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Effect of organic and biofertilizers on growth, herb yield and volatile oil of marjoram plant grown in Ajloun region, Jordan

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This experiment was carried out during two successive seasons of 2007/2008 and 2008/2009 in Ajloun region, to study the effect of organic manure (farmyard manure and poultry manure at a rate of 75 m³/ha as fertilizer of each) and biofertilizers (Nitroboein and Halex-2 at a rate of 988 gm/ha of each as well) as their interactions on growth, yield of herb and volatile oil of *Majorana hortensis* L., using drip irrigation system. The results indicated that the application of a poultry manure as an organic fertilizer to marjoram plants recorded the maximum values of herb fresh and dry yield, N, P and K contents and its uptake by herb in the early cut, volatile oil percentage, oil yield/ plant and per hectare, while in the late cut, the highest values in this respect were obtained by farmyard manure application. Marjoram transplant treated with halex-2 biofertilizers gave the highest values of herb fresh and dry yield, N, P and K contents and its uptake by herb in the early cut, in addition to, volatile oil percentage, oil yield/ plant per hectare, while plants treated with nitroboein biofertilizers gave intermediate values at different cut in this respect. The combination of poultry manure and halex-2 proved to be the superior treatments, which reflected in the greatest influence upon all the studied characters, that is, fresh and dry herb yields, mineral contents, highest volatile oil percentage and oil yield/ plant of marjoram plant, followed by FYM combined with the same biofertilizers and may be recommended under similar conditions of this work.

Key words: Biofertilizers, Jordan, halex-2, marjoram, nitroboein, organic, poultry manure, volatile oil.

INTRODUCTION

Marjoram (*Majorana hortensis*, L.), is a perennial herbaceous plant belonging to Lamiaceae family. It has been cultivated in the Mediterranean countries and is still widely cultivated today (Ietswaart, 1980; Verma et al., 2010). It is one of the most commercially important medicinal plants. The active principles are found chiefly in the aerial. The drug has agreeable spicy smell and taste. It is a popular flavoring used to flavor meat dishes and in

making salamis and other sorts of sausages. It contains up to 2% of essential oil (Baratta et al., 1998). The active principles in the drug oil help in digestion, relieve flatulence, act as an intestinal antispasmodic and stimulate the secretion of bile (Frantisek and Voclav, 1975). The major components of Marjoram oil are α -Pinene, β -Pinene, limonene, 1,8 cineole, linalool, terpinene-4-ol, α -Terpinene, Linalyl/acetate and eugenol (Demetzos and Perdetzoglou, 2001; Juliano et al., 2000). Some of these components are used for scented cosmetics and others are used for flavoring pharmaceuticals such as D-limonene and Linalool

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(Refaat, 1988).

Organic manures and biofertilization are very important for medicinal and aromatic plants to produce the best product in both quantity and quality and it is also safe for human, animal and the environment. El-Ghadban et al. (2002) reported that plants amended with the highest level of compost (37 m³/ha.) either alone or in conjugation with a mixture of N₂-fixing bacteria showed increments in plant growth characters on marjoram plants. Eid and El-Ghawwas (2002) stated that, plant height, number of branches/plant as well as fresh and dry weights of marjoram plants were significantly increased when plants were treated with microbein and nitrobein compared with untreated plants. In addition, Mahfouz (2003) found that, the highest values of *Majorana* plant height, number of branches, fresh and dry weights of root were recorded for the treatment of biofertilizer plus full dose of N and P than the other treatments and control plants. Moreover, Dewidar (2007) found that the combination between compost and biofertilizers increased plant fresh and dry weights in the fourth cut compared to the other treatments of marjoram plants.

The different organic fertilization treatments tended to increase the contents of N, P and K in the plant herb (Kandeel and Abou Taleb, 2002; Moheesn, 2002; El-Sayed et al., 2002) on sweet basil. Kandeel and Sharaf (2003) on marjoram plants, stated that, the highest oil percentage and oil yield/ ha were obtained with plants inoculated pre-sowing with three bacterial partners (biological fertilizers) and half of the recommended field rate of the inorganic N, P and K fertilization. Abo (2008) found that the highest values of the herb fresh and dry weight, and yield of oil/plant of marjoram were obtained when applied to poultry manure combined with phosphorein or yeast biofertilizers at 25 m³ ha.

The aim of this work was to study the effects of the application of organic manures (farmyard manure or poultry) and biofertilizer (nitrobein and halex-2) on the vegetative growth, oil yield and chemical composition of marjoram (*Majorana hortensis*, L.).

MATERIALS AND METHODS

This experiment was carried out during two successive seasons of 2007/2008 and 2008/2009 in Ajloun region, to study the effect of organic manure and biofertilizers as well as their interactions on the growth, yield of herb and volatile oil of *Majorana hortensis*, L. grown in sandy soil, using drip irrigation system.

