



Asian Journal of Plant Sciences

ISSN 1682-3974

science
alert

ANSI*net*
an open access publisher
<http://ansinet.com>



Research Article

Antibacterial and Antioxidant Abilities of Extracts and Essential Oil of *Perilla frutescens*

¹Tran Ngoc Quy, ¹Ngo Nguyen Thu Ngan, ¹Ly Huynh Kim Trong, ¹Ngo Tan Minh, ¹Huynh Tien Dat, ¹Nguyen Le Kim Ngoc, ¹Tran Thanh Toi, ²Tran Thanh Men, ¹Do Tan Khang and ³Nguyen Van Ay

¹Institute of Food and Biotechnology, Can Tho University, Xuan Khanh, Ninh Kieu, Can Tho, Vietnam

²College of Natural Sciences, Can Tho University, Xuan Khanh, Ninh Kieu, Can Tho, Vietnam

³College of Agriculture, Can Tho University, Xuan Khanh, Ninh Kieu, Can Tho, Vietnam

Abstract

Background and Objective: *Perilla frutescens* (Lamiaceae family) is an annual herbaceous species of the mint family originating from East Asia. This study aimed to evaluate some biological activities of *Perilla frutescens* extracts and essential oils collected at three locations, Can Tho City, An Giang and Vinh Long Province. **Materials and Methods:** Fresh perilla plants were collected from perilla farmers in Can Tho City (CT), An Giang (AG) and Vinh Long (VL) provinces. Samples were extracted using ethanol as solvent and isolated essential oil. Antioxidant activity was evaluated by two assays DPPH and ABTS. The antibacterial activity was conducted on *Escherichia coli*, *Staphylococcus aureus* and *Propionibacterium acnes* by disc diffusion method. **Results:** Quantitative results of total polyphenols and total flavonoids showed that the Can Tho treatment was the highest with 12.78 mg GAE/g extract and 65.99 mg QE/g extract. Essential oils contain a total of 38 volatile compounds. The major predominant compounds were perillaldehyde, 2-Furyl n-pentyl ketone, β -Caryophyllene, β -Linalool, valeric acid pent-2-en-4-ynyl ester and limonene in variable levels. **Conclusion:** All extracts and essential oils of *P. frutescens* leaves have revealed potent antioxidants and antibacterial activities. The different ecological factors determine the content and various biological activities.

Key words: Antibacterial, antioxidant, essential oil, *Perilla frutescens*, perillaldehyde, polyphenols

Citation: Quy, T.N., N.N.T. Ngan, L.H.K. Trong, N.T. Minh and H.T. Dat *et al.*, 2024. Antibacterial and antioxidant abilities of extracts and essential oil of *Perilla frutescens*. Asian J. Plant Sci., 23: 184-192.

Corresponding Author: Nguyen Van Ay, College of Agriculture, Can Tho University, Xuan Khanh, Ninh Kieu, Can Tho, Vietnam

Copyright: © 2024 Tran Ngoc Quy *et al.* This is an open access article distributed under the terms of the creative commons attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Perilla has the scientific name *Perilla frutescens* and belongs to the Lamiaceae family. *Perilla* is an annual herbaceous species of the mint family originating from East Asia. It is widely distributed throughout countries with hot and humid tropical climates such as India, China, Japan, Thailand and Vietnam¹. The leaves contain about 3.1% protein, 0.8% fat, 4.1% carbohydrate and 1.1% ash. The plant yields 0.3-1.3% essential oil, which contains 20% citral^{1,2}.

Perilla contains several antioxidant compounds, which play an important role in preventing diseases caused by free radicals such as aging, cancer and dermatitis^{3,4}. *Perilla* contains several organic natural compounds including phenolic acids, flavonoids, triterpenes, carotenoids, phytosterols, fatty acids, tocopherols and policosanols. Moreover, several researchers have been captivated by the pharmacological properties of many individual compounds such as rosmarinic acid, perillaldehyde, luteolin, apigenin, tormentic acid and isogomaketone^{2,5,6}. It is rich in perillaldehyde and limonene. They are volatile compounds with potent inhibition of the growth of *Salmonella* as well as many pathogenic microorganisms in food such as *Staphylococcus aureus* bacteria, *Escherichia coli*, *Salmonella typhimurium*, *Vibrio parahaemolyticus*, *Trichophyton mentagrophytes*, staphylococci, dysentery bacilli and colonic bacilli⁷. Many recent studies on the antibacterial and antioxidant activities available in plants are being carried out for application in the food, pharmaceutical and cosmetic industries. This study aimed to investigate the antibacterial and antioxidant capacities of extracts and essential oils of *Perilla frutescens* leaves.

MATERIALS AND METHODS

Study area: The study was conducted from April 2023 to October 2023 in Institute of Food and Biotechnology, Can Tho University, Vietnam.

Sampling method: Fresh perilla plants were collected from perilla farmers in Can Tho City (CT), An Giang (AG) and Vinh Long (VL) Province. *Staphylococcus aureus* and *Escherichia coli* were provided by the Molecular Biology Department of Can Tho University.

