Original Article

Investigating the Effect of Drought Stress and Vermicompost Biofertilizer on Morphological and Biochemical Characteristics of *Thymus vulgaris L.*

Ghodrat alla Sharafi[1], Mahdi Changizi [2]*, Masoud Rafiee[3], Masoud Gomarian[2], Shahab Khagani[2]

[1] Department of Medicinal plants, Faculty of Agriculture, Islamic Azad University, Arak Branch, Arak, Iran. [2] Department of Agronomy and plant Breeding, Faculty of Agriculture, Islamic Azad University, Arak Branch, Arak, Iran. [3]Seed and Plant Improvement Research Department, Lorestan Agricultural and Natural Resources Research and Education Center, AREO, Khorramabad, Iran.

Abstract

This experiment was conducted to investigate the effect of drought stress and vermicompost biofertilizer on morphological and biochemical characteristics of Thymus vulgaris. It was done under water stress conditions in a greenhouse in Khorramabad Agricultural and Natural Resources Research Center. A factorial trial in a completely randomized design was carried out in three replications in a pot experiment. The factors including vermicompost fertilizer levels at 0, 30, 50, and 75% of pot volume and water stress including irrigation after depleting (85%) , 70% (mild stress), and 55% (severe stress) of moisture content of the field capacity were considered. The results showed that the morphological traits .including plant height, number of branches, plant fresh weight, plant dry weight and root length, were decreased with drought stress increase. Vermicompost consumption increased the morphological traits of the plant. Drought stress reduced the amount of Cymene in the air; but, at all levels of moisture stress, with increasing vermicompost consumption, the amount of Cymene was increased. The amount of secondary metabolites of Alfa-terpinen, Gamma-terpinen, thymol and carvacrol was decreased significantly with increasing drought stress. The amount of secondary metabolites and essential oil was increased significantly with increasing vermicompost consumption. The secondary metabolites were measured using GC MASS. According to the results of this study, severe stress in *Thymus vulgaris L* reduced growth and decreased shoot and morphological levels, which may be due to the plant's efforts to survive and achieve moisture. The secondary metabolites were decreased due to severe stress and improved with decreasing stress intensity due to increased shoots and increased metabolite production. Vermicompost biofertilizers increased the production of secondary metabolites by compensating for drought stress and gradually releasing the necessary plant elements.

Keywords: Drought stress, Morphological traits, Secondary metabolites, Thymol, Carvacrol

INTRODUCTION

There are different reports on Thymus species; but, considering the least amount of morphological diversity, 215 species of this species have been reported by Morales (2002) [1]. Thyme is widely used throughout the world for its aroma and medicinal properties. The presence of secretory glands on the leaves and flowers surface is the main cause of the aroma and medicinal properties of the plant. Thyme is native to the Mediterranean countries and is sometimes found in the wild. This plant grows on slopes between rocks in different Mediterranean regions, especially in France, Portugal, Spain, Italy, Greece and some parts of Asia [2]. Thyme essential oil due to its high content of thymol and carvacrol has the highest antioxidant properties and can be used as natural antioxidants [3]. Currently, the pharmaceutical industry in a number of Western countries has manufactured numerous drugs from the active ingredients of this plant and marketed them to the pharmaceutical market [4].

One of the factors that increase the secondary metabolites in medicinal plants including thyme is moisture stress. Any soil moisture deficiency will further reduce cell inflammation, division, and development, especially in shoots and leaves [5].

Address for correspondence: Mr. Mahdi Changizi. Department of Agronomy and plant Breeding, Faculty of Agriculture, Islamic Azad University, Arak Branch, Arak, Iran. E-mail: mchangizi47 @ gmail.com

How to cite this article: Sharafi, Gh. A., Changizi, M., Rafiee, M., Gomarian, M., Khagani, Sh. Investigating the Effect of Drought Stress and Vermicompost Biofertilizer on Morphological and Biochemical Characteristics of *Thymus vulgaris L.* Arch Pharma Pract 2019;10(3):137-45.

Therefore, the first noticeable effect of water deficit on Thyme can be identified by decreasing plant height, shrinkage of leaves and decreasing number of shoots [6, 7] . Maleki Lajier (2011) evaluated ornamental potential, drought tolerance (irrigation intervals of 8, 16 and 24 days) and freezing (open air conditions and freezing chamber) of some Thyme species (*Thymus kotschyanus*, *T. sepyllum* and *T. vulgaris)* and an endemic species (*T. daenensis*) for cultivation in green space as cover plants [8]. The researchers attributed the decrease in growth factors, water potential, division, cell enlargement, and stomatal closure to the 10-day treatment compared to the control, which ultimately led to reduced growth. Decrease of essential oil yield due to decrease in soil moisture can be due to the detrimental effect of drought stress on growth and yield of vegetative body by Hassani and Omid Beigi (2003) [9]; Rafat and Saleh (1997) [10] Basil; Letchamo et al. (1994) [11] Thyme; and Salehi et al. (2011) chamomile. Hosseini et al. (2016) [12] in the study of morphological, physiological and secondary metabolites of thymol and carvacrol in thyme under different salinity levels (control, 50, 100, 150 mM) found that salinity on plant height, number of lateral stems, weight of fresh and dry vegetative organs, fresh and dry root weight, root length, leaf length and width had significant effects. Increasing salinity levels increases the amount of sodium, ionic leakage and proline in plant tissues and on the other hand decreases the amount of potassium, relative water content, chlorophyll and carotenoid. Thymol and carvacrol, the most important constituents of this plant, were increased with increasing salinity up to 100 mM NaCl compared to the control.

