## T.C. SIIRT UNIVERSITY GRADUATE SCHOOL OF NATURAL AND APPLIED SCIENCES DEPARTMENT OF HORTICULTURE

## EFFECTS OF SALICYLIC ACID APPLICATION ON COLD TOLERANCE AND GENE EXPRESSION IN PEPPER (*Capsicum annuum* L.) SEEDLING.

**MASTER'S DEGREE** 

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### THESIS ACCEPTANCE AND APPROVAL

The thesis study entitled "Effects of Salicylic Acid Application on Cold Tolerance and Gene Expression in Pepper (*Capsicum annuum* L.) Seedling." prepared by Mohammed Ahmed Ahmed has been accepted as a Master's Thesis at Siirt University, Institute of Science and Technology, Department of Horticulture, by unanimity of votes by the following January on 04/01/2019.

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I confirm the above conclusion.

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### THESIS NOTIFICATION

I hereby declare under the penalty of perjury that this paper is my unique authorial work, which I have done on my own. All the information reference and literature used or excerpted through the explanation of this work are correctly cited and listed incomplete reference to the owing caue.

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Be a scientist... If you cannot be educated, if you cannot love scientists, if not you cannot hate them.

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# ABBREVIATIONS AND SYMBOLS

<u>Abbreviation</u>	Explanation
SA	: Salicylic Acid
LT	: Low Temperature
L	: Liter
mmol	: Millimol
Тетр.	: Temperature
Repl.	: Replication
h	: Hour
g	: Gram
mg	: Milligram
cm	: Centimeter
ml	: Milliliter
m	: Meter
mm	: Millimeter
T.S.S	: Total Soluble Solids
NaOH	: Sodium hydroxide
μm	: Micron
ASA	: AcetylSalicylic Acid
SAR	: Systemic Acquired Resistance
MSL	: Mean Sea Level
IBA	: Indoul Butric Acide
Mg	: Micrograms
ROS	: Reactive Oxygen Species
RNS	: Reactive Nitrogen Species
JA	: Jasmonic Acid
RLWC	: Relative leaf Water Contents
MDA	: Malondialdehyde
NR	: Nitrate Reductase
BR	: Brassinolide
ASA	: Acetylsalicylic Acid
AOX	: Alternative Oxidase
PSB	: Potato Sucrose Broth
IAA	: Indole-3- Acetic Acid
GA <sub>3</sub>	: Gibberellic Acid
CKs	: Cytokinins
ABA	: abscisic acid
GSH N-Cl	: Glutathione
NaCl	: Sodium chloride
Cd	: Cadmium
RCPD GB	: Randomized Complete Parcel Design
GD BW	: Glycine betaine : Bacterial Wilt
HR	
GTA	: Hypersensitive response : Gentisic acid
-	: parts-per-million
ppm CaHB <sub>1</sub>	: Capsicum annuum home box 1
	: Using Visible
NO <sub>2</sub> -Tyr	: Tyrosine nitration
1102-1 yl	

SOD	: Superoxide dismutase
NPR1	: Non-expression of pathogenesis-related genes1
PCR	: Polymerase Chain Reaction
ALA	: Aminolifulcin acid
W1A2	: Water stress1 x Applcation2
W1	: Water stress1
W2	: Water stress2
W3	: Water stress3

<b>Explanation</b>
: Centigrade : Percent

## ÖZET

### YÜKSEK LİSANS

## SALİSİLİK ASİT UYGULAMALARININ BİBER (*Capsicum annuum* L.) FİDELERİNDE SOĞUĞA TOLERANTLIK VE GEN İFADESİ ÜZERİNE ETKİLERİ

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Bu proje, salisilik asidin düşük sıcaklık (0 <sup>o</sup>C) koşullarında yetiştirilen biber fidelerinin gelişimine katkısını araştırmak amacıyla geliştirilmiştir. Araştırma, Siirt Üniversitesi Ziraat Fakültesi Bahçe Bitkileri Bölümü araştırma-deneme alanındaki kontrollü bitki yetiştirme dolabında yürütülmüştür. Bitkisel materyal olarak, örtüaltı yetiştiriciliğinde kullanılan Urartu F1 biber çeşidi (kapya) kullanılmıştır.

Farklı salisilik asit dozu olarak; 0,01 ve 0,05 mmol dozları kullanılmıştır. 0 mmol salisilik asit dozu ise kontrol grubu olarak kullanılmıştır. Uygulama sıklığı olarak; 1 kez, 2 kez ve 3 kez şeklinde uygulanmıştır. Ayrıca 3 farklı soğuk uygulama süresi de araştırılmıştır; 24 saat, 48 saat ve 72 saat olmak üzere. Deneme tesadüf parsellerinde ve 3 tekrarlamalı olarak dizayn edilmiştir.

Biber fidelerinde, fidelerde kayıp ağırlık oranı, fidelerde kayıp boy oranı, fidelerde turgorite durumu, yaşayan fide oranı, soğuk zararı belirtileri ve uygulama sonrası fidelerin gelişmelerine devam etme oranı ile salisilik asidin soğuk uygulamasına maruz bırakılan biber fidelerinde WRKY genleri üzerindeki etkileri de incelenmiştir.

Araştırma sonunda; fidelerde Turgorite ve Soğuk Zararı sonuçları arasında istatistiki anlamda bir farkın olmadığı tespit edilmiştir. Salisilik asidin 0,01 mmol dozu, fidelerin boy ve ağırlıklardaki kayıp oranında pozitif etki gösterirken, 0,05 mmol dozu ise Turgoritenin korunmasında en pozitif etkiyi göstermiştir. Salisilik asidin kullanılmadığı kontrol grubu ise fidelerde soğuk uygulaması sonrası solgunluk oranında pozitif sonuca sahip olmuştur. Düşük sıcaklıklarda bekletme süreleri arasında, 24 saatlik bekletme, solgunluk dışındaki diğer tüm ölçümlerde en iyi süre olarak kaydedilmiştir. 3 kez uygulama yapılan sıklık uygulaması diğer (1 kez ve 2 kez) uygulamalardan daha iyi sonuç verdiği belirlenmiştir. Uygulanan her iki salisilik asit uygulaması göz önüne alındığında biber bitkisinin WRKY geninin ifadesi açısından 0,05 mmol, SA uygulamasında daha net ve anlaşılır bir tepki verdiği bulunmuştur

Anahtar Kelimeler; Düşük sıcaklık, Salisilik asit, Biber, WRKY genleri

#### ABSTRACT

#### **MS THESIS**

### EFFECTS OF SALICYLIC ACID APPLICATION ON COLD TOLERANCE AND GENE EXPRESSION IN PEPPER (*Capsicum annuum* L.) SEEDLING.

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This project was developed to investigate the contribution of salicylic acid to development of pepper seedlings grown in low temperature (0  $^{0}$ C) conditions. The research was carried out in the controlled plant growing cabinet in the research- investigation area of the Department of Horticulture, Faculty of Agriculture, Siirt University. As a vegetable material, Urartu F1 pepper type (capia) which is used in greenhouse cultivation has been used.

As a dose of different salicylic acid; 0.01 and 0.05 mmol doses were used. The dose of 0 mmol salicylic acid was used as control group. Application frequency; It was applied 1 time, 2 times and 3 times.

3 different cold application times were also investigated; 24 hours, 48 hours and 72 hours. The experiment was designed in randomized plots and 3 replications.

In the pepper seedlings, the effect on the weight loss of the seedlings, the rate of loss in the seedlings, the turgority of the seedlings, the rate of living seedlings, the signs of cold damage and the rate of continuing the development of seedlings and the effect of the seedlings on the WRKY genes were investigated.

At the end of the research; there was no statistical difference between the results of Turgorite and Cold Loss in seedlings. The 0.01 mmol dose of salicylic acid showed a positive effect on the rate of seedlings while the 0.05 mmol dose showed the most positive effect on turgorite protection. The control group, which did not use salicylic acid, had the most positive result after the application of cold in the seedlings. Between holding times at low temperatures, the 24-hour hold is recorded as the best time for all measurements except wilt. It has been determined that the frequency application performed 3 times is better than the other (1 time and 2 times) applications. Considering the administration of both salicylic acids, it was found that 0.05 mmol dose had a clearer and more understanding response to the expression of WRKY gene.

Key words; Low temperature, Salicylic asid, Pepper, WRKY genes

### **1. INTRODUCTION**

Low temperatures diminish the biosynthetic action of plants; they bring out aggravation in essential capacities and productivity and they may dispense lasting wounds that at long last achieve death. The survival limit of a plant species or variety in a particular environment is determined by as far as possible to which its metabolic procedures keep on working under low-temperature stress and by its cold resistance, the two of which are attributes of its ecophysiological constitution (Larcher, 1968).

The low temperature (LT) is the environmental stress that affects crop production and quality. Regulates the expression of a number of proteins, metabolites and many genes (Mitchell and Moyle, 1967).

Low temperature (LT) is a natural factor that has a huge impact on plant development influencing photosynthesis, take-up of water and supplements, among others. Numerous monetarily critical products, for example, cotton, maize, pepper, rice, soybean, tomato, some tropical organic products (e.g. bananas, papayas and mangoes) and subtropical natural products (e.g. grapes, oranges) are LT touchy, which influences their generation and quality (Sharma et al., 2005).

LT influences pepper vegetative advancement and generation by irritating the capacity of the blossom female organs and the number of reasonable dust grains per bloom (Polowick and Sawhney, 1985; Pressman et al., 1998, 2006; Shaked et al., 2004). Therefore, natural products from plants that have been set under low night temperatures 14 °C or less as a rule are twisted and seedless causing noteworthy prudent misfortunes. Considering the imperative agronomical importance of pepper (Mateos, 2006).

The impact of this kind of stress has been learned at various levels from entire plants to single particles. Be that as it may, depending either on the kind of plants (yearly, half-yearly, bushes or trees) or the force and term of the introduction of plants to LT, the procedures utilized by plants can change. In this way, it has been demonstrated that LT directs the statement of numerous qualities (Shinozaki, et al., 2003), and there are biochemical changes that influence the level of various proteins, lipids, and metabolites. These incorporate the amassing of cry protective peptides, amalgamation of low atomic cry protective sugars (praline and raffinose), liquid catalyst proteins, dehydrins, ROS searching compounds and solvent cancer prevention agents (Thomashow, 1999; Hannah et al., 2005; Sharma et al., 2005; Renaut et al., 2006; Lütz, 2010).

The campaign organized by the Institute of Field Crops 40 years ago proved high accuracy. That the original home of the pepper is located in the central regions of North and South America, and there is a natural pepper (*Capsicum annuum* L.) and various forms in the south of Mexico and Central America and Guatemala as for the following species are found in South America:

C. bolivianum Hazen., C. pubescence R. C. columbianum Hazenb., C. Peruvianum Hazenb.

And spread pepper crop outside the American continent after discovered. And that he was transported to Europe at the end of the fifteenth century, it was first transported to Bulgaria in the middle of the 16th century by the coming to buy land and has spread cultivation to increase its nutritional value. In the eight and nineteenth centuries, Bulgarian farmers played the hikers have a role in spreading it in southern and central Europe as a vegetable crop (Kinkov et al., 1974).

The spread of pepper in Asia with little information but the areas cultivated with pepper in Japan, Korea, India, and Vietnam are cultivated in a few areas. And China exports few quantities of red pepper sauce and the Asian continent prefers to growing varieties small hot fruits, except for the countries near the east that planted large pepper fruits. It also cultivated pepper in the northern regions of Africa such as Marrakech, Tunisia, Egypt and other and the presence of some varieties of pepper small and large fruits in Ethiopia, where it moved to Europe. And there is no economic importance of pepper in Australia. The countries exporting the largest quantities of fresh and red green peppers are Italy, Bulgaria, Hungary, and Spain. Pepper possesses a high biological quality and its ability to charge and store, it has made him one of the important vegetable crops and in the future is expected to increase the area cultivated with pepper and its consumption (Kinkov et al., 1974).

Pepper (*Capsicum annuum* L.) an individual from the *Solanaceae* family, is an imperative harvest, its organic products being the second overall consumable vegetables and phenomenal wellsprings of numerous fundamental supplements for people, particularly vitamin C, Magnesium,  $\beta$ -carotene, Iron, potassium, vitamin B and calcium.

Moreover, some pepper cultivars contain huge amounts of capsaicinoids, a gathering of sharp phenolic-determined mixes with solid physiological and pharmacological properties (Topuz and Ozdemir, 2007).

Sweet pepper (*Capsicum annuum*, L) is known as a most loved and broad vegetable yield over the world, its fruit wealthy in cancer prevention agents, vitamins, and minerals for human eating routine and sound (Mateos et al., 2003).

Along these lines, the developing worldwide request of pepper natural products suggests a few methodologies to expand edit generation and organic product quality through particular horticultural treatment hones (Pascual et al., 2010) or elevating the examination to enhance the plant protection from ecological burdens. Pepper plants are initially from tropic areas and require high-temperature conditions for their advancement. Subsequently, the ideal development temperature is in the vicinity of 25 and 30 °C, such that temperature changes influence an assortment of physiological capacities and morphological improvement. At the point when temperature diminishes underneath 15 °C, pepper development is decreased, and sprout and organic product generation stop (Mercado et al., 1997).

Many studies have tended to use phenolic compounds, including salicylic acid. First isolated in 1828 in the city of Munich, German from the bark of the willow tree it is called salicilin acid, and ten years later by the scientist, it has been called salicylic acid of the by the scientist of the Raffaele Birla in 1874, it was produced in Germany in a commercial manner in a pharmaceutical form by the company Bayer in 1898 under the name Aspirin. Salicylic acid is a crystalline powder that melts at a temperature of 107-109 °C. It is an average solubility in water and so much solubility in organic solvents, characterized as a rapid transition in parts of the plant from the treated areas to other areas (Popova et al., 1997; Raskin, 1992).

Salicylic acid which is an optional plant item performs essential activities in the development and advancement procedures of plants. It is a powerful flagging particle in plants and is engaged with inspiring reactions to biotic and abiotic stresses (Krantev et al., 2008).

These activities incorporate practicing a thermogenic impact (Ansari and Misra, 2007), expanding thermotolerance (Jabbarzadeh et al., 2009), empowering extrinsic

root arrangement (Kling and Meyer, 1983), demonstrating herbicides impact (Shettel and Balke, 1983), lessening leaf shed (Ferrarese et al., 1996), giving protection against pathogens (Alvarez, 2000), manages ethylene biosynthesis (Huang et al., 1993; Srivastava and Dwivedi, 2000) and changing the quality and amount of proteins (Doares et al., 1995). It has been asserted (Ray, 1986) that SA and comparative phenolic mixes practice their impact of giving protection against various anxiety factors in plants corrosive (Apte and Laloraya, 1982) and cytokinins (Ray et al., 1983).

These perceptions and reports on numerous other physiological impacts achieved by SA conjured in a few scientists this substance may be another plant development controller (Hayat et al., 2007). It was indicated that SA could exercise such an impact on NRA in the intervention of plant hormones (Schneider and Wightman, 1974), (Fariduddin et al., 2003) announced expanded NR movement due to low groupings of SA while higher fixations were seen to be inhibitory to NR action in *Brassica juncea*. An investigation by Shettel and Balke, (1983) demonstrated positive connection be tween's chlorophyll substance and aggregate nitrogen in cucumber cotyledons. Additionally, the increment in nitrogen substance and chlorophyll content at bringing down groupings of SA demonstrates this assumes an administrative part amid the biosynthesis of dynamic photosynthetic colors. It is known at the show that plants under anxiety age more quickly than those under ideal conditions (Vaadia et al., 1961).

Moreover, cytokinins are known to have a deferring impact on proteolytic action and leaf senescence (Nooden et al., 1997). It has been contended that the decrease in plastid arrangement and chlorophyll-carotenoid combination in plants presented to push comes about because of the amassing of abscisic corrosive (Duysen and Freeman, 1976) or the lessening in cytokinins levels (Itai and Benzino, 1973).

In another examination completed on plates of culled rice leaves, ethylene biosynthesis was observed to be repressed in 2 h following SA organization (Huang et al., 1993).

In still another investigation performed in spinach (*Spinacea oleracea* L.) suspension culture, ethylene amassing repressed chlorophyll creation (Dalton and Street, 1976; Miguel et al., 2003) watched that stem width and stature of the plants expanded by applying 10-10 and 10-8 M SA. Additionally, use of 10-8 and 10-6 M SA

expanded new stem weight, dry stem weight, and root length. The economically accessible type of salicylic corrosive is acetyl salicylic corrosive (ASA) (Mitchell and Broadhead, 1967).

SA was seen to lessen leaf zone (optional leaf), root development, and in addition protein and chlorophyll (a+b) sum parallel to an expansion in its focus in grain plants which were produced from grain seeds sprouted in SA arrangements of fluctuating fixations and developed in SA-containing milieu (Ananieva et al., 2002).

Khan et al. (2003) found that splashing minute fixations (10-5 mol L<sup>-1</sup>) of SA and ASA on the leaves prompted an expansion in the general photosynthetic yield of soybean and corn. It was accounted for in a similar report that stomatal portability and transpiration expanded while chlorophyll sum stayed unaltered. In thinks about where one-week-old corn and bean seedlings were ASA 50, 250 and 1000 ppm to the root (Canakci and Munzuroglu, 2002) or the leaf (Canakci and Munzuroglu, 2000) caused an expansion in crisp weight reduction and a lessening in transpiration in high focuses. It has been accounted for in an investigation utilizing circles got from essential leaves of one-month-old bean seedlings that chlorophyll and b amount diminished, carotenoid sum stayed unaffected while crisp weight reduction and protein pulverization expanded parallel to the expansion in ASA focus 100, 250 and 500 ppm (Canakci, 2003). ASA was found to prompt the conclusion of stoma pores in high fixations (Saavedra, 1978).

Ecological anxiety adversely influences the elements of the plasma layer which can be measured as the level of lipid peroxidation. The measure of malondialdehyde (MDA) mirrors the level of lipid peroxidation. Concentrate on the impact of SA in connection to oxidative anxiety demonstrates that the level of MDA in leaves stayed unaltered (Gautam and Singh, 2009), diminished (Krantev et al., 2008) in a relationship with poisonous worry in connection to different SA focuses. The situation being what it is, the target of the present investigation is to inspect the physiological and some biochemical impacts of low and high centralizations of salicylic corrosive which manufactures protection against biotic and abiotic worry in plants, on pepper seedlings without some different anxiety factor.

Salicylic acid plays an important role in plant metabolism, it is an organic acid as antioxidants (Rao et al., 2000).

SA it is from chemical compounds have been accounted for as resistance inducers in plants. SA is well regarded as one of the key components for transferring defense alerts. It is realized that SA is a natural product of various plants and incites protection from viral contamination. SA is a complete set of systemically acquired resistance genes stimulates physiological processes, for example, development, stomata conclusion, transpiration rate, photosynthesis and antioxidant capacity (Radwan et al., 2008).

SA is an endogenous development controller of phenolic nature and plays an important role in abiotic push resilience by expanding the activity of antioxidant enzymes and detecting excess reactive oxygen species (ROS), which brought about improving the physiological processing and upgrading of plant development (He and Zhu, 2008).

