

Effect of organic and bio-fertilizer application on growth and productivity of lovage (*Levisticum officinale*, Koch) plant under sandy soil conditions

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ABSTRACT

The present study was carried out during the two successive seasons of 2014 & 2015 and 2015& 2016 at the Experimental Station of Medicinal and Aromatic Plants of Belbis desert, EL-Sharkiya Governorate, Egypt. The objective of this work was to study the effect of organic and bio fertilizers and their combinations on vegetative growth, roots yield, essential oil productivity and some chemical constituents of *Levisticum officinale*, Koch) plants. The 2nd cut gave the highest mean values of number of branches / plant as well as fresh and dry weights of herb during both seasons. All organic and bio-fertilizer treatments had a pronounced effect on growth and yield characters as well as essential oil production of herb and roots of lovage plants. The combination between poultry manure 10m³/fed. + bio-fertilizer followed by the cattle manure 10 m³/fed. + bio-fertilizer resulted in the maximum values of herb fresh weight, herb dry weight, fresh weight of roots , dry weight of roots as well as oil yield in the herb and root. The plants treated with bio-fertilizer only gave the highest essential oil percentages in the herb and root .The main compounds in lovage herb oil samples studied were α -terpinyl acetate, β -phellandrene, α -myrcene and dl-limonene. On the other hand, the main chemical constituents of roots essential oil were falcarinol, trans ligustilide, β -pinene and sabinene.

Key words: Lovage , organic fertilizer , bio fertilizer

Introduction

Medicinal and aromatic plants play a significant role in the life of people who as by 80% of global population benefit from their medicinal therapeutic effects as reported by WHO (2008). These plants synthesize substances that are useful to the maintenance of health in humans and other animals and there is a return back to use herbs in pharmaceuticals. Lovage (*Levisticum officinale*, Koch) is a sturdy perennial plant belongs to Family Apiaceae, native to the Eastern Mediterranean Region only in South-Eastern Europe and South west from Asia, and is grown in Iran and Afghanistan. Lovage plant is tall up to 1.5 meters at flowering with yellow flowers, leaves are bright greenish yellow, stems are erect, round, thick and hollow. The fruit is yellow-brown, 5 to 7 mm. long, compressed, and has sharply keeled to winged ribs. The roots, are composed of a vertical rhizome, 5 to 10 cm. long and 2 to 4 cm. thick, from which spring forth numerous lateral rootlets, up to 20 mm. long and a few millimeters thick. Besides, all parts of the plant have a strong flavor which generally characterizes celery and contains a volatile oil, which can be obtained by steam distillation. The leaves are eaten as salad, or used dry as a tea. The roots and fruit are used as a stimulant aromatic, and as a carminative, diuretic, veiny, spasmolytic activity and emmenagogue. The fruits are used for flavoring specially in candy and in liqueurs of French type. Also, used in upsets of the stomach and fever, especially for cases of colic and flatulence in children (Guenther, 1961).

Manures and biofertilization are very important for medicinal and aromatic plants which can help increasing the yield without causing the damage associated with chemical fertilizers and reduce production cost, environmental pollution and get the best product of both quantity and quality. Also, continuous usage of inorganic fertilizer affects soil structure. Therefore, manures can serve as alternative to mineral fertilizers for improving soil structure and microbial biomass. Therefore, the trend now is using the bio and organic fertilizers. Organic fertilizers are obtained from animal sources such as animal manure or plant sources which contains the main plant nutrients nitrogen (N), phosphorus (P), potassium (K), and micronutrients that are released throughout the growing season. The plant nutrients are released when organic matter decomposes and is changed into humus. In sandy

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soil the humus makes the sand particles stick together, this reduces the size of the spaces (pores). Compost that derived from organic fertilizers is another source of organic material that is commonly used in sustainable soil management. Compost helps the soil hold both water and air for plants and makes trace elements or micronutrients available to plants. Compost also reduces soil pH and increases the electrical conductivity and the ability of absorbing soil nutrients (Davarnjad *et al.*, 2002). Many investigators indicated that organic fertilization increases soil organic matter, specially for the sandy soils in Egypt to improve the physical, chemical and biological properties. In this concern, Shahram and Kourosh (2011) indicated that organic and bio fertilizers are a good substitute for inorganic fertilizers for medicinal plants, Leite *et al.* (2005) on marigold reported that combining between organic and bio-fertilizers is useful tool for optimizing plant production and quality, as well as Sakr (2005) stated that fertilization of Alexandrian senna plants with poultry manure + cattle manure + biofertilizer resulted in the best results for the vegetative growth characteristics and chemical composition. Also, Saeed and Rezvani (2010), reported that the highest thousands kernel weight of cumin belonged to compost treatment, Saber and Khalid (2011) on chamomile recorded that, adding compost to liquid compost had a positive effect on growth characters and essential oil compared with chemical fertilizers.

