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Effects of Drought Stress on Biomass, Essential Oil Content, Nutritional Parameters, and Costs of Production in Six Lamiaceae Species

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Abstract: Lamiaceae is one of the largest families of aromatic plants and it is characterized by the presence of external glandular structures which produce essential oils highly valued in cosmetics and medicine. Plants of *Lavandula latifolia*, *Mentha piperita*, *Salvia sclarea*, *Salvia lavandulifolia*, *Thymus capitatus*, and *Thymus mastichina* were grown for one year. In order to evaluate the effects of drought stress, plants were subjected to two water treatments (100% ETo and 70% ETo, including the rainfall during the experimental period). At the end of the experiment, the biomass, the essential oil content, and leaf nutrients concentration were assessed for each water treatment and species studied. At the end of the experiment, *L. latifolia*, *M. piperita* and *T. capitatus* plants showed a significant fresh weight reduction under drought stress conditions whereas the other species studied remained unchanged. With respect to dry weight, only *L. latifolia* plants showed a reduction under water deficit conditions. As far as essential oil content was concerned, *L. latifolia* and *S. sclarea* plants had a reduction under water deficit conditions. Leaf nutrient concentration showed different trends between species considering the nutrient assessed. The economic viability of the growth of this species will be dependent on the benefits achieved which are related to yield production obtained and the price accorded for both raw material and the essential oil extracted.

Keywords: *Lavandula latifolia*; *Mentha piperita*; *Salvia lavandulifolia*; *Salvia sclarea*; *Thymus capitatus*; *Thymus mastichina*

1. Introduction

Water deficit or drought stress is considered as one of the most restrictive factors in plant growth and yield in many areas of the world with agricultural production [1]. Under drought stress, water potentials in the root zone become sufficiently negative, resulting in a reduction of water availability which affects the plant growth and development [2].

Drought stress generates changes at physiological and metabolic levels such as stomatal closure and reduction in photosynthesis rate and crops growth [3]. Under these adverse conditions, the amount of oxidized reduction equivalents (NADP⁺) working as electron acceptors is reduced. Sequentially, large amounts of NADPH + H⁺ are accumulated, resulting in a high over-reduced state. As a consequence, it is necessary to have a high level of consumption of NADPH + H⁺ in a plant, for instance, through the biosynthesis of highly reduced secondary compounds, such as phenols, terpenoids, alkaloids, cyanogenic glycosides and glucosinolates, which affect essential oil percentage and essential oil content in aromatic plants [4]. For instance, changes in the essential oil content and composition in Lamiaceae species under drought conditions have been noted by different researchers (Khorasaninejad et al. [5]; Nowak et al. [6]). The Lamiaceae (Labiatae) plant family is considered one of the largest families among the dicotyledons with more than 240 genera. This family is highly valued in terms of cosmetic and medicinal value due to the presence of external glandular structures that produce volatile oil [7].

In this experiment, we have focused on six species of the Lamiaceae family commonly grows in the Mediterranean region: *Lavandula latifolia*, *Mentha piperita*, *Salvia lavandulifolia*, *Salvia sclarea*, *Thymus capitatus* and *Thymus mastichina*. *Lavandula latifolia* Med. It is an aromatic shrub (30–80 cm high) with evergreen leaves and purple flowers [8]. The traditional uses of this aromatic plant have been as a raw material in perfumery and cosmetics and in pharmaceutical preparations for its sedative and antihypertensive effects [9]. *Mentha piperita* L. (peppermint) is commercially the most important mint species [10]. It is a perennial shrub (50–90 cm high) with branched stems which are often purplish or tinged violet, dark or light green leaves short-petioled, and the flowers are purple or pinkish [11]. It is frequently used as a flavoring agent for foods and beverages [12]. *Salvia lavandulifolia* Vahl. (Spanish sage) is a plant native in Spain and it is also distributed in North and South America, Europe and Asia [13,14]. It is a small woody herbaceous perennial shrub up to 17–100 cm with simple green or light gray leaves and mauve-blue flowers [15]. This aromatic species has medicinal uses as an antiseptic, spasmolytic, analgesic, sedative, and antioxidant [16]. *Salvia sclarea* L. (clary sage or clear eye) is a perennial plant native to southern Europe and central Asia [17]. It is a shrub that grows up to 60–100 cm with large hairy leaves and small blue, white or purple flowers [18]. It is widely valued for its aromatic and medicinal properties [19]. *Thymus capitatus* (L.) Hoff. et Link. syn. *Thymbra capitata* (L.) Cav., syn. *Coridothymus capitatus* (L.) Reichb. fil. is a native plant to the Mediterranean Basin region [20]. It is a shrub characterized by erect sparring branches (20–40 cm high), small linear and glandulous spotted leaves, and ovoid terminal inflorescences with pink flower corolla [21]. This species can be used in aromatherapy and it is also an excellent option for environmental restoration, forestry, and xerogardening [22]. *Thymus mastichina* L. (Spanish marjoram) is distributed mainly in the Iberian Peninsula [23]. It is characterized by leaves distributed in opposite pairs and small zygomorphic and bilabiate flowers [24]. It is an endemic thyme with a high added value due to its use as spices and/or medicinal herbs, with several pharmacological properties, such as antispasmodic, antiseptic, antitussive actions [25].

