

## Research Article

# The antifungal activity of some chemical salts against *Fusarium oxysporum* f. sp. *radicis-cucumerinum* causing cucumber root and stem rot disease

Zinab Mirzadeh Abgarmi<sup>1</sup>, Mousa Najafiniya<sup>2\*</sup> and Hesamodin Ramezani<sup>1</sup>

1. Department of Agriculture, Payame Noor University, PO Box. 193595-3697, Tehran, Iran.

2. Plant Protection Research Department, South Kerman Agricultural and Natural Resources Research and Education Center, AREEO, Jiroft, Iran.

**Abstract:** Cucumber root and stem rot disease caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (*Forc*) is one of the most important diseases of cucumber in Iran. In this regard, this study aimed to investigate the antifungal activity of some chemical salts against *Forc* in *in vitro* and greenhouse conditions. The experiment layout in the *in vitro* condition was based on a factorial experiment using a completely randomized design including three replications. Treatments included potassium sorbate (PS), mono-potassium phosphite, ammonium bicarbonate, salicylic acid (SA), di-potassium hydrogen phosphate (DHP), and fungicide carbendazim (CAR) (Bavistin WP 60%) at 0, 0.5, 2, 4, and 5 g/l. In a greenhouse condition, the experiment was conducted using a randomized complete block design with three replications. The laboratory and greenhouse experiments showed that the treatments had a significant effect on fungal growth inhibition and disease severity. In *in vitro* condition, the highest control of the fungus was attributed to PS and fungicide CAR. Conversely, DHP had the least control (8.74%) on the growth of *Forc* at 0.5 g/l. In a greenhouse condition, CAR prevented disease symptoms for one month. However, PS and SA controlled mycelial growth with an efficiency of 61.19 and 39.2%, respectively. Accordingly, it seems that PS and SA are fungitoxic against *Forc* and can control root and stem rot disease in the greenhouse by root and foliar application.

**Keywords:** disease control, fungicide, induced resistance, integrated management

## Introduction

Regarding cucumber production in the world, Iran ranks third after China and Turkey with a total annual production of 1.6 million tons per year. Cucumber *Cucumis sativus* L. root and stem rot disease caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* Vakalounakis (*Forc*) is a

relatively important disease, which was first reported in Greece. This disease is currently one of the most destructive greenhouse cucumber diseases worldwide (Vakalounakis, 1996; Vakalounakis and Fragkidakis, 1999; Moreno *et al.*, 2001; Karaca and Kahveci, 2010; Garibaldi *et al.*, 2016; Soyly and Incekara, 2017; Yadav *et al.*, 2019). It was reported in British Columbia (10% losses), Canada (25% losses) in 1994, and later in Ontario (35% losses) in 2000 (Yadav *et al.*, 2019). According to Yadav *et al.* (2019), in addition to cucumber, the *Forc* can infect melon *Cucumis melo*, watermelon *Citrullus lanatus*, and

Handling Editor: Naser Safaie

\*Corresponding author: m.najafinia@areeo.ac.ir

Received: 25 February 2020, Accepted: 28 October 2020

Published online: 20 November 2020

sponge gourd *Luffa aegyptiaca*. Based on a report, the disease was observed in the greenhouses of Jiroft, Yazd, and Varamin in Iran during 2002-2003 (Shahriari and Zare, 2006). It was further found that some greenhouses are highly infected in Iran while cultivating susceptible cultivars such as Negin and Royal (Najafiniya and Shahabi, 2019). However, the rate of the disease incidence in Iran was reported to be in the range of 20-60% (Shahriari and Zare, 2006; Yousefi *et al.*, 2010; Najafiniya *et al.*, 2018). According to another study, *Forc* is pathogenic on cucumber and sponge gourd but not on *Cucurbita* spp. (Najafiniya and Mirzadeh Abgarmi, 2020). It has been shown that different *F. oxysporum* pathotype can survive in the soil and above the ground as chlamydospores that are either free or embedded in the infected plant debris (Shlevin *et al.*, 2003). The induction of plant resistance against plant pathogens can be provided using different agents such as chemical salts. In addition, resistance inducers improve plant growth and thus increase the yield (Abdel Monaim, 2010). Evidence suggests that potassium mono-hydrogen phosphate inhibits the growth of *Fusarium solani*, *F. oxysporum*, and *Pythium* sp. at the rate of 4% in the culture medium (Abdel Kader *et al.*, 2012). The antifungal activity of 26 chemical salts against *Fusarium* root rot of onion was investigated and the results showed that ammonium bicarbonate (AB) completely inhibits the mycelial growth (Trukkan, 2013). Further, Stromberge and Brishammar (1991) reported the use of potassium salts (e.g.,  $\text{KNO}_3$  or  $\text{K}_2\text{HPO}_4$ ) as an abiotic agent for inducing resistance against plant pathogens. Also, several reports have demonstrated that chemical salts are promising candidates for controlling fungal diseases. The antifungal properties of various salts against some *Fusarium*-related diseases have been reported in onion, melon, cyclamen, and asparagus as well (e.g., Elmer, 2002; Mills *et al.*, 2004; Arslan *et al.*, 2009). Moreover, Yousefi *et al.* (2010) concluded that salicylic acid (SA) could induce and increase the resistance of cucumber plants against *Fusarium* root and stem rot disease. Considering the above-mentioned findings, the present study was aimed to investigate the antifungal activity of