Biofertilization with a group of living cells of micro-organism like bacteria, algae and fungi alone or in combination enhances the productivity of soil by fixing atmospheric nitrogen or by solubilizing soil phosphorus or by stimulating plant growth through production of phytohormones such as gibberellins, cytokinins and IAA that act as growth regulators and production of bio-control agents against soil-borne phytopathogens and consequently increase formation of metabolites which encourage the plant vegetative growth and enhance the meristematic activity of tissues to produce more growth (Ali *et al.*, 1987).

The positive effect of bio-fertilizers on vegetative growth, yield and oil productivity in several plants was revealed by Mahfouz and Sharaf-Eldin (2007) on fennel. Ismail (2007) on dragonhead plant, Amran (2013) on *Pelargonium graveolens*, El-Khyat (2013) on *Rosmarinus officinalis*, Ahmed and Kibret, (2014) and Mady and Youssef (2014) on dragonhead plants.

Organic and bio fertilizers in comparison with the chemical fertilizers have lower nutrient content and are slow release but they are as effective as chemical fertilizers over longer periods of use (Naguib, 2011 and Mohamed *et al.*, 2015). In this respect, Abd El- Wahab (2013) showed that organic fertilizers enhanced vegetative growth parameters and essential oil productivity of *Origanum syriacum* var. *Sinaicum* plants.

The target of this work was to estimate the benefits of adding organic and bio fertilizers on growth and essential oil productivity and get organic production of lovage plants.

Materials and Methods

The present study was carried out during the two successive seasons of 2014& 2015 and 2015& 2016 at the Experimental Station of Medicinal and Aromatic Plants of Sekem Company, Belbis, EL-Sharkiya Governorate, Egypt. The objective of this work was to study the effect of organic and bio fertilizers, cuts and their combinations on vegetative growth, yield, essential oil productivity and some chemical constituents of *Levisticum officinale*, Koch) plants.

Plant materials:

Lovage seeds were obtained from Germany by ENZA ZADEN Co. Schifferstadter Straße 67125 Dannstadt-Schauernheim Germany as an average weight of 1000 seeds was 01.18 gm.

Experimental procedure:

Seeds were sown on 23rd and 30th September during 1st and 2nd seasons, respectively directly in the reclaimed sandy soil plains. The distance between rows was 15 cm and 20 cm between hills (33 plant / m² equal 138600 plants/ fed). The plants were thinned to two plants per hill after three weeks from germination. Drip irrigation was used with drippers (4 liter / hour /hill) in the whole period of both seasons for only one hour every two days.

The analysis of soil and water:

All soil and water samples were analyzed in the Desert Research Center laboratories according to Rainwater and Thatcher (1960) as presented in Tables (1&2). Soil samples representing the experimental area were taken at 0-30 cm depth. The water analysis previously illustrated was taken from the irrigation water used.

Table 1: The physical and chemical properties of the experimental soil

Soil type	pH	E.C. (dSm ⁻¹)	O.M. (%)	Cations (meq/l)				Anions (meq/l)			TDS (mg/l)	N (mg/l)	P (mg/l)	K (mg/l)
				Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	HCO ₃ ⁻	SO ₄ ⁻	Cl ⁻				
Fine Sandy	7.5	0.93	1.9	2.98	1.07	4.72	0.59	2.45	3.36	3.55	584.9	71.2	5.12	65.4

Table 2: Water analysis of the irrigation water

TDS mg/l	pH	EC (dSm ⁻¹)	Soluble cations (mg/l.)				Soluble anions (mg/l.)			
			Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	CO ₃ ⁻	HCO ₃ ⁻	SO ₄ ⁻	CL ⁻
233.6	7.2	397	33.2	11.0	33.0	5.0	21.0	112.9	44.5	29.6

Organic fertilizer:

The organic fertilization treatments included the following:

- 1- Compost manure at the rate of 10 m³ per feddan.
- 2- Cattle manure at the rate of 10 m³ per feddan.
- 3- Poultry manure (PM) at the rate of 10 m³ per feddan.

