

RESPONSE OF PEANUT TO DIAZOTROPHIC INOCULATION AND N FERTILIZATION IN SANDY SOILS IN PRESENCE OF NEMATODES- CONTROLLING AGENTS

A. F. SHAHABY¹, A. G. RAHAL², HEBA SH. SHEHATA²,

AND M. FAYEZ¹

¹ Department of Microbiology, College of Agriculture, Cairo University, Giza 12613, Egypt.

² Agricultural Microbiology Research Department; Soils, Water and Environment Research Institute; ARC; Giza 12613, Egypt

(Manuscript received 11 November 1998)

ABSTRACT

A field trial was executed in Ismailia sandy soil. Biomass, nodulation, N₂-fixation and yield of inoculated peanut cv. Giza 5 were monitored in presence of N fertilizer and nematodes-controlling agents (mocap and nemaless). Two rhizobial inocula were used, okadin (Single culture of *Bradyrhizobium* spp.) and rhizobacterin (dual inoculum of *Bradyrhizobium* spp. and *Azotobacter chroococcum*). Results demonstrated that higher biomass yield (2.44 - 4.95 ton fed⁻¹) was reported by okadin inoculation against 2.06-2.58 ton fed⁻¹ due to rhizobacterin. Peanuts treated with mocap (synthetic nematicide) and nemaless (bio-product) were higher in biomass than those untreated. Okadin supported higher nodule biomass compared to rhizobacterin whether N fertilization was applied or not, such effect was more pronounced with nematodes-controlling agents-treated plants. Nematodes-controlling agent supported better nodulation of uninoculated plants where nodule numbers increased by 21.3 and 5.6% due to treatment with mocap and nemaless, respectively. Nemaless-treated peanuts bore nodules with a dry weight of 1.42 g plant⁻¹ compared to 1.13 g plant⁻¹ for those supplied with mocap. Rhizobacterin, in absence of N fertilizer, produced the highest pod yield (1.94 ton fed⁻¹) while the lowest (1.65 ton fed⁻¹) was recorded by either untreated plants or those fertilized with 45 kg N fed⁻¹. Nemaless-treated peanuts did produce superior pod yield approximating 23% higher than that due to mocap treatment. Inoculated plants, with rhizobacterin in particular, received no N-fertilizer exhibited higher acetyle reducing activity on their roots compared to those simultaneously fertilized with 45 kg N fed⁻¹. Protein and peel yields considerably varied depending upon treatment.

INTRODUCTION

Desert soils are faced by several adversal and detrimental conditions, i.e. poor nutrients, low organic carbon, low precipitation, high salinity and/or drought (Shahaby, 1988 and Athar, 1998). Great efforts had been done, during, last decades, for minimizing losses and increasing nitrogen use efficiency in desert soils.

Diazotrophs could be used as biofertilizer to substitute a part of nitrogen requirement for peanut (*Arachis hypogaea*) nutrition and increasing fertility of newly reclaimed desert soils. The concept of biofertilization is not new to Egyptian agriculture. Recently, the application of bioorganic farming attracted the attention to biofertilizers. Most agronomists are now in favour of the use of biofertilizers to conserve environment and reduce agricultural costs.

Peanut is an important legume crop in newly reclaimed sandy soils in Egypt. Legume cultivation in these soils, undoubtedly, requires careful management, but once limiting factors are defined, adequate input strategies should allow maximization of biological nitrogen fixation (BNF) process. Although nitrogen is an important nutrient in such poor sandy soils, its application is considered of economical and environmental risks. Many problems are encountered in peanut production in sandy soil, including nematodes. Nematicides applied for nematodes control, may exhibit side effects on beneficial nontarget microorganisms in the ecosystem (Yueh and Hensley, 1993 and El-Sherif *et al.*, 1995). Fayez (1990) pointed out that nematicides promoted growth of corn (*Zea mays*) inoculated with *Azospirillum lipoferum*. Therefore, the effects of nematicides on *Bradyrhizobium* spp. demand the elucidation of possible interactions of nematodes-controlling agents with legume inoculants of N₂-fixers.

The purpose of this study is to evaluate the effects of two commercial rhizobial inocula, okadin (single culture of *Bradyrhizobium* spp.) and rhizobacterin (dual inoculum of *Bradyrhizobium* spp. and *Azotobacter chroococcum*) on peanut growth, nodulation, nitrogen fixation and yield in presence of N fertilizer and nematodes-controlling agents in sandy soils.

MATERIALS AND METHODS

Site and crop

This study was conducted at Ismailia Experimental Station, Agricultural Research Center. The experimental area lies at 33° east and 30° south at an altitude of 13 m above sea level and characterized by the following climate; a) rainfall is scant, irregular and variable; b) temperature fluctuates from 13 to 23 °C in winter and 19 to 29 °C in summer; c) relative humidity is 50-70% and d) abundance of sunshine.

Peanut, *Arachis hypogaea* (cv. Giza 5) seeds were obtained from Field Crops

Research Institute, ARC. Initially, germination rate of seeds was determined, the estimate was 90%. Seeds were thoroughly washed in water prior to treatment to get rid of any traces of pesticides possibly added for pest control during storage.

Soil analysis :

soil mechanical and physico-chemical characteristics were: coarse sand 61.8%, fine sand, 36.4%, silt 0.9%, clay 0.9%, CaCO₃ 1.6%, total carbon 0.029%, total nitrogen 0.007%, E.C. (dSm⁻¹) 0.42 and pH 7.7. Cations and anions (meq l⁻¹) were: Ca⁺⁺ 0.37, Mg⁺⁺ 0.31, K⁺, 0.13, Na⁺ 0.56, HCO₃⁻ 0.41, Cl⁻ 0.36; and SO₄=0.61 (piper, 1950 and Jackson, 1973).

Inoculation

Single and dual diazotrophic inocula were used to inoculate peanut. Rhizobacterin (dual inoculum of *Bradyrhizobium* spp. and *Azotbacter chroococcum*) and okadin (single culture of *Bradyrhizobium* spp.) are microbial preparations, the first is produced by General Organization for Agriculture Equalization Fund in Egypt (GOAEF) and the second is prepared by Agricultural Microbiology Research Department, Soils, Water and Environment Research Institute, ARC, Ministry of Agriculture.