The effects of drought stress on crops is a focus of research especially with medicinal and aromatic plants in arid areas in order to improve the yield and the production of essential oils. Therefore, the present study aimed to discern the effects of drought stress on yield, essential oil content, and leaf nutrient concentration in six Lamiaceae species.

2. Materials and Methods

2.1. Experimental Site and Growth Conditions

Seeds of all species were provided by the Institute of Research and Training in Agriculture and Fishery (IFAPA), located in Las Torres-Tomejil, Sevilla, Spain. Seeding was performed in polyethylene trays with alveols of 150 mL of volume filled with a mixture of black and blonde peatmoss (1:1). The substrate humidity levels were maintained by capillary action. The seedbed was placed in a

greenhouse ubicated at the facilities of the IFAPA (Sevilla, Spain) where the temperature was regulated at 25 °C in the daytime and 15 °C at night. When plants already had the fifth leaf completely spread, they were carried to the Institute of Research and Training in Agriculture and Fishery (IFAPA), located in La Mojonera, Almería, Spain (2°41' W, 36°47' N) for the development of the experiment. The experiment was conducted from July of 2011–June of 2012 in an experimental plot with a final harvest at the end of the experimental period. Almería is characterized by having a tropical and subtropical steppe climate (BSk) as stated by the Köppen classification, with a yearly average temperature of 19 °C (12 °C and 22 °C during winter and summer, respectively). The number of sunny hours per years is 2965 h, the average relative humidity is around 70%, and annual rainfall is scarce, ranging from 200–250 L m⁻² [26]. The chemical composition of the soil at the study is presented in Table 1.

Table 1. Chemical soil properties.

Chemical Soil Properties	
Organic matter (%)	0.51
EC (dS m ⁻¹)	0.64
pH	8.76
Available nutrients	
N-NO ₃ ⁻ (ppm)	30.50
N-NH ₄ ⁺ (ppm)	0.12
P (ppm)	45.53
K (mg kg ⁻¹ soil)	175.00
Ca (mg kg ⁻¹ soil)	1892.00
Mg (mg kg ⁻¹ soil)	281.00

The main characteristics of the irrigation water were the basic pH (8.02) and the low electrical conductivity (0.64 dS m⁻¹), which is suitable for growing aromatics herbs crops. The chemical composition of the irrigation water is presented in Table 2.

Table 2. The chemical composition of irrigation water.

pH	EC (dS m ⁻¹)	mmolc L ⁻¹										
		CO ₃ ²⁻	HCO ₃ ⁻	Cl ⁻	SO ₄ ²⁻	NO ₃ ⁻	H ₂ PO ₄ ⁻	NH ₄ ⁺	Ca ²⁺	Mg ²⁺	Na ⁺	K ⁺
8.02	0.64	0.20	3.30	1.17	1.09	0.07	0.00	0.00	2.09	3.40	1.03	1.02

