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Influence of water stress and storage time on preservation of the fresh volatile profile of three basil genotypes

María J. Jordán [a,*], María Quílez [a], María C. Luna [b], Farzaneh Bekhradi [b,c], José A. Sotomayor [a], Pedro Sánchez-Gómez [d], María I. Gil [b]

[a] Murcia Institute of Agri-Food Research and Development (IMIDA), c/Mayor s/n, 30150 La Alberca, Murcia, Spain
[b] Research Group on Quality, Safety and Bioactivity of Plant Foods, Food Science and Technology Department, CEBAS-CSIC, Espinardo, Spain
[c] Department of Horticultural Sciences, University College of Agriculture and Natural Resources, University of Tehran, Iran
[d] Vegetal Biology Department (Botany), University of Murcia, Campus de Espinardo s/n, E-30100 Murcia, Spain

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ABSTRACT

The main goal of the present study was to describe the volatile profile of three different basil genotypes (Genovese and Green and Purple Iranian), and the impact that water stress (75% and 50% field capacity) and storage time (up to 7 days) have under mild refrigerated conditions. The chromatographic profile pointed to three different chemotypes: linalool/eugenol, neral/geranial, and estragol, for Genovese, Green, and Purple genotypes, respectively. Water stress depleted the volatile profile of these three landraces, due to a reduction in the absolute concentrations of some of the components related to fresh aroma (linalool, nerol, geraniol and eugenol). The stability of the basil volatile profile during storage varied depending on the water stress that had been applied. Concentration reductions of close to 50% were quantified for most of the components identified in the Purple genotype.

1. Introduction

Basil (*Ocimum basilicum* L.), a herbaceous aromatic plant that may be annual or perennial, is native to tropical regions of India, Southern Asia and Africa (Putievsky & Galambosi, 1999). Because of its unique flavor, basil leads the market among aromatic and culinary species, and, accordingly, it is cultivated for commercial purposes in several countries, including India, France, Morocco, Iran, Egypt, Hungary, Indonesia, Italy, United States, Greece and Israel (Shatar et al., 2007; Srivasta, 1980). The aroma of this herbaceous plant can be attributed to the essential oil synthesized and stored in the glandular trichomes of the leaves (Sangwan, Farooqi, Shabih, & Sangwan, 2001). As is common in aromatic plants, basil essential oil exhibits a complex and variable composition, depending on the geographical origin of the plant, and therefore, on its genetic inheritance. As a consequence, several major volatile components that define the basil essential oil chemotype have been described, including citral, eugenol, linalool, 1,8-cineole, methylchavicol, and methyl cinnamate (Lee, Umano, Shibamoto, & Lee, 2005; Vieira & Simon, 2000).

Due to its commercial value, both researchers and growers have focused their attention on production of the essential oil with its characteristic volatile profile, and many bibliographic references are related to the effects that different agronomic practices, *in vitro* cultures and different extraction methods have on the quantitative composition of basil essential oil. Among them, it is interesting to note the research related with *in vitro* cultures (Bhuvaneshwari et al., 2016), the use of growth regulating agents (Mirzajani, Hadavi, & Kashi, 2015), harvest period and cultivation conditions (Yaldiz, Gul, & Kulak, 2015), water stress (Ekren et al., 2012; Radácsi et al., 2010), salt stress (Tarchoune, Baâtour, Harrathi, & Cioni, 2013a; Tarchoune, Baâtour, Harrathi, & Hamdaoui, 2013b), plant nutrition (Bihter, Bintuð, Özgür, & Dilek 2016), hybridization (Santos da Costa et al., 2014), UV elicitation (Bertoli et al., 2013), number of cuts (Nicoletto, Santagata, Bona, & Sambo, 2013), extraction processes (Chenni, El Abed, Rakotomanomana, Fernandez, & Chemat, 2016) and drying methods (Pirbalouti, Mahdad, and Craker (2013), Alves et al., 2015).

To analyze the essential oil volatile profile of different species, several analytical methods have been developed, including hydrodistillation, solvent extraction or simultaneous distillation–extraction (SDE), and headspace methods (Lucchesi, Chemat, & Smadja, 2004). Of these, the simplest method is hydrodistillation; however, it is well known that, occasionally, some essential oil

* Corresponding author.
E-mail address: mariaj.jordan@carm.es (M.J. Jordán).

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constituents are not present in the plant itself but are artifacts of the extraction process (Kubeczka, 2010). These difficulties can be minimized by the application of headspace-solid phase microextraction (HS-SPME) (Alpendurada, 2000), a sample preparation method that is not only environmentally friendly but also avoids most of the factors responsible for the transformation of the analyzed components (Chen, Poon, & Lam, 1998). The main limitation in the application of standard HS-SPME for evaluating plants as sources of essential oils are the quantitative differences in volatile components to those obtained by direct analysis of essential oil obtained by steam distillation from the same plant (Kovacevic & Kac, 2001). Because of this, Dawidowicz, Szewczyk, and Dybowski (2016) proposed a modified application of HS-SPME for quality evaluation of essential oil plant materials. These authors demonstrated that the quantitative relations of essential oil components established by HS-SPME procedure and direct analysis of essential oil/steam distillation are similar when the plant material in the HS-SPME process is replaced by its suspension in oil of the same physicochemical character as that of the SPME fibre coating. In contrast, extracts obtained by solvent extraction with different organic solvents may not be considered as true essential oils; however, they usually possess aroma profiles that are almost identical to the raw material from which they have been extracted (Kubeczka, 2010).

A list of the most commercial basil species was published by Makri and Kintzios (2007). This list included Sweet (*Ocimum basilicum*), Genovese (*Ocimum basilicum genovese*), Bush or Greek (*Ocimum basilicum minimum*), Purple (*Ocimum basilicum purpurascens*), Lettuce-leaf (*Ocimum basilicum crispum*), Scented (*Ocimum basilicum odoratum*), Holy (*Ocimum canum* or *Ocimum sanctum*), Camphor (*Ocimum kilimandscharicum*), Peruvian (*Ocimum micranthemum*) and Thrysiflora (*Ocimum thrysiflora*), most of which are cultivated for the production of essential oil, their dried leaves, or as an ornamental.

In the global market, the quality of fresh basil is conditioned by its color and aroma retention (Makri & Kintzios, 2007), however, fresh basil has a very short shelf-life because it is a highly perishable, chilling-sensitive herb (Aharoni et al., 2010). For this reason, we previously studied the effect of deficit irrigation (75% and 50% of field capacity) on the postharvest quality of different basil genotypes (Bekhradi et al., 2015), finding that although the aroma loss (measured on a 1–9 hedonic scale) increased slightly during storage, it was well preserved (in terms of the non-development of bad odors) until the end of the storage. Thus, as a fresh herb, basil can be exposed to water stress during cultivation without diminishing its subsequent sensory quality characteristics (darkening, dehydration and aroma loss) during storage for 7 days compared to a control group. Contrary to this and related to basil essential oil production and volatile profile changes during storage, Da Silva et al. (2005) concluded that there was a linear decrease in the essential oil content linked to an increase in the relative concentrations of eugenol and linalool in the case of fresh basil shoots packaged in PVC and stored for up to 9 days at 10 °C. In this line, Silva Rosado, Brasil Pereira Pinto, Vilela Bertolucci, Ramos de Jesus, and Alves Barreto (2013) assessed the qualitative and quantitative changes in the essential oil from dry leaves of sweet basil stored in different types of packaging at −20, 4, and 25 °C over a twelve-month period. The essential oil content decreased at a rate of 0.1% per month of storage. However, as regards the volatile profile, the relative concentration of the major components identified (linalool and geraniol) did not change during the whole storage period, although the concentrations of the minor constituents varied significantly. However, Di Cesare, Viscardi, Fusari, and Nani (2001) found that storage at −20 °C for 9 months caused a quantitative decrease in the characteristic volatile compounds, eugenol and linalool, accompanied by the disappearance of the green note compounds (*Z*)-2-penten-1-ol, hexanal, (*E*)-2-hexenal and (*Z*)-3-hexen-1-ol.

