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# Antibacterial And Antibiofilm Activity *Staphylococcus aureus* From Plants Containing Essential Oils: A Mini-Review

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**Abstract:** Biofilm is a collection of microbial cells irreversibly attached to a surface and encased in an Extracellular Polymeric Substance (EPS) matrix produced by itself and shows phenotypic changes such as changes in growth rates and changes in gene transcription from planktonic cells or their free cells. Bacteria that often cause biofilms are *Staphylococcus aureus*, which is widely distributed in nature. Some live like normal flora in humans in the axilla, inguinal and perineal region, and anterior nostrils. Approximately 25-30% of humans carry *Staphylococcus aureus* in the nasal cavity and skin. This review aims to determine the antibacterial and antibiofilm activity of plants containing volatile oil compounds against *Staphylococcus aureus*. The method used in this review is a narrative review approach. This method is a secondary research method that does not have specific guidelines in its preparation (non-systematic review) by collecting data or sources related to a particular topic obtained from various sources such as journals, the internet, and other libraries. From hundreds of journals, 20 journals have the activity of inhibiting *Staphylococcus aureus*, the conclusion obtained after reviewing hundreds of literature that essential oils have inhibitory activity and even destroy biofilms.

**Keywords:** Biofilm; *Staphylococcus aureus*; Essential Oil; Antibacterial; Antibiofilm.



## 1. Introduction

Traditional healing derived from plants is the embodiment of the active participation of citizens in solving health problems and has been recognized its role by various nations in improving the level of health of citizens. The World Health Organization (WHO) recommends using traditional medicine, including herbal medicine, to maintain citizens' health, prevent and cure diseases, especially chronic diseases, degenerative diseases, and cancer [1]. In addition to medicinal plants used as a cure for degenerative diseases, the city of Samarinda began to create efforts to build the independence of the most important treatment of drugs by using available resources, among others, through the use of traditional plants [2]. Native Indonesians have for generations used the advantages of medicinal plants to cure degenerative diseases. Currently, the urban population is already aware of the use of medicinal plants to cure degenerative diseases experienced by both themselves. Microorganisms tend to attach to something surface, multiply, and attach mossy rocks into biofilms. This creates environmental interactions between different species. Biofilms are generally built by one type of microbe. Still, in nature,

biofilms are often found in the form of dual-species or multispecies of bacteria, fungi, algae, yeast, along with foreign substances [3].

The growth of biofilm formed by microbes is currently recognized as the primary mediator of inflammation, estimated that 80% of all inflammatory events are caused by the microbes that make the biofilm [4,5]. Inflammation of biofilm-forming microbes is detrimental to humans because antimicrobial use is shrinking due to the prevalence of antimicrobials against microbial resistance [6]. Biofilms are the unity of a group of organisms such as fungi, bacteria, viruses present on the surface of microbial cells surrounded by a matrix of extracellular polymeric substances [7,8]. Bacteria make up heterogeneous biofilms in space and time. Biofilms continue to grow, which are affected by internal and external processes. Biofilms can be found on the surface of medical devices, bacterial endocarditis, and cystic fibrosis. Biofilms that have been formed can cause antibiotic resistance [9]. *S. aureus* is a globular and gram-positive bacterium, widely distributed in, and some live like normal flora in humans in the axial, inguinal, and perineal areas and the anterior nostrils. Approximately 25-30% of humans carry *S. aureus* in the nasal cavity and skin [10]. *S. aureus* causes the widespread infectious syndrome. Skin infections can occur in moist warm conditions or when the skin is open due to diseases such as eczema, surgical wounds, or intravenous devices [11]. *S. aureus* infection can also come from direct contamination of the wound, for example, a postoperative infection of *S. aureus* or an infection accompanying trauma. If *S. aureus* spreads and bacteriemia occurs, endocarditis, acute hematogenous osteomyelitis, meningitis, or lung infection can occur.

