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#### Research article

# Optimizing growth, yield, and water use efficiency of *Allium hirtifolium* with salicylic acid under water stress condiions

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#### ABSTRACT

Water stress significantly limits the growth and productivity of crops, particularly medicinal plants in arid and semi-arid regions. This study explores the use of salicylic acid (SA) as a means to enhance shallot (Allium hirtifolium) resistance to water stress, as well as improve growth, yield, and water use efficiency (WUE) under various irrigation levels. Conducted over three consecutive growing seasons in a field (2016-2019), The study was conducted as split plot based on randomized complete block design with four replications. Irrigation as the main factors included 100 % (full irrigation), 75 % and 50 % of plant water requirement and no irrigation (dryland) and SA foliar application as sub-factors including 0, 0.5, 0.75 and 1 mM in this study. After ensuring uniformity of variance of experimental errors for three years, composite analysis of data was performed for three years. The results of combined analysis of three-years data showed that water stress reduced several growth parameters, including plant height, leaf dimensions, bulb size, onion and grain weight, and yield. However, SA application notably mitigated these adverse effects. Specifically, applying 1 mM SA in dryland conditions, increased the pod diameter and 1000-seed weight by 10.17 % and 19.97 %, respectively. Also, in the condition of 50 % plant water requirement, 1 mM SA enhanced onion dry weight, daughter onion weight and plant height by 12.41 %, 21.68 % and 19.18 % respectively. Furthermore, shallot yield increased by 15.12 % in dryland and by 29.4 % under 50 % of the plant's water requirement with 1 mM SA. The WUE in the treatment of 50 % of the water requirement of the plant and the use of 1 mM SA increased by 19.1 % compared to the non-use. These findings suggest that applying 1 mM SA can be a viable strategy for improving the growth, yield, and water use efficiency of shallot plants under waterstressed environments.

## 1. Introduction

For many years, natural products obtained from medicinal plants have been widely consumed due to their therapeutic properties [1]. The importance of natural products obtained from medicinal and aromatic plants is due to the presence of active compounds such

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as essential oils [2]. Medicinal plants present secondary metabolites that are potential sources for the production of new drugs of natural origin. The secondary compounds of plants have important ecological functions in plants [3]. Shallot (*Allium hirtifolium*) is an Iranian native plant belonging to the Alliaceae family, containing saponins, sapugenins, flavonoids (such as quercetin and camferol) and sulfur compounds such as diallyl thiosolfinate or allicin. The properties of this plant have reported antibacterial, antifungal, antiviral effects [4], liver protection [5], anticancer [6], immunomodulators and wound healing [7,8]. Iranian shallot plant (*Allium hirtifolium*) is one of the most valuable medicinal and industrial plant species, currently at high risk of endangerment due to excessive exploitation.

The severity, frequency, and duration of drought are likely increasing as a result of climate change. Liu et al., 2021 [9] and Kusvuran et al., 2021 [10] and therefore the production and yield of plants will be more affected by water scarcity. Exposure to water stress has been reported to lead to serious physiological and biochemical disturbances [11].

Water stress has a negative effect on plant metabolic activity and growth by restricting root water availability and uptake [12]. Water stress can cause several molecular, biochemical, physiological and morphological changes in plants El–Metwally et al. 2021 [13], and Salem et al., 2022 [14]. Water deficit stress by changing the color of leaves and then their premature aging reduces leaf area index and ultimately decreases photosynthesis and plant yield [15]. It is important to try to find solutions that can reduce the effects of water deficit stress on plant growth and yield and increase plant yield. Nowadays, the use of plant growth regulators to improve the growth, yield and quality of medicinal plants is under study. Plant hormones can be considered as a powerful and sustainable tool in reducing the adverse effects of stress on plant growth and metabolic activities.

SA and its derivatives are among the compounds that act as plant growth regulators and affect various and extensive processes such as germination, ion uptake and transport, membrane permeability and photosynthesis in plants [16]. SA plays a key role in increasing the production of secondary metabolites of pharmacological value, and by speeding up the formation of secondary metabolites, it decreases the time to reach large amounts of metabolites [17]. The endogenous biosynthesis of SA, as a plant hormone [18], is affected by various stresses, including water stress [19]. This regulator can reduce plant susceptibility to environmental stresses by stimulating plant defense mechanisms, regulating opening and closing openings and transpiration rates, and improving plant photosynthesis rate [20]. SA is a water-soluble phenolic compound and antioxidant that is an important messenger to activate plant defense systems against living and non-living stresses. Its effectiveness in enhancing plant tolerance to various stresses has been well-documented [16, 21,22].

The external application of SA facilitates growth, regulates photosynthesis and increases the activity of enzymatic and non-enzymatic antioxidants, particularly under adverse environmental conditions [23,24]. Reports indicate that external SA consumption improves growth, increases water stress tolerance and It can also reduce reduces the harmful effects of oxidative stress at different stages of plant growth [21,25].

In several studies, the effect of SA on the production of many bioactive compounds in medicinal plants has been confirmed [26,27]. Foliar spraying of 1.5 mM SA in Echinacea purpurea plant increased the growth of both aerial organs and roots under water stress conditions [28]. In another study, foliar application of corn plants with SA under water stress increased plant dry weight, plant height, leaf number, leaf area and chlorophyll content [29]. Research has shown an increase in grain yield in SA application on soybeans Razmi et al., 2017 [30], millet Kolupaev et al., 2011 [31] and sunflower Hussain et al., 2009 [32]. Positive effects of SA on growth and yield of rosemary and lemon under water stress have also been reported [21]. Foliar application of SA in rapeseed under water stress conditions increased grain yield of rapeseed by 31 % [33]. Also, in another study, foliar application of SA significantly increased the number of pods per canola plant [34]. It seems that by foliar application of SA, water balance in the plant is maintained and the conditions for inoculation of more flowers are provided, resulting in an increase in the number of reproductive units in the plant [35]. Application of 1.5 mM concentration of SA increased the number and size of pepper fruits and total produced yield by enhancing stomatal conductance and leaf chlorophyll content under drought stress conditions [36]. According to Kazemi [37], SA (0.75 mM) foliar application improved strawberry growth parameters including leaf area, leaf number and photosynthetic rate. Similarly, Mardani et al. [38], found that increasing SA concentration from 0.5 to 1 mM increased the leaf area and root dry weight of deficit irrigated cucumber plants and according to their final results, spraying SA at 0.75 mM was the best treatment to decrease damages induced by water deficit conditions on cucumber seedlings. Moreover, the application of 2 mM SA showed a notable positive effect on both grain yield and quality. In zoysiagrass, 0.5 mM SA acted as a growth regulator under drought stress by enhancing photosynthetic rate and antioxidant enzyme activities while reducing lipid peroxidation [23].

With climate change and increasing water scarcity, especially in arid and semi-arid regions, farmers are often compelled to irrigate crops with less than optimal water, subjecting plants to water stress and reducing crop yields. Growth regulators can play a crucial role in mitigating these effects. While extensive research has examined the impact of various growth regulators on the morphophysiological traits, growth, and yield of crops, there is limited research on their effects on medicinal plants. Notably, no studies have yet investigated the effects of different concentrations of SA on the growth characteristics of shallot, a valuable medicinal plant. This study hypothesizes that applying SA under water stress conditions can mitigate the negative impacts of water deficit and enhance water use efficiency in shallot. Specifically, the application of appropriate concentration of SA is expected to improve plant height, leaf dimensions, bulb size, dry weight, yield components and finally overall yield under water stress conditions.

