

Aromatic and cannabinoid profiles of *Cannabis* inflorescences and seed oils: A comprehensive approach for variety characterization

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ABSTRACT

In this study, an exhaustive analysis of the species *Cannabis sativa* L. was carried out, focusing on the terpenoid and cannabinoid profiles of inflorescences from five different varieties and four intra-specific hybrids. In addition, terpenoid and cannabinoid compositions of seed oils from these same *Cannabis* varieties were examined. Aromatic compounds were analysed using gas chromatography-mass spectrometry (GS-MS) while cannabinoids were by high-performance liquid chromatography coupled to diode array detector (HPLC-DAD). Principal component analysis (PCA) and hierarchical analysis were performed to determine the relationship between aromatic compounds within varieties for classification. A total of 71 naturally occurring aromatic compounds were identified, including 27 terpenoids and 10 isomers. Terpenoid clustering from the different inflorescence groups confirmed the commercial aromatic description of each *Cannabis* variety; however, they were not associated with the clusters observed in the cannabinoid profiles. Seed oils contained trace amounts of both aromatic compounds and cannabinoids, suggesting a migration of components during the industrial extraction process. This study contributes to understanding cannabis chemistry and emphasizes the importance of ongoing research of cannabis-derived products throughout comprehensive analysis of cannabinoid content, and especially aromatic profile in varieties. It can guide the development of targeted breeding programs, complex classification of varieties and formulation of cannabis-based products tailored to specific applications.

1. Introduction

Cannabis (*Cannabis sativa* L.) is a plant of significant historical and scientific importance, extensively studied for its medicinal potential. Belonging to the Cannabaceae family and originating from the Central Asia region, cannabis has been cultivated globally due to its remarkable adaptability and resource-efficient growth (Naz et al., 2017).

In 1753, Carolus Linnaeus described the hemp plant cultivated in Europe, which he named *Cannabis sativa*. Its appearance was very tall (2–4 m) and little branched, with separate internodes, low leaf density, thin leaflets, and slow maturation. Among its uses was the production of grain and fibre. In 1785, Jean-Baptiste Lamarck described in India what he believed to be another species in the genus *Cannabis*, which he named *Cannabis indica*. The plants had a more compact, rounded appearance,

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with high leaf density, with large leaves and wide leaflets, with highly developed inflorescences and shorter maturation. Later, it was determined that botanically there is only one species, *Cannabis sativa* L. The variation of the morphological characteristics indicated above does not allow differentiation at the specific level, although some taxonomists insist on distinguishing subspecies such as *C. sativa* subsp. *sativa* and *C. sativa* subsp. *indica*. In addition, today the existence of multiple "intervarietal hybrids" or "intraspecific hybrids" makes it impossible a classification according to the appearance of the buds, shape, colour or size of the leaves and plants (McPartland, 2017; McPartland and Guy, 2017).

Throughout centuries, the therapeutic botanical properties of cannabis have been harnessed, primarily through the consumption of its inflorescences, which contain a high content of secondary metabolites (Andre et al., 2016; Birenboim et al., 2022; García-Valverde et al., 2020; Rice and Koziel, 2015). The main therapeutic properties of cannabis are attributed to cannabinoids, such as Δ^9 -tetrahydrocannabinol (Δ^9 -THC) and cannabidiol (CBD) (Andre et al., 2016; Gallily et al., 2018; Richins et al., 2018; Russo and Marcu, 2017). These compounds interact with the CB1 and CB2 receptors found in the central and peripheral nervous systems, exerting diverse therapeutic effects (Russo and Marcu, 2017).

These cannabinoids were often considered the main chemicals involved in the therapeutic properties and psychoactive effects associated with cannabis, remaining the only ones screened when assessing cultivar varieties (Solowij et al., 2019). However, increasing evidence supports the relevance of terpenes, including monoterpenes and sesquiterpenes, which contribute to its distinctive aroma and serve as natural defence mechanisms against predators, growth modulation, disease resistance, attraction of pollinators, plant-plant communication, and antioxidant properties (Hanuš and Hod, 2020; Nonier et al., 2004; Richins et al., 2018; Sommano et al., 2020).

In this sense, the analysis of terpenes may contribute to specific cultivars classifications which may be complementary to the determination of cannabinoids. Unlike cannabinoids, terpenoids may be used as biomarkers of chemosystematics studies to characterize plant samples because they do not vary among generations of the same strains, the sex, age, and part of the plant (Birenboim et al., 2022). Moreover, recent scientific studies have shed light on the pharmacological benefits of terpenoids, unveiling their antimicrobial, antioxidant, and anti-inflammatory properties (Booth et al., 2017; Fishedick, 2020; Gallily et al., 2018). Also, the entourage effect, arising from the synergistic interaction between cannabinoids and terpenoids, is believed to enhance the therapeutic efficacy of cannabis-derived products (Birenboim et al., 2022).

Although there are complex legal considerations surrounding the use of cannabis and cannabis-derived products, such as cannabis seed oils, there is a growing interest among consumers in utilizing cannabis dietary supplements, especially for their CBD content (Pavlovic et al., 2018). Among these products, seed oils and preparations have received attention due to their ability to adjust the individual administration dose required throughout the treatment period and their enhanced bioavailability of cannabinoids and non-cannabinoids like omega-3 fatty acids, terpenes and flavonoids (Aiello et al., 2020; Andre et al., 2016; Citti et al., 2019). Despite their bioactive potential, cannabis and cannabis-derived products are often limited in their use due to the psychoactive compound Δ^9 -THC, which is known for its intoxicating effects and is classified as a controlled substance in many jurisdictions (Fishedick, 2020; García-Valverde et al., 2020).

Overall, the presence of Δ^9 -THC in cannabis leads to regulatory restrictions on all cannabis-derived products, regardless of their Δ^9 -THC content (Andrews and Paterson, 2012). This limitation affects the commercial availability and consumption of other cannabis-derived products that may not contain Δ^9 -THC or have lower Δ^9 -THC concentrations that allowed regulations (Al Bakain et al., 2020; Salazar-Bermeo et al., 2023). The aim is to ensure public safety, prevent misuse, and comply with legal frameworks governing the use of psychoactive

substances like Δ^9 -THC (Hall, 2018).

The analysis of terpenoids and cannabinoids and their relationship between the *Cannabis* plant and other derived products, such as seed oils, is of utmost importance in classifying *Cannabis* varieties and accurately assessing the presence of Δ^9 -THC, even in products derived from plants with high Δ^9 -THC content (Aiello et al., 2020a; Citti et al., 2019; Gouvêa-Silva et al., 2023). By employing comprehensive analytical techniques such as gas chromatography-mass spectrometry (GC-MS) and high-performance liquid chromatography (HPLC), the chemical composition of *Cannabis* varieties can be thoroughly examined, enabling the identification and quantification of terpenoids and cannabinoids (Al Bakain et al., 2020; Birenboim et al., 2022; Citti et al., 2019; Delgado-Povedano et al., 2020).

This approach allows for strain characterization and quality control, facilitating the classification of *Cannabis* varieties based on their specific chemical profiles. Furthermore, by analysing seed oils derived from these *Cannabis* varieties, it becomes possible to determine the transfer of bioactive compounds, including Δ^9 -THC, into the oil matrix and it ensures that cannabis-derived products, even those without detectable Δ^9 -THC levels, meet the necessary standards and comply with legal requirements (Aiello et al., 2020a; Al Bakain et al., 2020; Citti et al., 2019; Gouvêa-Silva et al., 2023; Salazar-Bermeo et al., 2023).

The aim of this study was to analyse the chemical diversity exhibited by nine *Cannabis* varieties, shedding light on the distinctive profiles displayed by inflorescences and seed oils. These findings will not only aid in strain characterization and quality control but also provide valuable insights for targeted breeding programs and the development of cannabis-based products tailored to specific therapeutic and industrial applications (Calvi et al., 2018).

2. Experimental

2.1. Plant material

All nine (9) inflorescence samples of cannabis were purchased from T.H. Seeds (Amsterdam, Netherlands), that controlled the cultivation conditions of each variety. Fresh inflorescences from five plants per cultivar, at their optimal maturity (end of September), were used for this study. Five (5) *Cannabis* varieties [Bubble gum (BB), Stracciatella (STC), Kitne 2 (K2), Gelato 33 (G33), and Birthday cake (BC)] and four (4) intraspecific hybrids [Nicole x Bubble gum (BVD), Gelato 33 x Straw Banana Cream (SBC), L.A. S.A.G.E. (LS), and French cookies (FC)] were analysed. Their commercial aromatic description was determined by trained panellists from T.H. Seeds (Table 1). The samples were freeze dried and stored at $-20\text{ }^{\circ}\text{C}$ until analysis.

The oil from germinated seeds of the same varieties were also provided by T.H. Seeds and were obtained by cold-press procedures. The samples were maintained at $-20\text{ }^{\circ}\text{C}$ until further analysis.

Table 1
Predominant aromatic description of each variety.

