PRELIMINARY STUDY ON THE ESSENTIAL OIL OF PALMAROSA CULTIVATED UNDER GACP-WHO GUIDELINES IN HANOI: CHEMICAL COMPOSITION AND ANTIBACTERIAL ACTIVITY

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Abstract:

Palmarosa (Cymbopogon martini) is a plant species with a high geraniol content, widely applied in the production of cosmetics, pharmaceuticals, and aromatherapy. In this study, palmarosa essential oil cultivated under GACP-WHO guidelines in Hanoi, was extracted and analyzed to determine its chemical composition and preliminarily evaluate its antibacterial activity. The essential oil was extracted by steam distillation, its chemical constituents were identified using gas chromatography–mass spectrometry (GC-MS), and antibacterial activity was tested against three bacterial strains-Escherichia coli, Staphylococcus aureus, and Bacillus subtilis-using the agar diffusion method. The results showed an extraction yield of 1.3252%, which was higher than that of the conventionally cultivated sample from Đak Lak (1.25%). The major constituents were geraniol (76.86%), geranyl acetate (13.69%), and linalool (3.43%). At a concentration of 10 µL/mL, the essential oil exhibited remarkable antibacterial activity, with inhibition zone diameters of 2.9 cm against E. coli, 2.3 cm against S. aureus, and 2.1 cm against B. subtilis after 48 hours. These findings confirm the potential application of palmarosa essential oil cultivated under GACP-WHO guidelines in the development of natural-based cosmeceutical products, aligning with the sustainable healthcare trend.

Keywords: *Cymbopogon martini; GACP-WHO; GC-MS; Antibacterial activity; Palmarosa; Essential oil.*

1. Introduction

In recent years, the pharmaceutical and cosmetic industries have shown a marked shift toward the use of natural raw materials. Consumers increasingly demand not only efficacy but also safety and environmental sustainability in personal care products. Within this context, plant-derived essential oils have emerged as a promising group of bioactive ingredients, particularly those rich in monoterpenoids with well-documented biological activities such as antibacterial, anti-inflammatory, and antioxidant effects. Due to these properties, essential oils are extensively utilized in cosmetics, pharmaceuticals, and aromatherapy, while also becoming a focal point of global research aimed at developing natural, safe, and sustainable products with significant therapeutic and commercial value (Bakkali et al., 2008).

Palmarosa (*Cymbopogon martini* (Roxb.) W. Watson), belonging to the family Poaceae Barnhart, originates from India, where it has long been used both in Ayurvedic traditional medicine and in essential oil production. Botanically, palmarosa is a perennial tufted grass that can grow up to 1.5 m in height, characterized by pale pink to reddish stems, glabrous leaf sheaths, long narrow leaves with a smooth texture, and a mild fragrance reminiscent of rose when crushed. Its inflorescences are small, paniculate, and turn dark red upon drying. In Vietnam, palmarosa was first introduced for experimental cultivation in 1982 in the Central Highlands (Dak Lak), and later expanded to other regions owing to its

economic value and adaptability to diverse ecological conditions. Importantly, the aerial parts of the plant (stems, leaves, and flowers) are the main sources of essential oil, which is particularly rich in geraniol—a compound of high commercial value due to its strong antibacterial, antifungal, and antioxidant properties (Rao et al., 2005). Beyond its medicinal applications, palmarosa essential oil is widely employed in perfumery, skincare products, soap manufacturing, and food preservation (Hussain et al., 2008). The Good Agricultural and Collection Practices (GACP) guidelines established by the World Health Organization (WHO) provide a framework to ensure quality, safety, and sustainability in the cultivation and harvesting of medicinal plants. Implementation of these standards enables comprehensive control of the production chain, from seed selection, cultivation techniques, harvesting, and primary processing to storage, thereby optimizing bioactive compound content while minimizing impurities and contamination.

For palmarosa, adherence to GACP-WHO standards not only improves extraction yield and essential oil quality but also provides significant added value, contributing to the establishment of Vietnamese medicinal plant branding and enhancing competitiveness in the international marketplace (Organizacion Mundial de la Salud., 2003). In Hanoi, GACP-WHO–compliant cultivation of palmarosa has been implemented, providing a scientific and practical foundation for investigating the chemical composition and biological activities of essential oil derived from this standardized source.

