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Antibacterial Potential of Freshener Water Based on Siwalan Coir Extract (*Borassus flabellifer*) and Lemongrass (*Cymbopogon citratus*) Against Airborne Bacteria

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Abstract

Air is an important part of life. The Quality of air can be seen from three parameters, namely chemical, physical, and biological parameters. One of the biological parameters of air quality can be seen from the presence of microorganisms such as bacteria. The presence of airborne microorganisms, especially in enclosed spaces can affect human health, one of which can cause respiratory infections. On the other hand, people often use air freshener products. Therefore, this research was conducted with the aim of analyzing the potential of *air freshener* based on siwalan (*Borasus flabellifer)* coir extract and lemongrass as antibacterial against airborne bacteria. This research is an experimental research with *a pre test-post test design.* The data obtained is in the form of the value of the Total Plate Count of indoor air bacteria before and after treatment using air *freshener* based on coir extract of siwalan and lemongrass. The results showed that both formulations (P1 and P2) of *air freshener* based on coir extract and lemongrass were able to reduce the number of airborne bacterial colonies in the room, with a significance value of 0.000<0.05.

Keywords: Air freshener, Antibacterial; Lemongrass; Siwalan.

INTRODUCTION

Air is one of the important parts in the life of living things. In addition to oxygen, air also contains fungi, bacteria, carbon monoxide, carbon dioxide and other gaseous materials (Abidin & Hasibuan, 2019). At normal limits, these substances will be neutralized (Dewi et al., 2021). However, if it has exceeded normal limits and can threaten human health, then the condition is said that air pollution has occurred (Arwini, 2019).

Some studies state that the level of air pollution indoors is greater than outdoors (Rahmawati & Khairina, 2020). WHO (2009) states that humidity and the presence of indoor microorganisms are the main causes of morbidity and mortality in the world.

The cleanliness level of room air greatly affects human health because 90% of human activities are carried out indoors (Bahri et al., 2021). Problems related to the level of room air quality include three parameters, namely chemical, physical, and biological parameters characterized by the presence of airborne microorganisms (Rahmawati & Khairina, 2020).

Microorganisms, especially in a closed room need to be paid attention to, considering the many symptoms of diseases such as eye irritation, acute respiratory infections, and skin diseases due to infection by airborne microorganism (Putra et al., 2018). In fact, the majority of cases of infection occur due to the spread of bacteria through the air (Nisyak & Hartiningsih, 2020). One of the microorganisms that cause infections that are often found in indoor air is *Staphylococcus aureus* (Nisyak et al., 2020). The microorganisms are able to survive in the air for four to six hours. Therefore, the use of natural antibacterial needs to be developed to reduce the spread of pathogenic bacteria in the air, for example in air freshener products.

Air freshener also acts as an air purifier. This product releases volatile compounds into the air, so that if made of chemicals it can trigger the release of secondary pollutants (Kim et al., 2015). These secondary pollutants can cause disorders of the nerves, hormones, lung and cardiovascular system, can also damage the ozone layer (Singer et al., 2006). Therefore, it is necessary to develop

air freshener with natural ingredients, such as siwalan coir and lemongrass (Yoviska et al., 2022).

Siwalan (*Borassus flabellifer*) coir waste from siwalan fruit that has not been widely used. Fariha et al. (2020) said that 65-75% of the weight of the fruit is siwalan coir. Siwalan coir contains natural antioxidants, volatile and non-volatile fatty acids (Dahlan, 2011). Antioxidants in coir siwalan can prevent cell damage due to free radicals (Idayati et al., 2014).

While lemongrass (*Cymbopogon citratus*) contains flavonoids, saponins, polyphenols, and essential oils (Departemen Kesehatan Republik Indonesia, 2008). The lemongrass oil contain of citral compounds, essential oils with geraniol content of 65-90%, geranyl acetate 3-8%, citronellol 11-15%, citronellyl acetate 2-4%, citronellal 30-45%, cavikol, citral, kadinol, eugenol, vanillin, cadinen, kamfen, elemol, and limonene which makes lemongrass can be used as air freshener (Zurairah et al., 2021). Citral compounds contained in lemongrass oil play an important role as antibacterial because they can damage bacterial cell membranes (Lu et al., 2018). Chamdit & Siripermpool (2012) in their research said that the combination of eugenol and lemongrass oil can damage the biofilm *of S. aureus.*

Therefore, this research was conducted with the aim of seeing the potential of air freshener made from siwalan coir and lemongrass as antibacterial against indoor air bacteria. Thus, this air freshener based on siwalan coir and lemongrass is expected to improve air quality based on biological parameters, besides being known to also provide a relaxing effect and give a distinctive aroma to the room (Yoviska et al., 2022).

