

Growth, Volatile Oil Production and Genetic Study of Some Fennel Cultivars Under Different Compost Levels in Sandy Soil

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Abstract:

The present work was conducted during the two successive seasons of 2011/2012 and 2012/2013 at the experimental farm of El-Quassassin Hort. Res. Station, Ismailia Governorate, and Biotechnology Laboratory, Horticulture Research Institute, Agricultural Research Centre, Egypt. The aim of this study was to investigate the effect of three compost fertilizer levels 4, 6 and 8 ton per Fadden using five cultivars of bitter fennel on growth, fruits yield and volatile oil production of fennel (*Foeniculum vulgare* Mill). These cultivars were Netherland, Indian, Azoricum, German and Local fennel. Several trails were studied including growth and yield production, biochemical (the volatile oil) and molecular genetic (RAPD- and ISSR-PCR) characteristics under Egyptian sandy soil. The results showed that increasing compost level progressively (form 4 to 8 ton/ Fed) and significantly increased the values of such parameters. Azoricum cultivar was superior to other cultivars under study, as it showed the best growth in terms, fruits yield, fruit volatile oil (%) and volatile oil production per plant and Fadden. The main compounds in all fennel volatile oils were: Anethole, Estragole, Fenchone and Limonene. The highest percentage of Anethole found in German cultivar, while the lowest percentage found in Local cultivar, where the highest percentage of Estragole (= Methyl chavicol) compound undesirable found in Local cultivar, while the lowest percentage found in Netherland and German cultivars. Random amplified polymorphic DNA (RAPD) and inter-simple sequence repeat (ISSR) molecular fingerprinting markers were employed as genetic markers for the assay of the genetic relationship of five fennel cultivars. In RAPD analysis, 10 selected primers displayed a total of 98 amplified fragments, in which 60 (61.22%) were polymorphic fragments. Thirty-one out of 98 RAPD-PCR fragments were found to be useful as cultivar-specific markers. The largest number of RAPD-PCR markers was scored for Indian variety (68 markers), while the lowest (49 markers) was scored for Netherland variety. In the meantime, the highest number of RAPD-PCR cultivar-specific markers was generated by primer OP-C04 (7 markers), while the lowest number of RAPD-PCR specific markers (1 markers) was generated by primers OP-A13 and OP-B04. In ISSR analysis, 4 of the tested ISSR primers generated variable banding patterns. A total of 26 out of 34 ISSR fragments were polymorphic. Eleven DNA amplified fragments were considered as cultivar-specific markers. The cultivars distribution on the consensus tree according to the banding patterns of RAPD differed from that based on ISSR. This may be due to the possibility that each technique of amplified different parts of the genome. Therefore, it would be useful to use a combination of the banding patterns of the two technique in order to use more segments sites of the genome that verify the validity of the consensus tree

Keywords: Fennel Cultivars, Essential Oil, DNA Fingerprinting And Genetic Relationship.

Introduction

Fennel is a plant belonging to the Umbelliferae (Apiaceae) Family, known and used by humans since antiquity. Because of its flavor, it was cultivated in the countries surrounding the Mediterranean Sea. Fennel is one of the oldest field crop used by the Egyptian for medicinal purposes. Most of the area cultivated with fennel is located in Mid-southern Egypt mainly, El-Fayom, Menia and Assiut Governorates. Only one strain of common or bitter fennel (*Foeniculum vulgare*, Mill.) was cultivated in Egypt for the national and international purposes. Exports of local fennel (*Foeniculum vulgare* Mill.) from Egypt over the past few years have been affected due to the high Estragole but low Anethole content of the oil. Therefore, fennel seeds were imported from different countries to investigate the adaptability of such strains in different locations in Egypt, in comparison with the local one (Shalaby et al., 2011). The volatile oil is used as flavoring agent, carminative antispasmodic, stomachic, diuretic, expectorant, aromatic and lactagogue. Fruits are used as spice, in pickles, candies, in liquors and in the preparation of alcoholic beverages. They are also used as remedy for jaundice and menstrual troubles (Kotb, F. T.1985). The cultivar yields big fruits and reasonable percentage of essential oil with particularly high Estragole content, but it is poor in Fenchone which is an important constituent of the fennel essential oil. These, along with some other components, provide the unique aroma and taste. Trans-anethole accounts for the anise taste, Estragole(=Methyl chavicol) compound undesirable, Fenchone the bitterness, and Limonene provides the citrus taste. Commonly, Trans-anethole was used for flavor in the food and liquors industry, which considered non-toxic (Barazani et al., 2002). Moreover, essential oil of fennel has been shown to have antioxidant, antibacterial and antiviral activities (Frag et al, 1989). The fresh leaves and dried fruits of this plant are used as a spice for meat, baked and confectionery products (Davis, 1972). Compost enhances the environmental sustainability of agriculture by decreasing chemical inputs and increasing soil organic matter (Mathur et al., 1993). Many research workers gained best growth, yield, oil percentage and yield and chemical constituents when used compost for several medicinal and aromatic plants, as (Ibrahim, 1999) on *Ocimum sanctum*; (Khalil, 2002) on rosemary (*Rosemarinus officinalis*); (Khalil et al., 2002) on *Tagetes erecta*; (Khalil and El-Sherbeny, 2002) on three *Mentha* species; (Naguib and Aziz, 2004) on *Hyosyamus muticus* and (El-Sherbeny et al., 2005) on *Sidritis montana*. Germplasm is a vital source in generating new

plant types having desirable traits that help in increasing crop production with quality and thus improve the level of human nutrition. The genetic diversity is analyzed by using morphological as well as genetic based tools, DNA techniques (Bennici et al., 2003) and advanced molecular methods etc. (Barazani et al., 2002; Shiran et al., 2007). The organic fertilizers is utilized for the change of soil texture, supplying nutrients to the growing plants and organic acids, enhancing nutrients uptake as reported by Lampkin (1990), Mohamed and Matter (2001), Badran (2002) and Yousef (2002) and the organic fertilizers consider save for human health. Organic fertilization is also one of the methods used to reclaim sandy desert land and to improve the chemical and physical characteristics of the soil (Gomaa1995, Yousef 2002 and Yousef et al. 2008).

Random amplified polymorphic DNA (RAPD) markers are easier and quicker to use and are preferred in application where the relationships between closely related breeding lines are of interest (Hallden et al., 1994). ISSR-PCR is a genotyping technique based on variation found in the regions between microsatellites. It has been used in genetic fingerprinting (Blair et al., 1999), gene tagging (Ammiraju et al., 2001), detection of clonal variation (Leroy and Leon, 2000), cultivar identification (Wang et al., 2009), phylogenetic analysis (Gupta et al., 2008), detection of genomic instability (Anderson et al., 2001), and assessment of hybridization (Wolfe et al., 1998) in many plant and animal species.

The aims of this study were therefore to study the effect of organic compost on growth and volatile oil quantity of some cultivars of bitter fennel; determine the genetic variability of some fennel cultivars by RAPD and ISSR analysis; determine whether secondary metabolites such as volatile compounds would be used as taxonomic markers in these cultivars and elucidate relationships between genetic and chemical diversity by comparing their hierarchical structures.