Different organic fertilizers were added before planting in each season in only one dose. The organic fertilizers were obtained from Sekem Company. The chemical characteristics of compost, cattle manure and poultry manure are shown in Table (3). Common cultural practices were applied throughout the experiment, including weed control etc. were similarly done whenever needed. Organic fertilizer was analyzed in the Desert Research Center laboratories.

Table 3: Chemical analysis of compost, cattle manure and poultry manure

Character	pH	E.C (dSm ⁻¹)	C/N ratio	Moisture (%)	Organic carbon (%)	Organic matter (%)	N (%)	P (%)	K (%)	Ca (%)	Fe (ppm)	Mn (ppm)	Zn (ppm)	Cu (ppm)
Compost	8.10	3.10	1:16	30.11	20.19	45.33	1.3	1.1	1.51	5.73	2800	130	70	4.5
Cattle manure	7.36	5.22	1:15	55.45	18.25	51.21	1.5	0.8	1.45	8.77	2660	120	50	12
Poultry Manure	6.96	4.10	1:11	20.67	38.11	60.15	3.5	2.3	1.7	6.67	2400	150	170	15

Bio-fertilizer:

The biofertilizer was added as a mixture of 5 strains of bacteria namely: *Azotobacter chroococcum*, *Azospirillum lipoferum*, *Bacillus polymixa*, *B. megatherium* and *Pseudomonas fluorescense* (1X10⁸ / ml) obtained from Desert Research Center, El-Mataria, Egypt. The biofertilizer (1 L mixture of 5 strains of bacteria +20 l of water) was added as a soil drench four times per season at monthly intervals started from planting the seeds.

The treatments were conducted as follows:

- 1- Compost at 10 m³/fed. (Control)
- 2- Compost at 10 m³/fed. ++ biofertilizer.
- 3- Cattle manure at 10 m³/fed.

- 4- Cattle manure at 10 m³/fed. + bio-fertilizer.
- 5- Poultry manure at 10m³/fed.
- 6- Poultry manure at 10m³/fed. + bio-fertilizer.
- 7- Bio-fertilizer only.

Harvesting:

Two cuttings were carried out, the first one was 90 days after sowing and the second cutting was 70 days after the first one and roots were harvested after the second cut.

Statistical analysis:

The experimental layout was factorial in a complete randomized block design with three replicates, each replicate contained 10 plants, the main factor was fertilization treatments while the sub plot was the cuts. Data from all experiments were subjected to analysis of variance using Costat Statistical Software (1986). Means of all data were compared by L.S.D. method according to (Snedecor and Cochran, 1968).

Data recorded:

The following parameters were recorded during each cut:

Vegetative characteristics:

- Plant height (cm.).
- Number of branches / plant.
- Fresh and dry weights of herb (g/plant).
- Fresh and dry weights of roots (g / plant).

Determination of essential oil:

- Essential oil percentage in dry herb.
- Oil yield per dry herb (ml/plant).
- Essential oil percentage in dry roots.
- Oil yield per dry roots (ml / plant).

Volatile oil percentage and content of dry herb and roots for each sample were determined using Clavenger apparatus according to (British Pharmacopoeia, 1963). The extracted essential oil was dehydrated over anhydrous sodium sulphate and stored at freezer till used for gas chromatography-mass spectrometry (GC-MS) analysis.

GC-MS analysis: samples of oil for herb and roots were conducted according to (British Pharmacopoeia, 1963) GC/ Mass analysis of volatile oil of each treatment was performed with specification of the apparatus used according to (Robert Adams, 1995). GC/ Mass analysis of volatile oil for roots and herbs. The essential oil was analyzed on a VG analytical 70- 250S sector field mass spectrometer, 70 eV, using a SPsil5, 25 m x 30 m, 0.25 µm coating thickness, fused silica capillary column, injector 222°C, detector 240°C, linear temperature 80°–270°C at 10°C/min. Diluted samples (1/100, v/v, in *n*-pentane) of 1 µl were injected, at 250 °C, manually and in the split less mode flame ionization detection (FID) using the HP Chemstation software on a HP 5980 GC with the same type column as used for GC/MS and same temperature program. Identifications were made by library searches (Adams, 2007) combining MS and retention data of authentic compounds by comparison of their GC retention indices (RI) with those of the literature or with those of standards available in our laboratories (Adams, 2007).

