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ORIGINAL ARTICLE

The Effectiveness of Palm Kernell Shell Liquid Smoke (Elaeis Guineensis Jacq) as Antiseptic Against Microbials

Efektivitas Asap Cair Kulit Sawit (Elaeis Guineensis Jacq) sebagai Antiseptik terhadap Mikroba

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ABSTRACT

Background

Bacteria and fungi that enter through the body get intermediaries from the condition of hands that are not washed clean. Alcohol-based antiseptics are usually used for quick and effective hand washing, but repeated use can cause irritation and dryness of the hands. Natural components can be processed to make non-alcoholic substitutes, such as liquid smoke from palm oil's outer shell (Elais guineensis Jacq). It is known that liquid smoke contains antibacterial phenolic chemicals.

Methods

This research design is true experimental by testing the effectiveness of palm oil outer shell liquid smoke as an antiseptic, which is carried out in vitro using inhibitory tests and in vivo using palm swabs of respondents by looking at the effects before and after use. In vitro results formed the largest inhibition zone size using grade 2 liquid smoke with a concentration of 75%, with an inhibition zone size of Staphylococcus aureus ATCC 25923 of 15.6 mm; Escherichia coli ATCC 25922 of 5.8 mm; Candida albicans ATCC 10231 of 3.3 mm; and Aspergillus flavus ATCC 9643 of 33.3 mm.

Results

In vivo, results obtained through the respondent's palm swab showed an average value of 89.84% decrease in bacterial colonies and a 77% decrease in fungal colonies after using liquid smoke antiseptic. The results of the organoleptic assessment showed that 92% of respondents liked the color, 75% liked the smell, 75% felt that it did not cause dryness, and 92% felt that there were no side effects of use.

Conclusions

The liquid smoke of the outer shell of palm oil has the potential as an antiseptic.

Keywords: Antiseptic; Liquid Smoke; Palm Kernell Shell.

ABSTRAK

Latar Belakang

Bakteri dan jamur yang masuk melalui tubuh mendapatkan perantara dari kondisi tangan yang tidak dicuci dengan bersih. Antiseptik berbasis alkohol biasanya digunakan untuk mencuci tangan dengan cepat dan efektif, namun penggunaan secara berulang mengakibatkan iritasi dan kekeringan pada tangan. Komponen bahan dasar alami dapat diolah untuk membuat pengganti antiseptik non-alkohol contohnya asap cair yang terbuat dari kulit luar sawit (*Elais guineensis* Jacq). Diketahui bahwa asap cair mengandung bahan kimia fenolik antibakteri.

Metode

Rancangan penelitian ini adalah eksperimental murni dengan menguji efektivitas asap cair kulit luar sawit sebagai antiseptik yang dilakukan secara *in vitro* menggunakan uji daya hambat dan *in vivo* menggunakan *swab* telapak telapak tangan responden dengan melihat efek sebelum dan sesudah penggunaan.

Hasil

Secara *in vitro* terbentuk ukuran zona paling besar menggunakan asap cair *grade* 2 konsentrasi 75% dengan ukuran zona hambat *Staphylococcus aureus* ATCC 25923 sebesar 15.6 mm; *Escherichia coli* ATCC 25922 sebesar 5.8 mm; *Candida albicans* ATCC 10231 sebesar 3.3 mm; dan pada *Aspergillus flavus* ATCC 9643 sebesar 33,3 mm. Hasil *in vivo* melalui *swab* telapak tangan responden diperoleh nilai rata-rata penurunan jumlah koloni bakteri sebesar 89,84%, dan nilai rata-rata penurunan jumlah koloni jamur sebesar 77% setelah menggunakan antiseptik asap cair. Hasil penilaian organoleptik didapatkan 92% responden menyukai warna, 75% responden menyukai aroma, 75% responden merasa tidak menimbulkan kekeringan, dan 92% responden merasa tidak ada efek samping penggunaan.

Kesimpulan

Asap cair kulit luar sawit berpotensi sebagai antiseptik.

Kata Kunci: Antiseptik; Asap Cair; Kulit Sawit.

