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# Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on growth, yield and bunch quality of Crimson Seedless grapevines

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## ABSTRACT

This investigation was conducted for two successive seasons (2016 & 2017) to disclose the effect of inoculation with Arbuscular Mycorrhiza fungi (AM) and effective micro-organisms (EM) on growth, yield and bunch quality of Crimson Seedless grapevines. The chosen vines were seven-year-old, grown in a clay loamy soil, in a private vineyard located at Aga, Dakahlia governorate, spaced at 1.75 X 2.5 meters apart and irrigated by flood irrigation system, cane-pruned and trellised by the telephone shape system. The vines were pruned during the first week of March with bud load (72buds/vine).

Eight treatments were applied as follows; control (untreated vines), inoculation with Arbuscular mycorrhiza (AM) at 250g inoculum/vine, inoculation with effective micro-organisms (EM) at 10cm/vine, inoculation with effective micro-organisms (EM) at 20cm/vine, inoculation with effective micro-organisms (EM) at 30cm/vine, inoculation with AM + EM at 10cm/vine, inoculation with AM + EM at 20cm/vine and inoculation with AM + EM at 30cm/vine.

The results showed that inoculation with arbuscular mycorrhiza fungi (AM) and all different tested doses of the effective micro-organisms (EM) had the best results as compared to untreated vines (control) in both seasons. Dual inoculation of AM plus EM at 30cm/vine resulted in the best vegetative growth aspects namely shoot length, number of leaves, leaf area and coefficient of wood ripening, increased leaf content of total chlorophyll and mineral elements including percentages of total nitrogen, phosphorus and potassium, ensured the highest yield and good components and improved bunch quality attributes namely the physical and chemical characteristics of berries of Crimson Seedless grapevines.

***Key words:*** Mycorrhiza fungi, Crimson, grapevines, inoculation, effective micro-organisms

## Introduction

Biofertilizers are commonly called microbial inoculants which are capable of mobilizing important nutritional elements in the soil from non-usable to usable form by the crop plants through their biological processes. During the last decade, biofertilizers have been extensively used as an eco-friendly approach to minimize the use of chemical fertilizers, improve soil fertility status and for the enhancement of crop production by their biological activity in the rhizosphere (Ram Rao *et al*., 2007).

Micro-organisms in the soil play a vital role in nutrients cycling and mediate various processes through their interactions with plats and other soil organisms. The importance of micro-organisms (Fungi, bacteria, actinomycetes and yeast) can hardly be overstated. Yet their presence and activities are often disregarded in conventional agricultural systems, which rely heavily on non-sustainable inputs of energy, fertilizers and pesticide (Zarb *et al*., 1999). Some micro-organisms, particularly beneficial bacteria and fungi can improve plant performance and, consequently, enhance yield (Evelin, *et al*., 2009).

Arbuscular mycorrhiza fungi (AM) are obligatory symbiotic soil fungi, which colonize the roots of most plants (Douds & Millner, 1999). These fungi form mutualistic relationships with more than 80% of terrestrial plants (Ulrich *et al*., 2002) and provide the host with mineral nutrients in exchange for carbohydrates (Tahat *et al*., 2008 and Javaid, 2009). Generally, plants inoculated with AMF are more efficient in water and nutrient acquisition by increasing water uptake (Graham *et al*., 1987) and enhancing absorbtion of phosphours and other relatively immobile micronutrients cations, particularly zinc and copper (Marin *et al*., 2003), thus resulting in an improved plant growth (Oseni *et al*., 2010),

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producing plant growth hormones such as auxines, cytokinines and gibberellins (Zhang *et al*., 2008) and enhancing crop productivity by enhancing tolerance to various biotic and abiotic stress factors (Al-Garni, 2006; Khaosaad *et al*., 2007 and Javaid & Riaz, 2008).

Effective micro-organisms is a mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast, actinomycetes fermenting fungi that can be applied through the inoculation to increase the microbial diversity of the soil, this in turn can improve soil quality and fertility which enhances the growth, yield and quality of crops (Higa and Kinjo, 1991).

Therefore, the main objective of this study was to disclose the effect of inoculation with Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on vegetative growth, yield and bunch quality of Crimson Seedless grapevines.

## Materials and Methods

This investigation was conducted for two successive seasons (2016 & 2017) to disclose the effect of inoculation with Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on growth, yield and bunch quality of Crimson Seedless grapevines. The chosen vines were seven-year-old, grown in a clay loamy soil (Table, 1), in a private vineyard located at Aga, Dakahlia governorate, spaced at 1.75 X 2.5 meters apart and irrigated by flood irrigation system, cane-pruned and trellised by the telephone shape system. The vines were pruned during the first week of March with bud load (72buds/vine).

**Table 1:** Some physical and chemical analysis of the experimental soil

| | | |
|---|---|---|
| Physical | Sand (%) | 34.1 |
| | Silt (%) | 25.6 |
| | Clay (%) | 40.3 |
| | Texture | Clay loam |
| Chemical | Organic carbon (%) | 1.13 |
| | PH (1:25) | 7.74 |
| | EC (ds.m) | 1.23 |
| | Ca $Co_3$ (%) | 2.43 |
| | N (%) | 0.24 |
| | P (%) | 0.11 |
| | K (%) | 0.32 |

Ninety-six uniform vines were chosen on the basis their growth depending on weight of prunings and trunk diameter of the vine as indirect estimates for vine vigour. Each four vines acted as a replicate and each three replicates were treated by one of the following treatments.

