Effect of Biofertilizers as a Partial Substitute for Nitrogen Fertilzier on Vegetative Growth, Yield, Fruit Quality and Leaf Mineral Content of Two Seedless Grape Cultivars II: Fruit Quality and Leaf Mineral Content

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Abstract: Controlling chemical fertilization, especially N fertilizer is very important for reducing environmental pollution and obtaining safe food. Using biofertilizers has been a good method in this respect. This study was initiated as an attempt for replacing the excessive application of mineral nitrogen partially with four biofertilizers namely Nitrobeine, Rhizobacterine, Biogen and active dry yeast for achieving an economical yield and obtaining berries characterized with higher quality and safe produce. Nitrogen was applied at zero% up to 100% of the recommended nitrogen dose for Thompson seedless and Flame seedless cultivars with or without biofertilizers or biofertilizers alone during 2003 and 2004 seasons. Increasing N rates from zero% to 100% caused a significant increase in cluster length and width, berry length and diameter, volume and weight of 100 berries, T.S.S, T.S.S / acid ratio and a significant decrease in total acidity in both cultivars. Biofertilizer treatments resulted in the highest values of physical properties and T.S.S / acid ratio. However; they gave significant decrease in juice acidity as compared with the control. The treatment of 50% nitrogen fertilizer plus 20 gm Biogen gave the highest values of the studied characteristics. In the contrary, the treatment zero% gave the least value. Both N rate and Biofertilizers treatments caused significant increase in N, P and K of leaf petiole of the two cultivars as compared with the control in both seasons. Generally, results proved that, using nitrogen at 40 units for Thompson seedless and 30 units for Flame seedless plus Biogen led to a considrable increase in yield and improved fruit quality as compared with using 80 units for Thompson seedless and 60 units for Flame seedless without Biofertilizers. Hence, it could be concluded that using Biofertilizers can reduce the need for about 50% of the recommended nitrogen dose.

Key words: Grapes · Vitis vinifera · Biofertilizers · Fruit quality · Leaf mineral content

INTRODUCTION

Grape (Vitis vinifera L.) ranks first among fruit crops grown all over the world. It is the second fruit crop in Egypt after citrus and mainly consumed as fresh table grapes. Fertilization, especially nitrogen, is considered as an important practice during the growing season [1]. Mineral fertilizers and other chemicals that commonly used in agricultural production, not only have harmful effects on the environment, but also they can alter the composition of fruits, vegetables and root crops [2]. Biofertilizers are very safe for human, animal and environment and mainly comprise nitrogen fixers such as Rhizobium, Azotobacter, Azosprillum, Azola, Blue-green

algae, Phosphate dissolvers and Silicate bacteria [3, 4]. Average berry weight of Thompson seedless and Flame seedless grapevines was increased when N level was increased [5, 6]. Total soluble solids (TSS) content was much lower in the higher applied nitrogen concentrations with an undesired effect on increasing titrtable acidity [5, 7-10]. Soil nitrogen fertilization increased the total nitrogen content in leaf petioles [11-13, 6]. Using yeast as a soil or a foliar application on Thompson seedless grapevines increased berry weight and berry size and TSS compared with the control On the contrary, acidity was significantly decreased on Flame seedless grapevines [9, 6]. The application of biofertilizers, Biogen, Rhyzobacterine and Microbene; alone or in combination

with the mineral N fertilizer on Flame seedless and Crimson seedless grapevines resulted in a positive significant effect on berry weight [14, 15]. Phosphorene significantly increased P and K content of leaf petiole while nitrobeine or Rhizobacterine increased N of leaf petiole [16]. Using of the yeast on Flame seedless cultivar significantly improved the leaf content of N, P and K [9]. Furthermore, the combined application of Biofertilizers with mineral N caused a significant increase in the percentage of N, P and K in the leaves as compared with the addition of mineral N only [14, 17]. The aim of this investigation was to study the effect of using commercially available biofertilizers (Biogen, Rhizobacterine, Nitrobeine and active dry yeast), ammonium nitrate (33% N) with different levels and their interaction on fruit physical and chemical characteristics and leaf mineral content of Thompson seedless and Flame seedless grapes.

MATERIALS AND METHODS

A field study was conducted at the experimental farm of the Horticulture Research Station, Ali Moubark Village, Beheira governorate. The present work was carried out during 2003 and 2004 seasons on three years-old Thompson seedless and Flame seedless grapevines. Experimental vines were chosen as to be similar in growth as possible. They were planted in a sandy soil at 2.0×3.0 m apart under drip irrigation system. The cane system of training was applied on Thompson seedless. Number of the retained eyes on each vine was around 80 depending on knowledge that grape cultivars which the basal buds are less fruitful may be trained with cane system [18].

Flame seedless vines were trained to the double cordon system. Number of retained eyes on each vine was 60. Pruning for both cultivars was carried out at the first week of January in both seasons. Vines were sprayed with Dormex (4% v/v) at the last week of January. All vines of both cultivars were subjected to the standard horticultural practice. The vines were fertilized by the recommended doses of phosphorus (40 units of P₂O₅) as calcium super phosphate 15.5% and potassium (100 units of K₂O) as potassium sulphate 48%. The recommended doses of nitrogen were 80 units for Thompson seedless and 60 units for Flame seedless as ammonium nitrate 33% per feddan. Thus the experiment was comprised of 15 treatments for both cultivars (3 levels of nitrogen × 4 biofertilizers × 4 replicates in addition to the control as shown in Table (1).