Pest-controlling agents

Nematodes possibly existing in soil were controlled via two different agents. The first is mocap which is a synthetic chemical product, (O-ethyl- S, S-dipropyl phospho-dithioate), added at the rate of 15 kg fed⁻¹ prior to planting. The second is nemaless, a biological preparation, produced by Soils, Water and Environment Research Institute, ARC as biofertilizer and nematodes-controlling compound as well. Nemaless water- solution (1:100 v/v) was dropped on soil surface adjacent to root system. Treatment was repeated for 120 days at monthly intervals.

Experimentation

All the recommended agricultural practices (ploughing, compacting, fertilization and watering) were employed. Plots of 10.5 m² (3.0 x 3.5 m) were supplemented with superphosphate and potassium sulfate at the rates of 200 and 50 Kg fed⁻¹, respectively. Nitrogen as ammonium sulfate was added at the rate of 90 and 45 Kg fed⁻¹ in three equal doses: before planting, 45 and 75 days after. Inoculation was carried out by mixing seeds with Arabic gum and either rhizobacterin or okadin. Seeds were drilled in rows 50 cm apart and 3 m long at the rate of 10 Kg fed⁻¹.

Plants were then thinned to 10 plants per row. Mocap and nemaless were added to soil as previously mentioned. Seven treatments were applied: 1) 0 N (control), 2) 90 Kg N fed^{-1} , 3) 45 Kg N fed^{-1} , 4) Okadin, 5) Rhizobacterin, 6) 45 Kg N fed^{-1} + Okadin, and 7) 45 Kg N fed^{-1} + Rhizobacterin.

After 45, 75 and 110 days of sowing, peanut plants were uprooted and determined for shoot height, dry weight (at 80°C for 48 h) and nitrogen content (Bremner, 1965). Roots were washed free of soil and a part was selected to determine number and dry weight of nodules. Another set of roots was measured for acetylene reducing activity according to Dart *et al.* (1972). Pod yield was determined at harvest. Dehydrogenase and nitrogenase activities of soil of the different treatments were estimated according to Skujins (1973) and Hardy *et al.* (1973), respectively.

Data analysis

Data were statistically analyzed by least significant difference according to Steel and Torrie (1980).

RESULTS AND DISCUSSION

Plant growth

Shoot height of peanut plants hardly fluctuated among the various treatments (Fig.1). Regarding nematodes- controlling agents- untreated plants, the tallest were those inoculated okadin and 45 Kg N fed^{-1} fertilization regime. Inoculation with rhizobacterin decreased the peanut development by 6% compared to okadin. Plants inoculated with diazotrophs and received no N-fertilizer were significantly similar in heights to those supplemented with N-fertilizer. Irrespective of inoculation and fertilization, no significant response to the applied nematodes-controlling agents was observed, although reductions of 2.9 and 4.4 % were scored in shoot length due to mocap and nemaless treatments, respectively.

Biomass yield was increased continuously and progressively with plant age. No significant differences in biomass yield of the 45-day old peanuts were recorded between the applied treatments (Table 1). Response of the 75-day old leguminous plant to mineral N fertilization was more obvious in presence of nematodes - controlling agents. For better understanding of this observation, it has to be mentioned that while such agent-untreated plant showed no increase in dry matter production, those treated with mocap and nemaless were 6.5 and 14.7% higher than

the control. This phenomenon was extended to the 110-day old plants with higher response whereas the percentage increases raised to 52.4 and 84.1, respectively. Inoculation with diazotrophs (Okadin or rhizobacterin) had no effect on biomass yield of young plants, such effect was more pronounced with older ones. In this respect and in absence of N fertilizer, averages of 30.3, 12.7 and 40.9% over control were recorded for untreated-, mocap- and nemaless-treated 110-day old peanuts. The respective increases in presence of a rational dose of N fertilizer were 44.8, 72.0 and 0.7%. These findings indicate that nemaless seems to be more effective than mocap for nematodes control particularly in non N fertilized Plants. Regardless of plant age, the superior plant vigorousness (average of 946.7 g/ 10 plants) was scored for peanuts supplemented with 90 kg fed⁻¹ and treated with nemaless. Similar yield (average of 947.3 g/10 plant) was obtained by okadin-inoculated plants received mocap and 45 kg N fed⁻¹. In general, plants treated with mocap and nemaless were 19.9 and 24.4% higher than untreated ones.

Table 1. Biomass yield (g/10 plants) of peanut inoculated with diazotrophs and received mineral N and nematode- controlling agents.

Treatments	Harvest (days)		
	45	75	110
Untreated plants			
0 N, Control	116 0	734.7	773 5
45 kg N fed ⁻¹	125.2	518.1	739 5
90 kg N fed ⁻¹	165 7	473 3	1210 6
Okadin	117 1	777 1	1158 0
Rhizobacterin	142 1	666.3	859.2
45kg N fed ⁻¹ + Okadin	166 0	537 3	1107.3
45kg N fed ⁻¹ + Rhizobacterin	107 6	509 6	1132 0
Mocap-treated plants			
0 N, Control	117 3	848 2	907 7
45 kg N fed ⁻¹	189 7	895 1	924 8
90 kg N fed ⁻¹	91 3	903 7	1383 7
Okadin	94 3	740 0	1015 5
Rhizobacterin	145 3	553 8	1020 8
45kg N fed ⁻¹ + Okadin	76 7	785 3	1980 0
45kg N fed ⁻¹ + Rhizobacterin	107 0	623 2	1142.4
Nemaless-treated plants			
0 N, Control	231.3	572.7	1115 3
45 kg N fed ⁻¹	139 3	546.8	1079.7
90 kg N fed ⁻¹	129 3	657.0	2053.7
Okadin	136.7	559.9	2063 4
Rhizobacterin	92 1	784 6	1079 5
45kg N fed ⁻¹ + Okadin	340 0	523 8	1161 8
45kg N fed ⁻¹ + Rhizobacterin	73 3	674 9	1084 9

(LSI) at 0.05 is 288.7 and at 0.01 is 381.8

Plant nitrogen content

Table (2) demonstrates that peanut plants of the control treatment contained the lowest quantities of N in their shoot tissues (0.3 - 7.5 g/10 plants) and symptoms of N deficiency appeared as early as 40 days after sward establishment, this was more obvious with nematodes controlling agent-untreated plants. In general, plant nitrogen content increased as plant age increased due to plant biomass increases. Inocula applied resulted in higher N assimilations by growing plants although the effect was insignificant in most cases. The impact of inoculation was more pronounced when combined with the rational N fertilization level of 45 kg N fed⁻¹ with percentage increases of 140.5, 187.5 and 9.9 over only inoculated plants for nematodes-controlling agent untreated mocap and nemaless-treated peanuts, respectively. Rhizobacterin supported higher N yield than okadin. As expected, plants supplemented with full N dose (90 Kg N fed⁻¹) showed the highest N accumulation. Reducing the fertilization rate to 45 kg N fed⁻¹, in absence of diazotrophs, lowered the N yield by 2.6 - 49.0% for the various treatments being the worst with plants of the control treatment. Regardless of inoculation and fertilization treatments, nemaless-treated peanuts scored the highest N yield average of 10.6 g/10 plants.

