is resistant to ampicillin trimethoprim-sulfamethoxazole and ciprofloxacin; and Staphylococcus aureus which is resistant to oxacillin, vancomycin, clindamycin, and levofloxacin. In Indonesia, Syzygium polyanthum (Wight) Walp. Bay leaf is one of the plants that is often used as spices. Its bay leaf contains secondary metabolites that have an antibacterial function. This study analyzed the antibacterial activities of bay leaf extract against Staphylococcus aureus and Escherichia coli. This research used a systematic literature review with a literature search strategy using Google Scholar and PubMed. The literature discusses bay leaf extract, Staphylococcus aureus, Escherichia coli, and antibacterial activity. From this study, 14 research articles described the antibacterial activity of bay leaf extract by inhibiting the growth of Staphylococcus aureus and Escherichia coli in the medium, strong, and very strong categories. Bay leaf extract can inhibit the growth of Staphylococcus aureus and Escherichia coli because it contains phytochemical compounds with antibacterial properties, including alkaloids, flavonoids, steroids/triterpenoids, saponins, tannins, phenols, essential oils, and carbohydrate.

Keywords: Antibacterial activity; Bay leaf extract; Escherichia coli; Staphylococcus aureus

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INTRODUCTION

Antibiotic resistance is a global health problem that seriously impacts the level of morbidity, mortality, and health costs. According to the Antimicrobial Resistant in Indonesia (AMRIN Study) research, it was reported that there was bacterial resistance to antibiotics, including Escherichia coli resistance to ampicillin (73%), trimethoprim-sulfamethoxazole (56%), ciprofloxacin (22%) and Staphylococcus aureus resistance to oxacillin (40%), vancomycin (40%), clindamycin (50%) and levofloxacin (50%) (Suciari et al., 2017). Staphylococcus aureus and Escherichia coli are normal flora bacteria that can be pathogenic. Based on priority data on resistant bacteria in the World Health Organization, Staphylococcus aureus and Escherichia coli are included in the critical and high categories, which means they have entered high numbers for antibiotic resistance (WHO, 2019).

In treating infections caused by bacteria, the primary choice is the administration of antibiotics. However, research conducted by AMRIN-Study shows that the rate of antibiotic resistance is still high. Therefore, the global issue of antibiotic resistance needs
to find a solution. If the antibiotic's resistance exists, alternative medicine with herbal plants that have antibacterial properties and benefits is used (Parathon et al., 2017).

In Indonesia, the Bay plant is a distinctive and easy-to-find plant often used as a spice (Tammi et al., 2018). Bay leaf or *Syzygium polyanthum* (Wight) Walp. contains secondary metabolites with functions as antibacterial substances, namely saponins, tannins, alkaloids, flavonoids, and steroids (Hosaina et al., 2020). Bay leaf extract is known to have benefits as an antibacterial, but this knowledge is not widely known, and there is no literature review data in the discussion. Therefore, this study was used to test the antibacterial activity of bay leaf extract against *Staphylococcus aureus* and *Escherichia coli* along with the phytochemical compounds contained in a bay leaf or *Syzygium polyanthum* (Wight) Walp. as an antibacterial compound through the Systematic Literature Review methodology.

MATERIALS AND METHODS

a. Materials

We conducted this review using electronic databases such as PubMed and Google Scholar. The search terms include bay leaf extract, *Staphylococcus aureus*, *Escherichia coli*, and antibacterial activity. Database search after 2011, no language restrictions. Assessment of the quality of a methodology taken from JBI (Joanna Briggs Institute), which is a research organization and international development at the University's Faculty of Health Sciences Adelaide. JBI (Joanna Briggs Institute) has provided access to and serves more than 90 countries (The Joanna Briggs Institute, 2017). After conducting a quality assessment, researchers took data from 14 research articles and classified important data in research articles using tables.

b. Method

The method used to analyze the data is a Systematic Literature Review with PICO analysis (Population: antibacterial; Intervention: Bay Leaf Extracts (Syzygium polyanthum); Comparison: Staphylococcus aureus and Escherichia coli; Outcome: inhibiting the growth of bacterial). Next, the article selection process was carried out using a PRISMA-P (Preferred Reporting Items for Systematic Reviews and Meta-Analyses Protocols) flow diagram.

