

EFFECT OF LICORICE, FENUGREEK EXTRACTS AND GA3 ON YIELD OF CARAWAY *Carum carvi* L.¹

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Abstract:

A field experiment was conducted at the Dept. of Field Crop Sciences ,College of Agriculture, University of Baghdad, Baghdad, Iraq on a silty loamy clayey soil during the two successive seasons 2007/08 and 2008/09 to investigate the effect of licorice and fenugreek extracts and GA3 on yield characters of caraway *Carum carvi* L. An RCBD design with three replicates was used under factorial arrangement by two factors, first factor levels were water, licorice ,fenugreek extracts and GA3.Second factor levels, were the stage of spraying for one time at vegetative, flowering stage and vegetative+ flowering stages. The results revealed that spraying of GA3 at flowering stage gave the highest conc. of carvone of (53.08%) for each season and oil of 3.00%, in the second season. The spraying at vegetative+ flowering stage gave the highest oil% of 2.17 in the first season. But in the second season was 2.79% under the spraying on vegetative stage. The fenugreek was superior in fruit yield and essential oil yield of 502.7 and 530.74 kg.ha⁻¹ and 9.74 and 13.78 L.ha⁻¹. For each season, respectively. The spraying on vegetative stage had the highest fruit yield and essential oil yield of 486.8 and 485.19kg.ha⁻¹ , 10.43 and 13.68L.ha⁻¹. For each season, respectively. It was concluded that plant extracts were effective in increasing the yield traits.therefore; it could be used as plant growth regulators.

تأثير مستخلصي (عرق السوس والحلبة) والجبرلين في حاصل الكراوية *Carum carvi* L.

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المستخلص:

نفذت تجربة حقلية في حقل قسم علوم المحاصيل الحقلية-كلية الزراعة جامعة بغداد في تربة طينية مزيجة غرينية خلال موسمي ٢٠٠٧ و٢٠٠٨، على التوالي لتقييم تأثير الرش بمستخلصي عرق السوس والحلبة ومقارنتها بالرش بالماء والجبرلين في صفات حاصل الكراوية. استخدم تصميم القطاعات العشوائية الكاملة بثلاثة مكررات ضمن ترتيب التجارب العاملية بعاملين. اذ تضمن الاول الماء و عرق السوس والحلبة والجبرلين، اما العامل الثاني فقد تضمن وقت الرش في المرحلة الخضريه وفي مرحلة الازهار وفي المرحلة الخضريه+مرحلة الازهار. اظهرت النتائج ان معاملة الرش الجبرلين في مرحلة الازهار اعطى اعلى معدل لتركيز مركب الكرفون ٥٣.٠٨ %، لكلا الموسمين، بالتتابع ونسبة زيت طيار ٣.٠٠ % في الموسم الثاني. اما الرش على المجموع الخضري+المجموع الزهري فقد اعطى اعلى معدل لنسبة الزيت ٢.١٧ % في الموسم الاول بينما تفوقت معاملة الرش على المجموع الخضري بنسبة زيت طيار ٢.٧٩ % في الموسم الثاني. تفوق مستخلص الحلبة في حاصل الثمار والزيت الطيار بمعدلات ٥٠٢.٧ و ٥٣٠.٧ كغم.هـ^{-١}، و ٩.٧٤ و ١٣.٧٨ لتر.هـ^{-١}، لكلا الموسمين، بالتتابع. امتلكت معاملة الرش على المجموع الخضري اعلى معدل لحاصل الثمار والزيت الطيار ٤٨٦.٨ و ٤٨٥.١٩ كغم.هـ^{-١}، و ١٠.٤٣ و ١٣.٦٨ لتر.هـ^{-١}، لكلا الموسمين، بالتتابع. يمكن ان يستنتج ان مستخلصي الحلبة وعرق السوس تأثير فعال في زيادة الحاصل لذا يمكن ان يستخدم كمحفزات لنمو النبات.

1: Part of dissertation of the first author.

Introduction:

Caraway (*Carum carvi* L.) belongs to the traditional minor crops which have been grown in Bohemia since the end of the 19th century. Harvesting area has fluctuated around 2,500 ha in recent years, with a yield of about 0.8 t/ha on average (Kamenik, 2001). A large part of the seed production is exported. This plant has been used since ancient times especially in the treatment of digestive disorders which was known worldwide as antibacterial (Singh et al., 2002), antiulcerogenic (Khayyal et al., 2001) and antiproliferative (Nakano et al., 1998). Caraway fruits are traditionally used to treat diabetes, cardiovascular diseases and hypertension (Eddouks et al., 2002). The aetheric oil, which is extracted from the seed, is used in the processing of perfume, liqueur and pharmaceutical products. This reflects the importance of carvone, the main component of volatile oil (Laribi et al., 2009 & 2010), the aromatic ketone, carvone, is the essential component of the oil.

The oil content of caraway seed fluctuates considerably through the year under the influence of climatic conditions and the degree of maturity of the harvested product (Dijkstra and Speckmann, 1980).

