AntibacterialTestofVariousIngredientsof“Indonesian Jamu”

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Abstract. Jamu is an Indonesian traditional beverage, made from various Indonesian spices, and believed to possess antibacterial properties. The aim of this research was to determine whether typical Indonesian Jamu ingredients had antibacterial compounds. The study was performed to determine the activity of the filtrate of various ingredients in inhibiting *Staphylococcus aureus* ATCC 25922 and *Escherichia coli* ATCC 25923 growth. Ingredients used in Jamu were *Alpinia galanga*, *Zingiber officinale*, *Curcuma domestica*, *Tamarindus indica*, *Kaempferia galanga*, *Curcuma xanthorrhiza*, *Syzygium aromaticum*, *Syzygium polyanthum*, *Myristica fragrans*, *Cinnamomum burmannii, Cymbopogon citratus*, and *Boesenbergia pandurata*. The concentration used for the respective filtrates was 10% (w/v). Filtrates were made according to common Jamu production, i.e. by blending the ingredients together with distilled water. Sensitivity tests were done by agar diffusion method. Qualitative analysis based on diameter of inhibition zone was conducted to determine the antibacterial activity of all Jamu ingredients. Results showed that the Jamu ingredient with the best antibacterial content for inhibiting *E. coli* was *Kaempferia galanga* (9 mm), while for inhibiting *S. aureus* it was *Tamarindus indica* (8 mm). The results indicate that Jamu ingredients have antibacterial compounds as expected by most Indonesian people.

Keywords: Jamu, antibacterial, *Escherichia coli*, jamu, *Staphylococcus aureus.*

INTRODUCTION

The abundance of biodiversity in Indonesia is one natural resource that can be developed, especially in the health and pharmaceutical sectors. Indonesia has a high number of medicinal plants, and most of them have been widely used and mixed as medicines by Indonesia people since ancient times. Jamu is a term for a traditional herbal medicine from Indonesia. Currently, Indonesian Jamu is recognised by the government and has been developed on an industrial scale.[1,2,3] The medicinal plants that are known Jamu ingredients are *Curcuma domestica*, *Curcuma xanthorrhiza*, *Kaempferia galanga*, *Syzygium aromaticum*, and *Zingiber officinale*.[4]

Cinnamomum burmannii (bark and leaves) and *Tagetes erecta* (leaves) from Indonesia have been reported to be antimicrobial agents against *Bacillus subtilis*, *Salmonella typhimurium*, and *Candida albicans*.[5,6] Another research study also reported that *Cinnamomum burmannii* essential oil can inhibit the growth of *Staphylococcus aureus* and *Candida albicans*.[7] *Cymbopogon citratus, Cymbopogon nardus,* and *Cymbopogon schoenanthus* have antifungal activities that can control superficial mycosis.[8] *Syzygium aromaticum* from the *Myrtaceae* family has known potential antifungal, antiviral, and antibacterial agents based on previous research.[9,10] It also has antimutagenic, antioxidant, anti-inflammatory, and antiparasitic compounds.[11] *Zingiber officinale* belongs to the *Zingiberaceae*

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family and has antibacterial activity to inhibit the growth of *Staphylococcus aureus* and *Streptococcus pyogenes*.[12, 13] Ginger has an essential oil that is known as gingerol.[14]

Based on this, it can be determined that the ingredients of Jamu have great potential and should be developed. There is lot of research that refers to the extracts of Jamu ingredients. However, there is little information about the potential of Jamu ingredients based on their water filtrates. Thus, the objective of this study was to screen the antibacterial activity of some natural medicinal plant/herb filtrates. The antibacterial activity test used the distilled water filtrates of Java galangal, fingeroot, cinnamon, turmeric, Java ginger, lemon grass, galangal, nutmeg, clove, bay leaf, tamarind, and ginger against a Gram-positive standard strain of bacteria represented by *Staphylococcus aureus* ATCC 25922 and a Gram-negative standard represented by *Escherichia coli* ATCC 25923.

EXPERIMENTAL DETAIL

Plant Filtrates Preparation

The plant materials used in the study consisted of leaves (*Cymbopogan citratus* and *Syzygium polyanthum*), fruits (*Myristica fragrans*, *Tamarindus indica*, and *Syzygium aromaticum*), stems (*Cinnamomum burmannii*), and roots (*Alpinia galangal*, *Boesenbergia pandurata*, *Curcuma domestica*, *Kaempferia galangal*, *Curcuma xanthorrhiza*, and *Zingiber officinale*). These were collected from the traditional market in Surabaya, Indonesia. The fresh plant materials were finely ground using a blender. One gram of each plant material was weighed in an Erlenmeyer flask, to which 100 mL of distilled water was added for pre-filtrate. The Erlenmeyer flask was placed in dark for three days at room temperature. The mixture was filtered using Whatman No. 1 filter paper. The filtrates were kept at 4°C until used in the experiment.

Microorganisms

Two reference bacteria, *Escherichia coli* ATCC 25923 and *Staphylococcus aureus* ATCC 25922, were used during the study. The tested strains were obtained from the Biology Department of Airlangga University, cultured in nutrient broth at 37°C and stored in nutrient agar slants at 4°C.