Table 2. Nitrogen content(g/10 plants) of 45-,75- and 110-day old peanuts of different treatments

Treatments	Harvest		
	45 day	75 day	110 day
	Untreated plants		
Control	0.6	2.3	3.4
45 kg N fed ⁻¹	3.5	9.5	15.5
90 kg N fed ⁻¹	5.1	15.4	30.4
Okadin	0.4	2.0	1.8
Rhizobacterin	4.5	4.9	8.8
45kg N fed ⁻¹ + Okadin	1.2	4.3	14.3
45kg N fed ⁻¹ + Rhizobacterin	3.6	7.5	22.5
	Mocap treated plants		
Control	0.3	2.8	4.2
45 kg N fed ⁻¹	4.5	9.1	11.7
90 kg N fed ⁻¹	3.0	15.0	30.0
Okadin	0.9	1.8	5.5
Rhizobacterin	0.7	3.6	6.5
45kg N fed ⁻¹ + Okadin	1.3	9.0	19.5
45kg N fed ⁻¹ + Rhizobacterin	2.6	4.5	18.2
	Nemaless-treated plants		
Control	1.2	4.8	7.5
45 kg N fed ⁻¹	3.6	16.0	26.2
90 kg N fed ⁻¹	3.6	18.7	37.5
Okadin	3.7	8.3	11.3
Rhizobacterin	4.2	8.2	13.0
45kg N fed ⁻¹ + Okadin	2.5	6.3	14.3
45kg N fed ⁻¹ + Rhizobacterin	2.3	8.9	18.9

(LSD) at 0.05 is 6.8 and at 0.01 is 9.0

Nodulation

Considerable high numbers of nodules were formed on roots of non-inoculated peanuts indicating that native rhizobia of the leguminous plant are present in the experimental site in high population. Nematodes-controlling agents supported better nodulation of non-fertilized uninoculated plants where nodule numbers increased by 21.3 and 5.6% due to treatment with mocap and nemaless, respectively. Mineral N fertilization, in the majority of cases, did inhibit peanut nodulation particularly with plants received neither mocap nor nemaless. Plants inoculated with diazotrophs beared the highest numbers of nodules on their root system being 250,189 and 196 nodules per plant in average for untreated-, mocap-and nemaless-treated plants, respectively (Fig.2). Fertilization with 45 kg N fed^{-1} decreased nodule production except for nemaless-received plants. Irrespective of treatment and sampling date, nodule formation was promoted due to nemaless application with an average increase of 4.3%, such effect was not observed with mocap.

As expected, nodule weights progressively increased with plant age. Okadin supported higher nodule biomass compared to rhizobacterin, whether N fertilizer was applied or not, such effect was more pronounced with nematodes-controlling agent-treated plants. No significant differences in nodule biomass were obtained by the 45- and 75-day old peanut plants, thereafter, differences were more obvious. As to the 110-day old plants, okadin inoculated peanuts and received nemaless produced the superior nodule biomass of 4.53 g plant^{-1} compared to others (Fig. 3). Irrespective of treatment, nemaless-received plants formed nodules with average dry weight of 2.94 g plant^{-1} while those treated with mocap did produce nodules of 2.28 g plant^{-1} .

Inoculation with diazotrophs simultaneously with rational N fertilization resulted in enhancing all plant parameters measured, good development and biomass production of peanut. Such promoting effect extended to nodulation and nitrogen content of plants. Inoculation of peanuts with rhizobia-containing preparations resulted in good biomass and nitrogen yields, a finding in harmony with results of Kang *et al.* (1991) and Carter *et al.* (1994). Barbour *et al.* (1991) mentioned that evidence from *Rhizobium*-legume symbiosis suggests that motility and chemotaxis are involved in the early stages of the interaction by allowing the bacterium to spread through soil, effectively locate infection sites, nodulate the plant more efficiently and more effectively compete for nodule formation. Likewise, motility has been reported to be a factor in competition for nodulation between *Rhizobium* strains. Le-

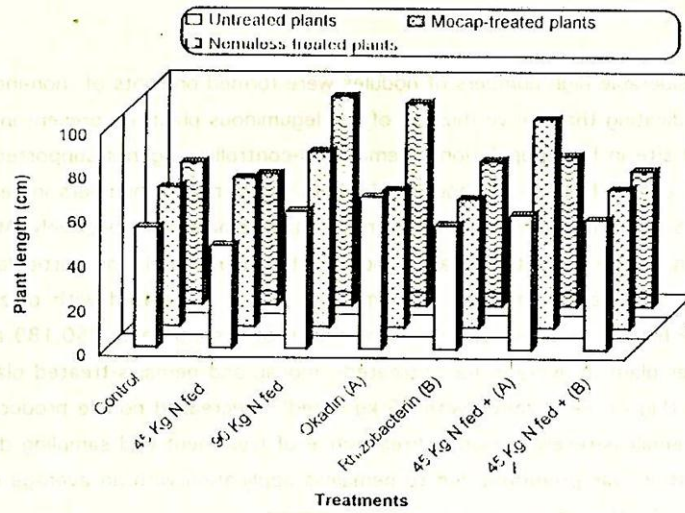


Fig 1. Hight of 110-day old peanut plants as affected by different treatments

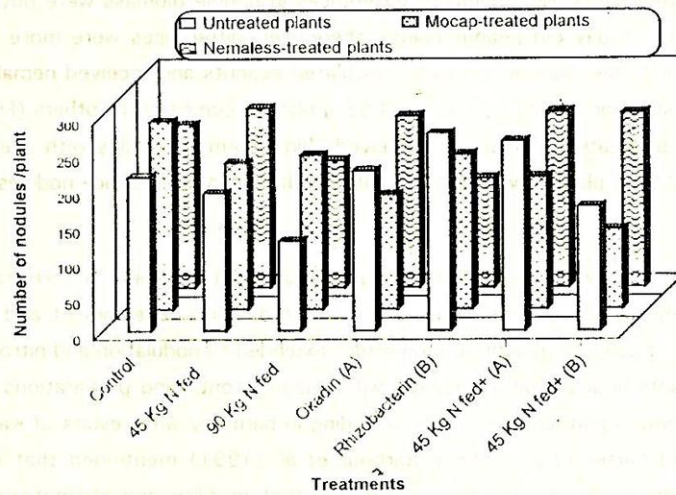


Fig 2. Average numbers of nodules of inoculated and N fertilized peanuts in presence of mocap and nemaless.