Preparation of perilla extract and essential oil: The dried leaves (2 kg) of each accession (CT, AG, VL) were extracted

with ethanol 96% with a ratio of 1:5. The *Perilla frutescens* extract (PFE) were filtered by filter paper and then evaporated in a rotary evaporator 70°C to remove the solvent. Ethanol extraction was repeated three times.

The extraction of essential oil (EO) from each accession involved a 4 hrs hydrodistillation process using a Clevenger-type apparatus. Precisely measured dried leaves (0.5 kg) were placed in a round-bottomed flask filled with distilled water (3 L) following the method described by Ahmed and Tavaszis-Sarosiin⁸. After collection, the essential oils underwent a drying process with anhydrous sodium sulfate to eliminate residual water. Subsequently, the essential oils were preserved in sealed dark vials and stored at 4°C until analysis.

Determination of total polyphenol content (TPC): The total polyphenol content of PFE was determined based on the method of Folin-Ciocalteu⁹. Briefly, 100 µL gallic acid solution 10-100 µg/mL (or extract 1 mg/mL) were mixed with 250 µL of 10% Folin-Ciocalteu reagent and these mixtures were reacted at room temperature for 5 min. Then, after 200 µL of 2% Na₂CO₃ was added, the mixtures were reacted in darkness for 45 min. The absorbance was measured using a plate reader spectrophotometer (Multiskan, Thermo Fisher Scientific, Tanglin Rd, Singapore) at 725 nm. The TPC results were expressed as mg gallic acid equivalents (GAE)/g extract. Each sample was analyzed in triplicate.

Determination of total flavonoid content (TFC): The total flavonoid content of PFE was determined based on method of Djeridane *et al.*¹⁰. Briefly, 100 µL quercetin solution 10-100 µg/mL (or extract 1 mg/mL) was mixed with 100 µL of 2% AlCl₃ and these mixtures were reacted at room temperature for 15 min. The absorbance was measured using a spectrophotometer at 430 nm. The TFC results were expressed as mg quercetin equivalents (QE)/g extract. Each sample was analyzed in triplicate.

Gas chromatographic-mass spectrometric analysis: The analysis of essential oils was conducted through GC-MS utilizing an Agilent Technology 6890 N instrument, which was equipped with an HP-5MS capillary column (30 m × 0.25 mm i.d. × 0.25 µm) and an Agilent Technologies MS 5975 inert mass selective detector. The initial temperature was set at 60°C and maintained for 5 min, followed by a gradual increase at a rate of 3°C/min until it reached 240°C. Helium was used as the carrier gas at a flow rate of 1 mL/min, while the injector and detector temperatures were set at 250°C. The split ratio was adjusted to 1:30 and the injection volume was 0.2 µL. Prior

to injection, the crude essential oils underwent dilution (10 μ L EO/1 mL hexane). The percentage composition of the essential oil was calculated based on the GC peak areas, with the acquisition mass range set at 40-400 m/z and an ionization energy of 70 eV.

DPPH radical scavenging activity: The DPPH scavenging was modified from the described method of Phuyal *et al.*¹¹. About 100 μ L of PfE 0.5 mg/mL (or EOs 0.2-1%) was added in 100 μ L DPPH 100 μ g/mL and then incubated at 30°C in darkness for 30 min, followed by absorbance measurement at 517 nm. Gallic acid was used as a positive control. The percentage of inhibition (I%) was calculated by formula:

$$I (\%) = \frac{A_o - A_s}{A_o} \times 100$$

Where:

A_o = Control reaction absorbance

A_s = Testing specimen absorbance

ABTS radical scavenging activity: The ABTS scavenging was modified from the ABTS cation decolorization assay¹². The stock solution was mixed including ABTS (7 mM) with potassium persulfate (2.4 mM) and allowed to react for 16 hrs in the dark at room temperature. The mixtures were diluted with methanol to an absorbance of 0.70 ± 0.02 at 734 nm. While 10 μ L of extract 0.5-2 mg/mL (or essential oils 0.2-1%) was allowed to react with 190 μ L of the ABTS reagent, after 7 min, followed by absorbance measurement at 734 nm. Gallic acid was used as a positive control. The percentage of inhibition (I%) was calculated by formula:

$$I (\%) = \frac{A_o - A_s}{A_o} \times 100$$

Where:

A_o = Control reaction absorbance

A_s = Testing specimen absorbance

Antibacterial activity: The antimicrobial assay was modified from the disc diffusion method of Mahesh and Satish¹³. The 50 μ L of bacterial suspension (10^6 CFU/mL) was spread onto the Tryptic Soy Agar medium. Then, the sterilized paper discs (6 mm in diameter) were placed on the surface and impregnated with 10 μ L of PfE 1-10 mg/mL (or essential oils 0.01-100%). Streptomycin (1 mg/mL) was applied as a positive control. The plates were incubated for 24 hrs at 37°C. After incubation, the inhibition zones were measured in millimeters.

