International Journal of Pure and Applied Zoology
Volume 5, Issue 2, pp: 85-91, 2017
http://www.ijpaz.com

ISSN (Print) : 2320-9577
ISSN (Online): 2320-9585

Research Article

INVESTIGATION OF MORPHOLOGICAL AND PHYTOCHEMICAL CHANGES AND TOLERANCE THRESHOLD OF CHAMOMILE UNDER DROUGHT STRESS CONDITIONS

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Article History: Received 26th May 2017; Accepted 18th July 2017; Published 21st July 2017

ABSTRACT

In order to investigate the effect of different levels of cattle manure under drought stress on yield and yield components of German chamomile a field experiment was conducted as a split-plot based on a randomized complete block design with three replications. Treatments were included five levels of drought stress as irrigation after of (50 (S1), 80 (S2), 110 (S3), 140 (S4) and 170 (S5) mm evaporation from evaporation pan) as main plot and three levels of cattle manure (0 (V1), 5 (V2) and 10 ton ha^{-1} (V3)) as sub plot. The results indicated that the main effect of drought stress and cattle manure was significant on all measured characteristics. Also the interaction of drought stress and cattle manure was significant on plant height, biological yield, essential oil content and essential oil yield. The highest flower yield (277.61 kg ha^{-1}) was achieved from treatments of 50 mm evaporation from evaporation pan (among the main effects of drought stress), while the highest flower yield (223.66 kg ha^{-1}) from treatments of 10 ton ha^{-1} among the cattle manure levels. Also the most essential oil content (0.945%) was observed at the interaction of 10 ton ha^{-1} cattle manure and 170 mm evaporation from evaporation pan. Based on the above, using of cattle manure under drought stress led to improving of morphological and phytochemical characteristics of chamomile.

Keywords: Cattle manure; Chamomile; Drought stress; Essential oil

INTRODUCTION

German chamomile (*Matricaria chamomila L.*) is one of the most valuable medicinal plants of composite family having many applications in pharmaceutical industries, nutritional and cosmetic. Generally, the essential oil content in medicinal plants changes due to genetic and environmental conditions. Irrigation different regimes had significant effect on flower yield, %age and yield essential oil (Hornok L, 1992). Decreasing of water content in plant tissues under drought stress conditions leads to growth limiting and some physiological and metabolic changes. In the conducted studies on chamomile were found that chamomile yield affected by cultivar, weather conditions and available water content at the room environment. Among the environmental hindering factors of growth and crops, horticultural and medicinal yield, drought is considered to be the most important factor of production losses, especially in arid and semi-arid (Reddy et al., 2004). In discussing of medicinal plant production, the actual value is given to product quality and production stability and product quantity placed on secondary importance. Studies conducted indicated that using of sustainable agriculture systems due to compliance with normal conditions and product quality originality provides the best conditions for the plant's production and the maximum active ingredient is produced under these circumstances (Darzi et al., 2008). One of the main pillars in sustainable agriculture uses organic manure at agronomic ecosystems. Crops production under drought weather conditions is required applying of appropriate management procedures. Using of organic manure as soil supplement and factors that caused improving of physical, chemical and biological properties of soil, can be one of the reduction strategies of the impact of adverse drought stress. For this purpose, several studies have been done on the effect of organic fertilizers on quality and quantity of medicinal plants. Water stresses is highly relevant with plants nutrition. One of the most negative impacts of drought stress is reduction availability and absorption of different nutrient elements for plants (Pirzad et al., 2006). Plant with appropriate nutrition, have better resistance to drought. Fertilizer management can had greatly effect crops production under arid region (Mohammadkhani et al., 2007). Organic manure especially cattle manures have much amounts of organic matter than fertilizer chemicals, and as resource of rich nutrients, especially nitrogen, phosphorus and potassium.

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Cattle manure cannot supply all the nutritional requirements of plants, but by improving the soil physical structure somewhat will cause balance in the soil chemical (Chaudhry et al., 1999). Investigation the effect of combined fertilizer chemical, compost and cattle manure on quantitative and qualitative yield of chamomile was found that by application of chemical fertilizers under mild stress conditions increased flower yield and essential oil and also in the second year application of cattle manure and compost was significantly increased quantitative and qualitative yield of chamomile (Ahmadian et al., 2009). In other studies interaction of cattle manure, fertilizer chemical and drought stress on essential oil, yield and chemical compounds of cumin indicated that cattle manure application leads to increasing quantitative yield and improving of qualitative yield under stress (Akabarinia, 2003). Although yet has been done extensive research on the effect of drought stress on crops, but few studies have been conducted about the interaction behaviour of some medicinal and aromatic plants such as chamomile under water deficit conditions. Aim of this research was investigating the effects of drought stress and cattle manure on morphological indexes, essential oil and tolerance threshold of German chamomile.