INTRODUCTION

Indonesia has a high prevalence of infectious diseases, some of which, such as tuberculosis, diarrhea, and typhoid fever, are often found in society and contribute to the high mortality rate.[1] Fungi and bacteria are the most important causative agents of these infectious diseases.[2] Bacteria often infect humans. Staphylococcus aureus and Escherichia coli. S. aureus can trigger infections such as pneumonia, acne, boils, and urinary tract infections. Other bacteria, namely E. coli, cause diarrhea and sepsis.[3] Cases of Extraordinary Events (KLB) of diarrhea were recorded 21 times and spread to 12 provinces with 1.725 infected people, followed by the death of 34 people.[4]

Apart from bacteria, the fungi that often infect are Candida albicans and Aspergillus flavus.[5] Candida albicans trigger an infection of the hair, skin and nails called candidiasis. The prevalence in Indonesia for this disease is recorded at 20-25%.[6] Aspergillus flavus can be the main factor causing cancer due to aflatoxin.[7] In Kenya in 2004, aflatoxin poisoning occurred in which 317 people were poisoned, and 215 people died.[8] Poor hand hygiene is a transmission of pathogens to enter the body's tissues.[9]

Antiseptic hand sanitizer is a hand-cleaning product that can quickly reduce the number of microbes because it contains alcohol.[10] Prolonged use of alcohol-based antiseptics causes skin

dryness and irritation.[11] Natural compounds derived from plants can substitute safe non-alcoholic antiseptics.[12]

Oil palm outer skin (Elais Guinness Jacq) is a biomass material that can be pyrolyzed into products such as charcoal and liquid smoke because it contains organic matter such as cellulose, hemicellulose, and lignin.[13] Lignin undergoes pyrolysis, decomposing into less complex substances such as phenol and its derivatives. Phenol is an active ingredient with antimicrobial, antioxidant and antibacterial effects.[14]

Liquid smoke is indeed known to have antimicrobial properties. It can potentially replace alcohol as a basic ingredient for making antiseptics. Still, laboratory tests are needed to validate the feasibility of using liquid smoke as an antiseptic. Based on the description above, researchers are interested in testing the effectiveness of palm oil outer skin liquid smoke as an antiseptic compared to alcohol against microbes.

METHODS

The design of this study used pure experimental research carried out in vitro and in vivo, liquid smoke testing at the Rajawali Health Institute microbiology laboratory from January to February 2022. The in vitro test was carried out by an inhibition test to assess the antimicrobial effectiveness of liquid smoke against microbes, while the in vivo test palm swabs were carried out on respondents before treatment and after treatment with liquid smoke.

The in vitro test subjects of this study were Staphylococcus aureus ATCC 25923, Escherichia coli ATCC 25922, Candida albicans ATCC 10231 and Aspergillus flavus ATCC 9643, which were purchased from the West Java Provincial Health Laboratory and the Faculty of Medicine, Padjadjaran University. Meanwhile, the in vivo test subjects of this study consisted of 24 respondents who were determined using the Federer Formula, taken with the criteria of respondents who had not washed their hands, had no allergies to antiseptics, and had no wounds on the palms of their hands.

The process for making grade 1 and grade 2 palm outer shell liquid smoke was obtained by redistilling the previous grade liquid smoke made in the School of Life Sciences and Technology, ITB campus laboratory.

In the in vitro test, the inhibition test was carried out by placing paper discs dripped with liquid smoke on the surface of the nutrient agar medium, which had been planted with bacterial cultures. In contrast, the potato dextrose agar medium, which had been planted with mushroom cultures, was made by adding wells with liquid smoke. The inhibition test was carried out at concentrations of 35%, 50%, and 75% liquid smoke of the outer skin of the palms grades 1 and 2. The inhibition zone results were measured with callipers and compared for each concentration at each grade to take the grade concentration that optimally inhibited microbes.

The in vivo test was carried out by administering liquid smoke to the palms of the respondents according to the grade and optimal concentration of the in vitro test results. The number of research samples was 24 respondents divided into two groups, each consisting of 12 people for giving liquid smoke as a test material and 70% alcohol as a control. Respondent's palm swabs were carried out before and after administering the antiseptic. The suspension of the swab

was planted on nutrient agar medium and potato dextrose agar medium by pour plate. Colonies growing on the swab result medium before the use of antiseptics were counted and compared after the use of antiseptics. The number of colonies was counted using the Standard Plate Count method.

The results of the in vivo test were then related to the organoleptic assessment of the liquid smoke that the respondent used. The data was obtained using a questionnaire sheet filled in by the respondent. Some assessment parameters include color, aroma, dryness, and itching and irritation effects after using liquid smoke. The rating scale starts from 0 (dislike), 1 (fair), 2 (like), and 3 (like very much).

RESULTS

Liquid smoke from the outer skin of grade 1 and grade 2 palm oil which had been obtained, was observed for several physical properties. The physical properties of liquid smoke from the outer skin of the palm are presented in Table 1.

