



Antimicrobial Activity of Edible Plant Extracts Against Skin Infection Pathogens

Sirilak Sanpa & Sirikarn Sanpa*

School of Medical Sciences, University of Phayao, Phayao, 56000 Thailand

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Abstract

Antimicrobial activity of four edible plant extracts including ginger, galangal, lemongrass and tree basil have been investigated. For this purpose, the extract of plants was acquired using ethanol and distilled water. The inhibitory effect on six skin infection pathogenic microorganisms, i.e., *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Pseudomonas aeruginosa*, *Propionibacterium acnes* and *Candida albicans* were performed by disc diffusion, agar well diffusion and microdilution methods. From the results, ethanolic extract of galangal showed the best antimicrobial activities against all test pathogens in terms of the minimum inhibitory concentration and minimum bactericidal concentration, ranging from 0.49-15.62 mg/ml and 0.49-62.5 mg/ml, respectively. Therefore, the aqueous extract has lower antimicrobial activity compared with the ethanolic extract. The ethanolic extracts of ginger, galangal and lemongrass could inhibit *P. acnes* at the concentrations lower than 1 mg/ml. The outcome of this study suggested that edible plant extracts could possibly be applied as a natural antimicrobial agent and combined with other materials for further applications.

Introduction

Skin infection pathogens are important problems in both hospital and community. Skin and skin-structure infections aetiology is dominated by *Staphylococcus aureus* and other common bacteria such as *Pseudomonas aeruginosa* (Lipsky et al., 2007; Livermore et al., 2015). *Propionibacterium acnes* is the major bacteria causing acne, which is one of the most common and chronic skin problems (Vora et al., 2018). This group of microorganisms is often mentioned in health and beauty products. Synthetic drugs or antibiotics induced mutations in the genetic composition of these microorganisms, leading

them to be resistant to drugs or antibiotics (Cohen, 1992). In recent years, an increasing number of human pathogenic microorganisms poses a concern on antibiotic resistance strains. For this reason, bioactive compounds isolated from plants may offer a new solution to this problem by being a novel antimicrobial agent. The search for new antibacterial agents should be conducted and studied among various plant families. Due to their therapeutic properties, plants have been widely used in many pharmaceutical industries. Medicinal plants have been known to produce compounds with therapeutic properties such as antidiabetic, antioxidant, antibacterial, anti-inflammatory, antipyretic and gastroprotective

effects (Gupta et al., 2016).

There was a report on seven Cameroonian dietary plants that display their inhibitory effect on the multidrug-resistant Gram-negative bacteria (Djeussi et al., 2013). Various Thai edible plants should be studied on their biological activities. Ginger (*Zingiber officinale* Roscoe) is an edible plant that has been widely used all over the world. It belongs to the Family Zingiberaceae. It possesses antimicrobial activity and can be used to treat bacterial infection (Tan & Vanitha, 2004). Chemical composition of ginger shows that it contains over 400 different compounds. The major constituents in ginger rhizomes are carbohydrates, lipids, terpenes and phenolic compounds (Grzanna et al., 2005). Ginger contains terpene components including zingiberene, β -bisabolene, α -farnesene, β -sesquiphellandrene α -curcumene, phenolic compounds including gingerol, paradols, and shogaol. The antimicrobial activity of methanol and n-hexane extract of ginger may be due to the gingerol and shogaol as active ingredients (Hasan et al., 2012). Galangal (*Alpinia galanga* (L.) Willd.) belongs to the Family Zingiberaceae and has also been used as a traditional medicine for treatment of stomachache, a carminative and diarrhea (Oonmetta-aree et al., 2006). The essential oil from *A. galanga* consists of cineole, 4-allylphenylacetate, α -farnesene, (2, 6-dimethylphenyl) borate and α -pinene (Hamad et al., 2016). Lemongrass (*Cymbopogon citratus* (DC.) Stapf) belongs to the Family Gramineae and is commonly used in folk medicine for treatment of nervous and gastrointestinal disturbances, and as an antispasmodic, analgesic, anti-inflammatory, anti-pyretic, diuretic and sedative (Santin et al., 2009). The major constituents of *C. citratus* essential oils are geranial, neral and myrcene (Bassolé et al., 2011). Tree basil (*Ocimum gratissimum* L.) is an herbaceous plant that belongs to the Labiatae family. Chemical analysis of the essential oil demonstrated the presence of Eugenol and other compounds such as 1, 8-cineole and β -selinene (do Nascimento Silva et al., 2016). Since a large number of different chemical compounds is presented in this extract, their combined mechanism of actions can affect multiple target sites against the bacterial cells. Therefore, due to their availability and affordable production cost, Thai edible plants with medicinal properties should be further investigated for antimicrobial applications.