This component is produced commercially in the Netherlands and Germany as antisprouting of potato (Oosterhaven et al., 1995) and has medicinal properties (Iacobellis et al., 2005). The flowering initiation occurred as multicomponents and multisteps mechanism without the intervene of endogenous and exogenous factors, depending on the action of phytohormones and sugars (Samuoliene et al., 2008).

GA3 improved the performance of the plant more efficient, some of the characteristics of vegetation (the source) and thus positively reflected in volatile oil% through the revitalization of the effectiveness of the enzymes, especially Nitrate Reductase enzyme and improve the efficiency of photosynthesis (Shah, 2007). Many literatures referred to the effectiveness of plant extracts in the growth of many crops, where the natural product

that was consisting of fenugreek, tea and rosemary was effective in improving crops and protection them from diseases (Hochenhull et al., 2007). Spraying the onion plants, or soaking seeds with licorice extract gave the highest leaf area (Al-Marsoumi and Al-Sahaaf, 2001). Furthermore, spraying cucumber plant with the same extract increased the leaf area and the number of branches and the total content of chlorophyll pigment in the leaves (Hussein, 2002 & AL-Jebouri et al., 2010). However, licorice extract had significant effect in reducing physiological cracking of the fruits of pomegranate and reduced the damage caused by the blight of the sun and increased the marketable fruits (Al-Sahaaf et al., 2002).

Spraying the carnation plants with licorice extract at conc. of 3 g / liter led to increase lengths of plants, stem diameter and increasing the longevity of leaf and cut flowers of carnation (Muhammed-Sharif, 2002). There had been several attempts to find an alternative to chemical compounds without side effects in humans and the environment, especially, those were used with medicinal and aromatic plants. Therefore, this experiment was carried out to know the impact of the fenugreek and licorice extracts spray at different stages time of growth that caused the effects compared with GA3 and water on yield of caraway.

Materials and methods:

Field application:

A field trial was carried out in the experimental field of Field Crops Dept., College of Agriculture, University of Baghdad / Abu Ghraib, for two seasons (2007 & 2008), arranged as a factorial experiment in Randomized Complete Blocks Design (RCBD) with three replicates. The study included two factors, the first factor levels were GA3, fenugreek, licorice with conc. of 500 mg . L⁻¹ for GA3 and 500 mg . L⁻¹ for fenugreek and licorice respectively and water as control, while the second factor levels were the spraying on shoot after 45 days from emergence (time I).

The second time was spraying on stage of flowering in the same concentrations above. Third time was spraying on shoot + flowering stage at the concentration of (250 +250) mg . L⁻¹ for GA3, fenugreek and licorice, respectively. The experiment soil was plowed, and then partitioned to plots of (3 × 3) m, which included (4rows). The distance between rows was (75) cm and the distance between plot and another was (0.5 m) and between blocks was (1m). Seeds of caraway sowed on (5/11/2007) in the first season and (27/10/2008) in the successive season, at depth of (1-2) cm a long of drills (rows).

The experiment was fertilized as recommended, as nitrogen fertilizer was added in two batches after the first emergence and the second after 45 days of emergence, and the phosphate fertilizer was added in the form of triple superphosphate (52% P₂O₅) incorporately in soil, at rate of 70 kg. P / ha. Soil irrigation and weeding were conducted out as required for this crop. Plants were harvested on 30/5/2008 and 31/5/2009 for the two seasons, respectively after the maturity of the fruits (turned brown), before the fully dryness.

Extracts processing:

Powders of licorice and fenugreek were bought from the local market in adequate quantities. 100 grams of each powder were added to 150 ml distilled water and placed in an electric mixer for one minute. Then, the extract was filtered by filter paper (Whatman no.1) using the suppression Bouchner fennel to get rid of plant residues in the volumetric flask.

Process was repeated several times to get the right amount. And then placed in a water bath for the concentration of their access to material resembled jelly or mortar. Then, they were put in the refrigerator at a temperature of 5 ° ± 2 until the preparation of concentrations studied in the experiment.

Extraction and quantification of the volatile oil:

Water Distillation Method:

The device used Clevenger connected flask (2 L), as weight of 50 grams of dry fruits that were crushed in electric miller, and then placed in a beaker and 500 ml of distilled water were added. The distillation process was done by heating the flask continuously for two and a half hour for each sample to extract the volatile oil from samples until formed two layers of water and oil. layers separated by oil collection tube, and water is at the bottom and oil to the top because it is lighter than water.

After the separation of oil of each sample put in dark sealed bottles. The amount of oil measured by a digital balance (1260MP-Sartorius, Germany), then bottles stored on the temperature (4 °) until the measurement of the proportion of oil and the estimate of some active components of oil (Rozek,2007).

Analysis of the components of volatile oil by GC-FID:

Essential oil was analyzed using a Hewlett-Packard 6890 gas chromatograph equipped with a flame ionic detector (FID) detector, a capillary injector, an auto sampler and a relay control for automated operation. Samples were injected (injection temperature, 270°C) into capillary column is a polar column HP Innowax (PEG) (30 cm × 0.25 mm i.d., liquid phase DB-1 film thickness 0.25 µm).The carrier gas was helium (He) and operated at split ratio of 10:1 with flow rate of 2.0 mL / min.The analyses were performed using the following temperature program: oven temperature isotherm at 35° C for 10min.,from 35 to 205° C. at the rate of 3°C min-1 and isotherm at 225°C during 10 min. The temperature of the incubator detector was 225 and 250° C, respectively (Zoubiri and Baaliouamer, 2010).