Any tissue or device of the body can be infected by *S. aureus* bacteria and cause the onset of disease with typical signs, namely inflammation, necrosis, and abscess formation [12,13]. *S. aureus* is the second-largest inflammatory bacterium in the oral cavity after Streptococcus alpha bacteria. *S. aureus* causes various types of oral cavity inflammation, such as parotitis, cellulitis, angular cheilitis, and periodontal abscesses [14,15]. Essential oils are present in various organs, such as in the hair of the glands, in the parenchyma cells, in the cavities of schizogenous and listens. Essential oils can be created directly by protoplasm due to the decomposition of resin arrangements from cell chambers or by hydrolysis of certain glycosides [16]. Essential oils are used as raw materials in various industries, such as perfume, cosmetics, pharmaceuticals, and flavoring agents in the food and beverage industry. Essential oils and their constituent components are commonly used in various products, such as cosmetic products, hygiene products, food manufacturing, medicine, fragrance products, and agriculture. According to Ali, essential oils are also essential for therapy, aromatics, and perfumes and used for spirituality [15]. Sourced from the above explanation, researchers are interested in researching with literature review methods or literature review with narrative review approach to see the antibiofilm and antibacterial activity of *S. aureus* from plants containing essential oils with in vitro methods.

**Table 1.** Inclusion journal results.

| Plant Species | Research Methods | Results and Conclusions |
|---|---|---|
| *(Cyperus rotundus L.)* | The research methods in this journal use (MHA) media Mueller Hinton Agar, Trypticase Soya Broth (TSB), Mueller Hinton Broth (MHB), ethyl acetate, toluene, DMSO, vanillin sulfate stain holder, tween 80, Na2SO4 dryer, sterile aquades, violet crystal 1%, glucose 2%, solution ½ McFarland I. | Tuber essential oil provides Minimum Bland Level (KHM) at a concentration of 0.05% inhibiting bacterial growth by 90.68%. Minimum Kill Rate (KBM) at a concentration of 12.5%, killing bacteria by 99.95% of bacterial colonies. Tuber essential oil also provides biofilm inhibition of 89.01% at a concentration of 0.09% [17]. |
| *Cryptocarya massoia (Oken) Kosterm.* and *Cinnamomum verum* | Kirby-Bauer method aims to obtain test concentrations from a combination of masoyi essential oil and cinnamon as an antibacterial. | The study obtained results in antibacterial activity in the combination of 5% masoyi and 10% cinnamon by |