Data analysis: The collected data were processed using Microsoft Excel 2016 software. The statistical Minitab 16 software was used to analyze the variance and test the difference between the mean of the treatments at a 5% significance level.

RESULTS

TPC and TFC in perilla extract: Table 1 shows that the total polyphenol content in perilla leaf extract in three places ranged from 7.79-12.78 mg GAE/g extract, the flavonoid content ranged from 60.09-65.99 mg QE/g extract. In particular, the Can Tho and An Giang extracts had similar total polyphenol and flavonoid content, statistically significantly different from the Vinh Long treatment at the 5% significance level through Tukey's test.

Identification of essential oil constituents: Table 2 displays the compositions of essential oils (EO) extracted from perilla leaves. A total of thirty-eight volatile components were identified and quantified across three essential oils. The highest number of components, comprising approximately 100%, was observed in EOCT with thirty compounds. Following closely, EOAG exhibited twenty-two components, also constituting 100% of the oil. In contrast, the lowest number of components, fifteen-six in total, was found in EOVL, representing 100% of its composition. In the EOVL, 2-Furyl n-pentyl ketone (67.28%), beta-Caryophyllene (8.28%) and β -Linalool (5.89%); 2-Furyl n-pentyl ketone (68.39%), β -Caryophyllene (9.85%) and valeric acid, pent-2-en-4-ynyl ester (5.38%) in EOAG; limonene (33.55%), perillaldehyde (19.73%) and 2-Furyl n-pentyl ketone (16.69%) in EOCT were detected.

DPPH radical scavenging activity: Perilla extract showed a high ability to remove DPPH radicals at a concentration of 0.5 μ g/mL. All three treatments at low concentrations could eliminate DPPH free radicals with the percentage of inhibition ranging from 72.76-80.92% (Table 3). Among them, the PfEAG had the highest percentage inhibition of 80.92%, a statistically significant difference with the other two treatments at the 5% significance level. The linear regression equation for the percentage of gallic acid inhibition was $y = 11.908x + 3.437$ ($R^2 = 0.9905$) and IC_{50} was 3.91 ± 0.01 μ g/mL.

The oils exhibited varying degrees of scavenging ability. The EOAG and EOVL exhibited strong radical scavenging effects at 1% with percentage inhibition of $61.82 \pm 0.15\%$ and $60.36 \pm 0.23\%$, respectively, which was low to that of the gallic acid (Table 4). Notably, EOAG had the strongest activity ($IC_{50} = 0.68\%$).

Table 1: Total polyphenol and total flavonoid content in three perilla extracts

Sample	TPC (mg GAE/g extract)	TFC (mg QE/g extract)
PFECT	12.78±0.108 ^a	65.99±1.95 ^a
PFEAG	11.88±0.360 ^a	65.05±0.72 ^a
PFEVL	7.79±0.859 ^b	60.09±0.56 ^b

Values assigned by the same superscript alphabets in a column were not significantly different at p<0.05

Table 2: Qualitative and quantitative results of volatile compounds present in the EOs

No.	Volatile components	Mean of GC-MS area (%)		
		EOVL	EOAG	EOCT
1	1-Octen-3-ol	1.97	1.46	2.62
2	Limonene	2.44	0.40	33.55
3	β-Linalool	5.89	4.41	3.56
4	2-Furyl n-pentyl ketone	67.28	68.39	16.69
5	Perillaldehyde	1.92	1.3	19.73
6	Methyl geranate	0.48	0.46	0.11
7	β-Elemene	0.24	0.31	0.13
8	β-Caryophyllene	8.28	9.85	6.68
9	Germacrene D	0.28	0.38	0.22
10	alpha-Caryophyllene	1.08	1.51	0.79
11	trans-alpha-Bergamotene	0.48	0.52	0.1
12	alpha-Farnesene	0.13	0.24	0.1
13	alpha-Cubebene	-	0.06	0.06
14	3-Octanol	-	0.17	1.41
15	alpha-Thujene	-	0.90	1.34
16	1-Ethoxy-2-methoxy-4-methylbenzene	0.26	0.47	-
17	4-(2-Methylcyclohex-1-enyl)-but-2-enal	2.08	1.89	-
18	5-Methyl-3a,7a-dihydro-1H-indene-1,7(4H)-dione	1.51	1.26	-
19	Isovalerone	5.68	-	-
20	Valeric acid, pent-2-en-4-ynyl ester	-	5.38	-
21	alpha-Murolene	-	0.13	-
22	β-Bisabolene	-	0.04	-
23	delta-Cadinene	-	0.47	-
24	alpha-Pinene	-	-	0.43
25	β-Phellandrene	-	-	1.59
26	β-Myrcene	-	-	1.17
27	3-Carene	-	-	0.55
28	alpha-Terpinolene	-	-	0.21
29	alpha-Terpineol	-	-	0.40
30	2,2-Dimethyl 1-3-heptanone	-	-	0.47
31	Perilla alcohol	-	-	0.14
32	β-Citronellol	-	-	0.5
33	β-Citral	-	-	0.09
34	6,6-Dimethyl-2-vinylidene bicyclo 0,6[3.1.1]heptane	-	-	0.6
35	alpha-Methylcinnamaldehyde	-	-	3.69
36	Perillic alcohol	-	-	2.31
37	Eugenol	-	-	0.06
38	7-Methyl-1,2,3,5,8,8a-hexahydronaphthalene	-	-	0.7
	Total	100	100	100
	No. of compounds	16	22	30