The use of biofertilizers is very important for improving the quantitative and qualitative of performance of medicinal plants. valdabadi and Aliabadi Farahani (2011) showed that biofertilizers had a positive and significant effect on the quantitative and qualitative traits of black seed [13]. Valadabadi et al. (2001) concluded that seed inoculation with biofertilizers increased the uptake of nutrients by the root and thereby increased growth factors. Monjanata et al. (2002) concluded in an experiment that the effect of biofertilizers on growth, yield and essential oil content of medicinal plants was significant, improving their medicinal properties as well as aromatic compounds in plants [14].

A study by Seghatoleslami (2013) showed that the effect of biofertilizer on height, number of shoots per plant, seed oil percentage and germination percentage was not significant [15]; while, manure had significant effect on these traits. Shalan et al. (2005) showed that the application of biofertilizers such as pseudomonas aeruginosa increased the growth factors in medicinal plants, which is favorable for bacterial activity due to unfavorable soil conditions (pH, EC and texture) [16]. Zahir et al. (2004) showed that regulation of growth hormones was induced by the effects of biofertilizers modifies plant growth [17]. Carlydac et al. (2007) stated that the use of biofertilizers was a factor in reducing chemical fertilizers and as a result of this reduction, negative environmental effects would be eliminated [18].

Yeganehpour et al. (2015) in a study of the effect of drought stress and biofertilizer on yield of coriander showed that dehydration decreases, and in contrast, biofertilizer neutralizes the effect of stress and equilibrium or increases seed yield. Yeganehpour et al. (2017) demonstrated the effective role of biofertilizers in stress conditions to improve the quality and composition of oil in coriander [19]. Yeganehpour et al. (2016) showed that incorporation of the biological, chemical and salicylic acid fertilizer increased the number of umbrella in coriander because they were effective in improving morphological and physiological parts of this plant [20]. The researchers also found that drought would reduce number of umbrellas in coriander. Jafarzadeh et al. (2014) studied the effect of drought stress and nitrogen biofertilizer on evergreen medicinal plant showing that plant height, flower yield and some biochemical properties are affected by the interaction of treatments [21]. Yeganehpour et al. (2017) showed that application of biofertilizers and drought stress improved the quality of essential oil components and constituent oils (tyrocellicic acid, oleic acid, linoleic acid, palmitic acid, stearic acid, lenolenic acid, etc.) [19]. Yeganehpour (2016) showed that with increasing drought due to increased irrigation intervals, yield components and yield of coriander were decreased, but the biofertilizer use compensated for this decrease [20]. A wide variety of research has been done on the effects of drought stress and vermicompost biofertilizer on different plants, but comprehensive research has not been done on the effect of these factors on thyme in the region, which is new. The use of biofertilizers is very important for improving the quantitative and qualitative performance of medicinal plants and on the other hand, due to the reduction of water in most parts of the world in the production of medicinal plants, the effect of water scarcity on the quantitative and qualitative performance of medicinal plants, especially medicinal plants.

Recently, due to the side effects of chemical drugs, special attention has been paid to medicinal plants. Therefore, in this study, the effect of vermicompost biofertilizers on the quantitative, qualitative, morphological and biochemical characteristics of *Thymus vulgaris* under drought stress was investigated in order to reduce the negative effects of vermicompost and its effects on drought stress. It also determined the amount of drought that would increase production of secondary metabolites. The purpose of this study was to evaluate the effect of vermicompost and drought stress on morphological and biochemical characteristics of *Thymus vulgaris*.

Materials and Methods

The experiment was conducted in the spring of 2018 in Khorramabad with 33 degrees 20 minutes north latitude, 48 degrees 18 minutes east longitude and 1171 meters above sea level. Khorramabad has a subtropical climate with hot and dry summers. According to the long-term average minimum, maximum and average temperatures are 9.2, 25.2 and 17.2 ° C, respectively, with a mild climate with an average annual rainfall of 525 mm and an average relative humidity of 46.7%.

The experiment was conducted as a factorial experiment in a completely randomized design with three replicates as a pot experiment. Factors include vermicompost fertilizer rate factor: 0 (V1: control), 30 (V2), 50 (V3) and 70 (V4) percent pot volume, and drought stress after irrigation after 85 discharge (D1: control, conditions). Normal moisture), 70 (D2: moderate stress), and 55 (D3: severe stress) were the percentages of field moisture content.

Table 1- The characteristics of the vermicompost used

Cu	Zn	Mn	Fe	Mg	Ca	K	P	N	EC	**PH**
			Mg/kg						ds/m	
30	125	650	1500	1200	33000	4400	4230	6600	4.9	**8.3**

In this study, soil nutrients and vermicompost were measured prior to the experiment. Seeds were sown in plastic pots with a span diameter of 20 cm and a height of 15 cm. The soil was first sterilized at 121 ° C for 1.5 h and the pots were disinfected with alcohol after washing with ordinary water. After preparing the pots and sterilizing the pots, according to the different treatments, seed pits were created and then the seeds were sown. Twenty seeds were planted in each pot, and after seed germination and emergence, the plants were thinned out in several stages, eventually reaching 8 in each pot. Until one month after planting (6-8 foliage planting), the pots were irrigated with equal amounts and from this stage onwards irrigation stresses began.

Morphological traits including plant height, branch number, plant fresh weight, plant dry weight and root length and biochemical traits including secondary metabolites of cymene, alpha-terpinene, gama- terpinene, thymol, carvacrol, and essential oil content were measured. Morphological traits including plant height (with ruler), number of branches, fresh weight of plant (using digital scale), plant dry weight (using digital scale), and root length (with ruler) were measured.