2.2. Experimental Design and Irrigation Treatments

The experiment was designed in a randomized block design with four replications and two irrigation regimen per species. Each experimental plot size was 5.5 m × 4.5 m, and in each plot the distance between lines was 50 cm for *L. latifolia*, *M. piperita*, *S. lavandulifolia* and *S. sclarea* (planting density of 1.45 plants per m²) and 25 cm for *Thymus capitatus* and *Thymus mastichina* (planting density of 2.90 plants per m²). Two irrigation (I) levels were based on the difference between reference evapotranspiration demand (ET_o) and rainfall. ET_o was calculated daily by the method of Penman Monteith [27] in this experiment: 100% ET_o and 70% ET_o were considered. ET_o was calculated using data from a weather station placed in the facilities at IFAPA (Almería, Spain). The 100% ET_o indicates that the plants were fully irrigated without any water stress throughout the experimental period. The treatment with 70% ET_o received only 70% related to the irrigation amount applied in the 100% ET_o irrigation. These irrigation levels were applied throughout the growing seasons for the experimental period following the recommendations given by local growers. All water treatments were irrigated on the same day. In addition, rainfall amounts were recorded throughout the experimental period (Table 3). An Irritrol-JRMAX-4 controller (Irritrol Systems, Riverside, CA, USA) was used to check the opening and closing of the valves. The drip lines had emitters spaced 25 cm apart with an

emitter flow rate of 3 L h^{-1} . In each experimental plot were installed flow meters in the hose nozzle to determine the volume of irrigation water applied.

Table 3. Monthly values of rainfall (R), evapotranspiration (ET_o), and irrigation (I) in each water treatment throughout the experimental period (years 2011 and 2012).

Year	Month	R (L m ⁻²)	ET _o (L m ⁻²)		I (L m ⁻²)	
			100%	70%	100%	70%
2011	July	0.2	120.6	84.4	120.4	84.2
2011	August	0	180.5	126.3	180.5	126.3
2011	September	5	136.5	95.5	131.5	90.5
2011	October	29.6	90	63	60.4	33.4
2011	November	47.2	60.8	42.5	13.6	0.0
2011	December	2.4	42.5	29.7	40.1	22.6
2012	January	22.4	46.5	32.5	24.1	10.1
2012	February	8.6	64.7	45.2	56.1	36.6
2012	March	13	94.3	66.0	81.3	53
2012	April	9.4	125	87.5	115.6	78.1
2012	May	0.4	162.2	113.5	161.8	113.1
2012	June	3.4	172.2	120.5	168.8	117.1
	Total	142	1296	907	1154	765

2.3. Biomass, Water Content, and Essential Oil Content Determinations

The dates of harvests were between May and June depending on the species considered. For each species and treatment, measurements of fresh and dry weights were evaluated by destructive harvests of sixteen randomly selected plants from the central rows of each plot. Plants were harvested manually in order to avoid mechanical damages 10 cm above the plant's neck and immediately were weighed in order to determine the fresh weight (FW). To calculate the essential oil content expressed in mL kg⁻¹ DW or mL m⁻², four randomized subsamples of 125 g of vegetal material (sample of 500 g in total) per plot were distilled after being dried in the shadows with air circulation for 2 days. The distillation process was conducted using a vapor dragging with a distiller of the Clavenger type. Distillation time was set at 1.5 h per sample. The remaining material of each plant was then wrapped in a clean paper bags, labeled, and oven-dried at 65 °C for 48 h to determine the respective dry weight (DW) following the methodology established by Corell et al. [28]. Fresh and dry weights were expressed in kilograms per m² and were used to determine the water content (-).

2.4. Leaf Nutrient Concentration

The oven-dried samples of the aerial part of the plant were ground to a fine powder in a mill (Grindomix GM 200; Retsch, Haan, Germany), cleaning carefully between each sample. Each sample was then split up in two subsamples. The first subsample was used to determine the soluble N-NO₃⁻ by HPLC methodology, as described by Csaky and Martinez-Grau [29]. The other subsample was mineralized with 96% sulfuric acid in the presence of P-free hydrogen peroxide (30% weight/volume) at 300 °C (humid mineralization) for the determination of organic N [30], total P [31], and K⁺ [32] concentration. The total N concentration was calculated as the sum of the organic N and N-NO₃⁻ concentration.

2.5. Economic Analysis

An economic analysis was carried out to discern the effects of changes in the amounts of irrigation water applied and the resulting yields on the costs of production and the gross revenue according to the methodology reported by Nakawuka et al. [33].

2.6. Statistical Analysis

The data were analyzed through one-way analysis of variance (ANOVA) and least significant difference (LSD) tests ($P < 0.05$) in order to assess the differences between treatments using Statgraphics Centurion XVI.II (Statpoint Technologies, Inc. Warrenton, VA, USA).

