

ROLES OF MYCORRHIZAAND SALICYLIC ACID IN CARRYING THE CARNATION PLANT DAINTHUS CARYOPHULLUS TO WATER STRESS

Mahmood Shaker Ahmed and Sawsan Abdullah Abdul Latif

Department of Gardening and Garden Engineering, Faculty of Agricultural Engineering Sciences, University of Baghdad, Iraq.

Abstract

The experiment was carried out in the facilities of the Department of Horticulture and Garden Engineering, Faculty of Agriculture, University of Baghdad, Jadriya for the period from 15-8-2017 until 15-8-2018. The roots of the plants were vaccinated with the Mycorrhiza by use the method of Pad with two levels M0 (0) and M1 (20g) for each seedlings. The plants were sprayed with Salicylic acid on the vegetative with concentrations of SA0 (0 mg L⁻¹), SA50 (50 mg L⁻¹) SA100 (100 mg L⁻¹) SA150 (150 mg L⁻¹). The plants were exposed to three irrigation separators (D2 two days, D4 four days, D6 six days). The results showed that the Inoculation of plant roots with Mycorrhiza led to increase the leaf area, chlorophyll content, number of spores, reduction of antioxidant enzymes of catalase and peroxidase with a total of 799cm², 313.9mg, 100g⁻¹, 8.26mg, 100g⁻¹, soft weight, 125.5 days, 21.69%, 10.51 days, 113.6cm, 2,197.60, 10g, 1 dry soil, 1.03g, 114.2m and 82.7m respectively. The majority of the vegetative, root and floral indices showed insignificant differences in the results of the interaction between the irrigation separators at 2 days after irrigation and the Mycorrhiza (M1D2) between irrigation separators (irrigation after 4 days) and Mycorrhiza (M1D4).

Key words: clover, Mycorrhiza, Salicylic acid, irrigation separators.

Introduction

The role of cut flowers, especially cloves, is important in the international trade arena, with the Netherlands' exports reaching 281 million picking flowers each year and in Colombia, 6.284 million flowers per year. The global competition in carnation production is competition in the agricultural, industrial and economic fields, the problem of economic returns in different countries such as the Netherlands, which is ranked first economically, depends on trade in cut flowers and comes mainly for export (Mekonnen, Hoekstra & Becht, 2012). The cloves family includes about 2100 species and 89 genus, Dianthus has 300 species, all of which grow in North Africa, Asia and Europe. The clove plant is a special breeding plant. It needs agricultural operations to strengthen, squash, and fertilize. It also needs irrigation whenever the land is dried. The cloves are affected by dryness and thirst. Branch formation Particularly in the vegetative growth season as well as the survival of small-sized seedlings, the plants undergo various environmental stresses, including unusual temperatures, inappropriate chemical and physical conditions, exposure to high humidity and increased

toxicity. However, long-term water stress reduces the growth of the plant and crop yields more than all other stresses combined (Sairam, Rao & Srivastava, 2002).

As a result of the scientific and technical development and the need to find scientific means to rationalize the consumption of water used in irrigation recently emerged scientific techniques in the rationing of water quantities that have deteriorated significantly, these techniques adopted to reduce the severity of the harmful effects of water shortage is the use of bio-fertilization of the Mycorrhizae is a fungus with plant roots to increase nutrient uptake and increase the susceptibility of plants to the tolerance of algal stresses It is the most common fungus, penetrating about 95% of the plant roots. The microorganisms have many characteristics that have made it a magnet for researchers (Ikeda & Nakagawa, 2003). The most important is the ability to penetrate the root shell cells easily and have a symbiotic relationship with the plant, so carbon takes the energy source. In contrast, Nitrogen, zinc and manganese. They also play a role in protecting plants from the vital stresses of dehydration, salinity and other factors and reduce the need for plants to water (Levitt, 2015). The microorganisms are re-aggregating soil granules due to their exposure to Globen. It is considered a desirable condition in soil construction. The increase in plant growth rates in Mycorrhiza was observed for non-pollinated plants in water-stressed areas (Wigley *et al.*, 2017).