Table 1. Physical properties of liquid smoke from the outer skin of the palm

Characteristic	Palm Oil Outer Shell Liquid Smoke 35%		Palm Oil Outer Shell Liquid Smoke 50%		Palm Oil Outer Shell Liquid Smoke 75%	
	Grade 1	Grade 2	Grade 1	Grade 2	Grade 1	Grade 2
Color	Yellow	Yellow	Yellow	Yellow	Yellow	Yellow
Scent	Strong	Strong	Strong	Strong	Strong	Strong
Transparency	Clear	Clear	Clear	Clear	Clear	Clear
pH	3.4	3.1	3.2	3.0	3.1	2.9

pH = degree of acidity

The results of the observations showed that in terms of color, grade 1 and grade 2 at various concentrations that had been made had a yellow color with clear transparency, had a characteristic pungent odor, and had a pH of grade 1 at a concentration of 35%, namely 3.4; pH concentration 50% 3.2; and pH concentration of 75% 3.1. Whereas in grade 2, the concentration of 35% is 3.1, the pH of the 50% concentration is 3.0, and the 75% concentration is 2.9. The pH of liquid smoke ranges from 2.9 to 3.4, with the pH obtained in grade 2 being lower (acidic) than grade 1 liquid smoke, although the difference seems very slight, namely 0.2 to 0.3.

The test microbes used in this study were S. aureus ATCC 25923, E. coli ATCC 25922, C. albicans ATCC 10231, and A. Flavus ATCC 9643, identified macroscopically and microscopically. The identification of microbes is presented in Table 2.

Table 2. Identification of microbes

No.	Microbial Type	Cell Morphology	Colony Morphology
1	Staphylococcus aureus ATCC 25923	Coccus, purple in color which is gram positive	Colonies are round, white, convex elevation with flat sides
2	Escherichia coli ATCC 25922	Bacillus, red in color which is gram negative	Colonies are round, white, convex elevation with flat sides
3	Candida albicans ATCC 10231	Oval, purple in color which is gram positive	Colonies are round, yellowish white in color, smooth texture, convex elevation
4	Aspergillus flavus ATCC 9643	Conidia globosa, conidiophores clearly visible, round vesicles	Colonies in the form of granules like a velvet, greenish yellow in color

Note: ATCC = *American Type Culture Collection*

The identification results showed that S. aureus was a gram-positive bacterium and E. coli was a gram-positive bacterium. Meanwhile, C. albicans forms round colonies with oval-shaped cell morphology and is gram-positive, while A. flavus forms granular colonies like velvet with cell morphology forming conidia globosa.

The in vitro test showed that the higher the concentration of the liquid smoke of the outer skin of the palm used, the greater the zone of inhibition formed against S. aureus, E. coli, C. albicans, A. flavus. The results of the inhibition test of

The liquid smoke of the outer skin of the palm are presented in Figure 1. The results of the zone of inhibition of liquid smoke of grade 1 palm outer skin on the growth of S. aureus at a concentration of 35% were 3.1 mm, 50% concentration was 5.5 mm, and 75% concentration was 10.1 mm.

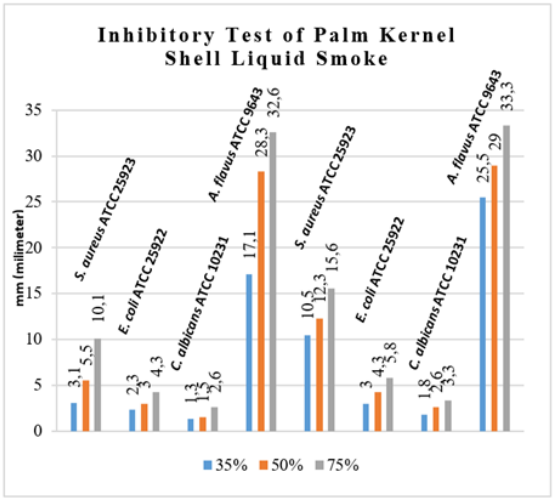

Figure. 1. Test the inhibition of liquid smoke of the outer skin of grade 2 palm oil with a concentration of 35%, namely 10.5 mm, a concentration of 50%, 12.3 mm, and a concentration of 75%, 15.6 mm. The results of the zone of inhibition of liquid smoke of grade 1 palm outer skin on the growth of E. coli at a concentration of 35% were 2.3 mm, 50% concentration was 3.0 mm, and 75% concentration was 4.3 mm. In grade 2, the 35% concentration is 3.0 mm, the 50% concentration is 4.3 mm, and the 75% concentration is 5.8 mm.

