

THE EFFECT OF FOLIAR SPRAY OF SALICYLIC ACID AND POTASSIUM SULPHATE ON BIOCHEMICAL CHARACTERISTICS OF GRAPE BUD IN DORMANT PERIOD

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Abstract. In order to study the biochemical changes, increase of resistance to cold stress and possibility of reducing primary buds death of grape, an experiment was conducted in a factorial design based on randomized complete block design with three replications and two vines per plot in the city of Dena in 2014. The first factor was salicylic acid (SA) at concentrations of 0, 150, 300 and 450 mg/l, and the second factor was potassium sulphate (K_2SO_4) at the levels of 0, 1, and 2%. Sampling was performed after leaves drop and start of dormancy, endodormancy stage, and before the growth of buds in the following year, and the related characteristics were recorded in each stage. The results showed that different levels of SA, dates of sampling, interaction of SA and sampling date, interaction of SA and potassium sulphate, and interaction of potassium sulphate and sampling date had significant effect on Malondialdehyde at the level of 1%. Furthermore, Malondialdehyde was affected by interaction of SA and potassium sulphate, and interaction of potassium sulphate and sampling date at probability level of 5%. The results also showed that SA at different levels had significant effect on catalase enzyme at 1%. Interactions between SA and potassium sulphate, and between SA and sampling date caused a significant effect on the amount of peroxidase at 5 and 1% levels, respectively. Moreover, superoxide dismutase was affected significantly by sampling date and SA at the level of 5%.

The effects of different levels of potassium sulphate, dates of sampling, and interaction of sampling date and SA on relative water content were significant at 1%, while interaction of SA, potassium sulphate and sampling date had significant effect at 5%. The effects of different levels of potassium sulphate, interaction of potassium sulphate, SA and sampling date on electrolyte leakage were significant at 5%, while at the level of 1%, just the effect of sampling date was significant. Sampling date and interaction of SA and sampling date had significant effect on the amount of antioxidant activity at 1%.

Keywords: catalase, peroxidase, superoxide dismutase, antioxidant activity, electrolyte leakage, malondialdehyde, relative water content, grape.

1. INTRODUCTION

Cultivation area of grape in Yasouj province, especially Dena county, is rising due to the desire of farmers and economical value of this crop as one of the main sources of income of the people living in this province. In the meantime, winter cold annually causes severe damage to the vines of the the region, and places significant limitations on the growing of vine. Reduction of yield is the result of damage to the primary bud tissue and growth of weak lateral buds. Growth of secondary bud, production of thin shoots, increase of cold damage, and reduction of yield are the main problems of growing Askari grape in the city of Dena. Pretreatment of grape with SA reduced the amount of malondialdehyde (MDA) and other aldehydes under weather conditions (Wang et al., 2005). It is reported that free radicals increase in the atmosphere under stress and cause damage to cell membrane, because the amounts of MDA and electrolyte leakage of the membrane increase (EL-Tayeb, 2005). Salicylic acid is a well-known endogenous growth regulator affecting various physiological functions and biochemical activities in plants. As a molecular signal, SA plays an important role in creating a defensive response against various biotic and abiotic stresses (Arfan et al., 2007; Wang et al., 2010). Plant physiological processes, growth, development, fertility, and response to abiotic stress are affected by SA (Arfan et al., 2007). Salicylic acid changes the activity of antioxidant enzymes, which play an important role in the protection of plants against oxidative damage by detoxification of strong oxidizing radicals (Munns and Tester, 2008). High activity of antioxidant enzymes improves plant resistance to oxidative damage caused by reactive oxygen species (Gapinska et al., 2008). It was reported that relative water content (RWC) in chrysanthemum increased by foliar application of SA (Vahdati et al., 2012). The interaction among SA, proline and soluble sugar accumulation, and increase in RWC

can be attributed to osmotic regulation by accumulation of proline and soluble sugars. It was reported that treatment of SA caused a significant increase in soluble sugar and proline under drought stress by osmotic regulation (Demiralay et al., 2013). The effect of potassium ion on the increase of yield and cold tolerance was studied by Devi et al. in 2012. It was shown in Panax ginseng tree that high concentration of potassium ion activated antioxidant system and increased transcriptional levels of secondary metabolites related to ginsenoside, which is associated with cold tolerance. Cold stress may disrupt the photosynthesis and reduce the effect of antioxidant enzymes, resulting in the accumulation of ROS (reactive oxygen species production) such as superoxide radicals (O_2^-), hydrogen peroxide (H_2O_2), hydroxyl radicals (OH^-) (Mittler, 2002; Xiong et al., 2002; Suzuki and Mittler, 2006). It has been reported that potassium increased survival of plants under cold stress by increasing antioxidant levels and reducing the production of ROS (Devi et al., 2012; Cakmak, 2005). Foliar application of SA and potassium sulphate can result in the increase of photosynthesis in the autumn, before harvest and the onset of dormancy, increase of soluble and insoluble carbohydrates, enhancement of resistance to environmental stresses such as cold stress, reduction of bud necrosis, and increase of yield (quantitative and qualitative characteristics). The aim of the present research was to study biochemical changes, increase of cold hardiness and the possibility of reducing primary bud death of grape using SA and potassium sulphate foliar spray.

