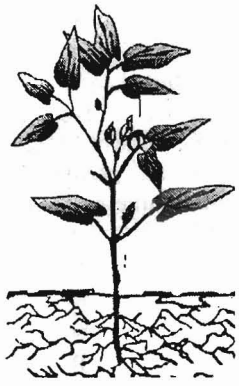

465
Annals
Agric. Sci.,
Ain Shams Univ.,
Cairo, 52(2), 465-484, 2007

EFFECT OF SOME CHEMICAL AND BIO – FERTILIZERS ON PEPPERMINT PLANTS GROWN IN SANDY SOIL
2. EFFECT ON ESSENTIAL OIL PRODUCTION , CHEMICAL COMPOSITION AND ANATOMICAL FEATURES

[38]

Swaefy Hend[1], M.F.; Weaam R.A. Sakr[1]; A.Z. Sabh[2] and A.A. Ragab[3]

1. **Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, Egypt.**
2. **Agricultural Botany Department, Faculty of Agriculture, Cairo University, Giza, Egypt.**
3. **Agricultural Microbiology Department, Soil, Water and Environment Research Institute, Agricultural Research Center, Giza, Egypt**

Keywords: Chemical fertilizers, Biofertilizers, Peppermint, Essential oil

ABSTRACT

This study was carried out at the Experimental Nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, during the two successive seasons of 2005 and 2006 to investigate the effect of chemical fertilizers (N and P) and / or some microbial strains (*Rhizobium leguminosarum* bv. *phaseoli*, *Azotobacter chroococcum* and *Bacillus megatherium* var. *phosphaticum*) as biofertilizers or as a plant growth promoting rhizobacteria (PGPR) on the oil yield, chemical composition and anatomical features of *Mentha piperita*, L. plants grown in sandy soil. The obtained results indicated that the treatment of 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate (50% NP) in addition to *Azotobacter* inoculation gave the highest total oil yield per plant and per feddan followed by the treatment of the same amount of chemical fertilizers plus *Rhizobium* inoculation. The enhancement of oil yield of fresh herb as a result of treating peppermint plants with *Rhizobium* or *Azotobacter* in addition to 50% NP was correlated with increasing the vascular tissues of both stem and leaf blade, also increased palisade tissue thickness in the leaf blade. Plants received 50% NP in addition to any of bio-fertilizers increased total carbohydrates content compared to those received 50% NP only.

INTRODUCTION

Peppermint (*Mentha piperita*, L.) plant is one of the most important aromatic plants in Egypt. The plant grows in different soils, preferring those deep and fertile. The leaves are used for flavouring purposes. An essential oil, obtained from the leaves, is used in perfumery and soap making industries. The oil is also used medicinally as stimulants, carminative and for sickness and vomiting **(Pandey, 1982 and Bown, 1996).**

Organically- grown agricultural products are considered to be healthier, and cause less risk to the environment, since their tissues contain lower levels of chemical residues. Also, factors such as escalating N fertilizer costs, soil structural degradation, environmental pollution, and sustainable land use have generated a growing interest in natural N fixation as a method of providing plants with their N requirements **(Subba Rao, 1984).** In this

(Received September 17, 2007)
(Accepted October 22, 2007)

respect, **Omar *et al* (1991)** reported that inoculation of wheat with *Bacillus polymyxa* can save 41.6% of the nitrogen fertilizer. Also, **Salah *et al* (1998)** on datura and **Kandeel *et al* (2002)** on *Ocimum basilicum*, L. recorded appreciable improvement on number of branches as well as fresh and dry weights of vegetative parts due to using N fixer bio-fertilizer. **El- Khyat and Zaghloul (1999)** recorded similar observations on *Carum carvi*, L. plants by using N fixer bio-fertilizer + compost application.

Biofertlizers had favorite effects on oil yield as reported by **Maheshwari *et al* (1995)** on *Cymbopogon maritini* var. *motia* plants, since they found that application of *Azotobacter chroococcum* resulted in 10.3-39.6% and 11.7-35.2% higher herbage and oil yields, respectively, than the control. Also, **AL-Qadasi (2004)** obtained the highest oil yield per plant and per feddan from basil plants received full NPK plus mixture of bio-fertilizers at 150 ml/plant.

An explanation was suggested by **Lazarovits and Nowak (1997)** who reported that the stimulatory effects of microorganisms may result from indirect action by acting as bio-control agents and reducing diseases, liberation of antibiotic substances that kill noxious bacteria.

Regarding the anatomical studies, **Medani *et al* (2000)** studied the effect of bio-fertilizers (mixture of *Azotobacter, Azospirillum* and *Bacillus*) combined with mineral N fertilizer (ammonium sulphate, 20.5%N) at full recommended dose (80 kg/fed.), two-thirds dose (53.33 kg/fed.) and one-third dose (26.66 kg/fed.) on sugar beet plants. They found that plants treated with bio-fertilizers combined with N at its two-thirds dose had a thicker leaf blade represented in palisade and spongy tissues than the control. On the other hand, midrib thickness and vascular bundle width were slightly decreased by 1.70 and 2.96%, respectively, as bio-fertilizers combined with N at its two-thirds dose in comparison with those of full recommended N-dose. **Sakr-Weaam (2001)** stated that, treating peppermint plants with NPK at 900 kg/fed./season increased number of glandular hairs/mm^2 on lower surface of leaf blade up to 41.1% more than the untreated control plants. Also, **Ramadan *et al* (2003)** stated that sugar beet plants treated with 50% mineral fertilizers in combination with a mixture of bio-fertilizers as nitrogen fixers (*Azotobacter spp.* and *Azospirillum spp.*) and as phosphate dissolving bacteria (*Bacillus spp.*) had a stimulative effect on leaf blade structure, which was attributed to the increase in thickness of leaf lamina and midvein by 49.8% and 18.4% more than the control plants, respectively. Such thicker lamina produced by this treatment was mainly due to the increase in thickness of both palisade and spongy tissues by 130.0 and 52.4% over the control, respectively. Also, the dimensions of the midvein were increased due to the increment in its length by 22.0% and its width by 22.1% more than the control plants. On the other hand, vessel diameter was decreased by 5.0% compared with the control plants.

Agamy (2004) treated sweet fennel (*Foeniculum vulgare*, Mill.) plants with mixed bio-fertilizers (*Azospirillum spp., Azotobacter spp., Pseudomonas spp.* and *Bacillus spp.*) as well as mineral fertilizers (NPK) at the rate of 0, 25, 50 and 100% of the recommended dose, each either alone or combined with mixed bio-fertilizers. They revealed that, the combination between bio-fertilizers and 100% mineral fertilizers realized the highest increase in number and diameter of oil glands. Also, thickness of leaf blade tissues represented in midvein, palisade and spongy tissues were increased with application of bio-fertilizers in combination with mineral fertilization. **Hassan *et al* (2006)** on moghat plants (*Glossostemon bruguieri*, Desf) stated that plants obtained from seeds treated with mixture of nitrogen fixers (*Azotobacter spp and Azospirillum spp.*) and phosphate dissolving bacteria (*Bacillus spp.*) and received half of the recommended dose of mineral fertilizers from nitrogen and phosphorus (NP) showed a prominent increase in thickness of both midvein and lamina of the leaf by 7.88 and 19.14%, respectively more than the control (plants received 100% of the recommended dose of mineral fertilizers). Also, the increase in lamina thickness was accompanied with 21.20% and 23.54% increments in thickness of palisade and spongy tissues compared with the control, respectively. Likewise, the midvein bundle was increased in size by 10.60% more than that of the control. Moreover, xylem vessels had wider cavities, being 40.53% more than the control.

This study was aimed to investigate the effect of some chemical and bio-fertilizers on the oil production, chemical composition and anatomical structure of *Mentha piperita*, L. plants and to assess the possibility of using these bio-fertilizers to reduce the need for chemical NP fertilization and act as a plant growth promoting rhizobacteria (PGPR).

MATERIALS AND METHODS

This study was conducted at the Experimental Nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, during the two successive seasons of 2005 and 2006.

On March 15th, (in both seasons), rhizomes of *Mentha piperita*, L. were obtained from the Experimental Farm of Medicinal and Aromatic Plants, Faculty of Pharmacy, Cairo University, Giza. The rhizomes (12 cm long, with 8 – 10 leaves) were planted in clay pots (30 cm – diameter) filled with a sandy soil. The mechanical and chemical analyses of the sandy soil **(Table 1)** were carried out before planting according to **Chapman and Pratt (1961).**

Prior to planting, the rhizomes used for planting 108 pots (6 treatments) were separately inoculated with *Rhizobium leguminosarum* bv. *phaseoli*, *Azotobacter chroococcum* or *Bacillus megatherium* var. *phosphaticum* [each at 10^8 colony forming units (CFU)/g carrier] at the rate of 300 g/fed as bio-fertilizers. The rhizomes were inoculated by wetting their bases, then immersing them in the bio-fertilizers. A basal dressing of the previous bio-fertilizers were repeated after 90 days from planting, at the rate of 300g/fed, and the pots were irrigated immediately thereafter, while vermiculite was added alone at the same rate to pots receiving no bio-fertilization.