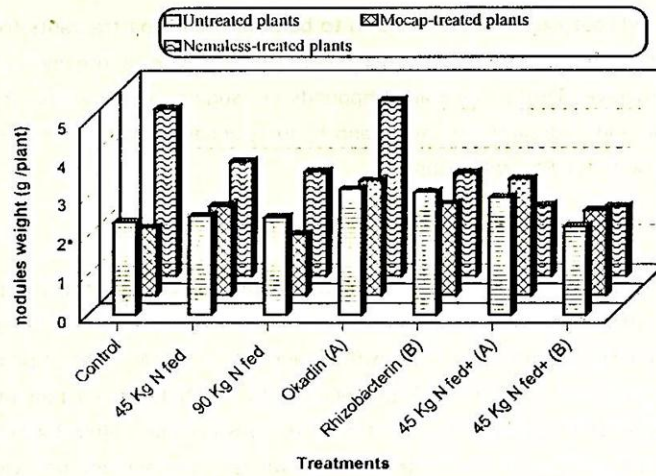


Fig 3. Nodules biomass of 110-day old peanuts as affected by the different treatments.

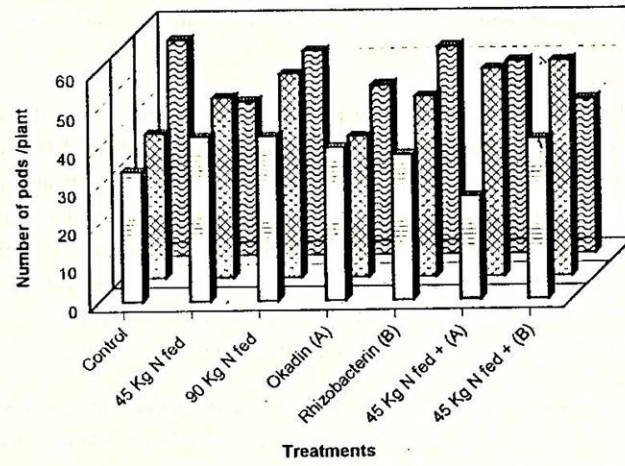


Fig 4. Number of pods produced by peanuts as affected by different treatments.

gume seed and root exudates were shown to be potent chemoattractants for specific rhizobia since exudates are known to contain a wide range of organic compounds (Curl and Truelove, 1986). Several compounds i.e. sugars, amino acids, organic acids, aromatic and hydroaromatic acids and hydroxycinnamic acids have been shown to be attractants for *Rhizobium* spp.

Peanut yield

Pod production by peanut was found to be nematodes controlling agents-dependent rather than inoculation- or fertilization-dependent (Fig. 4). Untreated plants produced 34 - 56 pods per plant with an average of 43. Number of pods slightly increased to the maximum of 50 per plant with full N fertilized peanuts. Other treatments resulted in pod production of 27 - 56 pods per plant. Rhizobacterin seems more favourable for pod production than okadin whereas the average pod yields were 46 for the former and 42 for the latter.

As to total pod yield, Table (3) demonstrates that slight fluctuations, of no significance, were recorded among the experimental treatments. Rhizobacterin in absence of N fertilizer produced the highest pod yield with an average of $1.94 \text{ ton fed}^{-1}$ while the lowest yield of $1.62 \text{ ton fed}^{-1}$ was obtained by either untreated plants or those supplied with 45 kg N fed^{-1} . Nemaless-treated peanuts, generally, produced superior yield approximating 26.8% higher than that resulted by mocap-treated ones.

Plenty of seeds were produced by peanut plants of the different treatments (Table 3). Control plants produced seed yields varied from 0.89 to $1.05 \text{ ton fed}^{-1}$. Fertilization with 45 kg fed^{-1} resulted in yield of 0.76 - 1.25 with an average of $0.94 \text{ ton fed}^{-1}$. Leveling the N fertilization regime to $90 \text{ kg N ton fed}^{-1}$ gave average yield of $1.13 \text{ ton fed}^{-1}$. Inocula applied did increase the yields to 1.29 and 1.0 ton fed^{-1} for rhizobacterin and okadin, respectively, although the differences were statistically insignificant. Treatment with nematodes-controlling agents magnified the seed yields to 11.1 - 45.5% over the untreated plants.

Peanut peels are secondary products which could successfully be used for animal feeding as well as inocula carriers preparation. Table (3) demonstrates that considerable variations in peel weights were observed among the different treatments. Plants inoculated with rhizobacterin and received no nitrogen fertilizer produced peel yield of 653 kg fed^{-1} comparable to that due to full N fertilization regime (average of 660 kg fed^{-1}). Reducing mineral N level did not lower the production. Nemaless-treated plants produced superior peel yield of 18.9% higher than mocap-

treated ones. Peanuts received no nematodes-controlling agents were the inferior in respect with an average yield of 543 kg fed⁻¹

Mineral N fertilization showed no significant influence on weight of 100 seeds of peanut plants (Table 4). On the contrary, diazotrophs inoculation obviously increased the seed weight. Irrespective of nematodes-controlling agents, okadin resulted in 12.2% increase over control while rhizobacterin showed lower promoting effect (increase of 11.3%). Response to inoculation combined with N fertilization was lower, i.e. increases were 5.7 and 4.3%, respectively.

Table 3. Total pod, seed and peel yields of peanut inoculated, N fertilized and received nematodes-restricting agents.