some chemical salts such as di-potassium hydrogen phosphate (DHP), potassium sorbate (PS), AB, SA, and mono-potassium phosphite (MPP), along with carbendazim (CAR) fungicide (Bavistin 60% WP) against *Fusarium oxysporum* f. sp. *radicis-cucumerinum* causal agent of cucumber root and stem rot disease in laboratory (*in vitro*) and greenhouse conditions in Jiroft, Kerman, Iran.

### Materials and Methods

To this end, the isolates of *F. oxysporum* f. sp. *radicis-cucumerinum* were obtained from the Plant Protection Research Department, South Kerman Agricultural and Natural Resources Research and Education Center, AREEO, Jiroft, Iran according to previous work (Najafiniya *et al.*, 2018). To ensure and obtain active pathogenic isolates, the target isolates were inoculated to cucumber seedlings using the root dip method and pathogen re-isolation in duplicate. For greenhouse experiments, *Fusarium* inoculum was prepared as follows.

Wheat grains were soaked overnight using sterile water and then sifted to remove excess water, and finally, spread on a clean cloth to evaporate the surface water. Next, 100 to 150 grams of wheat grains were poured into 500 ml flasks, and their lid was plugged with the cotton and aluminum foil. Next, the flasks were sterilized in an autoclave for half an hour (at the atmospheric pressure of 1.5 and an approximate temperature of 121 °C). After cooling the contents of the flasks at room temperature, four to five active mycelial discs were cut from seven-day-old *Forc* isolates, added to each flask, and kept at  $25 \pm 2$  °C for 14-21 days to colonize the substrate. Then, the colonized grains were air-dried and blended to make a powder. Eventually, for each 2 kg of the substrate, 75 g of this powder were used for the inoculation of pots (Fallah poor *et al.*, 2013).

### Antifungal activity of chemical salts in *in vitro*

This assay was performed to determine the antifungal activity of the target chemical salts to

control the mycelial growth of *Forc* in *in vitro* as compared to CAR fungicide (Bavistin WP 60%). For this purpose, a five mm (in diameter) disc from the target fungal mycelium was placed on the salt-treated PDA medium in a Petri plate and incubated at  $25\text{ }^{\circ}\text{C} \pm 2$ . Then, the mean diameter of the colony was measured 96 hours after plating using a ruler from the bottom of the Petri plate (Roustaei and Mohammadian, 2005). The following formula was used to measure the control efficiency of the radial growth.

$$\text{Control efficiency (\%)} = \frac{CE_c - CE_t}{CE_c} \times 100$$

Where  $CE_c$  and  $CE_t$  are colony diameter in control and treatment, respectively.

This part of the study was conducted in a factorial experiment based on a completely randomized design with six chemical salts (factor a) in five different concentrations (factor b) for a total of 30 treatments in three replications. In addition, three 9 cm Petri plates containing 12-15 mL PDA medium treated with chemical salts were considered for each experimental plot (Roustaei and Mohammadian, 2005). Chemical salts: DPHP, PS, AB, MPP, SA, and CAR fungicide were used. The recorded data (colony diameter in mm 96 hours after plating) were statistically analyzed, and finally, the control efficiency and mean comparison of all treatments were grouped using Duncan's multiple range tests.

#### Antifungal activity of chemical salts in greenhouse condition

The efficiency of the selected treatments from the *in vitro* experiment was evaluated in reducing the disease severity of the cucumber root and stem rot using the cucumber seedlings of the susceptible Royal cultivar (Najafiniya and Shahabi, 2019) planted in *Forc* inoculated pots (plastic pots 20 cm in diameter and 2-3 kg in capacity) in a randomized complete block design. Transplant production was conducted in transplant trays. Royal cucumber seedlings

were treated with the desired chemical salts while growing up to the two to four true leaf stage. Then, the seedlings were root dipped for 15 minutes at the desired concentration of the target treatments and then transferred to plastic pots according to the modified method of El-Mohamedy *et al.* (2014). The pots contained autoclaved substrate consisting of the river sand, peat moss, and field soil (1: 1: 1) which were inoculated by *Forc* (75 g of inoculum containing spores and mycelium fragments for each 2 kg of the substrate). Three cucumber seedlings were planted per pot, and three pots were considered for each experimental plot. Additionally, the roots of the positive (inoculated with *Forc* but no the chemical salt) and negative (without *Forc* and the chemical salt) controls were immersed in sterile distilled water for 15 minutes and then planted in the pots. After the inoculation, the pots were irrigated and kept in greenhouse condition set at  $23 \pm 3\text{ }^{\circ}\text{C}$ , 65-70% RH and LD 14:10 h. One week after the root treatment with different compounds, the spraying of the experimental plots of cucumber in the greenhouse (3 times) with target treatments were performed once a week, followed by recording the data one week after each spraying. It is noteworthy that the inoculated seedlings were continuously evaluated for the signs of an infection. The applied compounds for treating the roots and spraying cucumber seedlings included DPHP (5 g/l), PS (2 g/l), AB (5 g/l), MPP (5 g/l), SA (5 g/l), and CAR (2 g/l WP 60%), as well as a positive control with the pathogen (distilled water) and negative control without a pathogen (distilled water).

