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Effect of glutathione and ascorbic acid on some physical characteristics of seedlings of grape plant Halawani cultivar Vitis vinifera L.

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Abstract

This experiment was conducted in the saran-covered plastic house of the College of Agriculture and Marshes, Thi-Qar University, on 20.02.2022, to study the effect of spraying with glutathione and ascorbic acid on the physical and chemical properties of the grape plant *Vitis vinifera* L at one year age. The effect of glutathione compound at four concentrations (0, 75, 150, 225) mg.l^{-1} and ascorbic acid at four concentrations also (0, 50, 100, 150) mg.l^{-1}, and the interaction between them was studied and implemented as a factorial experiment with two-factors (4×4×3) by following the randomized complete block design (RCBD) with three replications (48 seedlings per replicate). The averages were tested using the Revised least significant difference R.L.S.D at the probability level of 5%. The most important results obtained indicate that the effect of spraying with glutathione significantly on the studied vegetative characteristics, where the spraying treatment (G3) with a concentration of (225 mg l^{-1}) was significantly superior to the control treatment (G0) in(Stem diameter, number of branches, dry weight, wet weight), which gave the highest averages, respectively ((24,901 mm, 7.942 branches. plant-1, 1.596 g, 0.403 g, 20.903), respectively. As for ascorbic acid, its effect was significant. In the studied vegetative traits, the spraying treatment (3C) with a concentration of (150 mg.L-1) was superior in (plant height (cm), number of leaves (leaf. plant-1), leaf area (cm2), which gave the highest averages, respectively (25.110 mm, 25.110 mm, 7.509 branches. plant-1,1.649 g, 0.392, 20.767).

As for the two interactions (glutathione + ascorbic acid) at a concentration of (225 mg.l-1 + 150 mg.l-1), the treatments showed clear statistical differences, as the treatments excelled in the vegetative and chemical characteristics (stem diameter, number of branches, wet weight, weight). Dry, chlorophyll content of leaves) was a significant effect on the control treatment

I. INTRODUCTION

The grape (*Vitis vinifera* L.) is classified within the European grape group and belongs to the Vitaceae family, which includes (14 genera), the most important of which is the genus (Vitis). The number of grape species is estimated at about (700 species) and (10000 varieties) of grapes are planted on a large scale in the world, and the cultivation of grapes spreads between latitudes (20-50) north of the equator (20-40) south of the equator, the Food and Agriculture Organization issued Global Almanac on the area under cultivation of grapes in the world; It was estimated at about (7598,570 hectares). As for the global production, it is estimated at about (67557199 tons), as for Iraq, the number of trees reached (3552782 trees),

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the production rate of one tree is (34,640 kg), and the total production rate is approximately (123083 tons), and the number of planted varieties is about (70 varieties), most of which are concentrated In Northern Iraq (Abdul-Qader, 2006).

Grapes are one of the important horticultural crops, whether in Iraq or the world, and the most widespread in the world; This is due to its nutritional value and economic return. Grapes are one of the most common summer fruits in the world; It comes in second place after citrus in terms of production, and is one of the most beloved fruits by many people (Qassem et al., 2012).

Grapes have great economic importance in the reclamation and investment of various types of lands, including sandy, fertile, and shallow lands. It is also important in stabilizing the soil and preventing erosion compared to other types of fruits. The economic importance of grapes is due to its great financial return and the continuity of fruit for decades; As it reaches (50-60) years or more. Grapes have a high nutritional value, as their fruits contain sugars, vitamins, organic acids, mineral salts, proteins, fats and others, as well as their importance in medical uses in the treatment of diseases. It bears transportation and storage (Al-Saeedi, 2014)

There is great interest at the present time focused on the external addition of compounds and vitamins to improve plant growth and development, as they are natural products, including glutathione, which is used in biostimulants and antioxidants that have a role in curbing the oxidation and reduction processes in the plant cell. plant tissues, and it has multiple functions in plant development and growth that other antioxidants cannot perform or contribute, such as detoxification and stabilization or oxidation balance within the plant cell (Noctor et al., 2012). And also ascorbic acid (vitamin C), which is made vitally in high-end plants and affects plant growth and development,