Materials and methods

1-Plant materials:

This study was conducted to investigate the effect of different levels of compost fertilizer using five fennel cultivars on growth, fruits production, volatile oil percentage and volatile oil yield. These cultivars were Netherland, Indian, Azoricum, German, and Local fennel. This investigation was carried out during the two successive seasons of 2011 / 2012 and 2012/2013 at the experimental farm of El-Quassassin Horticultural Research Station, Ismailia Governorate, Agricultural Research Center, Egypt. Table 1 shows the mechanical and chemical analyses of farm

The mechanical analysis		The chemical analysis	
Sand	89.92 %	Macro elements (ppm)	
Silt	4.0 %	Nitrogen	81
Clay	6.08 %	Phosphorus	23
The soil was sandy in texture		Potassium	108
		Micro elements (ppm)	
Field capacity (F.C.)	11.20 %	Fe	2.0
Welting point (W.P.)	2.20 %	Cu	---
Organic matter	0.42 %	Zn	0.26
PH (1 soil : 2.5 d.w.)	8.1	Mn	0.8
E.C. (mmohs/cm)	0.21	Anion (mq/100 g soil)	
		Cl ⁻	0.5
			1.0
		HCO ₃	0.97
		SO ₄	
		Cations (mq/100 g soil)	1.0
		Ca ⁺⁺	0.4
		Mg ⁺⁺	0.76
		Na ⁺	0.31
		K ⁺	2.6
		CaCO ₃ (meq/100 g soil)	

Table (1) : The mechanical and chemical analysis of the experimental soil.

Seeds of bitter fennel obtained from Sekem Company, were sown on 1 November of both years. Seedlings were thinned to single plants and irrigated 15 days after sowing. The experimental unit was 5.4 m²; every unit contained three dripper lines with 3m length. Every experimental unit contained 30 plants (about 22222

plants per Fadden). The experiment was carried out using three replicates in a randomized split plot design where the levels of compost were the main plot and varieties of fennel were the subplot. The compost fertilizer was obtained from Arab Organization for Industrialization (A.O.I.); the chemical composition of the compost fertilizer is

Results	Compost characteristics
The color	Dark brown
The smell	Acceptable
The Strength	Spongy
Wet weight per cubic meter (kg / m ³)	510
Dry weight per cubic meter (kg / m ³)	375
The moisture	26.60
PH (1:10)	8.36
Total nitrogen (%)	1.50
Ammonium nitrogen (ppm)	57
Nitrate nitrogen (ppm)	95
Organic matter (%)	35.3
Carbon organic (%)	20.5
The ash (%)	64.7
C:N ratio	1:13.7
Total phosphor (%)	0.38
Total potassium (%)	0.63
Humic acid for organic matter (%)	19.6
Grass seeds	No
Nematode	No
Parasites	No

Table (2): The chemical composition of compost fertilizer

The organic fertilization treatments as compost fertilizer were applied at the rate of 4, 6 and 8 ton per Fadden. The data of plant height (cm), herb fresh and dry weights (g/plant), number of umbels /plant and number of flowers/umbel were recorded at full flowering stage. The fruits yield (g/plant) and fruits yield (kg/fed) were recorded when harvested at fruit maturity stage.

Determination of volatile oil content and composition:

Volatile oil percentage was determined in fruits according to the method described in the General Medical Council (1963). Volatile oil yield per plant was calculated by multiplying volatile oil percentage by fennel fruit yield/plant and expressed as ml/plant. Samples taken for the volatile oil obtained in the second season were analyzed using DsChrom 6200 Gas Liquid Chromatography equipped with a flame ionization detector for separation of volatile oil constituents. The analysis conditions were as follows:-

The chromatograph apparatus was fitted with capillary column BPX-5, 5% phenyle(equiv.) polysillphenylene-siloxane 30m X 0.25 mm ID X 0.25µm film. Temperature program ramp increase with a rate of 10°C/ min from 70° to 200° C. Flow rates of gases were nitrogen at 1 ml / min, hydrogen at 30 ml/ min and 330 ml / min for air. Detector and injector temperatures were 300°C and 250°C, respectively. The obtained chromatogram and report of GC analysis for each sample were analyzed to calculate the percentage of main components of volatile oil. Volatile oil yield/feddan was calculated by multiplying oil (%) by fennel fruit yield.

Rapd -pcr analysis

Polymerase chain reaction (pcr).

In order to obtain clear reproducible amplification products, different preliminary experiments were carried out in which a number of factors were optimized. These factors included PCR temperature cycle profile and concentration of each of the template DNA, primer, MgCl2 and Taq polymerase. A total of twenty random DNA oligonucleotide primers were independently used according to Williams et al. (1990) in the PCR reaction. Only ten primers succeeded to generate reproducible polymorphic DNA products.

The PCR amplification was performed in a 25 µl reaction volume containing the following: 2.5 µl of dNTPs (2.5 mM), 1.5µl of Mg Cl2 (25 mM), 2.5 µl of 10x buffer, 2.0 µl of primer (2.5 µM), 2.0 µl of template DNA (50 ng/µl), 0.3 µl of Taq polymerase (5 U/µl) and 14.7 µl of sterile ddH2O. The reaction mixtures were overlaid with a drop of light mineral oil per sample. Amplification was carried out in Techni TC-512 PCR System. The reaction was subjected to one cycle at 95 °C for 5 minutes, followed by 35 cycles at 96 °C for 30 seconds, 37 °C for 30 seconds, and 72 °C for 30 seconds, then a final cycle of 72 °C for 5 minutes. PCR products were run at 100 V for one hour on 1.5 % agarose gels to detect polymorphism between the fennel cultivars under study. Only ten primers succeeded to generate reproducible polymorphic DNA products. Table 3 lists the base sequences of these DNA primers that produced informative polymorphic bands. The PCR products were separated on a 1.5 % agarose gels and fragments sizes were estimated with two 100bp ladder markers (1000, 900, 800, 700, 600, 500, 400,300,200 and 100bp) and (3000,2500,2000,1500,1000,750,500,250bp).

ISSR-PCR Analysis

Polymerase Chain Reaction (PCR).

ISSR-PCR reactions were conducted using four primers. Amplification was conducted in 25 µl reaction volume containing the following reagents: 2.5 µl of dNTPs (2.5 mM), 2.5 µl MgCl2 (2.5 mM), and 2.5 µl of 10 x buffer, 3.0 µl of Primer (10 pmol), 3.0 µl of template DNA (25 ng/ µl), 1 µl of Taq polymerase (1U/ µl) and 12.5 µl of sterile dd H2O. the PCRs were programmed for one cycle at 94° C for 4 min. followed by 45 cycles of 1 min. at 94 °C, 1 min. at 57 °C, and 2 min at 72 °C the reaction was finally stored at 72 °C for 10 min. The PCR products were separated on a 1.5 % agarose gels and fragments sizes were estimated with the 100bp ladder marker. Only four primers succeeded to generate reproducible polymorphic DNA products. Table 3 lists the base sequences of these DNA primers that produced informative polymorphic bands.

