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Original Research Article

Determination of Effects of Some Plant Activators against Potato Virus Y in Pepper*

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ABSTRACT

In this work, the effects of the plant activators Messenger, Actigard, ISR-2000 and Crop-Set against Potato Virus Y (PVY) in pepper (*Capsicum annuum*) were evaluated. In surveys conducted in Adana and Mersin provinces of southern Turkey in 2015, PVY was detected in symptomatic peppers by DAS-ELISA tests. The PVY isolate used in this study was isolated from pepper plants in Adana. Effects of the activators were evaluated in a climate chamber. Pepper seedlings were treated by Messenger, Actigard, ISR-2000 and Crop-Set and then the activator-treated peppers were inoculated by the PVY isolate 72 and 96 hrs after the activator treatments. In the assay, symptom expression time, plant length, leaf width, root length, lignification (lignin accumulation) and H_2O_2 accumulation were measured until the flowering period. The data was analyzed by using statistical methods. Messenger, Actigard, ISR-2000 and Crop-Set delayed symptom expression time by 12, 10, 8 and 3 days, respectively. Moreover Actigard, Messenger and ISR-2000 increased leaf surface area and plant height. Among all treatments, only inoculations with PVY 72 hrs after Crop-Set treatments increased the root length. Activator treated plants showed lignification and H_2O_2 accumulation. Best treatment times for lignification and H_2O_2 accumulation were determined as 72 hrs for Messenger, 96 hrs for Actigard, Crop-Set and ISR-2000.

Keywords: Pepper, PVY, Actigard, Messenger, ISR-2000, Crop-Set, Activator, Lignification, Hydrogen peroxide accumulation

INTRODUCTION

There are many pathogens that infect pepper plants. Plant viruses are among the serious threats for peppers. The most common pepper viruses are Potato Virus Y (PVY), Tobacco Etch Virus (TEV), Pepper Veinal Mottle Virus (PepVMV) and Cucumber Mosaic Virus (CMV) which transmit non-persistently by aphids, and persistently thrips-transmitted Tomato Spotted Wilt Virus (TSWV) in the Mediterranean basin. PVY, CMV, TEV and Beet Western Yellows Virus (BWYV) are major pepper viruses in Turkey. However, Tobacco Mosaic Virus (TMV), Potato Virus X (PVX), TSWV and Pepper Mild Mottle Virus (PMMoV) exist in Turkey (Arli-Sokmen and Sevik 2005; Ozaslan et al. 2006; Sevik 2011; Buzkan et al. 2013).

PVY that is a widespread worldwide and in Turkey is the one of most important plant viruses affecting *Solanaceous* crops (10-100% yield loss). It is a member of the most important and large family "*Potyviridae*" of the genus "*Potyvirus*" (De Bokx and Huttinga 1981; Büchen-Osmond 1987). The methods of management for plant virus diseases are insufficient or ineffective compared to other plant pathogens. Hence, there has been increased interest in Systemic Acquired Resistance (SAR) against plant diseases (Saravanakumar et al. 2007). Although many studies focused on SAR against several fungal diseases, just a few studies have been performed for control of plant viruses. Furthermore, there is very little information about stimulation of SAR via plant activator against plant virus diseases.

SAR is the stimulation of the plant's defense mechanism by biotic or abiotic agents (activators) and it can be expressed as the activation of resistance mechanism which is in the passive state of the plant (Van Loon et al. 1998). It is stimulated by activators and various defense mechanisms such as hypersensitivity reaction, lignification, papilla formation, pathogenicity related proteins (PR) synthesis, phytoalexins synthesis, H_2O_2 production are stimulated by them. Lignification is one of the mechanisms for disease resistance in plants and occurs in a variety of plant-microbe interactions including viruses (Ryals et al. 1996). Lignification occurs around the lesions promoted by viruses on many hypersensitivity reacting hosts (Collendavelloo et al. 1983). H_2O_2 plays an essential role in plant defence responses against pathogens. It directly inhibits the growth and reduces the viability of many plant pathogens. It indirectly plays a role in strengthening RNA silencing to restrict

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systemic viral infection (Balázs et al. 2003, Liao et al. 2013). At low concentrations, H_2O_2 provides a diffusible signal for the initiation of antioxidant and pathogenesis-associated genes in adjacent plant tissues. Low concentrations of H_2O_2 suppress necrotic disease symptoms caused by viral, bacterial, and fungal infections by increasing plant antioxidant capacity. However, high concentrations of H_2O_2 cause death (necrosis) of the plant as well as invading pathogen cells (Hafez et al. 2012). SAR-induced plants are capable of prolonged protection against the pathogens range of several weeks to months (Schonbeck et al. 1993).

