

Chemical Components of Essential Oils From the Leaves of Seven Species Belonging to Rutaceae Family from Binh Chau-Phuoc Buu Nature Reserve, Vietnam

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Summary

Several plant species of the Rutaceae family are medicinal plants, oil bearing and food crops. To provide more information for utilization of some species of this family in Binh Chau-Phuoc Buu Nature Reserve, we extracted essential oils from the leaves of seven species of the Rutaceae family: *Acronychia pedunculata* (L.) Miq., *Atalantia citroides* Pierre ex Guillaumin, *Clausena excavata* Burm.f., *Glycosmis pentaphylla* (Retz.) DC., *Luvunga scandens* (Roxb.) Buch.-Ham. ex Wight & Arn, *Melicope pteleifolia* (Champ. ex Benth.) T.G. Hartley, and *Micromelum* sp., via hydrodistillation, and identified their components using GC/MS analysis. A total of 60 compounds were identified from essential oils of seven species. The main components of the essential oils isolated from five species, including *A. pedunculata*, *C. excavata*, *M. pteleifolia*, *G. pentaphylla*, and *Micromelum* sp., were caryophyllene (57.63% and 55.41% in *A. pedunculata* and *C. excavata*, respectively), 1,9-decadiene (32.59%, *M. pteleifolia*), β -ocimene (23.10%, *G. pentaphylla*), and 3-carene (58.03%, *Micromelum* sp.). Additionally, this study revealed the chemical composition of essential oils of *L. scandens* and *A. citroides* for the first time. The main constituent of *A. citroides* was 7-oxabicyclo[4.1.0] heptane, 3-oxiranyl- (53.91%) and that of *L. scandens* was caryophyllene (34.66%). These findings provide the basis for further application of these species in medicine.

Key words

Rutaceae, essential oils, gas chromatography-mass spectrometry, Binh Chau-Phuoc Buu Nature Reserve

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INTRODUCTION

The Rutaceae family has about 155 genera with over 1600 species distributed in tropical and subtropical regions (Zhang et al., 2009). Several species of this family are medicinal plants, food producing plants, spice plants and essential oil crops (Doreen et al., 2011; Aldona et al., 2016). Some species belonging to the Rutaceae family have biologically-active compounds such as saponins, steroids, cardiac glycosides, alkaloids, and flavonoids (Sekar et al., 2011). In Vietnam, over 100 species of the Rutaceae family have been recorded from the northern to the southern regions (Pham, 2000; Ban, 2005). Furthermore, the chemical constituents of some plants of this family have been identified. However, most of those studies have only analyzed components of individual species collected in Northern or North Central regions of Vietnam (Leclercq et al., 1994; Tran et al., 2007; Pham et al., 2007; Lesueur et al., 2008; Tran et al., 2015).

Binh Chau-Phuoc Buu Nature Reserve is the only remaining sandy forest along the coastline of Vietnam and home to several species of the Dipterocarpaceae family, which is located in Xuyen Moc District, Ba Ria-Vung Tau Province, Vietnam. In the report of the Management Board of Binh Chau-Phuoc Buu Nature Reserve, 796 plant species belonging to 142 genera have been recorded in the area (Van et al., 2019). To date, there are many studies conducted in this area to evaluate the diversity of plant species or record the new species for the flora of Vietnam (Bui et al., 2019; Ton et al., 2019). However, the comparison of chemical constituents of plant species of specific genus has not been studied yet. Previously, we recorded and collected 7 species of the Rutaceae family in this area, including *Acronychia pedunculata* (L.) Miq., *Atalantia citroides* Pierre ex Guillaumin, *Clausena excavata* Burm.f., *Glycosmis pentaphylla* (Retz.) DC., *Luvunga scandens* (Roxb.) Buch.-Ham. ex Wight & Arn, *Melicope pteleifolia* (Champ. ex Benth.) T.G. Hartley and *Micromelum hirsutum* Oliv. In this study, we analyzed and compared the chemical constituents of the essential oils isolated from the leaves of these species to provide more information for utilization of the natural resource from Binh Chau-Phuoc Buu Nature Reserve.