Statistical analysis:

The data was subjected for statistical analysis using the statistical analysis program Genstat. Means were compared using the application of two-way analysis of variance (ANOVA) followed by testing of less significant difference LSD for means. Differences amongst individual means were significantly considered at level of ($P < 0.05$).

Results:

Volatile oil (%):

Results in (table-1) showed a significant effect of the spraying materials in the oil %, whereas GA3 had the highest oil % of 2.6 and 3.00 %, followed by fenugreek extract 1.94 and 2.6 %, and licorice extract 1.94 and 2.4 %, for the two seasons, respectively. While spraying of water gave the lowest oil %, in the first season of 1.86 %, but it was outweighed on the licorice extract in the second season of 2.49 %. It was observed from the results of the table that there was a significant difference among the stages of spraying in oil %, the spraying at vegetative + flowering stage had highest oil % of (2.17 %), in the first season, followed by spraying on the vegetative stage of (2.14 %), but spraying at the vegetative stage was significantly superior in the second season of 2.79 %, followed by spraying at the vegetative + flowering stage (2.56), which did not differ significantly from spraying at the flowering stage (2.52), in the second season. Whilst spraying on the flowering stage gave lowest oil% (1.95%) in the first season.

The results of (table-1) revealed that there was a significant interaction between material and spraying stages in oil%, which spraying of GA3 at the vegetative + flowering stage gave the highest oil% (2.92%) in first season, while spraying of GA3 at vegetative stage gave the highest oil% (3.6%) in the second season, however, spraying of water at flowering stage gave the lowest percentage of oil of 1.78 and 2.35%, for each season, respectively.

Yield of fruits (kg / ha):

Results of (table-2) showed that the total yield of fruits ($\text{kg} \cdot \text{ha}^{-1}$) was significantly affected by spraying materials for both seasons. Whereas, spraying of fenugreek had the highest yields of fruits (552.7 and 530.74 $\text{kg} \cdot \text{ha}^{-1}$), followed by spraying of water treatment (418 and 440.12 $\text{kg} \cdot \text{ha}^{-1}$), but the treatment of spraying with licorice gave the lowest yields of the fruits reached to 377.2 and 336.93 $\text{kg} \cdot \text{ha}^{-1}$, for the two seasons, respectively. Results indicated significant differences amongst stage times of spraying in the total yields of the fruits, for both seasons.

That spraying at vegetative stage gave the highest yields of the fruits (486.8 and 485.14 $\text{kg} \cdot \text{ha}^{-1}$), followed by spraying at flowering stage (447.1 and 429.63 $\text{kg} \cdot \text{ha}^{-1}$), while treatment of spraying at both stages (vegetative + flowering stage) gave the lowest yields of fruits (350.1 and 355.37 $\text{kg} \cdot \text{ha}^{-1}$), for the two seasons, respectively. The results of the table revealed that there was a significant difference between the materials and stages of growth interaction in the yields of fruits, for both seasons. Spraying of GA3 at flowering stage gave the highest yield of fruits (532.6 $\text{kg} \cdot \text{ha}^{-1}$), in the first season, followed by spraying of fenugreek at vegetative stage (523.7 $\text{kg} \cdot \text{ha}^{-1}$).

In the second season, spraying of fenugreek at vegetative stage gave the highest yield (549.63 $\text{kg} \cdot \text{ha}^{-1}$), followed by spraying of the fenugreek at flowering stage (548.52 $\text{kg} \cdot \text{ha}^{-1}$), however, spraying of licorice at vegetative + flowering stage gave the lowest yields of fruits reached to 259.3 and 243.71 $\text{kg} \cdot \text{ha}^{-1}$, for the two seasons, respectively.

Yield of volatile oil ($\text{L} \cdot \text{ha}^{-1}$) :

Results of (table-3) stated a significant effect of spraying materials on volatile oil yield ($\text{L} \cdot \text{ha}^{-1}$) for the two seasons, where the spraying of fenugreek and GA3 gave highest yield of the volatile oil (9.74 $\text{L} \cdot \text{ha}^{-1}$)

for both of them in the first season, but spraying of fenugreek outweighed on GA3 of 13.78 (L . ha⁻¹) in the second season, followed by spraying of GA3 of 11.84 (L . ha⁻¹). However, spraying of water at first season or licorice at second season gave the lowest yield of volatile oil (7.82 and 8.10 L . ha⁻¹). Also, the results showed that yield of volatile oil was affected by growth stage spray.

Spraying at vegetative stage gave the highest yields of volatile oil (10.43 and 13.68 L . ha⁻¹), followed by spraying at the flowering stage (8.72 and 10.78 L . ha⁻¹), whilst the spraying at vegetative+ flowering stage had lowest yield of volatile oil (7.36 and 9.14 L . ha⁻¹), for the two seasons, respectively. Moreover, the results showed that there was a significant difference between the spray of materials and stages of spraying in yield of volatile oil (L . ha⁻¹). Whereas, the interaction of GA3 X vegetative stage gave the highest yield (13.48 and 18.03 L . ha⁻¹), while the interaction of licorice X vegetative+ flowering stage gave the lowest yield (5.42 and 5.61L . ha⁻¹),for each season, respectively.

