Journal of Convergence for Information Technology
Vol. 12. No. 2, pp. 192-197, 2022
e-ISSN 2586-4440
DOI : https://doi.org/10.22156/CS4SMB.2022.12.02.192

Comparison Of Antibacterial Activity Of 10 Essential Oils And Oxacillin Against *Staphylococcus Epidermidis*

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표피포도상구균에 대한 에센셜 오일 10종과 옥사실린의 항균비교

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Abstract Although various studies have focused on the relationship between essential oils (EOs) and skin flora, there are only few comparative studies on the antibacterial properties of EOs and their efficacy against common microorganisms. In this study, we tested the antibacterial activity of 10 different EOs against *Staphylococcus epidermidis(S. epidermidis)* with oxacillin as control. Optical density was used to measure the activity of solutions containing the EOs and *S. epidermidis* at two different concentrations. Three EOs (palmarosa, lemongrass, and Mellissa True) had higher antibacterial activity than oxacillin, but their concentrations had negligible effect on antibacterial activity.

Key Words : *Staphylococcus Epidermidis*, Essential Oils, Oxacillin, Alternative Medicine, Phytochemical, Antibiotics

요 약 다양한 연구가 에센셜 오일과 피부 상재균에 관한 초점을 맞추고 있지만, 에센셜 오일의 항균 특성과 미생물에 대한 효능 비교 연구는 거의 없다. 이 연구에서 우리는 옥사실린을 대조군으로 사용하여 표피포도상구균에 대한 10가지 에센셜 오일의 항균 활성을 테스트했다. 두 가지 다른 농도에서 에센셜 오일과 표피포도상구균을 함유한 용액의 활성을 측정하기 위해 광학 밀도(OD)를 사용했다. 옥사실린에 비해 에센셜오일 3종(팔마로사, 레몬그라스, 멜리사 트루)이 항균활성은 더 높았으나 농도는 항균활성에 미미한 영향을 미치는 것으로 나타났다.

주제어 : 표피포도상구균, 에센셜 오일, 옥사실린, 대체의학, 파이토케미컬, 항생제

1. Introduction

The skin serves as the primary barrier in protecting the body from external threats like chemical irritants and microscopic pathogens[1]. It receives tactile input from the external stimuli and plays a role in regulating body temperature. Skin is colonized by microorganisms with various characteristics, like *Staphylococcal* species (*S. epidermidis* and *S. aureus*), *Pseudomonas aeruginosa*, *Escherichia coli*, and *Candida albicans* [2,3]. Although skin flora is generally harmless, under specific stimuli, these microorganisms can break into the body and lead to infection[1]. Although *S. epidermidis* has a low chance of

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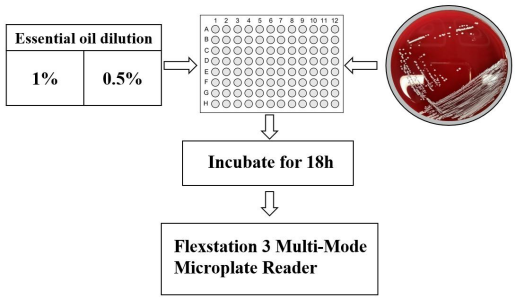
Received December 2, 2021 Revised January 17, 2022
Accepted February 20, 2022 Published February 28, 2022

becoming pathogenic, immunocompromised individuals are at increased risk of developing infections secondary to *S. epidermidis* [4]. Because multiple disorders can lead to a compromised immune system, *S. epidermidis* has become an increasingly important target of study[5,6].

Essential oils (EOs) have natural antibacterial and antioxidant properties[7]. Among various EOs, lemongrass (LG) has antibacterial properties against *Propionibacterium acnes* and *S. epidermidis,* the former being a primary cause of acne [8,9]. Another EO with similar properties is Rosemary verbenone (RM), which has antifungal, antiviral, antibacterial, anti- inflammatory, and antioxidant properties[10,11]. RM has been used effectively in the clinic to treat atopic dermatitis [12]. Palmarosa (PR) is yet another EO with antibacterial activity specific against *P. acnes* [13]. PR also has antibacterial activity against *C. albicans, S. aureus*, and *E. coli* [14]. Oxacillin, an antibiotic similar to penicillin, is effective in treating select *S. epidermidis* infections[15,16]. However, no study has compared the antibacterial properties of oxacillin and EOs. In this study, antibacterial activity against *S. epidermidis* was compared between oxacillin and 10 different EOs.

2. Methods

EOs were provided by the Certification Academy for Holistic Aromatherapy, and 10 Eos were randomly selected for analyses. Table 1 shows the types of EOs in this study. Oxacillin (2 mg/ml) was purchased from Sigma-Aldrich (Steinheim, Germany). A Flexstation 3 Multi-Mode Microplate Reader (Molecular Devices, California, USA) was used along with its spectrophotometer absorption feature to read absorption of all samples at 600 nm wavelength. *S. epidermidis* (KCTC 14990) was purchased from the Korean Collection for Type Cultures (KCTC). *S. epidermidis* was cultured on a nutrient agar plate (MB-N1036, KisanBio, Seoul, Korea) by incubating for 18 h at 37°C. The cultured strains were calculated for their colony-forming units (CFU)/ml according to a 1 × 10^6volume ratio and the CLSI 0.5 McFarland standard, using nutrient broth (Difco, Claix, France). The standard concentration bacteria lsuspension (100 ml) was dispensed into a 96-well plate. EOs were added at concentrations of 1% and 0.5%. Oxacillin (2 mg/ml) was used as a control, and optical density (OD) measured at 600 nm wavelength was used as an endpoint. The experimental method is shown in Fig. 1 and Table 1 This process was repeated four times to increase the accuracy of the results. This study was conducted with the approval of the Ethics Committee of Dankook University (IRB File No. NON2021-002) and in accordance with the Declaration of Helsinki.