SA is one phytohormone and the most important growth organizations. It has control of plant development, biochemical and physiological procedures reaction in the plant under environmental pressures like photosynthesis, proline, nitrogen metabolism, production of glycine betaine (GB), metabolism and under environmental stress may provide security for plants against abiotic stress to an anti-oxidant defense system (Fayez and Bazaid, 2014; Khan et al., 2015).

SA in many physiological processes are regulated in a molecule signal produced in the plant for a dose of phenolic nature (Shakirova et al., 2003).

Aspirin, an exchange name for acetylsalicylic corrosive (ASA), is a subordinate of SA and when connected exogenously, it experiences unconstrained hydrolysis and changed over to SA (Popova et al., 1997).

The external application of SA has been reported to have an effect on a wide range of physiological processes including increased tolerance of cold germination in peppers, it also shows its role in the plant's tolerance to non-vital pressures (Korkmaz, 2005).

SA is known to have an important role in membranes by modifying the oxidation balance, and thus resist the negative effects of intermediate oxygen resulting from oxidative stress (Yang et al., 2004).

To reduce the tolerability of plants for salinity the external application of SA was used extensively (Jayakannan et al., 2015).

To increase the state of abiotic stress, SA interferes with various physiological responses such as lipid oxidation-reduction, oxidative system regulation, ion prohibiting, osmotic adjustments and kinases synthesis (Jayakannan et al., 2013).

SA plays roles in the growth and development of plants and is a phenolic plant through the organization of seed germination and vegetative growth. SA the activation of biochemical pathways has a regulatory effect on them in the plants associated with tolerance in plants. Plant growth in under abiotic stress conditions of salicylic acid has an improved effect which was associated in turn membrane stability, photosynthesis, nutrient uptake, growth, water relations, inhibition of ethylene biosynthesis and stomata regulation (Khan et al., 2003; Stevens et al., 2006).

SA is prerequisite for the synthesis of oxygen and/or cytokinin when it is sustainable, SA its increase flower life, induces flowering, and increases cell metabolic rate and retards senescence (Metwally et al., 2003).

To study its effect as a chemical, SA was used by many researchers to resist plants against many pathological factors (Chérif et al., 1992; Buck et al., 2008).

SA is one of the substances similar to hormone and its role is important in the direction the physiological process like ion uptake and transport, stress tolerance as well as membrane permeability and photosynthesis (Noreen et al., 2009).

And when SA was used at a concentration of 50 ppm the result to promote the development and growth of the plant (Azooz and Youssef, 2010), protects the plants from oxidative damage and expanded protection from abiotic worries in numerous plants (Moosavi, 2012).

And when the concentration of 1 mM SA protects the plant from damage with the increased oxidative enzyme in the antioxidant activities and controls the physiological adaptation and a decrease in the level of lipid peroxide (Orabi et al., 2010).

SA treatment led to an increased number of pods and seeds/plant and increase the length of the plant as well as the weight of 100 seeds under drought stress in bean plants (Ali and Mahmoud, 2013).

SA is a compound naturally used in the fight against diseases, post-harvest in fruits such as tangerine (Zheng and Zhang, 2004) and sweet cherry (Yao and Tian,

2005) and apples (Yu and Zheng, 2006) and strawberries (Zhang et al., 2004) and others, and SA her direct effect on pathogens as well as induced systemic resistance (Uquillas et al., 2004).

SA is one of the plant phenolic derivatives widely used in plant species, and the primer for its production within the plant (Lee et al., 1995). It is classified as a group of plant hormones because of the physiological roles of ions in the growth and flowering of plants and absorption of ions. It also affects the movement of holes and the production of ethylene in plants (Shudo, 1994). It also accelerates the formation of chlorophyll and carotene, accelerating the process of photosynthesis and increasing the activity of certain enzymes (Hayat et al., 2007). Studies have indicated that many plants respond to the treatment with salicylic acid (Martin et al., 2003).

The treatment of gloxinia with a concentration of 20 mg/L of SA resulted in a significant increase in the paper area, while the concentration of 10 mg /L increased the number of leaves, he pointed out that the spraying of plants Jaafari (*Tagetes erecta*), by SA led to a moral increase in the soft and dry walnut and roots length (Jabbar and Saeed, 2017).

And studied the effect of salicylic acid spray on two varieties of African violet plant (*Viola odorata*) noticed a significant increase in the number of leaves compared to non-treatment plants, adding that the treatment led to an increase in the diameter of flowers and the number of flower buds and early flowering (Jabbarzadeh et al., 2009).

Salicylic acid (willow acid) is an aromatic carboxyl acid, its chemical installation (C<sub>6</sub>H<sub>4</sub>(OH)COOH), colorless and extracted naturally from some plants such as white willow and eucalyptus meadows, it can also be manufactured in the laboratory and is the main compound of several drugs known especially aspirin (D'Maris et al., 2011).

Plays many important plant growth regulators roundabout in regulating the growth under tensile environmental. SA is a phenolic compound commonly produced by the plant and it works as an organizer for growth (Aberg, 1981), it can be in the class of plant hormones (Raskin, 1992). The processing SA affects the vital processes in the plants, which include the effectiveness of antioxidant enzymes (Almagro et al., 2009). The germination of the seed (Basra et al., 2007). And the closure of the stomata (Saavedra, 1979). And take the ions and transportation (Alpert et al., 1981). And

permeability of the membrane (Barkosky and Einhelling, 1993). And disease resistance (Park et al., 2009). And photosynthesis and growth rate (Khan et al., 2003). And it stimulates the increase in the content of dissolved proteins and proline (El-Tayeb, 2005).

Several studies have also shown the effect of salicylic acid, in improving growth and obtained for many plants as an internal growth regulator of phenolic nature contribute to give protection against dispersion vital and abiotic for plant as well as the organization of the physiological processes of the plants such as ion absorption, photosynthesis, Heat regulation of flowering, nitrate representation and ethylene production (Fariduddin et al., 2003).

Note that when spraying the coriander plants with SA at a concentration of 0.01 mM has caused a significant increase in the number of inflorescences flowers, the reason for the concentration of spraying 0.01 mM a significant increase in the number of seeds plant and seed yield compared to those plants that were not sprayed (Hesami el at., 2013).

SA is a natural phenolic and is widely available in the plant kingdom. It is characterized as an internal growth organization. It is proven that it regulates many physiological processes of the plant under stress conditions such as photosynthesis, breathing, nutrient absorption, opening and closing of holes, and improves plant tolerance for salinity by increasing non-enzymatic antioxidants such as superoxide enzyme dismutase, catalase enzyme, and peroxidase enzyme (Tufail et al., 2013).

SA has raised the interest of researchers during the last 20 years, due to its importance in bearing plant conditions of water stress, salt stress, heat stress, and Heavy metal stress (Hovrath et al., 2007).

Galal, (2012) found that salicylic acid is used in the agricultural medium of the jujube plant with concentrations of 10-25-50 mg/L his positive effect. Where the highest percentage for the survival of plants emerged alive at concentration 10 mg/L salicylic acid and as well good response to a germination branch and rooting with the same emphasis.

And reported that there was an increase in salinity tolerance in corn plant when using salicylic acid (Hussein et al., 2007).

SA is one of the most common phenolic compounds produced by the plant widely, it has important physiological roles in plant growth, flowering, and ion absorption and it has an impact on the movement of stomata, and it works to speed up the formation of pigments and carotene and accelerate the process of photosynthesis and increasing the activity of some important enzymes, and due to the physiological roles of many of salicylic acid in plant growth and its development and reveals a number of natural plant hormones (Fariduddin et al., 2003).

Al-Shabbani et al. (2013), indicated many studies to the response of many medicinal plants to the treatment with salicylic acid, have noted that the spraying of black cumin (*Nigella sativa* L.) plants of salicylic acid with concentrations of 12 and 20 ml/L by two from sprays the first, one after the planting three weeks and the second, two months after the first spray, results led to a significant increase in plant height and number of branches and leaves and the weight dry the total (vegetative and root), and the number of flowers and the weight of 1000 seeds.

Sprayed the dill plant species local of Salicylic acid, and the concentration of 40 mg/L led to a significant increase in diameter and stem and the proportion of dry matter in the stems and roots (Yasin, 2016).

Spray plants coriander (*Coriandrum sativum* L.) with four SA concentrations 0, 4000, 8000, 10000 and 12000 mM/L, when the high concentration 12000 mM/L has caused a significant increase in dry weight of the plant, while the sprayed plants with a low concentration of 4000 mM/L significantly outperformed the pilot oil ratio (Pouyanfar et al., 2014).

Al- Doughji et al. (2017) found that significant increase in the soft weight of the vegetative group, and the number of flowers inflorescences, and the number of seeds per inflorescence, and weight 100 seed and production of the plant seeds, and the pilot oil when the coriander plants sprinkle the local species with salicylic acid with at concentrations of 20 and 35 mg/L compared to those that were not sprayed.

It has attracted the attention of researchers in the past few years, the active role for plant growth hormone salicylic acid in improving plant tolerance for environmental stresses and different biological stresses through its effect on the internal content of the plant from plant hormones (Naji, 2013). The treatment of plants with salicylic acid is one of the projects and solutions to reduce the impact of the drought phenomenon of drought and semi-drought areas in the world to increase plant resistance to dry stress conditions (Aldesuquy et al., 2012).

One of the positive effects of acid salicylic is to increase the sum of seeds in natural conditions and the conditions of stress and increase the proportion of oxygen and abscisic acid reduction (Hayat et al., 2007).

SA has a role in the association with amino acids such as Arginine, which is one of the sources of proline in the plant and increases the absorption of mineral elements such as phosphorus (Haddad et al., 2008).

Also, the salicylic acid role in the effectiveness of metabolism nitrogen representation and increases the effectiveness of an enzyme nitrate reductase, metabolism nitrogen is also an influential factor in the accumulation of proline acid and contributes to the increase in the percentage of gibberellins, which have a role in the flowering process, through its association with anthesin and the production of the hormone flowering florigen, which has a role in urging flowering (Hassanein et al., 2010).

Scientific research has indicated that spraying plants with low concentrations of salicylic acid it can stimulate endure of vital and abiotic stresses such as cold and drought tolerance high temperature and resistance to fungal, bacterial and viral diseases (Al-Hamdani et al., 2017).

Salicylic acid can also have a positive effect on growth and production, partly due to its effect in controlling the production of some plant hormones (Shakirova et al., 2003).

In addition, found that salicylic acid an important role in regulating physiological processes in plants such as opening process the process of opening and closing of stomata and absorption and transfer of ions and inhibition of ethylene manufacturing and bearing stress and maintain the permeability of cellular membranes and improve process photosynthesis and growth process (Gharib, 2006; Hayat et al., 2010).

Temperature is a major factor in abiotic stress and to determine agricultural productivity and crop productivity. The rate is reduced and the amount of absorption of

water and nutrients from cold stress, leading to cell drying and starvation and called extreme forms of cold stress stresses frozen and cause the formation of ice in the cell fluid, which leads to dehydration and death in plants. Low- temperatures to promote the accumulation of endogenous free SA in grape berry and wheat (Scott et al., 2004; Wan et al., 2009; Kosova et al., 2012).

The concentration of 0.5 mM SA would prefer to use cold tolerance for both the cucumber, corn, and rice preferred (Kang and Saltveit, 2002).

In winter wheat leaves grown at low temperatures, when sprayed with salicylic acid, the influence of external factors decreased and also the decreased freezing injury (Taşgin et al., 2003).

Chilling damage in freshly harvested green bell pepper (*Capsicum annuum* L.) was reduced by methyl SA and methyl (Fung et al., 2004) .Increase in response to low-temperature pressure in rice to express alternative oxidase (AOX) (Ito et al., 1997).

The use of a 0.5 mM concentration of SA by spraying leaves or irrigation roots of banana seedlings for one day led to improved cold tolerance (Kang et al., 2003).

Improves the cold resistance of the plants of tomatoes and beans, when soaked their seeds in the solution of aspirin or SA solution at a concentration of 0.1–0.5 mM before agriculture (Senaratna et al., 2000).

The treatment of salicylic acid is effective in relieving damage to the cooling, which is one of the most serious post-harvest losses in peach fruit. Curiously, the combination of ultrasonic and ultrasound treatment greatly inhibited the chilling injury of peach fruits compared to SA treatment alone (Yang et al., 2012).

The utilization of low centralizations of SA to tomato fruits products reduced the chilling damage and the occurrence of rot amid low-temperature stockpiling (Ding et al., 2002).

It was spring and winter wheat that was consistently used in aqueous solution for salicylic acid identical permanently due to low temperature (Horvath et al., 2007).

Leading to plant protection against biological pathogens, with the participation of salicylic acid in the regulation of protein expression associated with diseases (D'Maris et al., 2011).

SA plays an important role in the regulation of plant growth, and maturity, Walnmo, and respond to non-critical pressure (Rivas-San and Plasencia, 2011; Hara et al., 2012).

But high concentrations of salicylic acid may cause cell death or exposure to abiotic stresses, in general, may lead low concentrations of salicylic acid may enhance antioxidant capacity in plants (Hara et al., 2012).

Salicylic acid has important physiological roles such as increasing the plant's ability to withstand the stresses resulting from high temperature and low temperatures (Senaratna et al., 2000).

It also has important effects on the physiological activities related to the growth and development of plants under normal conditions without stress the need to control the absorption and transfer of ions and the permeability of cellular membranes, accelerate the formation of chlorophyll and carotene dyes, accelerate the carbon process and increase the activity of some important enzymes (Fariduddin et al., 2003; Hayat et al., 2007).

The external addition of SA has effectively stimulated the formation of phenolic compounds and the building of multi-phenolic materials (polyphenolic) by increasing enzyme activity Phenylalanine Ammonia Lyase (PAL) which leads to increased accumulation of dissolved phenolics (Kovacik et al., 2009).

SA is a natural compound used to combat post-harvest diseases as in the fruits *Citrus clementina* (Zheng and Zhang, 2004), apple (Yu and Zheng, 2006), sweet cherry (Yao and Tian, 2005), strawberry (Zhang et al., 2004), as it has a direct impact on the pathogens as well as Induction systemic resistance (Uquillas et al., 2004).

Salicylic acid (salicylic acid or willow acid) it is a carboxylic acid aromatic, colorless, produces naturally from some plants such as white willow and coriander meadows, and can workmanship as well as in the laboratory, it is the main compound of many known drugs, especially aspirin (D'Maris et al., 2011).

The treatment of roots of rice plants prior to SA and cadmium (Cd) exhibit the level of enzymatic and non-enzymatic antioxidants increases in the stem and roots this

reduces oxidative damage in terms of low level, H<sub>2</sub>O<sub>2</sub>, and increased transpiration (Guo et al., 2009).

It found that treatment with Cd raise the acid level of internal salicylic, which may work directly antioxidant to Scavenging ROS and / or it may be amended indirectly the oxidative-oxidative balance through the activation of antioxidant responses (Popova et al., 2009).

Gharib, (2006) found that salicylic acid spraying plants sweet basil and marjoram (*Majorana hortensis*), planted in the pot 40 cm diameter, it led to an increase of moral in the content of the papers of total amino acids.

Ali and Jaafar (2013) found that the sprayed plants salicylic acid on ginger plants (*Zingiber officinale* Roscoe), it has led to a significant increase in the content of the papers of total chlorophyll was 390.39  $\mu$ g/L compared with plants which did not sprinkle and which gave 269.23  $\mu$ g/L.

The use of SA in the agricultural amid of the Jujube plant at the concentrations 5, 25 and 50 mg/L, it has had a positive impact, where the highest rate for the survival of plants emerged surviving at the concentration of 10 mg/L SA, as well as a good response in the composition of branches and rooting at the same concentration (Ghaleb et al., 2010).

The beginning was a reference to a relationship between SA resistance in 1983 induced systemic, this was not confirmed until 1990, when it turns out that the salicylic acid the plant is produced locally at the site of infection as well as in phloem fabric in the distant leaves from the injury site which prompted the belief that this acid, it gives the start signal in the Systemic Acquired Resistance (SAR) (Metrau, 2001).

Conchic et al. (2007) pointed that the treatment of melon seeds (*Cucumis melo*) in the salicylic acid led to the induction development systemic resistance by increasing enzymes (peroxidases and chitinases), the post-harvest diseases less analogy treatment comparison.

The possibility of using SA and ASA, in the form of treatment seeds of lupine as an effective way to combat root rot disease lupine, the results showed an increase in the enzyme alcaitniz (El-Mougy, 2004). In addition, it has a role in transmittance and the transfer of ions and participates in stimulating certain changes in leaves anatomy and the installation of chloroplasts and participate in signals events (endogenous) and enters into the defense against internal etiology (Hayat et al., 2007).

The experiment was designed to study the effect of different concentrations of salicylic acid on resistance to low temperature. The aim of this study to find out the answer to these questions;

- 1. To determine whether the negative effects of low temperatures on pepper plants are eliminated with salicylic acid.
- 2. Determination of the possibilities of growing hot peppers in low temperature vegetables.
- 3. Determination of the most suitable salicylic acid doses.
- 4. To observe the effects of salicylic acid applications on different low temperature periods.
- 5. Application of salicylic acid in pepper seedlings and studying the effects of pepper seedlings on gene expression constitute the specificity of this study.

### 2. LITERATURE RESEARCH

It could be recommended that foliar spraying with SA at 100 ppm and chelated zinc at 50 ppm can be used to increase the final yield and fruit quality of sweet pepper plant during the low temperatures of autumn plantations. In the experiment about effect of foliar application of salicylic acid and chelated zinc on growth and productivity of sweet pepper (*Capsicum annuum* L.) under autumn planting. The field experiment was conducted to study the effect of foliar application with 50 and 100 ppm of SA and 50 and 100 ppm chelated zinc (Zn) and their combination on some growth aspects, photosynthetic pigments, minerals, endogenous phytohormones, fruiting and fruit quality of sweet pepper. Results indicated that different applied treatments significantly increased all studied growth parameters, namely, number of branches and leaves per plant, leaf area per plant and leaf dry weight. Besides, the two concentrations of each applied salicylic acid or chelated zinc obviously increased photosynthetic pigments, N, P, K, Zn, total sugars, total free amino acids and crude protein concentrations in leaves of treated plants as compared with those of untreated ones (El-Yazied, 2011).

It was established that SA had a bidirectional physiological effect on the seedlings in a concentration-dependent manner. And the effect of different concentrations of salicylic acid 0. 0.3. 1.5. 5 and 10 ml on the growth and some other parameters of pepper (*Capsicum annuum* L. cv.) seedlings was investigated. The results were, in concentrations 0.3 and 1.5 mM increased the length and proportion of chlorophyll and protein, while the concentration of 10 mM SA increases the length of the paper. But, concentrations of 5 and 10 mM have inhibitory effects on the seedlings to varying degrees (Canakci, 2011).