The disease severity index (DSI) was evaluated 60 days after planting and a zero to three scoring system was used in this regard (Vakalounakis, 1996; Pavlou *et al.*, 2002). Healthy plants with no symptoms of the disease, plants with dead and dying leaves, plants with crown and stem rot, and complete plant death received a score of 0 to 32, respectively, and then the following formula was used to calculate the DSI in each plot:

$$Ds = \sum_{i=1}^{ni} \left( \frac{ni \times vi}{N \times V} \right) \times 100$$

Where DS, ni, vi, N, and V represent: disease severity, the number of plants infected with a similar score, the disease score of 0-3 for each treatment, the total number of plants, and the highest disease score, respectively. Finally, the obtained data were analyzed by SAS software and the mean comparison was conducted by Duncan's multiple range tests.

## Results

The results of the analysis of variance in the *in vitro* experiment showed that the effect of CAR fungicide and the chemical salts on the mycelial growth inhibition of *Forc* was significant at the level of 0.01% ( $F = 243.8$ ,  $df = 5, 60$ ,  $p < 0.01$ ). The results further demonstrated that the effect of the applied concentrations and the interaction effect of chemical salts and their concentration were statistically significant (Table 1).

Table 2 presents the mean comparison of the separate effect of different chemical salts (Factor a) and different dosages (factor b) on the colony diameter of *F. oxysporum* f. sp. *radicis-cucumerinum*. Based on the results the mean radial growth rate in the control was

34.3 mm. At 0.5 g/l, the minimum radial growth rate was 0, 0, and 10.3 mm in PS, CAR, and SA after 96 hours of incubation, respectively. Furthermore, the maximum observed radial growth rate was 36.6, 31.3, and 30 mm in AB, DPHP, and MPP, respectively (Table 2). The results revealed that PS and CAR without any mycelial growth represented 100% control efficiency 96 hours after the treatment at 0.5 g/l. Moreover, the control efficiency of SA, MPP, DPHP, and AB was 69.97, 12.53, 8.74, and -6.68%, respectively. The results of the fungal radial growth and control efficiency of different chemical salts are shown in Table 2.

**Table 1** Mean squares of the antifungal effect of different chemical salts on growth rate of *Fusarium oxysporum* f. sp. *radicis-cucumerinum* *in vitro* condition.

Sources of variation	df	MS (colony diameter after 96 h)
Chemical salts (A)	5	1.3762**
Chemical salt dosages (B)	4	2.9240**
A × B	20	0.1932**
Error	60	0.0068
CV = 6.2212		

All data have been transformed, \*\* = significant at level of 0.01.

**Table 2** Mean comparison (main data) of colony diameter and inhibitory efficiency of different chemical salts on *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (96 hours after treatment).

Chemical compounds	0 (g/l)		0.5 (g/l)		2 (g/l)		4 (g/l)		5 (g/l)	
	CD <sup>1</sup>	CE <sup>2</sup>	CD	CE	CD	CE	CD	CE	CD	CE
DPHP	34.3 <sup>ab</sup>	0	31.3 <sup>bcd</sup>	8.74	29.6 <sup>bcd</sup>	13.70	27.7 <sup>cde</sup>	19.53	30.0 <sup>ab</sup>	12.53
AB	34.3 <sup>ab</sup>	0	36.6 <sup>a</sup>	6.68	23.0 <sup>efg</sup>	32.90	27.3 <sup>cde</sup>	20.40	28.0 <sup>bc</sup>	18.36
MPP	34.3 <sup>ab</sup>	0	30.0 <sup>bcd</sup>	12.53	28.0 <sup>cde</sup>	18.36	21.8 <sup>de</sup>	36.44	22.6 <sup>bc</sup>	34.11
SA	34.3 <sup>ab</sup>	0	10.3 <sup>f</sup>	69.97	5.0 <sup>i</sup>	85.42	14.6 <sup>e</sup>	57.43	0 <sup>e</sup>	100
PS	34.3 <sup>ab</sup>	0	0 <sup>g</sup>	100	0 <sup>i</sup>	100	0 <sup>f</sup>	100	0 <sup>e</sup>	100
CAR	34.3 <sup>ab</sup>	0	0 <sup>g</sup>	100	0 <sup>i</sup>	100	0 <sup>f</sup>	100	0 <sup>e</sup>	100

<sup>1</sup> CD: Colony diameter (mm), <sup>2</sup> CE: Control efficiency (%), Means in a column followed by the same letter (s) are not significantly different (Duncan's multiple ranges test  $P < 0.05$ ).