The inhibition test results of grade 1 palm outer skin liquid smoke on the growth of C. albicans were a concentration of 35%, namely 1.3 mm, a concentration of 50%, 1.5 mm, and a concentration of 75%, which was 2.6 mm. In grade 2, the concentration of 35% is 1.8 mm, 50% is 2.6 mm, and 75% is 3.3 mm. The results of the inhibition zone of liquid smoke of grade 1 palm outer skin on the growth of A. flavus at a concentration of 35% were 17.1 mm, 50% concentration of 28.3 mm, and 75% concentration of 32.6 mm. In grade 2, the concentration of 35% is 25.5 mm, 50% is 29.0 mm, and 75% is 33.3 mm.

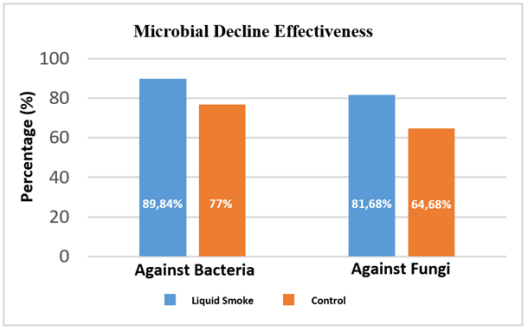

Figure 2. Percentage of microbial reduction effectiveness

Based on in vivo tests, the liquid smoke of the palm's outer skin can inhibit the growth of bacteria and fungi. The percentage of microbial reduction effectiveness is presented in Figure 2.

Showed that 75% liquid smoke of the outer skin of the palm was effective as an antiseptic because it can inhibit the growth of bacteria and fungi with an average decrease in the number of bacterial colonies of 89.84% and an average decrease in the number of fungal colonies of 77%. Meanwhile, 70% alcohol as a comparison antiseptic control had an average decrease in bacterial colonies of 77.00% and an average decrease in fungal colonies of 64.68%.

The assessment of the liquid smoke antiseptic of the outer skin of the palm was carried out after the respondent used it by filling out a questionnaire. The results of the questionnaire on the use of liquid smoke antiseptic for the outer skin of the palm are presented in Figure 3.

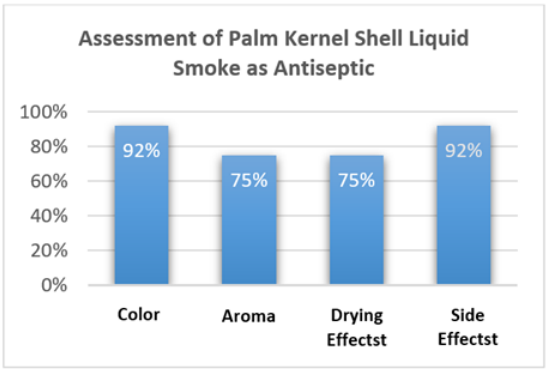

Figure 3. Evaluation of the liquid smoke antiseptic of the outer skin of the palm

Based on the data in Figure 3, it shows that 92% of respondents like color, 75% of respondents can accept aroma, 75% of respondents have no problems related to dryness caused, and 92% of respondents do not get side effects such as itching, burning or red rashes on their skin. Skin from liquid smoke of the outer skin of the palm 75%.

DISCUSSION

Grade 1 and grade 2 palm bark liquid smoke both have a clear yellow color and a pungent aroma, like liquid smoke in general, and have a pH of grade 1 at a concentration of 35%, namely 3.4, a pH of 50% concentration, 3.2, a pH concentration of 75%. 3.1. While the pH at grade 2 concentration

of 35% is 3.1, the pH of the 50% concentration is 3.0, and the pH of the 75% concentration is 2.9. Grade 1 liquid smoke is the result of purification by redistillation from grade 2 liquid smoke which has a clearer color, lighter aroma, and a high content of phenol and acid compounds, so grade 1 liquid smoke has a lower pH value than other grades. [15] However, based on Table 1. grade 2 liquid smoke is liquid smoke with the lowest pH (acidic) compared to grade 1 liquid smoke. This is due to the temperature and time of distillation will affect the pH value; liquid smoke distilled at a higher temperature high will result in a lower pH due to increased levels of acid and phenolic compounds. [16]

The quality of liquid smoke can be determined by its pH value. The lower the pH, the higher the quality, and the lower pH of liquid smoke indicates the high content of phenolic and acidic compounds that can be used to prevent microbial activity. [17]

The macroscopic observation of S. aureus ATCC 25923 showed that the colonies appeared round, with convex elevations, and white. The colonies on E. coli ATCC 25922 are small, rounded, convex elevations with smooth margins. E. coli ATCC 25922 belongs to a group of gram-negative bacteria that retain the red color of the safranin substance, whereas S. aureus ATCC 25923 retains crystal violet as the main substance, which does not fade due to the cell wall of gram-positive bacteria. [18]