Determination of photosynthetic pigments and minerals:

Chlorophyll (a), chlorophyll (b) and carotenoids contents (mg/g fresh leaves) were determined as described by AOAC (1980) after one month from the last treatment. The percentage of N,P,K, and total carbohydrates were determined in the dry leaves during flowering stage, where total nitrogen was determined using the modified microKjeldahl method according to A.O.A.C. (1980). Other nutrients (P and K) contents were determined after wet digestion according to the method described by Chapman and Pratt (1961), while total carbohydrates percent was determined according to Dubois *et al.*, (1956).

Results and Discussion

1- Growth and yield characteristics:

Data presented in Tables (4, 5 and 6) exhibit the effects of seven fertilizer treatments (various kinds of organic fertilizers and / or biofertilizer) on vegetative growth and yield characters.

1-1- Effect of cuttings:

It is clear from data tabulated in Tables (4,5) that cutting had a pronounced effect on plant height, number of branches / plant as well as fresh and dry weights of herb (g / plant) during both seasons. The maximum mean values of plant height were obtained during 1st cut while the 2nd cut gave the highest mean values of number of branches / plant as well as fresh and dry weight of herb (g / plant) during both seasons.

Table 4: Effect of fertilization on plant height and number of branches of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters	Plant height (cm.)			Number of branches / plant		
	1 cut	2 cut	Mean	1 cut	2 cut	Mean
Treatments	First season 2014/2015					
F1 (Control)	56.75	53.50	55.13	3.50	4.50	4.00
F2	62.50	59.00	60.75	5.50	6.75	6.13
F3	67.50	63.50	65.50	6.00	7.00	6.50
F4	70.00	65.00	67.50	6.50	7.50	7.00
F5	68.75	63.75	66.25	6.00	7.75	6.88
F6	71.75	60.75	66.25	6.75	7.50	7.13
F7	52.00	43.00	47.50	3.00	3.50	3.25
L.S.D. at 0.05%						
Fertilization	1.71			0.53		
Cuts	1.25			0.60		
Fertilization * cuts	1.55			0.75		
Second season 2015/2016						
F1 (Control)	56.00	52.00	54.00	3.25	4.00	3.63
F2	61.00	58.00	59.50	5.00	6.00	5.50
F3	66.25	62.25	64.25	5.75	6.50	6.13
F4	68.00	64.00	66.00	6.00	7.00	6.50
F5	67.50	62.00	64.75	6.20	7.00	6.60
F6	70.50	61.50	66.00	6.00	7.00	6.50
F7	50.00	41.00	45.50	3.25	3.50	3.38
L.S.D. at 0.05%						
Fertilization	1.66			0.51		
Cuts	1.15			0.57		
Fertilization* cuts	1.89			0.81		

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

Table 5: Effect of fertilization on herb fresh and dry weights / plant of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters	Herb fresh weight / plant (g)			Herb dry weight /plant (g)		
	1cut	2 cut	Mean	1cut	2 cut	Mean
Treatments	First season 2014/2015					
F1 (Control)	25.00	30.00	27.50	3.23	4.32	3.78
F2	30.33	40.00	35.17	4.35	5.25	4.80
F3	40.33	45.00	42.67	5.40	6.11	5.76
F4	50.00	65.00	57.50	6.66	8.24	7.45
F5	55.00	60.00	57.50	7.41	7.87	7.64
F6	60.00	60.00	60.00	7.75	7.85	7.80
F7	20.00	25.00	22.50	2.33	3.35	2.84
L.S.D. at 0.05%						
Fertilization	4.22			1.11		
Cuts	4.93			0.98		
Fertilization * cuts	2.61			0.37		
Second season 2015/2016						
F1 (Control)	28.00	34.00	31.00	3.66	4.55	4.11
F2	35.33	45.00	40.17	4.61	5.73	5.17
F3	45.33	50.00	47.67	5.88	7.54	6.71
F4	55.00	60.00	57.50	7.12	7.60	7.36
F5	60.00	65.00	62.50	7.71	8.33	8.02
F6	65.00	65.00	65.00	8.43	8.40	8.42
F7	20.00	25.00	22.50	2.41	3.42	2.92
L.S.D. at 0.05%						
Fertilization	4.92			0.88		
Cuts	4.75			0.88		
Fertilization* cuts	2.65			0.39		