Treatments	None	Mocap	Nemaless
	Pod Yield (ton fed ⁻¹)		
Control	1.40	1.27	2.19
45 kg N fed ⁻¹	1.45	1.51	1.90
90 kg N fed ⁻¹	1.48	1.96	1.94
Okadin	1.34	1.47	2.14
Rhizobacterin	1.57	1.66	2.60
45kg N fed ⁻¹ + Okadin	1.08	2.10	1.76
45kg N fed ⁻¹ + Rhizobacterin	1.79	1.63	1.70
L S D 0.05		0.56	
0.01		0.74	
	Seed Yield (ton Fed ⁻¹)		
Control	0.92	0.89	1.05
45 kg N fed ⁻¹	0.76	0.82	1.25
90 kg N fed ⁻¹	0.90	1.18	1.32
Okadin	0.76	0.95	1.30
Rhizobacterin	1.05	1.01	1.81
45kg N fed ⁻¹ + Okadin	0.70	1.32	1.00
45kg N fed ⁻¹ + Rhizobacterin	1.22	1.00	1.31
L S D 0.05		0.85	
0.01		1.14	
	Peel	Weight (kg fed ⁻¹)	
Control	480	380	440
45 kg N fed ⁻¹	690	690	650
90 kg N fed ⁻¹	580	780	620
Okadin	580	520	840
Rhizobacterin	520	650	790
45kg N fed ⁻¹ + Okadin	380	780	760
45kg N fed ⁻¹ + Rhizobacterin	570	630	470
L S D 0.05		360	
0.01		400	

Nitrogen content of seeds

As shown in Table (4), very low N content was scored in seeds of non-fertilized uninoculated plants (0.26 - 0.46%). Addition of 90 Kg N fed⁻¹ into soil resulted in highly significant increases in seed N of up to 900%, the corresponding increases with the rational N dose were less. In general, N fertilization supported higher seed N accumulation than diazotrophs inoculation. No significant differences were reported between both inocula applied. Again, nematode-treated peanuts produced seeds of the richest N content (average of 2.67%) followed by mocap-received ones (1.9%).

Dry matter yield

Table (4) reveals that the highest dry matter yield (average of 3.69 ton fed⁻¹) was produced by full N fertilized peanuts followed by those inoculated with okadin in combination with 45 kg N fed⁻¹ fertilization regime (average of 3.40 ton fed⁻¹). Reducing the N fertilizer quantity to 45 kg fed⁻¹ decreased the total biomass yield by 40.7% compared to full dose of N. Among the inocula applied, higher yields (2.44 - 4.95 ton fed⁻¹) were obtained by okadin inoculation against 2.06 -2.58 ton fed⁻¹ due to rhizobacterin. The promoting influence of diazotrophs on total yield was more obvious in presence of a rational N fertilization dose.

Fertilizer (kg N fed ⁻¹)	Inoculum	Dry Matter Yield (ton fed ⁻¹)
0	Control	0.26
0	Okadin	0.46
0	Rhizobacterin	0.46
45	Control	2.06
45	Okadin	3.40
45	Rhizobacterin	2.44
90	Control	3.69
90	Okadin	3.69
90	Rhizobacterin	3.69
90	Nematode	2.67
90	Mocap	1.90

Table 4. Weight of 100 seeds (g), grain nitrogen percentage and dry matter yield (ton fed⁻¹) of peanuts as affected by inoculation, N fertilization and nematodes-controlling agents.

Treatments	None	Mocap	Nemaless
	Weight of 100 Seeds		
Control	78.54	71.35	75.67
45 kg N fed ⁻¹	74.93	69.89	78.50
90 kg N fed ⁻¹	81.82	77.90	75.30
Okadin	88.10	89.30	81.40
Rhizobacterin	87.40	82.80	81.50
45kg N fed ⁻¹ + Okadin	83.00	83.85	73.68
45kg N fed ⁻¹ + Rhizobacterin	82.07	76.75	78.50
L. S. D. 0.05	8.78		
0.01	11.68		
	Grain nitrogen percentage		
Control	0.46	0.26	0.45
45 kg N fed ⁻¹	2.60	2.65	3.33
90 kg N fed ⁻¹	3.06	2.58	4.04
Okadin	2.00	2.21	2.55
Rhizobacterin	1.35	2.03	2.40
45kg N fed ⁻¹ + Okadin	1.86	1.85	2.80
45kg N fed ⁻¹ + Rhizobacterin	1.71	1.71	3.15
L. S. D. 0.05	1.21		
0.01	1.64		
	Dry matter yield (ton fed ⁻¹)		
Control	1.86	2.18	2.68
45 kg N fed ⁻¹	1.77	2.22	2.59
90 kg N fed ⁻¹	2.91	3.22	4.93
Okadin	2.77	2.44	4.95
Rhizobacterin	2.06	2.47	2.58
45kg N fed ⁻¹ + Okadin	2.66	4.75	2.79
45kg N fed ⁻¹ + Rhizobacterin	2.72	2.74	2.60
L. S. D. 0.05	1.04		
0.01	1.39		

To optimize crop productivity in the newly reclaimed sandy soils, the complementation of diazotrophic inoculation and adequate supply of mineral fertilizers was the major target of the present study. As the experimental field harboured relatively low densities of diazotrophs (ca. 10^5 cells g^{-1}), inoculation together with reasonable dose of N fertilization resulted in good development and dry matter yield of peanut. Such promoting effect extended to nitrogen content and pods yield of plants. These data are similar to that obtained by other researchers (Carter *et al.*, 1994 and Dashti *et al.*, 1997).

Dehydrogenase activity

Dehydrogenase activity is graphically illustrated in Fig.(5). After 110 days of planting, variation in enzyme activities were recorded depending upon treatment. Untreated soil exhibited dehydrogenase activity of $42.9 \mu g$ TPF g^{-1} indicating that soil of the experimental site harbours dense population of microorganisms. Among the applied treatments, the lowest dehydrogenase activities were scored in N fertilized soils (average of $26.2 \mu g$ TPF g^{-1}). Diazotrophic inoculations increased the activity over N fertilization. Okadin overcame rhizobacterin in providing favourable conditions for soil microbial proliferation and consequently higher dehydrogenase activity.

The data indicate that inoculation with either *Rhizobium* or free-living N_2 -fixing bacteria improve soil bacteria proliferation and in turn dehydrogenase activity (Skujins, 1973), plant growth and yield.