, and plays an important role as a catalyst in enzymatic reactions in the electron transfer system and metabolic processes. Ascoric acid participates in many activities including photosynthesis, cell wall growth, cell elongation, resistance to environmental stresses, biosynthesis of ethylene, gibberellins, anthocyanins and proline pigments (Galal et al., 2000). Blokhina et al. (2003) that ascorbic acid is one of the most widely available antioxidants that protect plant cells, and it has been considered as a regulator of cell division and differentiation, adding that it is used in a wide range of important functions as a defensive antioxidant, photoprotection and regulation of photosynthesis and growth.

II. MATERIALS AND METHODS

A The experiment location

The experiment was carried out in the saran-covered plastic house of the College of Agriculture and Marshes, University of Dhi-Qar, during the growing season 2021-2022. A number of one-year-old grape seedlings were selected to know the effect of spraying with glutathione and ascorbic acid on the physicochemical characteristics of grapes grown in Dhi-Qar governorate. An agricultural growth media consisting of mixed soil and peat moss was used in a ratio of 1:3. A random sample was taken from the river mixture and a sample of peat moss and analyzed in the laboratories of the Directorate of Agriculture of Dhi-Qar to identify some chemical properties of soil and peat moss.

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Table (1) the chemical and physical properties of soil properties

Ph		٧,٥
EC	**ds.m^{-1}**	٢,٥١
Texture	**Silty loam**	**loamy sand**
Sand	**mg l^{-1}**	٦٩,٢٤
Silt		١٥,٤٤
Clay		١٥,٣٢
N		٢٥,٨٩
P		٧,٥٥
K		١١٦,٧٨

B Study Factors:

The study included a factorial experiment with two factors:

Factor I: Glutathione complex in four concentrations (0, 75, 150, 225) mg.l^{-1}

Factor 2: ascorbic acid in four concentrations (0, 50, 100, 150) mg.l^{-1}

C Dates and times of spraying

Grape seedlings, Halawani cultivar, were sprayed with glutathione at concentrations (0, 75, 150, 225) mg.l^{-1} and ascorbic acid at concentrations (0, 50, 100, 150) three times and according to the following dates:

First spray: On (25.03.2022) after the vegetative buds have opened

Second spray: 15 days after the first spray (10.04.2022)

Third spray: 15 days after the second spray (25.04.2022)

Measurements were taken a month after the third spray (25.05.2022).

A 5-liter capacity hand sprayer was used after adding the Tween-20 % to the spray solutions at a concentration of 0.1% to reduce the surface tension.

D studied traits

1: Diameter of the stem (mm) :-

The diameter of the seedling was measured using a vernear.

2: Number of side branches (branch. plant-1):-

According to the number of lateral branches per plant by calculating the number of branches on the main stem.

3: Wet and dry weight of leaves (gm):-

Paper samples were taken from grape seedlings from the vegetative growths located after the fifth leaf from the growing top, at a rate of five whole leaves for each repeater. dry.

4: Determination of the percentage of total chlorophyll (mg/g fresh weight):

The content of leaves of chlorophyll a, chlorophyll b and total chlorophyll in the leaves was estimated based on the method presented before the scientist (Dere, S., Gunes 1998) by taking (1 g) from the plant model, the method was cut into small pieces and crushed in a ceramic mortar with the presence of (10 ml) of acetone with a concentration of 80%. The filtrate

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was then separated from the sediment using a centrifuge at 3000 rpm for 15 minutes. The process of separating the filtrate from the precipitate was repeated several times until the green dye disappeared from the precipitate, after which the optical density of the filtrate was measured. by a spectrophotometer at two wavelengths (645, 663 nm).