In this study, four plant activators called Messenger, Actigard, ISR-2000 and Crop-Set were used. Actigard, S-methyl 1,2,3-benzothiadiazole-7-carbothioate (Acibenzolar-S-Methyl), is the most studied plant activator in the world (Roberts and Hutson 1999). In the world market Acibenzolar-S-Methyl is sold under different trade names such as Bion 5% WP, BTH (Agrochemical) and Actigard 50 WG (Syngenta). Actigard 50 WG (Syngenta) was registered as a plant protection product against some fungal and bacterial plant pathogens, and a virus (Iris Yellow Spot Virus) in the USA (Anonymous 2017). Actigard is not a registered product in Turkey, but Bion is registered.

Messenger includes natural harpin protein isolated from *Erwinia amylovora*. The active ingredient of ISR-2000 is a fermentation product of *Lactobacillus acidophilus* containing plant extract, yeast extract, riboflavin, benzoic acid, nicotinamide and thiamine. ISR-2000 increases the activity of enzymes such as chitinase, gluconase and peroxidase in plants. After the stimulation has occurred, the plant remains at the highest level of alert against a possible subsequent attack and thus can defend itself against pathogenic invasion (Tosun and Ergun 2002; Koca 2003). The active ingredient of Crop-Set is a liquid fermentation product of *L. acidophilus*-containing plant extract, manganese sulphate ($MnSO_4$), ferric sulphate [$Fe_2(SO_4)_3$] and copper sulphate ($CuSO_4$). The Crop-Set safely improves the ability of the plant to use nutrients, optimize plant yield, improves fruit quality and homogeneity (Anonymous 2015).

It is known that an effective control method against viruses that also include PVY currently does not exist. The aim of this study is to develop an effective management approach against to PVY using some plant activators in pepper plants.

MATERIALS AND METHODS

Virus Isolates

Surveys were conducted in pepper fields in Adana and Mersin provinces of Turkey from April to June 2015. In the surveys, symptomatic plants showing PVY-like symptoms including leaf mosaic, chlorosis, leaf vein banding, and leaf deformation were selected and their leaves were collected. The plant materials were kept at -20°C until used.

Virus Detection

The collected leaf samples were tested by enzyme-linked immunosorbent assay (ELISA) (Clark and Adams 1977) by using ten antisera kits specific to PVY, Alfalfa Mosaic Virus (AMV), CMV, PVX, Pepper Mottle Virus (PepMoV), TEV, TMV, TSWV, PMMoV and Tomato Yellow Leaf Curl Virus (TYLCV). The test was conducted according to the instructions of the manufacturer (Bioreba).

Host Reaction

PVY positive leaf samples in DAS-ELISA were used for mechanical inoculation. For biological identification of PVY, following indicator plants were used: *Nicotiana tabacum* cv Samsun, *Chenopodium murale*, *Solanum lycopersicon*, *Capsicum annuum.* PVY inoculum was prepared with 0.1 M Sorensen phosphate buffer (pH 8.0) (1/10 g/ml) containing Celite® 545 (0.5 g/20 ml). Then, the PVY inoculum inoculated to the indicator plants by the cotton bud. Two minutes after inoculation, the plants were washed with water. Indicator plants were maintained under climate chamber at about 24°C.

Plant Material and Growth Conditions

In the experiments, cv 'Demre Sivrisi' pepper plants were used. Demre Sivrisi was selected because it is susceptible to PVY and widely produces both open field and greenhouse cultivation (Çelik et al. 2013). The pepper plants were maintained in a climate chamber at about 22-24°C, 70% moisture under a photoperiod of 16/8 hrs.

Plant Activator Treatments

Actigard, Messenger, ISR-2000 and Crop-Set were applied to pepper plants having 2 cotyledons and 2 true leaves (Table 1).

