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ORIGINAL PAPER

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DETERMINATION OF SOME AGRONOMICAL CHARACTERISTICS AND ESSENTIAL OIL VARIATION IN DIFFERENT LOCAL *Ocimum basilicum* L. ECOTYPES UNDER SEMI-ARID CLIMATIC CONDITIONS

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ABSTRACT

The study aimed to determine agronomic characteristics and essential oil components of different basil ecotypes in semi-arid climatic conditions of South Eastern Anatolia, Diyarbakir, Turkey. Two-year harvest data about fresh and dry herb yield, dry leaf yield, essential oil content and its components from the plants of year 2015 and 2016 was analyzed in this study. Essential oil components were detected by gas chromatography/mass spectrometry (GC/MS). The resulting outcomes demonstrated that both ecotypes and harvests had important effects on fresh and dry herb, dry leaf yield and essential oil contents of sweet basil. The highest dry leaf yield was noted from green leafy ecotypes and from second harvest stage. Twenty-three constituents were detected in the essential oil of *O. basilicum* ecotypes. The main components of basil essential oil were linalool, methyl chavicol, neral, geranial and methyl cinnamate that differed according to ecotypes and harvests during experimental years 2015 and 2016. Purple leafy basil ecotypes were determined as linalool rich, while greenish leaf ecotypes were abundant in methyl chavicol. Silbe – green ecotype contains higher neral and geranial levels than the other ecotypes. It was concluded that basil plant could be grown successfully and harvested two or more times to prefer for maximum dry leaf yield and essential oil contents under semi-arid climatic conditions.

Key words: agronomy, basil, harvest stages, dry leaf yield, volatile oil, linalool

INTRODUCTION

Genus *Ocimum* (*Lamiaceae*) constitutes 30 species spread all over tropical and subtropical areas. They have high economic significance, the most developed and used species include *O. americanum* L., *O.* × *citriodorum* Vis., *O. gratissimum* L., *O. basilicum* L., *O. tenuiflorum* L. and *O. minimo* L. [Labra et al. 2004, Da Costa et al. 2015]. Among these species, *O. basilicum* L. is the most utilized and its popularity is based on the types of essential oils and their constituents that give a particular fragrance to the condiments.

Albeit, it is generally utilized as therapeutic herb to treat kidney disorders, migraines, coughs, loose bowels and worms [Simon et al. 1990, Patil et al. 2011]. Moreover, it is also a source of biologically dynamic and active constituents of fragrance and essential oils. These could also be used as nematocidal, antibacterial, and insect repellent compounds [Sagdic and Ozcan 2003, Patil et al. 2011]. Use of antioxidants

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obtained from various natural materials is recommended to prevent harmful effects of synthetic chemicals [Ozcan and Akgul 1995]. Basil essential oils are broadly utilized as a part of flavors for bakery products and prepared merchandise like hot dogs, meat, mixed green servings, ice creams, beverages, and desserts. Furthermore, they are also used in pharmaceutical industry for preparations used in dental and oral therapy [Simon et al. 1990, Zheljazkov et al. 2008, El-Dakar et al. 2015].

Relying on factors like season and locality, the essential oil yield from various basil plant parts could shift between 0.15 and 1.59% [Ozcan and Chalchat 2002]. The chemical analysis of basil extracts reveals the availability of saponins, tannins, flavonoids and unstable terpenes [Lorenzi and Matos 2002]. Major chemotypes of basil essential oil are described by high centralization of linalool, methyl chavicol, methyl cinnamate or eugenol [Sarrou et al. 2016]. The most significant basil essential oil in the European markets principally constitute linalool (40.5 to 48.2%) and methyl – chavicol (28.9 to 31.6 %) [Fleisher 1981, Charles and Simon 1990].

The chemotype grouping in a view of only one noteworthy volatile oils is problematic as the species most often contain two mixes in almost equal amounts with variable and broad chemo-toxic ranges. European basil chemotype has (–) linalool and methyl chavicol as the major oil constituents. The Reunion basil chemotypes have methyl cinnamate as a major constituent, while tropical basil chemotypes have methyl cinnamate as the significant constituent. Another basil chemotype developed in North Africa, Russia, Eastern Europe, and various Asian regions has eugenol as the major constituent [Marotti et al. 1996].

It is accounted that basil species could be grown under a wide scope of climatic and natural (ecological) conditions, yet they could be grown best under warm environments [Chang et al. 2005]. Basil is broadly grown in irrigated and warm mild localities of Diyarbakir province, along the edges of Tigris River at the historic Hevsel Gardens.

The cost of dried basil herb (purple shade) in the market achieves values up to US$ 20.00/kg. This value makes the development and production of basil under irrigated semi-arid areas of Diyarbakir, that it makes an attractive and promising option for small farmers.

The ideal harvest stage for essential oil extraction is at the time of blooming, when the oil content and favored essential oil contents synthesis is the most elevated [Zheljazkov et al. 2008]. There are no previous reports about the agronomic properties and fundamental oil constituents of local basil ecotypes developed under south-eastern Anatolian Turkey. Along these lines, the aim of the present study was to evaluate essential oil contents and some agronomic properties of various local basil ecotypes reaped at two stages under semi-arid climatic conditions.

MATERIAL AND METHODS

Plant materials. *Ocimum basilicum* ecotypes seeds were provided from two different locations and named the same according to the place from where they were obtained (Dicle – Purple, Dicle – Green, Silbe – Purple, Silbe – Green). Voucher specimens of *Ocimum basilicum* were deposited vide No. DUZF 0048–51 at the Herbarium of the Medicinal and Aromatic Plants, Faculty of Agriculture, Dicle University, Diyarbakir.