| Plant Species | Research Methods | Results and Conclusions |
|---|---|---|
| | Antibacterial activity is carried out by determining the KHM50, KHM90, and KBM by the microdilution method. | inhibiting the formation of biofilms in test bacteria through the decomposition of active biofilms to be less active [18]. |
| *Ocium Sanctum L.* | Test antibacterial activity using well diffusion methods. The suspension of the test bacteria is calculated over the Muller Hinton Agar plate medium, then made a negative control well hole using sterile aqua dest, while the positive control used is chloramphenicol. It is then uncredited at 370C for 24 hours. Furthermore, the diameter of the bland zone is observed and measured using the mouthpiece term. | Results from the study showed that basil leaf extract (*Ocimum sanctum L.*) might inhibit the growth of both bacteria tested. The bland bacterial zone is formed by *Staphylococcus aureus* ATCC 25923 [19]. |
| *Lippia alba* | Essential oils are extracted from parts three of L. alba specimens by hydrodistillation and analyzed with gas chromatography combined with a mass spectrometer. Minimum Bland Concentration (MIC) and Minimum Bacterial Concentration (MBC) are determined by microdilution methods. For antibiofilm tests, biomass formation in biofilms is evaluated by microtiter-plate technique with violet crystal test (CV) and analyzed the viability of bacterial cells. | The oil and its main components exhibited antibacterial activity, and the lowest MIC and MBC values were 0.5 mg/mL-1 when LA1EO and citral were used. The inhibitory potential (100%) of *S. aureus* biofilm formation at a concentration of 0.5 mg/mL-1 of all essential oils was observed. this is evidenced by the destruction of biofilm cells with concentrations of 1 mg/mL-1, 2 mg/mL-1, and 0.5 mg/mL, respectively. The results obtained in this study showed promising antibacterial and antibiofilm potential of L. alba essential oil against *S. aureus* [20]. |
| *Cinnamomum verum* | The CLSI M7-A10 micro atmospheric method is performed at minimum concentration inhibitors (MIC) and minimum bactericidal concentrations (MBC). The effect of essential oils on biofilms has been calculated and visualized using XTT and Scanning Electron Microscope (SEM). In-vitro toxicity is evaluated using Human Keratinocytes (HaCaT). Chemical analysis of essential oils is performed using Gas Chromatography-Mass Spectrometry (GC-MS). | All strains of bacteria tested were sensitive to cinnamon oil vapor. EO shows 0.5 and 1.0 mg/mL of MIC and MBC against all strain tests. Minimum Biofilm Barrier and Biofilm Eradication Concentration (MBIC50 and MBEC) are 1.0 and 4.0 mg/mL. SEM shows cellular shrinkage, cell wall damage, and decreased biofilm density. Cinnamon oil showed no toxicity to HaCaT cells at any concentration tested [21]. |
| There are 4 types of spices used, namely fennel green (*Pimpinella anisum L.*), *cinnamon* (*Cinnamomumzeylanicum*), cloves (*Syzygium aromaticum*), and cumin (*Cuminum cyminum L.*) | The inhibitory effects of essential oils are evaluated with Agar Well Diffusion, assay determinants, and Minimum Concentration Inhibition (MIC). The most active essential oils, cinnamon, and cloves were tested on adult biofilms 18, 24, 48, 72 hours. | Cinnamon and cloves showed the best results showing significant activity against all bacteria tested. Regarding biofilms, the results suggest that *Cinnamomum zeylanicum* Oil may be a helpful approach to damaging biofilms produced by gram-negative bacteria tested [22]. |
| Essential oils (EO) from *Eucalyptus globulus* LABILL. Cns globulus and from mediterranean native aromatic plants - *Thymus mastichina L., Mentha pulegium L., Rosmarinus officinalis L., Calamintha nepeta (L.) SAVI ssp. nepeta, Cistus ladanifer L., Foeniculum vulgare L., Dittrichia viscosa (L.)* | It uses various methods, namely, agar disc diffusion methods, microdilution methods, violet crystal tests, and Live/Dead staining for biofilm formation assessment. The potential for synergy is assessed by the chessboard method. | EO from *R. officinalis* and *C. ladanifer* indicates domination in monoterpene hydrocarbons (> 60%); EO of *C. nepeta, M. pulegium, T. mastichina, E. globulus*, and F. *vulgare* are rich in oxygenated monoterpenes (62-96%) while EO of D. viscosa consists mainly of oxygenated sesquiterpenes (54%). All EO's exhibit antimicrobial activity; *M. pulegium* and *E. globulus* generally have the most potent antimicrobial activity. The EO of *C. nepeta* is most promising in inhibiting the formation of biofilms. The combination of *D. viscosa/C. nepeta* and *E. globulus/T. mastichina* synergizes against *Staphylococcus aureus* [23]. |
| *Achillea biebersteinii* | The minimum biofilm bland concentration (MBIC) test was conducted using a biofilm inoculator with a 96-well plate with peg led. The minimum bland (MIC) concentration is determined in regular microtiter plates using a double dilution series. | *Achillea biebersteinii* essential oil showed good activity against all bacteria tested. MIC values are in the range of 0.125 - 1 mg / mL while MBIC values are in the range of 0.125 -4 mg / mL. The mechanism of working of *Achillea biebersteinii* essential oil is associated with a strong increase in membrane permeability of 260 nm absorbent materials and potassium ions from cell membranes. *Achillea biebersteinii* essential oil can inhibit early adherence to methicillin-resistant *Staphylococcus aureus* (ATCC 43300) at sub-inhibitor concentrations. *Achillea biebersteinii* essential oil has the potential to be used as an effective antibacterial and antibacterial agent that functions by damaging the permeability of cell membranes resulting in cell death. |
| lemongrass (*Cymbopogon flexuosus*) and grapefruit (*Citrus paradisi*) | Antimicrobial susceptibility screening is done using disk diffusion methods. All strains tested were susceptible to lemongrass, grapefruit, | Compared to other EO tested, lemongrass exhibited the most effective antimicrobial and anti-biofilm activity. Significance and Impact of The Study: The influence of |

