



In Vitro and in Vivo Evaluating the Efficacy of Salicylic Acid and Gibberellic Acid Against *Fusarium verticillioides* in Garlic (*Allium sativum* L.)

Nebbache Saloua^{1,2*}, Sennoussi Mohammed Mourad^{1,2}, Oufroukh Ammar³

¹ Department of Biology, faculty of exact sciences, natural and life Sciences, Larbi-Ben-M'hidi University of Oum El Bouaghi, BP 358, Constantine Road, 04000, Algeria,

² Laboratory of Biomolecules and Plant Amelioration, Larbi-Ben-M'hidi University of Oum El Bouaghi, BP 358, Constantine Road, 04000, Algeria,

³ National Institute of Agronomic Research of Algeria (INRAA / UR Constantine), 10 rue Ben Mouloud (Ex. Basset), Constantine 25000, Algeria.

ABSTRACT

In Algeria, fungal diseases are the most important biotic limiting growth of garlic (*Allium sativum* L.). The application of salicylic acid is known as a plant hormone that has the signaling role in defence responses, including systemic resistance acquired. Gibberellic acid (GA3) is a very powerful hormone whose natural appearance in plants controls their development. This study aimed to evaluate the affectivity of certain concentrations of salicylic acid (SA) and gibberellic acid (GA3) under laboratory and greenhouse conditions against the phytopathogenic fungus *Fusarium verticillioides* and on the height of garlic (*Allium sativum* L.). The results showed that the inhibitory effect of (SA) on the development of this phytopathogen increased with increasing concentration. In the case of gibberellic acid, the concentrations 100 ppm and 150 ppm were the most inhibitory, its effect was more marked on the fungus during the first three weeks of infection. Inhibition decreases sharply during the fourth week of infection. Gibberellic acid at 150 ppm was an inhibitor over the first, second and third weeks. For morphological parameters, the results showed that the concentration of 150 mg / l recorded the highest values of the height of garlic for both growth regulators. In vitro, the diameter of the colony decreases significantly to 200 mg / l for salicylic acid and 150 mg / l for gibberellic acid. Therefore, our results showed that garlic treatment with (SA) and (GA3) was effective in reducing infection of the disease and increasing yield.

Keywords: *Allium sativum* L, *Fusarium verticillioides*, Salicylic Acid, Gibberellic Acid, Fungal Disease.

Corresponding author: Nebbache Saloua

e-mail ✉ nebachosaloua@yahoo.fr

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1. INTRODUCTION

Garlic (*Allium sativum* L.) is an important vegetable crop which belongs to the genus *Allium* and the family Liliaceae. It is the second most widely used cultivated *Allium* after onion (Bose and Som, 1990). It has been used since ancient times for medicinal and culinary purposes all over the world. It is propagated exclusively by vegetative means (cloves), making it inefficient to multiply and difficult to improve through conventional plant breeding (Harsh et al., 2014). In Algeria, garlic is considered the second crop of bulbs grown after the onion. Progressive growth in productivity occurred between 2010 (6099.3 kg / ha) and 2016 (10971.6 kg / ha) (Faoprds, 2017).

Fusarium is one of the aggressive telluric fungi that cause wilt and rot on many cultivated plant species. *Fusarium* species are described as the common seed-borne fungi of considerable economic importance which could be transmitted by seeds. Pathogens of Seed-borne are known as the first responsible for the reduction of seed growing and germination, and thus can affect production of this crop (Ignjatov, 2016). Lević et al.

(2009) indicated that significant diseases are periodically caused by the *Fusarium* species, especially the wilting diseases on onion, garlic and tomato. It is difficult to control this pathogen due to its persistence in soil and wide host range. Most of the chemicals that have been used in the treatment of wilt disease have gative impacts on the environment and wildlife. For this reason, many studies are being conducted to find alternative solutions such as the use of biotic and abiotic treatments to induce resistance in plant species. Enhancing resistance to disease in tissues far from the induced site is known as the phenomenon of systemic acquired resistance (SAR). This phenomenon is observed in a number of species which is recently subjected to deep reviews (Hammerschmidt, 1999).

Treatment with growth regulators would increase the plant production and productivity (Moon and Lee, 1980). Gibberellic acid plays an important role in bulb development in garlic (Rahim, 1988). GA3 is one of the available compounds that is produced by fungi. GA3 plays crucial roles in different cellular processes such as the elongation of stems, germination of seeds, flowering, fruiting, etc. (Paroussi et al., 2002; Davies, 2005; Tuluze and Celik, 2006).

Salicylic acid (SA) induces responses plant defence (Malamy and Klessing, 1993). Salicylic acid was found in leaves and reproductive organs from the 34 most important cultured

plants, his content increases significantly in infected plants (Raskin, 1992). Induction of acquired systemic resistance by exogenous salicylic acid is a known phenomenon (Friedrich et al., 1993).