No	RAPD Primer code	Sequence	No	RAPD Primer code	Sequence
1	OP-A02	5'TGCCGAGCTG3'	6	OP-C04	5'CCGCATCTAC 3'
2	OP-A10	5'GTGATCGCAG 3'	7	OP-C05	5'GATGACCGCC 3'
3	OP-A13	5'CAGCACCCAC 3'	8	OP-G14	5'GGATGAGACC3'
4	OP-B04	5'GGACTGGAGT 3'	9	OP-K10	5'GTGCAACGTG3'
5	OP-C02	5'GTGAGGCGTC 3'	10	OP-M15	5'GACCTACCAC3'
No	ISSR Primer code	Sequence	No	ISSR Primer code	Sequence
1	14A	5'CTCTCTCTCTCTCTTG 3'	3	HB-09	5' GTGTGTGTGTGTGC 3'
2	44B	5'CTCTCTCTCTCTCTGC 3'	4	HB-11	5'GTGTGTGTGTGTGTGCC 3'

Table(3): List of the used RAPD and ISSR primer names and their nucleotide sequences.

Statistical analysis:

The experimental design was factorial experiment between compost fertilizer and the fennel varieties in split plots with three replicates. The compost fertilizers were arranged in the main plots, while the fennel cultivars were assigned at random in the sub plots. The data were statistically analyzed according to Steel and Torrie (1960) and L.S.D. at (5% level) for comparison the means of different treatments. The DNA bands generated by each primer were counted and their molecular sizes were compared with those of the DNA markers. The bands scored from DNA profiles generated by each primer were pooled together. Then the presence or ab-

sence of each DNA band was treated as a binary character in a data matrix (coded 1 and 0, respectively) to calculate genetic similarity and to construct dendrogram tree among the fennel cultivars under study. Calculation was achieved using Dice similarity coefficients (Dice, 1945) as implemented in the computer program SPSS-10.

Results and discussion

Effect of different compost levels on growth, fruits and Volatile oil production of fennel plant:

Data in Table (4) and Table (5) show that organic compost significantly increased fennel growth parameter, fruit production, volatile oil % and volatile oil yield per plant (ml) and per feddan (L) of fennel.

Level of compost	Plant height (cm)		F.W of herb (g)		D.W of herb (g)		No of umbels/plant		No of flowers/umbel	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
4 ton/fed	88.80	82.40	124.51	113.04	24.34	21.16	11.53	10.00	19.13	16.13
6 ton/fed	94.00	89.13	157.25	146.14	30.98	26.44	16.93	13.26	21.47	19.26
8 ton/fed	105.46	100.06	180.73	169.33	38.74	31.44	20.66	19.40	23.87	23.13
L.S.D at 5%	1.80	4.61	3.55	6.23	0.98	1.37	1.17	0.78	1.24	0.30

Table (4): Effect of different compost levels on growth of five cultivars of fennel during the two seasons 2011/2012 and 2012/ 2013 over all cultivars

Increasing compost level progressively and significantly increased the values of such parameters. Wherever the highest values of increase resulted by the highest level of compost (8 ton/Fed): plant height (105.46, 100.06 cm), herb fresh (180.73, 169.33g) and dry weights (38.74, 31.44g) of plant, number of umbels /plant (20.66,19.40) and number of flowers/umbel(23.87,23.13) in the first and second season respectively. On the other side the lower percentage of increase resulted by the lower level of compost (4 ton/Fed.): (88.80, 82.40 cm) for plant height, (124.51, 113.04g)

for fresh weight of herb, (24.34, 21.16g) for dry weights, (11.53, 10.00) for number of umbels /plant and (19.13, 16.13) for number of flowers/umbel in the first and second season respectively.

Such results on fennel are in the same line with many researchers on different plants, (Kandil, 2002) on fennel (*Foeniculum vulgare* Mill.); (Khalil et al., 2002) on *Tagetes erecta*; (Khalil and El-Sherbeny, 2003) on three *Mentha* species, who reported that compost at different levels significantly increased the vegetative growth characters including plant height, number of branches, fresh and dry weight of herb during vegetative growth and flowering stage.

Level of compost	yield of fruits per plant (gm.)		yield of fruits per Fed (kg.)		Volatile oil (%)		Volatile oil yield per plant (ml.)		Volatile oil yield per Fed. (L.)	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
4 ton/fed	50.00	40.78	1100.00	897.16	1.62	1.85	0.82	0.76	18.11	16.69
6 ton/fed	56.73	49.22	1248.13	1082.84	2.07	2.32	1.25	1.16	27.40	25.56
8 ton/fed	61.00	54.97	1342.00	1209.41	2.39	2.62	1.52	1.46	33.43	32.06
L.S.D5%	1.67	1.85	36.79	40.62	0.06	0.05	0.07	0.05	1.45	1.13

Table (5): Effect of different compost levels on fennel yield traits as fruit yield, volatile oil % and volatile oil yield per plant (ml.) and /feddan (L) of fennel plant during the two seasons 2011/2012 and 2012/ 2013 over all cultivars :

Data in Table 5 revealed that the differences between the various levels of compost were significant in most cases. The highest level of compost (8ton /Fed.) increased yield of fruits per plant, yield of fruits per feddan, volatile oil (%),volatile oil yield per plant and volatile oil yield per Fed, by (61.00g, 1342.00 Kg, 2.39%, 1.52ml and 33.43L.) respectively in the first season and (54.97g, 1209.41Kg, 2.62%, 1.46ml and 32.06L.) successively in the second one, where as the values of increase due to the lower level of compost (4 ton/fed.) were [(50.00,40.78g), (1100.00,897.16Kg), (1.62,1.85%), (0.82,0.76ml) and (18.11,16.69L)]for the same parameter in the two seasons, consecutively. It is clear that volatile oil yield per plant (ml) and per feddan (L.) attained a parallel trend to volatile oil (%). The three compost levels significantly raised volatile oil yield. Raising compost levels progressively increased fennel volatile oil yield. These results agree with those obtained by (Ibrahim, 1999) on *Ocimum sanctum*; (Ibrahim and

Ezz El-Din, 1999) on catnip (*Nepta cataria L.*); (Khalil, 2002) on *Rosemarinus officinalis*; (Khalil et al., 2002) on *Tagetes erecta*; (Khalil and El-Sherbeny, 2003) on three *Mentha* species and (El-Sherbeny et al., 2005) on *Sidritis montana L.*, who mentioned that compost addition markedly improved volatile oil %, productivity and volatile oil yield.

2 Performances cultivars for growth , fruits and Volatile oil production of fennel plant over all levels of fertilizers at the two years :

Data in Table 6 and Table 7 show significant differences in plant height (cm/plant) among different cultivars. Netherland cultivar was generally the shortest (75.11 and 64.78cm) followed by Local cultivar (87.56 and 80.33cm) then Indian cultivar (91.22 and 82.78cm) after that German cultivar (108.33 and 108.66cm) and the tallest cultivar was Azoricum (118.22 and 116.11cm) in both seasons.