Eight treatments were conducted as follows:

1. Control (untreated vines)
2. Inoculation of arbuscular mycorrhiza (AM) at 250 g inoculum/vine
3. Inoculation of effective micro-organisms (EM) at 10cm/vine
4. Inoculation of effective micro-organisms (EM) at 20cm/vine
5. Inoculation of effective micro-organisms (EM) at 30cm/vine
6. AM + EM at 10cm/vine
7. AM + EM at 20cm/vine
8. AM + EM at 30cm/vine

Mycorrhiza spores were originally extracted from Egyptian soils. Soil drench was made around the roots of the vine at the second week of January. Spores of AM-mycorrhiza including the following genera Glomus, Gigaspora and Acaulospora were added after propagation. Extraction and counting of identified mycorrhiza spores occurred according to the method described by (Massoud, 1999) where the soil mass was gently removed from root system of each vine (250g), suspended and then sieved using the wet sieving and decanting technique. Five sieves (400, 250, 150, 75 and 65 mesh size) were used. The remained fractions were transferred into a glass bottle and diluted with water. The number of spores was estimated by spreading certain volume of mycrrhizal spore suspension onto a squared Petri–dish which was divided into squares from the base. The number was recorded using a binocular microscope (30-50X) (Daft and Hogarth 1983).

Mixed spores of AM-mycorrhiza genera via Glomus spp., Gigaspora spp. and Acaulospora spp., were prepared after extraction and mixed with sand as a carrier (40-50 spore/g inoculum) and then added to the soil at once during winter agricultural management at the rate of (100g inoculum/ m long) so each vine 2.5 m around needs 250 g inoculum.

Effective micro-organisms were used as trade mark (EM) constitute of mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast and Streptomycetes.

Components of Effective micro-organisms (EM)

| Total bacterial | Lactic acid bacteria | Yeasts | Streptomycetes |
|---|---|---|---|
| 2.5 – 9.6 X $10^4$ cfu/ml | 6.6 – 9.9 X $10^6$ cfu/ml | $10^5$ - $10^6$ cfu/ml | 8.5 X $10^3$ cfu/ml |

Three doses of the effective micro-organisms application (10, 20 or 30cm/vine) were soil drench application through the four phenological stages: the 1st date (after bud burst stage), the 2nd date (at fruit set stage), the 3rd date (after two weeks of fruit set stage), and the 4th date (veraison stage).

The following parameters were adopted to evaluate the tested treatments:-

*Yield and physical characteristics of bunches:*

Representative random samples of six bunches/vine were harvested at maturity when TSS reached about 16-17% according to Tourky *et al*., (1995). The following characteristics were determined:

Yield/vine (kg) was determined as number of bunches/vine X average bunch weight (g). Also, average bunch weight (g), bunch length and width (cm) were determined.

*Physical characteristics of berries:*

Berry weight (g), berry size ($cm^3$) and berry dimensions (length and diameter) (cm) were determined.

*Chemical characteristics of berries:*

Total soluble solids in berry juice (T.S.S.) (%) by hand refractometer and total titratable acidity as tartaric acid (%) (A.O.A.C. 1985). Hence TSS /acid ratio and total anthocyanin of the berry skin (mg/100g fresh weight) according to Husia *et al*. (1965) were calculated.

*Morphological characteristics of vegetative growth*

At growth cessation, the following morphological studies were conducted on four fruitful shoots/the considered vines.

Average shoot length (cm).

Average number of leaves/shoot.

Average leaf area ($cm^2$) of the apical $5^{th}$ and $6^{th}$ leaves using a CI-203- Laser Area-meter made by CID, Inc., Vancouver, USA.

Total leaf area/vine ($m^2$) was determined by multiplying average number of leaves/shoot by average leaf area then by the number of shoots per vine.

Coefficient of wood ripening was calculated by dividing length of the ripened part of the shoot by the total length of the shoot according to Bouard (1966).

*Leaf content of total chlorophyll and mineral elements.*

Total chlorophyll was measured by using nondestructive Minolta chlorophyll meter SPAD 502 (Wood *et al.,* 1992).

Mineral elements (%) were determined as total nitrogen (%) (Pregl, 1945), phosphorus (%) (Snell and Snell 1967) and potassium (%) (Jackson, 1967).

*Statistical analysis:*

The complete randomized block design was adopted for the experiment. The statistical analysis of the present data was carried out according to Snedecor and Cochran (1980). Averages were compared using the L.S.D. values at 5% level.

**Results and Discussion**

**1. Yield and physical characteristics of bunches**

As shown data in Table (2), it is evident that yield and bunch physical characteristics expressed in bunch weight and dimensions were significantly increased by inoculation with arbuscular mycorrhiza (AM) and all different tested doses of the effective micro-organisms (EM). Dual inoculation of AM plus EM at 30cm attained significantly the highest values of these parameters followed by dual inoculation of AM plus EM at 20cm. On the other hand, control had the lowest values of these ones in both seasons.

Yield produced in response to inoculations could be mainly attributed to the enhancement effect of AM and EM inoculations on bunch weight. The positive effect of AM inoculations can be explained by those mycorrhiza funguses, which are able to absorb and translocate elements to host root tissues, in addition, they can also break down certain complex minerals and organic substances in the soil and make them available to their hosts (Mona, 2001).

