

Effect of Water Deficit Stress on Yield and Essential Oil Components of *Rosa damascena* Mill

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ABSTRACT

To investigate water deficit stress on yield and essential oil components in *Rosa damascena* Mill., an experiment was conducted in a completely randomized block design with three replications in Karaj/ Iran. The treatments included 90 mm (severe stress) and 45 mm (mild stress) of cumulative evaporation from the surface of the class A evaporation pan that was compared to the control treatment (no stress). Combined analysis performed in the years. The studied properties included flower yield, essential oil percent, essential oil yield, and some essential oil components in the *Rosa damascena*. The essential oil of fresh flowers was extracted using the water distillation method by the Clevenger apparatus. The essential oil components were identified using GC and GC/MS. The results showed that the effect of year and water deficit stress was significant on flower yield, essential oil yield, and some components in the essential oil. Maximum flower yield was obtained in the second year without stress conditions (1208 g/plant), significantly different from the others. In the essential oil, citronellol, and geraniol increased due to water deficit stress and included 24.48 and 2.89%, respectively. Applying water deficit stress reduced the amount of nonadecane, eicosane, and heneicosan. By applying different levels of water deficit stresses can be modified, and manage the components in the essential oil of the *Rosa damascena*.

Keyword: Medicinal Plants, Rosaceae, Karaj, Iran

INTRODUCTION

Rosa damascena Mill. belongs to the *Rosaceae* family, whose products include dry buds, petals, rosewater, and essential oils. The essential oil is the most critical secondary metabolite of the damask rose [1]. The rose flowers are processed into rose oil, rose concrete, and rose water [2]. Rose petals usually contain little essential oil than other essential oil plants. The amount of essential oil in *Rosa damascena* petals is about 0.03%. Due to the lack of synthetic and natural alternatives, rose essential oil is one of the most expensive essential oils on the world market [3]. The rose essential oil is used in food, perfumery, health products, cosmetics, and pharmaceutical industries [2, 4, 5]. Although *Rosa damascena* needs sufficient water, this plant is usually resistant to water deficit stress. It can also be established and grown in almost arid regions with an annual rainfall of less than 250 mm [6]. In medicinal plants, secondary metabolites must be considered in addition to the amount of production. Successful rose cultivation can be done by knowing the water needs and supply during critical growth. Different amounts of water deficit stress affect the quantity and quality of plants. Water deficit does not always reduce plant yield and quality. By managing the application of water stress and reducing water consumption, an acceptable yield is obtained from cultivation. In the Karaj region, the yield of yarrow (*Achillea millefolium*) increased with mild water stress but decreased with severe stress [7]. Water deficit stress decreased the essential oil yield in rosemary and Japanese mint. It increased the percentage of essential oil and the main components of essential oil in sweet basil and American basil [8]. In *Satureja hortensis* L., severe deficit stress increased essential oil content compared to mild stress. Carvacrol increased in mild stress, and α -terpinene decreased in severe and mild stress [9]. In addition to the essential oil and its compounds, deficit stress also affects other plant compounds. Water stress activates the antioxidant mechanism [10]. The absorption of nutrients is related to the amount of irrigation water. It was reported in a study, the optimum flower yield was achieved with the potassium silicate spraying treatment at an irrigation level equal to 50% of the PWR [11]. It has been reported that increasing irrigation water amounts and nitrogen doses significantly increased the rose flower yield [12].

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This study investigated the effect of water deficit stress on flower yield, amount of essential oil, and components in *Rosa damascena* essential oil.

MATERIAL AND METHODS

In order to investigate the water deficit stress on yield and essential oil components in *Rosa damascena* Mill., an experiment was conducted at the Alborz Research Center (affiliated with the Research Institute of Forests and Rangelands) in Karaj/ Iran. Table 1 shows some of the soil characteristics of the region.

