# EXAMINING THE EFFECT OF SALICYLIC ACID AND JASMONIC ACID ON THE EXPRESSION OF PR1, NPR1, PAL AND WRKY GENES OF MELON AGAINST ZUCCHINI YELLOW MOSAIC VIRUS

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#### Abstract

Zucchini yellow mosaic virus (ZYMV) is one of the critical viruses of the cucurbits with worldwide spread leading to significant damage to them. The induction of pathogen-related genes with a role in disease resistance can affect the severe reduction of the disease. The evidence shows that plant hormones are effective in expressing these genes among plants. Furthermore, it stimulates the plant immune system through the activation of transcription of genes involved in the plant defense mechanism and leads to long-term resistance against a wide range of pathogens like viruses. The study examined the expression of PR1, NPR1, PAL, and WRKY genes in melon plants treated with salicylic acid at a concentration of 3 mM and jasmonic acid at a concentration of 0.5 mM and the control plant after inoculation using ZYMV in four different time intervals using Quantitative Real-time PCR (QRT-PCR) technique. The results revealed that treatment with these two hormones, particularly salicylic acid, led to great changes in the expression of these genes following inoculation of the virus into the plant. For the first time in Iran, the study examined the induction of PR1, NPR1, PAL, and WRKY genes in melon resistance to ZYMV.

**Keywords**; Pathogenesis-related proteins, resistance genes, systemic acquired resistance, QRT-PCR

## Introduction

Melon is a plant from the cucurbits genus whose fruit is large, sweet, and juicy. The plant is short and its stems lie on the ground. Melon is a plant from the Cucurbitaceae family with the scientific name Cucumis melo var. indoors. Melon is one of the summer crops. Iran is the third-largest producer of these summer crops in the world after China and Turkey with an annual production of more than 1.5 million tons of melons, yet it is the twelfth exporter of this crop (FAO, 2012).

The members of the C. Melo species are highly susceptible to viral diseases. Potyviruses, especially ZYMV, are the largest and most economical group of viruses damaging the production of these products. The virus has spread to all summer crop areas of Iran and is considered a limiting factor in the cultivation of cucurbits in summer and autumn. ZYMV infection leads up to 100% damage in some cases causes (Bananej et al., 2008). The widespread distribution of ZYMV in Iran and its high losses in cucurbits production areas necessitates finding a suitable approach to control the virus.

Disease resistance strategy is a key component of modern agriculture decreasing using chemicals and enhancing healthy agriculture. Plants defend themselves against pathogens through structural mechanisms and induced resistance (Sticher et al., 1997; Heil & Bostock 2002). Using genetic engineering is an efficient approach for producing pathogen-resistant plants (Lillemo et al., 2008; He et al., 2009). Here, resistance genes, especially the ones synthesizing pathogenesis-related protein (PR), have received the most attention, able to be used in the production of transgenic plants that are resistant to stressors (Van Loon & Pieterse 2006; Kogel & Langen 2005). Salicylic acid has a key role in the response to biological stresses and the induction of systemic acquired resistance (SAR) against viral diseases (Flacioni et al., 2014) proven to be compatible with plants in non-biological stresses (Kang et al., 2014). This hormone betters resistance to biological and abiotic stresses by expressing pathogen-related protein (PRs) and antioxidant enzymes (Hayat et al., 2009; Mahmoodi Jaraghili et al., 2016). Jasmonic acid (JA) and its methyl ester can increase plant resistance to biological and abiotic stresses (El-Khallal, 2007). The accumulation of salicylic acid (SA) and JA in plant tissues is one of the first events of hypersensitivity reaction (Anderson et al., 2006), induced by the invasion of pathogens leading to the production of reactive oxygen species (ROS). Although these radicals act as secondary peaks at first in inducing resistance,

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their accumulation in high concentrations ends in extensive damage to plant tissues (Yadava et al., 2015). Plant mechanisms against pathogens are regulated by a complex network of signal transduction pathways containing interacting molecules such as SA, JA, ROS, and ethylene (Kunkel & Brooks, 2002).

