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## Agronomic Performance and Essential Oil Yield of Greek Oregano Under Different Irrigation Management

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### Abstract

The effects of two treatments of irrigation and non-irrigation on dry weight yield, oil yield and composition of Greek oregano (*Origanum vulgare* ssp. *hirtum* (Link) Letswaart) during the second and third seasons from its field establishment (first year) for two harvest each year on the first full blooming on June and the second on September were examined. Average dry weight increased significantly from about 120 g in the first to about 284 g in the second season for irrigated cultivation and from 44 to 155.5 to not -irrigated, as regards as oil concentration increased significantly from about 5.5% to 6.3% for irrigation treatment and there was no significant difference for not irrigated treatment 6.6 to 5.8 (v/w) for the first and the second year correspondingly and it was higher in the inflorescences (upper part) when compared with leaves (bottom part) and for the first harvest on June. The chemical composition of oregano Essential Oil (EO) was determined by Gas Chromatography-Mass Spectrometry analysis (GC-MS). Results showed that *Origanum vulgare* EO was rich in monoterpene and the major constituent was carvacrol ranging from 73.3% to 83% among treatments and period of harvests. It was followed by paracymenthene (3.6-10, 2) and  $\gamma$ -terpinene (3.1-10.1) thymol was detected at low levels (0.20-1.44%). Overall, oregano is adaptable to the climatic conditions of the area both in terms of dryness and yield of essential oil and it could be proposed to be cultivated in the area perennially mainly irrigated.

**Keywords:** Greek oregano; *Origanum vulgare* ssp. *Hirtum*; Irrigation; Essential oil concentration; Essential oil composition; Dry weight

### Introduction

The use of aromatic and medicinal plants for aromatic, medicinal, and cosmetic goals has a long tradition in Europe [1,2]. Apart from playing a central role in the natural biodiversity of many countries, these plants contribute largely to national and local economies, with a constantly increasing role in human nutrition over the last decades [3]. The aromatic and medicinal plants and their products not only serve as valuable source of income for small scale farmers, but in many cases also provide valuable foreign exchange by way of exports [1,4,5]. Greece, among several other countries of the Mediterranean Basin, produces hundreds of aromatic plants species [6,7]. Enriching knowledge on the role of cultivation techniques for increasing yield and quality of production may increase opportunities for expanding the areas of cultivation of these plants, seeking novel crop opportunities of an increase economic profit. In this context, improving technical and economic results is a necessary objective in a continuous process of production technologies optimization for use of these plants as alternative crops.

The genus *Origanum* belongs to the family of Lamiaceae and includes many species that are commonly found as wild plants in the Mediterranean areas [8]. Greek oregano (*Origanum vulgare* subsp. *hirtum*), a hardy perennial plant, is a subspecies of common oregano that thrives naturally in almost every region of Greece, on sunny and dry non-cultivated areas. Because of the special composition of the essential oil, the leaves of *Origanum* plants are widely used as a popular spice in a variety of foods, drawing attention of consumers due to the antimicrobial, antifungal, and

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antioxidative effects on human health [9-12]. The quality of oregano is determined mainly by the essential oil content and the composition of the essential oil. Both parameters may vary considerably depending on genotypes, climate conditions, and nutrient supply during the cultivation [13,14]. In addition, the components of oregano EO seem to be determined to a greater extent by genotype, while environment conditions account only for smaller variation in the components of the essential oil [14]. Cultivation of oregano can be a profitable business for growers as they can get up to 2500 Euros per hectare per year, which is a value considerably higher than for most field and horticultural crops [15]. This fact, together with the limited product availability and the low cost of cultivation, can be important in cultivating oregano in areas dominated by winter wheat [16]. Studies on oregano plants in Greece showed that the subspecies *hirtum* (*O. vulgare* ssp. *hirtum* (Link) Letswaart, syn.: *Origanum heracleoticum*) contained a high amount of essential oil. The content of essential oil as high as 8% with carvacrol as dominant component (95%) was reported for this subspecies [17]. Its EO contains more than 60 ingredients, most of which possess important antioxidant and antimicrobial properties [18-22].

