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Efficacy of coffee peel extract as natural antimicrobial in coconut oil soap to against staphylococcus aureus

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Abstract. The aim of the study was to evaluate the efficacy of coffee (*Coffea canephora*) peel extract as natural antimicrobial in coconut oil soap to against *Staphylococcus aureus*. *S. aureus*, a pathogenic bacteria, was common to be found on human skin. By using Minimum Inhibitory Concentration (MIC) and inhibition zone, the antimicrobial activity of coffee peel extract were $16,7 \pm 7,22\%$ and $0,38 \pm 0,21$ mm, respectively. Phytochemical compounds of coffee were caffeic acid, catechin, coumaric acid, gallic acid, hesperetin, kaempferol, hesperidin, luteolin, quercetin. Antimicrobial activity of coffee peel extract showed to be potential as natural antimicrobial in coconut oil soap to inhibit *S. aureus*.

1. Introduction

Indonesia has various types of coffee which was processed on its seed and produced coffee peel as waste. This waste was wasted and disposed of from the source of human activity or a natural process that is no longer used and has no economic value. According to data from the Central Statistics Agency [1] in East Java, the production of coffee was 65,474 tonnes in 2017 and producing coffee waste to reach for 41%.

One of the efforts to reduce the accumulation of waste, namely by making the skins of coffee as antimicrobial compounds that can inhibit the growth of pathogenic bacteria. Coffee peel was extracted by using the maceration in order to extract bioactive compounds. Saleem and Saeed [2] reported that the extracted coffee peel has components as antimicrobial, antioxidant and anti-inflammatory.

Antimicrobials were compounds that can inhibit growth or kill bacteria that can harm human [3]. Bacteria that can harm humans are bacteria that can contaminate food and can cause disease after consuming contaminated food [4,5]. One of these pathogenic bacteria is *Staphylococcus aureus* which may cause infection to humans. *S. aureus* bacteria can cause poisoning if the amount exceeds 1×10^5 CFU / g in traditional sausages on the market [6]. This bacteria was natural flora in human skin. Antimicrobial inhibition in the growth of pathogenic bacteria requires an antimicrobial role [7]. Antimicrobial ability to fight and inhibit bacterial growth can be measured in vitro using Minimum Inhibitor Concentration and disc diffusion method to determine the antimicrobial activity. However, there was still limited study about coffee peel extract as natural antimicrobial in coconut soap to against



S. aureus. The aim of this present study was to evaluate the efficacy of coffee (*Coffea canephora*) peel extract as natural antimicrobial in coconut oil soap to against *Staphylococcus aureus*.

2. Material and methods

2.1. Coffee peel extraction

Coffee peel was obtained from PUSLIT Jember (8-11 months, Arabica variety) and was extracted by using maceration method. These was cut and weighed of 25 grams. These were diluted into 96% ethanol solution (1: 2) (v/v) and was covered with cap. The mixtures were incubated using shaker incubator for 14 hours at 100 rpm. The filtrate was obtained by filtering the solution and separated from solute. These was evaporated by using vacuum rotary evaporator at 70 °C for 5 hours.

2.2. The strains of microorganism

Pure culture of *S. aureus* (Nusaroma, Gajah Mada University, Yogyakarta) was cultured on Mannitol Salt Agar (Himedia, India) for 24 until 48 hours at 37°C. The colonies of *S. aureus* were appeared as yellow colonies. By using streaking method, a single colony was purified onto culture media namely tryptic soya agar (TSA) (Himedia, India).

2.3. Minimum Inhibitory Concentration (MIC)

MIC was measured as done by Budiati *et al.* [8,9]. Cultures were grown at 37 °C for 16 hours in Luria Broth (LB, Himedia, India) in shaking incubator. The cultures were diluted and compared the turbidity. This has to be set at 0.5 of McFarland scale. A serial concentration was prepared on Mueller Hinton Broth (Oxoid, United Kingdom). A total of 20 µL of the aliquot was artificially contaminated to the serial dilution. These were incubated at a temperature of 37 °C for 24 hours. MIC value (%) describes the lowest concentration of the bacteria might grow. These values are calculated as mean values of three measurements. The experiment was done in triplicate. A 15 µl aliquot of 10% DMSO was dropped onto a serial dilution as negative control. Penicillin G (10 unit/ml) was used as positive control.

