## Determination of some bioactive activities of a mixture of perilla and coriander essential oils

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### Abstract

The mixture of perilla and coriander essential oils is slowly mixed with essential oil perilla and essential oil coriander at a 1:1 (v/v) ratio. The mixture of perilla and coriander essential oils has a light yellow color, a characteristic aroma, and is transparent. The goal of this study was to determine some biological activities of the essential oil mixture perilla and coriander, laying the groundwork for the use of this essential oil mixture in food processing and preservation. The antioxidant capacity of the mixture of perilla and coriander essential oils has been determined to be 68.16±0.07%. The antibacterial activity against Bacillus mesentericus was the highest. This was followed by strains from different species of microorganisms: Escherichia coli, Micrococcus luteus, Salmonella enterica, Streptococcus pneumonia, Staphylococcus aureus, Bacillus cereus, Lactobacillus acidophilus and Bacillus subtilis with diameters of 44.34±0.16 mm, 42.17±0.28 mm, 41.32±0.19 mm, 39.86±0.21 mm, 39.62±0.15 mm, 38.45±0.27 mm, 37.54±0.13 mm and 37.23±0.18 mm, respectively. The essential oils perilla and coriander have a strong inhibitory effect on alpha-glucosidase. Their IC<sub>50</sub> values were 0.79 $\pm$ 0.02 µg/mL (the mixture of essential oil perilla and coriander),  $0.84\pm0.02 \ \mu g/mL$  (essential oil perilla) and  $0.91\pm0.03$  µg/mL (essential oil coriander), while the IC<sub>50</sub> value of the positive control acarbose was 145.32±9.24 µg/mL. Moreover, all samples have stronger alpha-amylase inhibition than alpha-glucosidase. The mixture of essential oils perilla and coriander, essential oil perilla and essential oil coriander exhibited strong inhibitory activity against alpha-amylase with IC<sub>50</sub> values of  $0.18\pm0.01$ ,  $0.23\pm0.01$  and  $0.26\pm0.02$  µg/mL, respectively. These research results provide a good basis for later processing and preservation.

### 1. Introduction

Essential oils have an important role in many areas of life, such as food, cosmetics and pharmaceuticals. Essential oils have a characteristic, attractive aroma. Adding essential oils to food not only contributes to creating a characteristic, and attractive aroma for food, but essential oils also have the effect of preserving food due to their ability to preserve food and their antioxidant and antibacterial properties. Perilla and coriander are grown a lot in Vietnam. These two vegetables, currently grown in Vietnam, are mainly used to eat raw or as food seasoning. Research and production of essential oils from perilla and coriander are modest. Essential oil perilla is extracted and recovered from the perilla.

Essential oil perilla contains the chemical components perillene, 1-octen-3-ol,  $\alpha$ -copaene, benzaldehyde, linalool,  $\beta$ -caryophyllene, perillaketone, and spathulenol (Baser et al., 2003). Essential oil coriander is extracted and recovered from the coriander. Essential oil coriander chemical components contains such as gthionodecalactone, 1,2-decanediol, n-cetyl alcohol, 2,4dimethylheptane, 2-decenoic acid, 2-dodecanal, acaryophyllene, α-pinene, capric acid, nonanoic acid, nonanol, octanoic acid and oleic acid (Bhuiyan et al., 2009).

According to some studies, the essential oils perilla and coriander have biological activities as well as high antioxidant capacities (Shyamapada *et al.*, 2015; Linyu **RESEARCH PAPER** 

et al., 2021) and high antibacterial ability (Shyamapada et al., 2015; Li-Yun et al., 2016; Ahmed et al., 2020). That is very important when adding these essential oils to food. There are currently very few studies on the biological activity of a mixture of essential oils, particularly perilla and coriander. Therefore, the purpose of this study is to determine some biological activities of the mixture of essential oils perilla and coriander, thereby forming the basis for the aroma combination between essential oils perilla and coriander. As a result, the mixture of essential oils perilla and coriander can be used more effectively in food processing and preservation. Therefore, the research to determine some biological activities of a mixture of essential oils perilla and coriander is both scientific and practical in nature.

### 2. Materials and methods

### 2.1 Materials

Steam distillation after drying with Na<sub>2</sub>SO<sub>4</sub> yielded the essential oils perilla and coriander Na<sub>2</sub>SO<sub>4</sub>. The sample was stored in the Department of Food Science and Technology, University of Science, Vietnam National University, Hanoi.