In addition, the water supply is one of the most determinative cultivation conditions which significantly affect the yield and essential oil content of various spices and herb crops [23-26]. In most cases oregano plants must be irrigated during the cultivation period to obtain a good yield. For example during cultivation of *Origanum dictamnus* in Crete (Greece), irrigation was necessary for two harvests in 1 year [27,28]. Practically, the time at which the plants are irrigated is important for the efficiency of irrigation. For example, appropriate irrigation strategies showed a great potential for improvement of the yield of monoterpenes in field-grown spearmint and rosemary [26].

Experimental data on field performance of Greek oregano are limited. Thus, the objective of this study was to determine biomass and essential oil production of Greek oregano under field conditions and determine whether this aromatic plant has potential to be a viable niche opportunity for alternative cultivation not only for the Greek producers but also for farmers from regions that have a similar climate with the Greek Mediterranean climate with a dry-warm summer. For example all the areas around the Mediterranean Sea or even in areas with prolonged drought in summer months.

## Materials and Methods

### Crop management and experimental procedures

Oregano seedlings were purchased from a certified nursery in northern Greece. The transplantation production system is common for the commercial production of aromatic plants and was selected to ensure that plants of the same genotype for each species are studied. The experiments were conducted at the experimental field of Democritus University of Thrace experimental farm in Orestiada, northern Greece (41°30'N, 26°32'E, 22 m elevation) on a fertile silty clay loam soil (8% sand, 52.4% silt, and 39.6% clay) with pH (1:1 H<sub>2</sub>O) 6.7 and organic matter content 1.01%. The field was in fallow the previous year. Seedbed preparation consisted of standard conventional soil tillage in fall 2011 followed by chisel ploughing and disc harrowing before transplanting. Seedlings were transplanted in rows on 10 May, 2012. Row spacing was 80 cm apart and within row spacing (plant-to-plant distance) was 60 cm, resulting in a population density of 20,800 plants per ha. Seedlings were transplanted in eight rows of 20 m. Four rows were left rain fed, except for the time of establishment, whereas the other four rows were irrigated every

15 days from mid-May to mid-September of each year with a drip irrigation system for four hours. No type of fertilization was applied at any growth stage of the plants. The area was maintained free of weeds with manual weeding within and between rows. The basic weather data for each growing season were collected from a meteorological station near the experimental site (Table 1). Multiple harvests of plant material were conducted to evaluate biomass and essential oil production. The first harvest took place on 15 September, 2012. Plants were cut at the ground level, separated in upper and lower vegetative parts (samples cut in half), and weighed. Then, the plant parts were oven-dried at 70°C until constant weight and the dry weight was determined. Additionally, plant height, canopy diameter, and number of stems were recorded. Plant parts which were intended for distillation for essential oil were dried hanging in dark room for fifteen days in 30°C. Harvests were repeated from June to July 2013 and 2014 during full blooming of plants and in September 2013 and 2014, after the dry period of summer.

### Isolation of essential oils

Dried plant samples were used to determine the essential oil concentration of the various plant parts by hydro-distillation, using a Clevenger-type apparatus. Ground sample of 150 g was placed in the apparatus with 1500 ml of deionized water. The mixture was heated at 100°C for 3 h. Dehydration was achieved by using anhydrous magnesium sulfate (Sigma- Aldrich Co.). After filtration (Glass microfiber GF/C, Whatman), the volume of the essential oils produced was measured and expressed as mL/100 g dry weight of plant material. All essential oils were stored in closed dark vials at 4°C until further analysis.