RESULT

In this study, data were obtained from 14 research articles extracted, synthesized, and continued with a discussion of the results. The antibacterial activity test will be known through the diameter of the inhibition zone formed after being given a test treatment using bay leaf extract. The size of the inhibition zone produced by the antibacterial was categorized into four divisions, namely extreme inhibition zone (> 20 mm), strong inhibition zone (10-20 mm), moderate inhibition zone (5-10 mm), and weak inhibition zone 5 mm (Davis and Stout, 1971).
<table>
<thead>
<tr>
<th>No</th>
<th>Author and year</th>
<th>Phytochemicals</th>
<th>Extract</th>
<th>Antibacterial test</th>
<th>Effective concentration</th>
<th>Positive control</th>
<th>Inhibition zone</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Faizah et al., 2020.</td>
<td>Saponin, alkaloid</td>
<td>Methanol 96%</td>
<td>Solid dilution</td>
<td>1 g/mL</td>
<td>Cotrimoxazole</td>
<td>S. aureus 13.7 mm, E. coli 9 mm</td>
</tr>
<tr>
<td>2</td>
<td>Maharani et al., 2017.</td>
<td>Flavonoid, tannin, essential oil, and alkaloid</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>10%</td>
<td>-</td>
<td>S. aureus 10 mm, E. coli 9.31 mm</td>
</tr>
<tr>
<td>3</td>
<td>Ramadhania et al., 2018.</td>
<td>Saponin, tannin, alkaloid, triterpenoid, flavonoid, polyphenol, and essential oil</td>
<td>Methanol 96%</td>
<td>Broth dilution</td>
<td>S. aureus 23.16 µg/mL, E. coli 10.28 µg/mL</td>
<td>Ampicillin</td>
<td>S. aureus 23.16 mm, E. coli 35.01 mm</td>
</tr>
<tr>
<td>4</td>
<td>Ramli et al., 2017.</td>
<td>-</td>
<td>Ethanol</td>
<td>Disk diffusion</td>
<td>10%</td>
<td>Chlorhexidine</td>
<td>S. aureus 9.33 mm, E. coli 7 mm</td>
</tr>
<tr>
<td>5</td>
<td>Gusmiah et al., 2014.</td>
<td>Tannin, flavonoid, and essential oil</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>10%</td>
<td>-</td>
<td>S. aureus 9.78 mm</td>
</tr>
<tr>
<td>6</td>
<td>Tammi et al., 2018.</td>
<td>Alkaloid, tannin, flavonoid, and essential oil</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>100%</td>
<td>Chloramphenicol</td>
<td>22.75 mm, 33.5 mm</td>
</tr>
<tr>
<td>7</td>
<td>Nordin et al., 2019.</td>
<td>-</td>
<td>Methanol</td>
<td>Disk diffusion</td>
<td>100%</td>
<td>Amoxicillin</td>
<td>13.50 mm, 21.81 mm</td>
</tr>
<tr>
<td>8</td>
<td>Putriani et al., 2021.</td>
<td>Flavonoid, saponin, and tannin</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>80%</td>
<td>Chloramphenicol</td>
<td>16.6 mm, 21.3 mm</td>
</tr>
<tr>
<td>9</td>
<td>Sari, 2019.</td>
<td>Polyphenol, flavonoid, tannin, saponin, essential oil, and alkaloid</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>100%</td>
<td>Amoxicillin</td>
<td>20 mm, 22.9 mm</td>
</tr>
<tr>
<td>10</td>
<td>Trisnawati et al., 2020.</td>
<td>Steroid, flavonoid, and phenolic</td>
<td>Methanol 95%</td>
<td>Disk diffusion</td>
<td>10%</td>
<td>Ampicillin</td>
<td>9 mm, 17 mm</td>
</tr>
<tr>
<td>11</td>
<td>Afifah et al., 2021.</td>
<td>Phenolic, flavonoid, tannin, and alkaloid</td>
<td>Chitosan nanoparticles</td>
<td>Disk diffusion</td>
<td>0.2%</td>
<td>Tetracycline</td>
<td>11.22 mm, 8.56 mm</td>
</tr>
<tr>
<td>12</td>
<td>Evendi, 2017.</td>
<td>Alkaloid, flavonoid, saponin, tannin, and steroid</td>
<td>Methanol</td>
<td>Disk diffusion</td>
<td>200 µg/well</td>
<td>Chloramphenicol</td>
<td>12.11 mm, 100 mm</td>
</tr>
<tr>
<td>13</td>
<td>Utami et al., 2020.</td>
<td>Tannin, flavonoid, essential oil</td>
<td>Ethanol 96%</td>
<td>Disk diffusion</td>
<td>75%</td>
<td>Ciprofloxacin</td>
<td>20 mm, 31 mm</td>
</tr>
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</table>
### DISCUSSION