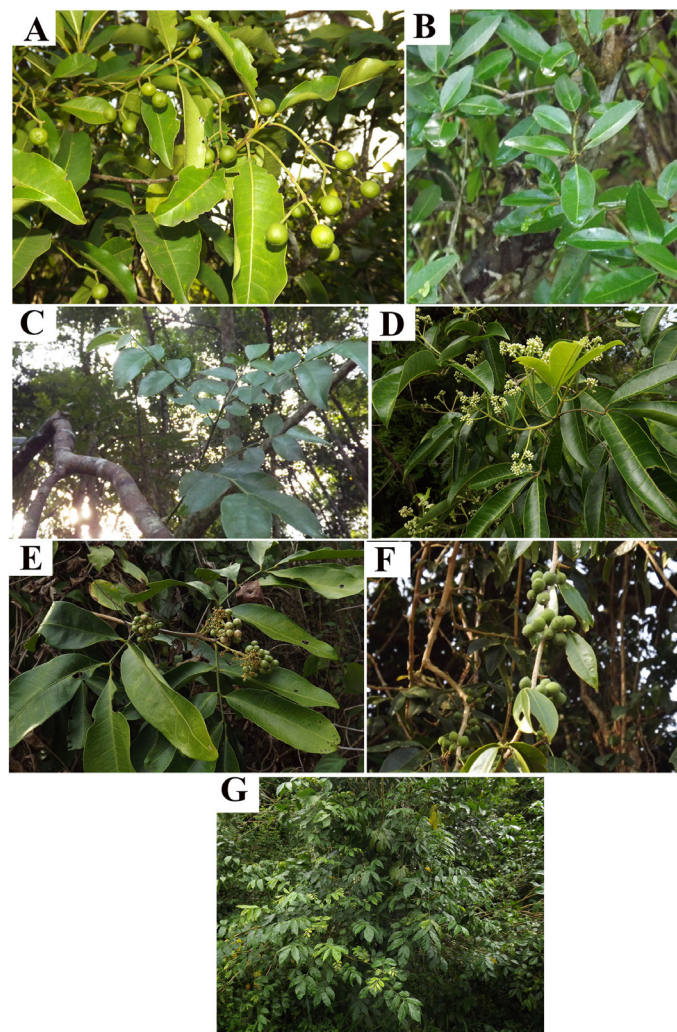
MATERIALS AND METHODS

Plant Materials

The fresh leaves of seven plant species (10 kilograms for each species), including *A. pedunculata*, *A. citroides*, *C. excavata*, *M. pteleifolia*, *G. pentaphylla*, *L. scandens* and *Micromelum* sp. were collected from Binh Chau-Phuoc Buu Nature Reserve, Bung Rieng Ward, Xuyen Moc District, Ba Ria-Vung Tau Province. The specimens were collected by Mr. Le Van Son and the collected sites were described in Table 1. All vouchered specimens were deposited at the Herbarium of Binh Chau-Phuoc Buu Nature Reserve (Table 1). The specimens were identified by Dr. Hong-Thien Van, the botanist from the Institute of Biotechnology and Food-technology, Industrial University of Ho Chi Minh City in January 20, 2020.

Hydrodistillation of the Essential Oils and Yield Calculation

The hydrodistillation of the essential oils was performed immediately after the specimens were collected in Binh Chau-Phuoc Buu Nature Reserve. Five hundred grams of fresh leaves of each of the seven studied species were pulverized and introduced into a 2-liter flask. Distilled water (1 liter) was then added until the leaves were completely submerged. Hydrodistillation processes were performed in a Clevenger type apparatus at 100°C for 4 hours at normal pressure. The essential oils were collected in the receiver arm of the apparatus and transferred into clean and dark bottles. These oils were then dried by Na₂SO₄ and stored at 4°C (Tran et al., 2018). The experiments were performed in triplicate. The oil yields were determined using the equation $RO = M/B_m \times 100\%$, where M is the weight of the extracted oil (g) and B_m is the initial leaf biomass (g) (Olivia et al., 2014).



A. *Acronychia pedunculata*, B. *Atalantia citroides*, C. *Clausena excavata*, D. *Melicope pteleifolia*, E. *Glycosmis pentaphylla*, F. *Luvunga scandens*, G. *Micromelum* sp.

Figure 1. Seven studied species in their habitat

Table 1. Detailed information of collected site of 7 studied species in Binh Chau-Phuoc Buu Nature Reserve

| Code of Samples | Vouchered specimens | Scientific names | Coordinates |
|-----------------|---------------------|-------------------------------|----------------------------|
| TH1 | Le 401 | <i>Glycosmis pentaphylla</i> | 10°32'46"N; 107°30'27"E |
| TH2 | Le 402 | <i>Acronychia pedunculata</i> | 10°32'55"N; 107°29'12"E |
| TH3 | Le 404 | <i>Clausena excavata</i> | 10°32'47"N; 107°28'57"E |
| TH4 | Le 415 | <i>Luvunga scandens</i> | 10°32'46"N; 107°30'27"E |
| TH5 | Le 419 | <i>Micromelum</i> sp. | 10°32'51"N; 107°25'56"E |
| TH6 | Le 432 | <i>Melicope pteleifolia</i> | 10°31'23"N; 107°31'02"E |
| TH7 | Le 433 | <i>Atalantia citroides</i> | 10°30'28"N; 107°30'28"E |

