

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.

*¹Mervat, S. Rizk-Alla and ²Hager, I. Tolba

¹*Horticultural Research Institute, ²Microbiology Dep., Soils, Water & Environment Research Institute, Agricultural Research Center, Giza, Egypt.

Abstract: The current research was carried out during two successive seasons (2007 and 2008) on ten years old Black Monukka grapevines to disclose the role of some natural soil conditioners namely, humic acid (HA), Nile fertile (NF) and AM fungi (AM) in a single application or in combined mixture growth, root density and distribution, yield and quality of Black Monukka grapevines grown under calcareous soil in a private vineyard in Nobarria at Cairo-Alexandria Desert Road; the results showed that all different soil conditioners were effective but the treatment of humic acid at 15 ml/ vine (HA1) + Nile fertile at 200 g/ vine (NF1) + AM fungi gave the best results in comparison with other treatments and control. This treatment enhanced the growth characters namely total leaf area/ vine, shoot diameter and coefficient of wood ripening, total chlorophyll, NPK% of the leaves and total carbohydrates of the canes. Also, the vines of this treatment produced the heighest fibrous root fresh weight, larger number and longest fibrous root. With respect to microbiological activity in the rhizosphere, it was noticed that the best AM infection %, no. of AM spores /g dry soil, total microbial count, phosphatase and dehydrogenase enzymes activity were obtained by the same treatment. From the economic point of view, this treatment was accompanied by the highest yield and best its components namely physical and chemical characteristics of bunches and berries. Under such promising treatment the adverse effects of calcareous soil on growth and production of vines could be overcome. [Journal of American Science. 2010;6(12):253-263]. (ISSN: 1545-1003).

Keywords: grapevine; humic acid (HA); Nile fertile (NF); AM fungi (AM); microbiological activity

1. Introduction:

In Egypt different types of new reclaimed soils particularly calcareous soils have an alkaline reaction. Application of some mineral fertilizers may be ineffective because of the expected problem of its availability, where phosphorus are fixed in insoluble forms and become unavailable to plants. Phosphorus deficiency may be limit plant growth than do nitrogen deficiency (Awad, 1999). In addition, grape tree grown on calcareous soil developed in many instances chlorotic symptoms. This lime induce chlorosis is due it iron (Fe) deficiency whereas, micronutrients e. g. Fe, Mn and Zn are unavailable under condition of high soil pH and CaCO₃ content (Clark and Kajiura, 1986).

Humic acids (HA) have been found to a profound effect on not only the biological activity and soil structure, but also on the plant itself. This is due to their positive effect on plant nutrients uptake, increasing the productivity of fruit crops due to the conversion of unavailable minerals into soluble forms that plants can use (El Fakharani, 1999). Agricultural (HA) are reputed to enhance drought tolerance, seed germination and overall plant performance (Morales-Payar and Stall, 2003).

Cheng *et al.*, (1998) reported that, humic acids decrease the loss of soil moisture, enhance

water retention, increase the ability rate of wheat leaves for photosynthetic process and also increase root growth in a manner similar to auxins. Where the soil pH is high, correction of Fe, Mn and Zn deficiency can be attempted by adjusting pH with acidifying compounds such as elemental sulphur (Clark and Smith, 1986). Whereas, sulphur is oxidized by soil microorganisms, such as *Thiobacillus* spp. Bacteria which, consider as the most important microorganisms involved in the bioleaching of sulphide compounds to sulphuric acid in amount enough to decrease soil pH, improve availability of most soil nutrients and uptake by plant, enhancing root development and increasing the activity of soil microorganisms (Kassem *et al.*, 1995). Ibrahiem (2003) found that fertilizing the vines by Nile-fertile (263 g/vine) gave the highest yield and best quality of berries of flame seedless grapevines.

Arbuscular mycorrhizal (AM) fungi are ubiquitous soil organisms that can form mutualistic associations with the roots of the majority of vascular plant species (Lindermann, 1988). The establishment of AM association is often beneficial for plant nutrition especially enhance absorption of phosphours and other relatively immobile micronutrients cations, particularly zinc and copper (Marin *et al.*, 2003) also AM fungi produce plant

growth hormones such as auxines, cytokinines and gibberellins (Zhang *et al.*, 2008), increasing water uptake (Graham *et al.*, 1987). AM fungi may also confer tolerance and resistance to various abiotic and biotic stresses to the host plant (Colla *et al.* 2008).

Black monukka is one of the table grape cultivars; ripens in mid July to late August, seedless, sweet, crispy, purplish-black coloured, tender skin. The production of small to medium berries and loose bunches are negatively reflected on bunch quality (Harry *et al.*, 1991)

Therefore, the main objective of this study is to evaluate the possible effects of humic acid, Nile fertile, AM fungi and their combination on growth, vine nutritional status, yield/vine and quality of black Monukka grapevines grown in a calcareous soil.

2. Materials and Methods

The current study was carried out during two successive seasons (2007 and 2008) in a private vineyard in Nobaria using 10 years old Black Monukka grapevines. The chosen vines grown in a calcareous soil (Table 1).

Table (1). Main chemical and physical characteristics of the experimental soil.

Particle size distribution%	
Coarse sand	11.75
Fine sand silt	37.10
silt	18.03
Clay	33.12
Texture class	Sandy clay loam
Chemical analysis	
pH(1:2.5 soil water suspension)	8.2
Calcium carbonate (%)	25.8
EC (dS/m)	1.48
Ca ⁺⁺ (meq/L)	4.13
Mg ⁺⁺ (meq/L)	1.12
Na ⁺ (meq/L)	6.46
K ⁺ (meq/L)	1.43
CO ³⁻ (meq/L)	0.00
HCO ³⁻ (meq/L)	1.47
Cl ⁻ (meq/L)	6.90
SO ⁴⁻⁻ (meq/L)	4.77
Organic matter (%)	0.43
Total N (%)	0.053
Total P (ppm)	616
Available N (ppm)	25.9
Available P (ppm)	4.35

1- Lay out of the experiment:

Spaced between each vine lines at 2X 2.5 meters a part and irrigated by the drip irrigation system, cane-pruned and trellised by the double T shape system. The vines were pruned during the second week of January with bud load of 60 buds/vine. 144 uniform vines were chosen. Each four vines acted as a replicate and each three replicates by one.