**Table 2:** Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on yield and bunch physical characteristics of Crimson Seedless grapes during 2016 and 2017 seasons

| Treatments | Yield/vine (kg) | | Average bunch weight (g) | | Average bunch length (cm) | | Average bunch width (cm) | |
|---|---|---|---|---|---|---|---|---|
| | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 |
| Control (untreated vines) | 12.94 | 13.86 | 457.2 | 476.3 | 23.83 | 24.71 | 16.71 | 16.92 |
| Arbuscular mycorrhiza (AM) at 250g inoculum/vine | 13.47 | 15.05 | 469.5 | 490.1 | 24.15 | 25.07 | 17.09 | 17.29 |
| Effective micro-organisms (EM) at 10cm/vine | 13.16 | 14.21 | 461.8 | 481.7 | 23.92 | 24.83 | 16.82 | 17.04 |
| Effective micro-organisms (EM) at 20cm/vine | 13.29 | 14.46 | 464.6 | 485.2 | 23.97 | 24.89 | 16.89 | 17.11 |
| Effective micro-organisms (EM) at 30cm/vine | 13.33 | 14.72 | 466.2 | 487.4 | 24.08 | 24.94 | 16.98 | 17.23 |
| AM + EM at 10cm/vine | 13.65 | 15.39 | 473.9 | 494.7 | 24.23 | 25.13 | 17.23 | 17.38 |
| AM + EM at 20cm/vine | 13.77 | 15.78 | 478.1 | 499.4 | 24.32 | 25.25 | 17.35 | 17.49 |
| AM + EM at 30cm/vine | 14.02 | 16.17 | 485.2 | 506.8 | 24.38 | 25.32 | 17.44 | 17.61 |
| LSD at 0.05 | 0.23 | 0.31 | 6.7 | 7.1 | 0.05 | 0.06 | 0.07 | 0.11 |

With respect to Effective micro-organisms, it is a mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast, actinomycetes fermenting fungi that can be applied through the inoculation to increase the microbial diversity of the soil, this in turn can improve soil quality and fertility which enhances yield (Higa and Kinjo, 1991).

The obtained results are in harmony with those achieved by several investigators who reported that mycorrhiza fungi increase fruit weight and yield of some grape cultivars; Abd El-Wahab *et al*., (2008) and Rizk-Alla and Tolba (2010) on Black Monukka grapevines and Abd El-Wadoud (2016) on Flame seedless grapevines.

As for the effect of EM, Sabry, *et al*., (2009) on Red Globe grapevines, Abd El-Hameed *et al*., (2010) on Thompson Seedless grapevines and Abd El-Wadoud *et al*., (2014) on Red Globe grapevines pointed out that EM application as soil drench increased significantly bunch weight and yield/ vine.

**2. Physical characteristics of berries:**

Data presented in Table (3) showed that inoculation with arbuscular mycorrhiza (AM) and all different tested doses of the effective micro-organisms (EM) improved significantly the physical characteristics of berries namely average berry weight, average berry size and average berry dimensions i.e. length and diameter. The highest values of these determinations were obtained from the dual inoculation of AM plus EM at 30cm followed by dual inoculation of AM plus EM at 20cm, whereas the lowest values was obtained from control in both seasons.

The positive effect of AM and EM inoculations on berry physical properties could be attributed to that AM inoculation can be explained by those mycorrhiza funguses, which are able to absorb and translocate elements to host root tissues, in addition, they can also break down certain complex minerals and organic substances in the soil and make them available to their hosts (Mona, 2001). With respect to effective micro-organisms is a mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast, actinomycetes fermenting fungi that can be applied through the inoculation to increase the microbial diversity of the soil, this in turn can improve soil quality and fertility which enhances quality of crops (Higa and Kinjo, 1991).

The obtained results are in agreement with those given by Abd El-Wahab *et al*., (2008) and Rizk-Alla and Tolba (2010) on Black Monukka grapevines and Abd El-Wadoud (2016) on Flame seedless grapevines they found that mycorrhiza fungi had the best physical characteristics of berries.

**Table 3:** Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on berry physical characteristics of Crimson Seedless grapes during 2016 and 2017 seasons

| Treatments | Average berry weight (g) | | Average berry size ($cm^3$) | | Average berry length (cm) | | Average berry diameter (cm) | |
|---|---|---|---|---|---|---|---|---|
| | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 |
| Control (untreated vines) | 3.96 | 4.03 | 3.81 | 3.87 | 2.41 | 2.43 | 1.68 | 1.72 |
| Arbuscular mycorrhiza (AM) at 250g inoculum/vine | 4.09 | 4.17 | 3.97 | 4.04 | 2.45 | 2.47 | 1.71 | 1.75 |
| Effective micro-organisms (EM) at 10cm/vine | 3.99 | 4.08 | 3.85 | 3.93 | 2.42 | 2.45 | 1.69 | 1.73 |
| Effective micro-organisms (EM) at 20cm/vine | 4.01 | 4.11 | 3.88 | 3.96 | 2.43 | 2.46 | 1.70 | 1.73 |
| Effective micro-organisms (EM) at 30cm/vine | 4.04 | 4.13 | 3.91 | 3.99 | 2.43 | 2.47 | 1.70 | 1.74 |
| AM + EM at 10cm/vine | 4.13 | 4.22 | 4.02 | 4.09 | 2.46 | 2.48 | 1.72 | 1.75 |
| AM + EM at 20cm/vine | 4.15 | 4.25 | 4.04 | 4.13 | 2.47 | 2.50 | 1.72 | 1.76 |
| AM + EM at 30cm/vine | 4.18 | 4.29 | 4.08 | 4.18 | 2.50 | 2.52 | 1.74 | 1.79 |
| LSD at 0.05 | 0.02 | 0.03 | 0.03 | 0.04 | 0.02 | 0.01 | 0.01 | 0.02 |

As for the effect of EM, Sabry, *et al*., (2009) on Red Globe grapevines, Abd El-Hameed *et al*., (2010) on Thompson Seedless grapevines and Abd El-Wadoud *et al*., (2014) on Red Globe grapevines pointed out that EM application as soil drench increased significantly the physical properties of berries i.e. weight, size, length and diameter.