MATERIALS AND METHOD

In order to investigate the effect of different levels of cattle manure under drought stress on yield and yield components of German chamomile, a series of experiments was conducted as split-plot based on a randomized complete block design with three replications. The annual rainfall average 63 mm, minimum and maximum average of annual temperature 16 and 30°C respectively, and in terms of the climate is considered warm and arid regions. Treatments were included five levels of drought stress as irrigation after of (50 (S1), 80 (S2), 110 (S3), 140 (S4) and 170 (S5) mm evaporation from evaporation pan) as main plot and three levels of cattle manure (0 (V1), 5 (V2) and 10 ton ha^{-1} (V3)) as sub plot. Drought stress was considered as major factor and cattle manure as subplots. For the implementation of experiment land preparation operations was conducted with favourable weather conditions in end of April and preparation operations of soil substrate was done by using mechanized equipment (plows and disc). For sowing this plant in early of March was done planting in nursery and so in the spring seedlings were transferred to the main land. The dimensions of each main plot and each subplots was 3 × 5 and 2.5 × 2 m, respectively. Subplots were included two heaps with four rows and distance of 0.1 m between plants on row with a distance of 0.3 m between rows. Distance between main plot, subplot and between rows was 2, 0.5 and 3 m, respectively. The early of March is transferred seedlings to the main land and irrigation was conducted as regular until 5% flowering (flowering beginning). Actions of irrigation time were done by using evaporation pan that already was stationed in the field. Irrigation of plots different levels of drought stress was conducted by using data collected from class A evaporation pan and table of evaporation calculation from class A evaporation pan. Drought stress treatment continued until 80% field flowering and at the time the sampling was done from different plots considering the effect of marginal plot. At every turn flowers harvest, were counted number of flowers 12 plants and the mean it was considered as number of flowers per plant of first harvest and until last harvest (fourth harvest) thus was action and flowers number total in all of harvest, were recorded as the number of flowers per plant. After the harvest of each harvest, flowers were dried for seven days and were calculated flower yield. In order to determination of essential oil content, after flowers harvest and drying in shade from each plot a random sample preparation that after milling, was distillation by Clevenger apparatus and by using method of water distillation (Zeinali et al., 2008). The essential oil yield% was calculated by dry sodium sulfate. After determination of essential oil efficiency, it yield was calculated by product of flower yield and essential oil efficiency. Data analysis was done by using SAS software and means comparison of treatments was conducted LSD test at 1% probability levels.

RESULTS AND DISCUSSION

Plant height

Analysis of variance showed that cattle manure and drought stress ($P \geq 0.01$) and as well as their interaction ($P \geq 0.05$) had effect significant on plant height (Table 1). Results of cutting indicated that in all levels of drought stress, the effect of cattle manure had significant on plant height (Figure 1). Mean comparison interaction of drought stress and cattle manure revealed that in each levels of drought stress, the highest and the least plant height related to application of the most levels using of cattle manure (10 ton ha^{-1}) and non-using

Table 1. Analysis of variance measured traits German chamomile under the effect of drought stress and vermicompost (**, * and ns are significant at 0.01 and 0.05 probability level and non-significant, respectively).

S.O.V	d.f	Plant height	Number of branches	Main stem	Number of flowers per plant	Yield Flower	Biological yield	Harvest index	Essential oil content	Essential oil yield
Replication	2	0.35	2.86	0.68	51.81	134.79	7682.75	0.9	0.0011	0.017
Drought stress (S)	4	714.31**	455.07**	69.81**	12095.96**	20694.30**	1079797.96**	16.39**	0.0181**	0.815**
Main error	8	0.99	3.81	0.16	36.82	31.02	8721.36	2.34	0.0009	0.004
Vermicompost V)	2	94.68**	30.86**	2.15**	528.26**	218.16**	54158.95**	3.92**	0.1319**	0.749**
S × V	8	2.82*	0.81ns	0.37ns	23.01**	32.82ns	1291.23*	0.89ns	0.0172**	0.088**
Sub error	20	1.16	1.22	0.23	17.13	20.71	525.94	0.26	0.0036	0.013
C.V	-	2.74	7.46	9.74	3.97	2.06	1.82	3.16	7.88	7.24