This experiment included six treatments, which were the combinations of two organic manures, that is, farmyard manure (FYM) and poultry manure, and two biofertilizers, that is, nitrobein and halex-2 beside control. These treatments were arranged in a split plot in a complete randomized block design with three replicates. Organic fertilizers were randomly arranged in the main plots and biofertilizers were randomly distributed in the sub plots.

Seeds of marjoram were obtained from International Center for Agricultural Research in the Dry Areas (ICARDA). On October 1st from each season, the seeds of marjoram were sown on the foam trays (216 eyes) containing peat moss: vermiculite (1:3 by volume).

On December 1st from each season, uniform seedlings of about 15 cm length were transplanted in the experimental area at 25 cm apart. The experimental unit area was 12.6 m². It contains three dripper lines with 6 m length each and 60 cm distance between each dripper lines. Organic manure was added during the soil preparation before planting at a rate of 75 m³/ha for both FYM and poultry manure. The nitrobein contains *Azotobacter*, a nitrogen fixing bacteria; Halex-2 contains a mixture of N-fixing bacteria of the genera *Azotobacter*, *Azospirillum* and *Kelbsiella*. The nitrobein and Halex-2 were added at a rate of 400 g per hectare of each. Both biofertilizers were dissolved in 5 l water and little Arabic gum (20%) as adhesive agent was used and the roots of transplants were dipped for 3 m in this suspension before transplanting.

Data recorded

The following data were recorded at harvesting stages for two cuts at 1 May and 5 October for the first season and at 10 May and 20 October for the second season (after 75% of flowering). Harvesting the plants was done at 15 cm above soil surface. Random samples of ten plants from each sub plot in both cuts were taken and the following parameters were determined.

Vegetative growth

For vegetative growth of plants, there were changes in Plant height (cm), number of branches/ plant, herb fresh and dry weight / plant (g).

Nitrogen, phosphorus and potassium content and its uptake

The dry weight of herb was finely ground and wet digested with sulfuric acid and perchloric acid (3:1). Nitrogen, phosphorus and potassium were determined as dry weight basis according to the methods described by Bremner and Mulvaney (1982), Olsen and Sommers (1982) and Jackson (1970), respectively. Uptake of N, P and K were calculated.

Oil percentage in air-dried herb

The volatile oil from air-dried herb of marjoram plant was isolated by water distillation, using 25 g dried herb according to Balbaa et al. (1981), while essential oil yield/plant (ml/100 g) was determined according to Güenther (1972). The oil percentage (%) of plant dry weight (air dried) and the estimated essential oil yield/ ha was thus calculated.

Statistical analysis

The data were subjected to proper statistical analysis of variance according to Snedecor and Cochran (1980) and means separation were done according to New L.S.D. at 5 % level of probability.

RESULTS AND DISCUSSION

Vegetative growth

Effect of organic fertilizers

Plant height, number of branches of marjoram, was

Table 1. Chemical properties of the soil before transplanting and farm yard manure (FYM) as well as poultry manure during 2007/2008 and 2008/2009 seasons.

Parameters (%)	Soil		FYM		Poultry manure	
	1 st season	2 nd season	1 st season	2 nd season	1 st season	2 nd season
O M	0.06	0.07	13.18	13.57	61.00	59.18
Total N	4.23	3.98	0.76	0.79	2.47	2.29
Total P	3.71	4.27	0.12	0.13	0.23	0.22
Total K	11.36	10.18	0.63	0.66	0.99	0.97

Table 2. Effect of organic sources and biofertilizers and their interaction on plant height (cm) and number of branches /plant of marjoram plants during 2007/2008 and 2008/2009 seasons.2

Treatments	Plant height (cm)				Number of branches /plant			
	1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
Effect of organic fertilizer								
Farmyard manure	45.82	45.74	44.00	42.08	31.24	60.64	31.25	55.87
Poultry manure	49.53	43.21	49.71	40.82	37.19	51.51	40.81	46.12
New LSD at 0.05	3.06	NS	2.57	NS	2.20	2.46	2.79	3.43
Effect of biofertilizers								
Without	45.08	42.45	42.93	39.65	24.26	52.99	29.29	47.22
Nitrobein	47.94	44.51	47.80	39.80	33.59	54.51	35.55	51.06
Halex-2	50.42	46.47	49.83	44.89	44.79	60.73	43.25	54.71
New LSD at 0.05	1.58	1.53	1.32	1.25	1.13	1.27	1.44	1.77

significantly affected by addition of organic fertilizer source in the two cuts in both seasons, except plant height at the second cut in both seasons. It is clear from the data in Table 2 that fertilization of marjoram plants with poultry manure resulted in the maximum values of 49.53 and 49.71 cm for plant height and 37.19 and 40.81 cm for number of branches in the 1st cut, while application of farmyard manure recorded 60.64 and 55.87 cm for number of branches in 2nd cut in the first and second seasons, respectively (El-Ghadban et al., 2002). Therefore, it could be concluded that poultry manure had a favorable effect on vegetative growth in the early growing season, while farmyard manure had a favorable effect on the late growing season. Farmyard manure is generally applied in large amounts due to the long period required for mineralization of nutrients into plant available forms.