2. MATERIALS AND METHODS

Plant materials

The study focused on *Allium sativum*, a highly nutritious and widely consumed foodstuff in Algeria. It is cultivated for easy preservation in the dry state and occupies an area of increasing importance in agro-economy, but its place in the production system is still low compared to other legumes and cereals.

Fungal material

The fungi *Fusarium verticillioides* was isolated in the laboratory of NIAR (National Institute of Agronomic Research-Constantine, Algeria), from different parts of the garlic plant (roots, stems and leaves).

The strain was purified and cultured in Petri dishes on PDA medium (Potato Dextrose Agar) and stored in a refrigerator at 4 ° C. in Petri dishes for the short term and inclined tubes (18 × 180 ml) for long term.

Laboratory bioassay

Standard solution of Salicylic acid and gibberellic acid was prepared. Certain quantities of this solution were mixed up in a milieu of PDA according to the used concentrations.

Salicylic acid (0, 100, 150, 200 mg / l) and gibberellic acid (0, 100, 150, 200, 250 mg / l) were added to the PDA medium. The whole is autoclaved for 20 minutes. 121 ° C. Then, the solutions obtained were distributed on Petri dishes with three repetitions for each concentration. After curing the media, the center of each Petri dish was vaccinated with a 0.5 cm disc taken from the edge of a growing colony of *Fusarium verticillioides*. All plates were cultured at a temperature of 25 ° C. The diameter of the colonies was measured daily until the growth of pathogens reached the edges of the box.

The rate of diameter growth of the fungus pathogen was calculated according to the following equation:

$$\% \text{ of inhibition} = \frac{\text{Growth average of control} - \text{Growth average of treatment}}{\text{Growth average of control}} \times 100$$

Field experiment

Soil preparation

So that our support was light while keeping a good retention of the water we made it of earth clay, sand and potting soil with the proportions 1: 1/2: 1.

The soil-sand-loam mixture was sterilized by autoclaving at a high temperature for 20 minutes.

32 pots were filled with this sterile mixture. The pots were divided into two lots of 16 pots. The batch of plants treated with SA and the batch of plants treated with GA3.

Sterilization of the seeds

For this phase, we took the large uninfected seeds, the selected garlic seeds were sterilized with sodium hypochlorite (0.5%) then washed with distilled water and dried with sterile filter paper for avoid contamination with other pests.

Preparation of salicylic acid and gibberellic acid and seed treatment

Salicylic acid (SA) and gibberellic acid (GA3) were applied in four concentrations (0ppm, 100ppm, 150ppm, 200ppm). Garlic seeds were emerged in each prepared concentration for both hormones for 24 h. Control seeds were emerged in distilled water.

Seed planting

The treated seeds were transplanted into the 32 pots with a seedling per pot, so we had 16 pots for each phytohormone.

The pots were placed in the greenhouse where the conditions were favourable and the irrigation was maintained twice a week at maximum hydration.

Infection of seeds by fungi

When the symptoms of *Fusarium verticillioides* appeared on the plants like yellowing of the leaves and stems, wilting of the plants, the percentage of infection was measured for each treatment compared to healthy plants.

The measured parameters: Measurements were taken weekly.

- The height of the plant (cm) was measured using a long ruler with 4 repetitions per treatment.
- The percentage of the infection: the degree of infection was calculated by the number of control leaves compared to the number of infected leaves for each treatment.

The following equation was used:

$$\% \text{ of infection} = \frac{\text{Degree of infection of control plants} - \text{Degree of infection of treated plants}}{\text{Degree of infection of control plants}} \times 100$$

All results were presented as means of four values.

Statistical processing of data

The statistical significance of the results obtained during this experiment was tested by an analysis of variance carried out using SPSS software. Least significant difference was employed to test for significant difference between treatments at $p \leq 0.05$.

3. RESULTS AND DISCUSSION

1-Laboratory bioassay

A. Effect of salicylic acid on mycelial growth of *Fusarium verticillioides*:

The results in Table 1 showed that the high concentration of SA (200 mg / l) significantly affected the decrease in growth of *Fusarium verticillioides*. The diameter of the colonies was reduced at this concentration compared to the control; this reduction was remarkable from the 3rd day after treatment (1.4 ± 0.00 cm and 1.8 ± 0.00 cm) until the 9th day after treatment by salicylic acid (3.2 ± 0.00 cm and 4.5 ± 0.00 cm). The diameter of the colony revealed the smallest values compared to the control in the 6th day (3.8 ± 0.2 cm and 4.1 ± 0.00cm) and the 7th day after treatment (3.95 ± 0.25 cm), which differed from the control (4.5 ± 0.00 cm) and from 100 mg / l (4.35 ± 0.15 cm). The cause of this inhibition could be attributed to the high concentration of SA in the nutrient medium (PDA). The increase of SA and its accumulation in the

body of living beings could poison them while the SA is transformed into a toxic composition (Glucoside SAG) that could not be excluded by living organisms (Naji, et al., 2012).