Table 1. Physical and chemical characteristics of the soil used for growing *Mentha piperita*, L plant during 2005 and 2006 seasons.

Physical characteristics							
Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)	Soil texture	Field capacity (% V)		
32.5	62.1	1.7	3.7	Sandy	15.8		
Chemical characteristics							
pH	Organic matter (%)	$CaCO_3$ (%)	EC (dS/m) (1:2.5)	CEC (meq/100 g)	Available macro - nutrients (ppm)		
					N	P	K
7.4	1.23	0.43	0.76	5.1	19.8	3.6	98.1

Chemical N and P fertilizers were added using ammonium sulphate (20.5% N) and calcium super-phosphate (15.5% P_2O_5). Ammonium sulphate at 350 Kg/fed and calcium super-phosphate at 200 Kg/fed (100% NP) as a recommended rate reported by **Swaefy Hend (1996)** were added to 18 un-inoculated pots. Also, 50% of the recommended rate (ammonium sulphate at 175 Kg/fed and calcium super-phosphate at 100 Kg/fed) were applied to 18 un-inoculated pots. Half the number of inoculated pots (54 pots) received 50% of chemical fertilizers (NP). The above rates were divided into two doses: the first dose was added after one month from planting, and the second one was added two months later (two weeks after the first cut). Eighteen un-inoculated pots were left without NP chemical fertilization (control). All pots were supplied with potassium sulphate (48% K_2O) at the rate of 60 Kg/fed, divided into two equal doses. The first dose was applied as a basal dressing one month after planting and the second one was added two weeks after the first cut.

The layout of the experiment was a randomized complete blocks design. The experiment included 9 treatments, with 3 blocks (replicates), each block consisting of 54 planting pots (six pots for each treatment).

In each season, two cuts were taken on June 1st and August 15th by cutting the vegetative parts of all plants, 5 cm above the soil surface.

Oil yield per plant (ml) and per feddan (L) were determined based on oil% and herb fresh weight per plant. The yield per feddan was calculated with the assumption that the normal plant density is 21000 plants/ feddan. In addition, samples of fresh leaves were chemically analyzed to

determine chlorophyll a, b and total carotenoids contents (mg/g fresh weight) using the method described by **Nornai, 1982**, then total chlorophylls were calculated. Total carbohydrates content (%) was determined in samples of dried herb using the method described by **Dubois *et al* 1956**. Also, samples of dried herb were digested to extract nutrients as described by **Piper, 1947**. The extract was analyzed to determine its contents of N (using the modified micro-Kjeldahl method as described by **Pregl, 1945**), and phosphorus (according to **Jackson 1967),** while potassium was estimated photometrically using Jenway flamephotometer according to **Chapman and Partt (1961)**.

The data of oil yield per plant and per feddan were subjected to an analysis of variance, and the differences between means were compared using the "Least Significant Difference (LSD)" test at 0.05 level, as recommended by **Snedecor and Cochran (1982).**

For anatomical studies, specimens of selected treatments according to their beneficial effects on plants (100% NP, *Rhizobium* + 50% NP, *Azotobacter* + 50% NP) in addition to the control were taken directly before the second cut of the second season (plants were 150 days old) from the middle of the fifth internode from the tip of shoot (stem) as well as the middle of the leaf blade on the fifth node. Specimens were kept for killing and fixation for at least 48 hours in F.A.A. (10 ml. formalin, 5 ml glacial acetic acid and 85 ml ethyl alcohol 70%). The fixed materials were then washed in 50% ethyl alcohol, dehydrated in normal butyl alcohol series and finally embedded in paraffin wax of 56 ° C m.p. Sections, 20 µ thickness, were stained with safranin/light green combination, then cleared in xylene and mounted in Canada balsam **(Sass, 1967 & Nassar and El-Sahhar, 1998).** The prepared slides were microscopically examined to detect the anatomical manifestations of the chosen treatments and measurements were taken. Averages of 6 readings from 3 slides were calculated and photomicrographs were taken.

An impression of the lower epidermis (surface) of the fifth leaf blades from the tip of shoot (stem) were prepared during the second cut of the second season according to the method described by **El-Sgai and Sabbour (2000) & Sakr-Weaam (2001).** Slides were microscopically examined and the mean number of glandular hairs/ microscopic field was accounted as well as mean diameter of gland (µ) was determined for each choice treatment and control.

RESULTS AND DISCUSSION

1- Oil yield/plant

In the first cut of each season **(Table, 2)**, the plants received chemical fertilization at 350 Kg/fed ammonium sulphate and 200 Kg/fed calcium super- phosphate gave the significantly higher oil yield per plant (0.563 and 0.834 ml in the first and second seasons, respectively) compared to most of the other treatments. In the second cut of each season, the plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Azotobacter* inoculation gave the significantly higher oil yield per plant (1.902and 2.258 ml in the first and second seasons, respectively), compared to the other treatments, in most cases.

Generally, data presented in **Table (2)** showed that in the first season, the plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Azotobacter* inoculation gave the significantly higher total oil yield per plant (2.347 ml/plant) and the total oil yield per feddan (49.29 ml/fed), compared to the other treatments in most cases. Also, in the second season this treatment gave insignificantly lower total oil yield per plant (2.826 ml) and the total oil yield per feddan (59.35 liter) as compared to the highest total oil yield per plant (2.890 ml) and the total oil yield per feddan (60.69 liter) which resulted from the plants received 350 Kg/fed ammonium sulphate and 200 Kg/fed calcium superphosphate.

From the above mentioned results, it can be noticed that plants which received chemical fertilization at 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate, in addition to *Azotobacter* inoculation, gave the highest total oil yield per plant and total oil yield per feddan followed by the treatment of the same amount of chemical fertilizer plus *Rhizobium* inoculation.

These results are almost in harmony with those obtained by **Maheshwari *et al* (1995)** on *Cymbopogon maritini* var. *motia*, they found that application of *Azotobacter chroococcum* resulted in 10.3 - 39.6% and 11.7 - 35.2% higher herbage and oil yields, respectively, than the control. Also, **AL-Qadasi (2004)** obtained the highest oil yield per plant and per feddan from basil plants received full NPK plus mixture of bio-fertilizers at 150 ml/plant.

Table 2. Effect of some chemical and bio-fertilizers on essential oil yield/plant, (ml) and oil yield/ feddan (L) of *Mentha piperita*, L. plants during 2005 and 2006 seasons.

fertilization treatments	Essential oil yield/plant (ml)						oil yield/ feddan (L/fed)	
	First season			Second season				
	First cut	Second cut	Total oil yield/ plant	First cut	Second cut	Total oil yield/ plant	First season	Second season
Control	0.202	0.497	0.699	0.226	0.604	0.830	14.68	17.43
NP50%	0.506	1.655	2.153	0.500	1.771	2.270	45.21	47.67
NP100%	0.563	1.642	2.205	0.834	2.056	2.890	46.31	60.69
Rhizobium (Rh)	0.307	1.095	1.402	0.289	1.053	1.342	29.44	28.18
Rh + NP50%	0.531	1.587	2.118	0.491	2.127	2.618	44.48	54.98
Azotabacter (Az)	0.354	1.035	1.389	0.328	1.516	1.844	29.17	38.72
Az + NP50%	0.445	1.902	2.347	0.568	2.258	2.826	49.29	59.35
Bacillus (B)	0.386	1.045	1.427	0.475	1.160	1.635	29.97	34.34
B + NP50%	0.444	1.376	1.826	0.544	1.699	2.243	38.35	47.10
LSD $_{0.05}$	**0.060**	**0.230**	**0.219**	**0.100**	**0.250**	**0.279**	**5.48**	**6.98**

NP100% = Ammonium sulphate at 350 Kg/fed and calcium super-phosphate at 200 Kg/fed
NP50% = Ammonium sulphate at 175 Kg/fed and calcium super-phosphate at 100 Kg/fed
Rh= *Rhizobium leguminosarum* bv . *phaseoli* Az = *Azotobacter chroococcum*
B = *Bacillus megatherium* var. *phosphaticum*

2- Leaf pigments

a -Total chlorophylls content

Data recorded in **Table (3)** showed the total chlorophylls (a+b) in leaves of *Mentha piperita*, L. plant. In the first cut of both seasons and the second cut in the second season, fertilization treatments increased total chlorophylls (a+b) contents, compared to the unfertilized control plants, in most cases.

In both seasons, plants received chemical fertilization at 350 Kg/fed ammonium sulphate and 200 Kg/fed calcium super-phosphate gave higher content of total chlorophylls (a+b) than that recorded with the plants received chemical fertilization at 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate as well as unfertilized control plants .

In the first season, plants received *Bacillus* inoculation only as a bio- fertilizer gave the highest total chlorophylls (a+b) content in both cuts (2.91 and 1.63 mg/g fresh weight in the first and second cuts, respectively), compared to the other treatments. On the other hand, in the first season, the plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate gave the lowest total chlorophylls (a+b) in the first cut (1.45mg/g f.w.), compared to the other fertilization treatments. Also, the plants received *Azotobacter* only gave the lowest total chlorophylls (a + b) in the second cut (1.11 mg/g f.w.), compared to the other treatments.