Table 1 Soil properties in Alborz Research Station, Karaj

Properties	No.1	No.2	No.3
Deep (cm)	0-10	10-20	>20
Ec (dSm-1)	2.01	3.76	2.73
pH	7.48	7.75	8.04
OC (%)	0.65	0.67	0.65
Total N (%)	0.063	0.056	0.067
P (Ava)	15.4	9.03	11.95
K (Ava)	222.7	230	218.6
Fe (Ava)	-	5.9	-
Cu (Ava)	-	0.62	-
Sand (%)	32.4	39	27
Loam (%)	40.3	34.7	44.7
Clay (%)	27.3	26.3	28.3
Soil texture	Loam clay-Loam	Loam	Clay-Loam
Bulk density (gcm-3)	1.45	1.48	1.46

The experiment was conducted in the field condition in a completely randomized block design with three replications. The treatments included 90 mm (severe stress) and 45 mm (mild stress) of cumulative evaporation from the surface of the class A evaporation pan that was compared to the control treatment (no stress). Combined analysis performed in the years. The studied properties included flower yield, essential oil percentage, essential oil yield, and some essential oil components in *Rosa damascena*.

Irrigation was done by the drip method. Nine plants were considered for each treatment. In each experimental plot, seedlings (prepared in October of the previous year) were transferred to the field in the second half of March. The distance between the two plants in the row and between the two rows was two meters.

After harvesting flowers in May, the essential oil of fresh flowers was extracted using the water distillation method by the Clevenger apparatus. The components in the essential oil were identified using Gas chromatography and Gas Chromatography-Mass spectrometry.

Gas Chromatography (GC)

The oils were analyzed by GC, using a Thermo-UFM ultra-fast gas chromatograph equipped with a DB-5 fused silica column (10 m × 0.1 mm i.d., film thickness 0.4 µm). The oven temperature was 60–280 °C at a rate of 40 °C/min and then held at 280 °C for 3 min. Detector (FID) temperature and injector temperature were 280 °C; helium (purity 99/99%) was used as carrier gas with a linear velocity of 0.5 ml/min. Quantification data was obtained from GC-FID area percentages without the use of correction factors.

Gas Chromatography-Mass Spectrometry (GC-MS)

GC-MS analysis is performed on a Varian 3400 GC-MS system equipped with a DB-5 fused silica capillary column (30 m × 0.25 mm i.d.; film thicknesses 0.25 µm). The temperature was programmed from 60 °C to 250 °C at a rate of 3 °C min⁻¹; Injector and transfer line temperatures were 260 °C and 270 °C, respectively; acquisition mass range of 40–340 amu; ionization voltage of 70 eV; the carrier gas was helium with a linear velocity of 31.5 cm/s, split ratio 1/60, ionization energy 70 eV; scan time 1 s and mass range of 40–340 amu.

In this research, a comparison of means was done with LSD test, and statistical calculations were done with SAS software.

RESULTS

According to the analysis of variance, the effect of year, water treatments, and their interaction on fresh and dry flower weight and flower essential oil yield were significant at a 5% level (Table 2).

Between the years, the highest fresh flower weight (726.4 g/plant), dry flower weight (167.06 g/plant), and essential oil yield (0.51 g/plant) were observed in the second year, which were significant compared to the first year. Among the water treatments, the control treatment showed the highest fresh flower weight (898.3 g/plant), dry flower weight (206.6 g/plant), and essential oil yield (0.58 g/plant) which were significant compared to other treatments (Table 3). The interaction effect of the treatments showed that the highest fresh flower weight (1208.8 g/plant), dry flower weight (278.02 g/plant), and essential oil yield (0.71 g/plant) were observed in the second year and under stress-free conditions, which were significantly different from other treatments (Table 4).

Table 5 shows the essential oil components of *Rosa damascena*. According to the variance analysis, the year's effect was significant on citronellol, geraniol, heptadecane, eicosane, and heneicosane at 1% and tricosane at 5% level. The effect of water treatment was significant on geraniol at 1% and on citronellol, nonadecene, nonadecane, eicosane, and heneicosane at a 5% level. Interaction of year and water treatments was significant on citronellol and geraniol at 1% level and on heptadecane, nonadecene, nonadecane, eicosane, heneicosane, and tricosane at 5% level (Table 6).