3. Results

3.1. Biomass, Water Content, and Essential Oil Content Determinations

Drought stress reduced the fresh weight in *L. latifolia*, *M. piperita*, and *T. capitatus*, whereas *S. lavandulifolia*, *S. sclarea*, and *T. mastichina* did not vary the fresh weight under drought conditions. With respect to dry weight, *L. latifolia* was the only species which showed a significant decrease under water deficit environments while the rest of species remained unchanged. The water content did not show any changes in *L. latifolia*, *S. lavandulifolia*, *S. sclarea*, and *T. mastichina* under drought stress, whereas *M. piperita* and *T. capitatus* plants decreased the water content when they were subjected to drought conditions.

As far as essential oil content (expressed in mL kg⁻¹ DW) was concerned, drought stress only decreased the essential oil content in *S. sclarea* plants, whereas the rest of the species studied showed no changes in essential oil content under drought conditions. Nevertheless, considering the essential oil content (expressed in mL m⁻²), *S. sclarea* and *L. latifolia* showed a reduction of 36% and 53% of the essential content, respectively. In the case of *L. latifolia*, the reduction of the essential oil content (expressed in mL m⁻²) was related to the decrease of the dry weight under drought conditions (Table 4).

Table 4. Fresh weight and dry weight, water content, and essential oil content in the species studied under different drought conditions.

Species	Treatments	FW (Kg m ⁻²)	DW (Kg m ⁻²)	% Water Content	Essential Oil Content (mL kg ⁻¹ DW)	Essential Oil Content (mL m ⁻²)
<i>L. latifolia</i>	100%	1.41 ± 0.13 a	0.50 ± 0.05 a	64.91 ± 6.07 a	28.49 ± 2.01 a	14.19 ± 1.21 a
	70%	0.84 ± 0.06 b	0.34 ± 0.04 b	59.99 ± 6.05 a	26.69 ± 2.12 a	9.09 ± 0.92 b
<i>M. piperita</i>	100%	1.26 ± 0.12 a	0.33 ± 0.03 a	77.49 ± 6.24 a	22.18 ± 1.88 a	7.28 ± 0.68 a
	70%	0.92 ± 0.09 b	0.30 ± 0.03 a	65.19 ± 5.93 b	21.34 ± 2.08 a	6.54 ± 0.61 a
<i>S. lavandulifolia</i>	100%	0.57 ± 0.05 a	0.15 ± 0.01 a	73.40 ± 6.85 a	24.14 ± 2.18 a	3.62 ± 0.34 a
	70%	0.63 ± 0.05 a	0.16 ± 0.01 a	74.44 ± 7.23 a	24.75 ± 2.22 a	3.86 ± 0.38 a
<i>S. sclarea</i>	100%	0.71 ± 0.07 a	0.14 ± 0.01 a	80.42 ± 8.11 a	27.68 ± 2.42 a	3.88 ± 0.36 a
	70%	0.70 ± 0.07 a	0.14 ± 0.01 a	80.13 ± 7.80 a	13.05 ± 1.14 b	1.81 ± 0.17 b
<i>T. capitatus</i>	100%	1.57 ± 0.11 a	0.53 ± 0.05 a	66.34 ± 6.07 a	46.36 ± 4.59 a	24.57 ± 2.44 a
	70%	1.08 ± 0.10 b	0.50 ± 0.05 a	53.64 ± 5.04 b	44.65 ± 3.97 a	22.32 ± 2.18 a
<i>T. mastichina</i>	100%	0.78 ± 0.06 a	0.24 ± 0.02 a	69.48 ± 6.65 a	63.54 ± 6.17 a	15.25 ± 1.41 a
	70%	0.81 ± 0.07 a	0.25 ± 0.02 a	68.60 ± 6.58 a	67.39 ± 6.41 a	16.65 ± 1.48 a

Values are the means ± standard deviation of sixteen plants per treatment in fresh weight (FW), dry weight (DW) and water content and in the case of oil content (expressed in mL kg⁻¹ DW or mL m⁻²), the number of representative samples was four. Means within a column within a species without the same letter are significantly different at $P < 0.05$ (ANOVA and LSD test).