The statistics of the results were significant from the 2nd day to the 9th day [(F 2 DAT = 13.4 df = 3, p = 0.001); (F9DAT = 655535, df = 3, p <0.000)].

Table 1. Mycelial growth of *Fusariumverticillioides* under treatment by salicylic acid

SA concentration	half-diameter of colony cm (mean±SE)									
	1 DAT	2 DAT	3 DAT	4 DAT	5 DAT	6 DAT	7 DAT	8 DAT	9 DAT	10 DAT
0 ppm	0,65±0,05 ^a	1,1±0,1 ^b	1,8±0,00 ^c	2,5±0,00 ^c	3,05±0,05 ^{bc}	4,1±0,00 ^c	4,5±0,00 ^a	4,5±0,00 ^a	4,5±0,00 ^a	4,5±0,00 ^a
100 ppm	0,6±0,1 ^a	1,25±0,05 ^b	1,95±0,05 ^{bc}	2,5±0,00 ^{ab}	3±0,00 ^b	4,05±0,05 ^{ab}	4,35±0,15 ^{ab}	4,5±0,00 ^a	4,5±0,00 ^a	4,5±0,00 ^a
150 ppm	0,55±0,15 ^a	1±0,00 ^a	2,45±0,05 ^{ab}	2,9±0,00 ^b	3,35±0,05 ^a	3,8±0,2 ^b	3,95±0,25 ^c	4,3±0,2 ^{bc}	4,5±0,00 ^a	4,5±0,00 ^a
200 ppm	0,45±0,05 ^a	1±0,00 ^a	1,4±0,00 ^a	1,8±0,00 ^a	2,5±0,00 ^{abc}	2,8±0,00 ^a	3±0,00 ^{bc}	3,2±0,00 ^d	3,4±0,00 ^a	4,25±0,25 ^{ab}
P value	NS	0,001	<0,000	<0,000	<0,000	<0,000	<0,000	<0,000	<0,000	NS
F	2,33	13,4	452	65535	298	103,7	64,23	116,75	655535	3
SEM	0,0875	0,0375	0,025	0	0,025	0,0625	0,1	0,05	0	0,0625

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test (P < 0.05). P probability, S.E.M standard error mean, DAT days after treatment.

As an antibiotic against microorganisms, salicylic acid can be characterized as having two main effects on the pathogen. One is the inhibition of hyphal growth, sporulation and conidia germination; another is the stimulation of the production of mycotoxins and enzymes related to pathogenesis (Hong-Sheng, et al., 2008).

In the present study, salicylic acid caused a strong inhibition of hyphal growth. Growth of the mycelium at the highest concentration of salicylic acid (200 mg L⁻¹) in PDA decreased by 33.33% and 28.88 % in the 6th and 7th days respectively (Fig. 1). The result was consistent with the study that salicylic acid with a concentration of 270 mg L⁻¹ exhibited direct fungitoxicity on *Moniliniafructicola* and significantly inhibited mycelial growth of the pathogen in vitro (Yao and Tian, 2005). Plant resistance to biotrophic pathogens is thought to be mediated by salicylic acid signaling (Loake and Grant, 2007). From current results, pathogen-induced salicylic acid not only improves plant resistance to the pathogen, but also inhibits the growth of the pathogen.

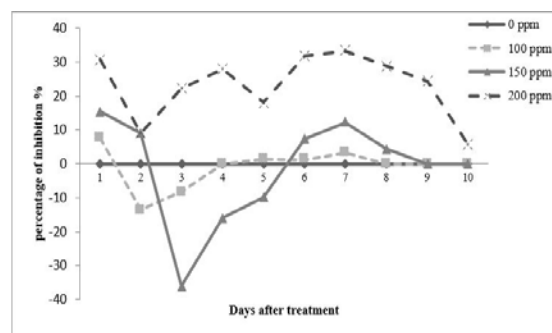


Figure 1. Percentage inhibition of three concentrations of SA on *Fusariumverticillioides*

B. Effect of gibberellic acid on mycelial growth of *Fusariumverticillioides*

Inhibition of *Fusariumverticillioides* growth by gibberellic acid was observed in the 2nd day (1.1 ± 0.5 cm) while control was (1.65 ± 0.05 cm) and in the 3rd day at a concentration of 200 mg L⁻¹ of gibberellic acid (1.5 ± 1 cm) relative to the control (2.55 ± 0.05 cm) (Fig 2). Growth of hyphae (colony diameter) of *Fusariumverticillioides* was stimulated at low concentrations of gibberellic acid (100 mg L⁻¹), then it decreases significantly with the concentration 150 ppm at the 8th day (3.85 ± 0.15) with a half-diameter of 4.1 ± 0, 1 cm (control), while inhibited at the same concentration of gibberellic acid (150 mg L⁻¹) with half-diameter downward from 4.2 ± 0.00 cm (control) to 4.05 ± 0.15 cm on the 9th day after treatment.(Tab 2).The results' statistics in 8th and 9th day were significant [(F8DAA = 11.4, df = 3, p = 0.002); (F9DAA = 21.66, df = 3, p ≤ 0.000)].

