Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Biomedicine

journal homepage: www.elsevier.com/locate/apjtb

Original article http://dx.doi.org/10.1016/j.apjtb.2017.09.009

# Chemical profiling and antimicrobial activity of essential oil from *Curcuma aeruginosa* Roxb., *Curcuma glans* K. Larsen & J. Mood and *Curcuma* cf. *xanthorrhiza* Roxb. collected in Thailand

Nararat Akarchariya, Sasithorn Sirilun, Jakaphun Julsrigival, Sunee Chansakaowa*

*Department of Pharmaceutical Sciences, Faculty of Pharmacy, Chiang Mai University, Chiang Mai 50200, Thailand*

ARTICLE INFO

*Article history:*
Received 30 Jun 2017
Received in revised form 3 Aug 2017
Accepted 8 Sep 2017
Available online 20 Sep 2017

*Keywords:*
*Curcuma aeruginosa* Roxb.
*Curcuma glans* K. Larsen & J. Mood
*Curcuma* cf. *xanthorrhiza* Roxb.
Essential oil
Antimicrobial activity

ABSTRACT

**Objective:** To investigate chemical constituents and new antimicrobial agents among essential oils from the rhizomes of *Curcuma aeruginosa* (*C. aeruginosa*) Roxb., *Curcuma glans* K. Larsen & J. Mood and *Curcuma* cf. *xanthorrhiza* Roxb.

**Methods:** The essential oils were obtained by hydro-distillation and analyzed by gas chromatography/mass spectroscopy. Agar-well diffusion assay was used to study the antimicrobial activity and also broth-micro dilution techniques were examined for minimum inhibitory concentration (MIC) against four bacterial strains and yeast.

**Results:** The gas chromatography/mass spectroscopy analysis showed monoterpenes predominantly (88.53%) in the rhizome oil of *Curcuma* cf. *xanthorrhiza*. Sesquiterpenes (50.10%) was the most abundant component in the essential oil of *C. glans*, while monoterpenes (45.55%) and sesquiterpenes (45.81%) were found in *C. aeruginosa* with a significant amount. The major components of *C. aeruginosa* were characterized as camphor (29.39%) and germacrone (21.21%). Germacrone (15.76%), $\beta$-pinene (9.97%) and camphor (9.96%) were found as major compounds in the rhizome oils of *C. glans* while $\alpha$-terpinolene (24.86%) and *p*-cymen-*7-ol* (12.17%) were found as major compositions in *Curcuma* cf. *xanthorrhiza*. The essential oils were tested against four bacterial strains and yeast. As a result, the rhizome oil of *C. aeruginosa* exhibited potent activity against *Staphylococcus aureus* [inhibition zone (21.94 ± 0.24) mm, MIC 125 μg/mL], *Bacillus cereus* [inhibition zone (20.83 ± 0.36) mm, MIC 125 μg/mL], and *Candida albicans* [inhibition zone (11.60 ± 0.30) mm, MIC 250 μg/mL].

**Conclusions:** The essential oils from three *Curcuma* species possessed greater activity against the gram-positive bacteria (*Staphylococcus aureus* and *Bacillus cereus*) than gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*). The results suggest that the essential oils from the fresh rhizome of *Curcuma* spp. might be a potential source of natural antimicrobial substances.

## 1. Introduction

The members in Zingiberaceae have been recognized in widely uses of food, medicine, and traditional knowledge [1]. *Curcuma* is a perennial rhizomatous herb, which is one of the largest genera in the Zingiberaceae family, with about eighty species distributed mostly in Southeast Asia, Papua New Guinea and Northern Australia [1,2]. Among them, 43 species are normally found in Thailand [1–3]. Most species in *Curcuma* are usually aromatic in at least one part. In Thailand, various kinds of *Curcuma* are well known for their medicinal values or as ornamental plants, especially Turmeric [*Curcuma longa* (*C. longa*) L.] and Siam Tulip (*Curcuma alismatifolia* Gagnep.) [1–6]. Previous reports of phytochemical studies and bioactivities of *Curcuma* species presented several monoterpenoids in essential oils, diarylheptanoids of which curcuminoids are the significant bioactive compounds in some species and phenolics in extracts. The utilization of the

*Corresponding author: Sunee Chansakaow, Department of Pharmaceutical Sciences, Faculty of Pharmacy, Chiang Mai University, Chiang Mai 50200, Thailand.
Tel: +66 5394 4399
Fax: +66 5394 4390
E-mails: chsunee@gmail.com, sunee.c@cmu.ac.th (S. Chansakaowa).