Of the 14 research articles that were successfully reviewed, there were three research articles by Ramadhania et al. (2018) and Tammi et al. (2018), Witari et al., (2019), which resulted in extreme inhibition zones, eight research articles by Faizah et al. (2020), Maharani et al., (2017), Nordin et al., (2019), Putriani et al., (2021), Sari, (2019), Afifah et al., (2021), Evendi (2017), and Utami et al., (2020) produced a solid inhibitory zone. Three research articles, Ramli et al. (2017), Gusmiah et al. (2014), and Trisnawati et al. (2019), resulted in a medium inhibition zone. Research articles with intermediate results are caused because the effective concentration tested is only 10%. In contrast, research articles with powerful category results are tested with high effective concentrations. Therefore, it can be concluded that the effective concentration is directly proportional to the resulting inhibition zone. The higher the test concentration, the higher the inhibition zone results produced. Based on the above, the researchers saw the antibacterial activity and the potential that bay leaf extract could function and be used as an antibacterial drug.

Based on research by Faizah et al. (2020), Maharani et al. (2017), and Ramli et al. (2017), a comparison of the antibacterial activity of bay leaf extract against *S. aureus* and *E. coli* bacteria was carried out. The comparison showed that the inhibition zone of bay leaf extract against *S. aureus* was greater than that of *E. coli*. This is because the treatment given is bay leaf extract, which can inhibit bacterial growth. In addition, the content of flavonoid and alkaloid phytochemical compounds has a role in disrupting and damaging bacterial cell walls. *S. aureus* is a gram-positive bacterium with a cell wall structure consisting of a layer of peptidoglycan and teichuronic acid with or without an envelope. In contrast, the cell wall structure of *E. coli* is a gram-negative bacterium with an outer cell wall structure and lipopolysaccharide. Lipopolysaccharides can inhibit the penetration of foreign or antimicrobial compounds from entering bacteria (Ramli et al., 2017). Based on this, gram-negative bacteria have a cell wall layer that is more complex than gram-positive, making it more difficult for bay leaf extract to produce antibacterial activity against gram-negative bacteria. Based on this, the results of the inhibition zone of bay leaf extract against *S. aureus* are more excellent than *E. coli* because *S. aureus* has a cell wall structure simpler than E.coli, which, has only a single plasma membrane surrounded by a thick cell wall of peptidoglycan. (Faizah et al., 2020).

Putriani et al. (2021) conducted two types of treatment: the administration of bay leaf extract and mango Bacang leaf extract as an antibacterial against *S. aureus* bacteria. In this study, a growth inhibition test was carried out and resulted in a different inhibition zone diameter in each treatment. The result of the inhibition zone produced in the bay leaf extract was 16.6 mm, and the mango Bacang leaf extract was 16.3 mm at the same concentration of 80%. The difference in these results indicates that the antibacterial properties and inhibition zone produced by the bay leaf extract are more effective than the mango Bacang leaf extract; this can be caused by the antibacterial compounds contained in the bay leaf extract.

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<tr>
<td>14</td>
<td>Witari et al., 2019.</td>
<td>Essential oil, phenolic, flavonoid, saponin, and tannin</td>
<td>Ethanol 96% Disk diffusion 30% Tetracycline</td>
</tr>
<tr>
<td></td>
<td></td>
<td>22.50 mm 23.25 mm</td>
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</table>
being more significant than the Mango Bacang extract such as flavonoids, saponins, and tannins. (Putriani et al., 2021).