Figure 1. Palmarosa (*Cymbopogon martini* (Roxb.) W. Watson) cultivated under GACP-WHO guidelines in Hanoi

2. Research overview

International studies have demonstrated that palmarosa (*C. martini*) essential oil exhibits strong biological activities, particularly its ability to inhibit a wide range of microorganisms, including both Gram-positive and Gram-negative bacteria, as well as several common pathogenic fungi (Onawunmi, 1989; Prabuseenivasan et al., 2006). Its chemical composition is dominated by geraniol, which typically accounts for 70–80% of the oil, alongside other key constituents such as geranyl acetate, linalool, and additional monoterpenoids that contribute to both its characteristic fragrance and its bioactivity (Lawrence, 1996).

In Vietnam, research on palmarosa has mainly addressed agronomic traits and small-scale extraction processes, providing useful baseline information but offering little insight into its phytochemical profile (Thuan, 2007). However, there has been limited investigation integrating chemical composition analysis with antibacterial evaluation of palmarosa cultivated under GACP-WHO standards, particularly in the northern regions.

In light of this gap, the present study was undertaken with three primary objectives: (1) extraction of essential oil from palmarosa cultivated in Hanoi, under GACP-WHO guidelines; (2) etermination of its chemical composition using gas chromatography–mass spectrometry (GC-MS); and (3) evaluation of its antibacterial activity against selected reference microorganisms, namely *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis*.

3. Material and methods

3.1. Plant material

The plant material used in this study was palmarosa (*C. martini* (Roxb.) W. Watson) cultivated in Hanoi. The initial plant stock was imported from India and propagated vegetatively from elite mother plants to ensure genetic uniformity and consistent raw material quality. The entire cultivation and maintenance process was conducted in compliance with GACP-WHO guidelines, ensuring safety, sustainability, and control of bioactive compound levels. Harvesting was performed at the full-flowering stage, which corresponds to the peak essential oil and geraniol content, as recommended by Rao et al. (Rao et al., 2005).

3.2. Sample preparation and essential oil

extraction

3.2.1. Sample preparation

The aerial parts of the plant, including stems, leaves, and inflorescences, were collected and processed immediately to minimize essential oil loss. Fresh material was cut into 3–5 cm fragments to increase the surface area for extraction. Samples were weighed to determine fresh biomass for yield calculation. All distillation procedures were carried out within 4 h of harvesting to preserve oil yield and chemical integrity, while minimizing evaporation or degradation of volatile constituents.

3.2.2. Distillation procedure

Essential oil was extracted by steam distillation following the procedure of Hussain et al. (Hussain et al., 2008) with minor modifications. Distillation was performed using a 20 L glass apparatus, with 5.5 kg of fresh material per batch. Heating was provided by a 2 kW electric source and maintained at a steady boil throughout the process. Distillation was conducted for 3.5 h from the onset of boiling. After distillation, the oil layer was separated from the condensate using a separatory funnel, dried over anhydrous sodium sulfate to remove residual moisture, and stored in dark glass vials at 4°C under airtight conditions to minimize oxidation and volatilization.

The extraction yield (H%) was calculated as:

$$H\% = \frac{x}{m} \times 100\%$$

Where:

x is the weight of essential oil obtained

m is the weight of fresh plant material

3.3. GC-MS analysis

Chemical composition was determined by gas chromatography–mass spectrometry (GC-MS). Analyses were performed using an Agilent HP 6890 GC system equipped with an HP 5973 mass selective detector and an HP-5MS capillary column (60 m × 0.25 mm × 0.25 µm). The temperature program was set as follows: initial hold at 40°C, then increased at 3°C/min to 230°C, and held for 10 min. Injector and detector temperatures were set at 230°C and 250°C, respectively. Helium was used as the carrier gas at a flow rate of 1.0 mL/min with a split ratio of 1:50. One microliter of the essential oil, diluted in n-hexane, was injected for each run.

The identification of chemical constituents was based on comparison of retention times and mass spectra (MS) of the chromatographic peaks with reference spectra available in the Wiley/ChemStation HP library. The relative percentage of each compound in the total essential oil was calculated from the corresponding peak areas in the chromatogram.

