Suad Abd Said Aljalaly et al /J. Pharm. Sci. & Res. Vol. 10(12), 2018, 3286-3290

ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in

The effect of Salicylic acid, fertilizer NPKZn and water stress on the pepper plant *Capsicum annuum* L.

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Abstract

An Experience blastic pots 8 kg capacity in the agricultural season of 2016-2017 was carried out in the green house of Biology Department, College of Education for Pure Science– Ibn AL–Haitham, Baghdad University. The aim of this experiment is to study the effect of NPKZn fertilizer at 160 kg And salicylic acid with concentrations(20,30) mg.L^{-1} In addition to the treatment of control of both factors and their interactions under the effect of water stress (irrigation every day and every 6 days and every 12 days) in some qualities (chlorophyll content and the effectiveness enzyme of (SOD, CAT , POD) (unite mg $protein^{-1}$), and proline content(µg dry $weight^{-1}$)) in the pepper shoot, The design experiment was design of sectors For full randomisation (RCBD) and three replications, the results confirmed that the water stress was significantly increased all studied traits except recipe chlorophyll content has been reduced, fertilization and spraying salicylic acid and reduced their interaction moral qualities of all except chlorophyll content rose compared with the control treatment.

Key words: Salicylic acid , NPKZn fertilizer, water stress

INTRODUCTION

The water stress increases the ethylene and the abscisic acid, which closes the stomata and leaks potassium ions from the cells of the guard and increases the free radicals Reactive oxygen species (ROS), including hydrogen peroxide H2O2 [1]. Salicylic acid is an internal growth regulator that gives partial signals to regulate plant growth. [2]. Systemic Acquired Resistance (SAR) Treatment with salicylic acid and its derivatives has an important role in plant tolerance for abiotic and biotic stress by increasing the resistance of the plant to Systemic Acquired Resistance (SAR) by stimulating or altering the internal paper dissection and giving internal signals to withstand a large number of stresses [3]. Nitrogen is a major nutrient needed by the plant in large quantities. It is very important in the functions it performs[4],. Nitrogen enters the synthesis of amino acids, proteins, enzymes, porphyrins, cytochrome, and some plant hormones such as IAA [5]., Phosphorus enters the building of phospholipids, nucleic acids (RNA and DNA), nuclear proteins [6],. energy compounds, enzymatic correlations and the formation of esters with hydroxyl groups of sugars and alcohols [7]. The potassium component plays a large and important role in most of the vital activities within the plant. [8]., Plays a role in stabilizing cytoplasmic pH (pH) between 7 and 8, activates more than 120 anzema within the plant, And its role in the mechanism of opening and closing the stomata and regulating the plant's osmatic Potential, which increases the efficiency of the plant to withstand water stress and Transmission of photosynthesis process[7]., Zinc plays a clear role in the regulation of hormones in the plant and is an adjunct to the activity of more than 300 enzymes [9]. And it is an important element in the building of nuclear acids and found that 3000 of the upper plant proteins contain zinc [10] ,. And is involved in signal transfer proteins [11]. It is very important to build hormones such as alcohols, gibberellins and cytokines. Zinc enhances the level of antioxidants within plant tissues [12] ,. Zinc plays a vital role in and plant cell protection against oxidizing stresses [13].

MATERIALS AND METHODS

The experiment was carried out in the greenhouse . The experiment was designed according to Randomized Completely Block Design (RCBD) with three replications, It includes 54 pots, Experiment includes three factors NPKZn fertilizer is two levels (Addition of 160 $Kg.H^{-1}$and non-addition), Salicylic acid concentrations (0, 20 and 30) mg.L^{-1}, irrigation periods (every day, every 6 days and every 12 days) and three replicates.Pots stuffed with soil (After being dried , tanned , sifted and weighed). After packing After filling, add 0.64 gm of NPKZn fertilizer,a according to the experimental units, and then planted three seedlings on 4/2/2017(The seedlings were brought from one of the nurseries in Baghdad), and then irrigated to reach (50%) of the field capacity. The watering process was based on the experimental treatments of watering every day, 6 and 12 days. After 34 days of planting on 6/3/2017, the plants were sprayed with two salicylic acid concentrations, prepared after preparation of the stock solution of acid . Total chlorophyll content was estimated in plant leaves after 73 days of seedling cultivation using a chlorophyll meter. Paper samples were taken after 18 days of acid spraying. Prepare leachate from wet leaves to estimate the effectiveness of enzymes