Among the years, the highest citronellol (24.92%), geraniol (2.67%), and heptadecane (3.84%) were observed in the second year. The maximum amount of eicosane (4.48%), heneicosane (26.53%), and tricosane (7.86%) were observed in the first year, significantly different from the other year. Among the water treatments, the highest amounts of citronellol (24.48%), geraniol (2.89%), and nonadecene (5.44%) were observed in the control treatment. The highest amount of heneicosane (22.14%) was observed in mild stress. The highest amount of nonadecane (42.47%) and eicosane (3.96%) were observed in severe stress. Water treatments did not significantly affect heptadecane and tricosane (Table 7). The interaction of year and water treatments showed that the highest citronellol (27.95%) and geraniol (5.5%) were observed in the second year and non-stress conditions. Most heptadecane (4.11%) was observed in the second year and mild stress conditions which was significantly different from the first year. The highest nonadecene (5.97%) was observed in the first year and non-stress conditions. In both years, non-stress conditions significantly differed from severe stress on nonadecene. The highest nonadecane (42.82%) was observed in the second year, and severe stress was significantly different from the non-stress condition in the first year. The highest eicosane (5.33%), heneicosane (30.50%), and tricosane (7.99%) were observed in the first year and severe stress condition (Table 8).

Table 2 Analysis of variance in the year and water treatment effects on yield and essential oil of damask rose

S.O.V.	df	Mean of squares				
		Fresh weight flowers	Dry weight flowers	Dry to wet weight of flowers	Percentage of essential oil	The yield of essential oils
Year	1	621922.26 *	32899.69 *	ns 0.0036	0.000066 ns	0.36 *
Error (Ea)	4	24859.24	1315.1	0.017	0.00034	0.066 *
Water treatments	2	601840 *	31837.34 *	ns 0.0065	0.00037 ns	0.23 *
Water × Year	2	101004.9 *	5343.16 *	ns 0.0067	0.00056 ns	0.00031 *
Error (Eb)	8	54557.3	2886.1	0.032	0.00066	1.114
Total	12	-	-	-	-	-

- **, *, and ns, respectively show significance at 1% and 5% probability levels and no significant difference

Table 3 Comparison of means in the year and water treatment effects on yields and essential oil of damask rose

Treatments		Fresh weight flowers (g/plant)	Dry weight flowers (g/plant)	Dry to wet weight of flowers	Percentage of essential oil	The yield of essential oil (g/plant)
Years						
	First-year	354.6 b	81.56 b	0.23 a	0.06 a	0.23 b
	Second year	762.4 a	167.06 a	0.28 a	0.06 a	0.51 a
Water treatments						
	Control	898.3 a	206.60 a	0.23 a	0.07 a	0.58 a
	Mild stress	427.2 b	98.24 b	0.22 a	0.06 a	0.33 b
	Severe stress	296 b	68.08 c	0.28 a	0.05 a	0.2 b

- Similar letters in each column indicate no significant difference

Table 4 Comparison of means interaction in the year and water treatment effects on yields and essential oil of damask rose

Treatments		Fresh weight flowers (g/plant)	Dry weight flowers (g/plant)	Dry to wet weight of flowers	Percentage of essential oil	The yield of essential oil (g/plant)
Years	Water treatments					
First	Control	587.72 ab	135.18 b	0.230 a	0.08 a	0.44 ab
First	Mild stress	375.57 b	86.38 c	0.227 a	0.05 a	0.19 bc
First	Severe stress	100.50 b	23.12 d	0.228 a	0.05 a	0.05 c
Second	Control	1208.8 a	278.02 a	0.230 a	0.06 a	0.71 a
Second	Mild stress	478.73 ab	110.11 bc	0.207 a	0.07 a	0.47 ab
Second	Severe stress	491.53 ab	113.05 bc	0.332 a	0.06 a	0.35 b

- Similar letters in each column indicate no significant difference

Table 5 A number of essential oil components of *Rosa damascena* Mill.