GC-Analysis of volatile oil:

The results of chromatogram graphs (Fig.1-12) from gas chromatography analysis of the oil extracted from caraway seeds consisted of (8-11) compounds, which three of those compounds were identified by availability of standards. Spraying of GA3 had the highest concentration of carvone compound (53.08%), followed by spraying of fenugreek at vegetative stage gave concentration of 52.62%, then spraying of licorice at vegetative stage concentration of 52.16%, respectively. Whilst spraying of fenugreek on the shoot + flowering gave lowest concentration (36.13%).

Gas chromatography analysis stated that spraying with water had highest concentration of limonene (54.08 , 56.8 and 52.13%) sprayed at vegetation + flowering stage, respectively. Whilst spraying of GA3 and plant extracts did not

increase limonene. The spraying of fenugreek gave the lowest concentration of limonene reached to 40.01%. There is no trace of a compound carvocrol .spraying of water on flowering stage had concentrations 1.27 %, and the spraying fenugreek on the shoot gave concentration of 0.02 and the spraying of fenugreek on the shoots + flowering stage had concentration 6.61% and the spraying fenugreek on flowering stage had concentration was 7.03.it was obtained the highest concentration by spraying of licorice on flowering stage of 0.3%.

Discussion:

Transport of sucrose from leaves to apical meristem and increase in gibberellins concentration is observed during flowering induction (Samuoliene et al., 2008).

Evidence indicated that many of plant developmental and physiological processes are regulated in response to other signaling molecules, such as sucrose or gibberellins. Therefore, the flowering initiation acted as multicomponent and multistep mechanism and without endogenous and exogenous factors, depending on the action of phytohormones and sugars (Samuoliene et al.,2008).

It may be due to the reason that GA3 efficiently improve the performance of the plant, some of the characteristics of vegetation (the source) and thus positively reflected volatile oil% through the revitalization of the effectiveness of the enzymes, especially enzyme Nitrate Reductase and improve the efficiency of photosynthesis (Shah, 2007). On the other hand, It may be due to the different physiological needs associated with environmental determinants of each stage.

Fenugreek was outweighed due to it has increased the effectiveness of plant growth through some of the attributes (Tables 1 and 2) because its content compounds may have a role as hormones catalyst for plant growth, although GA3 had good performance of plant growth through some recipes, but the effect decreased probably because of the inhibitory effect of male

flowers (Jadoua and Atiyah, 1990).

Spraying with licorice root extract has led to increase the flowering% of onion plant (Al- Marsoumi and Al-Sahaaf, 2001) and cucumber (AL-Jebouri et.al.2010), these attributed to that licorice extract similar to behavior of GA3 in stimulating flowering, which contained mevalonic acid that improve the vegetative growth as a result of stimulating the enzymes that necessary to convert complex compounds into simple compounds, and exploited in the processing of the energy required for plant growth, this may be due to extract contains compounds, which could stimulate the emergence of floral buds.

Also, it may be due to the correlation between fruit yield components

characteristics and proliferative vegetative growth that is required different physiological and environmental factors in different stages of the spraying. It is known that yield of volatile oil affected by the oil % and fruits yield, the yield of fruits necessarily caused superiority in yield of volatile oil. The above spraying materials at vegetative were affected volatile oil % and yield of fruit. There was indicator that the different components of volatile oil affected by being different abiotic environmental determinants of their impact on genetic factors, since it is believed that stimulate the production of secondary compounds and the environmental factors which in turn owns physiological and environmental role in the prevention of photophosphorylation.