The varieties	Plant height (cm)		F.W of herb (g)		D.W of herb (g)		No of umbels/plant		No of flowers/umbel	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Netherland	75.11	64.78	118.22	106.71	22.56	19.29	14.00	10.89	13.78	13.33
Indian	91.22	82.78	119.32	110.51	28.71	20.60	15.67	13.89	20.00	17.66
Azoricum	118.22	116.11	202.25	189.42	39.00	34.82	17.11	16.78	24.67	22.55
German	108.33	108.66	235.15	212.97	45.80	39.91	13.67	12.00	23.11	19.66
Local	87.56	80.33	95.88	94.59	20.71	17.12	21.44	17.56	25.89	24.33
L.S.D 5%	2.01	3.30	3.23	3.48	0.89	0.50	1.01	1.40	1.84	1.06

Table (6): Performances cultivars for growth of fennel plant during the two seasons 2011/2012 and 2012/ 2013 using the three levels of fertilizers:

Concerning the fresh and dry weight, data in Table (3) represent that, in general, the Local cultivar recorded the lowest value (95.88 and 94.59 g) in fresh weight and (20.71 and 17.12g) in dry weight in both seasons. While German cultivar recorded the highest value (235.15 and 212.97 g) in fresh weight and (45.80 and 39.91g) in dry weight in the first and second season respectively. Taking number of umbels (umbels/plant) into consideration, data in Table (3) reveal significant differences among all cultivar. General-

ly, it was noticed that Local cultivars was superior in number of umbels/plant (21.44 and 17.56 umbels/plant). Moreover, it was found that Netherland and German cultivars were the lowest in number of umbels/plant (14.00 and 10.89 umbels/plant), (13.67 and 12.00umbels/plant) in the first and second season respectively. As for the number of flower (flowers/plant), it was found also that Local cultivar was superior in number of flowers/plant (25.89 and 24.33 flowers/plant) and Netherland cultivar recorded the lowest value (13.78 and 13.33 flowers /plant) in both seasons.

The cultivars	yield of fruits		yield of fruits		Volatile oil (%)		Volatile oil yield		Volatile oil yield	
	per plant (gm.)		per feddan (kg.)				per plant (ml.)		Per fed. (L.)	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Netherland	49.50	42.59	1089.00	936.96	1.61	1.88	0.81	0.81	17.75	17.82
Indian	55.94	44.91	1230.70	988.04	1.92	2.14	1.08	0.98	23.78	21.51
Azoricum	73.00	57.83	1606.00	1272.33	2.82	3.03	2.14	1.77	46.98	38.92
German	47.55	41.24	1046.20	907.38	1.99	2.20	0.96	0.93	21.10	20.39
Local	53.55	55.04	1178.20	1210.97	1.81	2.04	1.00	1.15	21.95	25.23
L.S.D at 5%	2.47	1.59	54.29	35.02	0.04	0.06	0.04	0.05	0.82	1.02

Table (7): performances cultivars for fruits production, volatile oil % and volatile oil yield per plant (ml.) and per feddan (L.) of fennel plant during the two seasons 2011/2012 and 2012/ 2013 using three levels of fertilizers:

The data on fruits yield/plant and fruits yield/feddan as shown in Table 4, show significant differences in these parameters. It was noticed that the German cultivar showed the lowest value of both parameters (47.55 and 41.24g) for fruits yield/plant and (1046.20 and 907.38 Kg) for fruits yield/feddan in both seasons. Furthermore, Azoricum cultivar showed the highest value in the same parameters (73.00 and 57.83g) for fruits yield/plant and (1606.00 and 1272.33 Kg) for fruits yield/feddan in both seasons. Table 4, demonstrate the volatile oil of the cultivar. It is generally noticed that, the Azoricum exhibited the highest values in volatile oil percentage (2.82 and 3.03%) in the first and second seasons respectively. Moreover, Netherland cultivar exhibited the lowest values (1.61 and 1.88%) in both seasons. Concerning volatile oil yield/plant and volatile oil yield/feddan, data in table 4, show that generally speaking, the Azoricum cultivar was the highest values (2.14 and 1.77 ml) for volatile oil yield/plant and (46.98 and 38.92L) for volatile oil yield/feddan in the first and second seasons respectively. Also, Netherland cultivar was the lowest values (0.81 and 0.81ml) for volatile oil yield/plant and (17.75 and 17.82 L) for volatile oil yield/feddan in the first and second seasons respectively. The results revealed that Azoricum cultivar was the highest values in volatile oil percentage, fruits yield/plant and fruits yield/feddan. It is in the same line with Lal (2007), Lopes et al. (2010) and Safaei et al. (2011) which reported that there is a positive and significant correlation between volatile oil content and grain yield of fennel. It is clear that, the growth of different cultivars of fennel could be arranged in a descending order as follows; Azoricum; German; Local; Indian then Netherland from production point of view. Generally, it can be concluded that Azoricum and German cultivars were the suitable for sandy soil

conditions.

The mean effect interactions at the different compost levels and the cultivars of fennel on growth, fruits and volatile oil production at the two years:

Tables 8 and Table 9 conclude that, plant height (cm) of the studied cultivars. Significant differences were noticed among compost levels and the studied varieties. Also, the Azoricum cultivar combined with the highest level of compost fertilizer (8 ton/fed) were superior, it recorded (125.33, 122.00cm) in first and second season respectively. While Netherland cultivar was the shortest (66.00, 55.67cm) when applied the compost fertilization level at (4 ton/fed) in first and second season respectively. Generally, increasing compost level led to increase plant height in both seasons. There was a significant interaction between compost levels and cultivars of fennel for fresh and dry weight, (Table 8). German cultivar when applied the highest level of compost (8 ton/fed) appeared the highest value (266.56, 239.10g) for fresh weight and (50.60, 44.93g) for dry weight in first and second seasons respectively. Fresh and dry weight was gradually reduced when compost level was reduced. The results found that Local cultivar combined with compost fertilizer level (4 ton/fed) exhibited the lowest value (74.13, 66.07g) for fresh weight and (15.40, 11.90g) for dry weight in first and second seasons respectively. As for number of umbels/plant and number of flowers/plant, it was noticed that Local cultivar was superior in number of umbels/plant (27.00, 23.66) and in number of flowers/plant (29.00, 29.00) in both seasons. At the same time, German cultivar recorded the lowest value (9.67, 7.00) for number of umbels/plant and Netherland cultivar recorded the lowest value (11.67, 10.33) for number of flowers/plant in both seasons.

Level of compost	The cultivars	Plant height (cm)		F.W of herb (g)		D.W of herb (g)		No of umbels /plant		No of flowers/umbel	
		1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
4 ton/fed	Netherland	66.00	55.67	93.00	83.37	17.60	15.00	10.67	8.00	11.67	10.33
	Indian	81.67	71.00	102.46	85.80	20.70	16.56	11.67	10.00	17.33	15.00
	Azoricum	116.67	111.33	155.23	144.16	29.03	26.50	11.67	12.00	22.33	18.66
	German	101.33	104.33	197.73	185.83	39.00	35.86	9.67	7.00	21.33	16.00
	Local	78.33	69.67	74.13	66.07	15.40	11.90	14.00	13.00	23.00	20.66
6 ton/fed	Netherland	75.00	65.00	124.63	110.16	23.36	19.53	14.00	10.66	14.67	14.00
	Indian	90.67	80.33	114.16	113.40	22.26	20.90	16.00	13.00	20.00	18.00
	Azoricum	112.67	115.00	212.13	197.66	41.33	35.10	17.00	15.66	24.33	22.33
	German	105.33	107.33	241.16	214.00	47.80	38.93	14.33	11.00	22.67	18.66
	Local	86.33	78.00	94.17	95.50	20.13	17.73	23.33	16.00	25.67	23.33
8ton/fed	Netherland	84.33	73.67	137.03	126.60	26.70	23.33	17.33	14.00	15.00	15.66
	Indian	101.33	97.00	141.33	132.33	43.16	24.33	19.33	18.66	22.67	20.00
	Azoricum	125.33	122.00	239.40	226.43	46.63	42.86	22.67	22.66	27.33	26.66
	German	118.33	114.33	266.56	239.10	50.60	44.93	17.00	18.00	25.33	24.33
	Local	98.00	93.33	119.33	122.20	26.60	21.73	27.00	23.66	29.00	29.00
LSD at 0.05		3.48	5.72	5.60	6.03	1.54	0.87	1.79	2.43	3.18	1.83