The treatments were in split plots in randomized completely blocks design. Soil analysis was carried out according to Wilde et al. [19] and the obtained data are shown in Table (2). The application of nitrogen fertilizer followed the same normal application practices as follow: 15% after bud burst and before flowering, 50% after flowering (at veraison) and 35% after harvest. Bio-fertilizers, Nitrobeine, Rhizobacterine and Biogen at 20 g / vine were mixed with the organic matter and the mixtures were added to the soil before the second and the third irrigations. Active dry yeast was applied to the soil at 16 g / vines (sugar was added to the yeast solution at the rate of 3 kg $\frac{1}{2}$ kg of dry yeast for activating their reproduction). The phosphorus was applied during winter. Potassium was added as follows: 50% during winter and 50% during the growing seasons.

Table 1:The treatments of the experiment for Thompson seedless and Flame seedless grapevines.

Treatment number	Nitrogen doses	Biofertilizers
1	Zero nitrogen	Nitrobeine
2		Rhizobacterine
3		Biogen
L		Active dry yeast
5		Control
	50% of the recommended Nitrogen	Nitrobeine
		Rhizobacterine
1		Biogen
		Active dry yeast
0		Control
1	100% of the recommended nitrogen	Nitrobeine
2		Rhizobacterine
3		Biogen
4		Active dry yeast
15		Control

Table 2: Chemical and mechanical analysis of the soil at the experimental site.

Soluble cations (meq / L)				Soluble anions (meq / L) Mechanical analysis									
Soil	EC												
depth (cm)	dcm^{-1}	pН	Ca ⁺²	Mg^{2+}	Na ⁺	K	HCÔ₃	CL^{-}	SO ⁻²	Sa%	Si%	Cl. %	Tex.
0 - 30	1.38	9.16	1.25	0.60	1.60	0.20	1.18	1.8	0.75	90.9	3.6	55	Sand.
30 - 60	1.32	9.25	1.10	0.55	1.44	0.15	1.02	1.6	0.63	91.5	2.8	5.7	Sand.

Physical and Chemical Properties of Grapes: Cluster width, cluster length, weight of 100 berries (g) and volume of 100 berries (ml) was recorded at harvest. Berry diameter in (cm) was measured by venire caliper and berry length in (cm), were determined. Total soluble solids percentage was determined by a hand rafractometer, titrtable acidity as grams of tartaric acid / 100 ml Juice according to A. O. A. C. [20] and total soluble solids / acid ratio (T.S.S / acids / ratio) was calculated.

Leaf Mineral Analysis: Leaf samples were taken at first week of August for both seasons from the most recent fully matured leaves $(5-7^{th})$ leaves from shoot tips). The petioles of leaves were dried at 70° C until a constant weight, then ground to a powdery mixture and 0.2 g was taken from each treatment for N, P and K determination. Total nitrogen was determined according to Pregl [21], potassium was Flame photometrically determined by using a bye unican sp 1990 Atomic absorption spectrometer according to Brandifeld and Spincer [22]. Phosphorus was colorimetrically determined according to Murphy and Riley [23].

Statistical Analysis: The treatments were arranged in split plots in completely randomized blocks design and analyzed according to Snedecor and Cochran [24]. Least significant differences were used to compare between treatment means according to Walter and Duncan [25].

RESULTS AND DISSCUSSION

Effect of Nitrogen and Biofertilizers on Physical Properties of Grape Berries

Berry Length and Berry Diameter: Data listed in Tables 3 and 5 showed that, berry length and berry diameter of both Thompson seedless and Flame seedless grapes were increased significantly by increasing nitrogen doses in both seasons. The highest value of berry length was recorded by vines received 100% of the recommended nitrogen. Regarding the effect of biofertilizers, data revealed that Biogen and active dry yeast significantly increased berry length and berry diameter of both cultivars. However, no significant differences were found between Rhizobacterine, Nitrobeine and the untreated vines. The interaction

between the two studied factors was significant as shown in Tables 4 and 6. The highest values of berry length and berry diameter were obtained by vines received 50% of the recommended nitrogen plus Biogen. However, the least value of berry length was observed in vines of the control.

Weight of 100 Berries and Volume of 100 Berries:

As shown in Tables 3 and 5, significant differences among nitrogen doses applied to Thompson seedless and Flame seedless grapes were reported as concerns weight of 100 berries and 100 berries volume during 2003 and 2004 seasons. The highest weight and volume, of 100 berries were obtained by treatment of 100% of the recommended nitrogen. It was clear that use of biofertilizers led to a significant increase in weight and volume of 100 berries in comparison with the untreated vines. The highest value was obtained by treatment of Biogen followed by active dry yeast, Rhizobacterine and Nitrobeine, respectively. The interaction between the two studied factors was significant as shown in Tables 4 and 6. The highest weight and volume of 100 berries were obtained by vines received 50% of the recommended nitrogen plus Biogen. However, the least weight of 100 berries was given by the control for both the two cultivars. These findings could be attributed to the effect of nitrogen on stimulating vegetative growth which increased carbohydrate formation in addition to its direct effect on stimulating fruit growth. These results are in agreement with those of Chadha and Singh [26] and Ali - Mervet [27] on Flame seedless grapevines.