--: Absent

Table 3: DPPH Inhibition percentage of three perilla extracts at a concentration of 0.5 µg/mL

Treatment	Percentage of inhibition (%)
PFECT	75.28±1.41 ^b
PFEAG	80.92±0.87 ^a
PFEVL	72.76±0.60 ^b

Values assigned by the same superscript alphabets in a column were not significantly different at p<0.05

Table 4: IC₅₀ values DPPH scavenging of three essential oils

Sample	Linear regression equation	IC ₅₀ (%)
EOVL	$y = 47.52x + 11.85$ ($R^2 = 0.97$)	0.80 ^b
EOAG	$y = 38.19x + 12.12$ ($R^2 = 0.99$)	0.68 ^c
EOCT	$y = 42.06x + 21.23$ ($R^2 = 0.98$)	>1 ^a

Values assigned by the same superscript alphabets in a column were not significantly different at $p < 0.05$

Table 5: IC₅₀ values ABTS scavenging of three extracts and gallic acid

Sample	Linear regression equation	IC ₅₀ value (µg/mL)
PfECT	$y = 22.821x + 19.885$ ($R^2 = 0.9852$)	1.32 ^d
PfEAG	$y = 17.509x + 13.432$ ($R^2 = 0.9915$)	2.09 ^c
PfEVL	$y = 14.537x + 16.958$ ($R^2 = 0.9791$)	2.27 ^b
Gallic acid	$y = 1.0764x + 0.0734$ ($R^2 = 0.9792$)	46.38 ^a

Values assigned by the same superscript alphabets in a column were not significantly different at $p < 0.05$

Table 6: IC₅₀ values ABTS scavenging of three essential oils

Sample	IC ₅₀ (%)
EOVL	0.80 ± 0.01 ^c
EOAG	0.99 ± 0.01 ^a
EOCT	0.86 ± 0.01 ^b

Values assigned by the same superscript alphabets in a column were not significantly different at $p < 0.05$

Table 7: Value of antibacterial zones diameter of essential oil treatments

Concentration (%)	Diameter of resistance zones (mm)					
	<i>Staphylococcus aureus</i>			<i>Propionibacterium acnes</i>		
	EOCT	EOAG	EOVL	EOCT	EOAG	EOVL
0.01	6.00 ^f	6.00 ^f	6.00 ^f	6.00 ^e	6.00 ^{de}	6.00 ^e
0.1	7.33 ^{ef}	6.67 ^{ef}	7.67 ^e	7.33 ^d	6.67 ^d	7.33 ^d
1	9.33 ^d	10.33 ^{cd}	9.33 ^d	9.33 ^{bc}	9.67 ^{bc}	9.33 ^{bc}
10	11.67 ^{bc}	11.67 ^{bc}	11.33 ^{bc}	10.67 ^b	10.67 ^b	10.33 ^b
100	15.33 ^a	12.67 ^a	14.67 ^a	16.33 ^a	12.33 ^a	15.33 ^a
Streptomycin	18	17	18	17	18	18

Values assigned by the same superscript alphabets in a column were not significantly different at $p < 0.05$

Table 8: Value of antibacterial zones diameter of extract treatments

Concentration (mg/mL)	Diameter of antibacterial zones (mm)							
	<i>Escherichia coli</i>				<i>Staphylococcus aureus</i>			
	1	2.5	5	10	1	2.5	5	10
Can Tho	4.33 ^b	6.33 ^b	8.67 ^{ab}	10.67 ^a	5.67 ^b	7.00 ^b	8.00 ^b	9.67 ^{ab}
An Giang	4.33 ^b	5.00 ^c	6.67 ^c	10.00 ^a	5.67 ^b	6.67 ^b	7.67 ^b	9.00 ^{bc}
Vinh Long	3.67 ^b	5.33 ^{bc}	7.67 ^{bc}	10.33 ^a	5.33 ^b	6.33 ^b	7.33 ^b	8.33 ^c
Streptomycin	9.88 ^a	9.89 ^a	9.89 ^a	9.89 ^a	10.45 ^a	10.45 ^a	10.45 ^a	10.45 ^a

Values assigned by the same superscript alphabets in a column were not significantly different at $p < 0.05$

ABTS radical scavenging activity: The linear regression equation of gallic acid was $y = 1.0764x + 0.0734$ ($R^2 = 0.9792$), equivalent to an IC₅₀ value of 46.63 µg/mL. The results showed that all extracts could eliminate ABTS free radicals with IC₅₀ values ranging from 1.32 to 2.27 µg/mL, as shown in Table 5. Among them, the PfECT showed good results with the lowest IC₅₀ value, 1.32 µg/mL. This meant that the PfECT had better antioxidant capacity than in the remaining extracts.