MATERIALS AND METHODS

This research used an experimental design conducted for 3 months (May-July 2023) at the Biological Laboratory of Universitas PGRI Ronggolawe.

Research Instruments

The tools in this research were beakers, erlenmeyers, test tubes, split funnels, petri dishes, digital balances, drip pipettes, distillation and soxhletation device sets, *autoclaves*, incubators, *colony counters*, space thermometers, hygrometers, glass bowls, statifs, scissors, insulation, *hot plates*, masks, *glove*, glass bottles, tissues, paper labels and *fiber reed sticks*. While the ingredients used were siwalan coir, lemongrass, distilled water, 96% ethanol, 90% isopropyl alcohol, and Nutrient Agar (NA) media.

Procedures

Air Freshener Making Process

The making process of air freshener was carried out referring to Yoviska et al. (2022)'s research includes the manufacture of siwalan coir and lemongrass extracts. Siwalan and lemongrass are dried 3 days and blended. After that, it was dissolved with ethanol and distilled at a temperature of 70^0C. Formula 1 (P1) was made with a composition of 11.8% siwalan coir extract, 0.2% lemongrass extract, 0.2% isopropyl alcohol, and 87.6% distilled water. While formula 2 (P2) was 7.6% coir extract, 0.3% lemongrass extract, 0.3% isopropyl alcohol, and 91.7% distilled water.

Measurement of Room Environmental Factors

Indoor environmental factors measured include air humidity and temperature. Air temperature was measured using a room thermometer and air humidity was measured using a hygrometer.

Air Freshener Antibacterial Test

In this research, a room with a size of 4x4m with air conditioning was used. The number of airborne bacterial colonies was identified in the room by air capture method with a petri dish containing 10mL of sterile NA media placed in an open room for 10 minutes. Then, the petri dish is closed and incubated for 24 hours in an incubator at 27°C. Furthermore, quantitative tests are carried out by calculating the Total Plate Count (TPC) of bacteria.

The treatment of air freshener of coir extract of siwalan and lemongrass was carried out for 3 days. Furthermore, every 24 hours during the treatment, air bacteria are taken by the same method when the initial identification of the number of colonies.

Data analysis

The data were statistically analyzed using the *Paired T Test* to determine the ability of *air freshener* coir extract and lemongrass in reducing the number of airborne bacterial colonies in space. While the effect of application time was tested using *one way Anova* with a significance level of 95% (α 0.05). Before the *Anova one-way* test, a normality test was carried out on the data.

RESULTS AND DISCUSSION

Indoor Environmental Factors

Environmental factors are important parameters to measure. This is because the growth of bacteria, especially in a room is strongly influenced by environmental factors. Environmental factors that can be measured in this research include air humidity and temperature. The results of measuring air humidity and room temperature in this research are shown in Table 1 below.

Table 1. Room Air Temperature and Humidity.

Room	Air Temperature (ºC)	Humidity (%)
Room 1	20	26,7
Room 2	20	28,2

Temperature is a factor that affects bacterial growth directly. Temperature affects the work of enzymes in bacterial cells, where enzymes are catalysts in biochemical reactions in bacterial cell metabolism. In Table 1 it appears that the air temperature of the two rooms used in this research is on average 20^0C. This temperature is in the optimum temperature range for the growth of mesophyll group bacteria (Lestari & Hardisari, 2019). However, this conditions has met the requirements for healthy air temperature for the room, where the requirements for air temperature in a healthy room are based on Peraturan Menteri Kesehatan Republik Indonesia (2011) is 18-30^0C.

In the air humidity parameter, it is known that the air humidity in room 2 is slightly higher than the air humidity in room 1. However, the humidity value in both spaces in this research was lower than the humidity for indoor healthy air based on Peraturan Menteri Kesehatan Republik Indonesia (2011). The government through Peraturan Menteri Kesehatan Republik Indonesia (2011) had set the air humidity for healthy air requirements in the room is 40-60%. However, Astuti, Hastutiningrum, & Sudarsono (2022) mentioned that very dry air can inhibit the growth of bacteria.