Table 1. Plant activators used in this study.

Plant Activators	Active Ingredients	Company
Actigard	1,2,3-benzothiadiazole-7-carbothioic acid-S-methyl ester	Syngenta
Messenger	Harpin protein 3% WG	Plant Health Care
ISR-2000	*Lactobacillus acidophilus,* yeast extract, plant extract and benzoic acid	Improcrop
Crop-Set	*Lactobacillus acidophilus,* plant extract, manganese sulphate, iron sulphate and copper sulphate	Improcrop

The plant activator treatments and the virus inoculations were carried out when the first true leaves were fully formed. The plant activators used in the experiments were prepared by diluting with water at the doses given in Table 2.

Table 2. Treatment doses of plant activators.

Plant Activators	Recommended Dose	Dose 1	Dose 2	Dose 3	Dose 4	Dose 5
Actigard (g/l)	0.17	0.04	0.08	0.17	0.34	-
Messenger (g/l)	0.15	0.075	0.15	0.3	0.6	1.2
ISR-2000 (ml/l)	1	0.25	0.5	1	2	4
Crop-Set (ml/l)	0.6	0.15	0.3	0.6	1.2	-

According to the instructions of the producers of the activators, the pepper plants were applied 2 more times with an interval of 14 days after the first treatment date of the activators. The virus inoculation was performed 72 hrs (72 hrs ATAs) and 96 hrs (96 hrs ATAs) after treatment of the activators. The PVY isolate (Adana) was mechanically inoculated to pepper plants, using 0.1 M Sorensen phosphate buffer (pH 8.0) containing 0.5 g/20 ml Celite® 545. The virus symptoms were recorded 2-3 weeks after mechanical inoculation. PVY detection was confirmed by DAS-ELISA. In this study, the evaluation stage of the results began with the observation of the first symptom in pepper plants that were not treated by the plant activators, inoculated only by the virus. The activator treatments continued until the blooming of the pepper plant.

Growth Measurements

The measurements were started with the beginning of flowering of the pepper plants. The following measurements were taken: plant height, root length and leaf area. The distance between the root crown and the top point of the plants was measured and the average plant length was determined. The roots of the plants removed from the pots were washed and measured. The average root length was determined. Each fifth leaf from the top point of the plant was cut and the leaf area was measured via computer-connected HP Deskjet F2180 scanner (Flaudung and Ritter 1991; Tsuda 1999).

Histochemical Staining

Root, stem and leaf tissues of the plants were used as materials in histochemical staining. Histochemical staining was performed to determine lignification and H_2O_2 accumulation in the infected tissues. For determination of lignification, tissue pieces of 3-5 mm were taken from root, stem and leaves of pepper plants. Chlorophyll in the

tissues was removed in 100% methanol containing 1% phloroglucinol at room temperature (20°C) for a night. Bleached tissues were stored in chloral hydrate (2.5 g / ml) for at least 24 hrs to ensure clearness of the tissues. The tissue sections were placed on a sterile lamina and 1-2 drops concentrated HCl solution were added and waited for 10 min. At the end of the waiting period, a few drops of 50% glycerol solution were applied to the tissue and a coverslip was placed over it. The samples were examined under a light microscope, in which lignified structures were observed to have a dark pink color.

To determine hydrogen peroxide accumulation, tissue pieces of 3-5 mm were cut from root, stem and leaves of pepper plants as in the phloroglucinol/HCl test. The tissues were kept in the dark for 15-25 min in the DAB solution. Chlorophyll in the tissues was eliminated in 100% methanol by maintaining at room temperature (20°C) for a night. The bleached tissues were immersed in chloral hydrate (2.5 g/ml) for at least 24 hrs to ensure clearness of the tissues. The tissue sections were placed on a sterile lamina, 1-2 drops of 50% glycerol solution were dropped and the coverslip was placed over it. The samples were examined under a light microscope, H_2O_2 accumulation was observed to have a brown color (Gahan 1984; Vallet et al. 1996; Soylu 1999).

Statistical Analysis

The study was conducted as 3 replicates (5 pots per replicate, 1 plant per pot) for each treatment and dose according to completely randomized experimental design. Analysis of variance (ANOVA) was performed and mean values were separated using the least significant difference (LSD) test ($P \leq 0.05$).