The oils exhibited varying degrees of ABTS scavenging ability with IC₅₀ values ranging from 0.80 ± 0.01 - 0.99 ± 0.01 % as shown in Table 6. Among them, EOVL showed the strongest

free radical scavenging activity with the lowest IC₅₀ value of 0.80 ± 0.01 %. The EOCT followed with an IC₅₀ value of 0.86 ± 0.01 % and the EOAG had the highest IC₅₀ value of 0.99 ± 0.01 %. Thus, the essential oil treatment with the best antioxidant capacity was EOVL, followed by EOCT and the lowest was EOAG.

Antibacterial ability: The antimicrobial activities of perilla essential oils and extracts were shown in Table 7 and 8. Results demonstrated that concentration was proportional to the value of zone inhibition. The EOCT had higher activity than the

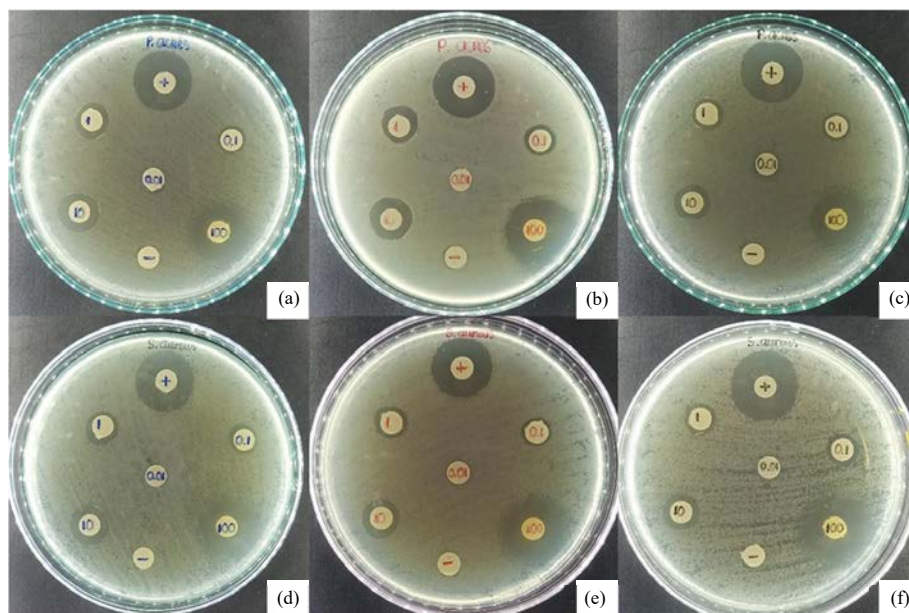


Fig. 1 (a-f): Antibacterial zones diameter of essential oils, (a-c) *Propionibacterium acnes*, (d-f) *Staphylococcus aureus*, (a, d) EOCT, (b, e) EOAG and (c, f) EOVL

other oils, 15.33 mm for *S. aureus* and 16.33 mm for *P. acnes* (Table 7 and Fig. 1(a-f)). This showed that the geographic conditions played an important role in the antibacterial activity of essential oils. The PfECT showed the highest antibacterial zone 10.67 mm for *E. coli* and 9.67 mm for *S. aureus*. The solvent DMSO used to dissolve the extracts and essential oils always gave negative results.

DISCUSSION

The concentration of the extract was proportional to the percentage of inhibition. In this experiment, perilla extracts and essential oils showed the ability to DPPH and ABTS radicals scavenging. According to a research on the ability to inhibit DPPH and ABTS radicals by Gaber *et al.*¹⁴, there was positive correlation between the antioxidant activity and the bioactive compound concentration in tested extract samples.

The PfECT had the strongest antioxidant activities that could be explained by the total polyphenol and flavonoid content. The PfECT was higher than the remaining treatments (12.78 mg GAE/g extract and 65.99 mg QE/g extract). Therefore, the content of total polyphenols and total flavonoids were an important indicator to evaluate the antioxidant capacity of a raw material. Besides, *Perilla frutescens* contain a large amount of anthocyanin, 44.66 units/g of fresh weight and there were 22 anthocyanins

were identified in the leaves of *Perilla frutescens*⁵. Anthocyanins are glucosides made of glucose, galactose, combined with colored aglycones (anthocyanidin). Anthocyanins have absorption wavelengths in the visible range, maximum absorption at wavelengths of 510-540 nm. The anthocyanin molecule contains many conjugated double bonds and also contains hydroxyl groups at positions C-3 of ring C, C-3', -4' and -5' in ring B, which is strongly effective in capturing free radicals and the oxidation chain reaction¹¹.