The bacterial strains tested (*Bacillus mesentericus*, *Escherichia coli, Micrococcus luteus, Salmonella enteric, Streptococcus pneumonia, Staphylococcus aureus, Bacillus cereus, Lactobacillus acidophilus*, and *Bacillus subtilis*) were obtained from the Institute for Quality Testing and Inspection, which is located at number 7, niche 168/21, line Nguyen Xien, Ha Dinh, and Thanh Xuan, Hanoi, Vietnam.

### 2.2 Method of making the mixture of perilla and coriander essential oils

The mixture of perilla and coriander essential oils was mixed in a ratio of 1:1 (v/v). From this mixture, 20 mL was aspirated into a 100 mL conical flask. Next, 20 mL of coriander essential oil was added to the conical flask. The conical flask was sealed, thoroughly shaken, and then placed in the magnetic stirrer. The solution was stirred at 150 rpm for 2 mins. The perilla and coriander essential oils were then poured into a dark glass bottle with a volume of 50 mL, closed tightly, and stored frozen (Loi *et al.*, 2017).

### 2.3 Determination of the antioxidant activity of a mixture of perilla and coriander essential oils

The free radical scavenging activity of a mixture of perilla and coriander essential oils was measured using 1,1-diphenyl-2-picrylhydrazol (DPPH). A 0.5 mM solution of DPPH in methanol and a 0.005 M acetate buffer (pH 5.5) were prepared. An aliquot of 0.1 mL of

the essential oil solution was added to the tube containing 2 mL of acetate buffer, 1.9 mL of methanol, and 1 mL of DPPH solution. In the blank tube, DPPH was removed; in the control tube, 1 mL of DPPH was added to the tube containing 2 mL acetate buffer and 2 mL methanol. The mixture was shaken immediately after adding DPPH and allowed to stand at room temperature in the dark. The decrease in absorbance at 517 nm was measured after 32 mins until the reaction reached a plateau. Vitamin E with a concentration of 0.5 mM was used as a positive control, and its free radical scavenging activity was performed in parallel in the same experiment. These experiments were run in duplicate.

The inhibitory percentage of DPPH was calculated as follows:

Scavenging effect (%) =  $[(A_o - (A - A_b)) / A_o] \times 100\%$ 

Wherein  $A_o$  is the value of absorbance of the control at the wavelength of 517 nm, A is the value of absorbance of the sample at the wavelength of 517 nm, and  $A_b$  is the value of absorbance of the blank at the wavelength of 517 nm (Choi *et al.*, 2000; Teresita *et al.*, 2000; Miroslava *et al.*, 2020; Kumar *et al.*, 2022).

### 2.4 Determination of the antibacterial ability of the mixture of essential oils of perilla and coriander

Antibacterial activity was determined by the agar diffusion method. A 50  $\mu$ L mixture of the essential oils perilla and coriander was placed in wells on plates containing the bacterial strains tested. The activity was estimated by the diameter of the antibacterial round (mm), which was calculated by the formula D-d(mm), wherein D was the diameter of the antibacterial round (mm) and d was the hole diameter (mm) (Cui *et al.*, 2017).

2.5 Determination of the alpha-glucosidase and alphaamylase inhibitory activities of a mixture of perilla and coriander essential oils

2.5.1 Determination of the alpha-glucosidase inhibitory activity of a mixture of perilla and coriander essential oils

2.5 mM p-nitrophenyl- $\alpha$ -D-glucopyranoside, 250 µL of products (the concentrations were varied from 1-1000 µM) in dimethyl sulfoxide, and 0.3 U/mL alphaglucosidase in phosphate buffer, pH 6.9. Control tubes contained only dimethyl sulfoxide, enzyme, and substrate, whereas acarbose replaced the product solution in positive controls. The absorbance of the resulting pnitrophenol was determined at 405 nm and was considered directly proportional to the activity of the enzyme. The products were tested for alpha-glucosidase inhibition activity at different concentrations (0.01-0.015)  $\mu$ g/mL). Each sample generation was done in triplicate. Inhibition percentages by-products and acarbose were Alpha – glucosidase inhibition activity (%) =  $\frac{A_{405 \text{ (control)}} - A_{405 \text{ (essential oil)}}}{A_{405 \text{ (control)}}} \times 100$ calculated using the following equation:

Where:  $A_{405 \text{ (control)}}$  is the absorbance of the control at wavelength 405 nm. At wavelength 405 nm,  $A_{405 \text{ (essential}}$ oil) is the absorbance of the mixture of essential oil perilla and coriander, essential oil perilla and essential oil coriander. The IC<sub>50</sub>, which is the concentration of the sample required to inhibit 50% of the enzyme, was determined for each sample. All products were compared on the basis of their IC<sub>50</sub> values, estimated from the dose -response curves. Different concentrations of plant extracts range from 10 µg/mL to 100 µg/mL (Hichri *et al.*, 2019; Ojaha *et al.*, 2020).