DPHP: Di-potassium hydrogen phosphate, AB: Ammonium bicarbonate, MPP: Mono-potassium phosphite, SA: Salicylic acid, PS: Potassium sorbate, CAR: Carbendazim WP 60%.

The results at 2 g/l showed that the minimum radial growth rate belonged to PS (0 mm) CAR (0 mm), and SA (5 mm) while the maximum radial growth rate was observed in DPHP (29.6 mm). Additionally, the control efficiency of PS, CAR, and SA was 100, 100, and 85.42%, respectively, and that of DPHP at 2 g/l was 13.70% (Table 2). The results at 4 g/l indicated that the maximum amount of control efficiency was related to PS, CAR, and SA with an average of 100, 100, and 57.43%, respectively, whereas the minimum control efficiency was found in DPHP (19.53%). As regards the results at 5 g/l, the maximum amount of control efficiency (100%) was detected in PS, CAR, and SA without any radial growth. However, the minimum control efficiency was found in DPHP, AB, and MPP at the rate of 12.53, 18.36, and 42.85%, respectively.

#### Antifungal activity of chemical salts in greenhouse condition

The results of greenhouse experiments are summarized in Tables 3 and 4. The results of the analysis of variance (Table 3) showed that the effect of different chemical salts (i.e., DPHP, MPP, PS, SA, & AB) and CAR fungicide on the reduction of the disease severity, of cucumber root and stem rot caused by *Forc*, was statistically significant ( $F = 85.99$ ,  $df = 7, 14$ ,  $p < 0.01$ ).

Based on the effects of chemical salts on the DSI the highest DSI was observed in positive control and treatments related to DPHP and AB

with an average of 17.3, 14.83, and 7.7% one week after the first spraying, respectively. In other treatments, no disease was recorded after one week. The control efficacy after the first foliar application was the lowest in DPHP and AB treatments with 14.27 and 57.22%, respectively (Table 4). Based on the results of the effects of chemical salts on the DSI, the highest DSI was detected in DPHP, AB, and MPP with a DSI of 42.9, 34.8, and 33.16% one week after the 2<sup>nd</sup> spraying, respectively, as compared to the positive control. On the other hand, the lowest DSI (0%) was observed in PS and CAR which placed them in the same group. The SA with an average DSI of 19.73% statistically ranked in the next group. After the 2<sup>nd</sup> spraying, the highest control efficiency (100%) was related to PS and CAR (Table 4).

**Table 3** Analysis variance of the effect of different chemical salts on disease severity caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* in cucumber under greenhouse condition.

Sources of variation	1 <sup>st</sup> spray 2 <sup>nd</sup> spray 3 <sup>rd</sup> spray			
	df	MS	MS	MS
Block	2	1.52 <sup>ns</sup>	8.03 <sup>ns</sup>	18.86 <sup>ns</sup>
Treatment	7	97.31 <sup>**</sup>	1552.51 <sup>**</sup>	3479.71 <sup>**</sup>
Error	14	0.19	10.71	40.46
CV (%)		25.37	15.84	10.14

\*\* = significant at level of 0.01, ns = non-significant.

**Table 4** Mean comparison of disease severity and control efficacy of chemical compounds on *Fusarium oxysporum* f. sp. *radicis-cucumerinum* one week after each spraying time in greenhouse condition.

Treatments	1 <sup>st</sup> spray		2 <sup>nd</sup> spray		3 <sup>rd</sup> spray	
	Disease severity (%)	Control efficiency (%)	Disease severity (%)	Control efficiency (%)	Disease severity (%)	Control efficiency (%)
AB	7.4 <sup>b</sup>	57.22	34.8 <sup>b</sup>	22.07	77.63 <sup>c</sup>	16.35
DPHP	14.83 <sup>b</sup>	14.27	42.9 <sup>a</sup>	3.94	96.16 <sup>a</sup>	-4.07
PS	0 <sup>c</sup>	100	0 <sup>d</sup>	100	35.76 <sup>c</sup>	61.19
MPP	0 <sup>c</sup>	100	33.16 <sup>b</sup>	25.75	83.86 <sup>bc</sup>	9.56
SA	0 <sup>c</sup>	100	19.73 <sup>b</sup>	55.82	56.5 <sup>d</sup>	39.2
Negative control	0 <sup>c</sup>	100	0 <sup>d</sup>	100	0 <sup>f</sup>	100
Positive control	17.3 <sup>a</sup>	0	44.66 <sup>a</sup>	0	92.93 <sup>ab</sup>	0
CAR	0 <sup>c</sup>	100	0 <sup>d</sup>	100	38.86 <sup>c</sup>	58.20

Means in a column followed by the same letter (s) are not significantly different (Duncan's multiple ranges test,  $P < 0.05$ ). AB: Ammonium bicarbonate, DPHP: Di-potassium hydrogen phosphate, MPP: Mono-potassium phosphite, SA: Salicylic acid, PS: Potassium sorbate, CAR: Carbendazim WP 60%.