Noted (Meena, Maurya, Verma & Meena, 2016) that microorganisms play an important role in increasing plants' tolerance to thirst, improving water relationships and increasing root efficiency in the ease of water withdrawal from the soil by changing the structural structure of plant roots by providing the main and secondary nutrients and hormones of dioxins, Gibberellins and cytokines Ethylene, salicylic acid, acidic acid, organic acids, and the secretion of enzymes and clumps that create them in the rhizosphere, the vital exchange zone between roots and soil. Salicylic acid is a non-enzymatic antioxidant and is taken by the plant as a defensive method against the biological and non-biotic stresses. Salicylic acid acts as an adjunct to many antioxidant enzymes such as catalase and peroxidase. It enters the mechanism of action of these enzymes and has a regulatory role for many phytochemical processes of plants Under stress conditions including photosynthesis, respiration and absorption of nutrients and the mechanism of opening and closing of holes and break down the path

of the amino acid ACC, which is the basis in the construction of ethylene and works to increase the activity of non-enzymatic antioxidants Which works to increase the viability of the plant to withstand these stresses and antibiotics is an enzyme super oxide Desmyutz (Super oxide dismutase) enzyme catalase and peroxidase (Stuper-Szablewska, Kurasiak-Popowska, Nawraca³a & Perkowski, 2017).

Materials and methods

The experiment was conducted in the fields of horticulture and garden engineering - Faculty of Agriculture - University of Baghdad from 15-8-2017 to 15-8-2018, it was took small pens 10cm by five-decade equal lengths and lengths of carnation plants class "Fadi" Cultivation of the plant, by exposing the plants to three breaks for irrigation using the weight method and depending on the sensor of the moisture content, Three levels of the first interval, which is watering after two days (D2), second interval is watering after four days (D4) and watering after six days (D6) For the purpose of rationing the quantities and economy of irrigation water, the roots of the plants were fertilized with Mycorrhiza at two levels (20.0g per plant pillow Pad method M0 and M1. The plants were sprayed with salicylic acid with three doses of four concentrates (150, 100, 50.0

 Table 1: Effect of Mycorrhiza mushroom, irrigation interval and salicylic acid in the leaf area (cm²) of clove plant *Dianthus* caryophyllus "Fadi"

MXD	Salicylic acid SA				Irrigation interval	Mycorrhiza	
	SA-150	SA-100	SA-50	SA0	(D)day	М	
658	502	793	810	528	D2 two days	M0(Comparison)	
638	472	785	810	487	D4 four days		
242	230	249	257	235	D6 Six days		
1005	893	1038	1174	915	D2 two days	M1(Mycorrhiza)	
996	886	1037	1159	904	D4 four days		
395	325	441	455	359	D6 Six days		
13.9	27.9				LSD		
	551	724	777	571	Salicylic averages		
		11	1.37		LSD		
		Salicy	lic X Mycorr	hiza			
Mycorrhiza Average	SA-150	SA-100	SA-50	SA0			
513	402	609	626	417	M0		
799	701	839	929	726	M1		
8.0		16	5.1		LSD		
		Salicylic	X Irrigation	interval			
Average Irrigation	SA-150	SA-100	SA-50	SA0			
interval							
832	697	915	992	721	D2		
817	679	911	984	695	D4		
319	278	345	356	297	D6		
9.9		19	9.7		LSD		

mg/l⁻¹) in conjunction with the start of the strains, The field experiment was carried out according to the design of the complete random sectors RCBD of the three factors respectively and three replicates, the mean of the treatments compared by selecting the least significant difference LSD at a significant level of 0.05.

Study Indicators:

leaf area (**Cm**²): It was took 10 leaves of each plant in experimental unit, selected plants and used the program Digimizer Scanner (CARVALHO, Toebe, Tartaglia, Bandeira & Tambara, 2017) The results were calculated according to the following equation:

Leaf area plant (cm^2) = leaf area for leaves rate $(cm^2) \times Number$ of leaves per plant

Chlorophyll in the leaf (Mg 100 g⁻¹ **soft weight):** Taking 1g of leaves full of breadth of the second pair at the beginning of the flowering and estimated the total chlorophyll, extracted the pigments by Crushed 1g dry weight of the leaves in acetone alcohol at 80% concentration using the optical spectrometer spectrophotometer with a wavelength of 645 and 665 nanometers Total chlorophyll = 20.2D(645) + 8.02D(665)(Zhang *et al.*, 2018).

Number of spores (Spor. 10 Cloud¹ Dry soil): calculated depend (Posada, de Prager, Heredia-Abarca,

& Sieverding, 2018).

Assessing the effectiveness of the catalase enzyme (CAT) Catalase (Unit.Mol): The efficacy of CAT enzyme is estimated By spectrophotometer according to the (Arsenov *et al.*, 2017).

Determination of the effectiveness of peroxidase Peroxidase (Unit. Liters): Effectiveness of the enzyme was estimated POD According to the method described by (Belinky, Lasser & Dosoretz, 2017).