Cluster Width and Cluster Length: As shown in Tables 7 and 9, cluster width and cluster length of both Thompson seedless and Flame seedless grapevines were significantly affected by nitrogen doses during 2003 and 2004 seasons. The highest value of cluster width was recorded by vines received 100% of the recommended nitrogen. As concerns the effect of Biofertilizers, data showed that the highest values of both cluster width and cluster length for both cultivars were recorded by vines received Biogen in both seasons. The interaction between the two studied factors was significant as shown in Tables 8 and 10. The highest values of cluster width and length were obtained by vines received 50% of the recommended nitrogen in the presence of Biogen.

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Table 3: Effect of nitrogen and Biofertilizers on some berry characteristics of Thompson seedless grapevine in 2003 and 2004 seasons

	Berry dian	Berry diameter (cm)		Berry length (cm)		reight (g)	100 berries v	100 berries volume (cm) ³	
Treatments	2003	2004	2003	2004	2003	2004	2003	2004	
$\overline{N_0}$	0.84	0.83	1.17	1.30	129.50	133.05	119.30	121.40	
½ N	0.91	0.89	1.27	1.40	141.20	145.10	135.55	136.25	
1 N	0.95	0.94	1.38	1.46	146.05	151.70	139.65	139.65	
$LSD_{0.05}$	0.03	0.05	0.06	0.05	1.77	4.98	2.84	2.68	
\mathbf{B}_1	0.94	0.87	1.20	1.32	133.00	136.38	127.08	130.16	
B_2	0.94	0.88	1.18	1.33	138.00	142.00	132.58	135.66	
B_3	0.99	0.96	1.41	1.61	147.58	153.33	141.41	144.33	
B_4	0.95	0.94	1.30	1.45	143.50	148.08	137.66	139.41	
B_5	0.83	0.79	1.17	1.30	129.50	131.80	121.08	125.83	
$LSD_{0.05}$	0.08	0.08	0.091	0.11	3.19	4.44	4.30	4.71	

 N_0 = Zero nitrogen, B1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 4: The interaction effect of nitrogen and Biofertilizers on some berry characteristics of Thompson seedless grapevine in 2003 and 2004 seasons

		2003 season				2004 season			
Treatments		Berry diameter (cm)	Berry length (cm)	100 berries weight (g)	100 berries volume (cm) ³	Berry diameter (cm)	Berry length (cm)	100 berries weight (g)	100 berries volume (cm) ³
Nitrogen b	oiofertilizers								
0	\mathbf{B}_1	0.81	1.22	122.00	115.00	0.77	1.25	125.50	113.25
	B_2	0.82	1.15	130.50	121.50	0.80	1.22	135.00	125.50
	\mathbf{B}_3	0.87	1.40	139.00	124.20	0.85	1.50	144.25	129.75
	B_4	0.93	1.32	135.70	122.50	0.90	1.40	140.00	126.75
	B_5	0.82	1.10	120.20	113.00	0.87	1.12	120.50	111.75
1/2	\mathbf{B}_{1}	0.85	1.17	138.20	133.50	0.87	1.27	143.25	132.50
	B_2	0.95	1.20	146.00	138.70	0.97	1.37	150.50	141.75
	\mathbf{B}_3	1.02	1.45	157.20	144.50	1.07	1.75	164.75	147.50
	B_4	0.98	1.37	152.70	141.20	1.05	1.50	155.25	146.75
	\mathbf{B}_{5}	0.95	1.20	136.70	131.70	0.90	1.40	141.75	129.75
1	\mathbf{B}_1	0.90	1.20	138.70	136.20	0.95	1.37	136.75	131.75
	B_2	0.90	1.20	137.50	137.50	0.90	1.40	140.50	139.75
	\mathbf{B}_3	0.97	1.40	146.50	143.20	0.87	1.60	151.00	143.75
	B_4	0.92	1.20	142.00	140.20	0.90	1.37	149.00	141.75
	\mathbf{B}_{5}	1.02	1.22	140.50	134.20	0.92	1.40	148.25	140.00
LSD _{0.05}	0.13	0.15	5.54	7.45	0.15	0.20	7.70	8.16	

Table 5: Effect of nitrogen and Biofertilizers on some berry characteristics of Flame seedless grapevine cultivar in 2003 and 2004 seasons