Peer review under responsibility of Hainan Medical University. The journal implements double-blind peer review practiced by specially invited international editorial board members.

Zingiberaceous plants can be attributed to biological activities which are antimicrobial, antioxidant, antityrosinase activities, and anti-inflammatory effect of some species which were reported in the previously published data [7–9]. The antimicrobial properties of essential oils and plant extracts have been recognized for many years. The members of the family Zingiberaceae were also reported as natural antimicrobial agents, especially their essential oils [10–14]. From folk wisdom, many of *Curcuma* species usually known as a medicine were used to alleviate antidiarrhea, infectious wound or abscess [14]. Three species, *Curcuma aeruginosa* (*C. aeruginosa*) Roxb., *Curcuma glans* (*C. glans*) K. Larsen & J. Mood and *Curcuma* cf. *xanthorrhiza* (*C.* cf. *xanthorrhiza*) Roxb., were found in Thailand especially in the North with the history of their utilization which followed the indigenous wisdom. *C. aeruginosa* is known as Kamindam in Thailand [3]. The ethnomedical uses of this plant included a treatment of flatulence, dyspepsia, diarrhea and parasitic infection [5,6]. Traditionally, the rhizome of *C. glans* has been used medicinally to treat a sore throat, tonsillitis, wound or abscess in mouth, throat, and nose and herpes simplex virus [5]. *C.* cf. *xanthorrhiza*, Wan-Salika-Linthong in Thai, is known as a kind of sacred plant. In the northern part of Thailand have been used the fresh rhizome or dried powder for skin disease [6]. The practical uses of some species especially *C. glans* are based on the knowledge and experiences of the ancestor without sufficient scientific support. The present study is focused on the study of chemical constituents and to investigate the antimicrobial activities of three species of *Curcuma* compared with the well-known species *C. longa*.

## 2. Material and methods

### 2.1. Plant materials

Rhizomes of *C. aeruginosa* (voucher number WP.5811), *C. glans* (WP.5810) and *C.* cf. *xanthorrhiza* (WP.5818) were collected from an herbal market at Chiang Dao district, Chiang Mai Province, Thailand. The plants were identified by taxonomists and voucher specimens have been deposited at the Queen Sirikit Botanic Garden Herbarium and Faculty of Science, Chiang Mai University, Thailand.

### 2.2. Extraction of essential oil

The fresh rhizome of each sample (3 000 g) was cleaned with tap water, reduced their size and then subjected to hydrodistillation for 5 h using a cleavenger-type apparatus. The essential oils were dried over anhydrous sodium sulfate and stored in air-tight containers at 4 °C before analyzed and tested.

### 2.3. Gas chromatography/mass spectroscopy analysis

A Shimadzu GCMS-QP 2010 Plus system was used, with a mass-selective detector with electron impact ionization. The samples were separated using a DB-5 MS capillary column (5% phenylmethylpolysiloxane, 30 m × 0.25 mm, 0.25 μm film thickness) with helium as the carrier gas (0.99 mL/min). The temperature program used for analysis was as follows: the initial temperature was 70 °C and programmed to 150 °C with the rate of 5 °C/min held for 5 min, and then ramped to 230 °C with the rate of 3 °C/min and kept constant for 3 min. The split flow ratio was 1:100. The injection temperature was 150 °C and the gas chromatography/mass spectroscopy interface temperature was set at 250 °C.