Variety	Type	Aromatic description
BB	Sativa	Fruity smell, floral, berry-like, sweet
BVD	Hybrid	Citrus smell, floral, berry-like, sweet
STC	Sativa	Candy, sweet, creamy, cocoa
K2	Sativa	Hash-like scent, pungent
G33	Sativa	Earthy, gassy, sweet, creamy, herbal, mint
SBC	Hybrid	Citric, sweet, earthy
LS	Hybrid	Pine, sage, sweet
BC	Sativa	Creamy, vanilla, baked, citrus, sweet,
FC	Hybrid	Grape, fruity, baked, creamy, sweet, gassy, earthy

Varieties: Nicole x Bubble gum (BVD), Bubble gum (BB), Stracciatella (STC), Kitne 2 (K2), Birthday cake (BC), Gelato 33 (G33), Gelato 33 x Straw Banana Cream (SBC), L.A.S.A.G.E. (LS), and French cookies (FC).

2.2. Reagents

Certified cannabinoid standards: Δ^9 -THC, tetrahydrocannabinolic acid (THCA), tetrahydrocannabivarin (THCV), CBD, cannabidiolic acid (CBDA), cannabinol (CBN), cannabigerol (CBG), cannabigerolic acid (CBGA) and cannabichromene (CBC) were purchased from Cayman (Barcelona, Spain), Sigma Aldrich (Madrid, Spain), and Cerilliant Corporation (Madrid, Spain). Membrane filters (0.45 μm) were supplied by Merck (Darmstadt, Germany). Ethanol (99.8%), acetonitrile (HPLC grade), acetone ($\geq 99.9\%$), and methanol (HPLC grade) were purchased from PanReac (Barcelona, Spain). Distilled water used in this study was purified by Milliport-Q system. Thermal desorption tubes Supelco Tenax TA were obtained from Sigma Aldrich.

2.3. Extraction of phytochemicals

2.3.1. Extraction of terpenoids from inflorescences and seed oils

The trapping of aromatic compounds from inflorescence and seed oil samples was conducted in a Tenax TA adsorbent tube. Briefly, 50 mg of dry inflorescence or seed oil were homogenised with 5 mL solution of distilled water/methanol/acetone (5:4:1) for 60 min at 50 °C and coupled to a nitrogen stream at a flow rate of 30 mL/min. After sample trapping, the tube was desorbed in thermal desorption unit (TDU) on a GC-MS system. The sorbent tubes were conditioned for a minimum of 15 min at 330 °C prior to each use according to (Fischedick, 2020; García-Valverde et al., 2020; Zhai and Granvogl, 2019).

2.3.2. Extraction of cannabinoids

For cannabinoid extraction, the inflorescences were manipulated according to (Salazar-Bermeo et al., 2023; UNODC, 2014). The dried material was grounded and extracted with 99.8% ethanol using sonication for 15 min. For seed oil samples, 100 mg of oil were mixed with 10 mL of isopropanol. Then, 10 μL of each sample were diluted in 990 μL of 99.8% of ethanol. The extracts were 0.45 μm -filtered, transferred to a vial, and stored at -20 °C until analysis.

2.4. Analysis of aromatic compounds by GC-MS

For the analysis of aromatic compounds in both inflorescences and seed oils from cannabis, the Shimadzu QP 2010 Plus, equipped with a TDU and DB-5 ms capillary column (30.00 m x 0.25 mm x 0.25 μm). Helium was used as carrier gas at constant flow of 1.01 mL/min and linear velocity of 36.2 cm/s. The oven temperature was programmed from 40 °C (6 min) to 200 °C (8 min) at 10 °C/min and to 240 °C (2 min) at 30 °C/min (33.33 min total). The injector was set at 30 °C and the inlet mode was split. A mass range of 40–400 m/z , the filament voltage was 70 eV while interface and ion source temperature was 230 °C (Rice and Kozziel, 2015). After analysis, GC-MS Solution Software vs. 4.52 (Shimadzu Corporation) was used for data acquisition, data processing and instrument control. The substances were identified by means of a mass with more than 95% of similarity according to The National Institute of Standards and Technology (NIST, 2017) database (NIST, 2017) and Scientific Working Group for the Analysis of Seized Drugs database (SWGDRUG, 2021). The mean values of the percentage area (triplicate) of every compound peak were used for statistical analysis.

The odour description of each identified compound was acquired from Flavornet database (Acree and Arn, 2004), Nature Derived Bioactive Molecules Database of Bangladesh (GreenMol BD) (Hosen et al., 2022), Food database (FOODB) (TMIC, 2021), Human Metabolome database (HMDB) (Wishart et al., 2022) and The Good Scents Company database (TGSC) (TGSC, 2021).

2.5. Analysis of cannabinoids by HPLC coupled to diode array detector (DAD)

The cannabinoids were separated using an Agilent series 1200 apparatus (Santa Clara, California, USA), which is coupled with a temperature-controlled autosampler, binary pump, and DAD. The separation was achieved on a Poroshell column 20 SB-C18, 4.6 x 150 mm, 2.7 μm . Under gradient conditions at 0.5 mL/min, the mobile phase compositions were 0.1% formic acid in water (A) and 0.1% formic acid in acetonitrile (B). The gradient used to accomplish the separation was as follows: 0–8 min, 65% B, 8–12 min, 65–95% B, and 12–13 min, 95% B. After each run, a 5 min column re-equilibration was carried out. The injection volume was 2 μL , and quantification was performed at 214 nm with pure cannabinoid standards (Salazar-Bermeo et al., 2023). The results were expressed as mg per g of sample and were used for statistical analysis.

2.6. Data processing

The results were processed using PAST (PALEontological STATistics) 4.12b statistical software. Hierarchical clustering analysis was performed using Ward's method, an agglomerative clustering algorithm with Euclidian distance index along with principal component analysis (PCA). Averaged values of each terpenoid or cannabinoid in the inflorescences or seed oils of the nine *Cannabis* varieties assessed were used for the analysis. All the assays were performed by triplicate independently ($n = 3$).

3. Results

3.1. Identification of aromatic compounds and classification of Cannabis varieties

3.1.1. Aromatic compounds in cannabis inflorescences

Overall, GC-MS analysis identified a total of 71 natural aromatic compounds in cannabis inflorescences (Figs. 1 and 2) and Table 2. In addition to these, 3 phthalate esters were identified that would have migrated from the plastic packaging films that normally protect the inflorescences during storage and transport. A total of 16 compounds were ruled out as they are not typically found in cannabis and could have resulted from contamination or degradation during thermal desorption. Out of the 71 naturally occurring aromatic compounds identified, 27 were terpenoids, 10 isomers, 6 benzenoids, 8 alcohols and polyols, 8 aldehydes, 5 acids and esters, 4 alkane hydrocarbons, and 3 classified as organic compounds. Notably, benzyl alcohol and diethyl phthalate were the most prevalent compounds in most samples, belonging to the benzenoid group, followed by α -bergamotene, *trans*-caryophyllene, α -humulene, linalool, β -myrcene, *trans*-2-pinanol, and β -selinene as prevalent terpenes and terpenoids. *n*-Decanal, nonanal, and undecanal were the primary aldehydes. *n*-Hexanoic acid was the predominant acid and 1-methoxy-2-propyl acetate represented the major organic compound.

The hierarchical clustering analysis (Fig. 3A) revealed a clear division of aromatic compounds into two main clusters. Cluster CI consisted of a single compound, diethyl phthalate, while cluster CII further divided into two sub-clusters, CIIA and CIIB. CIIB, in turn, displayed additional complexity with two groups, CIIB1 and CIIB2. CIIB1 was composed of aromatic compounds commonly found in cannabis samples, such as β -myrcene, *trans*-caryophyllene, benzyl alcohol, limonene, nonanal, *n*-decanal, and L-limonene. CIIB2, on the other hand, encompassed various sub-clusters with compounds present in low percentage ratios. CIIA clustered two compounds: tetraethylene glycol and pentaethylene glycol. Regarding the samples, two main clusters were observed. CMI composed solely by BC variety while CMII was subdivided into two subclusters, one of them being composed solely by LS.