2. MATERIALS AND METHODS

This experiment was performed in a vineyard in Sisakht city (545946-3414228 UTM) to investigate biochemical changes and cold resistance (by measuring the intended characteristics). At the end of August 2014, leaf and soil samples were prepared from the vineyard and were transferred to

the laboratory for the subsequent analysis. Based on the results obtained from the analysis of the samples, fertilizing nutrition was performed during winter. The experiment was conducted in a factorial based on randomized complete block design with three replications and two vines per plot in a 15-year-old vineyard in 2014. The effects of SA at 4 concentrations (0, 150, 300 and 450 mg/l) and sulphate potassium at three levels (0, 2 and 3%) were investigated. Biochemical characteristics were measured at three stages: after leaves drop and the onset of dormancy, endodormancy stage, before the buds growth in the following year. Statistical analysis was performed by using software MSTATc, and statistical design was a split plot in time.

2.1 Antioxidants

The extract of 100 mg buds was prepared. To separate insoluble materials, the extract was centrifuged for 5 min at 3500 rpm. An appropriate amount of transparent solution was mixed with 800 µl half-mM DPPH solution, and the rate of light absorbance was measured at the wavelength of 517 nm after the maintenance of the samples in the dark for 30 min. Besides, light absorbance of control samples was measured (Abe et al. 1998).

$$\text{Antioxidant activity (\%)} = \left\{ \frac{\text{control-sample}}{\text{control}} \right\} \times 100$$

2.2 Enzymes extraction

In order to measure catalase(cat), peroxidase(pod) and superoxide dismutase(sod) enzymes, extraction was performed according to the method described. 100 mg buds powdered in liquid nitrogen were homogenized with 1000 µl of very cold potassium phosphate buffer (pH = 7/8, 50 mM) containing 50 mM EDTA, and the homogeneous solution was centrifuged for 20 min at 130000 rpm and 4 °C. The clear supernatant obtained was used to determine enzyme activity.

2.3 The assay of superoxide dismutase(sod) enzyme

The reaction mixture containing potassium phosphate buffer (pH=7.5, 50 mM), 0.1 mM EDTA, 75 µmol nitroblue tetrazolium (NBT), 13 mM methionine, 4 mM riboflavin (the last additive), and plant extract. Superoxide dismutase enzyme activity was assessed by using NBT photoreduction assay at the wavelength of 560 nm (Beauchamp and Fridovich 1971).

2.4 The assay of catalase enzyme activity

1 ml reaction mixture of catalase enzyme assay contained potassium phosphate buffer (pH=7, 10 mM), 33 mM hydrogen peroxide and the enzyme extract. Reduction of hydrogen peroxide was recorded every 5 s by reducing the OD at 240 nm for 60 s. In the present experiment, the activity of the enzyme was estimated from the amount of H₂O₂ changes. Each µmol hydrogen peroxide destroyed in a minute is equal to one unit of enzyme activity. The extinction coefficient in this reaction was 40 mM.cm⁻¹ (Velikova et al, 2000).

2.5 Peroxidase enzyme assay

The reaction mixture included potassium phosphate buffer (pH=6, 20 mM), 1 mmol H₂O₂ and 5 µmol guaiacol, which was combined with 20 µl plant extract and 10 µl hydrogen peroxide, and light absorbance was recorded every 5 s at the wavelength of 490 nm for 60 s. The slope of the graph obtained represents the amount of activity and the presence of enzyme in the sample. In this reaction, guaiacol is converted into tetraguaiacol. The extinction coefficient in this reaction was considered 26/6 mM.cm⁻¹ (Srinivas et al. 1999).

2.6 Electrolyte leakage (EL)

Firstly, bud sections were washed in distilled water. Then, the buds were shaken in 25 ml distilled water in a bottle for 18 hr by a shaker, and electrical conductivity of the samples (EC₁) was measured. Next, the bottles were autoclaved for 15 min at 121 °C. After cooling, electrical conductivity (EC₂) was measured again. Finally, EL was calculated based on the following equation (the modified Marcum method, 1998).

