Essential Oil Chemotypes of Four Vietnamese *Curcuma* Species Cultivated in North Alabama

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**Abstract:** *Curcuma* (turmeric) species are important culinary and medicinal plants, and the essential oils of *Curcuma* rhizomes have demonstrated promising pharmacological properties. The essential oils (EOs) of *Curcuma* species possess a wide variety of pharmacological properties, including anti-inflammatory, anticancerous, antiproliferative, hypcholesterolemic, anti-diabetic, anti-inflammatory, hypotensive, antioxidant, antimicrobial, antiviral, antithrombotic, antityrosinase, and cyclooxygenase-1 (COX-1) inhibitory activities, among others have been attributed to the essential components of *Curcuma* species. *Curcuma* oils are also known to enhance immune function, promote blood circulation, accelerate toxin elimination, and stimulate digestion. *C. longa* (turmeric) and *C. zedoaria* (zedoary) are the most extensively studied species of *Curcuma* due to their high commercial value. There is some interest in expanding the cultivation of *Curcuma* species to regions in North America where the climate is favorable. The purpose of this work was to examine the rhizome essential oil compositions of four species of *Curcuma* (*C. aromatica*, *C. caesia*, *C. longa*, *C. zanthorrhiza*) that were obtained from Vietnam and cultivated in North Alabama. The rhizome essential oils were obtained by hydrodistillation and analyzed by gas chromatographic techniques. The essential oils of *C. aromatica* were dominated by curzerenone (14.7–18.6%), germacrone (10.7–14.7%), 1,8-cineole (5.2–11.7%), and an unidentified component (8.7–11.0%). The major components in *C. longa* rhizome oil were *α*-turmerone (8.3–36.1%), *α*-turmerone (12.7–15.2%), *β*-turmerone (5.0–15.4%), *α*-zingiberene (4.6–13.9%), and *β*-sesquiphellandrene (4.6–10.0%). The essential oils of *C. caesia* and *C. zanthorrhiza* were rich in curzerenone, curdione, and germacrone. These adapted turmeric varieties in North Alabama have potential use for medical purposes and medicinal plant oil market demands in the U.S.

**Keywords:** turmeric; *Curcuma aromatica*; *Curcuma caesia*; *Curcuma longa*; *Curcuma zanthorrhiza*; chemical composition; enantiomeric distribution; chiral

1. Introduction

There are currently 93 recognized species of *Curcuma* L. (Zingiberaceae) [1]. These perennial rhizomatous herbs originated in subtropical and tropical areas of Asia, Australia, and South America [2], and a number of *Curcuma* species are cultivated in large scales in India, Nepal, Pakistan, Bangladesh, Indonesia, Malaysia, and Thailand [3]. *Curcuma* species are herbaceous perennial herbs with tuberous rhizomes (underground stems). Among them, some important species such as *Curcuma amada* Roxb. (mango ginger), *Curcuma angustifolia* Roxb. (wild arrowroot), *C. aromatica* Salisb. (wild turmeric), *Curcuma caesia* Roxb. (black turmeric), *Curcuma decipiens* Dalzell, *Curcuma kwangsiensis* S. G. Lee & C. Valeton, *Curcuma longa* L. (Zingiberaceae) [1]. These perennial rhizomatous herbs originated in subtropical and tropical areas of Asia, Australia, and South America [2], and a number of *Curcuma* species are cultivated in large scales in India, Nepal, Pakistan, Bangladesh, Indonesia, Malaysia, and Thailand [3]. *Curcuma* species have economical value as they are used in medicine, cosmetics, and both the floricultural and culinary industries [4]. Turmeric is mainly used for culinary, medicinal, and aromatic purposes. Its
rhizomes are the ancient colorful spice source and have a bitter and pungent taste and a pepper-like aroma. Turmeric is also known as the “Golden Spice of India” [5] or “Kitchen Queen” [6]. For example, it has been used in curries in India; in Japan and Korea it is popularly served as a herbal tea; and it is used as a preservative and a coloring agent in mustard sauce, cheese, butter, and chips in the Western world [7]. Curcuminoids and the essential oil of turmeric are associated with a myriad of medicinal, culinary and industrial properties of curcuma species [8], which are derived from the underground plant part, rhizomes (actually the stem), which are tuberous, with a rough and segmented skin. The primary rhizome is known as “mother rhizome” or bulb, and is pear-shaped in the center (Figure 1). The branches of mother rhizomes are the secondary rhizomes, called lateral or “finger rhizomes” [9].

Though turmeric has been known for its multiple uses for over 4000 years in India [10], its use as a medicinal and health supplement in the United States is of recent origin. The interest in turmeric in the U.S. has been increasing over the past two decades mainly due to a large number of scientific publications on its medicinal benefits [8]. To meet the growing demand for turmeric, the U.S. imports 90% of its market demand mainly from India. The U.S. import market was estimated at USD 87.28 million in 2018 [11]. The large market for turmeric in the United States suggests that there is opportunity for cultivation of turmeric in this country provided varieties with high curcumin yield and desirable essential oil composition are available.

Curcuma aromatica Salisb. (wild turmeric) is found naturally in South Asia, including southern China, Bhutan, Myanmar, India, Nepal, Sri Lanka [12], and Vietnam [13], and is widely cultivated in China, India, and Japan [14]. The plant is used in traditional medicines throughout its range for its wound-healing, anti-inflammatory, anti-tumor, immunomodulatory, antimicrobial effects and as an antidote for snake venom [15–17]. The rhizome essential oils are generally dominated by camphor, curzerenone, germacrone, curdione, and 1,8-cineole [14].

Curcuma caesia Roxb. (black turmeric) grows wild in northeastern and central India, Malaysia, Thailand, and Indonesia [14,18]. The rhizome of C. caesia has been used as a traditional medicine to treat leprosy, bronchitis, asthma, cancer, epilepsy, fever, wounds, im-
potence, fertility, vomiting, and pain [19]. Curcuma caesia is considered to be endangered in its native range in India [18], however, it has been underexplored in terms of cultivation and commercialization [20]. The major components in the rhizome essential oil of C. caesia from northeastern India were camphene, 1,8-cineole, camphor, borneol, (E)-β-caryophyllene, and ar-turmerone, which defined two chemotypes, a camphor/ar-turmerone chemotype and a 1,8-cineole/(E)-β-caryophyllene chemotype [20].

Curcuma longa L. (turmeric) is cultivated worldwide, especially in tropical countries in Asia, Australia, and the Neotropics [9]. It is a well-known medicinal agent and culinary ingredient. In addition to curcumin and other non-volatile curcuminoids, the essential oil of turmeric has been employed in the treatment of various maladies in humans and animals [21]. Turmeric essential oils are made up of hundreds of components, the major components, however, are α-turmerone, β-turmerone, ar-turmerone, β-sesquiphellandrene, α-zingiberene, germacrone, terpinolene, ar-curcumene, and α-phellandrene [3,22].

Curcuma zanthorrhiza Roxb. (Javanese turmeric) is often referred to in the literature as Curcuma xanthorrhiza Roxb., however, that name is not recognized by World Flora Online [23]. The plant is native to Indonesia, although is also cultivated in Malaysia, the Philippines, Thailand, Vietnam, and to a lesser extent in China, India, Japan, and South Korea [24]. Traditional medicinal uses of the plant include treatment for stomach illness, liver ailments, constipation, bloody diarrhea, dysentery, arthritis, rheumatism, fevers, hemorrhoids, vaginal discharge, and skin eruptions [24].

As part of our program investigating potential cultivation of Curcuma in Alabama, Curcuma aromatica (both green- and white-colored rhizomes), C. caesia (black-colored rhizome), C. zanthorrhiza (lime-green rhizome), and C. longa (both yellow-, and red-colored rhizomes), obtained from Vietnam, were cultivated in North Alabama (Figure 1). The rhizome essential oils were obtained by hydrodistillation and analyzed by gas chromatographic methods. Both the “mother” or main rhizomes as well as the “daughter” or finger rhizomes were obtained and analyzed. The six Curcuma varieties used in this study were selected out of 64 genotypes according to three criteria: high yield but low curcuminoid content (variety, CL56), high yield but no curcumin content (CA22, CA46, CC38, and CZ44), and high yield and high curcumin content (CL63) based on unpublished data by the authors, Lam Duong and S.R. Mentreddy at Alabama A&M University.

