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Effect of Spraying Chitosan and Calcium Silicate on Yield and Berry Quality of Black Monukka Grapevines

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Abstract: This two-year trial conducted during 2021 and 2022 experimental seasons in a vineyard located at Tamay, Minia governorate, was investigating the effect of spraying different concentrations of chitosan ($C_{56}H_{103}N_9O_{39}$) at 500 and 1000 ppm and calcium silicate (Ca_2SiO_4) at 0.1 and 0.2 % alone or combined with each other on yield and berry quality of Black Monukka grapevines. Ten-year-old vines grown in sandy soil, spaced at (2.0 x 3.0 m.), drip irrigated and trellised by Spanish parron were used in this trial. The results of the experimental treatments revealed that the higher concentration of chitosan at 1000 ppm + calcium silicate at 0.2 % treatment was the best, positively affecting all growth parameters, yield, chemical characteristics of berries and leaves along with improving berry firmness, adherence and berry shattering % followed by the lower concentration of chitosan at 500 ppm + calcium silicate at 0.1 % over the other treatments and the control.

Key words: Black Monukka cv. · Chitosan · Calcium silicate · Yield · Berry quality

INTRODUCTION

Black Monukka is one of the table grape cultivars which holds a significant promise for commercial purpose. The production of loose clusters and high berry shattering are negatively reflected on productivity [1].

However, direct application of calcium at pre-veraison and post-veraison phases were found to increasing berry firmness and breaking force [2]. Therefore, plant Ca requirements must be continually obtained from external sources. It can be said that the most effective grapevine treatment increasing berry Ca concentration is foliar application.

It was found in a study done by Hocking [3] on the effects of high and low calcium supply in grapevines that low Ca showed early berry softening and berry weight loss while high Ca treatment delayed the berry development. These results indicated that in low calcium grown berries, berry weight loss was due to higher post-veraison berry transpiration. In opposite, high calcium grown vines reduced transpiration and net assimilation rates compared to low calcium grown vines.

Recently, chitin and its derivatives like chitosan have shown great effect in being an alternative to mineral fertilizers due to their natural properties. Chitosan provide coating to the skin of fruits which helps to reduce transpiration and control weight loss to slow down ripening and expand shelf life by controlling respiration rate and ethylene production [4].

Chitosan is a natural compound prepared mainly from chitin, which is the main component of the skeleton of crustaceans and it is used as a cheap polymer non-toxic and safe for health [5]. Many researchers have recently urged to the use of chitosan for the agricultural and horticultural purposes, primarily for plant defense, for yield increase and enhancement of the plant development, as this glucosamine polymer influences the biochemistry of the plant cell [6 , 7].

MATERIALS AND METHODS

The present investigation has been carried out during the two successive growing seasons of 2021and 2022 in a vineyard located at Tamay, Minia governorate 28.1003° N and 30.7582° E. Ten-year-old Black Monukka grapevines spaced at 2 x 3 m. grown in a sandy soil were used in this investigation. Vines were cane pruned and trellised by Spanish parron system with a bud load of 96 buds/vine [8 canes x 12 buds]. Pruning was carried on the 15 of January and irrigated via a drip irrigation system. Sixty-three uniform vines were chosen for this study (7 treatments x 3 replicates x 3 vines/replicate).

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The experiment was carried out on the same vines for both seasons and were carefully selected to be nearly uniform in vigor as possible and received common horticultural practices recommended by the Ministry of Agriculture. All vines except for the control were sprayed twice during the growing season, the 1st at shoot length 15 cm and the 2nd after berry set. The following treatments were adopted with respect to spraying treatments for both seasons as follow:

- Control
- Chitosan at 500 ppm
- Chitosan at 1000 ppm
- Calcium silicate at 0.1%
- Calcium silicate at 0.2 %
- Chitosan at 500 ppm + calcium silicate at 0.1 %
- Chitosan at 1000 ppm + calcium silicate at 0.2 %

Measurements

Yield and its Components: Representative random samples of 15 clusters /treatment [5 cluster from each replicate] were collected when clusters reached their full color and total soluble solids reached about 16-19 Brix, according to Badr and Ramming [8].