Organic fertilization is a very important method of providing plants with their nutritional requirement without having an undesirable impact on the environment (El-Sayed et al., 2002). The increase in marjoram plant height due to application of poultry manure might be attributed to the effect of organic fertilizer that improves physical, chemical, and biological properties of soil; that is, increasing soil organic matter, cation exchange

capacity, water holding capacity and availability of mineral nutrients and, this in turn, increases plant height. These results are in harmony with those obtained by El-Ghadban (1998) and Abo (2008) on marjoram. They indicated that, fertilization with organic manures at the highest rate increased plant growth.

Effect of biofertilizers

It is clear from the data in Table 2 that the sources of biofertilizers had a significant effect on plant height and number of branches/plant in both seasons. Application of biofertilizers as halex-2 gave the highest values in this respect, compared to the other treatments. The increment in vegetative growth due to biofertilizers application might be due to the vital role of bacteria present in the applied biofertilizer and capable of contributing some hormone substances, that is, gibberellins, auxins and cytokinins (Cacciari et al., 1989). These phytohormones may stimulate the cell elongation and development and hence plant growth (Paleg, 1985). These results agreed with those obtained by Abo El-Ala (2002), Kandeel and Sharaf (2003) and Mahfouz (2003) on marjoram. They reported that, the highest values of

Table 3. Effect of organic sources and biofertilizers and their interaction on plant height (cm) and number of branches /plant of marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	Plant height (cm)				Number of branches /plant			
		1 st season		2 nd season		1 st season		2 nd season	
		1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
FYM	Without	42.67	43.18	41.42	40.12	21.41	62.22	23.20	55.97
	Nitrobein	46.84	45.91	44.33	39.94	30.68	56.65	30.68	54.20
	Halex-2	48.78	48.10	46.23	46.16	41.63	63.08	39.88	57.44
Poultry manure	Without	47.49	41.71	44.44	39.19	27.12	43.76	35.38	38.48
	Nitrobein	49.03	43.11	51.26	39.65	36.50	52.38	40.42	47.92
	Halex-2	52.05	44.83	53.43	43.61	47.96	58.37	46.63	51.98
New LSD at 0.05 level		2.74	2.65	2.30	2.17	1.97	2.20	2.50	3.07

Table 4. Effect of organic sources and biofertilizers on herb fresh weight of marjoram plants during 2007/2008 and 2008/2009 seasons.

Treatments	Herb fresh weight							
	(g/plant)				Ton/ha			
	1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
Effect of organic fertilizer								
FYM	83.43	59.98	87.68	75.53	5.772	4.148	6.066	5.226
Poultry manure	108.53	50.46	112.69	52.20	7.509	3.491	7.796	3.612
New LSD at 0.05 level	3.75	3.44	3.79	2.90	0.105	0.096	0.106	0.081
Effect of biofertilizers								
Without	76.87	49.72	85.14	54.91	5.317	3.439	5.890	3.798
Nitrobein	96.42	54.53	101.13	62.69	6.671	3.773	6.998	4.336
Halex-2	114.64	61.41	114.30	74.00	7.932	4.247	7.907	5.120
New LSD at 0.05 level	1.94	1.77	1.95	1.50	0.054	0.049	0.054	0.042

plant height and number of branches, were recorded for the treatment of biofertilizers in comparison to the other treatments (without biofertilizers) plants.

The results of the interaction between organic manure and biofertilizers are shown in Table 3. The results indicated that plant height and number of branches/plant were significantly increased by application of organic manure and biofertilizers. Addition of poultry manure to marjoram plant combined with halex-2 biofertilizers recorded the maximum values of plant height and branches number/plant in the 1st cut in both seasons, while application of FYM conjugation with halex-2 gave the highest values of these parameters in the 2nd cut in both seasons.

Herb fresh weight

Effect of organic fertilizers

Presented data in Table 4 shows that herb fresh weight/plant and hectare were significantly increased by

application of organic manure in the 1st and 2nd cut in both seasons. Application of poultry manure as an organic manure recorded 108.53 and 112.69 g/plant; 7.509 and 7.796 ton/ha in 1st cut in the first and second seasons, respectively. While application of organic manure as the source of farmyard manure significantly increased herb fresh weight /plant and hectare in the 2nd cut and recorded 59.98 and 75.53 gm/plant , 4.148 and 5.226 ton/ha during the first and second seasons, respectively .

It could be concluded that the increment in plant fresh weight may be attributed to the increase in both plant height and number of branches/plant as already discussed.