# 2.5.2 Determination of the alpha-amylase inhibitory activity of a mixture of perilla and coriander essential oils

The alpha-amylase inhibitory activity of a mixture of perilla and coriander essential oils was determined using the assay described by Hichiri et al. (2019). In dimethyl sulfoxide, different concentrations of the essential oils perilla and coriander were prepared, ranging from 10 to 100 g/mL. The mixture of essential oils perilla and coriander, essential oils perilla and coriander (250µl) of each concentration, and 250 µl of alpha-amylase isolated from Aspergillus oryzae (Sigma-Aldrich) (0.1U/mL) were taken and incubated at 25°C for 10 mins. After the pre-incubation, 250 µl of 1% starch solution in 0.02 M sodium phosphate buffer (pH 6.9) was added to each tube at regular time intervals. After the incubation at 25°C for 30 minutes, the reaction was stopped with 0.1 mL of dinitrosalicylic acid reagent. The test tubes were then incubated in a boiling water bath for 5 minutes and cooled to room temperature. The reaction mixture was then diluted with the addition of 5 mL of distilled water, and the absorbance was measured at 540 nm. The readings were compared with the control, which contains dimethyl sulfoxide and buffer instead of sample extract. In the positive test, the acarbose replaces the extract. The % inhibition activity of a mixture of perilla and coriander essential oils was calculated using the following formula:

 $\label{eq:alpha} Alpha - amylase inhibition activity \ (\%) = \frac{A_{540 \ (control)} - A_{540 \ (essential \ oil)}}{A_{540 \ (control)}} \times 100$ 

Where  $A_{540 \text{ (control)}}$  is the absorbance of the control at wavelength 540nm and  $A_{540 \text{ (essential oil)}}$  is the absorbance of a mixture of perilla and coriander essential oils at 540 nm (Hichri *et al.*, 2019; Ojaha *et al.*, 2020).

### 3. Results and discussion

3.1 The antioxidant activity of the mixture of perilla and coriander essential oils

Antioxidant activity is an important parameter to evaluate the quality of the mixture of perilla and coriander essential oils. The results of determining the antioxidant activity of the mixture of perilla and coriander essential oils are shown in Table 1.

Table 1. The antioxidant activity of the mixture of perilla and coriander essential oils.

Tested substances	Result (%)
The mixture of perilla and coriander essential oils	68.16±0.07
Ascorbic acid	56.24±0.03

The DPPH free radical scavenging activity of the mixture of essential oils perilla and coriander was  $68.16\pm0.07\%$ , and this value was a bit higher than that of ascorbic acid at 56.24±0.03%. Compared with the research results of Miroslava et al. (2020), who reported that free radical resistance of essential oil coriander was 51.05%, the mixture of essential oils perilla and coriander was more resistant to free radicals than essential oil coriander. Therefore, it can be seen that the combination of two essential oils, perilla and coriander, has a synergistic effect, increasing the anti -free radical effect.

### 3.2 The antibacterial activity of the mixture of perilla and coriander essential oils

The agar diffusion method was used to estimate the antibacterial potentials of the mixture of perilla and coriander essential oils. Tested microorganisms used in this experiment were *Bacillus mesentericus, Escherichia coli, Micrococcus luteus, Salmonella enterica, Streptococcus pneumonia, Staphylococcus aureus, Bacillus cereus, Lactobacillus acidophilus* and *Bacillus subtilis.* Table 2 shows the diameters of the antibacterial activity rounds of a mixture of perilla and coriander essential oils against these bacteria.

Table 2. The antibacterial activity diameters of the mixture of perilla and coriander essential oils.