Among the proteins related to pathogenicity, PR1 is the key in resistance to different pathogens (Van-Loon & Pieterse, 2006). Although no specific biochemical function has been stated for it, many researchers have reported its antimicrobial role (Rauscher et al., 1999; Sels et al., 2008). This protein is effective in the membrane and may have a role in thickening the cell wall and preventing pathogen spread in the apoplast. It has been identified that PR1 genes in tobacco are induced by signaling systems that depend on SA and JA or ethylene pathways. JA- or ethylene-dependent pathways have a role in inducing the expression of some alkaline PR1 genes, whereas the SA-dependent pathway is effective in inducing acidic PR1 genes (Vidhyasekaran, 2002).

Non-expresser of pathogenesis-related protein 1 (NPR1) known as Non-inducible immunity (NIM1) and Salicylic acid-insensitive (SAI1) is a key regulator making the plants resistant to a wide range of pathogens with a key role in SAR and ISR. This gene affects the downstream pathway of the SA signal (Bai et al., 2011). NPR1 protein group has been isolated in tobacco, Arabidopsis, wheat, barley, and many other plants so far (Glazebrook et al., 1996). NPR1 gene is naturally expressed in Arabidopsis and can be stimulated by SA treatment or a variety of analogues or by the pathogen. Nonetheless, normal expression of NPR1 in the absence of stimulants does not result in expressing PRs genes, which shows that increased expression of NPR1 protein is necessary along the SAR pathway (Yuan et al., 2007). The increase in the expression of NPR1 in tomatoes increases the resistance to the tomato mosaic virus (Lin et al., 2004).

Based on the reports on the involvement of phenylpropanoid biochemical pathway derivatives against different biological and abiotic stresses (Campbell & Elis 1992), one can mention the study of the changes in important enzymes in this pathway, including phenylalanine ammonia-lyase (PAL). PAL gene has a key role in plant resistance by interfering with the synthesis pathways of phenylpropanoids and isoflavones that have phytoalexin activity. This gene is involved in the biosynthesis pathway of SA and other defense-related compounds and is a key signaling compound for the activation of defense-dependent genes, catalysts, and transcription factors (Stotz et al., 2004).

Changes in intrinsic signaling pathways like SAR and ISR can activate some transcription factors and increase the expression of a large number of defense genes. The main problems of activating all signaling pathways are the reduction in cost-effectiveness and potential performance related to the structural expression of a large number of genes, so the genes activating parts of the pathway or amplify the pathway are ideal candidates. Potential candidate genes for genetic engineering include transcription factors like WRKY, ERF1, and NPR1. WRKY gene family shows the key group of transcriptional regulatory factors (Zhang & Wang, 2005). Binding transcription factors to DNA is the key to the activation of PRs genes (Van-Loon & Pieterse, 2006). Some WRKY genes have been identified in different plants. Increased expression of the WRKY8 gene in plants of the Cruciferae family has resulted in resistance to tobacco mosaic virus (TMV) (Chen et al., 2013). The increased expression of the ptrWRKY1 gene in poncire has been reported against the citrus thyristor virus (Sahin-Cevik et al., 2014). It has been suggested that WRKY is a transcriptional regulator in SA and JA-dependent signal cascades (Song & Goodman, 2001).

Few studies have been carried out on the use of plant hormones to induce the expression of genes associated with pathogenicity and resistance in plants. Hence, the purpose of the study was to examine the expression of PR1, NPR1, PAL, and CmWRKY6 genes at the level of vector prescription treated with SA and JA hormones in melon so that it is possible by better understanding the induction pathway and response of melon to ZYMV.