	Berry diameter (cm)		Berry lengt	Berry length (cm)		100 berries weight (g)		100 berries volume (cm) ³	
Treatment	2003	2004	2003	2004	2003	2004	2003	2004	
$\overline{N_0}$	1.20	1.40	1.30	1.45	210.55	215.30	208.45	211.35	
½ N	1.33	1.60	1.40	1.60	237.35	241.00	224.45	230.65	
1 N	1.43	1.66	1.48	1.63	243.30	247.85	235.10	246.35	
$LSD_{0.05}$	0.09	0.05	0.068	0.02	1.29	3.34	7.95	9.16	
\mathbf{B}_1	1.31	1.35	1.38 c	1.53	223.91	228.58	222.66	229.75	
B_2	1.33	1.39	1.45	1.55	231.00	234.8	225.91	231.91	
\mathbf{B}_3	1.56	1.64	1.45	1.65	245.58	248.83	237.25	238.08	
B_4	1.45	1.50	1.47	1.57	238.33	241.0	230.83	232.50	
\mathbf{B}_{5}	1.18	1.20	1.33	1.49	218.16	224.58	217.66	223.33	
LSD _{0.05}	0.10	0.10	0.11	0.09	6.89	5.97	8.58	13.60	

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 6: The interaction effect of nitrogen and Biofertilizers on some berry characteristics of Flame seedless grapevine in 2003 and 2004 seasons

		2003 season				2004 season			
Treatmen	ts	Berry diameter (cm)	Berry length (cm)	100 berries weight (g)	100 berries volume (cm) ²	Berry diameter (cm)	Berry length (cm)	100 berries weight (g)	100 berries volume (cm) ²
Nitrogen	Biofertilizers								
0	\mathbf{B}_1	1.20	1.32	200.00	196.20	1.42	1.42	203.25	210.00
	\mathbf{B}_2	1.22	1.37	211.20	204.00	1.45	1.50	215.75	213.00
	\mathbf{B}_3	1.45	1.47	230.00	220.20	1.60	1.55	233.75	225.00
	B_4	1.30	1.37	219.00	208.00	1.57	1.45	223.75	216.50
	\mathbf{B}_{5}	1.12	1.25	192.00	185.00	1.35	1.35	200.00	203.50
1/2	\mathbf{B}_1	1.37	1.47	238.70	231.00	1.65	1.55	242.50	235.00
	B_2	1.40	1.47	243.00	231.70	1.70	1.60	246.25	239.50
	\mathbf{B}_3	1.60	1.62	254.50	246.00	1.85	1.75	257.50	258.00
	B_4	1.50	1.52	250.20	241.00	1.72	1.62	253.75	244.50
	\mathbf{B}_{5}	1.25	1.35	230.00	222.00	1.52	1.47	235.00	226.25
1	\mathbf{B}_{1}	1.36	1.47	235.00	229.00	1.50	1.62	240.00	236.25
	B_2	1.37	1.50	238.70	233.00	1.62	1.62	241.75	236.25
	B_3	1.47	1.52	244.00	236.20	1.77	1.65	246.25	241.25
	B_4	1.39	1.52	238.70	233.70	1.67	1.60	242.50	237.50
	\mathbf{B}_{5}	1.32	1.40	233.20	227.00	1.57	1.65	238.75	230.50
LSD _{0.05}	0.17	0.18	11.94	14.88	0.17	0.16	10.35	23.55	

Table 7: Effect of nitrogen and Biofertilizers on some cluster characteristics of Thompson seedless grapevines in 2003 and 2004 seasons

	Cluster length (cm)	-	Cluster width (cm)	
Treatments	2003	2004	2003	2004
$\overline{N_0}$	21.25	20.96	10.52	10.66
½ N	23.29	23.95	11.14	11.66
1 N	23.51	25.90	11.57	12.35
$LSD_{0.05}$	0.72	1.34	0.27	0.54
B_1	22.65	23.02	10.69	11.30
B_2	23.30	24.51	10.85	11.57
B_3	24.69	26.71	11.93	12.80
B_4	24.30	25.20	11.30	11.99
B_5	20.82	21.57	10.62	10.65
LSD _{0.05}	1.41	1.25	0.52	0.73

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 8: The interaction effect of nitrogen and Biofertilizers on some cluster characteristics of Thompson seedless grapevine in 2003 and 2004 seasons

		2003 season		2004 season	
Treatments		Cluster length (cm)	Cluster width (cm)	Cluster length (cm)	Cluster width (cm)
Nitrogen	biofertilizers				
0	\mathbf{B}_1	22.40	10.55	22.62	10.72
	B_2	24.40	10.30	24.62	10.87
	\mathbf{B}_3	24.30	11.22	26.05	11.87
	B_4	24.00	10.40	25.15	11.05
	\mathbf{B}_{5}	21.40	10.15	21.37	10.30
1/2	B_1	22.80	10.77	23.12	11.37
	B_2	22.60	11.37	24.72	11.80
	B_3	25.50	12.45	27.55	13.30
	B_4	24.50	12.37	26.20	12.97
	$\mathrm{B}_{\scriptscriptstyle{5}}$	22.00	10.30	22.90	10.97
1	B_1	22.80	10.55	23.32	11.05
	B_{2}	22.90	10.88	24.20	12.05
	B_3	24.20	12.12	26.55	13.22
	B_4	24.30	11.12	25.70	11.75
	\mathbf{B}_{5}	22.60	11.62	23.45	12.62
LSD _{0.05}	2.44	0.89	2.17	1.25	