The dendrogram displayed the highest distance for diethyl phthalate

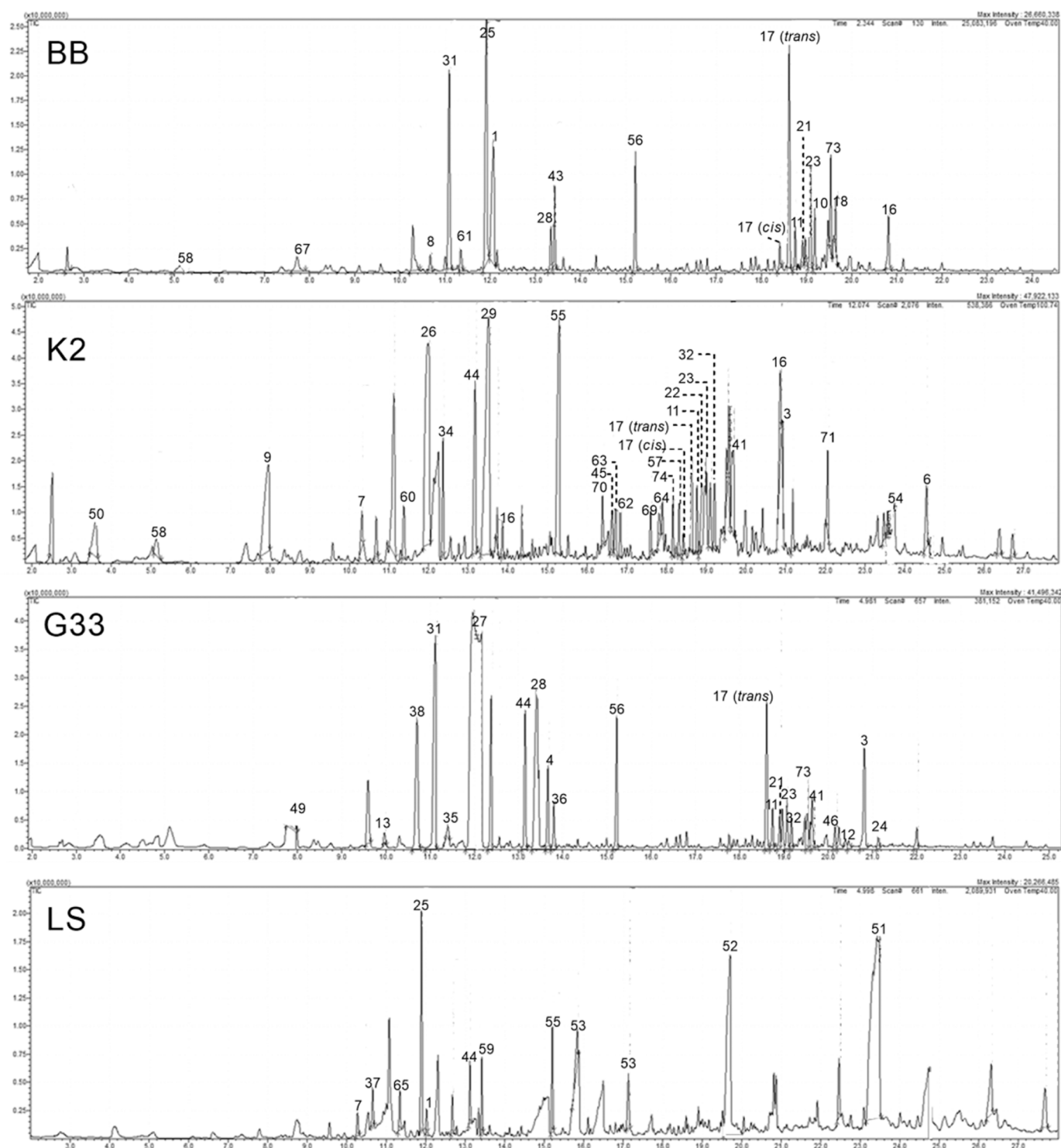


Fig. 1. Representative chromatograms of aromatic profile identified in inflorescences of 4 *Cannabis* varieties: Bubble gum (BB), Kitne 2 (K2), Gelato 33 (G33), and L. A. S.A.G.E. (LS). Numbers in each peak represent the aromatic compounds found in samples and are listed in Table 2.

among other aromatic compounds, followed by β -myrcene, limonene, *trans*-caryophyllene, and pentaethylene glycol as compounds with the highest percentage ratios. Most aromatic compounds in CIIB2 showed a low percentage ratio, resulting in close distances among them. Furthermore, the clustering analysis (Fig. 3B) revealed the similarity among the cannabis samples. BC variety displayed the highest distance index compared to other samples, particularly with the LS (86.39).

Conversely, BB and STC varieties showed the lowest distance index (13.24), indicating the closest aromatic profiles between two varieties.

BVD and G33 varieties displayed a moderate distance index (28.48), while SBC and FC showed a similar distance index (31.15). The remaining *Cannabis* varieties exhibited distance indexes ranging from 33 to 60 points.

The PCA (Fig. 3C) supported the clustering results and explained the variance of results (63.82%) principal component 1 (PC1) and principal component 2 (PC2), with BC variety showing the highest distance from other samples due to its high content of diethyl phthalate, locating in the negative PC1. LS variety also exhibited distance from other samples due

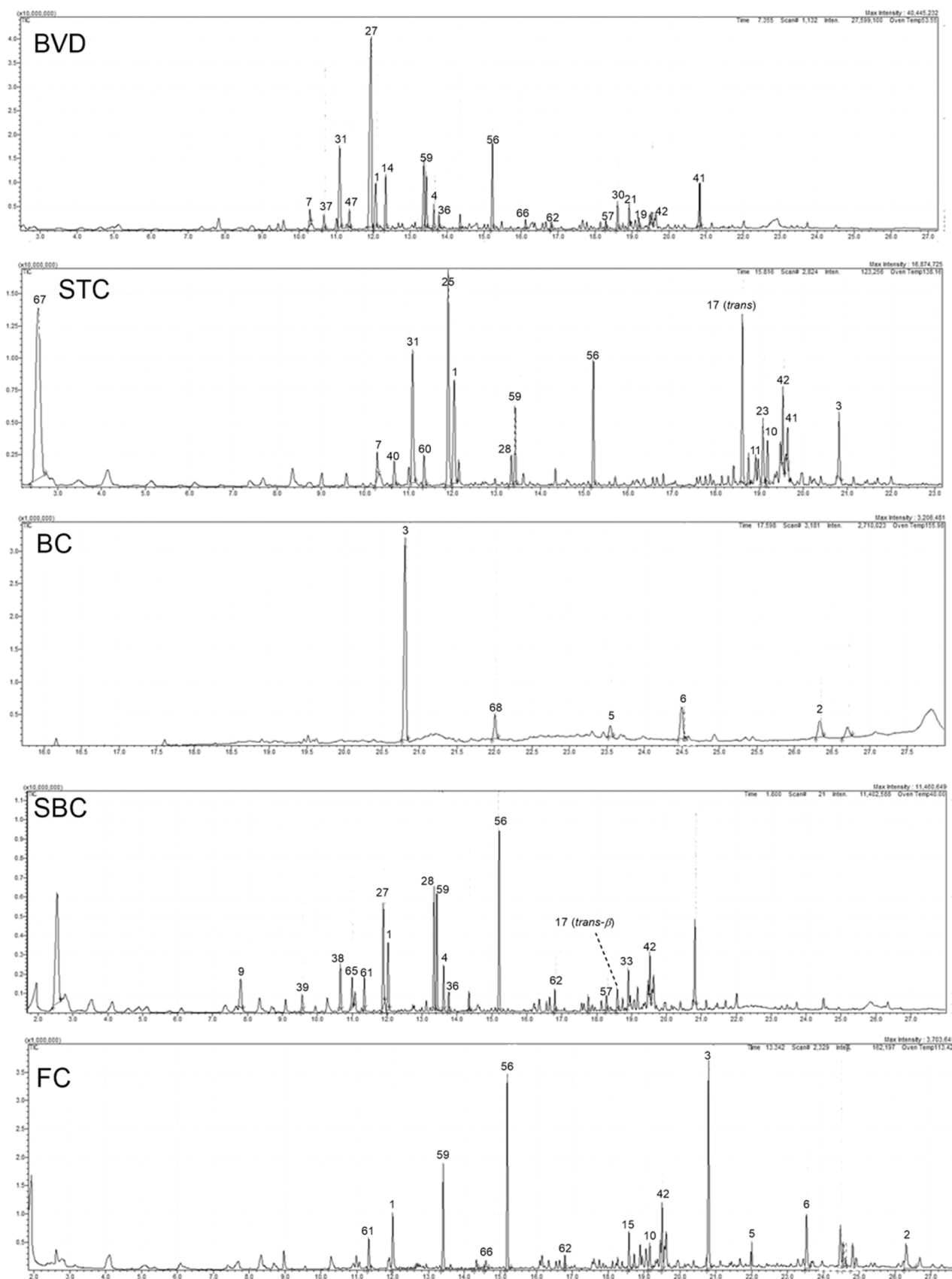


Fig. 2. Representative chromatograms of aromatic profile identified in inflorescences of 5 Cannabis varieties: Nicole x Bubble gum (BVD), Stracciatella (STC), Birthday cake (BC), Gelato 33 x Straw Banana Cream (SBC), and French cookies (FC). Numbers in each peak represent the aromatic compounds found in samples and are listed in Table 2.

Table 2
Aromatic profile of identified compounds in Cannabis varieties.