| Plant Species | Research Methods | Results and Conclusions |
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| | bergamot, and lime EOs with an inhibition zone varying from 2 85 to 8 to 60 cm even though they were resistant to lemon EO. Lemongrass EO inhibits biofilm formation at 0 125% (v/v) as measured by colorimetric tests and at 0 25% (v/v) no observed metabolic activity is determined by 2,3-bis (2- methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxamide (XTT). Grapefruit EO does not show anti-biofilm activity. After being exposed to EO disorders of widespread lemongrass staph. The aureus biofilm is shown under an electron microscope scan. | lemongrass highlights its EO potential against antibacterial-resistant *Staph. aureus* in a health care environment [24]. |
| citronella (*Cymbopogon nardus*) | evaluation of antimicrobial and antibacterial action of *C. nardus* essential oils (EOCN) and geraniol on Gram-negative and positive bacteria from the determination of minimum bland concentration (MIC) and minimum bactericidal concentration and inhibition of biofilms. | It produces 41 mm halo in S. aureus, which is susceptible with MIC values of 0.5 and 0.25 mg/mL for EOCN and geraniol with bactericidal effects. Antibacterial action has been confirmed, EOCN and geraniol decreased the biomass of *S. aureus* biofilms by 100% between concentrations of 0.5 and 4 mg/mL. The decrease in cell viability is 0.25 and 1 mg/mL, from EOCN and geraniol. EOCN and geraniol are proven to be good antibacterials against S. aureus, promising antibacterials against *S. aureus* [25]. |
| Lippia alba Essential Oil, Citral, and Carvone | It is extracted from three air parts of the L. alba specimen by hydrodistillation and analyzed with gas chromatography combined with a mass spectrometer. Minimum Concentration Barrier (MIC) and Minimum Bacterial Concentration (MBC) are determined by microdilution methods. For antibiofilm tests, biomass formation in biofilms is evaluated by the microtiter-plate technique with violet crystals (CV). Tests and viability of bacterial cells are analyzed. | The oil and its main components exhibited antibacterial activity, and the lowest MIC and MBC values were 0.5 mg/mL-1 when LA1EO and citral were used. The inhibitory potential (100%) of *S. aureus* biofilm formation at a concentration of 0.5 mg/mL-1 of all essential oils was observed. this is evidenced by the destruction of biofilm cells with concentrations of 1 mg/mL-1, 2 mg/mL-1, and 0.5 mg/mL, respectively. The results obtained in this study showed promising antibacterial and antibiofilm potential of L. alba essential oil against *S. aureus* [20]. |
| Peppermint (*Mentha ×piperita L.*), coriander (*Coriandrum sativum L.*), and anise (Pimpinella anisum L.) | Minimum bland concentration (KHM) tests were conducted by double dilution method, and MTT tests against gram-positive *Staphylococcus aureus*) and gram-negative (*Escherichia coli*) bacteria, biofilm growth and development were assessed using violet crystal reduction (CV) and XTT tests. | All EO's (at MIC values of 0.08%- 0.63%) and 8 of 14 plant extracts (at MIC values of 2-4 mg/ml) inhibit bacterial cell attachment in both bacteria.. CV and XTT reduction tests for plant extracts and EO with bacterial attachment inhibition of at least 50% showed that coriander EO had the highest antibacterial activity against biofilms formed by both test bacteria (*S. aureus* and *E. coli*) at the lowest MIC values of 0.08% and 0.16% (v/v), respectively, indicating further investigation due to the high potential of oil antibiofilm activity [26]. |
| *Thymus vulgaris* | Analysis of thyme essential oil is performed using GC/MS analysis. Clinical isolates are isolated using differential diagnostic nutrient media. Antibacterial susceptibility is identified with the help of disc diffusion tests. Agar diffusion tests determine the sensitivity of microorganisms to plant extracts. The antibiofilm activity of the extract was tested in a standard 96-well microtitration plate. | The results proved a broad spectrum of antibacterial activity from thyme essential oil. The highest antimicrobial activity was registered against typical and clinical strains of *S. aureus* and microscopic Candidagenus fungi. Thyme essential oil is certain to show high antibacterial formation activity against *S. aureus* [27]. |
| *Mentha piperita* and *Zataria multiflora* | *Mentha piperita* and *Zataria multiflora* essential oils are obtained using a Clevenger tool. Bacterial cultures are prepared as standard samples. Finally, antimicrobial and anti-biofilm activity is determined by microdilution methods. | The results showed that the lowest concentration of *Zatflora zataria* essential oil was 1.25 mg/ml, while the remaining bacteria were inhibited at a concentration of 2.5 mg/ml. The lowest and highest concentrations of bland were found at 1.25 and 5 mg/ml against *Pseudomonas aeruginosa* [28]. |
| peppermint essential oil | It is investigated by determining electrical conductivity, cell membrane integrity, cell viability, and bacterial morphology. In addition, quantification tests of violet crystals and the number of plate colonies are used to evaluate the effect of PEO in inhibiting and disabling the biofilm *S. aureus.* | The results showed that PEO affects the permeability and integrity of *S. aureus* cell membranes, as evidenced by increased relative electrical conductivity and leakage of nucleic acids, proteins, and ATP. Decreased cell viability and further changes in cell morphology confirm cell membrane damage to *S. aureus* by PEO. In addition, PEO inhibits biofilms formation, and inactive mature biofilms are formed by *S. aureus* [29]. |
| *Eucalyptus staigeriana* | The antimicrobial activity of EOdlES against gram-positive and gram-negative is determined using the disc diffusion method. Micro broth dilution techniques evaluate the minimum bland | EOdlES can inhibit the formation of biofilms but have little or no ability to inhibit the formation of previous biofilms. The study suggests that EOdlES promises an alternative to |