Table (8): the mean interactions at the different compost levels and the fennel cultivars on vegetation growth during the two seasons 2011/2012 and 2012/ 2013:

Data in Table 9 revealed that Azoricum cultivar produced the highest value in yield of fruits per plant (79.67, 65.90gm), yield of fruits per feddan (1752.60, 1449.80kg.), volatile oil (3.50, 3.71%), volatile oil yield per plant (2.81, 2.36ml.) and volatile oil yield per feddan (61.75, 51.99 L) in both seasons at rate (8 ton/fed) of compost , While German cultivar recorded the lowest value in

yield of fruits per plant (44.33,34.86gm), yield of fruits per feddan (975.33, 767.07kg) and Netherland cultivar represented the lowest value in volatile oil (1.43 , 1.66%), volatile oil yield per plant (0.65 , 0.60ml.) and volatile oil yield per feddan (14.23 , 13.27 L) in both seasons at rate (4 ton/fed) of compost.

Level of compost	The cultivars	yield of fruits per plant (gm.)		yield of fruits per feddan (kg.)		Volatile oil (%)		volatile Oil yield per plant (ml.)		volatile Oil yield per fed. (L.)	
		1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
4 ton/fed	Netherland	44.50	36.20	979.00	796.40	1.43	1.66	0.65	0.60	14.23	13.27
	Indian	54.17	38.03	1191.60	836.73	1.65	1.88	0.89	0.72	19.51	15.77
	Azoricum	60.00	51.20	1320.00	1126.40	1.85	2.04	1.11	1.04	24.42	22.95
	German	44.33	34.86	975.33	767.07	1.71	1.91	0.76	0.67	16.65	14.74
	Local	47.00	43.60	1034.00	959.20	1.50	1.74	0.72	0.76	15.77	16.72
6 ton/fed	Netherland	48.67	42.93	1070.60	944.53	1.66	1.92	0.80	0.83	17.67	18.19
	Indian	53.67	47.00	1180.60	1034.00	1.87	2.10	1.01	0.99	22.22	21.85
	Azoricum	79.33	56.40	1745.30	1240.80	3.12	3.36	2.49	1.90	54.78	41.80
	German	48.00	41.23	1056.00	907.13	1.92	2.15	0.93	0.89	20.53	19.65
	Local	54.00	58.53	1188.00	1287.73	1.81	2.04	0.99	1.20	21.78	26.33
8ton/fed	Netherland	55.33	48.63	1217.30	1069.93	1.74	2.05	0.97	1.00	21.34	22.00
	Indian	60.00	49.70	1320.00	1093.40	2.24	2.44	1.35	1.22	29.63	26.92
	Azoricum	79.67	65.90	1752.60	1449.80	3.50	3.71	2.81	2.36	61.75	51.99
	German	50.33	47.63	1107.30	1047.93	2.34	2.54	1.19	1.22	26.11	26.77
	Local	59.67	63.00	1312.60	1386.00	2.13	2.34	1.29	1.48	28.31	32.63
	LSD at 0.05	3.48	5.72	94.03	60.64	0.07	0.10	0.06	0.08	1.42	1.77

Table (9): The mean interactions at the different compost levels and the fennel cultivars for fruits production, volatile oil % and volatile oil yield per plant (ml.) and /feddan (L) of fennel, during the two seasons 2011/2012 and 2012/ 2013:

It was found that increasing fertilization levels gave the highest values of the studied parameters. These results are in agreement with those obtained by Haridy et al. (2001) on lemongrass and El-Ghadban et al. (2002) on *Origanum majora*. In this respect, it is possible that the favorable effect of compost on growth characteristics may be due to their ability to enhance the physical, chemical and biological properties of the soil. A similar suggestion was made by Hanafy et al. (2002) on rocket plants. The ratios of volatile oil from all cultivars under this study were between (1.43 and 3.71%); our results are in line with (Miraldi, 1999) who mentioned that volatile oil ratio in sweet and bitter fennel samples were found on an average to be 3.26% and 1.47%, respectively. As known,

amounts of volatile oil in fennel, like other aromatic plants, can be influenced by a lot of factors such as climatic and environmental conditions, season of collection and the stage of ripening of the fruits (Arslan et al., 1989; Miraldi, 1999).

Table 10 showed the volatile oil composition (%) for five cultivars of fennel at rate of compost (6 ton /fed) in the second season. The results found presence of 11 compounds, five are monoterpene hydrocarbons comprising between (6.95%) in German cultivar volatile oil and (17.1%) in Local cultivar volatile oil and six oxygenated compounds comprising between (82.89%) in Local cultivar volatile oil and (93.04%) in German cultivar volatile oil.

Peak No.	components	Retention time(min)	Netherland	Indian	Azoricum	German	Local
Monoterpene hydrocarbons							
1	3-Carene	5.882	2.53	1.41	0.67	0.8	2.15
2	α -pinene	6.513	0.89	0.33	0.12	0.08	0.57
3	B-Myrecene	6.58	0.21	0.09	0.04	0.03	0.08
4	P-cymene	7.36	0.98	0.26	0.11	0.92	0.4
5	Limonene	7.418	9.81	9.05	6.19	5.12	13.9
	Total hydrocarbons		14.42	11.14	7.13	6.95	17.1
Oxygenated monoterpenes							
6	Fenchone	8.45	4.77	4.48	5.5	3.79	5.99
7	Camphor	9.352	0.33	0.16	0.14	0.07	0.25
8	Estragole	10.163	24.4	27.42	39.89	26.92	57.96
9	α -Fenchyl acetate	10.63	0.37	0.12	0.15	-----	0.45
10	Anisaldehyde	11.128	3.41	0.01	0.01	0.02	0.01
11	Trans anethole	11.44	52.29	56.67	47.18	62.24	18.23
	Total oxygenated		85.57	88.86	92.87	93.04	82.89
	Total identified		99.99	100	100	99.99	99.99
	Unknown		0.01	0.00	0.00	0.01	0.01

Table (10): Volatile oil composition (%) for five cultivars of fennel (*Foeniculum vulgare* Mill.), at rate of compost (6 ton /fed) in the second season.

The major components in fennel fruits volatile oil were Trans-anethole, it represented (52.29%, 56.67%, 47.18% and 62.24%) in Netherland, Indian, Azoricum and German cultivars respectively, the highest values of Anethole found in German cultivar, while the lowest values found in Local cultivar.