$$\text{EL(\%)} = (E_1/E_2) \times 100$$

2.7 Malondialdehyde (MDA)

Extract of 2.0 g bud tissue was centrifuged at 10000 rpm for 5 hr. 4 ml of 20% TCA solution containing 0.5% thio-barbituric acid (TBA) added to 1 ml of the supernatant obtained from the centrifugation. The obtained mixture was placed in hot bath at 95 °C for 30 min and immediately cooled in ice, and the mixture was centrifuged again for 10 min at 10000 rpm. Light absorbance of the solution was measured at the wavelength of 532 nm using spectrophotometer. The compound for absorption in this wavelength was red complex (MDA-TBA). Absorbance of other non-specific pigments was determined at the wavelength of 600 nm and deducted from this value. To calculate MDA

concentration, the extinction coefficient of 155 mM.cm⁻¹ was used (Heath and Packer 1968).

$$\text{MDA}(\%) = \frac{(\text{OD}_{532} - \text{OD}_{600}) \times 1000}{(155 \times 0.2)}$$

2.8 Relative water content(RWC)

To calculate RWC, the buds were first weighted (FW). The weight of samples was then measured after 4 hr of soaking in water (TW). The samples were dried at 75 ° C for 48 hr and weighted (DW). Relative water content of the buds was calculated based on the following formula (Barrs and Weaterley 1962).

$$\text{RWC}(\%) = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100$$

3. RESULTS AND DISCUSSION

Grape buds were taken separately in three stages (onset of dormancy, endodormancy and before breaking bud dormancy) for each treatment. The major characteristics including malondialdehyde, catalase, peroxidase, superoxide dismutase, relative water content, electrolyte leakage and antioxidant activity were measured at each stage. Each trait was analyzed by using statistical split-plot design in time. The results of the analysis of variance related to the measured traits in grape buds in three stages are shown in Table 1.

Table 1. ANOVA results of experimental factors on different char

Factor	DF	Mean squar						
		MDA	CAT	POD	SOD	RWC	EL	Antioxidant
Block	2	0.106 ^{ns}	0.056 ^{ns}	0.829 ^{ns}	0.216 [*]	3.025 ^{**}	0.940	0.075 ^{ns}
SA	3	1.628 ^{**}	1.294 ^{**}	0.145 ^{ns}	0.131 [*]	0.415 ^{ns}	1.702	0.021 ^{ns}
K	2	0.030 ^{ns}	0.095 ^{ns}	0.038 ^{ns}	0.023 ^{ns}	2.976 ^{**}	3.195	0.048 ^{ns}
SA×K	6	0.613 [*]	0.450 ^{ns}	0.734 [*]	0.086 ^{ns}	0.369 ^{ns}	2.374	0.049 ^{ns}
Error	22	0.170	0.245	0.265	0.039	0.304	0.877	0.031
Date	2	8.968 ^{**}	0.117 ^{ns}	0.290 ^{ns}	0.343 [*]	6.982 ^{**}	37.327	0.187 [*]
SA×Date	6	2.125 ^{**}	0.349 ^{ns}	0.575 ^{**}	0.150 ^{ns}	1.757 ^{**}	0.960	0.084 ^{**}
K×Date	4	0.533 [*]	0.162 ^{ns}	0.181 ^{ns}	0.042 ^{ns}	0.071 ^{ns}	3.610	0.037 ^{ns}
SA×K×Date	12	0.525 ^{**}	0.191 ^{ns}	0.269 ^{ns}	0.142 ^{ns}	0.608 [*]	2.177	0.022 ^{ns}
Error	48	0.201	0.252	0.168	0.098	0.287	1.687	0.020
Cv(%)		15.83	20.06	14.33	25.15	7.06	29.21	1.62

ns: Difference is not significant, *: Difference is significant at five percent, **: Difference is significant at one percent

Potassium sulphate at 1% had the greatest effect on the reduction of MDA average level in the three sampling dates. Increase in the percentage of potassium sulphate in the second sampling date led to the decrease in MDA concentration, so that 2% potassium sulphate used in the second sampling

Malondialdehyde (MDA): Based on the variance analysis (Table 1), different levels of SA, date of sampling, SA interaction with sampling date, SA interaction with potassium sulphate and date sampling had signif

icant effect on the level of MDA at 1%. The interaction of SA and potassium sulphate, potassium sulphate and sampling date had a significant effect on MDA at probability level of 5%. Potassium sulphate had no significant effect on the level of MDA (Table 1). The average of MDA concentration decreased along with the increase in the concentration of SA. Increasing the concentration of SA from 150 to 450 mg/l decreased MDA level in the first and second sampling dates but not in the third sampling date, in which MDA enhanced along with the increase in SA concentration. Salicylic acid at concentration of 450 mg/l caused the greatest effect on reducing the average concentration of MDA in the first, second and third sampling dates. Concentration of MDA was significantly affected by the date of sampling, and the second sampling date had greater effect on MDA than the first and third sampling dates (Figure 1).