The field experiments were conducted during 2015−2016 at the Department of Field Crops, Dicle University, Diyarbakir (latitude 37°53'N, longitude 40°16'E, altitude 680 m). The experimental area soils had pH of 7.45, with 1.16% organic matter.

Two-year field experiment was conducted with local basil varieties in Diyarbakir a semi-arid climatic region. Seeds were sown on seedbed tubes containing soil, sand and burnt farmyard manure (1 : 1 : 1) mixture, and placed in a greenhouse. The maintenance works, such as weed control and irrigation, were applied on a regular basis. The seedlings that reached 10–15 cm were planted on 18 May during the first year and 9 May during the second year with 70 × 20 cm row spacing. The three times replicated field experiment was established as Randomized Complete Block design. The area of one plot was 6.3 m^2 (2.1 × 3 m^2). The soil of each experimental plot was watered soon after planting to avoid wilting of the transplanted material. These were watered by sprinkler irrigation and hand weeded when required during experiment. The content of soil samples was salt with 0.16% content, pH of 7.46, available phosphorus (P_2O_5) with 1.45 kg ha^{-1}, potassium (K_2O) (8.2 kg ha^{-1}), available lime ($CaCO_3$) in 11.02% and

Table 1. Phenological and climatic data of different sweet basil ecotypes grown in semi-arid conditions in 2015 and 2016 years

Phenological data	Years	
	2015	2016
Sowing to seed bed	24 March	15 March
Transplanting to the field	18 May	9 May
Beginning of blooming	10 July	25 June
Cutting I	14 July	30 June
Cutting II	18 August	29 July
Days from planting to harvest (I)	57	52
Days to first to second harvest (I–II)	36	29
Climatic data		
Mean temperature (°C)		
March	7.6	9.3
April	12.1	15.2
May	18.9	19.3
June	25.6	26.1
July	30.9	31.0
August	30.1	31.2
Mean humidity (%) (March–August)	52.3	45.3
Total rainfall (mm) (March–August)	196.5	100.4

organic matter (0.48%). No fertilizers were added to any of the soils at the experiment area.

Aerial parts of basil were harvested at the beginning of flowering both during 2015 and 2016. Two harvests were realized in a year and first and second harvests were made on 14th July and 18th August in 2015 and 30th June and 29th June in 2016. The experiment was evaluated for plant height, number of branches, fresh herb, dry herb and dry leaf yield and essential oil contents as agronomical characteristics (Table 1). The plants were cut 10 cm above the ground. Fresh herbages were dried shady and under well ventilated room temperature (20−25°C).

Isolation of the essential oil. Dried finely ground leafy stems of the plants (20 g) were chopped and hydro-distilled for 3 h with Clevenger type apparatus to extract the oils that were dried over anhydrous sodium sulphate (Na_2SO_4) [Ozcan and Chalchat 2002] .

Gas chromatography–mass spectrometry (GC–MS) analysis. GC–MS analyses were done at the laboratory of Plant Physiology, the Department of Biology, Sutcu Imam University, Kahramanmaras, Turkey. GC/MS analyses were done with Agilent GC – 6890 II series coupled with Agilent 5975C Mass Spectrometer. Column: HP – 88, 100 m × 250 μm × 0.20 μm film thickness. The GC/MS temperature was adjusted from 70°C (1 min) to 230°C (20 min) with rate of change of 10°C/min. The injection temperature remained 250°C. Injection volume was 1.0 μL. Carrier gas was He. Injection mode was split (20 : 1). MS interface temperature was 250°C; MS mode remained EI; detector voltage: 70 eV; mass range of 35–400 m/z with scan speed (amu/s). The components of the oil were detected by mass spectra and compared with reference compounds of pure authentic samples, available in our laboratories, and with those stored in HPCH1607, Willey7n.1 and NIST08 libraries. Retention indices (RI) were computed from gas chromatograms by logarithmic interpolation between n–alkanes. The homologous series of n–alkanes C7 – C40, Supelco, USA were used as standard. Retention indices were calculated as HP – 88

capillary column. The analyses of all samples were replicated thrice for GC/MS analysis.

Statistical analysis. MSTAT – C" computer software package was used to statistically analyze the experimental results by subjecting the data to analysis of variance (ANOVA) for split plot design. The means of treatments were compared using the Least Significant Difference (LSD) test at 0.05 probability level.

RESULTS AND DISCUSSION

Field studies. Agronomic properties of *O. basilicum* plant and their chemical compositions are shown in Tables 2, 3 and 4. The results clearly specified significant differences among treatments for effects of ecotypes, significant interaction ($P < 0.05$) for year × ecotypes and year × harvests for fresh herb yields.

Plant height of basil ecotypes showed significant differences among each other. Green leafy ecotypes were clearly taller compared to purple ones. Plant heights of green leafy ecotypes were determined as 63.4 cm for Silbe and 58.8 cm for Dicle. The purple ecotypes were small compared to the green ecotypes. There were significant differences among harvest periods for plant height. The means of the second year were higher compared to the means of the first year.