## 2. Materials and methods

The present study was conducted in split plot design with four replications in Lorestan province (Aleshtar), latitude  $33^{\circ}$  86 min north and  $48^{\circ}$  and 26' east longitude of  $48^{\circ}$  and 26' minutes east. Land preparation operations including plowing in autumn and two perpendicular disks and using trowel for land leveling before planting was performed. In this research, irrigation levels at four levels of

non-irrigation (dryland), full irrigation (100 %), 75 % and 50 % of plant water requirement were investigated as main factor, and foliar application of SA at four levels of 0 (control), 0.5, 0.75 and 1 mM as sub-factor. Irrigation treatments were applied about 50 days after seeds emergence (simultaneously with emergence of flowering stem). Prior to the implementation of the irrigation treatments, no irrigation was conducted, following the customary practices of the region. Each subplot consisted of 6 rows of planting with a distance of 50 cm and a length of 6 m. The spacing between the plants on the planting lines was 10 cm. The distance between main plots was 2 m, 1 m and 2 m between blocks. Shallot planting was done in November by linear method. Before planting, soil sampling was performed to determine soil physical and chemical properties from 0 to 30 cm depth (Table 1). Based on the results of soil analysis, application of super-phosphate fertilizers (50 kg ha-1), potassium sulfate (100 kg/ha) before planting and nitrogen fertilizer (urea) were applied in two stages (100 kg ha-1) in April and May. To calculate water requirement, daily meteorological parameters recorded in Aleshtar synoptic station were used. Water requirement of shallot plant was determined based on FAO's Penman Monteith method [39]. The meteorological data of the area of the experiment during the growing period of the plant is listed in Table 2 for three years.

Water requirement of plant was calculated by calculating evapotranspiration under standard conditions (ETC) according to relation 1. Reference evapotranspiration and evapotranspiration were obtained from the relationship of 2 and crop coefficient (KC) at the beginning, middle and end of growth. The irrigation cycle was considered constant and the required water volume of each plot was obtained from daily water requirement multiplier (ETC) to the time of irrigation in the area of each plot and considering irrigation efficiency [40]. In the deficit irrigation treatments, the volume of actual water requirement was multiplied by the deficit coefficient of each treatment and the calculated water volume for each plot was determined. Then, water was distributed evenly in each plot using a water tank with a volume meter in its output.

$$ET_c = ET_o \times K_c n \tag{1}$$

$$ET_0 = \frac{0.408 \ \Delta (R_a - G) + y \frac{900}{T + 273} \ U_2(es - e_a)}{\Delta + y(1 + 0.34U_2)} \tag{2}$$

Where  $ET_0$  evapotranspiration reference plant (mmd $^{-1}$ ), KC vegetation coefficient,  $R_a$  net radiation at vegetation surface (MJm $^{-2}d^{-1}$ ), G heat flux into the soil (MJm $^{-2}d^{-1}$ ), T average air temperature at 2 m (°C),  $U_2$  wind speed at 2 m (ms $^{-1}$ ), es saturated vapor pressure (kPa) at 2 m altitude, ea real vapor pressure (kPa) at 2 m elevation,  $\Delta$  steam pressure curve (kPa°C $^{-1}$ ) and y Humidity factor (kPa°C $^{-1}$ ). After the irrigation treatments, SA was applied once during the early stages of flowering stem emergence. To prepare the SA solution treatments (MERK, Germany), the required amount of 0.138 g was accurately measured using a precision scale with an accuracy of 0.001 g to create a 1 mM solution. After complete dissolution, the solution was adjusted to the desired volume with distilled water. For the 0.75 mM and 0.5 mM treatments, 0.103 g and 0.069 g of SA were used, respectively. To ensure the full effectiveness of the treatment, the SA solution was sprayed under windless conditions using a 20-L rechargeable Hardy backpack sprayer manufactured in Denmark. The amount of water used at each irrigation time during different stages of growth of shallot plant for different irrigation treatments during three years of experiment is mentioned in Table 3.

## 2.1. Measurement of agronomic traits

In flowering stage, plant height was measured from the ground to the highest point of the plant using a cloth measuring tape. After emergence stage of flowering stem, the number of leaves per 10 plants was counted and their mean was recorded as number of leaves per plant. The length of leaves was measured from tip to point of intersection with stem and width of leaves from the widest part of the middle leaves using a ruler with an accuracy of 0.1 cm.

In every three years, harvest was carried out after full maturity and about the end of June. To measure the traits, such as the number of capsules per plant, the number of grains per capsule, the weight of 1000 seeds, the number of bulbs per plant and the weight of bulbs, 10 plants were randomly selected from each plot at the plant maturity stage. The average values for each trait were then recorded. To measure the weight of 1000 grains, digital scale with accuracy of 0.001 was used. For measuring the length of onion, the diameter of mother and daughter bulbs and the diameter of the bulb were used with high accuracy.

In order to determine dry weight of onion samples, each plot was separately dried in oven with temperature of 85  $^{\circ}$ C for 72 h and then weighed and expressed on a gram per square meter basis. To measure the grain yield and onion weight of shallots, harvesting was conducted over an area of 10 m<sup>2</sup> after removing two side rows, and half a meter from both the beginning and end of each plot to

**Table 1**Physical and chemical characteristics of the soil of the experiment site during three years.

Texture	Fe	Mn	Cu	Zn	K (av.)	P (av.)	pН	EC	Organic carbon	Organic matter	Sand	Silt	Clay	Year
	mg/kg	3					ds/m		%					
silty clay loam	2.21	4.06	0.48	0.75	242.8	17.9	7.14	1.21	1.52	1.62	18	52	30	2016–2017
Silty clay loam	2.44	3.98	0.66	0.71	255.4	19.1	7.25	1.04	1.38	1.42	13	55	32	2017–2018
Silty clay loam	1.96	4.11	0.59	0.69	247.6	18.7	7.06	1.11	1.42	1.55	20	42	38	2018–2019

**Table 2**Average monthly meteorological data of Aleshtar city meteorology station.

month/Year	Average temperature (oC)	Relative humidity (%)	Maximum wind speed (m $\rm s^{-1}$ )	Rainfall (mm)		
	monthly	monthly	monthly	Maximum	monthly	
22 October 2016–November 20, 2016	12.1	45.9	15	20	12.3	
23 October 2017-November 21, 2017	11.9	44.5	12	20	10.3	
23 October 2018-November 21, 2018	10.3	55	15	20	109.7	
21 November 2016-December 20, 2016	4	57.9	11	30	53.6	
22 November 2017-December 21, 2017	4.9	61.7	17	30	39.7	
22 November 2018-December 21, 2018	6.1	72.8	14	30	123.7	
21 December 2016-January 19, 2017	4.2	66.4	13	20	115.2	
22 December 2017-January 20, 2018	4.6	62.4	16	20	52.1	
22 December 2018-January 20, 2019	3.5	66.2	20	20	126.5	
20 January 2017-February 18, 2017	-0.1	71	10	21	101.1	
21 January 2018-February 19, 2018	4.5	64.8	21	21	81.4	
21 January 2019-February 19, 2019	3.7	68.9	21	21	128.3	
19 February 2017-March 20, 2017	5.4	62.2	18	30	72.9	
20 February 2018-March 20, 2018	8.2	63.8	17	30	43.2	
20 February 2019-March 20, 2019	4.1	69.1	17	30	102.4	
21 March 2017-April 20, 2017	3.3	66.1	14	23	114.1	
21 March 2018-April 20, 2018	12	61.2	21	23	70.7	
21 March 2019-April 20, 2019	9.1	68.2	21	23	319.2	
21 April 2017-May 21, 2017	16.5	55.6	11	28	35.8	
21 April 2018-May 21, 2018	13.4	71.3	23	28	208.5	
21 April 2019-May 21, 2019	13.8	59	98	28	12.4	
22 May 2017-June 21, 2017	19	46.5	10	21	0.1	
22 May 2018-June 21, 2018	19.2	55.5	18	21	6.7	
22 May 2019-June 21, 2019	20.1	51.4	17	21	0	