N ^o *	Compound	RT ^{***} (min)	Odour description ^{**}
	Benzenoids and benzoyl derivatives		
1	Benzyl alcohol	12.05	Floral, rose, phenolic. Balsamic
2	Dibutyl phthalate	26.32	Faint fruity, green odour
3	Diethyl phthalate	20.84	Odourless to mild baked and must
4	Durene	13.73	Sweet, herbal, earth, phenolic, green
5	2-Ethylhexyl salicylate	23.54	Mild orchid, sweet, balsam
6	Isobutyl phthalate	24.51	Mild ester, fruity
7	Phenylmethanal	10.29	Almond, nutty, cherry and benzaldehyde-like
8	Pseudocumene	10.67	Phenol, earth, floral, green
9	o-Xylene	7.88	Geranium
	Terpenes/terpenoids		
10	Alloaromadendrene	19.19	Woody
11	α -Bergamotene	18.76	Infusion, woody, tea
12	Calacorene	20.46	Woody
13	Camphene	9.97	Woody, herbal, fir needle
14	(+)- 3-Carene	12.54	Citrus, herbal, solvent, resinous, phenolic, cypress
15	Caryophyllene	18.58	Sweet, woody, spice, clove, dry
16	Caryophyllene oxide	20.84	Sweet, fresh, dry, woody, spicy
17	cis-Caryophyllene	18.42	Sweet, woody, spice, clove, herbal
	trans-Caryophyllene	18.63	herbal
	trans- β -Caryophyllene	18.60	Sweet, woody, spice, clove, herbal
18	Eremophilene	19.65	Mild wood
19	β -Farnesene	19.18	Woody, citrus, herbal, sweet
20	Fenchol	13.67	Camphor, borneol, pine, woody, dry, sweet, lemon
21	Geranyl acetone	18.93	Fresh, rose, leaf, floral, green, magnolia
22	α -Guaiene	18.83	Sweet, woody, balsam, peppery
23	α -Humulene	19.10	Woody, oceanic-watery, spicy-clove
24	Humulene-oxide	21.14	Herbal
25	Limonene	11.93	Terpene, pine, herbal, sweet, citrus and peppery
26	D-Limonene	19.99	Sweet, orange, citrus
27	L-Limonene	11.93	Terpene, pine, herbal, peppery, citrus
28	Linalool	13.34	Citrus, floral, sweet, bois de rose, green, blueberry
29	L-Linalool	13.45	Fresh, floral, woody, natural, deep lavender
30	Longifolene-(V4)	18.61	Sweet, woody, rose, medical, pine
31	β -Myrcene	11.12	Must, herbaceous, woody with a rosy nuance
32	Nealloocimene	19.19	Citric, herbal
33	Nerylacetone	18.91	Fatty, metallic, geranium
34	trans- β -Ocimene	12.37	Sweet, herbal
35	α -Phellandrene	11.41	Citrus, terpenic, slightly green, black pepper-like
36	trans-2-Pinanol	13.80	Green, pine, fatty, woody
37	L- β -Pinene	10.32	Dry, woody, fresh, pine, hay, green, resinous
38	2- β -Pinene	9.58	Pine, green
39	D- α -Pinene	9.58	Terpenic, aromatic minty, floral
40	β -Pinene	10.67	Dry, woody, resinous, pine, hay, green
41	α -Selinene	19.96	Herbal, amber
42	β -Selinene	19.54	Herbal, woody
43	γ -Terpinene	13.43	Sweet, citrus, lime nuances
44	α -Terpinolene	13.15	Fresh, woody, sweet, pine, citrus
45	α -Thujol	16.72	Minty, camphorous, spicy
46	Valencene	20.16	Sweet, fresh, citrus, grapefruit, woody, orange
	Alcohols and polyols		

Table 2 (continued)

N ^o *	Compound	RT ^{***} (min)	Odour description ^{**}
47	Dipropylene glycol	11.36	Sweet, fruity, mild alcohol
48	n-Heptadecanol-1	24.57	Fatty, waxy, fruity
49	1-Hexanol	7.97	Pungent, fusel, oily, fruity, alcoholic, sweet
50	2-Methyl-1-butanol	3.60	Winey, onion, fruity, alcoholic, whiskey
51	Pentaethylene glycol	23.42	Odourless to mild sweet
52	Tetraethylene glycol	19.70	Odourless to mild sweet
53	Triethylene glycol	15.84	Odourless to mild sweet
54	Hexaethylene glycol	23.57	Mild sweet
	Aldehydes		
55	Decanal	15.29	Sweet, aldehydic, waxy, orange peel, floral
56	n-Decanal	15.22	Sweet, aldehydic, waxy and citrus rind
57	Dodecanal	18.30	Sweet, aldehydic, citrus with floral nuances
58	n-Hexanal	5.14	Green, fruity and clean with a woody nuance
59	Nonanal	13.42	Waxy, aldehydic, rose, fresh, peeled orange
60	n-Octanal	11.34	Aldehydic, waxy, orange with peeled nuance
61	Octanal	11.40	Waxy, citrus or fruity
62	Undecanal	16.81	Fresh, clean, citrus, waxy
	Acids and esters		
63	cyclohexyl ester-Acetic acid	16.63	Solvent-like and fruity sweet
64	n-Decanoic acid	17.83	Rancid, sour, fatty, citrus
65	n-Hexanoic acid	11.17	Sour, fatty, sweat, cheesy
66	Nonoic acid	15.35	Waxy, green and cheesy
67	1-Methoxy-2-Propyl acetate	7.73	Sweet, ether-like
	Organic compounds		
68	Limonene diepoxide	22.01	Menthol, sweet, woody
69	5-Ethoxy-2-Methylpyridine	17.60	Nutty, strong, raw, potato, roasted, earthy
70	hexadecyl-Oxirane	16.38	Herbal, lavender
	Alkane hydrocarbons		
71	(R,R)- 3,8-Dimethyldecane	22.06	Burn, sugary
72	Hentriacontane	22.01	Odourless to fuel-like
73	Pentadecane	19.54	Alkane, waxy
74	n-Tetradecane	18.16	Mild waxy

*Numbers indicate the peak of each compound in each sample chromatogram (Figs. 1 and 2).

**All odor descriptions were obtained from Flavornet, GreenMol BD, FOODB, HMDB and TGSC databases. Discarded compounds presented odorless properties or were simpler structures that belonged to more complex compounds (data not shown).

***RT: Retention time.

to the presence of pentaethylene glycol and tetraethylene glycol, locating in the negative PC2. BVD and G33 varieties were closely related, in line with their shared content of L-limonene, as observed in the clustering analysis (Fig. 3A). The other Cannabis variety samples (SBC, STC, FC, K2, and BB) demonstrated minimal differences in their aromatic profiles, consistent with the clustering analysis and distance matrix.

3.1.2. Aromatic compounds in cannabis seed oils

The aromatic content of cannabis seed oil from the same varieties was assessed using GC-MS analysis (Fig. 4). A lower number of aromatic compounds (22 compounds) were identified in the seed oil samples compared to the inflorescences. The hierarchical clustering analysis (Fig. 4A) revealed the formation of two clusters, CI and CII; the last one primarily consisted of trans- β -caryophyllene. Cluster CI further divided into two sub-clusters, CIA and CIB. CIB included β -myrcene, (-)-guaiaol, and (-)- α -bisabol. CIA contained other aromatic compounds, with CIA1 comprising compounds commonly found in cannabis and CIA2 consisting of only cis-ocimene.