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

Table 6: Effect of fertilization on fresh and dry weights of roots / plant of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters	Fresh weight of roots / plant (g)			Dry weight of roots /plant (g)		
	First season	Second season	Mean	First season	Second season	Mean
Treatments						
F1(Control)	15.50	16.00	15.75	4.00	4.50	4.25
F2	25.00	27.50	26.25	5.10	5.75	5.43
F3	30.00	29.75	29.88	6.12	6.10	6.11
F4	35.50	37.00	36.25	7.00	7.55	7.28
F5	33.50	32.50	33.00	6.34	6.15	6.25
F6	35.00	35.00	35.00	7.21	7.15	7.18
F7	10.50	11.00	10.75	2.50	2.61	2.56
L.S.D. at 0.05%						
Fertilization	4.12		4.16	0.72		0.69

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

1-2-Effect of fertilizers:

All the applied fertilizer treatments significantly increased growth and yield characters (Tables 4 and 5) in the two seasons. However, cattle manure at 10 m³/fed. + bio-fertilizer gave the highest value of plant height (67.50 cm) in the 1st season while, cattle manure at 10 m³/fed. + bio-fertilizer or poultry manure at 10m³/fed. + bio-fertilizer gave the highest one (66.0 cm) in the 2nd season.

Regarding the effect of fertilizer treatments on number of branches/ plant, it can be noticed that, number of branches reached to its maximum values (7.13 and 6.60) as a result of poultry manure 20m³/fed. + bio-fertilizer and poultry manure at 10m³/fed. during 1st and 2nd seasons, respectively. It is obvious from data in Table (5) that, poultry manure at 10m³/fed. + bio-fertilizer increased herb fresh weight to 60 and 65 g / plant and herb dry weight to 7.80 and 8.42 g / plant during 1st and 2nd seasons, respectively.

Table (6) exhibited data on fresh and dry weights of roots as influenced by the different fertilizer treatments during both seasons. Cattle manure at 10 m³/fed. + bio-fertilizer treatment gave the highest values of roots fresh weight which recorded 35.5 and 37.0 g / plant for first and second seasons, respectively. On the other hand, roots dry weight reached at its maximum values (7.21 and 7.55 g / plant) as a result of poultry manure at 10m³/fed. + bio-fertilizer and cattle manure at 10 m³/fed. + bio-fertilizer treatments for 1st and 2nd seasons, respectively. Generally, the lowest mean values of the above characters were obtained as a result of bio-fertilizer only.

1-3- Effect of interaction treatments between fertilizer and cuts:

Data tabulated in Tables (4 and 5) indicate that, the combination treatments had a significant effect on plant height (cm), number of branches / plant as well as herb fresh and dry weights (g / plant). Plant height reached to its maximum mean values (71.75 and 70.50 cm) as a result of the interaction treatment between 1st cut and poultry manure at 10m³/fed. + bio-fertilizer followed by the interaction between 2nd cut and cattle manure at 10 m³/fed. + bio-fertilizer which recorded (65.00 and 64.00 cm) for 1st and 2nd seasons, respectively. Regarding the effect of combination treatments on number of branches, data in Table (4) shows that the combination between second cut and poultry manure at 10m³/fed., gave the highest value of branches number (7.75) during the first season. Plants receiving cattle manure 20 m³/fed. + bio-fertilizer, poultry manure at 10m³/fed., or poultry manure at 10m³/fed. + bio-fertilizer under 2nd cut gave the highest mean value of number of branches (7.00) during the 2nd season.

It is obvious from data of the first season, in Table (5), that the combination between 2nd cut and cattle manure at 10 m³/fed. + bio-fertilizer resulted in the maximum values of herb fresh weight (65.00g/plant) and herb dry weight (8.24 g / plant) compared with other treatments. In the second season, the combination between 1st cut or 2nd cut with poultry manure at 10 m³ / fed. + biofertilizer as well as the combination between 2nd cut with 10 m³ poultry manure only gave the highest values of herb fresh weight (65.00 g / plant). Regarding herb dry weight, the results in Table (5), clear that the combination between 1st cut with poultry manure + biofertilizer produced the highest values of herb dry weight (8.43 g / plant) followed by the combination treatment between 2nd cut with the same fertilizer treatment which recorded (8.40 g / plant) where there were non significant differences between both treatments.

2- Essential oil percentage and yield (ml / plant):

Results tabulated in Table (7) clear that, fertilizer treatments had a significant effect on herb essential oil percentage and oil yield (ml/plant) during both seasons.

Essential oil percentage reached maximum values (0.85 and 0.86 %) as a result of biofertilizer treatment only followed by compost at 10 m³ / fed, as recorded 0.83 for each seasons.