- Yield per vine (kg)
- Average cluster weight (g)
- Average berry weight (g)
- Average berry size (cm^3)
- Berry firmness and adherence (g/cm^2) by using PHSH-PULL (Dynamometer Model DT101).
- Berry shattering %: Shatter potential was measured by dropping clusters from a standard height and percentage of shattered berries per cluster was recorded by Dokoozlian [9].

Chemical Characteristics of Berries:

- Total soluble solids (TSS %) and titratable acidity as gram of tartaric acid per 100 ml of juice were determined according to AOAC [10] then TSS / acid ratio was calculated.
- Total anthocyanin in berry skin (mg/100g) using spectrocolourimeter at 250 μm according to Yildiz and Dikmen[11].

Vegetative Growth Parameters:

- Leaf area (cm^2): Samples of 30 leaves taken from the fifth and the sixth positions from the apex were randomly collected from each treatment from the fruiting shoots for leaf area determination at harvest (using leaf area meter, Model CI 203, U.S.A.).
- Shoot length (cm): it was determined by measuring the fruiting shoots at the growth cessation.
- Total chlorophyll content (SPAD) were measured at harvest in the mature leaves of the fifth and the sixth positions from the apex from the fruiting shoots by using the nondestructive Minolta chlorophyll meter model SPAD 502 [12].
- Percentage of N, P, K and Ca content in leaf petioles: were estimated in the leaf opposite to the cluster at full bloom, N (%) content were measured according to Hesse [13]; P % was measured referring to the method of Schouwenburg and Walinga [14]; K % was determined using a flame photometer [15] and Ca % by atomic absorption spectrometry [16].

Statistical Analysis: The complete randomized block design was adopted for this experiment. The statistical analysis of the present data was carried out according to Snedecor and Cochran [17]. Averages were compared using the new L.S.D. values at 5% level.

RESULTS AND DISCUSSION

Yield

Average Yield/vine (Kg), Average Cluster Weight (g), Average Berry Weight (g) and Average Berry Size (cm^3): Data of the average yield per vine and its attributes as affected by all treatments are clearly displayed in Table (1) showing significant differences between them and the control. Generally, all parameters in term of average yield/vine, average cluster weight, average berry weight and average berry size increased by treating the vines with the combined treatments, single treatments and the control respectively, with a superiority of the higher concentrations. Results showed that the combined treatment of Chitosan at 1000 ppm + Calcium silicate at 0.2 % significantly produced the highest yield followed by the lower concentration at 500 ppm chitosan and 0.1 % Calcium silicate in both seasons. It is also noticeable that cluster weight, berry weight and average berry size followed that same trend. This detection is being linearly related to those of Al-ahmadi [18] who reported that chitosan stimulates plant growth by enhancing cell division similar to gibberellins. In addition, increasing concentrations of chitosan caused a progressive promotion on yield and cluster weight of Flame seedless grapevines [19]. In another trial done by Bedrech and Farrouh [20] it was stated that chitosan at 5 cm^3/ L has been shown an increase in average yield, cluster weight and berry size in relation to the control.

Table 1: Effect of different concentrations of chitosan and calcium silicate on yield and berry quality of Black Monukka grapevines during the two successive seasons 2021 and 2022

Treatments	Average yield (Kg)		Cluster weight (g)		Berry weight (g)		Berry size (cm³)	
	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022
Control	22.7	23.0	590.7	593.0	3.4	3.5	3.2	3.4
Chitosan at 500 ppm	25.5	26.3	628.8	633.5	4.1	4.1	3.9	4.0
Chitosan at 1000 ppm	26.2	26.9	645.0	639.4	4.3	4.4	4.2	4.3
Calcium silicate at 0.1%	24.4	24.9	608.6	610.5	3.6	3.6	3.5	3.4
Calcium silicate at 0.2 %	25.2	25.5	620.9	626.6	3.7	3.8	3.6	3.7
Chitosan at 500 ppm + Calcium silicate at 0.1 %	26.8	27.5	663.3	665.9	4.6	4.8	4.4	4.5
Chitosan at 1000 ppm + Calcium silicate at 0.2 %	27.5	28.2	686.7	698.4	4.9	5.1	4.6	4.7
New L.S.D at 5%	0.4	0.5	11.2	12.3	0.1	0.1	0.1	0.1