Antibacterial Assay

The antimicrobial activity of Jamu ingredient filtrates were tested using a disk diffusion assay, modified from Bauer *et al.*[15] Bacterial strains were grown on nutrient agar at 37°C for 24 h then suspended in buffer saline (0.85% NaCl) and adjusted to a turbidity of a 0.5 McFarland standard (10^8 CFU/mL). The bacterial suspension (1000 µL) was inoculated in Petri dishes. The dissolution of the plant filtrates was facilitated with the addition of 1% (v/v) distilled water, which affected the growth of microorganisms (as shown by our control experiments). Briefly, the nutrient agar medium (25 mL) was poured into Petri dishes under aseptic conditions in a laminar air flow hood. The Petri dishes were kept in the laminar air flow chamber for solidification of the media. The Petri dishes were then kept in laminar air flow for drying. Once dried, sterile 6 mm filter paper discs (Difco) were placed in the Petri dishes and 25 µL of the test solution was loaded on each disc. The diameters of inhibition zones were measured in mm after incubation at 37°C for 24 h. All experiments were conducted in triplicates for each treatment against each bacteria. Distilled water was used as a negative control and chloramphenicol was used as a control standard.

RESULTS AND DISCUSSION

The current study was designed to investigate the antibacterial activity of some *Jamu* ingredient filtrates: *Alpinia galanga, Boesenbergia pandurata, Cinnamomum burmannii, Curcuma domestica, Curcuma xanthorrhiza, Cymbopogan citratus, Kaempferia galanga, Myristica fragrans, Syzygium aromaticum, Syzygium polyanthum, Tamarindus indica,* and *Zingiber officinale* against *Escherichia coli* ATCC 25923 and *Staphylococcus aureus* ATCC 25922. The antibacterial activity of the tested plant filtrates was qualitatively assessed by the presence or absence of inhibition zones. The results are represented in Table 1.

According to the results given in Table 1, the distilled water filtrates of *Curcuma domestica*, *Curcuma xanthorrhiza*, *Kaempferia galanga*, and *Tamarindus indica* showed antibacterial activity against *Escherichia coli*

ATCC 25923 and *Staphylococcus aureus* ATCC 25922. Their effect was comparable to the antibiotic used as control. In addition, *E. coli* ATCC 25923 was strongly influenced, with a mean inhibition zone of 9 mm, by *Kaempferia galanga* filtrate. This result is very interesting because *E. coli* is the most common cause of opportunistic infections. Meanwhile, *Staphylococcus aureus* ATCC 25922 was strongly influenced, with a mean inhibition zone of 9 mm, by *Cymbopogan citratus*. *S. aureus* is a pathogenic bacteria that is resistant to antibiotics because it has the ability to survive in the air for days.[16]

TABLE 1. Antibacterial activity of Jamu ingredients

Name of sample	Inhibition zone diameter (mm)	
	Antibacterial activity	
	***Escherichia coli* ATCC 25923**	***Staphylococcus aureus* ATCC 25922**
Alpinia galangal	-	7± 0.14
Boesenbergia pandurata	-	-
Cinnamomum burmanni	-	7,5± 0.07
Curcuma domestica	8± 0.14	7± 0.42
Curcuma xanthorriza	7.8± 0.42	7± 0.35
Cymbopogan citratus	8± 0.35	9± 0.21
Kaempferia galangal	9± 0.14	7.5± 0.21
Myristica fragrans	-	-
Syzygium aromaticum	-	-
Syzygium polyanthum	-	-
Tamarindus indica	7± 0.28	8± 0.49
Zingiber officinale	-	-
*Chloramphenicol	15± 0.88	10± 0.21

*Positive control of bacteria

The distilled water filtrate of *Alpinia galanga* and *Cinnamomum burmannii* showed antibacterial activity against *Staphylococcus aureus* ATCC 25922. *S. aureus* and *E. coli* can be inhibited by ginger ethanolic extract.[17,13] Results showed that the *Cinnamomum burmannii* filtrate effect was stronger than *Alpinia galanga*. The essential oil of *Cinnamomum burmannii* causes membrane disruption in the bacteria.[18] This study demonstrated that the Gram-negative bacteria were more resistant to the plant filtrate than Gram-positive bacteria, as *Escherichia coli* ATCC 25923 exhibited more resistant than *Staphylococcus aureus* ATCC 25922 when they were tested with *Cinnamomum burmannii* filtrate. These essential oils have different sensitivities between Gram-negative and Gram-positive bacteria.[19] The membrane of Gram-negative bacteria is composed of peptidoglycan and the outer layer has a hydrophilic surface as a strong permeability barrier.[20] These results supported that essential oil constituents have antibacterial activity dependent on their hydrophobicity.[21]

Results showed that *Boesenbergia pandurata, Myristica fragrans, Syzygium polyanthum,* and *Zingiber officinale* distilled water filtrates had noticeable activity against *Escherichia coli* ATCC 25923 and *Staphylococcus aureus* ATCC 25922. Meanwhile, the antibacterial activity of *B. pandurata* ethanol extract and essential oils has been shown to inhibit *Listeria monocytogenes* and *Salmonella typhimurium*.[22] The essential oils of *B. pandurata* also obstructed growth of *Escherichia coli, Staphylococcus aureus, Bacillus cereus,* and *L. monocytogenes*. *B. pandurata* essential oil have the ability to change the permeability and to alter salt tolerance of the bacterial cell.[23] Many factors affect antibacterial activity from plant filtrates, which vary in different regions of the world, such as the effect of climate, soil composition, age and vegetation cycle stage, the quality, quantity and composition of filtrate product, and different type of bacterial strains.[24,25] Moreover, the process of filtering depends on the type of solvent.[26] These results indicated that Jamu ingredients have antibacterial compounds as expected by most Indonesian people. The compounds from Jamu ingredients are soluble in water. Therefore, it is important to do further studies to detect the specific active compounds.

SUMMARY

Various Jamu ingredients as distilled water filtrates showed promising antibacterial activity, especially *Curcuma domestica, Curcuma xanthorrhiza, Cymbopogan citrates, Kaempferia galangal,* and *Tamarindus indica*.

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