**3. Chemical characteristics of berries:**

As shown data in Table (4), it is noticed that chemical characteristics of berry *i.e.* total soluble solids, titratable acidity, TSS/acid ratio and anthocyanin were significantly improved by inoculation with arbuscular mycorrhiza (AM) and all different tested doses of the effective micro-organisms (EM). Dual inoculation of AM plus EM at 30cm attained significantly the highest values of TSS, TSS/acid ratio, anthocyanin content in berry skin and the lowest values of acidity in berry juice followed by dual inoculation of AM plus EM at 20cm, while control resulted in the lowest values of TSS, TSS/acid ratio, anthocyanin and the highest values of acidity in berry juice in both seasons.

The positive effect of AM and EM inoculations on berry chemical properties i.e. TSS%, TSS/acid ratio and anthocyanin content of berry skin and the negative effect on acidity% in the grape juice could be attributed to the absorbtion and translocation of elements to host root tissues by mycorrhiza fungi (Mona, 2001).

With Regard to Effective micro-organisms, it is a mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast, actinomycetes fermenting fungi that can be applied through the inoculation to increase the microbial diversity of the soil, this in turn can improve soil quality and fertility which enhances quality of crops (Higa and Kinjo, 1991).

The obtained results are in harmony with those achieved by several investigators who reported that mycorrhiza fungi produced the best chemical characteristics of berries of some grape cultivars; Abd El-Wahab *et al*., (2008) and Rizk-Alla and Tolba (2010) on Black Monukka grapevines and Abd El-Wadoud (2016) on Flame seedless grapevines.

As for the effect of EM, Sabry *et al*., (2009) and Abd El-Wadoud *et al*., (2014) on Red Globe grapevines pointed out that EM application as soil drench increased significantly TSS%, TSS/acid ratio and anthocyanin content in berry skin and decreased total acidity% of the juice.

**Table 4:** Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on berry chemical characteristics of Crimson Seedless grapes during 2016 and 2017 seasons

| Treatments | TSS (%) | | Acidity (%) | | TSS/acid ratio | | Anthocyanin (mg/100g F.W.) | |
|---|---|---|---|---|---|---|---|---|
| | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 |
| Control (untreated vines) | 16.1 | 16.5 | 0.59 | 0.57 | 27.3 | 28.9 | 31.2 | 31.8 |
| Arbuscular mycorrhiza (AM) at 250g inoculum/vine | 16.5 | 16.9 | 0.56 | 0.54 | 29.5 | 31.3 | 32.1 | 33.2 |
| Effective micro-organisms (EM) at 10cm/vine | 16.2 | 16.7 | 0.58 | 0.56 | 27.9 | 29.8 | 31.6 | 32.3 |
| Effective micro-organisms (EM) at 20cm/vine | 16.3 | 16.8 | 0.57 | 0.55 | 28.6 | 30.5 | 31.7 | 32.5 |
| Effective micro-organisms (EM) at 30cm/vine | 16.4 | 16.9 | 0.56 | 0.55 | 29.3 | 30.7 | 32.0 | 32.9 |
| AM + EM at 10cm/vine | 16.5 | 17.0 | 0.55 | 0.53 | 30.0 | 32.1 | 32.4 | 33.3 |
| AM + EM at 20cm/vine | 16.6 | 17.1 | 0.54 | 0.53 | 30.7 | 32.3 | 32.6 | 33.7 |
| AM + EM at 30cm/vine | 16.9 | 17.3 | 0.52 | 0.50 | 32.5 | 34.6 | 33.1 | 34.3 |
| LSD at 0.05 = | 0.2 | 0.1 | 0.01 | 0.02 | 1.5 | 1.9 | 0.4 | 0.5 |

**4. Morphological characteristics of vegetative growth**

As shown data in Table (5), it is noticed that some vegetative growth traits expressed as shoot length, number of leaves per shoot, leaf area and coefficient of wood ripening were significantly improved by inoculation with arbuscular mycorrhiza (AM) and all different tested doses of the effective micro-organisms (EM). Dual inoculation of AM plus EM at 30cm attained significantly the highest values of these parameters followed by dual inoculation of AM plus EM at 20cm, while control had the lowest values of these ones in both seasons.