All vines were subjected to the normal horticultural practices. Nitrogen (60 kg N/ feddan), potassium (100 Kg K₂O/feddan) and phosphorus as phosphoric acid (1 liter/2 weeks) were added.

Chelated Fe, Zn and Mn at rates of 200 g, 100 g and 100 g respectively per 600 liter water sprayed two times, once 10 days before anthesis and the other after fruit set (at 3-4 mm berry diameter).

The randomized complete block design was followed the tested treatments that evaluated through the following parameters:

Twelve treatments applied as follows:

- T1- Control (untreated vines).
- T2- P-humex acid at 15 ml/ vine (HA1).
- T3- P-humex acid at 30 ml/ vine (HA2).
- T4- Nile fertile at 200 g/ vine (NF1).

- T5- Nile fertile at 300 g/ vine (NF2).
- T6- AM fungi.
- T7- HA1 + AM fungi.
- T8- HA2 + AM fungi.
- T9- NF1 + AM fungi.
- T10- NF2 + AM fungi.
- T11- HA1 + NF1.
- T12- HA1 + NF1+ AM fungi.

Humic acid (HA) content in the liquid organic fertilizer was determined using BaCl₂ precipitation method as described by (Fataftah *et al.*, 2001). P-humex acid (HA) contains 25% humic acid, 2% N, 4% P₂O₅, 6% K₂O, 0.2% Fe, 8.9 pH, 45.22 organic matter, 300- 500 meg/ 100g CEC and 1.25 Kg/L density. Humic acid was added at the rate of 15 or 30 ml/vine (according to Ali *et al.*, 2006) on the soil surface after bud burst.

Nile fertile (NF) contains 38% S, some essential elements, (2.7 % N, 3.5% P, 1.2% K, 5% Ca, 2.7% Mg and 1% Fe) and sulphur bacteria, *Thiobacillus* spp. (10⁶ CFU/gm). Nile fertile was applied once during winter agricultural management by mixing with soil in the wetting zone adhesive to the root at a recommended rate 200 or 300 g /vine (Ibrahiem, 2003)

AM fungi inoculum: mycorrhizal spores that contained the mixture of the following genera; *Glomus*, *Gigaspora* and *Acaulospora* were extracted from Egyptian soil. Extraction and counting of identified mycorrhizal spores were carried out according to the method described by Massoud (2005). The extracted mycorrhizal spores were mixed with sterilized peat as a carrier (250 spore/gram) and then applied to the soil at a rate of (100 g inocula /m long) so each vine diameter was 2.5 m. so, each vine needed 250 g inoculums.

Growth characters:

Total leaf area /vine (m²) was determined by multiplying average number of the leaves/ shoot by average leaf area then by the number of shoots per vine. Ten shoots/ vine were labeled to determine the average shoot diameter (mm) and determine the wood ripening coefficient by dividing the length of the mature part of the shoot by total shoot length (Bouard, 1966).

Root system measurements:

Fibrous root density (root fresh weight and total number) was determined in soil samples taken with auger to make a hole of 10 cm in diameter (auger volume = 1153.8 cm³) and 30 cm depth from four directions at 50 and 100 cm away from vine trunk. Soil sample were taken in late November, average weight per hole was calculated as gm/ hole

according to Cahoom *et al.*, (1959). Average length of roots (cm) was determined.

Chemical determinations:

leaf content of total chlorophyll was measured using nondestructive Minolta chlorophyll meter SPAD 502 of the 5th and 6th leaves (Wood *et al.*, 1992) Cane content of total carbohydrates (%) was determined according to (Smith *et al.*, 1956) leaves opposite to the clusters were collected then dried to estimate N, P and K percentages according to (Jackson, 1973).

Yield, clusters and berry characteristics:

Clusters were picked when TSS of each treatment reached about 16- 17% (Tourky *et al.*, 1995). Yield/ vine (kg), cluster weight (g), berry weight (g), berry size (cm³). Shattering (%) was determined on clusters stored for seven days at room temperature (28 to 32 C^o), Shattering (%) was calculated by dividing weight of Shattering berries by the initial weight of the clusters. T.S.S. (%) was estimated by a hand refracto-meter, total titrable acidity as tartaric acid (%) A.O.A.C. (1985), T.S.S./ acid and total anthocyanin of berry skin (mg/ 100 g fresh weight) was estimated as described by Yildiz and Dikman (1990).

Microbiological parameter:

Arbiscular mycorrhizal infection was determined according to method described by (Massoud, 2005). Spores number after each season was also counted according to (Massoud, 2005). Total microbial count (--- X 10⁶ CFU/g soil), dehydrogenase (µg TPF/ g dry soil/ day) and phosphatase enzyme activity(IP /g/ dry soil/ day) were also determined according to the methods described by (Esher and Jensen 1972, Salman 1967 and Drobnikova 1961)

Data obtained were statistically analyzed according to Snedecor and Cochran (1980) using the New L.S.D. test for examining the significant differences between the studied treatments.

3. Results and Discussion:

1- Vegetative growth parameters:

Obtained data in table (2) showed, vegetative growth parameters of Black Monukka cv. as affected by humic acid, Nile fertile and AM fungi and their combinations. The data revealed that, all treatments improved the growth parameters compared to the control. However, it is worthy to notice that, the effect of single application of this soil conditioner was significantly lower compared to the combined application of these materials. However, the highest values of total leaf area/ vine, shoot diameter and coefficient of wood ripening were detected in case of vines treated with (HA1 +NF1+

AM fungi) amounting to 21.8 and 22.5 m² for total leaf area/vine, 8.0 and 8.6 mm² for shoot diameter and 0.86 and 0.89 for coefficient of wood ripening in both seasons respectively. While, the lowest values were recorded with the control 15.2 and 16.5 m² for total leaf area/vine, 6.0 and 5.6 mm² for shoot diameter and 0.63 and 0.61 for coefficient of wood ripening in both seasons respectively.