To determine the secondary metabolites of essential oils from GC Agilent 7890 A and MASS model 5975 C MSD with 70V electron ionization voltage equipped with HP-5 column 30 m long and 0.25 mm internal diameter and 0.25 mm thick were used. The temperature planning meter was adjusted from 50 to 240 by increasing the temperature to 8 ° C / min and the temperature of the injection chamber was 220. It should be noted that a library search was conducted at two Wiley sources, NIST. Helium gas was used as carrier gas at a rate of 1 ml / min and was injected using Splitless method. The amount and percentage of essential oils were measured using a mill, mixer, celevenger unit, apparatus and laboratory equipment. Initially, the dry thyme was milled and poured into the balloon with water. 100 grams of dry thyme and 3 liters of water were used in a 5 liter balloon. This method turned on the device and then cooled the tap to flow around the condenser. When the water and thyme began to boil, the vapor moved to the condenser, where it was cooled, and the resulting sweat, which is the liquid vapor with the essential oil, was poured into the pipette. It was poured through the back tube into the balloon but because of the essential oil and water (sweat) phase, the essential oil remained in the pipette. When the valve was finished, the pipette was opened and after extracting the water (sweat), the essential oil was poured into a special jar.

Analysis of variance was performed using SAS software and mean comparisons were made by LSD at 5% probability level. Charts were drawn using EXCEL software.

Results and Discussion

The effect of drought stress was significant on all morphological traits except root length. The effect of vermicompost on all morphological traits was significant. Interaction of drought stress and vermicompost fertilizer on all measured morphological traits was not significant.

Bush height

The simple effects of drought and vermicompost on plant height in Thyme were significant, but this effect was not affected by the interaction effect of drought in vermicompost (Table 2).

Table 2- Analysis of variance of morphological traits of Thymus vulgaris

The source of changes	degree of freedom	mean square Plant height	Number of branches	Fresh weight of plant	Dry weight of plant	**Length of roo**
Drought stress	2	18.91*	4.22*	8.37**	0.793**	**0.0741**
Vermicompost	3	100.4**	12.37**	3.77**	0.544**	**13.85****
Drought stress; vermicompost	6	8.576601	1.554695	1.087485	0.144663	**0.356774**
Error	24	4.919397	1.583611	0.607933	0.082114	**1.062714**
Coefficient of variation (%)		**8.04**	**10.33**	**11.9**	**12.7**	**8.34**

* And ** were significant at the 5% and 1% levels, respectively.

Plant height was decreased significantly with increasing drought stress and increased from 28.8 cm under normal moisture to 26.3 cm under severe drought stress. Plant height was increased significantly with increasing vermicompost consumption and reached its maximum value (31.9 cm) with 75% volume (23.9 cm) under non-use conditions (Figure 1).

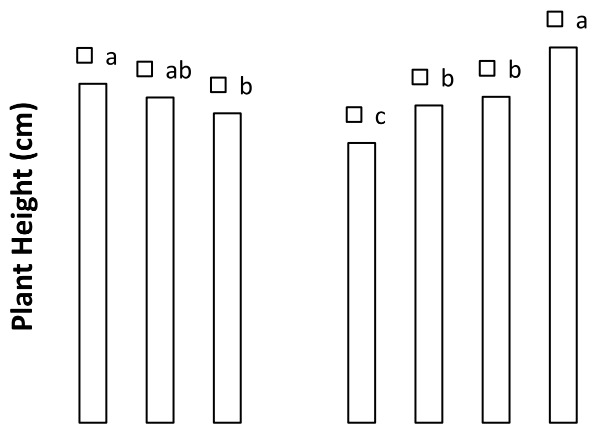

Figure 1- Comparison of the average effects of drought stress and vermicompost on plant height

Number of branches

The simple effects of drought stress and vermicompost on the number of branches per plant were significant, but this effect was not affected by the interaction effect of drought stress on vermicompost (Table 2).

The number of branches was decreased significantly with increasing drought stress and increased from 12.8 in normal to 11.6 in severe drought stress. The number of branches was increased significantly with increasing vermicompost consumption and reached its maximum value (13.8 branches) with 75% volume consumption (10.8) (10.8) (Fig. 2).

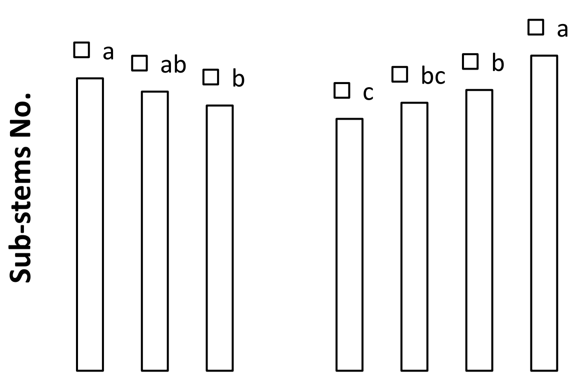

Figure 2- Comparison of the average effects of drought stress and vermicompost on the number of branches

Plant fresh weight

The simple effects of drought and vermicompost on fresh weight of Thymus vulgaris were significant, but this effect was not affected by the interaction effect of drought stress on vermicompost (Table 2).

Plant fresh weight was decreased significantly with increasing drought stress and reached 5.59 g under severe drought stress under normal moisture conditions. Plant fresh weight was increased significantly with increasing vermicompost consumption and reached its maximum value (7.35 g) with 75% volumetricity (5.35 g) under non-use conditions (Figure 3).