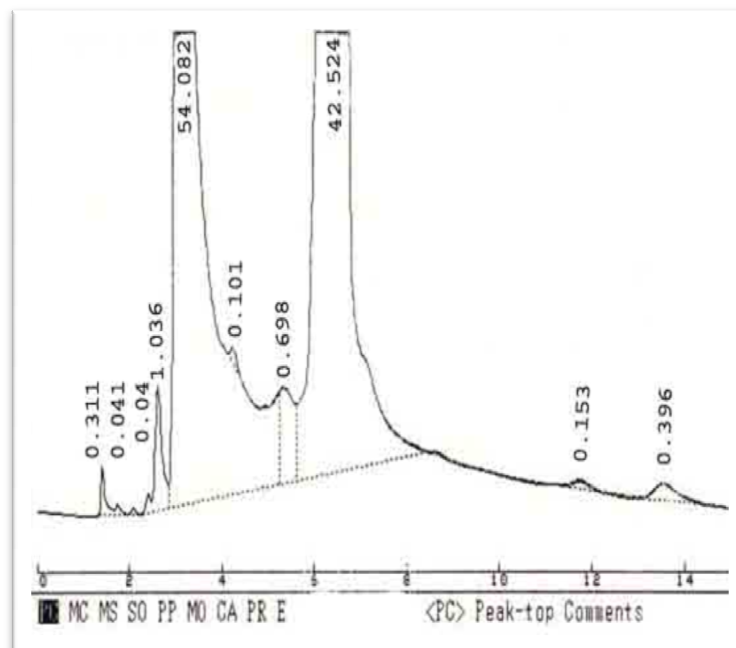


Figure-1: GC-Chromatogram of water at vegetative stage

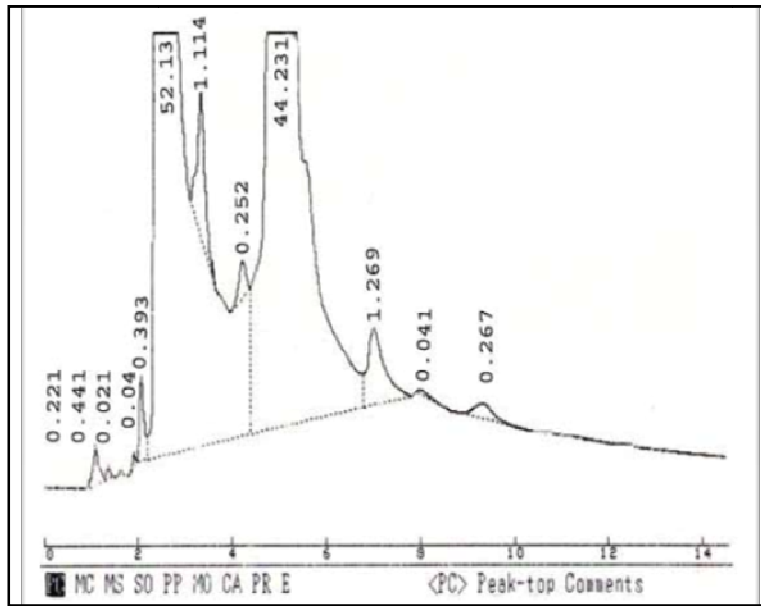


Figure-2: GC-Chromatogram of water at flowering stage

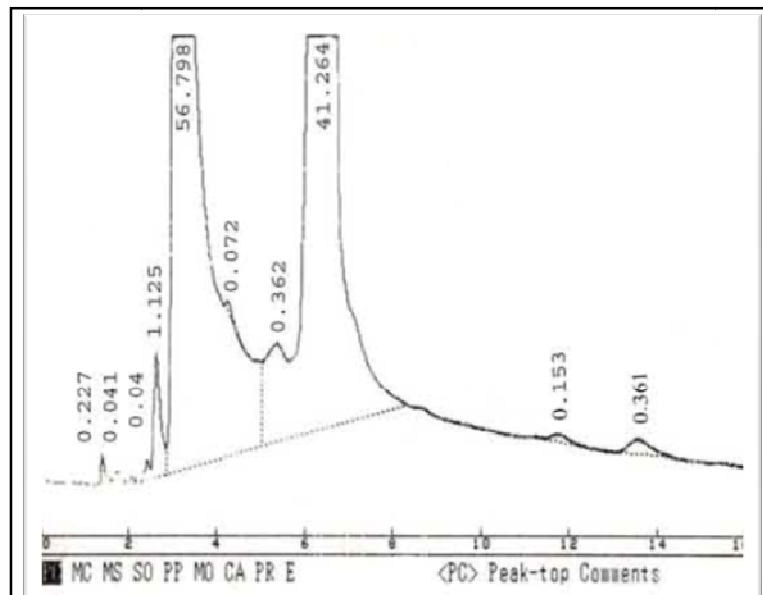


Figure-3: GC-Chromatogram of water at vegetative + flowering stage

Table-1: the effect of spray materials in different stage on volatile oil % for two seasons.

Spray stages	2007-2008				Means	2008-2009				Means
	water	fennugreek	GA ₃	licorice		water	fennugreek	GA ₃	licorice	
vegetative	1.95	1.89	2.75	1.96	2.14	2.35	2.6	3.6	2.6	2.79
flowering	1.78	1.95	2.14	1.93	1.95	2.73	2.5	2.54	2.3	2.52
vegetative + flowering	1.84	1.97	2.92	1.93	2.17	2.4	2.7	2.85	2.3	2.56
L.S.D interaction	0.04					0.23				
Means	1.86	1.94	2.6	1.94		2.49	2.6	3.00	2.4	
L.S.D	0.02				0.02	0.13				0.11

Table-2: the effect of spray materials in different stage on yield of fruits (kg/ha) for two seasons

Spray stages	2007-2008				Means	2008-2009				Means
	water	fenugreek	G-A ₃	licorice		water	fenugreek	G-A ₃	licorice	
vegetative	504.7	523.7	429.3	489.6	486.8	544.82	549.63	500.74	345.56	485.19
flowering	365.6	507.8	532.6	382.6	447.1	354.81	548.52	393.7	421.48	429.63
vegetative + flowering	383.7	475.7	280.7	259.3	350.1	420.74	494.07	262.96	243.71	355.37
L.S.D interaction	10.22					8.12				
Means	418.0	501.7	414.2	377.2		440.12	530.74	385.80	336.92	
L.S.D	5.90				5.11	4.69				4.06

Table-3: the effect of spray materials in different stage on yield of volatile oil (L/ha) for two seasons.