The importance of studying the volatile profile lies in the fact that it affects the specific aroma of a plant. As is known, variations in the aromatic profile of some species not only depend on the cultivar, but also on agronomical practices and storage time and conditions, among other factors, which affect the concentration of sensory-relevant aromatic components. Water stress and storage time are known to affect the relative chemical composition of basil essential oil, but, to the best of our knowledge, there are no references related to how water stress can contribute or not to the maintenance of the volatile profile of basil stored under mild refrigeration conditions. With this in mind, changes in the absolute quantitative volatile profile of three basil genotypes grown at three different watering levels and stored for up to 7 days were studied.

2. Materials and methods

2.1. Chemicals and reagents

Hexane was purchased from Panreac Quimica S.L.U. (Madrid, Spain). Anhydrous sodium carbonate, analytical grade, was acquired from Scharlau Chemie S.A. (Sentmenat, Spain). Pure reference standards for 2-octanol, α-pinene, β-pinene, β-myrcene, limonene, (*Z*)-β-ocimene, terpinolene, caryophyllene, α-farnesene, α-humulene, valencene, δ-cadinene, 1-octen-3-ol, (*Z*)-sabinene hydrate, linalool, borneol, nerol, geraniol, α-terpineol, citral, fenchone, camphor, bornyl acetate, eugenol, 1,8-cineole, estragol and eugenol methyl ether were supplied by Sigma-Aldrich (Madrid, Spain). Chavicol, (*Z*)-α-bisabolene and β-bisabolene were obtained from Parchem (New Rochelle, NY) and 2,4-dimethylbenzaldehyde from Acros Organics (Thermo Fisher Scientific, Geel, Belgium).

2.2. Plant material and experimental design

Commercial seeds from three basil (*Ocimum basilicum* L.) cultivars, Purple and Green from Ardestan and Mobarake (Isfahan province, Iran) and the Genovese variety Dolly from Enza Zaden España (Albujón, Murcia, Spain) were used for the development of this research. The experimental design was previously reported (Bekhradi et al., 2015), but the most relevant information is briefly detailed for the purpose of this study. This assay was performed in an experimental area of IMIDA (Murcia Institute of Agri-Food Research and Development) at Torreblanca (37°47′ N, 0°54′ W and 30 m above sea level) in the province of Murcia (Spain). One of the greatest problems that the southeast of the Iberian Peninsula has is its semi-arid climate and general lack of water. For this reason, the study consisted of applying three irrigation levels, the first to achieve 100% field capacity (FC) and two deficit irrigation treatments of 75% and 50% FC, in an attempt to ascertain whether water stress affects the volatile profile of stored basil.

Seeds were sown in commercial nursery trays under controlled temperatures (25–30 °C). After 10 days, the seedlings were transferred to 1.3-L pots filled with commercial peat moss at a density of 250 plants/m^2, with a total number of 684 pots. Three equally spaced plants per pot were grown in a greenhouse with controlled temperature and light intensity (25–30 °C and 145–118 W/m^2). Crop management followed the practices described in Bekhradi et al. (2015). An assay with a randomized split plot (irrigation being the main plot and the genotype the subplot) and three replications (per water treatment and genotype) was designed. Due to the different growth rates, harvesting time was set at 30, 36 and 43 days after transplanting for Purple, Green and Genovese genotypes, respectively. The shoots were harvested by cutting just above the peat moss. For each irrigation treatment, the shoots were prepared for storage and further analysis.

For storage of the fresh basil, three shoots per package were stored in perforated polypropylene trays (187 × 136 × 57 mm), sealed with 35-μm perforated polypropylene lids. The packages were stored with a 12 h photoperiod under commercial light conditions (6.7 ± 2 μmol m^{-2} s^{-1}) for up to 7 days at 12 °C.

2.3. Extraction of volatile compounds

To ascertain the effect of the deficit irrigation treatments and the storage time on the conservation of the fresh volatile profile, the profile was analyzed immediately after harvest (0 d) and after 5 and 7 days of storage. Fresh stems of individual plants were taken and the leaves from the first nodes were cut into pieces for analysis. For each genotype, 81 plants were harvested, corresponding to 9 plants per irrigation assay and per sampling day, making a total of 243 individual plants analyzed.

Volatile compounds were isolated using a liquid extraction technique. A standard internal solution (200 μL/L) of 2-octanol was added to 2 g of chopped fresh leaves. Volatile components were then extracted with 50 mL of hexane by stirring for 2 h with a magnetic stirrer at approximately 2 °C. The resulting mixture was centrifuged (4000 rpm) for 10 min and the organic layer was dried with sodium sulfate and concentrated to 1 mL using an evaporator (Syncore Polyvap R-96; Büchi, Flawil, Switzerland) operating at 40 °C under vacuum conditions. The extracts were kept in vials at −80 °C prior to their corresponding analysis.

2.4. Gas chromatography–mass spectrometry analysis

The qualitative and quantitative analyses of the volatile compounds were made using an Agilent Technologies 6890 N gas chromatograph (GC) (Santa Clara, CA) equipped with a 30 m × 0.25 mm i.d. HP-5 (crosslinked phenyl-methyl siloxane) column with 0.25 mm film thickness and a DB-Wax 52CB (polyethylene glycol/Carbowax) 30 m × 0.32 mm i.d. with 1.0 μm film thickness. Both columns were supplied by Agilent Technologies (Santa Clara, CA).

Helium was used as the carrier gas (constant pressure, β-ionone eluting at 27.60 min for HP-5MS column and 47.02 min for DBWax column) and the split ratio was set to 20:1 with 1 μL of injected sample. The GC was linked to an Agilent model 5972 inert mass spectrometry detector. The initial oven temperature was set at 60 °C then increased at 2.5 °C/min to 155 °C and finally raised to 250 °C at a rate of 10 °C/min. The injection port and the transfer line to the mass selective detector were kept at 250 and 280 °C, respectively.

The mass spectrometer was operated in electron impact ionization mode with an ionizing energy of 70 eV, scanning from m/z 50–350 at 3.21 scan/s. The quadrupole temperature was 150 °C and the electron multiplier voltage was maintained at 1300 V. The individual peaks were identified by the retention times and retention indices (relative to C6–C17 *n*-alkanes), compared with those of known compounds, and by comparison of mass spectra using the NBS75K library and spectra obtained from standards. Table 1 shows the compounds, along with their retention indices, and the target and qualifier ions considered for their positive identifications.