5: User design and statistical analysis:

A randomized complete block design was used, and the experiments used were Factorial Experiments, with two factors, the first factor being glutathione complex at four concentrations (0, 75, 150, 225) mg.l-1, and the second factor being ascorbic acid, which was used at four concentrations. Also (0, 50, 100, 150) mg.l-1 and with three replicates for each experimental unit 3 seedlings, so that each replicate would have 48 seedlings. The statistical analysis was conducted using the statistical program Genstat in the analysis of variance. As for the comparison between the means, the Revised least significant difference (R.L.S.D) was used at the probability level of 0.05 (Al-Rawi and Khalaf Allah, 2000).

III. RESULTS

1: the rate of increase in the diameter of the leg (mm):

The results in Table (1) showed the superiority of the treatment of adding glutathione (3G) to the plant at a concentration of (225) mg.L-1 by giving it the highest increase in stem diameter of (24.901 mm) significantly over the control treatment that It gave the least increase in leg diameter (21.627) mm. As for the addition of ascorbic spray to the plant, treatment (C3) with a concentration of (150) mg.L-1 was superior to the control treatment (C0), in which the increase in stem diameter was (25.110) mm compared to the control treatment (C0) in which the increase in the diameter of the leg reached (21.566)mm.

As for the interaction between the two factors of the study, the study treatment (225*150) mg.l-1 showed a significant superiority over the rest of the interactions by giving it the highest value for increasing the stem diameter amounted to (26,071) mm, while the treatment given In comparison, the lowest value for the increase in the diameter of the leg was (15.771) mm.

Table (1) The effect of glutathione and ascorbic and the interaction between them on the increase in stem diameter (mm) for Helwani cultivar seedlings)

Glutathione Average	Ascorbic acid concentration				Glutathione Concentration
	١٥٠	١٠٠	٥٠	0	
٢١,٦٢٧	٢٥,٧٠٣	٢٢,٠٩٢	٢٢,٩٤٢	١٥,٧٧١	٠
٢٢,٣٣٧	٢٣,٣٣٢	٢٣,٧٧٤	٢٠,٩٢٣	٢١,٣٢٢	٧٥
٢٤,٥٦٣	٢٥,٣٣٤	٢٣,٩٥١	٢٤,٨٢٤	٢٤,١٤٣	١٥٠

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٢٤,٩٠١	٢٦,٠٧١	٢٥,١٤٤	٢٣,٣٦١	٢٥,٠٣١	٢٢٥
	٢٥,١١٠	٢٣,٧٤٠	٢٣,٠١٢	٢١,٥٦٦	**Ascorbic acid average**
RLSD 0.05					
G*C=1.160		**C=0.580**		**G=0.580**	

2: the increase in the number of branches (branch. plant^{-1}):

The results in Table (2) indicate that the addition of glutathione had a significant effect on the rate of increase in the number of branches, as the treatment (G3) at a concentration of (225) mg.L-1 significantly outperformed the control treatment (G0).) by giving it the highest increase in the average number of branches, which reached (7,942) branch.plant-1. As for ascorbic, the results of the same table indicate the superiority of the treatment of (C3) at a concentration of (150) mg.L- 1 significantly over the comparison treatment (C0), as it gave the highest increase in the number of branches, which amounted to (7.509) branch.plant-1.

As for the binary interaction (adding the compound glutathione * ascorbic acid to the plant), it exhibited a significant positive behavior, as the interaction (225 * 150) mg.l-1 was superior to the rest of the interactions, and the increase in the number of branches was (8.723)) plant. branch-1, while the interaction treatment (0*0) gave the least number of branches, which amounted to (4.057) branch. plant-1.

Table (5) Effect of glutathione and ascorbic and the interaction between them on the increase in the number of branches (Branch.plant $^{-1}$) of grape seedlings variety (Halwani)

Glutathione Average	**Ascorbic acid concentration**				**Glutathione Concentration**
	١٥٠	١٠٠	٥٠	**0**	
٥,٥٦٣	**6.870**	**6.033**	**5.293**	**4.057**	٠
٥,٨٠١	**7.703**	**6.317**	**5.567**	**3.617**	٧٥
٦,٥٠٢	**6.743**	**6.527**	**6.590**	**6.140**	١٥٠
7.942	**8.723**	**8.333**	**7.943**	**6.767**	٢٢٥
	٧,٥٠٩	٦,٨٠٣	٦,٣٤٨	٥,١٤٥	**Ascorbic acid average**
RLSD 0.05					
G*C=0.448		**C=0.224**		**G=0.224**	