RESULTS AND DISCUSSION

PVY Symptoms on Indicator Plants

As a result of mechanical inoculation studies, the symptoms of PVY on indicator plants were observed 12-20 days after mechanical inoculation. PVY infected indicator plants were tested by ELISA for the virus confirmation. PVY on *N. tabacum* cv Samsun induced leaf mottling, interveinal clearing, leaf distortion and stunting; on *C. murale* PVY created no symptom; on *S. lycopersicon* PVY induced mild mosaic and yellowing; on *C. annuum* PVY caused stunting, leaf mottling and deformation. Similar results were previously reported by many researchers (Taraku and Juretic 1991; Nyamwamu et al. 2014).

The Effect of the Plant Activators on Symptom Development

In the assay of 72 hrs ATAs, the highest dose of Actigard (0.34 g/l) and the doses of Messenger (0.15, 0.3, 0.6 and 1.2 g/l) delayed significantly the symptom expression time compared with positive control**.** In the assay of 96 hrs ATAs all doses of Messenger, three doses of Actigard (0.08, 0.17, 0.34 g/l), all doses of ISR-2000 except 0.25 ml/l and Crop-Set's highest dose (1.2 ml/l) significantly delayed symptom development compared with positive control (Table 3).

Table 3. Comparison of the effect of the plant activators on symptom development.

Treatment and Dose	**Symptom Development (Day)**	
	72 hrs	**96 hrs**
0 (Positive Control)	14.13 d*	14.13 g
Actigard (g/l)		
0.04	15.06 d	15.33 defg
0.08	15.40 cd	16.13 cde
0.17**	16.13 cd	16.93 bc
0.34	24.86 ab	21.00 a
Messenger (g/l)		
0.075	14.93 d	16.06 cdef
0.15**	17.46 c	16.33 bcde
0.3	23.53 b	17.73 bc
0.6	23.93 b	17.13 bc
1.2	26.40 a	17.66 b
ISR-2000 (ml/l)		
0.25	15.33 cd	14.60 fg
0.5	16.20 cd	16.73 bcd
1**	15.80 cd	17.06 bc
2	15.86 cd	17.40 bc
4	16.00 cd	22.13 a
Crop-Set (ml/l)		
0.15	14.46 d	14.26 g
0.3	14.80 d	15.13 efg
0.6**	15.93 cd	14.93 efg
1.2	16.06 cd	17.33 bc

*Means within columns by unlike letters differ significantly according to LSD test (P≤0.05). **Recommended dose.

According to our findings, Messenger, Actigard, ISR-2000 and Crop-Set delayed symptom expression time by 12, 10, 8 and 3 days, respectively. These results are in agreement with Anfoka (2000), who showed that BTH delayed the development of CMV symptoms for up to 7 days. In a study conducted by Mejia et al. (2009), the tomato plants treated with ASM showed virus symptoms delayed by 7 days and a considerable reduction in disease severity. Similar results were reported by Çalışkan and Kamberoğlu (2010). The researchers reported that Actigard treatment 48 hrs after Zucchini Yellow Mosaic Virus (ZYMV) inoculation delayed symptom expression time in pumpkins. In addition, Messenger and ISR-2000 treatments (72 hrs after the virus inoculation) were effective on symptom expression significantly compared with positive control**.** ASM decreased the incidence of tomato plants infected with TSWV by 28% under high disease pressure (Momol et al. 2004). Madhusudhan et al. (2008) found that ASM reduced the concentration of Tomato Mosaic Virus (ToMV) in infected plants. The same investigators observed increased concentrations of RNA-dependent RNA polymerase **(**RdRp) in ToMV-infected, ASM-treated plants. The synthesized cRNAs were found to be effective owing to disrupting replication and translocation of the virus RNA. In this case, the delay of the symptom of PVY symptom in ASM treated plants may be related to the increase of PVY RdRp concentration.

The Effect of the Plant Activators on Plant Height

Plant height increased at all doses of Messenger and ISR-2000 in both treatments (72 hrs ATAs and 96 hrs ATAs). The differences were statistically significant. On the other hand, the highest dose (0.34 g/l) of Actigard only increased statistically plant height in both treatments (Table 4).