2.5. Quantification

For the purpose of quantifying the identified components, linear regression models were obtained using standard dilution techniques with 2-octanol as the internal standard. Samples were run in triplicate. Target and qualifier ions were used in the identification and quantification of each component by the mass spectrometry data system (Table 1). Standard reference compounds were used in all cases if commercially available. For the quantification of the eight compounds that were not available, linear regression of similar components was used. In this case α-bergamotene was substituted by α-farnesene; β-farnesene by α-humulene; germacrene D and germacrene A by valencene; γ-elemene by α-farnesene; γ-cadinene and τ-cadinol by δ-cadinene and 3,4-dimethylbenzaldehyde by 2,4-dimethylbenzaldehyde.

2.6. Statistical analysis

The data are reported as the mean ± standard deviation for 9 plants per watering level and storage time. ANOVA was used to compare irrigation treatments and storage times for each genotype. Means were compared by Tukey's HSD, and a probability level of $p < 0.05$ was adopted as the criterion for statistically significant difference. The results were processed by Excel and STATGRAPHICS Centurion XV.II programs.

3. Results and discussion

3.1. Volatile profile

The volatile profile is one of the most important factors related to basil quality. Based on this premise, for the first time, the absolute quantification of the aromatic fraction from three basil

Table 1
Kovat's Indices (KI), primary ions (T), and secondary ions (Q1 and Q2) used for identifying and quantifying volatile compounds.

Compunds	KI[a]	KI[b]	T	Q1	Q2
α-Pinene	912	1038	93	91	92
β-Pinene	952	1094	93	69	91
β-Myrcene	966	1172	93	69	91
Limonene	1008	1198	68	93	67
(*Z*)-β-ocimene	1031	1238	93	91	79
Terpinolene	1078	1299	93	121	91
Caryophyllene	1406	1581	93	133	69
α-Bergamotene	1422	1578	93	55	91
α-Humulene	1434	1675	93	88	121
β Farnesene	1443	1664	69	93	67
Germacrene D	1457	1724	161	79	91
γ-Elemene	1471	1644	93	55	91
Germacrene A	1478	1741	81	93	67
γ-Cadinene	1480	1749	161	91	105
β-Bisabolene	1487	1727	69	93	67
(*Z*)-α-Bisabolene	1529	1761	93	79	80
Alcohols					
1-Octen-3-ol	952	1456	57	72	55
(*Z*)-Sabinene hydrate	1053	1469	93	71	91
Linalool	1093	1552	71	93	55
Borneol	1170	1704	95	110	93
Nerol	1241	1806	69	67	93
Geraniol	1268	1854	69	67	93
α-Terpineol	1199	1695	59	93	121
τ-Cadinol	1617	2173	161	91	105
Aldehydes					
3,4-Dimethylbenzaldehyde	1223	1793	133	134	105
Neral	1253	1690	69	94	109
Geranial	1283	1731	69	84	94
Ketone					
Fenchone	1078	1389	81	80	84
Camphor	1144	1521	95	81	108
Ester					
Bornyl acetate	1295	1579	95	93	121
Phenols					
Chavicol	1270	2338	133	134	105
Eugenol	1359	2166	164	131	77
Ether					
1,8-Cineole	1011	1212	81	71	108
Estragol	1208	1660	148	147	121
Eugenol methyl ether	1400	2033	178	105	147

[a] HP-5 column.
[b] DB-WAX column.

genotypes grown under three different watering levels and stored refrigerated for up to 7 days are shown in Tables 2–4. The chromatographic analysis of the isolated volatile fractions allowed identification of a total of 26, 28, and 18 components that made up the major volatile profile for Genovese, Purple and Green Iranian genotypes, respectively. The finding that basil accumulates monoterpenes, sesquiterpenes and phenylpropanoids, while the main components of the essential oil are oxygenated monoterpenes and phenylpropane derivatives agrees with the observations of Bernhardt et al. (2015), and Hiltunen and Holm (1999).

For the Genovese genotype, the major components that defined the chemotype were the terpenoids linalool, 1,8-cineole and α-bergamotene, along with the phenylpropanoid eugenol (Table 2). These components could be related to the hay-like, sweet and spicy sensory attributes described for this variety by Bernhardt et al. (2015). The relative concentration of these components (eugenol > linalool > 1,8-cineole) in Genovese basil oil has been reported previously by Rojas et al. (2012). However, Tarchoune, Baâtour, Harrathi, and Hamdaoui (2013b) described a different relative abundance among the major components that comprise the chemotype of this genotype, linalool (45.9%), 1,8-cineole (16.7%) and eugenol (10.3%) being the most abundant compounds quantified. Variations between the orders of abundance among major components are probably more related to the genetic inheritance of the plants, i.e., to their respective origins, than to environment factors.

Purple Iranian genotype exhibited a single chemotype defined by estragol (methyl chavicol, Table 3). This epoxide gives a pleasant fruity/anise aromatic note to the plant, and, according to Al-Kateb and Mottram (2014), it could be considered as the component that defines the essential oil quality of basil, since it is one of the most important characteristic volatile compounds in this basil species. Linalool and (*Z*)-β-ocimene, also detected at high concentrations, are probably responsible for the pleasant fragrance of this variety. Similar to these results, Pirbalouti et al. (2013) published the chemical oil composition of this basil landrace. For these authors the main components quantified were methyl chavicol (65.63%), linalool (6.11%) and (*E*)-α-bergamotene (4.09%). However, studies carried out by Ekren et al. (2012) regarding the volatile profile of purple basil from Turkey showed that the major volatile components quantified in this genotype were linalool, eugenol and methylchavicol.

As regards the Green Iranian genotype, the component that defines the chemotype of this species was citral, considered as the sum of neral plus geranial (Table 4). Major components in the volatile profile also included the sesquiterpenes caryophyllene, germacrene D and (*Z*)-α-bisabolene. In line with the idea that genetic factors are the principal agents in aromatic plants that explain differences in the oil chemical profile, Pirbalouti et al. (2013) found that the major components that define the aromatic composition of the Green Iranian basil landrace were methylchavicol (44.93%), geranial (17.61%) and neral (15.04%).

Bernhardt et al. (2015) established the correlation between GC–MS profile and the sensory attributes of different basil gene bank accessions. For these researchers, the intense lemon odor described for this genotype is closely correlated with the presence of citral

Table 2
Effect of storage time, depending on the water stress applied, on the fresh volatile profile (mg/kg) of Genovese basil genotype.