### Analysis of essential oils

Essential oils were analyzed using a Hewlett Packard II 5890 Gas Chromatography (GC) system, equipped with a FID detector and HP-5ms capillary column (30 mx0.25 mm, film thickness 0.25 µm). Injector and detector temperatures were set at 220°C and 290°C, respectively. GC oven temperature was programmed from 60°C to 240°C at a rate of 3°C/min and held isothermally for 10 min. Helium was the carrier gas at a flow rate of 1 ml/min. Diluted samples (1/100 in diethyl ether, mg/l-1) of 1.0 µl were injected manually and in the splitless mode. Quantitative data were obtained electronically from FID area percent data without the use of correction factors. Qualitative analysis of the essential oils was performed using the same conditions with GC, in a Hewlett Packard II 5890 gas chromatograph equipped with Hewlett Packard II 5972 mass selective detector in the electron impact mode (70 eV). Identification of the major essential oil components was based on the comparison of their retention indices with those of authentic compounds by coelution and MS analysis. For the other components, tentative identification involved matching retention indices and recorded mass spectra with those obtained from NIST 98 and Wiley 275 libraries as well as from Adams CD computer library (Adams, 2007) [29]. For the identification of the main constituents of obtained essential oils the GC-MS analysis was used. The essential oils were diluted in hexane (Sigma-Aldrich Co). The chemical analysis of the essential oil was performed using Agilent Technologies CG-6890A and MS5975 with quadruple detector operating at 70 eV in electron ionization mode, equipped with HP-5 fused silica capillary column (300x0.25 mm i.d.; 0.25 µm film thickness). The carrier gas helium (He) was used at a constant flow rate of 1.0 mL min<sup>-1</sup>. The thermal program was 60-240°C at a rate of 3°C per min. The injected volume was 1,0 µL in split ratio

**Table 1:** Meteorological data for each growing season from a meteorological station near the experimental site.

Year		Month												TOTAL
		1	2	3	4	5	6	7	8	9	10	11	12	
2012	Monthly rainfall (mm)	41.7	36.8	7	61.0	74.0	4.0	0.8	0.4	2.0	40.0	20.0	160.0	447.7
	Min temperature	-12.6	-11.5	-5.1	-0.5	8.9	1.5	13.8	11.2	7.7	4	-1.9	-6.9	
	Max temperature	11.9	20	23.2	30.1	31.6	37.4	38.8	40.7	33.8	33.6	23.5	18.1	
2013	Monthly rainfall (mm)	132.6	70.8	52.4	47.0	24.8	57.4	13.6	5.0	17.4	41.6	72.6	7.4	542.6
	Min temperature	-7.7	-2.4	-2.9	2.4	6.2	10.2	13.1	14.7	7.2	-1.4	-2.8	-5.7	
	Max temperature	18.2	18.2	23.5	31.5	33.8	35.6	38.1	36.33	33.2	25.6	23.2	14.2	
2014	Monthly rainfall (mm)	92.6	2.4	99.2	58.0	36.2	57.8	168.8	62.8	98.2	117.8	54.8	121.2	969.8
	Min temperature	-4.3	-3.4	-0.8	3.2	4.9	10.1	12.3	12.8	5.9	2.4	-3.8	-5	
	Max temperature	17.7	19.6	25.4	24.4	30.7	34.2	34.9	35	29.8	28.4	19.7	15.	

30:1. The injector and detector temperatures were kept at 220°C and 250°C, respectively. The amount of each compound was expressed as a relative percentage of the total area of the chromatograms.

### Statistical analysis

All experimental data are presented as mean values with their corresponding standard deviation. Growth data as well as composition of the essential oils were compared for statistical differences among the various sampling periods by using the repeated measures ANOVA at an alpha level of 5%. All analyses were performed with the SPSS v20 statistical package (IBM SPSS Statistics for Windows, Armonk, NY: IBM Corp.).

## Results

In Table 2 the results of the oregano growth are presented based on the samples collected at different periods of the experimental culture. In the same table the results of the concentration of essential oils are also presented.

