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EFFECT OF FERTLIZATION WITH INORGANIC, ORGANIC AND BIOFERTILIZER ON GROWTH, YIELD AND VOLATILE OIL CONTSTITUENTS OF FENNEL (*Foeniculum vulgare*, MILL)

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ABSTRACT

A field trial was conducted during two consecutive seasons (2001/2002 and 2002/2003) at the Experimental Farm, Faculty of Agriculture, Ain Shams Univ., Shobra El-Kheima, Cairo, Egypt, to investigate the growth, productivity and quality of Fennel (*Foeniculum vulgare*, Mill) as affected by diversity sources of nitrogen fertilization. Ammonium nitrate was applied at the rate of 0, 30, 60 and 90 kg N/fed. as a source of inorganic nitrogen fertilization. Biogas manure used as organic fertilizer with the levels 0, 40 and 60 kg N/fed., and biofertilizer was applied as inoculation with *Bacillus polymyxa*. Irrespective of the concentrations, inorganic fertilizer surpassed all bio-organic fertilizers. The best records of growth parameters, oil percent, estimated yields of fruits and oil and nitrogen parameters were obtained by application 90 kg N/fed. of mineral nitrogen. Soil inoculation with *Bacillus polymyxa* significantly overcame the fruits inoculation with the same inoclum for the aforementioned parameters. Biogas manure application at 60 kg N/fed. produced the highest values of all tested parameters in comparison with other bio-organic treatments and control. The biofertilizers as such or combined with inorganic nitrogen augmented all tested parameters in comparison with uninoculated plants. Therewithal, soil inoculation with *Bacillus polymyxa* and supplemented with inorganic nitrogen 60 kg N/fed. produced the highest values of growth parameters, oil content, yields of fruits and oil in comparison with control, with exception plant fruit yield in the first season wherein 90 kg N/fed. of inorganic N and the same inoculum was the best treatment. On the other side, application of biogas manure at 60 kg N/fed. + 60 kg inorganic N/fed. significantly induced the best records of the most tested parameters. In addition, biogas manure (Bio2) produced the highest values of estimated volatile oil yield, nitrogen and protein contents and nitrogen uptake, when supplemented with 60kg N/fed. of inorganic nitrogen.

In concern to essential oil constituents GC analysis of the fruits oil, indicated that estragole, limonene, fenchone, anethole and γ-terpinene are the main compounds of the oil. It was found that the relative percentage of the oil constituents were fluctuated as a result of interaction among all fertilization sources. In terms of fenchone compound, which is responsible for fennel bitterness, the interaction between aforementioned nitrogen fertilizers reduced its relative percent, this mean more favorable taste for the essential oil if used for food dressing.

Counts of total microbial flora and bacilli influenced by inoculation with *Bacillus polymyxa* giving an additional increase in the rhizosphere of inoculated fennel plants. CO_2 evolution was assayed during the different stages of plant growth, it was found that using of inoculation, inorganic N-fertilizer and biogas manure gave higher rate of CO_2 evoluted from the rhizosphere of inoculated plants than those of uninoculated one and it was also noticed that soil inoculation treatment surpassed to fruit inoculation treatment for the previous tested parameter.

Keywords: Fennel (*Foeniculum vulgare* Mill), mineral nitrogen fertilizer, *Bacillus polymyxa*, biogas manure, essential oil.

INTRODUCTION

The bio-organic farming is a promising new concept in agriculture aims at getting clean, healthy and good quality for exportable agricultural products free from agrochemicals especially for medicinal and aromatic plants. Fennel (*Foeniculum vulgare* Mill) is one of the most important medicinal plants grow well under Egyptian conditions and plays an important role in the foreign exchange of the hard currency. Fennel has several medicinal purposes, i.e. carminative, sedative, stimulative, weak diuretic, laxative, emmenagogic, antispasmodic and flavoring agents (Stuart, 1982 , Chiej, 1984 and Charles *et al.* 1993). As well as the fruits are used in liqueur manufacture and as a condiment (Sturat, 1982). The major constituents of fennel essential oil such as anethole and limonene are also used as essence in cosmetics and perfumes and for some medicinal purposes (Stuart, 1982 and Marotti *et al.* 1993).

The intensive use of manufactured nitrogen fertilizers increased the crops productivity but with low quality which is not accepted for export (Lain *et al.* 1996 and Wang *et al.* 1996). Biofertilizers are the most reliable tools to reduce the rate of chemical fertilizers applied for medicinal and aromatic plants production, as well as decreasing agricultural costs and environmental pollution. Furthermore, *Enterobacter cloacae, B. polymyxa*, and the mixture inoculants appeared to increase the total-N (mg plant^{-1}) contents of plant material during early vegetative growth. Also, a potential grain yield response was observed for the plants inoculated with *B. polymyxa*..(Renato de Freitas, 2000). Rennie & Thomas (1987) observed that of 10 different cultivars of spring wheat inoculated with *B. polymyxa* C-11-25 and *Azospirillum brasilense* ATCC 29729, only one cultivar (Cadet; inoculated with *B. polymyxa* C-11-25) showed consistent plant yield responses due to inoculation. Thus, as suggested by other workers (Pedersen *et al.* 1978; Ruschel & Ruschel 1978; Rennie & Larson 1979; Baldani *et al.* 1983; Rennie *et al.* 1983), plant and/or bacterial genotype might be the controlling factor in an inoculation response.

The present study aims to investigate the effect of combination between inorganic N-fertilizer and either inoculation with *Bacillus polymyxa* or biogas manure on producing high quality yield and safety for human consumption of fennel and its oil content. Also, reducing the extensive use of manufactured nitrogen fertilizer which became a high source of pollution of water and soil to obtain a big yield and good quality.

MATERIALS AND METHODS

Plant materials and soil preparation

Two field Experiments were carried out at the Experimental Farm, Faculty of Agriculture, Ain Shams University, Shobra El-Kheima, Cairo,

Egypt, during the two successive seasons of 2001/2002 and 2002/2003. The experimental area was clay loam soil of the following characteristics: Sand, 24.5 %; silt, 22.9 %; clay, 52.6 %; organic matter, 1.24 %; $CaCO_3$, 1.2 %, total N, 104 (mg/100g); P, 3.2 (mg/100g) and K, 0.9 (mg/100g). In addition to, available Fe, 91.4 (ppm), available Mn, 35.6 (ppm), available Zn, 18.3 (ppm), available Cu 16.4 (ppm), pH 7.5 and EC(dSm^{-1}), 1.2 . The experimental field was divided into 60 plots, each plot was 6.0 m^2, comprising of 3 rows, 3 m long and 60 cm apart. Fruits of fennel (*Foeniculum vulgare* Mill) were obtained from a yield produced in the Experimental Farm, Faculty of Agriculture, Ain Shams University, during the previous season (2000/2001). Fruits were sown in the soil, immediately before irrigation in hills 30 cm apart with 4-5 fruits per hill on 24th October 2001 and 27th October 2002. The seedlings were thinned one month later to one plant/hill. The plants were fertilized with 100 kg superphosphate (15.5% P_2O_5) and 50 kg potassium sulphate (48% K_2O) per fedaan, which applied at the time of thinning. All agricultural management practices were carried out as usually recommended for fennel production in Egypt .