Table 3 The amount of water consumed by the shallot plant (irrigation and rainfall) in four treatments (I0 = no irrigation, I50 % = 50 %, I75 % = 75 % and I100 % = 100 % water requirement of the plant) during three crop years (2016–2017, 2017–2018 and 2018–2019.

Growth stages	2016-201	7			2017-201	8		2018–2019					
	I100 %	I75 %	I50 %	10	I100 %	I75 %	I50 %	10	I100 %	I75 %	I50 %	10	
	mm				mm				mm				
1	23	23	23	23	22	22	22	22	26	26	26	26	
2	41	41	41	41	28	28	28	28	46	46	46	46	
3	129.5	103	76.7	24	140.4	115.8	91.2	42	120	95	70	20	
4	24	18	12	_	22	16.5	11	_	28	21	14	_	
Total	217.5	185	152.7	88	212.4	182.3	152.2	92	220	185	156	92	

Growth stages: 1. beginning of the growth stage - Emergence 2. Emergence - Flowering stem 3. Flowering stem - Flowering 4. Flowering - Harvest.

account for edge effects. Seeds and bulbs were weighed with an accuracy of 0.01 g. Seed and onion yield were reported as g/m<sup>2</sup>.

#### 2.2. Water use efficiency Measurement

Water use efficiency was measured using relationship 3 and based on the ratio of economic yield to total water consumed for plants under different irrigation treatments [41].

$$WUE = GY/TWU$$
 (3)

In this regard, WUE Water use efficiency in  $kg/m^3$ , GY economic performance in kilograms per hectare and TWU total water consumed in cubic meters per hectare.

#### 2.3. Statistical analysis

After three years of experiments, separate analysis of variance was performed for each year. In the following, the test of variance uniformity of the experimental errors was performed for three years using Hartley's F test and after ensuring the uniformity of variance of the three years test errors, composite analysis of data was performed for three years. Analysis of variance was performed using SAS version 9.1 (SAS Institute Inc., Cary, NC, USA). The mean comparison of each treatment was analyzed using the LSD test at the 5 % probability level. Excel 2013 software was used to draw the charts.

### 3. Results

#### 3.1. Plant height

The results of analysis of variance showed that three interaction effects of SA, irrigation and year, as well as the double interaction effects of SA in irrigation and SA per year on plant height were significant (Table 4). Comparing the mean of treatments, it was found that the highest plant height belonged to full irrigation, and applying water stress and increasing its severity caused a significant decrease in plant height.

Application of different concentrations of SA in water deficit conditions caused some increase in this trait (Table 6), but application of different concentrations of this substance, especially 1 mM under rainfed conditions, significantly increased the height of shallot plant compared to control without SA. Among the studied treatments, the highest plant height (40.25 cm) was related to full irrigation (100 % of plant water requirement) and no foliar application of SA in the second year. The least amount of this trait was assigned to non-irrigation and non-SA treatment in each three years (Table 6). It seems that SA application is more effective on plant height in water deficit conditions (Table 6).

## 3.2. Number of leaf

The results of analysis of variance of the data showed that the simple effects of the years of experiment, irrigation levels, SA, and the double interaction of irrigation levels in SA and irrigation in the years of experiment, were significant on the number of leaf at the probability level of 1 % (Table 4). Comparison of mean data showed that full irrigation and foliar application of shallot plant with SA increased the number of leaf of shallot plant (Table 5). Incidence of water stress and increasing its severity significantly reduced the number of leaves in shallot plant so that the highest number of leaf was observed in plants under full irrigation. Plants under irrigation conditions of 75 % and 50 % of plant water requirement and rainfed conditions were in the next ranks. Among the treatments studied, the highest number of leaf with 5.68 leaves was obtained in full irrigation and foliar application of 1 mM SA and the lowest leaf content with 2.72 leaves in non-irrigation and non-SA application (Table 5).

Comparing the mean of double interaction of irrigation levels in experiment years, it was also found that in every three years of the study, any occurrence of water stress caused a decrease in the number of leaf in shallot plant. The highest number of leaf with 5.65 leaves belonged to irrigation treatment of 100 % of plant water requirement in the first year. The lowest rate of this trait was obtained with 3.3 leaves in plants under non-irrigation conditions (dryland) and in the first year. In general, the trend of leaf number changes under different irrigation leaf was constant in three years and no significant difference was observed between three years (Table 7).

## 3.3. Leaf length and width

The results of analysis of variance showed that the triple interaction effect of SA, irrigation and year on leaf length were significant (Table 4). It was found that in every three years of the research, water stress and increasing its severity significantly reduced leaf length, with full irrigation yielding the longest leaves, followed by 75 % irrigation. Non-irrigated plants had the shortest leaf length, differing significantly from other irrigation levels Additionally, applying SA, especially at 1 mM, under water deficit conditions (no irrigation and 50 % irrigation) increased leaf length, most notably in the third year in dryland plants (Table 6). Also, the results of analysis of variance showed that interaction of irrigation and SA levels on shallot leaf width were significant at the probability level of 1 % (Table 4). Adequate water supply significantly increased leaf width, while water deficit reduced it, with the narrowest leaves observed under rainfed conditions (Table 5). SA applications enhanced leaf width under water deficit conditions, though its effects diminished with increased water availability.

Table 4

Combined analysis of variance of the studied agricultural traits in A. hirtifolium under the influence of different levels of irrigation and foliar application with salicylic acid during three years of experiment (2016, 2017, and 2018).

Source	DF	Bush height	Number of leaf	Leaf length	Leaf width	Pod diameter	Diameter of mother onion	Diameter of daughter onion
Year (Y)	2	64.421**	1.0078**	29.12**	61.72**	8.783**	373.52**	7.81**
Error (year)	9	13.13	0.267	4.489	5.84	0.483	11.59	8.82
Irrigation (I)	3	2205.93**	37.119**	1657.26**	9949.43**	132.51**	8888.97**	1035.909**
Y*I	6	5.102 <sup>ns</sup>	0.333**	1.05 <sup>ns</sup>	5.48 <sup>ns</sup>	0.28 <sup>ns</sup>	67.59**	8.95**
Error I	27	5.278	0.114	2.302	4.78	0.178	33.71	2.25
Salycilic Acid (SA)	3	151.546**	2.85**	40.54**	174.61**	2.753**	165.56**	54.81**
I * S	9	35.204**	0.42**	3.65**	29.199**	0.36**	61.42**	7.07**
Y*SA	6	11.42**	0.112 <sup>ns</sup>	5.118**	3.39 <sup>ns</sup>	$0.132^{ns}$	40.118 <sup>ns</sup>	3.79**
Y*I*SA	18	5.232*	0.089 <sup>ns</sup>	1.805*	1.902 <sup>ns</sup>	0.088 <sup>ns</sup>	31.083 <sup>ns</sup>	2.19*
Error Total	108	2.63	0.07	0.96	3.13	0.084	22.46	1.23
CV%		5.29	5.98	3.59	4.45	3.14	11.89	6.9

ns, \*, \*\* non-significant, significant at P  $\leq$  0.05 and P  $\leq$  0.01, respectively.