The positive effect of AM and EM inoculations on vegetative growth parameters could be explained by that AM produced enzymes that enhance the respiration of the root (Edrees

1982). He also noticed an increase in plant growth due to the improved uptake of elements and the production of growth promoting substances. However, inoculation with AM gave more repaid growth and increased plant biomass, plant height, leaf number, leaf area than the non inoculated plants. In addition, Mona (2001) emphasized that mycorrhiza fungi are able to absorb and translocate elements to host root tissues. In addition, they can also break down certain complex minerals and organic substances in the soil and make them available to their hosts. Moreover, several researchers reported that AM mycorrhiza usually increase the growth of plants by enhancing nutrient uptake. There are three possible explanations for the greater uptake of mineral nutrients by mycorrhiza plants in comparison with non-mnycorrhizal ones (Abbot & Robson, 1982). First, mycorrhiza increase nutrient uptake by reducing the distance at which nutrients must diffuse to plant roots (Rhodes & Gerdemann, 1975). Secondly, mycorrhiza roots may differ from non-mycorrhiza roots in the relationship between rate of nutrient absorbtion and nutrient concentration at the absorbing surface (Cress *et al*., 1979). Finally, mycorrhiza hyphae may chemically modify the availability of nutrients for uptake by plants. From a consideration of the published evidence; it is likely that AM increase nutrient uptake from soil primarily by shortening the distance that nutrients must diffuse through soil to the root. It is likely, therefore, that effects of mycorrhiza in increasing nutrient uptake will be most marked for nutrients which move to roots principally by diffusion and for plant species with coarse roots and sparse, short root hairs (Baylis, 1975).

**Table 5:** Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on morphological characteristics of vegetative growth of Crimson Seedless grapevines during 2016 and 2017 seasons

| Treatments | Average shoot length (cm) | | Average number of leaves/shoot | | Average leaf area ($cm^2$) | | Average leaf area/vine ($m^2$) | | Coefficient of wood ripening | |
|---|---|---|---|---|---|---|---|---|---|---|
| | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 |
| Control (untreated vines) | 164.2 | 167.3 | 24.7 | 23.6 | 169.7 | 174.2 | 19.3 | 20.2 | 0.82 | 0.85 |
| Arbuscular mycorrhiza (AM) at 250g inoculum/vine | 172.9 | 178.6 | 25.6 | 25.5 | 178.2 | 183.4 | 22.5 | 23.6 | 0.85 | 0.88 |
| Effective micro-organisms (EM) at 10cm/vine | 166.5 | 171.7 | 24.9 | 24.0 | 172.3 | 176.6 | 20.2 | 20.8 | 0.83 | 0.86 |
| Effective micro-organisms (EM) at 20cm/vine | 169.2 | 173.5 | 25.3 | 24.8 | 174.3 | 179.8 | 20.7 | 22.3 | 0.84 | 0.86 |
| Effective micro-organisms (EM) at 30cm/vine | 171.7 | 176.3 | 25.5 | 25.2 | 174.9 | 180.5 | 21.8 | 22.7 | 0.84 | 0.87 |
| AM + EM at 10cm/vine | 174.6 | 179.3 | 25.9 | 25.9 | 179.4 | 185.6 | 23.2 | 24.6 | 0.87 | 0.89 |
| AM + EM at 20cm/vine | 177.9 | 183.5 | 26.2 | 26.3 | 182.6 | 189.2 | 24.5 | 24.8 | 0.88 | 0.91 |
| AM + EM at 30cm/vine | 179.6 | 186.7 | 26.5 | 26.8 | 184.8 | 192.3 | 25.0 | 25.3 | 0.90 | 0.94 |
| LSD at 0.05 = | 1.4 | 1.7 | 0.2 | 0.3 | 2.1 | 2.4 | 0.4 | 0.3 | 0.01 | 0.02 |

With respect to Effective micro-organisms (EM), it is considered as a mixed culture of beneficial micro-organisms primarily photosynthetic and lactic acid bacteria, yeast, actinomycetes fermenting fungi that can be applied through the inoculation to increase the microbial diversity of the soil, this in turn can improve soil quality and fertility which enhances the growth (Higa and Kinjo, 1991).

The obtained results are in harmony with those achieved by several investigators who reported that mycorrhiza fungi enhance vegetative growth of some grape cultivars; Abd El-Wahab *et al*., (2008) and Rizk-Alla and Tolba (2010) on Black Monukka grapevines and Abd El-Wadoud (2016) on Flame seedless grapevines.

As for the effect of EM, Sabry, *et al*., (2009) on Red Globe grapevines, Abd El-Hameed *et al*., (2010) on Thompson Seedless grapevines and Abd El-Wadoud *et al*., (2014) on Red Globe grapevines pointed out that EM application as soil drench increased significantly main shoot length, produced thicker canes, and improved vine vigour in comparison with the untreated vines (control).

## 5. Leaf content of total chlorophyll and mineral elements

Data presented in Table (6) revealed that inoculation with arbuscular mycorrhiza (AM) and all different tested doses of the effective micro-organisms (EM) increased significantly the leaf content of total chlorophyll and percentages of total nitrogen, phosphorus and potassium. The highest values of these determinations was obtained from the dual inoculation of AM plus EM at 30cm followed by dual inoculation of AM plus EM at 20cm, whereas the lowest values was obtained from control in both seasons.

The obtained results are in agreement with those achieved by Abd El-Wahab *et al*., (2008) and Rizk-Alla and Tolba (2010) on Black Monukka grapevines and Abd El-Wadoud (2016) on Flame seedless grapevines they found that mycorrhiza fungi improve leaf nutrient and chlorophyll contents.

As for the effect of EM, Sabry, *et al*., (2009) on Red Globe grapevines, Abd El-Hameed *et al*., (2010) on Thompson Seedless grapevines and Abd El-Wadoud *et al*., (2014) on Red Globe grapevines pointed out that EM application as soil drench increased significantly the leaf content of nutrients in comparison with the untreated vines (control).