2. Materials and Methods
2.1. Plant Material
The six Curcuma varieties used in this study were selected out of 64 genotypes according to three criteria: high yield but low curcuminoid content (variety, CL56), high yield but no curcumin content (CA22, CA46, CC38, and CZ44), and high yield and high curcumin content (CL63) based on studies at Alabama A&M University. The rhizomes of the six turmeric varieties belonging to Curcuma spp. were collected by Lam Duong from various locations in Vietnam: CA22 (Quang Nam province), CA46 (Gia Lai province), CC38 (Nghe An province), CL56 (Bac Giang province), CL63 (Quang Tri province), and CZ44 (Gia Lai province). The Curcuma rhizomes varieties were initially planted in a glass greenhouse at Alabama A&M University (Normal, AL, USA) and subsequently cultivated at the Alabama A&M Winfred Thomas Agricultural Research Station (Hazel Green, AL, USA, 34° 89′ N, 86° 56′ W) as previously described for C. longa cultivation [9]. Each of the fresh Curcuma rhizomes were collected on 18 March 2021, and stored at −20 °C until processed. The mother and daughter rhizomes were, separately, chopped and hydrodistilled using a Likens–Nickerson apparatus for 4 h (see Table 1).
Table 1. *Curcuma* rhizome hydrodistillation yields.

| Sample Code | Rhizome Color | *Curcuma* Species | Mother Mass (g) | Daughter Mass (g) | Essential Oil Yield (mg) | % Yield | Essential Oil Color |
|-------------|---------------|-------------------|----------------|-------------------|-------------------------|---------|---------------------|
| CA22        | Green         | *C. aromatica*    | 58.72          | 101.27            | 401.9                   | 0.68    | pale yellow         |
|             |               |                   |                |                   | 571.4                   | 0.56    | pale yellow         |
| CA46        | White         | *C. aromatica*    | 48.37          | 85.16             | 382.3                   | 0.79    | pale yellow         |
|             |               |                   |                |                   | 346.3                   | 0.41    | pale yellow         |
| CC38        | Black         | *C. caesia*       | 131.92         | 47.14             | 981.2                   | 0.74    | pale yellow         |
|             |               |                   |                |                   | 531.6                   | 1.13    | pale yellow         |
| CL56        | Yellow        | *C. longa*        | 73.70          | 85.76             | 431.7                   | 0.59    | colorless           |
|             |               |                   |                |                   | 527.0                   | 0.61    | colorless           |
| CL63        | Red           | *C. longa*        | 40.61          | 66.49             | 372.4                   | 0.92    | colorless           |
|             |               |                   |                |                   | 368.1                   | 0.55    | colorless           |
| CZ44        | Lime          | *C. zanthorrhiza* | 64.96          | 79.94             | 700.2                   | 1.08    | colorless           |
|             |               |                   |                |                   | 728.7                   | 0.91    | colorless           |

2.2. Gas Chromatographic Analyses

The *Curcuma* rhizome essential oils were analyzed by gas chromatography—mass spectrometry (GC-MS) as previously reported [3]: Shimadzu GCMS-QP2010 Ultra (Shimadzu Scientific Instruments, Columbia, MD, USA), electron impact (EI) mode (70 eV), scan range 40–400 m/z, scan rate 3.0 scans/s; ZB-5ms GC column (60 m length × 0.25 mm diameter × 0.25 μm film thickness) (Phenomenex, Torrance, CA, USA), He carrier gas, 208.3 kPa head pressure, flow rate 2.0 mL/min, injector temperature 260 °C, ion source temperature 260 °C, oven temperature program 50 °C to 260 °C at 2 °C/min then held at 260 °C for 5 min; 0.3 μL of 5% w/v solutions in CH2Cl2 were injected, split ratio 1:24. Essential oil components were identified by comparison of MS fragmentation and retention index (RI) with those provided in the databases [25–28].

Gas chromatography with flame ionization detection (GC-FID) was carried out as previously reported [29]: Shimadzu GC 2010 (Shimadzu Scientific Instruments, Columbia, MD, USA) equipped with flame ionization detector, ZB-5 capillary column (60 m × 0.25 mm i.d.; film thickness 0.25 μm) (Phenomenex, Torrance, CA, USA); oven temperature programmed as above for GC-MS; injector and detector temperatures 260 °C; He carrier gas, flow rate 1.0 mL/min; 0.3 μL of 5% w/v solution in CH2Cl2 were injected, ratio 1:31. The percent compositions of the essential oils were calculated from peak areas with quantification using the external standard method; calibration curves of representative compounds from each class were used for quantification.

Chiral GC-MS was carried out as previously reported [29]: Shimadzu GCMS-QP2010S instrument (Shimadzu Scientific Instruments, Columbia, MD, USA), Restek B-Dex 325 column (Restek Corporation, Bellefonte, PA, USA); oven temperature program 50 °C to 120 °C at 1.5 °C/min, then to 200 °C at 2.0 °C/min; 0.1 μL of 5% w/v solutions in CH2Cl2 were injected, with a split ratio of 1:25. The enantiomers were determined by comparison of retention times with authentic samples obtained from Sigma-Aldrich (Milwaukee, WI, USA) and the relative enantiomer percentages were calculated from peak integration.

2.3. Statistical Analysis

Analysis of variance was conducted by one-way ANOVA followed by the Tukey test using Minitab® 18 (Minitab Inc., State College, PA, USA). Differences at $p < 0.05$ were considered to be statistically significant. For the agglomerative hierarchical cluster (AHC) analysis, the 12 essential oil compositions were treated as operational taxonomic units (OTUs), and the concentrations (percentages) of 18 of the most abundant essential oil components (curzerenone, curdione, germacrone, ar-turmerone, 1,8-cineole, α-turmerone, unidentified (RI = 1778), β-turmerone (=curlone), β-sesquiphellandrene, α-zingiberene, iso-curcumenol, curcumene, trans-β-elemene, ar-curcumene, β-pinene, curcumene, cam-
phor, and curzerene) were used to determine the chemical associations between the *Curcuma* rhizome essential oil samples using XLSTAT Premium, version 2018.1.1.62926 (Addinsoft, Paris, France). Similarity was determined using the Pearson correlation, and clustering was defined using the unweighted pair-group method with arithmetic mean (UPGMA).

3. Results and Discussion

3.1. Chemical Compositions of Curcuma Rhizome Essential Oils

The fresh rhizome samples were hydrodistilled to give colorless or pale-yellow essential oils in yields ranging from 0.41% to 1.13% (Table 1). The chemical compositions of the *Curcuma* rhizome essential oils are compiled in Table 2. Gas chromatograms of each Curcuma variety are shown in Supplementary Figure S1.