| Plant Species | Research Methods | Results and Conclusions |
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| | concentration value (MIC). The effects of antibiofilm are assessed by the microtiter plate method. | controlling gram-positive foodborne and clinical resistant bacteria [30] |
| *Agastache rugosa* | Essential oils of leaves and flowers are evaluated with GC and GC-MS methods, and floral essential oils reveal the presence of 21 components; of which essential oils are identified, the main compounds are p-Menthan-3-one (48.8%) and estragole (20.8%). At the same time, essential leaf oil is a very effective antimicrobial activity with MIC ranging from 9.4 to 42 lg ml1 and antibacterial potential, antitumor activity for flower essential oil, and leaf essential oil. | Essential oils from *A. rugosa* have antimicrobial, antibiofilm, and cytotoxic activity. So our results offer a reliable basis for resource optimization [31]. |

Based on table 1, journal inclusion can be 20 journals, while the rest entered the criteria of journal exclusion because more researchers who make plant journals that contain essential oils as antibacterials only than as antibiofilm. The profile of the activity and mechanism of essential oils as antibiofilm is seen from the compounds in essential oils, which are dominated by compounds α-siperone, caryopilen oxide, and β-selinenes, and contain monoterpenoid compounds, namely mirtenol, β-pinen and trans-pinocarveol. Sesquiterpenes and monoterpenoids are terpenoid derivatives that have potential as terpenoids themselves can interfere with the release of planktonic cells from biofilms and increase the composition of fatty acids on the cell membrane of biofilms so that cells become more hydrophobic, which will further destroy the biofilm [32,33]. Terpene compounds are thought to interfere with membranes by their lipophilic compounds and thus inhibit the process of respiration and ion transport in bacterial cells from such biofilms. It is at this moment concluded that natural compounds such as terpenes can be used to prevent cell aggregation and the formation of biofilm formations [8,34–36]. While antibacterial, it is shown that essential oils are more effective against Gram-positive bacteria, especially against *Staphylococcus aureus*, than Gram-negative bacteria [37]. Spatulenol compounds, patkulanol, and Essential oils from A. rugosa have antimicrobial, antibiofilm, and cytotoxic activity. So our results offer a reliable basis for resource optimization. Contained in essential oils is classified as a derivative of phenol compounds. Phenol compounds are known to have antiseptic effects and work by damaging cell membranes [38,39]. Antibacterial mechanisms are thought to be due to the binding of phenol compounds to bacterial cells, interfering with membrane permeability and transport processes. This results in the loss of cations and macromolecules from the cell so that cell growth will be disrupted or die. At low concentrations, phenol compounds also result in protein denaturation, and at high concentrations will result in coagulation of proteins so that cells will die [4,8,35].

## 4. Conclusions

After reviewing hundreds of journals using the narrative review method, the results were obtained from google scholar exclusion journal 149, PubMed 58, web of science 200, Scopus 64, science direct 53. And inclusion as many as 20 journals both nationally and internationally. From the 20 inclusion journals that have been discussed, it was concluded that plants containing essential oil compounds have effectiveness as an antibacterial by inhibiting bacterial growth, bactericidal, and damaging the makeup of bacterial cells. As for its effectiveness as antibiofilm essential oil compounds can inhibit the preparation of the biofilm matrix, damage the biofilm matrix, inhibit the formation of biofilms, and destroy the biofilm.

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## Conflicts of Interest

The authors declare no conflict of interest.

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