In spite of the fact that SA may likewise make oxidative anxiety plants, somewhat through the aggregation of hydrogen peroxide, the outcomes distributed so far demonstrate that the preparatory treatment of plants with low groupings of SA may have an acclimation-like impact, causing upgraded resilience toward most sorts of abiotic worries due fundamentally to improved ant oxidative capacity. In the experiment of "Induction of Abiotic Stress Tolerance by Salicylic Acid Signaling" is about the impact of exogenous SA relies upon various factors, for example, the species and formative phase of the plant, the method of use, and the centralization of SA and its endogenous level in the given plant. Late outcomes demonstrate that not exclusively does exogenous SA application direct anxiety impacts, however abiotic push elements may likewise modify the endogenous SA levels in the plant cells. This survey looks at the parts of SA amid various abiotic stresses (Horváth et al., 2002).

Crop production and quality affect low temperature (environmental stress), it also affects the level of a number of proteins and metabolites and regulates the expression of many genes. In the experiment about metabolism of reactive oxygen species and reactive nitrogen species in pepper (*Capsicum annuum* L.) Plants under low-temperature stress. In a study used leaves pepper (*Capsicum annuum* L.) plant which is exposed to low temperatures 8 °C different time periods from 1 to 3 mint. After 24 hours of exposure at 8 °C showed clear symptoms on the pepper plants characterized by flaccidity of leaves and stem. And also, significant changes in photosynthesis of reactive nitrogen species (RNS) and reactive oxygen species (ROS) with an increase of both lipid peroxidation and protein tyrosine nitration (NO<sub>2</sub>-Tyr). During the second and third days at low temperature, pepper plants underwent cold acclimation by adjusting their antioxidant metabolism and reverting the observed nitrosative and oxidative stress (Airaki et al., 2012).

Recent studies suggest SA, as a key molecule in the signal transduction pathway of biotic stress responses, has already been well described, and also participates in the signaling of abiotic stresses. The protection can be provided and the SA application is external against and some stressors such as low or high temperature and heavy metals. In the experiment about induction of abiotic stress tolerance by salicylic acid signaling. Salicylic acid and results showed that so far published the initial treatment of plants with low concentrations of may have an effect such as acclimatization, resulting in enhanced tolerance towards most types and abiotic stresses mainly caused by enhanced antioxidant capacity. Abiotic stress factors may change, also the SA levels are subjective in plant cells (Horvath et al., 2007).

In the study of the effect of the proportion of red pepper germination (*Capsicum annuum* cv. Sena) seeds before and after seed, storage was investigated. At low temperatures incorporating 5-Aminolifulcin acid (ALA) only in a priming solution. In the experiment about promotion by the 5-aminolevulinic acid of pepper seed

germination and seedling emergence under low-temperature stress. Preparing pepper seeds within the sight of ALA improved final germination rate (FGP) and germination rate (MGT) at 15 °C compared with non-prepared seeds. The most noteworthy FGP was obtained from seeds prepared within the sight of 25 ppm and higher ALA concentrations while the most astounding MGT was gotten from seeds prepared in KNO<sub>3</sub> supplemented with 10 ppm ALA. These results demonstrate that preparing seeds in 25 ppm and 50 ppm ALA incorporated into the KNO<sub>3</sub> arrangement can be utilized as a compelling strategy to enhance low-temperature execution of red pepper seeds and that these seeds can be stored for 1 month at 4 °C or 25 °C (Korkmaz, 2005).

Appeared here to be powerful inhibitors of systemin-instigated and jasmonic acid (JA) - incited combination of proteinase inhibitor mRNAs and proteins. In the experiment about salicylic acid inhibits synthesis of proteinase inhibitors in tomato leaves induced by systemin and jasmonic acid. Heartbeat leaves were tomato (*Lycopersicon escolentum*) labeled with [35S] methionine, trailed by sodium dodecyl sulfate-polyacrylamide gel electrolyte, and inhibitory impacts SA were appeared to be particular to blend a couple of the Ca-actuated proteins that incorporate protease inhibitors. Here we report that the inhibition of combination of proteins, mRNAs and proteinase inhibitor by SA in both light and darkness likewise happens at a stage in the flag transduction pathway after JA blend yet going before the translation of the inhibitor qualities (Doares et al., 1995).

Late studies have investigated the prerequisite of SA for mounting the hypersensitive response (HR) against an attacking pathogen, where a specific cell death process is initiated at the site of endeavored contamination causing a limited injury. In the experiment about salicylic acid in the machinery of hypersensitive cell death and disease resistance. Results indicate that biochemical information proposes SA potentiates the flagged pathway for HR by influencing an early phosphorylation-delicate advance going before the pro-death signals, including those got from the oxidative burst. Accordingly, its primary relationship is between cell death and accumulation, SA activity is placed in the downstream feedback loop and the cell death source. In addition, the spatiotemporal patterns of the SA collection (non-homogeneous dissemination, biphasic kinetics) depicted in some HR lesions, it may also reveal important evidence to detect the complex cellular network, which was balanced with a

strict suspicion and pro functions, and anti-death in hypersensitivity to the cell death (Alvarez, 2000).

In studying the effect of SA on sunflower plants. In the experiment about effect of salicylic acid on pigment, protein content and peroxidase activity in excised sunflower cotyledons. The sunflower seedlings were grown in dark conditions for 9 days and then transferred to Petri dishes which contains a different percentage of SA 0.001, 0.1, 10, and 1000  $\mu$ M, and placed in the darkness of the room for 14 hours. Then incubated in light for 3 hours, carotenoid content, chlorophyll, and protein were examined. According to the results, the chlorophyll content is 1.5 times greater in 10  $\mu$ M, the protein amount increased 1.9, 2.3, and 1.7 fold in 0.001, 0.1 and 10  $\mu$ M (Cag et al., 2009).

The effect of salicylic acid treatment before harvest on the quality, quantity, and absorption of nutrients from the flower of roses. Using four different concentrations of SA 0, 50,100 and 150 ppm in experiment design were randomized and with 3 replicates. The temperature of the day was 28 °C, and at night it was 18 °C, the relative humidity in the greenhouse was between 60- 70. During the growth period, the plant was sprayed once every two weeks and was sprayed manually. Results salicylic acid had a significant effect on the ratio of chlorophyll and anthocyanin of the petal, nitrogen, potassium, and phosphorus, Plant production was 50 and 100 ppm, Compared to 0 and 150 ppm of salicylic acid. It is therefore important to use salicylic acid in the rose plant production Pre-harvest to improve the quality of rose (Fariduddin et al, 2003).

Ferrarese et al. (1996) carried out a research on a farm near Padova (Italy) for 3 days, in order to hold the ethylene eventually produced by the wounded leaves, at the instigation of papers and remove the leaves and keep the branches in air control containers. In the experiment about Cellulose involvement in the abscission of peach and pepper leaves is affected by salicylic acid. The data show that salicylic acid is able to reduce leaf breakage in each plant (peaches and peppers). Biochemical analysis has revealed that the enzyme, which usually does not increase after activating the breakage of leaves in plants treated with salicylic acid. And a significant increase in control stations in the levels of protein, cellulose and enzyme activity. The presence of salicylic

acid in the use of plants with external ethylene increases the expression of cellulose induced level in plants without salicylic acid.

The effect of salicylic acid on some biochemical and physiological properties of maize (*Zea mays* L.) seedlings under NaCl stress was studied. Stress and treatments were given in the presence and absence of 0.5 mL of salicylic acid and Presoaking treatments of NaCl 0, 50, 100 and 200 mM. In the experiment about salicylic acid-induced salinity tolerance in corn grown under NaCl stress. The results showed that the two-week maize seedlings exhibited a significant decrease in root length, dry weight, and shoot length and leaf area on 6 hours per 100 and 200 Mm NaCl stress. And increasing stress levels and Photosynthetic pigments decreased sharply. And seedlings that are pre-treated with salicylic acid 0.5 mM along with enhanced salinity levels in photosynthesis dyes and growth parameters photosynthesis dyes. The study concluded that 0.5 mL salicylic acid improves the ability of the corn plant to adapt to NaCl (Gautam and Singh, 2009).

In a study of the effect of SA and gentisic acid (GTA) on the growth rates of soybeans, maize, and photosynthesis under greenhouse conditions. Conductance and transpiration were also increased. The chlorophyll content of these compounds does not change. And treatment in some cases with these compounds led to an increase in the dry mass of the plant and leaf areas, however, root length and plant height were not affected (Khan et al., 2003).

In this study, an appropriate concentration of SA was determined on the rooting of the poinsettia (*Euphorbia pulcherrima*). Present study about demonstrated that there was an incredible variety in the majority of the measured characters at (P < 0.05) percent level. The results showed that salicylic acid was obtained, which increased the rooting rate. And has a positive effect on the use of salicylic acid. In this study, plant growth regulators salicylic acid have a profound effect on rooting of poinsettia (Kling and Meyer, 1983).

The metabolic and sedative changes in the pepper of plants grown at optimum temperature were studied. Plantation of pepper plants in the temperature of 29/20 and 25/14 (°C, day/night) were studied. The variables were previously related to cold acclimatization in temperate plants. At low temperatures showed cultivated plants

decreased by 50–70 % in the number of leaves and the length and dry weight compared to the high-temperature system. It was also shown in the cold system of plants grown an increased number of shoots in the armpits. And the content of proteins and chlorophyll decreased in both temperature treatments. The total nitrogen content was slightly higher at low temperature, but nitrate was lower. The plants grown in the low-night temperature have improved cooling when subjected to 4 nights at 6 °C. Nitrogen and carbon metabolism where differences of peppers plants for cold acclimation (Mercado et al., 1997).

In experiment of Aldesuquy et al. (2014) glycine betaine and salicylic acidinduced modification in water relations and productivity of drought wheat plants. The process of preparation of previous grains was depleted in SA or foliar application with glycine betaine mitigated the worry by keeping water inside leaves and subsequently recoup the turgidity of focused on plants by restricting the transpiration rate, stomatal closure, diminishing the abscisic acid (ABA) level and upgrading the growth promoters especially (indole-3- acetic acid (IAA), gibberellic acid (GA<sub>3</sub>), and cytokinins (CKs)) especially with the delicate cultivar. Besides, the impact was more articulated with glycine betaine + SA treatment. Furthermore, the effect was more pronounced with the treatment of glycine betaine+ acid salicylic. Cereal productivity appears to be positively correlated with IAA, GA<sub>3</sub>, and but negatively correlated with ABA, transpiration rate, and stomatal areas on wheat varieties.

The pepper plants (*Capsicum annuum* L.) are exposed to low temperatures 8 °C from 1 to 3 days to different time intervals. The main components of the metabolism of oxygen were analyzed as reactive nitrogen species (RNS) and reactive oxygen species (ROS) and nitrogen. Pepper plants showed clear symptoms 24 hours after exposure to low temperature, characterized by flaccidity of leaves and stems. There were also clear changes in the metabolism (RNS and ROS) with an increase in fat peroxide and protein tyrosine nitration (NO<sub>2</sub>-Tyr), thus pushing the plant to induces nitrosative and oxidative stress. The pepper plant underwent cold acclimatization during the second and third days at low temperatures by controlling the antioxidant metabolism and the nitrosative return and oxidative stress (Mitchell and Moyle, 1967).

The experiment was conducted on proteomic analysis of salicylic acid enhanced disease resistance in bacterial wilt affected chili (Capsicum annuum L.) crop. The experiment was carried out on seedlings of 3 weeks old. The seeds have been used to wilt susceptible from the fruit and vegetables. These seeds were planted in the potting pots (mixture of clay, sand and farmyard manure in 1:1:1 ratio) and was incubated at 28  $\pm$  2°C for 24- 48 hours, the bacterium was harvested in sterile distilled water by centrifugation, using visible UV spectra the bacterial suspension was adjusted to OD 600 = 0.01 to obtain the concentration of 1×108 cfu ml -1. Concentrations of 0.1, 0.25, 0.5, 0.75, 1 and 1.25 mM of SA aqueous solution were used and the seedlings (3-week old seedlings) were completely sprayed. Pepper plants were kept in the humidity chamber at 25  $\pm$ 1°C, and disease symptoms were recorded in 7 days of inoculation. Proteomic analysis revealed 25 differential expression proteins (which was more prominent in SA preparation challenges defying hot pepper samples), the differential articulation design uncovered that proteins related to pressure and protection, vitality and digestion, metabolism and energy, protein destination, protein synthesis, and storage and transcription-related were unregulated demonstrating the association of SA incited disease resistance in chilli seedlings. Discoveries from this examination will serve as the basis for designing disease-management strategies based on resistance conferred by SA, which could relevant to other biotic pressure influenced staple crops (Chandrasekhar et al., 2017).

In the experiment around *Capsicum annuum* home box 1 (*CaHB*<sub>1</sub>) is a nuclear factor that has roles in plant development, salt tolerance, and pathogen defense. The cultivation of tomato and pepper plants in pots and maintained under a 16-h photoperiod at 25 °C. The microscopic analysis was observed for one month or *CaHB*<sub>1</sub>-transgenic plants under a small light range and electron microscopy for communication, and pepper plants leaves were sprayed with chemicals leaves of 5 mM SA, our isolation will be carried out using a genome-wide assay, an expression *CaHB*<sub>1</sub> text levels found in flowers and stem while its low levels are both roots and leaves, GFP fluorescence was watched all through the cytoplasm and the core. Like other HD-Zip proteins, *CaHB*<sub>1</sub> were checked in pepper plants treated with biotic and abiotic push to comprehend the functional of  $CaHB_1$  in the stretch.  $CaHB_1$  articulation was firmly initiated when pepper leaves were inoculated with the pepper pathogen,  $CaHB_1$  transcripts expanded strikingly after inoculation, before infection indications were noticeable, and stayed hoisted until 72 h after inoculation. Minuscule examination uncovered that leaves from  $CaHB_1$ transgenic plants had thicker cell dividers and fingernail skin layers than those from controls. In addition,  $CaHB_1$ -transgenic plants showed upgraded resistance against Phytophthora infestans and expanded resistance to salt pressure. Thusly, our findings propose parts for  $CaHB_1$  being developed, salt pressure, and pathogen resistance (Oh et al., 2013).

Developed as guideline reproducing material in Turkey in carotenoid, capsaicinoid and ascorbic corrosive creation of ready products of five *Capsicum annuum* cultivars (730 F1, 1245 F1, Amazon F1, Serademre 8 and Kusak 295F1) were used. Seven primary carotenoids, five analogs of capsaicinoids and ascorbic corrosive were evaluated in the natural products developed for 2-year replication. In the experiment about assessment of carotenoids, capsaicinoids and ascorbic acid composition of some selected pepper cultivars (*Capsicum annuum* L.) grown in Turkey. The discoveries verified that the cultivars of 730 F1 and 1245 F1 had higher carotenoids (2310–2390 mg/kg in dry premise), capsaicinoids (471.3–688.1 mg/kg in dry premise), vitamin A (218.8–243.0  $\mu$ g RAE/100 g in wet premise) and vitamin C (63.1–64.9 mg/100 g in wet premise) content, with no noteworthy distinction among each of them. The cultivars which had higher capsaicinoids substance had higher ascorbic acids content too. With their high wholesome and useful segments, the cultivar of 730 F1 and 1245 F1 can be thought to be chosen reproducing the material for cultivar improvement (Topuz and Ozdemir, 2007).

In spite of the fact that SA may likewise make oxidative anxiety plants, somewhat through the aggregation of hydrogen peroxide, the outcomes distributed so far demonstrate that the preparatory treatment of plants with low groupings of SA may have an acclimation-like impact, causing upgraded resilience toward most sorts of abiotic worries due fundamentally to improved ant oxidative capacity. In the experiment about induction of abiotic stress tolerance by salicylic acid signaling. The impact of exogenous SA relies upon various factors, for example, the species and formative phase of the plant, the method of use, and the centralization of SA and its endogenous level in

the given plant. Late outcomes demonstrate that not exclusively does exogenous SA application direct anxiety impacts, however abiotic push elements may likewise modify the endogenous SA levels in the plant cells. This survey looks at the parts of SA amid various abiotic stresses (Alvarez, 2000).

Its capacity in physiological procedures identified with cell demise is still ineffectively comprehended, albeit broad information has depicted the key part of salicylic corrosive SA in flagging pathogen-induced sickness protection. Late investigations have investigated the prerequisite of SA for mounting the easily hypersensitive response (HR) against an attacking pathogen, where a specific cell demise process is enacted at the site of attempted disease causing a bound lesion. In the experiment about. Salicylic acid in the machinery of hypersensitive cell death and disease resistance. Biochemical information recommends that SA potentiates the flagged pathway for HR by influencing an early phosphorylation-delicate advance going before the age of genius demise signals, including those got from the oxidative burst. As needs are, the epistatic connection between cell passing and SA collection, broke down in crosses between sore copy mutants (unconstrained injury development) and the transgenic line (drained in SA) puts the SA action in a criticism circle downstream and upstream of cell demise (Mejía-Teniente et al., 2013).

In this experiment evaluated the endogenous  $H_2O_2$  production caused by SA,  $H_2O_2$  elicitors in (*Capsicum annuum* L.) and chitosan (QN). By searching around it oxidative and molecular responses in (*Capsicum annuum* L.). After hydrogen peroxide, salicylic acid, and chitosan foliar applications. In this search, we evaluated the endogenous  $H_2O_2$  production caused by SA,  $H_2O_2$  elicitors in (*Capsicum annuum* L). And has been determine the production of hydrogen peroxide after elicitation, catalase (CAT) and phenylalanine ammonia lyase (PAL) activities, their outcomes showed that 6.7- and 10-mM SA concentrations, and, 14 and 18 mM  $H_2O_2$  concentrations, initiated an endogenous  $H_2O_2$  and gene expression. Endogenous  $H_2O_2$  production monitored over several days, Results can be an indicator to determine the uses of applied opportunity in agriculture in order to maintain plant alarm systems from stress (Laura et al., 2013).

SA plays an essential part in the control of plant development, improvement, aging, and guard reaction. The part of SA in the plant– pathogen relationship has been

broadly researched. Notwithstanding resistance reactions, SA assumes a critical part in the reaction to abiotic stresses, including dry spell, low temperature, and saltiness stresses. In the experiment around "Regulation of water, salinity, and cold stress responses by salicylic acid". In this research, it has been recommended that SA has the immense agronomic potential to enhance the pressure resistance of horticultural imperative yields, the utility of SA is reliant on the grouping of the connected SA, the method of use, and the condition of the plants (e.g., formative stage and acclimation). For the most part, low concentrations of connected SA mitigate the affectability to abiotic stresses, and high concentrations of connected actuate elevated amounts of oxidative pressure, prompting a diminished resilience to abiotic stresses (Miura and Tada, 2014).

The most difficult dangers to the horticultural framework and economic yield of production plants, and form non-vital pressures such as (salinity, metals/metalloids, ozone, UV-B radiation, drought, and extreme temperatures). These worries (in isolation and/or combination) initiate various antagonistic impacts in plants, weaken biochemical/physiological and atomic procedures, and inevitably cause extreme decreases in plant development, improvement, and general profitability. In the experiment around salicylic acid-induced abiotic stress tolerance and underlying mechanisms in plants. Phytohormones have been perceived as a strong device for economically lightening unfavorable impacts of abiotic worries in crop plants. In this research, the importance of salicylic acid SA has been increasingly defined in improving plant abiotic stress-tolerance via stress by means of SA-mediated control of the main metabolic plant processes. Be that as it may, the essential biochemical/physiological and atomic instruments that conceivably support SA-initiated plant-resilience to major abiotic stresses remain least discussed. Based on the results reached by their research: (a) reviews recorded foundation and biosynthesis of SA under both ideal and upsetting conditions in plants; (b) fundamentally evaluates the part of SA in plants presented to major abiotic stresses; (c) cross-talks potential systems conceivably administering SA-instigated plant abiotic push resistance; lastly (d) quickly features real angles so far unexplored in the present setting (Khan et al., 2015).