Row	Essential oil components	RI (Retention index)
1	Citronellol	1230
2	Geraniol	1250
3	Heptadecane	1700
4	Nonadecene	1897
5	Nonadecane	1900
6	Eicosane	2000
7	Heneicosane	2100
8	Tricosane	2300

Table 6 Analysis of variance in the year and water treatment effects on essential oil components of damask rose

S.O.V.	df	Mean of squares							
		Citronellol	Geraniol	Heptadecane	Nonadecene	Nonadecane	Eicosane	Heneicosane	Tricosane
Year	1	922.56**	24.33 **	12.15 **	2.57 ns	83.43 ns	18.56**	751.93 **	75.88 *
Error (Ea)	4	66.93	4.56	0.19	0.67	40.69	0.27	8.54	5.72
Water treatments	2	203.45 *	8.66 **	0.22 ns	3.65 *	113.33 *	2.13 *	74.21 *	1.37 ns
Water × Year	2	106.64 **	4.87 **	0.25 *	0.95 *	18.57 *	0.65 *	19.63 *	1.36 *
Error (Eb)	8	41.23	9.79	0.11	0.53	22.95	0.23	17.20	13.11
Total	12	-	-	-	-	-	-	-	-

- **, *, and ns, respectively show significance at 1% and 5% probability levels and no significant difference

Table 7 Comparison of means in the year and water treatment effects essential oil components of damask rose

Treatment	Essential oil components (%)							
	Citronellol	Geraniol	Heptadecane	Nonadecene	Nonadecane	Eicosane	Heneicosane	Tricosane
Years								
First	10.60 b	0.34 b	2.01 b	5.20 a	36.85 a	4.48 a	26.53 a	7.86 a
Second	24.92 a	2.67 a	3.86 a	4.44 a	41.18 a	2.45 b	13.61 b	3.75 b
Water treatments								
Control	24.48 a	2.89 a	2.99 a	5.44 a	34.14 b	2.81 b	16.01 b	5.50 a
Mild stress	14.76 b	0.72 b	3.08 a	5.08 a	40.45 ab	3.64 a	22.14 a	6.35 a
Severe stress	14.06 b	0.91 b	2.71 a	3.94 b	42.47 a	3.96 a	22.06 b	5.56 a

- Similar letters in each column indicate no significant difference

Table 8 Comparison of means interaction in the year and water treatment effects on essential oil components of damask rose

Treatments		Essential oil components (%)							
		Citronellol	Geraniol	Heptadecane	Nonadecene	Nonadecane	Eicosane	Heneicosane	Tricosane
Years	Water treatments								
First	Control	21.01 ab	0.72 bc	1.96 b	5.97 a	30.28 b	3.50 bc	20.89 ab	7.71 a
First	Mild stress	8.51 c	0.3 bc	2.04 b	5.75 a	38.15 ab	4.63 ab	28.21 a	7.87 a
First	Severe stress	2.31 d	0 c	2.04 b	3.87 b	42.13 a	5.33 a	30.50 a	7.99 a
Second	Control	27.95 a	5.05 a	4.03 a	4.90 ab	37.99 ab	2.11 c	11.13 b	3.29 bc
Second	Mild stress	21.1 ab	1.13 b	4.11 a	4.41 ab	42.74 a	2.64 c	16.07 ab	6.24 ab
Second	Severe stress	25.81 a	1.81 b	3.39 a	4.02 b	42.82 a	2.61 c	13.62 b	1.05 c

- Similar letters in each column indicate no significant difference

Table 9 Correlation of some quantitative and qualitative characteristics of damask rose