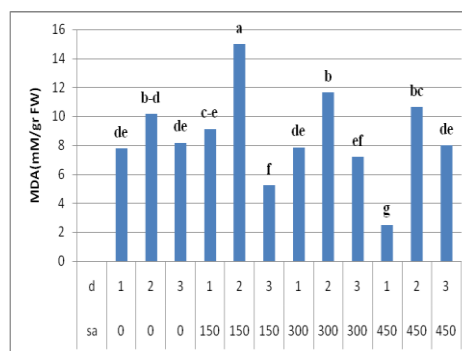


Figure 1. Effect of salicylic acid and sampling date on the amount of malondialdehyde (mM / gr FW)

date had the greatest effect on MDA reduction. Malondialdehyde production in the second sampling date, which was at endodormancy stage, was greater than the first and third sampling dates (Figure 2).

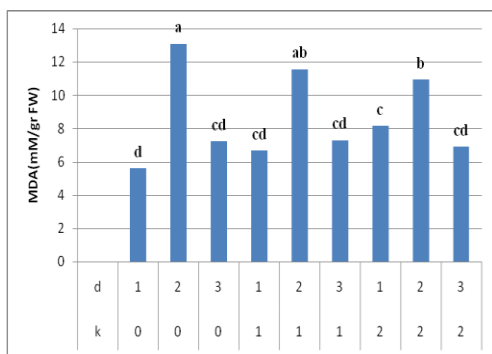


Figure 2. Effect of potassium sulphate and sampling date on malondialdehyde (mM / gr FW)

Evaluation of the means comparison related to the interaction of SA and potassium sulphate showed that increasing the concentration of SA from 150 to 450 mg/l decreased the amount of MDA, and the lowest MDA content was observed in the treatment

containing 450 mg SA and different levels of potassium sulphate. SA at low concentrations caused an increase in MDA content, while at the concentration of 450 mg/l, it reduced MDA (Figure 3).

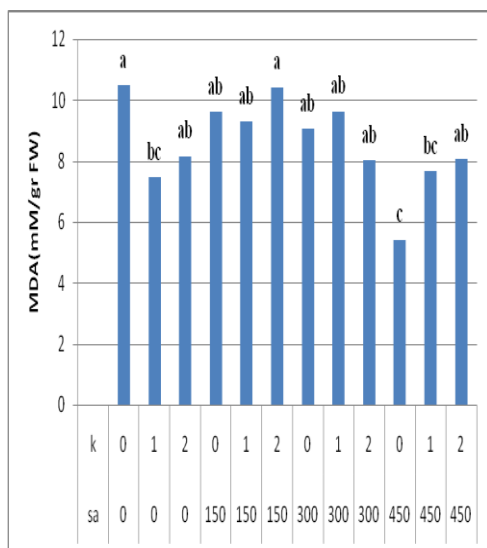


Figure 3. The effect of salicylic acid and potassium sulphate on malondialdehyde (mM/gr FW)

These results are in agreement with the previous studies. In grape, pretreatment with different levels of SA reduced MDA under environmental conditions (Wang et al., 2005). According to the results of this experiment, MDA decreased along with the increase in the concentrations of SA and potassium sulphate. It was reported that free radicals in the atmosphere increased under stress and caused damage to cell membrane due the increase in the amount of MDA and electrolyte leakage (EL-Tayeb, 2005).

Catalase (CAT): AS can be seen in variance analysis table (Table 1), different levels of SA had significant effect on catalase at 1% and other treatments did not have significant effect. Means comparison showed that SA at all the concentrations caused an increase in catalase, but the highest effect was due to the concentration of 450 mg/l (Figure 4). It was reported that pretreatment with SA activated antioxidant enzymes in maize (Janda et al., 1999, 2000) and bananas (Kang et al., 2003b) under cold stress. The greater increase in the activity of antioxidant enzymes SOD, CAT and APX after treatment with SA can be due to the metabolism of H₂O₂ produced by cold, thus resistance to cold stress is created (Kang et al., 2003b). Treatment of SA affected positively various parameters of fluorescence, and reduced those parameters related to electrolyte leakage. The reduction observed in catalase activity was accompanied with the increase in glutathione reductase and guaiacol peroxidase activity. In addition, SA and its derivatives such as benzaldehyde aspirin and coumaric acid also had a protective role against cold stress in maize (Janda et al., 1998, 2000; Horvath et al., 2002).

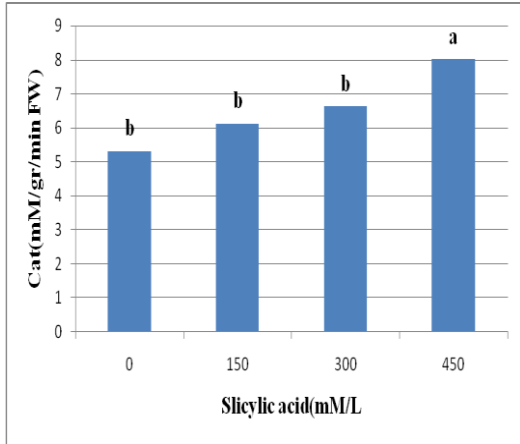


Figure 4. Effect of salicylic acid on the amount of catalase (mM/gr/min FW)

Peroxidase (POD): Table of ANOVA showed that the interaction of SA and potassium sulphate, and interaction of SA and sampling date had significant effect on the amount of POD at the levels of 5 and 1%, respectively. The results showed that POD activity increased by increasing the concentrations of SA. Salicylic acid at concentration of 450 mg/l resulted in the increase in the average amount of POD in three sampling dates when compared to the control. The highest effect of SA on POD was observed at the rate of 450 mg/l (Figure 5).