**Table 5**Combined analysis of variance of the studied agricultural traits in *A. hirtifolium* under the influence of different levels of irrigation and foliar application with salicylic acid during three years of experiment (2016, 2017, and 2018).

Source	DF	weight of daughter onion	Dry weight of onion	Onion yield (m²)	Number of capsules per bush	Number of grains per capsule	Thousand seed weight	Grain yield	Water use efficiency
Year (Y)	2	0.24**	8208.21**	318115.09**	1209.59**	1.089**	2.93**	1.53**	1.68**
Error(year)	9	0.14	454.49	623.33	58.066	0.066	0.107	0.05	0.032
Irrigation	3	184.45**	1443745.48**	11265507.08**	26442**	7.719**	93.048**	25.96**	25.284**
(I)									
Y*I	6	0.116**	630.729*	41552.17**	467.27**	0.054**	0.144 <sup>ns</sup>	0.078*	0.195**
Error I	27	0.37	242.25	567.02	98.506	0.012	0.172	0.038	0.015
Salycilic	3	0.844**	1450.804**	8196.77**	561.89**	0.137**	3.216**	0.6**	3.888**
Acid									
(SA)									
I * S	9	0.196**	229.490**	777.63 <sup>ns</sup>	64.053 <sup>ns</sup>	0.027**	0.484**	0.034**	0.050*
Y*SA	6	0.015 <sup>ns</sup>	41.786 <sup>ns</sup>	1325.08 <sup>ns</sup>	69.739 <sup>ns</sup>	0.017 *	0.051 <sup>ns</sup>	0.036*	2.066**
Y*I*SA	18	0.611**	127.87*	765.45 <sup>ns</sup>	65.436 <sup>ns</sup>	0.013*	0.113 <sup>ns</sup>	0.0149*	0.031 <sup>ns</sup>
Error Total	108	0.179	68.647	612.63	58.8	0.007	0.081	0.033	0.022
CV%		3.93	2.84	2.62	9.37	3.26	5.28	9.77	2.18

ns, \*, \*\* non-significant, significant at  $P \le 0.05$  and  $P \le 0.01$ , respectively.

Table 6
Comparison of the mean triple interaction of irrigation levels (100, 75, and 50 % of plant water requirement and non-irrigation) and salicylic acid foliar application (non-application, 0.5, 0.75, and 1 mmol) on the studied agricultural traits in shallot.

Experiment t	reatments							
Irrigation levels	Salicylic acid concentration	Leaf number	Leaf width	Pod diameter	Onion yield	Number of grains per capsule	Thousand seed weight	Water use efficiency
		cm	mm	mm	g.m <sup>-2</sup>		g	kg.m <sup>-3</sup>
100 %	0	5.44a	54.25a	11.24 ab	1404a	2.84bc	6.736 ab	6.42bc
	0.5	5.28 ab	53.58a	11.25 ab	1413a	2.85 ab	6.715 ab	6.43bc
	0.75	5.59a	55.25a	10.92b	1418a	2.8 ab	6.79 ab	6.44bc
	1	5.68a	55a	11.47a	1427a	2.87a	6.953a	6.65b
75 %	0	4.38cd	47.92c	9.61d	1211d	2.7c	6.137c	6.54bc
	0.5	4.56cd	48.67bc	9.99cd	1229bc	2.76bc	6.336bc	6.56b
	0.75	4.76c	49.92bc	9.69cd	1244bc	2.79 ab	6.304bc	6.64b
	1	4.82bc	50.17b	10.2c	1272b	2.8 ab	6.551abc	6.85a
50 %	0	3.86ef	26.33f	9.16h	623.2f	2.29f	4.026 fg	4.03ef
	0.5	4.35cd	30e	7.63gh	637.3f	2.31e	4.497ef	4.12e
	0.75	4.27de	31.75e	7.69g	642.3f	2.33e	4.791de	4.15e
	1	4.5cd	35.17d	7.89 fg	806.4e	2.51d	5.179d	4.8e
dryland	0	2.72g	20.75g	7.67g	419.8h	1.93g	3.37h	4.76d
	0.5	3.58f	24.83f	8.23ef	432.6h	1.9h	3.899gh	4.82d
	0.75	3.57f	25.33f	8.27ef	443.8h	1.99 fg	4.107 fg	5.11c
	1	3.65f	26.75f	8.45e	483.3g	2.06f	4.043 fg	4.95cd

Means in each column followed by similar letter(s) are not significantly different at 5 % probability level as determined by the LSD test.

## 3.4. Pod dimeter

The results of analysis of variance showed that the simple effects of the experiment years, irrigation and SA levels and the double interaction effect of SA in irrigation on the pod diameter in shallot plant were significant at the probability level of 1 % (Table 4). Comparing the mean of treatments, it was found that applying water stress and increasing its severity significantly reduced the diameter of the pod diameter. The maximum diameter belonged to full irrigation. Minimum diameter of pod was obtained in plants under water deficit condition (50 % of plant water requirement), which showed a significant difference with other irrigation levels. Application of all SA concentrations in water deficit conditions increased the values of this trait (Table 5). Among the studied treatments, the highest pod diameter (11.47 cm) was related to full irrigation (100 % of plant water requirement) and foliar application of 1 mM SA. The least amount of this trait (7.67 cm) was observed in irrigation treatment with 50 % of plant water requirement and no SA (Table 5).

## 3.5. Diameter of mother onion

Analysis of variance of experimental data showed that simple effects of irrigation and year levels on diameter of mother onion were significant at 1 % probability level (Table 4). The mean comparison showed that occurrence of water stress significantly reduced the

Table 7
Comparison of the mean triple interaction of irrigation levels (100, 75, and 50 % of plant water requirement and non-irrigation), salicylic acid foliar application (non-application, 0.5, 0.75, and 1 mmol) and year (2016, 2017, and 2018) on the studied agricultural traits in shallot.