The antioxidant effectiveness of an essential oil primarily stems from its major components. However, it could also result from the synergistic or antagonistic effects of minor components, along with potential interactions among the various compounds¹⁶. The essential oil of *Perilla frutescens* (L.) Britt contain perillaldehyde (7.01-82.12%), perilla ketone (75.09-97.69%), β -dehydro-elsholtzia ketone (67-79%), limonene (3.18-14.85%), shisofuran (7.71%), farnesene (Z, E, α) (1.03-6.86%), β -caryophyllene (1.28-6.88%) and trans-shisool (0.04-3.52%)¹⁷. The difference was explained by different ecological factors (cultivated conditions, soil, environment and weather) that determine the content and chemical composition of secondary metabolites, as a result, may lead to various biological activities.

These results were consistent with the study of Hao *et al.*¹⁸. The DPPH scavenging activity of perilla seed oil extracted by supercritical carbon dioxide had $IC_{50} = 7.01$ mg/mL. The DPPH scavenging activities of essential

oil from *Perilla frutescens* (L.) Britt obtained by ultrasonic-assisted hydro-distillation with natural deep eutectic solvents was 5 mg/mL¹⁹. The ABTS scavenging of purple perilla leaf extracts showed that the IC₅₀ value in 80% ethanol extract was 973.3 µg/mL, n-hexane (4.106 mg/mL), chloroform (2.760 mg/mL), ethyl acetate (320.0 mg/mL), n-butanol (728.3 mg/mL) and water (4.401 mg/mL)²⁰.

The perilla extracts and essential oils showed strong antibacterial activities. Antibacterial ability could be resulted from plant secondary metabolites such as alkaloids, flavonoids, tannins, etc., which were often present in plant extracts²¹. The antibacterial activity was attributed to the presence of bioactive components such as terpenoids, flavonoids, glycosides, alkaloids, quinine, phenol, tannins, saponins and coumarins²². According to research by Shang *et al.*²³ about the antimicrobial parameters of flavonoids from *P. frutescens* extract. The overall flavonoid compounds exhibited inhibitory effects against *E. coli* (7.48±0.80 mm), indicating that variations in bacteriostatic activity could be attributed to the presence of effective chemical compositions such as apigenin 7-O-caffeoylglucoside and scutellarein 7-O-diglucuronide. Besides, chlorogenic acid, homogentisic acid, myricetin, benzoic acid and naringenin are the dominant phenolic compounds in perilla leaves extract²⁴. Phenolic compounds can activate extracellular enzymes, thereby changing membrane permeability. Flavonoids have a strong antibacterial activity including mechanisms such as inhibition of nucleic acid synthesis, inhibition of cytoplasmic membrane function and inhibition of energy metabolism²⁵. The flavonoid group strongly inhibited *E. coli* bacteria through its ability to affect DNA gyrase²⁶. Chen *et al.*¹⁹ also indicated the potential of flavonoids from perilla leaves extract against *S. aureus* (15.47±0.35 mm) at the concentration 200 µg/mL.

These results of essential oils were consistent with the study on the effect of essential oil concentration on the inhibition of *S. aureus*²⁷. The antibacterial activity of biocomposite chitosan film containing *Perilla frutescens* L. Britt essential oil using the agar disc diffusion method. The inhibition zone diameter of the films with 1% oil was 20.153±0.006 mm and pure 1% oil was 19.697±0.031 mm for *S. aureus*. These results demonstrated that essential oils were an important antibacterial ingredient in this compound. In addition, the number of studies on *P. acnes* inhibitory activity has been limited. This study demonstrated that *Perilla frutescens* essential oil was a potential compound in the field of skincare cosmetics²⁸.

The hydrophobicity of essential oils was responsible for the disruption of bacterial cell structure. The selectively permeable membrane of the cell was required for numerous

molecules across the membranes, controlling metabolism and maintaining osmotic pressure. In reality, the mechanism of antibacterial activity of essential oils includes cell wall disintegration, such as phospholipid layer destabilization²⁹ and increased permeability that destroys the function and composition of the plasma membrane³⁰. The antibacterial ability of perilla essential oils was attributed to the perillaldehyde component which was a natural compound found abundantly in the herb perilla. Perillaldehyde was a volatile compound with potent antimicrobial³¹, antioxidants³² and insoluble in water³³. It was investigated for potential hypolipidemic, anti-inflammatory, neuroprotective, antidepressant-like and antifungal effects. Besides, the antibacterial activity was impacted by the interactions of a variety of compounds such as limonene, linalool and caryophyllene³⁴.

CONCLUSION

The volatile compounds isolated from essential oils of *P. frutescens* have been studied qualitatively and quantitatively, as well as TPC and TFC. There are a total of 38 volatile compounds were identified. The major predominant compounds detected in essential oils were perillaldehyde, 2-Furyl n-pentyl ketone, β-Caryophyllene, β-Linalool, valeric acid pent-2-en-4-ynyl ester and limonene in variable levels. All extracts and essential oils of *P. frutescens* leaves have revealed potent antioxidants and antibacterial activities. These results of the study will provide a database for further research on perilla and the foundation for the perilla application in medicinal herbs and cosmetics production.