**Table 6:** Effect of Arbuscular Mycorrhiza (AM) and effective micro-organisms (EM) on leaf content of total chlorophyll and mineral elements of Crimson Seedless grapevines during 2016 and 2017 seasons

| Treatments | Total chlorophyll (SPAD) | | N (%) | | P (%) | | K (%) | |
|---|---|---|---|---|---|---|---|---|
| | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 | 2016 | 2017 |
| Control (untreated vines) | 35.47 | 37.84 | 1.82 | 1.87 | 0.39 | 0.44 | 0.55 | 0.59 |
| Arbuscular mycorrhiza (AM) at 250g inoculum/vine | 37.06 | 39.61 | 1.85 | 1.90 | 0.42 | 0.47 | 0.58 | 0.62 |
| Effective micro-organisms (EM) at 10cm/vine | 35.99 | 38.65 | 1.83 | 1.88 | 0.40 | 0.45 | 0.56 | 0.60 |
| Effective micro-organisms (EM) at 20cm/vine | 36.32 | 38.97 | 1.84 | 1.88 | 0.41 | 0.46 | 0.56 | 0.61 |
| Effective micro-organisms (EM) at 30cm/vine | 36.85 | 39.28 | 1.84 | 1.89 | 0.42 | 0.46 | 0.57 | 0.61 |
| AM + EM at 10cm/vine | 37.43 | 39.89 | 1.86 | 1.91 | 0.43 | 0.48 | 0.59 | 0.63 |
| AM + EM at 20cm/vine | 37.79 | 40.37 | 1.87 | 1.91 | 0.44 | 0.49 | 0.60 | 0.64 |
| AM + EM at 30cm/vine | 38.96 | 42.13 | 1.89 | 1.94 | 0.47 | 0.51 | 0.62 | 0.67 |
| LSD at 0.05 = | 1.13 | 1.24 | 0.01 | 0.02 | 0.02 | 0.01 | 0.01 | 0.02 |

Data illustrated in Figures (1 & 2 & 3) indicated the existence of a highly positive correlation between total leaf area per vine ($m^2$) and yield (kg), between total leaf area per vine ($m^2$) and anthocyanin content of berry skin (mg/100g F.W.) and between total leaf area per vine ($m^2$) and coefficient of wood ripening in both seasons.

From the obtained results, it can be recommended the dual inoculation of arbuscular mycorrhiza (AM) at 250g inoculum/vine plus effective micro-organisms (EM) at the rate of 30cm/vine to obtain it on the optimum results in terms of mineral nutrients uptake, which reflected in enhancing growth and increasing yield besides improving the fruit quality attributes for Crimson Seedless grapevines.