Table 2. Mycelial growth of *Fusariumverticillioides* under treatment by gibberellic acid

GA3 concentration	half-diameter of colony cm (mean±SE)									
	1 DAT	2 DAT	3 DAT	4 DAT	5 DAT	6 DAT	7 DAT	8 DAT	9 DAT	10 DAT
0 ppm	0,8±0,00 ^a	1,65±0,05 ^{ab}	2,55±0,05 ^b	2,9±0,2 ^{bc}	3±0,2 ^c	3,4±0,1 ^c	3,85±0,05 ^{bc}	4,1±0,1 ^c	4,2±0,00 ^b	4,5±0,00 ^a
100 ppm	0,8±0,00 ^a	1,6±0,00 ^a	2,5±0,00 ^c	2,65±0,35 ^{abc}	3±0,2 ^b	3,45±0,25 ^{bc}	3,85±0,35 ^c	4,25±0,05 ^c	4,4±0,00 ^b	4,5±0,00 ^a
150 ppm	0,8±0,00 ^a	1,6±0,00 ^a	2,5±0,00 ^c	2,75±0,05 ^{ab}	2,85±0,15 ^{ab}	3,35±0,05 ^{bc}	3,7±0,1 ^{ab}	3,85±0,15 ^{ab}	4,05±0,15 ^{abc}	4±0,5 ^b
200 ppm	0,8±0,00 ^a	1,6±0,5 ^b	1,5±1 ^b	2,9±0,00 ^a	3,2±0,00 ^a	3,65±0,05 ^b	3,9±0,1 ^b	4,25±0,05 ^a	4,5±0,00 ^a	4,5±0,00 ^a
P value	NS	NS	NS	NS	NS	NS	NS	0,002	0,0003	NS
F	4,066	3,19	3,099	1,09	2,41	2,67	0,62	11,4	21,66	3
SEM	0,00	0,1375	0,2625	0,15	0,1375	0,1125	0,15	0,0875	0,0375	0,125

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test (P < 0.05). P probability, S.E.M standard error mean, DAT days after treatment.

Inhibition of gibberellic acid in the first weeks of fungus-infected garlic cultivation decreases with time. This phytohormone inhibited, *in vitro*, the mycelial growth of *Eutypalata* in both solid and liquid medium; this inhibition depends on the concentration and the antifungal effect was obtained from 2 mM. (Amorabé, et al., 2002).

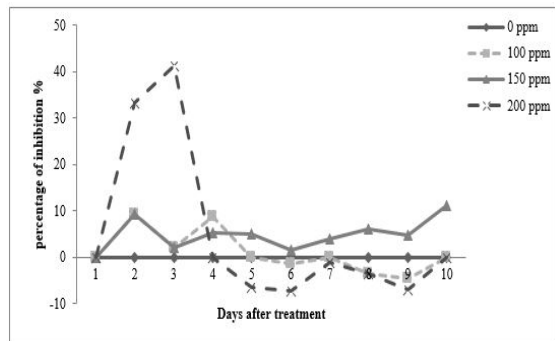


Figure 2. Percentage inhibition of three concentrations of GA3 on *Fusariumverticillioides*

Table 3. Effect of (SA) on the percentage of infection on *Alliumsativum* by *Fusariumverticillioides*

SA concentration	percentage of infection % (mean ± SE)			
	7 DAT	14 DAT	21 DAT	28 DAT
0 ppm	80,55±1,401 ^a	90±5,567 ^c	98,33±1,527 ^c	100±0,00 ^a
100 ppm	37,65±1,522 ^{ab}	46,66±2,081 ^{abc}	50±3 ^b	55,55±1,504 ^c
150 ppm	0±0,00 ^a	12±1,5 ^{ab}	12,23±1,250 ^{ab}	12,23±1,250 ^{bc}
200 ppm	10,55±1,101 ^b	11,75±0,661 ^{ab}	17±1 ^a	10±1,322 ^{ab}
P value	0,000	0,000	0,000	0,000
F	2820,35	434,12	1360,24	3897,63
SEM	1,006	2,452	1,694	1,019

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test ($P < 0.05$). P probability, S.E.M standard error mean. DAT days after treatment.

Phytohormones play an important role in the resistance of plants against diseases bacteria, viruses, fungi and pathogenic algae, this has been achieved by seed treatment. Salicylic acid is considered to be one of the plant hormones (Raskin, 1992) that play an important role in reducing damage from pathogens such as fungi. In addition, SA is an important factor that causes systematic acquired resistance (SAR) against different pathogens (Nie, 2006).