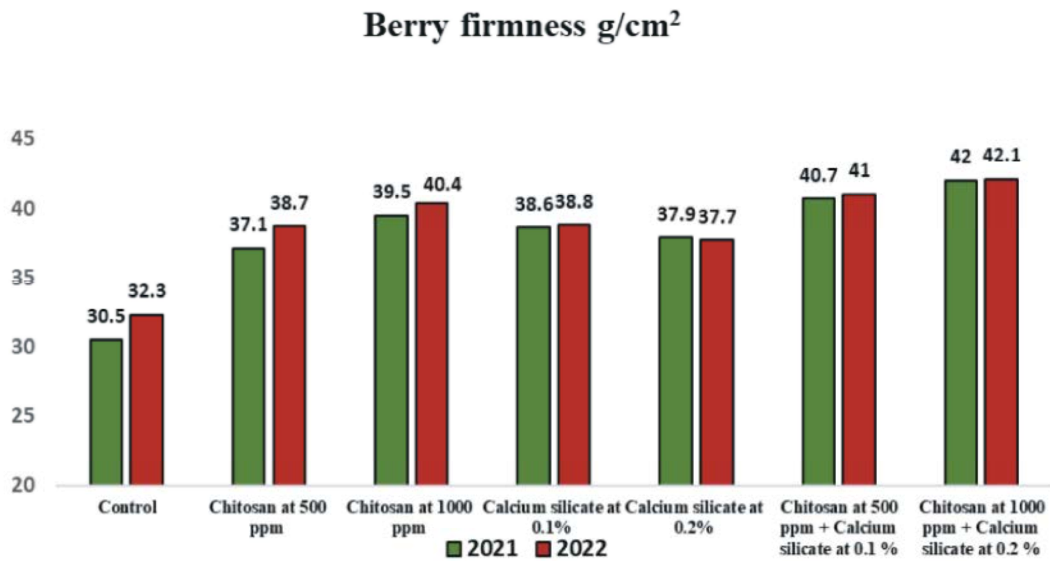

Fig. 1: Effect of different concentrations of chitosan and calcium silicate on berry firmness g/cm² of Black Monukka grapevines during the two successive seasons 2021 and 2022

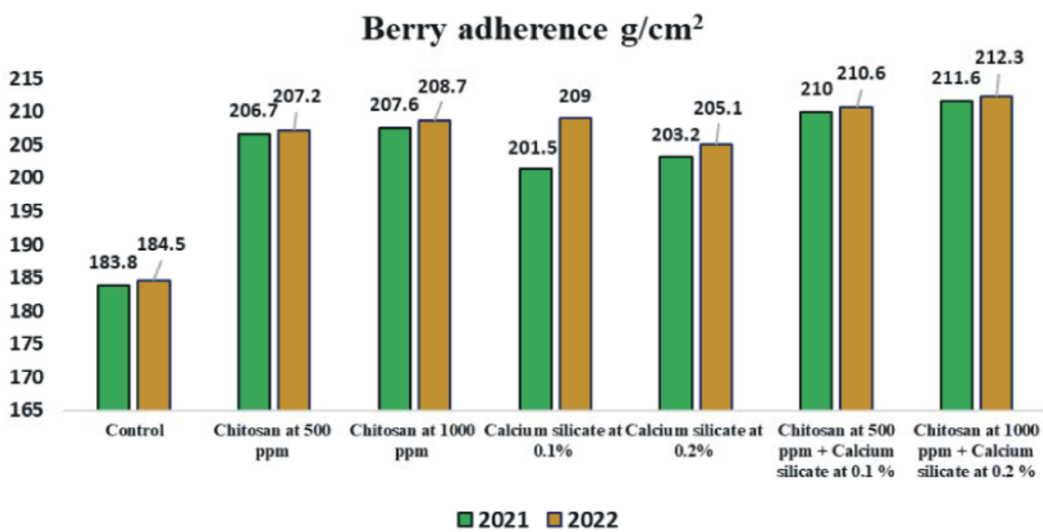

Fig. 2: Effect of different concentrations of chitosan and calcium silicate on berry adherence g/cm² of Black Monukka grapevines during the two successive seasons 2021 and 2022