Effect of biofertilizers

Data in Table 4 illustrate that, treatment of marjoram transplants with biofertilizers had a significant effect on herb fresh weight in both seasons. The use of halex-2

Table 5. Effect of interaction between organic sources and biofertilizers on herb fresh weight of marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	Herb fresh weight							
		(g/plant)				Ton/ha			
		1 st season		2 nd season		1 st season		2 nd season	
		1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
FYM	Without	67.25	56.01	69.95	66.07	4.652	3.874	4.840	4.571
	Nitrobein	81.36	58.41	86.80	73.64	5.629	4.040	6.004	5.095
	Halex-2	101.67	65.52	106.30	86.89	7.035	4.534	7.353	6.012
Poultry manure	Without	86.50	43.43	100.34	43.76	5.984	3.004	6.943	3.027
	Nitrobein	111.47	50.65	115.46	51.73	7.712	3.504	7.989	3.578
	Halex-2	127.63	57.30	122.29	61.11	8.831	3.963	8460	4.228
New LSD at 0.05 level		3.36	3.08	3.39	2.60	0.094	0.086	0.095	0.073

biofertilizers resulted in corresponding increase in herb fresh weight/ plant and per hectare, and recorded the maximum values 7.932 and 4.247 in the 1st and 2nd cut in the 1st season, 7.907 and 5.120 ton/ha in the 1st and 2nd cut in the 2nd season followed by nitrobein biofertilizers. This might be related to the favorable effects of halex-2 biofertilizers on vegetative growth (Table 2).

Biofertilizers are microbial inoculates used for application to either seed or soil for increasing soil fertility with the objective of increasing the number of such microorganisms and to accelerate certain microbial processes in the rhizosphere of inoculated plants or soil. Such microbiological processes can change unavailable forms of nutrients into available ones that can be easily assimilated by plants, and then increased herb fresh weight of marjoram (Subb, 1993). It is obvious from the data in Table 5 that interaction between organic manure and biofertilizers had a significant effect on herb fresh weight in the 1st and 2nd cut in both seasons. Poultry manure combined with halex-2 biofertilizers gave higher herb fresh weight and recorded maximum values of herb fresh weight/plant per hectare in the 1st cut in both seasons of the study, while farmyard manure combined with the same biofertilizers recorded the highest values in the 2nd cut in both seasons. Organic manure, combined with nitrobein biofertilizers, gave intermediate values in both seasons at different date of cut. On the other hand, the lowest values in the respect were obtained when the organic manure was added only. The results also indicated that the effect of organic manure sources and biofertilizers was greater and more pronounced when added in combination with biofertilizers than added alone. Therefore, a combination of organic manure and biofertilizers is suggested to be used in order to improve the fresh weight of marjoram plant.

The positive effect of the previous treatments on the fresh weight could be explained through its stimulatory effect on producing taller plants with more branches and

heavier fresh weights of marjoram plants. Moreover, the increase in fresh weight could be explained by increasing metabolic activities of the plant under the effect of the interaction between the bio and organic fertilizers that gave significant values for fresh weight in the two seasons.

Herb dry weight

Effect of organic fertilizer

It is clear from the data in Table 6 that the sources of organic fertilizer had a significant effect on the herb dry weight and per hectare in the two cuts of the plant during the experimental seasons. Moreover, application of poultry manure gave the highest values in this respect (2.244 and 2.920 ton/ha) in 1st and 2nd seasons at the 1st cut compared to the other treatments. While in the 2nd cut the highest values in this respect (2.478 and 2.238 ton/ha) in the 1st and 2nd seasons were obtained by application of organic manure as the sources of farmyard manure.

It could be concluded that the increment in plant dry weight may be attributed to the increase in plant height, number of branches/plant and plant fresh weight as already discussed. These results are in agreement with those results obtained by Eid and El- Ghawwas (2002) and Abo- (2008) on marjoram plants.

Effect of biofertilizers

Data in Table 6 showed that, treatment of marjoram transplants with biofertilizer significantly increased herb dry weight/ plant and per hectare and gave the same trend of herb fresh weight/plant and per hectare. The optimum values for herb dry weight were recorded for

Table 6. Effect of organic sources and biofertilizers on herb dry weight of marjoram plants during 2007/2008 and 2008/2009 seasons.

Treatments	Herb dry weight							
	(g/plant)				Ton/ha			
	1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
	Effect of organic fertilizer							
FYM	28.08	35.82	28.82	32.34	1.942	2.478	1.994	2.238
Poultry manure	32.42	21.00	42.20	21.11	2.244	1.452	2.920	1.460
New LSD at 0.05 level	2.71	3.37	2.96	3.31	0.076	0.095	0.082	0.092
	Effect of biofertilizers							
Without	23.11	24.74	31.72	21.15	1.598	1.712	2.194	1.462
Nitrobein	30.71	27.77	35.50	26.06	2.125	1.922	2.456	1.803
Halex-2	36.93	32.73	38.82	32.97	2.555	2.263	2.686	2.280
New LSD at 0.05 level	1.40	1.74	1.53	1.71	0.039	0.049	0.042	0.047