The essential oils from the green-colored mother and daughter rhizomes of *C. aromatica* (CA22) were dominated by curzerenone (18.6% and 14.7%, respectively), germacrone (14.7% and 10.7%, respectively), 1,8-cineole (11.7% and 6.6%, respectively), and an unidentified sesquiterpenoid (RI 1778, Supplementary Figure S2) (9.0 and 8.7%, respectively). Similarly, the white-colored mother and daughter rhizomes (*C. aromatica*, CA46) were dominated by the same components, curzerenone (14.9% and 14.8%), germacrone (14.5% and 12.5%), 1,8-cineole (10.2% and 5.2%), and the unidentified compound (RI 1778) (11.0% and 10.3%). The rhizome essential oil compositions of *C. aromatica* show wide variation depending on geographical location [14]. For example, camphor was found to be a major component of *C. aromatica* rhizome essential oils from India (18.8–32.3%), whereas 8,9-dehydro-9-formylcycloisolongifolene (2.7–36.8%) was a dominant compound in the essential oils from China. Camphor was relatively minor in *C. aromatica* cultivated in North Alabama in this work (1.4–2.5%) and 8,9-dehydro-9-formylcycloisolongifolene was not observed. Curzerenone, germacrone, and 1,8-cineole, however, are relatively concentrated in Indian *C. aromatica* rhizome essential oils [14]. The rhizome essential oil of *C. aromatica* from Thailand showed camphor (26.9%), *ar-curcumene* (23.2%), and xanthorrhizol (18.7%) as the major components [30], whereas the rhizome essential oil from *C. aromatica* cultivated in Japan revealed β-turmerone (32.2 and 44.0%), 1,8-cineole (7.5 and 25.3%), and germacrone (4.6 and 9.6%) to be major compounds [31]. Notably, curzerenone, 8,9-dehydro-9-formylcycloisolongifolene, *ar-curcumene*, and xanthorrhizol were not detected in the essential oils from Japan. A recent examination of *C. aromatica* from different regions of eastern and southern India revealed relatively low concentrations of curzerenone (0.0–1.2%), but high concentrations of xanthorrhizol (8.8–24.4%), camphor (4.1–18.1%), germacrone (3.5–21.9%), neocurdione (5.8–14.6%), and 1,8-cineole (3.7–11.9%) [32].

The black-rhizome (*C. caesia*, CC38) essential oil, on the other hand, was rich in curzerenone (26.1% and 29.1%), curdione (28.7% and 35.6%), as well as *iso-curcumene* (6.5% and 5.6%), for the mother and daughter rhizomes, respectively. In contrast, *C. caesia* rhizome essential oil from north India showed 8,9-dehydro-9-formylcycloisolongifolene, (11.7%), camphor (6.1%), 1,8-cineole (6.0%), and β-elemene (5.2%, reported as β-germacrene) as major components [33]. Neither curzerenone, curdione, nor *iso-curcumene* were reported in the essential oil from north India, and 8,9-dehydro-9-formylcycloisolongifolene was not found in the essential oil from North Alabama. Note that curzerenone was determined to be artificially elevated in *C. caesia* essential oil due to the Cope rearrangement of furanodienone [34].
Table 2. Chemical compositions of the rhizome (mother and daughter) essential oils of Curcuma species from Vietnam, cultivated in North Alabama.

| RI<sub>calc</sub> | RI<sub>db</sub> | Compound                  | Curcuma aromatica (Green Rhizome) | Curcuma aromatica (White Rhizome) | Curcuma coesia (Black Rhizome) | Curcuma zanthorrhiza (Lime Rhizome) | Curcuma longa (Yellow Rhizome) | Curcuma longa (Red Rhizome) |
|-----------------|-----------------|---------------------------|-----------------------------------|-----------------------------------|---------------------------------|-------------------------------------|-------------------------------|-----------------------------|
| 807             | 802             | 2-Hexanol                 | 0.2                               | 0.1                               | 0.2                             | 0.1                                 | 0.1                           | 0.1                         |
| 888             | 889             | 2-Heptanone               | —                                 | —                                 | 0.8                             | 0.1                                 | 0.1                           | 0.1                         |
| 897             | 900             | 2-Heptanol                | 0.2                               | 0.2                               | 0.2                             | 0.2                                 | 0.1                           | 0.2                         |
| 918             | 923             | Tricycylene               | tr                               | tr                                | tr                              | tr                                  | tr                            | tr                          |
| 926             | 925             | α-Thujene                 | 0.5                               | 0.1                               | 0.3                             | 0.1                                 | 0.1                           | 0.1                         |
| 933             | 933             | β-Pinene                  | 0.2                               | 0.2                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 949             | 950             | Camphene                  | 0.3                               | 0.4                               | 0.3                             | 0.3                                 | 0.1                           | 0.1                         |
| 962             | 960             | Benzaldehyde              | —                                 | —                                 | 0.3                             | 0.3                                 | 0.1                           | 0.1                         |
| 972             | 972             | Sabinene                  | 0.6                               | 0.6                               | 0.6                             | 0.6                                 | 0.6                           | 0.6                         |
| 979             | 978             | β-Pinene                  | 2.4                               | 6.2                               | 2.5                             | 5.9                                 | 0.3                           | 0.5                         |
| 980             | 979             | 2-Methyl-2-hepten-4-one   | —                                 | —                                 | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 984             | 981             | 6-Methyl-5-hepten-2-one   | —                                 | —                                 | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 986             | 986             | p-Methyl-3-ene            | 0.2                               | 0.3                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 989             | 989             | Myrene                    | 0.6                               | 0.6                               | 0.6                             | 0.6                                 | 0.6                           | 0.6                         |
| 1002            | 1004            | 2-Octanol                 | 0.1                               | 0.1                               | 0.1                             | 0.1                                 | 0.1                           | 0.1                         |
| 1007            | 1007            | α-Phellandrene            | 0.2                               | 0.3                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 1009            | 1009            | δ-3-Carene                | 0.2                               | 0.3                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 1017            | 1017            | α-Terpine                 | 0.2                               | 0.3                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 1024            | 1025            | p-Cymene                  | 0.2                               | 0.3                               | 0.2                             | 0.2                                 | 0.1                           | 0.1                         |
| 1029            | 1030            | Limonene                  | 0.6                               | 0.6                               | 0.6                             | 0.6                                 | 0.6                           | 0.6                         |
| 1033            | 1032            | 1,8-Cineole               | 0.8                               | 6.2                               | 10.2                            | 5.2                                 | 2.0                           | 0.5                         |
| 1035            | 1034            | (Z)-β-Ocimene             | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1038            | 1041            | 2-Heptyl acetate          | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1045            | 1045            | (E)-β-Ocimene             | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1057            | 1057            | γ-Terpine                 | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1085            | 1086            | Terpinolene               | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1090            | 1093            | p-Cymenene                | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1090            | 1099            | 2-Nonanal                 | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1099            | 1099            | Linalool                  | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1105            | 1105            | 2-Nonanol                 | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1138            | 1139            | Neopinone                 | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1142            | 1141            | (E)-Myroxide              | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1146            | 1145            | Camphor                   | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1146            | 1146            | Myrcene                   | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1158            | 1156            | Camphene hydrate          | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1162            | 1162            | Pinocarvone               | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1164            | 1165            | iso-Borneol               | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1170            | 1170            | δ-Terpine                 | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| 1174            | 1173            | Borneol                   | 0.8                               | 6.2                               | 0.5                             | 0.5                                 | 0.1                           | 0.1                         |
| Rלק | Rלקב | Компонент | Curcuma aromatica (Green Rhizome) | Curcuma aromatica (White Rhizome) | Curcuma caesia (Black Rhizome) | Curcuma zanthorrhiza (Lime Rhizome) | Curcuma longa (Yellow Rhizome) | Curcuma longa (Red Rhizome) |
|------|------|-----------|-----------------------------------|-----------------------------------|---------------------------------|-----------------------------------|------------------------------|-----------------------------|
| 1175 | 1176 | cis-Pinocamphone | — | — | — | — | 0.1 | 0.1 |
| 1180 | 1180 | Terpinen-4-ol | 0.3 | 0.2 | 0.3 | 0.2 | 0.2 | 0.1 |
| 1184 | 1188 | 4′-Methylacetophenone | — | — | — | — | — | — |
| 1187 | 1186 | p-Cymen-8-ol | — | — | — | — | 0.2 | 0.1 |
| 1189 | 1290 | Indole | — | — | tr | — | — | — |
| 1191 | 1190 | 2-Decanone | — | tr | — | — | 1.3 | 0.3 |
| 1195 | 1195 | α-Terpineol | 0.7 | 0.5 | 0.7 | 0.4 | 0.4 | 0.2 |
| 1203 | 1203 | p-Cumenol | — | — | — | — | — | — |
| 1206 | 1205 | Verbenone | — | — | — | — | — | — |
| 1209 | 1211 | 2-Decanone | — | — | — | — | — | — |
| 1232 | 1233 | 2-Nonyl acetate | — | — | — | — | — | — |
| 1243 | 1242 | Carvone | tr | — | — | — | — | — |
| 1274 | 1274 | Cyclooctyl acetate | — | — | — | — | — | — |
| 1291 | 1293 | 2-Undecanone | 0.1 | 0.1 | tr | 0.1 | 0.1 | 0.1 |
| 1310 | 1317 | 2-Undecanol | — | — | — | — | 0.2 | 0.1 |
| 1334 | 1335 | 3-Elemene | — | 0.2 | — | — | 0.3 | 0.2 |
| 1336 | 1339 | Piperitenone | — | — | — | — | 0.1 | 0.1 |
| 1343 | 1344 | Evodone | — | — | — | — | — | — |
| 1346 | 1348 | α-Cubebeene | — | — | — | — | — | — |
| 1368 | 1367 | Cyclostivene | — | tr | — | — | — | — |
| 1374 | 1375 | α-Copaene | — | — | — | — | — | — |
| 1381 | 1383 | cis-β-Elemene | 0.1 | 0.3 | 0.1 | 0.2 | 0.2 | 0.1 |
| 1390 | 1390 | trans-β-Elemene | 2.7 | 4.9 | 2.4 | 3.7 | 2.9 | 2.0 |
| 1393 | 1394 | Sativene | — | — | — | — | — | — |
| 1402 | 1405 | Sesquithujene | — | — | — | — | 0.1 | 0.2 |
| 1418 | 1417 | (E)-β-Caryophyllene | 0.4 | 1.4 | 0.4 | 1.1 | 0.9 | 0.8 |
| 1423 | 1425 | α-Sinensal | — | — | — | — | — | — |
| 1428 | 1427 | γ-Elemene | 0.3 | 0.5 | 0.3 | 0.4 | 0.3 | 0.2 |
| 1432 | 1432 | trans-α-Bergamotene | — | — | — | — | — | — |
| 1440 | 1442 | Guai-6,9-diene | — | — | — | — | 1.1 | 1.2 |
| 1442 | 1439 | (Z)-β-Farnesene | — | — | — | — | 0.1 | 0.1 |
| 1447 | 1447 | iso-Germacrene D | 0.1 | 0.2 | 0.1 | 0.1 | 0.1 | 0.1 |
| 1451 | 1452 | (E)-β-Farnesene | 0.1 | 0.3 | 0.1 | 0.2 | 0.1 | 0.1 |
| 1455 | 1454 | α-Humulene | 0.1 | 0.2 | 0.1 | 0.2 | 1.7 | 1.5 |
| 1459 | 1458 | allo-Aromadendrene | tr | 0.1 | tr | 0.1 | — | — |
| 1462 | 1462 | α-Acoradiene | — | — | — | — | — | — |
| 1472 | 1474 | Selina-4,11-diene | — | — | — | — | — | — |
| 1473 | 1473 | Dauca-5,8-diene | — | — | — | — | — | — |
| 1474 | 1475 | γ-Muurolene | 0.1 | 0.3 | — | — | tr | — |
### Table 2. Cont.