In an experiment by Al Shukri and Abbas (2016) around morphological and physiological effects of fertilizer NPKZn and salicylic acid on the growth of coriander plant (*Coriandrum sativum* L.) and used three levels of concentrations of SA 0, 15 and 30 mg/L, and spraying salicylic acid especially at 30 mg/L. The interaction between the factors gave the best values of studied parameters under 15, 30, mg/L salicylic acid.

In the experiment of Turk and Abu–Hinna (2016), the effects of adding salicylic acid in the potato plant (*Solanum tuberosum* L.) growth indicators were determined. And spraying a plant with three concentrations of SA 0, 75.150 mg/L. The results were significantly higher in concentration of 75 mg /L to giving the highest plant length and the largest number of leaves and the largest leaf area and the highest rate of tuber weight and the highest yield of one plant and the total yield of the unit of area and the content of the tuber from the original coupler treatment comparison that gave lower values as compared to the comparative treatment which gave lower values.

In the experiment of Al-Dabbagh and Al-Duleimi (2017) on pepper feeding in boron and SA in the production qualities and quality to installation genetically Mung bean (*Vigna radiata* L) were studied. And it used three concentrations of SA 0, 150, 300 mg/L, the results have been given that the concentration of 150 mg/L in both seasons gave the highest fertility rate in the pods as 77.53 and 69.76 % and the highest average yield of the seeds is up to 760 and 698.4 Kg/ ha and the concentration of SA 300 mg/L exceeded in the autumn season in the number of seeds by pod. The genotypes differed significantly in the studied traits except for the percentage of protein in the seeds and both seasons.

In a study of El Sayed et al. (2017) Increasing of resistance to Salt Stress of *Duranta plumieri* irrigated with seawater by using salicylic acid were investigated. The experiment was performed during the two consecutive seasons of (2014 and 2015) to estimate the effects of soil application foliar spraying plants with salicylic acid at 250 and 500ppm combined with irrigation. Results showed that the vegetative plant growth transactions were affected largely by salinity stress, plant height, decreased the number of branches/plants and shoot fresh and dry weight linearly with an increase in saline concentration. SA has been induced as a catalyst effect on all vegetative growth parameters in plants that are irrigated with all dilute seawater concentrations. SA has improved physiological processes in *Duranta* plants under salinity conditions by

significantly increasing carbohydrate and carotenoids contents total and reducing and non-reducing sugars.

In the experiment of Al-Dabbagh and Al-Duleimi (2017) the effect of foliar application salicylic acid in the Production and quality characteristics of genotypes of mung bean (*Vigna radiata* L.) was investigated. A field experiment was carried out in the spring and autumn seasons of the year 2016, in soil with a clay-silty texture. To determine the effect of foliar application with three concentrations of SA; 0,150 and 300 mg/ L in both seasons gave the highest fertility rate in the pods in the concentration of SA 150 mg/ L, and the highest average yield of the seeds and the protein content. And the concentration of SA 300 mg/L in the autumn season compositions genetic morally in studied qualities, the local genotype showed the highest mean number of seeds by pod and seed by yield 789.20 and 791.5 kg and protein with 210.20 and 187.30 kg. While the Indian genotype in both seasons showed the highest average length of the pod and 100 seed weight 6.74 and 7.02 g, for the two seasons respectively.

Ibrahim and Jadoua (2014) carried out experiment about effect of salicylic acid in some vegetative and fruit characteristics of (*Nigella sativa* L.) to find out the possibility of improving the growth and qualities seed yield off (*Nigella sativa* L.) plant, by using spraying it the plant growth regulator SA with concentrations 50, 100 or 200 mg/L.The results showed that the highest plant height was achieved in the SA 100 and 200 mg/L treatments with an increase of 9 % compared to the comparison treatment. Dry leaf weight increased by 43, 24 and 22 % significantly and total dry weight by (27, 13 and 8 %) at all salicylic acid concentrations respectively, and also increased chlorophyll content of leaves at (24,7 and 6 %), produce seeds increased by 29, 21 and 21 % respectively. Therefore, the concentration of 50 mg/L treatment is the best in increasing the indicators of growth and yield. Therefore, it can be concluded that SA at a concentration of less than 50 mg/L can produce positive results.

In an experiment of Al-Zyadi and Mohammad (2018), "Effect of planting date and salicylic acid spray on the growth of burdock plant (*Arctium lappa* L.) And its roots content of quercetin compound" were researched. The experiment was conducted in the 2015-2016 season, to study the three agriculture dates 15/9, 15/10 and 15/11 and the effect of spraying in three concentrations salicylic acid 50,100 and 200 mg/L addition to plant growth and the content of the rooted from composite quercetin. Results showed the superiority of cultivated plants in the first date 15/9 in plant height, the leaf area, the dry weight of vegetative total and the dry weight of root total, the results showed that the superiority of plants in the third date of 15/11 in giving their roots production higher than quercetin 6.428 mg plant dry weight. As he gave the superiority of plants salicylic acid treatment with concentration 100 mg/L in the leaf area, the plant height, dry weight of vegetative growth and dry weight of roots, as well as the content of roots of quercetin (6.366 mg.plant dry weight).

Anjum et al. (2016)'s study was conducted to assess the efficacy of (*Saccharomyces cerevisiae*) salicylic acid in the renewal of resistance in orange fruits against (*Penicillium digitatum*) researchers, used different concentrations of salicylic acid 1, 10, 100, 500 and 1000 ppm and cool the middle of the agricultural to 40-45  $^{\circ}$ C, and add salicylic acid to get the desired concentration. Achieved salicylic acid positive impact in the growth of fungus (*S. cerevisiae*) all concentrations used 1, 10, 100, 500 and 1000 ppm preparation of yeast, with an activation rate of 25.28 % compared to the comparison treatment. Also, the results showed for the development of resistance in orange fruits, it has outperformed the combination salicylic acid and yeast (*S. cerevisiae*) in the development of systemic resistance by increasing the effectiveness of the enzyme (peroxidase) in the fruits of analogy treatment of salicylic acid alone.

In a study by Hameed et al. (2015) to study the effect of water stress and the external application of glycine and salicylic acid on the growth and production of eggplant (*Solanum melongena* L.). It included global water stress and irrigation in three levels of 72, and 46.8, 21.6 liters/day, and spraying with an acidic mixture of glycine and salicylic acid at two levels (first spraying with water only and the second spraying with a mixture of 100 mg/L glycine and 100 mg/L salicylic acid), planted seedlings in a silt clay loam soil. Drip irrigation system was used. Eggplant seedlings were planted in two batches the first month after agriculture and the second two weeks after adding the first installment. The results showed a significant superiority for the treatment of irrigation on the first level W1 (water stress1) transactions W2 (water stress2) and W3 (water stress3) and all study indicators, in turn, outperformed the treatment of

interference W1A2 (water stress1 x Applcation2) on the rest of overlapping transactions.

In a study by Aziz et al. (2015) about the response of *Narcissus* plant to spraying with plant growth regulators salicylic acid the characteristics of the vegetative growth and syphilis and bulbs characteristics of plant daffodils (*Narcissus poeticus*), plants were sprayed with four concentrations of growth regulators, Salicylic acid concentrations were 0, 20, 40 and 80 mg/L. Sprayed the plants three times: The first two months after agriculture, and the second 20 days after the first spatter, the third was carried out 20 days after the second spatter. The results increased in plant height number of branches, plant at the 4.90 SA concentration and the ratio of chlorophyll 19.88 % and wet weight 25.78 gm and flower diameter 4.71 cm 2.95 weight wet and dry. The effect of overlap between the two studied factors is significant in most studied traits.

In the study by Ramadan et al. (2018) that about SA and acetylsalicylic induced resistance to *powdery mildew* disease of milk thistle (*Syllibium marianum*), plants from the fields of the Faculty of Agriculture and Forestry - the University of Mosul in February 18/2/2013. The plants were sprayed with materials and one was on 7/3/2013 and the second was 25/3/2013. Concentrations of salicylic acid and acetylsalicylic acid 1 and 5 10 mM were used. The results suggest that the use of different concentrations of SA and ASA, it led to the increase in fresh weight and dry Aloren and plant height, the reason for the concentration of 1 mM of ASA spray one to increase in fresh weigh 10.9 g. And dry weight differed significantly from the rest of the transactions except for 1 mM concentration of SA spray one.

In the experiment of Al-Jubouri and Hassan (2017) about the effect of salicylic acid on the growth and productivity indicators of ten genotypes of wheat (*Triticum aestivum* L.) under dry farming condition was studied. The experiment was carried out in the fields of a research station (Bany Maqan) in the district of Chamchamal of the Sulaymaniyah Research Directorate, in the winter farming season 2016-2017, and used in this study ten genotypes of wheat Rizgari, Adana 99, Arras, Milan, Ala, Eba 3 and Abu Ghraib. Three levels of salicylic acid for sowing seeds and spraying in the Branches:

• Without stimulation of seeds and not to spray acid.

- Soak the seeds with salicylic acid 50 ppm, and sprinkle on the total 100 ppm, at the elongation phase of the first node.
- And soak the seeds salicylic acid concentration of 200 ppm in the first node elongation stage.

The results showed superior treatment of activation SA1. Morally superior treatment of other activation in the proportion of (75.5 %), dry weight in both phases of elongation and the expulsion of spikes, yield, and chlorophyll content of the flag leaf, plant height and a number of kernels spike 45 pills, the biological yield is 15.73 tons and the grain yield is 5749.0 kg/ha. also showed compositions genetics differences morally in all the studied traits. And showed overlap between the genetic structures activation coefficients. The composition genetic of wheat sham 6, excellence morale, number of the spike, weight of 1000 grains, grain yield and harvest index in the activation treatment SA1. And Abu Ghraib was in superior plant height, number of spike grains, grain yield, and harvest index.

In the study by Yasin (2016) about the effect of salicylic acid vegetative growth and the oxidizing enzymes of the sunflower (*Helianthus annuus* L.) was investigated. In this experiment, the seeds are soaked with sunflower seeds with two concentrations of salicylic acid 30 and 60 mg/ L in addition to the comparative treatment with a view to knowing effects in growth visas, in addition to the comparative treatment with a view to knowing effects in growth and the total vegetative content of DNA and RNA and the content of the leaves from (peroxidase and catalyze).

Al-Kraidi et al. (2017) found the effect of the three planting dates of the corundum plant (*Coriandrum sativum* L.) the spraying with SA 0, 35 and 70 mg/L. The results most important results obtained were: plants sown on 20/10 gave a significant increase in plant height, fresh and dry weight of vegetative growth. Whereas plants sown on 10/10 gave a significant increase in weight of 100 seeds, seed yield, and volatile oil yield. Plants sprayed with salicylic acid gave a significant increase in dry weight of vegetative growth, and seeds number, the weight of 100 seeds, seed yield.

In experiment of Jassim and Al-Kooranee (2012) effect of SA on *macrophomina* and evolution of fever disease on Sun Flower plant (*Helianthus annus* L.) was found. Results experience recommended that all pots concentrations may be affected

significantly in reducing the percentage of injury. As for the concentrations of 200 and 250 mg /L salicylic acids effect of the dry weight increase total of vegetables and also an increase in the dry weight total of the roots. The study indicated that high concentrations of salicylic acid have been reduced by the number of stone bodies (sclerotia). While low concentrations of salicylic acid have significant differences with control treatment.

The researcher by Al-Lishi el at. (2012) found SA to the liquid media Potato Sucrose Broth (PSB) caused complete inhibition to the fungal growth at 3 and 5 mM of SA applied in greenhouse experiments, seeds were soaked mM of SA for 24 hours. As a protective treatment, then planted in soil contaminated with the pathogen caused the significant reduction in percent infection and disease severity of leaf spots. Dual treatment by soaking the seeds with the same concentrations. Along with spraying the shoot system was the most efficient in reducing percent infection by 100 % and disease severity of the disease and improving growth characters of the treated plants. Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

## **3. MATERIAL AND METHOD**

## **3.1. MATERIAL**

## **3.1.1.** The geographical structure of the experiment area;

This study was carried out in the horticulture department, agriculture faculty of Siirt University in 2018-2018. Siirt is a province and located between the 41°- 57° East longitude and 37° -55° northern latitudes in as its geographical location.

### **3.1.2.** Climatic and geographical characteristics;

## **3.1.2.1. Siirt province:**

In Siirt province, it dominates the continental climate which is the most important features of the four seasons, continental climate prevails. The summers are hot and arid, with no precipitation in June and October.

After the Southeastern Anatolia project started, some items related to climate change were realized.

After this period, more rainfall was observed in the spring and the humidity was 40 % in normal has reached over this rate.

Siirt province in eastern and northern regions that have much difference between day and night temperatures, the winter month is rainy and frosty and the southern and western regions are warmish. The highest temperature is 43.3 °C, the lowest temperature is -19.5 °C. The wind from the east and the north blows (Anonymous, 2003).

The 35-year average of the province of Siirt climate data are shown in Table (3.1) (Anonymous, 2003).

	Average of Temp. (°C)	Average of High Temp. (°C)	Average of low Temp. (°C)	Highest Temp. (°C)	Lowest Temp (°C)	Average of rainfall (mm)	Average of Relative Humidity (%)
January	2.3	6	-1.1	16.2	-19.3	114	77
February	4	8.2	0.1	19.6	-16.5	108	69
March	7.5	12.2	3.1	24.1	-10.1	109	65
April	13.3	18.4	8.2	29.5	-4.1	108	59
May	19.3	24.9	13.1	36.2	2.4	66.5	52
June	25.9	31.9	18.5	39.4	8.2	9.3	36
July	30.5	36.7	23.1	42.7	13.1	1.4	30
August	30.1	36.6	22.8	41.2	14.5	0.5	29
September	25.1	31.9	18.3	38.4	9	3.7	34
October	17.8	23.9	12.2	34.4	0.3	47.6	47
November	10.6	15.3	6.3	26	-14.1	86.6	64
December	4.9	8.7	1.4	18.4	-2.3	94.2	72
Total	191.3	254.7	126	366.1	-18.9	748.9	634

Table 3.1 The average of climatic data during the 35 years, in Siirt province

## **3.1.3. Plant Materials;**

In this experiment, pepper (*Capsicum annuum*) was used as plant material. Urartu F1 pepper variety which could be grown in the greenhouse was used as pepper variety. Pepper is grown at large area and needs a high temperature for flowering and fertilization. Urartu F1 has capita form and is grown in greenhouse in the winter. Capita peppers are rich in ascorbic acid and carotene. Grill it or just eat it raw. The Capita pepper is a delicacy of different southern cousins the features of Urartu F1 is given (Table 3.2).

Table 3.2	The features	of Urartu F1
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Variety	Fruit Weight	Growing Season	Period of Maturity	Disease Resistance HR	
Urartu F1	100-120gr	Winter in greenhouse	Late	Tm0 (TMV, ToMV, PMMoV)	



Figure 3.1. Urartu F1.

# 3.1.4. Growing Seedling;

The seedling which was used at this experiment were grown at seeding tray (7 x 10). Turba (pet moss) was used as growing or substrate. A plant growing cabinet was used for growing seedlings (Figure 3.2) the seeds were sown which (month) ago from low-temperature applications. For seedling a shaded seedling, the tunnel was used.



Figure 3.2. Put the pepper seedlings on the cooling device (Refrigerator).

## 3.1.5. The Chemicals Materials;

Salicylic acid was used as the chemical material for the resistance of pepper seedlings to low temperature (Figure 3.3). Polysorbate 20 is a hydrophilic nonionic surfactant generally used as emulsifiers, dispersing agent and solubilizer (Zielinski et al., 2015).



Figure 3.3. The chemicals materials.

# **3.2. METHODS:**

After 0  $^{0}$ C temperature application; do all seedlings maintaining turgescence, continue to normal growth, did cold application cause a non-irreversible wilting to seedlings, do all seedlings have cold damage on their leaves and stems? Also, are there differences in seedlings weight and lengths? The form of the experiment is given below (Table 3.3, 3.4 and 3.5).

## **3.2.1.** Salicylic Acid Application and Applications' Frequencies;

Three different treatments 0.01 mmol, 0.05 mmol, and 0 mmol salicylic acid applicated (Figure 3.3). Salicylic acid was 1, 2 and 3 times applicated for each dose. For control groups only distilled water was applicated each time. Polysorbate (Twin 20) was also used very very small quantity for adhesive. With small needles, very thin holes were opened in the leaves, making salicylic acid penetration easier (Figure 3.4).



Figure 3.4. Opening holes on leaves.

# **3.2.2.** Temperature and Temperature Durations.

0 <sup>0</sup>C was the application temperature for the seedlings to find out the effect of salicylic acid at low temperature. All seedlings were exposed to the cold application for 24, 48 and 72 hours. The temperature was measured with a maximum/minimum thermometer (Figure 3.5).



Figure 3.5. Maximum/minimum thermometer.

l. 24 Hours									
	20 Seedlings				20 Seedlings			20 Seedling	gs
1		1.Dose			2.Dose			3.Dose	
1. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol
	20 Seedlings			20 Seedlings			20 Seedlings		
20 5.1	1.Dose			2.Dose			3.Dose		
20 Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol
	20 Seedlings			20 Seedlings			20 Seedlings		
	1.Dose			2.Dose			3.Dose		
20 Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol

Table 3.3. The experiment format the application of salicylic acid at (24 hou	Table 3.3. The	experiment	format th	e application	of salicy	lic acid at	(24 hours
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					11. 48 H	Iours				
			20 Seedling	s		20 Seed	lings		20 Seedling	s
			1.Dose	!		2.Do	se		3.Dose	
	1. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
		0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol
	2. Repl.	20 Seedlings			20 Seedlings			20 Seedlings		
		1.Dose		2.Dose			3.Dose			
		1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
		0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol
		20 Seedlings			20 Seedlings			20 Seedlings		
			1.Dose		2.Dose			3.Dose		
	3. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times
		0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol

Table 3.4. The experiment format the application of salicylic acid at (48 Hours).

Table 3.5. The experiment format the application of salicylic acid at (72 Hours)

III. 72 Hours										
		20 Seedlin	ngs		20 Seedlings			20 Seedlings		
		1.Dose			2.Do	se		3.Dose		
1. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times	
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol	
	20 Seedlings			20 Seedlings			20 Seedlings			
	1.Dose			2.Dose			3.Dose			
2. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times	
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol	
	20 Seedlings			20 Seedlings			20 Seedlings			
		1.Dose		2.Dose			3.Dose			
3. Repl.	1 time	2 times	3 times	1 time	2 times	3 times	1 time	2 times	3 times	
	0 mmol	0 mmol	0 mmol	0.01 mmol	0,01 mmol	0,01 mmol	0,05 mmol	0,05 mmol	0,05 mmol	

# 3.2.3. Physical properties of the Seedlings

# 3.2.3.1. Seedling Weight (gr)

Before and after cold application, 5 randomly seedlings per applications were selected and then measured by scale sensitive to 0.01 g (Figure 3.6).