2.4. Disc diffusion assay

By using disc diffusion assay, the antimicrobial activity was measured as the method of Bauer [8]. Cultures were grown at 37 °C for 16 hours in Luria Broth (LB, Himedia, India) in shaking incubator. The cultures were diluted and the turbidity was set at 0.5 of McFarland scale. By using a sterilized cotton swab, the cultures were swabbed onto Mueller Hinton Agar (Oxoid, United Kingdom) and were let about 30 min. Twenty microliter essential oil was dropped on sterilized disc. This disc was then placed onto the agar. This was incubated for 18–24 h at 37 °C. The inhibition zones was measured in four different directions after the incubation period. This was recorded as milimeter. The experiment was done in triplicate. Fifteen microliter aliquot of 10% DMSO was dropped onto a sterile paper disc as negative control. Penicillin G (10 unit/ml) was used as positive control.

2.5. Identification bioactive compounds

Bioactive compounds were identified by using LC-MS as methods of Bduhafsdun *et al.* [10].

2.6 Application to coconut soap

The application to coconut soap was done by weighing 36.25 g of NaOH which is put into 53 ml of water. By pouring of 100 ml coconut oil, 60 ml palm oil and 100 ml olive oil into the container, adding 17 % (v/v) of coffee skin extract and coloring to taste, putting the coconut oil, palm oil and olive oil mixture in the container for about 10 minutes and mixing the cool NaOH into the oil mixture in a container and stirring until it is mixed homogeneously and forming a thick solution. The molding was done by pouring the solution into the silicon mold. The coconut soap was storing in a dry place at room temperature until it forms a solid. The ageing was done for 3 weeks and checking the pH of the soap using pH paper until it reaches a neutral pH (pH 6-7). The examination is carried out every 1 week to

find out that the pH of the soap reaches a neutral pH. Soap that has reached a neutral pH, the saponification process has occurred completely and there is no free alkaline contained so that the soap is safe to use. The finished product will go through a paper packaging and labeling process.

3. Results and Discussion

Coffee is one of the plantation commodities which has a high economic value among other plantation crops. In Indonesia, there are 2 types of coffee, namely arabica coffee and robusta coffee [11]. The part of the coffee used is the seed, while the skin becomes waste which only becomes animal feed and fertilizer. Yet according to Esquivel and Jiménez [9], coffee bean husk waste also inherits the content contained in coffee beans, namely as a natural antioxidant ingredient. According to Simanjuntak *et al.* [13], coffee skin waste contains secondary metabolites such as polyphenol compounds and caffeine which are antioxidants that are able to capture free radicals, so they can damage bacterial cells.

By using disc diffusion assay (inhibition zone) and MIC, the antimicrobial activity of coffee peel to against *S. aureus* was studied. This present study found that antimicrobial activity of coffee peel extract showed to be potential as natural antimicrobial (table 1).

Table 1. Inhibition zone and Minimum Inhibitory Concentration (MIC) of *S. aureus* on coffee peel extract

<i>Inhibition zone</i>	MIC
0,38 ± 0,21 mm	16,7 ± 7,22%

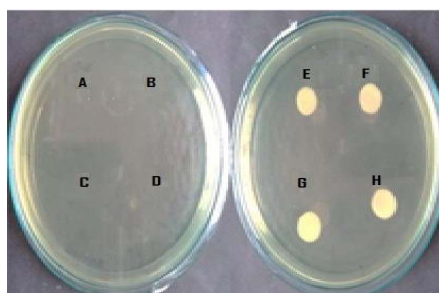
By using disc diffusion assay (inhibition zone) and MIC, the antimicrobial activity of coffee peel extracts to against *S. aureus* was studied. This present study found that coffee peel extract showed antimicrobial activity (table 1). This present study found that coffee peel extract composed by naringenin, quercetin, kaempferol, hesperetin, luteolin, gallic acid and coumaric acid as bioactive compound which may act as antimicrobial. According to Esquivel and Jiménez [12], the polyphenol compounds found in coffee bean husks are flavan-3-ol, hydroxynamic acid, flavonols, anthocyanidins, catechins, epicatechins, routine, tannins, ferulic acid.