Three different types of nitrogen fertilizations and their combination were applied for fertilizing the plants i. e., mineral nitrogen, organic and biofertilization. Ammonium nitrate (33.5% N) was applied to soil as side dressing with four levels (0, 30, 60 and 90 kg N/fed.) as a source of inorganic nitrogen fertilization. The amount divided into two portions, the first one at the time of thinning and the second 30 days later. Biogas manure was used as a source of organic fertilization, that obtained from Waste Recycling Center, Agricultural Research Center, Ministry of Agriculture. It was applied before sowing during soil preparation in two rates i. e., 40 units of N (2.9 kg/plot) and 60 units (4.351 kg/plot). The applied biogas manure had the following characteristics: organic matter %, 53.5; organic carbon %, 30.3; density, 410 kg/m^3; pH, 7.3; moisture content %, 23.44; EC (dSm^{-1}), 3.00; C/N ratio, 20:1; N %, 1.97; P %, 0.72; K %, 0.45; Fe (ppm), 315; Mn (ppm), 44; Cu (ppm), 6; Zn (ppm), 42.

Inoculants

Biofertilizer was applied as inoculants with Bacillus polymyxa. Two inoculation methods were used i.e., fruits inoculation (B.F.) and soil inoculation (B.S.). One strain of *Bacillus polymyx*a (BF26) previously isolated from rhizospheric soil of fennel was used . The strain was grown on Hino & Wilson's medium (Hino and Wilson,1958). Fruits of fennel(*Foeniculum vulgare*, Mill) were surface sterilized by immersing in 0.01% $HgCl_2$ solution (Rovira,1956), then washed repeatedly with sterile water and kept to dry . the sterilized fruits were soaked in the appropriate bacterial cell suspension(10^8 cells ml^{-1}) for 25 minutes and Arabic gum (16%) was used as an adhesive agent. *Bacillus polymyxa* strain (BF26) was grown in Hino & Wilson's liquid medium for 3 days to prepare the inoculum for the soil inoculation treatments by spraying the liquid culture immediately after sowing seeds in plots chosen for this method of inoculation at the rate of 40 L/fed., then covered with a thin layer of soil .

Experimental design

The experiment was designed as split plots, mineral nitrogen fertilizer as main plots and bio-organic fertilization (biogas manure and *Bacillus polymyxa*) as subplots. Three replicates were conducted to each treatment. Each replicate was comprised of 20 plots randomly distributed representing the various studied treatments.

At the harvest time on the third week of May 2002 and 2003 for the first and second season, respectively: plant height (cm), number of branches and umbels, plant and umbels dry weights (gm), dry fruits yield (gm/plant) and estimated fruits yield (kg/fed.) were recorded. The percentage contents of nitrogen and protein and nitrogen uptake (gm/plant) were recorded in the dry fruits in the second season according to the method described in A.O.A.C.(1970).

Essential oil extraction, measurement and analysis

The represented samples from the fruits of each treatment was subjected to hydro-distillation to record the volatile oil percentage for three hours using Clevenger apparatus according to the method described by Guenther (1961). The volume of the extracted oil was determined and recorded on the basis of oil volume to fruits dry weight (ml/100 g dry fruits) Estimated volatile oil yield (L/fed.) was also calculated. The essential oil constituents were analyzed and determined in the oil samples from the fruits at the second season. Samples were then dehydrated over anhydrous sodium sulfate and stored in silica vials with Teflon-sealed caps at 2°C in the absence of light for gas chromatographic analysis in the Institute of Plant Nutrition and Soil Science, FAL Braunschweig, Germany. The dehydrated oil of each treatment was separately subjected to GLC analysis with (Perkin Elmer Autosampler 2000). The extracted oil has been diluted with n-hexane, injected into a GLC using auto-sampler and the different compounds have been separated on a HP-INNOWAX (60X 0.25X 0.25 µm) capillary column. Helium was used as a carrier gas (flow rate 1.5 ml^{-1}). The temperature program was: 35°C to 230°C (2.5°C per min.) in course of time (92 min.). Injector and flame ionization detector temperatures were 250°C and 300°C, respectively. Area percentages were obtained using a PC program (Maestro Chromatography data system). The identification of these compounds was achieved by matching their retention times with those of authentic samples injected at the same conditions .

Microbiological analyses

Samples of fennel rhizospheric soil were collected after 50, 100 and 150 days and subjected to microbilogical determinations by counting total microbial flora on modified Bunt & Rovira medium (Abd El-Hafez,1966).and *Bacillus polymyxa* number were determined on Hino & Wilson's medium (Hino and Wilson,1958). CO_2 evolution were determined of the rhizospheric soil of fennel plants according to the method described by Pramer and Schmidt (1964) modified by Shehata (1972) .

Statistical analysis :

With the exception of the identification of essential oil constituents which was conducted only in the second season. Statistical analysis was made according to Snedecor and Cochran (1990) using M stat program version 4. Means of various treatments of the two seasons were compared by Duncan's Multiple Range Test (Waller and Duncan, 1969) at 5 % probability level.

RESULTS

The effect of different sources of fertilization on growth parameters of fennel (*Foeniculum vulgare* Mill) were shown in Table 1. Inorganic nitrogen surpassed bio-organic fertilization for growth parameters in both seasons. Except branches number in the second season, all growth parameters significantly augmented by increasing inorganic nitrogen level until 90 kg N/fed. in both seasons. In terms of biofertilizer and biogas significant increase in all growth parameters were obtained in both seasons. With the exception of plant height in the second season, addition of biogas manure at level 60 kg N/fed. (Bio2) significantly overcame other treatments of all tested parameters in both seasons. Soil inoculation with *Bacillus polymyxa* significantly surpassed the fruit inoculation with the same inoculum in both seasons.

Table (1): **The influence of biofertilizer, biogas manure and mineral fertilization on some fennel growth parameters.**

Parameters / Effective factor		Plant height (cm)		Branches (number/plant)		Umbels (number/plant)		Umbels dry weight (gm/plant)	
		1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season
* Min. ferti.	Zero	112.5 d	114.0 d	10.7 c	10.2 d	45.5 c	43.3 c	45.4 c	48.6 c
	30	116.9 c	117.1 c	12.4 b	11.9 c	47.7 b	49.7 b	58.3 b	60.9 b
	60	119.2 b	120.0 b	13.9 a	13.4 a	52.9 a	53.7 a	68.7 a	71.9 a
	90	121.6 a	122.0 a	14.3 a	12.4 b	54.2 a	55.0 a	69.3 a	73.0 a
** Bio-org. ferti.	Zero	107.7 d	107.3 d	9.9 d	10.2 c	43.7 e	46.0 d	53.5 e	55.4 e
	B.F.	118.0 c	119.3 c	12.1 c	11.8 b	49.2 d	48.2 c	58.1 d	58.3 d
	B. S.	119.4 b	120.3 bc	13.3 b	12.3 b	50.5 c	49.2 c	61.4 c	60.4 c
	Bio 1	120.7 ab	121.0 a	13.2 b	12.5 ab	52.0 b	51.8 b	62.9 b	70.2 b
	Bio 2	121.0 a	120.9 ab	13.3 a	13.1 a	53.7 a	54.2 a	66.1 a	72.3 a

Values within each column followed by the same letter are not statistically significant at 5% level.

Bio 1 = biogas manure (40 kg N/fed.) **B.F. = Fruits inoculation with *Bacillus polymyxa***

Bio 2 = biogas manure (60 kg N/fed.) **B.S.= Soil inoculation with *Bacillus polymyxa***

*** Min. ferti.= Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)**

**** Bio-org. ferti. = Bio-organic fertilizers=biofertilizer + biogas manure**

It was evident from the data presented in Table 2 that mineral nitrogen application significantly increased plant dry weight, fruit yield (gm/plant), oil content and estimated dry fruit yield (ton/fed.) in both seasons. With the exception of plant dry weight and fruit yield (gm/plant) in the first season, all previously mentioned parameters significantly increased by increasing inorganic nitrogen level until 90 kg N/fed. in both seasons.