Regarding the effect of fertilizer treatments on herb essential oil yield, it is clear that in the first season poultry manure at 10 m³ / fed. only or combined with biofertilizer produced the highest herb essential oil yield (0.056 ml / plant) then came in a descending order cattle manure at 10 m³ / fed. + biofertilizer treatment which led to 0.054 ml / plant. In the second season the results attained a similar

trend to that of the first season except for poultry manure at 10 m³ / fed. + biofertilizer as gave the highest herb essential oil yield (0.061 ml/ plant) followed by poultry manure only at 10 m³ / fed.

Concerning the effect of fertilizer treatments on root essential oil percentage and oil yield (ml / plant) , it is clear from data tabulated in Table (8) that plants fertilized with biofertilizer only produced the maximum values of essential oil percentage (1.45 and 1.46%) followed by compost at 10 m³ / fed. which recorded 1.43 and 1.42 % during first and second seasons, respectively. Regarding the effect of fertilizer treatments on root essential oil yield (ml / plant) , results presented in Table (8) show that the highest root essential oil yield (0.095 and 0.102 ml / plant) resulted from plants which received cattle manure at 10 m³ / fed. + biofertilizer followed by poultry manure at 10 m³ / fed. + biofertilizer which recorded 0.094 ml / plant during each seasons.

Table 7: Effect of fertilization on essential oil percentage in dry herb and oil yield per dry herb (ml) of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters Treatments	Essential oil percentage in dry herb			Oil yield per dry herb (ml)		
	First season	Second season	Mean	First season	Second season	Mean
F1 (Control)	0.83	0.83	0.83	0.031	0.034	0.033
F2	0.79	0.78	0.79	0.038	0.040	0.039
F3	0.75	0.76	0.76	0.043	0.051	0.047
F4	0.73	0.71	0.72	0.054	0.052	0.053
F5	0.73	0.72	0.73	0.056	0.058	0.057
F6	0.72	0.73	0.73	0.056	0.061	0.059
F7	0.85	0.86	0.86	0.024	0.025	0.025

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

Table 8: Effect of fertilization on essential oil percentage in roots and oil yield per roots (ml) of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters Treatments	Essential oil percentage in dry roots			oil yield per dry roots (ml)		
	First season	Second season	Mean	First season	Second season	Mean
F1 (Control)	1.43	1.42	1.43	0.057	0.064	0.061
F2	1.40	1.41	1.41	0.071	0.081	0.076
F3	1.38	1.37	1.38	0.084	0.084	0.084
F4	1.35	1.35	1.35	0.095	0.102	0.098
F5	1.33	1.33	1.33	0.084	0.082	0.083
F6	1.31	1.32	1.32	0.094	0.094	0.094
F7	1.45	1.46	1.46	0.036	0.038	0.037

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

3- Essential oil constituents:

The effect of some fertilizer treatments on the main chemical compositions of the essential oil from the herb and roots of lovage plants are given in Tables (9 and 10).

The main compounds in lovage herb oil samples studied were α -Terpinyl acetate (12.79 – 34.42%), β -phellandrene (14.93- 20.20%), α -myrcene (4.56 –5.20%) and dl-limonene (3.27 – 4.86%). A comparison between composition of lovage herb under the three fertilizer treatments (Table, 9) shows that plants which received compost at 10 m³ only gave the highest content of α -terpinyl acetate (34.42%) and β -phellandrene (20.20%). The maximum values of α -myrcene (5.20%) and dl-limonene

(4.86%) were obtained as a result of cattle manure at 10 m³ / fed. + bio-fertilizer and bio-fertilizer only, respectively.

From data tabulated in Table (10), it is clear that, the main chemical constituents of roots were falcarinol (19.40 – 24.50%), trans ligustilide (11.13 – 14.03%), β-pinene (7.13 – 10.77%) and sabinene (7.03 – 9.73%). It can be noticed that, bio-fertilizer only gave the highest values of falcarinol (24.50%) and trans ligustilide (14.03%) while the highest values of β-pinene (10.77%) and sabinene (9.73%) were obtained from plants fertilized by compost at 10 m³ / fed.

Table 9: Effect of fertilization on the relative percentage of essential oil of lovage herb.