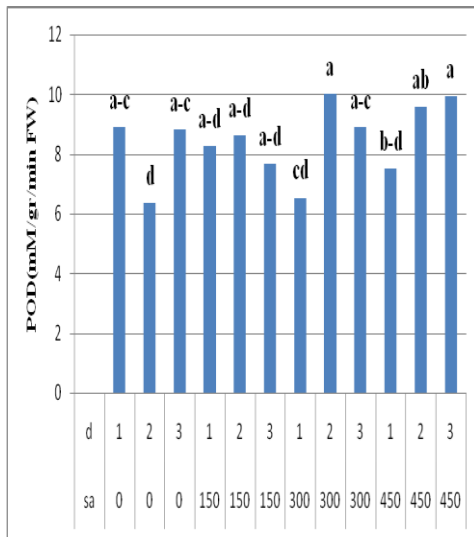


Figure 5. Effect of salicylic acid and sampling date on peroxidase (mM/gr/min FW) Interaction between SA at 450 mg/l and different levels of potassium sulphate (0 and 2%) had the reatest effect on the average rate of POD (Figure 6).

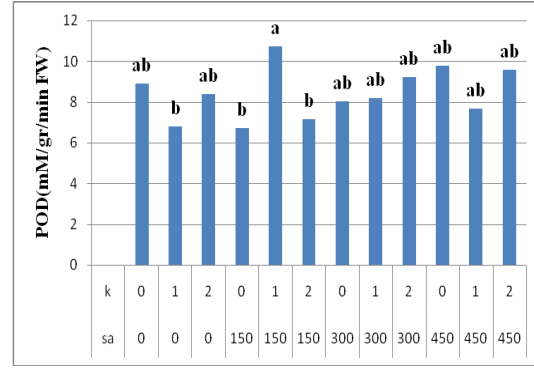


Figure 6. Effect of salicylic acid and potassium on peroxidase (mM/gr/min FW)

Peroxidase enzyme acts as an important catalyst in metabolic functions of plants. Plant behaviour has especial relationship with this enzyme. Physiologists and environmental scientists studied the response of plants to environmental factors, and introduced peroxidase as an appropriate indicator to assess the effects of environmental factors on the physiology of plants (Shirvani, 1998). Peroxidase is an oxidoreductase enzyme with important roles in plant's defense against external factors and the detoxification of reactive oxygen forms in the cell. It also reflects the metabolic response of plants to stressful factors (Shirvani, 1998). Peroxidase enzyme breaks toxic substance of hydrogen peroxide, which is produced in all physiological functions of plants, and converts it into water and oxygen. Plant resistance to death (due to the poisoning by the produced hydrogen proxise) is associated with the increase in peroxidase enzyme in infected plants. peroxidases of cell wall play an important role in subrification and cross-linking of cell wall compounds (Shirvani, 1998).

Superoxide dismutase (SOD): As the results in the table of ANOVA showed, SOD was significantly affected by just SA and sampling date at the level of 5% (Table 1). The highest SOD was observed in the second sampling date, the stage of endodormancy that plant increases antioxidant enzymes to neutralize the effect of free radicals. Salicylic acid at the concentration of 450 mg/l in the second sampling date had the greatest impact on SOD. Generally, increase in the level of potassium sulphate and SA led to the increase in the levels of SOD (Figure 7).

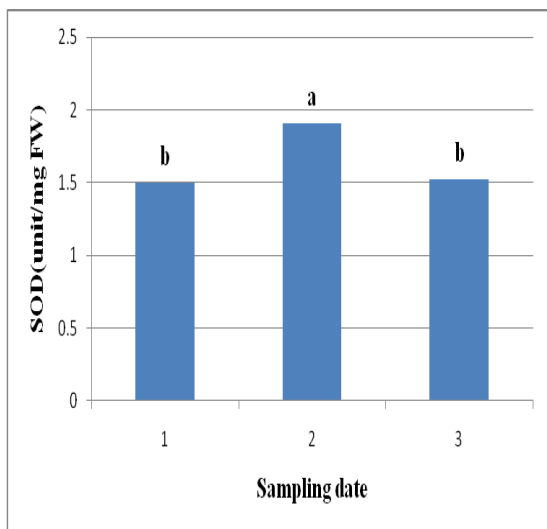


Figure 7. The effect of sampling date on superoxide dismutase (unit/mg FW)