Table 7. Effect of interaction between organic sources and biofertilizers on herb dry weight of marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	Herb dry weight							
		(g/plant)				Ton/ha			
		1 st season		2 nd season		1 st season		2 nd season	
		1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
FYM	Without	21.75	32.84	23.42	25.30	1.505	2.273	1.621	1.750
	Nitrobein	27.98	33.36	29.11	32.29	1.935	2.308	2.014	2.233
	Halex-2	34.51	41.25	33.92	39.43	2.388	2.854	2.347	2.728
Poultry manure	Without	24.46	16.63	40.02	17.00	1.692	1.151	2.770	1.176
	Nitrobein	33.43	22.17	41.88	19.83	2.312	1.534	2.899	1.371
	Halex-2	39.35	24.20	43.72	26.50	2.723	1.675	3.024	1.833
New LSD at 0.05 level		2.43	3.02	2.65	2.96	0.068	0.085	0.074	0.083

halex-2 biofertilizers (2.555 and 2.263ton/ha) in the 1st and 2nd cut in the 1st season (2.686 and 2.280 ton/ha) in the 1st and 2nd cut in the 2nd season. While nitrobein biofertilizers gave intermediate values in both seasons at different cut. It could be concluded that the increment in plant dry weight may be attributed to the increase in both plant height, number of branches/plant and plant fresh weight. These results agreed with those obtained by Abo-El-Ala (2002), Kandeel and Sharaf (2003), Mahfouz (2003) and Abo- (2008) on marjoram. They reported that, the highest values of plant height, number of branches, herb fresh and dry weights were recorded for the treatment of biofertilizer in comparison to the other treatments (without biofertilizers) plants.

The effect of interaction between organic and biofertilizers on herb dry weight of plant and per hectare

are presented in Table 7. It is clear that, the interaction between organic and biofertilizers had a significant effect on herb dry weight of plant and per hectare. Poultry manure combined with halex-2 biofertilizers gave higher herb dry weight and recorded maximum values of herb fresh weight / plant and per hectare (2.723 and 3.024 ton/ha) in the 1st cut in the 1st and 2nd seasons, while farmyard manure combined with the same biofertilizers recorded the highest values of 2.854 and 2.728ton/ha in the 2nd cut in the 1st and 2nd seasons. Organic manure, combined with nitrobein biofertilizers gave intermediate values in both seasons at different dates of cut. On the other hand, the lowest values in the respect were obtained when the organic manure was used only. The optimum stimulation effect which was recorded for poultry manure may be due to its high content of essential

Table 8. Effect of organic sources and biofertilizers on nitrogen, phosphorus and potassium contents (%) of marjoram plants during 2007/2008 and 2008/2009 seasons.

Treatments	N (%)				P (%)				K (%)			
	1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
Effect of organic fertilizer												
FYM	2.15	2.26	2.44	2.45	0.229	0.228	0.236	0.228	1.64	1.82	1.62	1.94
Poultry manure	2.64	2.51	2.60	2.50	0.258	0.243	0.246	0.247	2.35	2.38	2.43	2.29
New LSD at 0.05 level	0.13	0.06	0.08	NS	NS	NS	NS	NS	0.08	0.19	0.19	0.10
Effect of biofertilizers												
Without	2.17	2.37	2.38	0.23	0.220	0.223	0.223	0.223	1.76	1.91	1.96	2.17
Nitrobein	2.50	2.55	2.52	0.24	0.233	0.242	0.245	0.245	2.18	2.01	2.09	2.50
Halex-2	2.48	2.65	2.54	0.26	0.254	0.260	0.245	0.245	2.36	2.14	2.28	2.48
New LSD at 0.05 level	0.07	0.03	0.04	NS	0.028	0.023	NS	NS	0.04	0.09	0.09	0.05

macro-elements (N, P, K) comparing with the farmyard manure as shown in Table 1. These nutrients are important constituents of the macronutrients in the soil and are generally determined before the site is distributed in order to complete a site reclamation plan. This in turn, increases herb dry weight. In this concern, it appears that using biofertilizers in this study could supply marjoram plants with their need nutrients by cheap means and leads to a significant decrease in the production costs and the pollution rates in the soil, water and air. This indicated that, organic and biofertilizers have positive alternative to chemical one. They are very safe for humans and animals, and reduce pollution of environment. Dewidar (2007) and Abo (2008) found that the combination between organic and biofertilizers increased plant fresh and dry weights of marjoram plants.

N, P and K (%)

Effect of organic fertilizer

Data in Table 8 showed that N, P and K (%) in herb of marjoram proved a significant response with using poultry manure as an organic fertilizer at different cut in both seasons, except N (%) in the 2nd cut in the 2nd season and P (%) in different cutting stages in both seasons. Poultry manure were the superior source, which gave the highest values of N, P and K content of marjoram plants at different cutting stages in both seasons. Moreover the highest concentrations of N and K in the herb were obtained in the 1st cut in both seasons. The increment in N, P, K contents in different cuts of herb tissues of marjoram plants may explain the efficiency of suitable quantity of organic fertilizers that can attract and hold nutrients and water on its surface to supply the plants with suitable amounts for a longer time and may be due to release of some nutrients; such as, Fe, Zn and Mn through the breakdown of organic manure in the soil and

makes these elements in available forms and this in turn improves N, P, K and this reflects a beneficial effect on growth and dry weight. Moreover, the increments in NPK as a result of organic fertilizers application may be attributed to their favorable effect on marjoram vegetative growth (Table 2) and dry weight (Table 6) as mentioned earlier (Adholeya and Prakash, 2004).