### 2.4. Identification of components

The identification of volatile components was based on computer matching with WILLEY 7 and NIST 2005 Library, as well as by comparison of the mass spectra and Kovats retention indices with a series of *n*-alkanes ($C_8$–$C_{20}$).

### 2.5. Antimicrobial assay

#### 2.5.1. Microbial culture

The following bacterial species were used: gram-positive bacteria [*Staphylococcus aureus* (*S. aureus*) (ATCC 25923) and *Bacillus cereus* (*B. cereus*) (ATCC 11778)], gram-negative bacteria [*Escherichia coli* (*E. coli*) (ATCC 25922) and *Pseudomonas aeruginosa* (*P. aeruginosa*) (ATCC 27853)] and yeast [*Candida albicans* (*C. albicans*) (ATCC 90028)].

The antimicrobial assays were carried out by the agar disc-diffusion method and the microdilution method modified by Lorian in 1991, Sirilun in 2005 and Sirilun *et al*. [15–17]. These assays were used to determine the antimicrobial activity of the essential oils against human pathogenic bacteria and yeast.

#### 2.5.2. Disc diffusion method

Essential oil samples were aseptically prepared by steriled membrane filtration (Pall Gelman, 0.45 and 0.22 μm millipore). Bacteria and yeast were cultured in tryptic soy broth and sabouraud dextrose broth, respectively. The density of microbial test cultures was standardized using McFarland 0.5 turbidity standard at $1.0 \times 10^8$ CFU/mL of bacteria and $1.0 \times 10^6$ CFU/mL of yeast. The culture tests were swabbed and spread on the culture plate of Mueller-Hinton agar and Sabouraud dextrose agar for bacteria and yeast, respectively and allowed to dry for 15 min. The discs were loaded with 10.0 μL essential oils and placed on the medium surface. The positive controls were gentamicin (10 μg/disc) and cyclopiroxolamine (10 μg/disc) against bacterial and yeast growth, respectively. The negative control was dimethyl sulfoxide (DMSO) (10 μL/disc). After (24–48) h of incubation at (35–37) °C, the diameters of the growth inhibition zones were measured for each test in triplicate.

#### 2.5.3. Minimum inhibitory concentration (MIC) determination

MIC was analyzed using broth microdilution assay in Mueller-Hinton Broth and Roswell Park Memorial Institute 1640 (RPMI-1640) for bacteria and yeast test, respectively, described by Das *et al*. [18] with modification. The stock solutions of essential oil were prepared in DMSO (1.00 mg/mL), filtrated by using the sterile membrane (0.22 μm) and subsequent two-fold serial dilution as final concentration at (0.49–1 000.00) μg/mL in reaction mixture well. The microbial strains were grown and the density of cultures was standardized using McFarland 0.5 turbidity standard at $1.0 \times 10^8$ CFU/mL of bacteria and $1.0 \times 10^6$ CFU/mL of yeast. The positive control was gentamicin with stock concentration at 10 mg/mL and cyclopiroxolamine with stock concentration at 10 mg/mL to against bacterial and yeast growth, respectively. The negative

control was DMSO. After (24–48) h of incubation at (35–37) °C, the turbidity of the microbial growth was determined for each test in triplicate.

### 2.6. Statistical analysis

All experiments were conducted repeatedly in triplicate ($n$ = 3). Analysis of one-way-ANOVA were performed ($P < 0.05$) for each microbial species and overall activity of each sample (agar disc-diffusion assay). The differences among means were determined by Duncan's test ($P < 0.05$).

## 3. Results

The percentage oil yields of *C. aeruginosa*, *C. glans* and *C.* cf. *xanthorrhiza* were 0.35%, 0.32% and 0.29% v/w, respectively. The GC–MS analysis of the identified constituents is revealed in Table 1.

The rhizome oils showed monoterpenes and sesquiterpenes as major components with a different of their chemical profiles. The major components of *C. aeruginosa* were characterized as camphor (29.39%) and germacrone (21.21%). The most abundant compositions in the rhizome oils of *C. glans* were germacrone (15.76%), followed by $\beta$-pinene (9.97%) and camphor (9.96%).