Macroscopically, A. flavus ATCC 9643 has a textured velvet in the form of granules (granules) because it belongs to the mould group with mycelium thickening. Whereas C. albicans ATCC 10231 is a yellowish-white, round, convex elevation which belongs to the yeast group. Microscopic results of A. flavus ATCC 9643 show blue blisters resulting from the cotton blue component of LPCB. On C.albicans ATCC 10231, the purple color results from gram staining, which shows gram-positive due to the color defence even though it has been decolorized. [19]

Grade 1 and grade 2 palm outer shell liquid smoke inhibited S. aureus, E. coli, C. albicans, and A. flavus at various concentrations studied, namely 35% concentration, 50% concentration, and 75% concentration. The results of the inhibition test in Figure 1 shows that the inhibition zones formed tend to be larger at higher concentrations because the higher the concentration of the liquid smoke content, the higher the concentration of the active antimicrobial ingredients, making it more effective in disrupting microbial activity. [20] The outer shell of liquid smoke Palm oil contains antimicrobial compounds, including phenols, carbonyls, and acid compounds

At high concentrations, the mechanism of phenol as an antimicrobial work by agglomerating proteins in the cell wall through hydrogen bonds between hydroxyl groups which causes damage to the permeability of parts of the membrane and its function is unstable because the cell structure has changed, so that the components that secrete components inside the cell. Whereas at low concentrations, phenol can form weak hydrogen bonds with proteins, affecting nutrient transport in the bacterial cell wall. [21] Apart from phenolic compounds, as for the acidic compounds that affect the inhibition zone formed, the acidic compounds from liquid smoke can destroy protein compounds. , hydrolyzes fat, and acidifies the cytoplasm so that the microbial cell membrane becomes damaged, which disrupts the stability of the structure and function of the cell membrane as a transport medium for the active substances needed. [22]

Grade 2 liquid smoke from the outer skin of palm oil with a concentration of 75% is effective as an antiseptic because it can inhibit the growth of bacteria and fungi, as shown in Figure 2. The bioactive ingredients contained in liquid smoke, such as alcohol, acid, and phenol groups, together

inhibit the growth of microbes by interfering with the physiological structure of microbes. [23] Grade 2 liquid smoke is liquid smoke that has been redistilled so that the impurities such as tar and carcinogenic benzo(a)pyrene compounds have mainly been removed; this redistillation process causes the pH value to increase to the direction is more acidic due to the increase in the content of phenol and acid in liquid smoke.

Respondents' assessment of liquid smoke antiseptic outer shell at a concentration of 75% can be seen in Figure 3. The clear yellow color of grade 2 palm oil outer skin liquid smoke was liked by 92% of respondents; the public preferred the color of liquid smoke antiseptic, which was brighter and clearer than the color of dark liquid smoke 75% of respondents liked the aroma of liquid smoke from the outer skin of the palm but the remaining percentage of respondents did not, because the aroma was too strong, this is in line with research by Saepul et al. [19] that the pungent aroma of liquid smoke was disliked by 50% of the public because of its aroma stinging. Assessing the aspect of dryness, 75% of respondents did not feel that their skin was dry, and 92% of respondents did not feel any side effects such as heat and itching, according to Fitriani's research. [24] Most people like dryness and the side effects that arise after using liquid smoke antiseptic in the short term. And it is necessary to research the prolonged use of liquid smoke antiseptic. Factors of comfort and safety in using antiseptic products, such as in terms of color, aroma, and texture, and do not cause side effects on the skin must be prioritized[25]

The limitation of this study was not to carry out GC-MS analysis to measure the concentration of the compound components contained in the liquid smoke of the outer skin of the palm, so it is unknown what percentage of each compound component is present to support more complete research data. For future researchers, it is hoped that they will conduct further research regarding the effects of the long-term use of liquid smoke antiseptic for the outer skin of palm oil.

CONCLUSION

Based on the study's results, it can be concluded that liquid smoke of palm bark with an effective concentration of 75% grade 2 has antibacterial and antifungal properties and is effectively used as an antiseptic.

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AUTHORS CONTRIBUTION

ALS, ND, MFAI: Data collection, analysis and interpretation of results, preparation of manuscripts. FF, YM: Research concept and design, analysis and interpretation of results, manuscript finalization. AR: Research concept and design, analysis and interpretation of results, author correspondence, manuscript finalization. All authors agreed to the final drafting of the manuscript.

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CONFLICT OF INTEREST

The author declares no conflict of interest in this article.

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