Table 9: Effect of nitrogen and Biofertilizers on some cluster characteristics of Flame seedless grapevine in 2003 and 2004 seasons

	Cluster length (cm)		Cluster width (cm)	
Treatments	2003	2004	2003	2004
N_0	19.47	19.88	11.63	11.81
½ N	21.33	20.85	12.11	13.02
1 N	21.46	21.82	13.11	13.39
$LSD_{0.05}$	1.12	0.94	0.29	0.31
\mathbf{B}_1	19.14	20.30	11.87	11.95
B_2	21.02	21.56	12.38	12.50
B_3	22.25	22.72	13.50	13.70
B_4	21.16	22.05	13.05	12.97
B_5	19.14	19.79	11.76	11.90
LSD _{0.05}	0.85	0.94	0.45	0.46

 $1 N_0$ = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}N = 50\%$ of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 10: The interaction effect of nitrogen and biofertilizers on some cluster characteristics of Flame seedless grapevine in 2003 and 2004 seasons

		2003 season		2004 season		
Treatments		Cluster length (cm)	Cluster width (cm)	Cluster length (cm)	Cluster width (cm)	
Nitrogen	biofertilizers					
0	\mathbf{B}_1	18.30	11.30	18.82	11.62	
	B_2	20.30	11.55	21.07	11.95	
	\mathbf{B}_3	21.37	12.20	21.97	12.55	
	B_4	20.40	12.10	20.97	11.47	
	B_5	17.02	11.10	17.40	11.47	
1/2	\mathbf{B}_1	21.20	12.20	20.32	12.62	
	B_2	21.80	13.10	22.47	13.30	
	\mathbf{B}_3	22.97	14.60	23.40	14.72	
	B_4	22.30	14.10	22.97	14.22	
	\mathbf{B}_{5}	20.47	11.62	20.57	12.07	
1	B_1	21.47	12.1	20.22	12.40	
	B_2	21.90	12.50	21.15	12.87	
	\mathbf{B}_3	22.40	13.70	22.80	13.82	
	B_4	22.32	13.10	22.22	13.22	
	\mathbf{B}_{5}	22.20	12.60	22.72	12.80	
LSDaar	1 47	0.78	1.63	0.81		

However, the least cluster width was recorded by the control for both cultivars. The enhancement of vine growth and nutritional status certainly reflected their effect on improving yield as well as number of clusters / vine and cluster weight. Another interpretation of the positive role of N is in its beneficial effect of N in raising the number of reproductive shoots and berry set%. The present results are in agreement with those obtained by Gobara [28] and Ali – Mervet [27]. However, the effect of biofertilizers was studied by Papric [29], James [30] and Ahmed et al. [31, 32] who worked on Nitrobeine and active dry yeast. They reported that, the positive action of Nitrobeine in improving vine productivity may be attributed to reducing plant requirements of N and improving the availability of various nutrients. They also supported the effect of active dry yeast on growth, through its basic functions i.e. carbon dioxide production and formation of natural hormone namely cytokinins

which after its decomposition, it develops a wide group of amino acids and B vitamins.

Effect of Nitrogen and Biofertilizers on Some Chemical Properties of Berries

Total Soluble Solids (T.S.S%): Data in Tables 11 and 13 show the effect of nitrogen dose and biofertilizers on T.S.S% of Thompson seedless and Flame seedless grapes. The highest values of T.S.S% were obtained by vines received 100% of the recommended nitrogen in both seasons. Concerning the effect of biofertilizers, it is clear that the highest values of T.S.S% were recorded by vines received Biogen for both cultivars. The interaction between the two studied factors was significant as shown in Tables 12 and 14. The highest value of T.S.S was given by vines received 50% of the recommended nitrogen plus Biogen. However, the least value of T.S.S was recorded by the control.

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Table 11: Effect of nitrogen and biofertilizers on T.S.S, titratable acidity and T.S.S / acid ratio of Thompson seedless grapevine cultivar in 2003 and 2004 seasons

	T.S.S (%)		Total acidity (%	5)	T.S.S / acid rat	tio
Treatments	2003	2004	2003	2004	2003	2004
$\overline{N_0}$	17.83	18.30	0.65	0.66	27.49	27.86
½ N	18.32	18.55	0.61	0.63	29.74	29.25
1 N	18.59	18.82	0.60	0.61	30.67	30.42
$LSD_{0.05}$	0.20	0.20	0.0082	0.012	0.30	0.69
\mathbf{B}_1	18.09	18.68	0.64	0.66	27.56	28.15
B_2	18.24	18.77	0.62	0.64	29.20	28.97
B_3	18.76	19.06	0.58	0.60	32.25	31.60
B_{4}	18.38	18.95	0.61	0.63	29.88	30.00
\mathbf{B}_{5}	17.80	18.50	0.65	0.67	27.60	27.15
LSD _{0.05}	0.37	0.18	0.019	0.018	0.91	0.87

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}N = 50\%$ of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 12: The interaction effect of nitrogen and biofertilizers on T.S.S, titratable acidity and T.S.S / acid ratio of Thompson seedless grapevine in 2003 and 2004 seasons