Spray stages	2007-2008				Means	2008-2009				Means
	water	fennugreek	G-A ₃	licorice		water	fennugreek	G-A ₃	licorice	
vegetative	9.89	9.91	13.48	8.41	10.43	13.43	14.29	18.03	8.98	13.68
flowering	6.5	9.90	8.19	10.23	8.72	9.7	13.72	10.00	9.7	10.78
vegetative + flowering	7.06	9.39	7.57	5.43	7.36	10.11	13.34	7.5	5.61	9.14
L.S.D interaction	0.49					0.9				
Means	7.82	9.74	9.74	8.04		11.08	13.78	11.84	8.10	
L.S.D	0.25				0.24	0.52				0.45

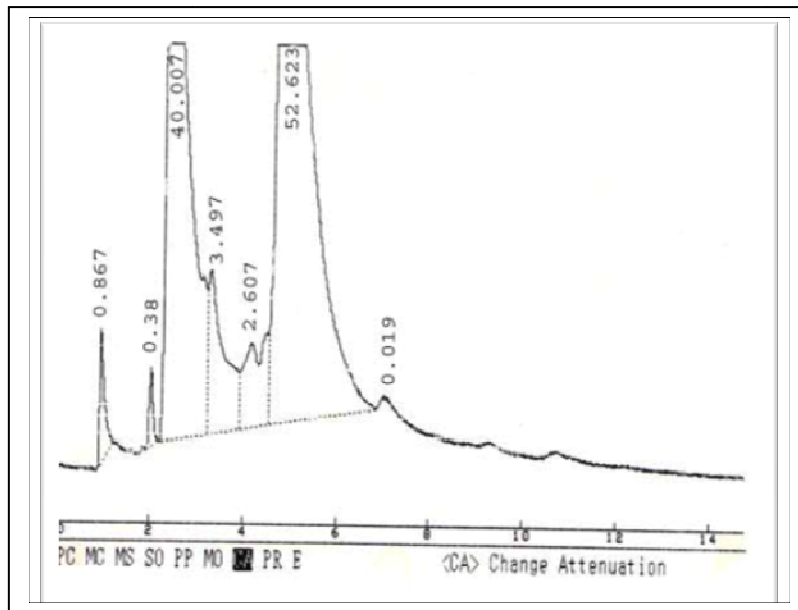