The obtained increasing in vegetative growth parameters recorded with the treatment (HA1 +NF1+ AM fungi) this might be due to the role of humic acid in modifications in the soil-root interface that make the nutrients more available to plants (El Fakharani, 1999) and humic acid improved plant vigour and health (Kassem *et al.*, 1995).

Table (2) Effect of soil condition with humic acid, nilefertile and AM fungi application on some characteristics of vegetative growth of Black Monukka grapevines during 2007 and 2008 seasons.

Treatments	Total leaf area/vine (m ²)		Shoot diameter (mm ²)		Coefficient of wood ripening	
	2007	2008	2007	2008	2007	2008
T1	15.2	16.5	6.0	5.6	0.63	0.61
T2	15.5	17.0	6.1	6.1	0.65	0.66
T3	15.8	17.3	6.2	6.4	0.67	0.70
T4	15.6	17.0	6.4	6.1	0.69	0.68
T5	16.4	17.5	6.6	6.4	0.70	0.70
T6	15.5	16.8	6.1	6.0	0.66	0.65
T7	16.6	18.0	6.7	6.7	0.73	0.73
T8	17.0	18.3	6.9	7.0	0.77	0.77
T9	17.5	18.9	7.2	7.4	0.77	0.80
T10	17.9	19.3	7.4	7.7	0.80	0.82
T11	18.6	20.7	7.7	8.1	0.82	0.85
T12	21.8	22.5	8.0	8.6	0.86	0.89
New L.S.D.at 5%	2.8	2.5	0.10	0.30	0.02	0.03

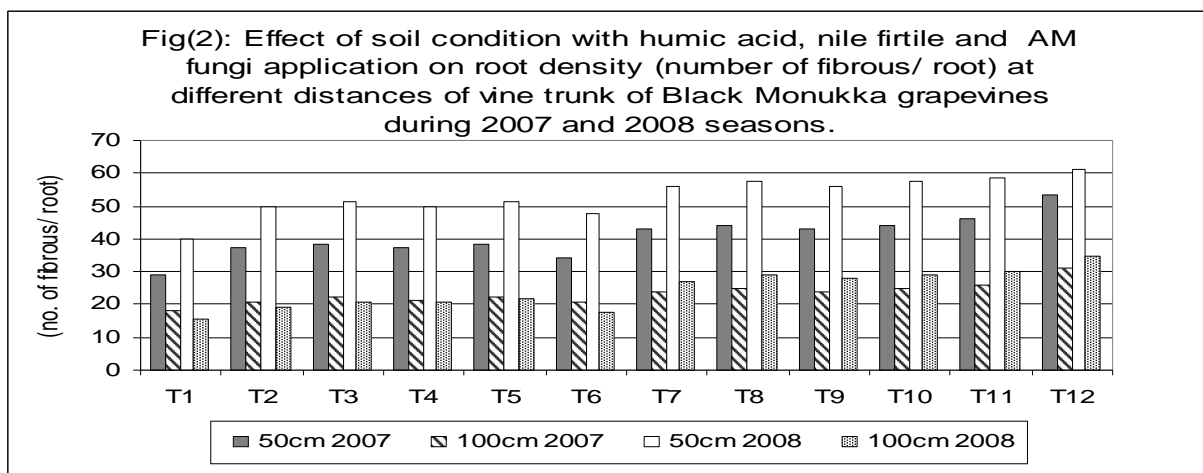
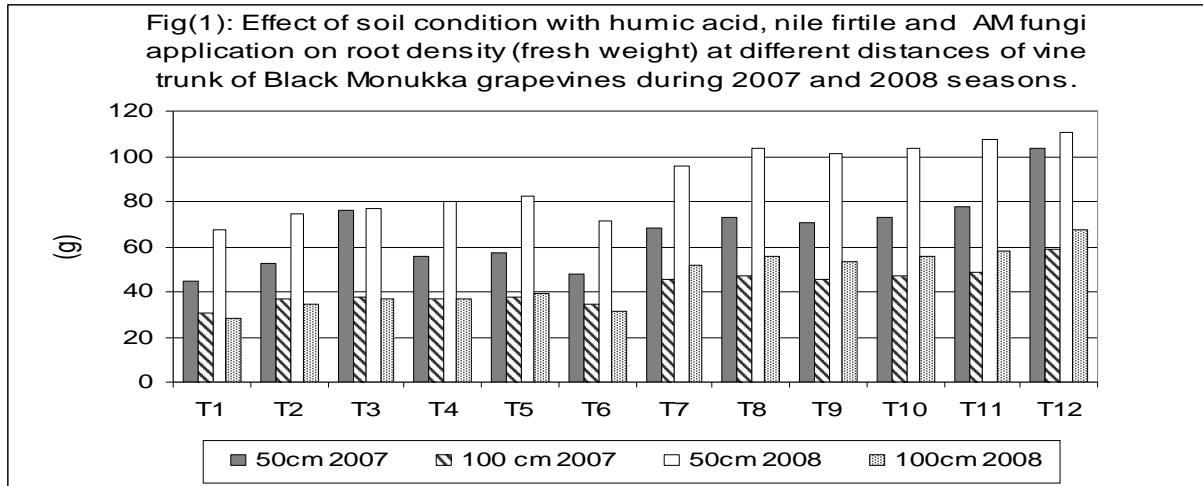
With respect to Nile fertile, it is considered as a soil conditioner, reducing pH, due to acid bacteria that *Thiobacillus* spp. bacteria where, the mechanisms for the growth of *Thiobacillus* spp. bacteria on sulphur is proposed. Initially, the bacteria attach to the sulphur particles and start oxidizing it and the number of attached bacteria increased with time, resulting an increase in the rate of generation of partially simple oxidized sulphur compounds. The sulphur compounds are consumed by plants that contribute on its growth and yield (Bhavaraju *et al.*, 1993). Also increased availability of most nutrients and increasing organic mater (Ibrahiem, 2003) could explain these results with respect to AM fungi that produced enzymes that enhance the respiration efficiency of the root (Zhang *et al.*, 2008) and he also noticed an increase in plant growth due to the improved uptake of elements. However, the colonization with AM fungi gave more repaid growth and increased plant biomass, plant height, leaf area

than the non inoculated plants. The findings are in the same line with those of El Shenawy and Fayed (2005), Hussien *et al.*, (2005), Omar (2005) and Ali *et al.*, (2006) who reported that humic acid enhance vegetative growth, Ibrahiem (2003) and Rizh-Alla *et al.*, (2006) who reported that Nile fertile recorded the maximum values of vegetative growth. As for the effect of AM fungi Abd El Wahab *et al.*, (2008) found that dual inoculation with AM fungi increased the vegetative growth parameters.