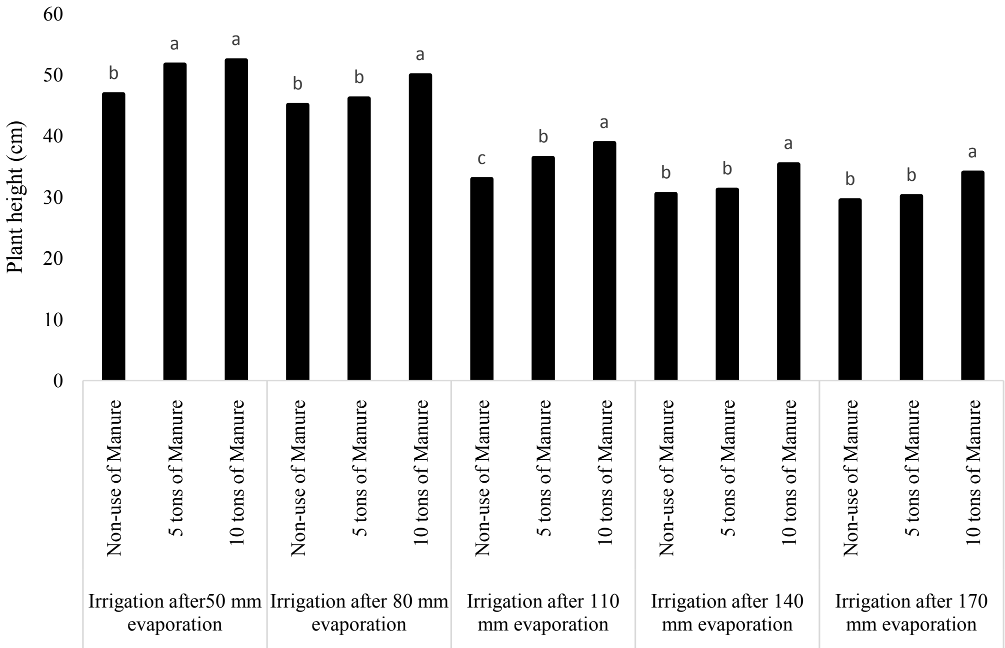

Figure 1. The interaction of drought stress and vermicompos on plant height of German chamomile.

of cattle manure, respectively. Also results showed that by increasing of drought stress, decreased plant height which increased with cattle manure application. The most and the least plant height in level of 10 ton ha^{-1} cattle manure was obtained from drought stress of 50 and 170 mm evaporation from evaporation pan, respectively. In treatments of non-using of cattle manure, drought stress had the most impact on reduction of plant height (Figure 1). At this study, it seems cattle manure *via* high strength of water absorption and optimal availability of macro and micronutrients had positive impact on photosynthesis rate and biomass production and have caused improving of plant height. In relation to decreasing of plant height in impact of water stress can stated that water restrictions caused reduction of division and cell expansion and from this through decreases organs growth and plant height. Thus such as negative impacts of drought stress, is reduction of plant height due to moisture available and nutrients uptake capability and growth reduction that the results of similar studies is reported by (Afzali et al., 2007) on chamomile, (Azizi et al., 2008) on chamomile and (Safikhani, 2005) on moldavian balm.

Number of lateral branches

Analysis of variance showed that the effect of cattle manure and drought stress was significant ($P \geq 0.01$) on number of lateral branches of chamomile (Table 1). Based on mean comparison increasing of drought stress had effect significant on reduction of lateral branches of chamomile, so that the most (24.11) and the least (8) lateral branches was obtained from treatments of 50 and 170 mm evaporation, respectively. On the other hand, with increasing of cattle manure, increased significantly the number of lateral branches so that the most (16.2) and the least (13.33) the number of lateral branches was obtained from treatments of 10 ton ha^{-1} and non-using of cattle manure, respectively. It seems cattle manure by providing nutrients and more water uptake makes increasing of lateral branches and subsequently increasing of the number of main stem and number of flowers per plant, that corresponded with results (Saeednejad and Rezvanimoghadam, 2011) in the using of bio-fertilizers such as vermicomposting on cumin. (Tahami et al., 2010) by investigate of organic manure and chemical fertilizer on basil stated that sheep manure caused production the most the number of lateral branches per plant, and at the same environmental conditions, providing nutrients for plants by different fertilizers can causes increasing of plant growth and, subsequently the number of lateral branch.

