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CHROMATOGRAPHIC SEPARATION AND IDENTIFICATION OF MANY FATTY ACID AND VOLATILE OIL COMPOUNDS FROM *CARUM CARVI* L. GROWING IN IRAQ

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Abstract

Natural products are considered one of the most important natural sources for producing effective compounds, especially in the various medical fields due to the nature of their composition, safety of use and ease of access to their sources in nature. One of these plants is caraway plant grown in Iraq. In this research, the fatty acids were separated from the seeds of caraway plant, calculating their concentration and diagnosing with GLC technology, which included: (Butyric acid, Undecanoic acid, Elaidic acid, Oleic acid, Lenolic acid, Arachidic acid, Linolenic acid, Erucic acid and Tricosanoi acid). Volatile oils were also separated from the seeds themselves by the light Kelvinger device and diagnosed with GLC technology. Chromatographic identification showed that the seeds contain a number of volatile oils, which included: (Camphor, Camphene, Linalool, Myrcine, Limonine, Terpinen and Sabinen).

Key words: caraway, fatty acids, volatile oils.

Introduction

Most wild and medicinal herbal plants contain active chemical compounds of great interest and importance resulting from secondary metabolism processes Inside the plant, and these compounds are known as natural products, and medicinal plants have been used for many years as a treatment for human diseases because they contain chemical components of effective therapeutic value, and according to the World Health Organization (WHO), about 70-80% of the world's population, in developing countries, relies mainly on traditional medicines as a primary means of health care, and the rest 20-30% of the population of developed countries indirectly benefit from natural products in health care (Ghourchian *et al.*, 2016). Treating many infectious diseases through bacterial antagonists of plant origin and they are highly effective without having any side effects as happens in chemically manufactured antibiotics, and in recent years one of the areas that attracted a great deal of attention is the potential pharmacological potential of antioxidants to control On diseases associated with oxidative damage, as many different plant extracts of plant chemicals that have antioxidant activity are very evident (Bhuiyan *et al.*, 2009). The need has become urgent in finding antibiotics that have a new synthetic chemical structure that differs from other antibiotics due to the spread of infectious diseases, as well as the development of resistance against antibiotics, due to its ability to form a biofilm envelope, which doubles its ability to resist antibiotics (Karaman *et al.*, 2003). Among the natural products used in the medical field are volatile oils that are used in the manufacture of antibiotics against bacteria and fungi, as well as fatty acid, which is an important source in the production of some vitamins and also acts as cholesterol regulators in the blood (Nascimento *et al.*, 2000). *Carum carvi* L. is one of the most important medicinal plants rich in active compounds, and it is an aromatic herbal plant belonging to the Apiaceae family, and because of its healing importance, the plant is often cultivated for the purpose of obtaining fruits or volatile oil (Lidefelt, 2014). It is an herbaceous plant around a height of between 30 and 80 cm, and its leg is thin, ribbed and the leaves are composed of filamentous leaves, positioned in parallel, white flowers are oval, and the fruits of the oval *Carum carvi* shape each fruit contains two small seeds inside it and have a deep root (Sedlakova *et al*., 2015). The Mediterranean basin is the original habitat of the plant, in addition to its spread in Asia, Europe and North Africa, as well as in narrow Norway and in mountainous regions, where it grows in sunny areas to a height of 2000 meters above

sea level. Sweden, the Netherlands, Norway, Germany, Poland, Russia, Morocco, Egypt, Syria and India grow One of the most productive countries for *Carum carvi* plants (Atal & Sood, 2015). The volatile oils, carvone and limonene, are the main chemical compounds of volatile oils for Carum carviplants, each with a ratio of 35% and 60%, respectively, as well as being rich in petroselinic acid, capric fatty acid, lauric, palmitic, linoleic, ter-terpinene, limonene and p-cymene, and the seeds contain 24.6 - 27.7% protein, vitamins C, E , B6, calcium, phosphorus, iron, magnesium, starch, sugars, other carbohydrates, tannins, phytic acid and nutritional fiber components (Laribi *et al.*, 2010). *Carum carvi* fruits also help increase the generation of breast milk and stimulate the mammary glands by eating hot water extract of the fruit powder, as well as appetizing and increases urine output and soothes abdominal pain, joints, muscles, treats hemorrhoids, treats chest diseases, cold, cough, and expectorant as well as helps to relax at psychological pressure, and the World Health Organization has recommended its use in pediatric medicines because of its analgesic effect, as it works to reduce the cumulative negative effects on the one hand and improve the taste on the other hand (Alessio, 2009), and it is also used in the treatment of acidity that occurs in the stomach (Eddouks, 2004). It is also included in the composition of the Cid water treatment, which is used to treat colic and expelling gases in children, in addition to using it as an anti bacterial (Iacobellis *et al.*, 2005). Caraway is recommended by the World Health Organization as it works to reduce the cumulative negative effects of chemicals and improve the taste on the other hand (Bown, 1995).