Gas Chromatography-Mass Spectrometry (GC/MS)

Analysis

The chemical constituents of essential oils from the leaves of the seven studied species were identified via the gas chromatography-mass spectrometry (GC/MS) analysis on an Agilent GC 7890B-MS 5975C. An HP-5MS capillary column (30 m x 250 μ m) coated with a 0.25 μ m film was used for separation. Helium (0.9 mL min⁻¹) was used at the pressure of 13.209 psi as the carrier gas. The injected volume was 0.2 μ L for each sample with split mode injector. The column temperature program was as follows: started at 50°C, then increased linearly to 320°C at 8°C min⁻¹. Inlet and MSD temperatures were set up at 250°C and 350°C, respectively. Compounds were identified by comparing their mass spectra with those contained in the NIST02 database. Quantification was performed using the relative peak area percentage. The relative percentage of each component was determined using the ratio between the peak area and the total area of all compounds. The retention indices (arithmetic indices) of the oil components were calculated relative to the homologous series of C₉-C₁₇ n alkanes by using the equation propounded by van den Dool and Kratz.

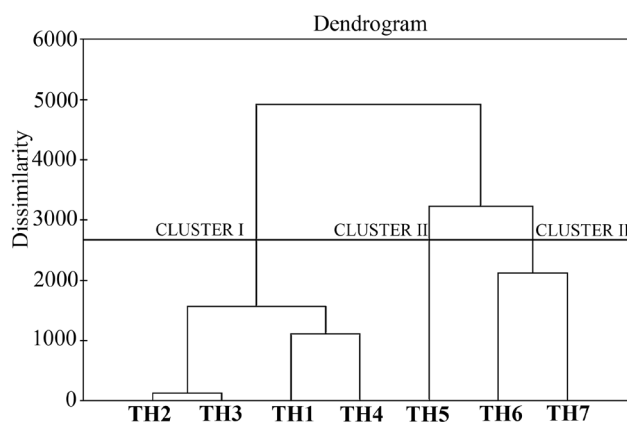
Data Analysis

The experiments of hydrodistillation and GC/MS analysis were conducted in triplicate and the results were expressed as mean \pm standard deviation. Data were statistically interpreted and calculated using Excel 2010 software.

Agglomerative Hierarchical Clustering (AHC) was performed to classify the essential oils isolated from the seven plant species into clusters according to their similarities in the concentration of the 60 compounds identified by GC/MS. Principal Component Analysis (PCA) was performed on the concentrations of the 60 compounds using covariance matrix to identify the main constituents of each of the seven essential oils to reveal the differences among them. The analyses were conducted using XLSTAT Sensory (Addinsoft, Boston, USA).