3.2. Aerial Part Nutrient Concentration

Leaf N concentration did not vary in *M. piperita*, *S. lavandulifolia*, *S. sclarea* and *T. capitatus* under drought stress, whereas *L. latifolia* and *T. mastichina* plants reduced leaf N concentration under drought conditions. With respect to leaf P concentration, there was a decrease in all the species under drought conditions, unless for *S. sclarea* plants, which remained without changes under drought stress. Concerning leaf K concentration, there were no differences under drought conditions in all the species studied (Table 5).

Table 5. Leaf nutrient concentration in the species studied under different drought conditions.

Species	Treatments	N	P	K
<i>L. latifolia</i>	100%	22.05 ± 2.01 a	1.56 ± 0.07 a	23.31 ± 2.01 a
	70%	17.57 ± 1.61 b	1.22 ± 0.05 b	22.69 ± 2.12 a
<i>M. piperita</i>	100%	32.05 ± 2.98 a	7.18 ± 0.53 a	20.63 ± 1.88 a
	70%	35.89 ± 3.12 a	4.69 ± 0.24 b	21.86 ± 2.08 a
<i>S. lavandulifolia</i>	100%	32.90 ± 3.06 a	5.56 ± 0.35 a	31.56 ± 3.18 a
	70%	33.48 ± 2.92 a	3.74 ± 0.23 b	33.41 ± 3.22 a
<i>S. sclarea</i>	100%	26.51 ± 2.23 a	5.04 ± 0.31 a	35.68 ± 3.42 a
	70%	28.19 ± 2.41 a	4.93 ± 0.30 a	33.59 ± 3.14 a
<i>T. capitatus</i>	100%	16.27 ± 1.39 a	1.41 ± 0.07 a	17.94 ± 1.59 a
	70%	15.42 ± 1.31 a	0.96 ± 0.04 b	18.98 ± 1.67 a
<i>T. mastichina</i>	100%	29.84 ± 2.47 a	5.68 ± 0.35 a	23.51 ± 2.07 a
	70%	23.83 ± 2.31 b	4.52 ± 0.28 b	22.89 ± 2.11 a

Values are the means ± standard deviation of four plants per treatment. Means within a column within a species without the same letter are significantly different at $P < 0.05$ (ANOVA and LSD test).

3.3. Economic Analysis

The costs of aromatic and medicinal production plants were classified into two different groups: Fixed costs or variable costs. Fixed costs for in this experiment are those costs independent of amounts of irrigation water applied, whereas variable costs are dependent on differences in the amounts of water applied. Fixed costs include field activities (soil preparation, planting, irrigation system, weeds, fertilization, insecticide application), harvests, and other costs such as land rent. The variable costs include the cost of water per ha and the volume of irrigation water applied (Table 6). The cost of water varies during the experimental period depending on the available water and the cost of pumping. The benefits are related to the yield production and the essential oil content of each species cultivated and the price of the raw material or the essential oil extracted which varies depending on the sales.

Table 6. Costs (€) per ha for the cultivation of the species selected in the experiment.

	Experimental Period
Initial activity	
Soil preparation	110
<i>Planting</i>	
• Vegetal material	1286
• Planting activities	567
<i>Irrigation system</i>	
• Establishment	2814
Total	4777
Annual activities	
• Repair and maintenance of irrigation system	1015
<i>Weeds</i>	
• Herbicide application	180
• Weeding	504
<i>Fertilization</i>	217
<i>Insecticide application</i>	80
<i>Actual harvesting</i>	600
<i>Transporting</i>	50
<i>Residue disposal and packing</i>	500
Total	3146
Other costs	
<i>Land rent</i>	600
Total	8523