Number of main stem per plant

Analysis of variance indicated that the effect of cattle manure and drought stress was significant ($P \geq 0.01$) on number of main stem, while their interaction was not significant on this trait (Table 1). Results of mean comparison showed increasing of drought stress lead to significant reduction in the number of main stem (Table 2). So that the most (8.44) and the least (2.55) the number of main stem was obtained from treatments of 50 and 170 mm evaporation from evaporation pan, respectively (Table 2). Also results of mean comparison showed by increasing of cattle manure, increased significantly the number of main stem, so that the most the number of main stem (5.26) related to treatments of 10 ton ha^{-1} cattle manure that had not significant difference with 5 ton ha^{-1} cattle manure and the least the number of main stem (4.53) was obtained from non-using of cattle manure. Arazmjoo et al. reported that by increasing of levels of drought stress from 90% to 50% of filed capacity the number of main stem German chamomile decreased from 11.93 to 9.07. Also these researchers reported that using of different fertilizer sources produces the number of different main stem per plant; so that chemical fertilizer, cattle manure and

Table 2. Mean comparison main effects of drought stress and vermicompost on measured traits of German chamomile (In each column and treatment similar letters indicates no significant difference in the level of 5% based on the LSD test).

Drought stress (mm evaporation)	Number of branches	Harvest index (%)	Flower yield (kg ha-1)	Main stem
50	24.11a	18.65a	277.61a	8.44a
80	20.22b	16.05b	261.87b	7.44b
110	12.88c	15.76b	209.94c	3.66c
140	8.77d	15.74b	175.66d	2.66d
170	8.0d	15.25b	174.63d	2.55d
Manure (ton ha^{-1})				
0	13.33c	15.93b	216.04c	4.53b
5	14.86b	16.06b	220.14b	5.06a
10	16.20a	16.88a	223.66a	5.26a

vermicompost produces 12.27, 10.78 and 9.23 main stem per plant, respectively.

Number of flowers per plant

Results of analysis of variance indicated that the effect of cattle manure, drought stress and their interaction was significant on number of flower per plant at 1% probability levels (Table 1). Mean comparison interaction of drought stress and cattle manure revealed that in each levels of drought stress the most and the least the number of flowers per plant related to the 12 highest levels of cattle manure (10 ton ha^{-1}) and non-using of cattle manure, respectively. Also results showed that by increasing of drought stress reduced the number of flowers per plant and by cattle manure application increased the number of flowers per plant; so that the most the number of flowers related to levels of 10 ton ha^{-1} cattle manure under drought stress 50 mm evaporation from evaporation pan. At treatments of non-using of cattle manure, drought stress had the greatest impact on the number of flowers per plant and the least the number of flowers related to levels of 170 mm evaporation from evaporation pan (Table 1). (Arazmjoo et al., 2010) reported that by increasing of drought stress levels from 90% to 50% of field capacity the number of flowers per plant decreased from 152.6 to 90.83. (Ahmadian et al., 2010) investigated the effect of chemical fertilizer, cattle manure and compost on yield components, some physiological characteristics and essential oil content under drought stress conditions reported that by increasing of drought stress levels from 90 to 50% of field capacity decreased the number of flowers in german chamomile from 158.91 to 99.77.