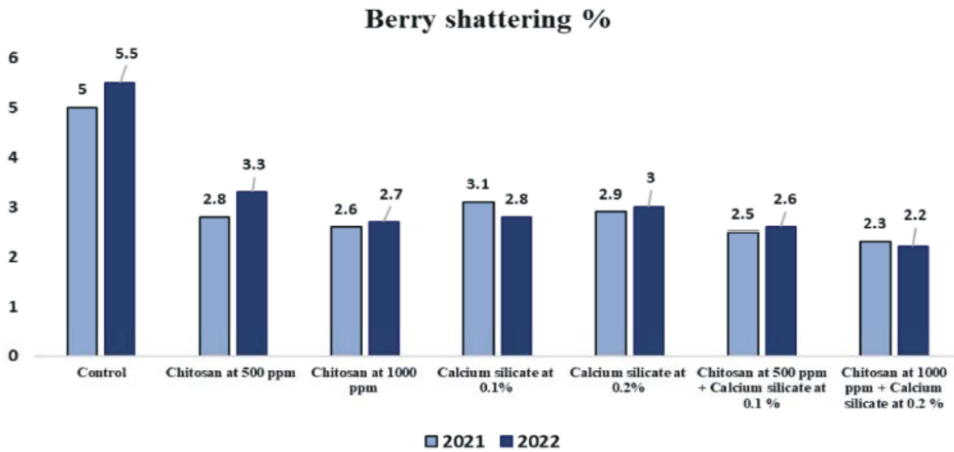

Fig. 3: Effect of different concentrations of chitosan and calcium silicate on berry shattering % of Black Monukka grapevines during the two successive seasons 2021 and 2022

Likewise, Bonomelli and Ruiz [21] mentioned that yield components including berry weight, cluster weight and thus total yield per vine were significantly influenced by the application of Ca sprays on leaves. In addition, previous studies have shown that potassium silicate applied on "Early sweet" grapevines enhanced yield/vine and average cluster weight [22].

Berry Firmness (g/cm²), Adherence (g/cm²) and Berry Shattering (%): In both years of the study, it was observed an improving in berry firmness, berry adherence and berry shattering % due to spraying both chitosan and calcium silicate in all treatments over the control (Figure 1, 2 and 3). Generally, treating the vine with the higher concentrations are more effective. However, best results were obtained from spraying the vine with Chitosan at 1000 ppm + Calcium silicate at 0.2 %. This result may be ascribed to silicon affecting activities of major cell wall from degrading enzymes such as poly galacturonase, cellulose and xylanase. Similarly, previous studies have shown that the silicon is benefits confers on plants, that enhanced berry firmness [23].

In respect to calcium effect on berry firmness Amiri *et al.* [24] mentioned that berry firmness was significantly influenced positively by Ca sprays. Additionally, it was stated that chitosan increased berry adherence and reduced berry shattering % [25, 26].

Chemical Characteristics of Berries

Total Soluble Solids (TSS %), Titratable Acidity (TA %), TSS / Acid Ratio and Total Anthocyanin: It was observed from the data in Table 2 that total soluble solids %, TSS / acid ratio and total anthocyanin recorded a significant increase, whereas titratable acidity % was reduced due to spraying Chitosan at 1000 ppm + Calcium silicate at 0.2 % followed by the lower concentration Chitosan at 500 ppm + Calcium silicate at 0.1 % in both seasons. Results are in accordance to previous studies which showed that chitosan sprays increased significantly TSS % while reducing total acidity % in addition to increasing the anthocyanins in berry skins [25, 27].