| RI<sub>calc</sub> | RI<sub>db</sub> | Compound | Curcuma aromatica (Green Rhizome) | Curcuma aromatica (White Rhizome) | Curcuma caesia (Black Rhizome) | Curcuma zanthorrhiza (Lime Rhizome) | Curcuma longa (Yellow Rhizome) | Curcuma longa (Red Rhizome) |
|------------------|-----------------|----------|-----------------------------------|----------------------------------|--------------------------------|-----------------------------------|--------------------------------|--------------------------------|
|                  |                 |          | CA22 (M)  | CA22 (D)  | CA46 (M)  | CA46 (D)  | CC38 (M)  | CC38 (D)  | CZ44 (M)  | CZ44 (D)  | CL56 (M)  | CL56 (D)  | CL63 (M)  | CL63 (D)  |
| 1476             | 1480            | β-Acoradiene | —        | —        | 1.8       | 2.5       | —         | —         | 1.9       | 2.3       | 3.5       | 3.6       | 2.6       | 2.4       |
| 1480             | 1480            | α-Curcumene  | 1.6      | 3.1      | —         | —         | —         | —         | 1.9       | 2.3       | —         | —         | —         | —         |
| 1480             | 1480            | Germacrene D | —        | —        | —         | —         | —         | —         | —         | —         | —         | —         | —         | —         |
| 1484             | 1483            | trans-β-Bergamotene | —    | —        | —         | —         | —         | —         | 0.1       | 0.1       | —         | —         | —         | —         |
| 1488             | 1489            | β-Selinene   | 0.3      | 0.4      | 0.3       | 0.4       | 0.2       | 0.2       | 0.2       | 0.2       | —         | —         | —         | —         |
| 1494             | 1493            | Curzerene    | 1.2      | 2.0      | 1.4       | 1.9       | 3.3       | 1.8       | 2.4       | 3.9       | —         | —         | —         | —         |
| 1495             | 1497            | α-Selinene   | 0.4      | 0.5      | 0.2       | 0.2       | —         | 0.1       | 0.1       | 0.1       | —         | —         | —         | —         |
| 1496             | 1496            | α-Zingiberene | —        | —        | —         | —         | —         | —         | 0.3       | 11.4      | 13.9      | 3.4       | 4.6       | —         | —         |
| 1497             | 1497            | α-Murolene   | 0.1      | 0.3      | 0.1       | 0.5       | —         | —         | 0.1       | 0.3       | —         | —         | —         | —         |
| 1504             | 1501            | β-Dihydroagarofuran | —  | —        | —         | —         | —         | —         | 0.3       | 11.4      | 13.9      | 3.4       | 4.6       | —         | —         |
| 1504             | 1502            | trans-β-Guaiene | —    | —        | —         | —         | —         | —         | 0.1       | 0.3       | —         | —         | —         | —         |
| 1505             | 1505            | α-Bulnesene  | 0.1      | 0.2      | —         | —         | —         | —         | —         | —         | —         | —         | —         | —         |
| 1507             | 1504            | iso-Daucene  | —        | —        | —         | —         | —         | —         | 0.1       | 0.1       | —         | —         | —         | —         |
| 1508             | 1508            | β- Bisabolene | 0.5      | 1.0      | 0.6       | 0.8       | —         | —         | 0.5       | 0.5       | 2.4       | 2.5       | 0.8       | 0.8       |
| 1513             | 1512            | γ-Cadinene   | 0.1      | 0.2      | 0.1       | 0.1       | —         | —         | 0.1       | 0.1       | —         | —         | —         | —         |
| 1517             | 1519            | α-Cubebol    | 0.2      | 0.4      | —         | —         | —         | —         | —         | —         | —         | —         | —         | —         |
| 1518             | 1518            | δ-Cadinene   | 0.3      | 0.9      | 0.3       | 0.5       | tr        | 0.1       | 0.3       | 0.6       | —         | —         | —         | —         |
| 1520             | 1518            | 7-epi-α-Selinene | —     | —        | —         | —         | —         | —         | tr        | —         | —         | —         | —         | —         |
| 1521             | 1523            | β-Guaiene    | —        | —        | —         | —         | —         | —         | 0.4       | 0.4       | —         | —         | —         | —         |
| 1522             | 1523            | β-Sesquiphellandrene | 1.1   | 2.5      | 1.6       | 1.9       | —         | tr        | 1.6       | 1.9       | 8.9       | 10.0      | 4.9       | 4.6       |
| 1528             | 1528            | (E)-γ-Bisabolene | —   | —        | —         | —         | —         | —         | 0.2       | 0.2       | 0.2       | 0.2       | —         | —         |
| 1549             | 1549            | α-Elemol     | 0.2      | 0.2      | —         | —         | —         | —         | —         | —         | —         | —         | —         | —         |
| 1555             | 1555            | 7-epi-cis-Sesquisabinene hydrate | — | —        | —         | —         | —         | —         | 0.5       | 0.4       | 0.2       | 0.1       | —         | —         |
| 1558             | 1560            | Germacrene B | 0.5      | 0.9      | 0.6       | 0.8       | 0.7       | 0.5       | 0.8       | 0.9       | 0.3       | 0.2       | —         | —         |
| 1560             | 1560            | (E)-Nerolidol | —     | —        | —         | —         | —         | —         | —         | 0.1       | tr        | 0.5       | —         | —         |
| 1578             | 1578            | α-Tumerol    | 0.3      | 0.3      | 0.3       | 0.3       | —         | —         | 0.3       | 0.3       | 0.3       | 0.3       | —         | —         |
| 1582             | 1587            | Caryophyllene oxide | 0.2   | 0.5      | 0.2       | 0.4       | 0.3       | 0.4       | 0.2       | 0.2       | 0.1       | 0.1       | 0.4       | —         |
| 1590             | 1590            | Globulol     | 0.2      | 0.3      | —         | —         | 0.3       | —         | 0.3       | —         | —         | —         | —         | —         |
| 1592             | 1591            | 7-epi-trans-Sesquisabinene hydrate | — | —        | —         | —         | —         | —         | 1.6       | 1.5       | 0.3       | 0.7       | —         | —         |
| 1592             | 1591            | cis-β-Elemenone | —    | —        | —         | —         | —         | —         | 0.2       | —         | —         | —         | —         | —         |
| 1595             | 1594            | Viridiflorol | 0.7      | 0.2      | 0.2       | 0.2       | 0.1       | 0.2       | 0.1       | 0.3       | —         | —         | —         | —         |
| 1596             | 1596            | trans-β-Elemenone | —   | 1.6      | 3.0       | 2.7       | —         | —         | 1.8       | 1.9       | 1.3       | 1.1       | 0.4       | —         |
| 1600             | 1601            | Humulene epoxide I | — | —        | —         | —         | 2.2       | 1.7       | —         | —         | —         | —         | —         | —         |
| 1600             | 1599            | anti,anti,anti-Heliofol-12-al B | —   | —        | —         | —         | —         | —         | 0.2       | 0.3       | 0.8       | 0.6       | —         | —         |
| 1602             | 1603            | Curzerenone   | 18.6     | 14.7     | 14.9      | 14.8      | 26.1      | 29.1      | 16.3      | 19.7      | —         | —         | —         | —         |
| 1611             | 1613            | Humulene epoxide II | —  | —        | —         | —         | 0.3       | 0.3       | 0.3       | —         | —         | —         | —         | —         |
### Table 2. Cont.