		2003 season			2004 season		
Treatments		T.S.S (%)	Total acidity (%)	T.S.S / acid ratio	T.S.S (%)	Total acidity (%)	T.S.S / acid ratio
Nitrogen	biofertilizers						
0	\mathbf{B}_1	17.5	0.66	26.20	18.50	0.68	27.15
	B_2	17.80	0.65	27.30	18.65	0.67	27.60
	\mathbf{B}_3	18.40	0.60	30.60	19.00	0.62	30.62
	B_4	18.00	0.64	27.90	18.80	0.66	28.22
	\mathbf{B}_{5}	17.50	0.68	25.40	18.20	0.70	25.70
1/2	\mathbf{B}_{1}	17.97	0.62	28.60	18.87	0.64	29.12
	B_2	18.60	0.61	30.50	18.95	0.63	30.07
	\mathbf{B}_3	19.20	0.56	34.10	19.25	0.58	32.75
	B_4	18.90	0.59	31.80	19.07	0.60	31.50
	\mathbf{B}_{5}	18.20	0.63	28.50	18.50	0.66	27.80
1	\mathbf{B}_{1}	17.95	0.64	27.90	18.67	0.66	28.20
	B_2	18.30	0.61	29.80	18.72	0.64	29.25
	\mathbf{B}_3	18.70	0.58	31.90	18.95	0.60	31.41
	B_4	18.25	0.61	29.90	18.97	0.62	30.27
	B_5	18.50	0.64	28.80	18.62	0.66	27.97
LSD _{0.05}	0.64	0.032	1.57	0.28	0.10	1.50	

Table 13: Effect of nitrogen and biofertilizers on T.S.S, titratable acidity and T.S.S / acid ratio of Flame seedless grapevine cultivar in 2003 and 2004 seasons

Treatment	T.S.S (%)		Total acidity (%	(6)	T.S.S / acid ratio	
	2003	2004	2003	2004	2003	2004
$\overline{N_0}$	18.01	18.34	0.76	0.74	23.52	28.10
½ N	18.69	18.81	0.72	0.70	26.22	29.20
1 N	19.15	19.31	0.68	0.66	27.66	30.20
$LSD_{0.05}$	0.35	0.32	0.032	0.034	1.33	0.95
\mathbf{B}_1	18.60	18.70	0.74	0.70	25.94	30.08
B_2	18.70	18.60	0.74	0.70	25.09	30.57
B_3	19.10	19.25	0.69	0.68	27.46	30.50
B_4	18.90	19.14	0.71	0.68	26.40	30.41
B_5	18.00	18.30	0.78	0.75	24.09	29.75
$LSD_{0.05}$	0.45	0.38	0.03	0.08	1.60	1.12

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 14: The interaction effect of nitrogen and biofertilizers on T.S.S, titratable acidity and T.S.S / acid ratio of Flame seedless grapevine in 2003 and 2004

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	2003 season			2004 season			
Treatments		Total acidity (%)	T.S.S / acid ratio	T.S.S (%)	Total acidity (%)	T.S.S / acid ratio	
biofertilizers							
\mathbf{B}_1	17.90	0.78	22.90	18.17	0.76	30.75	
\mathbf{B}_2	18.00	0.76	23.60	18.32	0.74	30.50	
\mathbf{B}_3	18.40	0.72	25.40	18.75	0.70	30.25	
B_4	18.20	0.75	24.10	18.72	0.72	30.75	
\mathbf{B}_{5}	17.70	0.82	21.40	17.90	0.79	29.75	
\mathbf{B}_{1}	18.72	0.71	26.30	19.30	0.69	29.75	
\mathbf{B}_2	18.92	0.71	27.50	19.35	0.68	30.50	
\mathbf{B}_3	19.77	0.67	29.20	20.02	0.65	30.75	
B_4	19.12	0.69	29.00	19.47	0.67	30.00	
\mathbf{B}_{5}	18.30	0.76	24.00	18.72	0.74	29.75	
\mathbf{B}_1	18.32	0.71	25.90	18.82	0.68	29.75	
\mathbf{B}_2	18.50	0.71	25.20	18.55	0.69	29.50	
\mathbf{B}_3	19.05	0.67	27.70	19.22	0.66	30.00	
B_4	18.88	0.69	27.50	19.07	0.66	29.75	
\mathbf{B}_{5}	18.53	0.70	25.80	18.97	0.68	30.00	
0.77	0.045	2.78	0.65	0.045	1.95		
	biofertilizers B ₁ B ₂ B ₃ B ₄ B ₅ B ₁ B ₂ B ₃ B ₄ B ₅ B ₁ B ₂ B ₃ B ₄ B ₅ B ₁ B ₂	T.S.S (%) biofertilizers B ₁ 17.90 B ₂ 18.00 B ₃ 18.40 B ₄ 18.20 B ₅ 17.70 B ₁ 18.72 B ₂ 18.92 B ₃ 19.77 B ₄ 19.12 B ₅ 18.30 B ₁ 18.32 B ₂ 18.50 B ₃ 19.05 B ₄ 18.88 B ₅ 18.53		$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	

Titratable Acidity: As shown in Tables 11 and 13, increasing nitrogen fertilizer doses caused a significant reduction in titratable acidity of both Thompson seedless and Flame seedless. The highest value of titratable acidity was obtained by vines receiving zero nitrogen in both seasons. As for the effect of biofertilizers, it was clear that they significantly decreased titratable acidity. The highest value of titratable acidity was recorded by untreated vines. The interaction between the two studied factors was significant as shown in Tables 12 and 14. The highest value of titratable acidity was given by vines of the control. However, the least value of titratable acidity was obtained by vines received 50% of the recommended nitrogen plus Biogen.

