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# Phytochemical Variations in *Ocimum basilicum* L. Cultivars: Essential Oil Composition and Multivariate Chemotype Differentiation

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**Abstract:** This study investigates the phytochemical variations among 34 cultivars of *Ocimum basilicum* to evaluate their essential oil composition and identify chemotaxonomic patterns. Understanding this variation is important for cultivar authentication, selection, and potential industrial or pharmacological applications. Essential oils were extracted via hydrodistillation and analyzed using gas chromatography-mass spectrometry (GC/MS). To assess chemical variation and group cultivars based on their volatile profiles, principal component analysis (PCA) and hierarchical cluster analysis (HCA) were applied. A total of 48 volatile compounds were identified across the cultivars. Linalool was the most abundant constituent, dominating in 28 cultivars with concentrations ranging from 24.2% to 81.1%. Methyl chavicol exhibited strikingly high levels in certain cultivars, reaching up to 82.0% in some samples. Cluster analysis revealed two major chemotype clades, with chemical similarity indices ranging from −33.07% to 90.84%, indicating substantial intra-specific variation and the potential for clear chemotaxonomic discrimination. The essential oil composition of *O. basilicum* cultivars exhibited marked variability, supporting classification based on chemotypic profiles. These findings enhance the understanding of chemotaxonomy within the genus and provide practical insights for cultivar selection in medicinal, aromatic, and commercial applications.

**Keywords:** *Ocimum basilicum*; essential oils; chemotaxonomy; GC/MS; multivariate analysis. 

## 1. Introduction

The genus *Ocimum* L. is classified within the family Lamiaceae, a taxonomically rich and pharmacologically significant family comprising aromatic herbs and shrubs with well-documented medicinal and culinary uses. Lamiaceae encompasses over 230 genera and approximately 7,000 species globally and is especially renowned for producing essential oils and phenolic compounds [1,2]. Within this family, *Ocimum* stands out as one of the most chemically diverse and widely utilized genera, colloquially referred to as the "king of herbs" due to its extensive cultural, therapeutic, and economic applications [3].

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The genus *Ocimum* comprises between 65 and 160 species, depending on the criteria used in morphological and molecular taxonomic classifications. These species are predominantly distributed throughout tropical and subtropical regions of Asia, Africa, and South America. The broad variation in reported species numbers reflects taxonomic challenges stemming from interspecific hybridization and pronounced morphological plasticity [3–6]. India is widely recognized as a primary center of origin and diversification for *Ocimum*, particularly for well-studied species such as *Ocimum tenuiflorum* L. (holy basil) and *Ocimum basilicum* L. (sweet basil), both of which are extensively cultivated for traditional and industrial use [7–9].

*Ocimum* species play a central role in traditional medicinal practices across various cultures. In Ayurvedic medicine, *O. tenuiflorum* (tulsi) is venerated as a sacred plant and is frequently used to manage respiratory disorders, gastrointestinal issues, skin conditions, and to enhance general immunity [10–12]. In African and South American ethnomedical systems, various *Ocimum* species are utilized as antimicrobial agents, insect repellents, and in healing rituals and spiritual practices [5,13,14]. The widespread use of leaves, seeds, and inflorescences in infusions, poultices, oils, and decoctions underscores the genus's longstanding cultural significance and therapeutic value [15–18].

A growing body of pharmacological research has substantiated many traditional uses attributed to *Ocimum* species. Investigations have confirmed their antimicrobial, antioxidant, anti-inflammatory, antidiabetic, adaptogenic, hepatoprotective, and immunomodulatory activities. These therapeutic effects are primarily linked to the synergistic actions of bioactive phytoconstituents such as eugenol, rosmarinic acid, ursolic acid, and apigenin [19–21]. Species of *Ocimum* are prolific producers of pharmacologically important secondary metabolites, including flavonoids, phenolic acids, terpenoids, alkaloids, and tannins. These compounds support plant defense mechanisms and ecological interactions and contribute directly to the plant's medicinal properties. Major identified metabolites include flavonoids (e.g., apigenin, luteolin), phenolic acids (e.g., rosmarinic, caffeic), and triterpenoids (e.g., ursolic acid, oleanolic acid), many of which have demonstrated significant biological activity, suggesting that Ocimum species could be therapeutically useful in humans [2,22].

Ocimum's defining phytochemical traits are its production of complex essential oils (EOs), which are synthesized in glandular trichomes located on aerial plant parts. These EOs comprise a rich and dynamic assemblage of volatile monoterpenes and phenylpropanoids, including linalool, eugenol, methyl chavicol, 1,8-cineole, and methyl eugenol. The composition and relative abundance of these constituents vary significantly between and within species, influenced by factors such as genotype, environmental conditions, developmental stage, and geographic origin [4,23–26].

This study aimed to analyze and characterize the volatile oil profiles of 34 *Ocimum basilicum* cultivars propagated from seeds. Essential oils were extracted via hydrodistillation and chemically profiled using gas chromatography-mass–mass spectrometry (GC/MS). To further explore interspecific variation and underlying chemotypes relationships, multivariate statistical methods including principal component analysis (PCA) and hierarchical cluster analysis (HCA) were employed. The resulting data provides a comprehensive overview of essential oil variation within the cultivars and offers novel insights into chemotaxonomic classification and potential applications in biological activity and industrial contexts.

## 2. Materials and Methods

### *2.1. Plant Materials*

The total of 34 *Ocimum* cultivars used in this study were propagated from seed obtained from commercial seed suppliers [Ball Seed Company (West Chicago, Illinois, USA), Harris Seeds (Rochester, New York, USA), Johnny's Selected Seeds (Winslow, Maine, USA), and Park Seed Company (Greenwood, South Carolina, USA)] (Table 1). Plants were field-grown during spring 2010 at the South Mississippi Branch Experiment Station (SMBES) in Poplarville, MS (30°50′26″N, long. 89°32′46″W; USDA hardiness zone 8b). Aboveground parts were harvested in mid-July 2010 and air-dried for three weeks inside an air-conditioned building (25°C max.). Voucher specimens were deposited at the SMBES for future reference.