#### Materials and methods

An experiment was carried out as three treatments and three replications to examine the effect of two plant hormones JA and SA on the expression of some genes examined in this study in melon plants infected with ZYMV. Firstly, the substrates were prepared for sowing melon seeds (ghasri danab variety 201, Anbar Sabz Company), so that some cultivable and fertile soil was prepared and sterilized for one hour at 121 °C and one-atmosphere pressure. Sterile soil was transferred to culture media (paper cups and plastic boxes) and melon seeds were cultured after surface disinfection with 1% sodium hypochlorite solution and rinsing with sterile distilled water (Veladi et al., 2013; Ibrahim 2012; Hussein et al., 2016). About 4 weeks after sowing the seeds, the seedlings were transferred to the main pots in the two-leaf stage. A particular space was selected and enclosed with a fabric net to prevent the entry of vector insects to prepare a suitable environment for seedling growth and apply various treatments in the greenhouse. The pots were sterilized with 5% sodium hypochlorite solution and soil in an autoclave. Overall, three treatments - ZYMV with SA, ZYMV with JA, and water (as a control) - were considered in three replications in a completely randomized design. Some seedlings were sprayed with JA solution at a concentration of 0.5 mmol. To dissolve JA, it was first dissolved in ethanol (4% by volume) and then brought to a certain volume with distilled water (Traw & Bergelson, 2003). Moreover, some seedlings were

treated with SA solution at a concentration of 3 mmol. To dissolve SA, it was first dissolved in ethanol (10% by volume) and then diluted to a certain volume with distilled water (Muchembled et al., 2006). The seedlings treated with JA were kept in the growth chamber for 7 days and those treated with SA for 2 days under the plastic cover. Some seedlings were considered as controls. After the time specified following spraying, the seedlings were mechanically inoculated with ZYMV (received from the Sari University of Mazandaran) propagated on the zucchini. Thus, one milliliter of inoculation buffer was used and the plant tissue was well ground inside a porcelain mortar and the resulting extract was used for inoculation for 0.1 g of plant tissue (Sudhaker et al., 2006). A sampling of leaf tissue of treated and control seedlings was done on 0, 4, 8, and 15 days post inoculation (dpi) and stored in a freezer at -70 ° C.

#### Extraction of total RNA from samples and cDNA fabrication

TRIZOL (Invitrogen Co.) buffer was used to extract RNA from samples stored in a freezer at -70  $^{\circ}$  C. Fermentas RQ1 RNase-free DNase kit was used to remove genomic DNA from RNA samples. RNA quantity and quality were assessed with agarose gel and spectrophotometer.

### Fabrication and evaluation of cDNA quality

AccuPowerR CycleScript RT PreMix (dN6) kit was used for CDNA synthesis. The samples were stored at -20 °C until use. PCR with Actin-specific primers was used to assess the quality of synthesized cDNA samples.

## Examining the expression of PR1, NPR1, PAL, and WRKY genes by Quantitative real-time PCR

Gene expression was evaluated using quantitative real-time PCR. Applied Biosystems® StepOnePlus <sup>TM</sup> Real-Time PCR was used for doing so. Maxima SYBR Green / R kit Thermo OX qPCR Master Mix (2X) was used in this reaction. The specific primer of the Actin gene was used as housekeeping and the primer of PR1, NPR1, PAL, and CmWRKY6 genes for cDNA amplification in RT-PCR to examine gene expression. Each PCR reaction was performed in a final volume of 15  $\mu$ l containing one  $\mu$ l of 11 templates, 8  $\mu$ l of cybergreen mixture, 1  $\mu$ l of each primer, and 4  $\mu$ l of 12 free water. PCR reaction has the initial step at 95°C for 10 minutes, then 40 cycles involving 95 ° C for 15 seconds, 60 ° C for one minute to connect the primers, then plotting the melting curve at 95 ° C for 15 seconds, 60 ° C for one minute and increasing the temperature of one ° C in each cycle to 95 ° C and keeping at this temperature for 15 seconds. The amplification was measured at the end of each cycle by the tool. The relative changes in the expression of the studied genes compared to the actin gene were normalized. Ultimately, the expression and changes in gene expression were measured according to formula 2- ( $\Delta \Delta CT$ ) (Livak & Schmittgen, 2001).

 $\Delta\Delta C_T = (\Delta C_T \text{ of the control sample} - \Delta C_T \text{ of the test sample})$ 

 $\Delta C_T = (C_T \text{ of housekeeping gene - } C_T \text{ of target gene})$ 

### **Results and discussion**

The study examined the expression pattern of several genes in the melon plant throughout 4 periods after inoculation with ZYMV in three treatments SA, JA, and control. Real-time PCR outcomes confirmed that the expression level of PR1, NPR1, PAL, and CmWRKY6 genes increased in both SA, JA, and control treatments after virus infection. Nonetheless, the study of changes in gene expression shows that the level of expression of these genes at various times after inoculation in these three treatments varies.