3.4. Antibacterial activity evaluation

3.4.1. Test microorganisms

Antibacterial activity was evaluated against three reference strains: *Escherichia coli* ATCC 25922 (Gram-negative), *Staphylococcus aureus* ATCC 25923 (Gram-positive), and *Bacillussubtilis* ATCC 6633 (Gram-positive).

3.4.2. Agar diffusion assay

The antibacterial assay was performed following the Clinical and Laboratory Standards Institute (CLSI, 2012) guidelines using the agar diffusion method.

Mueller-Hinton agar (MHA) was used as the growth medium, with incubation at 37°C for 24–48h.

For each test, 150 µL of bacterial suspension (24 h culture, 37°C, 150 rpm shaking) was spread on LB agar plates. Sterile paper discs were placed equidistantly on the agar surface, and 10 µL of each test sample was applied. The treatments included: (i) kanamycin at 10 µg/mL as the positive control, (ii) palmarosa essential oil diluted to 10 µL/mL, and (iii) methanol as the negative control. Plates were refrigerated for 2–3 h to allow diffusion before incubation at 37°C for 24 h.

Antibacterial activity was quantified by measuring the inhibition zone diameter (Dd), calculated as the difference between the total inhibition zone diameter (D, cm) and the disc diameter (d = 1 cm). An inhibition effect was considered positive when D–d > 0.0 cm. Values were reported as the mean of three replicates. Statistical significance was assessed by one-way ANOVA, with $p < 0.05$ considered significant.

4. Research results

4.1. Essential oil yield

Steam distillation of 5.5 kg of fresh palmarosa material yielded 72.89 g of pure essential oil, corresponding to an extraction yield of 1.3252%. This value was higher than that obtained from conventionally cultivated samples

in Dak Lak (1.25%) (Thuan, 2007) and comparable to the findings of Rao et al. (2005) under tropical humid climatic conditions (Table 1).

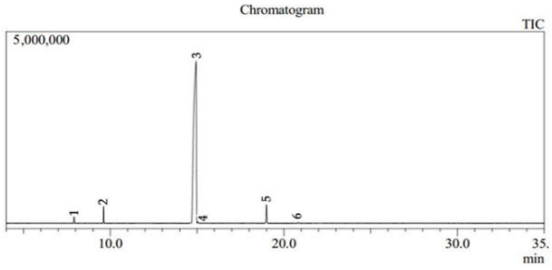
Table 1. Comparison of essential oil yield from palmarosa cultivated under GACP-WHO guidelines in Hanoi, with selected previous studies

Cultivation site	Cultivation conditions	Yield (%)	Reference
Ha Noi, Viet Nam	GACP-WHO	1.3252	This study
Dak Lak, Viet Nam	Traditional	1.25	(Thuan, 2007)
Hyderabad, India	Agricultural practices	1.30–1.35	(Rao et al., 2005)
Rajasthan, India	Traditional	1.15–1.20	(Hussain et al., 2008)

4.2 Chemical composition of the essential oil

The GC-MS chromatogram of palmarosa (*C. martini* (Roxb.) W. Watson) essential oil (Figure 2) revealed the presence of six major constituents. Among these, cis-geraniol predominated, accounting for 76.86% of the total peak area at a retention time of 14.650 min, thereby confirming its role as the characteristic and principal component of the oil. Other notable constituents included geranyl acetate (13.69%), β-linalool (3.43%), and α-citral (2.38%), whereas β-ocimene (1.96%) and isocaryophyllene (1.68%) were present at lower levels.

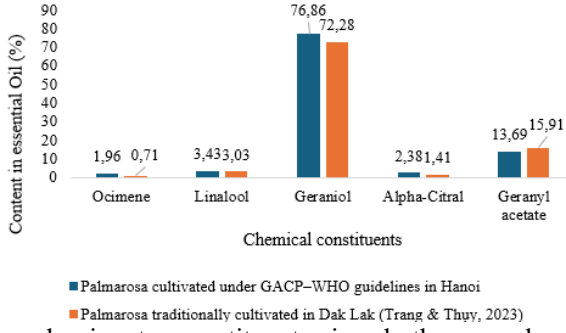
Figure 2. GC–MS Analysis of palmarosa (*Cymbopogon martini*) essential oil cultivated under GACP–WHO guidelines in Hanoi