Application of bio-organic fertilization significantly augmented all tested parameters in both seasons. As general trend biogas manure at 60 kg N/fed. overcame all bio-organic treatments in both season. Soil inoculation with *Bacillus polymyxa* surpassed fruits inoculation with the same inoculum.

Table (2): The influence of biofertilizer, biogas manure and mineral fertilization on plant dry weight, fruit yield, oil content and estimated dry fruit yield .

Parameters › Effective factor ˇ		Plant dry weight (gm)		Fruit yield (gm/ plant)		Oil content (mL/100g fruits)		Estimated dry fruit yield (ton/ fed.)	
		1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season
* Min. ferti.	Zero	189 7 c	195.1 d	40.4 c	40.8 d	1.719 d	1.724 d	0.904 d	0.896 d
	30	335.7 b	337 9 c	52.8 b	54.2 c	1.926 c	1.865 c	1.181 c	1.172 c
	60	415.7 a	395.1 b	62.1 a	54.3 b	2.059 b	1.996 b	1.354 b	1 387 b
	90	407 4 a	416 4 a	64.4 a	66.4 a	2 125 a	2.105 a	1.433 a	1 433 a
** Bio-org. ferti.	Zero	259.6 e	246.6 e	45.1 c	47 6 d	1.763 e	1.736 e	1 006 e	1 004 e
	B. F.	323.7 d	326.8 d	53 6 b	55 5 c	1.959 d	1.932 d	1.166 d	1.180 d
	B. S.	331 4 c	344 7 c	55 8 b	59.9 ab	2.007 b	1.977 b	1.249 c	1.261 c
	Bio 1	372.4 b	364 7 b	57 7 ab	58 7 b	1.990 c	1.954 c	1.279 b	1 281 b
	Bio 2	398.5 a	397.9 a	62 5 a	60 8 a	2 067 a	2.013 a	1.391 a	1.384 a

Values within each column followed by the same letter are not statistically significant at 5% level.

Bio 1 = biogas manure (40 kg N/fed.) B.F. = Fruits inoculation with *Bacillus polymyxa*

Bio 2 = biogas manure (60 kg N/fed.) B.S.= Soil inoculation with *Bacillus polymyxa*

* Min. ferti.= Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)

** Bio-org. ferti. = Bio-organic fertilizers=biofertilizer + biogas manure

The data presented in Table 3 showed that illustrate a general trend for estimated volatile oil yield, contents of nitrogen and protein and nitrogen uptake which significantly augmented with chemical nitrogen application. Except nitrogen uptake all aforementioned parameters significantly increased by increasing inorganic nitrogen level until 90 kg N/fed. Concerning of bio-organic fertilization, it was found that biogas manure with level 60 kg N/fed. surpassed other treatments. As for biofertilizer the same trend was obtained, wherein soil inoculation with *Bacillus polymyxa* overcame fruit inoculation with the same biofertilizer.

As for the impact of divers sources of fertilization and their effect on fennel growth parameters was shown in Table 4. The results within hand speak well about the combination between different doses of inorganic fertilization with *Bacillus polymyxa* or biogas manure. All aforementioned treatments significantly augmented fennel growth parameters in comparison with control in both seasons. Concern of biofertilizer, it was found that soil inoculation with *Bacillus polymyxa* with or without inorganic nitrogen overcame the fruits inoculation with the same inoculum in both season. On the other side, it was found that plants inoculated with *Bacillus polymyxa* and fertilized with 60 kg N/fed. produced the highest values of growth parameters compared with control in both seasons. Furthermore, the boifertilized plants that received mineral nitrogen (90 kg N/fed.)came at the second order.

Table (3): Effect of different fertilization sources on estimated oil yield, nitrogen content , protein content and nitrogen uptake

Parameters› / Effective factor˜		Estimated volatile oil yield (L/fed.)		Nitrogen. Content (%)	Protein content (%)	Nitrogen uptake (gm/plant)
		1 st season	2 nd season	2 nd season	2 nd season	2 nd season
Mineral* nitrogen fertilizer	Zero	15.7 d	15.6 d	4.58 d	28.56 d	1.88 d
	30	22.9 c	21.9 c	4.65 c	29.03 c	2.55 c
	60	28.0 b	27.5 b	4.70 b	29.34 b	3.42 a
	90	30.5 a	30.2 a	4.77 a	29.79 a	3.17 b
Bio-organic fertilizers	Zero	18.1 e	17.8 d	4.54 e	28.33 e	2.68 d
	B. F.	23.1 d	22.8 c	4.65 d	29.05 d	2.58 e
	B. S.	25.4 c	25.2 b	4.71 b	29.43 b	2.81 b
	Bio 1	25.7 b	25.3 b	4.69 c	29.32 c	2.76 c
	Bio 2	29.0 a	28.0 a	4.76 a	29.76 a	2.94 a

Values within each column followed by the same letter are not statistically significant at 5% level.

*** = Ammonium nitrate was added as mineral fertilization at the rates ((0, 30, 60 and 90 kg N/fed.)**

Bio1 = biogas manure (40 kg N/fed.) **B.F. = Fruits inoculation with *Bacillus polymyxa***

Bio2 = biogas manure (60 kg N/fed.) **B.S. = Soil inoculation with *Bacillus polymyxa***

Amongst biogas manure the data illustrated in Table 4 revealed that biogas manure significantly surpassed other treatments in both seasons. Except plant height and umbels number in both seasons, application biogas manure with level 60 kg N/fed. + mineral nitrogen (60 kg/fed.) produced the highest values of the tested parameters. Meanwhile the increasing values of plant height and umbels No. were not significant by increasing inorganic nitrogen level to 90 kg N/fed. + biogas manure 60 kg N/fed., with exception plant height in the first season.

The influence of divers fertilization sources and their effect on plant dry weight, fruit yield (gm/plant), oil content and estimated fruit yield (ton/fed.) were presented in Table 5. In comparison with control the interaction between the divers sources of fertilization produced highly significant effect on aforementioned parameters in both seasons. Increasing inorganic nitrogen level until 90 kg N/fed. significantly increased all tested parameters in both seasons. Once again amongst the combination between biofertilizer and mineral nitrogen, it was found that soil inoculation with *Bacillus polymyxa* and received 60 kg N/fed. produced the highest values of previously mentioned parameters in both seasons. With exception fruit yield (gm/plant) in the first season, wherein the comparison was in the favor the soil inoculation with *Bacillus polymyxa* + 90 kg N/fed. of inorganic nitrogen. In concern to interaction between inorganic nitrogen and biogas manure it was found that the highest values of all the tested parameters were obtained by applied biogas manure compared with other treatments in both seasons. The results presented in Table 5 revealed that applied biogas manure with level 60 kg N/fed. produced highly significant effect on all aforementioned parameters in comparison with untreated plants in both seasons.

Table (4): The associative combined influence of biofertilizer, biogas manure and mineral fertilization on some fennel growth parameters.