Several studies have shown that SA could stimulate resistance in plants such as *Arabidopsis* (Malamy and Klessing, 1992) and this because SA could stimulate a group of genes responsible for resistance mechanisms in plants (Staskawicz, et al., 1995). The results of this study were consistent with those of (Naji, et al., 2012) who treated legumes with SA and observed that resistance against *Rhizoctoniasolani* was increased. In addition, Bassa, et al. (2016) showed that the percentage of *Fusariumroseum* infection in Chickpea (*Cicerarietinum* L.) was 100% when treated with fungi, whereas these percentages decreased in treated plants by salicylic acid.

SA is well known for its endogenous signal molecule playing an important role in the development of systemic resistance in plants (Dempsey, et al., 1999). SA stimulated systemic

2- Field experiment

A. Effect of salicylic acid on the infection of garlic by *Fusariumverticillioides*

The results showed that the inhibitory effect of salicylic acid on the development of *Fusariumverticillioides* increased linearly with increasing concentration. For the four concentrations, the lowest values of the infection were 0% and 10%, which was observed at the concentration of 150 mg / l in the first week and 200 mg / l in the fourth week compared to the concentrations of 100 and 0 mg / l whose inhibition exerted by salicylic acid in the first days of culture diminishes with time.

This was explained by the efficacy of salicylic acid in the resistance to fungi *Fusariumverticillioides* by limiting its development when *Fusarium* accumulates in vessels of the xylem and prevents the passage of water to different parts causing wilting of the plant (Raskin, 1992).

The analysis of the variance performed on this parameter showed significant differences between the percentage of infection of the plants by the fungus and the four concentrations of salicylic acid (Tab. 3)

resistance against *Fusarium* wilt and greatly reduced the severity of the disease (Saikia, et al., 2003).

A similar response was observed in tobacco plants treated with 100 mM SA (Oostendorp, et al., 2001). The potentiated responses induced by a pathogen have been observed in many plant species pretreated with SA (Metraux, 2001; Zimmerli, et al., 2000)

SAR, biologically and chemically induced in plants, is associated with a capacity of plants to resist the attack of pathogens by an increased activation of cellular defence mechanisms (Metraux, et al., 2002).

B. Effect of gibberellic acid on the infection of garlic by *Fusariumverticillioides*

The best results were observed between the first and second week of the experiment. After three weeks, GA3 become less inhibitory.

For the concentrations used, 100 ppm and 150 ppm were the higher inhibitor. Its effect was more pronounced before three weeks of infection and decreases sharply in the fourth week of infection (12,45% and 20%). The 200 ppm concentration had little action. Gibberellic acid at 150 ppm was relatively inhibitory at the first, second and third week (Tab. 4).

Inhibition of gibberellic acid in the first weeks of fungus-infected garlic cultivation decreases with time.

The variance analysis performed on the percentage infection reveals significant differences between the different

concentrations of gibberellic acid and the plants infected with *Fusariumverticillioides*.

Table 4. Effect of (GA3) on the percentage of infection on *Allium sativum* by *Fusariumverticillioides*

GA3 concentration	percentage of infection % (mean ± SE)			
	7 DAT	14 DAT	21 DAT	28 DAT
0 ppm	43,33±1,522 ^{ab}	50±3,605 ^c	75±4,582 ^{abc}	100±0,000 ^a
100 ppm	34,33±1,527 ^b	41,66±1,755 ^{bc}	41,66±1,755 ^{bc}	60±3,605 ^{ab}
150 ppm	12,45±1,450 ^b	20±3,744 ^{ab}	30,66±0,650 ^b	55,5±4,272 ^{bc}
200 ppm	33,22±3,020 ^a	33,22±3,020 ^b	47,44±2,450 ^a	74,6±4,118 ^b
P value	0,000	0,000	0,000	0,000
F	128,83	49,83	140,01	100,03
SEM	1,88	3,039	2,359	2,999

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test ($P < 0.05$). P probability, S.E.M standard error mean. DAT days after treatment.

Fusarium wilt of plants increased with increasing culture time, after three weeks of culture, control plants (0 ppm GA3) completely withered in soil inoculated with *Fusarium* mycelia. On the other hand, seeds treated with the different concentrations of GA3 and grown in the soil inoculated with fungal mycelia were more resistant to wilt. These results are consistent with those of Al-Awlaqi (2011).

C. Effect of salicylic acid on the height of the plant

For this parameter, there was a remarkable increase at the level of 150 ppm for the plants infected by the fungi *Fusariumverticillioides*. The maximum average values were respectively 12.66 cm and 17.6 cm.

At the 100 ppm level, an estimated maximum value of 13.23 cm was observed during the fourth week of the experiment (Tab. 5).

Variance analysis shows significant differences between plant height and different concentrations of salicylic acid.