Figure 3.6. Seedling weight (befor and after) application.

# **3.2.3.2** Seedling length (cm)

5 seedlings per applications were randomly selected. The distance from the soil surface to the seedling's top was measured for each seedling by a ruler. All measurements were performed before and after cold application.



Figure 3.7. Seedling length (before and after) application.

## 3.2.3.3 Progess Rate 1

The ratio between the number of seedlings before and after the application was determined.

## 3.2.3.4 Wilting Rate

The ratio of seedling wilting which happen at cold application duration was determined

## 3.2.3.5 Cold Damage

Cold damage at Seedling at cold application was determined.

## 3.2.3.6 Progress Rate2

The ratio between the numbers of seedlings after the application during one week was determined

## 3.3. Experimental Design:

The experimental design used was a Randomized Complete Parcel Design (RCPD) with factorial. The treatment in each experiment had three replicates. Where was statistically analyze the data and compared means using "LSD"s Multiple Range Test at 0.05 and 0.01 levels.

# **3.4**. Some pictures of the test;



Figure 3.8. Planting seeds and pepper plant growth.



Figure 3.9. Preparation of salicylic acid in Laboratory.



Figure 3.10. Take the weights SA in the laboratory.



Figure 3.11. Materials in need it to prepare the acid salicylic in the test.



Figure 3.12. Process prepare concentrations salicylic acid used in the testing.



Figure 3.13. An irrigation process for seedlings pepper.



Figure 3.14. Preparation of the process of spraying the concentrations of salicylic acid on the pepper plant.



Figure 3.15. Make a hole in the leaves of the peppers before spraying the salicylic acid on it.



Figure 3.16. Conduct a salicylic acid spray on leaves of pepper plant.

## 4. RESULTS AND DISCUSSION

Where was statistically analyze the data and compared means using "LSD"s Multiple Range Test at 0.05 and 0.01 levels, the results obtained in the test as follows;

### 4.1. Rate of Lost Seedling Weight

#### 4.1.1. Doses (mmol)

The differences between the doses with respect to Least Square (Sq.) Mean were significant. According to the Table 4.1, Least Sq. Mean ranged between -45.98 - -63.10 percent. The average Least Sq. Mean of doses were -52.03 %. The highest Least Sq. Mean values were obtained from 0.01mmol dose as -45.98 %, and the lowest Least Sq. Mean values were obtained from control concentration of salicylic acid as -63.10 %, as in the lower table (Table 4.1).

Doses mmol	Least Sq. Mean %
0.01	-45.98 a
0.05	-46.99 a
Contr.	-63.10 b
Mean	-52.03

Table 4.1. Effect of salicylic acid doses on rate of weight

Researchers Joudi and Abbas (2016), pointed out that there are similar results compared to the test where the significant differences in the effect of salicylic acid concentrations as the concentration of (0) mM SA gave the highest value but was used on the potato crop while our results were on the pepper plant.

The researchers (Al-Kraidi el at., 2017) refers when spraying the coriander plant with a concentration of (70) mg/L of the salicylic acid to have a significant effect in the soft weight of total vegetative compared to the comparative treatment. When I compared this result with this study the total result is the same but we used a concentration of 0.1 mM on the pepper plant.

Also, researchers showed (Ibrahim and Jadoua, 2014) that the treatment SA at a concentration of 50 mg/L, it was significantly superior to increase the content of chlorophyll a rate of 12 %, while the two treatments did not differ significantly from

100 to 200 mg/L on a comparison transaction, but that SA has reduced chlorophyll content by increasing concentration. This is what much research has done that the low concentration of SA increases the plant content of chlorophyll (Borsani el at., 2001).

Hayat et al. (2007) noted in their research that the soft weight decrease in the potato plant at the high concentrations of acid salicylic, and the reason returned to the use of SA, it works to promote growth and thus to water absorption which is reflected positively on the fresh weight. When we compared this result with my study the total result is the same but was used on the potato crop while our results were on the pepper plant.

#### 4.1.2. Time (h)

Times (h)	Least Sq. Mean %
24	-13.80 a
48	-67.59 b
72	-74.60 b
Mean	-51.99

Table 4.2. Effect of salicylic acid times on rate of weight

The differences between the times of application cold with respect to Least Sq. Mean were compelling. According to the table 4.2, Least Sq. Mean ranged between -13.80 - -74.6 percent. The average Least Sq. Mean of times were -51.99 %. The highest Least Sq. Mean values were obtained from time 24 h as -13.80 %, and the lowest Least Sq. Mean values were obtained from time 72 h as -74.60 % (Table 4.2).

When processing the pepper seedlings with salicylic acid for 16 hours, the results obtained, it was the negative effect on fresh weight increase, and dry weight increase in the application 5- and 10-mM SA. When we compared this result with this study the result is different because the times used are different to each other's (24, 48 and 72 hours in the test) (Canakci, 2011).

## 4.1.3. Frequency

Frequencies	Least Sq. Mean %
1	-47.70 a
2	-54.31 a
3	-54.07 a
Mean	-52.03

Table 4.3. Effect of salicylic acid frequencies on rate of weight

The differences between the frequencies with respect to Least Sq. Mean were no significant. According to the Table 4.3, Least Sq. Mean ranged between -47.70 - -54.31 percent. The average Least Sq. Mean of frequencies were -52.03 %. The highest Least Sq. Mean values were obtained from frequency 1 as -47.70 %, and the lowest Least Sq. Mean values were obtained from frequency 3 as -54.31 % (Table 4.3).

In winter wheat leaves grow at low temperature. When it is sprayed with salicylic acid, the influence of external factors decreased and also the decreased freezing injury (Taşgin et al., 2003). These results obtained by the researcher in his research are similar to results of our study in our research. However, the salicylic acid spray was used once on the wheat crop, but we used 1-3 repetitions and on the pepper seedlings.

# 4.1.4. Frequency and Time (h)

Frequencies	Times (h)	Least Sq. Mean %
2	24	-10.98 a
3	24	-13.10 a
1	24	-17.32 a
1	48	-58.83 b
1	72	-66.93 bc
2	48	-71.44 bc
3	48	-72.50 bc
3	72	-76.50 c
2	72	-80.50 c
Me	-52.01	

The differences between the frequencies and times with respect to Least Sq. Mean were significant. With reference to the Table 4.4, Least Sq. Mean ranged between - 10.98 - 80.50 percent. The average Least Sq. Mean of frequencies and times were - 52.01 %. The highest Least Sq. Mean values were obtained from frequency 2 and time 24 h as -10.98 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and time 72 h as -80.50 % (Table 4.4). When the first three results are observed, 24 h applications were the best result and very different from the other applications.

Frequencies	Doses mmol	Least Sq. Mean
1	0,01	-41.35 a
1	0,05	-46.45 a
3	0,05	-47.15 a
2	0,05	-47.37 a
3	0,01	-47.88 a
2	0,01	-48.70 a
1	Contr.	-55.27 ab
2	Contr.	-66.86 b
3	Contr.	-67.17 b
Mea	n	-52.02

4.1.5. Frequency and Doses (mmol)

Table 4.5. Effect of interaction the frequencies and doses on rate of weight

The differences between the frequencies and doses with respect to Least Sq. Mean were significant. According to the Table 4.5, Least Sq. Mean ranged between -41.35 – 67.17 percent. The average Least Sq. Mean of frequencies and doses were -52.02 %. The highest Least Sq. Mean values were obtained from frequency 1 and dose 0.01mmol as -41.35 %, and the lowest Least Sq. Mean values were obtained from frequency 3 and control dose as -67.17 % (Table 4.5). At the point when watched the initial three outcomes, and one of the concentration at 0.01 mmol, and two other at 0.05 mmol applications were the best outcome, and were, altogether, different from other applications. While the last three outcomes have fewer outcomes.

Salicylic acid sprinkled on coriander plant with at concentrations of 20 and 35 mg/L. Got results on the significant increase in the soft weight of the vegetative group, and the number of flowers inflorescences, and the number of seeds per inflorescence,

and weight 100 seed and production of the plant seeds compared to those that were not sprayed (Al-Kraidi et al., 2017). The results of these research are smiliar compared to these results we concluded from the test but we used pepper plant while it is used on plant coriander.

Reached the researcher (El-Yazied, 2011) it could be recommended that foliar spraying with salicylic acid at 100 ppm, to increase the final yield and fruit quality of sweet pepper plant during the low temperatures of autumn plantations. This is what we applied to our study and of the stay of the seedling.

The researcher found (Jassim and Al-Kooranee, 2012) effect of salicylic acid on macrophomina and evolution of fever disease on Sun Flower plant. Results experience recommended that all pots concentrations may be affected significantly in reducing the percentage of injury. As for the concentrations of 200 and 250 mg /L salicylic acids effect of the dry weight increase total of vegetables and also an increase in the dry weight total of the roots. The study indicated that high concentrations of salicylic acid have been reduced by the number of stone bodies (sclerotia). While low concentrations of salicylic acid have significant differences with control treatment. Application of SA is different from our test, while our application of SA was sprayed on the pepper plant at the low temperature.

### 4.1.6. Frequency, Doses (mmol) and Time (h)

The differences between the frequencies, doses and times with respect to Least Sq. Mean were significant. With reference to the Table 4.6, Least Sq. Mean ranged between -9.84 - -96.50 percent. The average Least Sq. Mean of frequencies and doses and times were -52.02 %. The highest Least Sq. Mean values were obtained from frequency 2 and control dose and time 24 h as -9.48 %, and the lowest Least Sq. Mean values were obtained from frequency 3 and control doses and time 48 h as -96.50 %, as in the lower table (Table 4.6). It has been observed the first sixth results, 24 h applications were the best result and very different from the other applications. While the last sixth results have the worst results at the five concentration in the (contr.) and only one at 0.01mmol.

Frequencies	Doses mmol	Times (h)	Least Sq. Mean %
2	Contr.	24	-9.84 a
3	0.01	24	-10.57 a
2	0.05	24	-10.71 a
2	0.01	24	-12.40 a
3	0.05	24	-13.56 a
1	0.05	24	-14.16 a
3	Contr.	24	-15.18 a
1	Contr.	24	-16.20 a
1	0.01	24	-21.61 a
1	0.01	48	-23.01 a
2	0.05	48	-53.43 b
3	0.05	48	-55.32 bc
1	0.05	72	-58.60 bc
1	Contr.	72	-62.75 b-d
3	0.01	48	-65.69 b-e
2	0.01	48	-65.86b-e
1	0.05	48	-66.60 b-e
3	0.01	72	-67.37 b-e
2	0.01	72	-67.84 b-e
3	0.05	72	-72.56 b-f
2	0.05	72	-77.97 b-f
1	0.01	72	-79.44 c-f
1	Contr.	48	-86.87 d-f
3	Contr.	72	-89.83 ef
2	Contr.	48	-95.04 f
2	Contr.	72	-95.71 f
3	Contr.	48	-96.50 f
	Mean	I	52.02

Table 4.6. Effect of interaction the frequencies, doses and times on rate of weight

Fariduddin et al. (2003) concluded that SA has important effects on the physiological activities related to the growth and development of plants under normal conditions without stress the need to control the absorption and transfer of ions and the permeability of cellular membranes. These results what we reached in the test.

The field study has conducted the response of two types of basil to salicylic acid and their overlap in growth indicators and plant yield during growing seasons. The results local type significantly exceeded the foreign species during the two seasons, when this study SA spray with a concentration of 100 mg/L had a significant effect on leaf content of chlorophyll, carotene, phenols, amino acids, the propellant ratio of oil, pilot oil yield per unit area for seasons. The bilateral and trilateral interaction between the study factors showed a significant effect on the above characteristics (Al-Saadi et al., 2017). Application of SA is different from our test when the two types of basil through two seasons were used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

#### 4.1.7. Time (h) and Doses (mmol)

Table 4.7. Effect of interaction the times and doses on rate of weight

Times (h)	Doses mmol	Least Sq. Mean %
24	0,05	-12.81 a
24	Contr.	-13.74 a
24	0,01	-14.86 a
48	0,01	-51.52 b
48	0,05	-58.45 bc
72	0,05	-69.71 cd
72	0,01	-71.55 cd
72	Contr.	-82.76 de
48	Contr.	-92.80 e
M	ean	-52.02

The differences between the times and doses with respect to Least Sq. Mean were significant. According to the Table 4.7, Least Sq. Mean ranged between -12.81 - 92.80 percent. The average Least Sq. Mean of times and doses were -52.02 %. The highest Least Sq. Mean values were obtained from time 24 h and dose 0.05 mmol as -12.81 %, and the lowest Least Sq. Mean values were obtained from time 48 h and control dose as -92.80 % (Table 4.7). It can be seen that the first three results, 24 h applications and it exists all the concentration in were the best results are very different from the other applications. Whereas the last three results have the worst results at the most in the control concentration and the time in at 72 h applications.

The researcher concluded (Al-Hamdani el at., 2017) that spraying plants with low concentrations of salicylic acid it can stimulate endure of vital and abiotic stresses such as cold and tolerance high-temperature. This is what we applied to our effects of the salicylic acid application on cold tolerance and gene expression in pepper seedling in the test.

The researchers of Senaratna et al. (2000)'s study showed that plants did not show saturated seed high concentration of 1 mM at the SA any change in tolerance iced, while the low concentrations of 0.1-0.5 mM at the SA encouraged tolerance to sedative stress in bean and tomato.

SA induced resistance to (*Powdery mildew*) disease of milk thistle. The results suggest that the use of different concentrations of SA, it led to the increase in fresh weight and dry alone and plant height, and dry weight differed significantly from the rest of the transactions except for 1 mM concentration of SA spray one (Ramadan et al., 2018). Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

## 4.2. Rate of Lost Seedling Length

### 4.2.1. Doses (mmol)

The differences between the doses with respect to Least Sq. Mean were significant. With reference to the Table 4.8, Least Sq. Mean ranged between -51.95 - -65.39 percent. The average Least Sq. Mean of doses were -51.95 %. The highest Least Sq. Mean values were obtained from dose 0.01 mmol as -51.95 %, and the lowest Least Sq. Mean values were obtained from control dose as -65.39 % as in the lower table (Table 4.8).

Doses mmol	Least Sq. Mean %
0.01	-51.95 a
0.05	-59.02 ab
Contr.	-65.39 b
Mean	-58.79

Researchers reached to (Turk and Abu–Hinna, 2016) the results of their studies that spraying potato plants with high concentrations led to an increase in plant length. The results obtained by researchers, such as the results obtained from the research but used salicylic acid spray was on the potato plant.

Salicylic acid is a plant hormone that promotes growth under stress conditions and in its absence, it improves growth and increases total nitrogen intake and nitrate by increasing the effectiveness of nitrate reeducate at the same time, salicylic inhibits the formation of ethylene, which leads to increased plant height (Horvath el at., 2002) and (Noreen el at., 2009). The researcher concluded (Al Jabber, 2016) to study the effect of planting and spraying with salicylic acid in growth, leaf extract, seeds and oil of the Dill plant. Plants sprayed with salicylic acid gave a significant increase in plant height, numbers of lateral branches, leaves the number, number of inflorescences, inflorescence, the weight of 100 seeds, seed yield, volatile oil percentage and volatile oil yield. Where the experiment included the overlap between two factors: the method of cultivation lines or scattering and sprinkling with three concentrations of salicylic acid, 0, 30 or 60 mg/L and at two times, and three replicates. Its results differ from our results where the experiment was conducted on a Dill plant while we conducted the test in the laboratory and the pepper plant.

### 4.2.2. Time (h)

Table 4.9.	. Effect of	fsalicvlic	acid times	on rate	of length

Times (h)	Least Sq. Mean %
24	-28.60 a
48	-60.21 b
72	-87.55 c
Mean	-58.79

The differences between the times and doses with respect to Least Sq. Mean were significant. According to the Table 4.9, Least Sq. Mean ranged between -28.60 - -87.55 percent. The average Least Sq. Mean of times were -58.79 %. The highest Least Sq. Mean values were obtained from time 24 h as -28.60 %, and the lowest Least Sq. Mean values were obtained from time 72 h as -87.55 % (Table 4.9).

In the study of salicylic acid prevents, the synthesis of protein inhibitors in tomato leaves caused by system in and jasmonic acid (JA), by the researcher (Doares et al., 1995). He explained that strong inhibitors of acid-induced, and explained that the inhibition by the salicylic acid of proteinase inhibitor synthesis induced by system and JA. The researcher used the tomato leaves while we used pepper.

The results of the researcher (Mercado et al., 1997) were that in the low temperatures showed cultivated plants decreased by 50–70 % in the number of leaves and the length and dry weight compared to the high-temperature system. It was also shown in the cold system of plants grown an increased number of shoots in the armpits. Also, the content of proteins and chlorophyll decreased in both temperature treatments. The total nitrogen content was slightly higher at low temperature, but nitrate was lower. These results, are like the result that we obtained from the test, and an action the researchers' studies in the field, but we used the laboratory in the test.

## 4.2.3. Frequency

Table 4.10. Effect of salicylic acid frequencies on rate of length

Frequencies	Least Sq. Mean %
2	-57.85 a
1	-58.35 a
3	-60.15 a
Mean	-58.79

The differences between the frequencies with respect to Least Sq. Mean were no significant. According to the Table 4.10, Least Sq. Mean ranged between -57.85–-60.15 percent. The average Least Sq. Mean of frequencies were -58.79 %. The highest Least Sq. Mean values were obtained from frequency 2 as -57.85 %, and the lowest Least Sq. Mean values were obtained from frequency 3 as -60.15 % (Table 4.10).

The researchers (Shakirova et al., 2003) concluded that Salicylic acid increases plants resistance to inappropriate conditions especially against the stress that is the plant is exposed to stress (saline and drought), it works on the organization of some physiological processes of plant photosynthesis and transpiration. And the results of this conformity our results in the test while we used to tolerate pepper plant for low temperatures.

## 4.2.4. Frequency and Time (h)

Frequencies	Frequencies Times (h)	
2	24	-24.59 a
1	24	-29.75 a
3	24	-31.47 a
1	48	-58.82 b
2	48	-59.89 b
3	48	-61.91 b
1	72	-86.50 c
3	72	-87.08 c
2	72	-89.08 c
Me	an	-58.79

The differences between the frequencies and times with respect to Least Sq. Mean were significant. According to the Table 4.11, Least Sq. Mean ranged between -24.59 - -89.08 percent. The average Least Sq. Mean of frequencies and times were -58.79 %. The highest Least Sq. Mean values were obtained from frequency 2 and time 24 h as -24.59 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and time 72 h as -89.08 % (Table 4.11). It has been clearly observed that the first three results, 24 h applications were the best result and very different from the other applications, while the last three results have the worst results.