Parameters / treatment		Plant height (cm)		Branches (number/plant)		Umbels (number/plant)		Umbels dry weight (gm/plant)	
		1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season
* 0	Zero	98.7 i	97.0 h	7.1 l	7.5 h	37.0 m	38.0 g	37.9 m	37.7 n
	B.F.	112.0 g	113.7 f	10.4 j	10.7 fg	45.3 k	41.0 f	40.3 m	41.0 m
	B.S	115.3 f	117.3 e	11.7 hij	11.0 e	47.0 ijk	43.6 f	46.0 l	43.3 l
	Bio1	117.7 def	120.7 bcd	11.3 ij	10.7 fg	47.7 ijk	43.3 f	49.1 k	59.0 i
	Bio2	119.0 cde	121.3 bc	12.8defg	10.9 e	50.3 efg	52.3 d	53.7 ij	62.0 n
30	Zero	105.7 h	106.3 g	8.3 k	9.7 g	40.7 l	43.7 f	51.6 jk	53.4 k
	B.F.	117.3 ef	118.0 de	12.0 fghi	12.0 cdef	48.0 hij	49.0 e	56.0 l	55.0 jk
	B.S	121.0 bc	119.3 cde	13.3 cdef	12.8 bcd	49.3fghi	51.0 de	60.4 h	58.3 j
	Bio1	120.0 cde	119.7 cde	13.7 bcde	12.3 cde	49.0 ghi	51.1 de	59.9 h	69.0 efg
	Bio2	120.3 cd	122.0 abc	14.3 abc	13.0 bcd	51.0 efg	53.7 cd	63.3 g	71.0 de
60	Zero	110.7 g	109.7 g	11.8 ghij	12.0 cdef	46.3 jk	49.0 e	60.1 h	62.0 h
	B.F.	121.3 bc	123.0 ab	13.3 cdef	12.7 cd	52.0 de	52.3 d	69.7 cd	70.0 ef
	B.S	121.3 bc	124.7 a	14.4 abc	13.0 bcd	54.0 cd	53.6 cd	72.3 bc	79.3 j
	Bio1	121.3 bc	121.7 abc	14.7 ab	14.3 ab	55.0 bc	56.0 bc	69.1 d	75.1 i
	Bio2	121.7 bc	122.0 abc	15.3 a	15.0 a	57.0 ab	58.0 ab	75.3 a	82.7 a
90	Zero	115.7 f	117.3 e	12.3 fghi	11.7 d	50.7 efg	53.3 cd	64.3 fg	68.3 fg
	B.F.	120.3 cd	120.0 cd	12.6 efgh	11.8 d	51.0 efg	50.3 de	66.4 ef	67.1 g
	B.S	121.3 bc	121.3 bc	14.0 bcd	12.3 cde	52.0 de	51.6 de	68.3 cd	69.7 efg
	Bio1	123.7 ab	122.0 abc	13.0 defg	12.7 cd	56.3 abc	56.7 ab	73.4 ab	77.3 c
	Bio2	125.3 a	123.0ab	14.7 ab	13.3 bc	58.3 a	59.3 a	73.0 ab	79.7 b

Values within each column followed by the same letter are not statistically significant at 5% level.

* = Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)

Bio 1 = biogas manure (40 kg N/fed.) B.F. = Fruits inoculation with *Bacillus polymyxa*

Bio 2 = biogas manure (60 kg N/fed.) B.S.= Soil inoculation with *Bacillus polymyxa*

Table 6 indicates the impact of different fertilization sources on estimated oil yield (L/fed.), contents of nitrogen and protein and nitrogen uptake. With respect to estimated oil yield, significant increase over control were induced owing to the application of various fertilization sources in both seasons. Application of *Bacillus polymyxa* (either fruits or soil inoculation) + 60 kg N/fed. of inorganic nitrogen induced the highest values compared to untreated plants in both seasons. Furthermore, the biofertlized plants that received mineral nitrogen 90 kg N/fed. came at the second order for estimated oil yield. Meanwhile biogas manure 60 kg N/fed. (Bio2) + Inorganic nitrogen 90 kg N/fed. surpassed all other treatments for all the tested parameters. With respect of the contents of nitrogen and protein and nitrogen uptake significantly increase was obtained by increasing inorganic nitrogen level with or without *Bacillus polymyxa* inoculation or biogas manure. The highest values were obtained by application biogas manure (60 kg N/fed.) + mineral nitrogen (90 kg N/fed.).

Table (5): The interaction among inorganic fertilization, biofertilizer and biogas manure on plant dry weight, fruit yield, oil content and estimated fruit yield of fennel.

Parameters › Treatments		Plant dry weight (gm)		Dry fruit yield (gm/ plant)		Oil content (mL/100g dry fruits)		Estimated dry fruit yield(ton/ fed.)	
		1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season
0	Zero	107.3 j	108.3 n	33.0 j	33.7 k	1.401 o	1.410 j	0.728 o	0.733 p
	B.F.	193.7 i	196.0 m	37.0 ij	39.0 j	1.7757 n	1.784 h	0.826 n	0.833 o
	B.S.	205.3 hi	207.0 lm	40.2 hij	41.7 ij	1.800 m	1.790 h	0.887 m	0.893 n
	Bio1	215.3 gh	216.3 kl	43.0 ghi	43.3 hi	1.788 mn	1.794 gh	0.973 k	0.953 m
	Bio2	227.0 g	248.0 j	48.0 fgh	46.3 h	1.833 l	1.840 f	1.108 j	1.067 i
30	Zero	228.3 g	228.7 k	42.4 ghij	45.0 hi	1.775 n	1.703 i	0.950 l	0.942 m
	B.F.	326.3 f	232.7 h	50.7 efg	53.7 fg	1.890 jk	1.830 fg	1.138 i	1.127 k
	B.S.	344.7 e	356.0 g	56.4 cde	59.0 e	1.967 i	1.926 e	1.240 g	1.253 i
	Bio1	362.3 d	371.3 fg	54.4 def	56.0 efg	1.910 j	1.920 e	1.218 h	1.209 j
	Bio2	416.7 c	400.7 de	59.9 bcde	57.3 ef	2.086 fg	1.944 e	1.360 e	1.331 h
60	Zero	327.5 f	294.3 i	51.6 efg	52.7 g	1.875 k	1.830 fg	1.138 i	1.146 k
	B.F.	411.0 c	395.7 de	60.5 bcde	66.3 bcd	2.096 efg	2.119 b	1.408 d	1.390 f
	B.S.	415.0 c	422.0 c	63.3 abcd	70.0 ab	2.150 bc	2.125 b	1.428 c	1.469 d
	Bio1	455.0 b	410.0 cd	65.1 abc	65.0 cd	2.100 ef	2.000 d	1.428 d	1.446 e
	Bio2	476.0 a	489.3 a	71.3 a	72.3 a	2.217 a	2.180 a	1.563 a	1.584 a
90	Zero	375.3 d	355.0 g	53.5 def	59.0 e	2.001 h	2.000 d	1.208 h	1.196 j
	B.F.	363. d	383.0 ef	63.0 abcd	63.0 d	2.075 g	1.996 d	1.290 f	1.360 g
	B.S.	369.7 d	393.7 de	66.1 ab	67.3 bc	2.113 de	2.066 c	1.380 e	1.430 e
	Bio1	457.0 b	461.0 b	68.3 ab	70.3 ab	2.160 b	2.100 bc	1.496 c	1.518 c
	Bio2	473.0 a	453.7 b	70.0 a	67.3 bc	2.133 cd	2.086 bc	1.534 b	1.534 b

Values within each column followed by the same letter are not statistically significant at 5% level.

* = Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)

Bio 1 = biogas manure (40 kg N/fed.) B.F. = Fruits inoculation with *Bacillus polymyxa*

Bio 2 = biogas manure (60 kg N/fed.) B.S. = Soil inoculation with *Bacillus polymyxa*

The results within hand dealing with essential oil constituents compiled in Table 7 showed the relative percentage of the oil compounds of different treatments during the second season. The illustrated data indicated that 9 compounds were identified in the fennel fruits oil. In general, it was noticed that the relative percentage of the oil constituents were fluctuated as a result of interaction between all fertilization sources. The most important compounds were estragole and limonene, which accounted for 47.3 – 65.3 and 10.8 – 24.7 % respectively. The third main compound was fenchone, which is responsible for fennel bitterness it ranged from 3.21 to 9.9 %. The obtained data indicated that the relative percentage of fenchone decreased with interaction between inorganic fertilization and biofertilization or biogas manure compared with inorganic fertilizer alone. Meanwhile the relative percent of anethole (the fourth compound) ranged from 0.40 to 5.61 % followed by γ-terpinene, phellandrene, anis aldehyde, α-pinene and β-pinene. Their mean relative percentages were 3.35, 2.90, 2.51, 1.23, 0.65 and 0.55 % for other aforementioned compounds, consecutively. The total mean of identified compounds were 90.31 %, while unidentified ones reached 9.69 %.

Table (6): The combined impact of different fertilizers sources estimated volatile oil yield nitrogen content, protein content and nitrogen uptake of fennel .

Parameters› / Treatments *		Estimated volatile oil yield /fed.)		Nitrogen content (%)	Protein content (%)	Nitrogen uptake (gm/plant)
		1 st season	2 nd season	2 nd season	2 nd season	2 nd season
0	Zero	10.2 p	10.3 k	4.45 h	27.63 n	1.53 r
	B.F.	14.7 o	14.9 j	4.56 k	28.52 l	1.77 q
	B.S.	16.0 n	16.0 i	4.61 ij	28.81 i	1.92 p
	Bio1	17.4 l	17.2 h	4.58 jk	28.62 kl	1.98 o
	Bio2	20.3 k	19.6 g	4.68 gh	29.23 h	2.16 m
30	Zero	16.8 m	16.0 i	4.52 l	28.26 m	2.03 n
	B.F.	21.5 j	20.6 f	4.61 l	28.84 i	2.48 l
	B.S.	24.4 h	24.1 e	4.69 fgh	29.30 gh	2.77 l
	Bio1	23.3 l	23.2 e	4.68 gh	29.26 h	2.62 k
	Bio2	28.4 f	25.9 d	4.72 def	29.50 ef	2.87 h
60	Zero	21.3 j	21.0 f	4.59 ijk	28.67 jk	2.42 l
	B.F.	29.5 de	29.7 c	4.67 h	29.17 h	3.09 e
	B.S.	32.0 c	31.2 b	4.74 cde	29.63 de	3.32 c
	Bio1	30.0 d	28.9 c	4.71 efg	29.41 fg	3.06 f
	Bio2	32.7 b	32.1 b	4.77 bc	29.82 c	3.21 d
90	Zero	24.2 h	23.9 e	4.60 ij	28.76 ij	2.72 j
	B.F.	26.8 g	26.2 d	4.75 cd	29.69 d	2.99 g
	B.S.	29.2 e	29.5 c	4.80 b	30.00 b	3.23 d
	Bio1	32.3 bc	31.9 b	4.79 b	29.96 b	3.37 b
	Bio2	34.7 a	34.5 a	4.88 a	30.50 a	3.53 a

Values within each column followed by the same letter are not statistically significant at 5% level.

* = Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)

Bio 1 = biogas manure (40 kg N/fed.) B.F. = Fruits inoculation with *Bacillus polymyxa*

Bio 2 = biogas manure (60 kg N/fed.) B.S. = Soil inoculation with *Bacillus polymyxa*

Data recorded in Fig 1 clearly indicate that inoculation with *Bacillus polymyxa* increased the count of total microbial flora in the rhizospheric soil of fennel. Fruits inoculation treatment had a lower number of total microbial flora than soil inoculation treatment. The maximal count of total microbial flora in the fennel rhizosphere was found in the treatment of biogas manure at the rate of 60 kg N/ fed in the presence of the full dose of inorganic N-fertilizer (90 kg N/ fed) after 100 days of cultivation in the second season .

Data in Fig 2 show that the number of *Bacillus polymyxa* markedly increased in soil inoculated by spraying the diazotroph bacteria than seeds inoculated by immersing in such bacteria .Application of the full dose of inorganic N-fertilizer at the rate of 90 kg N/ fed and biogas manure at the full level (60 kg N/ fed) gave an additional increase of *B. polymyxa* number in fennel rhizosphere in the second season . Uninoculated treatments gave the lowest densities of *B.polymyxa* in the rhizosphere of growing plants in the absence or presence of inorganic N-fertilizer .

Table(7):The different constituents of the volatile oil of fennel as affected by different fertilizers sources.

Nitrogen fertilization	Bio-organic fertilization	α-pinene	β-pinene	Phellandrene	Limonene	γ-terpinene	Fenchone	Esragole	Anethole	Anis aldehyde	Total	Unidentified compounnds
* 0	Zero	0.75	0.62	0.96	20.90	3.10	9.31	58.4	4.08	-	98.12	1.88
	B.F.	0.38	0.33	4.34	18.60	3.64	6.33	62.7	0.40	-	95.72	4.28
	B.S.	0.30	0.29	2.37	13.96	2.95	4.76	50.4	0.64	4.73	80.40	19.6
	Bio1	0.90	0.76	0.69	19.20	0.05	4.81	53.6	5.06	-	85.10	14.9
	Bio2	0.78	0.83	3.31	20.40	3.41	6.94	51.0	4.36	2.63	93.66	6.34
30	Zero	0.81	0.58	1.20	21.90	0.61	5.88	61.0	5.10	0.08	97.16	2.84
	B.F.	0.39	0.33	3.85	18.90	6.07	5.40	53.1	5.61	0.05	93.70	6.30
	B.S.	0.63	0.43	-	10.80	-	4.86	51.3	2.21	-	70.20	29.8
	Bio1	0.78	0.73	2.19	11.40	2.18	4.95	50.1	4.30	1.11	77.74	22.3
	Bio2	0.99	0.65	3.27	14.80	3.56	3.21	47.3	3.20	0.76	77.74	22.3
60	Zero	0.81	0.76	2.28	18.30	-	9.90	56.3	3.96	0.83	95.14	4.86
	B.F.	0.27	0.23	-	24.70	-	7.93	59.8	0.73	-	93.66	6.34
	B.S.	0.38	0.32	4.45	16.60	4.55	4.95	62.0	2.02	-	95.27	4.73
	Bio1	0.84	0.75	1.80	17.50	3.10	6.41	65.3	2.10	-	97.80	2.20
	Bio2	0.87	0.69	1.10	20.20	3.84	4.10	63.2	4.83	-	98.83	1.17
90	Zero	0.77	0.67	2.30	16.30	-	8.18	56.4	5.31	0.51	90.44	9.56
	B.F.	0.39	0.33	3.71	14.90	2.86	8.35	59.2	4.96	-	94.70	5.30
	B.S.	0.38	0.33	4.25	22.90	2.61	3.48	60.3	2.96	-	97.21	2.79
	Bio1	0.78	0.66	-	22.90	1.78	6.10	63.3	0.96	-	96.48	3.52
	Bio2	0.80	0.61	0.63	11.40	2.10	5.81	51.2	4.21	0.4	77.16	22.8
Mean		0.65	0.55	2.51	17.83	2.90	6.08	56.8	3.35	1.23	90.31	9.69

* = Ammonium nitrate was added as mineral fertilization at the rates (0, 30, 60 and 90 kg N/fed.)