Bay leaf extract can inhibit the growth of *S. aureus* and *E. coli* bacteria due to phytochemical compounds. Based on data from 14 research articles, the phytochemical compounds in bay leaf extract are alkaloids, flavonoids, steroids/triterpenoids, saponins, tannins, phenols, essential oils, and carbohydrates. According to research by Maharani et al. (2017), alkaloids can form a disruptive mechanism for developing bacterial peptidoglycan, which makes the cell wall unable to create entirely and causes cell death. In addition, Evendi's research (2017) said alkaloids functioned as esterase inhibitors, DNA RNA polymerase, and cell respiration as DNA intercalation. Flavonoids are compounds that can cause protein denaturation and damage bacterial cell walls, according to Witari et al. (2019). In addition, according to Evendi (2017), tannins inhibit the adhesion of bacterial cells to their hosts and inhibit transport within the cell layer. The ability of phenol as an antibacterial is tremendous with the mechanism of disrupting bacterial cell tissue, inactivating enzymes, and denaturing proteins that cause bacteria to decrease their permeability so that they experience cell damage. Decreased permeability disrupts the transfer of critical organic ions, affecting the growth and death of bacterial cells (Purwantiningsih et al., 2014). Essential oils, generally known as essential oils, contain phenolic toxicity, which can cause the breakdown of proteins in cell walls, producing a tertiary structure of a protein using non-specific bonds. In addition, essential oils have citral and eugenol compounds as anesthetics and antiseptics that can kill and inhibit bacterial growth (Arum Samudra, 2014). The role of carbohydrates as an antibacterial is the glucose mechanism, which can cause an increase in cell osmotic pressure, driving hypertonic conditions. Hypertonic conditions in bacterial cells can cause cells to become dehydrated and cause cell death (Purnama, 2013).

The results of the inhibition zone in the study of Tammi et al. (2018), which used 96% ethanol solvent with an effective concentration of 100%, obtained a large inhibition zone of 22.75 mm. Maharani et al. (2017) conducted a study using the same solvent with a concentration of 10% and got an inhibition zone of 10 mm. Based on this, to find out the superiority of the solvent in producing a more significant inhibition zone, it is necessary to compare it with research using methanol solvent with the same effective concentration of 100%, namely the study conducted by Nordin et al., (2019). An investigation by Nordin et al. (2019) resulted in a smaller inhibition zone of 13.50 mm, and research conducted by Trisnawati et al. (2020) resulted in an inhibition zone of 9 mm. Therefore, it can be proven that ethanol has a higher level of polarity and a more significant advantage in producing an inhibitory area as a solvent for extraction. A study by Noviyanty (2019) stated that the higher the polarity of a solvent, the better the extraction ability and the resulting increased inhibition zone.

After reviewing 14 research articles, 4 out of 14 are arranged with the complete and appropriate information as researchers need. Full and proper information includes the data on the extract used, the effective concentration, the zone of inhibition produced by the administration of the effective concentration and positive control, and there is a repetition of treatment in each test group. A total of 4 research articles, including Tammi et al. (2018), Putriani et al. (2021), Sari et al. (2019), and Utami et al. (2020). In addition, there are 3 out of 14 research articles that have inhibition zone results with a powerful category, namely in a study conducted by Ramadhania et al. (2018) with an inhibition zone of 23.16 mm in *S. aureus* and 35.06 mm in *E. coli*, Tammi et al., (2018) with an inhibition zone of 22.75 mm, and Witari et al., (2019) with an inhibition zone of 22.50 mm.
CONCLUSION

Based on the results of this Systematic Literature Review research, it can be concluded that bay leaf extract (Syzygium polyanthum (Wight) Walp.) has antibacterial activity against S. aureus and E. coli. There are 3 out of 14 research articles that have inhibition zone results with a very strong category, namely in the research conducted by Ramadhania et al., (2018) with an inhibition zone of 23.16 mm in S. aureus and 35.06 mm in E. coli, Tammi et al., (2018) with an inhibition zone of 22.75 mm, and Witari et al., (2019) with an inhibition zone of 22.50 mm. Bay leaf extract contains phytochemical compounds that act as antibacterial, alkaloids, flavonoids, steroids/triterpenoids, saponins, tannins, phenols, essential oils, and carbohydrates.

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