Air Freshener Antibacterial Test

Air freshener antibacterial test based on coir and lemongrass against airborne bacteria in this research was conducted by comparing the TPC value of airborne bacteria before and after treatment. Identify the number of air colonies using the air capture method, where petri dishes containing sterile NA media are placed in each corner of the room with an open state for 10 minutes. This is done in order to get bacteria from the air. Furthermore, incubation is carried out for 24 hours at a temperature of 27ºC. The count of the number of bacterial colonies was carried out at hours 0, 24, 48, and 72 in 2 rooms, where in room 1 Formula 1 (P1) was applied and in room 2 Formula 2 (P2) was applied. Average data on the number of airborne bacterial colonies are presented in Table 2.

Table 2. Average Number of Airborne Bacterial Colonies in Room.

Room	Number of Bacterial Colonies after treatment at the hour of-			
	0	**24**	**48**	**72**
Room 1	14	13	11	9
Room 2	23	21	16	11

Data in Table 2 shows that air freshener applications based on coir and lemongrass are able to reduce the number of airborne bacterial colonies in space. This is due to the content of metabolite compounds, namely saponins, tannins, alkaloids, and triterpenoids that can inhibit bacterial growth (Pamput, 2023). While the citral component in lemongrass oil has an important role as antibacterial because it damages cell membranes in bacteria (Lu et al., 2018). However, the difference in formulations applied to Room 1 and Room 2, based on the results of statistical tests one*way anova* that has been carried out, did not show a significant difference in reducing the number of colonies of airborne bacteria in the room (sig. 0.246>0.05). This result is influenced by the composition of the active ingredients of the coir extract used. In P1, coir extract used was 11.8% and in P2 7.6%. While P1 contains lemongrass extract as much as 0.2% and P2 as much as 0.3%. This difference does not have a significant impact on the antibacterial content in it, so it does not provide a significant difference in reducing the number of airborne bacterial colonies in room.

While the percentage decrease in the number of airborne bacterial colonies after treatment using air freshener based on siwalan coir and lemongrass is shown in Figure 1. Based on Figure 1 and the results of *oneway Anova* analysis, it is known that the length of application of air freshener based on siwalan coir and lemongrass has a significant effect on the percentage of reducing bacterial colonies with a sig value of 0.000<0.05. This shows that the longer the application time, the greater the decrease in the number of airborne bacterial colonies in the room. This result occurs because the longer the application time, the contact of the active compounds contained in the *air freshener* will be more thorough to the airborne bacterial cells in the room.

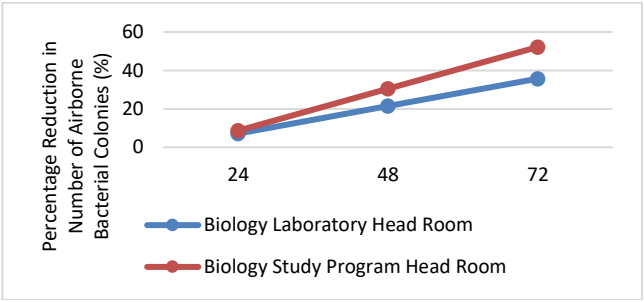

Figure 1. Percentage Decrease in the Number of Airborne Bacterial Colonies in the Room after Treatment with *Air Freshener* Based on Siwalan Coir and Lemongrass

CONCLUSIONS

The conclusion of this research is that formulation 1 (P1) and formulation 2 (P2) applied to two different rooms did not show a significant difference, but the two formulations from the results of one-way Anova analysis found that the length of application of air freshener based on siwalan coir and lemongrass had a significant effect on the percentage of reducing bacterial colonies with a sig value of 0.000<0.05. This explains that the longer the

application time, the number of airborne bacterial colonies in the room will also decrease greater.

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Authors' Contributions: The First Author monitored the preparation and implementation of research, evaluation for each stage, and data analysis; The Second Author tidied up administrative matters and prepared manuscripts; The Third Author recorded and managed administrative costs for research and coordinate the purchase of research tools and materials and documentation; The Fourth Author worked on the completion of the manuscript; The Supervisor played a role in directing, evaluating the results of teamwork, and finalizing article manuscripts.

Competing Interests: The authors declare that there are no competing interests.

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