RESULTS AND DISCUSSION

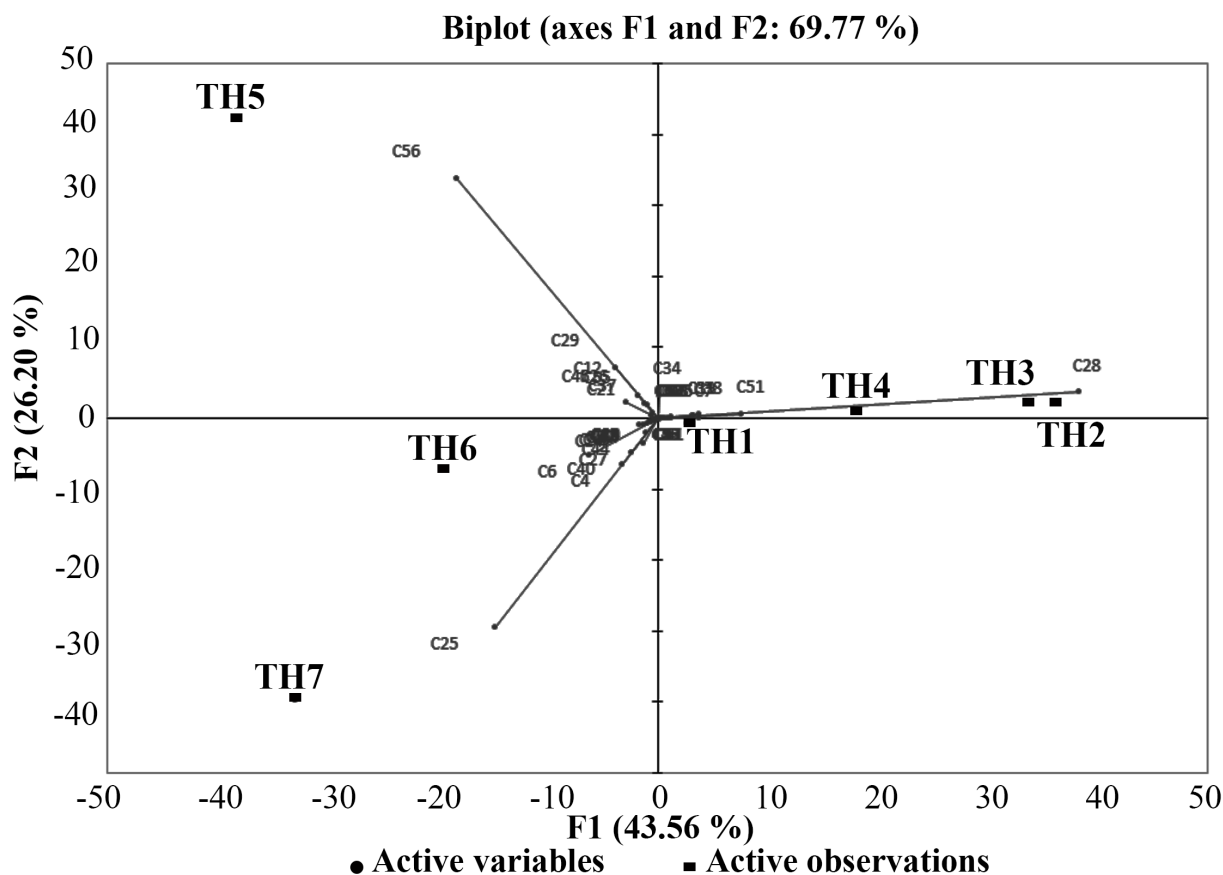
The average oil yields of *Glycosmis pentaphylla*, *Acronychia pedunculata*, *Clausena excavata*, *Luvunga scandens*, *Micromelum* sp., *Melicope pteleifolia*, *Atalantia citroides* varied significantly, with 0.0016 \pm 0.0001%, 0.032 \pm 0.0012%, 0.016 \pm 0.001%, 0.08 \pm 0.0173%, 0.016 \pm 0.0015%, 0.008 \pm 0.0038%, and 0.0016 \pm 0.0002%, respectively. Figure 2 presents the results obtained from clustering analysis and PCA. The chemical compositions of essential oils were divided into 3 clusters: cluster I contained 4 species (*A. pedunculata*, *C. excavata*, *G. pentaphylla*, and *L. scandens*) with the presence of caryophyllene and globulol; cluster II contained 1 species (*Micromelum* sp.) with high concentration of 3-carene (58.03%); cluster III contained 2 species (*M. pteleifolia*, and *A. citroides*) with the presence of 1,9-decadiyne, and 7-oxabicyclo[4.1.0] heptane, 3-oxiranyl-. Furthermore, cluster I could be divided into 2 subgroups: one subgroup characterized by high concentration of caryophyllene with 2 species, *A. pedunculata* and *C. excavata* (57.63% and 55.41%, respectively); the other has lower concentration of caryophyllene with 2 species, *G. pentaphylla* and *L. scandens* (16.02% and 34.66%).



Glycosmis pentaphylla (TH1), *Acronychia pedunculata* (TH2), *Clausena excavata* (TH3), *Luvunga scandens* (TH4), *Micromelum* sp. (TH5), *Melicope pteleifolia* (TH6), *Atalantia citroides* (TH7)

Figure 2. The AHC tree diagram presenting the three clusters of the essential oils of seven studied species

The chemical compositions of the essential oils of the five species, *A. pedunculata*, *C. excavata*, *M. pteleifolia*, *G. pentaphylla* and *Micromelum* sp. were identified in previous studies (Leclercq et al., 1994; Tran et al., 2007; Pham et al., 2007; Lesueur et al., 2008; Tran et al., 2015; Vignesh et al., 2016). However, those specimens were collected in different geographical regions of Vietnam or other countries. In previous studies, specimens of *A. pedunculata*, *C. excavata*, *M. pteleifolia* and *Micromelum* sp. were collected in the Red River delta and northern central coast regions of Vietnam (Vinh Phuc Province, Ninh Binh Province, and Nghe An Province) (Leclercq et al., 1994; Tran et al., 2007; Pham et al., 2007; Lesueur et al., 2008; Tran et al., 2015), whereas *G. pentaphylla* was collected in India (Vignesh et al., 2018). Note that, those specimens were collected in different geographical regions of Vietnam or other countries; therefore, the chemical composition of the specimens in this study maybe differ from the previous studies. In addition, the chemical compositions of essential oil of *L. scandens* and *A. citroides* have not yet been studied.