In the second season, combination between chemical and bio-ferilizers played a pronounced role in increasing total chlorophylls (a+b) contents. Plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Azotobacter* inoculation resulted in the highest total chlorophylls (a + b) contents in the first cut (3.74 mg/g f.w.), whereas that received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Rhizobium* inoculation resulted in the highest total chlorophylls (a + b) contents (1.88 mg/g f.w.) in the second cut. On the other hand, the unfertilized control plants gave the lowest total chlorophylls (a + b) contents in both cuts (1.28 and 1.03 mg/g f.w. in the first and second cut, respectively), compared to the other treatments in the second season. The obtained results are almost in harmony with those obtained by **Ragab and Rashad (2003),** they found that rhizobial inoculation enhanced the total chlorophylls (a+b) of sorghum plants and **Swaefy Hend and Milad (2006)** on *Euryops pectinatus* plants, they found that there was a significant increase in total chlorophylls content by using the combination of microbein, 50% of the soil chemical fertilizers and nofatrein.

Table 3. Effect of some chemical and bio-fertilizers on contents of total chlorophylls (a+b) and carotenoides (mg/g fresh weight) in leaves of *Mentha piperita*, L. plants during 2005 and 2006 seasons

Treatments	First season (2005)				Second season (2006)			
	Total Chlorophylls		Carotenoids		Total Chlorophylls		Carotenoids	
	First cut	Second cut	First cut	Second cut	First cut	Second cut	First cut	Second cut
Control	1.94	1.35	0.49	0.33	1.28	1.03	0.51	0.31
NP50%	1.45	1.25	0.52	0.28	2.03	1.54	0.32	0.28
NP100%	2.55	1.37	0.37	0.31	2.93	1.79	0.31	0.37
Rhizobium (Rh)	2.01	1.32	0.45	0.21	2.03	1.08	0.42	0.41
Rh + NP50%	2.78	1.12	0.17	0.16	2.80	1.88	0.24	0.52
Azotabacter (Az)	2.60	1.11	0.34	0.22	2.50	1.22	0.10	0.41
Az + NP50%	2.75	1.20	0.34	0.22	3.74	1.23	0.04	0.27
Bacillus (B)	2.91	1.63	0.16	0.32	2.90	1.59	0.10	0.30
B + NP50%	2.37	1.50	0.39	0.29	3.28	1.11	0.31	0.12

NP100% = Ammonium sulphate at 350 Kg/fed and calcium super-phosphate at 200 Kg/fed
NP50% = Ammonium sulphate at 175 Kg/fed and calcium super-phosphate at 100 Kg/fed
Rh= *Rhizobium leguminosarum* bv . *phaseoli* Az = *Azotobacter chroococcum*
B = *Bacillus megatherium* var. *phosphaticum*

b - Carotenoids content

Regarding the effect of fertilization treatments on carotenoids content, data presented in **Table (3)** showed that plants received fertilization treatments in both seasons gave lower carotenoids content, compared to the control plants, in most cases. Unfertilized control plants gave the highest carotenoids content in the second cut of the first season (0.33 mg/g f.w.) and the first cut of the second season (0.51 mg/ g f.w.). Plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate (50% NP) resulted in the highest carotenoids content in the first cut of the first season (0.52 mg/g f.w.), whereas the highest value in the second cut of the second season (0.52 mg/g f.w.) was recorded in plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Rhizobium* inoculation.

In the first season, it is clear that the lowest carotenoids content was recorded with plants inoculated with *Bacillus* only in the first cut (0.16 mg/g f.w.) and in plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium superphosphate with *Rhizobium* inoculation in the second cut (0.16 mg/g f.w.) . In the second season, it could be noticed that the lowest carotenoids content was recorded in plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Azotobacter* inoculation in the first cut (0.04 mg/g f.w.), and in plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to *Bacillus* inoculation in the second cut (0.12 mg/g f.w.).

3-Total carbohydrates content

Data presented in **Table (4)** revealed that, in both cuts of the two seasons, all fertilization treatments increased total carbohydrates content in herb dry weight as compared to the values recorded with the unfertilized control plants, in most cases. In both seasons, plants fertilized with 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate gave higher total carbohydrates content than that received any of biofertilizers only, in most cases.

Plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to any bio-fertilizer gave higher total carbohydrates content than that received any of the same bio-fertilizer only. In most cases, plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to any bio-fertilizer gave higher total carbohydrates content than that received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate only. Generally, it is evident that the plants received 350 Kg/fed ammonium sulphate and 200 Kg/fed calcium super-phosphate gave the highest total carbohydrates content, whereas the unfertilized control plants gave the lowest total carbohydrates content, in both seasons.

Table 4. Effect of some chemical and bio-fertilizers on total carbohydrates, nitrogen, phosphorus and potassium contents (% of dry weight) in the herb of *Mentha piperita*, L. plants during 2005 and 2006 seasons.

Fertilization treatments	Total carbohydrates%		N%		P%		K%	
	First cut	Second cut	First cut	Second cut	First cut	Second cut	First cut	Second cut
First season (2005)								
Control	20.02	24.70	0.80	0.96	0.12	0.15	1.80	2.20
NP 50%	27.95	30.03	1.20	1.12	0.08	0.14	1.70	1.79
NP 100%	30.94	36.92	1.41	1.82	0.09	0.13	1.30	1.95
Rh	21.06	27.04	0.84	0.98	0.11	0.14	1.90	2.10
Rh + NP 50%	28.34	32.63	1.04	1.21	0.07	0.20	1.68	1.89
Az	20.80	23.53	0.98	1.04	0.09	0.18	1.86	2.60
Az + NP 50%	29.12	34.45	1.68	1.54	0.06	0.11	2.80	1.70
B	27.04	25.74	1.12	1.34	0.09	0.15	1.85	2.19
B + NP 50%	28.47	29.77	1.35	1.73	0.06	0.14	1.89	2.10
Second season (2006)								
Control	20.25	23.10	1.02	1.23	0.17	0.13	2.10	2.01
NP 50%	25.20	27.90	1.39	1.84	0.10	0.11	1.80	1.75
NP 100%	32.70	30.75	1.88	2.28	0.11	0.09	1.45	1.60
Rh	24.75	25.35	1.30	1.64	0.15	0.15	1.92	2.20
Rh+NP 50%	30.15	28.95	1.70	1.95	0.09	0.11	1.70	1.76
Az	25.95	30.60	1.36	1.80	0.15	0.11	1.85	1.95
Az+NP 50%	30.90	37.20	1.81	2.11	0.15	0.12	2.14	1.80
B	20.40	24.75	1.19	1.51	0.13	0.11	1.79	2.11
B+NP 50%	22.65	32.10	1.48	1.86	0.19	0.13	1.85	1.93

NP100% = Ammonium sulphate at 350 Kg/fed and calcium super-phosphate at 200 Kg/fed
NP50% = Ammonium sulphate at 175 Kg/fed and calcium super-phosphate at 100 Kg/fed
Rh= *Rhizobium leguminosarum* bv . *phaseoli* Az = *Azotobacter chroococcum*
B = *Bacillus megatherium* var. *phosphaticum*

4-Contents of nutrients (N, P and K)

a- N content (% of herb dry matter)

Data presented in **Table (4)** revealed that, in both cuts of the two seasons, all fertilization treatments increased N% in the dry herb of *Mentha piperita* plants, as compared to the unfertilized control plants.

In both seasons, plants fertilized with 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate (50% NP) gave higher N% than that received any of bio-fertilizers only, in most cases. Plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to any bio-fertilizer gave higher N% than that received any of the same bio-fertilizer only. In most cases, plants received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate in addition to any bio-fertilizer gave higher N% than that received 175 Kg/fed ammonium sulphate and 100 Kg/fed calcium super-phosphate only. Generally, plants received 350 Kg/fed ammonium sulphate and 200 Kg/fed calcium super-phosphate gave the highest N%, whereas the unfertilized control plants gave the lowest N%, in the two cuts of both seasons.

b- P and K content (% of herb dry matter)

Data presented in **Table (4)** showed that, all fertilization treatments mostly decreased P% and K% in the dry herb of *Mentha piperita*, L. plants, as compared to the unfertilized control plants. This reduction in the P and K (as a percentage, calculated on the dry matter basis) was explained since the NP fertilization caused an increase in vegetative growth. This caused an increase in the uptake

of P and K (which can be confirmed by calculating the total amount of P and K in the herb, by multiplying the herb dry weight by the P and K percentage), compared to the control. However, the P and K percentage was reduced because the uptaken P and K was "diluted" in a high dry weight of herb (**Marschner, 1995**). The previous results are in harmony with the results obtained by **Abd-El Latif and Salem (2002) on** *Tagetes minuta*, L., **Kandeel *et al* (2002)** on *Ocimum basilicum*, L. and **Ragab and Rashad (2003)** on sorghum plants.