SIGNIFICANCE STATEMENT

Perilla frutescens is a type of vegetables in many countries. It is sometimes used as a herbal tea. Therefore, we would like to evaluate the antibacterial and antioxidant activities which are beneficial to human health. Essential oils contain a total of 38 volatile compounds. The major predominant compounds were perillaldehyde, 2-Furyl n-pentyl ketone, β-Caryophyllene, β-Linalool, valeric acid pent-2-en-4-ynyl ester and limonene. The tested extracts and essential oils of *Perilla frutescens* leaves have revealed potent antioxidants and antibacterial capacities.

ACKNOWLEDGMENT

This study was funded in part by the Can Tho University, Code: TSV2023-180.

REFERENCES

- Asif, M., 2012. Phytochemical study of polyphenols in *Perilla frutescens* as an antioxidant. *Avicenna J. Phytomed.*, 2: 169-178.
- Yu, H., J.F. Qiu, L.J. Ma, Y.J. Hu, P. Li and J.B. Wan, 2017. Phytochemical and phytopharmacological review of *Perilla frutescens* L. (Labiatae), a traditional edible-medicinal herb in China. *Food Chem. Toxicol.*, 108: 375-391.
- Bachheti, R.K., A. Joshi and T. Ahmed, 2014. A phytopharmacological overview on *Perilla frutescens*. *Int. J. Pharm. Sci. Rev. Res.*, 26: 55-61.
- Tian, J., X. Zeng, S. Zhang, Y. Wang, P. Zhang, A. Lü and X. Peng, 2014. Regional variation in components and antioxidant and antifungal activities of *Perilla frutescens* essential oils in China. *Ind. Crops Prod.*, 59: 69-79.
- Ghimire, B.K., J.H. Yoo, C.Y. Yu and I.M. Chung, 2017. GC-MS analysis of volatile compounds of *Perilla frutescens* Britton var. *Japonica* accessions: Morphological and seasonal variability. *Asian Pac. J. Trop. Med.*, 10: 643-651.
- Ahmed, H.M., 2019. Ethnomedicinal, phytochemical and pharmacological investigations of *Perilla frutescens* (L.) Britt. *Molecules*, Vol. 24. 10.3390/molecules24010102.
- Ahmed, H.M. and A.M.A. Al-Zubaidy, 2020. Exploring natural essential oil components and antibacterial activity of solvent extracts from twelve *Perilla frutescens* L. Genotypes. *Arabian J. Chem.*, 13: 7390-7402.
- Ahmed, H.M. and S. Tavaszi-Sarosi, 2019. Identification and quantification of essential oil content and composition, total polyphenols and antioxidant capacity of *Perilla frutescens* (L.) Britt. *Food Chem.*, 275: 730-738.
- Otang, W.M., D.S. Grierson and R.N. Ndip, 2012. Phytochemical studies and antioxidant activity of two South African medicinal plants traditionally used for the management of opportunistic fungal infections in HIV/AIDS patients. *BMC Complementary Altern. Med.*, Vol. 12. 10.1186/1472-6882-12-43.
- Djeridane, A., M. Yousfi, B. Nadjemi, D. Boutassouna, P. Stocker and N. Vidal, 2006. Antioxidant activity of some Algerian medicinal plants extracts containing phenolic compounds. *Food Chem.*, 97: 654-660.
- Phuyal, N., P.K. Jha, P.P. Raturi and S. Rajbhandary, 2020. Total phenolic, flavonoid contents, and antioxidant activities of fruit, seed, and bark extracts of *Zanthoxylum armatum* DC. *Sci. World J.*, Vol. 2020. 10.1155/2020/8780704.
- Re, R., N. Pellegrini, A. Proteggente, A. Pannala, M. Yang and C. Rice-Evans, 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biol. Med.*, 26: 1231-1237.
- Mahesh, B. and S. Satish, 2008. Antimicrobial activity of some important medicinal plant against plant and human pathogens. *World J. Agric. Sci.*, 4: 839-843.
- Gaber, N.B., S.I. El-Dahy and E.A. Shalaby, 2023. Comparison of ABTS, DPPH, permanganate, and methylene blue assays for determining antioxidant potential of successive extracts from pomegranate and guava residues. *Biomass Convers. Biorefin.* 13: 4011-4020.
- Jiang, T., K. Guo, L. Liu, W. Tian, X. Xie, S. Wen and C. Wen, 2020. Integrated transcriptomic and metabolomic data reveal the flavonoid biosynthesis metabolic pathway in *Perilla frutescens* (L.) leaves. *Sci. Rep.*, Vol. 10. 10.1038/s41598-020-73274-y.
- Wu, Z., H. Li, Y. Yang, Y. Zhan and D. Tu, 2013. Variation in the components and antioxidant activity of *Citrus medica* L. var. *sarcodactylis* essential oils at different stages of maturity. *Ind. Crops Prod.*, 46: 311-316.
- Ahmed, H.M., A.M. Al-Zubaidy and G. Othman-Qadir, 2022. Biological investigations on macro-morphological characteristics, polyphenolic acids, antioxidant activity of *Perilla frutescens* (L.) Britt. grown under open field. *Saudi J. Biol. Sci.*, 29: 3213-3222.
- Hao, L., C. Lv, X. Cui, F. Yi and C. Su, 2021. Study on biological activity of perilla seed oil extracted by supercritical carbon dioxide. *LWT*, Vol. 146. 10.1016/j.lwt.2021.111457.
- Chen, Z., K. Wu, W. Zhu, Y. Wang, C. Su and F. Yi, 2022. Chemical compositions and bioactivities of essential oil from perilla leaf (*Perillae folium*) obtained by ultrasonic-assisted hydro-distillation with natural deep eutectic solvents. *Food Chem.*, Vol. 375. 10.1016/j.foodchem.2021.131834.
- Jun, H.I., B.T. Kim, G.S. Song and Y.S. Kim, 2014. Structural characterization of phenolic antioxidants from purple perilla (*Perilla frutescens* var. *acuta*) leaves. *Food Chem.*, 148: 367-372.
- Mythili, T. and R. Ravindhran, 2012. Phytochemical screening and antimicrobial activity of *Sesbania sesban* (L.) Merr. *Asian J. Pharm. Clin. Res.*, 5: 179-182.
- Kaur, P., N. Kumar, T.N. Shivananda and G. Kaur, 2011. Phytochemical screening and antimicrobial activity of the plant extracts of *Mimosa pudica* L. against selected microbes. *J. Med. Plants Res.*, 5: 5356-5359.
- Shang, X., M. Zhang, J. Hu, Y. Zhang, L. Yang and X. Hou, 2023. Chemical compositions, extraction optimizations, and *in vitro* bioactivities of flavonoids from perilla leaves (*Perillae folium*) by microwave-assisted natural deep eutectic solvents. *Antioxidants*, Vol. 12. 10.3390/antiox12010104.
- Ghimire, B.K., J.H. Yoo, C.Y. Yu, S.H. Kim and I.M. Chung, 2019. Profiling volatile and phenolic compound composition and characterization of the morphological and biological activities of *Perilla frutescens* Britton var. *Japonica* accessions. *Acta Physiol. Plant.*, Vol. 41. 10.1007/s11738-019-2890-1.
- Cushnie, T.P.T. and A.J. Lamb, 2005. Antimicrobial activity of flavonoids. *Int. J. Antimicrob. Agents*, 26: 343-356.
- Wu, T., X. Zang, M. He, S. Pan and X. Xu, 2013. Structure-activity relationship of flavonoids on their anti-*Escherichia coli* activity and inhibition of DNA gyrase. *J. Agric. Food Chem.*, 61: 8185-8190.