Table 5. Effect of (SA) on the height of *Allium*

SA concentration	height of plant cm (mean ± SE)			
	7 DAT	14 DAT	21 DAT	28 DAT
0 ppm	6±0,721 ^a	7,2±0,4 ^b	8,9±0,2 ^c	9,9±0,9 ^{abc}
100 ppm	6,5±0,5 ^a	8,3±0,360 ^{ab}	10±1,322 ^{ab}	13,23±1,132 ^{bc}
150 ppm	8±0,888 ^{ab}	11,33±0,723 ^b	12,66±1,471 ^{ab}	17,6±0,953 ^c
200 ppm	6,16±0,196 ^b	6,43±0,902 ^a	11,46±0,866 ^a	13,73±0,832 ^a
P value	0,017	0,000	0,012	0,000
F	6,271	34,128	6,924	32,296
SEM	0,576	0,596	0,965	0,954

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test ($P < 0.05$). P probability, S.E.M standard error mean. DAT days after treatment

The growth regulators are involved in enhancing photosynthetic activity, efficient assimilation of photosynthetic products and it resulted in rapid cell division and cell elongation in the growing portions of the plant or stimulation of growth besides increasing uptake of nutrients (Singh et al., 2014).

Although plant height is a genetically controlled character in garlic, it can be altered by the use of various plant growth regulators (Nidhish et al., 2014).

Better salicylic acid efficacy in garlic has also been reported by Bideshki and Arvin (2010), which may be due to the accumulation of more chlorophyll content in plants treated with SA, which is responsible for improvement of the accumulation of fresh and dry matter by the final yield of the plant. These results are consistent with (Shuker, 1989).

D. Effect of gibberellic acid on the height of the plant

From our results, we find that there was a gradual increase in this parameter with each increase in GA3 concentration compared to controls up to 150 ppm, a decrease in this parameter was recorded at 200 ppm. The highest value was observed in the 150 ppm GA3 concentration (Tab. 6). The increase in plant height was decreased in the fourth week might be due to increase in temperature during this period and the effect of pathogenic fungus on plants.

At a concentration of 100 ppm, garlic has a mean maximum value of 14.06 cm for the height of the plant, but 14.03 cm was recorded for 200 ppm concentration, for 150 ppm recorded 15.66 cm as a maximum average value.

The variance analysis performed on plant height reveals significant differences between the different concentrations of

gibberellic acid and the height of plants infected with

Fusariumverticillioides

Table 6. Effect of (GA3) on the height of *Alliumsativum*.

GA3 concentration	height of plant cm (mean ± SE)			
	7 DAT	14 DAT	21 DAT	28 DAT
0 ppm	6,43±0,450 ^b	8,66±0,305 ^c	8,16±0,288 ^c	7,66±0,665 ^d
100 ppm	7±0,5 ^b	10,56±0,576 ^{abc}	14,06±0,602 ^b	14,03±0,550 ^c
150 ppm	8±0,4 ^b	9,9±0,838 ^{ab}	15,66±0,556 ^{ab}	15±0,866 ^{bc}
200 ppm	7,3±0,754 ^a	10±0,5 ^a	14,33±0,577 ^{ab}	12,4±0,529 ^a
<i>P</i> value	0,043	0,023	0,000	0,000
F	4,315	5,553	121,2	71,526
SEM	0,526	0,555	0,506	0,652

a, b, c, means, with different superscripts within a same line, are significantly different according to Tukey's HSD multiple range test ($P < 0.05$). *P* probability, S.E.M standard error mean. DAT days after treatment

Our results are similar to those of Gamie, et al. (1996). And it always comes down to the effect of gibberellins that increase the volume and number of plant cells and the composition of organic matter (photosynthesis) in the leaf (Abdul and Harris, 1987), the accumulation and storage of its materials increase the fresh weight of the parameters studied.

GA3 is an important hormone for stimulating plant growth. Similar results have already been reported for tomato (Groot, et al., 1987), pea (Swain, 1997) and cotton (Gokani and Thaker, 2002). In all these cases, gibberellin treatments caused growth increases, and resulted in rapid and improved fruiting of the plants. Gibberellin acts, in fact, by stimulation of meristematic dividing inter-stem internodes stems (Hooley, 1994). This stimulation itself comes from the coordinated effect of two independent actions: induction of meristematic proliferation and cell elongation (Inglese et al., 1998; Richards et al., 2001).

4. CONCLUSION

Our results showed that the concentration 150 mg / l for gibberellic acid records the highest values of plant height. These results are in conformation with the findings of Maurya and Lal. (1975); Bhople et al. (1999); Shakhda and Gajipara (1998); Saleh and Abdul (1980). The same concentration (150 mg / l) for salicylic acid gave the best heights of the plant.

Indeed, the results obtained showed that plants do not respond in the same way at the level of each treatment, with regard to gibberellic acid, the concentrations 100 ppm and 150 ppm were the most inhibitor. Its effect was more marked on the fungus during the first three weeks of infection. Inhibition decreases sharply during the fourth week of infection. The concentration of 200 ppm has little action. gibberellic acid at 150 ppm was an inhibitor relative to the first, second and third weeks. These results are consistent with those of Al-Awlaqi (2011).