*C. aeruginosa*, *C. glans*, *C.* cf. *xanthorrhiza*, were a broad inhibitory spectrum, against the growth of bacteria both of gram-negative and gram-positive and yeast. The activity of the essential oil was determined by comparison with positive control, gentamicin (bacterial tests) and cyclopiroxolamine (yeast test), and DMSO as a negative control. The activity of DMSO with low concentration was not detected against the growth of microbial tests (data was not shown). The MIC evidenced that the rhizome oil of *C. aeruginosa*, *C. glans*, and *C.* cf. *xanthorhiza* exhibited promising antibacterial activity against *S. aureus*, *B. cereus*, *E. coli* and *P. aeruginosa* with MIC values of (125–1 000) μg/mL and antifungal activity against *C. albicans* with MIC values of (250–500) μg/mL. The rhizome oil of *C. aeruginosa*, *C. glans*, *C.* cf. *xanthorhiza* and *C. longa* showed weak activity against the gram-negative bacteria *E. coli* (MIC 1 000 μg/mL). However, the *C. aeruginosa* showed strong activity in comparison to positive control against the growth of *S. aureus*, *B. cereus*, *P. aeruginosa* and *C. albicans*. It is interesting that the rhizome oil of *C. aeruginosa*, *C. glans*, and *C.* cf. *xanthorrhiza* showed activity against the growth of *S. aureus* and *C. albicans*. *Candida* species are the leading causes of nosocomial bloodstream infection and has become a major health problem as an opportunistic infection such as oral candidiasis (Fareid, 2014) [19]. The results indicate that the rhizome oils are potential oils with an ability to control opportunistic fungal *C. albicans* (Table 2). These results can be ascribed to them containing oxygenated monoterpenes, *1,8*-cineole (2.68%–4.87%) and camphor (2.49%–29.39%) in a great proportion. Some oxygenated monoterpenes especially *1,8*-cineole were previously observed to be the potential compounds for high antibacterial properties [20]. Additionally, germacrone was found as an important constituent in *C. aeruginosa* and *C. glans*. Germacrone possessed potent bacterial activity

**Table 1**
Chemical composition (%) of the essential oils from *C. aeruginosa* (CA), *C. glans* (CG) and *C.* cf. *xanthorrhiza* (CX).