| R_{calc} | R_{db} | Compound               | Curcuma aromatica (Green Rhizome) | Curcuma aromatica (White Rhizome) | Curcuma caesia (Black Rhizome) | Curcuma zanthorrhiza (Lime Rhizome) | Curcuma longa (Yellow Rhizome) | Curcuma longa (Red Rhizome) |
|----------|--------|------------------------|-----------------------------------|-----------------------------------|--------------------------------|-----------------------------------|-------------------------------|-----------------------------|
|          |        |                        | CA22 (M)                          | CA22 (D)                          | CA46 (M)                      | CA46 (D)                          | CC38 (M)                      | CC38 (D)                    |
| 1614     | 1610   | iso-Curzerenone         | —                                 | —                                 | —                              | —                                 | —                             | —                           |
| 1615     | 1615   | Zingiberenol            | 0.7                               | 0.8                               | 0.7                            | 0.8                               | —                             | —                           |
| 1621     | 1616   | iso-Curcumulene         | 4.0                               | 3.1                               | 3.7                            | 3.4                               | 6.5                           | 5.6                         |
| 1630     | 1629   | iso-Spathululon         | 0.7                               | 0.4                               | 0.8                            | 1.0                               | 0.5                           | 0.8                         |
| 1632     |        | Unidentified a          | 0.2                               | —                                 | 0.9                            | —                                 | 0.4                           | 0.5                         |
| 1636     |        | Unidentified a          | —                                 | —                                 | 0.1                            | 0.9                               | —                             | 0.3                         |
| 1643     | 1643   | \(\tau\)-Cadinol       | 0.3                               | 0.4                               | 0.4                            | 0.7                               | —                             | 0.3                         |
| 1644     | 1644   | \(\alpha\)-Aromadendrene epoxide | —                                 | —                                 | —                              | —                                 | —                             | —                           |
| 1646     | 1645   | \(\tau\)-Muurtrolol     | 0.3                               | 0.6                               | —                              | —                                 | —                             | —                           |
| 1648     | 1651   | \(\alpha\)-Muurtrolol   | —                                 | —                                 | —                              | —                                 | —                             | —                           |
| 1655     | 1655   | \(\alpha\)-Cadinol      | 0.3                               | 0.4                               | 0.4                            | 0.4                               | —                             | 0.7                         |
| 1659     | 1656   | \(\beta\)-Eudesmol      | —                                 | —                                 | —                              | —                                 | 1.7                           | 0.3                         |
| 1663     | 1664   | \(\alpha\)-Turmerone    | —                                 | 0.2                               | 0.3                            | 0.4                               | —                             | —                           |
| 1667     | 1668   | \(\alpha\)-Turmerone    | —                                 | —                                 | —                              | —                                 | —                             | 8.3                         |
| 1668     | 1667   | \(\alpha\)-Bisabolol    | —                                 | 0.4                               | —                              | 0.4                               | —                             | 2.9                         |
| 1695     | 1694   | Germacrene              | 14.7                              | 10.7                              | 14.5                           | 12.5                              | 4.9                           | 3.8                         |
| 1700     | 1699   | \(\beta\)-Turmerone (=Curline) | —                                 | —                                 | —                              | —                                 | —                             | 8.3                         |
| 1714     | 1712   | Curcuphenol             | —                                 | —                                 | —                              | —                                 | 5.0                           | 6.9                         |
| 1718     | 1713   | Curdione                | 1.3                               | 1.6                               | 1.3                            | 1.6                               | 28.7                          | 35.6                        |
| 1724     | 1723   | Curcumulone             | 4.6                               | 3.7                               | 7.1                            | 4.3                               | —                             | 19.8                        |
| 1733     | 1731   | Zerumbone               | 1.1                               | 0.7                               | 1.1                            | 1.0                               | —                             | 17.7                        |
| 1738     |        | Unidentified a          | 1.1                               | 0.7                               | 1.1                            | 1.0                               | —                             | 12.9                        |
| 1742     |        | Unidentified a          | —                                 | —                                 | —                              | —                                 | 0.1                           | —                           |
| 1743     | 1742   | (6S,7R)-Bisabolone      | 0.3                               | 0.2                               | 0.4                            | 0.3                               | —                             | 0.2                         |
| 1745     |        | Unidentified a          | 2.1                               | 1.6                               | 1.8                            | 1.9                               | 1.4                           | —                           |
| 1747     | 1750   | Xanthorrhizol           | —                                 | —                                 | —                              | —                                 | 0.6                           | 0.4                         |
| 1749     |        | Unidentified a          | —                                 | —                                 | —                              | —                                 | —                             | 0.2                         |
| 1752     |        | Unidentified a          | —                                 | —                                 | —                              | —                                 | 1.6                           | 0.6                         |
| 1771     | 1771   | trans-\(\alpha\)-Atlantone | —                                 | —                                 | —                              | —                                 | 2.1                           | 1.4                         |
| 1777     | 1775   | Curzerenone A           | —                                 | —                                 | —                              | —                                 | 0.2                           | 0.1                         |
| 1778     |        | Unidentified a          | 9.0                               | 8.7                               | 11.0                           | 10.3                              | —                             | 0.7                         |
| 1788     |        | Unidentified a          | 1.4                               | 1.6                               | 1.7                            | 1.5                               | 0.3                           | 0.3                         |
| 1827     |        | Unidentified a          | 1.6                               | 0.6                               | 1.0                            | 0.9                               | 0.1                           | 0.2                         |
| 1834     |        | Curcumulone             | 3.1                               | 3.1                               | 4.2                            | 3.8                               | 3.0                           | 4.5                         |
| 1836     |        | 4,5-Epoxygermacrone     | 2.1                               | 1.0                               | 1.3                            | 1.3                               | 0.3                           | 0.4                         |
| 1929     |        | Unidentified a          | —                                 | —                                 | —                              | —                                 | 4.4                           | 2.3                         |
| 1987     |        | Zederone               | 0.4                               | 0.4                               | 0.6                            | 0.6                               | 0.6                           | 0.9                         |
| 1992     |        | Unidentified a          | —                                 | —                                 | —                              | —                                 | 0.3                           | 0.5                         |