Likewise, some studies revealed that foliar applications of silicon significantly increased totat soluble sugars content and the of total soluble solids to titratable acidity ratio [28].

Vegetative Growth Parameters

Leaf Area (cm²) and Shoot Length (cm): It is obvious from the data displayed in (Table 3) that there are significant differences among treatments in the physical characteristics of the vegetative growth parameters in term of leaf area and shoot length. The stimulation of growth was proportional to the increase in the concentration where the highest values were recorded for vines that treated with the combined treatment of Chitosan at 1000 ppm + Calcium silicate at 0.2 % followed by the lower concentration Chitosan at 500 ppm + Calcium silicate at 0.1 % in both seasons. Similarly, in another trial El-Kenawy [25] found that the results of the single application of chitosan or in combination with calcium silicate were effective in increasing shoot length and leaf surface area, in both seasons. Moreover, it was informed that calcium plays an important role in cell division, as well as in the growth and development of fruit trees [29] which led in turn to increasing leaf area and shoot length.

Table 2: Effect of different concentrations of chitosan and calcium silicate on Total soluble solids (TSS %), titratable acidity (TA %), TSS / acid ratio and total anthocyanin of Black Monukka grapevines during the two successive seasons 2021 and 2022

	TSS (%)		TA (%)		TSS / acid Ratio		Anthocyanin mg/100 g. FW	
Treatments	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022
Control	15. 3	15.5	0.67	0.64	22.8	24.2	34.5	36.9
Chitosan at 500 ppm	17.0	17.1	0.53	0.52	32.0	32.8	42.0	42.3
Chitosan at 1000 ppm	17.5	17.6	0.50	0.49	35.0	35.9	42.6	42.8
Calcium silicate at 0.1%	16.0	16.1	0.60	0.57	26.6	28.2	40.4	41.0
Calcium silicate at 0.2 %	16.4	16.6	0.56	0.54	29.3	30.7	41.6	41.8
Chitosan at 500 ppm + Calcium silicate at 0.1 %	18.0	18.2	0.48	0.47	37.5	38.7	43.2	43.3
Chitosan at 1000 ppm + Calcium silicate at 0.2 %	18.6	18.7	0.46	0.45	40.4	41.5	43.7	43.9
New L.S.D at 5%	0.3	0.4	0.01	0.01	1.5	1.8	0.3	0.4

Table 3: Effect of different concentrations of chitosan and calcium silicate on leaf area (cm^2) and shoot length (cm) of Black Monukka grapevines during the two successive seasons 2021 and 2022.

	Leaf area (cm^2)		Shoot length (cm)	
Treatments	Season 2021	Season 2022	Season 2021	Season 2022
Control	22.5	23.1	143.5	144. 3
Chitosan at 500 ppm	25.2	25.7	154.0	156.8
Chitosan at 1000 ppm	26.4	27.8	157.8	160.7
Calcium silicate at 0.1%	23.3	24.1	147.0	148.7
Calcium silicate at 0.2 %	24.1	25.0	150.1	152.2
Chitosan at 500 ppm + Calcium silicate at 0.1 %	28.7	29.1	163.7	164.6
Chitosan at 1000 ppm + Calcium silicate at 0.2 %	30.1	32.3	167.5	169.0
New L.S.D at 5%	0.8	0.9	3.0	3.1

Total Chlorophyll Content (SPAD): Chlorophyll is an important photosynthetic pigment to the plant, largely determining photosynthetic capacity and hence plant growth. Significant differences between all treatments receiving chitosan and calcium silicate were observed according to the degree of concentration whereby the higher the chitosan and calcium silicate concentration the higher the chlorophyll content (Table 4). The application of Chitosan at 1000 ppm + Calcium silicate at 0.2 % showed an increase in leaf chlorophyll content over all other treatments in both seasons. Similar results were obtained from Sheikha and Al-Malki [30] revealing that treated the vines with chitosan has significantly enhanced the photosynthetic pigments concentration in leaves and activated the synthesis of carotenoids which in turn protect chlorophyll from oxidation and increase its content. Regarding the role of calcium silicate it was stated in a trial done by El-Kenawy [25] that the single application of chitosan or in combination with calcium silicate were effective in improving shoot length and leaf surface area of Thompson seedless grapevines.