The values in Table 2 represent the average of 4 measurements. With regard to plant growth, the most representative factor for estimation is to analyze the dry weight of the upper part of the plant (flowers) from which the oregano essential oil is produced. From Table 2 it appears that the dry weight of the upper part had a positive correlation with all the other growth-determining variables of the plant and therefore these variables were in covariance. From Table 2 it is clear that the irrigated crop prevailed against dry land in all harvesting periods, except for the year of installation where the harvest was negligible. Irrigated crop yields ranged from 0.9 g/plant in the year of installation and reached the maximum yield of 284.7 g/plant in June 2014 while yields for dry crops started at 1.5 g/plant in the year of installation and peaked at 155.5 g/plant in June 2014. For both treatments the maximum yield was in June 2014, in the first bloom of the second year.

As regards the quality of the oregano, the percentage of the essential oil and the upper part (flowers) and the bottom (stems) was studied, as well as an analysis of the essential oil. The content of essential oil is determined by the flowers of the plant. The strains have a much lower content of essential oil, and their quality is much lower than that of the flower oil, so the primary concern for analysis is the content of essential oil. The essential oil content of irrigated crops ranged from 4.3% in the year of installation to 6.3% in the June 2014 boom. For the dry crop, the minimum content was observed in the September harvest at 2.1% and the maximum yield in June 2013 with a percentage of 6.6%. As can be seen from Table 2, the maximum

yields for both treatments were in the June boom, while the figures for the two years did not show statistically significant differences between them, which mean that the first and the second year for both treatments the essential oil content is about the same. In the September harvests, there is a reduction of around 20% for irrigated crops, while for this drought the difference from the maximum to the minimum amount reaches over 60%. In the comparison between the two cultivation treatments, there is no clear precedence of one or the other treatment because there are differences in percentages but there are not statistically significant differences.

Finally, Table 2 also shows the nitrogen content of the upper and bottom that was analyzed in plants by the Kjeldahl method. The upper part for the irrigated crop ranged from 1.25% to 0.27% and from 0.65% to 0.25% for the bottom. The upper crop percentages ranged from 0.97% to 0.29% and from 0.72% to 0.22% for the bottom. For both treatments a corresponding picture was observed in the nitrogen levels where for the first year there were the maximum content for both the upper and the lower part and on the way there was a constant decrease until it reached the minimum in the last harvest in September 2014. A comparison between the two treatments from Table 2 shows that there were no statistically significant differences in percentages comparing the same per harvest period for both upper part and bottom of the plant.

In order to assess the quality of an aromatic plant and hence its essential oil, it is necessary to analyze its components by a gas chromatograph - mass spectrometer.

Through the GC/MS oil analysis, a total of 17 distinct components of the essential oil of oregano were semi-quantified by means of libraries of electronic libraries [29].

The total percentage of ingredients identified for each EO ranges from 96.6% to 99.2% which is considered to be quite high and which are able to give us a complete picture of the quality of each essential oil. Carvacrol is the predominant component of the essential oil of the Greek oregano [30,17], which is inextricably linked to its quality [6,31-33]. In our study, carvacrol was the main ingredient in both treatments. For irrigated crops the percentage of carvacrol started at 73.3% in June 2014 and reached 83% in September 2013. From Table 3 it appears that the percentages of carvacrol were higher at the September harvests at the second blossom of the plant. For dry cultivation, carvacrol ranged from 76.2% in June 2013 to 77.4% in June 2014, while the statistical analysis (Table 3) did not show any significant differences in the percentage of carvacrol for all harvests. Other components that were found to be in considerable quantities

**Table 2:** Growth and essential oil yield results from *Oregano vulgaris* L. collected in different periods (irrigated and non-irrigated treatments).