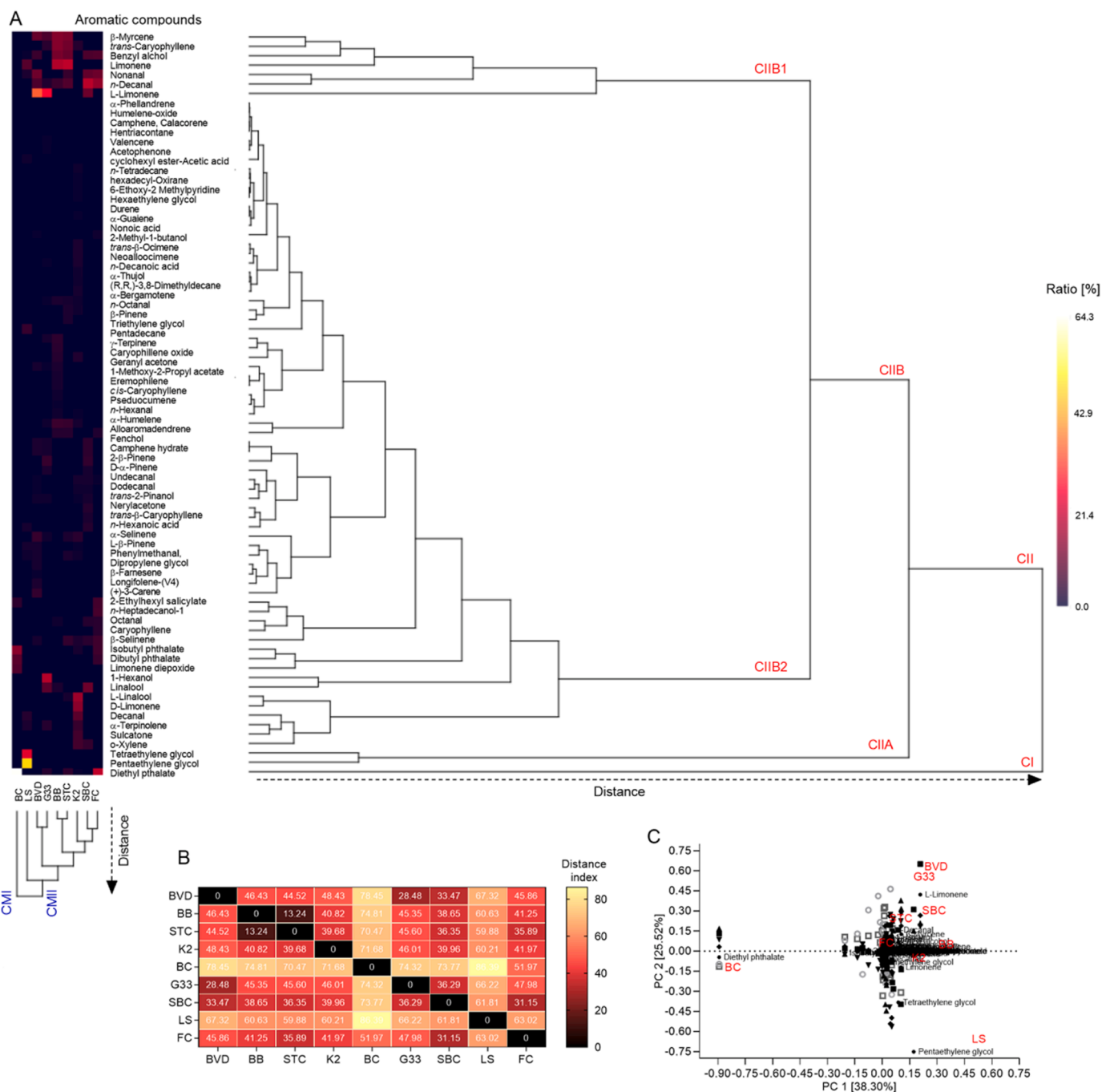


Fig. 3. Terpenoid profile of inflorescences of 9 *Cannabis* varieties: Bubble gum (BB), Nicole x Bubble gum (BVD) Stracciatella (STC), Kitne 2 (K2), Gelato 33 (G33), Gelato33 x Straw Banana Cream (SBC), L.A. S.A.G.E. (LS), Birthday cake (BC), and French cookies (FC). (A) Hierarchical clustering analysis of terpenoids from 9 *Cannabis* varieties. Color-coding consists of shades of yellow, red, and black, where higher percentage ratio of compounds stands for yellow tones while a lower percentage ratio of compounds stands for black tones. Main clusters between compounds are indicated in red while main clusters between samples are indicated in blue. (B) Distance index between inflorescences from different *Cannabis* varieties. Black tones indicate a low distance relationship among samples while yellow tones indicate a high distance relationship between samples. (C) Principal component analysis (PCA) biplot of terpenoid profile from inflorescences of 9 *Cannabis* varieties. Projection of the variables of the factor plane (PC 1 \times PC 2) considering the aromatic compounds quantified. *Cannabis* varieties are indicated in red while aromatic compounds are indicated in black.

While several of these compounds were also observed in the inflorescences, their ratios in the seed oils did not exceed 0.12%, indicating a lower content in the oils. *trans*- β -Caryophyllene, belonging to CII, exhibited the highest ratio percentage among most samples, particularly in FC variety. In contrast, *cis*-ocimene which was clustered in CIA2 was only present in five samples. In CIA1, most compounds were found at a ratio percentage of 0.04%, suggesting their close relationship due to their low presence in samples. However, α -terpinolene and

linalool, also found in CIA1, displayed a higher ratio percentage (approximately 0.08%), especially in samples from G33, LS, K2, and FC varieties, warranting their separation into a different sub-cluster within CIA1.

The varietal distribution (Fig. 4A) of the cannabis seed oil samples reflected a similar pattern observed in the aromatic profile of the inflorescences. Two clusters, CMI (including BB and BVD varieties) and CMII, were observed, with CMII further dividing into two sub-clusters,

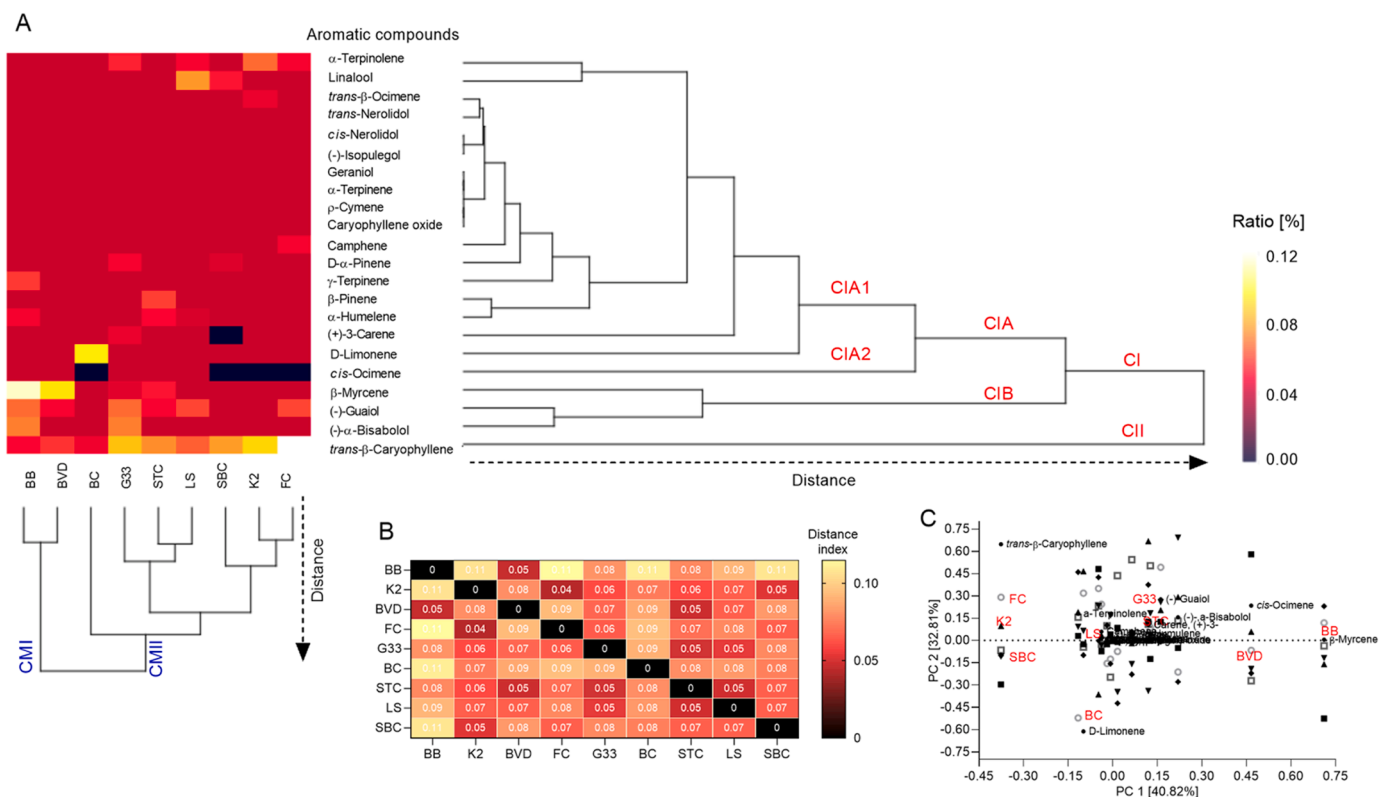


Fig. 4. Terpenoid profile of seed oils of 9 *Cannabis* varieties: Bubble gum (BB), Nicole x Bubble gum (BVD) Stracciatella (STC), Kitne 2 (K2), Gelato 33 (G33), Gelato33 x Straw Banana Cream (SBC), L.A. S.A.G.E. (LS), Birthday cake (BC), and French cookies (FC). (A) Hierarchical clustering analysis of terpenoids in inflorescences from 9 *Cannabis* varieties. Color-coding consists of shades of yellow, red, and black, where higher percentage ratio of compounds stands for yellow tones while a lower percentage ratio of compounds stands for black tones. Main clusters between compounds are indicated in red while main clusters between samples are indicated in blue. (B) Distance index between seed oils derived from different *Cannabis* varieties. Black tones indicate a low distance relationship among samples while yellow tones indicate a high distance relationship between samples. (C) Principal component analysis (PCA) biplot of terpenoid profile from seed oils of 9 *Cannabis* varieties. Projection of the variables of the factor plane (PC1 \times PC2) considering the aromatic compounds quantified. *Cannabis* seed oil varieties are indicated in red while aromatic compounds are indicated in black.

one of them being composed by BC variety. The distance index among the samples (Fig. 4B) indicated low distances between them, with values not exceeding 0.10. K2 and FC varieties exhibited the lowest distance index of 0.04, while BB variety displayed the highest distance index with other varietal samples, such as K2, FC, BC, and SBC (0.11). Similar to the inflorescence analysis, the seed oil from the BC variety exhibited a high distance index from other samples due to its D-limonene content. BB and BVD varieties showed a short distance index (0.05) due to their proximity in terms of β -myrcene content. Likewise, K2 and FC varieties exhibited a short distance index (0.04) due to their content of *trans*- β -caryophyllene and α -terpinolene, respectively.