27. He, W., C. Shi, X. Long, X. Liu and X. Zhao, 2020. Antimicrobial activity and mechanism of action of perilla essential oil against *Staphylococcus aureus*. E3S Web Conf., Vol. 145. 10.1051/e3sconf/202014501015.
28. Zhang, Z.J., N. Li, H.Z. Li, X.J. Li, J.M. Cao, G.P. Zhang and D.L. He, 2018. Preparation and characterization of biocomposite chitosan film containing *Perilla frutescens* (L.) Britt. essential oil. Ind. Crops Prod., 112: 660-667.
29. Gill, A.O. and R.A. Holley, 2006. Disruption of *Escherichia coli*, *Listeria monocytogenes* and *Lactobacillus sakei* cellular membranes by plant oil aromatics. Int. J. Food Microbiol., 108: 1-9.
30. Juven, B.J., J. Kanner, F. Schved and H. Weisslowicz, 1994. Factors that interact with the antibacterial action of thyme essential oil and its active constituents. J. Appl. Bacteriol., 76: 626-631.
31. Tian, J., Y. Wang, Z. Lu, C. Sun, M. Zhang, A. Zhu and X. Peng, 2016. Perillaldehyde, a promising antifungal agent used in food preservation, triggers apoptosis through a metacaspase-dependent pathway in *Aspergillus flavus*. J. Agric. Food Chem., 64: 7404-7413.
32. Omari-Siaw, E., Q. Wang, C. Sun, Z. Gu and Y. Zhu *et al.*, 2016. Tissue distribution and enhanced *in vivo* anti-hyperlipidemic-antioxidant effects of perillaldehyde-loaded liposomal nanoformulation against Poloxamer 407-induced hyperlipidemia. Int. J. Pharm., 513: 68-77.
33. Chu, Z.Y., Y.L. Li, L. Wang, S.Y. Wei, S.Q. Yang and H. Zeng, 2024. Perillaldehyde: A promising antibacterial agent for the treatment of pneumonia caused by *Acinetobacter baumannii* infection. Int. Immunopharmacol., Vol. 126. 10.1016/j.intimp.2023.111311.
34. Deba, F., T.D. Xuan, M. Yasuda and S. Tawatu, 2008. Chemical composition and antioxidant, antibacterial and antifungal activities of the essential oils from *Bidens pilosa* Linn. var. *Radiata*. Food Control, 19: 346-352.