**Table 1.** *Ocimum basilicum* cultivars used in the present study on essential oil composition, each identified by sample number, cultivar name used by the commercial seed supplier, and current botanical name

| Sample number | Cultivar name | Botanical name |
|---|---|---|
| **1** | Amethyst (Improved) | *Ocimum basilicum* 'Amethyst Improved' |
| **2** | Aristotle | *Ocimum basilicum* 'Aristotle' |
| **3** | Aroma2 | *Ocimum basilicum* 'Aroma 2' |
| **4** | Caesar | *Ocimum basilicum* 'Caesar' |
| **5** | Cardinal | *Ocimum basilicum* 'Cardinal' |
| **6** | Christmas | *Ocimum basilicum* 'Christmas' |
| **7** | Cinnamon | *Ocimum basilicum* 'Cinnamon' |
| **8** | Citriodorum | Ocimum × *africanum* (syn. *Ocimum* × *citriodorum*) |
| **9** | Citriodorum Mrs. Burns | *Ocimum* × *africanum* 'Mrs. Burns' (syn. *Ocimum* × *citriodorum* 'Mrs. Burns') |
| **10** | Fino Verde | *Ocimum basilicum* 'Fino Verde' |
| **11** | Genovese | *Ocimum basilicum* 'Genovese' |
| **12** | Italian large leaf | *Ocimum basilicum* 'Italian Large Leaf' |
| **13** | Mammoth | *Ocimum basilicum* 'Mammoth' |
| **14** | Marseille | *Ocimum basilicum* 'Marseille' |
| **15** | Minette | *Ocimum basilicum* 'Minette' |
| **16** | Mozzarella | *Ocimum basilicum* 'Mozzarella' |
| **17** | Napoletano | *Ocimum basilicum* 'Napoletano' |
| **18** | Nufar | *Ocimum basilicum* 'Nufar' |
| **19** | Osmin purple | *Ocimum basilicum* 'Osmin Purple' |
| **20** | Picolino | *Ocimum basilicum* 'Picolino' |
| **21** | Pistou | *Ocimum basilicum* 'Pistou' |
| **22** | Pluto | *Ocimum basilicum* 'Pluto' |
| **23** | Purple Ruffles | *Ocimum basilicum* 'Purple Ruffles' |
| **24** | Queenette | *Ocimum basilicum* 'Queenette' |
| **25** | Red Rubin | *Ocimum basilicum* 'Red Rubin' |
| **26** | Serata | *Ocimum basilicum* 'Serata' |
| **27** | Siam Queen | *Ocimum basilicum* 'Siam Queen' |
| **28** | Spicy Bush | *Ocimum basilicum* 'Spicy Bush' |
| **29** | Suberbo | *Ocimum basilicum* 'Superbo' |
| **30** | Sweet Thai | *Ocimum basilicum* 'Sweet Thai' |
| **31** | Thai Magic | *Ocimum basilicum* 'Thai Magic' |
| **32** | Valentino | *Ocimum basilicum* 'Valentino' |
| **33** | Dark Opal | *Ocimum basilicum* 'Dark Opal' |
| **34** | Genovese Compact (Improved) | *Ocimum basilicum* 'Genovese Compact Improved' |

### *2.2. Essential Oil Analysis*

Dried 300 grams of plant parts were submitted to hydrodistillation using a Clevenger-type apparatus, according to the European Pharmacopeia, and extracted with 2 L of water for 3 hrs. The percentage of oil yield was then calculated on a moisture-free basis (0.03%–0.33%, v/w). The essential oil was collected, dried using anhydrous sodium sulfate, and stored at 4 °C until used.

### *2.3. GC/MS Analysis*

Gas chromatography/mass spectrometry (GC/MS) analysis of the essential oils was performed using an Agilent 5975 GC/MSD system (Santa Clara, CA, USA), equipped with an Innowax FSC capillary column (60 m × 0.25 mm i.d., 0.25 μm film thickness). Helium was used as the carrier gas at a constant flow rate of 0.8 mL/min. The oven temperature program was as follows: initially held at

60 °C for 10 minutes, then increased to 220 °C at a rate of 4 °C/min and maintained at this temperature for another 10 minutes. Subsequently, the temperature was ramped to 240 °C at a rate of 1 °C/min. The injector temperature was set to 250 °C with a split ratio of 40:1. Mass spectra were acquired in electron impact (EI) mode at 70 eV, with a scan range of m/z 35–450 [27].

### *2.4. GC-FID Analysis*

Gas chromatography with flame ionization detection (GC–FID) was carried out on an Agilent 6890N system using the same column and temperature program as in the GC–MS analysis, to ensure consistent retention times. The FID temperature was maintained at 300 °C. Simultaneous auto-injection was applied to both systems. The relative percentages of individual components were calculated from GC–FID peak areas without the use of correction factors [27].

### *2.5. Identification of Essential Oil Components*

Compounds were identified by comparing their retention times and linear retention indices (LRIs) with those of authentic reference standards and a series of *n*-alkanes. Additionally, mass spectral data were matched against commercial databases (Wiley GC/MS Library, MassFinder Software 4.0) and an in-house library of essential oil constituents ("Başer Library") consisting of authenticated reference compounds and previously characterized essential oils [27].

### *2.6. Statistical Analysis*

Multivariate statistical analyses were conducted to assess interspecific chemical variability. All data were normalized before analysis. Principal component analysis (PCA) and hierarchical cluster analysis (HCA) were performed using Minitab 22 software (Minitab LLC, State College, PA, USA). Data was standardized prior to analysis to give equal weight to all variables. Ward's linkage method was applied with Pearson distance as the dissimilarity measure, as it emphasizes differences in the compositional profiles rather than absolute concentrations. The resulting dendrogram was generated based on all identified essential oil components (Tables 2, 3). Sample numbers were used in the dendrogram for clarity, with corresponding cultivar names provided in the figure caption [28].

## 3. Results and Discussion

### *3.1. Chemical Composition*

In this study, the chemical composition of essential oils obtained from 34 different *Ocimum* cultivars was analyzed, and the results are presented in Tables 2 and 3. A total of 48 volatile compounds were identified. Linalool emerged as a prominent constituent, being detected as the main compound in 28 cultivars, with the highest concentration in 'Amethyst Improved' (81.1%). However, linalool was not detected in 'Valentino'. Levels of linalool, the main compound, varied between 24.2% and 81.1% among the cultivars studied, highlighting its significant contribution to the overall aromatic profile of *O. basilicum* essential oils.

In contrast to linalool, methyl chavicol was dominant in a limited number of cultivars and is considered a potential chemotaxonomic marker. High concentrations of methyl chavicol were found in 'Cardinal' (46.0%), 'Mammoth' (12.8%), 'Napoletano' (10.2%), 'Nufar' (14.9%), 'Purple Ruffles' (14.3%), 'Queenette' (53.8%), 'Siam Queen' (82.0%), and 'Sweet Thai' (70.2%).

Additionally, certain cultivars exhibited notably high levels of specific compounds. For example, in 'Citriodorum', geranial (15.7%), nerol (15.5%), and caryophyllene oxide (11.3%) were found in substantial amounts. Methyl (*E*)-cinnamate was present at 15.0% and 24.8% in 'Christmas' and 'Cinnamon', respectively. Eugenol was detected in 'Aristotle', 'Genovese', 'Minette', 'Picolino', 'Pistou', and 'Spicy Bush', with respective concentrations of 9.0%, 18.1%, 12.3%, 10.4%, 11.7%, and 10.9%. Furthermore, $\alpha$-cadinol was identified at 15.7% in 'Valentino'. These compounds may serve as key chemical markers characterizing the essential oil profiles of their respective cultivars.