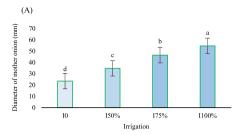
	Irrigation levels	Salicylic acid concentration	Plant height	Leaf length	Diameter of daughter onion	weight of daughter onion	Dry weight of onion	Number of grains per capsule	Grain yield
			cm	mm	mm	g	g.m <sup>-2</sup>		g
Y <sub>1</sub>	100 %	0	36.0а-е	34.64a	19.25bi	5.58 ab	459.4b	2.83d-i	2.00b
		0.5	37.75a-c	34.10a	21.50ac	5.548 ab	467.6 ab	2.85c-h	1.95bc
		0.75	36.75a-d	33.92a	20.50a-f	5.412 ab	469.5 ab	2.9b-f	2.01b
		1	37.75a-c	33.71a	19.25b-i	5.545 ab	464.9 ab	2.92b-e	2.13b
	75 %	0	34.25b-i	27.94b-e	17.25f-l	4.17d	381.9ef	2.66k-m	1.36d
		0.5	33.0c-j	27.31b-g	18.50b-j	4.338cd	383.9ef	2.73i-l	1.41d
		0.75	34.75b-h	27.67b-e	18.75b-j	4.452cd	386.0d-f	2.77g-k	1.39d
		1	34.5b-i	27.85b-e	19.75a-h	4.475cd	388.0d-f	2.82e-i	1.51d
	50 %	0	23.5no	24.34g-l	13.00m-p	2.072i	189.0h-k	2.25q-s	0.28h
		0.5	30.25g-l	24.24h- m	16.25i-m	2.21hi	183.2jk	2.27qr	0.32h
		0.75	29.75i-l	25.28e-j	16.00i-m	2.65ef	186.1h-k	2.29q	0.41gl
		1	30.0h-l	27.09c-h	16.50h-l	2.585e-h	206.9g-j	2.42p	0.4gh
	dryland	0	16.25q	18.00p-q	6.750t	1.108j-k	78.51n	1.91v-x	0.26h
	-	0.5	22.0op	19.85o-q	8.75q-t	1.315j	93.58mn	1.71y	0.78g
		0.75	22.250	20.58op	9.25q-t	1.395j	94.94l-n	1.96u-w	0.77 f
		1	23.25no	20.80n-p	11.0o-r	1.332j	94.93l-n	2.02uv	1.07e
$I_2$	100 %	0	40.25a	34.01a	20.0a-g	5.74a	476.9 ab	2.92bc	1.91c
		0.5	39.0 ab	34.99a	21.75 ab	5.81a	488.2a	2.72i-m	2.00b
		0.75	36.75a-d	35.40a	21.0a-d	5.747a	469.1 ab	2.96b	2.39a
		1	38.50 ab	35.96a	18.25c-j	5.637 ab	485.0a	2.95b-d	1.30e
	75 %	0	35.25b-f	28.98b-d	18.0d-k	4.3cd	419.4c	2.89b-f	1.61c
		0.5	35.0b-g	29.77bc	18.5b-j	4.537cd	406.2c-e	2.95bc	1.30e
		0.75	36.0a-e	29.68bc	19.0b-i	4.648c	409.9cd	2.97a	1.49d
		1	35.75a-f	30.22b	20.0a-g	4.57cd	423.1c	2.89b-g	0.29h
	50 %	0	29.25j-l	23.75i-n	14.25l-o	2.352f-i	198.6g-k	2.48p	0.37h
		0.5	29.0j-m	26.58d-i	16.75g-l	2.175i	211.2gh	2.5op	0.45g
		0.75	29.25j-l	26.50d-i	17.25f-l	2.625e-g	217.3g	2.52op	0.44g
		1	32.25d-j	27.65b-e	17.25f-l	2.888e	221.5g	2.67k-m	0.70g
	dryland	0	17.25pq	20.91n-p	7.75r-t	0.712k	97.93l-n	2.27tu	0.81 f
		0.5	23.75no	21.42l-o	10.0p-t	1.28j	111.3lm	2.14th	0.79f
		0.75	24.25m-o	21.31m- o	11.5n-q	1.33j	113.2lm	2.15r-t	1.09e
		1	27.25k-n	21.65k-o	12.0n-q	1.365j	120.01	2.22q-s	1.91c
$l_3$	100 %	0	34.25b-i	33.39a	20.0a-g	5.27b	477.7 ab	2.73i-l	1.97b
		0.5	36.00a-e	33.65a	20.75a-e	5.67 ab	474.8 ab	2.74h-l	1.99b
		0.75	36.5a-d	35.23a	21.76 ab	5.58 ab	479.6 ab	2.79f-g	2.1b
		1	38.0 ab	35.66a	22.75a	5.645 ab	483.5 ab	2.77h-k	1.39d
	75 %	0	31.00f-l	27.82b-e	15.5j-m	4.225d	378.5f	2.54n-p	1.21e
		0.5	31.25e-l	27.82b-e	16.25i-m	4.332cd	380.5f	2.61m-o	1.48d
		0.75	34.25b-i	28.96b-d	17.5e-l	4.45cd	384.0ef	2.65l-n	1.53d
	E0.0/	1	36.0a-e	30.07b	18.75b-j	4.455cd	392.8d-f	2.7j-m	0.28h
	50 %	0	26.75l-o	24.00i-m	15.5j-m	2.243g-i	179.9k	2.15r-t	0.27h
		0.5	29.25j-l	24.57f-k	14.75k-n	2.04i	185.6i-k	2.17r-t	0.38h
		0.75	29.75i-l	25.72e-i	16.0i-m	2.257f-i	191.1h-k	2.19q-s	0.37h
	المسامية المسام	1	32.00d-k	27.42b-f	16.75g-l	2.635e-g	209.3g-i	2.44p	0.66g
	dryland	0	14.50q	17.13q	7.5s-t	1.12j	90.10mn	1.82xy	0.76 f
		0.5	23.0no	19.97o-q	9.75p-t	1.265j	93.30mn	1.86w	0.75 f
		0.75 1	23.25no 24.25m-o	21.78k-o 22.68j-o	10.75p-s 11.75n-q	1.345j 1.34j	93.98mn 96.13l-n	1.87wx 1.93v-x	1.05e 1.97b

Means in each column followed by similar letter(s) are not significantly different at 5 % probability level as determined by the LSD test.

diameter of mother onion bulb so that the highest diameter of mother bulb (54.63 mm) was obtained in complete irrigation (100 % of plant water requirement) and minimum diameter of mother onion (23.48 mm) in rainfed conditions (Fig. 1A). Fig. 1B also shows that the highest value of mentioned trait (41.7 mm) was obtained in the second year and the lowest value (37.11 mm) in the third year.

## 3.6. Diameter and weight of daughter onion

The three interaction effects of SA, irrigation and year, as well as the double interaction of irrigation levels in SA, SA in experiment years and irrigation levels per year on diameter and weight of daughter onion were significant (Tables 4 and 5). The mean comparison



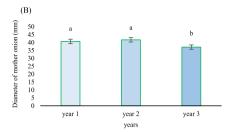


Fig. 1. The simple effect of different levels of irrigation, including I0 = no irrigation, I50 % = 50 %, I75 % = 75 % and I100 % = 100 % on the water requirement of the plant (A) and the years of the experiment, including year 1 = the first year, year 2 = the second year and year 3 = the third year (B) on the diameter of the mother bulb of the shallot plant.

showed that providing enough water for shallot increased the diameter and weight of daughter onion in this plant and the incidence of water stress and its severity significantly reduced the mentioned traits (Table 7). The highest diameter and weight of daughter onion were obtained under full irrigation conditions, which showed significant differences in all application or non-application levels of SA with other irrigation levels. Application of SA especially its concentration of 1 mM in water deficit conditions (dryland and 50 % of plant water requirement) significantly increased the diameter and weight of daughter onion. The results showed that foliar application of 1 mM SA in non-irrigated conditions in every three years of experiment, increased the diameter of daughter onion by 63, 54.8 and 56.6 % respectively and increased the weight of daughter onion by 20.2, 91.7 and 19.6 % in comparison with the control without spraying (Table 7). Although application of different concentrations of SA increased the diameter and weight of daughter onion, however, there was no significant difference between different treatments of SA application or non-application of SA.