The PCA (Fig. 4C) confirmed that most oil samples had low distances from one another, PC1 and PC2 explained 73.63% of variance. Among *cannabis* seed oil samples, the one from the BB variety stood out from the rest due to its β -myrcene content, while that from the BC variety showed distinctiveness due to its D-limonene content. FC, KD, and SBC varietal seed oils exhibited a close relationship, while the one from the BVD variety with *cis*-ocimene displayed relative differentiation. G33, LS, and STC varieties had seed oils with a more similar composition of terpenoids. These results indicate that although seed oils retain some aromatic compounds from their sources, their aromatic composition differs from that of the corresponding plants.

3.2. Description of aromatic compounds in *cannabis* inflorescences and seed oils

A search was carried out in several databases to identify the corresponding odour of each one of the identified compounds in the

inflorescences and seed oils of the nine tested *Cannabis* varieties, which would allow the subsequent determination of the key odorants in each variety (Table 2).

Analysis of inflorescences revealed that benzyl alcohol and diethyl phthalate were common in most samples, indicating floral, rose, phenolic, balsamic, and faint fruity odours in most of varieties. Dominant terpenes, including α -bergamotene, *trans*-caryophyllene, α -humulene, linalool, β -myrcene, *trans*-2-pinanol, and β -selinene, were also identified. These terpenes contributed to a complex aroma profile characterized by infusion, woody, tea, sweet, herbal, oceanic-watery, spicy-clove, citrus, floral, bois de rose or rosewood, green, blueberry, must, herbaceous, and rosy nuances. Aldehydes, namely *n*-decanal, nonanal, and undecanal, were the primary compounds found, imparting sweet, aldehydic, waxy, citrus rind, rose, orange peel, fresh, clean, and citrus aromas. The main acid detected was *n*-hexanoic acid, contributing sour, fatty sweat, and cheesy notes. Another significant compound was 1-methoxy-2-propyl acetate, adding a sweet aroma with ether-like characteristics.

Examining varietal inflorescences separately, BC variety sample exhibited diethyl phthalate as the main compound, suggesting a distinctive mild baked and musty aroma. The LS inflorescences showed a higher content of pentaethylene glycol, contributing to a notable mild sweet aroma differentiating it from other samples. G33 and BVD inflorescences contained L-limonene as the primary compound, resulting in pronounced pine, herbal, peppery, and citrus-like scents.

Among the samples, STC varietal inflorescences were the only to contain β -pinene, which may contribute to its unique woody, resinous, pine, and hay-like odour. K2 inflorescences exhibited distinctive

compounds such as α -guaiene, D-limonene, L-linalool, *trans*- β -ocimene, and α -thujol, contributing to a balsamic, peppery, deep lavender, minty, and camphor aroma. In the FC varietal sample, caryophyllene was present, accompanied by benzenoids, resulting in a specific aroma characterized by sweetness, spiciness, clove, mild floral, balsamic, and ester notes.

The seed oil samples of *Cannabis* varieties displayed lower contents of aromatic compounds, mainly terpenes. *trans*- β -Caryophyllene emerged as the prominent compound in most samples, followed by α -bisabolol, guaiol, and β -myrcene, conferring notes of sweetness, woodiness, spice, clove, herbal nuances, and a musty, herbaceous, woody aroma with a rosy undertone. Notably, α -terpinolene and linalool

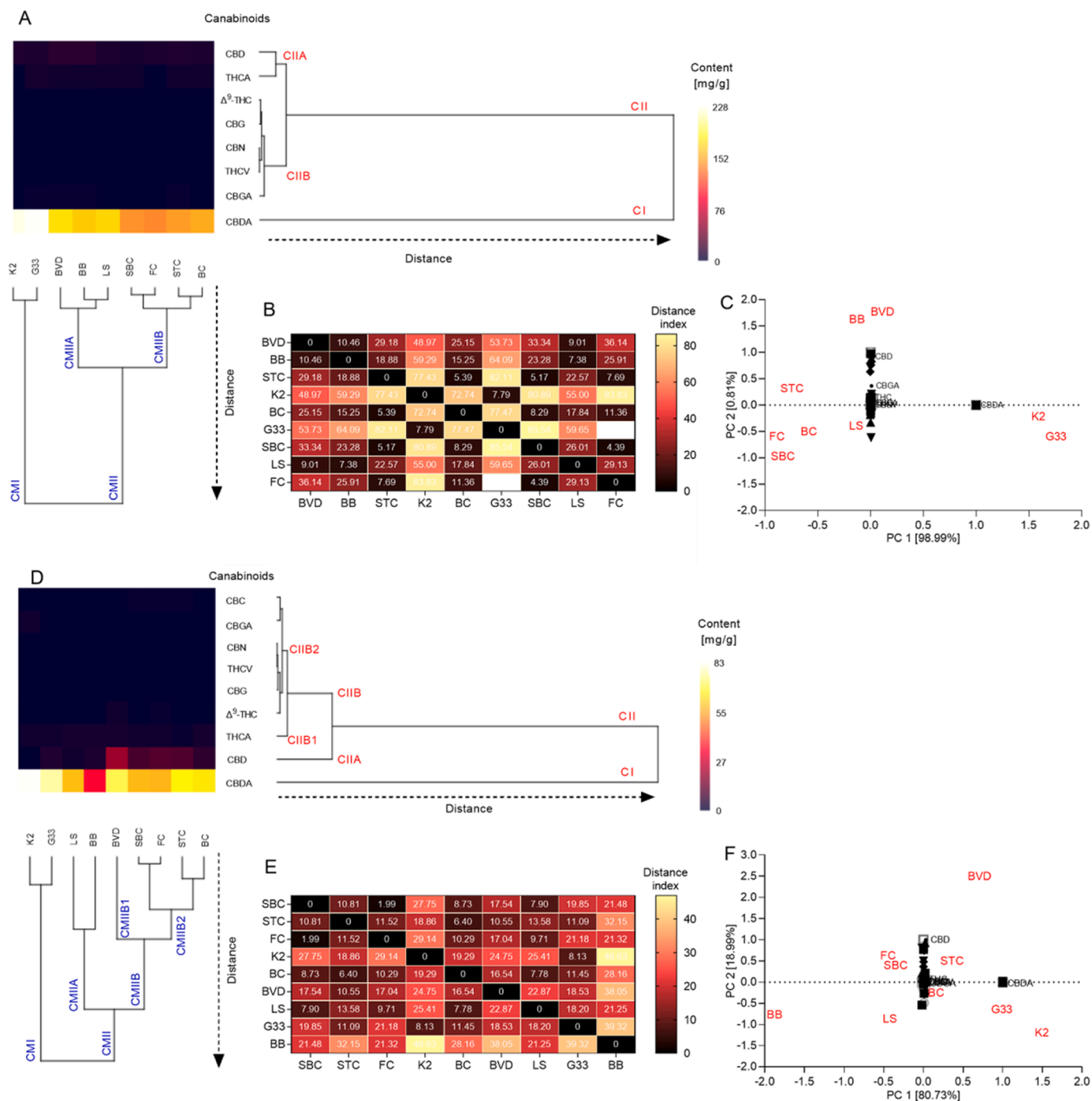


Fig. 5. Cannabinoid profile of inflorescences and seed oils of 9 *Cannabis* varieties: Bubble gum (BB), Nicole x Bubble gum (BVD) Stracciatella (STC), Kitne 2 (K2), Gelato 33 (G33), Gelato33 x Straw Banana Cream (SBC), L.A. S.A.G.E. (LS), Birthday cake (BC), and French cookies (FC). Hierarchical clustering analysis of cannabinoids in (A) inflorescences and (D) seed oils from 9 *Cannabis* varieties. Color-coding consists of shades of yellow, red, and black, where higher concentration (mg/g) of compounds stands for yellow tones while a lower concentration of compounds stands for black tones. Main clusters between compounds are indicated in red while main clusters between samples are indicated in blue. Distance index between samples of (B) inflorescences and (E) seed oils from different *Cannabis* varieties. Black tones indicate a low distance relationship among samples while yellow tones indicate a high distance relationship between samples. Principal component analysis (PCA) biplot of cannabinoid profile of (C) inflorescences and (F) seed oils of 9 *Cannabis* varieties. Projection of the variables of the factor plane (PC 1 \times PC 2) considering the cannabinoids quantified. *Cannabis* varieties are indicated in red while cannabinoids are indicated in black.

were present in high proportions in certain samples, lending citrus, floral, sweet, bois de rose, green, blueberry, fresh, woody, pine, and citrus-like aromas.