superoxide dismutase (SOD), catalase (CAT) and peroxidase (POD), one gram of cut plant leaves was crushed with 10 cm 3 of 0.1 mL potassium phosphate with cold pH 7.8, after being nominated with a cloth, it was filtered using a 4 ° C cooled centrifuge at 4000 rpm for 30 minutes, according to method [14] . The effectiveness of the SOD enzyme was estimated by the method of Nitro blue tetrazolium (NBT) and riboflavin and by method [15]. The effectiveness of the catalase enzyme was estimated according to method [16]. The effectiveness of the POD enzyme was estimated according to method [17], Determination of protein in leachate for plant extract and prepared by method [18] In the protein estimate. Proline was estimated using the method [19]. The data were statistically analyzed using the least significant difference at the 0.05 probability level to compare the average of the coefficients [24].

RESULTS

The results of Table (1) shown a significant decrease in the average content of chlorophyll content at water stress from 52.87 (spad) when irrigating every day to 36.08 (spad) during irrigation every 12 days**,** Table 2, 3, and 4 indicated that the average Antioxidative enzyme (SOD, CAT and POD) were high in water stress, For each 12-day irrigation, the mean values of the above-mentioned traits increased to 35.36, 36.34 and 38.76 units per gram $protein^{-1}$ compared to 9.44, 6.73, 9.65 and 26.07 (unit 1) Every day in respectively**.** The content of proline, a non-enzymatic antioxidant, increased from 26.07 (µg.gm dry $weight^{-1}$) per day to 38.89(µg dry $weight^{-1}$) at irrigation every 12 days. The results in the tables above showed a difference in the values of the treatments added to the compost by the non-composted treatments.

The treatment of the fertilizer was significantly higher than that for which fertilizer was not added. The average chlorophyll content increased from 42.69 to 44.49. The enzyme antioxidants

(SOD, CAT, POD) and non-enzymatic (proline) decreased from 23.56, 23.79, 23.68 and 33. 60) to (20.93, 16.33, 20.33 and 31.60), respectively.

The results showed in Table 1, 2, 3, 4, 5, and 6 that there were significant differences under the effect of spray treatments with salicylic acid when spraying was increased by 30 $mg.L^{-1}$of acid. The average chlorophyll content increased from 40.01 to 47.38 (spad).while the average properties ((SOD, CAT, POD) (18.61, 15.65, 19.19 and 30.08)was decreasing compared with 25.48, 24.49, 23.99 and 35.02 at zero concentration of acid. For NPKZn fertilizer at a level of 160 $Kg.H^{-1}$ significant effect in reducing the damage of water stress, As the results of the tables (1, 2, 3, 4, 5 and 6) showed that adding fertilizer at irrigation treatment once every 12 days increased the chlorophyll content significantly to 37.27 compared to 34.90 at zero concentration of fertilizer and the same period of irrigation, The addition of fertilizer resulted in a decrease in the average of the antioxidants of the enzyme (SOD, CAT, POD) and non-enzymatic (proline) of 37.28, 45.22, 41.04 and 40.41 (in addition to fertilizer and irrigation every 12 days) to 33.43, 27.46 و 36 47 and 37.37) at (add fertilizer and irrigation once every 12 days), respectively. The reduction of the effects of water stress by salicylic acid was apparent in the results of the above tables. Spraying plants exposed to severe stress, irrigation every 12 days at a concentration of 30 $mg.L^{-1}$ of acid, increased the chlorophyll content to 39.80 compared with 31.80 when the acid was not sprayed and under the same period of irrigation, while the average of the enzyme antioxidants decreased (SOD and CAT (POD) and non-enzymatic (proline) content in this treatment, spraying with smooth acid at a concentration of 30 $mg.L^{-1}$ irrigation every day (12 to 32.89, 30.22, 35.37 and 36.89) compared to 37.69, 42.97, 41.39 and 41.36 Of acid and the same treatment of irrigation. The results indicated in the tables above that the effect of the bilateral interaction between the level of fertilizer and the concentration of salicylic acid was significant and the reason for the increase in the average content of chlorophyll, at the level of fertilizer 160 Kg.H-1 concentration of 30 $mg.L^{-1}$of acid, the average content of chlorophyll 48.27 compared to 39.31 in the treatment of control, and the cause of the effect of the interference of the above-mentioned significant decrease in the averages of antioxidants enzyme(SOD and CAT (POD) and non-enzymatic (proline) content, at the level of compost 160 $Kg.H^{-1}$ and the concentration of 30 mg. L^{-1} of acid were the average of those characteristics (17.34, 13.87, 17.40 and 29.47) compared with (26.87, 29.81, 25.24 and 36.50) when the control treatment. The level of compost was 160 $Kg.H^{-1}$ and acid concentration are effective in reducing the negative impact of water stress, as the interaction between these three factors significantly, as indicated by the results of the tables above, at the level of fertilizer 160 $Kg.H^{-1}$. The concentration of chlorophyll is 40.60 as compared to 30.50 for non-fertilization and not to spray acid and under the irrigation period every 12 days. The values of enzyme antioxidants (SOD, CAT and POD) Enzyme content (proline) decreased from 38.47, 55.70, 43.28, and 43.60 in non-fertilization, acid spraying and irrigation period every 12 days to 30.32, 25.45, 32.53 and 35.56 at fertilization and spraying with a concentration of 30 $mg.L^{-1}$.