In Thailand, studies of the biological activity of plants have been widely reported (Komutiban, 2014; Sritubtim et al., 2014; Junsathian et al., 2018). Some Thai

medicinal plants were already screened and observed for their antibiofilm activity (Teapaisan et al., 2017) and anti-protozoa effect (Leesombun et al., 2017). However, the role of Thai edible plants that can provide useful medicinal properties especially the antimicrobial effect is not quite clear and still needs to be elucidated. The purpose of the present study was to evaluate the antimicrobial activity of the extracts from some edible plants including ginger (*Zingiber officinale* Roscoe), galangal (*Alpinia galanga* (L.) Willd.), lemongrass (*Cymbopogon citratus* (DC.) Stapf), tree basil (*Ocimum gratissimum* L.) against some skin infection pathogens.

Materials and methods

1. Preparation of crude extract

Four plants were purchased from local market in Phayao province, Thailand, from July to December 2017 including ginger (*Zingiber officinale* Roscoe), galangal (*Alpinia galanga* (L.) Willd.), lemongrass (*Cymbopogon citratus* (DC.) Stapf), tree basil (*Ocimum gratissimum* L.). Information on the plant material is shown in Table 1. Fresh plants were washed and dried at 40°C. After dried completely, the material was powdered for further extraction. Twenty grams of powdered plant material was extracted with 200 ml of 95% ethanol and distilled water at room temperature for 24 h maceration without shaking; this extraction process was repeated 3 times. The extract was then filtrated through Whatman No.4 filter paper and concentrated using a rotary evaporator. The extract yield was determined on a weight basis and the yield percentage was calculated as follows:

$$\% \text{ extraction yield} = (\text{mass of extract} / \text{mass of dry matter}) \times 100$$

Table 1 Edible plant materials and their extract yield percentage

| Scientific name | Common name | Part of sample | % extraction yield | |
|--|-------------|----------------|--------------------|-----------------|
| | | | Aqueous extract | Ethanol extract |
| <i>Zingiber officinale</i> Roscoe | Ginger | Rhizome | 17.16 | 10.66 |
| <i>Alpinia galanga</i> (L.) Willd. | Galangal | Rhizome | 18.06 | 14.80 |
| <i>Cymbopogon citratus</i> (DC.) Stapf | Lemongrass | Stem | 17.40 | 11.14 |
| <i>Ocimum gratissimum</i> L. | Tree basil | Leaf | 13.04 | 8.38 |

The crude extract was stored in a freezer until further use. The samples were re-dissolved using 95% ethanol or distilled water depending on their solvent. Three concentrations of the plant extract (100, 200 and 300 mg/ml) were used to test antimicrobial activity by disc diffusion and agar well diffusion methods.

2. Bacterial strains and growth conditions

Escherichia coli TISTR 117, *Staphylococcus aureus* TISTR 746, *Staphylococcus epidermidis* TISTR 518, *Pseudomonas aeruginosa* TISTR 1287 and *Candida albicans* TISTR 5554 were purchased from the culture collection of the Thailand Institute of Scientific and Technological Research (TISTR). *Propionibacterium acnes* was provided by Division of Microbiology and Parasitology, School of Medical Sciences, University of Phayao, Thailand. Bacteria (except *P. acnes*) and fungus were cultured on Mueller Hinton broth (MHB) for 24 h at 37°C and yeast extract-malt extract broth (YMB) for 48 h at 30°C, respectively. *P. acnes* was cultured on Brain Heart Infusion broth (BHI broth) for 48 h at 37°C under anaerobic condition. Before each experiment, cultures of the test microorganisms were suspended in MHB and YMB and the optical density of the suspension was adjusted to obtain the viable cell count of 10^6 CFU/ml.

3. Antimicrobial activity

3.1 Disc diffusion method

Disc diffusion method was carried out according to the standard method by Bauer et al. (1966). The microbial inocula were spread using the sterile cotton swab on Mueller Hinton agar (MHA) for bacteria and yeast extract-malt extract agar (YMA) for fungus. Sterile filter paper discs (6 mm) added with 20 µl of the extracts were placed on the cultured agar before incubation at 37°C for 24 h. *P. acnes* and *C. albicans* were incubated for 48 h at 37°C and 30°C, respectively. The experiments were performed in triplicate. Diameters of the inhibition zones were measured in millimeter. Antimicrobial activity of the controls (ethanol, tetracycline and Amphotericin B) against all the tested isolates were also determined.