**Table 2.** Chemical composition of essential oils from *Ocimum basilicum* cultivars numbered 1 to 17 (listed by sample number in Table 1)

| **Compound** | **RRI**[a] | **RRI**[b] | **1**[c] | **2**[c] | **3**[c] | **4**[c] | **5**[c] | **6**[c] | **7**[c] | **8**[c] | **9**[c] | **10**[c] | **11**[c] | **12**[c] | **13**[c] | **14**[c] | **15**[c] | **16**[c] | **17**[c] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $\alpha$-Pinene | 1032 | 1008–1039 [29] | 0.2 | 0.3 | 0.5 | 0.2 | 0.4 | 0.2 | 0.1 | 0.1 | 0.3 | 0.2 | 0.2 | – | 0.3 | 0.1 | – | tr | tr |
| $\beta$-Pinene | 1118 | 1085–1130 [29] | 0.2 | 0.6 | 0.9 | 0.4 | 0.7 | 0.3 | 0.1 | tr | 0.1 | 0.1 | 0.3 | – | 0.4 | tr | tr | tr | tr |
| Sabinene | 1132 | 1098–1140 [29] | tr | 0.2 | 0.2 | 0.1 | 0.2 | 0.1 | tr | tr | tr | 0.1 | tr | – | 0.1 | tr | – | tr | tr |
| 1,8-Cineole | 1213 | 1186–1231 [29] | 4.3 | 9.7 | **14.0** | 6.0 | **11.0** | 6.1 | 1.7 | 0.2 | 0.7 | 1.8 | 6.9 | 1.2 | 6.8 | 0.5 | 2.6 | 5.1 | 3.4 |
| *trans*-Linalool oxide (*Furanoid*) | 1450 | 1429–1481 [29] | 0.3 | 0.3 | 0.3 | 0.3 | 0.5 | 0.9 | 1.1 | 1.7 | 1.3 | 0.7 | 0.2 | 0.2 | 0.7 | 3.9 | tr | 0.2 | tr |
| *cis*-Linalool oxide (*Furanoid*) | 1478 | 1410–1478 [29] | 0.4 | 0.3 | 0.2 | 0.3 | 0.1 | 0.7 | 1.0 | 1.4 | 1.6 | 0.6 | 0.2 | 0.2 | 0.6 | 3.6 | tr | 0.2 | tr |
| Camphor | 1532 | 1481–1537 [29] | 1.0 | 2.0 | 0.4 | 0.6 | – | 0.5 | 0.6 | – | – | 0.9 | 0.8 | 0.3 | 0.5 | – | 1.6 | 0.5 | – |
| Linalool | 1553 | 1507–1564 [29] | **81.1** | **30.7** | **66.5** | **50.9** | **29.7** | **54.9** | **29.8** | 8.4 | **44.5** | **66.3** | **48.6** | **49.4** | **39.3** | **64.9** | **51.3** | **66.8** | **60.5** |
| Octanol | 1562 | 1519–1574 [29] | tr | tr | tr | tr | tr | tr | tr | 1.2 | 0.2 | tr | tr | tr | tr | tr | tr | tr | tr |
| *trans*-α-Bergamotene | 1568 | 1560–1590 [29] | 1.5 | 3.0 | 2.0 | 5.1 | 0.2 | tr | 0.4 | 2.0 | 2.5 | 1.8 | 4.1 | 7.6 | 6.9 | 0.2 | 3.1 | 0.7 | 1.4 |
| Bornyl acetate | 1591 | 1549–1597 [29] | 2.3 | 2.8 | 2.4 | 2.5 | 1.0 | 0.8 | 0.5 | – | – | 2.5 | 1.8 | 0.6 | 0.1 | – | 1.7 | 2.0 | 0.1 |
| $\beta$-Elemene | 1600 | 1565–1608 [29] | 0.6 | 3.8 | 1.0 | 2.2 | 0.5 | 2.1 | 2.2 | – | – | 1.5 | 1.0 | 1.6 | 1.6 | 1.0 | 3.1 | 2.7 | 0.7 |
| Terpinen-4-ol | 1611 | 1564–1630 [29] | tr | 0.2 | 0.6 | 0.6 | 0.5 | 3.6 | 1.6 | – | 6.1 | 3.4 | 0.7 | 2.0 | 4.1 | 1.9 | 6.2 | 0.4 | 0.2 |
| $\beta$-Caryophyllene | 1612 | 1569–1632 [29] | tr | 0.2 | – | – | – | – | 0.2 | 0.6 | 1.1 | tr | – | – | – | – | – | 0.4 | – |
| Hotrienol | 1616 | 1580–1616 [29] | tr | 0.5 | 0.1 | 1.0 | 0.1 | tr | 0.7 | tr | tr | 0.5 | 0.3 | tr | 0.5 | 2.6 | – | 0.4 | – |
| Methyl chavicol | 1687 | 1652–1690 [29] | tr | – | tr | tr | **46.0** | tr | 6.3 | tr | – | tr | tr | 6.2 | **12.8** | – | tr | – | **10.2** |
| α-Humulene | 1687 | 1637–1689 [29] | 0.3 | 1.4 | 0.1 | 1.2 | – | 0.4 | – | – | tr | 0.2 | 0.5 | – | – | 0.1 | 1.2 | 1.2 | – |
| Neral | 1694 | 1641–1706 [29] | tr | – | – | – | – | – | – | 9.6 | 6.9 | – | – | – | – | – | – | – | – |
| $\alpha$-Terpineol | 1706 | 1659–1724 [29] | 1.0 | 2.0 | 1.7 | 1.5 | 1.8 | 0.8 | 0.8 | 2.3 | 1.2 | 1.0 | 1.8 | 1.9 | 1.3 | 0.5 | 2.6 | 3.2 | 0.9 |
| Germacrene D | 1726 | 1676–1726 [29] | 0.4 | 1.3 | 0.8 | 1.6 | 0.2 | 1.3 | 0.8 | – | tr | 0.7 | 0.9 | 1.2 | 1.5 | 0.5 | 1.2 | 1.6 | 0.9 |
| $\delta$-Guaiene | 1733 | | 0.3 | 2.2 | 0.4 | 1.2 | 0.1 | 0.8 | 1.1 | – | – | 0.9 | 0.7 | 0.9 | 0.7 | 0.5 | 1.6 | 1.8 | 0.6 |
| Neryl acetate | 1733 | 1693–1740 [29] | – | – | – | – | – | – | – | 1.6 | 1.1 | – | – | – | – | – | – | – | – |
| Geranial | 1740 | 1680–1750 [29] | tr | – | – | – | – | – | – | **15.7** | **10.6** | – | – | – | – | – | – | – | – |
| $\beta$-Bisabolene | 1741 | 1698–1748 [29] | tr | – | – | – | – | – | | 0.1 | tr | – | – | tr | tr | – | – | – | – |
| Geranyl acetate | 1765 | 1728–1772 [29] | – | – | – | – | tr | – | – | 0.7 | 0.4 | 1.0 | – | – | – | 0.8 | – | – | – |
| $\delta$-Cadinene | 1773 | 1722–1774 [29] | tr | 0.3 | 0.3 | 0.4 | 0.1 | 0.2 | 0.4 | – | – | 0.2 | 0.3 | 0.3 | 0.4 | 0.3 | 0.2 | 0.5 | 0.3 |
| $\gamma$-Cadinene | 1776 | 1735–1782 [29] | 0.6 | 3.1 | 1.1 | 2.9 | 0.8 | 0.9 | 1.7 | – | – | 1.8 | 2.1 | 3.1 | 2.8 | 1.7 | 1.4 | 3.3 | 1.6 |
| (*E*)-$\alpha$-Bisabolene | 1784 | 1763–1786 [29] | – | – | – | – | – | – | – | 1.3 | 1.8 | – | – | – | – | – | – | – | – |