Anatomical studies

1-Anatomy of the stem

Microscopical counts and measurements of certain characters in transverse sections of the fifth internode of *Mentha piperita* stem treated with 100% NP, *Rhizobium* inoculation + 50% NP and *Azotobacter* inoculation + 50% NP in addition to the untreated plants, are given in **Table (5).** Likewise, microphotographs illustrating these treatments are shown in **Figure (1)**. Treating the plants with 100% NP increased dimensions of the stem by 15.6% over the control. The increase in stem dimensions could be attributed to the increase in pith dimensions by 15.4%, epidermis thickness by 16.7% and cortex thickness by 13.2% in sides and 23.5% in corners over the control. The increment of cortex thickness may be due to the increase in number of parenchyma layers in corners (16.7%) over the control, while such increment in corners may be due to the increase in thickness of collenchyma layers by 9.9% and parenchyma layers by 22.5%, as well as number of parenchyma layers by 16.7% over the control. *Mentha piperita* stem is quadrangular in transverse section, with well-defined groups of collenchyma in the 4 angles. Regarding the effect of 100% NP fertilization on the anatomical features of vascular bundles, it was found that such treatment did not affect the number of large bundles (4 bundles) but increased their dimensions by 7.6% in length and 19.2% in width, comparing with control plants, this could be attributed to the increase in thickness of phloem and xylem being, 23.7 and 4.3%, respectively, over the control. Increasing thickness of xylem tissue as a result to 100% NP treatment was mainly due to increasing number of xylem rows/bundle and number of xylem vessels/row as well as diameter of xylem vessel being 7.7, 16.7 and 17.2%, respectively, more than the control plants.

Table 5. Effect of some chemical and bio-fertilizers on anatomical features of the fifth internode of *Mentha piperita* stem directly before the second cut of the second season (Averages of 6 readings from 3 slides).

Treatments / Characters	Control	100% NP	± % to control	*Rhizobium* +50% NP	± % to control	*Azotobacter* +50% NP	± % to control
Dimensions of stem, µ	2988 x 2556	3312x 3096	+ 15.6	3060 x 2736	+ 4.6	3384 x 3060	+ 16.2
Dimensions of pith, µ	2304 x 2124	2628x 2484	+ 15.4	2376 x 2232	+ 4.1	2880 x 2412	+ 19.5
Epidermis thickness, µ	24	28	+ 16.7	24	0.0	24	0.0
Cortex thickness, µ :							
In sides	220	249	+ 13.2	220	0.0	220	0.0
In corners :	498	615	+ 23.5	528	+ 6.0	557	+ 11.9
Collenchyma	293	322	+ 9.9	264	- 9.9	293	0.0
Parenchyma	205	293	+ 42.9	264	+ 28.8	264	+28.8
No. of cortex layers :							
In sides	7	7	0.0	7	0.0	7	0.0
In corners :							
Collenchyma	10	10	0.0	10	0.0	12	+ 20.0
Parenchyma	6	7	+ 16.7	7	+16.7	7	+ 16.7
No. of large bundles	4	4	0.0	4	0.0	4	0.0
Dimensions of large bundle, µ							
Length	381	410	+ 7.6	440	+ 15.5	410	+ 7.6
Width	1467	1748	+ 19.2	1529	+ 4.2	1654	+ 12.8
Phloem thickness, µ	59	73	+ 23.7	88	+ 49.2	73	+ 23.7
Xylem thickness, µ	323	337	+ 4.3	352	+ 9.0	337	+ 4.3
No. of xylem rows / bundle	28	30	+ 7.7	32	+ 14.3	34	+ 21.4
No. of xylem vessels/ row	6	7	+ 16.7	8	+ 33.3	8	+ 33.3
Diameter of xylem vessel, µ	29	34	+ 17.2	39	+ 34.5	34	+ 17.2

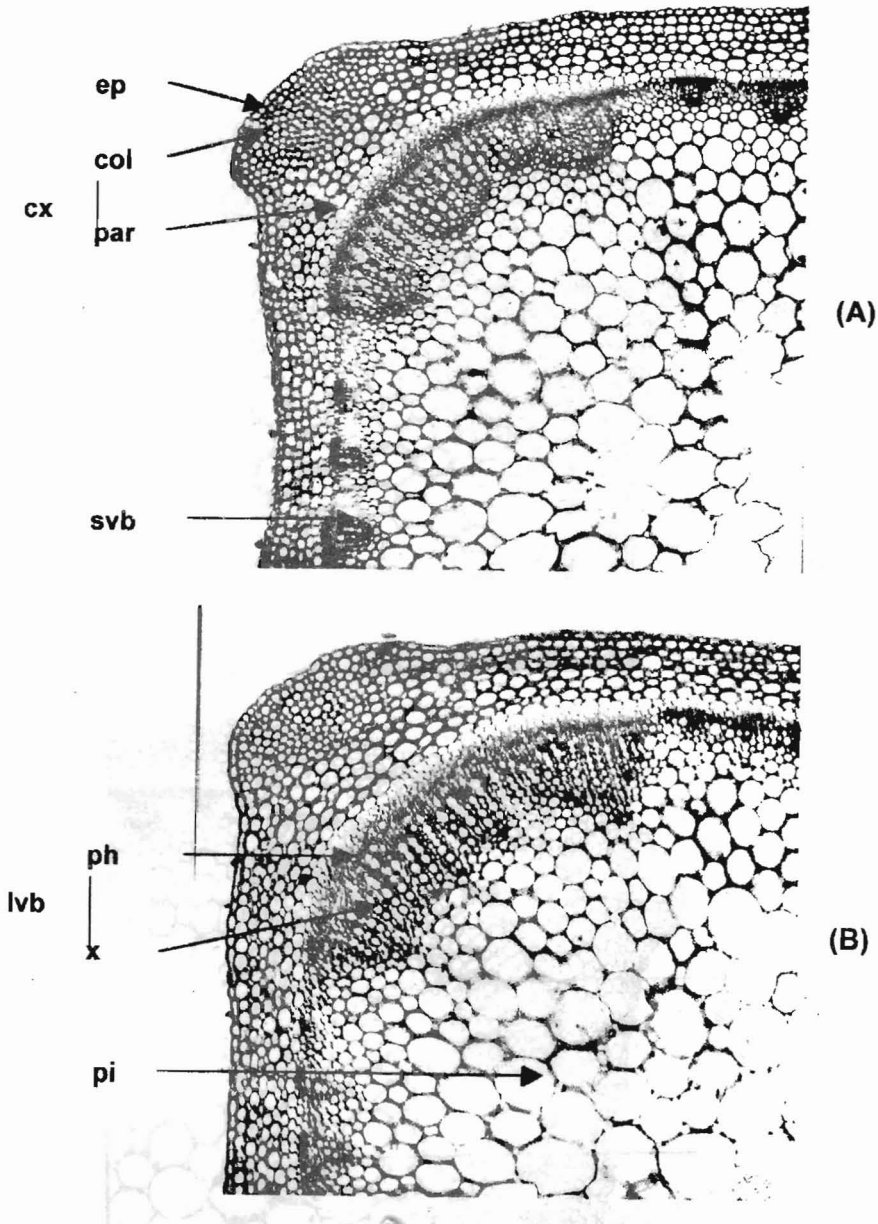

Fig. 1: Transverse sections through the median portion of the 5th internode of *Mentha piperita* plant. (X40)

A) Untreated plant

B) Plant treated with NP

***Details*: col, collenchyma; cx, cortex; ep, epidermis; lvb, large vascular bundle; par, parenchyma; ph, phloem; pi, pith; svb, small vascular bundle; x, xylem.**

Cont.