Salicylic acid has an inhibitory effect on the growth of fungus studied; this phytohormone is known as a plant hormone that has the signal role in defence responses, including systemic resistance acquired. The results showed that the inhibitory effect of (SA) on the development of *Fusariumverticillioides* increased linearly with increasing concentration.

Based on the current results, we could advise to use salicylic acid and gibberellic acid as a stimulating agent to reduce the

degree of infection by *Fusarium* diseases on the one hand, and on the other hand to increase the yield of garlic by immersing the seeds in the mentioned concentrations for 24 hours before planting.

REFERENCES

1. Abdul KS, Harris GP (1987). Control of flower number in the first inflorescence of tomato (*Lycopersicon esculentum* Mill). *Ann. Bot.* 42:1361-1367.
2. Al-Awlaqi MM (2011). In vitro growth and sporulation of *Fusariumchlamydosporum* under combined effect of pesticides and gibberellic acid. *Research Journal of Agriculture and Biological Sciences*, 7(6): 450-455, 2011
3. Ambarabé BE, Fleurat-Lessard B, Chollet JF, Roblin G (2002). Antifungal effects of salicylic acid and other benzoic acid derivatives towards *Eutypa* sp.: Structure-activity relationship. *Plant Physiol. Biochem*, 40(12), P 1051-1060.2.
4. Bassa N, Senoussi MM, Oufroukh A, Halis Y, Dehimat L (2016). Efficiency of Salicylic Acid in the Reduction of *Cicerarietinum* Infection by *Fusariumroseum*. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*. RJPBCS 7(1) Page No. 416
5. Bhople, S.R. Dod, V.N. Bharad, S.G. Gholap, S.V. and Jadhao, B.J., 1999. Seed production of onion asinfluenced by the application of growth regulators. *J. Soils & Crops*, 9(1): 78-79.
6. Bideshki A, Arvin MJ (2010). Effect of salicylic acid (SA) and drought stress on growth bulb yield and allicin content of garlic in field.
7. Bose TK, Som MG (1990). *Vegetables Crops in India*. NayaPrakash, Calcutta, India.
8. Davies PJ (2005). *Plant hormones, biosynthesis, signal transduction, action*. MA: Kluwer; p. 7.
9. Dempsey DA, Shah J, Klessig DF (1999). Salicylic acid and disease resistance in plants, *Crit. Rev. Plant Sci.* 18:547-575.
10. Faoprds (2017). *Production-statistics-crops-crops-processed*.