(Continued)

**Table 2. (continued)**

| Compound | RRI[a] | RRI[b] | 1[c] | 2[c] | 3[c] | 4[c] | 5[c] | 6[c] | 7[c] | 8[c] | 9[c] | 10[c] | 11[c] | 12[c] | 13[c] | 14[c] | 15[c] | 16[c] | 17[c] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Nerol | 1808 | 1752–1832 [29] | 0.3 | – | tr | – | tr | tr | 0.2 | **15.5** | 8.1 | 0.2 | 0.2 | 0.2 | 0.1 | 0.2 | tr | 0.1 | 0.3 |
| Geraniol | 1857 | 1795–1865 [29] | 0.3 | 0.2 | 0.1 | – | 0.1 | tr | 0.6 | 3.2 | 3.7 | 0.9 | 0.5 | 0.4 | 0.4 | 5.3 | 0.2 | 0.8 | 0.7 |
| Methyl (*Z*)-cinnamate | 1981 | | – | – | – | – | – | 3.2 | 3.2 | – | – | – | – | – | – | – | – | – | – |
| Caryophyllene oxide | 2008 | 1936–2023 [29] | tr | – | – | – | – | – | tr | 11.3 | 1.4 | – | – | – | – | – | – | – | – |
| Maaliol | 2012 | | 0.3 | – | tr | 1.6 | – | 0.4 | tr | – | – | – | – | 0.4 | 1.5 | tr | – | 0.1 | 0.6 |
| Methyl eugenol | 2030 | 1961–2033 [29] | tr | 6.4 | tr | 0.4 | 0.1 | tr | – | – | – | 0.2 | 0.3 | 0.6 | 0.4 | – | 0.9 | 0.4 | tr |
| (*E*)-Nerolidol | 2050 | 1995–2055 [29] | tr | 0.5 | tr | tr | – | 0.9 | 0.9 | – | 0.2 | 0.1 | tr | 0.4 | 0.3 | 0.2 | 0.3 | 0.4 | 0.7 |
| Humulene epoxide-II | 2071 | 2003–2071 [29] | – | 0.3 | – | tr | – | – | – | 2.0 | 0.2 | 0.1 | – | – | 0.2 | 0.1 | 0.5 | – | – |
| 1,10-*diepi*-Cubenol | 2080 | 2022–2074 [29] | 0.1 | 1.0 | 0.3 | 1.0 | 0.1 | 0.4 | 1.0 | – | – | 0.6 | 0.7 | 1.3 | 1.2 | 0.7 | 0.5 | 1.1 | 2.0 |
| Methyl (*E*)-cinnamate | 2096 | 2046–2105 [29] | – | – | – | – | – | **15.0** | **24.8** | – | – | – | – | – | – | – | – | – | – |
| Spathulenol | 2144 | 2074–2150 [29] | 0.1 | 1.2 | 0.3 | 0.6 | tr | 0.4 | 0.8 | – | – | 0.6 | 0.6 | 0.8 | 0.5 | 0.5 | 0.7 | – | 0.7 |
| Eugenol | 2186 | 2100–2198 [29] | 0.7 | 9.0 | 0.9 | 4.5 | – | 1.1 | 3.8 | – | – | 2.8 | **18.1** | 5.8 | 0.8 | – | **12.3** | – | 0.6 |
| $\tau$-Cadinol | 2187 | 2136–2198 [31] | 1.0 | 7.0 | 1.4 | 7.2 | 1.2 | 1.9 | 4.0 | – | tr | 3.5 | 4.3 | 8.7 | 8.2 | 3.9 | 2.3 | – | **12.3** |
| $\alpha$-Bisabolol | 2232 | 2178–2234 [29] | – | tr | – | – | – | tr | tr | 0.5 | tr | – | – | – | 0.2 | tr | – | – | – |
| $\alpha$-Cadinol | 2255 | 2180–2255 [29] | tr | 0.6 | – | 0.1 | – | tr | 1.5 | – | – | 0.2 | 0.2 | 0.1 | 0.7 | 0.4 | tr | – | 0.6 |
| Nerolic acid | 2308 | | – | – | – | – | – | – | – | 1.4 | tr | – | – | – | – | – | – | – | – |
| Anol | 2328 | | – | tr | – | – | – | – | 0.4 | – | – | – | – | tr | 0.5 | – | – | – | tr |
| Geranic acid | 2349 | 2292–2347 [30] | – | – | – | – | – | – | – | 5.6 | 0.5 | – | – | – | – | – | – | – | – |
| Caryophyllenol-II | 2392 | | – | – | – | – | – | – | – | 1.8 | – | – | – | – | – | – | – | – | – |
| | **Total** | | **97.3** | **87.3** | **96.5** | **94.4** | **95.4** | **98** | **92.3** | **87.2** | **94.5** | **95.4** | **96.3** | **95.4** | **96.4** | **94.4** | **95.5** | **93.9** | **99.3** |

**tr:** Trace (<0.1%); [a]: Relative retention indices (RRI) experimentally determined against *n*-alkanes; [b]: Relative retention indices reported in the literature [29–31]; [c]: calculated from FID data; **1:** Amethyst (Improved); **2:** Aristotle; **3:** Aroma2; **4:** Caesar; **5:** Cardinal; **6:** Christmas; **7:** Cinnamon; **8:** Citriodorum; **9:** Citriodorum Mrs. Burns; **10**: Fino Verde; **11:** Genovese; **12:** Italian large leaf; **13:** Mammoth; **14:** Marseille; **15**: Minette; **16:** Mozzarella; **17:** Napoletano; **18:** Nufar; **19:** Osmin purple; **20:** Picolino; **21:** Pistou; **22:** Pluto; **23**: Purple Ruffles; **24:** Queenette; **25:** Red Rubin; **26:** Serata; **27**: Siam Queen; **28:** Spicy Bush; **29**: Suberbo; **30:** Sweet Thai; **31:** Thai Magic; **32**: Valentino; **33:** Dark Opal; **34**: Genovese Compact (Improved).