In the control plant, the expression of the PR1a gene in response to viral infection increased slightly from day 4 after inoculation and continued until day 8 after inoculation. The expression rate of the gene at the peak time was about 1.72 times higher than the zero time. Then the expression rate on the 15th day after inoculation showed a decreasing trend.

In plants treated with SA hormone, PR1a gene expression in response to viral infection increased significantly 4 days after inoculation (4.43 times zero time). After that, the gene expression continued to increase up to 8 days after inoculation, and the maximum expression of this gene was observed on the same day (7.45 times zero time). The expression rate of this gene decreased after the peak time and on the 15th day after inoculation, it reached its lowest level of expression - about 1.81 times more than the expression of this gene at time zero.

In plants treated with JA, the expression of this gene in response to viral infection started to increase on day 4 after inoculation and its expression went on to increase until day 8 after inoculation, and the maximum expression at the same time was observed (about 2.36 times zero time). The expression rate of this gene diminished following the peak time and reached its lowest level of expression - about 0.28 times more than the expression of this gene at time zero - on the 15th day following inoculation. The rate of expression of this gene in plants treated with JA at the peak of expression (8 days after inoculation) was 1.37 times higher than the expression of this gene in the control plants in the same period (Figure 1).

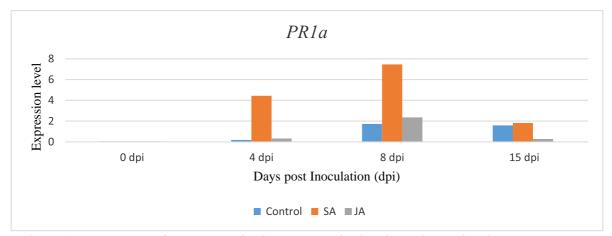


Figure 1- PR1a pattern of gene expression in control, salicylic acid- or jasmonic acid-treated plants at various times after inoculation with ZYMV

PR1 protein exists in various plant tissues like cell walls, vascular bundles, and vacuoles (Hoegen et al., 2002; Grunwald et al., 2003). PR1s are a key index of systemic acquired resistance (SAR) (Schulthesis et al., 2003). The role of the acidic PR1 gene in the defense of ethephon-treated tobacco against tobacco mosaic virus (TMV) has been proven (Bol et al., 1993). In this study, the expression of the PR1a gene increased on the first days after inoculation, which according to the analysis of sap extracted from the phloem vascular area of plants inoculated with the virus, showed that the first increase in SA accumulation on day 4 after virus inoculation. This increase in expression in all three treatments continued until the eighth day after inoculation. High expression of the PR1a gene in both control plants and plants treated with SA and JA hormones shows the induction of this gene in the interaction of ZYMV with melon. Accumulation of copies of this gene on the 15th day after inoculation in all three treatments had a decreasing trend, probably because of the reduced proliferation and movement of the virus in the treated plants. The expression level of this gene in plants treated with SA was significantly higher than control plants in all periods after inoculation. Hence, increasing the expression of this gene on the 8th day after inoculation, causes programmed cell death to prevent the development of pathogens in the host cell and finally results in plant resistance. Some findings of other scholars show an increase in the expression of this gene in plants after inoculation with the pathogen (Bol et al., 1990).

Moreover, given the increased expression of PR1a gene on day 4 after inoculation and its recognition as a marker of induced resistance stimulated by SA (Bol et al., 1993) as well as the scholars' findings according to which SA is usually effective in protecting plants against viral pathogens (Falcioni et al., 2014), one can conclude that ZYMV in interaction with melons, initially responds to SA pathogen attack by activating SA-dependent defense pathways.

In the control plants, the expression of the NPR1 gene in response to viral infection increased significantly on day 4 after inoculation (about 2.14 times zero time). The expression of this gene continued to increase until day 8 after inoculation and the maximum expression of this gene was at the same time (about 2.9 times zero time). The expression rate of this gene diminished following the peak time and on the 15th day after inoculation reached its lowest expression level, about 1.95 times higher than the expression of this gene at zero time.