Among all treatments studied, the highest diameter of daughter onion (22.7 mm) was allocated to full irrigation (100 % of plant water requirement) and application of 1 mM SA in the third year and the lowest of this trait (6.75 mm) was related to non-SA application under rainfed conditions and in the first year (Table 7). The highest weight of daughter onion (5.65 g) belonged to complete irrigation (100 % of plant water requirement) and application of 1 mM SA in the third year, and the lowest of this trait (0.71 mm) was obtained in the treatment of non-irrigation (dryland) in the second year (Table 7).

## 3.7. Dry weight of onion

Analysis of variance of the experimental data showed that the triple interaction of the year, irrigation levels and SA use and the double interaction effects of SA at irrigation and irrigation levels were significant in the years of experiment on dry weight of onion (Table 5). The results of the comparison of mean showed that with the incidence of water stress and increasing its severity the dry weight of onion significantly decreased (Table 7), so that in every three years the lowest value of this trait was obtained in rainfed conditions. Complete irrigation and irrigation with 75 % of plant water requirement had the highest dry weight of onion and their differences with the other two irrigation treatments were quite significant (Table 7).

In every three years of experiment, foliar application with SA under full irrigation and irrigation of 75 % of the plant water requirement had no significant effect on onion dry weight, but application of this substance under rainfed conditions and irrigation level of 50 % of the water requirement of the plant improved the dry weight of onion of shallot plant. In each three years of experiment, application of 1 mM SA concentration in rainfed conditions increased the dry weight of shallot onion by 21 %, 22.5 % and 6.7 % in

**Table 8**Comparison of the average main effects of irrigation levels (100, 75, 50 percent of plant water requirement, and no irrigation) and experimental years (Year 1: 2016–2017, Year 2: 2017–2018, and Year 3: 2018-1019) on agronomic traits and water use efficiency in *A. hirtifolium*.

Experime	ent treatments	Number of leaf	Number of capsules in plant	Number of grains in capsules	Onion yield (g.m²)	WUE (Kg.m3)
Years	Irrigation levels					
Year 1	100 %	5.65a	105.5a	2.88a	1415a	6.52b
	75 %	4.57bc	92.53bc	2.74b	1234b	6.64a
	50 %	4.2cd	85.78cd	2.31d	652.3d	4.12d
	dryland	3.3e	41.74f	1.9f	436.2e	4.88c
Year 2	100 %	5.49a	111.9a	2.89a	1426a	6.68 ab
	75 %	4.92b	92.58bc	2.92a	1243b	6.77a1
	50 %	4.44cd	80.56cd	2.54c	699.4c	4. 2bc
	dryland	3.43e	60.17e	2.15e	451.2e	4.77b
Year 3	100 %	5.36a	101.8 ab	2.75b	1406a	6.36b
	75 %	4.4cd	83.47cd	2.62c	1240b	6.75a
	50 %	4.1d	76.02d	2.24d	680.2c	4.16d
	dryland	3.42e	49.2ef	1.87f	447.2e	4.89c

Means in each column followed by similar letter(s) are not significantly different at 5 % probability level as determined by the LSD test.

comparison to the non-foliar application (Table 7).

### 3.8. Number of capsules per plant

The simple effects of years of experiment, irrigation levels, SA and the double interaction of irrigation levels per year on the number of capsules per plant shallot plant were significant at the probability level of 1 % (Table 5). The number of capsules in Shallot plant decreased dramatically with increasing water deficit levels. The lowest amount of this trait (41.74 capsules) was assigned to plants under rainfed conditions in the first year which had significant differences with other irrigation levels. Complete irrigation significantly increased the number of capsules per shallot plant in each three years compared to the non-irrigation, so that in the first, second and third year of the experiment, full irrigation increased the number of capsules by 153, 86 and 107 percent, respectively (Table 8).

SA increased the number of capsules per plant, so that the application of one mM SA increased the number of capsules per plant by 9.58 % compared to the non-consumption of SA. Other application concentrations of SA increased the number of capsules per plant compared to the insoluble spraying but could not cause a significant difference (Fig. 2).

## 3.9. Number of grains per capsule

The findings indicated that the interaction effects between irrigation levels, SA, and experiment years significantly impacted the number of grains per capsule in shallots (Table 5). Over the three-year study, water stress consistently reduced grain numbers per capsule, with the most grains (2.97 per capsule) observed in the second year under 75 % irrigation and 0.75 mM SA, while the fewest (1.71 per capsule) occurred in the first year under rainfed conditions with 0.5 mM SA (Tables 7 and 8).

Overall, the number of grains per capsule was notably higher under full irrigation and 75 % irrigation compared to rainfed and 50 % water levels, with the reduction under severe water stress most pronounced in non-irrigated plants (Table 6). Specifically, grain numbers per capsule increased by 48.7 % and 45.07 % with 1 mM SA under full and 75 % irrigation, respectively (Table 6). The highest grain count (2.92 per capsule) was recorded with 75 % irrigation in the second year, while the lowest (1.87 grains per capsule) was observed under dryland conditions in the third year (Table 8).

#### 3.10. Thousand seed weight

The variance analysis of data revealed that the main effect of experiment years with the interaction between SA and irrigation levels, significantly impacted the thousand-seed weight of shallot (Table 5). Full irrigation and foliar application of SA notably increased thousand-seed weight, while water stress and its intensification reduced it, with the highest weights observed under full irrigation. Plants receiving 75 %, 50 %, and no irrigation followed in weight rankings (Table 6).

At all irrigation levels, SA application improved seed weight, though significant increases were only seen under dryland and 50 % irrigation conditions compared to the control (Table 6). The highest thousand-seed weight across the three years occurred in the second year (5.648 g), significantly differing from the other years, while the other two years fell within the same statistical group (Fig. 3).

### 3.11. Grain yield

The analysis of variance indicated that the three-way interaction between SA significantly affected shallot grain yield (Table 5). The mean comparison showed that water stress consistently reduced grain yield, with greater stress levels leading to more pronounced decreases. Across the three years, full irrigation resulted in the highest grain yields, followed by 75 % and 50 % irrigation, with the lowest yields in dryland conditions.

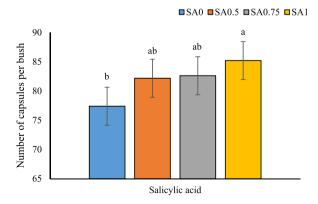


Fig. 2. Effect of salicylic acid concentrations (SA0 = non -use, Sa0.5 = 5.5 mmm, SA0.75 = 0.75 mM, SA1 = 1 mmmular) on the number of capsules per bush.

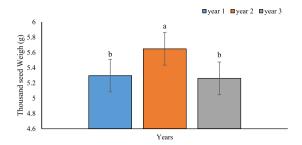


Fig. 3. The simple effect of the years of the experiment, including year 1 =first year, year 2 =second year and year 3 =third year on the Thousand seed weight of shallot plant.

SA application in every three years increased grain yield, especially under 50 % irrigation and rainfed conditions, although these increases were generally not statistically significant except in a few cases with 1 mM SA under dryland and 50 % irrigation levels (Table 7). The highest recorded grain yield (2.39 g) was under full irrigation with 0.75 mM SA in the second year, while the lowest yield (0.26 g) was observed under dryland conditions with no SA application in the first year (Table 7).