Table 2. Variation of plant height and number of branches of different local sweet basil ecotypes under semi-arid ecological conditions

Ecotypes		Plant height (cm)			Number of branches (pieces)		
		2015	2016	Mean	2015	2016	Mean
Dicle – Purple		42.0 de	39.0 e	40.5 D	10.6 abc	8.1 bc	9.3
Dicle – Green		65.9 a	51.6 c	58.8 B	10.5 abc	7.7 c	9.1
Silbe – Purple		47.9 c	46.5 cd	47.2 C	11.7 a	7.8 bc	9.7
Silbe – Green		67.7 a	59.1 b	63.4 A	9.2 abc	10.8 ab	10.0
Mean		55.9 A	49.07 B		10.5 A	8.6 B	
Cuttings		2015	2016	Mean			
I		50.0 b	50.5 b	50.2 B	7.3 c	9.7 b	8.5 A
II		61.8 a	47.6 b	54.7 A	13.7 a	7.6 bc	10.6 B
Mean		55.91	49.07		10.5	8.6	
Ecotypes × Cuttings		2015	2016	Mean	2015	2016	Mean
Dicle – Purple	I	36.1	42.7	39.4	6.9	8.8	7.8
	II	47.9	35.4	41.6	14.3	7.4	10.9
Dicle – Green	I	60.3	50.3	55.3	7.4	10.0	8.7
	II	71.5	52.9	62.2	13.6	5.4	9.5
Silbe – Purple	I	43.4	47.7	45.5	8.3	8.8	8.6
	II	52.5	45.4	48.9	15.0	6.8	10.9
Silbe – Green	I	60.3	61.4	60.9	6.5	11.0	8.8
	II	75.2	56.8	66.0	11.9	10.6	11.2
LSD_{Years}			**			**	
$LSD_{Ecotypes}$			4.0**			ns	
$LSD_{Cuttings}$			**			**	
$LSD_{Y \times E}$			5.7**			3.0**	
$LSD_{Y \times C}$			4.0**			2.1**	
$LSD_{E \times C}$			ns			ns	
$LSD_{Y \times E \times C}$			ns			ns	

*: 0.05; **: 0.01

The maximum plant height was recorded on second harvest during the first year as 61.8 cm. Vegetation period after first harvest during first year took longer time compared to the same for the second year (Tab. 1); whereas the least plant height was recorded during the second year at the second harvest (Tab. 2).

Number of branches in the first experimental year was recorded as 10.5 and for the second year as 8.6. Effect of cuttings was significant ($p < 0.01$) on number of branches. The highest number of branches was recorded from the second harvest as 10.6 and 8.5 for the first harvest (Tab. 2).

Year × ecotypes interaction was found significant on the fresh herb yield. The highest values obtained from Dicle – Green and Silbe – Green were recorded as 2851.4 and 2560.0 g m^{-2}, the least yields were obtained from the Dicle – Purple as 698.5 g m^{-2} and from the Silbe – Purple as 884.4 g m^{-2}. The green leafy basils are taller than the purple ones with more leaves. Both ecotypes with green leaf generate fresher herb

Table 3. Variation of fresh and dry herbage yield of different local sweet basil ecotypes under semi-arid ecological conditions

Ecotypes		Fresh herbage yield (g m^{-2})			Dry herbage yield (g m^{-2})		
		2015	2016	Mean	2015	2016	Mean
Dicle – Purple		1697.1 b	698.5 c	1197.8 B	302.8 b	112.5 d	207.7 B
Dicle – Green		2851.4 a	940.8 c	1896.1 A	549.2 a	165.4 cd	357.3 A
Silbe – Purple		1585.6 b	884.4 c	1235.0 B	272.7 bc	146.6 d	209.6 B
Silbe –Green		2560.1 a	1179.7 bc	1869.9 A	472.0 a	211.8bcd	341.9 A
Mean		2173.5 A	925.9 B		399.2 A	159.1 B	
Cuttings							
I		1664.7 b	918.7 c	1291.6 B	304.8 b	155.1 c	230.0 B
II		2682.4 a	933.2 c	1807.8 A	493.6 a	163.0 c	328.3 A
Mean		2173.5	925.90				
Ecotypes × Cuttings		2015	2016	Mean	2015	2016	Mean
Dicle – Purple	I	1151.8	798.5	975.2	210.9	128.0	169.4
	II	2242.3	598.5	1420.4	394.8	97.0	245.9
Dicle – Green	I	2177.5	844.7	1511.1	429.9	149.4	289.7
	II	3525.2	1037.0	2281.1	668.5	181.3	424.9
Silbe – Purple	I	1353.3	803.2	1078.2	210.4	131.8	171.1
	II	1818.0	965.6	1391.8	335.0	161.4	248.2
Silbe – Green	I	1976.1	1228.0	1602.1	368.0	211.3	289.7
	II	3144.2	1131.4	2137.8	576.0	212.3	394.2
LSD_{Years}			**			**	
$LSD_{Ecotypes}$			455.4**			87.3**	
$LSD_{Cuttings}$			**			**	
$LSD_{Y \times E}$			644.0**			123.5**	
$LSD_{Y \times C}$			455.4**			87.3**	
$LSD_{E \times C}$			ns			ns	
$LSD_{Y \times E \times C}$			ns			ns	

*: 0.05; **: 0.01

than purple leafy ecotypes. The total fresh herb yield (2682.4 g plant^{-1}) of basil plants was the maximum at second harvest during 2015, while the minimum yield (918.7 kg m^{-2}) was obtained at the first harvest during 2016. The second harvests yielded higher compared to the first harvests during both years (Tab. 3).

Similarly, high fresh and dry herb yield values were obtained from the green leafy ecotypes. The highest yield was obtained from the Dicle – Green ecotype during first year, while the minimum yield was obtained from the Dicle – Purple during second year.