Naringenin, eriodictyol and hesperetin compounds include flavonoid compounds from flavonone derivatives that are found in citrus fruits [14; 15; 16]. Naringenin has antibacterial activity that can increase membrane permeability and can change the morphology of *S. aureus* cells [17]. Eriodictyol compounds can provide antimicrobial effects on *S. aureus* bacteria [18], in a study of Mandalari *et al.* [19] said that eriodictyol compounds can inhibit bacteria through cell membranes. While hesperetin has a high level of antioxidant activity [20].

Quercetin compounds had a very strong antimicrobial mechanism by damaging the cell walls and membranes of pathogenic bacteria [21]. Coumaric acid inhibits the growth of pathogenic bacteria by disrupting the cell membrane of pathogenic bacteria effectively [22]. Moreover, luteolin showed good antibacterial activity by damaging cell membranes and hesperetin had high levels of antioxidant activity [20; 23]. The simultaneous reaction of these antimicrobial components can dissolve the phospholipid layer of the bacterial cell membrane so lysis may occur in bacterial cells which disturbs the stability of the cell membrane, this causes the cytoplasm to leak out of the cell and results in bacterial cell death [24]. Quercetin compounds have a mechanism. a very strong antimicrobial by damaging the cell walls and membranes of pathogenic bacteria [21]. Coumaric acid, effectively inhibits the growth of pathogenic bacteria by disrupting the cell membrane of pathogenic bacteria [22]. As well as luteolin compounds which show good antibacterial activity by damaging cell membranes and hesperetin which has a high level of antioxidant activity [20; 23]. The simultaneous reaction of these antimicrobial components can dissolve the phospholipid layer of the bacterial cell membrane resulting in lysis of bacterial cells which disrupts the stability of the cell membrane, this causes the cytoplasm to leak out of the cell and results in bacterial cell death [24].

Kaempferol, quercetin and myricetin compounds are flavonoid derivatives, namely flavonols [25]. Kaempferol compounds are present in all fruit waste extract samples, in the research of Górnjak *et al.* [26] that this compound can inhibit DNA gyrase from *E. Coli* bacteria. Quercetin has strong antibacterial properties because it can inhibit the growth of harmful bacteria and can damage cell walls and cell membranes [21]. Wang *et al.* [21] reported that *E. Coli* and *S. aureus* bacteria with certain doses of quercetin can damage the cell walls and cell membranes of the two types of bacteria. Meanwhile, myricetin can affect DNA by inhibiting DNA synthesis and can affect RNA polymerase [27].

Gallic acid and coumaric acid were phenolic acids that were a type of polyphenol which can be found in various fruits and vegetables [28; 29]. Sarjit *et al.* [30] revealed that gallic acid is not effective as an antimicrobial because there are several strains that are not sensitive to gallic acid so it is recommended to combine gallic acid with compounds that may be effective, the antimicrobial effect of gallic acid. in bacterial cells caused by pH. Meanwhile, coumaric acid according to study of Lou *et al.* [22] effectively inhibits the growth of *S. aureus* due to the capability of coumaric acid to increase the permeability of the plasma membrane. It can cause the barrier function to be lost and coumaric acid can disrupt the bacterial cell membrane and may bind the bacterial genomic DNA. Thus, it may inhibit cellular function which ultimately leads to cell death. The application of coffee peel extract in coconut soap was resulting to inhibit *S. aureus* in human skin (fig. 1). Coffee peel extract as natural antimicrobial in coconut oil soap was showed in fig 2.



Note : A = 100%; B = 50%; C = 25%; D = 12,5%; E = 6,25%; F = 3,125%; G = 1,56%; H = 0.78%

Figure 1. MIC coffee peel extract on *S. aureus*



Figure 2. Coffee Peel Extract as natural antimicrobial in coconut oil soap

4. Conclusion

Arabica coffee peel extract showed antimicrobial activity and it can be applied in coconut oil soap to against the growth of *S. aureus*.

5. Acknowledgment

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