Flower yield

Results of analysis of variance revealed that the effect of cattle manure and drought stress was significant on flower yield at 1% probability levels, while their interaction was not significant on this trait (Table 1). Mean comparison showed that by increasing levels of cattle manure increased dry flower yield of chamomile; so that the most (223 kg ha^{-1}) and the least (216 kg ha^{-1}) flower yield was obtained from treatments of 10 ton ha^{-1} cattle manure and non-using of cattle manure, respectively. Also mean comparison the effect of drought stress on dry flower yield indicated that by increasing of drought stress levels decreased flower yield of chamomile; so that the most (277 kg ha^{-1}) and the least (174 kg ha^{-1}) flower yield was obtained from treatments growth and produce of its constituent organs in different stages of vegetative and reproductive growth. The effect of drought stress on each of the constituent components of flower can lead to change in amount of flowers produce. In general flower yield in chamomile is the result of the interaction of components that formed each of them at different stages of vegetative and reproductive growth. In the meantime the number of stem and flowers per plant are considered as the most important flower yield components. (Arazmjoo et al., 2010) reported that by increasing of drought stress levels from control to 50% of field capacity, decreased flower yield to 18.1%.

Harvest index

Results of analysis of variance revealed that the effect of cattle manure ($P \geq 0.05$) and drought stress ($P \geq 0.01$) was significant on harvest index; but and their interaction was not significant on this trait (Table 1). Mean comparison the effect of drought stress on harvest index indicated that by increasing drought stress levels decreased harvest index (Table 2); so that among different levels of drought stress the most (18.65%) and the least (15.25%) harvest index was obtained from treatments of 50 and 170 mm evaporation from evaporation pan, respectively (Table 2). Also by increasing cattle manure levels increased harvest index of German chamomile (Table 2). Mean comparison revealed that by increasing cattle manure levels increased harvest index; so that the most (16.88%) and the least (15.93%) harvest index was obtained from treatment 10 ton ha^{-1} cattle manure and non-using of cattle manure, respectively (Table 2). In other study that by (Saeednejad and Rezvanimoghadam, 2011) was conducted on cuminum, the results showed that none of organic and cattle manure treatments had no significant the effect of on harvest index. In fact at these organic manure treatments by increasing biological yield, increased grain yield to certain ratio. These researchers reported that the most important external factor that causes changes of harvest index is changes at the fertilizer levels.

Biological yield

Results of analysis of variance revealed that the effect of cattle manure and drought stress ($P \geq 0.01$) and their interaction ($P \geq 0.05$) were significant on biological yield. Results of cutting revealed that in all drought stress levels the effect of cattle manure was significant on biological yield at 1% probability levels (Figure 2). Mean comparison the interaction of drought stress and cattle manure revealed that in each levels of drought stress the most and the least biological yield related to treatments of 10 ton ha^{-1} cattle manure and non-using of cattle manure, respectively. Also results showed that biological yield by increasing drought stress decreased and increased by cattle manure application, so that the most impact on biological yield related to levels of 10 ton ha^{-1} cattle manure under drought stress of 50 mm evaporation from evaporation pan. Using of cattle manure due to high water-holding ability, caused improving of soil physical conditions and indirectly involved in plant yield. Since the branches, leaves and shoots of plants under affected of root uptake activity and amount of transfer nutrients and water from the root can be argued that organic manure by increasing of amount of available nutrients and their gradual release lead to increasing of plant growth and increase biomass production.

Essential oil%

The analysis of variance revealed that the effect of cattle manure, drought stress and their interaction ($P \geq 0.01$) was significant on essential oil % of German chamomile (Table 1). Mean comparison the interaction of cattle manure and drought stress indicated that at drought stress levels 50, 140 and 170 mm evaporation from evaporation pan the most essential oil% related to levels of 10 ton ha^{-1} cattle manure and the least essential oil% was obtained from non-using of cattle manure (Figure 3). At drought stress levels of 50 mm evaporation from evaporation pan by increasing of cattle manure application increased essential oil% that could cause by increasing of leaching and result in increasing of nutrients losses at the time of non-application of cattle manure or small amounts of cattle manure, that by increasing cattle manure amount (10 ton ha$^{-1)}$ and with creating of suitable