Ecotypes and harvests stage significantly affected the dry leaf yield that varied and changed from 2015 to 2016. Effect of ecotypes showed that the highest total dry leaf yield (227.3 g m^{-2}) was determined from the Dicle – Green ecotype, while the minimum dry leaf yield was obtained from the Dicle – Purple ecotype as 136 g m^{-2}. Dry leaf yield showed variations between harvest stages. The maximum leaf yield was obtained from the second harvest as 224.7 g m^{-2}.

Table 4. Variation of fresh and dry leaf and essential oil content of different local sweet basil ecotypes under semi-arid ecological conditions

Ecotypes		Dry leaf yield (g m^{-2})			Essential oil content (%)		
		2015	2016	Mean	2015	2016	Mean
Dicle – Purple		194.4 b	77.5 c	136.0 C	0.61	0.72	0.66 c
Dicle – Green		343.2 a	111.4 c	227.3 A	0.90	0.95	0.92 ab
Silbe – Purple		200.4 b	100.8 c	150.6 BC	0.66	0.72	0.69 bc
Silbe – Green		275.4 a	126.9 bc	201.2 AB	0.91	1.13	1.02 a
Mean		253.4 A	104.1 B		0.77	0.88	
Cuttings							
I		166.6	99.1	132.9 B	0.67	0.84	0.76 b
II		340.2	109.2	224.7 A	0.87	0.92	0.89 a
Mean		253.4	104.1		0.77	0.88	
Ecotypes × Cuttings		2015	2016	Mean	2015	2016	Mean
Dicle – Purple	I	129.0	86.1	107.5	0.61 bcd	0.54 cd	0.58
	II	259.9	68.9	164.4	0.61 bcd	0.90 a-d	0.75
Dicle – Green	I	219.0	98.0	158.5	0.73 bcd	1.00 abc	0.86
	II	467.5	124.9	296.2	1.08 ab	0.90 a-d	0.99
Silbe – Purple	I	137.6	93.3	115.5	0.51 d	0.81 a-d	0.66
	II	263.3	108.2	185.7	0.81 a-d	0.63 bcd	0.72
Silbe – Green	I	180.9	119.1	150.0	0.85 a-d	1.01 ab	0.93
	II	370.0	134.7	252.4	0.98 a-d	1.25 a	1.11
LSD_{Years}			**			ns	
$LSD_{Ecotypes}$			52.8**			0.23**	
$LSD_{Cuttings}$			**			*	
$LSD_{Y \times E}$			74.7**			ns	
$LSD_{Y \times C}$			74.7**			ns	
$LSD_{E \times C}$			ns			ns	
$LSD_{Y \times E \times C}$			ns			0.46*	

*: 0.05; **: 0.01

Table 5. Major essential oil components of different sweet basil ecotypes harvested two cutting periods in 2015 year