**Table 3.** Chemical composition of essential oils from *Ocimum basilicum* cultivars numbered 18 to 34 (listed by sample number in Table 1)

| **Compound** | **RRI**[a] | **RRI**[b] | **18**[c] | **19**[c] | **20**[c] | **21**[c] | **22**[c] | **23**[c] | **24**[c] | **25**[c] | **26**[c] | **27**[c] | **28**[c] | **29**[c] | **30**[c] | **31**[c] | **32**[c] | **33**[c] | **34**[c] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| $\alpha$-Pinene | 1032 | 1008–1039 [29] | 0.2 | 0.3 | 0.2 | tr | 0.5 | 0.3 | 0.2 | 0.2 | 0.3 | tr | 0.3 | 0.3 | tr | tr | 3.7 | 0.4 | 0.5 |
| $\beta$-Pinene | 1118 | 1085–1130 [29] | 0.4 | 0.6 | 0.2 | 0.3 | 1.1 | 0.3 | 0.4 | 0.4 | 0.1 | tr | 0.3 | 0.6 | 0.4 | tr | tr | 0.7 | 0.6 |
| Sabinene | 1132 | 1098–1140 [29] | 0.1 | 0.1 | tr | tr | 0.3 | 0.1 | tr | 0.1 | tr | – | 0.1 | 0.1 | – | – | **39.9** | 0.2 | tr |
| 1,8-Cineole | 1213 | 1186–1231 [29] | 7.2 | 9.3 | 3.7 | 9.1 | **16.2** | 5.8 | 4.8 | 6.2 | 1.2 | 2.0 | 4.6 | 8.0 | **12.1** | 2.0 | – | 9.5 | 7.1 |
| *trans*-Linalool oxide (*Furanoid*) | 1450 | 1429–1481 [29] | 0.2 | 0.3 | 0.5 | – | 0.3 | 0.3 | – | 0.8 | 0.1 | – | 0.2 | 0.5 | – | tr | – | – | tr |
| *cis*-Linalool oxide (*Furanoid*) | 1478 | 1410–1478 [29] | 0.3 | 0.3 | 0.3 | – | 0.2 | 0.3 | tr | 0.8 | 0.1 | – | 0.1 | 0.5 | – | tr | – | 0.1 | 0.2 |
| Camphor | 1532 | 1481–1537 [29] | 0.3 | 1.3 | 0.5 | 1.8 | 2.3 | 0.2 | 2.2 | 1.7 | 1.1 | 2.6 | 0.5 | 0.6 | 2.7 | 0.2 | – | 1.8 | 0.2 |
| Linalool | 1553 | 1507–1564 [29] | **62.4** | **60.5** | **59.2** | **50.4** | **59.3** | **39.1** | 0.5 | **52.2** | **33.5** | 1.0 | **31.0** | **48.8** | 1.8 | **24.2** | tr | **67.5** | **76.2** |
| Octanol | 1562 | 1519–1574 [29] | tr | – | 0.2 | 0.3 | 0.3 | tr | – | 0.1 | 0.1 | – | 0.1 | 0.1 | – | – | – | – | – |
| *trans*-$\alpha$-Bergamotene | 1568 | 1560–1590 [29] | 0.2 | 0.5 | 3.2 | 2.8 | 1.4 | 7.5 | 1.9 | 3.4 | **10.0** | 3.9 | **10.7** | 4.6 | 0.4 | 5.3 | – | 0.3 | 1.3 |
| Bornyl acetate | 1591 | 1549–1597 [29] | 0.1 | 0.3 | 1.3 | 1.0 | 3.0 | 0.4 | 0.8 | 2.2 | 4.0 | tr | 2.4 | 2.8 | 0.9 | 1.6 | – | 0.7 | 0.9 |
| $\beta$-Elemene | 1600 | 1565–1608 [29] | 1.4 | 2.7 | 0.8 | 2.8 | 1.6 | 1.8 | 1.8 | 2.9 | 2.7 | 0.4 | 2.9 | 1.4 | 0.4 | 0.9 | – | 1.9 | 1.8 |
| Terpinen-4-ol | 1611 | 1564–1630 [29] | 0.2 | tr | 2.9 | 8.6 | 1.4 | 4.3 | 0.2 | 0.2 | 7.4 | tr | 4.9 | 0.6 | 0.5 | tr | tr | tr | 0.4 |
| $\beta$-Caryophyllene | 1612 | 1569–1632 [29] | 0.1 | 0.9 | – | – | 0.4 | – | 0.2 | 0.2 | 0.3 | – | tr | – | – | 0.3 | – | 0.8 | – |
| Hotrienol | 1616 | 1580–1616 [29] | tr | 0.5 | 0.4 | – | – | 0.4 | – | 0.9 | 0.2 | – | 0.3 | 1.0 | – | – | – | 0.1 | 0.3 |
| Methyl chavicol | 1687 | 1652–1690 [29] | **14.9** | tr | – | – | – | **14.3** | **53.8** | 1.0 | 0.2 | **82.0** | – | – | **70.2** | 0.2 | – | 1.8 | tr |
| $\alpha$-Humulene | 1687 | 1637–1689 [29] | – | 1.1 | 0.4 | 0.9 | 0.2 | – | – | 1.2 | 0.5 | – | 0.4 | 0.2 | – | tr | – | tr | 0.1 |
| $\alpha$-Terpineol | 1706 | 1659–1724 [29] | 0.9 | 2.3 | 1.1 | 2.0 | 1.6 | 1.8 | 1.7 | 1.8 | 1.1 | 0.4 | 1.7 | 1.9 | 1.2 | 1.1 | – | 1.5 | 1.8 |
| Germacrene D | 1726 | 1676–1726 [29] | 1.9 | 1.4 | 0.3 | 1.1 | 0.7 | 0.9 | 1.1 | 1.3 | 1.5 | – | 1.5 | 0.8 | – | 0.3 | – | 0.9 | 0.4 |
| $\delta$-Guaiene | 1733 | | 0.6 | 1.5 | 0.7 | 1.5 | 0.8 | 1.0 | 0.6 | 1.5 | 1.6 | – | 1.9 | 0.8 | – | 0.6 | – | 0.9 | 0.7 |
| Geranyl acetate | 1765 | 1728–1772 [29] | – | 0.4 | 0.4 | – | – | – | – | – | – | – | – | – | – | – | – | 0.3 | – |
| $\delta$-Cadinene | 1773 | 1722–1774 [29] | 0.2 | 0.3 | 0.2 | tr | 0.1 | 0.5 | 0.4 | 0.1 | 0.4 | tr | 0.4 | 0.3 | – | 0.2 | 1.1 | tr | 0.2 |
| $\gamma$-Cadinene | 1776 | 1735–1782 [29] | 1.3 | 1.0 | 1.8 | 0.8 | 1.0 | 2.1 | 4.0 | 1.2 | 3.3 | 1.1 | 3.2 | 2.4 | 1.2 | 1.6 | – | 0.2 | 1.0 |
| Nerol | 1808 | 1752–1832 [29] | tr | 0.2 | 0.1 | – | – | 0.4 | – | 0.2 | 0.1 | – | 0.1 | 0.2 | – | tr | – | 0.2 | 0.3 |
| Geraniol | 1857 | 1795–1865 [29] | tr | 1.3 | 0.8 | tr | tr | 1.0 | – | 0.4 | 0.3 | – | 0.3 | 0.4 | – | 0.3 | – | 0.5 | 0.7 |
| Caryophyllene oxide | 2008 | 1936–2023 [29] | – | 0.3 | 0.2 | – | – | – | 0.4 | 0.1 | – | 0.3 | – | – | – | 0.3 | tr | 0.1 | – |
| Maaliol | 2012 | | 0.1 | – | – | – | – | 0.2 | 0.7 | 1.0 | – | – | – | 0.7 | – | – | – | – | tr |

