

Thành phần hóa học của tinh dầu từ lá của loài hương nhu trắng và hoạt tính sinh học

TÓM TẮT

Ocimum gratissimum L. là một loài thực vật nhỏ thuộc chi *Ocimum* thuộc họ Lamiaceae. Loài này đã được nghiên cứu về hóa thực vật và hoạt tính sinh học. Trong nghiên cứu này, tinh dầu từ lá của loài *O. gratissimum* được chiết xuất bằng phương pháp chiết có hỗ trợ vi sóng. Các thành phần hóa học của dầu được xác định bằng phương pháp sắc ký khí - khối phổ (GC-MS). Dựa trên kết quả phân tích, 222 hợp chất đã được định danh. Thêm vào đó, tinh dầu đã chứng minh đặc tính kháng khuẩn đáng kể đối với khuẩn *Escherichia coli*, với vùng ức chế vi khuẩn (ZOI) trung bình là 12,4 mm ở thể tích 50 μ L. Những kết quả này cho thấy tinh dầu *O. gratissimum* là một chất kháng khuẩn tự nhiên, khiến nó trở thành một yếu tố triển vọng cho các ứng dụng sau thu hoạch và bảo quản thực phẩm.

Từ khóa: *Ocimum gratissimum*, essential oil, eugenol; antibacterial activity.

Chemical components of essential oil from leaves of *Ocimum gratissimum* and bioactivity

ABSTRACT

Ocimum gratissimum L. is a small plant belonging to *Ocimum* genus of the Lamiaceae family. It has been investigated about phytochemical and bioactivities. In this study, essential oil from leaves of *O. gratissimum* specie was extracted by microwave-assisted extraction methods. The chemical components of the oil were determined using gas chromatography - mass spectrometry (GC-MS) analysis. Basing on analytical result, two hundred and twenty-two compounds were identified. Furthermore, the essential oil demonstrated significant antibacterial properties against *Escherichia coli*, with an average zone of microbial inhibition (ZOI) of 12.4 mm at a volume of 50 μ L. These results showed that essential oil of *O. gratissimum* as a natural antibacterial agent, making it a promising factor for post-harvest and food preservation applications.

Keywords: *Ocimum gratissimum*, essential oil, eugenol, antibacterial activity.

1. INTRODUCTION

Medicinal plants have been used for thousands of years with various applications. They have been applied for treatments of various diseases due to their with diseases due to bioactivities. Among these medicinal agents are essential oils (EOs) ¹⁻² which is used as main flavorings in the food and pharmaceutical industries. ³⁻⁵ They possess a wide range of bioactivities, including antioxidant, antibacterial, antifungal, anticancer, anti-inflammatory, and immunomodulatory activities. ⁵⁻⁹ EOs are typically extracted through methods such as steam distillation, hydrodistillation, and other advanced distillation techniques. ¹⁰⁻¹¹ Up to now, there are approximately 3000 known Eos. ¹² They are extracted from different plant parts such as flowers, leaves, stems, roots, fruits and fruit-peels. ¹ While EO-containing plants belong to around 60 families, but only in some popular families include Alliaceae, Apiaceae, Asteraceae, Lamiaceae, Myrtaceae, Poaceae and Rutaceae. ¹³⁻¹⁴ *Ocimum gratissimum* L., is a small plant belonging to *Ocimum* genus in the Lamiaceae family. It is widely distributed across Africa, Asia, and South America. ¹⁵⁻¹⁶ In traditional medicine, it is used to treat diseases as cough, pneumonia, inflammation, anaemia, diarrhea, pains, and fungal and bacterial infections. ¹⁶ Up to now, numerous studies have published its bioactivities, such as anti-hyperglycaemic ¹⁷⁻¹⁸, hypoglycaemic ¹⁹, anti-inflammatory ²⁰, anti-diarrhoeal ²¹ anti-anaemic, hepatoprotective ¹⁶, anti-hypertensive ²², antibacterial activities. ²³

2. EXPERIMENTAL

2.1. Material

The leaves of *O. gratissimum* L. were collected in November 2023 in Kbang town, Gia Lai province, Viet Nam. The collected leaves were washed with water to remove any impurities. Next, the leaves were ground into powder using a blender. The powder was then packed in polyethylene bags under vacuum conditions and stored in a cool refrigerator compartment until needed.

2.2. Extraction of essential oils by microwave-assisted extraction

200 g fresh leaves were placed into a beaker, then added 600 ml deionized water. Next, beaker was put in ultrasonic bath for vibrating in thirty minutes. The resultant solution was transferred into a 500 ml round-bottom flask. Then, it was put in microwave oven. The extraction progress of essential oil was carried out using a Clevenger distillation system with supported by microwave oven. The oven was operated at a power of 480 W for one hour and forty-five minutes. Post-extraction, the essential oil was condensed and separated from water by anhydrous sodium sulfate.