After the 3<sup>rd</sup> spraying the highest DSI was found in DPHP (96.16%) and MPP (83.86 %) while the lowest DSI was observed in PS, CAR, and SA with an average of 35.76, 38.86, and 56.5%, respectively. The results further revealed that the highest control efficiency belonged to PS, CAR, and SA with an average of 61.19, 59.24, and 39.2%, and the lowest control efficacy belonged to DPHP and potassium phosphate with an average of -4.07 and 9.56%, respectively. Eventually, the treatments AB and MPP were considered in the same group. Although not statistically significant, they decreased the severity of the DSI (Table 4).

## Discussion

Based on the results of this study, the pathogenicity of *F. oxysporum* f. sp. *radicis-cucumerinum* (*Forc*) isolates on cucumber seedling were confirmed using the root dip method. The symptoms of the disease were observed five to seven days after inoculation and the pathogen was re-isolated. The incidence of the disease in the positive control treatment (in greenhouse experiments) also approved the pathogenicity of the applied isolates in this study. Therefore, the results of this study confirmed the antifungal activity of the chemical salts PS and SA on the radial growth rate of *Forc* in laboratory and greenhouse conditions. Chemical salts have been shown to have antifungal activity and act as resistance inducers against some fungal plant pathogens such as *Fusarium*-related disease (Arslan *et al.*, 2009; Yousefi *et al.*, 2010). *In vitro* condition, exposure of *Forc* to chemical salts in PDA medium significantly reduced the radial growth rate of the fungal colony and, in some cases, weakened the colony. Furthermore, PS and SA (at the concentration of 5 g/l) like carbendaizim, demonstrated a fungicidal effect on *Forc* and other salts represented fungistatic effects. The results of this experiment revealed that using PS had a higher growth inhibitory effect on *Forc* colony at all applied concentrations compared to SA and other compounds. Based on the

results of this study, the application of SA and PS through root feeding and foliar spray could induce resistance against *Forc* in cucumber. As regards SA, our results are in agreement with those of Mandal *et al.* (2009) on tomatoes. It was reported that while SA was applied on tomato as root feeding or foliar spray, the accumulation of SA inside the root increased up to 10 times higher than the control plants. Similarly, SA contents in the foliar spray-treated tomato were 8.7 times higher than the control plants (Mandal *et al.*, 2009). Regarding colony morphology on the PDA medium, the fungal colony appeared to be dense, cottony-like, with well-developed aerial mycelia in the control (check, no chemical salts) treatment. In the MPP treatment fungal colony had dispersed mycelia, adhered to the culture medium surface, and represented no aerial and cottony mycelium as in the check treatment, indicating that the applied chemical salt exerted no fungicidal effect although the radial growth rate of the target fungus demonstrated retardation. In the SA treatment, the colony morphology showed less aerial growth and density in comparison to the check treatment. Nonetheless, it indicated a good inhibitory effect and by increasing the concentration up to five g/l stopped the radial growth of the fungus, representing that it had a fungistatic effect at lower concentrations while having a fungicidal effect at higher concentrations. This is in line with the results of Mandal *et al.*, (2009) showing that the mycelial growth of *Fusarium* isolates in tomato was not significantly affected by SA at low concentrations. In the DPHP treatment, the aerial growth of mycelium was sparse and weaker compared to the control (fungistatic effect). As regards AB, the colony morphology was similar to the check treatment and demonstrated aerial growth and cottony mycelium on PDA. However, the radial growth of the target fungus was completely inhibited in the PS treatment and apparently exerted a fungicidal effect. The results of other studies have revealed that some chemical salts have no fungicidal effect despite hindering the fungus from utilizing nitrogen sources and producing

aerial mycelium by causing mutation, especially in *Fusarium*, leading to the production of thin mycelium and poor colony growth, and ultimately weakening the fungus (e.g., Puhalla, 1985; Corell *et al.*, 1987). Based on the results of this study, although some applied chemicals such as AB, DPHP, and MPP failed to inhibit the growth rate of the target fungus, they caused the production of the weakened fungal colony possibly due to the inability of the fungus to use nitrogen and other nutrients.

Although the laboratory assays of chemical salts are needed for screening and selecting chemical salts, *in vivo* tests are also required to confirm their efficacy in actual production. The results of the greenhouse experiment revealed that the application of chemical salts as the root treatment following by foliar application can perfectly control *Forc*. To the best of our knowledge, this is the first study that has focused on root feeding and the foliar application of chemical salts for controlling the *Fusarium* root and stem rot disease of cucumber. In the greenhouse, all seedlings were sprayed three times with selected concentrations from the preliminary *in vitro* tests. One week after the first spraying, the maximum DSI was observed in those seedlings which were treated with DPHP and confirmed the result of the *in vitro* condition. This may also be due to the wounds caused by the application of this salt on cucumber plant roots, which facilitates the entry of the pathogenic fungus. Based on the results of greenhouse experiments, no disease symptoms were observed regarding PS after the second spraying and 100% control efficiency was detected in comparison with the check treatment. On the other hand, in the course of time, some signs of infections were observed in the plants after the third spraying in the PS treatment. However, it was the best treatment after the CAR fungicide for reducing disease severity. The results of this study indicated that the root treatment of cucumber seedlings with PS before transplanting and spraying of plants once every two weeks was suitable for managing cucumber root and stem rot disease. Chemical salts may