4. Discussion

Under water deficit conditions, there is a decline in turgor which originates a decrease in both growth and cell development in the aerial part of the plant, especially in stems and leaves [34]. The results obtained in our experiment concerning to fresh weight reported that there was a different trend between the species studied, since under drought stress *L. latifolia*, *M. piperita* and *T. capitatus* showed a significant reduction in biomass, whereas *S. lavandulifolia*, *S. sclarea*, and *T. mastichina* remained without changes. The reduction of fresh biomass in the species studied in our experiment was in line with the results obtained in other Lamiaceae species by other researchers. For instance, Liu et al. [35] reported that water stress resulted in a reduction of fresh weight in *Salvia miltiorrhiza* plants when they were subjected to medium drought stress (MD) and severe drought stress (SD) (50% and 40% field water capacity, respectively). On the same note, Ekren et al. [36] noted a significant reduction of fresh weight in *Ocimum basilicum* plants under water deprivation, suggesting in this way that this species was sensitive to water stress. This decrease of fresh weight in the aerial part of the species studied in our experiment may be the result of differential growth in the root zone and consequently in its biomass as reported by Bettaieb et al. [37]. No differences in fresh weight in *S. lavandulifolia*, *S. sclarea*, and *T. mastichina* may be ascribed to the lower drought levels. In our experiment, no significant differences on dry weight in the species studied under drought conditions were found unless for *Lavandula latifolia*. No differences on dry weight in the species studied may be due to the process of drying since it reduces the differences in fresh weight between treatments due to the water evaporation in the samples. The reduction of dry weight under water deficit conditions in *L. latifolia* in our experiment was in line with the results obtained by other researchers in species of the Lamiaceae family. For instance, Delfine et al. [38] reported a decrease of the aerial part dry mass in *Mentha spicata* and *Rosmarinus officinalis* subjected to different watering regimes (daily restoring 0 (severe stress, S0), 50 (moderate stress, S50) and 100% (optimal conditions, S100) of the estimated plant water use. Similarly, Khorasaninejad et al. [5] carried out two experiments to discern the effects of drought stress on growth parameters in *Mentha piperita*, and the results obtained reported a significant reduction in dry weight. On the same hand, Govahi et al. [39] reported that *Salvia officinalis* plants grown under water deficiency showed a reduction of dry weight.

As far as water content was concerned, only *M. piperita* and *T. capitatus* plants showed a reduction of water content under drought conditions, while the other species remained without changes. No variations in water content in the species in our experiment may be ascribed to the lower drought levels imposed in our experiment. On the other hand, the reduction of water content in *M. piperita* and *T. capitatus* in our experiment was in line with the results reported in other experiments carried out with Lamiaceae species under drought conditions. For instance, Cermeño and Romero [40] evaluated the effects of different irrigation treatments based on the ETo levels in *Salvia sclarea* plants, and the results obtained reported a significant decrease in the water content under water deficit. Similarly, Bahreininejad et al. [41] reported the same trend in *Thymus daenensis* plants under water stress.

Drought stress generates an over-reduce state which involves the synthesis of secondary compounds, which affects the essential oil content [4]. In our experiment, in all species studied, only *L. latifolia* and *S. sclarea* plants showed a decrease of essential oil content under drought conditions. Investigating in foregoing literature, it was noted that there are contradictory results between our results and other reports as a consequence of differences in drought conditions and duration, the physiological status of the plant, plant species, and even cultivars of the same species. For instance, some researchers have reported a decrease in essential oil content in different Lamiaceae species such as *Mentha arvensis* [42], *Rosmarinus officinalis* [43,44], and *Salvia officinalis* [39]. Nevertheless, other researchers have reported an increase in essential oil content in Lamiaceae species under drought conditions. For instance, in one experiment with *Salvia officinalis* plants, Bettaieb et al. [37] reported that the essential oil content increased from 0.39% under 100% of field capacity (water control treatment) to 1.77 and 1.01% under moderate (50% of field capacity) and severe (25% of field capacity) water levels, respectively. A similar result has been reported earlier on other Lamiaceae species such as

Satureja hortensis [45] and *Lavandula angustifolia* [46]. The increase of essential oil content under drought conditions may be related to a higher density of oil glands, mainly due to the reduction in leaf area as a consequence of the stress generated by the water deficit.

Reviewing preceding experiments, there are few references about the effect of drought stress in leaf mineral composition in Lamiaceae species. It is worthy to mention that the leaf macronutrients concentration in our experiment differs depending on the Lamiaceae species studied. No differences in leaf N concentration in *M. piperita*, *S. lavandulifolia*, *S. sclarea* and *T. capitatus* may be ascribed to the lower drought levels imposed in our experiment. Nevertheless, the reduction of leaf N concentration in *L. latifolia* and *T. mastichina* was in line with the results obtained by Corell et al. [28] in *Salvia officinalis* where the increase of water stress in plants resulted in a reduction of leaf N concentration. On the same hand, Osuagwu et al. [47] reported a decline in leaf N concentration in *Ocimum gratissimum* plants when they were subjected to mild water stress (500 mL of water applied once per week) instead of the control treatment (750 mL of water applied three times per week). This reduction of leaf N concentration can be ascribed to a reduction in leaf Nitrate reductase (NR) activity which is joined to the photosynthesis activity and the availability of C skeletons, which are reduced under water deficit conditions [48]. Contrary to the results obtained in this experiment, other researchers have reported an increase of leaf N concentration in other crops under drought conditions such as *Pinus halepensis* [49] and *Leymu chinensis* [50]. This increase in leaf N concentration may be due to the translocation of nitrogen towards leaves for the synthesis of special protein in plants as a mechanism to overcome the effect of water deficit conditions as reported by Farooq et al. [51].