Figure-4: GC-Chromatogram of fenugreek at vegetative stage

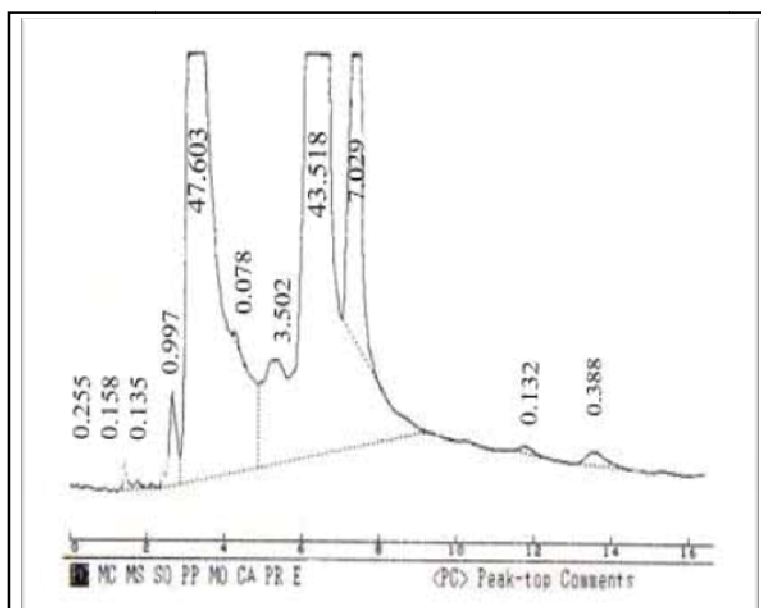


Figure-5: GC-Chromatogram of fenugreek at flowering stage

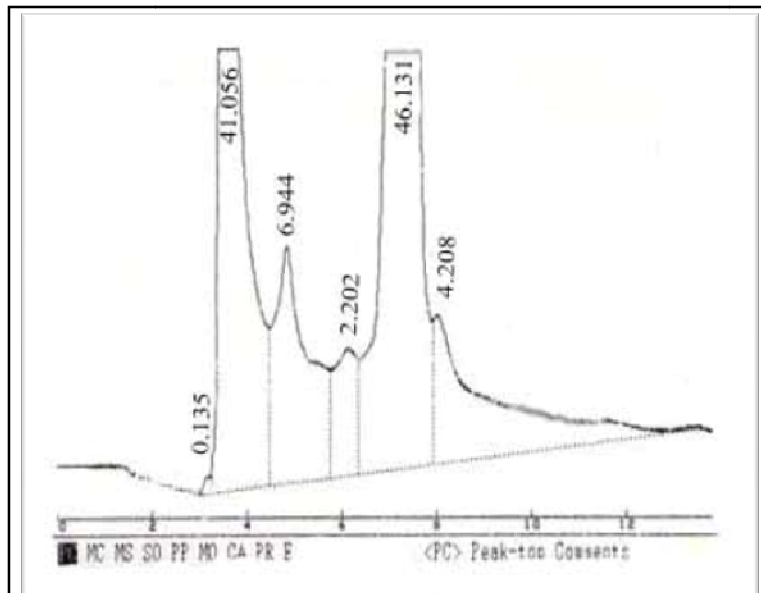


Figure-6: GC-Chromatogram of fenugreek at vegetative flowering stage

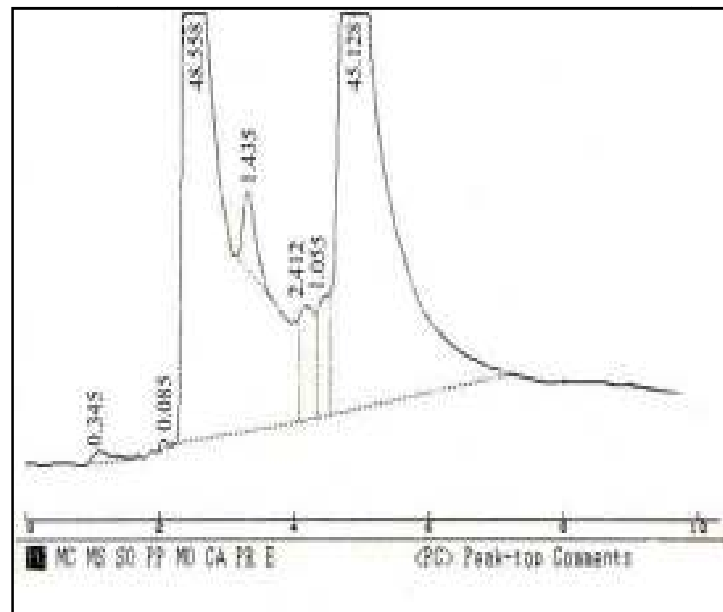


Figure-7: GC-Chromatogram of GA3 at vegetative stage

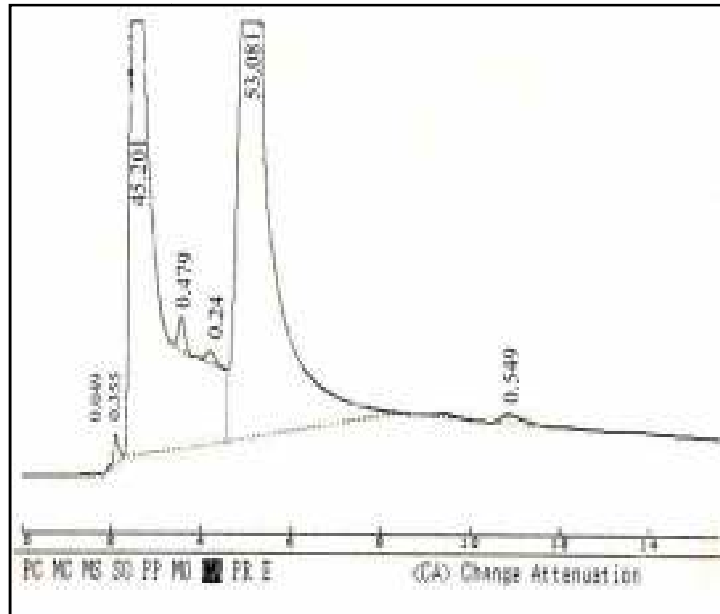


Figure-8: GC-Chromatogram of GA3 at flowering stage