2- Root system measurements:

a- fresh weight and numbers:

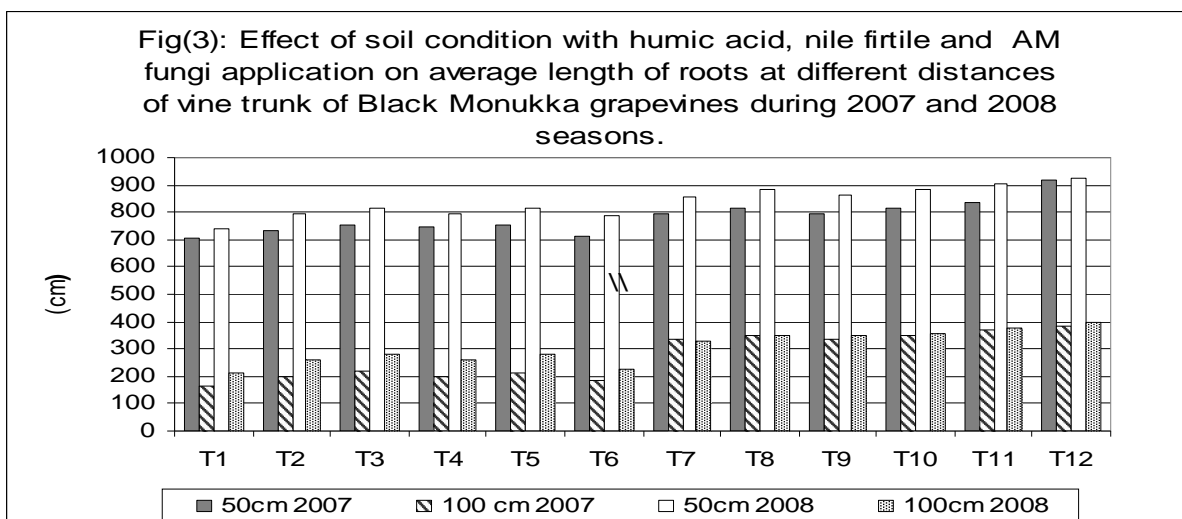
Data of figs (1 and 2) indicate that, vines of applied with (HA1 +NF1+ AM fungi) produced the highest fibrous roots fresh weight (gm/ hole) at 50 and 100 cm a way from vine trunk and gave highest numbers of fibrous roots in both seasons. On the other hand, untreated vines produced the least fibrous roots fresh weight. While, the other treatments were intermediate.



b- Root distribution:

Distribution of the roots in the soil profile are important to know the volume of wetted root zone, fig (3) show that the treatment (HA1 +NF1+ AM

fungi) recorded the longest roots. On the other hand untreated vines had the least values, while the other treatments gave intermediate values in this respect.



Generally, this treatment gave better and larger rooting area in soil. The greatest amount of roots and longest was observed in the 50 cm distance from the vine trunk compared with in the 100 cm distance from the vine trunk.

The obtained root results of this treatment could be attributed to the humic acid has high water-holding and affects the physicochemical properties of soil, which are important in controlling the uptake of nutrients, their retention and counteracting soil acidity (Hartwigsen and Evans, 2000).

The beneficial effect of Nile fertile on increasing the root formation could give an explanation for the present results. These results are in agreement with those obtained by Ahmed *et al.*, (1994) and Ibrahiem (2003).

Vitagliano *et al.*, (1999) showed that, AM fungi increased the growth of rooted cutting of olive

cultivar, via an increase in lateral root frequency. It has been recognized that AM fungi symbiosis play role in nutrient cycling in the ecosystem and also protect plants against environmental conditions and stress (Barea and Jeffries, 1995).

3- Chemical determination:

Results presented in Table (3) revealed that total chlorophyll, total carbohydrate content of the canes and percentages of total nitrogen, phosphorus and potassium of the leaves were increased significantly by the different treatments. Also, it can be noticed that the application of (HA1 +NF1+ AM fungi) generally resulted in higher values of these parameters as compared to uninoculated vines and the other single application of these materials in the both seasons.

Table (3) Effect of soil condition with humic acid, Nile fertile and AM fungi application on total chlorophyll, total carbohydrates, leaf N, P and K % contents of Black Monukka grapevines during 2007 and 2008 seasons.

Treatments	Total chlorophyll (mg/ g F.W)		Total carbohydrates (%)		N%		P%		K%	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
T1	31.5	33.8	23.9	24.1	1.79	2.42	0.30	0.22	1.25	1.35
T2	32.9	34.7	24.9	25.6	1.91	2.53	0.32	0.27	1.35	1.43
T3	34.0	35.5	25.7	26.7	2.00	2.63	0.35	0.31	1.45	1.58
T4	34.7	38.0	26.3	26.8	2.05	2.50	0.33	0.27	1.36	1.55
T5	34.9	38.2	26.8	26.9	2.07	2.63	0.37	0.32	1.46	1.59
T6	32.7	34.6	24.8	25.4	1.88	2.46	0.32	0.28	1.35	1.49
T7	36.0	39.0	27.7	27.6	2.15	2.73	0.40	0.38	1.57	1.67
T8	37.0	39.2	28.5	27.7	2.20	2.8	0.41	0.39	1.64	1.72
T9	37.5	40.0	28.8	27.9	2.27	2.83	0.40	0.41	1.66	1.74
T10	38.5	40.5	29.1	29.1	2.30	2.97	0.42	0.42	1.74	1.81
T11	38.8	41.2	29.6	30.3	2.33	3.15	0.43	0.44	1.84	1.90
T12	40.0	42.0	30.5	31.6	2.43	3.40	0.45	0.48	1.95	2.0
New L.S.D.at 5%	1.0	0.7	0.80	1.10	0.08	0.10	0.02	0.04	0.10	0.09