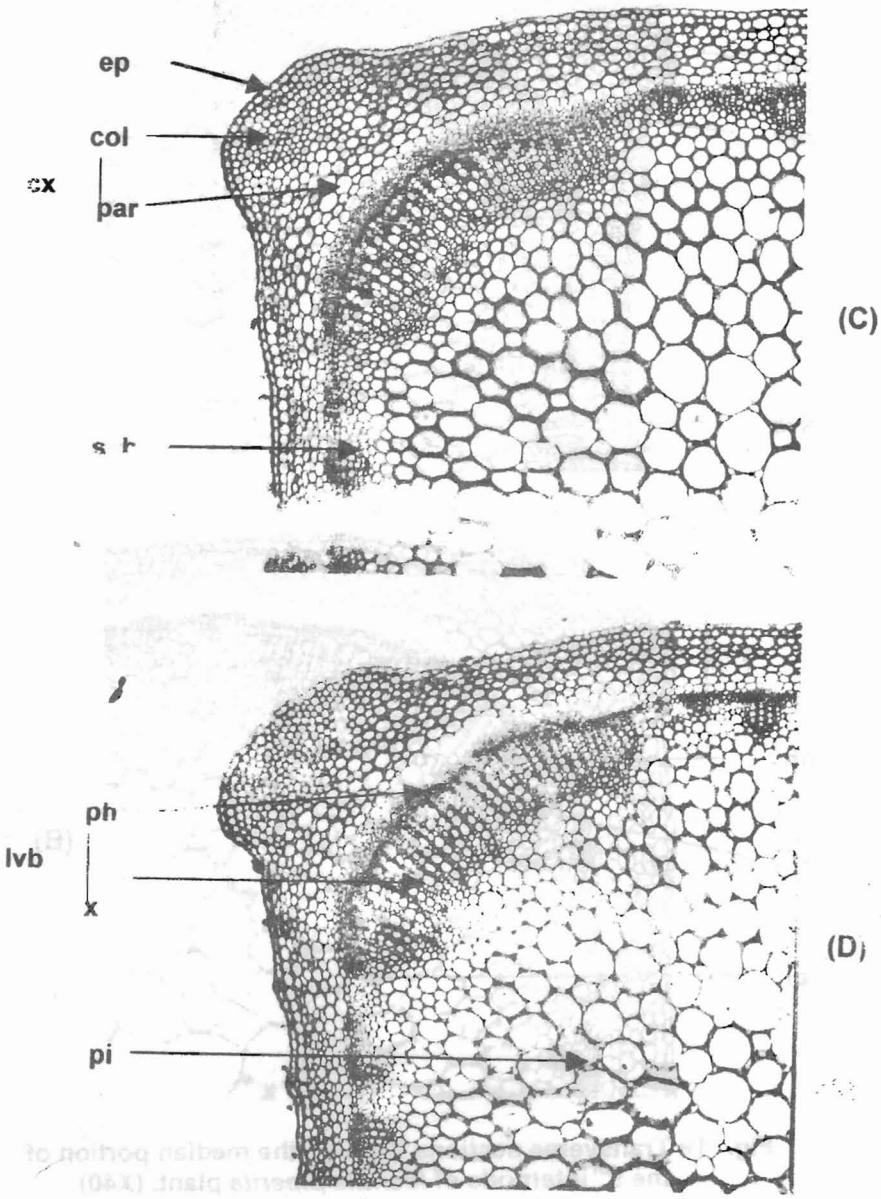

Fig. 1 - Cont. (X40)
C) Plant treated with *Rhizobium* + 50% NP
D) Plant treated with *Azotobacter* + 50% NP

Fertilization of *Mentha piperita*, L. with 50% NP in addition to the biofertilizers, *Rhizobium* or *Azotobacter* affected the anatomical features of stem **(Table 5 and Fig. 1)**. Relative to the control, both treatments exhibited wider stem dimensions being; + 4.6 and +16.2%. Such wideness may be due to the increase in pith dimensions by 4.1 and 19.5%, and cortex thickness in corner by 6.0 and 11.9% over the control in both treatments, respectively.

Both mixture treatments between chemical and bio-fertilizers did not affect cortex thickness in sides, as both thickness and number of layers were still constant, while the reason for increasing cortex thickness in corners of the two treatments (28.8% in parenchyma tissue) may be due to the increase in number of parenchyma layers by 16.7% for each treatment over the control.

In spite of constancy of number of collenchyma layers in corner comparing with the control plants (10 layers) in the first combination (*Rhizobium* + 50% NP), and increasing such number up to 12 layers (20%) in the second combination (*Azotobacter* + 50% NP), thickness of collenchymatous cortex in corners was decreased in the first combination by 9.9% below the control and still constant in the second combination.

Regarding the effect of chemical and bio-fertilization treatments as 50% NP plus either *Rhizobium* or *Azotobacter* inoculation on vascular system of *Mentha piperita* stems, the results indicated that mean dimensions of main four large bundles were increased as a result to both treatments by 15.5 and 7.6% in length and by 4.2 and 12.8% in width, respectively over the control. The increment of large bundle dimensions may be mainly due to the wide increase of phloem tissue thickness, being 49.2 and 23.7% in addition to xylem tissue thickness, being 9.0 and 4.3 % over the control, in both combination treatments, respectively. Increasing thickness of xylem tissue could be attributed to the increase in number of xylem rows/ bundle by 14.3 and 21.4%, number of xylem vessels per row by 33.3% and 33.3% as well as diameter of xylem vessel by 34.5 and 17.2% over the control for the combinations containing *Rhizobium* and *Azotobacter* bio-fertilizers, respectively.

2- Anatomy of the leaf blade

Transverse sections of *Mentha piperita* leaf blade as affected by 100% NP or 50% NP plus *Rhizobium* or *Azotobacter* inoculation as bio-fertilizers in addition to the untreated plants are presented in **Table (6)** and **Figure (2)**. It is realized that, relative to the control, 100% NP treatment increased thickness of leaf lamina by 9.1% and midvein dimensions by 11.6 in length and 14.6% in width. Though, NP treatment thickened the leaf blade that could be attributed to the increase in thickness of palisade tissue and lower epidermis, as well as dimensions of midvein region. These increments amounted to 43.0, 11.1, 11.6 (in length) and 14.6% (in width), respectively. On the other hand, such treatment decreased thickness of spongy tissue and upper epidermis by 21.4 and 7.7%, respectively. NP treatment did not affect either number of palisade or spongy tissue layers, being 1 and 4 layers, respectively. Increasing dimensions of midvein region could be attributed to increasing thickness and number of parenchyma below upper epidermis (74.2 and 50.0% , respectively), thickness of collenchyma and parenchyma above lower epidermis (9.1 and 16.3% , respectively) and dimensions of midvein vascular bundle (8.3 and 22.3% in length and width, respectively) over the control. The increase in dimensions of midvein vascular bundle may be mainly due to the increase in thickness of xylem tissue, number of xylem rows and number of xylem vessels/row, being 24.1, 10.0 and 25.0% over the control, respectively.

In the meantime, plants treated by 50% NP and either *Rhizobium* or *Azotobacter* inoculation showed a remarkable increase in midvein dimensions, being 27.0 and 38.5% in length and 41.1 and 61.8% in width over the control in *Rhizobium* and *Azotobacter* inoculation combinations, respectively, in spite of constancy the leaf lamina thickness in the first combination and its reduction in the second one (-9.0%) comparing with the control. The constancy of leaf lamina thickness in the first combination may be due to the decrease in thickness of upper epidermis (-15.4%) and spongy tissue (-12.2%) and increase in thickness of lower epidermis (11.1%) and palisade tissue (14.0%), while the reduction of lamina thickness in the second combination may be due to the decrease in thickness of both upper epidermis (-15.4%) and spongy tissue (-20.4%) compared to the untreated plants. The increase in the midvein dimensions as a result of first combination (*Rhizobium* + 50% NP) could be attributed to the increase in thickness of collenchyma tissue below upper epidermis (19.2%), parenchyma tissue below upper epidermis (132.3%) and its number of layers (50.0%), collenchyma tissue above lower epidermis (9.1%),

Table 6. Effect of some chemical and bio-fertilizers on anatomical features of leaf blade on the fifth node of *Mentha piperita*, L. stem directly before the second cut of the second season (Averages of 6 readings from 3 slides).

Characters / Treatments	Control	NP	± % to control	*Rhizo-bium* +50%NP	± % to control	*Azoto-bac-ter*+50 % NP	± % to control
Lamina thickness, µ	242	264	+ 9.1	242	0.0	220	- 9.1
Upper epidermis thickness, µ	26	24	- 7.7	22	- 15.4	22	- 15.4
Lower epidermis thickness, µ	18	20	+ 11.1	20	+ 11.1	20	+ 11.1
Palisade tissue thickness, µ :	100	143	+ 43.0	114	+ 14.0	100	0.0
No. of palisade tissue layers	1	1	0.0	1	0.0	1	0.0
Spongy tissue thickness, µ	98	77	- 21.4	86	- 12.2	78	- 20.4
No. of spongy tissue layers	4	4	0.0	4	0.0	4	0.0
Midvein:							
Dimensions of midvein, µ :							
Length	501	559	+ 11.6	636	+ 27.0	694	+ 38.5
Width	649	744	+ 14.6	916	+ 41.1	1050	+ 61.8
Collenchyma below upper epidermis:							
Thickness	52	52	0.0	62	+ 19.2	42	- 19.2
No. of layers	2	2	0.0	2	0.0	2	0.0
Parenchyma below upper epidermis:							
Thickness	62	108	+ 74.2	144	+ 132.3	192	+ 209.7
No. of layers	2	3	+ 50.0	3	+ 50.0	4	+ 100.0
Collenchyma above lower epidermis:							
Thickness	22	24	+ 9.1	24	+ 9.1	24	+ 9.1
No. of layers	1	1	0.0	1	0.0	1	0.0
Parenchyma above lower epidermis:							
Thickness	172	200	+ 16.3	243	+ 41.3	283	+ 64.5
No. of layers	5	5	0.0	6	+ 20.0	6	+ 20.0
Midvein vascular bundle:							
Dimensions of midvein bundle, µ :							
Length	120	130	+ 8.3	137	+ 14.2	154	+ 28.3
Width	350	428	+ 22.3	583	+ 66.6	739	+ 111.1
Xylem thickness, µ	54	67	+ 24.1	74	+ 37.0	80	+ 48.2
Phloem thickness, µ	20	20	0.0	22	+ 10.0	24	+ 20.0
No. of xylem rows	20	22	+ 10.0	30	+ 50.0	38	+ 90.0
No. of xylem vessels/ row	4	5	+ 25.0	5	+ 25.0	6	+ 50.0
No. of glandular hairs / field	15	21	+ 40.0	27	+ 80.0	26	+ 73.3
Diameter of glandular hair, µ	79	83	+ 5.1	76	- 3.8	73	- 7.6