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3: Leaves wet weight (g):

It is clear from the results shown in Table (3) that the treatment of adding glutathione to the plant ((G3) at a concentration of (225) mg.L-1 by giving it the highest increase in wet weight (1.596) g significantly over the control treatment Which gave the least increase in wet weight amounted to (0.761) g. As for adding ascorbic spray to the plant, the treatment (C3) with a concentration of (150) mg.L-1 outperformed the control treatment (C0), in which the increase in wet weight was (1.649) g compared to the treatment of The comparison (C0) in which the increase in wet weight was (0.861) g.

As for the interaction between the two factors of the study, the study treatment (225 * 150) mg.l-1 showed a significant superiority over the rest of the interactions by giving it the highest value for an increase in wet weight (1.976) g, while the treatment given The comparison has the lowest value for an increase in wet weight and it reached (0.520) g.

Table (3) Effect of glutathione and ascorbic and the interaction between them on wet weight (gm) in leaves of grape seedlings (Halwani)

Glutathione Average	Ascorbic acid concentration				Glutathione concentration
	١٥٠	١٠٠	٥٠	0	
٠,٧٦١	1.170	0.733	0.620	0.520	٠
1.004	1.660	1.106	0.696	0.553	٧٥
1.333	1.790	1.203	1.180	1.156	١٥٠
١,٥٩٦	1.976	1.853	1.343	1.213	٢٢٥
	١,٦٤٩	1.224	٠,٩٦٢	0.861	Ascorbic acid average
RLSD 0.05					
G*C=0.073		C=0.036		G=0.036	

4: Measuring the dry weight of leaves (g):

The results of Table (4) show that the addition of glutathione had a significant effect on the rate of increase in the dry weight of the leaves, as the addition treatment (G3) at a concentration of (225) mg.L-1 significantly outperformed the control treatment (G0) by giving it the highest increase in the dry weight rate, which reached (0.403) g. As for ascorbic, the results of the same table indicate that the addition treatment (C3) at a concentration of (150) mg.L-1 was significantly superior to the control treatment (C0), as it gave the highest increase in the dry weight of the leaves, which amounted to (0.392) g.

As for the two interactions (glutathione compound * ascorbic acid), it exhibited a significant positive behavior, as the interaction (225 * 150) mg.l-1 was superior to the rest of the interactions, and the increase in the dry weight of the leaves was (0.503) g Whereas, the interference treatment (0 * 0) gave the lowest leaf content of dry weight, which was (0.167) g.

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Table (4) Effect of glutathione and ascorbic and the interaction between them on dry weight (gm) in the leaves of grape seedlings variety (Halwani)

Glutathione average	Ascorbic acid concentration				Glutathione concentration
	١٥٠	١٠٠	٥٠	0	
٠,١٩٠	0.238	0.178	0.176	0.167	٠
٠,٢٤١	0.357	0.246	0.183	0.176	٧٥
0.383	0.468	0.416	0.412	0.232	١٥٠
0.403	0.503	0.476	0.347	0.282	٢٢٥
	٠,٣٩٢	٠,٣٢٩	0.279	0.214	Ascorbic acid average
RLSD 0.05					
G*C=0.004		C=0.002		G=0.002	

5: Total chlorophyll content of leaves (mg.g-1 fresh weight):

The results of Table (5)showed that the effect of glutathione on the rate of chlorophyll in the content of plant leaves was significant, and the treatment (G3 at a concentration of (225) mg.l-1 recorded the highest average of 20.903)) mg 100 g fresh weight that outperformed the control treatment that gave the lowest mean value 19.04 mg 100 gm fresh weight.

As for ascorbic, it had a significant effect on the rate of chlorophyll in the content of plant leaves, and the treatment (C3) gave a concentration of (150) mg.L-1 (20.767) mg 100 gm of fresh weight, which outperformed the comparison treatment that gave 19.108 mg 100 g fresh weight.