(Continued)

**Table 3. (continued)**

| Compound | RRI[a] | RRI[b] | 18[c] | 19[c] | 20[c] | 21[c] | 22[c] | 23[c] | 24[c] | 25[c] | 26[c] | 27[c] | 28[c] | 29[c] | 30[c] | 31[c] | 32[c] | 33[c] | 34[c] |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Methyl eugenol | 2030 | 1961–2033 [29] | 0.1 | 0.3 | 0.2 | – | 0.4 | 0.2 | 1.1 | 0.4 | 0.2 | 1.6 | 0.3 | 0.4 | 0.7 | **45.9** | – | 2.9 | tr |
| (*E*)-Nerolidol | 2050 | 1995–2055 [29] | 0.3 | – | 0.1 | tr | tr | 0.2 | – | 0.6 | 0.5 | – | 0.5 | 0.2 | – | tr | **17.8** | 0.2 | – |
| Humulene epoxide-II | 2071 | 2003–2071 [29] | tr | – | 0.1 | – | – | tr | 1.3 | 0.3 | tr | – | – | 0.1 | 0.7 | tr | – | 0.1 | – |
| 1,10-*diepi*-Cubenol | 2080 | 2022–2074 [29] | 0.4 | – | 0.6 | tr | 0.3 | 0.7 | 1.5 | 0.6 | 1.1 | 0.3 | 1.0 | 0.9 | 0.4 | 0.6 | – | 0.1 | 0.1 |
| 1-*epi*-Cubenol | 2088 | 2026–2090 [29] | – | – | – | – | – | – | – | – | – | – | – | – | – | – | 1.4 | – | – |
| Oplopenone | 2092 | 2049–2097 [29] | – | – | – | – | – | – | – | – | – | – | – | – | – | – | 7.0 | – | – |
| Methyl (*E*)-cinnamate | 2096 | 2046–2105 [29] | – | – | – | – | – | 0.1 | – | – | – | – | – | – | – | 0.1 | – | – | – |
| Hedycaryol | 2122 | 2037–2122 [30] | – | – | – | – | – | – | – | – | – | – | – | – | – | – | 2.1 | – | – |
| Spathulenol | 2144 | 2074–2150 [29] | 0.4 | – | 0.6 | tr | – | 0.3 | – | 1.2 | 1.2 | – | 1.1 | 0.6 | – | 0.5 | 3.3 | – | 0.1 |
| Eugenol | 2186 | 2100–2198 [29] | 0.3 | 2.6 | **10.4** | **11.7** | 2.7 | 2.4 | – | 1.8 | 8.2 | – | **10.9** | 3.4 | – | 5.1 | – | – | 0.7 |
| $\tau$-Cadinol | 2187 | 2136–2198 [31] | 1.8 | 3.2 | 4.0 | 0.6 | 1.3 | 4.5 | 9.2 | 3.7 | 7.3 | 2.3 | 7.3 | 5.6 | 2.1 | 4.2 | – | – | 0.5 |
| $\tau$-Muurolol | 2209 | 2143–2209 [30] | – | – | tr | – | – | 0.1 | – | – | 0.2 | – | – | – | – | – | 1.4 | – | – |
| $\alpha$-Bisabolol | 2232 | 2178–2234 [29] | – | – | 0.2 | – | – | – | – | – | 0.2 | – | 0.1 | tr | – | – | – | – | – |
| $\alpha$-Cadinol | 2255 | 2180–2255 [29] | 0.2 | – | 0.2 | – | tr | 0.5 | 0.2 | 0.5 | 0.3 | tr | 0.6 | 0.1 | – | 0.4 | **15.7** | – | – |
| Anol | 2328 | | tr | – | – | – | – | 0.2 | 0.2 | – | – | tr | – | – | – | – | – | – | – |
| (*E*)-*p*-Methoxy cinnamaldehyde | 2586 | | – | – | – | – | – | – | – | – | – | – | – | – | 2.7 | – | – | – | – |
| **Total** | | | **96.3** | **93.5** | **95.8** | **95.7** | **97.4** | **92.2** | **89.2** | **91.2** | **89.3** | **97.9** | **89.7** | **88.9** | **98.4** | **95.9** | **93.4** | **93.7** | **96.1** |

**tr:** Trace (<0.1%); [a]: Relative retention indices (RRI) experimentally determined against *n*-alkanes; [b]: Relative retention indices reported in the literature [29–31]; [c]: calculated from FID data; **1:** Amethyst (Improved); **2:** Aristotle; **3:** Aroma2; **4:** Caesar; **5:** Cardinal; **6:** Christmas; **7:** Cinnamon; **8:** Citriodorum; **9:** Citriodorum Mrs. Burns; **10**: Fino Verde; **11:** Genovese; **12:** Italian large leaf; **13:** Mammoth; **14:** Marseille; **15**: Minette; **16:** Mozzarella; **17:** Napoletano; **18:** Nufar; **19:** Osmin purple; **20:** Picolino; **21:** Pistou; **22:** Pluto; **23**: Purple Ruffles; **24:** Queenette; **25:** Red Rubin; **26:** Serata; **27**: Siam Queen; **28:** Spicy Bush; **29**: Suberbo; **30:** Sweet Thai; **31:** Thai Magic; **32**: Valentino; **33:** Dark Opal; **34**: Genovese Compact (Improved).

