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EFFECT OF MINERAL AND BIOLOGICAL NITROGEN FERTILIZATION ON THOMPSON SEEDLESS GRAPE TRANSPLANTS. III. EFFECT ON MICROBIAL ACTIVITY IN THE SOIL RHIZOSPHERE.

BY

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ABSTRACT

A pot experiment was carried out in 2001 and 2002 seasons on Thompson Seedless grape transplants to study the effect of two sources of nitrogen fertilizers (mineral and biological) on microbial activity in the rhizosphere of transplants. The experiment included three levels of mineral nitrogen (M_1=zero, M_2=5 and M_3=10g N / plant / year) and four levels of biological nitrogen (B_1=zero, B_2=50, B_3=100 and B_4=200ml) of liquid culture of free living nitrogen fixation bacteria *"Azotobacter chroococcum"*. Thus, the study is a factorial experiment in a randomized complete block design. The activity of *Azotobacter* expressed as counts of bacteria and CO_2 evolution in the rhizosphere were determined after 45, 90, 135 and 180 after inoculation time. Results proved that the levels of mineral nitrogen showed more or less similar values. On the contrary, the levels of biological nitrogen increased *Azotobacter* activity and the highest values were obtained by levels (B_3) &(B_4). Regarding the interaction between mineral and biological nitrogen, fertilizing with mineral nitrogen alone had no promising effect on both the count and CO_2 evolution. This is excepted because the soil of these treatments was not inoculated with *Azotobacter*. On the contrary, fertilizing with biological nitrogen alone increased counts of bacteria and CO_2 evolution compared with those of unfertilized plants. The promising biological nitrogen treatment was (M_1 x B_4) in the two seasons. However, other combinations [treatments (M_2 x B_3), (M_2 x B_4) and (M_3 x B_4)] gave the highest values of bacteria counts and CO_2 evolution

In other words, combining mineral with biological nitrogen showed more increase in the activity of the bacteria in the rhizosphere of transplants than adding any form alone and the recommended treatment was (M_2 x B_3)

Key words: Thompson Seedless, grape, *Azotobacter chroococcum*,mineral nitrogen, biological nitrogen, microbial activity, bacteria counts, CO_2 evolution

INTRODUCTION

The best known aerobic nitrogen-fixing bacteria are members of the genus *Azotobacter;* indeed *Azotobacter chroococcum* was the second free-living

nitrogen fixing microbe to be discovered after *Clostrdium pasteurianum*. Today "Azotobacter group" comprises several genera : *Azotobacter, Azomonas, Azotococcus, Beijerinckia* and *Derxia;* they are all rather similar in appearance and physiological characters and none is capable of growth without air. They also look rather alike : large granular microbes, spherical or ovoid, single or couple, capsulated, gram negative (Postgate,1978).

To measure the activity of *Azotobacter* in soil rhizosphere both the number of *Azotobacter* and CO_2 evolution are determined.

Abd-El-Malek (1971) in Egypt proved that "Most Probable Number (MPN)" method has been generally employed for counting *Azotobacter* while much of the information from abroad has been obtained from surface plate counts. The precision of two methods has been tested and it was found that the MPN estimation approximated the total cell count closely.

Alef and Nannipieri (1995) reported that soil respiration is one of the oldest and still the most frequently parameter used for quantifying microbial activities in soils. Respiration is the oxidation of organic matter by aerobic microorganisms and the end products of the process are carbon dioxide and water, so the metabolic activities of soil microorganisms can therefore be quantified by measuring CO_2 production.

Godara *et al.*, (1995), treated potted peach seedlings with different combinations of the following treatments: dipping the roots in a solution of *Azotobacter chroococcum*, inoculating the soil with VAM (*Glomus fasciculatum*), adding N fertilizer at 300 mg / kg soil. The data showed that *Azotobacter chroococcum* population in the rhizosphere increased up to 180 days then decreased; the population was higher with dual inoculation (*Azotobacter chroococcum* and *Glomus fasciculatum*) than with *Azotobacter chroococcum* treatment alone.

Thus the main goal of this work is to study the effect of two sources of nitrogen fertilizer (mineral and biological) on microbial activity in soil rhizosphere of transplants.