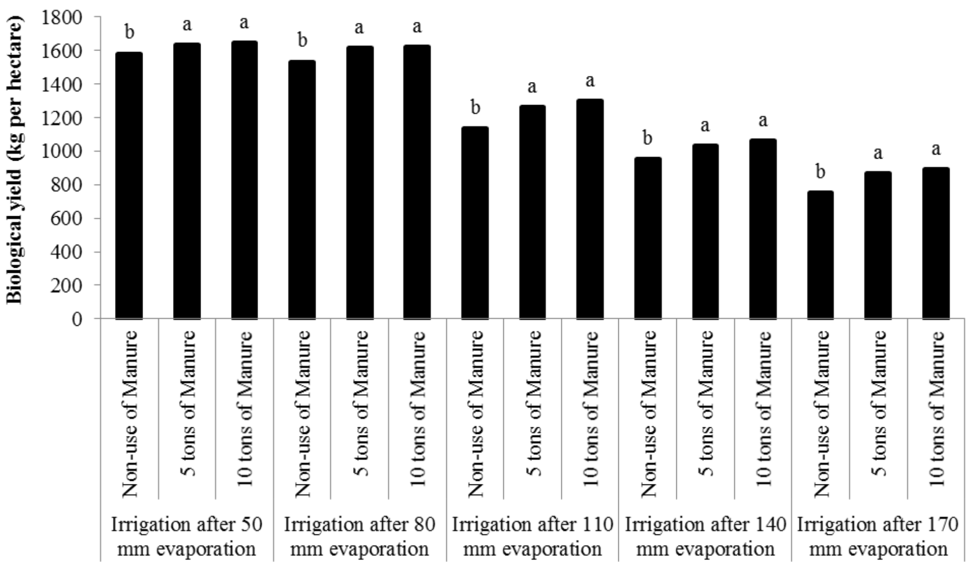

Figure 2. The interaction of drought stress and vermicompos on biological yield of chamomile.

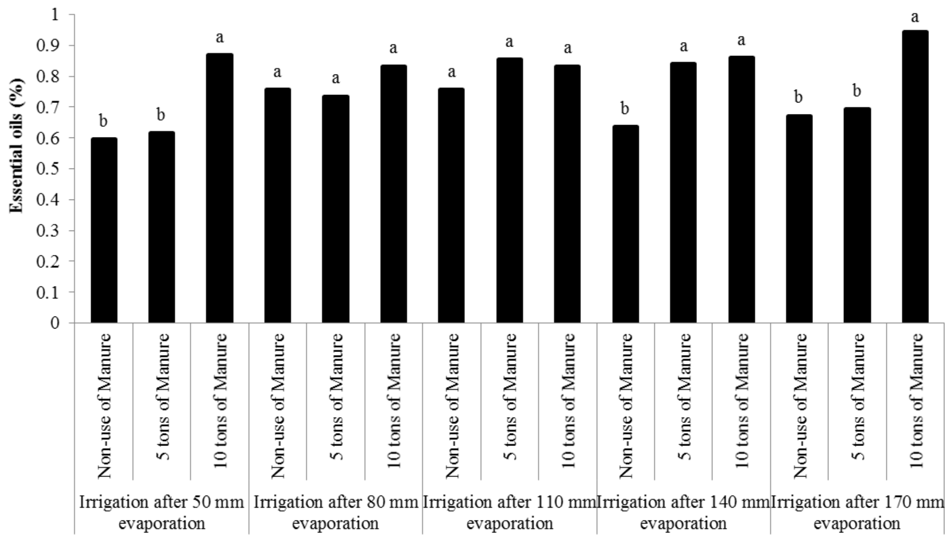

Figure 3. The interaction of drought stress and vermicompost on essential oil of chamomile.