Peak#	R.time	Area%	Height%	Name
1	8.059	1.96	2.33	.beta-Ocimene
2	9.546	3.43	10.11	.beta-Linalool
3	14.650	76.86	71.21	.cis-Geraniol
4	14.845	2.38	6.49	.alpha-Citral
5	18.865	13.69	9.86	Geranyl acetate
6	20.537	1.68	2.33	Isocaryophylene

The chemical composition of palmarosa essential oil cultivated under GACP-WHO guidelines in Hanoi was compared with that of conventionally cultivated samples from Dak Lak (Trang & Thụy, 2023), as presented in Figure 3.

Figure 3. Comparative chemical composition of palmarosa essential oils cultivated under GACP–WHO guidelines in Hanoi and traditionally grown in Dak Lak



As shown in Figure 3, geraniol was the predominant constituent in both samples. However, its content was higher in the present study (76.86%) compared with the Dak Lak sample (72.28%) (Trang & Thụy, 2023). In addition, other bioactive constituents such as linalool (3.43%) and α-citral (2.38%) were also present at higher levels compared with the Dak Lak sample (3.03% and 1.41%, respectively). Notably, the ocimene content in the investigated sample (1.96%) was nearly three times higher than that in the Dak Lak sample (0.71%). By contrast, geranyl acetate and isocaryophyllene were slightly lower in the investigated sample, at 13.69% and 1.68% compared with 15.91% and 1.83%, respectively.

4.3 Antibacterial Activity

Table 2 shows that palmarosa (*C. martini*) essential oil at a concentration of 10 µL/mL exhibited strong inhibitory activity against both Gram-positive and Gram-negative bacteria.

Table 2. Inhibition zone diameters (cm) of palmarosa essential oil (10 µL/mL) cultivated under GACP-WHO guidelines in Hanoi

Bacteria	Inhibition zone diameters (cm)				
	10h	12h	24h	36h	After 48h
B. subtilis	1.6	1.7	1.7	1.8	2.1
S. aureus	2.1	2.1	2.3	2.3	2.3
E. coli	2.7	2.9	2.9	2.9	2.9

Monitoring antibacterial activity over time revealed an increase in inhibition zone diameters. For *B. subtilis*, the inhibition zone expanded from

1.6 cm at 10 h to 2.1 cm at 48 h. For *S. aureus*, inhibition zones increased from 2.1 cm (10–12 h) to 2.3 cm, which then remained stable up to 48 h. In contrast, *E. coli* exhibited greater sensitivity, with inhibition zones of 2.7 cm at 10 h, increasing to 2.9 cm and remaining stable through 48 h.

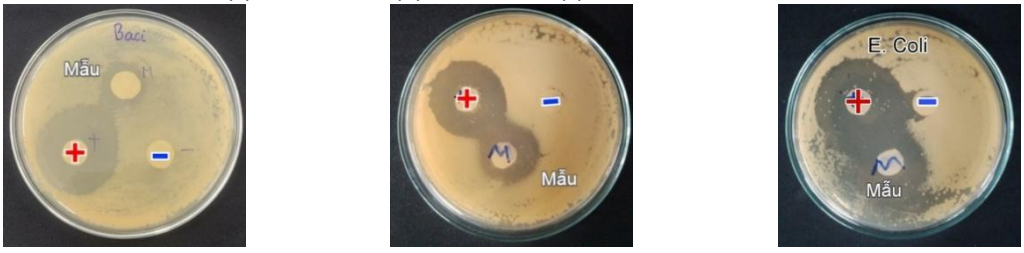
To further evaluate antibacterial efficacy, the inhibitory activity of palmarosa essential oil was compared with kanamycin (10 µg/mL) as a positive control (Table 3).