MATERIALS AND METHODS

The present study was conducted throughout two successive seasons (2001 and 2002) to investigate the effect of two sources of nitrogen (mineral and biological) on microorganisms activity in rhizosphere of Thompson Seedless grape transplants(*Vitis vinifera* L.). One- year- old transplants were planted in a plastic containers filled with sandy loam soil(about 22 kg / container) in a saran green house, Faculty of Agriculture, Ain Shams Univ., Shoubra EL- Khaima, Egypt.

The soil analysis indicated that the percentage of each of coarse sand, fine sand, silt, and clay were 3.7, 81.8, 3.2 and 11.4 % respectively in the first

season and 70.5, 11.2, 3.4 and 15.0 % respectively in the second season. Thus the soil texture in the two seasons was sandy loam. Chemical analysis indicated that pH 7.8 and EC (mmohs / cm) 1.05.

Ammonium sulphate (20.5%) was used as mineral nitrogen fertilizer. Mineral nitrogen treatments included three levels of nitrogen namely (M_1=zero, M_2=5 and M_3= 10g N / plant / year). Each nitrogen level was added at 20 applications at ten days intervals during the growing season from March to September in each season.

Biological nitrogen fertilizer involved a mixture of two local strains, (L_4 and L_6)of non symbiotic nitrogen fixers *Azotobacter chroococcum*. This culture was kindly provided from the Unit of Biofertilizers, Faculty of Agriculture, Ain Shams University.

Preparation of biological nitrogen fertilizer:-

Biological nitrogen fertilizer of *Azotobacter chroococcum* L_4 and L_6 was obtained separately as follows:-

Loop full of *Azotobacter chroococcum* L_4 culture slant, was inoculated into conical flask containing 1000 ml modified Ashby s' liquid medium (Abd El-Malek and Ishac, 1968)as shown in Table (1), then incubated at 28 ± 2 C° on rotary shaker (160 rpm). After 5 days incubation period, the examination of 1 ml of the culture indicated that it contained approximately 10^8 cells. From the above culture, 600 ml were transferred in a conical flask and completed to 6 liter by adding modified Ashby s' liquid medium. Thus the concentration of culture is (10%, v : v). After incubating the flask for 6 days at 28 ± 2 C°, it was found that 1 ml of the culture contained approximately 10^8 cells.

Another similar preparation was carried out but from L_6 strain. Thereafter a mixed culture was prepared by mixing the two strains of bacteria of bacteria (L_4 and L_6).

The treatment of the biological nitrogen fertilizer included four levels of nitrogen biofertilizer namely :

B_1 = the control which treated with tap water.
B_2 = treated with 50 ml of liquid culture.
B_3 = treated with 100 ml of liquid culture.
B_4 = treated with 200 ml of liquid culture.

It should be pointed out that The bacteria count in the stock fresh liquid culture was ca. 16.0 x 10^8 cell / ml. Before applying biological nitrogen fertilizer and to secure covering the soil rhizosphere with the bacteria, four holes (each of 15 cm in length x 1.5 cm in diameter) were dug in the soil of each pot, then the given amount of each level was applied to the surface of each pot. All biological nitrogen treatments were applied in the first week of April in each season.

Accordingly, the study involved three levels of mineral nitrogen and four levels of biological nitrogen in a factorial experiment in a randomized complete

block design. Each treatment was replicated five times and each replicate was represented by two plants

Microbial analysis:

For the microbial analysis, soil samples were taken at a depth of 15cm from the top of the soil in the pot to secure sampling from the rhizosphere zone. Soil samples were taken at four different dates after the inoculation data [after 45 days (May 15th), after 90 days (the first of July), after 135 days (August 15th) and after 180 days (the first of October) in each season]. On each sampling date, three soil samples (replicates) were taken from each treatment to determine cell number of azotobacters and CO_2 evolution as follows :

2-1 Determination of cell number of *Azotobacter chroococcum*:-

The method of "Most Probable Number (MPN)" was used to estimate the population density (Papen and Von Berg, 1998). From each soil sample 10 g fresh weight was suspended in 90 ml sterile tap water then shakened for 30 minutes at room temperature (25°C). The soil suspension was allowed to settle for 2 minutes before the supernatant containing the bacterial cells was decanted in a sterile conical flask. From this supernatant a series of dilutions (10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6} and 10^{-7}) were prepared. From each dilution a set of five test tubes were prepared by transferring 1 ml in each test tube containing 5 ml sterile "Modified Ashby's Liquid Medium"

All the above procedures were carried out under sterile conditions and the tubes were incubated at 28 ± 2 °C for 2 weeks.