The obtained results could be interpreted in view of the effect of humic acid and Nile fertile on enhancing the metabolism process of carbohydrates as well as its effect on reducing soil pH which by their turn could be responsible for increasing the availability of nutrients. Also, AM fungi produced enzymes that enhance the respiration of root (Edrees, 1982). AM fungi are able to absorb and translocate elements to host root tissues (Mona, 2001), AM fungi increase nutrient uptake by reducing the distance at which nutrients must diffuse to plant roots (Rhodes and Gerdemann, 1975). AM fungi improved nutrition mode possible by extensive hyphae network. This not only allows the plant to overcome phosphorus depletion from the zone around the root, but also

allows it to reach immobile phosphorus that the fungus can solubilize. This phenomenon is most apparent in soils low in phosphorus. (Zarb *et al.*, 1999).

The obtained results are nearly similar to those achieved by several investigators who reported that humic acid enhance these contents El Shenawy and Fayed (2005); Hussien *et al.*, (2005); Omar (2005) and Ali *et al.*, (2006).

As for the effect of Nile fertile, Ibrahiem (2003) and Rizk-Alla *et al.*, (2006) pointed that Nile fertile significantly increased these contents. In addition, some researcher found that AM fungi increased chlorophylls content of leaves El-Sharkawy (1989) and increased carbohydrate content of canes

and leaf mineral content Abd El-Wahab *et al.*, (2008).

4- Yield, cluster weight and some physical characteristics of berries:

Yield and cluster weight in general was significantly increased by all applications compared with control Table (4). The application of (HA1 +NF1+ AM fungi) resulted in the highest values of this estimate (9.0, 9.5 kg/ vine and 422.5, 451.3 g) for both seasons respectively. Whereas, the lowest values were obtained from untreated vines, this recorded (7.91, 8.36 kg/ vine and 383.7, 401.1 g) for both seasons respectively.

Also, this application gave the best results of physical properties of the berries in terms of increased berry weight, berry size and reduced berry shattering compared to the control. However, it is worthy to note that, the single application of humic

acid, Nile fertile or AM fungi were significantly lower compared to the combined application of these natural soil conditioner.

The beneficial effect of these materials on yield, cluster weight and some physical properties of berries could be attributed to its vital role in lowering soil pH. Consequently, vine growth and nutritional status (tables, 2 and 3) are being improved cluster weight, increasing yield/ vine and improved the physical properties of berries.

The obtained results are in agreement with those reported by Zhu and Zhu (2000); Hussien *et al.*, (2005) and Ali *et al.*, (2006) who found that humic acid applications significantly increased berry physical properties. As for the effect of Nile fertile Ibrahiem, (2003) found that Nile fertile improved physical properties of berries. Moreover, Abd El-Wahabe *et al.*, (2008) pointed out that yeast and AM fungi gave the best physical properties of berries.

Table (4): Effect of soil condition with humic acid, Nile fertile and AM fungi application on yield and some physical characteristics of berries of Black Monukka grapevines during 2007 and 2008 seasons.

Treatments	Yield/ vine (kg)		Cluster weight (g)		Berry weight (g)		Berry size (cm ³)		Shattering (%)	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
T1	7.91	8.36	383.7	401.4	2.03	2.34	1.89	2.28	24.67	24.95
T2	8.09	8.58	398.6	409.5	2.15	2.43	2.00	2.36	23.37	23.06
T3	8.20	8.72	403.2	415.0	2.26	2.52	2.10	2.41	22.10	21.39
T4	8.14	8.59	400.2	410.3	2.23	2.48	2.07	2.34	23.32	21.40
T5	8.24	8.71	404.6	416.1	2.25	2.50	2.10	2.40	22.06	21.39
T6	8.04	8.47	390.1	406.3	2.14	2.43	1.99	2.34	23.35	23.15
T7	8.39	8.89	409.5	423.5	2.39	2.61	2.21	2.48	20.76	19.59
T8	8.51	9.02	413.5	430.2	2.42	2.70	2.31	2.54	20.70	19.50
T9	8.64	9.13	410.0	436.1	2.44	2.65	2.34	2.59	20.72	19.55
T10	8.75	9.26	415.2	442.0	2.45	2.71	2.36	2.69	20.70	19.50
T11	8.86	9.38	417.7	446.0	2.48	2.80	2.39	2.74	20.69	19.46
T12	9.00	9.50	422.5	451.3	2.60	2.91	2.50	2.80	19.36	17.80
New L.S.D.at 5%	0.12	0.11	4.60	5.10	0.11	0.09	0.10	0.05	1.26	1.66

5- Chemical properties of berries:

Data in table (5) indicated that, there were significant differences between all treatments than control concerning the chemical properties of berries. Moreover, the application of (HA1 +NF1+ AM fungi) improved the chemical quality of berries in terms of increasing the total soluble solids, total soluble solids/ acid ratio and anthocyanin contents of berry skin and reducing the total acidity than the sole application of these materials in the two seasons of the study.

The enhancing of humic acid, Nile fertile and AM fungi on chemical properties of the berries may be ascribed to its role in achieving a good

balance between growths and fruiting through nutrients availability in soil which is reflected by its turn on increasing the accumulation of total carbohydrate and resulting the stimulation of ripening. These results are in accordance with those obtained by Li-Nan *et al.*, (1999) Zhu and Zhu (2000) and Ali *et al.*, (2006) who found that humic acid improved the chemical properties of berries. Moreover, Ibrahiem (2003) found that Nile fertile enhancing the chemical properties of berries. As for the effect of AM fungi, Abd El-Wahab *et al.*, (2008) pointed out that AM fungi gave the best chemical properties of berries.