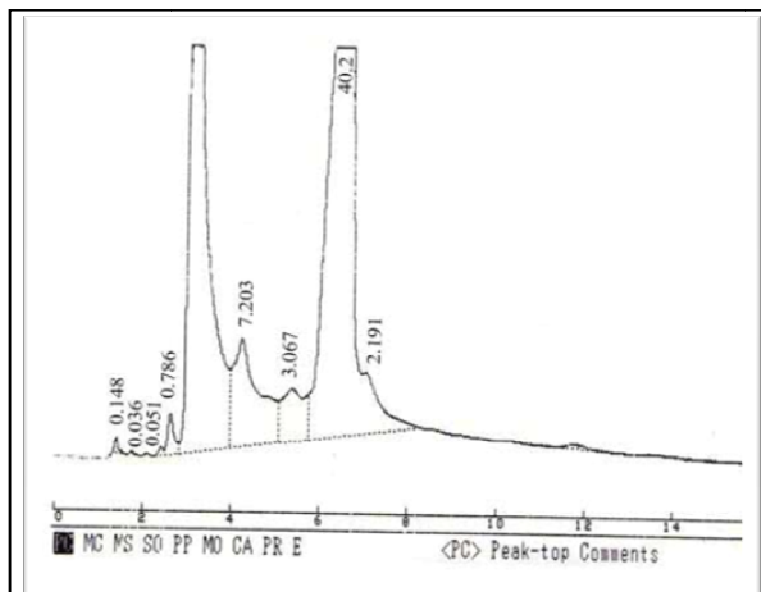


Figure-9: GC-Chromatogram of GA3 at vegetative + flowering stage

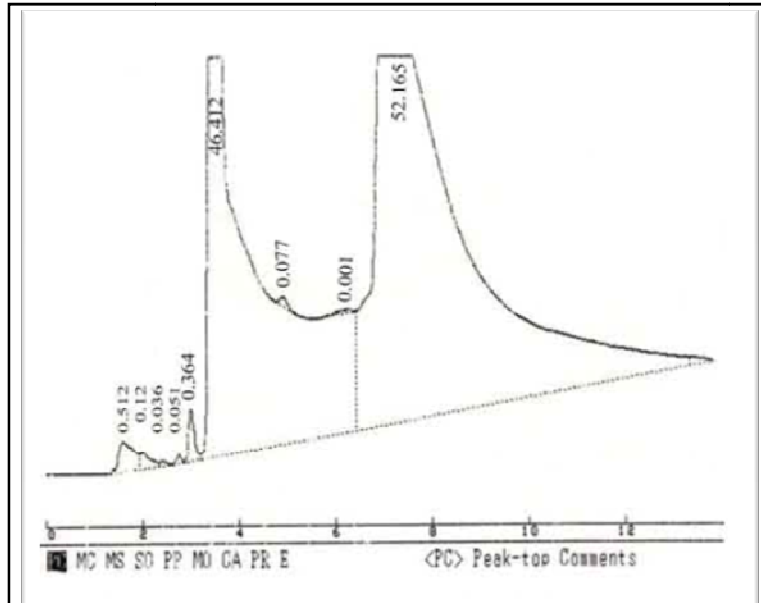


Figure-10: GC-Chromatogram of licorice at vegetative stage

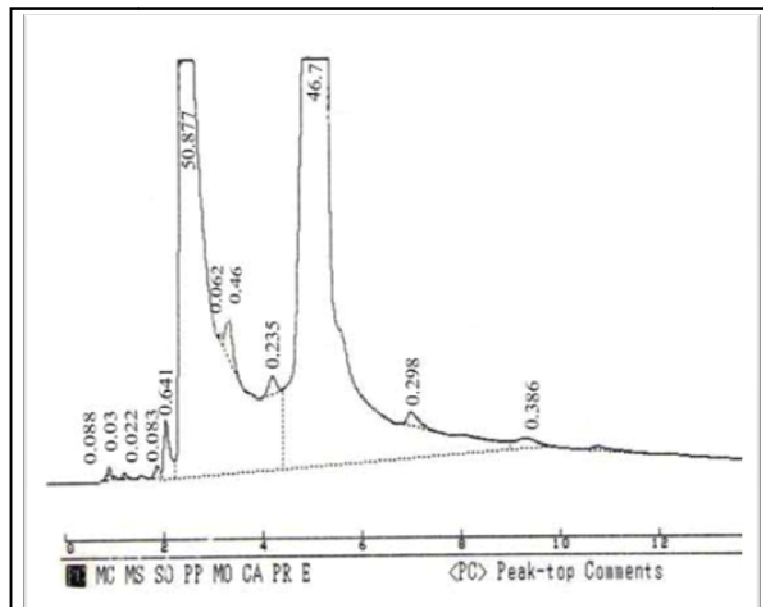


Figure-11: GC-Chromatogram of licorice at flowering stage

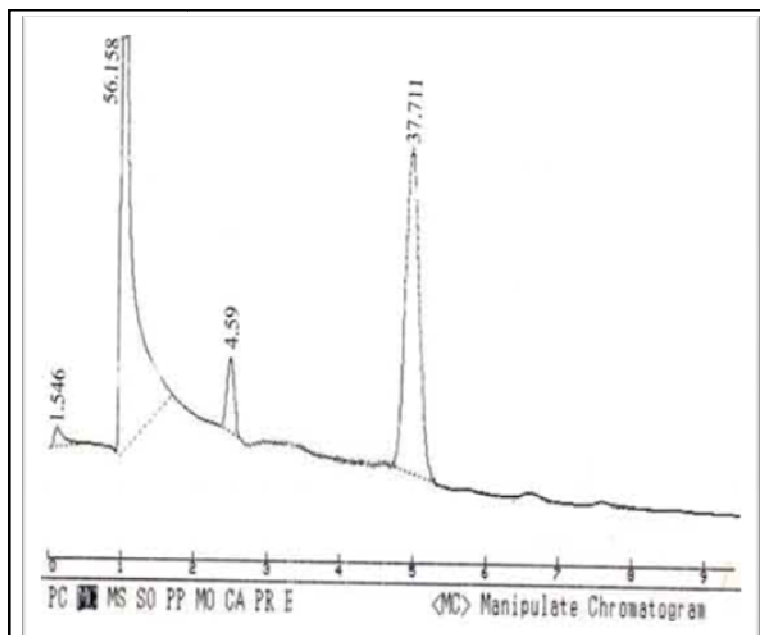


Figure-12: GC-Chromatogram of licorice at vegetative + flowering stage

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