Glycosmis pentaphylla (TH1), *Acronychia pedunculata* (TH2), *Clausena excavata* (TH3), *Luvunga scandens* (TH4), *Micromelum* sp. (TH5), *Melicope pteleifolia* (TH6), *Atalantia citroides* (TH7)

Figure 3. PCA biplot presenting the main compound constituents of the essential oils of seven studied species

The concentrations of chemical compositions of plant essential oils were found to vary depending on the geographical regions where they are cultivated. In the previous study, Hassiotis et al. (2010) indicated that the concentration of the main constituents of essential oil of *Lavandula angustifolia* Mill. collected at 2 locations, Kilkis and Kato Scholari, Greece, was different. The concentrations of linalyl acetate, linalool, 1,8-cineole, and camphor essential oil collected at Kato Scholari were 30.62%, 29.56%, 5.18%, and 4.03%, whereas those of essential oil collected at Kilkis were 26.92%, 16.78%, 5.55%, and 7.41%, respectively. These results were in line with Devkota et al. (2013) study in which the essential oils of *Centella asiatica* L. (Urb.) grown in 3 different soil conditions of Nepal (shady grassland, open grassland, and open agricultural land) had a variety in concentration of γ -caryophyllene (9.24-32.30%), β -caryophyllene (7.50-24.2%), β -farnesene (1.70-18.89%).

Table 2 showed that the chemical constituents of essential oils of the four species, *A. pedunculata*, *C. excavata*, *M. pteleifolia* and *G. pentaphylla*, were quite similar to those from previous studies. However, there was a significant difference in their concentrations compared to those in previous studies (Leclercq et al., 1994; Lesueur et al., 2008; Vignesh et al., 2016). The essential oil of *A. pedunculata* had the high concentration of caryophyllene (57.63%) and globulol (13.03%) while those in Lesueur et al. (2008) were 13.60% and 0.70%, respectively. Moreover, Lesueur

et al. (2008) showed that α -pinene was the main constituent of the essential oil of *A. pedunculata* but this compound only accounted for a small portion in the present study (1.57%). According to Leclercq et al. (1994), the essential oil of *C. excavata* contained caryophyllene (25.30%), globulol (0.90%), germacrene (11.80%) and β -phellandrene (9.20%). In this study, the concentrations of caryophyllene and globulol were 55.41% and 12.87%, respectively. Besides, germacrene and β -phellandrene were not identified in the essential oil of *C. excavata*. Additionally, the essential oil of *G. pentaphylla* collected at Binh-Chau_Phuoc-Buu Nature Reserve contained all three compounds of β -ocimene (23.10%), caryophyllene (16.02%), and γ -maaliene (9.53%), but only caryophyllene (7.47%) was identified in Vignesh et al. (2016). On the contrary, bicyclo [6.1.0] non-1-ene and benzaldehyde oxime were the main constituents of the essential oil of *G. pentaphylla* in previous study of Vignesh et al. (2016) whereas those were not found in this study. In 2017, Tran et al. (2007) suggested that (E)- β -ocimene was the main constituent of the essential oil of *M. pteleifolia* with a concentration of 24.40% while that was not found in this study. Besides, the main constituents of the essential oil of *M. pteleifolia* collected at Binh-Chau_Phuoc-Buu Nature Reserve, 1,9-decadiyne (32.59%) and patchoulane (10.04%), were not identified in the essential oil of *M. pteleifolia* in Tran et al. (2007).

Table 2. Chemical compositions in the essential oils from the leaves of 7 species of the Rutaceae family in Binh Chau-Phuoc Buu Nature Reserve