Control treatment (no SA) showed the greatest effect on the average amount of SOD. Superoxide dismutase decreased as the concentration of SA increased (Figure 8). It was reported by Yusuf et al. (2008) that spray of plants with SA increased the activity of SOD under stress. Pretreatment with SA activated antioxidant enzymes in maize (Janda et al., 1999, 2000) and bananas (Kang et al., 2003b) under cold stress. The higher increase in the activity of antioxidant enzymes such as SOD, CAT and APX after treatment with SA is associated with the metabolism of H_2O_2 caused by cold, thus creating resistance to cold stress (Kang et al., 2003b). One of the methods of increasing cold resistance is the use of growth regulators such as SA (Wang and Shao 2006).

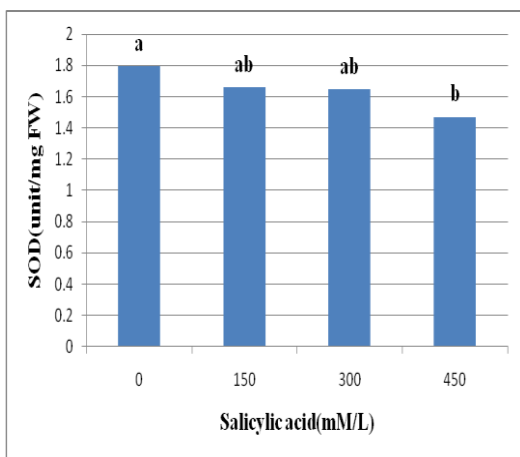


Figure 8. Effect of salicylic acid on superoxide dismutase (unit/mg FW)

Relative Water Content (RWC): According to the analysis of variance shown in table 1, the effects of different levels of potassium sulphate, dates of sampling, and interaction of SA and sampling date on RWC were significant at 1%, while the effects of the interaction of SA, potassium sulphate and sampling date were significant at the level of 5%. The analysis showed that the third sampling date had the greatest effect on RWC when compared with the two other dates (Figure 9).

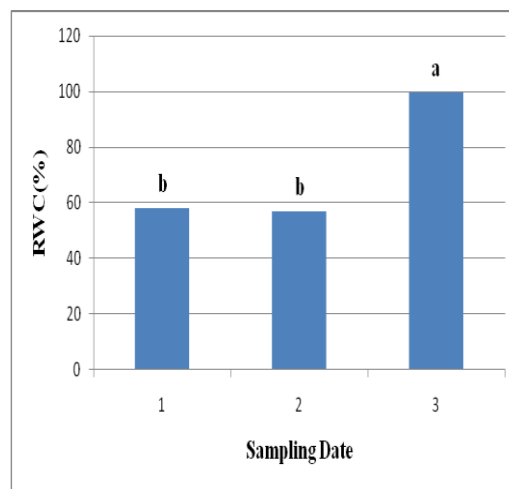


Figure 9. The effect of sampling date on relative water content (%)

Potassium sulphate at the levels of 0 and 2% had the greatest effect on RWC compared to the level of 1% with the lowest impact (Figure 10).

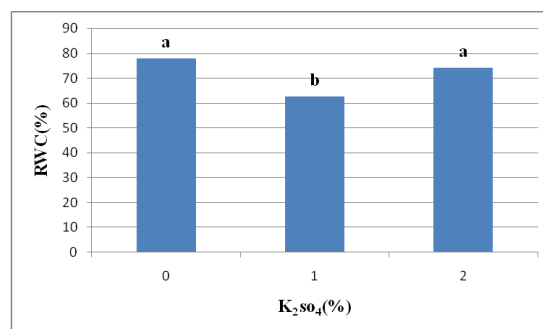


Figure 10. The effect of potassium sulphate on relative water content (%)

The highest RWC obtained in the grapes treated with SA at 450 and 150 mg/l, respectively (Figure 11). The increase found in RWC in the third sampling date (before breaking dormancy) is due to the increase in soluble sugars and proteins for the onset of bud activity. The reason for lower water content in the second sampling date is lower activity of buds which need lower amount of soluble sugars and proteins. An increase in the RWC was reported in chrysanthemum by spray of

salicylic acid (Vahdati et al., 2012). The interaction between SA and accumulation of proline and soluble sugar and increase of relative water content can be attributed to the osmotic regulation by accumulation of proline and soluble sugars. Demiralay et al. (2013) reported that under osmotic stress, treatment with SA significantly increased soluble sugars and proline through osmotic regulation.