The researchers (Aziz et al., 2015) pointed out to that the spraying of *narcissus* plant with salicylic acid led to an increase in plant height and also a significant increase in concentrations of 40 and 80 mg/I. These results, are likely similar to the result which we obtained from the test. They both show that the higher the concentration of salicylic acid, the greater the length of the plant.

#### 4.2.5. Frequency and Doses (mmol)

Frequencies	Doses mmol	Least Sq. Mean %	
1	0,01	-43.80 a	
2	0,05	-51.58 ab	
2	0,01	-53.18 a-c	
3	0,01	-58.88 b-c	
3	Contr.	-60.33 b-c	
3	0,05	-61.25 b-c	
1	0,05	-64.22 b-c	
1	Contr.	-67.04 cd	
2	Contr.	-68.79 d	
Mea	an	-58.79	

Table 4.12. Effect of interaction the frequencies and doses on rate of length

The differences between the frequencies and doses with respect to Least Sq. Mean were significant. With reference to the Table 4.12, Least Sq. Mean ranged between - 43.80 - 68.79 percent. The average Least Sq. Mean of frequencies and doses were - 58.79 %. The highest Least Sq. Mean values were obtained from frequencies 1 and dose 0.01 mmol as -43.80 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and control dose as -68.79 % (Table 4.12). At the point when watched the initial three outcomes, and two of the concentration at 0.01 mmol and one of the other at 0.05 mmol applications were the best outcome and altogether different from other applications. While the last three outcomes have fewer outcomes.

Khan et al. (2003) concluded in a study of the effect of salicylic acid (SA) and gentisic acid (GTA) on the growth rates of soybeans, maize, and photosynthesis under greenhouse conditions. The chlorophyll content of these compounds does not change. And treatment in some cases with these compounds led to an increase in the dry mass of the plant and leaf areas, however, root length and plant height were not affected. The results of this research are different from the results of our tests when the plants of soybeans and maize were used with SA, while we use SA was sprayed on the pepper plant at the low temperature.

The effect of adding salicylic acid in the potato plant (*Solanum tuberosum* L.) growth indicators was conducted by Al Jabber (2016). The results were significantly higher in concentration of 75 mg/L to giving the highest plant length and the largest

number of leaves and the largest leaf area and the highest rate of tuber weight and the highest yield of one plant and the total yield of the unit of area and the content of the tuber from the original coupler treatment comparison that gave lower values as compared to the comparative treatment which gave lower values. Its results differ from our results where the experiment was conducted on a potato plant while our use SA was sprayed on the pepper plant at the low temperature.

## 4.2.6. Frequency, Doses (mmol) and Time (h)

The differences between the frequencies, doses and times with respect to Least Sq. Mean were significant. According to the Table 4.13, Least Sq. Mean ranged between -19.31 - -95.65 percent. The average Least Sq. Mean of frequencies and doses and times were -58.79 %. The highest Least Sq. Mean values were obtained from frequency 1 and dose 0.01 mmol and time 48 h as -19.31 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and control dose and time 48 h as -95.65 % as in the lower table (Table 4.13). It is observed that the most of first sixth results when at concentration comparing and time at 72h and each repetition twice, applications were the best result and very different from the other applications, whereas the last sixth results have the worst results at the four concentration in the (contr.) and two at 0.01 mmol.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
1	0.01	48	-19.31 a
3	Contr.	24	-19.43 a
1	Contr.	24	-21.86 ab
2	Contr.	24	-22.27 ab
3	0.01	24	-22.46 ab
2	0.01	24	-23.28 ab
1	0.01	24	-27.05 ab
2	0.05	24	-28.21 a-c
1	0.05	24	-40.33 a-d
2	0.01	48	-44.75 b-e
2	0.05	48	-46.46 b-f
3	0.05	24	-52.53 c-g
3	0.01	48	-55.28 d-g
3	0.01	48	-60.26 d-h
1	0.05	48	-68.71 e-i
3	Contr.	48	-70.19 f-j
3	0.05	72	-75.94 g-k
2	0.05	72	-80.08 h-k
1	0.05	72	-83.64 h-k
1	0.01	72	-85.03 h-k
1	Contr.	48	-88.43 i-k
2	Contr.	48	-88.46 i-k
1	Contr.	72	-90.83 i-k
3	Contr.	72	-91.37 i-k
2	0.01	72	-91.50 i-k
3	0.01	72	-93.91 jk
2	Contr.	72	-95.65k
	Mean		-58.79

Table 4.13. Effect of interaction the frequencies, doses and times on rate of length

Al-Dabbagh and Al-Duleimi (2017) concluded productivity and quality of two genotypes of two mung bean (domestic and Indian) in order to determine the effect of paper feeding in three concentrations of salicylic acid (SA). Results showed gave a concentration of 150 mg/L SA in the two seasons the highest fertility in Alqrnat and the highest average quotient of seed, scored in the spring season, the highest weight of 100 seed while superiority in Autumnal in the sum of protein season, while the concentration of 300 mg/L SA may be greater than in the season In the number of seeds by pod. A field experiment was carried out in the spring and autumn seasons, while we conducted the test in the laboratory and on the pepper plant.

The found in an experiment (Al-Zyadi and Mohammad, 2018) results showed the superiority of cultivated plants in the first date in plant height, the leaf area, the dry weight of vegetative total and the dry weight of root total, and the superiority of plants in the third in giving their roots production higher than quercetin. As he gave the superiority of plants salicylic acid treatment with 100 mg/L concentration in the leaf area, the plant height, dry weight of vegetative growth and dry weight of roots, as well as the content of roots of quercetin plant dry weight. Application of SA is different from our test when the burdock plant was used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

Aziz et al. (2015) found about the response of *narcissus* plant to spraying with plant growth regulators salicylic acid the characteristics of the vegetative growth and syphilis and bulbs characteristics of plant daffodils. The results increased at the 80 mg/L SA in the plant height, the number of branches/plant, and the ratio of chlorophyll and wet weight and flower diameter, weight wet and dry. The effect of overlap between the two studied factors is significant in most studied traits. Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

## 4.2.7. Time (h) and Doses (mmol)

Table 4.14. Effect of interaction the times and doses on rate of length

Times (h)	Doses mmol	Least Sq. Mean %
24	Contr.	-21.19 a
24	0,01	-24.26 a
24	0,05	-40.36 b
48	0,01	-41.44 b
48	0,05	-56.82 c
72	0,05	-79.89 d
48	Contr.	-82.36 d
72	0,01	-90.15 d
72	Contr.	-92.62 d
Mean		-58.79

The differences between the times and doses with respect to Least Sq. Mean were significant. With respect to the Table 4.14, Least Sq. Mean ranged between -21.19 - 92.62 percent. The average Least Sq. Mean of times and doses were -58.79 %. The highest Least Sq. Mean values were obtained from time 24 h and control dose as -21.19 %, and the lowest Least Sq. Mean values were obtained from time 72 h and control dose as -92.62 % (Table 4.14). Observing the first three results, 24 h applications and it exists all the concentration in were the best results and very different from the other applications. While the last three results have the worst results at the most concentration in the (contr.) and the time in at 72 h applications.

In a study by Hameed et al. (2015) the effect of water stress and the external application of SA on the growth and production of eggplant were invstigatted. Results showed a significant increase for the first irrigation level W1 (water stress1) compared with level W2 (water stress2) transactions W3 (water stress3) for all indicators. Interacted treatment W1A2 (water stress1 x Applcation2) showed significant increase compared with other treatments. Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

## 4.3. Turgority

## 4.3.1. Doses (mmol)

As shown in the lower table (Table 4.15) there are no significant between salicylic acid concentrations compared to the comparative control dose.

Doses (mmol)	Least Sq. Mean %
0.01	la
0.05	1 a
Contr.	1 a

Table 4.15. Effect of salicylic acid doses on rate of turgority

## 4.3.2. Time (h)

As shown in the lower table (Table 4.16) there are no significant between the duration used in the research.

Times (h)	Least Sq. Mean %
24	1 a
48	1 a
72	1 a

## 4.3.3. Frequency

As shown in the lower table (Table 4.17) there are no significant when the frequencies of the spraying on the pepper plants.

Frequencies	Least Sq. Mean %
1	1 a
2	1 a
3	1 a

## 4.3.4. Frequency and Time (h)

As shown in the lower table (Table 4.18) there are no significant between the interaction of the frequencies and duration used in the test.

Frequencies	Times (h)	Least Sq. Mean %
1	24	1 a
1	48	1 a
1	72	1 a
2	24	1 a
2	48	1 a
2	72	1 a
3	24	1 a
3	48	1 a
3	72	1 a

Table 4.18. Effect of interaction the frequencies and times on rate of turgority

## 4.3.5. Frequency and Doses (mmol)

As shown in the lower table (Table 4.19) there are no significant between the interaction of the frequencies and doses used in the research.

Frequencies	Doses (mmol)	Least Sq. Mean %
1	0,01	1 a
1	0,05	1 a
1	Contr.	1 a
2	0,01	1 a
2	0,05	1 a
2	Contr.	1 a
3	0,01	1 a
3	0,05	1 a
3	Contr.	1 a

Table 4.19. Effect of interaction the frequencies and doses on rate of turgority

# 4.3.6. Frequency, Doses (mmol) and Time (h)

As shown in the lower table (Table 4.20) there are no significant between the interaction of the frequencies, doses, and duration used in the research.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
1	0,01	24	1 a
1	0.01	48	1 a
1	0,01	72	1 a
1	0,05	24	1 a
1	0,05	48	1 a
1	0,05	72	1 a
1	Contr.	24	1 a
1	Contr.	48	1 a
1	Contr.	72	1 a
2	0,01	24	1 a
2	0,01	48	1 a
2	0,01	72	1 a
2	0,05	24	1 a
2	0,05	48	1 a
2	0,05	72	1 a
2	Contr.	24	1 a
2	Contr.	48	1 a
2	Contr.	72	1 a
3	0,01	24	1 a
3	0,01	48	1 a
3	0,01	72	1 a
3	0,05	24	1 a
3	0,05	48	1 a
3	0,05	72	1 a
3	Contr.	24	1 a
3	Contr.	48	1 a
3	Contr.	72	1 a

Table 4.20. Effect of interaction the frequencies, doses and times on rate of turgority

# 4.3.7. Time (h) and Doses (mmol)

As shown in the lower table (Table 4.21) there are no significant between the interaction of the duration and doses used in the research.

Times (h)	Doses (mmol)	Least Sq. Mean %
24	0,01	1 a
24	0,05	1 a
24	Contr.	1 a
48	0,01	1 a
48	0,05	1 a
48	Contr.	1 a
72	0,01	1 a
72	0,05	1 a
72	Contr.	1 a

Table 4.21. Effect of interaction the times and doses on rate of turgority

Flaccidity (turgority) of leaves and stem are very clear symptoms in lowtemperature conditions (Mitchell and Broadhead, 1967; Airaki et al., 2012). Salicylic acid effects to vegetable growth (Turk and Abu-Hinna, 2016), SA led to increase in fresh weight (Noodén et al., 1997) and gives the highest plant length and the largest number of leaves and largest leaf area (Turk and Abu-Hinna, 2016). Applying SA gives the plant the regulation of water, salinity and cold stress (Miura and Tada, 2014). Also, salicylic acid could reduce leaf breakage (Ferrarese et al., 1996). SA is one of the substances like hormone and SA's role is important in the direction the physiological development (Noreen et al., 2009). Wheat leaves grown at low temperatures when sprayed with salicylic acid, the influence of external factors decreased and also the decreased freezing injury (Tasgin et al., 2003). Aldesuquy et al. (2012) found that investigated the effect of shikimic and salicylic acids at the concentrations of 0.4 and 0.7 mmol, respectively or their combination as phenolic compounds. The pathogen has caused a significant decrease in the relative water content of the infected leaves, as a result, increased water saturation deficit. SA helps to keep water inside the plant for example in the stem, leaves and the other tissues. And SA cause recoups of transpiration rate and stomatal closure (Aldesuquy et al., 2014).

#### 4.4. Progress Rate1

#### 4.4.1. Doses (mmol)

The differences between the doses with respect to Least Sq. Mean were significant. With respect to the Table 4.22, Least Sq. Mean ranged between 0.26 - 0.64 percent. The average Least Sq. Mean of doses were 0.45 %. The highest Least Sq. Mean values were obtained from dose 0.05 mmol as 0.64 %, and the lowest Least Sq. Mean values were obtained from control dose as 0.26 %, as in the lower table (Table 4.22).

Doses (mmol)	Least Sq. Mean %
0.05	0.64 a
0.01	0.44 b
Contr.	0.26 c
Mean	0.45

Table 4.22. Effect of salicylic acid doses on rate of progress rate 1

Canakci (2011) indicated that the salicylic acid is an effective precursor molecule that builds resistance against abiotic and bio-stress in plants, depending on the concentration of salicylic acid applied exogenously and plant type. However, high concentrations of salicylic acid and applied individually. This is what we applied to our tests, it was at low temperatures, the effect of salicylic acid on the pepper plant.

As a found (Galal, 2012) salicylic acid is used in the agricultural medium of the jujube plant with concentrations of 10, 25, and 50 mg / L his positive effect. Where the highest percentage for the survival of plants emerged alive at concentration 10 mg/L salicylic acid and as well good response to a germination branch and rooting with the same emphasis. These results are close to our results obtained from the test.

In experiment of Anjum et al. (2011) the effectiveness of bread yeast and salicylic acid against fungi (*Saccharomyces cerevisiae*) which cause of green mold on orange fruits was studied. Achieved salicylic acid positive impact on the growth of fungus (*S. cerevisiae*) all concentrations used. Also, the results showed for the development of resistance in orange fruits, it has outperformed the combination salicylic acid and yeast (*S. cerevisiae*) in the development of systemic resistance by increasing the effectiveness

of the enzyme (Peroxidase) in the fruits of analogy treatment of salicylic acid alone. The results of this experience are different from our results.

## 4.4.2. Time (h)

Table 4.23. Effect of salicylic acid times on rate of progress rate 1

Times (h)	Least Sq. Mean %
24	0.76 a
48	0.49 b
72	0.09 c
Mean	0.45

The differences between the times with respect to Least Sq. Mean were significant. According to the Table 4.23, Least Sq. Mean ranged between 0.08 - 0.76 percent. The average Least Sq. Mean of times were 0.45 %. The highest Least Sq. Mean values were obtained from time 24 h as 0.76 %, and the lowest Least Sq. Mean values were obtained from time 72 h as 0.09 % (Table 4.23).

Hayat et al. (2007) concluded that the salicylic acid play an important role in accelerating the formation of chlorophyll and carotene dyes, accelerate the carbon process and increase the activity of some important enzymes. This result shows how plant survives at the low temperature.

#### 4.4.3. Frequency

Table 4.24. Effect of salicylic acid frequencies on rate of progress rate 1

Frequencies	Least Sq. Mean %
3	0.49 a
1	0.45 b
2	0.41 b
Mean	0.45

The differences between the frequencies with respect to Least Sq. Mean were significant. According to the Table 4.24, Least Sq. Mean ranged between 0.41 - 0.49 percent. The average Least Sq. Mean of frequencies were 0.45 %. The highest Least Sq.

Mean values were obtained from frequency 3 as 0.49 %, and the lowest Least Sq. Mean values were obtained from frequency 2 as 0.41 % (Table 4.24).

SA is able to reduce the secretion in both pepper plants and peach, and biochemical analysis revealed that the enzyme Sulailaz does not increase after activating the secretion of paper in the treatment of SA, in return for, plants control stations show a marked increase in levels of both enzyme activity and protein cellulose (Ferrarese et al., 1996). The method use of SA is different from our test when the peach and pepper plants were used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

Frequencies	Times (h)	Least Sq. Mean %
1	24	0.81 a
3	24	0.79 a
2	24	0.70 b
3	48	0.50 c
1	48	0.49 c
2	48	0.48 c
3	72	0.20 d
1	72	0.03 e
2	72	0.03 e
Mea	an	0.45

#### 4.4.4. Frequency and Time (h)

	Table 4.25. Effect	of interaction th	he frequencies and	times on rate of progress rate 1
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The differences between the frequencies and times with respect to Least Sq. Mean were significant. According to the Table 4.25, Least Sq. Mean ranged between 0.81 - 0.03 percent. The average Least Sq. Mean of frequencies and times were 0.45 %. The highest Least Sq. Mean values were obtained from frequency 1 and time 24 h as 0.81 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and time 72 h as 0.03 % (Table 4.25). It is observed that the first three results, 24 h applications were the best result and very different from the other applications, while the last three results have the worst results at 72 h applications.

Popova et al. (2008) concluded that the pre-treatment with salicylic acid plant seeds split peas the harmful effect reduces on the growth and photosynthesis and interactions carboxylation, in addition to the content of chlorophyll. Application of SA is different from our test when the put pea's seed was used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

## 4.4.5. Frequency and Doses (mmol)

Frequencies	Doses (mmol)	Least Sq. Mean %
3	0,05	0.71 a
2	0,05	0.65 a
1	0,05	0.55 b
1	0,01	0.48 bc
3	0,01	0.45 c
2	0,01	0.41 c
3	Contr.	0.31 d
1	Contr.	0.30 d
2	Contr.	0.15 e
Ν	lean	0.45

Table 4.26. Effect of interaction the frequencies and doses on rate of progress rate 1

The differences between the frequencies and doses with respect to Least Sq. Mean were significant. With respect to the Table 4.26, Least Sq. Mean ranged between 0.15 - 0.71 percent. The average Least Sq. Mean of frequencies and doses were 0.45 %. The highest Least Sq. Mean values were obtained from frequency 3 and dose 0.05 mmol as 0.71 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and control dose as 0.15 % (Table 4.26). At the point when watched the initial three outcomes, the concentration at 0.05 mmol applications were the best outcome and altogether different from other applications, while the last three outcomes have fewer outcomes at the control doses.

### 4.4.6. Frequency, Doses (mmol) and Time (h)

The differences between the frequencies, doses and times with respect to Least Sq. Mean were significant. According to the Table 4.27, Least Sq. Mean ranged between 0.01 - 0.98 percent. The average Least Sq. Mean of frequencies and doses and times were 0.45 %. The highest Least Sq. Mean values were obtained from frequency 3 and dose 0.05 mmol and time 24 h as 0.98 %, and the lowest Least Sq. Mean values were obtained from frequency 1 and control dose and time 72 h as 0.01 %, as in the lower table (Table 4.27). The first sixth results, when concentration 0.05 mmol and most of the time at 24 h and each repetition, applications were the best results and very different from the other applications, while the last sixth results have the worst results at the time of them 72h.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
3	0.05	24	0.98 a
2	0.05	24	0.98 a
1	0.05	24	0.95 ab
3	0.05	48	0.94 ab
2	0.05	48	0.93 ab
3	0.01	24	0.90 ab
1	0.01	24	0.87 ab
2	0.01	24	0.83 b
1	0.05	48	0.69 c
1	Contr.	24	0.62 cd
1	0.01	48	0.51 de
3	Contr.	24	0.48 ef
2	0.01	48	0.38 fg
3	0.01	48	0.33 gh
2	Contr.	24	0.30 gh
1	Contr.	48	0.29 gh
3	Contr.	72	0.25 hi
3	0.05	72	0.23 h-j
3	Contr.	48	0.22 h-j
2	Contr.	48	0.15 i-k
3	0.01	72	0.12 j-l
1	0.01	72	0.06 kl
2	0.05	72	0.05 kl
2	0.01	72	0.03 kl
1	0.05	72	0.02 1
2	Contr.	72	0.02 1
1	Contr.	72	0.01 1
	Mean	I	0.45

Table 4.27. Effect of interaction the frequencies, doses and times on rate of progress rate1

According to the study of Senaratna et al. (2000)'s, when the improving the cold resistance of the plants of tomatoes and beans at the soaked their seeds in the solution of aspirin or SA solution at a concentration of 0.1 and 0.5 mmol before agriculture is the better application. The results of this research are different from our results because the

method of application is different where the seeds are immolersed tomatoes and beans in salicylic acid solution while the application method to spray the pepper plant.