| Retention time | KI[a] | KI[b] | Identified compound | CA | CG | CX | Chemical group |
|---|---|---|---|---|---|---|---|
| 3.458 | 905 | 904 | *2*-Heptyl alcohol | 0.15 | 5.04 | – | Alcohol |
| 3.960 | 940 | 940 | *α*-Pinene | 0.18 | 1.52 | 2.46 | Monoterpenes |
| 4.240 | 958 | 955 | Camphene | 1.17 | 1.38 | 3.19 | Monoterpenes |
| 4.702 | 985 | 983 | *β*-Pinene | 0.35 | 9.97 | 6.78 | Monoterpenes |
| 4.816 | 992 | 992 | *β*-Myrcene | – | – | 1.50 | Monoterpenes |
| 5.069 | 1007 | 1009 | *2*-Octanol | – | 0.39 | 0.39 | Alcohol |
| 5.190 | 1015 | 1013 | *a*-Phellandrene | – | – | 0.78 | Monoterpenes |
| 5.549 | 1037 | 1034 | *p*-Cymene | – | – | 8.09 | Monoterpenes |
| 5.627 | 1042 | 1040 | Limonene | 0.35 | 0.50 | 1.90 | Monoterpenes |
| 5.683 | 1046 | 1034 | *β*-Phellandrene | – | – | 0.32 | Monoterpenes |
| 5.736 | 1049 | 1048 | *1,8*-Cineole | 2.68 | 4.33 | 4.87 | Monoterpenes |
| 6.218 | 1076 | 1069 | γ-Terpinene | – | – | 0.40 | Monoterpenes |
| 6.819 | 1042 | 1079 | *α*-Terpinolene | – | – | 24.86 | Monoterpenes |
| 7.156 | 1118 | 1108 | *2*-Nonanol | 0.43 | 6.89 | – | Alcohol |
| 8.320 | 1139 | 1141 | Camphor | 29.39 | 9.96 | 2.49 | Monoterpenes |
| 8.692 | 1161 | 1159 | Borneol | 7.27 | 3.06 | 0.48 | Monoterpenes |
| 8.897 | 1173 | 1172 | *endo*-Borneol | 2.86 | 2.16 | 0.69 | Monoterpenes |
| 8.989 | 1179 | 1179 | Isopinocamphone | – | 0.12 | 0.44 | Monoterpenes |
| 9.068 | 1183 | 1182 | Terpinen-*4-ol* | 0.37 | 0.22 | 0.59 | Monoterpenes |
| 9.237 | 1192 | 1189 | *p*-Cymen-*7*-ol | – | – | 12.17 | Monoterpenes |
| 13.985 | 1392 | 1392 | *β*-Elemene | 1.35 | 2.32 | – | Sesquiterpenes |
| 14.704 | 1424 | 1424 | *trans*-Caryophyllene | 0.11 | 0.23 | 0.32 | Sesquiterpenes |
| 15.536 | 1461 | 1460 | *a*-Humulene | 0.58 | – | 0.60 | Sesquiterpenes |
| 16.429 | 1498 | 1480 | Curzerene | 4.84 | 1.00 | – | Sesquiterpenes |
| 16.718 | 1511 | 1504 | Germacrene A | 0.76 | 0.49 | – | Sesquiterpenes |
| 17.933 | 1564 | 1561 | Germacrene B | 5.20 | 0.43 | – | Sesquiterpenes |
| 19.770 | 1600 | – | Isocurcumenol | – | 2.43 | – | Sesquiterpenes |
| 20.031 | 1610 | 1617 | (+) Spathulenol | – | 0.92 | – | Sesquiterpenes |
| 21.530 | 1662 | 1666 | Isospathulenol | – | 0.18 | – | Sesquiterpenes |
| 22.473 | 1693 | 1693 | Germacrone | 21.21 | 15.76 | 0.74 | Sesquiterpenes |

a: Kovat retention index investigated relative to C8–C20 *n*-alkanes on DB-5 MS column; b: Kovat retention index from literature data (NIST Chemical Web Book).

**Table 2**
Antimicrobial screening with inhibition zone diameters (mm) of the rhizome oils of *C. aeruginosa* (CA), *C. glans* (CG) and *C.* cf. *xanthorrhiza* (CX), in comparison with G: Gentamicin (10 mg/mL), C: Cyclopiroxolamine (10 mg/mL) and *C. longa* (CL).

| Microorganism | CA | CG | CX | G | C | CL |
|---|---|---|---|---|---|---|
| *Staphylococcus aureus* | 21.94 ± 0.24a | 17.24 ± 0.07a | 11.53 ± 0.27a | 7.28 ± 0.09 | NT | 7.21 ± 0.12a |
| *Bacillus cereus* | 20.83 ± 0.36a | 7.28 ± 0.10 | 9.64 ± 0.45 | 9.58 ± 0.08 | NT | 4.87 ± 0.19a |
| *Escherichia coli* | ND | 3.35 ± 0.28 | 7.53 ± 0.20 | 7.21 ± 0.11 | NT | ND |
| *Pseudomonas aeruginosa* | 5.71 ± 0.13 | 3.56 ± 0.12 | 7.13 ± 0.39 | 7.60 ± 0.08 | NT | 5.29 ± 0.46 |
| *Candida albicans* | 11.60 ± 0.30a | 7.27 ± 0.17 | 7.29 ± 0.17a | NT | 10.54 ± 0.22 | ND |

a: means of individual trials within a column are significantly different ($P < 0.05$); ND = Not detected; NT = Not tested.

against gram-negative bacteria, *P. aeruginosa* (MIC 15.6 mg/mL; MBC 31.2 mg/mL) [21].