The highest value of chlorophyll content was at 30 $mg.L^{-1}$concentration of salicylic acid and the presence of fertilization and irrigation. Each day, the value of the fertilization was low. The highest antioxidant antioxidant values (SOD, CAT and POD) and non-enzymatic (proline) content were not added the fertilizer and not sprayed with salicylic acid and under the irrigation period every 12 days and the lowest values of those qualities At a concentration of 30 $mg.L^{-1}$of silicic acid and with the fertilization and when irrigating every day.

DISCUSSION

The decrease in chlorophyll content (table 1) is due to the fact that water stress increases the ratio of ethylene and abscisic acid, which closes the stomata, leaks potassium ions from the cells of the guard and increases the production of free radicals from the active oxygen group (ROS), mainly H2O2 Plastids, resulting in loss of plastids capacity, reduction in lengths, reduced keratin platelets and degradation of plastids by removal of the phytol part by the effect of Chlorophyllase enzyme, removal of the magnesium atom by Dechelatase-mg and paroxetine degradation by Dioxgenase [1].

The increase in the rest of the traits (Tables 2, 3, 4 and 5) is due to the fact that when the plant is subjected to abiotic stress, the effective oxygen species will increase their production. In order to reduce the destructive effects of these types of oxygen, the plant cells will in turn stimulate the antioxidant system, enzymatic oxidation and non-enzymatic antioxidants) [21].

Proline works as a defensive medium, combining harmful amino acids such as Asparatic acid and Glutamic acid, as well as antioxidants,

Proline has the ability to store excess nitrogen that causes leaf aging. It is nontoxic when collected at high concentrations. Proline is transported from place to place within the plant tissue and provides cells that need protein to build up the amino acids to produce energy during the dry period. The oxidation of each single molecule of proline produces(30) [22]. CAT catalyses hydrogen peroxide into water and oxygen, and oxidizes many hydrogen ions into its donor compounds such as methanol, ethanol, formic acid and phenols [23].

Pyroxidease can also oxidize phenolic compounds, lipids, amines and many other organic compounds and reduce the toxicity of hydrogen peroxide by converting it into water, it reduces the harmful effects of increased ROS [24].

The higher the average chlorophyll content (Table 1) and the lower the antioxidant averages (Tables 2, 3, 4 and 5) is due to the ability of Salsalic acid and its derivatives than the other plant hormones with unique qualities. The plant provides alternative ways of producing energy when the plant is exposed to drought stress through its (Hypersensitive Response)

Systemic Acquired Resistance (SAR) by stimulating the work of genes, changing protein metabolism and stimulating the production of enzymatic antioxidants. Salsalic acid is one of the most important non-enzymatic antioxidants [25].