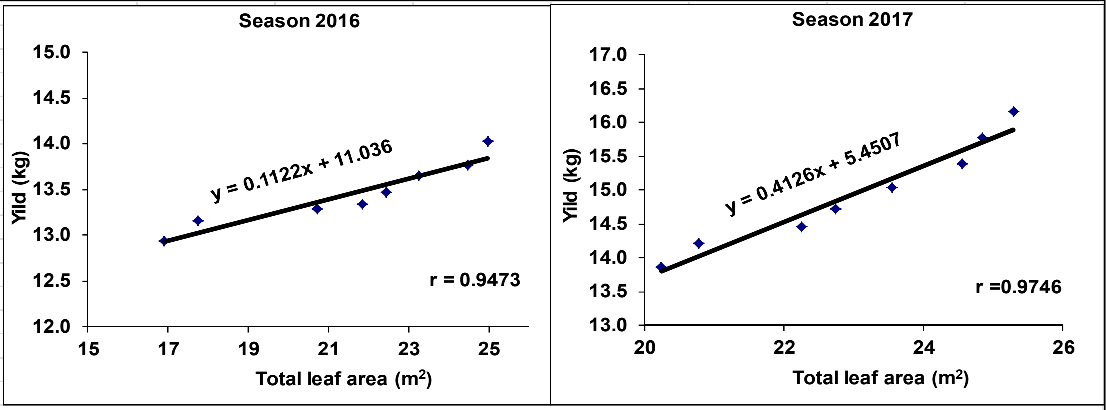


**Fig (1): Relationship between total leaf area/vine ($m^2$) and yield/vine (kg)**

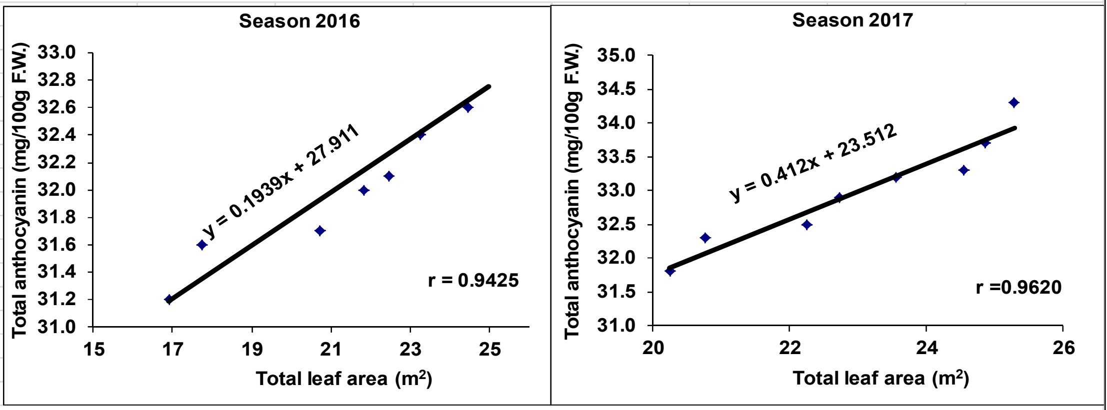


**Fig (2): Relationship between total leaf area/vine ($m^2$) and total anthocyanin (mg/100g F.W.)**

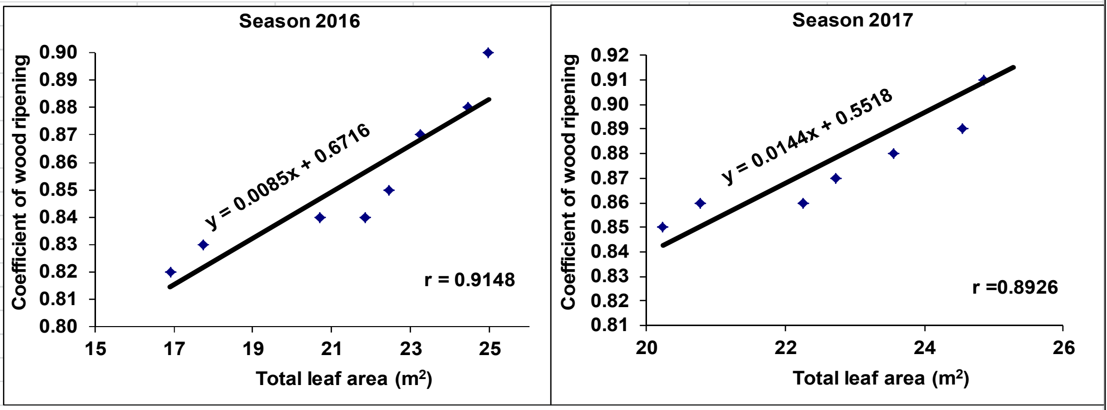


**Fig (3): Relationship between total leaf area/vine ($m^2$) and coefficient of wood ripening**

## References

Abbot, L.K. and A.D. Robson, 1982. The role of vesicular arbuscular mycorrhiza fungi in agriculture and the selection of fungi for inoculation. Aust. J. Agric. Res. 33:389-398.

Abd El-Hameed, H.M., S.A. Bondok and B.M. Selim, 2010. Trials for reducing in fertilizers by using EM in Thompson Seedless vineyards. The Sixth Inter. Conf. of Sustain. Agric. and Develop. Fac. of Agric., Fayoum Univ., 27-29-December, 143-152.

Abd El-Wadoud, M.Z., 2016. Effect of mycorrhiza fungi and yeast on growth, yield and bunch quality of Flame Seedless grapevines. Egyptian Journal of Applied Sciences, 31(11) 309-328.

Abd El-Wadoud, M. Z., M.N. Mohamed and A.S. Abd El-Rahman, 2014. Effect of some antioxidants and effective micro-organisms (EM) in improving growth and bunch quality of Red Globe grapevines. Egyptian Journal of Applied Sciences, 29(12) 648-662.

Abd El-Wahab, M.A., Shaker Gh. Sh. and O.N. Massoud, 2008. Role of Arbuscular Mycorrhiza (AM) and yeast in improving quality of Black Monukka grapes. J. Agric Sci. Mansoura Univi., 33 (1):393- 417.

Al-Garni, S.M.S., 2006. Increasing NaCl-salt tolerance of a halophytic plant Phragmites australis by mycorrhiza symbiosis. American-Eurasian Journal of Agricultural and Environmental Science 1: 119–126.

Association of Official Agricultural Chemists, 1985. Official Methods of analysis Published by A.O.A.C., Benjamin Franklin Station, Washington DC, USA.

Baylis, G.T.S., 1975. The Mongoloid mycorrhiza and mycotrophy in root systems derived from it. In: Endomycorrhizas. (Ed. F.E. Sanders; B. Mosse and P.B. Tinker), Academic Press, London, 373-375.

Bouard, J., 1966. Recherches physiologiques sur la vigne et en particulier pour I'aoutment des sarments. Thesis Sc. Nat Bordeaux-France. Pp.34.

Cress, W.A., G.O. Throneberry and D.L. Lindsey, 1979. Kinetics of phosphorus absorbtion by mycorrhiza and nonmycorrhiza tomato roots. Plant Physiol. 64: 484-492.

Daft, M.J. and B.C. Hogr彼arth, 1983. Competitive interaction amongst four species of Glomus on maize and onion. Trans - Brit. Mycol. Soc., 80: 339-345.

Douds, D.D. and P.D. Millner, 1999. Biodiversity of arbuscular mycorrhiza fungi in agroecosystems. Agriculture, Ecosystems and Environment 74: 77–93.

Edrees, M.H., 1982. Studies on growth of citrus seedlings, Ph. Thesis, Fac. Agric., Al-Azhar Univ., Cairo, Egypt.

Evelin, H., R.Kapoor and B.Giri, 2009. Arbuscular mycorrhiza fungi in alleviation of salt stress: a review. Ann Bot 104:1263–1280.