#### 3.12. Water use efficiency

The results of analysis of variance showed that dual interaction effects of SA application at irrigation, SA per year and irrigation per year on water use efficiency in shallot plant were significant at the probability level of 1 % (Table 5).

Comparing the mean of treatments showed that highest water use efficiency belonged to irrigation treatment of 75 % of plant water requirement and 1 mM SA ( $6.85 \text{ kg/m}^3$ ) and the lowest of this trait ( $4.03 \text{ kg/m}^3$ ) was allocated to treatment of 50 % of plant water requirement and no SA. Application of one mM SA in irrigation treatment of 50 % of plant water requirement increased water use efficiency by 21.09 % (Table 6). The interaction effects of years of experiment in SA showed that all concentrations of spraying in three years significantly improved water use efficiency compared to non-spraying conditions. In every three years of experiment, application of 1 mM SA was significantly superior to other treatments (Fig. 4).

Also, the results showed that in all three experiments, water stress of 50 % of plant water requirement decreased water use efficiency compared to other treatments (Table 9). Among the studied treatments, the highest water use efficiency  $(6.77 \text{ kg/m}^3)$  was assigned to 75 % of plant water requirement in the second year and the lowest water requirement was observed in irrigation treatment of 50 % of shallot plant water requirement  $(4.12 \text{ kg/m}^3)$  in the first year (Table 9).

## 3.13. Yield of onion

The variance analysis results showed that the interaction between irrigation levels and SA and also the interactions between SA and irrigation per year were significant on shallot onion yield (Table 5). Water stress markedly reduced yield, with the highest yields observed under full irrigation, followed by 75 % and 50 % of plant water requirements, and the lowest yields in dryland conditions. At all irrigation levels, applying SA, particularly at a concentration of 1 mM, significantly increased yield, especially under water-deficit

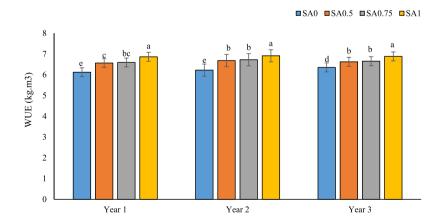


Fig. 4. Double interaction effect of years of experiment (year 1 = first year, year 2 = second year, and year 3 = third year) and concentrations of salicylic acid application (SA0 = no use, SA0 = 0.5 mM, SA0/75 = 0.75 mM, SA1 = 1 mM) on the efficiency of water consumption in shallot plants.

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Table 9
Comparison of the average main effects of salicylic acid application concentrations (SA0 = non-application, SA0.5 = 0.5 mmol, SA0.75 = 0.75 mmol and SA1 = 1 mmol) and experimental years (2016–2017, 2017–2018, and 2018-1019) on agronomic traits and water use efficiency in A. hirtifolium.

Years	Year 1					Year 2 Year 3						
Salicylic acid concentration	0	0.5	0.75	1	0	0.5	0.75	1	0	0.5	0.75	1
Onion yield (g m <sup>-2</sup> )	899.5e	919.8de	928.3cd	989.5b	918.1de	936.5cd	949.8c	1015a	926.3d	927.1cd	933.4cd	986.7b
WUE (g.m3)	6.12f	6.56e	6.59e	6.86b	6.22b	6.686d	6.72c	6.91a	6.35e	6.62e	6.65de	6.88b

Means in each column followed by similar letter(s) are not significantly different at 5 % probability level as determined by the LSD test.

conditions, enhancing yields by 15.12 % under rainfed and by 29.39 % at 50 % water levels (Table 6).

Yearly comparisons also revealed that water stress consistently lowered onion yield, with full irrigation increasing yields by 314–324 % over rainfed conditions across three years. Irrigation at 75 % of water needs raised yields by around 275–283 %, while 50 % irrigation still led to yield improvements of 50–55 % compared to rainfed levels (Table 8).

Additionally, the interaction effect of SA over three years consistently showed an increase in onion yield with rising SA concentrations. The highest yields were recorded with 1 mM SA, while the lowest were in the absence of SA. Over the three years, the 1 mM concentration enhanced yields by 10 %, 11 %, and 7 % compared to untreated plants. Lower SA concentrations (0.75 and 0.5 mM) provided smaller yield increases, 3 %, 3.5 % and 0.7 %, and 2.3 %, 2 % and 0.08 %, respectively (Table 9). Application of SA improves onion yield across all levels of irrigation applications, high concentration 1 mM SA1 significantly enhances yield under prevailing irrigation levels of I50 % and I75 %. This corresponded to the maximum values recorded for the treatment of 1 mM SA under both optimum I100 % and moderately stressed I75 % irrigation levels. Such a combination was, therefore, effective in minimizing the adverse effect of water stress on yield (Fig. 5). On the other hand, without SA application, SA0, a reduced yield was noted in all irrigation levels, ensuring the positive role of SA in enhancing water use efficiency and yield under water-limiting conditions (Fig. 5).

#### 4. Discussion

### 4.1. Yield

Water stress significantly impacts yield, especially with the intensifying effects of climate change [42]. Under water stress, reductions in plant water potential, weakens cell turgor, disrupts membrane integrity, leads to protein and pigment degradation, and damages chloroplast membranes through photooxidation and stomatal closure. Consequently, chlorophyll content decreases [43], photosynthesis diminishes [44], and cell growth is inhibited, ultimately reducing yield [45]. The decrease in grain yield under drought stress conditions largely stems from the detrimental impacts of stress on both vegetative and reproductive growth, as stomatal closure and reduced turgor pressure impair photosynthesis, nutrient uptake, and transport processes, ultimately affecting plant development and yield [46]. Similar reductions in grain yield due to water stress have been documented, particularly in fennel, by Fakheri et al. [47], and Mohtashami et al., [48]. It is also notable that the severity of yield reduction is directly related to the intensity of water shortage; in this study, the impact on growth and yield under water stress was especially marked. SA application appears to mitigate these adverse effects in shallot under water deficit, likely due to its role in activating plant defense mechanisms against both water and oxidative stress. Specifically, SA has been observed to enhance osmotic regulation and maintain plant water content (data not shown) [49]. This action supports chlorophyll retention (data not provided), leading to improved growth of photosynthetic tissues under stress, which ultimately increases photosynthesis and the allocation of assimilates within the plant. Eraslan et al., 2007 [50] observed similar findings, showing that SA application preserved leaf moisture content, thereby sustaining photosynthetic activity and mitigating water stress's negative effects on growth. This study's results align with such findings, underscoring SA's potential in enhancing shallot productivity under drought conditions.

## 4.2. Yield components

Leaf area development is crucial for enhancing photosynthesis and dry matter yield, making it essential to achieve optimal leaf area under varying environmental conditions [51]. In the present study, water stress resulted in a decrease in the number, length, and width of leaves, which aligns with the findings of Zhang et al., 2005 [52] who reported that water stress diminishes both the number and area of leaves in soybean plants. Similarly, Amiri Deh Ahmadi et al., 2011 [53] observed significant reductions in plant height, leaf count per plant, umbrella count per plant, seed count per plant, and the weight of a thousand seeds in the medicinal plant fennel under water stress. The insufficient photosynthetic materials necessary for embryonic growth and seed development appears to be a primary factor contributing to the decrease in the number of seeds produced under such conditions. This reduction in seed count ultimately leads to diminished seed yield in plants [54]. The authors contend that during water stress, certain flower buds do not receive adequate carbohydrates for seed development and filling, which inevitably results in fewer seeds per plant [55].