## 4. Discussion

The monoterpenes, α-pinene, β-pinene, sesquiterpenes and germacrone, are most often found in the rhizome oil of Zingiberaceous plants collected from Ban Thum, Chiang Dao district, Chiang Mai Province [6]. There are several reports in the chemical composition of the essential oil from the rhizome of *Curcuma* species [6–8,14,20,22], but none for *C. glans*. Chemical analysis of the essential oils from rhizomes of the aforementioned *Curcuma* species collected in Thailand and their antimicrobial activities compared to the worldwide-cultivated species, *C. longa*, were mentioned in this present study. Monoterpene and sesquiterpene hydrocarbons represented the most common chemical groups found in the essential oils from the rhizomes of the investigated *Curcuma* species [6]: *C. aeruginosa* (45.55%, 45.81%, respectively); *C. glans* (34.79%, 50.10%); and *C.* cf. *xanthorrhiza* (88.53%, 2.72%). Germacrone and camphor were found in the greatest amount in *C. aeruginosa* and *C. glans* while *C.* cf. *xanthorrhiza* found α-terpinolene and *p*-cymen-7-ol as major components. It has been reported that terpenes have antibacterial activity because of their bacteriostatic and bactericidal effects [23]. Many aromatic herbs (*i.e.* lemongrass, basil, and cinnamon, *etc.*) [23–25] including species of *Curcuma* [13,20–22,26] are comply with this basis.

This study is the first report of chemical compositions in the rhizome oil of *C. glans*. α-terpinolene (24.86%) and *p*-cymen-7-ol (12.17%) were found as major compositions in *C.* cf. *xanthorrhiza*. Previously reported by Helen *et al.* [27], xanthorrhizol was found in the rhizome oil of *Curcuma xanthorrhiza* (64.38%) as major compounds, followed by camphene (8.27%), curcumin (5.85%), and α-pinene (1.93%). Curcumene (41.40%) and xanthorrhizol (21.50%) were the major compositions in the rhizome oil collected in Netherlands [28]. Jarikasem *et al.* [20] reported that *1,8*-cineol (37.58%) and curzerenone (13.70%) were the highest proportions found in the rhizome oil collected in Thailand. The chemical profile of this present study revealed more similarity with Jarikasem *et al.* [20] than the samples collected in India and Netherlands [23,27]. Although the samples were collected in Thailand in the previous report [20], the quantity of chemical compositions showed differences with this present study might be affected by geography, harvesting period or variety. Three major compounds, germacrone (23.49%), curzerenone (11.78%) and *1, 8*-cineol (10.92%), were previously reported by Theanphong *et al.* [22]. Ethoxybenzene, santolinatriene, benzofuran, 6-ethenyl-4,5,6,7-tetrahydro-3,6-dimethyl-5-isopropenyl-, trans- were reported as major components by George [28] in the rhizome oil of *C. aeruginosa* from India. Sirat *et al.* [28] reported curzerenone and 1, 8-cineol as the principal compositions but has a low amount of germacrone. The present study showed differences in the quantity of chemical components found in the rhizome oil from those previous reports [28–30] as geography, climate, harvesting period may affect the chemical profile.

The antimicrobial activity could be ascribed to them containing monoterpenes and sesquiterpenes as major components, especially oxygenated monoterpenes, camphor, *1,8*-cineole sesquiterpenes and germacrone, which were previously observed to be the potential compounds for high antibacterial properties [21,25].

The essential oils from *Curcuma* species possessed greater activity against the gram-positive bacteria (*S. aureus* and *B. cereus*) than gram-negative bacteria (*E. coli* and *P. aeruginosa*). Interestingly, the rhizome oil of *C. aeruginosa*, *C. glans*, and *C.* cf. *xanthorrhiza* showed activity against the growth of *C. albicans* with an ability to control opportunistic fungal *C. albicans*. The results indicated that the essential oils of *Curcuma* species are able to have a potential for further application for the treatment of infectious diseases and as alternative natural products to substitute synthetic antimicrobial agents.