Variable	Irrigated cultivation					Non-Irrigated cultivation				
	Oct '12	June '13	Oct'13	June '14	Oct'14	Oct'12	June '13	Oct'13	June '14	Oct '14
Height(cm)	13±0.3 <sup>a</sup>	83.2±19 <sup>b</sup>	77±12.5 <sup>b</sup>	121±5 <sup>c</sup>	71.5±1.9 <sup>b</sup>	12.3±2 <sup>a</sup>	69.8±10.7 <sup>b</sup>	59.5±22.5 <sup>b</sup>	88.8±4.7 <sup>b</sup>	90±1.6 <sup>b</sup>
Diameter (cm)	6.2±0.9 <sup>a</sup>	66.2±28 <sup>b,d,f</sup>	77.5±18.9 <sup>b,f</sup>	150±46 <sup>e</sup>	85.2±3.8 <sup>b</sup>	6±0.8 <sup>a</sup>	41±15.7 <sup>c</sup>	46±11.5 <sup>d,c</sup>	102.5±15.1 <sup>b,f</sup>	100.2±2.5 <sup>f</sup>
Number of executives (g)	6.2±1.5 <sup>a</sup>	67±47 <sup>a,b,d,e</sup>	92.5±25 <sup>b,e</sup>	95.7±38 <sup>b,d,e</sup>	65±8.2 <sup>b,c</sup>	5±1 <sup>a</sup>	31.7±25 <sup>a,c</sup>	47.5±10.5 <sup>d,c</sup>	109±16 <sup>a</sup>	67±14.7 <sup>b,c,d</sup>
Fresh Weight Upper Part (g)	4.4±1.6 <sup>a</sup>	267±171.5 <sup>a,b,c,d,e</sup>	330±108 <sup>c</sup>	560±75 <sup>f</sup>	149±41 <sup>a,g</sup>	34±18.4 <sup>a,b</sup>	120±65.7 <sup>b,d,e</sup>	150±35.6 <sup>b,c,e</sup>	235.5±95.7 <sup>b,c,d,e</sup>	87.7±12 <sup>b,g</sup>
Fresh Weight Bottom (g)	6±0.7 <sup>a</sup>	168±42.5 <sup>a,b,c</sup>	206.7±24 <sup>e</sup>	400±53 <sup>d</sup>	66±15.9 <sup>c,f</sup>	3.7±2 <sup>a</sup>	75±26 <sup>c</sup>	82±18.5 <sup>b,c</sup>	197±113 <sup>b,e,f</sup>	66±4 <sup>b,f</sup>
Dry Weight Upper Part (g)	0.9±0.3 <sup>a</sup>	120±101.7 <sup>b,c,g,f</sup>	154.7±73 <sup>c,f</sup>	284.7±78 <sup>a</sup>	64.2±20.5 <sup>a,g</sup>	1.5±0.4 <sup>a,b</sup>	44±2 <sup>c,f</sup>	53±12.5 <sup>a,g</sup>	155.5±52 <sup>a,f,g</sup>	40.7±5 <sup>c,g</sup>
Dry Weight Bottom (g)	1.35±0.3 <sup>a</sup>	66.5±36.9 <sup>c,d,e</sup>	80±25.3 <sup>e,f</sup>	167.7±62 <sup>f</sup>	31.7±18 <sup>c,d</sup>	1±0.4 <sup>b</sup>	30.5±9.3 <sup>d</sup>	34±5.22 <sup>c,d</sup>	108.7±48 <sup>c,d,e,f</sup>	30.5±4.3 <sup>c,d</sup>
Percentage of Essential oil content Upper part %	4.3±0.25 <sup>a</sup>	5.5±0.43 <sup>d</sup>	5±0.26 <sup>a,g</sup>	6.3±0.2 <sup>c</sup>	4.8±0.5 <sup>a,f</sup>	3.8±0.1 <sup>a</sup>	6.6±0.3 <sup>c</sup>	2.1±0.14 <sup>a</sup>	5.8±0.5 <sup>b,c,f</sup>	4±0.5 <sup>a</sup>
Percentage of Essential oil content Bottom %	0.43 <sup>a</sup>	0.43 <sup>a</sup>	0.28± <sup>a</sup>	0.3±0.05 <sup>a,g</sup>	0.21 <sup>a</sup>	0.32±0.1 <sup>a,g</sup>	0.7±0.17 <sup>f</sup>	0.15±0.05 <sup>a</sup>	0.2 <sup>a</sup>	0.23±0.05 <sup>a</sup>
Nitrogen Upper part %	-	1.25±0.5 <sup>a</sup>	0.32±0.1 <sup>b,c,d</sup>	0.27 <sup>d</sup>	0.27±0.05 <sup>d</sup>	-	0.97±0.43 <sup>a,b</sup>	0.46±0.22 <sup>c,d</sup>	0.28±0.01 <sup>c,d</sup>	0.29 <sup>b,c</sup>
Nitrogen Bottom %	-	0.65±0.2 <sup>a</sup>	0.36±0.16 <sup>a,b,c</sup>	0.26±0.05 <sup>b</sup>	0.25±0.05 <sup>c</sup>	-	0.72±0.25 <sup>a</sup>	0.37±0.2 <sup>a,b,c</sup>	0.26 <sup>b</sup>	0.22 <sup>c</sup>