| Compounds | Code | RT | RI | The relative percentage (%) | | | | | | |
|--|------|-------|------|-----------------------------|------------|------------|------------|------------|------------|------------|
| | | | | TH1 | TH2 | TH3 | TH4 | TH5 | TH6 | TH7 |
| α -pinene | C1 | 8.30 | 980 | 2.33±0.12 | 1.57±0.02 | 1.6±0.1 | - | - | - | - |
| 3-carene | C2 | 8.58 | 1011 | - | - | 0.32±0.03 | - | 58.03±0.21 | - | - |
| β -terpinen | C3 | 9.24 | 1028 | - | 0.60±0.1 | - | - | - | - | - |
| Cyclopropane, 1,2- di- methyl-3- methylene - | C4 | 9.47 | 1062 | - | - | - | - | - | - | 11.92±0.44 |
| 1,8-nonadiyne | C5 | 9.56 | 1065 | - | - | - | - | - | 3.15±0.23 | - |
| 1,9-decadiyne | C6 | 9.69 | 1069 | - | - | - | - | - | 32.59±1.52 | 3.67±0.34 |
| Santolina triene | C7 | 10.06 | 1071 | - | - | 1.88±0.25 | - | - | 4.11±0.21 | - |
| β -ocimene | C8 | 10.72 | 1079 | 23.1±1.12 | 10.91±0.05 | - | - | - | - | - |
| Linalool | C9 | 10.90 | 1098 | 1.95±0.13 | - | 0.75±0.01 | - | - | - | - |
| Allo-ocimene | C10 | 11.51 | 1103 | 1.04±0.06 | - | - | - | - | - | - |
| β -myrcene | C11 | 11.60 | 1109 | 0.18±0.01 | - | - | - | - | - | - |
| Limonene | C12 | 11.94 | 1116 | 2.76±0.02 | - | - | 21.26±0.94 | - | - | - |
| Decanal | C13 | 12.70 | 1120 | 0.05±0.02 | - | - | - | - | - | - |
| Pent-2-ynal, 4,4-dimethyl- | C14 | 12.13 | 1128 | - | - | - | - | - | 2.27±0.27 | - |
| 2,3-hexadiene, 2- methyl - | C15 | 12.25 | 1147 | - | - | - | - | 5.51±0.1 | - | - |
| 2-octyne | C16 | 12.36 | 1136 | - | - | - | - | - | 1.32±0.08 | - |
| 1,7-octadiyne | C17 | 12.46 | 1139 | - | - | - | - | - | 0.87±0.22 | - |
| 6-nonynoic acid | C18 | 12.55 | 1141 | - | - | - | - | - | 1.12±0.12 | - |
| Santolina alcohol | C19 | 12.72 | 1143 | - | - | - | - | - | 1.41±0.2 | - |
| Cyclopropene, 1- methyl- 3- (2-methyl cyclopropyl)- | C20 | 12.77 | 1147 | - | 0.75±0.01 | - | - | - | - | - |
| 1,5-heptadiene, 2,5-di- methyl-3-methylene | C21 | 12.90 | 1149 | 0.09±0.03 | - | 1.12±0.03 | - | - | - | - |
| β -elemene | C22 | 13.04 | 1250 | 1.03±0.05 | - | - | - | - | - | - |
| Caryophyllene | C23 | 13.12 | 1251 | 16.14±0.29 | 57.63±0.01 | 55.41±0.04 | 34.66±0.02 | - | - | - |
| Copaene | C24 | 13.18 | 1353 | 0.58±0.03 | - | - | - | - | - | - |
| α -farnesene | C25 | 13.29 | 1355 | - | 0.24±0.02 | 0.35±0.02 | - | - | 1.44±0.02 | - |
| 7-octen-2-ol, 2-meth- yl-6-methylene | C26 | 13.36 | 1357 | - | - | - | - | 0.81±0.05 | 1.65±0.07 | - |
| Patchoulane | C27 | 13.43 | 1363 | - | - | - | - | - | 10.04±1.00 | - |
| 7-oxabicyclo[4.1.0] heptane, 3-oxiranyl- | C28 | 13.57 | 1365 | - | - | - | - | - | - | 53.91±0.25 |
| 2-methyl-1-phenyl-2-pro- pen-1-ol | C29 | 13.60 | 1368 | - | - | - | - | 3.57±0.02 | - | - |