No	Components	RI	Ecotypes							
			Dicle – Purple		Dicle – Green		Silbe – Purple		Silbe – Green	
			1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting
1	α -Terpinene	1316	0.13 ±0.02	0.14 ±0.03	0.24 ±0.11	0.59 ±0.33	0.10 ±0.00	0.16 ±0.06	tr.	0.07 ±0.01
2	1,8-Cineol	1365	0.52 ±0.00	0.70 ±0.08	1.41 ±0.57	1.89 ±0.57	1.25 ±0.31	2.02 ±0.45	0.09 ±0.01	0.12 ±0.01
4	Linalool	1628	52.83 ±6.18	37.15 ± 6.96	16.12 ±2.47	14.18 ±0.83	31.40 ±3.39	31.51 ±5.85	1.11 ±0.02	1.76 ±0.47
3	*Cis*-α-Bergamotene	1648	0.40 ±0.11	0.45 ±0.03	2.91 ±0.42	2.72 ±0.94	1.15 ±0.03	0.77 ±0.08	1.63 ±0.29	1.33 ±0.13
5	β-Elemene	1700	tr.	8.97 ±1.37	3.42 ±0.08	tr.	9.48 ±0.21	6.88 ±1.53	tr.	tr.
6	E-Caryophyllene	1770	0.41 ±0.02	0.65 ±0.27	0.83 ±0.06	0.45 ±0.07	2.48 ±0.74	1.77 ±0.31	2.46 ±1.00	2.29 ±0.16
7	α-Bulnesene	1815	1.69 ±0.58	2.21 ±0.61	1.14 ±0.02	0.95 ±0.04	2.55 ±0.50	1.63 ±0.25	1.17 ±0.02	0.94 ±0.16
8	α-Bisabolene	1837	0.19 ±0.01	0.25 ±0.09	0.33 ±0.06	0.33 ±0.11	1.61 ±0.27	1.45 ±0.31	3.96 ±1.42	3.41 ±0.08
9	γ-Cadinene	1870	2.78 ±0.35	3.14 ±0.69	2.57 ±0.11	2.42 ±0.73	4.30 ±0.66	2.70 ±0.84	0.36 ±0.00	tr.
10	β-Cubebene	1896	2.85 ±0.03	3.40 ±1.14	1.73 ±0.28	1.25 ±0.20	3.91 ±0.00	3.33 ±0.52	tr.	tr.
11	Germacrene D	1898	2.82 ±0.01	3.18 ±1.39	0.86 ±0.50	1.23 ±0.28	5.79 ±1.15	3.35 ±0.52	1.55 ±0.01	0.62 ±0.01
12	Methyl chavicol	1927	22.14 ±1.52	24.23 ±7.71	47.15 ±2.53	55.10 ±10.64	2.46 ±0.50	2.97 ±0.38	18.26 ±13.07	30.82 ±1.91
13	*Cis* geraniol (nerol)	2017	0.11 ±0.00	0.13 ±0.00	0.13 ±0.04	tr.	0.72 ±0.00	0.14 ±0.01	2.31 ±0.23	3.43 ±0.47
14	Geraniol	2074	tr.	0.29 ±0.09	tr.	tr.	1.78 ±1.08	1.15 ±0.02	1.59 ±0.85	1.33 ±0.01
15	Neral	2096	0.30 ±0.03	0.32 ±0.07	0.41 ±0.41	0.08 ±0.03	0.48 ±0.27	0.31 ±0.09	20.07 ±0.67	14.69 ±1.97
16	Geranial	2159	0.32 ±0.05	0.37 ±0.11	1.61 ±0.00	0.06 ±0.01	0.44 ±0.41	0.21 ±0.07	28.21 ±0.79	19.62 ±2.00
17	Epicubenol	2402	1.20 ±0.12	1.17 ±0.27	1.87 ±0.47	1.39 ±0.71	1.36 ±0.05	1.17 ±0.28	0.05 ±0.00	0.51 ±0.00
18	Methyl eugenol	2454	tr.	tr.	1.12 ±0.01	0.52 ±0.20	1.68 ±0.42	3.07 ±0.86	12.42 ±1.56	12.87 ±0.08
19	Epi-α-cadinol	2530	7.51 ±0.69	7.67 ±2.03	8.44 ±2.71	6.22 ±2.04	7.48 ±0.37	5.04 ±2.10	tr.	1.06 ±0.01
20	Spathulenol	2562	1.30 ±0.07	1.42 ±0.24	4.36 ±2.20	2.64 ±1.82	2.30 ±0.23	1.70 ±0.46	tr.	2.05 ±0.52
21	Caryophyllene oxide	2564	tr.	tr.	tr.	tr.	1.43 ±0.05	1.69 ±0.58	2.77 ±0.62	2.14 ±0.45
22	Methyl cinnamate	2630	0.23 ±0.00	0.29 ±0.05	2.17 ±2.20	1.92 ±0.00	12.96 ±7.67	20.66 ±9.60	0.25 ±0.00	0.19 ±0.64
23	Eugenol	2687	tr.	1.35 ±0.01	0.38 ±0.03	0.44 ±0.22	1.14 ±0.12	3.41 ±0.43	0.21 ±0.00	0.18 ±0.06
	Total		97.73	97.48	99.20	94.38	98.25	97.09	98.47	99.43
Grouped Components										
Monoterpene hydrocarbons			22.25	38.97	47.28	55.10	4.96	4.26	22.16	36.45
Oxygenated monoterpenes			54.33	24.65	21.96	18.72	46.63	54.87	49.73	35.58
Sesquiterpene hydrocarbons			8.32	19.07	12.93	8.12	25.48	18.53	9.58	7.97
Oxygenated sesquiterpenes			12.83	13.44	15.53	11.48	18.36	12.95	4.37	6.38
Phenylpropanoids			tr.	1.35	1.5	0.96	2.82	6.48	12.63	13.05

RI: retention index; tr.: trace

The essential oil contents obtained from aerial parts of basil ecotypes are shown in Table 4. Presented data indicate that the ecotypes, as well as the year of cultivation, had significant effect on the content of essential oil. Essential oil contents showed variations among ecotypes. The minimum oil content obtained from this study (Dicle – Purple as 0.58%) were found higher compared to the limit described by Turkish Standards Institute (0.3%) [TGK 2013]. Nurzynska-Wierdak et al. [2013] reported that essential oil content varied between 0.46 and 1.03% in two basil cultivars at different harvest stages. Moreover, they also reported that at the flower bud stage herb produce 40% less oil (0.65%) than at full bloom (1.03%) stage depending upon genetic factors. Basil ecotypes were harvested at full blooming stage in this study and the obtained results are in agreement with the results of Nurzynska-Wierdak et al. [2013]. Basil should be harvested at this optimal stage to obtain optimal dry leaf and high essential oil contents.

Light and temperature effect essential oil accumulations positively in many plant species. Moreover, it is reported that there is a close co-ordination between ontogeny in aromatic plants related to temperature and daylight [Sangwan et al. 2001].

First harvest was performed in June and second harvest in September during both experimental years. Essential oil content of the purple leafy ecotypes was found lower compared to the green leafy ecotypes. Moreover, second harvest gave high essential oil contents. Essential oil content of ecotypes changed between 0.66 and 1.02%. The Silbe – Green colored ecotypes showed the highest essential oil contents (1.02%). The second year harvest showed significantly improved performance in terms of oil contents compared to the oil contents during first year and affirmed the effects of significantly changing climatic conditions during two years. Day degree in semi-arid climatic conditions (Diyarbakir) reach up to 40°C. In basil growing, from the beginning of sowing (April) to the end of summer, temperature changed between 25 to 30°C and this temperature value was good for accumulating of essential oil. Yaldiz et al. [2015] reported that the highest essential oil contents were obtained from second harvest and the greenish basil type yielded more oil yield compared to the purple one under the Central Anatolia conditions. These results are in agreement with the results reported in this study.

Essential oil components. Chemical composition of the essential oils of four ecotypes of *Ocimum basilicum*, used in this study are given in Tables 5 and 6 in the order of the retention index of the constituents.