Estragole (= methyl chavicol) Compound undesirable (24.4%, 27.42%, 39.89% and 26.92%) in the same cultivars respectively, the highest values of Estragole found in Local cultivar, while the lowest values found in Netherland and German cultivars. Limonene represented (9.81%, 9.05%, 6.19% and 5.12%) in the same cultivars respectively, after that Fenchone at percentages (4.77, 4.48, 5.5 and 3.79) in the same cultivars respectively. Fenchone has a pungent and camphorate odour; it is present especially in bitter fennel. One of the main components of the fennel is 3-Carene which represents (2.53%, 1.41%, 0.67%, and 0.80%) in the same cultivars respectively. While, in Local cultivar the major components of its volatile oil were Estragole, Trans-anethole, Limonene, Fenchone and 3-Carene at percentages (57.96, 18.23, 13.9, 5.99 and 2.15) respectively. Furthermore, the minor compounds were α -Pinene, β -Myrecene, P-Cymene and Camphor at percentages (0.89, 0.21, 0.98, and 0.33) respectively, in Netherland cultivar. While their existed in Indian cultivar oil at percentages (0.33, 0.9, 0.26 and 0.16) respectively, in Azoricum cultivar oil at percentages (0.12, 0.4, 0.11 and 0.14) respectively, in German cultivar oil at percentages (0.03, 0.08, 0.92 and 0.07) respectively, and their percentages in Local cultivar were (0.57, 0.08, 0.4 and 0.25) respectively. Noteworthy differences were recorded in the percentage of Anisaldehyde an autoxidation product of Trans-anethole, ranged from 0.01% in Indian, Azoricum and Local cultivars to the intermediate 0.02% in German cultivar, up to the highest 3.41% in Netherland cultivar. Percentage of α -Fenchyl acetate ranged from 0.12% in Indian cultivar, 0.15% in Azoricum cultivar,

0.37% in Netherland cultivar up to the highest 0.45% in Local cultivar. However, it contents were not available in German cultivar volatile oil. Trans-Anethole, Estragole, Limonene and Fenchone were found to be main constituents in the studied cultivars (Table 10). Similar results were recorded by several researchers (Arslan et al., 1989; Charles et al., 1993). It can be seen in table 10 that Local cultivar volatile oil formed the highest percentage of Estragole, while the other cultivars volatile oil formed the highest percentage of T-anethole. These results are in the line with (Shalaby et al., 2011). It was reported that the chemical composition of bitter fennel volatile oil is very variable. The chemo cultivars and the environmental conditions cause this variability. The major components from these were found to be Methyl chavicol, Trans-anethole, Limonene, Fenchone, γ -terpinene, and piperitone oxide (Marotti. et al., 1994) *Foeniculum vulgare* var. presents great composition differences with varying populations with the aim of clarifying the status of var. *vulgare*, the proposed to subdivide it into three chemotypes according to their relative compositions (McDonald, 1999). They called them chemotype Estragole, chemotype Estragole/Anethole and chemotype Anethole. According to that our results divided into two chemotypes.

1- chemotype. Estragole (Estragole is the major compound) such as Local cultivar oil.

2- chemotype. Anethole (T-anethole is the major compound) such as Netherland, Indian, Azoricum and German cultivars volatile oil.

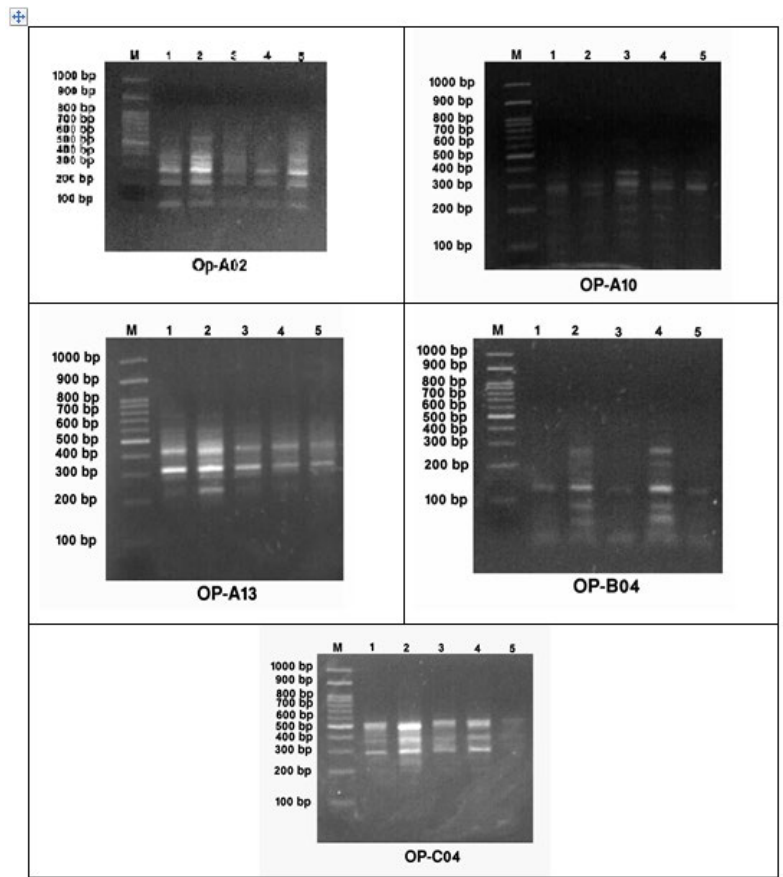
Molecular genetic identification

Randomly amplified polymorphic DNA (RAPD) markers

Table (11) and Figures (1 and 2) show the results of total amplified fragments (TAF), amplified fragments (AF) and specific markers (SM) for each cultivars of Fennel using RAPD-PCR analysis with ten random primers. A total number of 98 DNA frag-

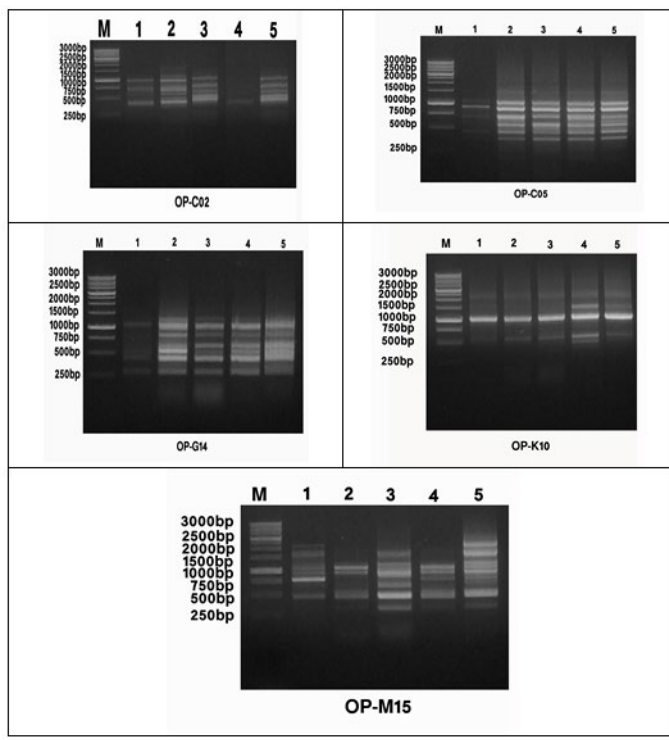
ments were detected, in which 60 (61.22%) were polymorphic fragments. However, 38 bands were common (monomorphic) for all cultivars. Polymorphism levels differed from one primer to another, i.e. The results found that (OP-C02, OP-M15, OP-B04, OP-C04 and OP-A10) primers exhibited high levels of polymorphism (90.91%, 78.57%, 75.00%, 72.73% and 70.00%) respectively. While, (OP-A02, OP-A13, OP-K10 and OP-C05) primers exhibited moderate level of polymorphism (62.50%, 55.56%, 62.50% and 54.55%), and primer OP-G14 represented the lowest level 37.50% as exhibited in Table (11). These results agree with the previously reported for other medicinal and aromatic species like *Lavandula angustifolia* (Echeverrigaray and Agostini, 2000) and *Ocimum gratissimum* (Vieria et al., 2001). The lowest num-