*Note:* a = tentative identification

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Table 2. Cont.

| RI<sub>calc</sub> | RI<sub>db</sub> | Compound | Curcuma aromatica (Green Rhizome) | Curcuma aromatica (White Rhizome) | Curcuma caesia (Black Rhizome) | Curcuma zanthorrhiza (Lime Rhizome) | Curcuma longa (Yellow Rhizome) | Curcuma longa (Red Rhizome) |
|------------------|----------------|----------|----------------------------------|----------------------------------|-------------------------------|----------------------------------|-------------------------------|-------------------------------|
|                  |                |          | CA22 (M)                         | CA22 (D)                         | CA46 (M)                      | CC38 (M)                        | CZ44 (M)                      | CL56 (M)                      |
|                  |                |          | CA22 (D)                         | CA46 (D)                         | CC38 (D)                      | CZ44 (D)                        | CL56 (D)                      | CL63 (M)                      |
|                  |                |          | CL63 (D)                         |                                  |                               |                                 |                               |                               |
| 2345             | —              | (E)-Labda-8(17),12-diene-15,16-dial | —                                | —                                | —                             | 1.0                             | 0.2                           | —                             |
|                  |                | Monoterpenes hydrocarbons           | 4.2                              | 8.9                              | 4.1                           | 8.1                             | 0.4                           | 0.5                           |
|                  |                | Oxygenated monoterpenoids           | 15.5                             | 10.8                             | 12.7                          | 8.1                             | 2.3                           | 0.6                           |
|                  |                | Sesquiterpenes hydrocarbons         | 10.3                             | 20.3                             | 10.7                          | 16.0                            | 11.6                          | 7.9                           |
|                  |                | Oxygenated sesquiterpenoids         | 52.8                             | 44.8                             | 54.0                          | 49.7                            | 76.1                          | 83.2                          |
|                  |                | Diterpenes                          | 0.0                              | 0.0                              | 0.0                           | 1.0                             | 0.0                           | 0.0                           |
|                  |                | Benzenoid aromatics                 | 0.0                              | 0.0                              | 0.0                           | traces                          | traces                        | traces                        |
| Others           | 1.0            | 1.6                                | 0.3                              | 0.6                              | 4.0                           | 0.9                             | 0.5                           | 0.6                           |
| Total identified | 83.8           | 86.4                               | 81.8                             | 83.4                             | 94.4                          | 93.2                            | 94.5                          | 91.4                          |

RI<sub>calc</sub> = Retention index determined with respect to a homologous series of n-alkanes on a ZB-5ms column. RI<sub>db</sub> = Retention index from the databases [25–28]. M = mother (main) rhizome. D = daughter (finger) rhizome. tr = trace (<0.05%). *Mass spectra of the unidentified components are presented in Supplementary Figure S2. The major components are highlighted in **bold font.**
Likewise, the lime-colored rhizome essential oils of *C. zanthorrhiza* (CZ44,) were also rich in curzerenone (16.3% and 19.7%) and curdione (19.8% and 17.7%), in addition to germacrone (11.3% and 11.1%) for the mother and daughter rhizomes, respectively. This composition is in marked contrast to that reported for *C. zanthorrhiza* from Thailand with 1,8-cineole (37.6%) and curzerenone (13.7%) as the major components [30]. 1,8-Cineole was in lower concentration (7.2% and 2.5% for the mother and daughter rhizomes) in *C. zanthorrhiza* in this work. A *C. zanthorrhiza* rhizome essential oils from Bogor, Indonesia, on the other hand, was dominated by xanthorrhizol (26.8%), β-curcumene (17.0%), *ar*-curcumene (15.1%), camphor (9.1%), and germacrone (5.4%) [35]. Another sample of *C. zanthorrhiza* from West Java, Indonesia, was composed of β-curcumene (23.4%), *ar*-curcumene (22.1%), curzerene (6.0%), camphor (5.0%), and xanthorrhizol (4.7%) as the dominant constituents [36]. Neither β-curcumene nor xanthorrhizol were detected in the essential oil sample cultivated in North Alabama.

The major components of the rhizome essential oils from *C. longa* CL56 (yellow-colored rhizome) were α-turmerone (12.7% and 14.1%), α-zingiberene (11.4% and 13.9%), *ar*-turmerone (8.3% and 12.6%), and β-sesquiphellandrene (8.9% and 10.0%). The red-colored rhizome variety of *C. longa* (CL63) also yielded essential oils rich in *ar*-turmerone (36.1% and 31.3%), and α-turmerone (15.2% and 13.0%), as well as β-turmerone (=curlone) (15.4% and 13.0%). By comparison, the rhizome essential oils of *C. longa* cultivated in North Alabama, reported previously, showed α-turmerone (13.6–31.5%), *ar*-turmerone (6.8–32.5%), β-turmerone (4.8–18.4%), *α*-phellandrene (3.7–11.8%), 1,8-cineole (2.6–11.7%), *α*-zingiberene (0.9–12.5%), and β-sesquiphellandrene (0.7–8.0%) [9]. Two distinct chemical variations were found in the previous examination of *C. longa* cultivated in North Alabama. One group was dominated by turmerones (α-turmerone, *ar*-turmerone, and β-turmerone), while the second group had lower concentrations of turmerones but high concentrations of α-zingiberene and β-phellandrene. Thus, the red-colored rhizome (CL63) belongs to the turmerone-rich chemical group, while the yellow-colored rhizome (CL56) belongs to the second group (high in *α*-zingiberene).

In order to place the volatile phytochemistry of the *Curcuma* rhizome essential oils in this study into perspective, an agglomerative hierarchical cluster analysis (HCA) was carried out based on the relative concentrations of the major components (Figure 2). There are two clearly defined clusters with at least 50% similarity based on the HCA: Cluster 1 is a cluster made up of CA22 (green rhizome), CA46 (*C. aromatica*, white rhizome) CC38 (*C. caesia*, black rhizome), and CZ44 (*C. zanthorrhiza*, lime rhizome), essential oils and defined by relatively high concentrations of curzerenone (14.7–29.1%), curdione (1.3–35.6%), and germacrone (3.8–14.7%); and Cluster 2, a cluster of CL56 (*C. longa*, yellow rhizome) and CL63 (*C. longa*, red rhizome) rhizome essential oils that were dominated by *ar*-turmerone (8.3–36.1%), α-turmerone (12.7–15.2%), and β-turmerone (5.0–15.4%) (see Table 2).

Interestingly, the volatile phytochemistry of *C. caesia* and *C. zanthorrhiza* rhizomes are very similar (about 90% similarity). Likewise, the green- and white-colored rhizome essential oils of *C. aromatica* are very similar (about 95% similarity). The yellow- and red-colored rhizome essential oils of *C. longa* showed somewhat lower similarity (about 60% similarity).