Similar superscript letters on a row indicate no significant differences (Repeated measures ANOVA, 5% alpha)

**Table 3:** Composition of essential oil from *Oregano vulgaris* L. collected in different periods (irrigated and non-irrigated treatments).

Compound	Irrigated				Non-irrigated		
	June 2013	October 2013	June 2014	October 2014	June 2013	June 2014	October 2014
1 α-thujene	1.4±0.1 <sup>a</sup>	0.3± 0.1 <sup>b,c</sup>	0.5 ±0.1 <sup>b,d</sup>	0.4±0.2 <sup>b,c,d,f</sup>	0.4 <sup>b</sup>	0.7±0.1 <sup>e</sup>	0.2±0.1 <sup>c,f</sup>
2 α-pinene	0.3±0.1 <sup>a,c</sup>	0.7±0.1 <sup>c</sup>	ND	0.7±0.3 <sup>a,c</sup>	0.1 <sup>a</sup>	ND	0.8±0.1 <sup>c,f</sup>
3 β- pinene	ND	ND	ND	0 <sup>a</sup>	ND	0.7±0.1 <sup>b</sup>	0 <sup>a</sup>
4 Octen-4-ol	ND	ND	0.3	ND	ND	ND	ND
5 Myrcene	1.2±0.1 <sup>a</sup>	1.4±0.4 <sup>a,c</sup>	1±0.1 <sup>b,c,d</sup>	1.1±0.1 <sup>a,d,f</sup>	0.5±0.2 <sup>b</sup>	ND	1 <sup>d,f</sup>
6 α-terpinene	1.1±0.2 <sup>a</sup>	0.2 <sup>c</sup>	0.7±0.1 <sup>a,d</sup>	0.4±0.2 <sup>b,c,d</sup>	0.7±0.2 <sup>a,b</sup>	0.5±0.1 <sup>b,d,e</sup>	ND
7 Paracymene	6.5±0.4 <sup>a,b</sup>	6.5±0.5 <sup>a,c</sup>	8±0.8 <sup>d</sup>	3.6±0.3 <sup>f</sup>	5.5±0.2 <sup>b</sup>	5.7±0.3 <sup>a,b,e</sup>	10.2±1 <sup>a</sup>
8 γ-terpinene	6±0.5 <sup>a</sup>	3.1±0.1 <sup>c</sup>	10.1±0.9 <sup>d</sup>	7±0.6 <sup>f</sup>	5±0.4 <sup>b</sup>	9.1±0.4 <sup>e</sup>	5.4±0.3 <sup>a,b</sup>
9 Borneol	0.2	ND	ND	ND	ND	ND	ND
10 Terpinen-4-ol	0.4±0.1 <sup>a</sup>	ND	0.2 <sup>b</sup>	ND	0.4±0.1 <sup>a</sup>	ND	ND
11 Thymol-methyl-ether	ND	ND	ND	ND	ND	0.2±0.1	ND
12 Carvacrol-methyl-ether	0.4±0.1 <sup>a</sup>	ND	ND	ND	0.4±0.2 <sup>b</sup>	0.7±0.2 <sup>b</sup>	0.2±0.1 <sup>c</sup>
13 Thymol	3.5±0.4 <sup>a</sup>	ND	0.2 <sup>c</sup>	ND	7.4±0.2 <sup>b</sup>	0.8±0.2 <sup>c</sup>	ND
14 Carvacrol	75.7±0.2 <sup>a</sup>	83±0.7 <sup>c</sup>	73.3±0.2 <sup>d</sup>	80.3±0.4 <sup>e</sup>	76.2±0.5 <sup>b</sup>	77.5±0.4 <sup>b</sup>	76.8±0.6 <sup>a,b,c</sup>