Table (5): Effect of soil condition with humic acid, Nile fertile and AM fungi application on chemical characteristics of berries of Black Monukka grapevines during 2007 and 2008 seasons.

Treatments	T.S.S. (%)		Acidity (%)		T.S.S./ Acidity		Anthocyanin (mg/100g F.W.)	
	2007	2008	2007	2008	2007	2008	2007	2008
T1	16.10	15.91	0.72	0.68	22.36	23.40	35.83	37.34
T2	16.27	16.11	0.70	0.67	22.24	24.04	36.24	37.78
T3	16.42	16.31	0.68	0.66	24.15	24.71	36.55	37.90
T4	16.40	16.30	0.69	0.65	23.77	25.08	36.33	37.83
T5	16.43	16.31	0.68	0.65	24.16	25.09	36.58	37.95
T6	16.25	16.11	0.70	0.67	23.21	24.04	35.52	37.46
T7	16.59	16.52	0.65	0.64	25.52	25.81	37.0	38.13
T8	16.61	15.54	0.64	0.63	25.95	26.25	37.2	38.26
T9	16.60	16.57	0.63	0.62	26.35	27.42	37.0	38.15
T10	16.65	17.00	0.62	0.62	26.85	27.42	37.3	38.27
T11	16.70	17.00	0.62	0.62	26.94	27.42	37.5	38.37
T12	16.80	17.2	0.61	0.60	27.54	28.67	37.8	38.5
New L.S.D.at 5%	0.15	0.20	0.02	0.01	0.50	1.10	0.30	0.12

6- Microbiological studies:

Regarding to data in table (6) all treatments caused a significant increase in AM infection % in comparison to control, the best increase was caused by the application of (HA1 +NF1+ AM fungi) in the two seasons of the study, this was recorded 95.33 and 100.00 respectively. This finding indicates that AM fungi may solubilize the surrounding weatherable mineral through excretion of organic acids such as α -ketoglutaric acid. These organic acids could exert a selective influence on soil microbial communities (Abd El-Wahab *et al.*, 2008).

Moreover, no. of AM spores in soil was significantly increased especially in treatment (HA1 +NF1+ AM fungi) in the two seasons of the study; this was recorded 1022.2 and 1340.0 respectively. These findings are in the same line with those obtained by (Turk *et al.*, 2006) who pointed out that AM fungi colonize plant roots and mainly in the surrounding soil extending the roots depletion zone around the root system and consequently completed its life cycle and obtained plenty of resting spores in soil.

The total microbial count was the same line of increasing especially in the application of (HA1 +NF1+ AM fungi) in the two seasons; this was recorded 81.60 and 93.61 respectively. These results are in agreement with those obtained by (Linderman and Pflieger, 1994) who explained that AM fungi is capable of increasing nutrient content which act as a

suitable media for most rhizospheric microorganisms in general. On the other hand, Godeas *et al.*, (1999) explained that the increases in populations of rhizospheric microorganism in roots of most plants are influenced by a combined inoculation of microorganism and AM fungi.

Also, phosphatase enzyme activity was significantly increased in all treatments especially in treatment (HA1 +NF1+ AM fungi) in the two seasons of the study, this was recorded 35.33 and 38.00 respectively. These results are in agreement with those obtained by (Hetick, 1989) who found that mycorrhizal hyphae can provide access to insoluble nutrient sources through enzyme activity or some physical or chemical modification of the rhizosphere. The inorganic phosphorus compounds can first be hydrolyzed by phosphatase enzyme which mostly originates from plant roots, through the action of fungi and bacteria.

The same increase was observed in dehydrogenase enzyme activity in all treatments in comparison to control where the best increase was caused by the application of (HA1 +NF1+ AM fungi) during the two seasons, this was recorded 113.67 and 133.67 respectively. The increase in dehydrogenase enzyme activity was attributed to the intense activity of microflora as a mixture of biomass (Massoud, 2005).

Table (6): Effect of soil condition with humic acid, nilefertile and AM fungi application on AM infection %, No of AM spores , No of AM spores and Phosphatase enzyme activity and Dehydrogenase activity in rhizospheric zone of Black Monukka grapevines during 2007 and 2008 seasons.

Treatments	AM infection %		No of AM spores /g dry soil.		Total microbial countX106 CFU/gdry soil		Phosphatase enzyme activity(P /g/ dry soil)		Dehydrogenase enzyme activity (µg TPF/ day/ g dry soil).	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
T1	30.00	34.67	27.9	41.3	40.30	41.25	10.23	10.57	40.33	44.33
T2	46.00	51.00	30.4	103.3	44.25	44.29	11.20	12.00	44.67	50.33
T3	60.00	60.00	32.1	111.7	44.56	46.32	15.60	16.87	53.33	55.67
T4	40.00	45.33	25.0	50.3	50.89	50.27	17.27	18.67	53.67	61.67
T5	50.67	55.33	40.0	101.0	55.67	57.41	19.50	22.00	90.33	95.67
T6	70.33	75.00	560.0	640.0	45.32	57.68	24.33	26.33	88.00	93.67
T7	85.33	87.33	708.9	783.3	80.59	87.55	29.00	30.33	87.67	90.33
T8	85.67	87.67	721.1	770.0	75.24	85.42	30.33	30.33	81.00	85.33
T9	70.33	80.67	558.9	633.3	70.26	77.49	25.00	27.67	88.00	89.67
T10	75.00	81.00	613.3	691.7	73.68	80.58	30.33	31.67	95.67	107.00
T11	80.67	81.33	334.4	400.0	80.50	87.22	33.00	34.67	110.33	121.33
T12	95.33	100.00	1022.2	1340.0	81.60	93.61	35.33	38.00	113.67	133.67
New L.S.D.at 5%	8.23	11.47	194.5	219.5	6.7	9.3	5.14	8.97	4.81	5.72

7-Economical evaluation of the recommended treatment (HA1 +NF1+ AM fungi) compared with control:

Data shown in table (7) clearly indicate that the application of (HA1 +NF1+ AM fungi) gave the

maximum net profit compared with the control in both seasons. The very slight raise in the cost of production/ feddan over control for this treatment is economically justifiend in view of the higher price of the yield obtained from this treatment.