Graham, J. H., J. P. Syvertsen and M. L. Smith, 1987. Water relations of mycorrhiza and phosphorous-fertilized on citrus under drought stress. New Phytologists, 105 (3): 411- 419.

Higa, T. and S.Kinjo, 1991. Effect of lactic acid fermentation bacteria on plant growth and soil humus formation. In: J.F. Parr, S.B. Hornick and C.E. Whitman (ed.) Proc. 1st. Intl. Conf. on Kysuei Nature Fanning. Oct. 17-21, 1989, Khan Kaen, Thailand. pp. 140-147.

Husia, C.L., B.S. Luh, and C.D. Chichester, 1965. Anthocyanin in free stone peach. J. Food Science, 30: 5-12.

Jackson, M.L., 1967. Soil Chemical Analysis. Printice-Hall Inc. Englewood Cliffs-N.S.

Javaid, A., 2009. Arbuscular mycorrhiza mediated nutrition in plants. J. Plant Nutr., 32: 1595–1618.

Javaid, A. and T. Riaz, 2008. Mycorrhiza colonisation in different varieties of Gladiolus and its relation with plant vegetative and reproductive growth. Int. J. Agric. Biol., 10: 278–282.

Khaosaad, T., Garcia-Garrido, J.M.S. Steinkellner, and H.Vierheilig, 2007. Take-all disease is systematically reduced in roots of mycorrhiza barley plants. Soil Biol. Biochem., 39: 727–734.

Marin, M., A. Mari, M.Ibrarra and L.Garacia-Ferriz, 2003. Arbuscular mycorrhiza inoculation of micropropagated persimmon plants. J. Hort. Sci. and Biotechnology, 78 (5):734- 738.

Massoud, O.N., 1999. Study on the effect of Azospirillum spp., VA- mycrrohizae and organic matter amended soil on the plants growth. M. Sc. Thesis, department of botany, Faculty of Science, El-Menoufia, University.

Mona, G.S., 2001. Response of banana and guava plants to some biological and mineral fertilizers. M. Sc. Thesis, Fac. Agric. Alexandria Univ. Egypt.

Oseni, T.O., N.S. Shongwe and M.T. Masarirambi, 2010. Effect of arbuscular mycorrhiza (AM) inoculation on the performance of tomato nursery seedlings in vermiculite. Int. J. Agric. Biol., 12: 789–792.

Pregl, F., 1945. Quantitative Organic Micro-Analysis. $4^{th}$ Ed J. and A. Churchill, Ltd., London.

Ram Rao, D.M., J. Kodandaramaiah, M.P.Reddy, R.S. Katiyar and V.K. Rahmathulla, 2007. Effect of AM fungi and bacterial biofertilizers on mulberry leaf quality and silkworm cocoon characters under semiarid conditions. Caspian J. Env. Sci. 5 (2): 111-117.

Rhodes, L.H. and J.W. Gerdemann, (1975). Phosphate uptake zones of mycorrhiza and non-mycorrhiza onion. New Phytol. 75: 555-561.

Rizk-Alla, M.S. and H.I. Tolba, 2010. The Role of some Natural Soil Conditioner and AM Fungi on Growth, Root Density and Distribution, Yield and Quality of Black Monukka Grapevines Grown on Calcareous Soil. Journal of American Science. 6(12):253-263.

Sabry, G. H., M.S. Rizk-Alla and M.A. Abd El-Wahab, 2009. Effect the application of some antioxidants and effective micro-organisms (EM) in improving growth and bunch quality of Red Globe grapevines. J. Agric. Sci. Mansoura Univ., 34(6): 5901-5921.

Snedecor, G.W. and Cochran, W.G., 1980. Statistical Methods. 7th ed., The Iowa State Univ. Press. Ames. , Iowa, U.S.A., pp. 593.

Snell, F.D. and C.T. Snell, 1967. Colorimeteric Method of Analysis. D. van Nestrant Company Inc., P. 551-552.

Tahat, M.M., S. Kamaruzaman, O.Radziah, J. Kadir and H.N Masdek, 2008. Response of (*Lycopersicum esculentum* Mill.) to different Arbuscular Mycorrhiza Fungi Species. Asian J. Plant Sci., 7: 479–484.

Tourky, M.N., S.S. El-Shahat and M.H. Rizk, 1995. Effect of Dormex on fruit set, quality and storage life of Thompson Seedless grapes (Banati grapes) J. Agric. Sci., Mansoura Univ., 20(12): 5139-5151.

Ulrich, H., J.Katharina and B. Hermann, 2002. Towards growth of arbuscular mycorrhiza fungi independent of a plant host. Appl. Environ. Microbiol., 68: 1919–1924.

Wood, C.W., D.W. Reeves and D.G. Himelrick, 1992. Relationships between chlorophyll meter readings and leaf chlorophyll concentration. N status and crop yield. A review: Proc. Agro. Soc. N.Z. 23: 1-9.

Zarb, J., C.Leifert and A.Litterick, 1999. Opportunities and challenges for the use of microbial inoculants in agriculture practice. Proceedings of the sixth international Conference on Kyusei Nature Farming. 28- 31. October. Pretoria, South, Afric.

Zhang, L., J.Zhang, P.Christie and X.Li, 2008. Pre-inoculation with Arbuscular mycorrhiza fungi suppresses root knot nematode (*Meloidogyne incognita*) on cucumber (*Cucumis sativus*). Biol Fertil Soils, 45:205–211.