Constituents	F1	F4	F7	Mean
Thujene	0.49	0.45	0.40	0.45
α-Pinene	3.35	3.12	2.99	3.15
Camphene	0.88	0.85	0.75	0.83
Sabinene	1.27	1.55	1.46	1.43
β-Pinene	1.16	1.32	1.43	1.30
α-Myrcene	5.07	5.20	4.56	4.94
α-Phellandrene	1.08	1.05	0.77	0.97
α-Terpinene	0.21	0.20	0.13	0.18
p-Cymene	0.69	0.64	1.16	0.83
dl-Limonene	3.96	3.27	4.86	4.03
β- Phellandrene	20.20	19.63	14.93	18.25
γ-Terpinene	1.25	0.90	0.89	1.01
Linalool	0.23	0.23	0.26	0.24
Pulegone	0.29	0.08	1.71	0.69
α-Terpinyl acetate	34.42	12.79	15.98	21.06
Geranyl acetate	2.04	2.32	2.78	2.78
Total	76.59	53.6	55.06	

F1= Compost at 10 m³/fed. (Control), F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

Table 10: Effect of fertilization on the relative percentage of essential oil of lovage roots.

Constituents	F1	F6	F7	Mean
α-Pinene	6.11	1.44	5.10	4.22
Camphene	1.47	0.39	1.60	1.15
Sabinene	9.73	7.13	7.03	7.96
β-Pinene	10.77	7.85	7.13	8.58
α-Myrcene	0.49	0.65	0.75	0.63
α-Phellandrene	0.35	0.14	0.32	0.27
α-Terpinolene	0.12	0.08	0.12	0.11
dl-Limonene	1.39	1.98	2.30	1.89
Trans ligustilide	12.61	11.13	14.03	12.59
Falcarinol	19.40	22.60	24.50	22.17
Total	62.44	53.39	62.88	

F1= Compost at 10 m³/fed. (Control), F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

4- Photosynthetic pigments:

Data presented in Table (11) exhibit the effect of fertilizer treatments on chlorophyll a, b and total carotenoids (mg / g fresh wt.) during the growing seasons of 2014 / 2015 and 2015 / 2016 seasons.

Cattle manure at 10 m³ / fed. + biofertilizer treatment gave the highest values of chlorophyll a (0.853 and 0.867 mg / g f.w) , chlorophyll b (0.577 and 0.587 mg/g f.w) and total carotenoids (0.570 and 0.583 mg / g fw) in the first and second seasons, respectively. In the following rank came cattle manure at 10 m³ / fed. only which gave chlorophyll a (0.814 and 0.853 mg/g) and chlorophyll b (0.547 and 0.570 mg / g) in the first and second seasons respectively. For total carotenoids, the second

rank came poultry manure at 10 m³ / fed. + biofertilizer which produced (0.566 and 0.571 mg /g) during first and second seasons, respectively.

Table 11: Effect of organic and bio fertilization on chlorophylls (a & b) in leaves of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters Treatments	Chlorophyll (a) mg/g. f.w.)			Chlorophyll (b) mg/g. f.w.)		
	First season	Second season	Mean	First season	Second season	Mean
F1 (Control)	0.675	0.691	0.683	0.445	0.452	0.449
F2	0.727	0.711	0.719	0.483	0.475	0.479
F3	0.814	0.853	0.834	0.547	0.570	0.559
F4	0.853	0.867	0.860	0.577	0.587	0.582
F5	0.744	0.749	0.747	0.470	0.477	0.474
F6	0.773	0.763	0.768	0.493	0.503	0.498
F7	0.653	0.673	0.663	0.430	0.437	0.434

F1= Compost 10 m³/fed. (Control), F2 = Compost 10 m³/fed. + bio-fertilizer, F3= Cattle manure 10 m³/fed., F4= Cattle manure 10 m³/fed. + bio-fertilizer, F5 = Poultry manure 10m³/fed., F6= Poultry manure 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

5- Total carbohydrate (%):

Table (12) presents data on total carbohydrate percentage as influenced by the different fertilizer treatments during 2014/2015 and 2015/2016 seasons. Cattle manure at 10 m³ / fed. + bio-fertilizer gave the highest values of (23.652 and 23.654 %) followed by cattle manure only at 10 m³ / fed which recorded (23.258 and 23.344 %) in the two seasons, respectively.

Table 12: Effect of fertilization on total carbohydrates %and carotenoids (mg/g f.w) of lovage plant during 2014/2015 and 2015/2016 seasons.