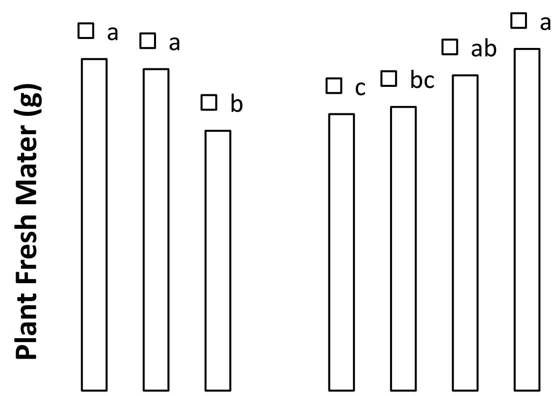

Figure 3- Comparison of the average effects of drought stress and vermicompost on plant fresh weight

Plant dry weight

Simple effects of drought and vermicompost on plant dry weight were significant in Thyme, but this effect was not affected by drought stress interaction in vermicompost (Table 2).

Plant dry weight was decreased significantly with increasing drought stress and increased from vermicompost consumption to 2.95 g under normal moisture conditions to 1.95 g under severe drought stress. 2 g in the non-consuming condition reached its maximum value (2.54 g) with 75% volumetric consumption (Figure 4).

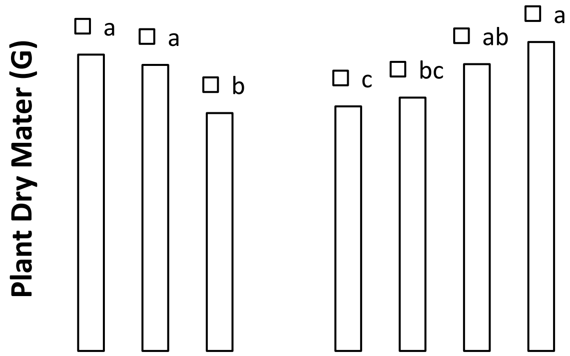

Figure 4- Comparison of the average effects of drought stress and vermicompost on plant dry weight

Root length

Only the simple effect of vermicompost on root length in Thyme was significant, but this trait was not affected by the simple effect of drought stress and interaction effect on vermicompost (Table 2).

Plant height was increased significantly with increasing vermicompost consumption and reached its maximum value (0.14cm) with 75% volumetricity (0.14cm) under non-application conditions (Figure 5).

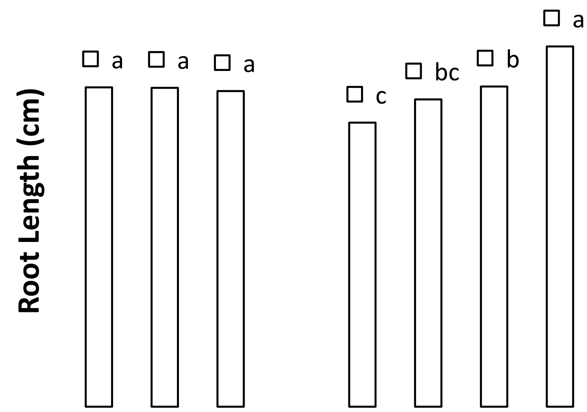

Figure 5- Comparison of the average effects of drought and vermicompost on root length

Thyme secondary metabolites

The effect of drought stress on all secondary metabolites and essential oil percentage was not significant. The effect of vermicompost on all secondary metabolites and essential oil percentage was significant. The interaction between drought stress and vermicompost fertilizer was not significant on cement and on other metabolites and essential oil percent.

Table 3- Analysis of variance of Thymus vulgaris secondary metabolites

		mean square					
The source of changes	degree of freedom	Symene	Alfaterpinen	Gamma-terpinen	thymol	carvacrol	**essential oil**
Drought stress	2	92.64**	10.15**	14.05**	1113.8**	15.2**	**0.017099**
Vermicompost	3	12.1**	2.90**	2.55**	192.8**	4.26**	**0.16****
Drought stress; vermicompost	6	1.08**	0.098498	0.17433	5.40938	0.181668	**0.016011**
Error	24	0.2389	0.041422	0.167289	3.977422	0.375933	**0.007594**
Coefficient of variation (%)		**8.855**	**9.86**	**12.4**	**3.84**	**8.74**	**11.4**

* And ** were significant at the 5% and 1% levels, respectively.

Symene

The simple effects of drought and vermicompost stress and the interaction effect of drought stress on vermicompost on cymene content of thyme were significant (Table 3). At all levels of moisture stress, the increase in vermicompost application increased the amount of aerial cymene. In normal humidity conditions, the highest and the lowest amount of cymene were obtained from 75% and non vermicompost application (9.97 and 6.77, respectively). In moderate drought stress conditions, maximum and minimum cymene content was obtained from 75% volumetric and no vermicompost application (4.62 and 8.23, respectively). In severe drought conditions, the highest and lowest cymene content was obtained from 75% volumetric and non vermicompost consumption (1.87 and 3.13, respectively) (Figure 6). Overall, drought stress reduced the amount of aerial cymene.

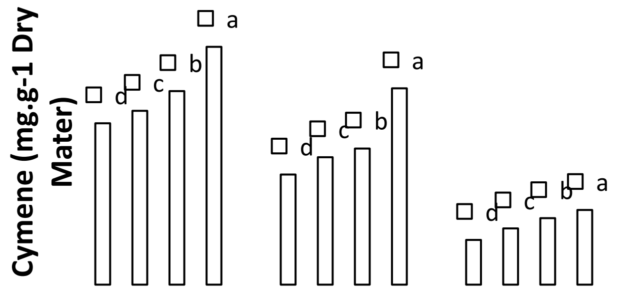

Figure 6- Interaction of drought stress in vermicompost on cymene content in Thyme

Alfaterpinen

The simple effects of drought stress and vermicompost on alpha-terpinene content in Thyme were significant but this effect was not affected by the interaction effect of drought stress on vermicompost (Table 3).