Leaf Petioles Content of N, P, K and Ca (%): Influence of chitosan and calcium silicate upon leaf chemical characteristics is given in Table (4). However, all treatments increased these parameters significantly compared to the control. The maximum significant content of nitrogen (N%), phosphorus (P%), potassium (K%) and calcium (Ca%) in leaf petiole was recorded with the higher concentration of Chitosan at 1000 ppm + Calcium silicate at 0.2 % treatment. Where the application of chitosan resulted in a progressive increase in these parameters due to role in enhancing the nutrient absorption capacity in plant. These results were related to the work of Mahmoud *et al*. [29] who found that the application of boric acid + chitosan was the treatment responsible for the higher increments in leaf petiole nutrients as compared with untreated vines.

Moreover, calcium leaf contents were even above the optimum values proposed due to spraying the vines with calcium silicate which led to increasing its contents where similar ranges were previously reported by El-Kenawy [25] who found that the results of the single application of chitosan or in combination with calcium silicate were effective in improving the percentages of N, P and K % in the leaf petioles as well as Ca % in both seasons. In another trial, Karimi *et al.* [31] declared that the effect of applying calcium sulfates to grapevine (*Vitis vinifera* L.) cv. Sultana has a positive effect on the content of leaf macro-elements especially, nitrogen and calcium content in leaves which were significantly higher compared to the control.

Table 4: Effect of different concentrations of chitosan and calcium silicate on the total chlorophyll content (SPAD) and the percentage of N, P, K and Ca of Black Monukka grapevines during the two successive seasons 2021 and 2022

	Total chlorophyll (SPAD)		Leaf petiole content							
			N %		P %		K %		Ca %	
Treatments	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season 2022	Season 2021	Season2022
Control	10.7	11.5	1.24	1.27	0.15	0.19	1.16	1.21	2.01	2.08
Chitosan at 500 ppm	12.5	13.2	1.62	1.67	0.32	0.34	1.34	1.40	2.41	2.45
Chitosan at 1000 ppm	13.0	13.8	1.71	1.79	0.37	0.38	1.38	1.45	2.51	2.55
Calcium silicate at 0.1%	11.1	12.3	1.32	1.39	0.25	0.25	1.23	1.29	2.20	2.22
Calcium silicate at 0.2 %	11.9	12.7	1.43	1.48	0.27	0.29	1.27	1.33	2.28	2.33
Chitosan at 500 ppm + Calcium silicate at 0.1 %	13.9	14.3	1.82	1.93	0.40	0.42	1.44	1.48	2.67	2.72
Chitosan at 1000 ppm + Calcium silicate at 0.2 %	14. 5	14.8	1.96	2.02	0.44	0.46	1.47	1.52	2.75	2.81
New L.S.D at 5%	0.5	0.5	0.08	0.09	0.02	0.03	0.01	0.02	0.06	0.08

CONCLUSION

In conclusion, we can deduce that treating the vines by a combination of chitosan at 1000 ppm and calcium silicate at 0.2 % is directly affecting the nutrients rate as well as improving their absorption efficiency. Hence, using chitosan as a coating material for fertilizers can control the release rate of the inorganic added nutrients, resulting in the prevention of excessive fertilization and improving the efficiency of fertilizers' uptake. Obtained results showed that the higher concentration of foliar spray of chitosan and calcium silicate improved yield, all growth parameters as well as enhancing the chemical characteristics of berries and leaves along with berry firmness and adherence and reducing berry shattering %. Therefore, chitosan can be used as a biodegradable biofertilizer to avoid the hazards of inorganic fertilizers overuse in horticulture besides the effect of silicate fertilizer which has a beneficial effect in increasing the grape quality.

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