3.2 Agar well diffusion method

Similar to the procedure used in the disc diffusion method, the fresh inocula were spread using a sterile cotton swab on MHA and YMA. A hole with a diameter of 6 mm is punched with a sterile cork borer on the medium. Fifty microliters of the extract were added into each hole. The bacteria were grown for 24-48 h at 37 °C whereas the fungus was grown for 48 h at 30 °C.

Inhibition zones were measured in millimeter.

3.3 Minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC)

The minimal inhibitory concentrations (MICs) of the plant extracts were determined according to Clinical and Laboratory Standard Institute (CLSI) (2016) guidelines. Plant extracts were prepared at different concentrations, ranging from 0.49 to 500 mg/ml in a 96-well microtiter plate. The microbial cells of 10^6 CFU/ml were inoculated in the microtiter plate followed by incubation at 37 °C for 24 h. After incubation, 10 µl of resazurin was added and incubated further for 2 h to evaluate the growth inhibition. Resazurin is a blue dye that can be reduced to pink color by viable cells. The lowest concentration causing the color change was considered as the MIC. The minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC) values were tested by subculturing the microorganisms from the MIC well onto the agar. The MBC and MFC values were defined as the lowest concentration of plant extract at which no bacteria and fungus growth was recorded.

4. Statistical analysis

Data are expressed as mean \pm standard deviation (SD). The data are subjected to analysis of variance (ANOVA) and Duncan's New Multiple range Test (DMRT). The difference level of $p < 0.05$ is considered significant.

Results and discussion

1. Plant extraction yield

Both of ethanolic and aqueous extracts of four edible plants including ginger, galangal, lemongrass and tree basil were examined for the antimicrobial activity against the skin infection pathogens. The extraction yields of plant materials with ethanol and water ranged from 8.38 to 14.80 % and 13.04 to 18.06 %, respectively. The highest yield of the plant extracts was obtained from galangal while tree basil gave the lowest extraction yield.

2. Antimicrobial activity of plant extract

The antimicrobial activities of edible plants at different concentrations against six microorganisms including three Gram-positive bacteria, two Gram-negative bacteria and one fungus were shown using the disc diffusion method (Table 2) and the agar well diffusion method (Table 3). Both methods present different ability of

Table 2 Antimicrobial activity of edible plant extracts using disc diffusion method

| Crude extract | | Concentration (mg/ml) | Diameter of inhibition zone (mm) (mean±SD) | | | | | |
|---------------------------|---------|--------------------------|--|-------------------------|-------------------------|-------------------------|-------------------------|------------------------|
| | | | <i>E. coli</i> | <i>S. aureus</i> | <i>S. epidermidis</i> | <i>P. aeruginosa</i> | <i>P. acnes</i> | <i>C. albicans</i> |
| Ginger | Aqueous | 100 | 6.00±0.00 ^c | 7.00±0.00 ^c | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ⁱ | 6.00±0.00 ^d |
| | | 200 | 10.33±0.58 ^b | 8.33±0.58 ^d | 6.00±0.00 ^d | 6.00±0.00 ^{ce} | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 10.67±0.58 ^b | 8.67±0.58 ^d | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | Ethanol | 100 | 7.33±0.58 ^d | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 9.00±0.00 ^g | 6.00±0.00 ^d |
| | | 200 | 9.33±1.15 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 8.33±0.58 ^c | 11.00±0.00 ^c | 6.00±0.00 ^d |
| | | 300 | 10.67±0.58 ^b | 6.00±0.00 ^f | 6.00±0.00 ^d | 8.00±0.00 ^c | 12.33±0.58 ^c | 6.00±0.00 ^d |
| Galangal | Aqueous | 100 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 7.00±0.00 ^d | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 10.00±0.00 ^b | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | Ethanol | 100 | 6.00±0.00 ^c | 19.33±0.58 ^c | 21.67±0.58 ^c | 6.00±0.00 ^c | 10.33±0.58 ^f | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 21.00±1.00 ^b | 24.33±1.15 ^b | 6.00±0.00 ^c | 11.33±0.58 ^d | 8.33±0.58 ^c |
| | | 300 | 6.00±0.00 ^c | 21.33±0.58 ^b | 24.33±0.58 ^b | 6.00±0.00 ^c | 15.00±0.00 ^b | 9.33±0.58 ^b |
| Lemongrass | Aqueous | 100 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | Ethanol | 100 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 7.00±0.00 ^b | 6.00±0.00 ^d |
| Tree basil | Aqueous | 100 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | Ethanol | 100 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 200 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| | | 300 | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d |
| Ethanol | | 6.00±0.00 ^c | 6.00±0.00 ^f | 6.00±0.00 ^d | 6.00±0.00 ^c | 6.00±0.00 ^j | 6.00±0.00 ^d | |
| Tetracycline (30 µg/ml) | | 27.33±1.53 ^a | 26.67±0.58 ^a | 29.33±1.15 ^a | 23.33±1.53 ^a | 32.00±0.00 ^a | ND | |
| Amphotericin B (10 µg/ml) | | ND | ND | ND | ND | ND | 21.00±2.00 ^a | |