Results and discussion

Leaf area (cm²):

The results from Table 1 showed the superiority of the M2 significantly compared with M0 which gave 799cm² in the leaf area, D2 was recorded clear significantly effect compared with the other treatments amounted 832cm². Salicylic acid concentration of 50 mgL⁻¹ showed a clear effect compared with the other treatments amounted 777.0cm². The interactions M1D2 and M1D4 showed a significantly excelled compared with the other treatments in the bilateral interaction between the Mycorrhiza and irrigation intervals reported the 1005 and 996 cm². As for the interaction between the irrigation intervals and Salicylic acid the interaction D2SA50 and D4SA50 had reached the significant level of 992 and 984 cm² respectively, in the

 Table 2: Effect of Mycorrhiza, irrigation interval and Salicylic acid levels on the of chlorophyll content (100 mg⁻¹) for clove *Dianthus* caryophyllus " Fadi "

MXD	Salicylic acid SA				Irrigation interval	Mycorrhiza	
	SA-150	SA-100	SA-50	SA0	(D)day	M	
176.4	160.2	184.0	192.7	168.5	D2 tow days	M0(Comparison)	
163.8	142.8	174.2	185.4	152.9	D4 four days		
116.3	101.5	124.9	134.1	104.5	D6 Six days		
398.9	264.2	472.9	552.7	306.0	D2 tow days	M1(Mycorrhiza)	
378.4	253.7	459.0	547.4	253.6	D4 four days		
164.3	165.2	168.5	175.3	148.0	D6 Six days		
7.1		14	.2		LSD		
	181.3	263.9	298.0	188.9	Salicylic averages		
		5	.8		LSD		
		Sal	icylic X My	corrhiza			
Average Mycorrhiza	SA-150	SA-100	SA-50	SA0			
152.2	134.8	161.1	170.8	142.0	M0		
313.9	227.7	366.8	425.1	235.8	M1		
4.1		8.	2		LSD		
		Salicyl	ic X interval	s Irrigation			
Average intervals	SA-150	SA-100	SA-50	SA0			
Irrigation							
287.6	212.2	328.5	372.7	237.2	D2		
271.1	198.2	316.6	366.4	203.3	D4		
140.3	133.4	146.7	154.7	126.2	D6		
5.0		10).1		LSD		

MXD	Salicylic acid SA				Irrigation interval	Mycorrhiza	
	SA-150	SA-100	SA-50	SA0	(D)day	Μ	
11.20	9.00	11.80	14.10	9.70	D2 two days	M0(Comparison)	
9.90	7.90	10.30	12.60	8.60	D4 four days		
5.90	4.80	6.40	6.50	6.10	D6 Six days		
262.50	185.10	281.10	372.30	211.60	D2 two days	M1(Mycorrhiza)	
242.40	167.10	250.40	369.20	182.90	D4 four days		
33.80	22.00	41.40	44.90	26.80	D6 Six days		
8.63	17.26			LSD			
	66.00	100.30	136.60	74.30	Salicylic averages		
		7.	.04		LSD		
		Salicy	lic X Mycor	rhiza			
Average Mycorrhiza	SA-150	SA-100	SA-50	SA0			
9.00	7.20	9.50	11.00	8.20	M0		
179.60	124.70	191.00	262.20	140.40	M1		
4.98	9.96	LSD					
		Salicy	lic X P hypho	en irrigation			
Average intervals Irrigation	SA-150	SA-100	SA-50	SA0			
136.80	97.00	146.50	193.20	110.70	D2		
126.10	87.50	130.40	190.90	95.80	D4		
19.90	13.40	23.90	25.70	16.50	D6		
6.10		12	.20		LSD		

 Table 3: Effect of Mycorrhiza, irrigation intervals and levels of Salicylic acid in the number of spores of the fungus Almekoraiza effect (spore 10 g⁻¹ soil dry) plant cloves Dinthus caryophyllus "Fadi"

 Table 4: Effect of Mycorrhiza mushroom and irrigation interval and Salicylic acid levels in the efficacy of the enzyme CAT (Unit. Mol) of the clove plant *Dianthus caryophyllus* "Fadi"