When exposed to stress and drought, Shikimic acid is converted to Chorismate by Chorismate Synthetase, IsoChorismate, Icsase, Salsalic SA, and Pyruvic Acid by BA2Hase. Salsalic acid plays a role in maintaining the stability of protoplasm and pyrofluoric acid. [26].

The decrease in the above studied traits is due to the fact that the use of fertilizers reduces the effect of salt stress and causes the increase of paper area and the content of chlorophyll and free and related hormones [27].

Table (1) the effect of Salicylic acid , fertilizer NPKZn and water stress in chlorophyll content (spad) in leaves of pepper plant					
Irrigation periods (day)	Salicylic acid concentrations ($mg.L^{-1}$)				Effect the average irrigation periods X level of fertilizer
	Level of fertilizer ($Kg.H^{-1}$)	0	20	30	
Every day	0	48.20	50.33	57.00	51.84
	160	49.02	52.55	60.10	53.89
Every six day	0	39.24	41.21	43.50	41.32
	160	40.00	42.80	44.10	42.30
Every twelve day	0	30.50	35.20	39.00	34.90
	160	33.10	38.10	40.60	37.27
Effect of the average concentrations of salicylic acid		40.01	43.37	47.38	2.37
L.S.D(0.05)		Effect salicylic acid effect			
		The interaction effect trio			
Effect salicylic acid concentrations X irrigation periods					
Irrigation periods		salicylic acid concentrations			Effect the average irrigation periods
		0	20	30	
Every day		48.61	51.44	58.55	52.87
Every six day		39.62	42.01	43.80	41.81
Every twelve day		31.80	36.65	39.80	36.08
L.S.D(0.05)		2.09			1.92
Effect level of fertilizer X salicylic acid concentrations					
level of fertilizer		Salicylic acid concentrations			Effect the average level of fertilizer
		0	20	30	
0		39.31	42.52	46.50	42.69
160		40.71	44.48	48.27	44.49
L.S.D(0.05)		3.28			1.83

Table (2) the effect of Salicylic acid , fertilizer NPKZn and water stress in the effectiveness enzyme of (SOD) (unite mg $protein^{-1}$) in leaves of pepper plant					
Irrigation periods (day)	Salicylic acid concentrations ($mg.L^{-1}$)				Effect the average irrigation periods X level of fertilizer
	Level of fertilizer ($Kg.H^{-1}$)	0	20	30	
Every day	0	14.83	10.06	5.90	10.26
	160	11.16	9.78	4.91	8.62
Every six day	0	27.32	23.86	18.27	23.15
	160	24.18	21.23	16.79	20.73
Every twelve day	0	38.47	37.91	35.45	37.28
	160	36.91	33.07	30.32	33.43
Effect of the average concentrations of salicylic acid		25.48	22.65	18.61	1.21
L.S.D(0.05)		Effect salicylic acid effect			
		The interaction effect trio			
Effect salicylic acid concentrations X irrigation periods					
Irrigation periods		salicylic acid concentrations			Effect the average irrigation periods
		0	20	30	
Every day		13.00	9.92	5.41	9.44
Every six day		25.75	22.55	17.53	21.94
Every twelve day		37.69	35.49	32.89	35.36
L.S.D(0.05)		1.93			1.24
Effect level of fertilizer X salicylic acid concentrations					
level of fertilizer		Salicylic acid concentrations			Effect the average level of fertilizer
		0	20	30	
0		26.87	23.96	19.87	23.56
160		24.08	21.36	17.34	20.93
L.S.D(0.05)		1.80			0.91

Table (3) the effect of Salicylic acid , fertilizer NPKZn and water in the effectiveness enzyme of (Cat) (unite mg protein^{-1}) in leaves of pepper plant