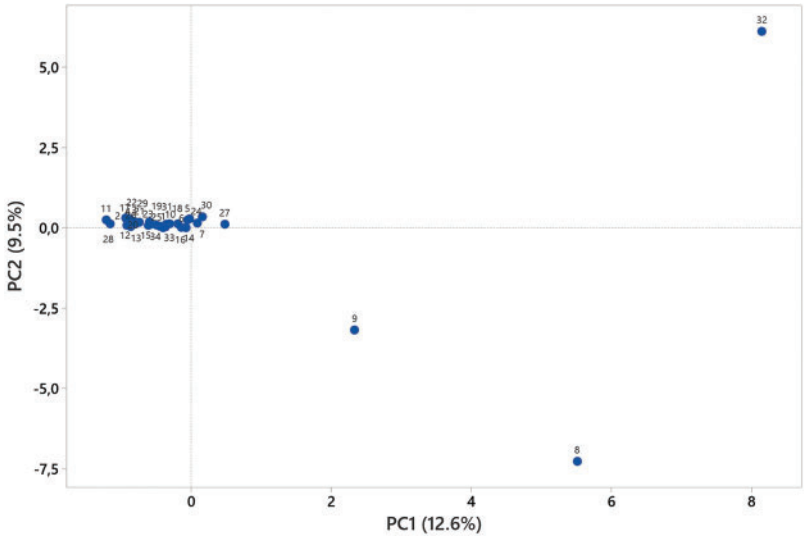


**Figure 1.** Principal component analysis of the major components of essential oils from *O. basilicum* cultivars. **1:** Amethyst (Improved); **2:** Aristotle; **3:** Aroma2; **4:** Caesar; **5:** Cardinal; **6:** Christmas; **7:** Cinnamon; **8:** Citriodorum; **9:** Citriodorum Mrs. Burns; **10:** Fino Verde; **11:** Genovese; **12:** Italian large leaf; **13:** Mammoth; **14:** Marseille; **15:** Minette; **16:** Mozzarella; **17:** Napoletano; **18:** Nufar; **19:** Osmin purple; **20:** Picolino; **21:** Pistou; **22:** Pluto; **23:** Purple Ruffles; **24:** Queenette; **25:** Red Rubin; **26:** Serata; **27:** Siam Queen; **28:** Spicy Bush; **29:** Suberbo; **30:** Sweet Thai; **31:** Thai Magic; **32:** Valentino; **33:** Dark Opal; **34:** Genovese Compact (Improved)

### *3.2. Multivariate Analysis*

Principal component analysis (PCA) and hierarchical cluster analysis (HCA) were used to characterize the volatile compounds of the essential oils of 34 *Ocimum* cultivars, with analyses conducted using Minitab 22 software. The PCA numbers of the essential oils for statistical analysis are given in Table 1. PCA analysis is illustrated in Figure 1, while HCA analysis is given in Figure 3. PCA was used to show the interrelationships among the cultivars of *Ocimum*. In addition, the cluster obtained was confirmed by PCA analysis to evaluate the accuracy of this classification. For the essential oils of *Ocimum*, all variables affected, PC1 (12.6%) and PC2 (9.5%), clarified 22.1% of the accumulated variation of the data analyzed.

To enhance visual separation and interpretability, a second PCA was conducted after excluding the essential oil data for samples 8, 9, and 32, identified as outliers in the initial model (Figure 1). The resulting biplot (Figure 2) revealed a more distinct clustering of the remaining *Ocimum* cultivars and provided clearer insights into the relationships between the samples and their principal volatile constituents. In this refined analysis, PC1 (22.11%) and PC2 (18.34%) jointly accounted for 40.45% of the total variance.

In PC1 (22.11%), positive scores were predominantly characterized by trans-$\alpha$-bergamotene, $\tau$-cadinol, methyl eugenol, and eugenol, whereas negative scores showed minor contributions from linalool. Meanwhile, PC2 (18.34%) was mainly defined by positive scores of linalool, methyl (*E*)-cinnamate, and 1,8-cineole. This distribution indicates that cultivars positioned on the positive side of PC1 tend to exhibit sesquiterpene- and phenylpropanoid-rich chemotypes, while those aligned with positive PC2 values are associated with oxygenated monoterpene-dominant profiles.

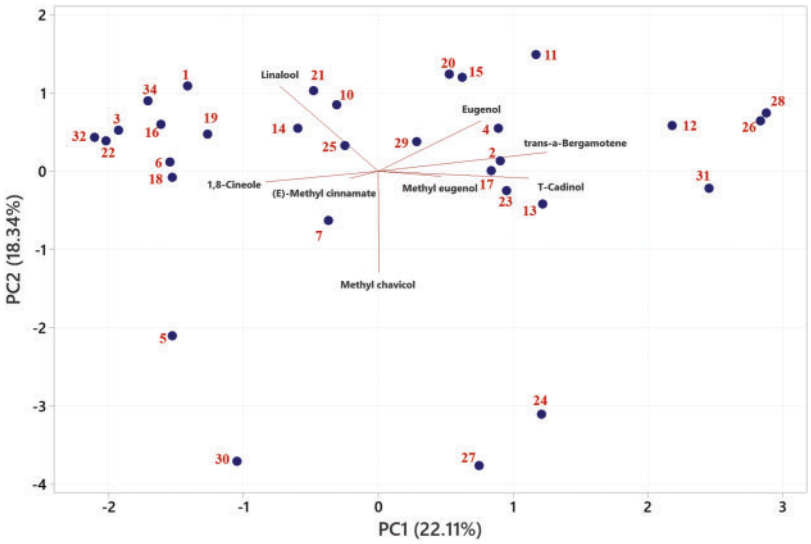


**Figure 2.** Principal component analysis (PCA) biplot of *Ocimum basilicum* cultivars based on essential oil composition (samples 8, 9, and 32 excluded). **1:** Amethyst (Improved); **2:** Aristotle; **3:** Aroma2; **4:** Caesar; **5:** Cardinal; **6:** Christmas; **7:** Cinnamon; **8:** Citriodorum; **9:** Citriodorum Mrs. Burns; **10:** Fino Verde; **11:** Genovese; **12:** Italian large leaf; **13:** Mammoth; **14:** Marseille; **15:** Minette; **16:** Mozzarella; **17:** Napoletano; **18:** Nufar; **19:** Osmin purple; **20:** Picolino; **21:** Pistou; **22:** Pluto; **23:** Purple Ruffles; **24:** Queenette; **25:** Red Rubin; **26:** Serata; **27:** Siam Queen; **28:** Spicy Bush; **29:** Suberbo; **30:** Sweet Thai; **31:** Thai Magic; **32:** Valentino; **33:** Dark Opal; **34:** Genovese Compact (Improved)

Cluster analysis of the essential oil profiles (Figure 3) revealed two main clades, with similarity values ranging from −33.07% to 90.84%. Notably, Amethyst Improved (PCA no: 1) and Genovese Compact Improved (PCA no: 34) exhibited the highest similarity (90.84%), indicating a strong chemotaxonomic relationship and a likely shared botanical origin. A high similarity (89.48%) was also observed between Amethyst Improved (PCA no: 1) and Aroma2 (PCA no: 3). In contrast, Amethyst Improved (PCA no: 1) and Citriodorum (PCA no: 8) were identified as the most dissimilar pair, with a similarity index of −33.07%. This negative value, obtained from Pearson correlation, reflects an inverse relationship in the relative distribution of major components between the two cultivars, highlighting pronounced differences in their essential oil composition and suggesting distinct chemotypes.

In a previous study conducted under Mediterranean environmental conditions in Türkiye, essential oils from various *Ocimum basilicum* cultivars were extracted from dried leaves using hydrodistillation. The oil yield ranged between 1.04 and 2.32 mL per 100 g of dry plant material. In terms of chemical composition, linalool was the dominant constituent in purple basil cultivars (23.7%–46.5%), methyl cinnamate was predominant in cinnamon basil (22.0%–44.1%), and citral was the major component in lemon basil cultivars (26.3%–44.4%) [32].