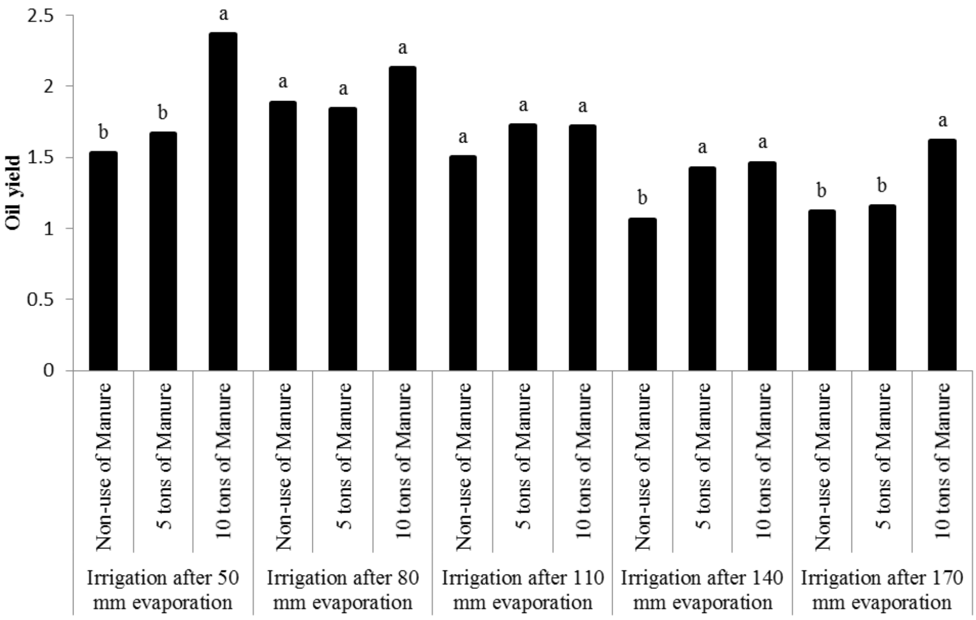

Figure 4. The interaction of drought stress and vermicompost on essential oil yield of chamomile.

environments for root growth and consequently improving of nutrients uptake, has increased essential oil%. In drought stress levels of 80 and 110 mm evaporation from evaporation pan (mild stress) by increasing of cattle manure amount was not significant increase that could due to availability to sufficient water and nutrients for all treatments levels. In related to the significance of interaction cattle manure and drought stress on essential oil% in drought stress 140 and 170 mm evaporation from evaporation pan (severe drought) could state that with regard to very important role of nutrients such as nitrogen and phosphorus in the essential synthesis cycle it seems high levels of cattle manure (10 ton ha^{-1}) by improving of physical conditions and critical processes of soil and in result improving of water and nutrients uptake especially nitrogen and phosphorus caused reduction the effect of water stress particularly at high stress levels that consequently has increased essential oil%. In fact, organic manures due to more moisture maintain in the soil and help to better absorption of nutrients and nutritional balance, assist the plant in stress conditions. It seems it is better in severe stress status to be used the greater amounts of organic fertilizers like vermicompost until reduced the negative effects of stress to the extent possible. Similar results reported by (Ahmadian et al., 2010) on German chamomile.

Essential oil yield

Results showed that the effect of cattle manure and drought stress ($P \geq 0.01$) and their interaction ($P \geq 0.05$) was significant on essential oil yield of chamomile shoot (Table 1). Mean comparison the interaction of cattle manure and drought stress revealed that in irrigation levels of 50, 140 and 170 mm evaporation from evaporation pan, the most the least essential oil yield related to levels of 10 ton ha^{-1} cattle manure and non-using of cattle manure, respectively. Since the essential oil yield is function of the essential oil content and flower yield in treatment of 170 mm evaporation from evaporation pan, drought stress by reduction of flower yield had the most impact on decreasing of essential oil yield, also at treatments of 50 mm evaporation from evaporation pan due to increasing of flower yield and essential oil content, has increased essential oil yield (Figure 4). Our results correspond with the results of (Salehi et al., 2011); they found that by increasing of organic manure application not only providing nutrients required increased for plants (especially nitrogen and phosphorus) but organic fertilizers with improving of physical conditions and critical processes of soil, by creating of a favourable environment for root growth caused increasing of dry matter production, flower yield and essential oil percent (%) in chamomile. Decreasing of essential oil yield in result soil moisture reduction may be due to the effect of deleterious the drought stress on growth and vegetative organs yield.

CONCLUSION

Recognition of environmental factors such as drought stress is an important factor in the success of medicinal plants planting and in the meantime identify fertilizer that can are compatible in the direction increasing of plant resistance to drought stress conditions and have been favourable effects on qualitative and quantitative indices of plants is necessary. With regards to the aim from chamomile planting is flower and essential oil yield, thus increasing of flower and essential oil yield under drought stress conditions is important by organic manure application. Based on the obtained results in this experiment can be stated that however, with the reduction of water consumption amount and consequently the incidence of drought stress is reduced flower and essential oil yield of German chamomile, but by cattle manure application (at the highest level of stress), can reduced partly the adverse effects of drought stress on chamomile yield. Decreasing of flower yield during drought stress even until irrigation levels of 170 mm, accompanied by increasing of essential oil. Therefore, it can be stated that during drought stress and reducing of the water amount, cattle manure has a positive impact on German chamomile and is better in the severe stress condition to be used greater amounts from organic fertilizers such as cattle manure until reduce the negative effects of stress to the extent possible.

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