15 Caryophyllene [E-]	1.7±0.6 <sup>a,b,c</sup>	1.6±0.1 <sup>a,b,c</sup>	1.8±0.3 <sup>c</sup>	1.4±0.3 <sup>a,b,c,e</sup>	1.5±0.4 <sup>b</sup>	1±0.1 <sup>a,b,e</sup>	1±0.2 <sup>e</sup>
16 α-humulene	ND	0.6±0.1 <sup>b</sup>	ND	ND	0.2 <sup>a</sup>	0.4±0.1 <sup>b</sup>	0.6±0.1 <sup>b</sup>
17 β-bisabolene	0.4±0.1 <sup>a</sup>	ND	0.5±0.3 <sup>a,c</sup>	0.5±0.3 <sup>a,b</sup>	0.8 <sup>b,c</sup>	ND	0.3 <sup>a,b</sup>
<b>Grouped comp.</b>							
Monoterpen hydrocarbons	16.6±0.1 <sup>a</sup>	12.6 <sup>c</sup>	20.4±1.6 <sup>d</sup>	13.3±0.1 <sup>c</sup>	12.3±0.1 <sup>b</sup>	17.1±0.1 <sup>a</sup>	17.8±0.1 <sup>b</sup>
Monoterpenyl alcohols	80±0.1 <sup>a</sup>	82.9 <sup>c</sup>	73.8±0.1 <sup>d</sup>	80.4±0.3 <sup>f</sup>	84±0.3 <sup>b</sup>	78.4±0.1 <sup>e</sup>	76.9 <sup>g</sup>
Sesquiterpens Hydrocarbons	2.1±0.1 <sup>a</sup>	1.6±0.1 <sup>c</sup>	2.4±0.2 <sup>b</sup>	2 <sup>a</sup>	2.4±0.1 <sup>b</sup>	1 <sup>d</sup>	1.4±0.2 <sup>c</sup>
Others	0.5 <sup>a</sup>	0.6±0.1 <sup>a</sup>	0.3 <sup>c</sup>	ND	0.9±0.1 <sup>b</sup>	1.3±0.1 <sup>d</sup>	0.8±0.1 <sup>b</sup>
TOTAL	99.2	97.7	96.9	95.7	99.6	97.8	96.9

ND: Not Detected. Similar superscript letters on a row indicate no significant differences (Repeated measures ANOVA, 5% alpha)

were p-cymene ranging from 3.6%-8% for the irrigated culture and 5.5%-10.2% for dry cultivation. Another component was γ-terpinene detected by 3.1%-10.1% and from 5%-9.1% respectively for the two treatments. The remaining components, except thymol, which were detected at 3.5% and 7.4% only for irrigated and dry crop irrigation in June 2013 respectively, were detected at levels below 1.7%, which means they play a minor role in the composition of the essential oil. We should mention that components which were in small concentrations are equally important in the case of the essential

oil activity (antimicrobial activity) since they are considered to be involved in the formation of the final active chemotype. However, since there are no detailed studies on the action of both the individual components that appear at low concentrations they often tend to be ignored.