Parameters Treatments	Total carbohydrats %			Carotenoids (mg/g . f.w.)		
	First season	Second season	Mean	First season	Second season	Mean
F1 (Control)	18.667	19.333	19.000	0.480	0.485	0.483
F2	22.661	22.667	22.664	0.490	0.495	0.493
F3	23.258	23.344	23.301	0.530	0.540	0.535
F4	23.652	23.654	23.653	0.570	0.583	0.577
F5	21.117	21.112	21.115	0.537	0.548	0.543
F6	21.367	21.355	21.361	0.566	0.571	0.569
F7	16.667	17.667	17.167	0.470	0.477	0.474

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

6- Nutrients Content (%):

Table (13) indicated that poultry manure at 10 m³ / fed. treatment produced the highest accumulation for nutrients of N, P and K for lovage plant during both seasons. The second rank of N (%) was obtained as a result of cattle manure at 10 m³ / fed + bio-fertilizer while compost at 10 m³ / fed + bio-Fertilizer treatment gave the second rank for both P and K (%) during both seasons.

Table 13: Effect of fertilization on N, P and K percentages of lovage plant during 2014/2015 and 2015/2016 seasons.

	N %		P %		K %	
	First season	Second season	First season	Second season	First season	Second season
F1 (Control)	1.343	1.339	0.119	0.120	1.983	2.000
F2	1.363	1.366	0.122	0.124	2.100	2.150
F3	1.303	1.309	0.121	0.122	2.033	2.047
F4	1.477	1.471	0.123	0.125	2.183	2.190
F5	1.453	1.449	0.120	0.121	2.053	2.063
F6	1.388	1.382	0.121	0.122	2.076	2.071
F7	1.341	1.337	0.112	0.114	1.782	1.771

F1= Compost at 10 m³/fed. (Control), F2 = Compost at 10 m³/fed. + bio-fertilizer, F3= Cattle manure at 10 m³/fed., F4= Cattle manure at 10 m³/fed. + bio-fertilizer, F5 = Poultry manure at 10m³/fed., F6= Poultry manure at 10m³/fed. + bio-fertilizer, F7 = bio-fertilizer only.

From the above results it can be concluded that organic and / or bio- fertilizer improved the productivity of lovage plants. In this connection, it appears that using bio-fertilizers in this study could supply plants with their need of nutrients by compost, cattle and poultry manure 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. The increase in lovage 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. It could be concluded that the increment in plant fresh and dry weight may be attributed to the increase in both plant height and number of branches/plant as already discussed. These results are in agreement with those results obtained by Eid and El- Ghawwas (2002), Abo- MAI (2008) and Ahmed *et al.* (2011) on marjoram plants. 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. Dewidar (2007) and Abo (2008) found that the combination between organic and biofertilizers increased plant fresh and dry weights of marjoram plants. The increment in N, P, K contents as a result of organic fertilizer 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 lovage vegetative growth and dry weight as mentioned earlier (Adholeya and Prakash, 2004). The significant effect of bio-fertilizers may be due to the effect of different strain groups and nutrients mobilizing microorganisms which help in availability of metals and their forms in the composted material and increased levels of extracted minerals (El-Kramany *et al.*, 2000). The favorable effect of organic fertilizer on essential oil percentage and yield can be explained in the light of 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 increase growth and dry matter accumulation and this in turn increased concentration of oil (Edris *et al.*, 2003; Lee *et al.*, 2004). They concluded that application of organic manure to marjoram plants had significantly, the highest increase in essential oil percentage. Bio-fertilizer had a significant effect on the essential oil percentage and oil yield. The beneficial effect of bio-fertilizers application on lovage 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 were agreeable with those obtained by Eid and El-Ghawwas (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 bio-fertilizers. The interaction between organic fertilizer and bio-fertilizer was of significant value and they increased essential oil percentage and oil yield/ plant in both seasons. The main constituents of herb essential oil

were α -terpinyl acetate, β -phellandrene, α -myrcene and dl-limonene. In this connection, hydrodistillation extractions of the aerial parts of lovage were analyzed by GC and GC/MS. In the oil, α -terpinyl acetate and β -phellandrene were the main constituents as confirmed by Keivandokh *et al.* (2006) and Azza and Hendawy (2010). For essential oil of roots, falcarinol, trans ligustilide, β -pinene and sabinene were the main constituents. These findings were in harmony with Ibrahim (1999) and Azza and Hendawy (2010). On the other hand, variation in principal components of essential oil from different European countries were determined by Ain *et al.* (2008). They mentioned that dominance of oil constituents was affected by the different geographical locations.

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