Genovese	100% field capacity			75% field capacity			50% field capacity		
Compound	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7
Terpenic hydrocarbons									
α-Pinene	2.0 ± 0.65	1.8 ± 0.70	2.1 ± 0.40	1.7 ± 0.31^{a}	2.3 ± 0.65^{b}	1.6 ± 0.45^{a}	2.3 ± 0.58^{a}	3.72 ± 0.65^{b}	2.4 ± 0.51^{a}
β-Pinene	3.6 ± 0.87^{a}	3.9 ± 1.01^{a}	7.4 ± 2.68^{b}	2.7 ± 0.60^{a}	8.2 ± 2.73^{b}	9.6 ± 2.91^{b}	6.3 ± 1.55^{a}	12.0 ± 1.82^{b}	6.6 ± 1.26^{a}
β-Myrcene	4.8 ± 0.82	4.5 ± 0.64	5.4 ± 0.57	5.1 ± 0.46	5.9 ± 1.57	5.1 ± 0.83	5.3 ± 0.76^{a}	10.1 ± 1.26^{c}	7.8 ± 1.84^{b}
Limonene	2.4 ± 0.65^{a}	12.4 ± 4.07^{b}	16.3 ± 3.93^{b}	2.3 ± 0.42^{a}	3.2 ± 0.55^{a}	14.0 ± 3.20^{b}	10.1 ± 2.24	12.2 ± 3.98	7.5 ± 1.70
(*Z*)-β-Ocimene	9.0 ± 1.92	8.0 ± 1.67	9.3 ± 1.52	8.7 ± 1,18^{b}	11.7 ± 1.73^{c}	5.9 ± 1.21^{a}	12.8 ± 1.16^{a}	16.3 ± 2.22^{b}	12.0 ± 1.44^{a}
Terpinolene	1.0 ± 0.35	1.3 ± 0.35	1.3 ± 0.29	1.0 ± 0.21	1.5 ± 0.54	1.2 ± 0.35	1.3 ± 0.33^{a}	2.1 ± 0.40^{b}	1.9 ± 0.50^{b}
Caryophyllene	1.6 ± 0.33	1.5 ± 0.46	1.4 ± 0.42	1.5 ± 0.44	1.6 ± 0.61	1.1 ± 0.47	1.7 ± 0.57	1.9 ± 0.26	2.0 ± 0.96
α-Bergamotene	30.7 ± 7.99	34.6 ± 13.80	23.2 ± 3.08	26.0 ± 7.51^{a}	35.5 ± 8.92^{b}	17.9 ± 4.44^{a}	29.1 ± 3.24	34.6 ± 6.67	25.5 ± 9.04
α-Humulene	6.6 ± 1.12	6.1 ± 1.09	6.6 ± 0.98	6.6 ± 0.61	7.2 ± 1.71	5.9 ± 2.23	7.9 ± 1.21^{a}	10.7 ± 1.30^{b}	9.7 ± 2.89^{ab}
β Farnesene	7.4 ± 2,61	9.6 ± 6.86	6.9 ± 5.13	10.5 ± 2.82^{b}	7.7 ± 1.75^{b}	3.9 ± 1.24^{a}	10.7 ± 4.45	10.9 ± 2.58	15.1 ± 3.77
Germacrene D	25.0 ± 5.83	17.9 ± 3.21	20.9 ± 4.82	18.6 ± 2.71^{b}	24.3 ± 2.38^{c}	14.4 ± 3.02^{a}	23.3 ± 6.11	22.8 ± 4.46	21.0 ± 4.92
γ-Elemene	17.7 ± 3.09	15.3 ± 3.39	13.7 ± 2.89	13.4 ± 1.81^{b}	16.1 ± 2.74^{b}	9.5 ± 2.41^{a}	16.6 ± 3.90	17.3 ± 4.33	22.0 ± 8.82
Germacrene A	11.2 ± 2.88	9.5 ± 1.99	8.9 ± 1.98	8.6 ± 1.61^{b}	9.6 ± 1.67^{b}	6.5 ± 1.91^{a}	12.4 ± 3.23	14.4 ± 1.97	11.4 ± 5.64
γ-Cadinene	7.0 ± 0.67	5.9 ± 1.64	6.2 ± 1.89	5.0 ± 1.12^{ab}	6.7 ± 1.88^{b}	4.5 ± 1.20^{a}	6.6 ± 1.88^{ab}	8.8 ± 2.07^{b}	5.1 ± 0.79^{a}
Alcohols									
1-Octen-3-ol	14.6 ± 1.40^{a}	18.0 ± 2.36^{b}	14.5 ± 1.64^{a}	13.5 ± 1.10^{a}	16.2 ± 1.86^{b}	15.1 ± 2.97^{ab}	12.9 ± 1.98^{a}	16.3 ± 2.02^{b}	14.8 ± 3.42^{ab}
(*Z*)-Sabinene hydrate	2.1 ± 0.48	1.8 ± 0.60	1.5 ± 0.33	1.6 ± 0.28^{a}	2.3 ± 0.65^{b}	1.5 ± 0.44^{a}	2.1 ± 0.53	2.9 ± 0.53	2.9 ± 0.94
Linalool	326.0 ± 34.75^{b}	247.5 ± 47.09^{a}	256.4 ± 39.23^{a}	222.1 ± 27.17^{a}	320.1 ± 19.65^{b}	190.9 ± 48.10^{a}	222.1 ± 32.18	273.4 ± 50.31	243.1 ± 56.39
α-Terpineol	8.9 ± 1.40	9.6 ± 1.65	8.5 ± 0.89	8.1 ± 0.96^{a}	11.7 ± 2.23^{b}	6.8 ± 0.67^{a}	11.9 ± 0.78	13.7 ± 1.50	13.0 ± 3.50
τ-Cadinol	12.5 ± 1.41^{b}	9.8 ± 1.66^{a}	10.6 ± 2.37^{ab}	10.6 ± 2.25	12.0 ± 2.47	9.2 ± 2.25	11.7 ± 2.16	14.6 ± 3.36	12.2 ± 3.66
Aldehydes									
3,4 Dimethylbenzaldehyde	0.7 ± 0.11^{a}	0.7 ± 0.23^{a}	2.2 ± 0.32^{b}	0.7 ± 0.11^{a}	1.6 ± 0.37^{b}	2.6 ± 0.31^{c}	0.6 ± 0.06^{a}	1.6 ± 0.49^{b}	1.5 ± 0.23^{b}
Geranial	1.1 ± 0.41	1.4 ± 0.40	2.7 ± 1.05	0.4 ± 0.22^{a}	0.2 ± 0.07^{a}	3.8 ± 0.99^{b}	1.0 ± 0.52^{b}	1.5 ± 0.35^{b}	0.1 ± 0.07^{a}
Ketone									
Camphor	5.7 ± 0.99^{a}	8.2 ± 1.72^{b}	5.5 ± 0.87^{a}	5.1 ± 0.72^{a}	9.2 ± 1.81^{b}	4.0 ± 0.43^{a}	6.8 ± 0.91^{a}	10.3 ± 1.55^{b}	7.5 ± 1.88^{a}
Ester									
Bornyl acetate	2.0 ± 0.65^{a}	2.5 ± 0.73^{ab}	3.5 ± 1.37^{b}	1.7 ± 0.84^{a}	2.7 ± 1.41^{a}	4.4 ± 1.83^{b}	3.6 ± 1.32	4.7 ± 1.81	3.6 ± 1.30
Phenols									
Chavicol	0.6 ± 0.06	0.5 ± 0.08	0.5 ± 0.08	0.4 ± 0.06^{a}	0.6 ± 0.04^{b}	0.4 ± 0.10^{a}	0.4 ± 0.09^{a}	0.6 ± 0.11^{b}	0.7 ± 0.15^{b}
Eugenol	150.3 ± 30.92	127.6 ± 12.43	139.6 ± 27.43	124.9 ± 11.72^{b}	143.9 ± 12.73^{c}	85.4 ± 16.92^{a}	151.8 ± 23.38^{a}	191.1 ± 20.74^{b}	133.3 ± 22.09^{a}
Ether									
1,8-Cineole	44.4 ± 6.80	47.3 ± 9.50	40.9 ± 7.95	44.6 ± 4,57^{b}	54.7 ± 8.10^{c}	36.4 ± 5.42^{a}	56.7 ± 9.29^{a}	85.6 ± 6.45^{b}	56.9 ± 6.01^{a}

[a]Values followed by different letters (considering the same hydric treatment) share significant differences at 95% Tukey's HSD (honestly significant difference).