In plants treated with SA hormone, the expression level of the NPR1 gene in response to ZYMV reached its maximum expression on day 8 after inoculation and a significant mutation was observed (15.6 times increase compared to zero time). The expression rate of this gene diminished following the peak time and on the 15th day after inoculation reached its lowest expression level, about 1.71 times higher than the expression of this gene at zero time. The expression rate of this gene in SA-treated plants at the peak of expression (day 8 after inoculation) is about 5 times higher than the expression of this gene in control plants at the peak of expression (day 8 after inoculation).

In the plants treated with JA, the expression of the NPR1 gene in response to viral infection increased from day 4 after inoculation (2.88 times compared to time zero), and then the expression of this gene continued to increase until day 8 after inoculation and the maximum expression of the gene was seen on the same day (about 3.75 times increase compared to zero time). The expression rate of this gene diminished following the peak time and

on the 15th day of infection reached its lowest level of expression, 0.93 times more than the expression of this gene at time zero. The expression rate of this gene in JA-treated plants at the peak of expression (day 8 after inoculation) is 1.29 times higher than the expression of the gene in control plants at the peak of expression (day 8 after inoculation) (Figure 2).

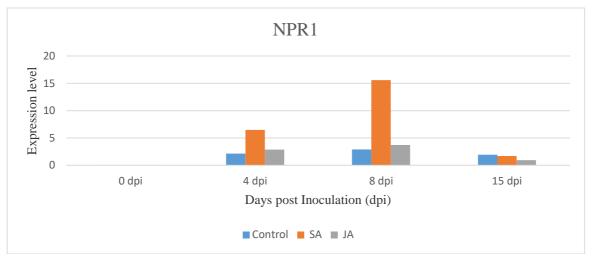


Figure 2- NPRI pattern of gene expression in control, salicylic acid, or jasmonic acid-treated plants at various times after inoculation with ZYMV.

NPR1 protein is a key regulator leading to the plants resistance to a wide range of pathogens with an important role in SAR and ISR. The gene affects the downstream pathway of the SA signal. The scholars have indicated that the expression of the NPR1 gene in SA and Arabidopsis treated with SA induces resistance to CMV cucumber mosaic virus and oilseed rape mosaic virus (ORMV) (Huang et al., 2005). In this study, the expression of the NPR1 gene in all three treatments increased after inoculation of plants with ZYMV, yet the expression varied in the treatments. As in the SA-treated plant, gene expression peaked on day 8 after inoculation in the control plant, yet its expression was much lower than in the SA treatment, which shows the significance of the SA hormone in the interaction of melon with ZYMV. Nonetheless, in JA treatment on the 8th day after inoculation, it had the highest expression, with a lower rate of expression compared to SA. Thus, the NPR1 gene expression rate in the plant treated with JA in all intervals is proportional to the plant treated with SA less and more than the control plant. The findings of other researchers show an increase in the expression of this gene after infection in plants (Glazebrook et al., 1996; Reuber & Ausubel 1996; Shah et al., 1997; Clarke et al., 2000; Kachroo et al., 2000). This increase may be justified by the fact that SA treatment increases the SA signal molecule which activates the NPR1 gene and thus increases PR1 gene expression inducing the SAR mechanism in the plant (Shah & Klessing 1991). Hence, the plant causes programmed cell death to prevent the development of the pathogen in the host cell leading to plant resistance by increasing the expression of this gene on the 8th day after inoculation.

In control plants, the expression of the PAL gene in response to viral infection started to increase on day 4 after inoculation that was about 0.9 times the zero time which was at its lowest expression level. After that, the expression of this gene continued to increase until the 8th day after inoculation, and the maximum expression of this gene was observed on the same day (about 5.25 times increase compared to zero time). The expression rate of this gene decreased slightly after the peak time and on the 15th day after inoculation, the expression rate of this gene reached about 3.56 times more than zero time.