The chlorophyll content increased 1.5 times, and the protein amount increased with SA appliyings. The results of this study differ from the results of our test. Because it was used to spray SA on the sunflower crop, while we used SA spray on the pepper plant (Cag et al., 2009).

Times (h)	Doses (mmol)	Least Sq. Mean %
24	0,05	0.96 a
24	0,01	0.86 b
48	0,05	0.85 b
24	Contr.	0.46 c
48	0,01	0.41 c
48	Contr.	0.22 d
72	0,05	0.10 e
72	Contr.	0.09 e
72	0,01	0.07 e
Me	ean	0.45

### 4.4.7. Time (h) and Doses (mmol)

 Table 4.28. Effect of interaction the times and doses on rate of progress rate 1

The differences between the times and doses with respect to Least Sq. Mean were significant. With respect to the Table 4.28, Least Sq. Mean ranged between 0.07 –0.96 percent. The average Least Sq. Mean of times and doses were 0.45 %. The highest Least Sq. Mean values were obtained from time 24 h and dose 0.05 mmol as 0.96 %, and the lowest Least Sq. Mean values were obtained from time 72 h and dose 0.01 mmol as 0.07 % (Table 4.28). It can be observed the first three results, at in the two time 24h applications and it two the concentration at 0.05 mmol in were the best results and very different from the other applications. While the last three results have the worst results at the all concentrations and the times in at 72 h applications.

In this study the researcher (Kling and Meyer, 1983) found that an appropriate concentration of SA was determined on the rooting of the poinsettia. The results showed that salicylic acid was obtained, which increased the rooting rate. And has a positive effect on the use of SA, plant growth regulators SA have a profound effect on rooting of

poinsettia. Application of SA is different from our test when SA was determined on the rooting of the poinsettia, while our use SA was sprayed on the pepper plant at the low temperature.

The researcher of Al Jabber, (2016) about the plants that were planted with salicylic acid outperformed the plant height, the number of lateral branches, the number of spores, the number of seeds per bulb, the weight of 100 seeds, and the seed yield. While cultivated excelled in a way prose plants significant for plant height and overtook plants sprayed with SA in plant height and number of branches and side number of inflorescences and the number of seeds per inflorescence and 100 seed weight and seed quotient. Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

#### 4.5. Wilting Rate

#### 4.5.1. Doses (mmol)

The differences between the doses with respect to Least Sq. Mean were significant. According to the Table 4.29, Least Sq. Mean ranged between 0.26 - 0.75 percent. The average Least Sq. Mean of doses were 0.55 %. The highest Least Sq. Mean values were obtained from control dose as 0.75 %, and the lowest Least Sq. Mean values were obtained from dose 0.05 mmol as 0.35 %, as in the lower table (Table 4.29).

Table 4.29. Effect of	doses salicylic acid	on rate of wilting rate

Doses (mmol)	Least Sq. Mean %
Contr.	0.75 a
0.01	0.55 b
0.05	0.35 c
Mean	0.55

Ding et al. (2002) concluded that the utilization of low concentrations of SA to tomato fruits products reduced the chilling damage and the occurrence of rot amid low-temperature stockpiling. These results are similar to our results but we used SA spray on the pepper plant.

Upgraded protection from bacterial wilt (BW) malady with no negative impact on plant development in SA-prepared chili during BW contamination. The differential articulation design uncovered that proteins related with pressure and guard, vitality, and digestion, protein blend, protein destination, and storage and transcription-related were unregulated demonstrating the association of SA prompted ailment opposition in bean chili seedlings, suggests this probably use as biomarkers in screening susceptibility of chili cultivars for wilt ailment (Chandrasekhar et al., 2017). Application of SA is different from this test, while we use SA was spraying on the leaves of pepper at the low temperature.

#### 4.5.2. Time (h)

Times (h)	Least Sq. Mean %
72	0.93 a
48	0.51 b
24	0.23 с
Mean	0.55

**Table 4.30.** Effect of salicylic acid times on rate of wilting rate

The differences between the times with respect to Least Sq. Mean were significant. With respect to the Table 4.30, Least Sq. Mean ranged between 0.23 - 0.93 percent. The average Least Sq. Mean of times were 0.55 %. The highest Least Sq. Mean values were obtained from time 72 h as 0.93 %, and the lowest Least Sq. Mean values were obtained from time 24 h as 0.23 % (Table 4.30).

Airaki et al. (2012) indicated that the pepper seedlings are exposed to low temperatures 8 °C during 1-3 days, caused an increase in the moisture content in both stem and leaves. These results are similar to our results.

An old have investigated the prerequisite of SA for mounting the easily hypersensitive response (HR) against an attacking pathogen, where a specific cell demise process is enacted at the site of attempted disease causing a bound lesion. That SA potentiates the flagged pathway by influencing an early phosphorylation-delicate advance going before the age of genius disappearance signals, including those got from the oxidative burst (Alvarez, 2000). Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

## 4.5.3. Frequency

Frequencies	Least Sq. Mean %
2	0.59 a
1	0.55 b
3	0.52 b
Mean	0.55

Table 4.31. Effect of salicylic acid frequencies on rate of wilting rate

The differences between the frequencies with respect to Least Sq. Mean were significant. According to the Table 4.31, Least Sq. Mean ranged between 0.52 - 0.59 percent. The average Least Sq. Mean of frequencies were 0.55 %. The highest Least Sq. Mean values were obtained from frequency 2 as 0.59 %, and the lowest Least Sq. Mean values were obtained from frequency 3 as 0.52 % (Table 4.31).

Jayakannan et al. (2013) concluded in his study that increasing the state of abiotic stress, SA interferes with various physiological responses such as lipid oxidation-reduction, oxidative system regulation, and ion prohibiting, osmotic adjustments and kinases synthesis. We introduced the low-temperature factor in our tests to the credibility of the results of this research and the results were good.

# 4.5.4. Frequency and Time (h)

Table 4.32. Effect of interaction the frequencies and times on rate of wilting rate

Frequencies	Times (h)	Least Sq. Mean %
2	72	0.97 a
1	72	0.97 a
3	72	0.85 b
2	48	0.51 c
1	48	0.50 c
3	48	0.50 c
2	24	0.30 d
3	24	0.21 e
1	24	0.19 e
Mea	n	0.55

The differences between the frequencies and times with respect to Least Sq. Mean were significant. With respect to the Table 4.32, Least Sq. Mean ranged between 0.19 - 0.97 percent. The average Least Sq. Mean of frequencies and times were 0.55 %. The highest Least Sq. Mean values were obtained from frequency 2 and time 72 h as 0.97 %, and the lowest Least Sq. Mean values were obtained from frequency 1 and time 24 h as 0.19 % (Table 4.32). It is observed the first three results, 72 h applications were the best result and very different from the other applications, while the last three results have the worst results at 24 h apps.

Utilizing leaves from pepper (*Capsicum annuum* L.) plants presented to low temperature 8 °C for various eras 1 to 3 d, and were examined, after 24 h of presentation at 8 °C, pepper plants displayed obvious indications portrayed by limpness of stems and takes off. This was joined by noteworthy changes in the digestion of RNS and ROS with an expansion of both protein tyrosine nitration (NO<sub>2</sub>-Tyr) and lipid peroxidation, demonstrating that low temperature initiates nitrosative and oxidative anxiety (Airaki et al., 2012) its results are close to our results, however, we used 3 frequencies and low temperatures (0 °C).

According to the findings of Al-Lishi et al. (2012) SA to the liquid media Potato Sucrose Broth (PSB) caused complete inhibition to the fungal growth at 3 and 5 mmol of SA applied in greenhouse experiments, seeds were soaked mmol of SA for 24 hrs. As a protective treatment, then planted in soil contaminated with the pathogen caused the significant reduction in percent infection and disease severity of leaf spots. Dual treatment by soaking the seeds with the same concentrations. Along with spraying the shoot system was the most efficient in reducing percent infection by 100 % and disease severity of the disease and improving growth characters of the treated plants. Application of SA is different from our test, while our use SA was sprayed on the pepper plant at the low temperature.

#### 4.5.5. Frequency and Doses (mmol)

Frequencies	Doses (mmol)	Least Sq. Mean %
2	Contr.	0.84 a
3	Contr.	0.73 b
1	Contr.	0.69 b
2	0,01	0.59 c
3	0,01	0.55 cd
1	0,01	0.52 d
1	0,05	0.45 e
2	0,05	0.35 f
3	0,05	0.28 g
М	ean	0.55

Table 4.33. Effect of interaction the frequencies and doses on rate of wilting rate

The differences between the frequencies and doses with respect to Least Sq. Mean were significant. According to the Table 4.33, Least Sq. Mean ranged between 0.28 – 0.84 percent. The average Least Sq. Mean of frequencies and doses were 0.55 %. The highest Least Sq. Mean values were obtained from frequency 2 and control dose as 0.84 %, and the lowest Least Sq. Mean values were obtained from frequency 3 and dose 0.05 mmol as 0.28 % (Table 4.33). At the point when watched the initial three outcomes, the concentration at (contr.) applications was the best outcome and altogether different from other applications, whereas the last three outcomes have fewer outcomes at the doses 0.05 mmol.

Sprayed the dill plant species local of SA at the concentration of 0, 20 and 40 mg/L and the concentration of 40 mg/L led to a significant increase in diameter and lenght and the proportion of dry matter in the stems and roots while spraying with a concentration of 20 mg/L led to significantly increase the pilot oil and composite corvon (Yasin, 2016). The results of this research are close to our results but we used a spray on dill plant while we used on the pepper plant.

Conchic et al. (2007) found that the treatment of melon seeds in the SA led to the induction development of systemic resistance the post-harvest diseases less analogy treatment comparison. The method of applying SA differs from our study method the

watermelon seeds were used to immolersion SA, while we using SA spray on the pepper plant.

### 4.5.6. Frequency, Doses (mmol) and Time (h)

The differences between the frequencies, doses and times with respect to Least Sq. Mean were significant. According to the Table 4.34, Least Sq. Mean ranged between 0.99 - 0.02 percent. The average Least Sq. Mean of frequencies and doses and times were 0.55 %. The highest Least Sq. Mean values were obtained from frequency 1 and control dose and time 72 h as 0.99 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and dose 0.05 mmol and time 24 h as 0.02 %, as in the lower table (Table 4.34). It is observed that the first sixth results, at each concentration twice and the time at 72 h and at each frequency, three repetitions, applications were the best results and very different from the other applications, while the last sixth results have the worst results at the concentration of them 0.05mmol.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
1	Contr.	72	0.99 a
2	Contr.	72	0.98 a
1	0.05	72	0.98 a
2	0.01	72	0.97 a
2	0.05	72	0.95 ab
1	0.01	72	0.94 ab
3	Contr.	72	0.90 ab
3	0.01	72	0.88 a-c
2	Contr.	48	0.85 b-c
3	Contr.	48	0.78 с-е
3	0.05	72	0.77 d-f
1	Contr.	48	0.71 e-g
2	Contr.	24	0.70 e-g
3	0.01	48	0.67 fg
2	0.01	48	0.62 gh
3	Contr.	24	0.52 hi
1	0.01	48	0.49 i
1	Contr.	24	0.38 j
1	0.05	48	0.31 j
2	0.01	24	0.17 k
1	0.01	24	0.13 kl
3	0.01	24	0.10 kl
2	0.05	48	0.07 kl
3	0.05	48	0.061
1	0.05	24	0.051
3	0.05	24	0.021
2	0.05	24	0.02 1
	Mean	1	0.55

Table 4.34. Effect of interaction the frequencies, doses and times on rate of wilting rate

Popova et al. (2008) concluded that SA may protect cells against oxidative damage and photosynthesis against (cadmium) Cd toxicity. Application of SA is different from our test when the seeds of maize were used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

Ghaleb et al. (2010) found that the use of salicylic acid in the agricultural amid of the Jujube plant, it has had a positive impact, where the highest rate for the survival of plants emerged surviving at the concentration of 10 mg/L salicylic acid, as well as a good response in the composition of branches and rooting at the same concentration. Application of SA is different from our test when amid of the Jujube plant was used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

### 4.5.7. Time (h) and Doses (mmol)

Times (h)	Doses (mmol)	Least Sq. Mean %
72	Contr.	0.96 a
72	0,01	0.93 a
72	0,05	0.90 a
48	Contr.	0.78 b
48	0,01	0.59 c
24	Contr.	0.53 c
48	0,05	0.15 d
24	0,01	0.13 d
24	0,05	0.03 e
M	ean	0.55

Table 4.35. Effect of interaction the times and doses on rate of wilting rate

The differences between the times and doses with respect to Least Sq. Mean were significant. With respect to the Table 4.35, Least Sq. Mean ranged between 0.03 - 0.96 percent. The average Least Sq. Mean of times and doses were 0.55 %. The highest Least Sq. Mean values were obtained from time 72 h and control dose as 0.96 %, and the lowest Least Sq. Mean values were obtained from time 24 h and dose 0.05 mmol as 0.03 % (Table 4.35). It has been observed that the first three results, 72 h applications and each the concentration of them were the best results and very different from the other applications, while the last three results have the worst results.

The content of ascorbate and glutathione (GSH) was studied in pepper leaves, to know how LT affects the status of non-enzymatic antioxidants. By the researcher (Airaki et al., 2012) and reached the results that the total ascorbate increased about 88 % and GSH increased about 35 % during the first day to LT compared in leaves exposed

with control stations, similar levels were maintained during the second and third days of LT treatment. These results, are likely the same as the results we have obtained.

The treatment soaks sesame seeds and for 2 hours in salicylic acid at a concentration of 4 mmol led to renewed resistance against rottenness coaly caused by a fungus (El-Fiki et al., 2004). Application of SA is different from our test when the seeds of sesame were used with SA, while our use SA was sprayed on the pepper plant.

Al-Shabbani et al. (2013) indicated effect the SA. Researchers have noted that the spraying of black cumin (*Nigella sativa* L.) plants of salicylic acid with concentrations of (12 and 20 ml/L) by two from sprays, results led to a significant increase in plant height and number of branches and leaves and the weight dry the total (vegetative and root), and the number of flowers and the weight of 1000 seeds. The results of her research were good and by two from sprays of the SA on black cumin while we used pepper and three sprays.

### 4.6. Cold Damage

### 4.6.1. Doses (mmol)

As shown in the lower table (Table 4.36) there are no significant between salicylic acid concentrations compared to the comparative control dose.

Doses (mmol)	Least Sq. Mean %
0.01	2 a
0.05	2 a
Contr.	2 a

Table 4.36. Effect of interaction the doses on rate of cold damage

It has been proven by a researcher (Tufail et al., 2013) on salicylic acid proven that it regulates many physiological processes of the plant under stress conditions such as photosynthesis, breathing, nutrient absorption, opening and closing of holes, and improves plant tolerance for salinity by increasing non-enzymatic antioxidants such as superoxide enzyme dismutase, catalase enzyme, and peroxidase enzyme.

The researcher concluded in his research that the concentration of 1 mmol of the SA protects the plant from damage with the increased oxidative enzyme in the

antioxidant activities and controls the physiological adaptation and a decrease in the level of lipid peroxide. This is what was concluded when the salicylic acid was applied at low temperatures in the test (Orabi et al., 2010).

## 4.6.2. Time (h)

As shown in the lower table (Table 4.37) there are no significant between the times used in the research.

Times (h)	Least Sq. Mean %
24	2 a
48	2 a
72	2 a

 Table 4.37. Effect of interaction the times on rate of cold damage

Results obtained by Airaki et al. (2012) when exposed pepper plant at low temperatures during 24-48 hours led to the production of a general imbalance of the ROS and RNS metabolism was produced which strongly suggests the induction of oxidative result of a rise in lipid peroxidation and protein tyrosine nitration.

#### 4.6.3. Frequency

As shown in the lower table (Table 4.38) there are no significant between the frequencies used in the research.

Frequencies	Least Sq. Mean %
1	2 a
2	2 a
3	2 a

Table 4.38. Effect of interaction the frequencies on rate of cold damage

## 4.6.4. Frequency and Time (h)

As shown in the lower table (Table 4.39) there are no significant between the interaction of the frequencies and times used in the research.

Frequencies	Times (h)	Least Sq. Mean %
1	24	2 a
1	48	2 a
1	72	2 a
2	24	2 a
2	48	2 a
2	72	2 a
3	24	2 a
3	48	2 a
3	72	2 a

Table 4.39. Effect of interaction the frequencies and times on rate of cold damage

Miura and Tada (2014) concluded SA mitigate the affectability to abiotic stresses, and high concentrations of connected actuate elevated amounts of oxidative pressure. This is what layer on the test to withstand LT at the spray SA on the pepper plant.

# 4.6.5. Frequency and Doses (mmol)

As shown in the lower table (Table 4.40) there are no significant between the interaction of the frequencies and doses used in the research.

Frequencies x	Doses (mmol)	Least Sq. Mean %
1	0,01	2 a
1	0,05	2 a
1	Contr.	2 a
2	0,01	2 a
2	0,05	2 a
2	Contr.	2 a
3	0,01	2 a
3	0,05	2 a
3	Contr.	2 a

Table 4.40. Effect of interaction the frequencies and doses on rate of lost seedling cold damage

The SA has important physiological roles such as increasing the plant's ability to bearing the stresses resulting from high temperature and low temperatures (Senaratna et al., 2000). These results what we reached in the test at the low temperature.

Khan et al. (2015) concluded that phytohormones have been perceived as a strong device for economically lightening unfavorable impacts of abiotic worries in crop plants. In this research, the importance of salicylic acid SA has been increasingly defined in improving plant abiotic stress-tolerance via stress by means of SA-mediated control of the main metabolic plant processes. This is what we applied to our study on the pepper plant at the low temperature.