The results of this study are consistent with research on oregano and fennel, which indicates that water stress adversely affects plant height, leaf area, and the number of capsules per plant [56,57]. Additionally, water stress led to a decrease in the diameter and weight of shallot bulbs, reflecting an economic yield loss. A common consequence of water deficit stress is the reduction in both fresh and dry weight of plants [51], which can be attributed to stomatal closure, along with reduced rates of photosynthesis and transpiration [51]. This phenomenon negatively impacts crop growth and productivity [58]. Following water stress, stomatal closure occurs, which is associated with a corresponding decline in photosynthesis and water-use efficiency [59]. The closing of stomata reduces photosynthesis and impairs the plant's ability to absorb water and essential nutrients from the soil, resulting in both water scarcity and nutrient deficiencies [51].

One study demonstrated that water stress decreased the photosynthesis rate due to a decline in the relative water content of leaves, leading to stomatal closure, reduced transpiration rates, decreased stomatal conductance, and a reduction in photosynthetic pigment content, ultimately resulting in decreased biomass and seed yield in urban balloon plants [60]. The application of certain growth regulators can significantly mitigate the effects of environmental stress on plants, enhancing their growth status [24]. SA is a multimodal regulator that participates in a wide range of growth, metabolic, and defense processes. This plant hormone modulates various plant responses and enhances plant immunity against diverse stresses [24]. In the present study, SA application effectively alleviated the adverse effects of water stress on shallot growth and yield. Specifically, the application of SA at a concentration of 1 mM

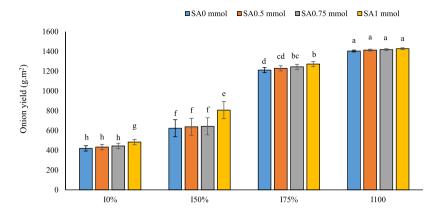


Fig. 5. Double interaction effect of y irrigation levels (I0 = no irrigation, I50 % = 50 %, I75 % = 75 % and I100 % = 100 %) and salicylic acid and concentrations of salicylic acid application (SA0 = no use, SA0 = 0.5 mM, SA0/75 = 0.75 mM, SA1 = 1 mM) on the onion yield in shallot plants.

resulted in increased plant height, leaf length, width, and count, as well as improvements in head diameter, bulb diameter, bulb weight, and overall onion yield under both moderate and severe stress conditions. SA plays a pivotal role in enhancing both fresh and dry weight of plants subjected to water stress [61].

To justify the observed effects of SA on the studied traits, it can be stated that this compound exogenously preserves the integrity of chloroplast and thylacoids [62]. Additionally, SA regulates the photosynthetic system, photosynthetic efficiency, pigment content, and stomatal conductance, thereby promoting the proper progression of plant growth and developmental stages [63]. Beyond its beneficial effects on plant growth under water stress, SA is also a principal plant growth regulator that plays a vital role in modulating photosynthesis during both normal and stressful conditions [24]. The application of SA in water-stressed plants leads to growth recovery, increased photosynthesis, and reduced oxidative stress [64]. This compound significantly contributes to increased photosynthesis by positively influencing photosynthetic processes, enzyme metabolism, and carbohydrate dynamics [65,66]. Increased growth in sunflowers following SA application has been attributed to its role in maintaining photosynthetic activity and ensuring appropriate distribution of photosynthates within the plants. Researchers have noted that foliar application of SA mitigates the negative impacts of water stress by bolstering the antioxidant defense system, stabilizing cell membranes, and enhancing photosynthesis along with physiological and biochemical processes [67].

Previous studies have reported that applying SA under water stress conditions resulted in an increased leaf count in violets and glucsinia [68]. Additionally, enhancements in plant height and leaf count in petunias were observed following SA treatment [69]. The beneficial effects of SA on grain yield and its components under stress conditions are likely linked to an increased transfer of photosynthetic materials to seeds during the grain-filling period, which culminates in an greater in grain weight, consistent with the findings from other researchers [70]. A study examining the impact of SA on maize growth and yield under water stress conditions revealed that this compound increased the number of reproductive units and enhanced seed filling capacity in the plants [71]. Van Hien et al. [72], demonstrated that the application of SA in *Brassica rapa* plant enhances flower count and longevity under water stress by regulating ion uptake through roots, improving stomatal conductance, and increasing osmotic potential. Moreover, this compound may elevate photosynthetic pigment content and optimize photosynthetic processes under stress conditions to maximize water utilization [24]. Other studies have also reported that SA application increases photosynthesis, stomatal conductance, gas exchange, CO<sub>2</sub> uptake rate, and chlorophyll content [73]. In research conducted by Ghasemzadeh and Jaafar 2013 [74], foliar application of different SA concentrations resulted in increased chlorophyll content, photosynthesis, and stomatal conductance under both normal and stress conditions.

## 4.3. Water use efficiency

Research has demonstrated that water stress significantly impacts photosynthetic parameters, including net photosynthesis rate, stomatal conductance, internal carbon dioxide concentration, WUE, and transpiration rate [75]. Under conditions of water stress, stomatal closure occurs gradually, leading to concurrent reductions in net photosynthesis and water use efficiency [59]. The observed low water use efficiency can be attributed to diminished photosynthetic carbon uptake due to reduced  $CO_2$  flow to the mesophyll and stomatal closure [76]. Reducing the number and size of leaves indicates a decrease in the capacity of photosynthetic organs, which ultimately leads to diminished photosynthesis and material accumulation. The current findings align with those of Xu et al. [77], who reported a reduction in the longitudinal growth of the stem of *Zoysia japonica* under various levels of water availability. Additionally, other studies have documented significant decreases in leaf area, head diameter, grain yield, and water use efficiency in sunflowers subjected to water stress [78].

Our results showed that SA application also enhanced water use efficiency. The observed improvement in this trait following SA application may be linked to the compound's ability to enhance the maintenance of relative moisture in plants by improving osmotic adjustment and reducing water loss through the regulation of stomatal conductance. Future studies can consider exploring the effects

of varying concentrations of SA, repeated applications at ten-day intervals, supplementary irrigation during rainy conditions, the use of other growth regulators, and applications at different growth stages of the plant.

#### 5. Conclusion

The results of the three-year study showed that incidence of drought stress reduced the amount of all traits related to the growth and yield of shallot plant. However, application of SA, especially 1 mM concentration, reduced and moderated the effects of stress on the mentioned traits. Application of this concentration of SA, especially in water deficit conditions, improved plant growth and seed and shallot onion yield. The morphological and physiological characteristics of shallots indicate a favorable economic performance when 50–75 % of the plant's water requirements are met. Thus, by enhancing plant resistance through the application of an appropriate concentration of SA during periods of water deficit, it is feasible to achieve acceptable yields while cultivating this plant in water-scarce regions.

## CRediT authorship contribution statement

**Peyman Yousefvand:** Writing – original draft, Software, Methodology. **Yousef Sohrabi:** Writing – review & editing. **Andrea Mastinu:** Writing – review & editing. **Gholamreza Heidari:** Writing – review & editing. **Weria Weisany:** Writing – review & editing, Software, Methodology.

## Data availability statement

Research data collected for this study will be made available if requested by contacting the corresponding author.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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