## Conflict of interest statement

We declare that we have no conflict of interest.

## Acknowledgments

This work was supported by the grants from the Biodiversity-Based Economy Development Office Public Organization -National Research Council of Thailand (Grant number. R000012298), Department of Pharmaceutical Sciences, the Faculty of Pharmacy and the Graduate School, Chiang Mai University, Chiang Mai, Thailand. Queen Sirikit Botanic Garden, Chiang Mai, Thailand for providing plants samples and plant identification.

## References

[1] Larsen K, Larsen SS. *Gingers of Thailand*. Chiang Mai: Queen Sirikit Botanic Garden; 2006.
[2] Sirirugsa P, Larsen K, Maknoi C. The Genus *Curcuma* L. (Zingiberaceae): distribution and classification with reference to species diversity in Thailand. *Gard Bul Singap* 2007; **59**(1&2): 203-20.
[3] Smittinan T. *Thai plant name, forest herbarium*. Bangkok: Forest Herbarium Royal Forest Department; 2014.
[4] Samarakoon K, Senevirathne M, Lee WW, Kim YT, Kim JI, Oh MC, et al. Antibacterial effect of citrus press-cakes dried by

high speed and far-infrared radiation. *Nutr Res Pract* 2012; **6**(3): 187-94.

[5] Inta A, Maknoi J, Pong-a-monkoon W. *Survey of Zingiberaceous plants in the northern part of Thailand in the research of traditional herbal products for Thai identity spa*. 2016, p. 61-122 [Unpublished data].

[6] Sookchot T. *Chemotaxonomy of medicinal and auspicious plants in Zingiberaceae sold at Ban Thum, Chiang Dao District, Chiang Mai Province.* [Master's thesis]. Department of Biology, Faculty of Science, Chiang Mai University; 2005.

[7] Angle GR, Vimala B, Bala N. Antioxidant and antimicrobial activity of essential oil from nine starchy *Curcuma* species. *Int J Curr Pharm Res* 2012; **4**(2): 45-7.

[8] Reanmongkol W, Subhadhirasakul S, Khaisombat N, Fuengnawakit P, Jantasila S, Khamjun A. Investigation the antinociceptive, antipyretic and anti-inflammatory activities of *Curcuma aeruginosa* Roxb. extracts in experimental animals. *Songklanakarin J Sci Technol* 2006; **28**(5): 999-1008.

[9] Chan EWC, Lim YY, Wong LF, Lianto FS, Wong SK, Lim KK, et al. Antioxidant and tyrosinase inhibition properties of leaves and rhizomes of ginger species. *Food Chem* 2008; **109**: 477-83.

[10] Chen IN, Chang CC, Ng CC, Wang CY, Shyu YT, Chang TL. Antioxidant and antimicrobial activity of Zingiberaceae plants in Taiwan. *Plant Foods Hum Nutr* 2008; **63**: 15-20.

[11] Duamkhanmanee R. The Efficiency test of the essential oil and extract of Greater Galangal, *Languas galanga* (L.) Stuntz on some plant pathogenic fungi. *Agr Sci J* 2008; **39**(3): 456-9.

[12] Oonmetta-aree J, Suzuki T, Gasaluck P, Eumkeb G. Antimicrobial properties and action of galangal (*Alpinia galanga* Linn.) on *Staphylococcus aureus*. *LWT-Food Sci Technol* 2006; **39**(10): 1214-20.

[13] Auta KI, Galadima AA, Bassey JU, Olowoniyi OD, Moses OO, Yako AB. Antimicrobial properties of the ethanol extracts of *Zingiber officinale* (Ginger) on *Escherichia coli* and *Pseudomonas aeruginosa*. *Res J Biol Sci* 2011; **6**(1): 37-9.