Bio 1 = biogas manure (40 kg N/fed.) B.F. = Fruits inoculation with *Bacillus polymyxa*

Bio 2 = biogas manure (60 kg N/fed.) B.S. = Soil inoculation with *Bacillus polymyxa*

It was also found that proliferation of total microbial flora and *B. polymyxa* number in the rhizosphere of fennel increased by growth of plants reaching its maximal levels after 100 days of cultivation and decreased thereafter in the second season (Fig 1& 2). CO_2 evolution from rhizosphere of fennel were periodically determined to evaluate microbial activities in the root zone as affected by application of bacterial inoculation in the presence or absence of mineral N-fertilizer and biogas manure . The obtained Data recorded in Fig 3 obviously show that CO_2 evolution positively increased in the rhizosphere of inoculated plants compared with uninoculated ones. Soil inoculation treatment gave a higher rate of CO_2 evolution than fruit inoculation treatment . The lowest levels of CO_2 evolution was found of the rhizosphere soil of uninoculated fennel plants (control) after 50 days of planting . Application cf biogas manure at the rate of 60 kg N/ fed. and the full dose of inorganic N-fertilizer (90 kg N/ fed) gave the highest CO_2 rates of fennel rhizosphere after 100 days of cultivation in the second season.

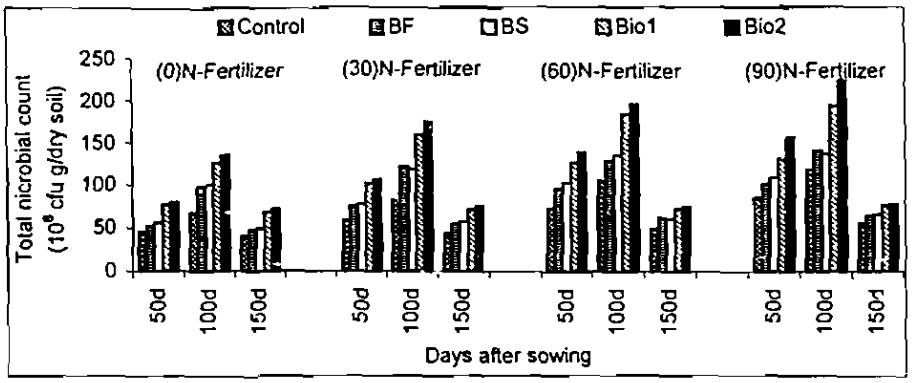

Fig.(1) Effect of inoculation with *Bacillus polymyxa*, mineral N-fertilizer and biogas manure on total microbial count of fennel rhizosphere in the second season .

Bio1 = biogas manure (40 kg N/fed.) B F = Fruits inoculation with *Bacillus polymyxa.*
Bio2 = biogas manure (60 kg N/fed.) B S = *Soil inoculation with Bacillus polymy*

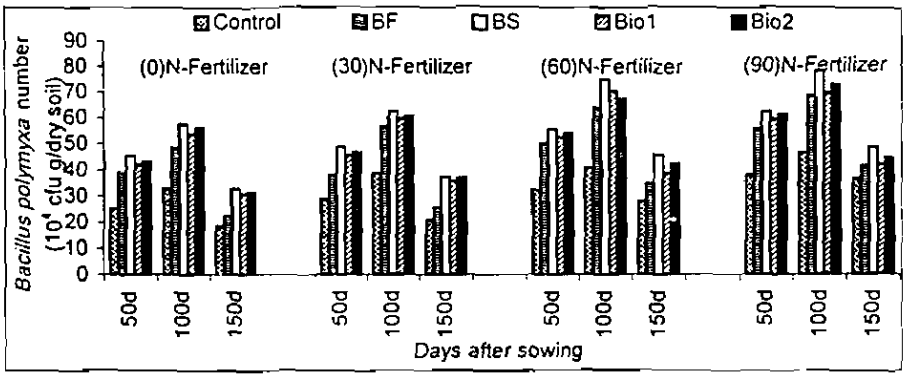

Fig.(2) Effect of inoculation with *Bacillus polymyxa*, mineral N-fertilizer and biogas manure on *Bacillus polymyxa* count of fennel rhizosphere in the second season

Bio1 = biogas manure (40 kg N/fed.) B F = Fruits inoculation with *Bacillus polymyxa.*
Bio2 = biogas manure (60 kg N/fed.) B S = Soil inoculation with *Bacillus polymyxa*

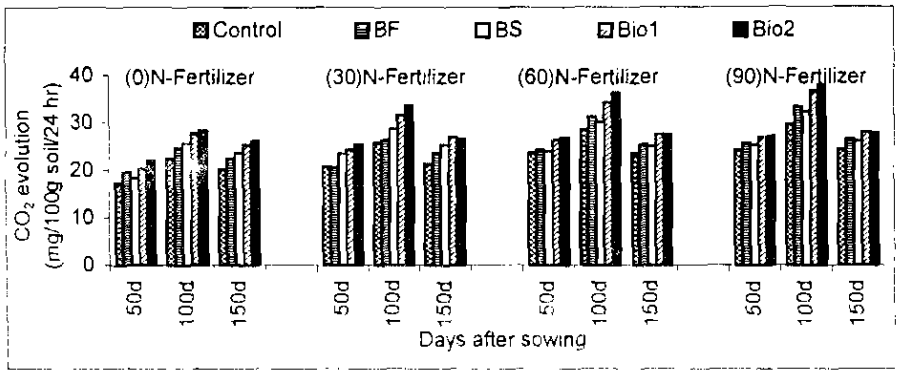

Fig.(3) Effect of inoculation with *Bacillus polymyxa*, mineral N-fertilizer and biogas manure on CO_2 evolution of fennel rhizosphere in the second season.

Bio1 = biogas manure (40 kg N/fed.) B F = Fruits inoculation with *Bacillus polymyxa*.
Bio2 = biogas manure (60 kg N/fed.) B S = Soil inoculation with *Bacillus polymyxa*

DISCUSSION

The application of bio organic farming technique aims to reduce the extensive use of synthetic fertilizers to get clean healthy, safe for human consumption and exportable agricultural product free from agrochemicals (Lampkin 1990). The results within hand obviously show the impact of inorganic fertilization and bio-organic fertilizer (Tables 1, 2 and 3) It was evident that inorganic ferdilization overcame the bio-organic fertilization in both seasons, with exception branches number in the second season. Mineral fertilization at 90 kg N/fed. produced the highest values of plant height, number of branches and umbels, dry weights of umbels and plant, fruit yield (gm/plant), oil content, estimated yields of fruit and oil in both seasons. The mean values of increasing percentages in the two season over control were 7.5, 27.6, 23.0, 51.5, 114.1 61.1, 22.8, 59.2 and 93.6 for all aforementioned parameters consecutively. In addition nitrogen and protein contents and nitrogen uptake significantly increased by increasing inorganic nitrogen levels until 90 Kg N/fed. They augmented over control by about 4.15, 4.3 and 68.6 % for all nitrogen tested parameters previously mentioned respectively. These results were in agreement with Sharma and Prasad, 1990; Hussien and Abou El-Maged, 1991 and Wagner, 1993 where they found that chemical nitrogen strongly affected on plant growth, fruit yield and essential oil of fennel.