The concentration of alpha-terpinene was decreased significantly with increasing drought stress and reached to 1.21 in severe drought stress from 3.04 in normal moisture conditions. The alpha-terpinene content was increased significantly with increasing vermicompost consumption. The highest and lowest levels of this secondary metabolite were obtained from 75% volumetric consumption and non-use of vermicompost (2.72 and 1.47, respectively) (Figure 7).

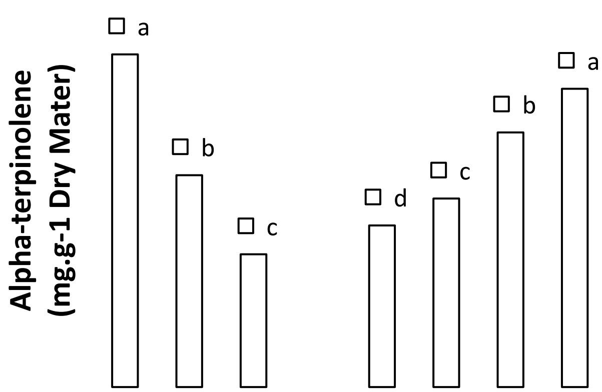

Figure 7- Simple effect of drought and vermicompost stress on alpha-terpinen content in Thyme

Gamma-terpinen

The simple effects of drought stress and vermicompost on the amount of gamma-terpinen in Thyme were significant but this effect was not affected by the interaction effect of drought stress on vermicompost (Table 3).

The gamma-terpinen content was decreased significantly with increasing drought stress and increased from 4.33 in normal to 2.17 in severe drought. Gametarpinene content was increased significantly with increasing vermicompost consumption. The highest and lowest levels of this secondary metabolite were obtained from 75% volumetric consumption and no vermicompost consumption (3.94 and 2.68, respectively) (Figure 8).

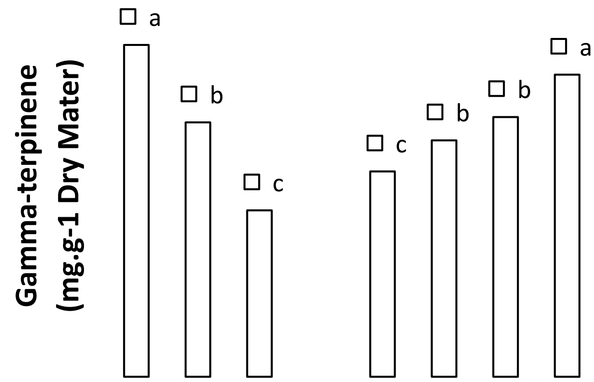

Figure 8- Simple effect of drought stress and vermicompost on gama-tarpinen content in Thyme

Thymol

The simple effects of drought stress and vermicompost on thymol content in Thyme were significant but this effect was not affected by the interaction effect of drought stress on vermicompost (Table3).

Thymol was decreased significantly with increasing drought stress and increased from 77.4 in normal to 59.3 in severe drought. Thymol was increased significantly with increasing vermicompost consumption. The highest and lowest levels of this secondary metabolite were obtained from 75% volumetric and non-vermicompost consumption (71/69 and 61/18, respectively) (Figure 9).

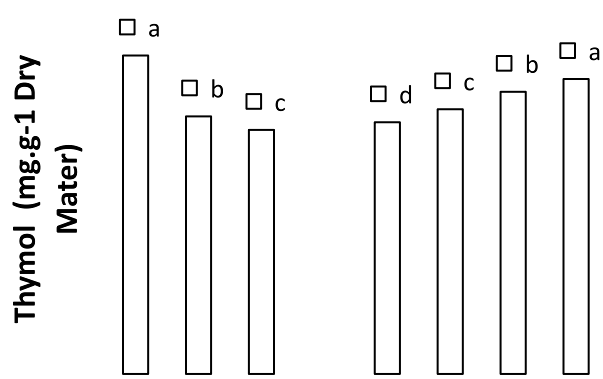

Figure 9- Simple effect of drought stress and vermicompost on thymol content in Thyme

Carvacrol

The simple effects of drought stress and vermicompost on carvacrol content in Thyme were significant but this effect was not affected by the interaction effect of drought stress on vermicompost (Table 3).

Carvacrol was decreased significantly with increasing drought stress and reached 5.97 in severe drought stress from 8.22 to normal conditions. Carvacrol was increased significantly with increasing vermicompost consumption. The highest and lowest levels of this secondary metabolite were obtained from 75% volumetric consumption and non-use of vermicompost (7.94 and 6.37, respectively) (Figure 10).

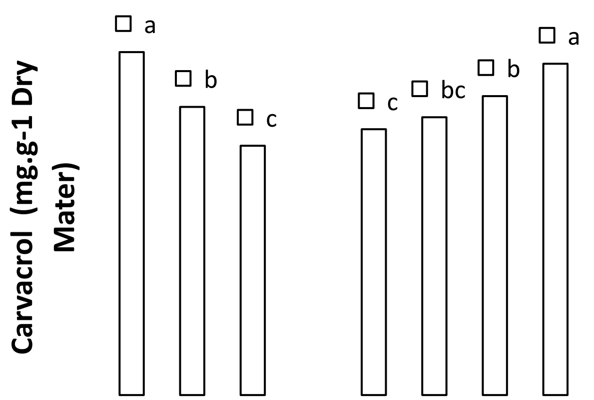

Figure 10- Simple effect of drought stress and vermicompost on carvacrol content in Thyme

Essential oil percentage

Only the simple effect of vermicompost on the essential oil content of thyme was significant, but this effect was not affected by the simple effect of drought stress and interaction effect of vermicompost (Table 3).