N_2 -Fixation activities

Acetylene reducing activity of soil sharply decreased from the 45- to the 75-day growth period (Fig. 6). At 45 days after planting, the highest activity for nematodes controlling agent-free soil (353.4 n moles C_2H_4 $g^{-1} h^{-1}$) was estimated with rhizobacterin treatment in absence of N fertilizer. Inoculation of N fertilizer-free soil with okadin resulted in nitrogenase activity of 207.6 n moles C_2H_4 $g^{-1} h^{-1}$. The lowest activity (100.7 n moles C_2H_4 $g^{-1} h^{-1}$) was scored for full N fertilized soil. In case of mocap treatment, superior activity of 302.2 n moles C_2H_4 $g^{-1} h^{-1}$ was obtained due to rhizobacterin inoculation simultaneously with 45 kg N fed^{-1} fertilization regime. Non-inoculated soil supplemented with a rational N fertilizer dose was the inferior (119.9 n moles C_2H_4 $g^{-1} h^{-1}$). In respect to nematode-treated soil, acetylene reducing activities of $109.2 - 268.8$ n moles C_2H_4 $g^{-1} h^{-1}$ were recorded being the maximum with okadin inoculation together with 45

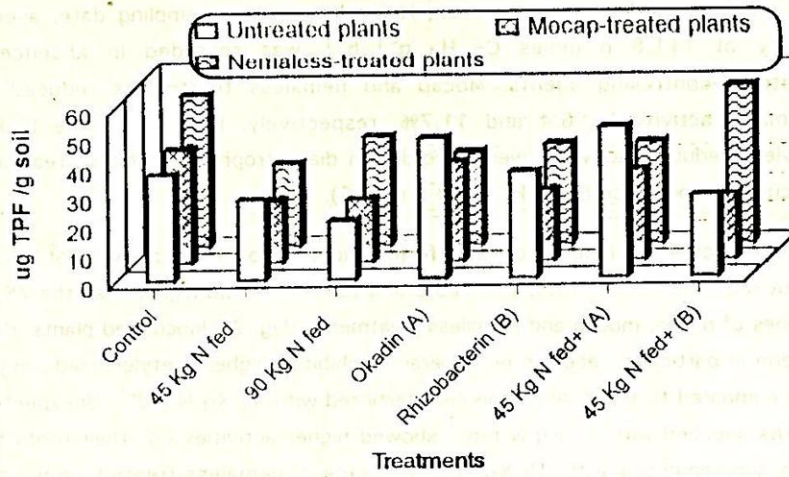


Fig 5. Dehydrogenase activity of soil cultivated with peanuts of the various treatments

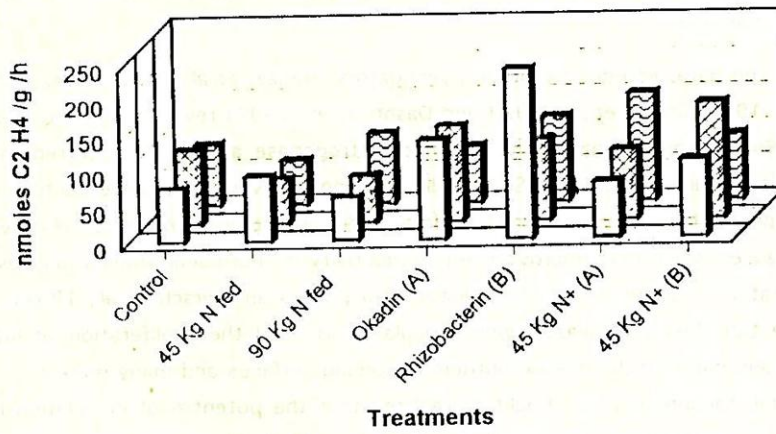


Fig 6. Average acetylene reducing activity of soils of the different treatments at 45- and 75-day growth period.

kg N fed⁻¹. Regardless of inoculation, N fertilization and sampling date, average activity of 111.8 n moles C₂ H₄ g⁻¹ h⁻¹ was recorded in absence of nematodes-controlling agents. Mocap and nemaless treatments reduced the enzymatic activity by 6.4 and 11.7%, respectively. In general, the highest acetylene reducing activities were recorded in diazotrophic inoculated treatments particularly in presence of 45 kg N fed⁻¹ (Fig. 6).

Irrpective of inoculation and fertilization, nitrogenase activity of 45-day peanut roots were 1.0 - 11.5, 2.1 - 29.2 and 2.0 - 10.4 fold higher than the 75-day old ones of nones, mocap and nemaless treatments (Fig. 7). Inoculated plants, rhizobacterin in particular, received no mineral N exhibited higher acetylene reducing activity compared to those simultaneously fertilized with 45 Kg N fed⁻¹. Unexpectedly peanuts supplied with 90 Kg N fed⁻¹ showed higher activities on their roots than those supplemented with 45 Kg N fed⁻¹, except nemaless-treated ones. Non-inoculated peanuts received no N showed the lowest activities of 987.2 µl C₂ H₄ g⁻¹ h⁻¹ in average. Nemaless supported slightly higher enzymatic activity than mocap whereas the mean records were 2091.0 µl C₂H₄ g⁻¹ h⁻¹ for the former and 1866.5 µl for the latter.

The data recorded by other investigators (Hegazi *et al.*, 1979; Fayez and Viassak, 1984; Carter *et al.*, 1994 and Dashti *et al.*, 1997) revealed that inoculation with *Rhizobium* or diazotrophs enhanced nitrogenase activity in root regions of Egyptian legumes or cereals. Such findings support th view that these bacteria produce plant phytohormones mainly indole acetic acid, gibberillin and cytokinine-like substances which may improve plant productivity by hormonal stimulation besides N₂-fixation (Tien *et al.*, 1979; Shahaby *et al.*, 1994 and Hirsch *et al.*, 1997). It is well established that diazotrophic inoculaton induced the proliferation of lateral roots and hairs which increase nutrient-absorbing surfaces and many more root tips available for infection. It should be realized that the potential of inoculation have been overestimated in the literature (Bockman, 1997). There are problems in both the establishment of the selected bacterial strains in the rhizosphere and the formation of an effective association with the roots of the required crop species (Athar, 1998). There is need for further information on the factors influencing diazotrophs-plant interactions before the agricultural potential of the bacteria inoculaton can be realistically evaluated.