As for biofetilizer the data presented in Tables (1, 2 and 3) revealed that all growth parameters and yield significantly increased by inoculation biofertilizer in both seasons. Soil inoculation with *Bacillus polymyxa* surpassed fruits inoculation. This could be attributed to the density of liquid culture applied to the soil in comparison with the low quantity of the same

inoculum applied to the fruits. Inoculation of *Bacillus polymyxa* in the soil leads to significant increase of plant height, Number of branches and umbels, dry weights of umbels and plant, fruit yield, oil content estimated yields of fruits and oil by about 11.49, 26.73, 11.15, 11.85, 33.56, 24.81, 13.83, 24.88 and 40.95 % over control, consecutively. These increases could be attributed to the growth hormones produced by this microorganism in its culture medium during its growth i.e. auxins, gibberellins and cytokinins (Azcon *et. al.* 1978). Also these results holds true with those obtained by Saleh et. al. (1998), they found that growth of *Datura* and its content of alkaloids were greatly improved by biofertilization. Furthermore Faid, (1994) found that biofertilization with nitrogen fixers gave an appreciable improvement of the growth and yield of different plants. Amongst nitrogen parameters boifertilization significantly augmented nitrogen and protein contents and nitrogen uptake. Furthermore these results were in agreement with those obtained by Renato De Freitas (2000), who suggested that rhizobacteria inoculants may be capable to enhance uptake of nutrients. However, these beneficial effects on plant growth may not be directly related to additional N_2 gains. In fact, it has been reported that *Azospirillum brasilense, Azotobacter* and *Bacillus* spp. are able to affect crop growth through mechanisms such as provision of plant growth regulators (Tien *et al.*,1979). Plant growth hormones may be supplied by *Azospirillum* and *Bacillus polymyxa* bacteria and/or roots as a reaction to bacterial infection at the root interface (Avivi & Feldman, 1982 and Kapulnik *et al.*,1985). In many cases, the shoot and root growth-altering effects induced by these rhizobacteria were comparable to those produced by additions of indole-3- acetic acid and gibberellic acid (Kapulnik *et al.*, 1985 and Kucey, 1988). Furthermore El Khayat and Zaghloul (1999) and khattab and Gomaa (2003) they found that nitrogen uptake increased by biofertilization application to caraway and genovese basil respectively.

Once again biogas manure significantly increased all growth parameters, yields of fruit and oil and nitrogen parameters in comparison with control in both seasons (Tables 1, 2 and 3). Biogas manure surpassed biofertilization treatments, this may be due to that biogas manure give similar amount of plant nutrient to the soil, or improve soil quality. This observation was in agreement with those obtained by Pareek *et. al.* (1984) who found that farmyard manure improved soil texture as well as increased oil yield of palmarosa grass by 50.6 % over control. Plant height, branches No., umbels No., dry weights of umbels and plant, fruit yield, oil content, estimated yields of fruits and oil and nitrogen uptake overcame control with application of 60 kg n/fed. by about 12.51, 30.69, 20.29, 27.09, 57.33, 32.79, 16.57, 38.11, 58.77 and 9.7 % consecutively. These findings were in agreement with those obtained by El-Khayat and Zaghloul (1999), they found that application of biogas manure augmented the highest values of oil percentage and nitrogen content of caraway.

In terms of interaction between three different sources of fertilization the data within hand in Tables (4, 5 and 6) revealed that, inoculation with *Bacillus polymyxa* + inorganic nitrogen (60 kg N/fed.) surpassed all other biofertilizer treatments. Except plant fruit yield in the first season, application of inorganic nitrogen (60 kg N/fed.) + soil inoculation with *Bacillus polymyxa*

produced the highest values for all the tested parameters in comparison with other biofertilizer treatments. Plant height, number of branches and umbels, dry weights of umbels and plant, dry fruit yield, oil content, estimated yields of fruits and oil significantly increased by about 25.96, 87.41, 43.47, 92.6, 288.2, 99.85, 52.1, 102.3 and 208.3 %, when soil inoculation with *Bacillus polymyxa* + inorganic nitrogen (60 kg N/fed.) in comparison with untreated plants, respectively in both seasons. These findings were in agreement with Kandeel and Sharaf (2003) they showed that, mixed inoculation with three biofrtilizers supplemented with half dose of NPK gave the highest and significant increase of growth parameters, oil content (%) and oil yield of marjoram, as well as, the N, P and K % in the plant herb. Kandeel *et. al.* (2002) reported that, inoculation of sweet basil with *Azotobacter* + *Azosopirillum* supplemented with half or full dose of the recommended mineral N-fertilizer, significantly increased plant growth, oil % and oil yield $plot^{-1}$ compared with uninoculated plants and given full dose of nitrogen. El-Sawy *et. al.* (1998) reported that inoculation with a mixture of *Azotobacter*, *Azosopirillum* and VAM amended with full dose of P as rock phosphate and inorganic N-fertilization, in combination with VAM inoculation, increased plant growth and khellin production of *Ammi visnaga* L. plant.

In terms of the interaction among biogas manure and inorganic nitrogen fertilizer, the highest values were obtained by applying biogas manure (60 kg N/fed.) + mineral nitrogen (60 kg N/fed.) for all tested parameters (Tables 4, 5 and 6), with exception of plant height, umbels number, estimated oil yield and nitrogen parameters, wherein the highest values were recorded by applying biogas manure (60 kg N/fed.) + inorganic nitrogen (90 kg N/fed.). These results are confirmed by those obtained by El-Khayat and Zaghloul (1999) who found that application of biogas manure produced the highest values of oil percentage and nitrogen of *Carum carvi* L.

With respect of the essential constituents GC analysis of the fennel fruits oil emphasized that the main oil components are estragole, limonene, fenchone, anethole, γ-terpinene, phellandrene and anis aldehyde. They are formed 90.7 % of the total oil constituents. These findings are confirmed with those obtained by Kandil, (2002), Braun and Franz (1999), Singh and Mahey(1994) and Embong et al. (1977),they found that these compounds are the major constituents of fennel oil. In concern to fenchone compound, which is responsible for fennel bitterness it was found that inorganic nitrogen fertilization alone increased its relative percent. Meanwhile the interaction between inorganic N and bio-organic fertilizers reduced the relative percent of fenchone, which means more favorable taste for the essential oil if it is used for food dressing. Thus bio-organic fertilizers may be an excellent sources for crop production with good quality. These findings are in agreement with Atta *et al.* (1999) they found that chemical fertilization increased fenchone relative percent of fennel meanwhile, organic fertilizer reduced its relative percent.

Amongst microbial densities in fennel rhizosphere reached their maximal levels in inoculated plants. It was also found that the highest numbers of total microbial flora and *B.polymyxa* in rhizosphere of growing plants were noticed after 100 days of cultivation then a decrease in their count was observed thereafter. Spraying of the N_2 fixing bacteria in soil

inoculation treatment increased the *Bacillus* number in fennel rhizosphere either with or without inorganic N-fertilizer than those obtained by fruits inoculation treatment. Rhizospheric soil of plants inoculated with diazotrophic bacteria in the absence or presence of inorganic N-fertilizer and biogas manure gave higher count of the *B. polymyxa* than uninoculated plants for both soil or fruits inoculation treatments .Rennie and Thomas (1987) reported that of 10 different cultivars of spring wheat inoculated with *B.polymyxa* C-11-25 and *Azospirillum brasilense* ATCC29729 only one cultivar (Cadet; inoculated with *B. polymyxa* C-11-25) showed consistent plant yield responses due to inoculation. Thus, as suggested by other workers (Rennie & Larson 1979; Baldani *et al.*, 1983 and Rennie *et al.*, 1983), plant and/or bacterial genotype might be the controlling factor in an inoculation response.