Table 3
Effect of the storage time, depending on the water stress applied, on the fresh volatile profile (mg/kg) of Green Iranian basil genotype.

Green Iranian	100% field capacity			75% field capacity			50% field capacity		
Compound	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7
Limonene	0.6 ± 0.21	0.8 ± 0.22	0.6 ± 0.06	1.1 ± 0.27	1.7 ± 0.91	2.2 ± 0.61	1.1 ± 0.33[a]	1.8 ± 0.33[b]	1.8 ± 0.41[b]
Caryophyllene	22.2 ± 4.89	25.4 ± 8.47	17.5 ± 3.79	35.1 ± 10.96	35.2 ± 9.09	30.4 ± 5.89	29.2 ± 8.13	33.9 ± 3.94	34.6 ± 10.8
α-Bergamotene	11.1 ± 3.11	13.1 ± 2.58	9.5 ± 2.06	16.2 ± 7.56	19.2 ± 4.04	13.4 ± 4.19	13.2 ± 4.45	18.3 ± 4.43	14.3 ± 2.90
α-Humulene	8.0 ± 2.24[a]	11.7 ± 1.95[b]	6.6 ± 1.76[a]	13.7 ± 3.11	13.2 ± 3.03	11.1 ± 2.02	11.7 ± 2.38	13.0 ± 2.75	12.2 ± 2.90
β-Farnesene	2.4 ± 1.22	2.5 ± 0.58	1.9 ± 0.42	3.6 ± 1.12	3.0 ± 1.10	3.2 ± 0.75	3.7 ± 1.91	4.3 ± 1.56	4.1 ± 1.54
Germacrene D	15.6 ± 5.36[ab]	20.4 ± 5.14[b]	11.1 ± 3.96[a]	25.0 ± 9.60	17.3 ± 2.71	18.5 ± 4.80	17.7 ± 3.04	22.7 ± 1.56	20.9 ± 4.03
Germacrene A	0.7 ± 0.42[b]	0.2 ± 0.40[a]	0.2 ± 0.27[a]	0.2 ± 0.48	0.23 ± 0.44	0.1 ± 0.26	0.3 ± 0.48	0.4 ± 0.49	0.1 ± 0.24
β-Bisabolene	0.8 ± 0.29	1.4 ± 0.71	0.7 ± 0.24	1.1 ± 0.95	1.04 ± 0.44	0.8 ± 0.36	0.9 ± 0.46	1.2 ± 0.40	0.9 ± 0.40
(*Z*)-α-Bisabolene	19.3 ± 4.51	17.6 ± 10.97	13.9 ± 2.84	24.7 ± 7.75	23.3 ± 9.23	21.6 ± 5.50	20.8 ± 5.49	25.9 ± 5.48	17.4 ± 2.50
Alcohols									
1-Octen-3-ol	3.2 ± 1.49	5.9 ± 3.36	1.7 ± 0.62	3.7 ± 1.32[b]	4.6 ± 0.85[b]	1.4 ± 0.47[a]	5.9 ± 1.98[b]	3.4 ± 0.94[ab]	1.6 ± 0.56[a]
Nerol	13.0 ± 3.42[b]	7.1 ± 1.56[a]	5.7 ± 1.86[a]	10.6 ± 4.41	8.2 ± 1.22	8.4 ± 2.70	8.2 ± 6.16	8.5 ± 10.42	9.2 ± 3.86
Geraniol	6.9 ± 2.97[b]	1.8 ± 0.68[a]	1.7 ± 1.08[a]	4.7 ± 0.64[b]	1.8 ± 0.74[a]	1.7 ± 0.81[a]	1.8 ± 0.62	0.8 ± 0.92	1.1 ± 0.51
Aldehydes									
3,4-Dimethylbenzaldehyde	1.9 ± 0.86[a]	3.3 ± 1.25[b]	0.9 ± 0.30[a]	2.7 ± 0.89	2.9 ± 0.59	2.5 ± 1.59	2.0 ± 0.50[b]	0.9 ± 0.11[a]	0.3 ± 0.40[a]
Neral	186 ± 53.1[a]	263 ± 59.4[b]	170 ± 59.9[a]	310 ± 89.3	302 ± 82.8	240 ± 60.2	293 ± 68.2	361 ± 87.9	247 ± 74.0
Geranial	219 ± 51.7[a]	306 ± 62.0[b]	165 ± 46.1[a]	329 ± 56.9	342 ± 86.5	254 ± 56.0	290 ± 73.0	359 ± 108	317 ± 83.7
Ketone									
Fenchone	2.4 ± 1.48[ab]	3.7 ± 1.32[b]	1.2 ± 0.33[a]	4.5 ± 1.16[b]	1.9 ± 0.68[a]	3.8 ± 1.03[b]	4.5 ± 1.54	4.9 ± 0.90	5.3 ± 1.09
Phenols									
Chavicol	0.1 ± 0.12	0.1 ± 0.11	0.1 ± 0.17	0.2 ± 0.21	0.2 ± 0.30	0.2 ± 0.20	0.1 ± 0.11	0.1 ± 0.16	0.1 ± 0.12
Ether									
Estragol	13.6 ± 16.0	10.7 ± 10.4	6.2 ± 4.09	12.0 ± 14.0	3.0 ± 3.01	8.6 ± 8.07	15.2 ± 13.6	22.3 ± 23.90	39.1 ± 22.5

[a]Values followed by different letters (considering the same hydric treatment) share significant differences at 95% Tukey's HSD (honestly significant difference).

Table 4
Effect of storage time, depending on the water stress applied, on the fresh volatile profile (mg/kg) of Purple Iranian basil genotype.