The percentage of essential oil was increased significantly with increasing vermicompost consumption. The highest and lowest levels of this secondary metabolite were obtained from 75% volumetric consumption and non-use of vermicompost (0.58 and 0.86, respectively) (Figure 11).

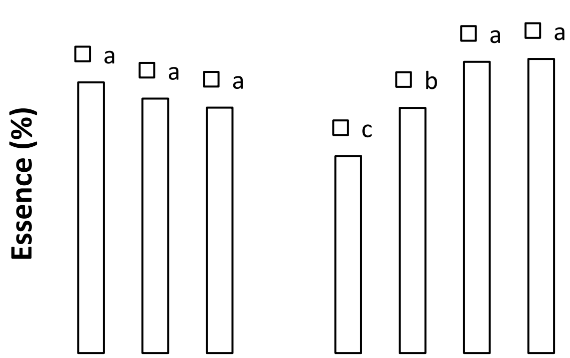

Figure 11- Simple effect of drought stress and vermicompost on essential oil percent in Thyme

Morphological characteristics of Thyme

The morphological traits of shoot and root of thyme including plant height, branch number, fresh plant weight, plant dry weight and root length were decreased with increasing drought stress. Vermicompost application increased morphological traits of the plant.

Decrease in plant height due to drought stress was reported by Zounemat Kermani and Asadi (1979) [22]; Khoshkhoi et al. (2012) [6]; and Erkusa et al. (2002) [7] in Thyme. Reducing cell division and expansion decreases plant growth and plant height. Vermicompost has a positive effect on photosynthesis and biomass production of chamomile and improves plant height through high water uptake and optimum availability of nutrients and nutrients.

Water deficit stress decreases growth due to lower Turgor pressure (Hsia Tc ,Xu,2000). Hosseini et al. (2016) have reported a decrease in the fresh and dry weight of Thymus vulgaris. Abbas Alipour et al. (2007) in the study of effects of drought stress on some morphological traits of chamomile in Shirazi reported that drought stress decreased chamomile dry weight [23].

Vermicompost provides nutrients and absorbs more water by increasing the number of branches and subsequently by increasing the number of main stems and the number of flowers per plant [24]. According to Tahami-Zandari et al. (2010), under the same environmental conditions, the availability of nutrients to the plant by different fertilizers can increase the plant growth and consequently the number of branches. In another study, the application of 30 ton / ha of manure increased the branches of chamomile [25]. Maleki Lajier (2011) in the evaluation of drought tolerance of some Thyme species found that the fresh weight of foliage was decreased due to stress and the species responded differently. Abbas Alipour et al. (2007) in the study of effects of drought stress on some morphological traits of chamomile in Shirazi reported that drought stress decreased chamomile dry weight. Falahi (2004) reported the positive effect of animal fertilizers on dry weight of chamomile plant [26], Delat (2000) [27] increased dry weight of lemongrass as a compost application and Arancon et al. (2004) increased dry weight of pepper plant by increasing vermicompost levels [28].

Ashouchi and Sharifi (2004) applied 25, 50, 75, and 100% of field capacity on pistachio, yarrow, sage, chamomile, and spring chamomile irrigation treatments, which increased drought stress, shoot weight and plant height and the studied plants were decreased [29].

Symon et al. (1992) investigated the effect of different aquatic regimes on the basil plant and reported that by decreasing the leaf water potential from -0.3 MPa (control) to -1.12 MPa (moderate water stress), the leaf essential oil content was increased from 3.1 to 6.2 µl / g dry leaf weight and leaf and stem dry weight was decreased with increasing water scarcity [30].

The decrease in the number of lateral branches due to water stress observed in this study has also been reported by Hosni and Omidibigi (2002) and Aslani et al. (2011) [31] in Basil, and Hassani (2006) in Badrshabu. Similar results have been obtained for increasing essential oil percentage by drought stress by Letchamow et al. (1994) and Hassani and Omidibigi (2002).

In another study on garlic (Allium sativum), the use of vermicompost significantly improved plant height [32]. In the research of Darzi et al. (2008) [33]

In a study, it was observed that the use of compost and vermicompost in potato (Solanum tuberosum L.) significantly increased the number of main and secondary branches per plant [34]. The effect of different types of organic and bio-fertilizers on increasing the number of main and sub-branches of the fennel (Foeniculum Vulgare L.) has also been reported to be significant [35].

Falahi (2004) [26].reported the positive effect of animal fertilizers on dry weight increase of chamomile (Chamaemelum nobile L.), Delat (2000) [28] increased dry weight of Melissa officinalis L., Compost and Arancon et al. (2004) [28]. dry weight of aerial parts of pepper (Capsicum annum L.) with increasing vermicompost levels.

Secondary metabolites

The results of this study showed that drought stress reduced shoots cymen content but at all levels of moisture stress, it increased shoot cement content by increasing vermicompost consumption. Secondary metabolites of alpha-terpinen, gamma terpinen, thymol and carvacrol were decreased significantly with increasing drought stress. Secondary metabolites of alpha-terpinen, gamma terpinen and essential oil content were increased significantly with increasing vermicompost consumption.

These results confirm that the greater use of vermicompost due to improved water storage capacity and balanced nutrient uptake has increased the rate of secondary metabolites in Thyme plant.

The production of secondary metabolites in medicinal plants is genetically controlled, but environmental factors, especially stressful conditions, play a major role in the quantity and quality of these substances.

Symon et al. (1992) investigated the effect of different aquatic regimes on the basil plant and reported that by decreasing the leaf water potential from -0.3 MPa (control) to -1.12 MPa (moderate water stress) leaf essential oil content was increased from 3.1 to 6.2 µl / g dry leaf weight and leaf and stem dry weight was decreased with increasing water scarcity.

One of the essential factors for success in cultivation of medicinal plants is manure management. Proper use of nutrients and nutrients not only plays a key role in enhancing the performance of medicinal plants, but also in improving the quantity and quality of the effective ingredients of the product produced [36].

There was an inverse correlation between dry leaf yield and essential oil percentage of basil, as the leaf yield per unit area was decreased and the control treatment had the highest essential oil percentage with lowest leaf yield per hectare. Similar results have been reported from the experiment of Tahami-Zarandi (2000) on basil of fennel. Another point is that the combined use of nitroxin, biophosphorus, and vermicompost, as well as other treatments containing vermicompost, had the highest essential oil content, even though they had the highest yield, indicating a positive effect on vermicompost. The increase was related to the essential oil of basil [37] Other research has also reported that the use of organic fertilizers increases the percentage of essential oils of medicinal plants [34].

Moradi (2004) reported that the use of organic and biological fertilizers significantly increased the yield of fennel essential oil compared to the control [35]. For example, the use of compost and vermicompost increased the essential oil yield by 49% compared to the control. He attributed the increase to higher seed yield and essential oil percentage than other treatments.

Ahmadian et al. (2009) in the study of the interaction between drought stress and manure application on the quantitative and qualitative characteristics of cumin, reported that consumption of 20 ton / ha of animal manure, while reducing the negative effects of drought stress, increased the amount of effective substance and improved the essential oil quality characteristics [36]. Cumin turned green and replaced by more irrigation at seed filling stage.

References

1. Morales R. The history, botany and taxonomy of the genus Thymus. Taylor and Francis, London; 2002 Sep 5.
2. Jamzad Z. Thymus and Satureja species of Iran. Research Institute of Forest and Rangelands, Tehran, Iran. 2009.
3. Mehran M, Hoseini H, Hatami A, Taghizade M. Investigation of components of seven Species of Thyme Essential oils and comparison of their antioxidant properties. Journal of Medicinal Plants. 2016 May 15;2(58):134-40.
4. Omidbaigi R. Production and processing of medicinal plants. Publications Astan Quds Razavi, Mashhad, 2005 : 438p [In Persian].
5. Sreevalli Y, Baskaran K, Chandrashekara RS, Kulkarni RN. Preliminary observations on the effect of irrigation frequency and genotypes on yield and alkaloid concentration in periwinkle. Preliminary observations on the effect of irrigation frequency and genotypes on yield and alkaloid concentration in periwinkle.. 2000;22(4a):356-8.
6. Khosh-Khui M, Ashiri F, Saharkhiz MJ. Effects of irrigation regimes on antioxidant activity and total phenolic content of thyme (Thymus vulgaris L.). Medicinal & Aromatic Plants. 2012;1(7):1-7.
7. Erkossa T, Stahr K, Tabor G. Integration of organic and inorganic fertilizers, effect on vegetable productivity. Rural poverty reduction through research for development and transformation. Deutscher Tropenttag. 2004.

8. Maleki Lajier, H. Evaluation of drought tolerance and frost tolerance of some Thymus spp species in green space. Ph.D. Thesis. Faculty of Agricultural Sciences. University of Guilan, 2011.
9. Hassani A, Omid Beigi, R, Heidari Sharifabad, H,. Investigation of Some Drought Resistance Indices in Basil. Gorgan Journal of Agricultural Sciences and Natural Resources. 2003; J. 10, p. 74-65.
10. Refaat AM, Saleh MM. The combined effect of irrigation intervals and foliar nutrition on sweet basil plants. Bulletin of Faculty of Agriculture, Cairo Univ.(Egypt). 1997.
11. Lechamo W, Marquard R, Hölzl J, Gosselin A. Effects of water supply and lighht intensity on growth and essential oil of two Thymus vulgaris selections. Angewandte Botanik. 19942003;68(3-4):83-8.
12. Hosseini, H., Mousavi, F.S., Fatehi, F., Qaderi, A. Changes in Phytochemical and Morpho-Physilogical Traits of Thyme (Thymus Vulgaris cv Varico 3) Under Different Salinity Levels. Journal of Medicinal Plants, 2017 ; 16(1): 34-22.
13. Valadabadi SA, Farahani HA. Investigation of biofertilizers influence on quantity and quality characteristics in Nigella sativa L. Journal of Horticulture and Forestry. 2011 Mar;3(3):88-92.
14. Manjunatha R, Farooqi AA, Vasundhara M, Srinivasappa KN. Effect of biofertilizers on growth, yield and essential oil content in patchouli (Pogostemon cablin Pellet.). Indian perfumer. 2002;46(2):97-104.
15. Seghatoleslami M. Effect of water stress, bio-fertilizer and manure on seed and essential oil yield and some morphological traits of cumin. Bulgarian Journal of Agricultural Science. 2013;19(6):1268-74.
16. Shaalan MN. Influence of biofertilizers and chicken manure on growth, yield and seeds quality of (Nigella sativa L.) plants. Egyptian Journal of Agricultural Research, 2005; 83: 811-828.
17. Zahir ZA, Arshad M, Frankenberger WT. Plant growth promoting rhizobacteria: applications and perspectives in agriculture. Advances in agronomy. 2004 Jan 1;81(1):98-169.
18. Karlidag H, Esitken A, Turan M, Sahin F. Effects of root inoculation of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrient element contents of leaves of apple. Scientia Horticulturae. 2007 Sep 11;114(1):16-20.
19. Yeganehpoor F, Zehtab-Salmasi S, Ghassemi-Golezani K, Shafagh-Kolvanagh J, Dastborhan S. The effect of water stress, salicylic acid and biofertilizer on quality of leaf and seed essential oil, and oil components of coriander. Net Journal of Agricultural Science. 2017;5(2):38-47.
20. Yeganehpour F. Zahhtab Salmasi S, Shaf Kalwang J. And Kazem Ghasemi Golzani K. Effect of drought stress, chemical and biological fertilizer and salicylic acid on seed yield and yield components in Coriander (Coriandrum sativum L.). Journal of Crop Production. 2016; 9 (4): 55-37.
21. Jafarzadeh L, Omidi H, Bostani A. The study of drought stress and Bio fertilizer of nitrogen on some biochemical traits of Marigold medicinal plant (Calendula officinalis L.). Journal of Plant Research (Iranian Journal of Biology). 2014 ; 27(2): 180-193.
22. Zounemat Kermani, M., Asadi R. Effect of Partial Root Zone Drying on the Quantitative Traits of (Thymus vulgaris L.) Journal of Water Research in Agriculture. 2018 ; 23 (1): 161-147.
23. Abbasalipour H. Houshmand S. Zeinali H. Tadayyon A. Effect of drought stress on some morphological characteristics of two genera of chamomile (Matricaria chamomile). 10th Congress of Agronomy and Plant Breeding, Tehran, University of Tehran Aboureihan, 2007.
24. Saidnejad A H, Rezvani Moghaddam PA. Effect of Biofertilizers and Chemical Fertilizers on Morphological Properties, Yield, Yield Components and Essence Percentage of Cumin (Cuminum cyminum). Journal of Horticultural Sciences (Agricultural Sciences and Industries). 2010 ; 24 (2): 148-142.
25. Jahan M, Koocheki A. Effect of organic production of german chamomile (Maricaria chamomilla L.) on it'; s chemical composition. In Agronomy and Horticulture. 2004 ; 61: 95-87.
26. Fallahi J. Effects of biological and chemical fertilizers on quality and quantity characteristics of chamomile. Msc Thesis, Faculty of Agriculture, Ferdowsi University of Mashhad, Iran. (In Persian with English Summary), 2004.
27. Delate K. Heenah mahyah student from herb trail. Leopold center for sustainable agriculture. Annual Reports, Jowa State University. Ames, IA. 2000.
28. Arancon NQ, Edwards CA, Bierman P, Welch C, Metzger JD. Influences of vermicomposts on field strawberries: 1. Effects on growth and yields. Bioresource technology. 2004 Jun 1;93(2):145-53.
29. Lebaschi MH, Sharifi AE. Growth indices of some medicinal plants under different water stresses. 2004 ; 20(3) : 269-241.
30. Simon JE, Reiss-Bubenheim D, Joly RJ, Charles DJ. Water stress-induced alterations in essential oil content and composition of sweet basil. Journal of Essential Oil Research. 1992 Jan 1;4(1):71-5.
31. Aslani Z, Hassani A, Sadaghiyani MR, Sefidkon F, Barin M. Effect of two fungi species of arbuscular mycorrhizal (Glomus mosseae and Glomus intraradices) on growth, chlorophyll contents and P concentration in basil (Ocimum basilicum L.) under drought stress conditions. Iranian Journal of Medicinal and Aromatic Plants. 2011;27(3):471-86.
32. Argüello JA, Ledesma A, Núñez SB, Rodríguez CH, Goldfarb MD. Vermicompost effects on bulbing dynamics, nonstructural carbohydrate content, yield, and quality ofRosado Paraguayo'garlic bulbs. Hortscience. 2006 Jun 1;41(3):589-92.
33. Darzi MT, Ghalavand A, Rejali F. Effect of mycorrhiza, vermicompost and phosphate biofertilizer application on flowering, biological yield and root colonization in fennel (Foeniculum vulgare Mill.). Iranian Journal of Crop Sciences, 2008 ; 10 (1): 109 - 88.
34. Atiyeh RM, Subler S, Edwards CA, Bachman G, Metzger JD, Shuster W. Effects of vermicomposts and composts on plant growth in horticultural container media and soil. Pedobiologia. 2000 Jan 1;44(5):579-90.
35. Moradi R. *Evaluation of biologic and organic fertilizers effects on grain yield, yield components and essence of fennel (Foeniculum vulgare)* (Doctoral dissertation, MSc dissertation, Faculty of Agriculture, Ferdowsi University of Mashhad, Iran. 2009.(in Persian)).
36. Ahmadian A, Ghanbari A, Galavi M, Siahsar B, Arazmjoo E. Effect of Different Irrigation and Fertilizer Regimes on Elemental Content, Essential Percentage and Chemical Compounds in Cumin. Iranian Journal of Crop Sciences, 2008 ; 40(1): 180-173.
37. Rezai moadab A,Nabavikalat CM,Sadrabadi hagigi R.Effect of vermicompost and biofertilizers on vegetable yield and essential oil of basil(*Ocomum basilicum* L.) in Mashhad.Juornal of Agricultural Ecology.2013.vul5..No 4.pp.350-362.