Table (1):Modified Ashby's Liquid Medium.

Constituents	g / l
Mannitol	10.00
Sucrose	10.00
K_2HPO_4	0.200
$MgSO_4.7H_2O$	0.200
NaCl	0.200
$CaSO_4$	0.100
$MnSO_4.4H_2O$	0.001
$FeCl_3.6H_2O$	0.001
$Na_2Mo_2O_4.2H_2O$	0.001
$CaCO_3$	5.00

After the incubation period, all tube sets of different dilutions were examined for the existence of the brown ring on the surface of the media which indicated the growth of bacteria (positive media tubes). For each soil sample, the dilution which showed negative effect (no brown ring in all the five tubes) was used as a control and the above three higher dilutions were selected and number of positive tubes [with brown ring] in each set (5 tubes) were counted and the corresponding MPN figure[*Azotobacter* (n)] was obtained from the statistical

table of " Most Probable Number" (Cochran, 1950). Therefore, total azotobacters count was obtained from the following equation :

$$Azotobacter(n) \times \frac{1}{dilution\ rate} \times soil\ mositur\%$$

2-2 Estimation of carbon dioxide evolution rate:-

The rate of CO_2 evolution was determined in the rhizosphere according to the method described by Promer and Schmidt(1964) and modified by (Shehata, 1972). In this procedure, 20 g fresh soil sample was placed in a cylindrical polyethylene bag (which allows passage of CO_2) and tied with a thin wire. Each bag was placed in 500 ml bottle containing 25 ml NaOH (0.05N). Bags were hanged in the bottle by tying the other end of the wire with the rubber stopper at the top of the bottle and fitted closely to avoid any loss of CO_2. Each treatment was represented by three bottles and the blank (with NaOH solution, but without soil sample) was also represented by three bottles. All bottles were then incubated at 30 °C for 72 hours.

After incubation, polyethylene bags were taken out from the bottles then 3 ml barium chloride solution (0.5 M) and 2-3 drops of phenolphthalein indicator were added to the solution in the bottle and then titrated by HCl (0.05M) under continuous stirring until the color changed from red to colourless.

The rate of CO_2 evolution was calculated by the following equation (Isermeyer, 1952 as quoted by Alef and Nannipieri (1995))

$$CO_2(mg)/sw/t = \frac{(v^\circ - v) \times 1.1}{dwt}$$

where, sw = the amount of soil dry weight in gram.
t = the incubation time in hours.
v° = amount (ml) of HCl used for the blank.
v = amount (ml) of HCl used for the soil sample.
dwt = the dry weight of 1 g moist soil.
1.1= the conversion factor (1 ml.05 M NaOH = 1.1 mg CO_2).

Data of microbial parameters in the two seasons were statistically analyzed by using the analysis of variance (Snedecor and Cochran, 1980). Means were differentiated by using Duncan's multiple range test at 5 % (Duncan, 1955).

RESULTS AND DISCUSSION

1-Effect on counts of *Azotobacter chroococcum* in rhizosphere of transplants:

Results in Table 2 show that the counts of *Azotobacter* were gradually increased from May (after 45 days from inculcation) and reached the maximum in August (after 135 days from inculcation). Thereafter the counts decreased at the end of the season in October (after 180 days from inculcation). This means that *Azotobacter* started their activity just after inoculation in the soil (in spring) and reached the maximum activity in summer then the activity reduced at the end of the fall.

Table (2) : Effect of mineral and biological nitrogen fertilization on counts of *Azotobacter chroococcum* (n x 10^4 cell g^{-1} dry soil) in the rhizosphere of Thompson Seedless grape transplants after different periods of inoculation in 2001 and 2002 seasons .