# 4.6.6. Frequency, Doses (mmol) and Time (h)

As shown in the lower table (Table 4.41) there are no significant between the interaction of the frequencies, doses and times used in the research.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
1	0.01	24	2 a
1	0.01	48	2 a
1	0.01	72	2 a
1	0.05	24	2 a
1	0.05	48	2 a
1	0.05	72	2 a
1	Contr.	24	2 a
1	Contr.	48	2 a
1	Contr.	72	2 a
2	0.01	24	2 a
2	0.01	48	2 a
2	0.01	72	2 a
2	0.05	24	2 a
2	0.05	48	2 a
2	0.05	72	2 a
2	Contr.	24	2 a
2	Contr.	48	2 a
2	Contr.	72	2 a
3	0.01	24	2 a
3	0.01	48	2 a
3	0.01	72	2 a
3	0.05	24	2 a
3	0.05	48	2 a
3	0.05	72	2 a
3	Contr.	24	2 a
3	Contr.	48	2 a
3	Contr.	72	2 a

Table 4.41. Effect of interaction the frequencies, doses and times on rate of cold damage

The spring and the winter wheat were consistently used in aqueous solution for salicylic acid identical permanently due to low temperature (Horvath et al., 2007). These results are similar to the results obtained by in the test but the salicylic acid spray was used on the wheat crop while we used SA spray on the pepper plant.

Horváth et al. (2007) found that using of exogenous SA could give insurance against a few sorts of stresses, for example, high or low temperature, heavy metals. The

impact of exogenous SA relies on various factors, for example, the species and formative phase of the plant, the method of use. Recent results show that not only does exogenous SA application moderate stress effects, yet abiotic stretch elements may likewise change the endogenous SA levels in the plant cells. These results were applied to the test at low temperatures.

### 4.6.7. Time (h) and Doses (mmol)

As shown in the lower table (Table 4.42) there are no significant between the interaction of the times and doses used in the research.

Times (h)	Doses (mmol)	Least Sq. Mean %
24	0,01	2 a
24	0,05	2 a
24	Contr.	2 a
48	0,01	2 a
48	0,05	2 a
48	Contr.	2 a
72	0,01	2 a
72	0,05	2 a
72	Contr.	2 a

Table 4.42. Effect of interaction the times and doses on rate of cold damage

Rivas-San and Plasencia (2011) indicated that SA plays a role during plant response to non-biological environments such as dehydration and cooling. This is what we found in the test.

It has been recommolended that SA has the immolense agronomic potential to enhance the pressure resistance of horticultural imperative yields, the method of use and the condition of the plants (Miura and Tada, 2014). This is what layer on the test to withstand low temperature at the spray SA on the pepper plant.

#### 4.7. Progress Rate2

#### 4.7.1. Doses (mmol)

The differences between the doses with respect to Least Sq. Mean were significant. According to the Table 4.43, Least Sq. Mean ranged between 0.49 - 0.60 percent. The average Least Sq. Mean of doses were 0.56 %. The highest Least Sq. Mean values were obtained from dose 0.01 mmol as 0.60 %, and the lowest Least Sq. Mean values were obtained from control dose as 0.49 %, as in the lower table (Table 4.43).

Doses (mmol)	Least Sq. Mean %
0.01	0.60 a
0.05	0.60 a
Contr.	0.49 b
Mean	0.56

Table 4.43. Effect of interaction the doses on rate of progress rate2

The concentration of 0.5 mmol SA would prefer to use cold tolerance for all of the cucumber, corn, and rice preferred (Kang and Saltveit, 2002). These results are consistent with our results.

In the study of salicylic acid prevents the synthesis of protein inhibitors in tomato leaves caused by system and JA, by Airaki et al. (2012) and explained that strong inhibitors of acid-induced, and explained that the inhibition by the salicylic acid of proteinase inhibitor synthesis induced by system and JA. The researcher used the tomato leaves while we used pepper.

Spraying the coriander plants with salicylic acid at a concentration of 0.01mmol has caused a significant increase in the number of inflorescences flowering, which led to a significant increase in the number of seeds plant and seed yield compared to those plants that were not sprayed (Hesami et al., 2013). The researcher used spray salicylic acid on coriander plants while we used to plant pepper.

## 4.7.2. Time (h)

Times	Least Sq. Mean %
24	0.88 a
48	0.80 b
72	0.01 c
Mean	0.56

#### Table 4.44. Effect of interaction the times on rate of progress rate2

The differences between the times with respect to Least Sq. Mean were significant. With respect to the Table 4.44, Least Sq. Mean ranged between 0.01 - 0.88 percent. The average Least Sq. Mean of times were 0.56 %. The highest Least Sq. Mean values were obtained from time 24 h as 0.88 %, and the lowest Least Sq. Mean values were obtained from time 72 h as 0.01 % (Table 4.44).

Jalal et al. (2012) pointed out that it was content in the leaf extracts from of 30day-old pepper plants exposed LT 8°C for 1-3 days, at catalase and superoxide dismutase (SOD) activities. And the variation from the control values was significant at (P > 0.05). The results of this research were similar to the results we obtained in our study.

Popova et al. (2008) concluded that Pre-treatment of seeds by SA reduced the negative effect of cadmium (Cd) on plant growth factors. Application of SA is different from our test when the seeds of maize were used with SA, while our use SA was sprayed on the pepper plant at the low temperature.

### 4.7.3. Frequency

Frequencies	Least Sq. Mean %
2	0.59 a
3	0.55 b
1	0.54 b
Mean	0.56

Table 4.45. Effect of interaction the frequencies on rate of progress rate2

The differences between the frequencies with respect to Least Sq. Mean were significant. According to the Table 4.45, Least Sq. Mean ranged between 0.54 - 0.59

percent. The average Least Sq. Mean of frequencies were 0.56 %. The highest Least Sq. Mean values were obtained from frequency 2 as 0.59 %, and the lowest Least Sq. Mean values were obtained from frequency 1 as 0.54 % (Table 4.45).

SA is one of the substances similar to hormone and its role is important in the direction the physiological process like ion uptake and transport, stress tolerance as well as membrane permeability and photosynthesis (Noreen et al., 2009). This is what we applied to our tests that salicylic acid has an effect on the tolerance of pepper plants to low temperatures.

Conchic et al. (2007) found that the treatment of melon seeds in the salicylic acid led to the induction development of systemic resistance by increasing enzymes (peroxidases and chitinases). The method of applying SA differs from our study method the watermelon seeds were used to immolersion SA, while we using SA spray on the pepper plant.

Frequencies	Times (h)	Least Sq. Mean %
2	24	0.90 a
3	24	0.90 a
2	48	0.87 a
1	24	0.85 a
1	48	0.78 b
3	48	0.75 b
3	72	0.01 c
1	72	0.01 c
2	72	0.01 c
Me	an	0.56

#### 4.7.4. Frequency and Time (h)

Table 4.46. Effect of interaction the frequencies and times on rate of progress rate2

The differences between the frequencies and times with respect to Least Sq. Mean were significant. According to the Table 4.46, Least Sq. Mean ranged between 0.01 - 0.90 percent. The average Least Sq. Mean of frequencies and times were 0.56 %. The highest Least Sq. Mean values were obtained from frequency 2 and time 24 h as 0.90 %, and the lowest Least Sq. Mean values were obtained from frequency 2 and time 72 h as 0.01 % (Table 4.46). It is observed that the first three results, 24 h applications were the

best result and very different from the other applications, whereas the last three results have the worst results, 72 h applications.

The SA is an endogenous development controller of phenolic nature and plays an important role in abiotic push resilience by expanding the activity of antioxidant enzymes and detecting excess ROS, which brought about improving the physiological processing and upgrading of plant development (He and Zhu, 2008). This is what we have reached in our results we tested.

Frequencies	Doses (mmol)	Least Sq. Mean %
2	0,05	0.65 a
2	0,01	0.62 ab
1	0,01	0.60 ab
3	0,05	0.59 b
3	0,01	0.58 b
1	0,05	0.56 bc
2	Contr.	0.51 cd
3	Contr.	0.49 d
1	Contr.	0.47 d
Me	ean	0.56

#### 4.7.5. Frequency and Doses (mmol)

Table 4.47. Effect of interaction the frequencies and doses on rate of progress rate2

The differences between the frequencies and doses with respect to Least Sq. Mean were significant. With respect to the Table 4.47, Least Sq. Mean ranged between 0.47 - 0.65 percent. The average Least Sq. Mean of frequencies and doses were 0.56 %. The highest Least Sq. Mean values were obtained from frequency 2 and dose 0.05 mmol as 0.65 %, and the lowest Least Sq. Mean values were obtained from frequency 1 and control dose as 0.28 % (Table 4.47). At the point when watched the initial three outcomes, at the first concentration at 0.05 mmol and also the doses second and three at 0.01 mmol applications was the best outcome and altogether different from other applications, whereas the last three outcomes have fewer outcomes at the control dose.

Hara et al. (2012) concluded that high concentrations of salicylic acid may cause cell death or exposure to abiotic stresses, in general, may lead low concentrations of SA may enhance antioxidant capacity in plants. SA has direct involvement in plant growth, thermogenesis, flower induction and uptake of ions, and affects ethylene biosynthesis, stomata movement (Hayat et al., 2007). These results for the thermogenesis we applied in the test to bear the pepper plant at low temperatures.

# 4.7.6. Frequency, Doses (mmol) and Time (h)

The differences between the frequencies, doses and times with respect to Least Sq. Mean were significant. According to the Table 4.48, Least Sq. Mean ranged between 0.01– 0.99 percent. The average Least Sq. Mean of frequencies and doses and times were 0.59 %. The highest Least Sq. Mean values were obtained from frequency 2 and dose 0.05 mmol and time 48 h as 0.99 %, and the lowest Least Sq. Mean values were obtained from frequency 1 and control dose and time 72 h as 0.01 %, as in the lower table (Table 4.48). It has been observed that the first sixth results, at each concentration twice and the most time at 24 h and at the frequency 2 and 3their three repetitions, applications were the best results and very different from the other applications, at a time the last sixth results have the worst results and at the time of them 72 h.

Frequencies	Doses (mmol)	Times (h)	Least Sq. Mean %
2	0,05	48	0.99 a
2	0,05	24	0.95 ab
3	0,01	24	0.95 ab
3	0,05	24	0.93 а-с
2	0,01	24	0.93 a-c
2	0,01	48	0.92 a-d
1	0,01	24	0.90 a-e
1	0,01	48	0.88 b-f
1	0,05	24	0.85 b-f
1	0,05	48	0.83 c-f
2	Contr.	24	0.82 d-f
3	0,05	48	0.81 d-f
3	Contr.	24	0.81 ef
3	0,01	48	0.79 fg
1	Contr.	24	0.78 fg
2	Contr.	48	0.69 gh
3	Contr.	48	0.64 h
1	Contr.	48	0.62 h
1	0,01	72	0.01 i
3	0,01	72	0.01 i
2	0,01	72	0.01 i
2	0,05	72	0.01 i
3	Contr.	72	0.01 i
1	0,05	72	0.01 i
3	0,05	72	0.01 i
2	Contr.	72	0.01 i
1	Contr.	72	0.01 i
	Mean	1	0.59

Table 4.48. Effect of interaction the frequencies, doses and times on rate of progress rate2

Sprayed plants salicylic acid to ginger plants (*Zingiber officinale* Roscoe), led to a significant increase in the content of the papers of total chlorophyll was compared with plants which did not sprinkle (Ali and Jaafar, 2013).

Fariduddin et al. (2003) concluded that SA had a significant effect on the ratio of chlorophyll and anthocyanin of the petal, nitrogen, potassium, and phosphorus, Plant

production was increasing at the 50 and 100 ppm, compared to 0 and 150 ppm of salicylic acid. It is therefore important to use salicylic acid in the rose plant production Pre-harvest to improve the quality of rose. Application of SA is different from our test when the plant of roses was used with SA.

### 4.7.7. Time (h) and Doses (mmol)

Doses (mmol)	Times (h)	Least Sq. Mean %
24	0,01	0.93 a
24	0,05	0.91 ab
48	0,05	0.88 ab
48	0,01	0.86 bc
24	Contr.	0.80 c
48	Contr.	0.65 d
72	0,01	0.01 e
72	0,05	0.01 e
72	Contr.	0.01 e
Mea	n	0.56

Table 4.49. Effect of interaction the times and doses on rate of progress rate2

The differences between the times and doses with respect to Least Sq. Mean were significant. According to the Table 4.49, Least Sq. Mean ranged between 0.01 –0.93 percent. The average Least Sq. Mean of times and doses were 0.56 %. The highest Least Sq. Mean values were obtained from time 24 h and dose 0.01 mmol as 0.93 %, and the lowest Least Sq. Mean values were obtained from time 72 h and control dose as 0.01 % (Table 4.49). It is observed that the first two results, 24 h applications and each the concentration of them were the best results and very different from the other applications, while the last three results have the worst results, at the time 72 h from the applications.

Content in the leaf extracts from pepper plants exposed low-temperature 8 °C for 1-3 days, to show the contents of the ascorbate and GSH at the total. The results showed increased activity of the enzyme catalase from 34 % to 36 % during the first two days and after that, it was not significantly affected by low temperatures (Airaki et al., 2012).

When you notice the results of this search, it appears clearly that it is almost similar to the results obtained in our test.

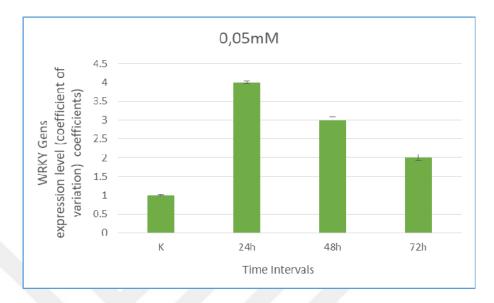
Aziz et al. (2015) investigated on the effect of spraying sour orange seedlings at the SA concentration of 50-100 mg/L twice between them a time interval of one month in some physiological traits of seedlings. At the seedling treatment concentration of 100 mg/L of the SA it has outperformed other transactions in height seedling and the number of its side branches, the diameter of its main stem, the average number of leaves, the soft and dry weight of its leaves, and the content of leaves of total chlorophyll. These results are close to our results, but spray orange seedlings at the salicylic acid twice while we used three sprays on the pepper plant.

SA is an intrinsic plant growth regulator that constitutes the corresponding response in many growth-development pathways in plants and thus affects plant growth and development (Hayat et al., 2010). Figure 4.1 and Figure 4.2 show how the changes in the gene level depend on the time of cold stress in salicylic acid applied pepper genotypes. As shown in Figure 1, leaf samples were taken after 24, 48 and 72 hours to measure the expression level of WRKY gene.

As a result of the analysis, it was found that the expression level of WRKY gene increased with the application of salicylic acid. This increase was highest in the first 24 hours exposed to cold stress, while gene expression decreased gradually in 48 and 72 hours (Figure 4.1). In many more studies, the expression of WRKY gene was found to be increased in the presence of salicylic acid. For example, in a study conducted on Arabidopsis, it was observed that the expression level of 49 of the 72 WRKY genes tested in salicylic samples increased (Dong et al., 2003). WRKY proteins are a group of transcription factors that appear as a gene superfamily in plants and play important roles in the regulation of defense response pathways in plants. WRKY proteins also play a role in other plant-specific functions such as trichome development and biosynthesis of secondary metabolites (Ramiro and Jalloul, 2010).

As shown in (Figure 4.1 and 4.2) leaf samples were taken after 24, 48 and 72 hours to measure the expression level of the WRKY gene.

As a result of the analysis, it was found that the expression level of WRKY gene increased with the application of salicylic acid.



**Figure 4.1.** Determination of the level of expression of WRKY gene in real-time polymerase chain reaction (PCR) at 0.05 mmol salicylic pepper.

This increase was highest for the first 24 hours exposed to cold stress, while gene expression was gradually decreased at 48 and 72 hours, but this decrease was not very sharp compared to 0.05 mmol application. Determined expression level of WRKY genes is close to each other.

Considering the administration of both salicylic acids, it was found that the pepper plant has a clearer and more understanding response to the expression of WRKY gene in the application of 0.05 mmol SA (Figure 4.1).

In Arabidopsis, the onset of Systemic acquired resistance (SAR) is initiated by the accumulation of the signal molecule, salicylic acid (SA).

Salicylic acid promotes nuclear translocation of transcription cofactor regulatory protein NPR1 (Non-expression of pathogenesis-related genes1) to activate most genes needed for disease resistance (Wang et al., 2006; Kinkema et al., 2000).

The expression of regulatory protein NPR1 has been shown to be under the regulation of WRKY transcription factors (Yu et al., 2001).

In another study, the WRKY59 gene was identified to be the direct target of SAdependent defense and NPR1 (Wang et al., 2006). WRKY59 also plays an important role in sustainable defense (Eulgem, 2005).



Figure 4.2. Determination of the level of expression of WRKY gene in real-time (PCR) at 0.01 mmol salicylic pepper.

### 5. CONCLUSIONS AND RECOMMOLENDATIONS

### 5.1. Conclusion

The results of this study in which the effects of salicylic acid (SA) applied to pepper seedlings grown in low temperature (0 °C) conditions can be summolarized as follows;

Three different doses of SA (0 mmol, 0.01 mmol and 0.05 mmol) were given from the leaf. Each dose was administered in three different frequencies (1 time, 2 times and 3 times spraying). Each application was exposed to cold (0 °C) in three different times (24 h, 48 h and 72 h).

The best dose of SA was 0.01 mmol in the rate of last seedling weight measurements, the worst result was obtained from the control group without SA treatment. The other two (1 and 2 times) frequency applications applied to the seedlings exposed to low temperatures were the highest in the rate of seedling weight. Rate of lost seedling lenght data, the best dose of 0.01 mmol dose, the most appropriate frequency of application 2 and 3 times were found to be.

Both dosing and SA applications with different frequencies and the exposure to cold during different time periods did not make any difference in protecting turgorit and cold Damage measurements of pepper seedlings.

When the rate of progressive seedlings expressing the proportion of the seedlings living in the cold application is examined, it is seen that the dose of 0.05 mmol SA dose and the application of the application three times as a dose give the best results. The control group, which had no SA administration, had the highest values in the wilting rate measurements. Among the progress rate2 data, which expressed the rates of continued improvement following the discontinuation of cold, both of the SA doses were in the best group. Two-time application was also the best frequency group.

Furthermore, when the application of 0.01 mmol and 0.05 mmol SA was evaluated for the expression of the WRKY gene of the pepper plant, it was found that the application of 0.05 mmol SA yielded very clear and observable results.

### 5.2. Recommolendation

Nowadays, as climate change accelerates and its impact is gradually increasing, researches on abiotic stresses are becoming increasingly important. The use of plant internal growth regulators that provide resistance or tolerance to low temperatures and the development of existing ones are more important. Vegetables grown in seasons without sufficient temperature values are expected to be low temperature resistant or tolerant, and both tolerant and resistant to the use of regulators that run WRKY proteins such as salicylic acid appear to be the right way to produce products naturally. According to this study, we may recommolend the use of 0.05 mmol salicylic acid.

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#### **APPENDICES**

### RESUME

#### PERSONAL INFORMATION

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# Education

Doktora	:	
Master Degree	: Siirt Unversity/ Faculty of Agriculture	
University	: Dohuk Unversity/ Faculty of Agriculture	2003
High School	: Brayety School	1999
Degree	: Name, Town, Province	Graduated

## Work Experience

Year	Institution	Mission

Area of Professional	: Agricultural Engineer – Duhok Polytechnic University
Foreign Languages	: Kurdish, Arabic and English
The Other Features	: Agriculture (Horticulture, Vegetables and Ornamental plants), Beekeepers, Cultivation under greenhouses, Designs and installation of furniture, Political.
PUBLICATIONS	:

