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Full Length Research Paper

Evaluation changing of essential oil of balm (*Melissa officinalis* L.) under water deficit stress conditions

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An experiment was carried out using a randomized complete block design with 4 replications to study the effect of water deficit stress on essential oil of balm. The factors studied included 5 levels of water deficit stress that were T_1 (100% field capacity), T_2 (80% field capacity), T_3 (60% field capacity), T_4 (40% field capacity) and T_5 (20% field capacity) respectively. The results showed that water deficit stress significantly affected biological yield, essential oil yield, essential oil %, leaf yield, height of plant, tiller number, stem diameter, stem yield and internode length in $P < 0.01$. Highest biological yield and height of plant were achieved under T_1, highest essential oil yield was achieved under T_3 and highest stem diameter and essential oil % were achieved under T_5. The results of this experiment showed that essential oil yield was reduced under water deficit stress but essential oil % increased under this condition.

Key words: Water deficit stress, essential oil, balm.

INTRODUCTION

Balm, *Melissa officinalis* L., a perennial herb native to southern climates of Europe and North America, is presently found in both wild and cultivated states. Several other species of *Melissa* have been reported from the Mediterranean and central Asian areas, but only *M. officinalis* L. is cultivated. The plant grows erect and reaches a height of 0.5 to 1 m. The highest levels of essential oil have been extracted in late summer from the lower parts of the plants (Kennedy et al., 2005). The essential oil contains geraniol, citronellol, cintronellal, linalool, eugenol acetate and nerol. The essential oil is often adulterated with mixtures of lemongrass, citronella or lemon oil. Oil of balm has also been shown to have antiviral, antibacterial and antispasmodic activity. Balm has been reported to be an insect repellent. As a medicinal plant, lemon balm has traditionally been employed against catarrh, fever (Kabala-Dzik et al, 1993), flatulence (Weizman et al., 1993) and headaches (Wake et al., 2000). Water deficit occurs when water potentials in the rhizosphere are sufficiently negative to reduce water availability to sub-optimal levels for plant growth and development (Aliabadi et al., 2008a). Drought stress is especially important in countries where crop agriculture is essentially rain-fed (Boyer, 1982; Ludlow and Muchow, 1990). Drought stress causes an increase in solute concentration in the environment, leading to an osmotic flow of water out of plant cells. This in turn causes the solute concentration inside plant cells to increase, thus lowering water potential and disrupting membranes along with essential processes like photosynthesis. These drought-stressed plants consequently exhibit poor growth and yield. In worst case scenarios, the plants completely die. Certain plants have devised mechanisms to survive under low water conditions. These mechanisms have been classified as tolerance, avoidance or escape (Kramer and Boyer, 1995; Neumann, 1995). Drought stress reduced dry matters of coriander by reduction in the area of the leaf, height of plant and lateral stem number (Aliabadi et al., 2008c). Droughted *M. officinalis* plants showed a significant decrease in the endogenous levels of photosynthetic pigments, the antioxidant function of α-tocopherol and the dissipation of excess excitation energy by the xanthophyll cycle may help to avoid irreversible damage to the photosynthetic apparatus that

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Table 1. Mean squares of some characteristics of balm.

Mean squares										
Essential oil yield	**Essential oil %**	**Stem yield**	**Leaf yield**	**Biological yield**	**Internode length**	**Stem diameter**	**Tiller number**	**Height of plant**	**df**	**Value Sources**
26.049	0.034	187357.18	3453762.1	11531168.779	2.352	0.851	56.191	197.831	4	Replication
2.158**	0.001**	38846.414**	323859.218**	137129.613**	0.186**	0.027**	0.149 **	7.97 **	3	Water deficit stress
0.842	0.001	10781.464	49149.644	18496.304	0.114	0.015	5.28	6.439	12	Error
10.65	9.13	4.74	7.56	2.68	7.38	3.32	4.85	4.43		CV (%)

** and * : Significant at 1 and 5% levels respectively.

decreased biological yield of balm (Munne-Bosch and Alegre, 1999). The results of a study showed that drought stress reduced flowering shoot yield, essential oil yield and internode length and increased essential oil % of coriander (Aliabadi Farahani et al., 2008b). Drought stress is the most important environmental factor that de-creases plants yield but in some medicinal plants increases essential oil. Therefore, this experiment was performed to study the effect of water deficit stress on essential oil of balm.

MATERIAL AND METHODS

This experiment was carried out using a randomized com-lete block design with 4 replications. The factors stu-died included 5 levels of water deficit stress that were T_1 (100% field capacity), T_2 (80% field capacity), T_3 (60% field capa-city), T_4 (40% field capacity) and T_5 (20% field capa-city) respectively. The field was prepared in an 8 m^2 area (4m × 2 m). In order to determine the soil moisture rate of 100% field capacity, 24 h after irrigation we selected 5 field soil samples by sampling drill, then samples were weighed by electrical scale and placed under 105°C in electrical oven for 48 h. Soil moisture rate of 100% field capacity was determined by the following formula.

$$\theta_w = \frac{\text{Moist soil weight-Dry soil weight}}{\text{Dry soil weight}}$$

We selected field soil samples for determination of soil moisture rate daily, than determined 80% field capacity, 60% field capacity, 40% field capacity and 20% field capa-city respectively. At the end of flowering stage, we selected 100 g flowering shoot from each plot for determination of essential oil % by Clevenger. Finally, essential oil yield determined by the following formula.

Essential oil yield = Essential oil percentage × Flowering shoot yield

At the end of growth stage for determination of some cha-racteristics of balm, we selected 10 plants from each plot and then determined biological yield, stem yield, height of plant, tiller number, leaf yield, stem diameter and internode length. Data were subjected to analysis of variance (ANOVA) using statistical analysis system and followed by Duncan's multiple range tests and terms were considered significant at $P < 0.05$ by MSTAT-C software.

RESULTS

The results showed that water deficit stress sig-nificantly affected biological yield, essential oil yield, essential oil %, leaf yield, height of plant, tiller number, stem diameter, stem yield and inter-node length in $P < 0.01$ (Table 1). Highest biolo-gical yield (6469 kg/ha), internode length (5.31 cm), leaf yield (3736 kg/ha), height of plant (65.32 cm) and stem yield (2733 kg/ha) were achieved under T_1, highest essential oil yield (12.97 kg/ha) was achieved under T_3 and highest essential oil % (0.3013%) and stem diameter (4.37 mm) were achieved under T_5 (Table 2) (Figures 1, 2 and 3).

DISCUSSION

As it was shown in the results of this study, drought stress had a negative effect on most of the morphological characteristics under study, but in terms of the essential oil %, it was completely different. This shows that in order to resist drought stress, the plant uses different ways. Great reduction in the length and width of the leaf and accordingly reduction in the area of the leaf, reduction in the height of plant and tiller number, all contribute to the reduction of plant's evapo-ration area and consequently reduction in the pro-duced dry matter is the final result of the reduction in the plant's photosynthesis which in turn, is the result of drought stress. Under drought stress, stomatals become blocked or half-blocked and this leads to a decrease in absorbing Co_2 and on the other hand, the plants consume a lot of energy to absorb water, these cause a reduction in producing photosynthetic matters.

Drought stress increased the amount of the %,

Table 2. Means comparison of some characteristics of balm.

Essential oil yield (kg/ha)	Essential oil (%)	Biological yield (kg/ha)	Stem yield (kg/ha)	Leaf yield (kg/ha)	Internode length (cm)	Stem diameter (mm)	Tiller number (n/plant)	Height of plant(cm)	Treatments
6.73 b	0.1041 c	6469 a	2733 a	3736 a	5.31 a	3.25 d	49.26 a	65.32 a	T_1
7.68 b	0.1274 c	6047 b	2600 b	3447 b	5.17 a	3.45 cd	50.38 a	61.51 ab	T_2
12.97 a	0.1456 b	5978 b	2537 b	3441 b	3.92 b	3.57 c	48.25 a	58.32 bc	T_3
8.71 ab	0.2813 ab	3094 c	2014 c	2583 c	3.85 b	4.05 b	48.35 a	54.63 c	T_4
3.97 c	0.3013 a	2319 d	1068 d	1464 d	3.57 b	4.37 a	40.91 b	45.92 d	T_5

Means within the same column and factors, followed by the same letter are not significantly difference ($P < 0.05$) using Duncan's multiple range test.

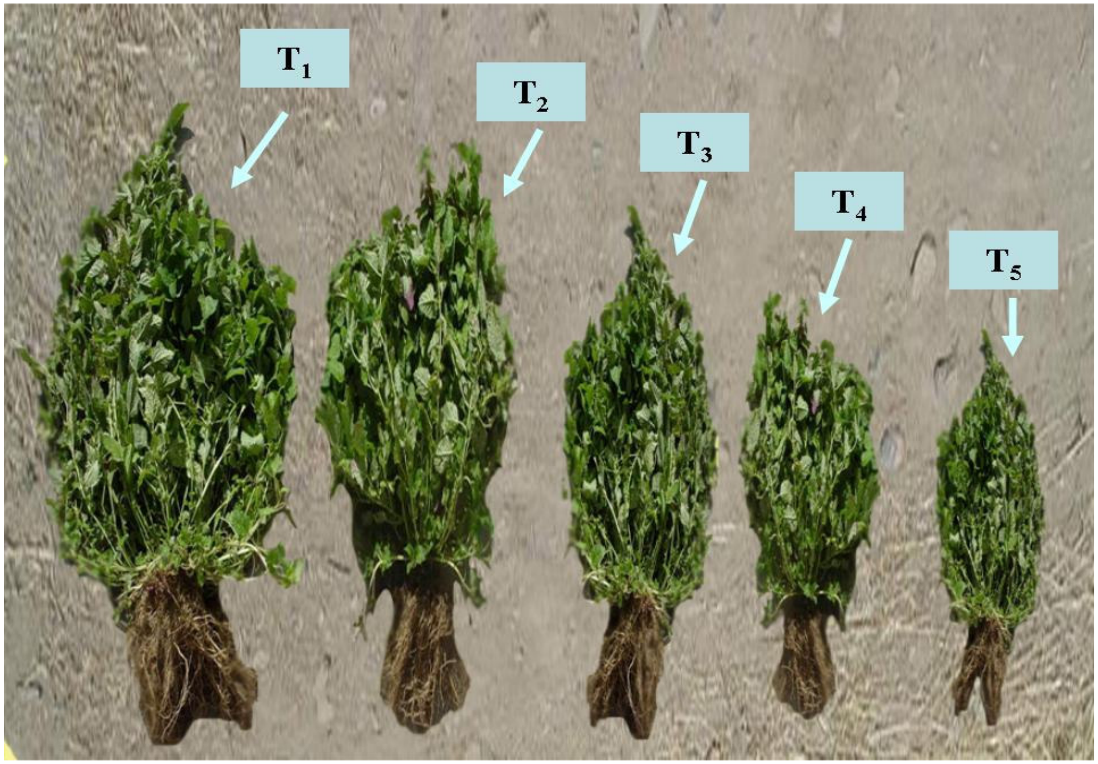

Figure 1. Height of plants under different levels of water deficit stress.

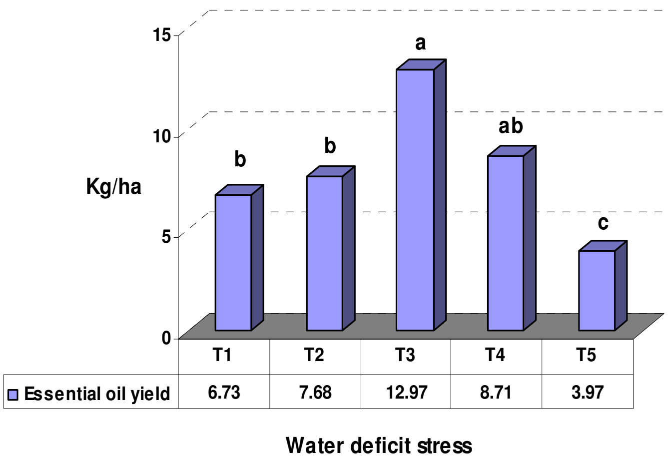

Figure 2. Essential oil yield under different levels of water deficit stress.

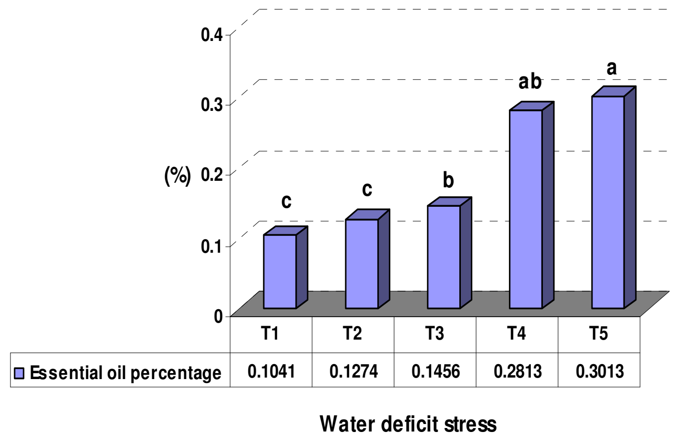

Figure 3. Essential oil percentage under different levels of water deficit stress.

essential oil because in case of stress, more metabolites are produced in the plants and substances prevent from oxidization in the cells. It was also seen that as the increase of drought stress, its height of plant, stem diameter and stem yield decreased. Shoot reduction could be due to the reduction in the area of photosynthesis, drop in producing chlorophyll, the rise of the energy consumed by the plant in order to take in water and to increase the density of the protoplasm and to change respiratory paths and the activation of the path of phosphate pentose or the reduction of the root deploy, etc. Examining the amount of essential oil % indicated that with exerting mild drought stress T2 and T3, the amount of essential oil % didn't change so much. But exerting the fairly large drought stress T4 resulted in the great increase of essential oil % and as drought stress T5 became more, it was reduced, but regarding the essential oil percentage, no significant difference was observed between treatments T4 and T5. This fact indicates that exerting mild drought stress T3 and fairly large drought stress T4, is proper to gain more essential oil %, for as drought stress rise, the flowering shoot yield decreased of balm, the interaction between the amount of the essential oil % flowering shoot yield is considered important as 2 components of the essential oil yield. As it can be seen in Table 2, by exerting stress, essential oil yield goes up first and then by the rise in drought stress, the amount of the essential oil drops. Our results were similar to the findings of Aliabadi Farahani et al. (2008b) and Munne-Bosch and Alegre (1999).

Conclusion

The investigation showed that essential oil yield of balm was reduced under water deficit stress. This condition can be the most important environmental factor for the increase of essential oil rate by control of irrigation.

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