1% and 0.5% concentrations of essential oils and S. epidermidis were dispensed in a 96-well plate, incubated for 18 h, and their absorbance measured.

Fig. 1. Experimental method.

Table 1. Classification of essential oils according to extraction site

Oils name	Botanical name	Origin	Abbreviation
Cypress	*Cupressus sempervirens*	Austria	CP
Lemongrass	*cymbopogon schoenathus*	Guatemala	LG
Manuka	*Leptospermum scoparium*	New Zealand	MK
Melisa True	*Melissa officinalis*	Italy	MS
Palmarosa	*Cymbopogon martini*	India	PR
Rosemary verbenone	*Rosmarinus officinalis*	France	RM

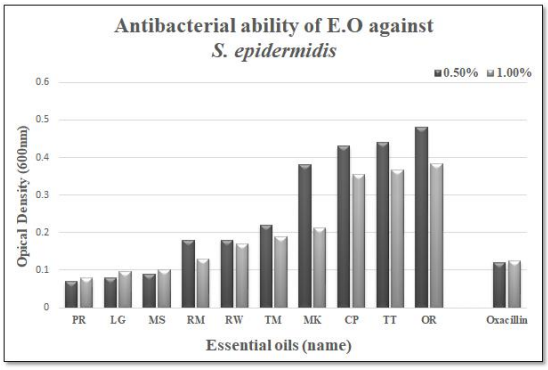
(Continued)

Table 1. Classification of essential oils according to extraction site.

Oils name	Botanical name	Origin	Abbreviation
Tea Tree	*Melaleuca alternifolia*	Australia	TT
Thyme white	*Thymus vulgaris*	Spain	TM
Orange Sweet	*Citrus sinensis*	Argentine	OR
Rosewood	*Aniba rosaeodora*	Brazil	RW

3. Results

More than 30% of the 10 EOs showed some level of antibacterial activity. three EOs showed higher antibacterial activity than oxacillin, as shown in Table 2 and Fig. 2. LG, Melissa True (MS), and PR showed higher antibacterial activity than oxacillin. Orange sweet had lower antibacterial activity than oxacillin.



Dark and light gray indicate 0.5% and 1% concentrations of EOs, respectively. Optical density values are shown in the graph. Palmarosa (PR), lemongrass (LG), and Melissa True (MS) showed better antibacterial activity than oxacillin against *S. epidermidis.*

Fig. 2. Comparison of antibacterial activity of whole essential oils and oxacillin.

Table 2. Optical density (%) of essential oils at 600nm wavelength.

Name	OD (0.5%)	OD (1.0%)
Lemongrass*	0.08	0.10
Mellissa true*	0.09	0.10
Palmarosa*	0.07	0.08
Oxacillin (control), 0.12		

(Continued)

Table 2. Optical density (%) of essential oils at 600nm wavelength.

Manuka	0.38	0.35
Cypress	0.43	0.17
Orange sweet	0.48	0.37
Rosemary	0.18	0.13
Rosewood	0.18	0.19
Tea tree	0.44	0.38
Thyme	0.22	0.21

* Higher antibacterial activity than oxacillin OD, optical density.

4. Discussion

Studies have shown that both RM and LG have excellent antibacterial activity against *S. epidermidis*[17,18], but in this study, PR showed the highest antibacterial activity among the tested compounds. PR and thyme white (TM) have previously shown high antibacterial activity against both *E. coli* and *S. epidermidis* strains[19]. In this study, PR showed the highest antimicrobial activity at both concentrations (0.5% and 1%). Furthermore, PR previously showed high antibacterial activity against gram-negative and positive bacteria[20]. Our results also showed high antibacterial activity of PR against *S. epidermidis*, a gram-positive bacterium.

LG exudes high antibacterial activity against *S. epidermidis*[21-24], and this was demonstrated in our study as well, with LG exhibiting high antibacterial activity against *S. epidermidis*. In previous studies, MS showed low antibacterial activity against several pathogenic strains[25]. However, one study reported that certain gram-negative pathogenic bacteria were highly sensitive to MS, including *P. aeruginosa, E. coli, Salmonella enteritidis, Salmonella typhi, E. coli* ATCC 25922, *Shigella sonnei* IPH -MR strain, and other Shigella strains[26]. In this study, MS showed higher antibacterial activity against *S. epidermidis* than oxacillin.

A previous study reported that oxacillin has high antimicrobial activity against *S. epidermidis* strain[27]. Here we found that PR, LG, and MS had higher antibacterial activity against *S. epidermidis* than oxacillin. Tea tree is also reported to have higher antibacterial activity than oxacillin against several pathogens [25], but this was not observed in our study. Previous studies also reported that cypress lacks antibacterial activity at a concentration of 10% against any strain[28].

EOs are reported to show a dose-dependent antibacterial activity, that is, their antibacterial activity increased with their concentrations[20]. In our study, we did not observe a difference in antibacterial activity EOs at different concentrations.

Further research should employ higher EO concentrations, different bacterial species, and comparative studies involving antibiotics and other EOs[29,30].

The authors declare that there are no conflicts of interest.

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