reduce disease severity by inducing plant resistance and enhancing plant growth in addition to their direct effect on fungal growth (laboratory results). Another reason for the reduction in the symptoms of cucumber root and stem rot disease and improvement of the seedling growth could be the nutritional effects of these chemical salts or the availability of those elements which cannot be absorbed directly. It also may increase the production of growth stimulants in the plant. The results of a previous study showed that the production of defense enzymes in the first 72 hours after the treatment increased by using resistance inducers while it decreased over time (Ghazimohseni *et al.*, 2014). SA, as an antimicrobial compound, is recognized as a disease-resistance inducing compound in various experiments. It has been reported to control the root rot and stem disease of cucumbers and to reduce the percentage of the disease (e.g., Yosefi *et al.*, 2010; Alizadeh and Salari, 2014). The findings of the present study showed that *Forc* was affected by several salts, and fungal growth was completely inhibited by the PS salt, which corroborates with the results of earlier studies (e.g., Mills *et al.*, 2004; Ghadiri *et al.*, 2013) in the case of *Fusarium* potato dry rot disease and *Fusarium* tomato disease (El-Mohamedy *et al.*, 2014). In conclusion, all the applied chemical salts in this study demonstrated antifungal activity against *Forc* although potassium sorbate in both *in vivo* and *in vitro* assays represented the best antifungal activity against *Forc*. Potassium sorbate is a useful agent for food and beverage preservation and has been used for a long time but concerns remain over their complete safety (Piper and Piper, 2017). It is considered as somewhat carcinogenic which is important to take into consideration, whether the substantial human consumption of these compounds could significantly increase levels of such damages in man is still unclear (Piper and Piper, 2017).

According to the results of this study, it is recommended that researchers apply PS and SA and also AB and MPP owing to their positive effect on disease severity, for treating cucumber seedlings via the root dip method before

transplanting and weekly foliar spraying (with SA, MPP, and AB) and biweekly (PS and CAR). In addition, our findings indicate the use of these chemical salts alone or in combination with fungicides can reduce the application of chemical pesticides and lead to the production of healthier products. Finally, our results showed that potassium sorbate and SA can be used in an integrated management program in order to control the *Fusarium* root and stem rot disease of cucumber in greenhouse condition.

### Acknowledgments

Authors are thankful to M. Azadvar, M. Sharif, and Ali Roshan for their help and technical assistance. This Study is part of the MSc thesis of the first author which has been submitted to the University of Payam Noor Shiraz.

### Declaration of conflicting interests

The authors state that there is no conflict of interest.

### Funding Acknowledgments

Authors are thankful to the South Kerman Province Agriculture Jihad Organization, Jirot, Iran for financial support of this project and to Iranian Research Institute of Plant Protection for its cooperation to conduct project No. 951001-107-16-70-24.

### References

- Abdel-Kader, M. M., El-Mougy, S. S., El-Gammal, N. G., Abd-El-Kareem, F. and Abd-Alla, M. A. 2012. Laboratory evaluation of some chemicals affecting pathogenic fungal growth. *Journal of Applied Sciences Research*, 8(1): 523-530.
- Abdel-Monaim, M. F. 2010. Induced systemic resistance in tomato plants against *Fusarium* wilt disease. Pages 253-263. In: *Proceedings of the 2<sup>nd</sup> Minia Conference for Agriculture and Environmental Science*, 22-25 March, 2010, Minia, Egypt.
- Alizadeh, H. R. and Salari, K. 2014. Induced resistances by  $\beta$ -amino butyric acid (BABA) against *Fusarium* stem and root rot of cucumber. *Iranian Journal of Plant Protection Science*, 45(2): 299-307 (In Persian with English summary).
- Arslan, U., Kadir, I., Vardar, C. and Karabulut, O. A. 2009. Evaluation of antifungal activity of food additives against soil borne phytopathogenic fungi. *World Journal of Microbiology and Biotechnology*, 25: 537-543.
- Correll, J. C., Klittich, C. J. R., and Leslie, J. F. 1987. Nitrate nonutilizing mutants of *Fusarium oxysporum* and their use in vegetative compatibility tests. *Phytopathology*, 77: 1640-1646.
- Elmer, W. H. 2002. Influence of inoculum density of *Fusarium oxysporum* f. sp. *cyclaminis* and sodium chloride on cyclamen and the development of *Fusarium* wilt. *Plant Disease*, 86: 389-393.
- El-Mohamedy, R. S. R., Jabnoun-Khiareddine, H. and Daami-Remadi, M. 2014. Control of root rot diseases of tomato plants caused by *Fusarium solani*, *Rhizoctonia solani* and *Sclerotium rolfsii* using different chemical plant resistance inducers. *Tunisian Journal of Plant Protection*, 9: 45-55.
- Fallah poor, A., Aminian, H., Sahebani, N. and Esmailzadeh Hosseini, S. A. 2013. Evaluation of resistance of 20 sesame germplasms to damping off caused by *Fusarium oxysporum* f. sp. *sesami* in Yazd region and investigation of phenylalanine ammonialyase (PAL) activity in resistant and susceptible germplasms. *Iranian Journal of Plant Pathology*, 49(4): 413-424.
- Garibaldi, A., Gilardi, G., Ortu, G., Gullino, M. L. 2016. First report of *Fusarium oxysporum* f. sp. *radicis-cucumerinum* causing wilt on cucumber (*Cucumis sativus*) in Italy. *Plant Disease* 100: 1791.
- Ghadiri, M. R., Dalili, A., Frotan, A., Zaker, M., Rahmanifard, B., Dalili, M. 2013. Study on antifungal activity of some salts on growth and dry rot development of *Fusarium solani* (Mart.) Sacc. *American-Eurasian Journal of Agricultural & Environmental Science*, 13(5): 668-672.