Purple Iranian	100% field capacity			75% field capacity			50% field capacity		
COMPOUND	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7	Day 0	Day 5	Day 7
Terpenic hydrocarbons									
α-Pinene	0.7 ± 0.49	0.6 ± 0.25	0.4 ± 0.21	0.7 ± 0.48	0.3 ± 0.18	0.7 ± 0.22	0.5 ± 0.23	0.7 ± 0.34	0.7 ± 0.35
β-Pinene	0.4 ± 0.24[a]	0.5 ± 0.22[a]	1.2 ± 0.63[b]	0.5 ± 0.26[a]	0.3 ± 0.27[a]	1.9 ± 0.92[b]	0.6 ± 0.13[a]	0.5 ± 0.20[a]	2.5 ± 1.30[b]
β-Myrcene	4.7 ± 2.64[b]	1.7 ± 0.88[a]	1.4 ± 0.65[a]	3.0 ± 1.55	2.2 ± 1.01	2.2 ± 0.74	2.3 ± 1.23	2.8 ± 1.09	1.6 ± 1.07
Limonene	2.0 ± 1.28[a]	1.3 ± 0.51[a]	10.3 ± 3.20[b]	1.6 ± 1.05[a]	1.1 ± 0.57[a]	22.5 ± 4.60[b]	1.6 ± 0.94[a]	2.1 ± 1.40[a]	18.5 ± 9.17[b]
(*Z*)-β-Ocimene	20.2 ± 8.11[b]	10.6 ± 3.34[a]	9.8 ± 4.31[a]	9.6 ± 5.06	8.5 ± 3.49	7.9 ± 2.30	8.7 ± 3.00	14.4 ± 7.91	6.4 ± 5.57
Caryophyllene	5.2 ± 2.37	3.4 ± 1.17	3.2 ± 1.29	7.5 ± 3.58	3.2 ± 1.37	4.7 ± 3.57	5.4 ± 1.60	4.8 ± 1.80	3.6 ± 1.29
α-Bergamotene	8.4 ± 5.11[b]	2.6 ± 2.23[a]	3.9 ± 2.22[a]	7.9 ± 7.89	4.9 ± 3.14	5.1 ± 2.69	7.3 ± 5.60	7.1 ± 3.68	3.6 ± 1.24
α-Humulene	2.8 ± 1.13	2.8 ± 1.64	1.8 ± 1.04	4.2 ± 2.51	2.3 ± 1.84	2.3 ± 1.08	3.9 ± 2.13	3.0 ± 1.39	2.7 ± 1.50
β-Farnesene	7.9 ± 3.37	7.1 ± 2.12	5.3 ± 3.31	10.2 ± 4.88[b]	4.1 ± 2.47[a]	4.0 ± 2.49[a]	7.3 ± 2.86[ab]	8.9 ± 3.93[b]	4.4 ± 2.73[a]
Germacrene D	7.2 ± 2.69[b]	6.6 ± 2.44[b]	4.0 ± 1.93[a]	8.7 ± 4.50[b]	4.4 ± 2.49[a]	5.4 ± 2.47[ab]	7.4 ± 2.93	6.1 ± 1.73	4.5 ± 2.79
γ-Elemene	3.7 ± 2.08[b]	1.3 ± 0.77[a]	0.5 ± 0.30[a]	1.2 ± 0.93	1.2 ± 1.33	0.9 ± 0.68	1.7 ± 1.27	1.4 ± 0.93	0.7 ± 0.65
Germacrene A	3.5 ± 2.08[b]	3.0 ± 1.22[b]	0.8 ± 0.77[a]	4.4 ± 2.79[b]	1.3 ± 0.85[a]	2.4 ± 1.36[ab]	3.9 ± 2.15[b]	3.3 ± 1.05[b]	1.0 ± 1.08[a]
γ-Cadinene	5.6 ± 2.56[b]	3.6 ± 1.81[ab]	1.9 ± 1.13[a]	4.5 ± 2.77	2.6 ± 1.51	3.8 ± 2.26	3.2 ± 1.82	3.0 ± 1.06	2.9 ± 1.29
1-Octen-3-ol	6.3 ± 3.10[b]	5.4 ± 1.33[ab]	3.3 ± 1.50[a]	8.0 ± 2.98[a]	4.8 ± 1.69[b]	4.5 ± 1.85[b]	5.3 ± 1.51[ab]	6.3 ± 1.51[b]	3.9 ± 1.12[a]
Linalool	63.0 ± 36.1[b]	47.4 ± 23.7[ab]	29.5 ± 9.33[a]	82.5 ± 42.7[b]	29.0 ± 5.98[a]	49.1 ± 20.1[ab]	60.0 ± 24.5	40.7 ± 15.9	41.7 ± 17.7
Borneol	0.5 ± 0.69	0.1 ± 0.11	0.1 ± 0.25	0.2 ± 0.17	0.1 ± 0.07	0.1 ± 0.15	0.3 ± 0.59	0.2 ± 0.17	0.2 ± 0.19
α-Terpineol	3.9 ± 2.88	3.4 ± 2.03	1.5 ± 1.29	4.2 ± 2.52[b]	1.7 ± 1.10[a]	2.1 ± 1.28[a]	3.5 ± 3.05	2.5 ± 0.90	2.2 ± 1.08
τ-Cadinol	14.0 ± 4.99[b]	7.4 ± 2.88[a]	3.8 ± 1.99[a]	8.9 ± 4.53	4.6 ± 2.71	6.1 ± 3.98	8.3 ± 3.86	7.0 ± 1.74	5.9 ± 3.59
Aldehydes									
3,4-Dimethylbenzaldehyde	2.9 ± 1.02[b]	2.4 ± 0.88[ab]	1.5 ± 0.70[a]	3.5 ± 0.86[b]	2.4 ± 0.37[a]	2.3 ± 0.91[a]	3.3 ± 1.24[b]	3.7 ± 1.26[b]	1.83 ± 0.83[a]
Geranial	n.d.	n.d.	3.0 ± 1.78	0.3 ± 0.68	0.2 ± 0.42	7.9 ± 4.38	n.d.	n.d.	7.4 ± 4.05
Ketone									
Fenchone	5.0 ± 6.09	4.2 ± 2.46	3.0 ± 3.81	8.3 ± 7.21	6.8 ± 4.97	5.5 ± 4.84	14.5 ± 9.83	18.0 ± 9.36	6.0 ± 4.29
Camphor	2.4 ± 3.96	0.7 ± 1.53	2.3 ± 2.67	0.1 ± 0.24	1.4 ± 1.53	1.6 ± 2.68	2.6 ± 4.25	1.2 ± 2.25	2.8 ± 2.82
Ester									
bornyl acetate	0.2 ± 0.24	0.9 ± 1.59	1.1 ± 0.69	0.1 ± 0.19[a]	0.1 ± 0.28[a]	2.3 ± 1.62[b]	n.d.	n.d.	2.0 ± 0.97
Phenols									
Chavicol	1.6 ± 1.09[b]	0.4 ± 0.18[a]	n.d.[a]	1.2 ± 0.72[b]	0.3 ± 0.29[a]	n.d.[a]	1.1 ± 0.48[b]	0.4 ± 0.26[b]	n.d.[a]
eugenol	1.7 ± 0.79[b]	n.d.[a]	n.d.[a]	1.0 ± 1.41[b]	n.d.[a]	n.d.[a]	n.d.[a]	0.6 ± 0.49[b]	n.d[a]
Ether									
1,8-Cineole	10.7 ± 5.91[b]	9.3 ± 1.50[b]	4.0 ± 2.33[a]	9.3 ± 6.28[b]	4.3 ± 2.69[a]	5.7 ± 2.62[a]	9.3 ± 3.13[b]	8.2 ± 3.97[b]	4.4 ± 2.95[a]
Estragol	243 ± 42.9[c]	153 ± 34.4[b]	115 ± 32.0[a]	279 ± 118[b]	166 ± 56.5[a]	174 ± 45.8[a]	242 ± 61.6[b]	244 ± 51.0[b]	123.3 ± 44.8[a]
Eugenol methyl ether	2.9 ± 1.12[b]	2.1 ± 0.93[ab]	1.5 ± 0.94[a]	4.2 ± 2.89	2.5 ± 1.78	2.7 ± 1.61	2.6 ± 0.92[ab]	4.0 ± 2.64[b]	1.6 ± 0.67[a]

[a]Values followed by different letters (considering the same hydric treatment) share significant differences at 95% Tukey's HSD (honestly significant difference).

among other minor components, such as isocitral, octanal, α-terpinene and β-cubebene.