11. Friedrich L, Vernooij B, Negrotto D (1993). Requirement of salicylic acid for the induction of systemic acquired resistance. *Science*, vol. 261, pp. 1473-1475.
12. Gamie AA, El Rahim GH, Imam MK, Abdoh AE (1996). Effect of sowing dates on yield and bulb quality in some onion cultivars grown by direct seeding. *Agric. Sci.* 27(2):101-110.
13. Gokani SJ, Thaker VS (2002). Role of gibberellic acid in cotton fiber development. *J. Agric. Sci.* 13: 255-260.
14. Groot SPC, Bruinsma J, Karssen CM (1987). The role of endogenous gibberellin in seed and fruit development of tomato: studies with a gibberellin-deficient mutant. *Physiol. Plant* 71:184-190.
15. Hammerschmidt R (1999). Induced disease resistance: how do induced plants stop pathogens? *PhysiolMol Plant Pathol*; 55:77-84.
16. Harsh DS, Sutanu M, Sanjay K (2014). Influence of plant bio-regulators on growth and yield of garlic (*Allium sativum* L.). *International Journal of Agricultural Sciences*.vol 10. Issue (2) :546-549.
17. Hong-Sheng W, Waseem R, Jia-Qin F, Yong-Gang S, Wei B, Dong-Yang L, Qi-Wei H, Ze-sheng M, Qi-Rong S, Wei-Guo M (2008). Antibiotic effect of exogenously applied salicylic acid on in vitro soilborne pathogen, *Fusarium oxysporum* f.sp. *niveum*. *Chemosphere*. 74, 45-50.
18. Hooley R (1994). Gibberellins: perception, transduction and responses. *Plant Mol. Bio.* 26:1529-1555.
19. Ignjatov M (2016). Occurrence, identification and phylogenetic analysis of *Fusarium proliferatum* on bean seed (*Phaseolus vulgaris* L.) in Serbia. Original scientific paper. *Ratar.Povrt.*, 53 (2) :42-45.
20. Inglese P, Chessa I, La Mantia T, Nieddu G (1998). Evolution of endogenous gibberellins at different stages of flowering in relation to return bloom of cactus pear (*Opuntia ficus - indica* L Miller). *Sci. Hort.* 73: 45-51.
21. Lević J, Stanković S, Krnjaja V, Bočarov-Stančić A (2009). *Fusarium* species: The occurrence and the importance in agricultural of Serbia. *Proceedings for Nature Sciences, Matica Srpska, Novi Sad*, 116 :33-48.
22. Loake G, Grant M (2007). Salicylic acid in plant defence - the players and protagonists. *Curr.Opi. Plant Biol.* 10, 466-472.
23. Malamy J, Klessing, DF (1992). Salicylic acid and plant disease resistance - *Plant J* 1992;2(5): 643-654.
24. Maurya AN, Lal S (1975). Effect of plant growth regulators on the growth and development of onion (*Allium Cepa* L.). *Bangladesh Hort.*, 3(2):11-16.
25. Metraux JP (2001). Systemic acquired resistance and salicylic acid, *Eur. J. Plant Pathol.* 107: 13-18.
26. Metraux JP, Nawrath C, Genoud T (2002). Systemic acquired resistance, *Euphytica* 124: 237-243
27. Moon W, Lee BY (1980). Influence of short day treatment on the growth and level of endogenous growth substances in garlic plants. *J. Korean Soc. Hort. Sci.*, 21: 109-18
28. Naji SJ, Jawadain Talib A (2012). Effect of Salicylic acid (SA) against the fungus *Macrophomina phaseolina* (Tassi) Goid and development of charcoal Rot disease on Sunflower *Helianthus annuus* L. *Basrah J AgrSci*; 25(2): 58-71.
29. Nidhish G, Dharminder K, Ramesh K, Sandeep K, Subhash S, Balbir D (2014). Growth and yield of garlic (*Allium sativum* L) as influenced by clove weight and plant growth regulators. *International Journal of Farm Sciences* 4(3):49-57, 2014
30. Nie X (2006). Salicylic acid suppresses Potato Virus Y Isolate N: O induced symptoms in tobacco plants. *Phytopathology* 96:255-263.
31. Oostendorp M, Kunz W, Dietrich B, Staub T (2001). Induced disease resistance in plants by chemicals, *Eur. J. Plant Pathol.* 107: 19-28.
32. Paroussi G, Voyiatzis DG, Paroussi E, Drogoudi PD (2002). Growth, flowering and yield responses to GA3 to strawberry grown under different environmental conditions. *SciHortic*, 96:103-13.
33. Rahim MA (1988). Control of growth and bulbing of garlic (*Allium sativum* L.). Ph. D Thesis, University of London.
34. Raskin IS (1992). Role of salicylic acid in plants. *Ann Rev. Plant Physiol. Plant Mol. Biol.*, 2:439-463
35. Richards DE, King KE, Ait-Ali T, Harberd NP (2001). How gibberellin regulates plant growth and development: a molecular genetic analysis of gibberellin signaling. *Plant Physiol. Plant Mol.Biol.* 52: 67-88.
36. Saikia S, Singh T, Kumar R, Srivastava J, Srivastava AK, Singh K, Arora DK (2003). Role of salicylic acid in systemic resistance induced by *Pseudomonas fluorescens* against *Fusarium oxysporum* f. sp. *ciceri* in chickpea, *Microbiol. Res.* 158:203-213.
37. Saleh MM, Abdul kS (1980). Effect of gibberellic acid and cycocel on growth, flowering and fruiting of tomato plant. *Mesopotamia J, agric.*, 15:137-161.
38. Shakhda KH, Gajipara NM (1998). A note on influence of IAA, IBA. And GA3 on growth and yield of onion (*Allium cepa* L.) *Veg. Sci.*, 25: 185-186. T
39. Shuker DA (1989). The role of some growth regulators in control of germination and vegetative characteristics, flowering and fruiting in pepper (*Capcium annuum*) plant. M.Sc. Thesis, coll. Of Edu. Salaheddin-Univ. (in Arabic).
40. Singh HD, Sutanu M, Sanjay K (2014). Influence of plant bio-regulators on growth and yield of garlic (*Allium sativum* L.). *International Journal of Agricultural Sciences*. Vol.10 No.2 pp.546-549 ref.16.
41. Staskawicz BJ, Ausubel FM, Baker BJ, Ellis JG, Jones JDG (1995). Molecular genetics of plant disease resistance. *Science*. 5;268(5211):661-7.
42. Swain SM, Reid JB, Kamiya Y (1997). Gibberellins are required for embryo growth and seed development in pea. *Plant J.* 12: 1329-1338.
43. Tuluze Y, Celik I (2006). Influence of subacute and subchronic treatment of abscisic acid and gibberellic acid on serum marker enzymes and erythrocyte and tissue antioxidant defense systems and lipid

- peroxidation in rats. *PesticBiochemPysiol.*86(2):85-92.
44. Yao HJ, Tian SP (2005). Effects of pre- and post-harvest application of salicylic acid or methyl jasmonate on inducing disease resistance of sweet cherry fruit in storage. *Posthar. Biol. Tech.* 35, 253-262.
 45. Zimmerli L, Jakab G, Metraux JP, Mauch-Mani B (2000). Potentiation of pathogen-specific defense mechanisms in *Arabidopsis* by γ -aminobutyric acid, *Proc. Natl. Acad. Sci. U.S.A.* 97, 12920-12925.