Table 3. Comparison of inhibition zone diameters (cm) between palmarosa essential oil and kanamycin at a concentration of 10 µL/mL

Bacteria	Sample	Inhibition zone diameters (cm)				
		10h	12h	24h	36h	After 48h
B. subtilis	Palmarosa oil	1.6	1.7	1.7	1.8	2.1
	Kanamycin	3	3	3.1	3.1	3.1
S. aureus	Palmarosa oil	2.1	2.1	2.3	2.3	2.3
	Kanamycin	2.6	2.6	2.9	2.9	2.9
E. coli	Palmarosa oil	2.7	2.9	2.9	2.9	2.9
	Kanamycin	2.9	3.1	3.1	3.1	3.1

The comparative analysis showed that kanamycin produced consistently larger inhibition zones than palmarosa oil across all tested strains. For *B. subtilis*, palmarosa oil yielded relatively small inhibition zones (1.6–2.1 cm) with gradual increases, whereas kanamycin produced stable inhibition zones of 3.0–3.1 cm. Against *S. aureus*, palmarosa oil showed moderate activity (2.1–2.3 cm), which was lower than that of kanamycin (2.6–2.9 cm). Notably, palmarosa oil exhibited strong activity against *E. coli* (2.7–2.9 cm), only slightly less than kanamycin (2.9–3.1 cm).

Figure 4. Inhibition zones of palmarosa essential oil (10 µL/mL) after 24 h incubation against three reference strains: (a) *B. subtilis*, (b) *S. aureus*, (c) *E. coli*.



(a) (b) (c)

5. Discussion

The results obtained from this study demonstrate that palmarosa essential oil cultivated under GACP-WHO guidelines in Hanoi exhibits superior quality characteristics compared with conventionally cultivated samples. The improved yield can be attributed to several key factors: harvesting at the optimal developmental stage, when essential oil content-particularly geraniol-is at its maximum; organic-oriented cultivation practices that minimize pesticide residues, thereby avoiding interference with secondary metabolite biosynthesis; and favorable climatic conditions in Hanoi, characterized by an average annual temperature of 23–25°C and relative humidity of 80–85%, which support palmarosa growth. Furthermore, implementation of GACP-WHO standards during cultivation and harvesting likely contributed to yield optimization, reduced contamination, and enhanced the overall quality of the final essential oil product.

The compositional profile highlights geraniol as the dominant compound, which determines both the distinctive fragrance and the biological potential of the essential oil. These findings are

consistent with international data reported by Lawrence (Lawrence, 1996), in which geraniol constituted more than 70% of high-quality *C. martini* essential oil. Geraniol, a monoterpenoid, is recognized for its prominent bioactivities, particularly its antibacterial effects mediated by disruption of bacterial cell membranes, leakage of intracellular components, and inhibition of protein biosynthesis. Additionally, geraniol exhibits significant anti-inflammatory activity through inhibition of NF-κB signaling and suppression of pro-inflammatory cytokine production (Aelenei et al., 2016; Chen & Viljoen, 2010). In parallel, linalool is a potent antioxidant that scavenges free radicals and protects skin cells from oxidative stress, thereby contributing to anti-aging effects (Kamatou & Viljoen, 2008). The co-occurrence of geraniol, geranyl acetate, and linalool in palmarosa essential oil suggests potential synergistic effects, enhancing antibacterial efficacy and skin-protective properties while providing a pleasant fragrance.

Overall, palmarosa essential oil obtained under the investigated conditions demonstrated advantages in terms of higher levels of geraniol, linalool, and α-citral, thereby enhancing its quality and application value, while the minor differences in other constituents did not substantially affect the general characteristics of the oil.

The antibacterial activity results demonstrate that palmarosa essential oil cultivated under GACP-WHO guidelines in Hanoi exhibits broad-spectrum antibacterial activity, effective against both Gram-positive and Gram-negative bacteria, while suggesting its potential as a safe source of bioactivity. Comparison with the results of Prabuseenivasan et al. (2006) indicated that the antibacterial efficacy of the GACP-WHO oil was comparable to, or in some cases superior to, international samples, particularly against *E. coli*. Although the overall antibacterial potency of palmarosa oil was weaker than that of kanamycin, its pronounced activity against *E. coli* underscores its potential as a natural agent for controlling certain Gram-negative bacteria.

This dual contribution of pharmacological and sensory value underscores the promise of palmarosa essential oil as a valuable natural ingredient for the development of pharmaceutical and cosmetic products. Such results highlight the potential application of palmarosa oil in diverse fields, including cosmeceuticals (e.g., antibacterial creams, mouthwashes, and soaps), food preservation (inhibition of spoilage microorganisms), and aromatherapy (stress reduction, sleep improvement).