Remark: Diameter of inhibition zone including disc diameter of 6 mm.

The different letters within the same column indicate statistically significant difference at 0.05 probability level.

ND; not determined.

diffusions. The disc diffusion method shows the capacity of antimicrobial agent to adsorb and diffuse through paper discs into the agar medium and inhibits the growth of the microbial strain whereas the agar well diffusion method shows the capacity of diffusion to agar medium.

As shown in Table 2 and 3, the ethanolic extract of galangal showed the most effectiveness of antibacterial activity against all microorganisms tested by both methods. The inhibition zone of the extracts ranged from 7 to 24 mm for the disc diffusion method and 7 to 29 mm for the agar well diffusion method. The results indicated that the inhibition zone variation was correlated with the concentration of the crude extracts.

At the concentration of 100 mg/ml, the microbial growth was inhibited by the ethanolic extracts of four plants using agar well diffusion method. While, at the same concentration using disc diffusion method, only the ethanolic extracts of ginger and galangal could inhibit the microbial growth. The aqueous extracts of lemongrass and tree basil extract could not inhibit all tested microorganisms using disc diffusion method. From

the results, the antibacterial activity depended on the concentration of the extract and the extraction solvent. Generally, high antibacterial activity was observed in high concentration of the extract. Accordingly, an increase in the extract concentration produced a relative increase in the diameter of inhibition zone. This effect could be from the major and minor chemical components of the extract, including the interactions between them. Our results agree with the study of Er et al. (2018) who demonstrated that inhibition zone diameter significantly indicated an increase parallel to the application dose.

From these results, the edible plant extract had more potential of antibacterial activity against Gram-positive than Gram-negative bacteria. In accordance with previous findings, higher antimicrobial activity against Gram-positive bacteria than Gram-negative bacteria may be due to the fact that the cell wall of Gram-negative bacteria is multilayered structure and composed of the outer membrane. Accordingly, antibacterial agents could not easily penetrate Gram-negative cells and inhibit their growth (Parekh & Chanda, 2011; Meeprathom et al., 2018).

Table 3 Antimicrobial activity of edible plant extracts using agar well diffusion method