Among cannabis seed oil samples, the one from the BB variety displayed a high proportion of β -myrcene, contributing to a musty, herbaceous, woody aroma with rosy nuances. The BVD varietal oil sample contained *cis*-ocimene, resulting in a sweet, herbal scent. BC sample exhibited a distinctive content of D-limonene, imparting a sweet, orange, or citrus-like scent. The oil samples of the FC, K2 and SBC varieties showed significant levels of β -*trans*-caryophyllene, accompanied by a sweet, woody, spicy, clove and herbal aroma, with the FC variety showing the highest concentration. BB and BVD seed oils showcased the highest levels of β -myrcene, amplifying the musty, herbaceous, woody scent. LS seed oil exhibited a high content of linalool, lending a citrus, floral, sweet, bois de rose, green, and blueberry aroma. G33 and STC oil samples exhibited similar substantial levels of *trans*- β -caryophyllene, bisabolol, guaial, D- α -pinene, and α -terpinolene, resulting in a terpenic, aromatic, minty, and floral scent.

3.3. Cannabinoid identification in cannabis inflorescences and seed oils

The cannabinoid content of inflorescences and seed oils from the same nine *Cannabis* varieties was analysed, and the results are presented in Fig. 5. Nine cannabinoids were analysed using pure standards.

Similar to the aromatic compound analysis, higher cannabinoid content was found in inflorescences compared to seed oils. In the inflorescences (Fig. 5A and Table 3), CBDA was the predominant cannabinoid, particularly in K2 and G33 varieties, with concentrations of nearly 228 mg/g. CBD and THCA showed lower concentrations, reaching about 10 and 3 mg/g respectively, while the remaining cannabinoids were present in the samples at concentrations of no more than 0.04 mg/g. CBC was not detected in samples while THCv was only detected in samples. The clustering analysis of inflorescences (Fig. 5A) revealed two clusters: CI, consisting of CBDA, and CII, further subdivided into two sub-clusters. CIIA sub-cluster included CBD and THCA, while CIIB comprised Δ^9 -THC, CBG, CBN, THCv, and CBGA. All compounds in CIIB exhibited low distances from one another, but three subgroups were identified: CIIB1 (Δ^9 -THC and CBG), CIIB2 (CBN and THCv), and CIIB3 (CBGA). All inflorescence samples displayed a Δ^9 -THC content ranging from 0.2% to 0.4% of total Δ^9 -THC.

Table 3

Cannabinoid content in both inflorescences (IF) and seed oils (SO) from nine *Cannabis* varieties.

		Cannabinoids [mg/g]								
Variety	Sample	CBDA	CBGA	CBG	CBD	THCV	CBN	Δ^9 -THC	THCA	CBC
BVD	IF	174.48	3.54	0.91	14.32	< 0.33	< 0.33	1.67	4.15	ND
	SO	57.23	0.55	< 0.33	9.46	< 0.33	< 0.33	0.32	1.61	1.22
BB	IF	164.03	3.35	0.9	14.12	< 0.33	< 0.33	1.67	3.76	ND
	SO	67.91	0.32	0.32	11.01	< 0.33	< 0.33	0.60	1.24	0.99
STC	IF	145.65	2.37	0.85	10.00	< 0.33	< 0.33	1.09	4.48	ND
	SO	56.4	< 0.33	< 0.33	11.21	< 0.33	< 0.33	0.67	1.56	1.00
K2	IF	222.96	1.35	0.93	9.07	< 0.33	< 0.33	0.11	0.58	ND
	SO	83.41	1.93	0.61	0.4	< 0.33	< 0.33	0.35	1.49	0.49
BC	IF	150.3	0.62	0.48	8.14	< 0.33	< 0.33	0.87	3.66	ND
	SO	64.9	< 0.33	< 0.33	5.53	< 0.33	< 0.33	0.47	1.17	ND
G33	IF	227.7	1.96	0.9	7.55	< 0.33	< 0.33	0.83	6.50	ND
	SO	70.14	< 0.33	0.45	21.16	< 0.33	< 0.33	1.76	1.17	ND
SBC	IF	142.2	0.78	0.58	6.54	< 0.33	< 0.33	0.64	4.29	ND
	SO	58.11	< 0.33	0.49	1.76	< 0.33	< 0.33	< 0.33	1.52	ND
LS	IF	168.11	0.56	0.56	8.75	< 0.33	< 0.33	0.97	4.51	ND
	SO	76.18	< 0.33	0.35	3.7	< 0.33	< 0.33	0.43	1.52	ND
FC	IF	139.15	0.38	0.49	8.05	< 0.33	< 0.33	0.40	1.55	ND
	SO	36.88	< 0.33	0.45	2.73	< 0.33	< 0.33	< 0.33	1.32	ND

Varieties: Nicole x Bubble gum (BVD), Bubble gum (BB), Stracciatella (STC), Kitne 2 (K2), Birthday cake (BC), Gelato 33 (G33), Gelato 33 x Straw Banana Cream (SBC), L.A.S.A.G.E. (LS), and French cookies (FC).

Cannabinoids: Cannabidiolic acid (CBDA), cannabigerolic acid (CBGA), cannabigerol (CBG), cannabidiol (CBD), tetrahydrocannabivarin (THCV), cannabinol (CBN), Δ^9 -tetrahydrocannabinol (Δ^9 -THC), tetrahydrocannabinolic acid (THCA), and cannabichromene (CBC).

Limit of quantification: 0.33 mg/g. ND: non-detected.

Regarding the clustering analysis of *Cannabis* varieties (Fig. 5A), two clusters were observed: CMI, comprising K2 and G33, which showed a small difference in cannabinoid content, likely due to their high CBDA content, and CMII, subdivided into two sub-clusters: CMIIA and CMIIIB. CMIIA was further divided into two groups: BVD and BB with LS, while CMIIIB also divided into two groups: SBC with FC and STC with BC.

The distance index among the samples of inflorescences (Fig. 5B) indicated that FC and SBC varieties exhibited the lowest distance value (4.39), followed for the value corresponding to the SBC and BC tandem (5.39). Among all *Cannabis* varieties, K2 and G33 showed the highest distance indexes in comparison to other samples but were close to each other in terms of their cannabinoid content. The PCA analysis (Fig. 5C) confirmed the relationship among all *Cannabis* varieties, with PC1 and PC2 explaining 99.8% variance of results. PC1 mainly influenced the dataset, with K2 and G33 varieties appearing more distinct from the other due to their CBDA content, while BB and BVD varieties also exhibited distinctness from the other due to their CBD content.

CBC along with CBGA were detected in cannabis seed oil samples below the limit of detection (0.033 mg/g). The hierarchical clustering analysis (Fig. 5D) of seed oil cannabinoids showed two clusters similar to those observed in the clustering analysis of inflorescences, with CI cluster contained high concentrations of CBDA, while CII was subdivided into two sub-clusters: CIIA, containing CBD, and CIIB that further subdivided into two subgroups: CIIB1 with THCA at concentrations below 2 mg/g in samples, and CIIB2, consisting of several cannabinoids at concentrations close to the limit of detection (0.033 mg/g).

Regarding the *Cannabis* varieties, the clustering analysis of seed oils exhibited two similar clusters to those obtained for samples of inflorescences. The seed oils from K2 and G33 varieties were clustered in CMI, while CMII was subdivided into two sub-clusters: CMIIA, comprising LS and BB varieties, and CMIIIB, that further subdivided into two groups: CMIIIB1, with BVD variety, and CMIIIB2, which was also subdivided into two subgroups composed by the varietal pairs SBC-FC and STC-BC, respectively.

Fig. 5E display the distance index between seed oils from different *Cannabis* varieties. K2 and G33 varieties exhibited the lowest distance index (8.13), which indicates similarity between their seed oils in agreement with the findings in Fig. 5B for inflorescences. The seed oil from BB variety appeared as the most distinct sample among all analysed, particularly in comparison to K2, with a distance index of more

than 46 points. Thus, the distance index between cannabis seed oils corroborated the observed distances among varieties in Fig. 5A.

In addition, Fig. 5F shows patterns where PC1 and PC2 explaining 99.72% variance of results which is similar to that observed in Fig. 4C. PC1 generates a remarked influence in dataset, with G33 and K2 varieties showing more distinct from the other due to their CBDA content, while BVD variety also exhibited distinctness from the other due to its CBD content.

4. Discussion

Because of its complex composition, which includes cannabinoids and aromatic compounds, *Cannabis* spp. are one of the most studied and commonly utilized plants (Naz et al., 2017). Quality control and standardization are crucial for the medicinal application of cannabis associated with various therapeutic benefits for human health being, therefore, necessary the development of reliable methods to classify the different *Cannabis* varieties (Al Bakain et al., 2020; Al Bakain et al., 2020). For this purpose, the aromatic profile database of screened compounds provided by this study could be very useful as a reference.