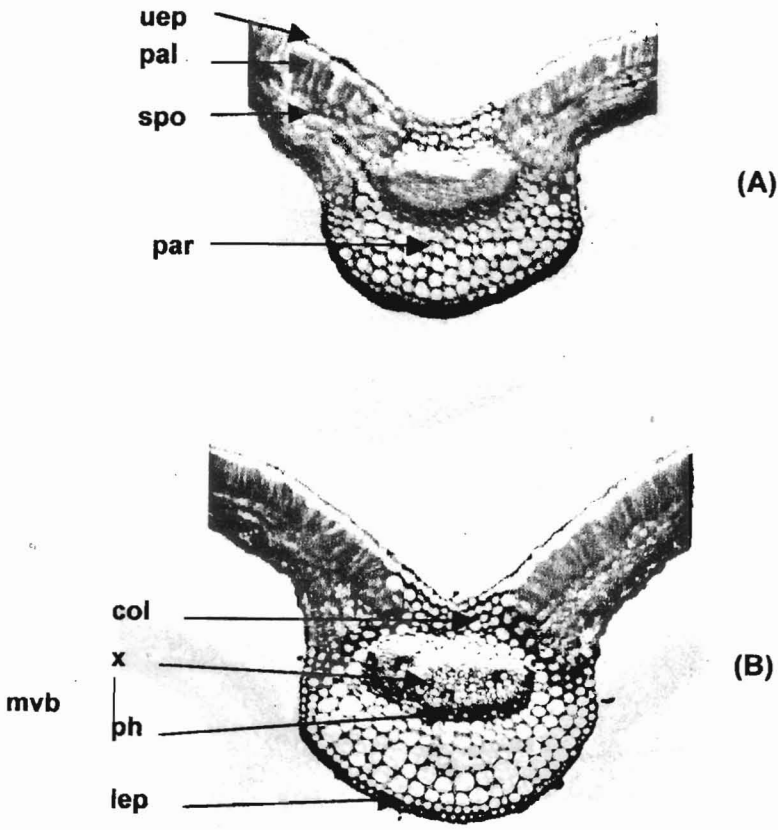

Fig. 2. Transverse sections of leaf blade of *Mentha piperita* plant through the midrib. (X40)

A) Untreated plant

B) Plant treated with NP

***Details*: col, colenchyma; lep, lower epidermis; mvb, midvein vascular bundle; pal, palisade tissue; par, parenchyma; ph, phloem; spo, spongy tissue; uep, upper epidermis; x, xylem.**

Cont.

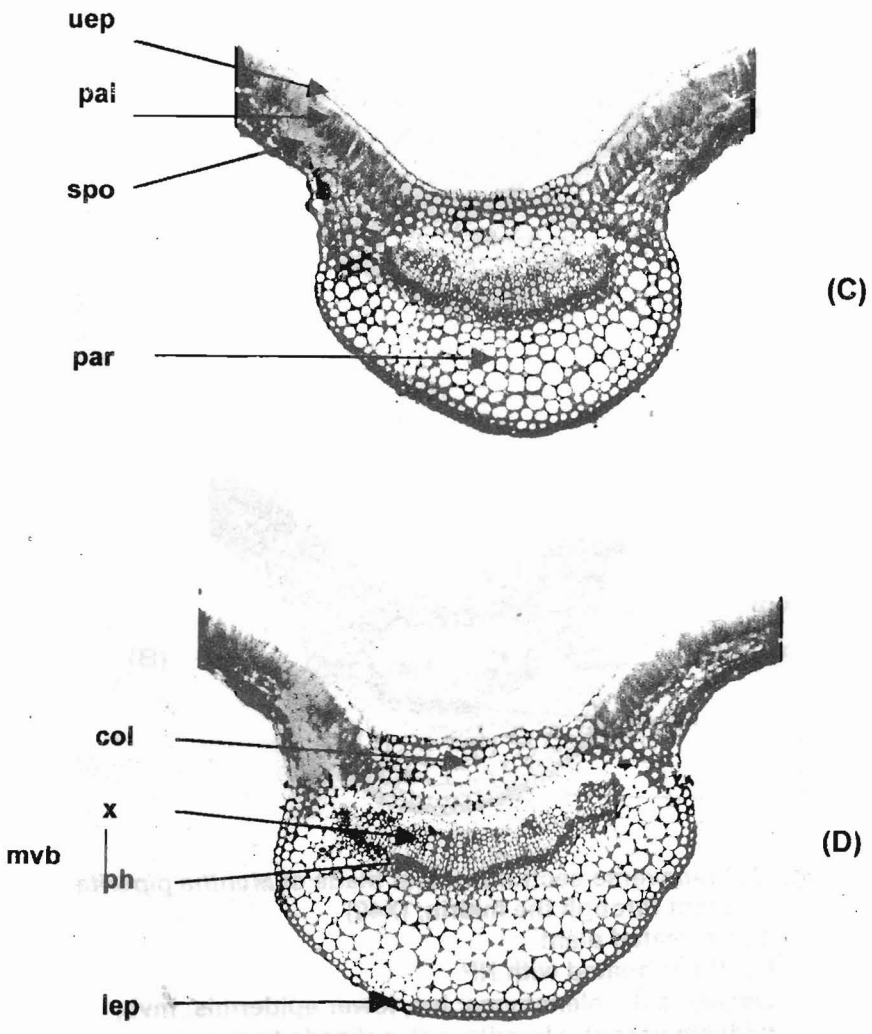

Fig. 2 - Cont. (X40)
C) Plant treated with *Rhizobium* + 50% NP
D) Plant treated with *Azotobacter* + 50% NP

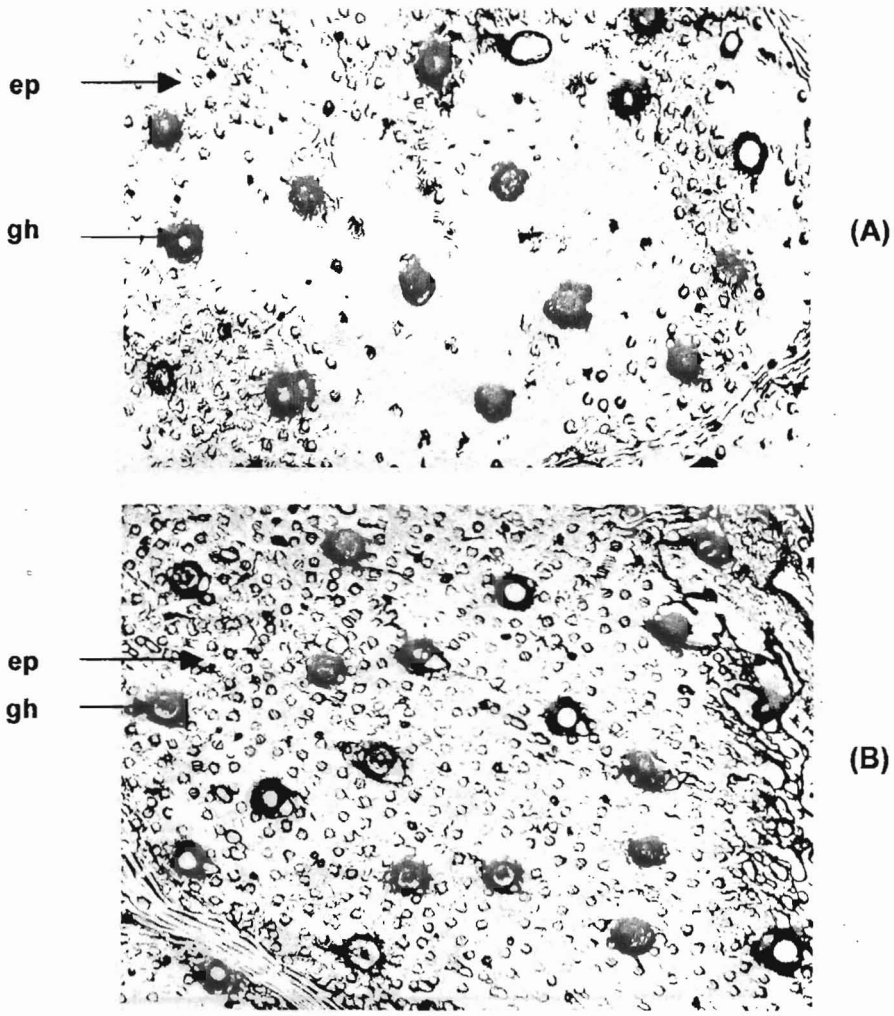

Fig. 3• The impressions of the lower epidermis of the 5th leaf blade of *Mentha piperita* plant. (X40)
A) Untreated plant
B) Plant treated with NP
***Details*: ep, epidermis; gh, glandular hair.**

Cont.