The results listed in Table (5) showed that the interaction had a significant effect, as the treatment (225 * 150) mg.L-1, which amounted to (21.603) mg 100 g fresh weight, outperformed the comparison treatment that gave the lowest value For this trait (18,520) mg 100 g fresh weight.

Table (5) Effect of glutathione and ascorbic acid and the interaction between them on total chlorophyll in leaves of grape seedlings variety (Halwani)

Glutathione Average	Ascorbic acid concentration				Glutathione concentration
	١٥٠	١٠٠	٥٠	0	
١٩,٠٤٧	19.700	19.263	18.703	18.520	٠
19.803	20.573	20.210	19.506	18.923	٧٥
٢٠,٣٩٥	21.190	20.900	20.420	19.070	١٥٠

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٢٠,٩٠٣	21.603	21.386	20.706	19.916	٢٢٥
	20.767	20.441	19.834	١٩,١٠٨	Ascorbic acid average
RLSD 0.05					
G*C=0.048		C=0.024		G=0.024	

It is noticed from the tables (1, 2, 3, 4, 5) that the following studied vegetative traits (stem diameter, number of branches, fresh weight of leaves, dry weight of leaves, content of leaves from chlorophyll) outperformed with a direct increase with increasing concentration of glutathione compound as it gave a concentration of 225 mg.l-1 is the highest averages in a row (24.901 mm, 7.942 branch.plant-1, 1.596gm, 0.403gm, 20.903 mg.g-1 fresh weight) compared to the comparison treatment. This is due to the fact that the glutathione compound has an active role in the various metabolic processes in the plant and thus increase the growth that It reflects positively on the activity of the vegetative system, which leads to an increase in the characteristics of the vegetative system as a result of cell expansion, and glutathione is small oxidizing and reducing molecules that have a role in the formation of salicylic acid and the defense signals of the plant, Salicylic acid works in maintaining auxins and inhibiting the enzyme IAA oxidase and also has a role in increasing gibberellin and cytokines and in increasing division in meristematic regions and thus increasing plant growth (Ghairb and Hegazi, 2010). This may be due to the fact that glutathione has a role in cell division and elongation, as glutathione is considered an antioxidant that works to protect cells from breakdown and maintains cells in their active form. They are glutamate, cysteine and glycine, and the amino acid cysteine is included in the composition of the sulfur bond, which is a sulfur donor, which works to protect cells from free radicals, as well as the important role of glutathione in the photosynthesis process, and a decrease in dry weight was observed with a decrease in glutathione in plants (Bianucci, 2008).). Also, treating the leaves with glutathione caused an increase in their total chlorophyll content because it caused an increase in the levels of Mg, which enters the synthesis of chlorophyll (Amini et al., 2011).

It was also shown that the significant increase that occurred in the tables (2,3,4,5,6) gave a concentration of 150 mg.L-1 in the traits (stem diameter, number of branches, fresh weight, dry weight, chlorophyll) the highest averages (25.110 mm). , 25.110 mm, 7.509 branch. plant-1, 1.649 g, 0.392, 20.767) compared to the comparison treatment, which gave the lowest values, and this is due to the fact that plant growth can be stimulated when treated with amino acids, which is due to its effect on building large molecules, for amino acids. that it increases DNA synthesis (Smith, 1985), , stimulating various processes associated with protein synthesis and stimulating cell division. Ascorbic acid plays a major role in promoting photosynthesis through the strong relationship between leaf surface area and its content of ascorbic acid. It may be attributed to stimulating growth through increased leaf development and increased photosynthetic surface area (El-Naggar and Sewedan, 2009). Which led to an increase in the efficiency of the photosynthesis process and consequently an increase in the wet and dry weight of vegetative growth and the percentage of carbohydrates in the leaves, thus increasing the number of cells in the leaves and their content of manufactured and stored materials, which increases the percentage of dry

matter in them. The reason may be attributed to the role of ascorbic acid in increasing the content of chlorophyll in leaves, which directly affects the photosynthesis process as chlorophyll is Chlorophyll is the main pigment in this cell

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