MXD	Salicylic acid SA				Irrigation interval	Mycorrhiza	
	SA-150	SA-100	SA-50	SA0	(D)day	Μ	
79.5	89.1	66.4	65.8	96.6	D2 two days	M0(Comparison)	
86.6	100.8	81.4	59.3	105.0	D4 four days		
349.2	343.8	332.9	327.8	392.2	D6 Six days		
56.0	59.9	49.9	46.3	67.9	D2 two days	M1(Mycorrhiza)	
66.4	72.4	57.1	51.8	84.2	D4 four days		
220.1	216.9	204.6	192.8	266.2	D6 Six days		
7.5	15.0				LSD		
	147.2	132.0	124.0	168.7	Salicylic averages		
		6.	.1		LSD		
		Sali	cylic X Myco	rrhiza			
Average Mycorrhiza	SA-150	SA-100	SA-50	SA0			
171.8	177.9	160.2	151.0	198.0	M0		
114.2	116.4	103.9	97.0	139.4	M1		
4.3		8.	7		LSD		
		Salicyl	ic X P hypher	n irrigation			
Average intervals	SA-150	SA-100	SA-50	SA0			
Irrigation							
67.7	74.5	58.1	56.1	82.3	D2		
76.5	86.6	69.2	55.5	94.6	D4		
284.7	280.4	268.8	260.3	329.2	D6		
5.3		10	.6		LSD		

MXD	Salicylic acid SA				Irrigation interval	Mycorrhiza	
	SA-150	SA-100	SA-50	SA0	(D)day	Μ	
81.7	85.9	69.5	63.2	108.4	D2 two days	M0(Comparison)	
83.0	91.5	72.8	60.8	106.8	D4 four days		
191.3	189.0	159.6	151.6	265.1	D6 Six days		
45.4	37.6	36.5	28.0	79.6	D2 tow days	M1(Mycorrhiza)	
57.5	66.2	50.5	47.0	66.5	D4 four days	-	
145.1	142.0	127.6	115.1	195.8	D6 Six days		
4.9		9	.9		LSD		
	102.0	86.1	77.6	137.0	Salicylic averages		
		4	.0		LSD		
			Sal	icylic X Mycor	rhiza		
Average Mycorrhiza	SA-150	SA-100	SA-50	SA0			
118.7	122.1	100.7	91.8	160.1	M0		
82.7	81.9	71.6	63.3	114.0	M1		
2.9		5	.7		LSD		
		•	Salicyl	ic X Irrigation	separator		
Average intervals	SA-150	SA-100	SA-50	SA0			
Irrigation							
63.6	61.7	53.0	45.6	94.0	D2		
70.3	78.9	61.7	53.9	86.7	D4		
168.2	165.5	143.6	133.3	230.4	D6		
3.5		7	.0		LSD		

Table 5: Effect of Mycorrhiza mushroom, irrigation interval and salicylic acid levels in the efficacy of the enzyme POD (1 liter) of cloves *Dianthus caryophyllus* "Fadi "

Triple interaction the leaf area increased in both interaction M1D2SA50 and M1D4SA50 reached 1038 and 1037 cm².

Chlorophyll content: (mg 100 gm⁻¹):

Table 2 shows that bio-fertilizers have a significant effect on chlorophyll content as for Mycorrhiza M1 gave 313.9 mg/100 gm⁻¹. While the D2 (Irrigation after two days) recorded the, the highest percentage of chlorophyll content in the leaves amounted 287.6 mg100g⁻¹. The of salicylic acid significantly effect on the percentage of chlorophyll rate, especially at the concentration of 50mgL⁻ ¹ reached 298.0 effect (mg100g⁻¹) where the interaction M1D2 (Mycorrhiza and irrigate after 2 days) compared with the other treatments amounted to 398.9 mg 100g⁻¹, followed by M1D4 (Mycorrhiza and irrigate after 4 days) amounted 376.4 (100 mg g⁻¹). As for the interaction between irrigation intervals and salicylic acid the interactions D2SA50 and D4SA100 recorded the highest percentage of Chlorophyll content with a significant difference from the rest of the treatments where reported 372.7 and 366.4 (100 mg g⁻¹).

The number of spores for the Mycorrhiza (Spor 10g ⁻¹ **dry soil):** Table 3 shows that the treated of *Glomus mosseae* with Mycorrhiza recorded the highest ratio of number of spores (197.60 spore 10g⁻¹ soil dry) with significantly excelled compared with the other treatments, interval treatment D2 showed significantly excelled where gave 136.80g. 10 spore⁻¹ soil dry, the results showed a significant effect of the spray Salicylic acid concentration of 50 mm.ltr⁻¹ where it appeared an increase in the number of spores of the Mycorrhiza in the soil reached 136.60 spore 10g⁻¹ soil dry, while the double interaction between the Mycorrhiza and the irrigation interval showed a significant effect on the number of spores in the soil, which recorded M1D2 a significantly exceeded compared with the other interactions where it reached 262.50 spore.10g⁻¹ dry soil, the two interactions D2SA50 and D4SA50 between the irrigation interval and the Salicylic acid levels showed a significant increase in the number of spores in the soil with 193.20 and 190.90 pug.10g⁻¹soil.