6. Conclusion and recommendations

This study successfully extracted and characterized the chemical composition of palmarosa (Cymbopogon martini (Roxb.) W. Watson) essential oil cultivated under GACP-WHO guidelines in Hanoi, Vietnam. The extraction yield was 1.3252%, which was higher than that obtained from conventionally cultivated samples in other regions. GC-MS analysis revealed geraniol (76.86%) as the predominant component, accompanied by geranyl acetate (13.69%) and linalool (3.43%). The high proportion of geraniol observed in this study exceeded values reported in several previous studies, underscoring the superior quality of the essential oil derived from GACP-WHO cultivation practices. Furthermore, the oil demonstrated broad-spectrum antibacterial activity, with particularly strong effects against *E. coli*, showing efficacy nearly comparable to the reference antibiotic kanamycin. Collectively, these findings highlight the potential of palmarosa essential oil cultivated under GACP-WHO guidelines as a promising natural source for the development of eco-friendly cosmeceutical products aligned with global trends in sustainable innovation.

Based on the present findings, several directions for future research and development are proposed: (1) Conduct in-depth studies on the antibacterial mechanisms of geraniol, as well as the potential synergistic effects of secondary constituents, to elucidate pharmacological pathways and broaden the scope of applications. (2) Implement clinical trials of cosmeceutical formulations containing palmarosa essential oil in order to comprehensively assess efficacy, safety, and tolerability in human subjects. (3) Establish an integrated value chain encompassing GACP-WHO–based cultivation, harvesting, and processing through to product commercialization.

This approach not only ensures enhanced quality and value-added benefits but also supports sustainable livelihoods for local farmers and strengthens the competitiveness of Vietnamese enterprises in the global medicinal and cosmeceutical markets.

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NGHIÊN CỨU SƠ BỘ TINH DẦU SẢ HOA HỒNG TRỒNG THEO HƯỚNG DẪN GACP-WHO TẠI HÀ NỘI: THÀNH PHẦN HÓA HỌC VÀ HOẠT TÍNH KHÁNG KHUẨN

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Tóm tắt: *Sả hoa hồng (Cymbopogon martini) là loài thực vật chứa hàm lượng geraniol cao, được ứng dụng rộng rãi trong sản xuất mỹ phẩm, dược phẩm và liệu pháp hương thơm. Trong nghiên cứu này, tinh dầu sả hoa hồng trồng theo hướng dẫn GACP-WHO tại Hà Nội được chiết xuất và phân tích nhằm xác định thành phần hóa học cũng như bước đầu đánh giá hoạt tính kháng khuẩn. Tinh dầu được chiết xuất bằng phương pháp cất kéo hơi nước, phân tích thành phần hóa học bằng sắc ký khí khối phổ (GC-MS), và thử nghiệm hoạt tính kháng khuẩn trên ba chủng vi khuẩn gồm Escherichia coli, Staphylococcus aureus và Bacillus subtilis theo phương pháp khuếch tán trên đĩa thạch. Kết quả cho thấy hiệu suất chiết đạt 1,3252%, cao hơn so với mẫu trồng thông thường tại Đắk Lắk (1,25%); các thành phần chính bao gồm geraniol (76,86%), geranyl acetate (13,69%) và linalool (3,43%). Ở nồng độ 10 µL/mL, tinh dầu thể hiện khả năng ức chế vi khuẩn rõ rệt với đường kính vòng vô khuẩn đạt 2,9 cm đối với E. coli, 2,3 cm đối với S. aureus và 2,1 cm đối với B. subtilis sau 48 giờ. Những kết quả này khẳng định tiềm năng ứng dụng tinh dầu sả hoa hồng trồng theo hướng dẫn GACP-WHO trong phát triển các sản phẩm dược mỹ phẩm có nguồn gốc thiên nhiên, phù hợp với xu hướng bền vững trong chăm sóc sức khỏe.*

Từ khóa: *Cymbopogon martini; GACP-WHO; GC-MS; Kháng khuẩn; Sả hoa hồng; Tinh dầu.*