- Ghazimohseni, V., Sabbagh, S. K., Esmaeilzadeh Bahabadi, S. and Ghorbani, M. 2014. Application of silicon in induction of systemic resistance against *Fusarium* wheat head blight disease. *Biological Control of Pest and Plant Disease*, 2(3): 128-137.
- Karaca, G., and Kahveci, E. 2010. First report of *Fusarium oxysporum* f. sp. *radicis-cucumerinum* on cucumbers in Turkey. *Plant Pathology*, 59(6): 1173-1174.
- Mandal, S., Mallick, N., Mitra, A., 2009. Salicylic acid-induced resistance to *Fusarium oxysporum* f. sp. *lycopersici* in tomato. *Plant Physiology and Biochemistry* 47(7): 642-649. doi:10.1016/j.plaphy.2009.03.001.
- Mills, A. A. S., Platt, H. W. and Hurta, R. A. R. 2004. Effect of salt compounds on mycelial growth, sporulation and spore germination of various potato pathogens. *Postharvest Biology and Technology*, 34: 341-350.
- Moreno, A., Alferez, A., Aviles, M., Dianez, F., Blanco, R., Santos, M., and Tello, J. C. 2001. First report of *Fusarium oxysporum* f. sp. *radicis-cucumerinum* on cucumber in Spain. *Plant Disease*, 85(11): 1206-1206.
- Najafiniya, M. and Mirzadeh Abgarmi, Z. 2020. Prevention of root and stem rot disease of cucumber caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* using grafting on cucurbits rootstocks. *Entomology and Phytopathology*, 88(1): 81-9.
- Najafiniya, M. and Shahabi, I. 2019. *Fusarium* Stem and root rot disease of cucumber and its control management. *Extension Journal of Greenhouse Vegetables*, 2(1): 63-72 (In Persian).
- Najafiniya, M., Shahabi, I. and Rezaee, S. 2018. Study isolates of *Fusarium* stem and root rot disease of greenhouse cucumber using pathogenicity tests, vegetative compatibility groups and molecular marker. *Journal of Plant Protection*, 32(1): 49-57.
- Pavlou, G. C., Vakalounakis, D. J. and Ligoxigakis, E. K. 2002. Control of root and stem rot of cucumber, caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum*, by grafting onto resistant rootstocks. *Plant Disease*, 86: 379-382.
- Piper, J. D., and Piper, P. W. 2017. Benzoate and sorbate salts: a systematic review of the potential hazards of these invaluable preservatives and the expanding spectrum of clinical uses for sodium benzoate. *Comprehensive Reviews in Food Science and Food Safety*, 16: 868-880.
- Puhalla, J. E. 1985. Classification of strains of *Fusarium oxysporum* on the basis of Vegetative Compatibility. *Canadian Journal of Botany*, 63: 179-183.
- Roustaei, A., and Mohammadian, M. 2005. Study of interaction between plant nutrition (N, P, K and Ca) and cucurbit wilt disease (*Phytophthora drechsleri* Tucker) in some cucumber cultivars. *Seed and Plant*, 21(3): 457-466.
- Shahriari, D. and ZARE, R. 2006. *Fusarium* stem and root rot of greenhouse- cucumber. 17<sup>th</sup> Iranian Plant Protection Congress. 2-5 Sep. 2006, Karaj Iran. P.191.
- Shlevin, E., Saguy, I. S., Mahrer, Y., Katan, J. 2003. Modeling the survival of two soilborne pathogens under dry structural solarization. *Phytopathology*, 93:1247-1257.
- Soylu, E. K., and Incekara, R. 2017. Biofungicidal activities of plant essential oils against cucumber root and stem rot disease caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum*. *Journal of Plant Pathology*, 99(2): 437-444.
- Stromberge, A. and Brishammar, S. 1991. Induction of systemic resistance in potato (*Solanum tuberosum* L) plants to late blight by local treatment with *Phytophthora infestans*, *Phytophthora cryptogea* or dipotassium phosphate. *Potato Research*, 34: 219-225.
- Turkkan, M. 2013. Antifungal effect of various salts against *Fusarium oxysporum* f. sp. *cepae*, the causal agent of *Fusarium* basal rot of onion. *Journal of Agricultural Sciences*, 19: 178-187.
- Vakalounakis, D. J. 1996. Root and stem rot of cucumber caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* f. sp. Nov. *Plant Disease*, 80:313-316.
- Vakalounakis, D. J. and Fragkiadakis, G. A. 1999. Genetic diversity of *Fusarium*