Essential oils are complex natural organic compound mixtures that are characterized by fairly high concentration (20−70%) of 2 or 3 major components as compared to other but available trace amount components. They are predominantly made of terpenic hydrocarbons and terpenoids (oxygen containing hydrocarbons) [Pandey et al. 2014]. In this study, among all the constituents, the oxygenated monoterpenes and monoterpene hydrocarbons were found in higher amount (Table 5 and 6). Oxygenated monoterpenes content of green leafy ecotypes were lower than purple ones. The highest oxygenated monoterpenes were recorded from the Silbe – Purple ecotype as 66.5% during the first experimental year. The green leafy ecotypes were rich with respect to monoterpene hydrocarbons. Dicle – Green ecotype has high monoterpene hydrocarbons and the rate of it increased at second harvest stage during both years.

Twenty three components were identified during 2015 and 2016 (Tables 5 and 6). Essential oil components percentage varied according to harvest period and ecotypes. For the purple leafy ecotypes, linalool, methyl chavicol, epi-α-cadinol, β-elemene, α-bulnesene, β-cubebene, γ-cadinesene, methyl cinnamate, and for green leafy ecotypes, methyl chavicol, linalool, neral, geranial, methyl eugenol, epi-α-cadinol, α-bisabolene were the main components in the same order, during both years (Table 5 and 6).

Linalool, methyl chavicol, neral, geranial, methyl eugenol, epi-α-cadinol and methyl cinnamate together constituted 58.62−85.25% during 2015 and 66.28−85.21% during 2016, respectively (Tables 5 and 6). Linalool is a monoterpene found in most aromatic plant essential oils. It is the major constituent of *O. basilicum* oil [Silva et al. 2015]. Linalool content was found as major component in purple basil ecotypes, whereas green ecotypes linalool content was found quite low. Among ecotypes, the higher linalool content (37.15−52.83%) was obtained from Dicle – Purple for 2015, and compared to the oil content (28.42−32.90%) noted during 2016. First year linalo-

Table 6. Major essential oil components of different sweet basil ecotypes harvested two cutting periods in 2016 year

No	Components	RI	Ecotypes							
			Dicle – Purple		Dicle – Green		Silbe – Purple		Silbe – Green	
			1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting
1	α-Terpinene	1316	0.07 ±0.05	2.00 ±0.01	2.20 ± 0.21	1.69 ±0.82	0.15 ±0.02	0.12 ±0.07	0.20 ±0.00	0.04 ±0.00
2	1.8-Cineol	1365	3.27 ±1.40	2.57 ±0.13	2.88 ±0.41	1.74 ±0.35	5.70 ± 0.84	5.71 ±3.20	0.09 ±0.00	0.07 ±0.04
4	Linalool	1628	42.13 ±0.56	36.86 ±5.13	8.97 ±3.87	5.48 ±3.89	28.42 ±1.47	32.90 ±7.21	0.61 ±0.06	3.41 ±0.88
3	*Cis*-α-Bergamotene	1648	0.54 ±0.35	0.32 ±0.15	1.18 ±0.48	1.96 ±0.01	1.00 ±0.02	0.56 ±0.02	1.10 ±0.23	1.12 ±0.18
5	B-Elemene	1700	tr.	tr.	1.91 ±0.91	0.94 ±0.00	8.12 ±1.38	7.50 ±0.10	0.56 ±0.00	0.18 ±0.00
6	E-Caryophyllene	1770	0.62 ±0.12	0.71 ±0.13	0.31 ±0.89	0.36 ±0.02	1.80 ±0.28	1.46 ±0.30	2.15 ±0.31	1.90 ±0.27
7	α-Bulnesene	1815	1.47 ±0.60	1.50 ±0.01	1.11 ±1.25	0.21 ±0.11	2.04 ±0.20	1.53 ±0.39	tr.	1.16 ±0.20
8	α-Bisabolene	1837	0.18 ±0.00	0.88 ±0.01	0.24 ±0.08	0.49 ±0.00	1.17 ±0.20	1.22 ±0.28	2.62 ±0.12	2.53 ±0.51
9	γ-Cadinene	1870	1.91 ± 0.78	1.63 ±0.01	1.51 ±0.56	1.58 ±0.02	2.66 ±0.25	2.21 ±0.47	0.31 ±0.00	tr.
10	β-Cubebene	1896	2.31 ±1.12	2.09 ±0.05	1.06 ±0.01	0.41 ±0.01	tr.	tr.	tr.	tr.
11	Germacrene D	1898	2.58 ±1.06	2.10 ±0.05	0.82 ±0.30	0.65 ±0.02	4.01 ±0.54	3.15 ±0.84	1.13 ±0.45	1.00 ±0.01
12	Methyl chavicol	1927	31.18 ±3.07	36.15 ±7.43	70.38 ±4.63	75.20 ±2.22	12.65 ±5.02	10.63 ±5.66	54.71 ±1.74	45.04 ±8.87
13	*Cis* geraniol	2017	0.68 ±0.82	0.09 ±0.01	tr.	tr.	tr.	tr.	1.51 ±0.36	2.47 ±1.12
14	Geraniol	2074	0.67 ±0.26	0.39 ±0.12	tr.	0.57 ±0.00	0.41 ±0.00	0.61 ±0.00	2.20 ±0.73	1.82 ±1.30
15	Neral	2096	0.27 ±0.00	0.16 ±0.06	tr.	0.12 ±0.04	tr.	0.13 ±0.05	11.33 ±0.35	14.26 ±0.81
16	Geranial	2159	0.15 ±0.12	0.29 ±0.014	0.05 ±0.01	0.10 ±0.03	tr.	0.11 ±0.04	14.72 ±1.25	18.37 ±0.82
17	Epicubenol	2402	0.97 ±0.10	0.79 ±0.08	0.43 ±0.01	0.64 ±0.00	1.39 ±0.86	0.76 ±0.20	0.26 ±0.00	0.07 ±0.04
18	Methyl eugenol	2454	0.39 ±0.11	0.51 ±0.14	0.47 ±0.01	0.49 ±0.06	2.32 ±0.26	3.02 ±1.17	0.55 ±0.00	0.76 ±0.10
19	Epi-α-cadinol	2530	5.26 ±2.69	5.19 ±0.65	2.98 ±1.15	3.73 ±0.16	4.87 ±0.44	4.90 ±0.87	0.99 ±0.00	0.99 ±0.00
20	Spathulenol	2562	tr.	tr.	1.15 ±0.02	1.14 ±0.03	tr.	tr.	tr.	2.36 ±0.03
21	Caryophyllene oxide	2564	tr.	tr.	tr.	tr.	tr.	tr.	0.90 ±0.00	1.06 ±1.13
22	Methyl cinnamate	2630	2.67 ±0.58	3.93 ±0.007	0.27 ±0.00	0.09 ±0.00	17.86 ±1.74	18.87 ±4.23	tr.	0.30 ±0.00
23	Eugenol	2687	0.37 ±0.16	0.45 ±0.20	tr.	0.07 ±0.00	1.32 ±0.05	2.09 ±0.50	0.06 ±0.00	0.07 ±0.02
	Total		97.69	98.61	97.92	97.66	96.05	97.48	96.00	98.98
Grouped Components										
Monoterpene hydrocarbons			32.53	45.81	70.38	75.77	13.06	11.24	58.42	49.33
Oxygenated monoterpenes			48.56	36.63	14.37	9.22	52.29	57.84	26.95	36.45
Sesquiterpene hydrocarbons			7.03	7.13	7.32	5.95	16.79	14.48	6.74	6.89
Oxygenated sesquiterpenes			8.81	8.08	5.38	6.16	10.27	8.81	3.28	5.48
Phenylpropanoids			0.76	0.96	0.47	0.56	3.64	5.11	0.61	0.83