Table (7): Some economical data on costs and profit per fed of the recommended treatment (humic acid, nile fertile and AM fungi) compared to control.

Season	Price of humic acid (L.E.)	Price of nile fertile (L.E.)	Price of AM fungi	Labour cost (L.E.)	Total cost (L.E.)	Increase in yield over control per fed (kg)	Price of increase in yield over control (L.E.)	The net profit (L.E.)
2007	160	70	200	50.0	480	763.0	2098.25	1618.25
2008	160	84	-----	50.0	294	798.0	2234.4	1940.4

As a conclusion it could be concluded that, adding humic acid (HA) at 15 cm/ vine plus nile fertile (NF) at 200 g/ vine and AM fungi (AM) this recommended under the calcareous soil having high pH values to obtain the highest yield besides improving the fruit quality and improving microbiological activity.

Corresponding author

Mervat, S. Rizk-Alla
Horticultural Research Institute
Agricultural Research Center, Giza, Egypt.

References

1. Abd El-Wahab, M. A.; Shaker, S. H. and Massoud, O. N. (2008): Rool of Arbuscular Mycorrhiza (AM) and yeast in improving quality

of Black Monukka grapes. J. Agric Sci. Mansoura Univ., 33 (1): 393- 417.

- Ahmed, F. F.; Mansour, F. M. and EL-Shamaa (1994): How much sulphur in necessary for Thompson seedless grapevines? Minia J. Agric. Res. and Dev. 16 (2): 287- 298.
- Ali, M. A.; Rafaat, S. S. and El-Shal, S. A. (2006): the role of humic acid in reducing mineral fertilizer rates applied in vineyards. Egypt. J. of Appl. Sci. 21 (11):14- 21.
- A.O.A.C. (1985): Association of Official Agricultural Chemist Official methods of Analysis. 12th Ed. Washington D.C., U.S.A.
- Awad, S. M. (1999): Response of "Flame" grape transplants to microrhizal inoculation and phosphorus fertilization. Egypt. J. Hort., 26 (3): 421-433.

6. Barea, J. M. and Jeffries, P. (1995): Arbuscular mycorrhizas in sustainable soil plant system. In. A. Varma and B. Hock (Editors) Mycorrhiza structure, function. Molecular Biology and Biochemistry, Springer- Verlag Heidelberg pp: 521-560.
7. Bhavaraju, S. R.; Modak, J. M.; Kumar, R. and Gandhi, K. S. (1993): Dissolution of Sulphur Particles by *Thiobacillus ferrooxidans*: Substrate for Unattached Cells. Biotechnology and Bioengineering, Vol. 41, Pp. 612-616.
8. Bouard, J (1966): Recherches physiologiques sur la vigne et en particulier pour l'acutment des sarments. Thesis Sc. Nat Bordeaux-France. Pp. 34.
9. Cahoon, G. A.; Morton, E. S.; Jones, W. W. and Garber, M. J. (1959): Effect of various types of nitrogen fertilizers on root density and distribution as related to water infiltration and fruit yield of Washington navel orange in a long term fertilizer experiment. Proc. Amer., Soc. Hort. Sci., 74: 289- 299.
10. Cheng, F. J.; Yang, D. Q. and Wu, Q. S. (1998): Physiological effects of humic acid on drought resistance of wheat. Chinese J. of Applied Ecology, 6 (4): 363- 367.
11. Clark, C. J. and Kajiura, I. (1986): Nutritional disorders in persimmon. Growing Today. April: 47-50.
12. Clark, C. J. and Smith, G. S. (1986): leaf analysis of persimmon. Growing Today February: 15- 17.
13. Colla, G.; Roupael, Y.; Cardarelli, M.; Tullio, M.; Rivera, C. M. and Rea, E. (2008): Alleviation of salt stress by arbuscular mycorrhizal in zucchini plants grown at low and high phosphorus concentration. Biol Fertl Soils 44:501–510.
14. Drobnikova, V. (1961): Factors influencing the determination of phosphatase in soil. Folia Microbial. 6: 260- 269.
15. Edrees, M. H. (1982): Studies on growth of citrus seedlings, Ph. thesis, Fac. Agric., Al-Azhar Univ., Cairo, Egypt.
16. El Fakharani, Y. M. (1999): combined effect of P fertilization and organic manuring on barley production in sandy soils. Fayoum J. Agric. Res. and Dev., 13 (2): 81- 97.
17. El-Sharkawy, S. A. (1989): Studies on mycorrhizal responses on citrus plants. M. Sc. thesis, Fac. Agric., Zagazig Univ., Zagazig, Egypt.
18. El-Shenawy, I. E. and Fayed, T. A. (2005): Evaluation of the conventional to organic and bio-fertilizers on 'Crimson Seedless' grapevines in comparison with chemical fertilizers. I- Vegetative growth and nutritional state. Egypt. J. Appl. Sci., 20 (1): 192- 211.
19. Esher, H. and Jensen, V. (1972): Aerobic chemo-organotrophic bacteria of Danish beech forest, OIKOS, 23: 248-260.
20. Fataftah, A. K.; Watia, D. S.; Ains, B. G. and Kotob, S. I. (2001): Comparative evaluation of known liquid humic analysis method. Arctech Inc., Chantilly, V. A. 20165, U.S.A.
21. Graham, J. H.; Syvertsen, J. P. and Smith, M. L. (1987): Water relations of mycorrhizal and phosphorous-fertilized on citrus under drought stress. New Phytologists, 105 (3): 411- 419.
- Harry, A.; Fred, J. and Elam, P. (1991): Growing quality table grapes in the home garden. University California pp: 1-30.
22. Godeas, A.; Fracchia, S.; Mujica, M. T. and Ocampo, J. A. (1999): influence of soil impoverishment on the interaction between *Glomus mosseae* and saprobe fungi. Mycorrhiza. 9: 185- 189.
23. Harry, A.; Fred, J. and Elam, P. (1991): Growing quality table grapes in the home garden. University California pp: 1-30.
24. Hartwigsew, L. M. and Evans, C. L. (2000): Effect of humic acids on growth and biomass of olive plants. Acta Hort., 453: 75- 80.
25. Hetrick, B. A. D. (1989): Acquisition of phosphorus by VA mycorrhizal fungi and the growth responses of their host plants. pp: 205-226. In: L. Boddy, R. Marchant, and D. J. Reid (eds.), Nitrogen, Phosphorus, and Sulphur utilization by fungi. Cambridge University Press, New York, NY, USA.
26. Hussien, A. M.; El Maghraby, T. A.; Sherif, H. M. and El Shal S. A. (2005): Effect of liquid organic fertilization techniques on yield and chemical composition of pear and apricot trees grown in sandy soils at South Tahrir province. Fayoum J. Agric. Res. And Dev., 19 (2): 224-238.
27. Ibrahiem, Alia H. (2003) Effect of sulphur sources the productive efficiency of flame seedless grapevines grown under sandy soil and drip irrigation conditions. Egypt. J. Appl. Sci., 18: 1-4.
28. Jackson, M. L. (1973): Soil chemical analysis. Constable and Company Ltd. London, 175-280.
29. Kassem, A. A.; Kassem, H. A.; Kamal, H. M. and Shalaby, M. M. (1995): the influence of nitrogen fertilizer and/or sulphur application on guava trees grown in alkaline sandy soil. J. Agric. Res., 20 (3): 1203- 1222.
30. Li-Nan, X.; Wang, B. L. and Lu, B. W. (1999): Study of the effect of apple liquid fertilizer on the growth and fruit development of Starkinson apple variety. China- Fruits, 4: 20-21.