Table 4. Comparison of the effect of the plant activators on plant height.

Treatment and Dose	Plant height (cm)	
	72 hrs	**96 hrs**
0 (Negative Control)	21.46 a*	21.46 b
0 (Positive Control)	17.73 fg	17.73 ef
Actigard (g/l)		
0.04	15.33 i	15.06 h
0.08	16.13 hi	15.40 gh
0.17**	16.93 gh	16.13 fgh
0.34	21.00 abcd	24.86 a
Messenger (g/l)		
0.075	21.10 abc	21.66 b
0.15**	19.66 cde	21.66 b
0.3	19.63 de	21.50 b
0.6	19.30 e	21.06 bc
1.2	19.30 e	20.73 bc
ISR-2000 (ml/l)		
0.25	19.83 bcde	19.66 cd
0.5	21.06 abcd	20.76 bc
1**	21.00 abcd	21.36 bc
2	21.83 a	21.03 bc
4	21.23 ab	20.50 bc
Crop-Set (ml/l)		
0.15	16.60 ghi	16.93 efg
0.3	19.03 ef	18.16 de
0.6**	18.86 ef	15.70 gh
1.2	17.73 fg	16.60 efgh

*Means within columns by unlike letters differ significantly according to LSD test (P≤0.05). **Recommended dose.

This study also showed that phytotoxicity was seen in the plant with increasing doses of Actigard. The most suitable dose of the plant height enhancer was 2 times the recommended dose for Actigard. Similarly, in a study conducted by Mandal et al. (2008), phytotoxicity was observed with the increase of ASM concentration against to TSWV. According to Godard et al. (1999), the height of BTH treated plants was reduced at a rate of 5.9-38.3%. On the other hand, Anfoka (2000) showed that the growth of BTH treated plants was unaffected. Nair et al. (2007) reported that ASM treated plants reduced plant growth. The combined use of ASM and plant growth-promoting rhizobacteria (PGPR) was more effective than only ASM treatment. Dereboylu and Tort (2010) showed that Crop-Set treated plants increased plant height up to 18.3%. In our study, the Crop-Set did not have any effect on plant height.

The Effect of the Plant Activators on Leaf Area

All doses of Actigard extended the leaf area in the both treatments (72 hrs ATAc and 96 hrs ATAc). In the assay of the virus inoculation performed 72 hrs after treatment of Messenger's (72 hrs ATMe) lowest dose (0.075 ml/l) the leaf area was extended. Similarly, in the inoculation performed 96 hrs after Messenger treatment (96 hrs ATMe) all doses of the activator increased the leaf area. In the assay of the virus inoculation performed 72 hrs after the Crop-Set treatment (72 hrs ATCs) (0.6 ml/l) the leaf area was extended. In the assay of the virus inoculation performed 96 hrs after the ISR-2000 treatments (96 hrs ATIs) (0.25 ml/l and 0.5 ml/l) the leaf area was enlarged (Table 5).

Table 5. Comparison of the effect of the plant activators on leaf area.

Treatment and Dose	**Leaf area (cm^2)**	
	72 hrs	**96 hrs**
0 (Negative Control)	17.05 b*	17.05 b
0 (Positive Control)	12.81 ghi	12.81 ef
Actigard (g/l)		
0.04	19.86 a	22.54 a
0.08	15.82 bc	22.28 a
0.17**	15.30 bcde	21.15 a
0.34	15.63 bcd	21.44 a
Messenger (g/l)		
0.075	14.73 cdef	16.73 b
0.15**	13.36 fghi	17.80 b
0.3	13.54 efghi	16.36 bc
0.6	13.31 fghi	14.83 cd
1.2	11.75 ijk	14.76 cd
ISR-2000 (ml/l)		
0.25	13.56 efghi	16.56 bc
0.5	14.16 cdefgh	18.07 b
1**	13.95 defgh	14.00 de
2	12.31 hij	12.42 efg
4	12.55 ghi	12.82 ef
Crop-Set (ml/l)		
0.15	10.70 jk	11.03 fg
0.3	10.40 k	10.69 g
0.6**	14.64 cdef	12.75 ef
1.2	12.54 ghi	13.94 de

*Means within columns by unlike letters differ significantly according to LSD test ($P \leq 0.05$). **Recommended dose.