In our experiment, drought stress resulted in a decrease in leaf P concentration in all species besides *S. sclarea*, which remained without changes. A significant decrease in leaf P concentration was also reported by Osuagwu et al. [47] in *Ocimum gratissimum* plants subjected to mild water stress. Comparing the ranges of leaf P concentration in the species selected in our experiment with the few results reported by other researchers, it can be highlighted that the range of leaf P concentration in *Mentha piperita* in our experiment (from 4.7 to 7.2 mg g⁻¹ DW) was higher than the value reported by Kizil et al. [52], who noted that the leaf P concentration in *Mentha piperita* was 1.1 mg g⁻¹ DW. The similar values of leaf P concentration in *S. sclarea* may be because the levels of drought essayed did not been too high to modify the nutritional status of this species.

No differences in leaf K concentrations in all the species studied in our experiment may be ascribed as it was already mentioned that the different species were subjected to lower drought levels to result in variations of leaf K concentration. The results recorded in our experiment were contradictory to the results reported by Bahreininejad et al. [41], who noted a reduction of leaf K concentration when *Thymus daenensis* plants were grown under drought conditions (20, 50 and 80% soil water depletion). Similarly, Khalid et al. [53] reported that other Lamiaceae species such as *Ocimum basilicum* and *Ocimum americanum* grown under water stress conditions showed a decline in leaf K concentration. The reduction in leaf K concentration under drought stress conditions may be explained by the fact that the water scarcity affects stomatal regulation, resulting in reduced photosynthetic capacity and also the uptake of K ions in order to retain and regulate turgidity and stomatal control as reported by Sarani et al. [54].

The results achieved in this experiment from an economical point of view have reported that the cost of production in € per ha is 8523 €, without including the cost of the water applied (variable cost), therefore the viability of the cultivation of the aromatic species will be dependent mainly on the benefits obtained throughout the cultivation period, such as the number of harvestings per year, yield obtained, and the duration of the crop. The price of the water is variable between different areas since in some cases there is a cost only related with the pumping costs whereas in other cases it is necessary to include the price of alternative water, such as the desalinated water, with an average cost ranging from 0.30–0.80 € per m³ of water supplied. The saving of water without differences in yields between water treatments means a reduction of 3890 m³ per ha, which involves a saving of 3100 € approximately considering the highest price of the desalinated water, which may be crucial in order to

determine the economic viability of this crop under water deficit conditions. In addition, this water saving is crucial especially in arid areas such as the Almeria region.

5. Conclusions

Drought stress triggered different responses in the Lamiaceae species was studied. At the end of the experimental period, only *L. latifolia*, *M. piperita* and *T. capitatus* plants reduced the fresh dry weight, and with respect to dry weight, only *L. latifolia* plants. The essential oil content only decreased in *L. latifolia* and *S. sclarea* plants under water deficit conditions. Leaf nutrient concentrations showed different trends between species considering the nutrient assessed. From an economic point of view, the growth of the species studied can be feasible when the benefits are higher than the costs (fixed and variables) and the benefits will be dependent of the yield production obtained and the price accorded for both raw material and essential oil extracted. Finally, considering the results obtained, we recommended the growth of *M. piperita*, *S. lavandulifolia*, *T. capitatus*, and *T. mastichina* plants because there are no differences in essential content but if there is a water-saving rate of 30%. Nevertheless, in the case of *L. latifolia* and *S. sclarea* plants, the reduction of the water volume supplied for their growth should be carried out according to the price of the essential oil.

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Abbreviations

The following abbreviations are used in this manuscript:

DW	Dry weight
EC	Electrical conductivity
ETo	Evapotranspiration demand
FW	Fresh weight
K	Potassium
MD	Medium drought
N	Nitrogen
P	Phosphorous
R	Rainfall
SD	Severe drought

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