31. Lindermann, R. G. (1988): Mycorrhizal interaction with the rhizosphere microflora: the mycorrhizosphere effect. *Phytopathology* 78:366–371.
32. Linderman, R. G. and Pflieger, F. L. (1994): General summary, p. 337- 344. In F.L. Pflieger and R. G Linderman,
33. Marin, M.; Mari, A.; Ibrarra, M. and Garacia-Ferriz, L. (2003): Arbuscular mycorrhizal inoculation of micropropagated persimmon plants. *J. Hort. Sci. and Biotechnology*, 78 (5): 734- 738.
34. Massoud, O. N. (2005): Microbiological and chemical evaluation of compost and its application in organic farming. Ph. D. thesis, department of botany, faculty of science, El-Menoufia, University.
35. Mona, G. S. (2001): Response of banana and guava plants to some biological and mineral fertilizers. M. Sc. Thesis, Fac. Agric Alexandria Univ. Egypt.
36. Morales-Payar, J. P. and Stall, W. M. (2003): Effect of substrates, boron and humic acid on the growth of papaya transplants. *Proc. Fla. State Hort. Soc.* 116: 28-30.
37. Omar, A. L. (2005): Fertilization of Thompson seedless vines with mineral and organic sources of nitrogen. *J. Agric. Sci. Mansoura Univ.*, 30 (12): 7855- 7862.
38. Rizk-Alla, M. S.; Abd El-Wahab, M. A. and Ghobrial G. F. (2006): Alleviating the harmful effects of irrigation water salinity on Thompson seedless grape rootings through the application of Nile fertile as a natural soil conditioner. *Egypt. J. of Appl. Sci.*, 21 (6): 25- 31.
39. Rhodes, L. H. and Gerdemann, J. W. (1975): Phosphate uptake zones of mycorrhizal and non-mycorrhizal onion. *New phytol.* 75: 555-561.
40. Salman, A. C. (1967): Über die mikrobielle Aktivität und ihre Beziehung zu Fruchtbarkeitsmerkmalen. Einiger Ackerböden unter besonderer Berücksichtigung der Dehydrogenaseaktivität (TTC-Reduktion). Diss Gießen, Ph.D. Thesis, W. Germany.
41. Smith, F.; Gilles, M. A.; Hamilton, J. K. and Gedess, P. A. (1956): Colorimetric methods for determination of sugar and related substances. *Anal. Chem.* 28: 350- 356.
42. Snedecor, G. W. and Cochran (1980): *Statistical methods*. 7th Ed. IOWA State Univ. Press, U. S. A.
43. Tourky, M. N.; El-Shahat, S. S. and Rizk, M. H. (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.
44. Turk, M. A.; Assaf, T. A.; Hameed, R. M. and Al-Tawha, A. M. (2006): Significance of mycorrhizae. *World journal of agricultural sciences*. 2 (1): 16-20.
45. Vitagliano, C.; Citrinesh, A. S.; Metzidakis, I. T. and Voyiatzis, D. G. (1999): Plant growth of *Olea europaea* L. as influenced by arbuscular Mycorrhizal fungi. *Proceedings of the 3rd international symposium on olive growing*. Chania, Crete Greece, 22-26 September 1997, Volume 2, *Acta Horticulture*. No 474: 357- 361.
46. Wood, C. W.; Reeves, D. W. and Himelrick, D. G. (1992): Relationships between chlorophyll meter readings and leaf chlorophyll concentration. N status and crop yield. A review: *Proc. Agro. Sco. N. Z.* 23:1-9.
47. Yilidiz, F. and Dikman, D. (1990): the extraction of anthocyanin from black grapes and black grape skins. *Doga Derigisi*, 14 (1): 57-66.
48. Zarb, J., Leifert, C. and 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, Africa.
49. Zhang, L.; Zhang, J.; Christie P. and Li, X. (2008): Pre-inoculation with Arbuscular mycorrhizal fungi suppresses root knot nematode (*Meloidogyne incognita*) on cucumber (*Cucumis sativus*). *Biol Fertil Soils*, 45:205–211.
50. Zhu, R. F. and Zhu, R. E. (2000): Effect of (Opal) HA-2002 liquid fertilizers on the growth and fruiting of pear trees. *South China Fruits*. 29 (2): 43- 47.

6/23/2010