In a study conducted on *Ocimum basilicum* grown in Djibouti, hydrodistillation of the flowering aerial parts yielded an essential oil rich in monoterpenoids. Among the 37 identified compounds, linalool (41.2%) and estragole (30.1%) were reported as the dominant constituents. Although oil yield was not explicitly stated, the extraction was performed using a traditional distillation setup. These results indicate that the essential oil profile of Djiboutian basil is characterized as a linalool/estragole chemotype [33].

*O. basilicum* plants cultivated in the Dak Lak province of Vietnam were subjected to hydrodistillation, yielding an essential oil with a reported content of 0.55%. GC-MS analysis revealed 34 volatile compounds, with estragole (73.66%) as the predominant component, followed by 1,8-cineole (6.41%), *trans*-$\alpha$-bergamotene (3.97%), linalool (2.25%), and fenchol (1.64%). These findings suggest that the basil essential oil from Dak Lak represents an estragole-rich chemotype [34].

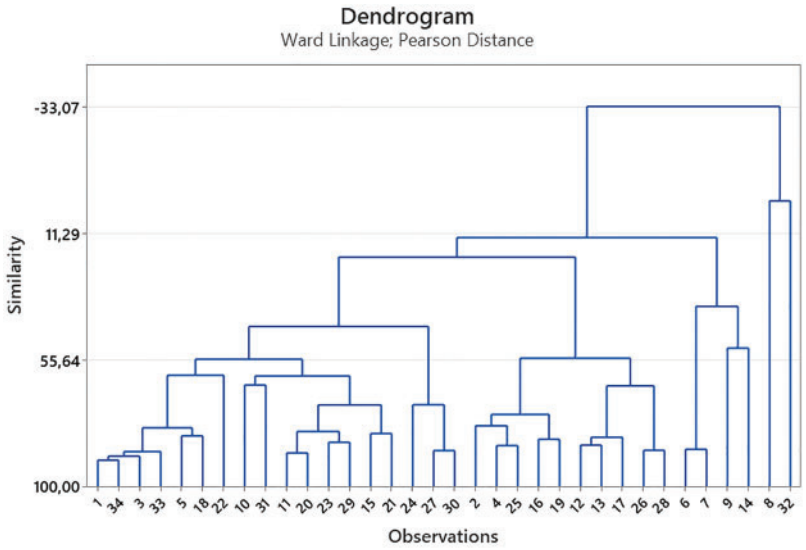


**Figure 3.** Hierarchical cluster analysis (HCA) of essential oil compositions from 34 *Ocimum* cultivars using Ward's linkage method and Pearson distance (dissimilarity measure). **1:** Amethyst (Improved); **2:** Aristotle; **3:** Aroma2; **4:** Caesar; **5:** Cardinal; **6:** Christmas; **7:** Cinnamon; **8:** Citriodorum; **9:** Citriodorum Mrs. Burns; **10:** Fino Verde; **11:** Genovese; **12:** Italian large leaf; **13:** Mammoth; **14:** Marseille; **15:** Minette; **16:** Mozzarella; **17:** Napoletano; **18:** Nufar; **19:** Osmin purple; **20:** Picolino; **21:** Pistou; **22:** Pluto; **23:** Purple Ruffles; **24:** Queenette; **25:** Red Rubin; **26:** Serata; **27:** Siam Queen; **28:** Spicy Bush; **29:** Suberbo; **30:** Sweet Thai; **31:** Thai Magic; **32:** Valentino; **33:** Dark Opal; **34:** Genovese Compact (Improved)

A 2024 study on the "Nufar" cultivar of *Ocimum basilicum* in Colombia examined essential oils extracted from leaves using microwave-assisted hydrodistillation. Samples were collected from three municipalities in Tolima: Honda, Mariquita, and Espinal. Across these sites, the chemical profiles showed similar compositions. The dominant constituents were linalool (37.9%–41.1%) and estragole (24.5%–33.6%), with additional monoterpenes such as 1,8-cineole present in minor amounts. These results highlight that the 'Nufar' cultivar possesses a linalool- and estragole-dominant oil profile [35].

In Tunisia, two *O. basilicum* varieties (including Genovese) were evaluated at different phenological stages for their essential oil profiles. Hydrodistillation was performed for three hours using a Clevenger-type apparatus, with an average yield of approximately 1.5%. In the vegetative phase, the oil contained sesquiterpenes such as germacrene D and bicyclogermacrene, along with oxygenated monoterpenes like linalool (~15%) and 1,8-cineole (6.36%). At full flowering, the composition changed dramatically, with linalool (up to 54.7%) and eugenol (up to 50.8%) becoming the main components. These findings underscore the significant impact of phenological development on basil oil composition [36].

An essential oil obtained from fresh *O. basilicum* leaves cultivated in Sudan was analyzed following hydrodistillation. A total of 41 compounds were identified, accounting for approximately 97% of the total oil composition. The oil was particularly rich in oxygenated monoterpenes, with methyl cinnamate (25.3%), linalool (19.1%), and estragole (12.3%) as the main constituents. Notable sesquiterpene hydrocarbons included $\alpha$-bergamotene (5.3%), germacrene (4.6%), and $\gamma$-cadinene (2.8%), while $\tau$-cadinol (4.3%) was the major oxygenated sesquiterpene. These results confirm the presence of a methyl cinnamate-dominant chemotype in Sudanese basil [37].

A study conducted in Oman evaluated fresh and dried *O. basilicum* samples sourced from various regions (Oman, India, Belgium, Egypt, UAE). Essential oils were extracted using microwave-assisted hydrodistillation. The highest yield was recorded from fresh conventional Omani basil (0.67%, or 675 µL/100 g), although it contained fewer volatile components compared to other samples. GC-MS analysis revealed 14 common compounds across all origins, with $\beta$-linalool consistently dominating the oil profile. Depending on the sample origin, linalool ranged from 31.7% to 80.5%. Notably, Omani

and Indian basil exhibited a linalool-rich chemotype, whereas Belgian organic basil showed a higher proportion of estragole. These results highlight the strong influence of origin and post-harvest treatment on basil essential oil composition [38].

This study provides a comprehensive chemotypic assessment of essential oil compositions across 34 seed-propagated *O. basilicum* cultivars. The application of GC/MS profiling and multivariate statistical analyses revealed significant intraspecific chemical variation, driven by variation in main volatile compounds such as linalool, methyl chavicol, and eugenol. Principal component and hierarchical cluster analyses allowed for identifying distinct chemotypes, supporting the use of essential oil composition as a robust chemotaxonomic tool.

These results underscore the chemical complexity and cultivar-dependent nature of *O. basilicum*, with implications for its industrial, pharmacological, agricultural, and horticultural applications. The chemotypic classification proposed here enhances our understanding of phytochemical variation within the cultivars and offers a valuable reference for future breeding, authentication, and quality control strategies in basil-based products. Given the global demand for high-value aromatic plants, this work contributes timely and applicable insights into the selection and standardization of basil cultivars for targeted uses.

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