2.3. GC-MS analysis

Essential oil was analyzed by Agilent GC 8890-MS 7010B triple quadrupole (USA) equipped with

a HP-5ms Ultra Inert column (30 m x 250 µm x 0.25 µm). The analytical parameters were set as follows: the carrier gas (He) with a flow rate of 0.9999 mL/min; the split ratio, 5:1; injection volume, 0.1 µL; injection temperature, 250 °C; oven temperature, progressing from 40 to 320°C at a rate of 5 °C·min⁻¹ and from 40 to 300 °C at a rate of 58 °C/min, and then hold at 300 °C for 5 min; and the ionization mode, electronic impact at 70 eV.

Compounds were identified by comparing their mass spectral fragmentation patterns to the standard library, with reference to the MS database (NIST 20 software, USA).

2.4. Determine antibacterial activity of essential oil by disc diffusion method

The antibacterial activity of the essential oil was assessed using the well-diffusion method²⁴ against the pathogenic microorganism *E. Coli*. Pure cultures of organisms were subcultured on sterilized Macconkey Agar for *E. Coli*. A cork drill was used to puncture gel and make 8 mm diameter wells. Then, 10 µL, 20 µL, 30 µL of essential oil were added into each well using a micropipette. After 24 h of incubation at 35°C for *E. Coli*, the zone of inhibition was calculated by measuring its diameter around each well to the nearest millimeter. The antibacterial activity was evaluated by determining the zone of microbial inhibition (ZOI) according to the formula:

$$\text{ZOI (mm)} = D - d$$

where D: diameter of inhibition zone (mm), d: diameter of the well (mm).

All experiments were carried out in triplicate.

3. RESULTS AND DISCUSSION

3.1. Results about extraction of essential oil

The essential oil was successfully extracted using a microwave-assisted hydrodistillation technique. The plant material was mixed with deionized water. Power consumption of microwave oven was 480 W, which is similar to high-average level of household microwave oven. The extraction time was one hour and forty-five minutes. The volume of essential oil obtained was 6 ml. This result was significant because progress of extraction

was shorter than other extraction methods like steam distillation and hydrodistillation. Source heat of microwave made distillation happened quickly due to closure of oven. Furthermore, ray of microwave made powder of leaves suspended in water easily. So, progress of distillation and extraction of essential oil were convenient.

3.2. Chemical component of essential oil

To identify the components of the isolated essential oil, a GC-MS analysis was conducted. The compounds found to be present in essential oil are reported in appendix. The results in reference 1 showed that, essential oil contained 227 compounds. Among these compounds, there were known antioxidant compounds as (-)-globulol, acetyl eugenol, eugenol, methyleugenol, etc... Eugenol, in particular, was found to be present in the essential oil with amount percent was 54.28 %. This compound possesses various bioactivities, such as antibacterial, antiviral, antifungal, anticancer, anti-inflammatory and antioxidant.²⁵ Additionally, derivatives of eugenol such as acetyl eugenol and methyleugenol have also demonstrated significant bioactivities. Acetyl eugenol is a phytochemical compound with ability against infectious diseases and tumors.²⁶ Methyl eugenol exhibit anti-quorum sensing activity.²⁷ Furthermore, terpene compounds were found in essential oil as squalene (23.99 %), supraene (31.16 %).

3.3. Biological activity

Table 1. Result about test antibacterial activity of EO to *E. Coli*

	Volume H ₂ O	Volume EO		
	H ₂ O	10µL	30µL	50µL
ZOI (mm)	0	9	13	14
	0	11	12	13
	0	13	14	13

The antibacterial ability of essential oil from leaves of *O. gratissimum* was evaluated against *E. Coli* to be presented in table 1. The result showed that the oil exhibited the highest inhibitory effect on *E. Coli* at a volume of 50 µL, with an average ZOI was 12.4 mm. As mentioned in section 3.2, the essential oil was particularly rich in eugenol and its derivatives such as acetyl eugenol, methyl eugenol, which have been recognized for their significant antibacterial properties.^{26, 28-29}

4. CONCLUSIONS

The essential oil was extracted from leaves of *O. gratissimum* using microwave-assisted hydrodistillation. The maximum volume of essential oil obtained from the extraction process was 6 ml. The GC-MS analysis revealed that the essential oil was rich in eugenol, constituting approximately 54.28% of the oil's composition. In addition, the essential oil exhibited significant antibacterial activity against *E. Coli* with ZOI is 12.4 mm at a volume of 50 µL. This result proved that the potential of the essential oil from *O. gratissimum* as a natural source for postharvest and food preservation purposes in the future.

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