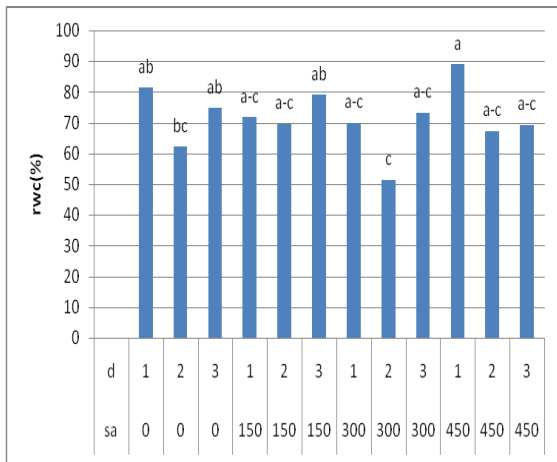


Figure 11. The effect of interaction between salicylic acid and sampling date on relative water content (%)

Electrolyte leakage (LE): Table of variance analysis showed that the effects of different levels of potassium sulphate, and interaction of SA and potassium sulphate on LE were significant at 5% level, and the effect of sampling date was significant at the level of 1% (Table 1). The results indicated that the lowest LE percentage was related to the second sampling date, and the highest rate was observed in the first and third sampling dates (Figure 12).

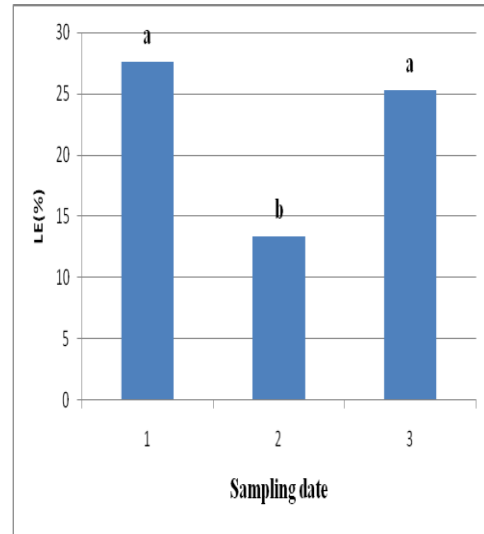


Figure 12. The effect of sampling date on electrolyte leakage percentage

Furthermore, the highest percentage of EL was associated to the treatment containing potassium sulphate at 1%, and the lowest rate was related to

potassium sulphate at 0 and 2% in all the three sampling dates (Figure 13).

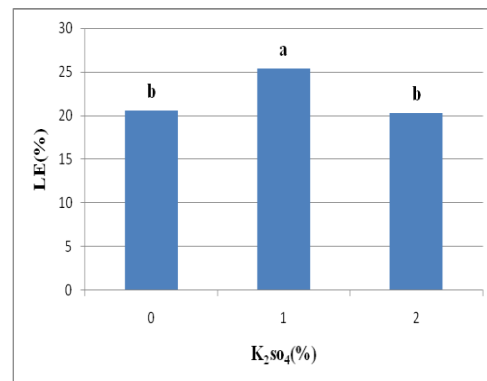


Figure 13. Effect of potassium sulphate on electrolyte leakage

Interaction between SA at 300 mg/l and potassium sulphate at 1% caused the lowest production of electrolyte leakage in grape bud, followed by the interaction of SA at 450 mg and potassium sulphate at 0 and 2%, interaction of SA at 0 and 300 mg and potassium sulphate at 0%, and interaction of SA at 0 mg and potassium sulphate at 2%, respectively (Figure 14). As one of the most prominent inorganic secretions in plants, potassium plays a key role in the creation of osmotic regulation ability even under drought conditions (Marschner, 2012). Measurement of electrolyte leakage in tissues under cold stress is an acceptable criterion for assessing

plant resistance to low temperatures (Ghasemi et al., 2012). When the plant tissues are damaged by cold, oxygen free radicals such as superoxide, hydrogen peroxide and hydroxyl radicals accumulate and cause damage to the membrane lipids and fatty acids (Hana and Bischoff 2004). Continuation of this trend results in higher destruction of the cell membrane and release of water from intracellular into extracellular space, leading to water-soaking phenomenon and increase of electrolyte leakage (Azzarello et al., 2009). Treatment of SA positively affected various parameters of fluorescence and reduced those parameters associated with electrolyte leakage (Janda et al., 1998, 2000; Horvath et al., 2002).