Effect of biofertilizers

The obtained results in Table 8 indicated that the different sources of biofertilizers had a significant effect on N, P and K contents in the two cuts in both seasons, except N (%) in the 2nd cut in the 2nd season and P (%) in 1st and 2nd cut in the 2nd season. It is clear that, generally, treatment of marjoram transplant with halex-2 biofertilizers in the two cuts in both seasons gave the highest content of N, P and K in herb of marjoram plants without significant differences with nitrobein biofertilizers in most cases, specially in 1st cut with regard to N (%), while the lowest value of N, P and K content was recorded in plants unfertilized with biofertilizers. The significant effect of biofertilizers may be due to the effect of different strain groups and nutrients mobilizing micro-organisms which help in availability of metals and their forms in the composted material and increased levels of extracted minerals (El-Kramany et al., 2000). As for the mineral content in the herb of marjoram plant as affected by the interaction between organic and biofertilizers, data in Table 9 obviously showed that organic and biofertilizers had significant effect on N, P and K contents in herb of marjoram plants in the two cut in both seasons, except P (%) in the 2nd cut in the 1st season and two cut in the 2nd season. Poultry manure as an organic fertilizer source combined with halex-2 or nitrobein biofertilizers, significantly increased N, P and K contents in herb and recorded the maximum values in this respect without significant differences between the two cuts in both

Table 9. Effect of interaction between organic sources and biofertilizers on nitrogen, phosphorus and potassium contents (%) of marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	N (%)				P (%)				K (%)			
		1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
		1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
FYM	Without	1.91	2.11	2.35	2.39	0.209	0.220	0.222	0.226	1.41	1.42	1.48	1.69
	Nitrobein	2.25	2.38	2.44	2.49	0.223	0.212	0.228	0.227	1.64	1.85	1.64	1.94
	Halex-2	2.29	2.28	2.54	2.48	0.254	0.251	0.259	0.232	1.87	2.19	1.74	2.18
Poultry manure	Without	2.56	2.23	2.39	2.37	0.243	0.220	0.224	0.220	2.19	2.10	2.33	2.23
	Nitrobein	2.70	2.62	2.66	2.54	0.264	0.253	0.256	0.264	2.34	2.50	2.39	2.24
	Halex-2	2.65	2.69	2.75	2.59	0.268	0.257	0.261	0.259	2.52	2.53	2.55	2.39
New LSD at 0.05 level		0.12	0.05	0.07	0.07	0.049	NS	NS	NS	0.07	0.17	0.17	0.09

seasons. On the other hand, the minimum values in this respect were obtained by addition of organic manure as the source of farmyard manure without biofertilizers. Organic manure contains microorganisms such as *Azotobacter*, *Azospirillum*, etc., which fix N and release phytohormones as GA, IAA, Cytokinin,

N, P and K uptake by plant effect of organic fertilizer

As for the mineral uptake by herb of marjoram plant as affected by application of organic manure, data in Table 10 showed that poultry manure significantly increased N, P and K uptake by herb in the early cut, while farmyard manure recorded the maximum values in the late cut in both seasons. It could be concluded that the increment in N, P and K uptake by herb plants may be attributed to the increase in the plant dry weight and N, P and K (%) in herb. These results coincide with those reported by Aflatuni et al. (1993) and Abo- (2008). They found that application of organic manure increased N, P and K contents in herb tissues of marjoram. etc., which are necessary for stimulating plant growth, dry matter content, and absorption of nutrients (Reynders and Vlassak, 1982).

Effect of biofertilizers

The effect of biofertilizers sources on N, P and K uptake by herb of marjoram plants are presented in Table 10. The results indicated that the different sources of biofertilizers had a significant effect on N, P and K uptake in the two cuts in both seasons. Generally, treatment of marjoram transplant with halex-2 in the two cuts in both seasons gave the highest uptake of N,P and K by herb of marjoram plants, while, nitrobein biofertilizers gave

intermediate values in the two cuts in both seasons. On the other hand, the lowest values in this respect were obtained in the absence of biofertilizers in the two cut in both seasons. These results agreed with those obtained by Kandeel and Sharaf (2003). They indicated that biofertilizers stimulated N, P and K contents in herb tissues of marjoram plants. The interaction between organic manure sources and bio-fertilizers had a significant value and increased N, P and K uptake at two cuttings stage, reaching the maximum value for the combination of poultry manure and halex-2 or nitrobein bio-fertilizers in the early cut in both seasons, while the combination between farmyard manure and halex-2 bio-fertilizers recorded the maximum values in this respect in the late cut in both seasons. On the other hand, the lowest values of N, P and K uptake were recorded by farmyard manure in the absence of bio-fertilizers in the two cutting stages in both seasons (Table 11).