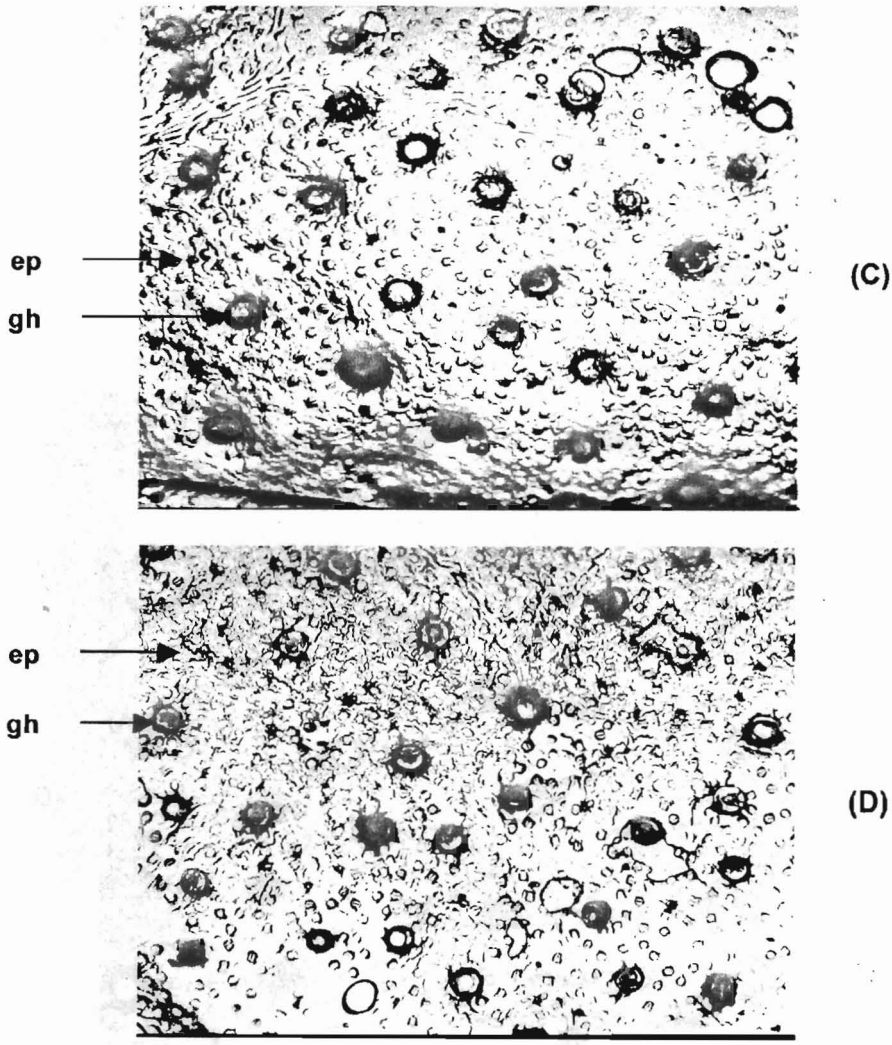

Fig. 3 • Cont. (X40)
C) Plant treated with *Rhizobium* + 50% NP
D) Plant treated with *Azotobacter* + 50% NP

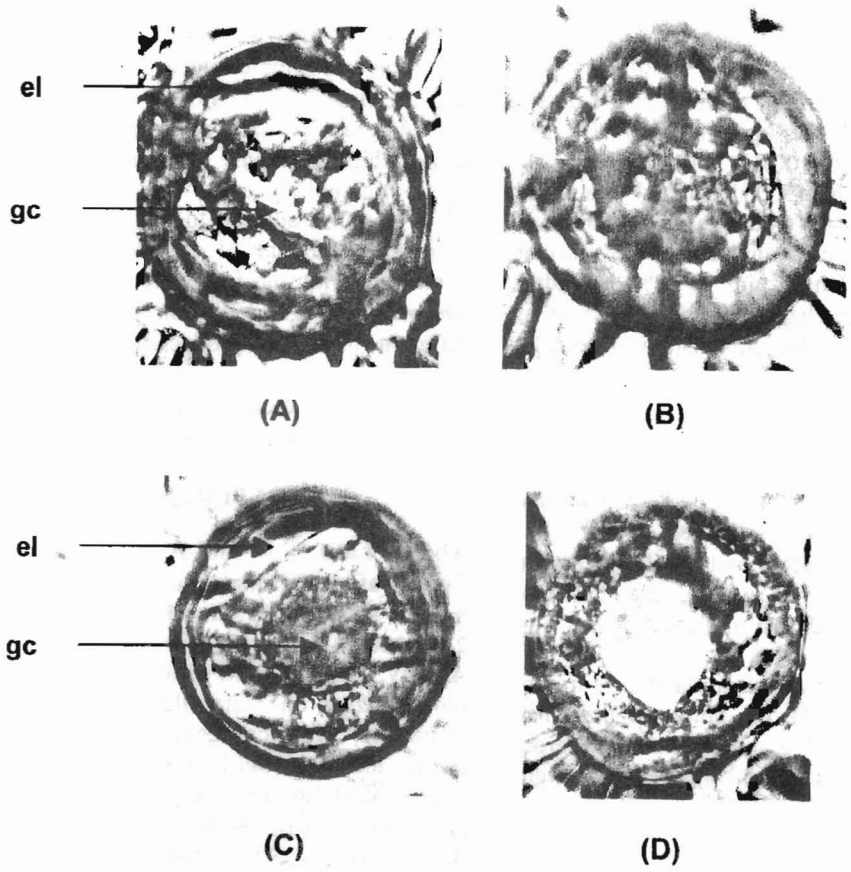

Fig. (4) Magnified glandular hairs showing the size of glands on the lower epidermis of *Mentha piperita* leaf blade. (X400)
A) Untreated plant
B) Plant treated with NP
C) Plant treated with *Rhizobium* + 50% NP
D)Plant treated with *Azotobacter* + 50% NP
***Details*: el, epithelial layer; gc, glandular cavity.**

parenchyma tissue above lower epidermis (41.3%) and its number of layers (20.0%) and dimensions of midvein vascular bundle (14.2% in length and 66.6% in width) over the control. While the increase in the midvein dimensions due to the second combination (*Azotobacter* + 50% NP) could be attributed to the increase in thickness of parenchyma tissue below upper epidermis (209.7%) and its number of layers (100%), collenchyma tissue above lower epidermis (9.1%), parenchyma tissue above lower epidermis (64.5%) and its number of layers (20%) and dimensions of midvein vascular bundle (28.3% in length and 111.1% in width) over the control. The characteristics of all vascular tissues shared in increasing dimensions of vascular bundles of bio-fertilizer combinations i.e., xylem tissue thickness (37.0 and 48.2%), phloem tissue thickness (10 and 20%) , number of xylem rows (50 and 90%) and number of xylem vessels/row (25 and 50%) over the control, in *Rhizobium* and *Azotobacter* inoculation, respectively.

The effect of chemical and bio-fertilizers on glandular hairs of *Mentha piperita* leaf blade (the lower epidermis) are shown in **Table (6)** and **Figures (3 and 4)**.

Regarding number of glandular hairs/ microscopic field (**Figure, 3**), the untreated plant had the lowest number (15 glandular hairs), whereas *Rhizobium* inoculation plus 50% NP had the highest one, followed by *Azotobacter* inoculation plus 50% NP, then NP treatment, being 80.0, 73.3 and 40.0% over the control.

On the other hand, diameter of glandular hair (**Figure, 4**) increased by 5.1% over the control as NP fertilizer was used, while such diameter was decreased due to using 50% NP in addition to *Rhizobium* or *Azotobacter* inoculation being 3.8 and 7.6% below the control, respectively.

It is clear from the above results that, both number and diameter of glandular hairs affected the oil yield of *Mentha piperita*, L. plant as the increase in diameter of glandular hair (83 μ) due to NP treatment compensated the shortage of glandular hairs number (21 glandular hairs). In addition, increasing number of glandular hairs as the bio-fertilizers *Rhizobium* or *Azotobacter* plus 50% NP were applied (27 and 26 glandular hairs, respectively) compensated the shortage of glandular hairs diameters of both treatments (76 and 73 μ, respectively).

CONCLUSION

It is clear from the previous results that, the enhancement oil yield of fresh herb as a result of treating *Mentha piperita* plants with *Rhizobium leguminosarum* bv . *phaseoli* or *Azotobacter chroococcum* biofertilizer in addition to 50% NP was correlated with increasing the vascular tissues of both stem and leaf blade, consequently improving water and soluble salts as well as food transfer into all plant organs. Moreover, these treatments also increased palisade tissue thickness in the leaf blade which is mainly responsible for photosynthesis of carbohydrates.

It could be advised to apply 50% of the recommended amount of chemical fertilizer (NP), combined with bio-fertilizers of *Azotobacter* or *Rhizobium* to get high oil yield insignificantly different than that produced by using the full amount of the recommended chemical fertilizers, but with much more oxygenated compounds and healthier environment.

REFERENCES

Abd-El Latif, T.A. and A.G. Salem (2002). Influence of bio-fertilizer and NPK rates on *Tagetes minuta*, L. **J. Agric. Sci., Mansoura Univ., 27(4): 2475-2487.**

Agamy, R.A. (2004). Effect of mineral and/or bio-fertilizers on morphological and anatomical characters, chemical constituents and yield of sweet fennel (*Foeniculum vulgare*, Mill. cv. Dulce) plants grown in calcareous soil. **Egypt. J. Appl. Sci., 19 (3): 55-75.**

AL-Qadasi, A.S.S. (2004). Effect of Bio-fertilization on *Ocimum basilicum*, L. Plant. pp. 115-119. M.Sc. Thesis , Fac. Agric., Cairo Univ., Egypt.