| Compounds | Code | RT | RI | The relative percentage (%) | | | | | | |
|--|------|-------|------|-----------------------------|------------|------------|------------|------------|-----------|-----------|
| | | | | TH1 | TH2 | TH3 | TH4 | TH5 | TH6 | TH7 |
| Viridiflorene | C30 | 13.63 | 1369 | - | 2.73±0.05 | 3.05±0.05 | - | - | 9.40±0.56 | 5.21±0.41 |
| 1,4-pentadiene | C31 | 13.67 | 1371 | - | - | - | - | 12.18±0.68 | - | - |
| 1,3,6-octatriene, 3,7-dimethyl-, (z) | C32 | 13.67 | 1372 | - | 1.56±0.01 | 9.94±0.08 | - | - | - | - |
| α-selinene | C33 | 13.70 | 1374 | - | 0.83±0.04 | - | - | - | - | - |
| α-humulene | C34 | 13.75 | 1376 | 4.51±0.09 | 0.31±0.03 | - | 10.85±1.33 | 5.23±0.28 | - | - |
| Alloaromadendrene | C35 | 13.79 | 1378 | 6.12±0.03 | 3.67±0.03 | - | - | - | - | - |
| γ-elemene | C36 | 13.89 | 1378 | - | - | - | 25.74±0.54 | - | - | - |
| γ-gurjunene | C37 | 13.95 | 1379 | 0.95±0.03 | - | - | - | - | - | - |
| Cis-α-bisabolene | C38 | 13.98 | 1379 | - | - | 4.39±0.04 | - | - | - | - |
| 6-deoxysorbitol pentaacetate | C39 | 14.05 | 1380 | - | - | - | - | 1.41±0.2 | - | - |
| Cis-muurolo-3,5-diene | C40 | 14.11 | 1382 | - | - | 1.41±0.04 | - | - | - | - |
| 4-propyl-1,6-heptadien-4-ol | C41 | 14.12 | 1383 | - | - | - | - | - | - | 8.84±0.86 |
| γ-maaliene | C42 | 14.17 | 1384 | 9.53±0.02 | - | - | - | - | - | - |
| Oxirane, 2,2'-[1,4-butanediylbis(oxy-methylene)]bis- | C43 | 14.20 | 1385 | - | - | - | - | - | 1.92±0.09 | - |
| δ-cadinene | C44 | 14.23 | 1386 | 3.64±0.03 | 1.14±0.05 | - | 2.71±0.1 | - | - | - |
| Propanamide, 2,3,3,3-tetrafluoro-2-heptafluoropropoxy-N-(2,3-dimethyl-5-oxo-1-phenyl-3-pyrazolin-4-yl) | C45 | 14.29 | 1392 | - | - | - | - | - | - | 3.88±0.87 |
| 1-(1-adamantyl)-1-phenylethanol | C46 | 14.55 | 1394 | - | - | - | - | 6.16±0.31 | 2.26±0.18 | 2.16±0.12 |
| Spathulenol | C47 | 14.61 | 1397 | - | - | - | 3.97±0.5 | - | - | - |
| 3-cyclohexen-1-carboxaldehyde, 3,4-dimethyl | C48 | 14.67 | 1398 | - | - | - | - | - | 1.05±0.2 | - |
| Caryophyllene oxide | C49 | 14.71 | 1399 | 6.10±0.25 | - | 0.35±0.03 | - | - | 7.25±0.34 | - |
| 7-propylidene-bicyclo[4.1.0] heptan | C50 | 14.78 | 1400 | - | 0.58±0.01 | 0.70±0.18 | - | - | 5.68±0.2 | - |
| Globulol | C51 | 14.82 | 1402 | 7.56±0.21 | 13.03±0.08 | 12.87±0.38 | - | - | - | - |
| α-cadinol | C52 | 14.85 | 1402 | 5.09±0.09 | - | - | - | - | - | - |
| 2-methyl-1-nonene-3-yne | C53 | 14.92 | 1403 | - | - | 2.01±0.08 | - | - | - | - |
| 1,5,9,13-tetradecatetraene | C54 | 14.95 | 1404 | - | - | - | - | - | - | 0.83±0.18 |
| 2-heptenoic acid, 7-(methylene-cyclopropyl)-, methyl ester | C55 | 14.99 | 1405 | 2.48±0.02 | 0.23±0.02 | - | - | 3.19±1.01 | - | - |

| Compounds | Code | RT | RI | The relative percentage (%) | | | | | | |
|---|------|-------|------|-----------------------------|------------|------------|------------|------------|------------|------------|
| | | | | TH1 | TH2 | TH3 | TH4 | TH5 | TH6 | TH7 |
| β -chamigrene | C56 | 15.01 | 1408 | - | 1.74±0.03 | - | - | - | - | - |
| β -selinene | C57 | 15.08 | 1410 | 1.57±0.06 | - | - | - | - | - | - |
| 1H,15H-hexadecamethyl- loctasiloxane | C58 | 18.29 | 1581 | - | - | - | - | 1.47±0.47 | - | 3.11±0.14 |
| 2- (octyloxycarbonyl) benzoic acid | C59 | 19.67 | 1614 | - | - | 1.03±0.07 | - | - | - | - |
| Bis (2-ethylhexyl) phthalate | C60 | 19.78 | 1616 | 3.17±0.02 | 1.00±0.11 | - | - | - | - | - |
| Monoterpene hydrocarbons | | | | 29.52±1.09 | 15.23±0.05 | 15.56±0.42 | 21.26±0.94 | 58.03±0.20 | 9.77±0.35 | 0.00 |
| Oxygenated monoterpenes | | | | 1.95±0.13 | 0.00 | 0.75±0.01 | 0.00 | 0.81±0.05 | 4.98±0.32 | 8.84±0.86 |
| Sesquiterpene hydrocarbons | | | | 43.12±0.22 | 68.03±0.03 | 64.61±0.13 | 73.96±1.88 | 5.23±0.28 | 20.84±0.25 | 5.21±0.41 |
| Oxygenated sesquiterpenes | | | | 18.72±0.51 | 13.03±0.08 | 13.22±0.39 | 0.00 | 6.16±0.32 | 9.51±0.45 | 2.16±0.12 |
| Non-terpenes | | | | 5.69±0.01 | 1.97±0.10 | 3.05±0.12 | 0.00 | 27.33±2.08 | 41.05±2.14 | 77.32±1.25 |
| Total identified (%) | | | | 98.99±0.76 | 97.53±0.13 | 97.19±0.16 | 95.21±2.80 | 97.56±2.26 | 86.16±2.18 | 93.54±1.82 |