Soil nematodes represent a major problem encountered in crop production. Indeed, nematicides applied for nematodes control may exhibit side and deleterious ef-

ffects on beneficial soil biota in the environment. Biological control, a modern concept for pest suppression, aims at using a variety of bio-agents having antibiotic activity against protozoa, fungi and insects. In the present study, a bio-product, nemaless was introduced to peanut plants to restrict root-knot nematodes possibly existing in the field. This obviously reflected on plant growth where treated peanuts were the best in biomass and nitrogen yield. In this respect, studies of other investigators proved that volatile substances produced during the metabolic activity of the bacteria were involved in the inactivation of nematodes (Zavaleta-mejia *et al.*, 1989). It was also reported that NO_3^- or NO_2^- produced during the decomposition of nitrogenous substances were the principal compounds responsible for these population of nematodes. Other alkaline metabolic products have been recognized as having nematotoxic properties (Rodriguez *et al.*, 1994). Among the biological products, those rich in ammonifying bacteria seem to be practical since bacteria are primary consumers of root exudates which are known to contain amino acids and likely to be converted to ammonia along the root providing return nitrogen to plant, as well as a micro provide shield the root. Theoretically, if it is possible to provide the introduced rhizobacteria with a sufficient and continuous supply of organic nitrogen, they would be able to maintain a microzone around the growing root that could be taken into consideration. On the other hand, the increasing environmental, economic and human health problems inherent to pest control practices demand further research on the use of organic amendments to manage the nematode and disease problems of crop production.

For better management of desert environments, it is recommended to employ the cropping systems applied in Nile Valley farms which permit intensive cropping with two crops per year and the application of plant litter to sandy soils. This is expected to result in a potential N_2 -fixation and nutrients pool which, in turn, increase plant productivity. Application of integrated BNF-fertilization systems and selection for potent bacterial symbionts is a must for better manipulation of desert sandy soils.

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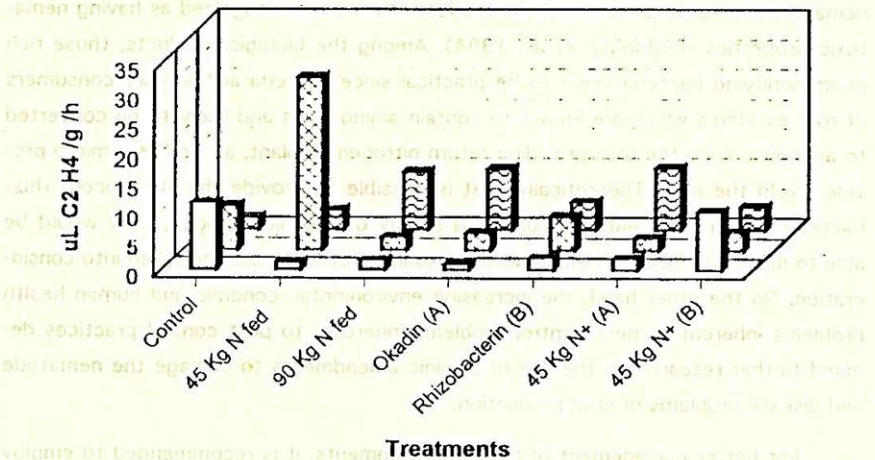


Fig 7. Nitrogenase activity of peanut roots of the different treatments of 45-day old plant. Values represent folds of activity over 75-day old plants.

REFERENCES

1. Athar, M. 1998. Drought tolerance by lentil rhizobia (*Rhizobium leguminosarum*) from arid and semiarid areas of Pakistan. Lett. Appl. Microbiol., 26:38-42
2. Barbour, W.M., D. R. Harmann, and G. Stacey. 1991. Chemotaxis of *Bradyrhizobium japonicum* to soybean exudates. Appl. Environ. Microbiol., 58:2635-2639.
3. Bockman, O.C. 1997. Fertilizers and biological nitrogen fixation as sources of plant nutrients: Perspectives for future agriculture. Plant and Soil, 194:11-14
4. Bremner, J.M. 1965. Inorganic forms of nitrogen. In: Methods of Soil Analysis II, C.A. Black (ed.), PP. 1179-1237. Amer. Soc. Agron. Inc., Madison, Wisconsin, USA.
5. Carter, J.M., W.K. Gardner, and A. H. Gibson. 1994. Improved growth and yield of faba beans (*Vicia faba* cv. Fiord) by inoculation with strains of *Rhizobium leguminosarum* biovar *viceae* in acid soils in south-west Victoria. Aust. J. Agric. Res., 45(3): 613-623.
6. Curl, E.A., and B. Truelove. 1986. Root Exudates in the Rhizosphere. Springer-Verlag, Berlin.
7. Dart, P.J., J.M. Day, and D. Harris. 1972. Assay of nitrogenase by acetylene reduction. Technical Report No. 149 from FAO/IAEA " Use of isotopes for study of fertilizer utilization by legume crops "pp. 85-100.
8. Dashti, N., F. Zhang, R. Hynes, and D.L. Smith. 1997. Application of plant growth promoting rhizobacteria to soybean (*Glycine max* L.Merr.) increases protein and dry matter yield under short-season conditions. Plant and Soil, 188:33-41.
9. El-Sherif, M.A., A.H. Ali, and M.I. Barakat. 1995. Suppressive bacteria associated with plant parasitic nematodes in Egyptian agriculture. Jap. J. Nematol., 24(2): 55-59
10. Fayez, M., 1990. Interactions of some nematicides with *Azospirillum lipoferum* and the growth of *Zea mays*. Z. Pflanzennahr. Bodenk., 153:219-223.
11. Fayez, M., and K. Vlassak. 1984. Effect of inoculation with *Azospirillum brasilense* on barely grown under semi-arid conditions. Zbl. Microbiol., 139:359-365.