[14] Akarchariya N. *Chemical constituents and antimicrobial activity of some Zingiberaceous plants* [Master's thesis]. Department of Pharmaceutical Science, Faculty of Pharmacy, Chiang Mai University; 2017.

[15] Lorian EV. *Antibiotic in laboratory medicine*. 3rd ed. Baltimore: William & Wilkins; 1991.

[16] Sirilun S. *Fermentation kinetics of Morinda citrifolia Linn and antimicrobial activity of its products.* [Master's thesis]. Department of Pharmaceutical Science, Faculty of Pharmacy, Chiang Mai University; 2005.

[17] Sirilun S, Chaiyasut C, Kantachote D, Luxananil P. Characterisation of non human origin probiotic *Lactobacillus plantarum* with cholesterol-lowering property. *Afr J Microbiol Res* 2010; **4**(10): 994-1000.

[18] Das K, Tiwari RKS, Shrivastava DK. Techniques for evaluation of medicinal plant products as antimicrobial agent: current methods and future trends. *J Med Plant Res* 2010; **4**(2): 104-11.

[19] Fareid MA. *In vitro*: evaluation of inhibitory activity of some plant extracts against oral Candidiasis. *N Y Sci J* 2014; **7**(12): 66-76.

[20] Jarikasem S, Thubthimthed S, Chawananoraseth K, Suntorntanasat T. Essential oils from three *Curcuma* Species collected in Thailand. In: Başer, Franz G, Cañigueral S, Demirci F, Craker LE, Gardner ZE, editors. *Perspectives in natural product chemistry*. Chiang Mai: Wocamp III; 2005, p. 37-41.

[21] Diastutia H, Syahb YM, Juliawatyb LD, Singgih M. Antibacterial activity of germacrone sesquiterpene from *Curcuma xanthorrhiza* rhizomes. *Alchemy J Penelit Kim* 2016; **12**(2): 103-11.

[22] Theanphong O, Mingvanish W, Kirdmanee C. Chemical constituents and biological activities of essential oil from *Curcuma aeruginosa* Roxb. rhizome. *BHST* 2015; **13**(1): 6-16.

[23] Uribe S, Ramirez J, Pena A. Effects of beta-pinene on yeast membrane functions. *J Bacteriol* 1985; **161**(3): 1195-200.

[24] Silveira SM, Júnior AC, Scheuermann GN, Secchi FL, Vieira CRW. Chemical composition and antimicrobial activity of essential oils from selected herbs cultivated in the South of Brazil against food spoilage and foodborne pathogens. *Ciência Rural* 2012; **42**(7): 1300-6.

[25] Soković M, Glamočlija J, Marin PD, Brkić D, Griensven LJLDV. Antibacterial effects of the essential oils of commonly consumed medicinal herbs using an *in vitro* model. *Molecules* 2010; **15**: 7532-46.

[26] Revathi S, Malathy NS. Antibacterial activity of rhizome of *Curcuma aromatic* and partial purification of active compounds. *Indian J Pharm Sci* 2013; **75**(6): 732-5.

[27] Mary Helen PA, Susheela Gomathy K, Jayasree S, Nizzy AM, Rajagopal B, Jeeva S. Phytochemical characterization and antimicrobial activity of *Curcuma xanthorrhiza* Roxb. *Asian Pac J Trop Biomed* 2012; **2**(2): 637-40.

[28] Sirat MH, Jamil S, Hussain J. Essential oil of *Curcuma aeruginosa* Roxb. from Malaysia. *J Essent Oil Res* 1998; **10**: 453-8.

[29] Zwaving JH, Bos R. Analysis of the essential oils of five *Curcuma* species. *Flavour Fra J* 1992; **17**(1): 19-22.

[30] George M, Britto JS. Phytochemical and antioxidant studies on the essential oil of the rhizome of *Curcuma aeruginosa* Roxb rhizome. *Int Res J Pharm* 2015; **6**(8): 573-9.