Note: *Glycosmis pentaphylla* (TH1), *Acronychia pedunculata* (TH2), *Clausena excavata* (TH3), *Luvunga scandens* (TH4), *Micromelum* sp. (TH5), *Melicope pteleifolia* (TH6), *Atalantia citroides* (TH7). RT: retention time, RI: Kovats retention index

In the present study, the chemical compositions of the essential oils isolated from leaves of *L. scandens*, *A. citroides* and *Micromelum* sp. have been identified for the first time. The main constituents of the essential oil of *A. citroides* were 7-oxabicyclo[4.1.0] heptance, 3-oxiranyl- (53.91%) and cyclopropane, 1,2-dimethyl-3-methylene- (11.92%). Additionally, the essential oil of *L. scandens* consisted of caryophyllene (34.66%), γ -elemene (25.75%), limonene (21.26%), and α -humulene (10.85%). Furthermore, 3-carene (58.03%) and 1,4-pentadiene (12.18%) were identified as the major constituent of essential oil *Micromelum* sp.

In this study, we found the presence of some bioactive compounds in chemical composition of these species such as caryophyllene, limonene, 3-carene, which suggests these species as sources to extract the bioactive compounds or to use as remedies for treatment of some diseases. For example, caryophyllene, which is found in *G. pentaphylla*, *C. excavata*, *A. pedunculata*, *L. scandens*, exhibits the anti-microbial, anti-inflammatory, and anti-cancer effects (Dahham et al., 2015). According to Miller et al. (2013), limonene, which is one of the main components of *L. scandens*, possesses the chemopreventive and chemotherapeutic activities in breast cancer. Moreover, 3-carene, the main component of *M. hirsutum*, is reported for its anti-inflammatory and antinociceptive effects as well as its role in differentiation process of osteoblastic cells (Jeong et al., 2008; Huang et al., 2019). These findings provide the basis for the further application of these species in medicine.

CONCLUSIONS

The present study identified a total of 60 compounds of the essential oils from the leaves of seven species of the Rutaceae family collected in Binh-Chau-Phuoc-Buu Nature Reserve. The results suggested that the concentration of the main constituents of the essential oil of *Acronychia pedunculata*, *Clausena excavata*,

Melicope pteleifolia and *Glycosmis pentaphylla* was significantly different from that reported in previous studies. In brief, the main constituents of *G. pentaphylla* essential oil were β -ocimene (23.10%), caryophyllene (16.14%), γ -maaliene (9.53%), globulol (7.56%) whereas 1,9-decadiene (32.59%), patchoulane (10.04%), viridiflorene (9.40%), caryophyllene oxide (7.25%) were the major compounds in essential oils of *M. pteleifolia*. The essential oil of *A. pedunculata* possessed a high amount of caryophyllene (57.63%), β -ocimene (10.91%), Globulol (13.03%). On the other hand, the main constituents of *C. excavata* essential oil were caryophyllene (55.41%), Globulol (12.87%), 1,3,6-Octatriene, 3,7-dimethyl-, (z) (9.4%). Furthermore, this study analyzed the chemical composition of the essential oils of *Luvunga scandens*, *Atalantia citroides* and *Micromelum* sp. for the first time. Accordingly, the main constituents of *L. scandens* essential oil were caryophyllene (34.66%), γ -elemene (25.74%), limonene (21.26%), α -humulene (10.85%) while those of *A. citroides* essential oil were 7-oxabicyclo[4.1.0] heptance, 3-oxiranyl (53.91%), cyclopropane, 1,2- dimethyl- 3- methylene (11.92%), 4-propyl-1,6-heptadien-4-ol (8.84%). Furthermore, the essential oil of *Micromelum* sp. was found to be rich of 3-carene (58.03%), 1,4-pentadiene (12.18%), 1-(1-adamantyl)-1-phenylethanol (6.16%). We found the presence of some bioactive compounds in chemical components of these species, which provides the basis for further application of these species in medicine.

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