In the plants treated with SA hormone, PAL gene expression increased significantly on day 8 after inoculation, and the maximum expression of this gene was observed at the same time in response to viral infection (about 15.34 times increase compared to time zero). The expression rate of this gene decreased slightly after the peak time the expression rate of this gene was about 6.28 times more than zero time and on the 15th day after inoculation. The lowest expression of the gene on day 4 after inoculation was about 2.53 times higher than the zero time. Moreover, the expression rate of this gene in plants treated with SA at the expression peak (on the 8th day after inoculation) was 2.9 times higher than the expression of this gene in control plants at the peak of

expression (on the 8th day after inoculation).

In JA-treated plants, the expression of the gene increased slightly on day 4 after inoculation (about 1.76 times the increase over time zero) in response to viral infection and then increased up to day 8 after inoculation that was the significant and the maximum expression of this gene (about 11.81 times higher than zero time). The expression rate of this gene on the 15th day after inoculation decreased slightly compared to the 8th day after inoculation to its expression level, about 10.44 times higher than zero time. Furthermore, the expression rate of this gene in plants treated with JA at the peak of expression (8 days after inoculation) was 2.24 times higher than the expression of this gene in control plants at the peak of expression (on the 8th day after inoculation) (Figure 3).

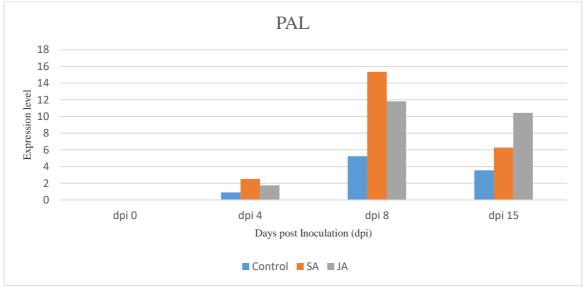


Figure 3- PAL pattern of gene expression in control plants, treated with salicylic acid, jasmonic acid at various times after inoculation with ZYMV.

The enzyme phenylalanine ammonia-lyase is the first and the key enzyme in the phenylpropanoid pathway that converts L-phenylalanine to ammonium ions and Trans cinnamic acid by deamination (Brooks et al., 2005). In most cases, the variations in the level of expression of this enzyme are related to the susceptibility of the various cultivars of a plant to disease (Xu et al., 2010). In different studies, the increased PAL gene expression has been related to the increased PRs gene expression as well as rapid induction of cell death (Fitzgerald et al., 2004). Given the information gathered and the points stated on examining the role of PAL, one can be stated that this gene has a key role in the resistance of SA-treated tobacco to TMV virus (Malamy et al., 1990; Yalpani et al., 1991). The increase in the expression of the PALs gene in the SA-treated maize plant increases resistance to sugarcane mosaic virus (SCMV) by increasing the accumulation of antimicrobial phenolic compounds as well as stimulating lignin accumulation (Yuan et al., 2019).

As seen, the expression of the gene in plants treated with SA and JA is relatively higher in peak days. Based on the previous studies, the addition of SA and JA (as a treatment) by binding to membrane receptors led to the production of oxygen, NO, protein kinases, SA, and methyl jasmonate (Raman and Ravi, 2011). SA and JA possess specific signaling pathways, yet each can inhibit or stimulate the other. The need for JA during R-gene resistance to the virus is very complicated and different. The study showed that the expression path of the PAL gene in plants treated with JA on the 15th day after inoculation was significantly higher than SA on the 15th day after inoculation, which indicates the importance of JA in increasing the expression of this gene. Hence, it has been indicated that JA signaling is required for resistance induced by the N gene to the tobacco mosaic virus (Liu et al., 2004). The value of JA and its metabolic precursor exophytoditoic acid increases in TMV-resistant tobacco plants (Dhondt et al., 2000).

In the control plants, the expression level of the CmWRKY6 gene in response to viral infection started to increase on the 4th day after inoculation and continued to increase until the 15th day after inoculation. The gene expression rate at the peak time was about 2.43 times higher than the zero time.

In SA hormone-treated plants, the expression of this gene in response to viral infection increased maximally on day 4 after inoculation (about 8.24 times higher than time zero). The expression rate of this gene decreased slightly after the peak time, after which on the 8th day after inoculation, the expression of this gene decreased slightly (about 4.18 times the increase compared to the zero time). The expression rate of this gene on the 15th day after inoculation reached its lowest level of expression, which is about 2.81 times higher than the expression of this gene at time zero. The expression rate of this gene in SA-treated plants at the peak of expression (day 4 after inoculation) is 3.39 times higher than the expression of this gene in control plants at the peak of expression (on the 15th day after inoculation).