Essential oil (%), oil yield / plant and oil yield per hectare

Effect of organic fertilizers

The presented data in Table 12 indicates the effect of organic sources, on volatile oil percentage, oil yield/ plant and oil yield per hectare of marjoram plant (Gewaily et al., 2006). It is clear that sources of organic fertilizers reflected a significant effect on volatile oil yield/ plant and oil yield per hectare in herb at different cuts date in each experimental season, except in the two cut in the 2nd season as for volatile oil percentage. It also showed that volatile oil percentage, oil yield/ plant and oil yield per hectare were at the uppermost values by adding organic manure as the source of poultry manure (1.93 and 1.62%) for volatile oil percentage in the 1st and 2nd cut in

Table 10. Effect of organic sources and biofertilizers on N, P and K uptake by marjoram plants during 2007/2008 and 2008/2009 seasons.

Treatments	Uptake (mg/ plant)											
	N				P				K			
	1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
Effect of organic fertilizer												
FYM	611.75	809.13	707.41	795.52	65.17	82.17	68.74	73.98	470.29	662.29	471.41	637.86
Poultry manure	857.19	534.23	1090.93	530.98	84.38	51.62	103.66	52.80	769.85	505.25	1016.09	485.55
New LSD at 0.05 level	143.5	128.51	132.91	140.21	NS	23.87	21.02	16.32	130.85	NS	163.29	140.78
Effect of biofertilizers												
Without	520.81	531.89	753.42	503.79	52.45	54.42	70.82	47.29	421.17	407.78	639.54	403.34
Nitrobein	766.08	687.41	912.15	653.85	75.33	63.41	86.79	62.82	620.57	585.71	739.17	535.31
Halex-2	916.53	795.74	1031.93	832.11	96.56	82.87	100.98	80.06	818.48	757.82	852.53	746.46
New LSD at 0.05 level	74.63	66.37	68.63	69.90	12.13	12.36	10.86	8.43	67.59	86.71	84.34	72.55

Table 11. Effect of interaction between organic sources and biofertilizers on N, P and K uptake by marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	Uptake (mg/ plant)											
		N				P				K			
		1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	
FYM	Without	415.43	692.92	550.37	604.67	45.46	72.25	51.99	57.18	306.68	466.33	346.62	427.57
	Nitrobein	629.55	793.97	710.28	804.02	62.40	70.72	66.37	73.30	458.87	617.16	477.40	626.43
	Halex-2	790.28	940.50	861.57	977.86	87.66	103.54	87.85	91.48	645.34	903.38	590.21	859.57
Poultry manure	Without	626.18	370.85	956.48	402.90	59.44	36.59	89.64	37.40	535.67	349.23	932.47	379.10
	Nitrobein	902.61	580.85	1114.01	503.68	88.26	56.09	107.21	52.35	782.26	554.25	1000.93	444.19
	Halex-2	1042.78	650.98	1202.30	686.35	105.46	62.19	114.11	68.64	991.62	612.26	1114.86	633.35
New LSD at 0.05 level		129.2	114.9	118.8	121.0	21.10	21.42	18.83	14.63	117.01	150.11	146.0	125.6

the 1st season, (0.628, 0.581 and 0.837, 0.570 mg) for oil yield /plant in the 1st and 2nd cut in the

1st and 2nd seasons, respectively, (43.426, 40.174 and 57.928 and 39.425 kg) for oil yield/ha in 1st and

2nd cut in the 1st and 2nd seasons, respectively. These results can be explained in the light of

Table 12. Effect of organic sources and biofertilizers and their interaction on volatile oil percentage, oil yield / plant and oil yield /ha of marjoram plants during 2007/2008 and 2008/2009 seasons.

Treatments	Volatile oil percentage				Oil yield/plant (ml/100 g)				Oil yield (kg/ha)			
	1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
	Effect of organic fertilizer											
FYM	1.393	1.400	1.649	1.598	0.391	0.294	0.475	0.337	27.062	20.341	32.882	23.339
Poultry manure	1.936	1.621	1.984	1.762	0.628	0.581	0.837	0.570	43.426	40.174	57.928	39.425
New LSD at 0.05 level	0.325	0.201	NS	NS	0.108	0.106	0.110	0.118	0.222	0.171	0.295	0.214
	Effect of biofertilizers											
Without	1.432	1.339	1.654	1.545	0.331	0.331	0.525	0.327	22.897	22.921	36.299	22.607
Nitrobein	1.606	1.511	1.846	1.715	0.493	0.420	0.655	0.447	34.125	29.032	45.341	30.922
Halex-2	1.956	1.681	1.950	1.781	0.722	0.550	0.757	0.587	50.000	38.066	52.376	40.626
New LSD at 0.05 level	0.168	0.103	0.205	0.132	0.065	0.054	0.056	0.060	0.114	0.087	0.151	0.110

facts that, using poultry manure, led to increased organic matter, availability of nutrients, nitrogen fixation, rizosphere microorganisms that release phyto hormones, and substances which lead to increased growth and dry matter accumulation and this in turn increased concentration of oil (Edris et al., 2003; Jung et al., 2004). These results are in line with those obtained by Elazam (2008) on marjoram plants. He concluded that application of organic manure to marjoram plants had significantly, the highest increase in essential oil percentage.