With regard to the separation into grouped ingredients, monoterpen alcohols (oxygenated monoterpenes) were in abundance at rates ranging from 73.8%-82.9% and from 76.9-84% for irrigated



and dry crops, respectively. Monoterpen hydrocarbons ranged from 12.6%-20.4% and 12.3%-17.8% and sesquiterpenic hydrocarbons from 1.6%-2.4% and from 1%-2.4% respectively for the two treatments.

## Discussion

In table 2 the results of the growth of oregano cultivation and, as mentioned above, the most representative factor to estimate is to analyze the dry weight of the upper part of the plant (flowers) from which the oregano essential oil is produced. From Table 2 it appears that average yields ranged from 40.7 g/plant to 284.7 g/plant for both crops (except year of installation). The yields are considered to be quite high compared to the yields reported by Baranauskienė et al., (2003) [34] for fresh whole plant which yielded from 150 g/plant to 400 g/plant or 8 to 18 t/ha when we have a yield of 150 g/plant to 1060 g/plant of fresh weight in 2013 and 2014 for both crops. In the harvests of the first blossom in June the lowest yield is 200 g/plant in the dry crop. As for the quality of the oregano as mentioned above, the percentage yield of the essential oil and its composition are very significant. The content of EO is determined by the flowers of the plant. The essential oil content varied from 2.1% in the installation year to 6.6% for both treatments. Corresponding performance rates have been reported by other investigators [4,30,32,35]. The percentages we received are considered quite high especially for the maximum yields compared to the results from other species of oregano such as *O. vulgare* ssp. *vulgare* and *O. vulgare* ssp. *viridulum* with an essential oil content of 0.3% to 0.8% (Kokkini & Vokou, 1989; Kokkini et al., 1994) [17,32]. There are, of course, researchers who have reported rates of 8% in Greek oregano [17]. In addition to the essential oil content as mentioned above, to assess the quality of an aromatic plant and hence its essential oil, it is necessary to analyze its components with a gas chromatograph- mass spectrometer. In our study, the carvacrol was the main ingredient in both treatments. Carvacrol rates ranged from 73.3% to 83%. Similar figures have been published by other researchers, Karamanos & Sotiropoulou [35] report rates from 56.4% to 84.88% and Azizi et al., (2009) [36] rates from 70% to 77.4%. Published reports with much lower carvacrol levels from 3% to 68% have also been reported, [2,37] while a species of Greek oregano has been found to have a percentage of carvacrol of up to 95% [17]. Other components that are found to be satisfactory are p-cymene ranging from 3.6% to 10.2%. At lower rates the results of Azizi et al. (2009) [36] varied in 3 species of oregano, i.e., from 4.3% to 5.3% Karamanos & Sotiropoulou (2013) [35] reported rates from 4.19% to 21.4%. With regard to the classification into grouped ingredients, monotonic alcohols (oxygenated monoterpenes) were in abundance at rates ranging from 73.8% to 84%. Monotone hydrocarbons are present at rates ranging from 12.3% to 20.4% and sesquiterpenes hydrocarbons from 1% to 2.4% and from 1% to 2.4%. Corresponding results have been provided by Pesavento et al., (2015) [38] with oxygenated monoterpenes, 77.2%, monerpene hydrocarbons with 19.2% and sesquiterpenes hydrocarbons with 2.9%.

## Conclusion

In conclusion, the oregano is adaptable to the climatic conditions of the area both in terms of dryness and yield of essential oil and it could be proposed to cultivate in the type of areas perennially mainly irrigated and to produce two harvests in June and September and also the essential oil of oregano is of high quality so it has high commercial value.

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