RI: retention index; tr.: trace

ol content decreased towards the end of vegetation. The highest values were obtained at the first harvest stage. Linalool content of the Dicle – Purple during 2015 was higher compared to that of year 2016.

Methyl chavicol content of the ecotypes during 2015 ranged 2.46−55.10% at different harvest or development stages. The highest value (55.10%) was noted from the Dicle – Green ecotype, while the lowest value (2.46%) noted from Silbe – Purple ecotypes. Second year, methyl chavicol value also ranged 10.63−75.20%. The highest value was obtained from the Dicle − Green ecotype at second harvest. The Green ecotypes, Dicle – Green and Silbe – Green, are methyl chaviol chemotypes, especially the Dicle – Green ecotype produced high methyl chavicol during both experimental years. Methyl chavicol and linalool contents of the Dicle – Purple ecotype were very close to each other by contrast with Vieira and Simon [2006], who reported that concentration of methyl chavicol, also called estragole, is variable depending one ecotypes and shows a negative correlation with the concentration of linalool.

Although the basil oil has a high content of citral (geranial and neral), it was not detected in significant amounts in the oil of different ecotypes, except the Silbe – Green. Among ecotypes, the highest neral and geranial were obtained from the Silbe – Green. First year, neral contents changed between 0.08% to 20.07% and second year changed between 0.09−11.33%, in the same order. Geranial content, during 2015, changed between 0.06−28.21%, and it had variation of 0.05−18.37% during the second year. Neral content at the first harvest were more compared to those obtained after the second harvest during both years.

Methyl eugenol was low in the essential oil of basil ecotypes, except the Silbe – Green. Its values changed with respect to years and the highest value was obtained from the Silbe – Green ecotype. In the first experimental year, it was found at the percentage of 12.42− 12.87%; on the other hand during second year quite low amount (0.55−0.76%) of Methyl eugenol was recovered.

Epi-α-cadinol content during 2015 changed between 1.06−10.01% and 0.99−5.26% during 2016. The highest value was obtained from the Dicle – Green ecotype as 10.1% during the first year.

Methyl cinnamate content of different basil ecotypes changed between 0.19–20.66% during 2015 and 0.09−18.87% in second year. The highest values were obtained from the Silbe – Purple ecotypes during both years as 12.96−26.64% and 17.86−18.87%, in the same order.

Significant differences were observed when the main components were evaluated statistically for 2015 and 2016 (Tables 7 and 8). The highest linalool content (52.83%) was obtained from Dicle – Purple ecotype at first cutting, the lowest value (1.11%) was obtained from the Silbe – Green ecotype at the first cutting for 2015. Linolool content was high in Dicle − Purple at first cutting and was low in the Silbe − Green at first cutting. When the ecotypes and forms are evaluated for methyl chavicol, Dicle – Green at second cutting gave the highest rate (55.10%), the Silbe – Purple at first cutting gave the lowest rate (2.46%) for 2015. During 2016, the highest value was found in second cutting of the Dicle – Green; the lowest value was (10.63%) found in second cutting of the Silbe – Purple. The highest epi-α-cadinol content for 2015 was 8.44% in first cutting of the Dicle – Green ecotype, was 5.26% in first cutting of the Dicle – Purple for 2016. The lowest epi-α-cadinol content for 2015 was trace in first cutting of Silbe – Green ecotype, was 0.99% in both first and second cutting of the Silbe – Green for 2016. Methyl cinnamate content of Silbe – Purple had the highest content at second cutting both 2015 and 2016, and also, the Dicle – Purple and the Silbe – Green ecotypes had the lowest values in both cuttings for 2015. The Silbe – Green and the Dicle – Green ecotypes had the lowest content for 2016.