In JA-treated plants, the expression of the gene in response to a viral infection on the 4th day after inoculation increased drastically, and then the expression of this gene continued to increase until the 8th day after inoculation and the maximum expression this gene was also observed on the same day (about 3.94 times increase compared to time zero). The expression rate of the gene diminished after the peak time and on the 15th day after inoculation, it reached its lowest expression level, which is about 1.17 times more than the expression of this gene at zero time. The expression rate of this gene in JA-treated plants at the peak of expression (on the 8th day after inoculation) was 1.62 times higher than the expression of this gene in control plants at the peak of expression (on the 15th day after inoculation) (Figure 4).

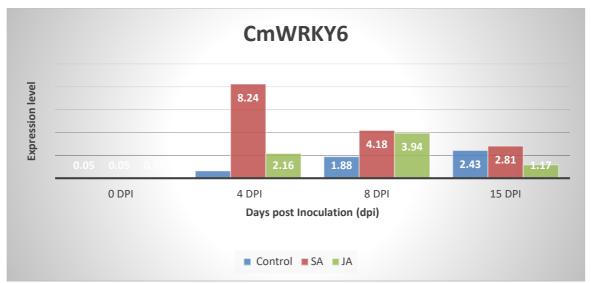


Figure 4- Cm WRKY6 pattern of gene expression in control plants, treated with salicylic acid, jasmonic acid at various times after inoculation with ZYMV.

Binding of the factors WRKY TFs transcription to SA and JA is important for activation of downstream genes of PRs and NPR1 (Huang et al., 2016). Suppression of these transcription factors prevents the expression of PR genes (Boyle et al., 2009). Increased expression of WRKY Group III genes in tomatoes results in the resistance to Tomato yellow leaf curly virus (TYLCV) (Huang et al., 2016). In transgenic pepper plants, the CaWRKY1 gene increases the resistance of the pepper plant to Pseudomonas syringe and tobacco mosaic virus (TMV) by increasing the hypersensitivity reaction and eventually cell death (Oh et al., 2008).

In this study, considering the maximum expression of the WRKY gene in plants treated with SA on day 4 after virus inoculation and other results, one can be stated that this gene has a key role in defense signaling pathways and gene expression like PR1. The findings of other studies showed an increase in the expression of this gene after infection in plants (Pandey & Somssich, 2009). Significant differences in WRKY gene expression in the control and plants the ones treated with SA at all times indicate the important role of SA in defense against ZYMV in melon.

Viruses are destructive pathogens and our knowledge of virus pathogenicity and plant host resistance is not understood completely yet. The plants use a complex network of signaling pathways to increase resistance responses. The ability of NPR1 to participate in SA-dependent signaling pathways points to the importance of NPR1 as a key convergence point in the development of systemic resistance pathways. According to our

findings, SA hormone can induce more PR1, PAL, and CmWRKY6 transcription genes compared to JA in melon. In general, one can state that these hormones are effective as bioinducers and make the melon plant resistant to cucurbits yellow mosaic virus by inducing the gene expression system and producing enzymes. Given the greater effect of SA hormone, one can use it in the management of this disease by treating it on the plant or producing transgenics of the melon where the production of this hormone is higher or transgenics with higher levels of NPR1 and PAL genes. The studies on SA as well as plant defense enhance our understanding of related physiological and molecular processes. SA interacts positively and negatively with many hormones of the plant and other signal molecules, affecting not only the defense but also the regulation of plant growth and development processes. A better understanding of the systemic resistance signaling pathway can lead to safer methods of discussion that are richer than what protects the product. Induction of systemic acquired resistance to control field plant contamination is achieved by the use of SA derivatives such as benzothiadiazole. Systemic acquired resistance can be a great goal for controlling the damage of plant diseases, including viruses. The ability to target this pathway to reduce dependence on toxic chemicals is used to protect plants from the insects that carry the viruses without jeopardizing the health of the environment and humans.

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