3.2. Impact of water stress on volatile profile

To assess the effect of the water stress on the volatile profile, only data from freshly picked plants were considered. The heterogeneity among plants was evident, as can be observed in Tables 2–4. The chemical variability detected among individual plants that were subjected to the same watering level did not allow for a real assessment of the impact of water stress on the volatile profile of these basil genotypes.

Genovese basil seems to be susceptible to water stress, as can be observed in Fig. 1(a). In this genotype, water stress led to an increase in the concentration of some monoterpene hydrocarbons (β-pinene, limonene and (*Z*)-β-ocimene), along with the sesquiterpene α-humulene, the ester bornyl acetate, the terpenic alcohol α-terpineol, the ketone camphor and the ether 1,8-cineole. These increases were accompanied by a decrease in the concentration of the alcohol linalool and the phenolic component chavicol. Studies carried out by Radácsi et al. (2010) also found that the concentration of linalool decreases with water stress. Contrary to our findings, studies conducted by Simon, Reiss-Bubenheim, Joly, and Charles (1992), and later by Khalid (2006) regarding the effect of water stress on sweet basil essential oil composition showed that linalool and methylchavicol increased their relative concentrations as water stress increased, while the relative proportion of sesquiterpenes decreased.

Alcohols are the components normally related with the freshness and balsamic aroma of the essential oils of certain herbs. For example, Wojtowicz, Zawirska-Wojtasiak, and Przygoński (2007) found that (*Z*)-sabinene hydrate was the dominant aroma in marjoram (*Origanum majorana* L.) despite the fact the linalool was quantified as the major volatile compound. In the same line, Bernhardt et al. (2015) did not describe linalool as a special contributor to the sensory profile of basil. However, the role of every component in the pleasant aroma of aromatic herbs depends on its threshold and the relative concentration of its components. In fact, in the case of linalool, some authors affirm that there is a good correlation between the linalool content and any perceived fruity-flowery flavor (Ortiz, Graell, & Lara, 2011).

Terpenic hydrocarbons have been reported to impart a spicy and woody aroma (Jordán, Margaría, Shaw, & Goodner, 2003). Taking into account these considerations, and the fact that the volatile compounds present at the highest concentrations are not always the main contributors to oil aroma (Erickson & Covey, 1980), our results suggest, contrary to those published by Simon et al. (1992) and Khalid (2006), that water stress depleted the aroma quality of the Genovese basil genotype. In order to confirm this result, further studies on the key aroma compounds in this variety are necessary.

For Iranian Green basil genotype (Fig. 1b), concentrations of nerol and geraniol decreased significantly as water stress increased. This agrees with Harley (2013), who affirmed that under a moderate stress, non-oxygenated terpenes are not influenced, while oxygenated ones can be drastically reduced. Although these components are minor components in the aromatic fraction of this landrace, their contributions to the overall flavor of basil needs to be considered. These components have been described as having a very floral aroma that contributes to the aromas of various foods and beverages, such as fruits, juices, wines and beers. In fact the olfactory description for nerol and geraniol are citrus-rose and rose, with odor thresholds of 80 and 7 μg/L, respectively. To tentatively know their proximal OAV (odor activity value) levels, the positive incidence of the presence of these two monoterpene alcohols on the desirable aroma of basil can be assumed. In fact, they are both reported as being among the major contributors to the intense sweet odor of basil (Bernhardt et al., 2015).

Water stress increased the levels of neral and geranial in the volatile profile of this genotype. Citral is the mixture of isomers that clearly defines the lemon odor in many plants and fruits. An increase in their concentration should improve the desirable aroma of basil. At the same time, components that also increased their presence were fenchone (camphor-like smell), 1-octen-3-ol (earthy type odor) and the terpenic hydrocarbons limonene (citrus type flavor), caryophyllene (spicy flavor), α-humulene (bitter and woody smell) and germacrene D (woody type odor). Considering that all these components are special contributors to the spicy and woody aroma of basil, the higher level of citral (to the detriment of nerol and geraniol concentrations) means that the aroma of this genotype is sensitive to deficit irrigation, an irrigation level equivalent to 75% of field capacity being sufficient to cause such changes.

Contrary to that observed in Green and Genovese genotypes, hardly any statistically significant changes were detected in the volatile profile of Purple Iranian after the water stress treatments. For this genotype the chemical variability detected among individual plants did not permit the real changes associated with deficit irrigation to be detected. As can be observed in Fig. 1c, only γ-elemene, τ-cadinol and eugenol exhibited statistically significant differences in concentration between the water stress levels applied. Although this effect was barely detectable at a quantita-

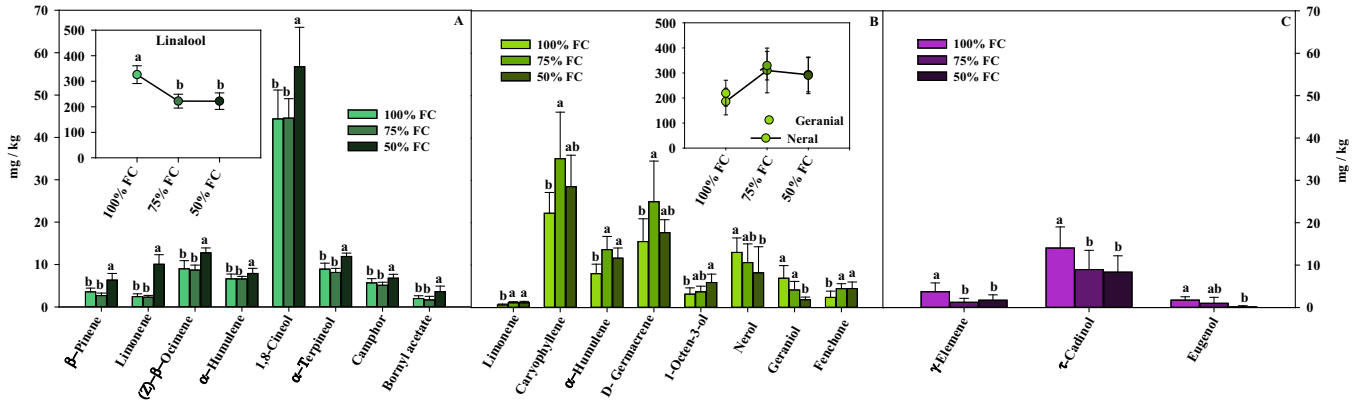

Fig. 1. Effect of water stress on the volatile profile of freshly picked Genovese, Green Iranian and Purple Iranian basil plants. Bars are the mean standard deviation of 9 replicates. Bars with different letters are significantly different ($P < 0.05$) according to Tukey's HSD (Honestly Significant Difference). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

tive level, eugenol is considered as one of the components conferring the spicy odor and aroma (taste) to this plant, despite only being present at low concentration. Bekhradi et al. (2015) did not report changes in the sensory perception of Purple basil grown at different irrigation levels, although in the present study some modifications were attributed to the water stress. Studies carried out by Ekren et al. (2012) on the effect of watering level on the volatile composition of Purple basil essential oil pointed to a positive effect of water stress on the essential oil composition. According to these authors, this positive effect would be related to a decrease in the relative concentrations of linalool and eugenol, and an increase in methylchavicol. These results are hard to extrapolate, since no statistical analyses were reported by these authors regarding the effect of water stress on the essential oil composition.