T.S.S / Acid Ratio: Data in Tables 11 and 13 show the effect of mineral nitrogen and biofertilizers on T.S.S / acid ratio of both Thompson seedless and Flame seedless grapes during 2003 and 2004 seasons. The highest value of T.S.S / acid ratio was obtained by vines given 100% of the recommended nitrogen. As for the effect of biofertilizers, it was obvious that Biogen gave the highest value of T.S.S / acid ratio in both seasons for Thompson seedless cultivar. However, for Flame seedless cultivar both Biogen and active dry yeast gave the highest values of T.S.S / acid ratio in both seasons. The interaction between the two studied factors was significant as shown in Tables 12 and 14. The highest value of T.S.S / acid ratio was obtained by vines received 50% of the recommended nitrogen plus Biogen.

However, the least value of T.S.S / acid ratio was obtained by vines of the control. The reduction of total soluble solids and increment of the total acidity in response to the application of N at higher rates might be ascribed to the great depletion of the total carbohydrates which makes them unavailable for the stimulation of ripening. These results are in harmony with those obtained by Abha *et al.* [33] and Ahmed *et al.* [34]. On the other hand, the positive action of biofertilizers on the quality of the berries could be attributed to their effect on increasing carbohydrates and accelerating cluster ripening. These results are coincided with those obtained by El-Sayed [6] who worked on the bioform of N and Ahmed *et al.* [35].

Effect of Nitrogen and Biofertilizers on N, P and K Leaf Content: It was obvious from the data in Tables 15 and 17 that, N content of both Thompson seedless and Flame seedless leaf significantly increased with increasing N rate in both seasons. The high value of leaf N, P and K content in both seasons was recorded by vines received 100% of the recommended doses. As for the effect of Biofertilizers, in general, the highest value of leaf N, P and K content was recorded by vines receiving Biogen followed in a descending order by active dry yeast, Rhizobacterine and Nitrobeine in both seasons. As for the interaction between the two studied factors it was significant as shown in Tables 16 and 18. The high value of leaf N and P and K content was recorded by vines receiving Biogen plus 100% of the recommended nitrogen,

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Table 15: Effect of nitrogen and biofertilizers on leaf content of N, P and K of Thompson seedless grapevine in 2003 and 2004 seasons

	N (%)		P (%)		K (%)	
Treatment	2003	2004	2003	2004	2003	2004
$\overline{N_0}$	1.301	1.411	0.143	0.150	1.114	1.190
½ N	1.587	1.688	0.213	0.224	1.243	1.254
1 N	1.705	1.802	0.248	0.259	1.268	1.287
$LSD_{0.05}$	0.017	1.019	0.009	0.009	0.015	0.05
\mathbf{B}_1	1.469	1.567	0.192	0.201	1.188	1.215
\mathbf{B}_2	1.537	1.636	0.205	0.208	1.207	1.272
\mathbf{B}_3	1.640	1.737	0.223	0.237	1.255	1.285
B_4	1.585	1.693	0.209	0.223	1.234	1.257
\mathbf{B}_{5}	1.421	1.534	0.176	0.185	1.156	1.189
LSD _{0.05}	0.021	0.030	0.009	0.011	0.014	0.058

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, D_5 = 100% of the recommended nitrogen

Table 16: The interaction effect of nitrogen and biofertilizers on leaf content of N, P and K of Thompson seedless grapevine in 2003 and 2004 seasons

		2003 season			2004 season		
Treatments		N (%)	P (%)	K (%)	N (%)	P (%)	K (%)
Nitrogen	Biofertilizers						
0	\mathbf{B}_1	1.252	0.132	1.100	1.352	0.140	1.147
	B_2	1.302	0.150	1.120	1.407	0.145	1.290
	\mathbf{B}_3	1.390	0.165	1.160	1.507	0.175	1.200
	B_4	1.355	0.150	1.140	1.462	0.162	1.185
	\mathbf{B}_{5}	1.205	0.117	1.050	1.325	0.130	1.127
1/2	\mathbf{B}_1	1.502	0.205	1.220	1.600	0.210	1.137
	\mathbf{B}_2	1.605	0.210	1.242	1.702	0.222	1.250
	\mathbf{B}_3	1.722	0.230	1.290	1.805	0.255	1.302
	B_4	1.652	0.222	1.272	1.755	0.235	1.282
	\mathbf{B}_{5}	1.452	0.197	1.190	1.577	0.200	1.200
1	\mathbf{B}_1	1.652	0.240	1.245	1.750	0.255	1.260
	\mathbf{B}_2	1.605	0.255	1.260	1.800	0.257	1.277
	\mathbf{B}_3	1.810	0.275	1.317	1.900	0.282	1.355
	B_4	1.750	0.255	1.290	1.862	0.272	1.305
	\mathbf{B}_{5}	1.607	0.215	1.230	1.700	0.227	1.240
LSD _{0.05}	0.037	0.016	0.024	0.013	0.018	0.005	