The Effect of the Plant Activators on the Root Length

Only Crop-Set (0.6 ml/l) treatment increased the root length in the assay of 72 hrs ATCs. All other treatments were found ineffective. In assays of 96 hrs ATAs, all doses of the activators were found to have no positive effects on root development (Table 6).

Table 6. Comparison of the effect of the plant activators on root length.

Treatment and Dose	Root Length (cm)	
	72 hrs	**96 hrs**
0 (Negative Control)	30.86 ab*	30.86 a
0 (Positive Control)	27.46 cde	27.46 bcdefg
Actigard (g/l)		
0.04	25.03 efg	23.63 i
0.08	25.13 efg	23.73 i
0.17**	25.46 defg	24.83 ghi
0.34	23.70 gh	24.26 gi
Messenger (g/l)		
0.075	25.10 efg	30.26 ab
0.15 **	23.63 gh	28.53 abcd
0.3	22.06 h	27.96 abcdef
0.6	24.76 fg	29.56 abc
1.2	24.30 fgh	27.10 cdefgh
ISR-2000 (ml/l)		
0.25	28.26 bc	27.16 cdefgh
0.5	27.53 cde	28.40 abcde
1**	26.40 cdef	25.23 fghi
2	27.56 cde	25.46 efghi
4	27.86 cd	26.26 defghi
Crop-Set (ml/l)		
0.15	26.63 cdef	29.86 abc
0.3	27.93 cd	27.73 bcdefg
0.6**	32.80 a	27.43 bcdefg
1.2	28.46 bc	27.96 abcdef

*Means within columns by unlike letters differ significantly according to LSD test (P≤0.05). **Recommended dose.

Similarly, Kiracı and Padem (2015) found that Crop-Set and phosphorus treatments increased the average root length by 16.8%. On the other hand, Zhou et al. (2015) reported that HapG (Harpin) significantly promoted the growth of roots of TMV-infected tobacco plants. In our study, the Harpin had not any effect on root development and even caused root shortening. Csinos and Pappu (2001) pointed out that plants treated with 1.0 g of ASM had shorter roots than plants treated with 0.25 g ASM.

Determination of Lignin Synthesis

Actigard and ISR-2000 promoted higher degree lignification than Messenger and Crop-Set in the assay of 72 hrs ATAs. Actigard induced higher lignification than Messenger, ISR-2000 and Crop-Set in 96 hrs treatment. Lignification was the highest in root, stem and leaf in 72 and 96 hrs ATAs (Figure 1).

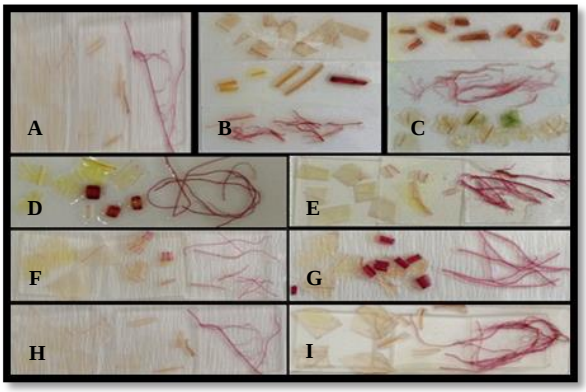

Figure 1. Localization of lignin compounds after treatment of the plant activator and PVY inoculation. (**A:** Control, **B:** Actigard 72hrs 0.34 g/l, **C:** Actigard 96 hrs 0.34 g/l, **D:** Messenger 72 hrs 1.2 g/l, **E:** Messenger 96 hrs 1.2 g/l, **F:** ISR-2000 72 hrs 4 ml/l, **G:** ISR-2000 96 hrs 4 ml/l, **H:** Crop-Set 72 hrs 1.2 ml/l **I:** Crop-Set 96 hrs 1.2 ml/l, leaf-stem-root in order)

In a study of ZYMV in squash plants, lignification was found to be greater 72 hrs ATI application compared to Actigard (Kashtban and Elibüyük 2010).