3.3. Impact of storage on volatile profile

As expected, the stability of the volatile profile of basil during 7 days of storage at 12 °C depended on the water stress that had been applied to the plants and the genotype under study. As mentioned above, the watering level affected the volatile profile of all three basil genotypes, and the different ratios between the absolute concentrations of the aromatic components affected the behavior during storage, depending on the metabolism of the plant.

For example, in the case of the Genovese basil genotype (Table 2) plants grown under a watering level equivalent to 100% field capacity (no water stress), terpenic hydrocarbons hardly varied between the different storage times, and only β-pinene and limonene increased in concentration with storage time (at Days 7 and 5, respectively). Along with the variations observed in these two components in the water stress study, this finding suggests that the concentration of these two terpenes can be considered as stress indicators for these plants. Regarding alcohols, 1-octen-3-ol, linalool and τ-cadinol suffered variations during storage. It is interesting to note that the concentration of linalool decreased to a statistically significant degree, coinciding with increased concentrations of β-pinene and limonene. This can be attributed to the fact that linalool is the precursor of a wide range of monoterpenes (Lasky & flavouring synthetics, 2009). Other components that showed modified concentrations during storage were 3,4-dimethylbenzaldehyde, bornyl acetate and camphor. This last ketone evolved in a similar way to 1-octen-3-ol, since the concentration of both components had increased by Day 5, but had fallen again to the initial level by Day 7. When the water stress was evaluated, variations in the concentrations of the volatile components were more evident, and were especially noticeable in plants grown under a watering regime equivalent to 75% of field capacity. It is also interesting to note that most of the components showing modified concentrations during storage reached their highest levels at Day 5, after which they fell again, so that by Day 7 they were present in the same or even lower concentrations than those detected at harvest. The same phenomenon was observed for α-pinene, β-pinene, (*Z*)-β-ocimene, 1-octen-3-ol, 3,4-dimethylbenzaldehyde, camphor, chavicol, eugenol and 1,8-cineole, in genotypes cultivated under both water stress treatments (75% and 50% of FC).

As regards terpenic hydrocarbons, at 75% FC the components that also followed this line were α-bergamotene, germacrene D, γ-elemene and germacrene A. Once again, a correlation between β-pinene and linalool was observed. In this case, both components showed a similar tendency to that observed in plants not subjected to any water stress, i.e. an increase in β-pinene concentration at Day 7, coinciding with a decrease in the linalool level. However, similarly to (*Z*)-sabinene hydrate and α-terpineol, the linalool concentration had increased by Day 5 of storage. This reflects the description given in the above paragraph since, after applying a light water stress to the plants, it was observed that the fifth day of storage was crucial for any quantitative changes occurring in the volatile profile of these plants. At the same time, components that had increased in concentration by Day 7 of storage were β-pinene, limonene, geranial and bornyl acetate.

The plants grown at 50% FC showed changes similar to those mentioned above during storage, although fewer terpenic hydrocarbons and alcohols were affected. In this case, the concentrations of other components not mentioned before, including β-myrcene, terpinolene and α-humulene, increased by Day 5 of storage, before falling again by Day 7. Unlike in the other two treatments, the concentration of limonene did not change with storage time, probably due its high concentration at harvest, as a consequence of the water stress. The concentration of geranial changed during storage in plants that had been submitted to both water stress levels. However, and probably due to the heterogeneity among individual plants, the same tendency was also detected in plants not subjected to deficit irrigation.

The importance of knowing these variations that take place in the volatile profile during storage is based on the fact that aroma is one of the most appreciated food characteristics, and volatile flavor compounds play a key role in determining the perception and acceptability of products by consumers. Changes in the aroma of sweet basil during storage at low temperature have not been studied in depth. Our results suggest that storage reduces the aromatic quality of this genotype, which was particularly evident on Day 5, especially in plants that had previously been submitted to a water stress equivalent to 75% FC.

The absolute volatile profile of the Green Iranian basil genotype also exhibited changes during storage (Table 3), but for this landrace the high intraspecific variability detected among individual plants made it difficult to ascertain statistically significant variations. The most significant changes were observed in plants grown at 100% FC. In this case, as detected for the water stress treatments, the concentration of the terpenic alcohols, nerol and geraniol, decreased during storage in favor of an increase in their corresponding aldehydes, neral and geranial, at Day 5 of storage. According to Iijima, Wang, Fridman, and Pichersky (2006), the increase in geranial concentration could be explained by the action of dehydrogenases in the glands of sweet basil that transform the alcohol geraniol into the corresponding aldehyde geranial. Another component that also decreased in concentration during storage was 1-octen-3-ol, following all three treatments. Thus, as a major conclusion for this landrace, the effect of storage time on the volatile profile depended on the watering level applied to the plants, since (probably due to the previous water stress) changes were only detected in plants watered at 100% FC. Components sensitive to this effect were those that defined the chemotype of this landrace, neral and geranial, along with the alcohols 1-octen-3-ol, nerol and geraniol.

By contrast, Purple Iranian genotype was the most sensitive of the three landraces to storage, although, as mentioned above, it was the most stable genotype in the face of water stress. According to the results shown in Table 4, storage dramatically damaged the volatile profile of this genotype from Days 5–7, when the concentrations of most of the components identified decreased to a statistically significant extent. The great variability detected among individual plants did not permit the true assessment of this phenomenon in some of the components analyzed, although the volatile concentrations showed a tendency to decrease in all of them, with the exception of β-pinene and limonene, whose concentrations increased with stress, as described for the Genovese genotype. The fact that only very low modifications in the volatile profile were recorded for Purple Iranian genotype after the application of water stress, meant that the effects of storage time

appeared quite similar for the different watering levels. An increased sample size, i.e. ignoring the water stress effect and grouping the 81 plants as a homogeneous group, verifies this statement: storage damages the aromatic quality of this *Labiatae*. In line with this, the concentrations of estragol and linalool, components that define the chemotype of Purple basil, had fallen by half by Day 7 of storage.

4. Conclusions

Water stress not only depletes the fresh volatile profile of these three landraces but also influences the stability of this profile during storage. In the Genovese genotype this effect was most evident in plants submitted to a water stress equivalent to 75% FC, and after five days of storage. For the Green Iranian landrace, changes in the volatile profile during storage were patent in plants watered at 100% FC. Finally, for the Purple Iranian genotype, the water stress barely induced changes in the volatile profile, but storage time dramatically damaged it from Days 5–7. As changes in the aromatic fraction of basil may lead to consumer rejection, even if the other parameters still meet the expected requirements, water stress should be avoided in all three landraces studied. In addition storage conditions and times should be selected in such a way that the volatile profile is maintained to the greatest extent possible.

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