Treat .of Bio. N* (ml/plant)	Treatments of Mineral nitrogen (g N /plant)															
	Days after inoculation															
	After 45 days				After 90 days				After 135 days				After 180 days			
	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean
	2001 season															
0 (B_1)	1.40f	1.78f	1.60f	1.59D'	3.30g	4.40g	3.37g	3.69C'	7.30f	8.66f	9.70f	8.55D'	5.00f	3.93f	2.70f	3.88D'
50 (B_2)	5.20e	6.80de	8.70cd	6.90C'	24.00f	24.70f	37.30e	28.67B'	46.67e	50.00e	96.67d	64.45C'	35.67e	52.00d	76.67c	54.78C
100(B_3)	4.60e	11.30bc	19.70a	11.87B'	33.70e	55.30c	82.67a	57.22A'	113.3d	170.00c	208.33ab	163.88B'	83.67c	97.30b	136.67a	105.88A'
200(B_4)	15.30b	20.30a	18.30a	17.3A'	50.00d	72.00b	53.30cd	58.43A'	166.67c	213.33a	180.00bc	186.67A'	83.00c	110.0b	53.00d	82.00B
Mean	6.13C	10.05B	12.08A		27.75C	39.10B	44.16A		83.49B	110.50A	123.68A		51.84B	65.81A	67.26A	
	2002 season															
0 (B_1)	1.96d	1.47d	1.03d	1.49C'	3.03f	1.40f	1.60f	2.01C'	4.50f	4.70f	4.93f	4.71D'	1.53f	2.10f	4.00f	2.54D'
50 (B_2)	8.53d	5.90d	3.37d	5.93C'	27.0b-d	17.33e	23.0o-e	22.44B'	61.33e	123.33d	60.47e	81.71C'	33.00d	47.00c	16.00e	32.00C
100(B_3)	13.33cd	32.33ab	8.77d	18.14B'	33.33b	49.00a	20.00de	34.11A'	56.00e	183.33a	160.0b	133.11A'	20.67e	63.00b	45.00c	42.89B
200(B_4)	38.00a	34.33a	22.00bc	31.44A'	31.33bc	28.0b-d	46.67a	35.33A'	61.00e	163.33b	145.0c	123.11B'	34.00d	93.33a	50.30c	59.21A
Mean	15.45A	18.51A	8.79B		23.67A	23.93A	22.82A		45.71C	118.67A	92.60B		22.30C	51.36A	28.83B	

* Biological nitrogen = nitrogen fixation bacteria (*Azotobacter chroococcum*).

In each month in each season, means of each of mineral and biological nitrogen levels or their interactions having the same letters are not significantly different at 5% level

Results in Table 2 show that counts of *Azotobacter chroococcum* were affected significantly by levels of mineral nitrogen, biological nitrogen and their interaction.

Effect of mineral nitrogen levels:

In both the two seasons, level (M_1) gave the least significant values in any given date. However, in the first season values were increased gradually by adding mineral nitrogen and the highest counts values were obtained by level M_3 although the differences between levels M_2 and M_3 were not significant in the first half of the growing season. In the second season level M_2 gave higher values than those of level M_3.

Therefore, it seems that mineral nitrogen fertilizer irrespective of level of application increased *Azotobacter* counts and level (M_2) may be promising in this respect.

Effect of biological nitrogen levels :

In the two seasons, level (B_1) gave the least significant values of azotobacters counts. However, *Azotobacter* counts in most cases were gradually increased by increasing the level of biological nitrogen up to 200 ml / plant (level B_4).

The interaction between mineral and biological nitrogen:

In the two seasons, unfertilized transplants [treatment (M_1 x B_1)] gave the lowest values of *Azotobacter* counts in any given date except after 45 & 90 days from inoculation in the second season.

In the two seasons, fertilization with mineral nitrogen alone [treatments(M_2 x B_1) and (M_3 x B_1)] gave more or less similar values as that of unfertilized treatment. This is excepted because the soil of these treatments was not inoculated with *Azotobacter*.