ber of polymorphic fragments was detected for primer OP-M15 (3 out of 14 amplified bands); while the highest number of polymorphic fragments was detected for primer OP-C02 (10 out of 11 amplified bands). Cultivar-specific markers generated from RAPD-PCR analysis are shown in Table (11). Thirty-one out of 98 RAPD-PCR fragments were found to be useful as cultivar-specific markers. The largest number of RAPD-PCR markers was scored for Indian cultivar (68 markers), while the lowest (49 markers) was scored for Netherland cultivar. In the meantime, the highest number of RAPD-PCR cultivar-specific markers was generated by primer OP-C04 (7 markers), while the lowest number (1 markers) was generated by primers OP-A13 and OP-B04. Moreover, OP-G14 was generated no RAPD-PCR cultivar-specific markers.



1-Netherland 2-India 3-Azoricum
4-German 5-Local

Fig. 1: RAPD-PCR analysis of five fennel cultivars cultivated under Egyptian sandy soil condition. (Second season)(Using ladder markers from 100bpto 1000bp)



1-Netherland 2-India 3-Azoricum
 4-German 5-Local

Fig. 2: RAPD-PCR analysis of five fennel cultivars cultivated under Egyptian sandy soil condition. (Second season)(Using ladder markers from 250bp to 3000bp)

Primers code	Range of M.S.	TAF	MF	PF	SM	Polymorphism (%)
RAPD primers						
OP-A02	128-440	8	3	5	3 (273)-(283)-(278)-(0)-(0)bp	62.5
OP-A10	121-348	10	3	7	3 (0)-(325)-(121)-(117)bp	70.00
OP-A13	210-625	9	4	5	1 (0)-(210)-(0)-(0)-(0)bp	55.56
OP-B04	65-264	8	2	6	1 (0)-(155)-(0)-(0)-(0)bp	75.00
OP-C02	254-1129	11	1	10	5 (574)-(1129,879,553,497)-(0)-(0)-(0)bp	90.91
OP-C04	205-481	11	3	8	7 (344)-(403,251,235,205)-(438,333)-(0)-(0)bp	72.73
OP-C05	325-1311	11	5	6	4 (0)-(558)-(449)-(0)-(1087,583)bp	54.55
OP-G14	174-1097	8	3	5	0	37.50
OP-K10	206-3092	8	3	5	2 (0)-(0)-(0)- (505)-(530) bp	62.50
OP-M15	168-2516	14	11	3	5 (0)-(0)-(599)-(784,339)-(2516,1228)bp	78.57
Total RAPD primers		98	38	60	31	
ISSR primers						
14A	279-332	2	2	0	0	0
44B	238-515	9	2	7	2 (0)-(0)-(429)-(0)-(238)bp	77.78
HB-09	207-518	8	3	5	2 (0)-(399)-(207)(0)-(0) bp	62.50
HB-11	102-631	15	1	14	7 (631,357)-(0)-(107)-(596,294,114)-(102) bp	93.33
Total ISSR primers		34	8	26	11	
Total		132	46	86	59	

Table (11): Species-specific RAPD and ISSR markers for five fennel cultivars genotypes.
 TAF = Total Amplified Fragments, MF= Monomorphic Fragments, PF= Polymorphic Fragments, SM= Specific Markers.

In conclusion, all of the ten primers used allowed enough distinction among the cultivars under study. These cultivar-specific markers can be used in subsequent experiments to detect molecular markers for polymorphic genes with economic importance among these and other cultivars. Similar finding were obtained in mints by Hassan (2005) and Momeni et al., (2006) and in other genera (Choi et al., 1999 and Benedetti et al., 2000).

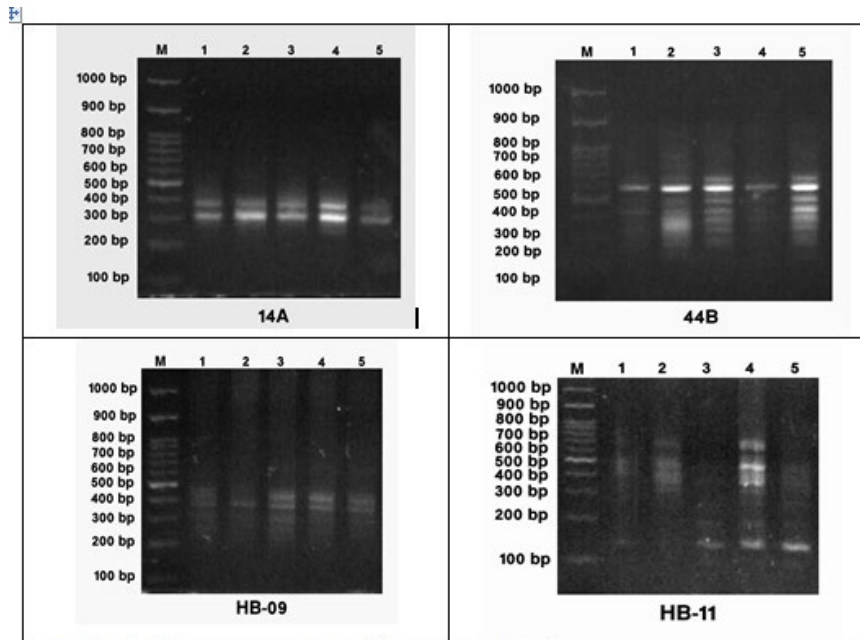
Genetic similarity and cluster analysis based on RAPD markers:

Genetic similarities among the five fennel cultivars were estimated according to the RAPD data by using UPGMA computer analysis (Table 12 and Fig. 1 and Fig. 2). Table 12 showed that the most two closely related cultivars were Indian and Local with the highest similarity index (1.000). On the other hand, the results indicated that the two most distantly related cultivars were Netherland and Local with low similarity index (0.411). The results showed that there was no similarity between Azoricum cultivar and Local cultivar. A dendrogram for the genetic relationship

among the five genotypes of fennel cultivars genotypes is exhibited in Fig. 4, which separated them into two major groups. The first group included Indian cultivar only, while the second group included two subgroups, the first subgroup involved German cultivar only and the other subgroup included Local, Azoricum and Netherland genotypes.