In this study, the aromatic and cannabinoid composition of inflorescences and seed oils from nine commercial *Cannabis* varieties were investigated. The results demonstrated that inflorescences contained a greater number and abundance of aromatic compounds compared to seed oils. Each variety exhibited distinct chemical profiles, with some compounds being unique to specific varieties. The commercial descriptions of the varieties generally matched the aromatic compounds detected, indicating that the aroma of *Cannabis* varieties can be attributed to the specific compounds detected in varieties (Hanus and Hod, 2020; Naz et al., 2017; Sommano et al., 2020). For instance, the inflorescences of certain varieties with descriptions of citrus, herbal, and pine fragrances were found to contain L-limonene, which aligns with their commercial descriptions.

Similar correlations were observed for other varieties based on their aromatic profiles like inflorescences of BC and LS varieties. BC variety was the most different from the other varieties, clustering by itself due to its noted content of diethyl phthalate. LS variety inflorescence also demonstrated to be distinctive from the other samples due to its content of pentaethylene glycol and tetraethylene glycol. Interestingly, the main aromatic compound found in both varieties are characterized as odourless to mild baked or mild sweet scents, respectively, which may indicate that although these varieties showcased other compounds in their aromatic profile, these compounds can be perceived by the nose distinctively, given their commercial description matched with the aroma of these compounds.

Seed oils, on the other hand, exhibited a lower abundance of aromatic compounds, which may be attributed to their naturally lower presence in this product. The extraction and purification techniques used for seed oils can also affect the composition of minor compounds (Pavlovic et al., 2018). It should be noted that the aromatic compounds detected in seed oils did not match the aromatic description of their respective inflorescences (Naz et al., 2017).

In terms of cannabinoid content, all inflorescences contained between 2–4% of total Δ^9 -THC but higher CBD content, classifying them as high-CBD chemical phenotypes (Naz et al., 2017). The hierarchical analysis and PCA revealed that CBDA (98.99% of variance) was the essential component distinguishing each variety from the others. The presence of diverse terpenes and other aromatic compounds played a significant role in the aromatic diversity of *Cannabis* varieties, reinforcing that these compounds are valuable indicators for differentiation (Birenboim et al., 2022; Naz et al., 2017).

It is crucial to recognize and consider these differences when evaluating the aromatic profiles of cannabis-derived products. Seed oils may not accurately represent both the aromatic and cannabinoid composition of inflorescences. Traditional extraction methods for seed oils do not consider the presence of cannabinoids and aromatic compounds

which are often regarded as oil contaminants (García-Valverde et al., 2020; Hanus and Hod, 2020; Nonier et al., 2004; Richins et al., 2018; Sommano et al., 2020).

The analysis of seed oils revealed fewer aromatic compounds, primarily terpenes, compared to the inflorescences of the nine *Cannabis* varieties examined (Naz et al., 2017). The most abundant aromatic compounds in the seed oils were *trans*- β -caryophyllene and β -myrcene. It is worth noting that cannabis has been reported to contain more than 100 aromatic compounds across various varieties (Calvi et al., 2018; Russo and Marcu, 2017). However, many of these compounds are low-molecular-weight compounds that easily volatilize at room temperature, and factors such as light exposure, temperature, humidity, and storage conditions can modify or degrade these volatile compounds (Gallily et al., 2018; Nonier et al., 2004; Richins et al., 2018; Russo and Marcu, 2017; Solowij et al., 2019). It may explain why several aromatic compounds detected in the inflorescences were not found in seed oils.

Hierarchical analysis and PCA of the aromatic compounds in seed oils resulted in two main clusters, but the maximum distance between samples was only around 48 points, indicating that these compounds alone might not be sufficient for accurate classification of cannabis samples. Furthermore, several seed oils exhibited low distance indexes, suggesting that the aromatic composition in these samples was similar and might be negatively influenced during the industrial extraction process from raw material (Andre et al., 2016; Citti et al., 2018).

In contrast, the analysis of cannabinoids in seed oils using hierarchical analysis and PCA revealed a different pattern, with distinct clustering of samples. However, similar to the inflorescences, CBDA was the predominant cannabinoid in the seed oils, while total Δ^9 -THC content remained below 0.2% in all samples. These results are relevant considering the limitations imposed on cannabis-derived products due to Δ^9 -THC content, indicating that not all cannabis-derived products will necessarily maintain the composition of their source material, which can affect their commercialization (Citti et al., 2019; Leizer et al., 2015).

Although both the aromatic and cannabinoid composition represent a small fraction of the components in cannabis seed oils, obtained results suggest that they actively contribute to the chemical variability and potential biological activities of the final product. It has been reported that certain aromatic compounds and cannabinoids found in cannabis seed oils may participate in specific biological activities, potentially enhancing their overall effects (Citti et al., 2019; Citti et al., 2018; Leizer et al., 2015).

Among the wide range of cannabis-derived products available, dried cannabis inflorescences are the most frequently consumed form due to their high concentration of bioactive compounds (Birenboim et al., 2022). However, there is still scepticism regarding the nutritional and therapeutic value of seed oils derived from cannabis, mainly because the potential risk associated with its toxicological properties and its synthetic derivatives (Andre et al., 2016; Citti et al., 2019), despite the fact that some studies have reported low concentrations of cannabinoids in these products (Andre et al., 2016; Citti et al., 2018).

The current regulatory classification of cannabis and its products does not consider differences in the composition and concentration of less prominent cannabinoids or other phytochemicals apart from CBD and Δ^9 -THC (Birenboim et al., 2022; Citti et al., 2018). Thus, it is important to explore classification approaches that focus on the cannabinoid composition, including total Δ^9 -THC (Δ^9 -THC+THCA) and total CBD (CBD+CBDA), as well as other naturally occurring bioactive cannabinoids such as CBG and cannabigerovarin (CBGV) (Birenboim et al., 2022). Even more when the quantitative structure-activity relationships, particularly 3-dimensional (3-D) conformation-specific bioactivities of cannabinoids, are not well known (Salha et al., 2023). Likewise, terpenoids are aromatic compounds whose presence and concentration in *Cannabis* varieties depend on their genetic background (Russo and Marcu, 2017). These compounds are known to exert pharmacological activities and modulatory effects on cannabinoids

(García-Valverde et al., 2020; Pavlovic et al., 2018; Russo and Marcu, 2017). Thus, their analysis can contribute to the accurate classification of *Cannabis* varieties and support the use of other cannabis-derived products such as seed oil.

The presence of bioactive compounds, both terpenoids and cannabinoids, in cannabis inflorescences and seed oils, predicts potential future applications in medicine, nutraceuticals, and wellness products (Leizer et al., 2015). Nevertheless, further research is necessary to fully understand the mechanisms of action and potential therapeutic benefits of these compounds (Booth et al., 2017; Gallily et al., 2018; Namdar et al., 2019; Richins et al., 2018). There is also a need to optimize extraction and purification methods leading to the development of safe high-quality, standardized cannabis-derived products with consistent compositions and specific concentrations of terpenoids and cannabinoids (Al Bakain et al., 2020; Leizer et al., 2015; Salazar-Bermeo et al., 2023).

In conclusion, the analysis of aromatic compounds highlighted the diverse array present in cannabis inflorescences, contributing to their unique aromas and flavours. GS-MS analysis allowed to characterize and categorize aromaticity in the sample set tested, proving to be a useful tool for the classification of *Cannabis* varieties. The observed differences between seed oils and their aromatic counterparts underscore the need for careful consideration of oil extraction process. Furthermore, the analysis of cannabinoids revealed the complexity of cannabis chemistry, with the cannabinoid content alone being insufficient for accurately classifying *Cannabis* varieties.

These findings highlight the importance of considering a broad range of compounds in the analysis of cannabis and its derived products. By doing so, it becomes possible to overcome the stigma associated with Δ^9 -THC content and ensure the accurate characterization and assessment of their potential applications. This study contributes to the growing body of knowledge on cannabis chemistry and highlights the need for further research to explore the role of terpenoids and less prominent cannabinoids, mainly CBD and CBG, in enhancing the therapeutic efficacy of cannabis-derived products. Ultimately, this knowledge can guide the development of targeted breeding programs and the formulation of cannabis-based products tailored to specific therapeutic applications.

CRedit authorship contribution statement

De la Torre Rosa: Conceptualization, Validation. **Martínez-Madrid María Concepción:** Conceptualization, Supervision, Validation, Visualization. **Martin-Bermudo Francisco:** Conceptualization, Validation. **Aguado Manuel:** Conceptualization, Validation. **Saura Domingo:** Conceptualization, Project administration, Supervision, Validation, Writing – original draft. **Moreno-Chamba Bryan Mauricio:** Formal analysis, Investigation, Methodology, Software, Writing – original draft. **Valero Manuel:** Conceptualization, Supervision, Validation, Visualization, Writing – review & editing. **Martí Nuria:** Conceptualization, Project administration, Supervision, Validation, Writing – original draft. **Hosseinian Farah:** Conceptualization, Validation. **Salazar-Bermeo Julio:** Formal analysis, Investigation, Methodology, Software, Writing – original draft.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

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