Generally, fertilization with biological nitrogen alone[treatments (M_1 x B_2), (M_1 x B_3) and (M_1 x B_4)] increased *Azotobacter* counts and in most cases treatment (M_1 x B_4) gave the highest values of *Azotobacter* counts in the two seasons. In other words the higher the inoculation level, the higher was the count of *Azotobacter* in the rhizosphere.

Regarding other combinations, it is observed in the two seasons that under the second and third levels of mineral nitrogen, increasing the level of biological nitrogen up to 200 ml / plant increased azotobacters counts. So, the highest values were obtained by treatments (M_2 x B_3), (M_2 x B_4) and (M_3 x B_4).

Therefore, it could be concluded that *Azotobacter* counts were affected significantly by different treatments. Mineral fertilization alone had slight or no effect on *Azotobacter* counts but biological fertilization alone increased *Azotobacter* counts more than those obtained by mineral fertilization alone and the higher the concentration of inoculation, the higher was the count of *Azotobacter*. However, combining mineral fertilization with biological fertilization induced more increase in *Azotobacter* counts when compared with biological fertilization alone.

In This respect, Awasthi *et al.*, (1996), indicated that the spore number and root colonization increased in the rhizosphere of one year old peach seedlings with the increase in period after inoculation with VAM and *Azotobacter* strain. However, mineral fertilizers application increased the spore number and root colonization.

1-2Effect on respiration of microorganisms in the rhizosphere of transplants:

Generally, results in Table 3 show that CO_2 evolution of microorganisms (mainly *Azotobacter chroococcum*) in the rhizosphere was increased gradually from the beginning of the growing season in May (after 45 days from inoculation) and attained higher values in August (after 135 days from inoculation). The evolution of CO_2 decreased slightly after 135 days from inoculation and reached the lowest values in October (after 180 days from inoculation).

Results in Table 3 show that CO_2 evolution was affected significantly by levels of mineral and biological nitrogen and their interaction.

Effect of mineral nitrogen levels:

In both the two seasons, level (M_1) gave the least significant values in most cases. In the first season, level (M_2) and (M_3) increased CO_2 evolution however, level (M_2) gave the highest values in most cases. In the second season, the three nitrogen levels behaved similarly on the first and second dates. However, on the latter two dates the second and third levels showed higher values than that of the lower level (M_1).

Therefore, it is clear that different levels of mineral nitrogen showed more or less similar values of CO_2 evolution but level (M_2) and (M_3) showed higher values than that of level (M_1).

Effect of biological nitrogen levels :

The low level of biological nitrogen (B_1) showed the least values in any given date in the two seasons. The CO_2 evolution was increased by increasing the concentration of biological nitrogen. However, in most cases, the highest values were obtained by levels (B_3) and (B_4) and the differences between them were too small and insignificant.

The interaction between mineral and biological nitrogen:

In the two seasons, transplants fertilized with mineral nitrogen alone [treatments(M_2 x B_1) and (M_3 x B_1)] had similar effect as that of the unfertilized plants (M_1 x B_1).

Fertilizing with biological nitrogen alone [treatments(M_1 x B_2), (M_1 x B_3) and (M_1 x B_4)] in both the two seasons, increased CO_2 evolution gradually as the rate of biological nitrogen increased as compared with unfertilized plants(M_1 x B_1). Almost the highest values were obtained by treatment (M_1 x B_4).

Table (3) : Effect of mineral and biological nitrogen fertilization on the rate of CO_2 evolution (mg CO_2 /g / 72h) of microorganisms in rhizosphere of Thompson Seedless grape transplants after different periods of inoculation with *Azotobacter chroococcum* in 2001 and 2002 seasons .