There are some significant differences in the concentrations of the major components in Cluster 1 (Figure 3). The concentration of curzerenone is significantly greater in *C. caesia* than in either *C. aromatica* or *C. zanthorrhiza*. The concentrations of curdione in *C. aromatica* are significantly lower than those in either *C. caesia* or *C. zanthorrhiza*. Germacrone was significantly lower in *C. caesia* than in either *C. aromatica* or *C. zanthorrhiza*. 

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There are some significant differences in the concentrations of the major components in Cluster 1 (Figure 3). The concentration of curzerenone is significantly greater in *C. caesia* than in either *C. aromatica* or *C. zanthorrhiza*. The concentrations of curdione in *C. aromatica* are significantly lower than those in either *C. caesia* or *C. zanthorrhiza*. Germacrone was significantly lower in *C. caesia* than in either *C. aromatica* or *C. zanthorrhiza*. 

In order to place the volatile phytochemistry of the *Curcuma* rhizome essential oils in this study into perspective, an agglomerative hierarchical cluster analysis (HCA) was carried out based on the relative concentrations of the major components (Figure 2). There are two clearly defined clusters with at least 50% similarity based on the HCA: Cluster 1 is a cluster made up of CA22 (green rhizome), CA46 (*C. aromatica*, white rhizome) CC38 (C. caesia, black rhizome), and CZ44 (C. zanthorrhiza, lime rhizome), essential oils and defined by relatively high concentrations of curzerenone (14.7–29.1%), curdione (1.3–35.6%), and germacrone (3.8–14.7%); and Cluster 2, a cluster of CL56 (C. longa, yellow rhizome) and CL63 (C. longa, red rhizome) rhizome essential oils that were dominated by *ar*-turmerone (8.3–36.1%), α-turmerone (12.7–15.2%), and β-turmerone (5.0–15.4%) (see Table 2).

Interestingly, the volatile phytochemistry of *C. caesia* and *C. zanthorrhiza* rhizomes are very similar (about 90% similarity). Likewise, the green- and white-colored rhizome essential oils of *C. aromatica* are very similar (about 95% similarity). The yellow- and red-colored rhizome essential oils of *C. longa* showed somewhat lower similarity (about 60% similarity).

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are the concentrations of *C. longa*.

A cluster made up of CA22 (green rhizome), CA46 (white rhizome), and CA22 (green rhizome) is very similar (about 90% similarity). Likewise, the green- and white-colored rhizome essential oils of *Curcuma aromatica* are very similar (about 95% similarity). The yellow- and red-colored rhizome essential oils in *Curcuma aromatica* or *C. zanthorrhiza* showed somewhat lower similarity (about 60% similarity), they are comparable to the respective chemical profiles of the yellow- and red-colored rhizomes of *Curcuma longa*, which are notably different (60% similarity), they are comparable to the respective chemical profiles of *C. longa* from tropical Asian collections [3].

There are some significant differences in the concentrations of the major components (Figure 3). The significant differences between the essential oils of yellow- and red-colored *C. longa* are the concentrations of α-turmerone (much higher in the red rhizome variety) and β-turmerone (also higher in the red rhizome variety). The concentrations of α-turmerone in the red and yellow varieties are not significantly different (Figure 4).

Figure 2. Dendrogram obtained from the agglomerative hierarchical cluster analysis of the rhizome essential oil compositions of Vietnamese *Curcuma* species cultivated in North Alabama.

Figure 3. Comparison of the main chemical components of *Curcuma aromatica*, *Curcuma caesia*, and *Curcuma zanthorrhiza*. For each chemical component, bars with the same letter are not significantly different at $p \leq 0.05$.

The significant differences between the essential oils of yellow- and red-colored *C. longa* are the concentrations of α-turmerone (much higher in the red rhizome variety) and β-turmerone (also higher in the red rhizome variety). The concentrations of α-turmerone in the red and yellow varieties are not significantly different (Figure 4). Nevertheless, although the compositions of yellow- and red-colored rhizomes of *C. longa* are notably different (60% similarity), they are comparable to the respective chemical profiles of *C. longa* from tropical Asian collections [3].
Figure 3. Comparison of the main chemical components of Curcuma aromatica, Curcuma caesia, and Curcuma zanthorrhiza. For each chemical component, bars with the same letter are not significantly different at $p \leq 0.05$.

The significant differences between the essential oils of yellow- and red-colored C. longa are the concentrations of $\alpha$-turmerone (much higher in the red rhizome variety) and $\beta$-turmerone (also higher in the red rhizome variety). The concentrations of $\alpha$-turmerone in the red and yellow varieties are not significantly different (Figure 4). Nevertheless, although the compositions of yellow- and red-colored rhizomes of C. longa are notably different (60% similarity), they are comparable to the respective chemical profiles of C. longa from tropical Asian collections.

Figure 4. Comparison of the main chemical components of Curcuma longa. For each chemical component, bars with the same letter are not significantly different at $p \leq 0.05$.

3.2. Enantiomeric Distribution of Terpenoids in Curcuma Essential Oils

The enantiomeric distributions of terpenoid components in Curcuma rhizome essential oils have been determined by enantioselective GC-MS (Table 3). Although only found in trace quantities, when detected by chiral GC-MS (C. aromatica, C. caesia), the $(-)\alpha$-thujene predominated. $(-)\alpha$-Pinene was the dominant enantiomer in all samples. $(-)\beta$-Pinene also predominated in all samples, but was especially dominant in the essential oils of C. zanthorrhiza (CZ44) and C. longa (CL56 and CL 63). ($+$)-Camphene was the dominant enantiomer in C. aromatica, C. caesia, and C. zanthorrhiza.

$\alpha$-Phellandrene and $\delta$-3-carene, only detected in the essential oils of C. longa, were both exclusively the ($+$) enantiomers. $(-)$-Limonene was the major stereoisomer in the Curcuma essential oils, but was nearly racemic in C. zanthorrhiza. $(-)$-Sabinene predominated is all Curcuma rhizome essential oils where it was detected. $(-)$-Linalool was the major enantiomer in nearly all Curcuma samples, but was particularly abundant in C. caesia (CC38), C. zanthorrhiza (CZ44), and the yellow-rhizome C. longa (CL56). Interestingly, however, the red-rhizome C. longa (CL63) exhibited ($+$)-linalool as the major enantiomer. Likewise, $(-)\alpha$-terpineol was the dominant enantiomer in all Curcuma essential oils. Camphor was not found in C. longa, but ($+$)-camphor was the dominant enantiomer in C. aromatica, C. caesia, and C. zanthorrhiza. The major enantiomer of terpinen-4-ol in Curcuma essential oils was $(-)$-terpinen-4-ol, although the distribution was nearly racemic in C. zanthorrhiza.

$\delta$-Elemene was nearly racemic in all of the Curcuma essential oils, whereas trans-$\beta$-elemene was exclusively the $(-)$ enantiomer. Both (E)$\beta$-caryophyllene and $\delta$-cadinene were 100% $(-)$ enantiomers, while germacrene D and $\beta$-bisabolene were exclusively the dextrorotatory stereoisomers.
Table 3. Enantiomeric distribution of terpenoid components in Vietnamese *Curcuma* rhizome essential oils cultivated in North Alabama.