Effect of biofertilizers

The presented data in Table 12 showed clearly that, the treatment of transplants of marjoram with biofertilizers had a significant effect on the volatile oil yield/ plant and oil yield per hectare in herb at different cuts during the experimental seasons. Application of halex-2 bio-fertilizers to transplants

gave the highest concentration of oil in herb of plants (1.95 and 1.68, 1.95 and 1.78%) in the 1st and 2nd cut in the 1st and 2nd seasons respectively and there was insignificant differences between nitrobein bio-fertilizers, (0.722 and 0.550, 0.757 and 0.587 mg) for oil yield per plant in the 1st and 2nd cut in the 1st and 2nd seasons, respectively, (50 and 38.066, 52.376 and 40.626 kg) for oil yield per hectare in the 1st and 2nd cut in the 1st and 2nd seasons, respectively. On the other hand, the lowest values in this respect were obtained in the untreated plants with bio-fertilizers.

The beneficial effect of bio-fertilizers application on marjoram plant may be correlated with improving plant growth, dry matter production and mineral content, then improved and increased volatile oil in herb tissues. These results ensure the beneficial effects of halex-2 and nitrobein bio-fertilizers for enhancing oil yield. These results were agreeable with those obtained by Eid and El-Ghaw was (2002) and Mahfouz (2003) on marjoram plants. They concluded that the

significantly highest increase in essential oil percentage, oil yield /plant and ha were obtained by application of biofertilizers. The interaction between organic manure fertilizer and biofertilizer was of significant value and there was increased volatile oil percentage, oil yield/ plant and oil yield per hectare at two cuts date in both seasons (Table13). The maximum values were obtained by the combination of poultry manure and halex-2 biofertilizer in both seasons while, the lowest values of these parameters were recorded with farmyard manure only, in both seasons (Gharib et al., 2008). Moreover, the highest concentration of volatile oil was obtained when plants were harvested at an early date (1st cut) in both seasons. On the other hand, the lowest concentration of oil in herb tissues was obtained in the 2nd cut in both seasons. Marjoram oil contains major components containing α - Pinene, β - Pinene, Limonene, 1,8 cineole, Linalool, Terpinene-4-ol, α -Terpinene, Linalyl/acetate and Eugenol. Some of these components are used for

Table 13. Effect of organic sources and biofertilizers and their interaction on volatile oil percentage, oil yield / plant and oil yield /ha of marjoram plants during 2007/2008 and 2008/2009 seasons.

Organic sources	Biofertilizers	Volatile oil percentage				Oil yield/ plant (ml/100 g)				Oil yield (kg/ha)			
		1 st season		2 nd season		1 st season		2 nd season		1 st season		2 nd season	
		1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut	1 st cut	2 nd cut
FYM	Without	1.164	1.193	1.668	1.383	0.253	0.198	0.391	0.235	17.517	13.726	27.028	16.267
	Nitrobein	1.368	1.351	1.630	1.788	0.383	0.300	0.474	0.355	26.482	20.722	32.830	24.532
	Halex-2	1.649	1.655	1.649	1.623	0.569	0.401	0.559	0.430	39.373	27.710	38.701	29.759
Poultry manure	Without	1.700	1.484	1.639	1.707	0.416	0.487	0.656	0.432	28.770	33.720	45.383	29.880
	Nitrobein	1.846	1.671	2.062	1.642	0.617	0.557	0.864	0.530	42.697	38.568	59.750	36.685
	Halex-2	2.263	1.707	2.250	1.940	0.890	0.704	0.984	0.765	61.613	48.719	68.062	52.925
New LSD at 0.05 level		0.291	0.180	0.355	0.230	0.097	0.095	0.098	0.105	0.198	0.152	0.263	0.191

scencing cosmetics and others are used for flavoring pharmaceuticals such as D-limonene and Linalool (Refaat, 1988). These results are in harmony with those obtained by Abo (2008) who found that the highest values of yield of oil/plant of marjoram were obtained when poultry manure was applied at 25 m³/ha combined with phosphorein or yeast biofertilizers. Finally it could be concluded that the interaction between poultry manure at a rate of 75 m³/ha and combined with halex-2 and nitrobein biofertilizers are adjusted as the superior treatments for increasing plant growth, herb dry weight and oil yield of marjoram plants (Banchio et.al., 2008).

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