- oxysporum* isolates from cucumber: Differentiation by pathogenicity, vegetative compatibility, and RAPD fingerprinting. *Phytopathology*, 89:161-168.
- Yadav, K., LalMeena, N., Yadav, R. and Dudi, K. 2019. Morphological, cultural and pathogenic variability among the different isolates of *Fusarium oxysporum* f. sp. *radices cucumerinum* causing root and stem rot of cucumber: A review. *Journal of Pharmacognosy and Phytochemistry* 8(5): 426-429.
- Yousefi, H., Sahebani, N., Mirabulfathi, M., Faravardeh, L. and Mahdavi, V. 2010. The effects of salicylic acid and *Bacillus subtilis* on cucumber root and stem rot caused by *Fusarium oxysporum* f. sp. *radices cucumerinum*. *Iranian Journal of Plant Pathology*, 46(4): 293-308 (Persian with English abstract pp.85-87).

## اثر ضدقارچی برخی نمک‌های شیمیایی در کنترل *Fusarium oxysporum* f. sp. *radicis*- *cucumerinum* عامل بیماری پوسیدگی ریشه و ساقه خیار

زینب میرزاده آبگرمی<sup>۱</sup>، موسی نجفی‌نیا<sup>۲</sup> و حسام‌الدین رضاعی<sup>۱</sup>

۱- گروه کشاورزی، دانشگاه پیام نور، صندوق پستی ۳۶۹۷ - ۱۹۳۵۹۵، تهران، ایران.

۲- بخش تحقیقات گیاه‌پزشکی، مرکز تحقیقات و آموزش کشاورزی و منابع طبیعی جنوب استان کرمان، سازمان تحقیقات، آموزش و ترویج کشاورزی، جیرفت، ایران.

پست الکترونیکی نویسنده مسئول مکاتبه: m.najafinia@areeo.ac.ir

دریافت: ۶ اسفند ۱۳۹۸؛ پذیرش: ۷ آبان ۱۳۹۹

**چکیده:** بیماری پوسیدگی ریشه و ساقه خیار ناشی از قارچ *Fusarium oxysporum* f. sp. *radicis*-*cucumerinum* یکی از مهم‌ترین بیماری‌های خیار در ایران می‌باشد/ این تحقیق به‌منظور بررسی تأثیر برخی نمک‌های شیمیایی در کنترل قارچ عامل بیماری پوسیدگی ریشه و ساقه خیار در شرایط آزمایشگاه (درون شیشه) و گلخانه انجام شد. در شرایط آزمایشگاه اثر تیمارها شامل پتاسیم سورات، فسفیت پتاسیم، بیکربنات آمونیوم، سالیسیلیک اسید، دی پتاسیم هیدروژن فسفات و کاربندازیم در غلظت‌های ۰/۵، ۲، ۴ و ۵ گرم در لیتر توسط آزمایش فاکتوریل در قالب طرح کاملاً تصادفی ارزیابی شد. در شرایط گلخانه، آزمایش در قالب طرح بلوک‌های کامل تصادفی با هشت تیمار در سه تکرار انجام شد. نتایج شرایط آزمایشگاه و گلخانه نشان داد، تیمارها تأثیر معنی‌داری بر کاهش رشد قارچ و شدت بیماری دارند. در آزمایشگاه بیش‌ترین تأثیر در مهار رشد قارچ، مربوط به ترکیبات پتاسیم سورات و قارچ‌کش کاربندازیم بود و در کلیه غلظت‌های مورد آزمایش، رشد قارچ را ۱۰۰ درصد کنترل نمودند. دی پتاسیم هیدروژن فسفات با غلظت نیم در هزار با کارایی کنترل ۸/۷۴ درصد کم‌ترین تأثیر را در مهار قارچ نشان داد. در شرایط گلخانه در تیمار قارچ‌کش کاربندازیم هیچ‌گونه علائم بیماری پس از یک ماه مشاهده نشد. اما پتاسیم سورات و سالیسیلیک اسید به‌ترتیب شدت بیماری را نسبت به شاهد به‌میزان ۶۱/۱۹ و ۳۹/۲ درصد کاهش دادند و در رده‌ی بعدی قرار گرفتند. نتایج این تحقیق نشان داد ترکیبات مذکور اثر قارچ‌کشی علیه عامل بیماری دارند. این ترکیبات بی‌خطر می‌توانند برای کنترل بیماری پوسیدگی ریشه و ساقه خیار به‌صورت تیمار ریشه و محلول‌پاشی در شرایط گلخانه استفاده شوند.

**واژگان کلیدی:** کنترل بیماری، قارچ‌کش، مدیریت تلفیقی، مقاومت القایی