| Crude extract | | Concentration (mg/ml) | Antimicrobial activity | | | | | |
|---------------------------|---------|-------------------------|-------------------------|-------------------------|--------------------------|--------------------------|-------------------------|-------------------------|
| | | | <i>E. coli</i> | <i>S. aureus</i> | <i>S. epidermidis</i> | <i>P. aeruginosa</i> | <i>P. acnes</i> | <i>C. albicans</i> |
| Ginger | Aqueous | 100 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 200 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 300 | 6.00±0.00 ^h | 6.00±0.00 ^h | 7.00±0.00 ^{kl} | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | Ethanol | 100 | 12.67±0.58 ^d | 10.67±0.58 ^c | 11.33±1.53 ^{fg} | 9.33±0.58 ^{de} | 13.67±0.58 ^c | 8.00±0.00 ^f |
| | | 200 | 13.67±0.58 ^c | 11.67±1.15 ^c | 12.33±0.58 ^{ef} | 9.67±0.58 ^{cd} | 14.33±0.58 ^c | 8.00±0.00 ^f |
| | | 300 | 14.33±0.58 ^b | 13.67±1.53 ^d | 13.00±0.00 ^{de} | 10.33±0.58 ^{bc} | 15.33±0.58 ^d | 8.00±0.00 ^f |
| Galangal | Aqueous | 100 | 6.00±0.00 ^h | 6.00±0.00 ^h | 10.67±0.58 ^{gh} | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 200 | 6.00±0.00 ^h | 6.00±0.00 ^h | 11.67±0.58 ^{fg} | 6.00±0.00 ^h | 7.00±0.00 ^j | 6.00±0.00 ^h |
| | | 300 | 6.00±0.00 ^h | 6.00±0.00 ^h | 13.67±1.15 ^d | 6.00±0.00 ^h | 10.00±0.00 ^g | 6.00±0.00 ^h |
| | Ethanol | 100 | 9.67±0.58 ^f | 20.67±1.53 ^c | 26.33±1.15 ^c | 9.33±0.58 ^{de} | 14.33±0.58 ^c | 20.00±0.00 ^c |
| | | 200 | 12.00±0.00 ^c | 23.33±1.15 ^b | 28.33±0.58 ^b | 9.67±0.58 ^{cd} | 21.67±1.15 ^c | 20.33±0.58 ^c |
| | | 300 | 13.33±0.58 ^c | 24.33±1.15 ^b | 29.00±1.00 ^b | 11.00±0.00 ^b | 23.33±1.53 ^b | 22.00±1.00 ^b |
| Lemongrass | Aqueous | 100 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 200 | 6.00±0.00 ^h | 6.00±0.00 ^h | 7.67±0.58 ^k | 9.33±0.58 ^{de} | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 300 | 6.00±0.00 ^h | 6.00±0.00 ^h | 8.33±0.58 ^{kl} | 10.67±0.58 ^b | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | Ethanol | 100 | 8.67±0.58 ^s | 8.67±0.58 ^{fg} | 8.00±1.00 ^{jk} | 8.00±0.00 ^f | 9.00±0.00 ^h | 9.67±0.58 ^c |
| | | 200 | 9.33±0.58 ^f | 9.33±0.58 ^f | 9.33±0.58 ^{ij} | 8.33±0.58 ^f | 9.67±0.58 ^{gh} | 9.67±0.58 ^c |
| | | 300 | 9.67±0.58 ^f | 9.33±1.53 ^f | 9.67±0.58 ^{hi} | 8.66±0.58 ^f | 11.00±0.00 ^f | 11.00±0.00 ^d |
| Tree basil | Aqueous | 100 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 7.00±0.00 ^g |
| | | 200 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 7.00±0.00 ^g |
| | | 300 | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 7.00±0.00 ^g |
| | Ethanol | 100 | 8.00±0.00 ^s | 7.67±0.58 ^s | 7.67±1.53 ^k | 7.00±0.00 ^g | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 200 | 8.00±0.00 ^s | 7.67±0.58 ^s | 7.67±0.58 ^k | 7.00±0.00 ^g | 6.00±0.00 ^j | 6.00±0.00 ^h |
| | | 300 | 8.00±0.00 ^s | 8.00±0.00 ^s | 8.00±0.00 ^{jk} | 7.00±0.00 ^g | 6.00±0.00 ^j | 6.00±0.00 ^h |
| Ethanol | | 6.00±0.00 ^h | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | 6.00±0.00 ^j | 6.00±0.00 ^h | |
| Tetracycline (30 µg/ml) | | 35.00±1.00 ^a | 35.33±0.58 ^a | 31.67±1.53 ^a | 32.33±1.53 ^a | 34.00±0.00 ^a | ND | |
| Amphotericin B (10 µg/ml) | | ND | ND | ND | ND | ND | 23.00±2.00 ^a | |

Remark: Diameter of inhibition zone including well diameter of 6 mm.

The different letters within the same column indicate statistically significant difference at 0.05 probability level.

ND; not determined

3. Minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC) and minimum fungicidal concentration (MFC)

MIC is the lowest concentration of the extract that can inhibit the microbial growth after incubation. The MIC of four edible plants against skin infection pathogens were determined using the microdilution method. From this study, the observed MIC, MBC and MFC of the extracts ranged from 0.49 to 500 mg/ml. The MIC value of galangal for the ethanolic extract was 0.49 to 15.63 mg/ml and the MIC on *S. aureus* was 0.98 mg/ml. This finding is consistent with the studies from Oonmetta-aree et al. (2006) and Mayachiew & Devahastin (2008), which indicates that galangal ethanolic extract has the strongest antibacterial activity. The ethanolic extract of ginger shows the highest antibacterial activity on *P. acnes* with the value of 0.49 mg/ml.