Inter Simple Sequence Repeats (ISSR) markers:

The four ISSR primers succeeded in amplifying DNA fragments for the five fennel cultivars genotypes (Fig.3). Polymorphism levels differed from one primer to another, i.e. HB-11 and 44A primers exhibited high levels of polymorphism (93.33% and 77.78%) respectively, while, HB-09 primer exhibited low level of polymorphism (62.5%) as exhibited in Table 11. The number of total amplified fragments (TAF), polymorphic fragments (PF), monomorphic fragments (MF) and specific markers (SM) for each primer of the four primers are shown in Table 11. 14A Primer showed two DNA fragments with molecular size ranging from 279 to 332bp (Fig.3 and Table 11), those two fragments were monomorphic, and there was not any polymorphic fragments or specific markers.



1-Netherland 2-India 3-Azoricum
4-German 5-Local

Fig. 3: ISSR-PCR analysis of five fennel cultivars cultivated under Egyptian sandy soil condition. (Second season)(Using ladder markers from 100bp to 1000bp)

44B primer showed nine DNA fragments with molecular sizes ranging from 238 to 515bp, seven fragments were polymorphic (77.78 %), and two of them were positive species- specific markers at 429bp for Azoricum genotype and 238bp for Local genotype. HB-09 primer showed eight DNA fragments with molecular size ranging from 207 to 518bp, five fragments were polymorphic (62.50 %), and two of them were positive species- specific markers at (399bp) for Indian genotype and at (207bp) for Azoricum genotype. HB-11 primer showed fifteen DNA fragments with molecular size ranging from 102 to 631bp, fourteen fragments of

them were polymorphic (93.33 %), and seven of them were positive species- specific markers at (631 and 357bp) for Netherland genotype, (107bp) for Azoricum genotype,(596, 294 and ,114bp) for German genotype and at 102bp for Local genotype.

Genetic similarity and cluster analysis based on ISSR markers:

According to ISSR results, the most two closely related cultivars were Indian and German (Table 13) with the highest similarity index (1.000). On the other hand, the most two distantly related cultivars were German and Local with low similarity index (0.550) and the two cultivars located very far were Azoricum and Local variety with similarity index (0.000).



Fig. (4): A dendrogram illustrates the genetic distance for five fennel cultivars genotypes based on RAPD data.

	Netherland	Indian	Azoricum	German
Netherland				
Indian	0.763			
Azoricum	0.552	0.936		
German	0.768	0.732	0.746	
Local	0.411	1.000	0.000	0.604

Table (12) Similarity value (Pairwise comparison) of five fennel cultivars genotypes based on RAPD data.

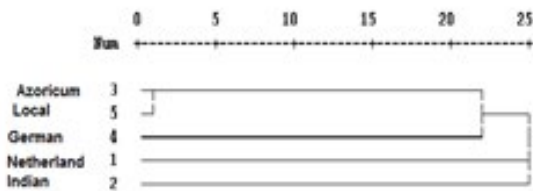


Fig. (5): A dendrogram illustrates the genetic distance for five fennel cultivars genotypes based on ISSR data.

	Netherland	Indian	Azoricum	German
Netherland				
Indian	0.835			
Azoricum	0.753	0.835		
German	0.920	1.000	0.908	
Local	0.835	0.679	0.000	0.550

Table (13) Similarity value (Pairwise comparison) of five fennel cultivars genotypes based on ISSR data

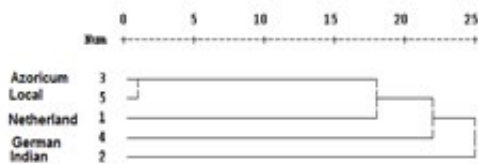


Fig. (6): A dendrogram illustrates the genetic distance for five fennel cultivars genotypes based on over-combination of RAPD and ISSR analysis.

	Netherland	Indian	Azoricum	German
Netherland				
Indian	0.860			
Azoricum	0.681	1.000		
German	0.904	0.893	0.888	
Local	0.604	1.000	0.000	0.650

Table (14) Similarity value (Pairwise comparison) of five fennel cultivars genotypes based on over-combination of RAPD and ISSR analysis.

Figure 5: indicated that the dendrogram revealed one main group of three cultivars including two subgroups. Subgroup 1 included both Azoricum and Local and subgroup 2 included German cultivar only. The remaining cultivars (Indian and Netherland) represented distant sequences.

Combined identification based on RAPD and ISSR analyses: Varieties distribution on the consensus tree according to the banding patterns of RAPD differed from that based on ISSR banding patterns, which may be due to that each technique, amplified different parts of the genome. So, it is better to use the combination of the banding patterns of the two techniques to use more segments of the genome that will increase the validity of the consensus tree. Results of the combined data as shown in Fig. 6 and Table 14 exhibited that the most closely related cultivars were Indian and both of Local and Azoricum with the highest similarity index (1.000). On the other hand, the two most distantly related cultivars were

Netherland and Local with low similarity index (0.604) and also the two cultivars located very far were Azoricum and Local variety with similarity index (0.000).

The results of the consensus tree indicated that the tree divided the cultivars into two main clusters, the first included cultivars Indian and German. The second one divided into two subgroups, the first one included cultivars Local and Azoricum and the other included Netherland. This study provides evidence that RAPD and ISSR polymorphisms could be used as efficient tools for the detection of similarities and phylogenetic relationships of the studied genotypes. The same conclusion was obtained by several authors (Alexander, 2002; Abdel-Tawab, et al., 2001 and Heikal, et al 2007). RAPD technique also is an effective technique in studying inter and intra specific variation in fennel. These results are in accordance with (Fu et al., 2003) who reported that out of 92 RAPD primers, 64 gave polymorphism which indicated that

51.2% of total diversity was among populations and 48.8 % within populations of *Changium smyrnioides* Wolff (Apiaceae)

The conclusion

- Increasing compost fertilizer level progressively and significantly increased the values of such studied vegetative and yield trials. Wherever the highest values of increase resulted by the highest level of compost (8 ton/ Fed.) in sandy soils gave the highest values of growth, fruits and volatile oil yield in fennel plant.

- Azoricum cultivar gave the highest values of vegetative growth, fruit yield and volatile oil content, while German cultivar gave the highest values of Anethole were main constituents in volatile oil, Estragole (= methyl chavicol) Compound undesirable found in Local variety, while the lowest percentage found in Netherland and German cultivars.

- Can expand the cultivation of cultivars fennel of German and Azoricum and breeding operations with a Local cultivar to produce a distinctive cultivar between the previous cultivars.

- According to the dendrogram of RAPD, ISSR and the combination between them the results revealed that Netherland existed alone in group or sub group of each dendrogram. This would explain the growth results of this study where Netherland showed the lowest value in most growth parameter (plant height, No of umbels/plant, No of flowers/umbel, volatile oil (%), volatile oil yield per plant, volatile oil yield per). Also, the results indicated that Netherland recorded the highest percentage of Anisaldehyde oil (3.41%). Such results are found for the German cultivar. From the dendrogram, it would be observed that German cultivar existed alone in a group or in a subgroup. This would explain the results indicated that German cultivar recorded the highest value of fresh and dry weight. Also α - Fenchyl acetate oil existed in all studied cultivars except Geman cultivar. It can be observed from similarity tables that Local cultivar and Azoricum cultivar represented distant sequences. It may explain that Local cultivar scored the highest value of estragole oil (57.96%) while, Azoricum recorded (39.89%) and Azoricum cultivar scored higher trans-anethole percentage than Local cultivar (47.18% and 18.23%) respectively. so that, they could be induced into a breeding program in the future for commercial production.

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