CO_2 evolution as an indication for microbial activity in fennel rhizosphere was assayed during the different stages of plant growth. In all cases, inoculation and fertilization with mineral N-fertilizer and biogas manure increased the rates of CO_2 evolution . Concerning inoculation with *B.polymyxa* bacteria, it was found that CO_2 evolution rates were lower in the rhizosphere of growing plants produced from fruits inoculated with asymbiotic bacteria than those of plants grown in soil inoculated by spraying the same bacteria . Increasing the levels of biogas manure and inorganic N-fertilizer resulted in an increase in the rates of CO_2 emission . These results were in accordance with those of Nannipieri *et al.*,(1990) who reported that the metabolic activities of soil microorganisms can be quantified by measuring the CO_2 production .Suttner and Alef (1988) found that CO_2 evolution associated significantly with soil organic matter(C_{org}) and most microbial parameters. Abo El- Ala (2002) mentioned that application of biofertilizers and inorganic nitrogen and phosphorus fertilization significantly increased the levels of CO_2 evolution in the rhizosphere of marjoram plants in comparison with uninoculated plants at the 2nd season.

In conclusion, the soil inoculation with *Bacillus polymyxa* and supplemented with mineral nitrogen (60 kg N/fed.) was the best treatment to obtain high productivity and quality of fennel. Furthermore application of biogas manure at level 60 kg N/fed. and 60 kg inorganic N/fed. giving the best growth parameters, yield and good quality of fennel in comparison with inorganic fertilizer alone.

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تأثير التسميد المعدني والعضوي والحيوي على نمو ومحصول ومكونات الزيت الطيار لنبات الشمر

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أجريت تجربة حقلية خلال موسمي ٢٠٠١/٢٠٠٢ ، ٢٠٠٢/٢٠٠٣ بالمزرعة التجريبية الخاصة بكلية الزراعة جامعة عين شمس لدراسة إنتاجية ونوعية محصول الشمر تحت تأثير تنوع مصادر التسميد النتروجيني. ولقد تم إضافة نترات الأمونيا كمصدر للسماد النتروجيني المعدني بمعدلات صفر ،٣٠ ،٦٠ ،٩٠ كجم ن/فدان وسماد البيوجاز كمصدر للسماد العضوي بمعدلين هما ٤٠ ،٦٠ كجم ن/فدان . وكذلك تم التلقيح الميكروبي بالباسيلس بوليمكسا كمخصب حيوي . وبدون النظر إلى التركيزات كانت النتائج كما يلي : تفوق التسميد النتروجيني المعدني بسماد نترات الأمونيوم على الأسمدة العضوية والحيوية ، وكانت أفضل استجابة لقياسات النمو ومحتوى الثمار من الزيت ، ومحصول الفدان من الثمار والزيت الطيار، وكذلك محتوى الثمار من النتروجين والبروتين ، والنتروجين الممتص عند إضافة السماد المعدني بمعدل ٩٠ كجم نيتروجين/فدان.

و بالنسبة للتسميد الحيوي تفوق حقن التربة بالباسيلس بوليمكسا عن تلقيح الثمار بنفس الميكروب حيث كانت هناك زيادات معنوية في القياسات سابقة الذكر ، كما أظهرت النباتات التي تم تلقيحها بالمخصب الحيوي بإضافة أو عدم إضافة التسميد النتروجيني المعدني تفوقا ملحوظا في قياساتها عن النباتات غير الملقحة. وكذلك أدي استخدام سماد البيوجاز بمعدل ٦٠ كجم ن/فدان إلي الحصول علي أعلي النتائج في جميع المعاملات المختبرة متفوقا علي التلقيح بالمخصب الحيوي. ولقد أدى تلقيح التربة بميكروب باسيلس بوليمكسا مع إضافة السماد النيتروجيني المعدني بمعدل ٦٠ كجم ن/فدان إلى إعطاء أفضل النتائج في قياسات النمو، ومحتوى الزيت الطيار ، ومحصول الفدان من الثمار و الزيت الطيار مقارنة بالنباتات غير المعاملة(كنترول) . باستثناء محصول النبات من الثمار في الموسم الأول فقد أعطي استخدام السماد المعدني بمعدل ٩٠ كجم ن/فدان وحقن التربة بالسماد الحيوي أفضل المعاملات كماأعطي احسن محصول من الثمار.

وعلى الجانب الأخر تم الحصول علي أعلي النتائج عند استخدام سماد البيوجاز بمعدل ٦٠ كجم ن/فدان مع السماد النتروجيني المعدني بمعدل ٦٠ كجم ن/فدان في معظم القياسات تحت الدراسة ولكن عند إضافة سماد البيوجاز (٦٠ كجم ن/فدان) مع إضافة (٩٠ كجم ن/فدان) من السماد المعدني كانت هي الأفضل في الحصول على زيادة معنوية في محصول الفدان من الزيت ، ومحتوى الثمار من النيتروجين والبروتين ، و النيتروجين الممتص.

وبالتحليل الكروماتوجرافي الغازي لمكونات الزيت الطيار في الثمار وجد ان الإستراجول ، والليمونين ، والفنشون والانيثول ، و جاما تربنين ، والفلاندرين هي مركبات رئيسية للزيت . ولقد أدت المعاملات المختلفة للتسميد النتروجيني متعدد المصادر إلي اختلاف نسب مكونات الزيت الطيار ، كما أظهرت النتائج وجود اختلاف في نسب المركبات المكونة للزيت الطيار نتيجة لاختلاف مصادر الأسمدة النتروجينية والتداخل بين المعاملات .

ولقد أدى استخدام السماد النتروجيني المعدني مع سماد البيوجاز في وجود المخصبات الحيوية إلى حدوث تأثيرا إيجابيا من الناحية الغذائية علي مركب الفنشون المسئول عن المرارة في نبات الشمر حيث أدى التفاعل بين هذه الأسمدة إلي تقليل نسبة الفنشون حيث ينعكس ذلك على خفض معدل المرارة في النباتات المعاملة عنها في حالة النباتات غير المعاملة مما يحسن من طعمه عند استخدامه في التوابل المضافة للأغذية .

تأثرت أعداد الميكروبات الكلية ، والعصويات بالتلقيح الميكروبي بالباسيلس بوليمكسا وأدى ذلك إلى زيادة هذه الأعداد في ريزوسفير نباتات الشمر الملقحة. كما تم قياس معدل انطلاق ك ا٢ أثناء مراحل النمو المختلفة للنباتات، ولقد أدى استخدام التلقيح البكتيري والتسميد النتروجيني المعدني و سماد البيوجاز إلى الحصول على أعلى معدل انطلاق لغاز ك ا٢ من ريزوسفير النباتات الملقحة عنه في حالة النباتات غير الملقحة، كما تفوقت معاملة التربة الملقحة بالرش البكتيري بالنسبة لمعدل غاز ك ا٢ المتصاعد عنها في حالة معاملة الثمار الملقحة عن طريق الغمس في اللقاح البكتيري.

ولهذا فإنه تحت ظروف الزراعة المصرية أثبتت النتائج أنه يمكن تقليل استخدام الأسمدة الكيماوية والاستعاضة عنها بالسماد العضوي (البيوجاز) أو المخصبات الحيوية في الحصول على محصول جيد قابل للتصدير آمن للاستهلاك الأدمي .