Treat .of Bio. N* (ml/plant)	Treatments of Mineral nitrogen (g N / plant)															
	Days after inoculation															
	After 45 days				After 90days				After 135 days				After 180 days			
	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean	0(M_1)	5(M_2)	10(M_3)	Mean
2001 season																
0 (B_1)	5.22h	4.87i	5.46g	**5.18D**	5.00h	6.73f	5.50g	**5.74C**	6.05f	5.60f	4.83g	**5.49C**	4.96i	4.26j	4.26j	**4.49D**
50 (B_2)	7.15e	7.56d	7.63d	**7.45C**	9.71e	9.93de	10.40c	**10.01B**	11.50e	13.51c	11.41e	**12.14B**	10.26g	11.53e	10.80f	**10.86C**
100(B_3)	6.98f	8.84b	8.83b	**8.22B**	10.03oe	11.50b	13.13a	**11.55A**	12.25d	15.37b	16.80a	**14.81A**	12.13c	12.33bc	12.43b	**12.30A**
200(B_4)	8.38c	8.99a	8.90ab	**8.76A**	10.20cd	10.20cd	9.93de	**10.11B**	13.75c	16.16a	15.10b	**15.00A**	11.80d	12.90a	10.00h	**11.57B**
Mean	**6.93C**	**7.57B**	**7.71A**		**8.74B**	**9.59A**	**9.74A**		**10.89C**	**12.66A**	**12.04B**		**9.79B**	**10.26A**	**9.38C**	
2002 season																
0 (B_1)	5.90e	5.50e	4.98e	**5.46C**	4.86d	5.20d	3.98e	**4.68C**	6.00e	6.50e	5.99e	**6.16D**	4.43de	4.27e	5.73d	**4.81D**
50 (B_2)	8.70b-d	7.95d	7.43d	**8.03B**	9.99b	9.00c	9.73b	**9.57B**	13.18c	10.16d	14.90b	**12.75C**	9.99bc	9.23c	9.26c	**9.49C**
100(B_3)	9.62a-c	10.17a	8.53cd	**9.44A**	10.36b	11.56a	9.84b	**10.59A**	13.42c	17.89a	15.21b	**15.51A**	9.00c	12.46a	10.00bc	**10.49B**
200(B_4)	9.80a-c	10.00ab	9.99ab	**9.93A**	10.12b	9.83b	11.33a	**10.43A**	15.21b	15.00b	14.95b	**15.05B**	10.03bc	13.10a	11.03b	**11.39A**
Mean	**8.51A**	**8.41A**	**7.73B**		**8.83A**	**8.90A**	**8.72A**		**11.95C**	**12.39B**	**12.76A**		**8.36B**	**9.77A**	**9.01B**	

* Biological nitrogen = nitrogen fixation bacteria (*Azotobacter chroococcum*).

In each month in each season, means of each of mineral and biological nitrogen levels or their interactions having the same letters are not significantly different at 5% level

Other combinations, gave variable results in the two seasons. However, it is clear that CO_2 evolution was increased by combining levels(M_2)or (M_3) of mineral nitrogen with levels (B_3) or (B_4) of biological nitrogen.

Thus, it is clear that mineral nitrogen alone gave low values of CO_2 evolution as those of the unfertilized transplants in the two seasons. This is excepted because transplants of these treatments were not inoculated with *Azotobacter*.

On the contrary, biological nitrogen alone especially the high levels [(B_3) or (B_4)] or other combinations especially [treatments (M_2 x B_3), (M_2 x B_4) and (M_3 x B_4)] created more increase in CO_2 evolution.

In other word, fertilizing with biological nitrogen alone or combined with mineral nitrogen fertilization increased CO_2 evolution in the rhizosphere of transplants which indicated a high activity of azotobacters.

These results are in accordance with those of Abo El- Ala (2002) and Kandeel and Sharaf (2003), who reported that statistical main effects on the rate of CO_2 evolution due to application of multi-biofertilizer inoculants in the presence of half or full dose of mineral nitrogen and phosphorus was significantly higher, compared with that of uninoculated marjoram plants (*Majorana hortensis* L.).

GENERAL CONCLUSION

With respect to the microbial activity it could be concluded that, *Azotobacter* activity as measured by the count of bacteria or CO_2 evolution in the rhizosphere of transplants, was higher at the beginning of the growing season than that at the end of the growing season in October. In other words activity of microorganism including the inoculated *Azotobacter chroococcum* seems to be related with degrees of temperatures which were suitable during spring and summer but activity reduced in early autumn when temperature decreased. However, in both the two seasons, fertilizing with mineral nitrogen alone had similar effect on microbial activity as those of the unfertilized treatment. However, biological nitrogen alone increased microbial activity and the highest values were obtained by the highest rate of *Azotobacter* inoculation, level (B_4). Combining mineral with biological nitrogen gave more increase in microbial activity.