Bown, D. (1996). Encyclopedia of Herbs and Their Uses. pp. 311-314. Dorling Kindersley, London.

Chapman, H.D. and P.F. Pratt (1961). Methods of Soil, Plants and Water Analysis. pp. 60-69. Univ. Calif., Division of Agric. Sci.

Dubois, M.; K.A. Gilles; J.K. Hamilton; P.A. Rebors and F. Smith (1956). Colourimetric method for determination of sugars and related substances. **Annal. Chem., 28 (3): 350-356.**

El-Khyat, A.S. and R.A. Zaghloul (1999). Bio-fertilization and organic manuring efficiency on growth and yield of caraway plants (*Carum carvi*, L.). **Annals of Agric. Sc., Moshtohor, 37(2): 1379-1397.**

El-Sgai, M.U. and A.M. Sabbour (2000). Comparative studies on the anatomical structure of stem and leaf of three genera of Lamiaceae. **J. Agric. Sci., Mansoura Univ., 25 (6): 3215–3229.**

Hassan, H.R.; Nassar Dalia, M.A. and M.H.A. Abou Bakr (2006). Effect of mineral and bio-fertilizers on growth, yield components, chemical constituents and anatomical structure of moghat plant (*Glossostemon bruguieri*, Desf.) grown under reclaimed soil conditions. **J. Agric. Sci. Mansoura Univ., 31 (3): 1433-1455.**

Jackson, M.L. (1967). Soil Chemical Analysis. pp. 144-197. Printice-Hall, India.

Kandeel, A.M.; Naglaa, S.A. and A.A. Sadek (2002). Effect of bio-fertilizers on the growth, volatile oil, yield and chemical composition of *Ocimum basilicum*, L. plants. **Annals Agric., Sci., Ain Shams Univ., Cairo, 47 (1):351-371.**

Lazarovits, G. and J. Nowak (1997). Rhizobacteria for improvement of plant growth and establishment. **Hort. Sci., 32 (2): 188-192.**

Maheshwari, S.K.; S.K. Gangrade and R.K. Shama (1995). Differential responses of *Azotobacter* and nitrogen on biomass and oil yield of palmarosa. **Crop Res. Hisar., 10 (3): 356-359.**

Marschner H. (1995). Mineral Nutrition of Higher Plants, 2nd Ed, pp 465-466. Academic Press, Harcourt Brace and Co. Publishers. New York.

Medani, R.A.; S.A. Mohamed; M.A. El-Yazal, and S.A. Mahfouz (2000). Growth, yield and chemical composition of sugar beet (*Beta vulgaris*, L.) plants as affected by specific isolated biofertilizers in relation to nitrogen application. **Annals Agric. Sci., Moshtohor, 38(4): 2019-2038.**

Nassar, M.A. and K.F. El-Sahhar (1998). Botanical Preparations and Microscope Analysis (Microtechnique), pp. 43-67 & 90-126. Academic Bookshop, Dokki, Cairo, Egypt (in Arabic).

Nornai, R. (1982). Formula for determination of chlorophyll pigments extracted with N.N. dimethyl formamide. **Plant Physiol., 69:1371-1381.**

Omar, M.N.A.; M.H. Hegazy; R.A. Abd El-Aziz; M.S.M. Abo Soliman and M.M. Sobh (1991). Effect of inoculation with rhizobacteria on yield of wheat under graded level of nitrogen fertilization. **Annals Agric. Sci., Ain Shams Univ. Cairo, 36 (1): 99-104.**

Pandey, B.P. (1982). Taxonomy of Angiosperms (Systematic Botany). pp. 414-422. Chand & Company LTD. Ram Nagar, New Delhi.

Piper C.S. (1947). Soil and Plant Analysis. pp. 258 - 275. Univ. of Adelaide, Adelaide, Australia.

Pregl P. (1945). Quantitative Organic Micro-analysis, 4th Ed., pp. 78-85. Churchill Publishing Co., London.

Ragab, A.A. and M.H. Rashad (2003). Enhancement of nutrients uptake and metabolism efficiency of sorghum (*Sorghum bicolor*, L.) plants by biofertilizers under water stress. **Zagazig J. Agric. Res., 30 (1):147-166.**

Ramadan, B. S. H.; H. R. Hassan and F. A. Abdo (2003). Effect of minerals and bio-fertilizers on photosynthetic pigments, root quality, yield components and anatomical structure of sugar beet (*Beta vulgaris*, L.) plants grown under reclaimed soils. **J. Agric. Sci. Mansoura Univ., 28 (7): 5139-5160.**

Sakr-Weaam R.A. (2001). Effect of Some Organic and Inorganic Fertilizers on *Mentha piperita*. pp. 37 and 126–134. M.Sc. Thesis, Fac. Agric., Cairo Univ, Egypt.

Salah, E.A.; T.H. Nokhal; M.A. Borollosy; I. Fendrik; M.S. Sharaf and M. El-Sawy (1998): Effectiveness of dual inoculation with diazotrophs and vesicular arbuscular mycorrhizae on growth and medicinal compounds of *Datura stramonium*. **Arab Univ. J. of Agric. Sci., 6 (2): 343-355.**

Sass, J. E. (1967). Botanical Microtechnique. 3rd Ed. pp. 5-77. Oxford and IBH Publishing Co., Calcutta, New Delhi, India.

Snedecor, G.W. and W.G. Cochran (1982). Statistical Methods. pp. 507. The Iowa State Univ. Press., Aonas, Iowa, USA.

Subba Rao, N.S. (1984). Bio-fertilzers in Agriculture. 3rd Ed, pp. 1-13, 83, 132 & 153-165. Oxford and IBH Publishing Co., New Delhi, India.

Swaefy-Hend, M.F.A. (1996). Effect of Chemical Fertilization and Some Trace Elements on Growth and Productivity of *Mentha piperita* L. Plant. pp. 31-37. M.Sc. Thesis, Fac. Agric., Cairo Univ., Egypt.

Swaefy-Hend, M.F.A. and S.M.N. Milad (2006). Effect of chemical and bio-fertilization on growth and chemical composition of *Euryops pectinatus*. **J. Agric. Sci. Mansoura Univ., 31(11): 7329-7345.**

حوليات العلوم الزراعية
جامعة عين شمس، القاهرة
مجلد(٥٢)، عدد (٢)، ٤٦٥-٤٨٤، ٢٠٠٧

تأثير بعض الأسمدة الكيماوية والمخصبات الحيوية على نباتات النعناع الفلفلي النامية في أرض رملية

٢- التأثير على محصول الزيت الطيار و التركيب الكيماوي للنبات و الخصائص التشريحية

[٣٨]

هند مصطفى فهمى سويفى[١] - وئام رمضان عرفة صقر[١] - عاطف زكريا سبع[٢] - عاطف عبد العزيز رجب[٣]

١- قسم بساتين الزينة - كلية الزراعة - جامعة القاهرة - الجيزة - مصر

٢- قسم النبات الزراعى - كلية الزراعة - جامعة القاهرة - الجيزة - مصر

٣- قسم الميكروبيولوجيا الزراعية- معهد بحوث الأراضي والمياه والبيئة- مركز البحوث الزراعية- الجيزة- مصر

أجريت هذه الدراسة بمشتل التجارب بقسم بساتين الزينة، كلية الزراعة، جامعة القاهرة، الجيزة، خلال الموسمين المتتاليين ٢٠٠٥ و ٢٠٠٦. وذلك بهدف دراسة تأثير الأسمدة الكيماوية النتروجينية والفوسفاتية مع أو بدون التلقيح ببعض السلالات الميكروبية التالية *Rhizobium leguminosarum* bv. *phaseoli*, *Azotobacter chroococcum* and *Bacillus megatherium* var. *phosphaticum* على كلا من محصول الزيت و التركيب الكيماوي و الخصائص التشريحية لنباتات النعناع الفلفلي النامية فى تربة رملية. أظهرت النتائج المتحصل عليها أن استخدام ١٧٥ كجم/ فدان سلفات أمونيوم و ١٠٠كجم/ فدان سوبر فوسفات الكالسيوم بالإضافة إلى التلقيح بالأزوتوباكتر أدى إلى الحصول على أعلى محصول كلي للزيت الطيار للنبات و للفدان و يليها المعاملة بنفس الكميات من الأسمدة الكيماوية مع التلقيح بالريزوبيم.

كذلك وجد أن الزيادة في محصول الزيت الطيار للعشب الأخضر الطازج للنباتات المعاملة بالريزوبيم أو الأزوتوباكتر بالاضافة إلى ٥٠% من التسميد الكيماوي الموصى به كانت مرتبطة مع زيادة الأنسجة الوعائية في كل من الساق ونصل الورقة وكذلك زيادة سمك النسيج العمادي في نصل الورقة.

أدى استخدام ٥٠% من التسميد الكيماوي مع التلقيح بأي من المخصبات الحيوية المستخدمة في الدراسة إلى زيادة المحتوى الكلى للكربوهيدرات بالمقارنة بالنباتات التي تم تسميدها ب ٥٠% من السماد الكيماوي فقط.

تحكيم: ا.د عواض محمد قنديل
ا.د جلال الدين علي النجدي