12. Hardy, R. W.F., R.C. Burns, and R.D. Holsten. 1973. Application of the acetylene ethylene assay for measurement of nitrogen fixation. *Soil Biochem. Biochem.* 5:47-81.
13. Hegazi, N.A., M. Monib, and K. Vlassak. 1979. Effect of inoculation with N_2 -fixing spirilla and azotobacters on nitrogenase activity on root of maize grown under subtropical conditions. *Appl. Environ. Microbiol.*, 38(4): 621-625.
14. Hirsch, A.M., Y. Fang, S. Asad, and Y. Kapulnik. 1997. The role of phytohormones in plant-microbe symbioses. *Plant and Soil.* 194:171-184.
15. Jackson, M.L. 1973. *Soil Chemical Analysis*. Pertic-Hall of Indian Private limited, New-Delhi.
16. Kang, U.G., Y.T. Jung, H.G. Park, I.S. Kim, G.J. Baek, and M. G. Ban. 1991. Inoculation response of peanut in different soil textural families. *Res. Crop Exp. Stn, RDA, Milyang, Korea.*
17. Piper, C.S. 1950. *Soil and Plant Analysis*, (1st Ed.), p:37-79. Interscience Publishers, Inc. New York, USA.
18. Rodriguez, C.F.A., A.G. Gonzalez, J.R. Lopez, C.D. Ciocco, B.J.C. Pacheco, and J.L. Parada. 1994. *Azospirillum brasilense* and *Bacillus polymyxa* inoculation on yield response of field grown wheat in an Argentine semi arid region. In: *Proceeding of the 6 th Int. Symp. on Nitrogen Fixation with Non-legumes*, (Eds.N.A.Hegazi, M. Fayez, and M. Monib). The American University in Cairo Press, Cairo.
19. Shahaby, A.F. 1988. Associative nitrogen fixation with C_4 grasses of the northern Chihuahuan Desert. Ph.D. Dissertation (Biology) New Mexico State University, Las Cruces, New Mexico, USA
20. Shahaby, A.F., G. Amin, and G.M. Khalafalla. 1994. Response of rice and tomato seedlings to inoculation with diazotrophs and their culture filterates. In : *Proceeding of the 6th Int. Symp. on Nitrogen Fixation with Non-Igumes*, (Eds.N.A. Hegazi, M. Fayez, and M. Monib). The American University in Cairo Press, Cairo, Egypt.
21. Skujins, J.J. 1973. Dehydrogenase: An indicator of biological activities in arid soils. *Bull Ecol. Res. Commun. (Stockholm)*, 17:235-241.

22. Steel, R.G.D., and J.H. Torrie. 1980. Principles and Procedures of Statistics, 2nd., Ed., McGraw-Hill Book Co., New York.

23. Tien, T.M., M.H. Gaskins, and D.H. Hubbell. 1979. Plant growth substances produced by *Azospirillum brasilense* and their effect on the growth of pearl millet (*Pennisetum americanum* L.). Appl. Environ. Microbiol., 37:1016-1024.

24. Yueh, L.Y., and D.L. Hensley. 1993. Pesticide effect on acetylene reduction and nodulation by soybean and lima bean. J. Am. Soc. Hort. Sci., 118 (1): 73-76.

25. Zavaleta-Mejia, M. E., S.V. Gundy, and G.S. Van. 1989. Volatile toxicity of *Serratia marcescens* Bizio and other bacteria on the root-knot nematode *Meloidogyne incognita* (Kofoid and White) Chitwood. Revista Mexicana de Fitopatologia, 7(2):188-194.

استجابة الفول السوداني للتلقيح البكتيري والتسميد النتروجيني في الأراضي الرملية في وجود المواد المانعة لنمو النيما تودا

أحمد فاضل الشهابي^١، أحمد غنيم رحال^٢،

هبة شحاتة شحاتة^٢، محمد فايز^١

١ قسم الميكروبيولوجي - كلية الزراعة - جامعة القاهرة.

٢ قسم بحوث الميكروبيولوجيا الزراعية - معهد بحوث الاراضي والمياه والبيئة - مركز
البحوث الزراعية - الجيزة ١٢٦١٢ - مصر.

أجري هذا البحث بهدف تقييم التلقيح البكتيري للفول السوداني (صنف جيزة ٥) بنوعين من اللقاح هما العقدين (مزرعة مفردة من نوع برادى ريزوبيوم) والريزوباترين (مزرعة مزدوجة مكونة من نوعى البرادى ريزوبيوم والازوتوباكتر كروكوم) فى وجود التسميد النتروجيني والمواد المانعة لنشاط النيما تودا فى تجربة حقلية تمت فى اراضي محافظة الاسماعلية الرملية وفى هذه التجربة تم تقدير الوزن الجاف للنباتات ونشاط تثبيت النتروجين الجوى والمحصول .

أوضحت النتائج ان التلقيح بالعقدين اعطى محصول عالى لوزن النباتات (٢,٤٤ - ٤,٩٥ طن /فدان) بينما اعطى التلقيح بالريزوباترين ٢,٠٦ الى ٢,٨٥ طن /فدان. واعطت نباتات الفول السودانى سواء المعاملة بمبيد النيما تودا موكاب (مبيد مخلق) أو المعامله بالنيمالس (منتج حيوى) اوزان نباتات جافة اعلى من النباتات غير المعاملة . كذلك ادى التلقيح بالعقدين الى زيادة الوزن الجاف للعقد الجذرية مقارنة بالنباتات الملحقه بالريزوباترين سواء سممت بالنتروجين او لم تسمد. وكان هذا التأثير اكثر وضوحا مع النباتات المعاملة بالمواد لنشاط النيما تودا. كذلك ادى استخدام المواد المانعة لنشاط النيما تودا الي تحسين تكوين العقد في حالة النباتات غير الملحقه حيث زاد عدد العقد بمقدار ٢١,٣ و ٥,٦% نتيجة المعاملة بالموكاب والنيمالس علي التوالي. وادت معاملة نباتات الفول السوداني بالنيمالس الي الحصول علي وزن جاف للعقد ١,٤٢ جم/نبات مقابل ١,١٣ جم/نبات للنباتات المعاملة بالموكاب. كذلك ادى التلقيح بالريزوباترين مع عدم التسميد النتروجيني الي الحصول علي اعلى محصول للقرون ١,٩٤ طن/فدان. مقابل ١,٦٥ طن/فدان للنباتات الغير معاملة أو النباتات المسمدة بمقدار ٤٥ كجم ن/فدان. واعطت النباتات المعاملة بالنيمالس محصول قرون اعلى بمقدار ٢٣% عن النباتات المعاملة بالموكاب. أظهر النباتات وخصوصا الملحقه بالريزوباترين والغير مسمدة اعلى نشاط لاختزال الأستلين علي جذورها مقارنة بالنباتات الملحقه والمسمدة بمقدار ٤٥ كجم ن/فدان. ولكن اختلف كثيرا كل من محصول القشر والبروتين الكلي في الفول السوداني حسب المعاملة المستخدمة.