Determination of catalysis efficacy (unit.): The results of Table 9 indicated a decrease in the efficacy of the enzyme CAT When treated with *Mycorrhiza* M1 Amounted 114.2. As for intervals for irrigation D2 the effectiveness of the catalase was decreased was 67.7 units. The effectiveness of the enzyme was decreased at the concentration of the 50 mg. L⁻¹ and amounted 124.0 units. The effectiveness of the enzyme was decreased when the interaction between the microorganisms and irrigation (M1D2) reached 56.0 units mol, insignificantly

different was recorded compared with the interaction M1D4, as for the double interaction between the irrigation interval and the salicylic acid levels, the catalase enzyme was increased CAT when the D6SA0 amounted to 329.2 units.

Determination of the effectiveness of peroxidase (unit of liters): Table 10 shows that the microorganisms play a large and effective role in reducing the effectiveness of the antioxidant enzymes POD when treated with microorganisms M1 82.7 units per liter, and the effectiveness of the enzyme peroxidase at the interval D2 at 63.6 units per liter. At salicylic acid levels, the enzyme activity decreased in 50 mg Concentration. L⁻¹ reached 77.6 units per liter, The treatment of the interaction between the microorganism and the interval treatment D2 reached 45.5 units per liter. As for the double interaction between the irrigation interval and the levels of salicylic acid, the enzyme's effectiveness was decreased during the treatment D2SA50 reached 45.6 units.

References

- Arsenov, D., M. Zupunski, M. Borisev, N. Nikolic, S. Orlovic, A. Pilipovic and S. Pajevic (2017). Exogenously applied citric acid enhances antioxidant defense and phytoextraction of cadmium by willows (Salix spp.). *Water, Air and Soil Pollution*, **228(6)**: 221.
- Belinky, P., H. Lasser and C. Dosoretz (2017). Methods of producing lignin peroxidase and its use in skin and hair lightening: Google Patents.
- Carvalho, J.O., M. Toebe, F.L. Tartaglia, C.T. Bandeira and A.L. Tambara (2017). Leaf area estimation from linear measurements in different ages of Crotalaria juncea plants. *Anais da Academia Brasileira de Ciências*, 89(3): 1851-1868.

- Ikeda, M. and S. Nakagawa (2003). The Corynebacterium glutamicum genome: features and impacts on biotechnological processes. *Applied microbiology and biotechnology*, **62(2-3)**: 99-109.
- Levitt, J. (2015). *Water, radiation, salt, and other stresses* (Vol. 2): Elsevier.
- Meena, V.S., B.R. Maurya, J.P. Verma and R.S. Meena (2016). Potassium solubilizing microorganisms for sustainable agriculture: Springer.
- Mekonnen, M., A.Y. Hoekstra and R. Becht (2012). Mitigating the water footprint of export cut flowers from the Lake Naivasha Basin, Kenya. *Water resources management*, **26(13):** 3725-3742.
- Posada, R.H., M.S. de Prager, G. Heredia-Abarca and E. Sieverding (2018). Effects of soil physical and chemical parameters, and farm management practices on arbuscular mycorrhizal fungi communities and diversities in coffee plantations in Colombia and Mexico. *Agroforestry systems*, 92(2): 555-574.
- Sairam, R.K., K.V. Rao and G. Srivastava (2002). Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. *Plant Science*, **163**(5): 1037-1046.
- Stuper-Szablewska, K., D. Kurasiak-Popowska, J. Nawraca³a and J. Perkowski (2017). Response of non-enzymatic antioxidative mechanisms to stress caused by infection with Fusarium fungi and chemical protection in different wheat genotypes. *Chemistry and Ecology*, **33(10):** 949-962.
- Wigley, P., S. Turner, C. George, D. Wright, T. Williams, K. Roberts and G. Hymus (2017). Agriculturally beneficial microbes, microbial compositions, and consortia: Google Patents.
- Zhang, J.Y., D.L. Pan, Z.H. Jia, T. Wang, G. Wang and Z.R. Guo (2018). Chlorophyll, carotenoid and vitamin C metabolism regulation in Actinidia chinensis' Hongyang'outer pericarp during fruit development. *PLoS ONE*, **13(3)**: e0194835.