Tested microorganisms	Diameter of an antibacterial	
	round (mm)	
Bacillus mesentericus	45.28±0.23	
Escherichia coli	44.34±0.16	
Micrococcus luteus	$42.17 \pm 0.28$	
Salmonella enterica	41.32±0.19	
Streptococcus pneumoniae	39.86±0.21	
Staphylococcus aureus	39.62±0.15	
Bacillus cereus	38.45±0.27	
Lactobacillus acidophilus	37.54±0.13	
Bacillus subtilis	37.23±0.18	

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The results showed that the mixture of perilla and coriander essential oils possessed antibacterial activity against all microorganisms tested. Among them, the antibacterial activity against Bacillus mesentericus was the highest. This was followed by strains of different microorganisms: Escherichia species of coli, Micrococcus luteus, Salmonella enterica, Streptococcus pneumonia, Staphylococcus aureus, Bacillus cereus, Lactobacillus acidophilus, and Bacillus subtilis. With antibacterial circle diameters of 44.34±0.16 mm, 42.17±0.28 mm, 41.32±0.19 mm, 39.86±0.21 mm, 39.62±0.15 mm, 38.45±0.27 mm, 37.54±0.13 mm and 37.23±0.18 mm, respectively. In this study, the antibacterial activity of the mixture of essential oils perilla and coriander was comparable to that of the mixture of essential oils peel lime and orange against test strains of microorganisms Escherichia coli and Staphylococcus aureus (Loi et al., 2017). This research result is also consistent with the research results of (Shyamapada et al., 2015; Li-Yun et al., 2016; Ahmed et al., 2020).

### 3.3 Determine the alpha-glucosidase and alpha-amylase inhibition activities of a mixture of perilla and coriander essential oils

It is critical to determine the alpha-glucosidase and alpha-amylase inhibition activities of the mixture of essential oils perilla and coriander. This is the scientific foundation for using the essential oils perilla and coriander in food processing and preservation. The inhibitory ability of the mixture of essential oils perilla and coriander on alpha-glucosidase and alpha-amylase was investigated in this study, and the results are shown in Table 3.

The effectiveness of the alpha-amylase and alphaglucosidase inhibitors of the mixture of perilla and coriander essential oils was compared using their IC<sub>50</sub> values, as shown in Table 3. High values of IC<sub>50</sub> indicate low inhibitory activity. The essential oils perilla and coriander have a strong inhibitory effect on alphaglucosidase. Their IC<sub>50</sub> values were 0.79±0.02 µg/mL (the mixture of perilla and coriander), 0.84±0.02 µg/mL (essential oil perilla), and 0.91±0.03 µg/mL (essential oil coriander), while the IC<sub>50</sub> value of the positive control acarbose was 145.32±9.24 µg/mL. Moreover, all samples have stronger alpha-amylase inhibition than alpha-glucosidase. The mixture of essential oil perilla essential oils exhibited strong inhibitory activity against alpha-amylase with IC<sub>50</sub> values of  $0.18\pm0.01$ ,  $0.23\pm0.01$  and  $0.26\pm0.02$  µg/mL, respectively. All the mentioned oils showed stronger inhibition activity in comparison with the reference drug, acarbose (IC<sub>50</sub> =  $81.56\pm1.03$  µg/mL).

### 4. Conclusion

Research results have determined the antioxidant capacity of the mixture of perilla and coriander essential oils, with the highest activity against Bacillus mesentericus and the largest antibacterial ring diameter. The mixture of perilla and coriander essential oils has alpha-glucosidase inhibitory strong activity, and all samples were more potent inhibitors of alpha-amylase than alpha-glucosidase. The mixture of perilla and coriander essential oils, perilla essential oil, and coriander essential oil all exhibited strong alpha-amylase inhibitory activity with corresponding IC<sub>50</sub> values. Thus, the combination of two essential oils perilla and coriander has a synergistic effect, increasing the anti-free radical effect.

### **Conflict of interest**

The author declares the research results in this article to be completely honest. The data has never been used or rotated from other research projects in any form.

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Table 3. Determine the alpha-glucosidase and alpha-amylase inhibition activities of a mixture of perilla and coriander essential oils.

Experimental samples	IC <sub>50</sub> (μg/mL)	IC <sub>50</sub> (µg/mL)
	Alpha-glucosidase inhibition	Alpha-amylase inhibition
Mixture of perilla and coriander essential oils	$0.79{\pm}0.02$	0.18±0.01
Perilla essential oil	$0.84{\pm}0.02$	0.23±0.01
Coriander essential oil	0.91±0.03	$0.26{\pm}0.02$
Acarbose	145.32±9.24	81.56±1.03

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