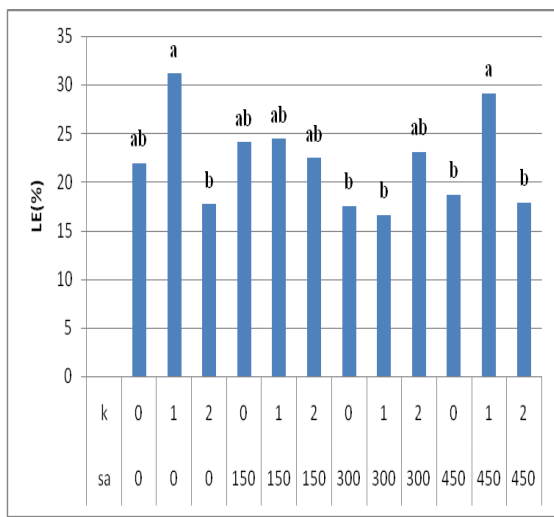


Figure 14. Effect of salicylic acid and potassium sulphate on ion leakage

Antioxidant activity: According to ANOVA table, sampling date and the interaction of SA and sampling date significantly affected antioxidant

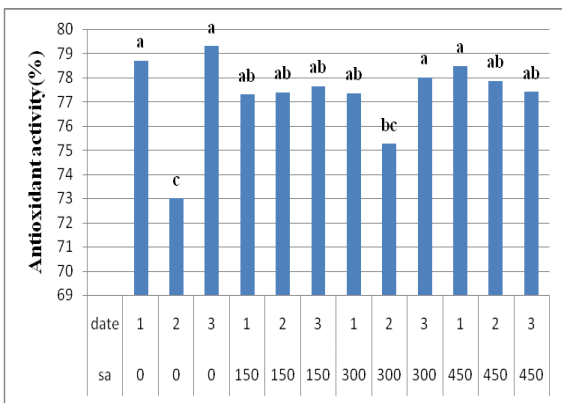


Figure 16. Interaction effect of salicylic acid and sampling date on antioxidant activity

activity at probability level of 1% (Table 1). The second sampling date showed the lowest percentage of antioxidant activity, and, on the contrary, the third sampling date exhibited the highest percentage of antioxidant activity (Figure 15).

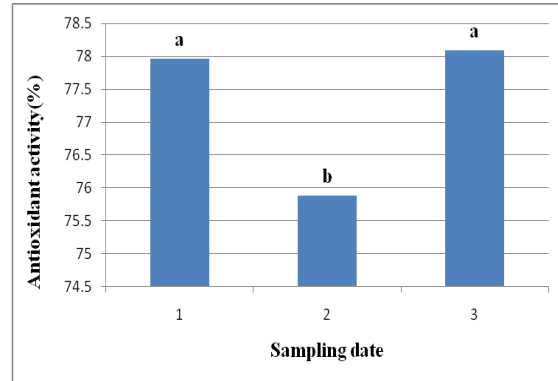


Figure 15. The effect of sampling date on the percentage of antioxidant activity

Regarding the interaction of SA and sampling date on antioxidant activity, the results showed that the highest percentage of antioxidant activity was attained in the treatment containing SA at 450 mg/l at the second sampling date. The average percentage of antioxidant activity in the treatment of 450 mg/l SA was higher than other concentrations. Salicylic acid at 450 mg/l in the

first, second and third sampling dates had reducing effect on the percentage of antioxidant activity. The lowest percentage of antioxidant activity in the second sampling date was related to the control (No SA). Salicylic acid at 150 mg had increasing effect on the percentage of antioxidant activity in different sampling dates (Figure 16).

Salicylic acid at 300 mg had the lowest effect on the production of antioxidant activity in the second sampling date. Generally, SA at 450 mg had the greatest effect on antioxidant activity. It was reported that SA affects fertility, growth, photosynthesis, plant water relations, and activities of antioxidant enzymes in plants exposed to biotic and abiotic stresses. Salicylic acid effectively reduced toxic effects caused by the exposure of plants to various abiotic stresses (Hayat et al., 2010). Cold stress limits plants growth and development and leads to the reduction of crop production, thus directly preventing metabolic reactions, and indirectly influencing osmotic pressure caused by cold, oxidative and other stresses. Positive effects of potassium ion on yield and cold tolerance were reported by Devi et al.

(2012). It was shown in *Panax ginseng* tree that high concentration of potassium ion activated antioxidant system and increased transcriptional levels of secondary metabolites relating to ginsenoside, which was associated with cold tolerance. Cold stress may cause disruption to the photosynthetic processes and reduce the effectiveness of antioxidant enzymes, thus leading to the accumulation of ROS (Mittler, 2002; Xiong et al., 2002; Suzuki and Mittler, 2006). Potassium increased the survival of plants under chilling stress by increasing antioxidant levels and reducing ROS production, (Devi et al., 2012; Cakmak, 2005).

4. CONCLUSION

In general, increasing the concentration of salicylic acid (450 mg), potassium sulphate (2%) and their interaction reduced the amount of malondialdehyde and electrolyte leakage, and increased catalase, peroxidase, superoxide dismutase, relative water content and antioxidant activity in grape buds on different sampling dates. Thus, the increase of antioxidant enzymes, relative water content and antioxidant activity, and decrease of electrolyte leakage and malondialdehyde in endodormancy stage led to the increased tolerance of grape buds to cold stress.

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