Consequently it could be concluded that, microbial activity was not affected significantly by mineral nitrogen fertilization but the application of *Azotobacter* alone to the rhizsphere increased the activity and more increase was obtained by the application of a combination of mineral and biological nitrogen. The maximum effect was obtained by treatments (M_2 x B_3) (M_2 x B_4) and (M_3 x B_4) which gave the highest values of microbial activity. No doubt the increase in microbial activity will increase fixed nitrogen in the rhizosphere and possible some nutrients and other biological substance which in turn will stimulate the absorption of such compounds and increase the growth of transplants. This results

could be early elucidated from the preceding data of the effect of mineral and biological nitrogen treatments on the vegetative growth (part I) and nutrient content (part II)on Thompson Seedless grape transplants.

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تأثير التسميد النتروجيني المعدني والحيوى على شتلات العنب البناتى صنف طومسون سيدلس

III- التأثير على النشاط الميكروبي في مجال الجذور.

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أجريت تجربة أصص خلال موسمي ٢٠٠١ - ٢٠٠٢ على شتلات العنب البناتى ' صنف طومسون سيدلس ' لدراسة تأثير مصدرين من التسميد النتروجينى (معدنى و حيوى) على النشاط الميكروبى فى مجال الجذور وقد اشتملت التجربة على ثلاثة مستويات من التسميد النتروجيني المعدنى (م١ = صفر، م٢ =٥٠، م٣ =١٠٠ جم ن / نبات / سنة) وأربعة مستويات من التسميد النتروجينى الحيوى (ح١ = صفر، ح٢=٥٠، ح٣= ١٠٠، ح٤=٢٠٠ مل من المزرعة السائلة ل *Azotobacter chroococcum*) فى تجربة عاملية فى تصميم قطاعات كاملة العشوائية.

و قد أوضحت النتائج أن النشاط الميكروبى (الذى تم التعبير عنه بأعداد البكتريا و ثانى أكسيد الكربون المنبعث فى المنطقة المحيطة بجذور النبات بعد ٤٥ ، ٩٠ ، ١٣٥ ، ١٨٠ يوم من التلقيح) وذلك خلال موسمى الدراسة أن العامل الاول وهو مستويات السماد المعدنى المختلفة قد فى حين أن العامل الثانى و هو مستويات السماد الحيوى قد زاد من النشاط الميكروبى بزيادة التركيز المستخدم من السماد الحيوى خاصة بالمستويات (ح٣)& (ح٤).

وبالنسبة للتفاعل وجد أن التسميد بمعاملات السماد المعدنى بمفرده لم يؤثر على كل من أعداد البكتريا و ثانى أكسيد الكربون المنبعث. و من جهة أخرى فأن التسميد بالسماد الحيوى بمفرده أدى الى زيادة أعداد البكتريا و كذلك ثانى أكسيد الكربون المنبعث مقارنة بالنباتات غير المسمدة وكانت أفضل معاملات التسميد الحيوى بمفرده هى (م١ × ح٤) و ذلك خلال الموسمين. أما بالنسبة للتفاعل فان التركيبات المختلفة وخاصة(م٢ × ح٣) ،(م٢ × ح٤)، (م٣ × ح٤) أعطت أعلى نشاط ميكروبى. ومن ذلك يستخلص ان التسميد بالتركيبات المختلفة بين السماد المعدنى والحيوى ادت الى زيادة نشاط البكتريا عما اذا اضيفت كل صورة بمفردها أفضل المعاملات الموصى بها هى التسميد بالمستوى الثانى معدنى مع الثالث حيوى (م٢ × ح٣). و بدون شك أن زيادة النشاط الميكروبى سوف يؤدى الى زيادة النيتروجين المثبت فى المنطقة المحيطة بالجذور بالإضافة إلى تيسير بعض العناصر الأخرى مما انعكس على زيادة نمو الشتلات.