According to the results from this study, the aqueous extract has lower antimicrobial activity compared with the ethanolic extract. It is possible that the ethanolic extract consists of more active compounds than the aqueous extract due to higher potency of the antimicrobial activity. The previously study showed that the main

compound of galangal ethanolic extract was D,L-10-acetoxychavicol acetate, which has high antibacterial effect against *S. aureus* (Oonmetta-aree et al., 2006). Moreover, the minor compounds of crude extract were identified by GC-MS, which are defined as p-coumaril diacetate, palmitic acid, acetoxyeugenol acetate, 9-octadecenoic acid, eugenol, b-bisabolene, b-farnesene and sesquiphellandrene. This ethanolic extract demonstrated both outer and inner membrane damages and disruption of the cytoplasmic membrane function (Oonmetta-aree et al., 2006).

The extracts were calculated for possibility to have a bactericidal or bacteriostatic effect. The extracts show bactericidal activity when the ratio of MBC/MIC ≤ 4 and bacteriostatic activity when the ratio is of MBC/MIC > 4 (Djeussi et al. 2013; Noumedem et al., 2013). Interestingly, bactericidal effect was obtained with the ethanolic and aqueous extracts from ginger, galangal, lemongrass and tree basil against all tested bacteria. This confirms that the studied extracts from edible plants exhibits bactericidal effect.

Generally, the inhibitory activity of essential oils was greater than that of ethanolic extracts. It has been shown

Table 4 MIC, MBC and MFC of edible plant extracts

| Crude extract | | Concentration of extract (mg/ml) | | | | | | | | | | | |
|---------------|---------|----------------------------------|------|------------------|------|-----------------------|-------|----------------------|------|-----------------|------|--------------------|-------|
| | | <i>E. coli</i> | | <i>S. aureus</i> | | <i>S. epidermidis</i> | | <i>P. aeruginosa</i> | | <i>P. acnes</i> | | <i>C. albicans</i> | |
| | | MIC | MBC | MIC | MBC | MIC | MBC | MIC | MBC | MIC | MBC | MIC | MFC |
| Ginger | Aqueous | 62.5 | 62.5 | 62.5 | 125 | 7.81 | 15.63 | 62.5 | 125 | 250 | 500 | 250 | 500 |
| | Ethanol | 0.49 | 0.49 | 0.98 | 1.95 | 0.49 | 0.49 | 31.25 | 125 | 0.49 | 0.49 | 15.63 | 31.25 |
| Galangal | Aqueous | 0.98 | 3.91 | 31.25 | 62.5 | 31.25 | 125 | 62.5 | 250 | 250 | 250 | 500 | 500 |
| | Ethanol | 0.98 | 0.98 | 0.98 | 1.95 | 0.98 | 0.98 | 15.63 | 62.5 | 0.98 | 0.98 | 0.49 | 0.49 |
| Lemongrass | Aqueous | 62.5 | 125 | 15.63 | 62.5 | 15.63 | 125 | 125 | 125 | 125 | 250 | 500 | 500 |
| | Ethanol | 1.95 | 3.91 | 31.25 | 125 | 7.81 | 7.81 | 31.25 | 125 | 0.98 | 0.98 | 15.63 | 62.5 |
| Tree basil | Aqueous | 250 | 500 | 250 | 250 | 250 | 250 | 250 | 250 | 125 | 500 | 62.5 | 500 |
| | Ethanol | 31.25 | 62.5 | 125 | 125 | 125 | 125 | 62.5 | 250 | 125 | 250 | 31.25 | 500 |

that the essential oil of galangal showed antimicrobial activities against bacteria, fungi, yeast and parasite (Farnsworth & Bunyapraphatsara, 1992). However, this result also supports that the ethanolic extract inhibited growth of bacteria and yeast. Moreover, the process to obtain ethanol extract by maceration extraction is simple and requiring less equipment. These support that aqueous and ethanolic extracts from plants are probably good alternative antimicrobial agents.

Conclusion

This study shows that the antimicrobial activity against all tested pathogens is highest for the ethanolic extract of galangal, followed by ginger and lemongrass, respectively. Furthermore, the extracts of ginger, galangal, lemongrass and tree basil exhibits bactericidal effects on all tested bacteria. The findings of this study suggest that edible plant extracts could possibly be used as a natural antimicrobial agent for further applications.

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