Table 17: Effect of nitrogen and biofertilizers on leaf content of N, P and K of Flame seedless grapevine in 2003 and 2004 seasons

	N (%)		P (%)		K (%)	
Treatment	2003	2004	2003	2004	2003	2004
$\overline{N_0}$	1.284	1.354	0.182	0.132	1.102	1.145
½ N	1.566	1.630	0.201	0.206	1.230	1.228
1 N	1.680	1.752	0.244	0.240	1.259	1.266
$LSD_{0.05}$	0.015	0.017	N.S	0.011	0.005	0.011
\mathbf{B}_1	1.446	1.522	0.265	0.180	1.176	1.186
\mathbf{B}_2	1.519	1.581	0.190	0.192	1.197	1.210
\mathbf{B}_3	1.616	1.681	0.218	0.222	1.246	1.257
\mathbf{B}_4	1.565	1.637	0.203	0.205	1.224	1.238
\mathbf{B}_5	1.403	1.470	0.169	0.161	1.145	1.173
LSD _{0.05}	0.018	0.027	N.S	0.007	0.017	0.013

 N_0 = Zero nitrogen, B_1 = Nitrobeine, B_2 = Rhizobacterine, B_3 = Biogen, B_4 = Active dry yeast, $\frac{1}{2}$ N = 50% of the recommended nitrogen, B_5 = the control, 1 N = 100% of the recommended nitrogen

Table 18: The interaction effect of nitrogen and biofertilizers on leaf content of N, P and K of Flame seedless grapevine in 2003 and 2004 seasons

		2003 season			2004 season		
Treatments		N (%)	P (%)	K (%)	N (%)	P (%)	K (%)
Nitrogen	biofertilizers						
0	\mathbf{B}_1	1.232	0.370	1.087	1.300	0.120	1.132
	B_2	1.282	0.127	1.110	1.352	0.132	1.140
	\mathbf{B}_3	1.382	0.160	1.147	1.452	0.165	1.182
	B_4	1.330	0.145	1.130	1.397	0.140	1.160
	\mathbf{B}_{5}	1.192	0.110	1.037	1.270	0.102	1.112
1/2	B_1	1.482	0.192	1.212	1.557	0.192	1.182
	B_2	1.587	0.205	1.230	1.650	0.200	1.232
	B_3	1.682	0.220	1.282	1.742	0.232	1.280
	B_4	1.642	0.210	1.260	1.712	0.222	1.260
	\mathbf{B}_{5}	1.435	0.180	1.180	1.487	0.182	1.185
1	B_1	1.625	0.235	1.230	1.710	0.230	1.245
	B_2	1.687	0.240	1.252	1.742	0.245	1.260
	B_3	1.785	0.275	1.310	1.850	0.270	1.310
	B_4	1.722	0.255	1.282	1.802	0.255	1.295
	\mathbf{B}_{5}	1.582	0.217	1.220	1.655	0.200	1.222
LSD _{0.05}	0.031	0.181	0.096	0.001	0.040	0.023	

followed by active dry yeast plus 100% of the recommended nitrogen respectively. On the other hand, the least value of leaf N content was recorded by vines of the control. These results were in agreement with those obtained by Wasnik and Bhargava [12] on Thompson seedless grapevines and Li et al. [36], on grapefruit. They mentioned that, increasing the rate of nitrogen fertilization caused a direct increase in the leaf nitrogen percentages. Regarding the effect of biofertilizers, data showed that all Biofertilizers treatments resulted in a significant increase in leaf N content than the untreated vines. This increment may be due to the action of biofertilizers converting the gaseous atmospheric nitrogen to the available form for plants. The highest value of leaf N content was recorded by vines fertilized with Biogen followed in descending order by active dry yeast, Rhizobacterine and Nitrobeine. The positive effect of biofertilizers in increasing nitrogen content was observed by Ahmed et al. [16], on grapevines, Mansour [37], on Anna apples, Samah [4] on Thompson seedless grapevines. Leaf phosphorus and potassium increment as a result of N treatments may be due to the effect of application in improving vegetative growth and root growth which means more absorption of nutrients from the soil. These results were in agreement with those obtained by El-Shazly and Abdel-Nasser [38] on Flame seedless grapevines, Abdel-Naby and Gomaa [39], on Balady mandarin and Taha et al. [40], on banana plants. The positive action of biofertoilizers can be interpreted by the results of Rodelas et al. [41] who suggested that plant

hormones released by microorganisms increase plant root growth causing, in turn, an increase in the plant root surface which improves nutrient absorption. In this respect, Bhardwi *et al.* [42] mentioned that the production of antimicrobial substances responsible for reducing plant root infection with pathogens make the plants more healthy and consequently increase their nutrient uptake.

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