The essential oil constituents vary among sweet basil cultivars, and the main oil constituents remain linalool, methyl chavicol, eugenol, 1,8-cineole, geranial, neral, methyl cinnamate [Nurzynska-Wierdak 2007]. In this study, linalool, methyl chavicol and methyl cinnamate were found as major compounds. Linalool was found dominant component of green leafy ecotypes, except for the Silbe – Green. The Purple ecotypes included linalool at high percentage. Similar results were reported by Nacar and Tansi [2000] as the existence of three main chemotypes of basil. The first group was the green leafy Greek variety and was defined as linalool chemotype. A second group included purple leafed Turkish cultivar as linalool and methyl cinnamate chemotypes and the third group with green

Table 7. Major essential oil components of different sweet basil ecotypes harvested two cutting periods in 2015 year

Components	Ecotypes							
	Dicle – Purple		Dicle – Green		Silbe – Purple		Silbe – Green	
	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting
Linalool	52.83 A*	37.15 B	16.12 D	14.18 D	31.40 C	31.51 C	1.11 E	1.76 E
Methyl chavicol	22.14 E	24.23 D	47.15 B	55.10 A	2.46 H	2.97 G	18.26 F	30.82 C
Epi-α-cadinol	7.51 B	7.67 B	8.44 A	6.22 C	7.48 B	5.04 D	0.00 F	1.06 E
Methyl cinnamate	0.23 D	0.29 D	2.17 C	1.92 C	12.96 B	20.66 A	0.25 D	0.19 D
LSD (0.05) (Ecotypes × Cuttings)	Linalool: 2.30		Methyl chavicol: 0.27		Epi-α-cadinol: 0.22		Methyl cinnamate: 0.37	

*Values fallowed by the same letter in a line do not differ significantly according to LSD

Table 8. Major essential oil components of different sweet basil ecotypes harvested two cutting periods in 2016 year

Components	Ecotypes							
	Dicle – Purple		Dicle – Green		Silbe – Purple		Silbe – Green	
	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting	1st cutting	2nd cutting
Linalool	42.13 A*	36.86 B	8.97 E	5.48 F	28.42 D	32.90 C	0.61 H	3.41 G
Methyl chavicol	31.18 F	36.15 E	70.38 B	75.20 A	12.65 G	10.63 H	54.71 C	45.04 D
Epi-α-cadinol	5.26 A	5.19 B	2.98 E	3.73 D	4.87 C	4.90 C	0.99 F	0.99 F
Methyl cinnamate	2.67 D	3.93 C	0.27 E	0.09 E	17.86 B	18.87 A	0.00 E	0.30 E
LSD (0.05) (Ecotypes × Cuttings)	Linalool: 0.97		Methyl chavicol: 0.26		Epi-α-cadinol: 0.05		Methyl cinnamate: 0.33	

*Values fallowed by the same letter in a line do not differ significantly according to LSD

leafy French varieties, was the linalool and methyl eugenol chemotype. Nurzynska-Wierdak et al. [2012] reported that basil genotypes could be distinguished; they differ in the amount and quality of the dominant components of the essential oil.

The leaves of the Dicle – Green ecotype are green initially, then transformed into purple color towards the end of development. For this reason, they are considered genetically close. The linalool percentage in the Dicle – Green ecotypes was recorded as 14.18−16.12% and was quite less compared to the linalool percentage in the Dicle – Purple ecotypes. Similarity in terms of leaf color this could be due to high cross pollination among *Ocimum* ecotypes [Smitha and Tripathy 2016].

It is commonly expressed that chemical composition of essential oils chemical composition is related to factors like origin, harvest time, climate and geography, with technological influence, parts used and stages of flowering, as well as variations due to the presence of chemotypes [Jankovsky and Landa 2002, Fraternale et al. 2004]. Previous studies show a large number of results on the subject with substantial variations in the constituents of chemotypes. There are many chemotypes of basil such as methyl cinnamate,

methyl chavicol, linalool, geranial, neral, eugenol etc. Wesolowska et al. [2012] reported that the main oil components of Europe originated basil varieties were linalool, methyl cinnamate and methyl chavicol. Their content changed between 24.60−47.75% for linalool, 5.57−20.21% for methyl chavicol and 18.73−21.90% for methyl cinnamate, respectively. The composition of oils reported in this study is in agreement with the literature mentioned above. Furthermore, the essential oil composition of basil ecotypes used in this study showed analogue to the composition of the commercial basil analyzed reported by Zheljazkov et al. [2008].

CONCLUSIONS

Basil is cultivated at the embankment of Tigris River in Diyarbakir province, Turkey since hundreds of years. This area is characterized by well irrigated and high moisture micro-climate, where the local farmers harvest basil more frequently than once a year.

The main compounds of four basil ecotypes oils were linalool, methyl chavicol and methyl cinnamate. Purple colored ecotypes were abundant in linalool and green ones had improved percentage of methyl chavicol. Results of the study showed that basil has high potential for cultivation in semi-arid climatic conditions successfully and could be harvested economically two times a year. Traditionally, purple leafy basil ecotypes are preferred over green leafy ones in the region. Greenish leafy ecotypes have more essential oil compared to purple ones. Therefore, these should be preferred for essential oil production under south-eastern Anatolian (Turkey), semi-arid climatic conditions.

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