Characteristics	Fw flower	Essential oil	Citronellol	Geraniol	Heptadecane	Nonadecene	Nonadecane	Eicosane
Fw flower	1							
Essential oil	0.58 *	1						
Citronellol	-0.39 ^{ns}	0.095 ^{ns}	1					
Geraniol	0.42 *	0.15 ^{ns}	-0.28 ^{ns}	1				
Heptadecane	-0.59 *	-0.71 **	0.47 ^{ns}	-0.18 ^{ns}	1			
Nonadecene	-0.51 *	-0.55 *	-0.22 ^{ns}	-0.35 ^{ns}	0.28 ^{ns}	1		
Nonadecane	0.61 *	-0.92 **	-0.25 ^{ns}	-0.13 ^{ns}	0.48 *	0.57 *	1	
Eicosane	-0.31 ^{ns}	-0.18 ^{ns}	0.013 ^{ns}	0.24 ^{ns}	0.16 ^{ns}	0.09 ^{ns}	0.17 ^{ns}	1

- **, *, and ns, respectively show significance at 1% and 5% probability levels and no significant difference.

Based on the characteristics correlation results, fresh flower weight had a positive and significant correlation with the percentage of essential oil, geraniol, and nonadecane at 5% and had a negative and significant correlation with heptadecane and nonadecene at the level of 5%.

The percentage of essential oil with heptadecane and nonadecane at 1% and with nonadecene at 5% showed a negative and significant correlation. Heptadecane and nonadecene had a positive and significant correlation with nonadecane at 5% (Table 9).

CONCLUSION

Flower Yield

According to the results, in the second year, the flower yield was higher than in the first year. The increase can be due to plant establishment or weather conditions in the second year. By applying mild and severe stresses, the yield of fresh flowers decreased by 36.1% and 82.9%, respectively, compared to non-stressed conditions. Drought stress is the main limitation in crop production and plant fertility [13]. Due to water deficit stress, nutrient uptake, and photosynthesis are reduced. Deficit water stress does not always reduce yield. The type of plant, environmental conditions, and stress level are the factors that determine the reduction of plant performance. According to research, the response of different genotypes of damask rose to the water stress in the early stages of growth showed that, after irrigation, stressed genotypes showed significant differences in vigor, viability, number of leaves, leaf area, number of branches, and length of the tallest branch [14, 15]. In the Karaj region, mild water stress increased the yield of yarrow (*Achillea millefolium* L.), but severe stress significantly reduced flower yield [7].

Essential Oil Content

In the experiment, the effect of year and drought stress was not significant on the percentage of essential oil, but the yield of essential oil was significant in years and water treatments. Essential oil yield depends on essential oil percentage and fresh flower yield. Therefore, an increase in each will increase the yield of the essential oil. In the experiment, an increase in flower yield increased the yield of essential oil.

The effect of drought stress on the percentage of essential oil is different in plant species. In some studies, water deficit stress increased the percentage of essential oil [8] and in others, it decreased [16, 17, 18]. In chamomile (*Matricaria chamomile*), drought stress caused a significant reduction in essential oil content [19].

Essential Oil Components

In this research, it was found that a number of essential oil components changed in the first and second years. According to a study, essential oil components are governed by environmental conditions [20]. For example, the percentages of citronellol and geraniol as the two important compounds of rose oil quality in the second year were more than their percentages in the first year [21].

In this study, the level of water deficit stress significantly affects the percentage of components in the essential oil. The highest percentage of citronellol, geraniol, and nonadecene was observed in the control treatment, and the highest tricosane was observed in the mild stress. The studies show the effect of water stress on the amount of essential oils in medicinal plants and their components [22]. It is noteworthy, the effect of drought stress on the amount and essential oil components is different in plant species [8, 9, 23].

In this study, it was determined that by applying correct management in irrigation and applying deficit water stress, it is possible to carry out targeted supplementary irrigation and control the quantitative and qualitative performance of medicinal plants.

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