Determination of H_2O_2 Accumulation

Actigard induced higher H_2O_2 accumulation than Messenger, Crop-Set and ISR-2000 in 72 hrs after the treatment. It also had higher H_2O_2 accumulation than Messenger, ISR-2000 and Crop-Set in 96 hrs after the treatment (Figure 2).

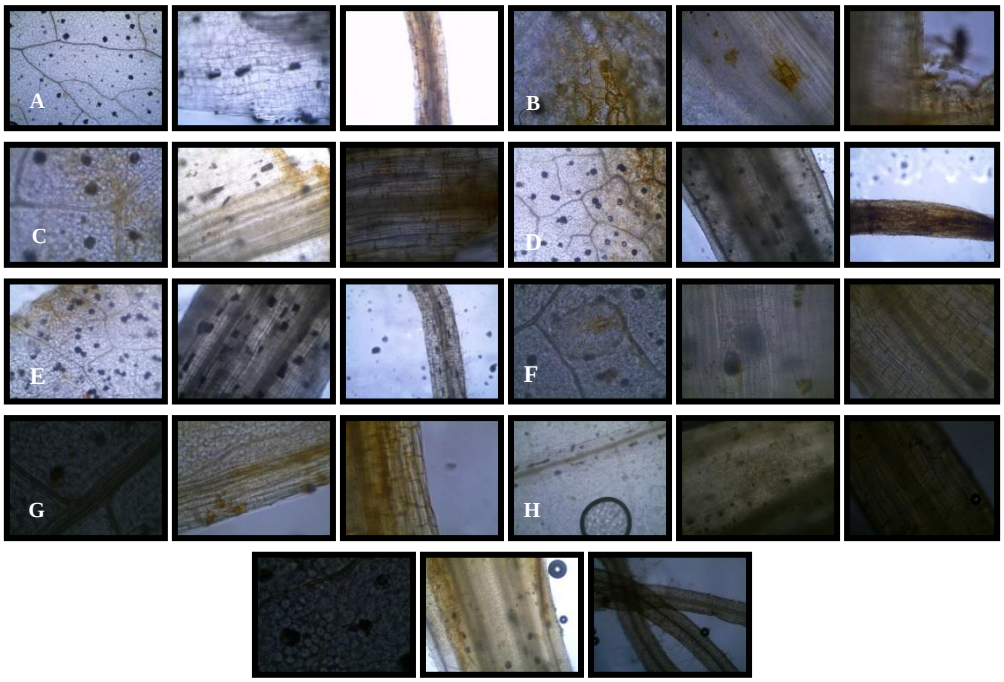

Figure 2. Localization of H_2O_2 after treatment of the plant activator and PVY infection. (**A:** Control, **B:** Actigard 72 hrs 0.34 g/l, **C:** Actigard 96 hrs 0.34 g/l, **D:** Messenger 72 hrs 1.2 g/l, **E:** Messenger 96 hrs 1.2 g/l, **F:** ISR-2000 72 hrs 4 ml/l, **G:** ISR-2000 96 hrs 4 ml/l, **H:** Crop-Set 72 hrs 1.2 ml/l **I:** Crop-Set 96 hrs 1.2 ml/l, leaf-stem-root in order)

As a result of the staining studies, it was found that the activator-treated plants were stimulated durability and the best treatment time was 96 hrs for Actigard, ISR-2000 and Crop-Set treatments while

Messenger treatment is set for 72 hrs. Similarly, Li et al. (2016) reported that H_2O_2 accumulated in the leaves of tomato plants treated with in *Enterobacter asburiae* BQ9 and inoculated with TYLCV. Çalışkan and Kamberoğlu (2010) recorded that the highest H_2O_2 accumulation in the plant activators (Actigard, Messenger and ISR-2000) used in the squash against ZYMV inoculation was observed to be 72 hrs.

CONCLUSIONS

Our results with the activator-virus combination revealed some differences from other studies. These differences may be due to differences in the plant species used, activator application doses and plant growth conditions. Our findings are promising for future studies to control PVY, which limits pepper cultivation, since this virus can not be controlled by chemical sprays. We suggest the combined use of the plant activators with insecticides that are specific to the aphids. Our observations imply that both the suppression of PVY in the plant and the transfer of the virus vectors to other plants can be prevented.

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