Irrigation periods (day)	Salicylic acid concentrations (mg.L^{-1})				Effect the average irrigation periods X level of fertilizer
	Level of fertilizer (Kg.H^{-1})	0	20	30	
Every day	0	8.84	7.00	4.96	6.93
	160	7.33	6.95	5.32	6.53
Every six day	0	24.88	20.50	12.32	19.23
	160	19.95	14.15	10.85	14.98
Every twelve day	0	55.70	44.96	34.99	45.22
	160	30.23	26.70	25.45	27.46
Effect of the average concentrations of salicylic acid		24.49	20.04	15.65	1.11
L.S.D(0.05)		Effect salicylic acid effect 1.71			
		The interaction effect trio 3.81			
Effect salicylic acid concentrations X irrigation periods					
Irrigation periods		salicylic acid concentrations			Effect the average irrigation periods
		0	20	30	
Every day		8.09	6.98	5.14	6.73
Every six day		22.42	17.33	11.59	17.11
Every twelve day		42.97	35.83	30.22	36.34
L.S.D(0.05)		2.39			0.96
Effect level of fertilizer X salicylic acid concentrations					
level of fertilizer		Salicylic acid concentrations			Effect the average level of fertilizer
		0	20	30	
0		29.81	24.15	17.42	23.79
160		19.17	15.93	13.87	16.33
L.S.D(0.05)		2.52			0.71

Table (4) the effect of Salicylic acid , fertilizer NPKZn and water stress In the effectiveness enzyme of (POD) (unite mg protein^{-1}) in leaves of pepper plant

Irrigation periods (day)	Salicylic acid concentrations (mg.L^{-1})				Effect the average irrigation periods X level of fertilizer
	Level of fertilizer (Kg.H^{-1})	0	20	30	
Every day	0	12.20	11.28	6.63	10.04
	160	11.90	9.86	6.00	9.25
Every six day	0	20.24	21.52	18.14	19.97
	160	16.83	15.34	13.66	15.28
Every twelve day	0	43.28	41.64	38.20	41.04
	160	39.49	37.40	32.53	36.47
Effect of the average concentrations of salicylic acid		23.99	22.84	19.19	3.01
L.S.D(0.05)		Effect salicylic acid effect 2.01			
		The interaction effect trio 5.18			
Effect salicylic acid concentrations X irrigation periods					
Irrigation periods		salicylic acid concentrations			Effect the average irrigation periods
		0	20	30	
Every day		12.05	10.57	6.32	9.65
Every six day		18.43	18.43	15.90	17.62
Every twelve day		41.39	39.52	35.37	38.76
L.S.D(0.05)		1.13			3.21
Effect level of fertilizer X salicylic acid concentrations					
level of fertilizer		Salicylic acid concentrations			Effect the average level of fertilizer
		0	20	30	
0		25.24	24.81	20.99	23.68
160		22.74	20.87	17.40	20.33
L.S.D(0.05)		3.04			1.31

Table (5) the effect of Salicylic acid , fertilizer NPKZn and water stress in proline content(µg.gm dry weight $^{-1}$) in leaves of pepper plant					
Irrigation periods (day)	Salicylic acid concentrations (mg.L^{-1})				Effect the average irrigation periods X level of fertilizer
	Level of fertilizer (Kg.H^{-1})	0	20	30	
Every day	0	30.10	27.22	23.06	26.79
	160	28.25	25.80	22.01	25.35
Every six day	0	35.80	33.16	31.80	33.59
	160	33.27	32.10	30.85	32.07
Every twelve day	0	43.60	40.41	37.22	40.41
	160	39.11	37.45	35.56	37.37
Effect of the average concentrations of salicylic acid		35.02	32.69	30.08	1.00
L.S.D(0.05)		Effect salicylic acid effect 1.81			
		The interaction effect trio 2.79			
Effect salicylic acid concentrations X irrigation periods					
Irrigation periods		salicylic acid concentrations			Effect the average irrigation periods
		0	20	30	
Every day		29.18	26.51	22.54	26.07
Every six day		34.54	32.63	31.33	32.83
Every twelve day		41.36	38.93	36.39	38.89
L.S.D(0.05)		1.87			1.08
Effect level of fertilizer X salicylic acid concentrations					
level of fertilizer		Salicylic acid concentrations			Effect the average level of fertilizer
		0	20	30	
0		36.50	33.60	30.69	33.60
160		33.54	31.78	29.47	31.60
L.S.D(0.05)		1.57			0.83

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