| Compound                  | *Curcuma aromatica* (CA22, Green Rhizome) | *Curcuma aromatica* (CA46, White Rhizome) | *Curcuma caesia* (CC38, Black Rhizome) |
|---------------------------|-------------------------------------------|-------------------------------------------|----------------------------------------|
|                           | (+) (−)                                   | (+) (−)                                   | (+) (−)                                 |
| **Mother**                |                                            |                                            |                                        |
| **Daughter**              |                                            |                                            |                                        |
| ** (+) **                  |                                            |                                            |                                        |
| **(−) **                  |                                            |                                            |                                        |
| α-Thujene                 | 14.1 88.9 12.5 87.5 15.9 84.1              | −                                         | 18.6 81.4                              |
| α-Pinene                  | 23.5 76.5 26.1 73.9 21.7 78.3              | 24.5 75.5                                 | −                                       |
| Camphene                  | 85.0 15.0 81.3 18.7 83.4 16.6              | 82.5 17.5                                 | −                                       |
| Sabinene                  | 28.3 71.7 36.6 63.4 33.7 66.3              | 63.0 37.0                                 | 17.8 82.2                              |
| β-Pinene                  | 47.8 52.2 43.3 56.7 41.3 58.7              | 60.0 40.0                                 | 43.5 56.5                              |
| α-Phellandrene            | −                                          | −                                         | −                                       |
| δ-3-Carene                | −                                          | −                                         | −                                       |
| Limonene                  | 19.2 80.8 32.2 67.8 19.7 80.3              | 33.8 66.2                                 | 19.4 85.1                              |
| Linalool                  | 39.7 60.3 20.3 79.7 48.5 51.5              | 29.2 70.8                                 | 0.0 100.0                              |
| Camphor                   | 99.1 0.9 99.2 0.8 99.1 0.9                 | 99.3 0.7                                  | 100.0 0.0                              |
| Terpinen-4-ol             | 30.7 69.3 37.0 63.0 29.9 70.1              | 39.4 60.6                                 | 25.1 74.9                              |
| δ-Elemene                 | 48.4 51.6 48.1 51.9 47.3 52.7              | 48.1 51.9                                 | 49.1 50.9                              |
| α-Terpineol               | 20.6 79.4 31.7 68.3 28.5 71.5              | 20.7 79.3                                 | 16.3 83.7                              |
| Carvone                   | 23.5 76.5 25.5 74.5                        | −                                         | −                                       |
| (E)-β-Elemene             | 0.0 100.0                                  | 25.5 74.5                                 | −                                       |
| (E)-β-Caryophyllene       | 0.0 100.0                                  | 0.0 100.0                                 | 0.0 100.0                              |
| Germacrene D              | 100.0 0.0 100.0 0.0 100.0 0.0              | 0.0 100.0                                 | 0.0 100.0                              |
| β-Bisabolene              | 100.0 0.0 100.0 0.0 100.0 0.0              | 0.0 100.0                                 | 0.0 100.0                              |
| δ-Cadinene                | 0.0 100.0                                  | 0.0 100.0                                 | 0.0 100.0                              |
| (E)-Nerolidol             | −                                          | −                                         | −                                       |

| Compound                  | *Curcuma zanthorrhiza* (CZ44, lime rhizome) | *Curcuma longa* (CL56, yellow rhizome) | *Curcuma longa* (CL63, red rhizome) |
|---------------------------|-------------------------------------------|----------------------------------------|-------------------------------------|
|                           | (+) (−)                                   | (+) (−)                                 | (+) (−)                             |
| **Mother**                |                                            |                                        |                                    |
| **Daughter**              |                                            |                                        |                                    |
| ** (+) **                  |                                            |                                        |                                    |
| **(−) **                  |                                            |                                        |                                    |
| α-Thujene                 | −                                          | −                                       | −                                    |
| α-Pinene                  | 21.6 78.4 11.7 88.3 11.9 88.1              | 11.4 88.6                               | 34.7 65.3                            |
| Camphene                  | 91.6 8.4 90.1 9.9                          | 9.9 90.1                                | 80.5 19.5                            |
| Sabinene                  | −                                          | 20.6 79.4                              | 19.5                                 |
| β-Pinene                  | 2.8 97.2 2.5 97.5 12.3 87.7              | 11.2 88.8                               | 24.2 75.8                            |
| α-Phellandrene            | −                                          | 100.0 0.0                              | 100.0 0.0                            |
| δ-3-Carene                | −                                          | 100.0 0.0                              | 100.0 0.0                            |
| Limonene                  | 41.3 58.7 42.7 57.3 12.5 87.5              | 12.2 87.8                              | 22.9 77.1                            |
| Linalool                  | 10.9 89.1 9.0 91.0 17.9 82.1              | 0.0 100.0                              | 82.0 18.0                            |
Table 3. Cont.

| Compound            | Curcuma zanthorrhiza (CZ44, lime rhizome) | Curcuma longa (CL56, yellow rhizome) | Curcuma longa (CL63, red rhizome) |
|---------------------|------------------------------------------|-------------------------------------|----------------------------------|
|                     | Mother                          | Daughter                          | Mother                          | Daughter                          | Mother                          | Daughter                          |
|                     | (+)       | (-)      | (+)       | (-)      | (+)       | (-)      | (+)       | (-)      | (+)       | (-)      | (+)       | (-)      |
| Camphor             | 99.6     | 0.4      | 99.7     | 0.3      | —         | —        | 26.4     | 73.6     | —         | —        | —         | —        |
| Terpinen-4-ol       | 42.6     | 57.4     | 45.7     | 54.3     | 11.0      | 89.0     | 26.0     | 74.0     | —         | —        | —         | —        |
| δ-Elemene           | 45.9     | 54.1     | 47.6     | 52.4     | 44.3      | 55.7     | 48.8     | 51.2     | —         | —        | —         | —        |
| α-Terpineol         | 17.3     | 82.7     | 15.3     | 84.7     | 11.0      | 89.0     | 10.6     | 89.4     | 41.5      | 58.5     | 13.1      | 86.9     |
| Carvone             | —        | —        | —        | —        | —         | —        | —        | —        | —         | —        | —         | —        |
| trans-β-Elemene     | 0.0      | 100.0    | 0.0      | 100.0    | 0.0       | 100.0    | 0.0      | 100.0    | —         | —        | 0.0       | 100.0    |
| (E)-β-Caryophyllene | 0.0      | 100.0    | 0.0      | 100.0    | 0.0       | 100.0    | 0.0      | 100.0    | 0.0       | 100.0    | 0.0       | 100.0    |
| Germacrene D        | 100.0    | 0.0      | 98.8     | 1.2      | 100.0     | 0.0      | 100.0    | 0.0      | —         | —        | 0.0       | 100.0    |
| β-Bisabolene        | 100.0    | 0.0      | 100.0    | 0.0      | 91.9      | 8.1      | 92.0     | 8.0      | 92.6      | 7.4      | 91.6      | 8.4      |
| δ-Cadinene          | 0.0      | 100.0    | 0.0      | 100.0    | —         | —        | —        | —        | —         | —        | 0.0       | 100.0    |
| (E)-Nerolidol       | —        | —        | —        | —        | —         | —        | 0.0      | 100.0    | 0.0       | 100.0    | —         | —        |

(+) = dextrorotatory enantiomer, (−) = levorotatory enantiomer.
4. Conclusions

The rhizome essential oils of C. aromatica, C. caesia, C. longa, and C. zanthorrhiza that were cultivated in North Alabama show wide variation in composition compared to essential oils from other geographical locations. Nevertheless, the essential oil yields and compositions provide evidence that Curcuma can be successfully cultivated in North Alabama and may provide additional sources of these species for both culinary and herbal medicinal uses. The knowledge of their relative oil yields and compositions could help in value-addition for either fresh rhizomes or dry herbal markets. The four species showed specific essential oil components, which are known to have extensive pharmacological activity separately or in combination with curcuminoids. The species can be used to tailor herbal medicines to combat particular ailments. The cultivation of specific varieties to cater to niche markets could not only benefit the farmers, but also have an impact on the socio-economic sustainability of rural Alabama in particular and the southeastern U.S. in general. As far as we are aware, this is the first report of the essential oil compositions, including enantiomeric distributions for these Curcuma species cultivated in North America. Among the four species, the C. longa species that combine high yield with high curcumin have been found to be suitable for cultivation. However, the remaining species have economic potential, for example C. zanthorrhiza is well known for its antimicrobial activity against common human pathogens to cater to herbal companies interested in varieties that are high in a certain essential oil component.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/horticulturae8050360/s1, Figure S1: Gas chromatograms of rhizome essential oils of Curcuma varieties cultivated in North Alabama; Figure S2: Mass spectra of unidentified components of Curcuma rhizome essential oils.

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