The aim of the research is to separate and diagnose fatty acids and volatile oils in the seeds of caraway plants and to determine their types in the seeds, for the purpose of studying them in other research

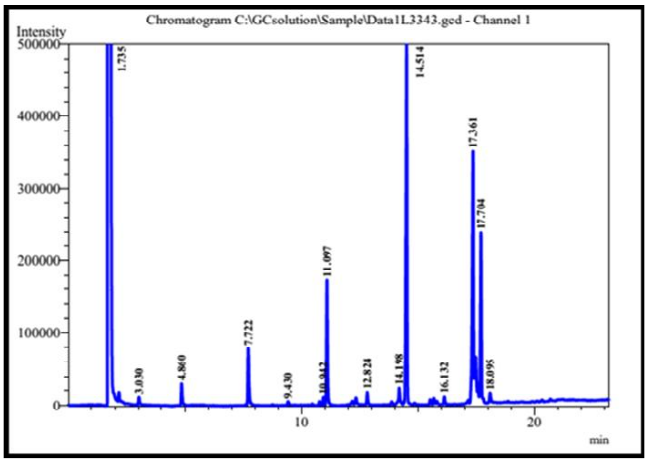

Fig. 1: standard curve of fatty acid compounds by GLC.

The taxonomic position of the plant (Sachan *et al.*, 2016):

Kingdom: Plantae

Subkingdom: Tracheobionta

Superdivision: Spermatophyta

Division: Magnoliophyta

Class: Magnoliopsida

Subclass: Rosidae

Order: Apiales

Family: Apiaceae

Genus: Carum

Species: carvi

Meterials and Methods

Collection of seeds:

Carum carvi seeds were collected from the Mosul Dam region and classified in the Directorate of the Medicinal Plants Development Project in the Mosul Dam of the Iraqi Ministry of Agriculture and Agricultural Reform. After that the seeds were cleaned from dust and so on, then they were ground and put in paper bags and kept in conditions away from moisture until use.

Preparation of Some Plant Extracts Using Continuous Soxholet Apparatus:

The seeds were crushed by an electrical mill, where 25 gm of the well-ground powder was placed in the Soxhlet batch system. 400 mL of ether petroleum was added to the flax seeds extracted oil. The extraction continued at a rate of 7 hours per day until the used solvent in the device became colorless. Finally, the extract was concentrated by a rotary vacuum evaporator (Al-Daody, 1998).

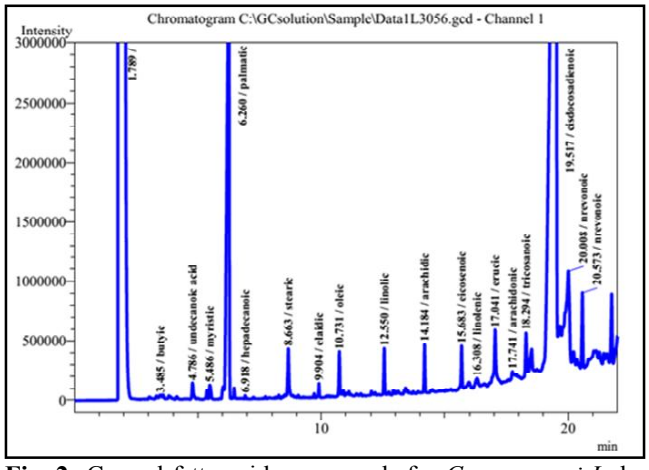

Fig. 2: Curved fatty acid compounds for *Carum carvi L.* by GLC.

Table 1: Fatty acids identified using the GCL technique for petroleum ether extract.

No.	Standard fatty acid compounds	Standard retention time (minute)	Petroleum ether	
			Millig-rams /ml	The retention time for the study sample (minute)
1	Butyric acid	3.485	0.0015	3.030
2	Undecanoic	4.786	0.0007	4.860
3	Elaidic	9.904	0.0002	9.430
4	Oleic	10.731	0.0001	10.942
5	Lenolic	12.550	0.0002	12.824
6	Arachidic	14.184	0.0002	14.198
7	Linolenic	16.308	0.0002	16.132
8	Erucic	17.041	0.002	17.361
9	Tricosanoic	18.294	0.0001	18.095

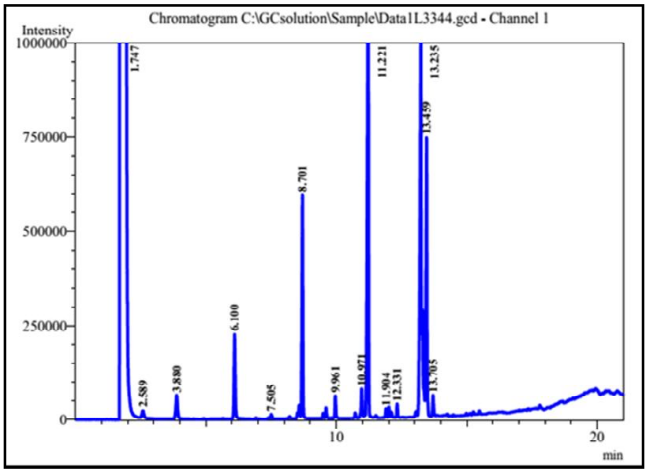

Fig. 2: Curved Essential Oils for *Carum carvi Seed*, Diagnosed with GLC Technology.

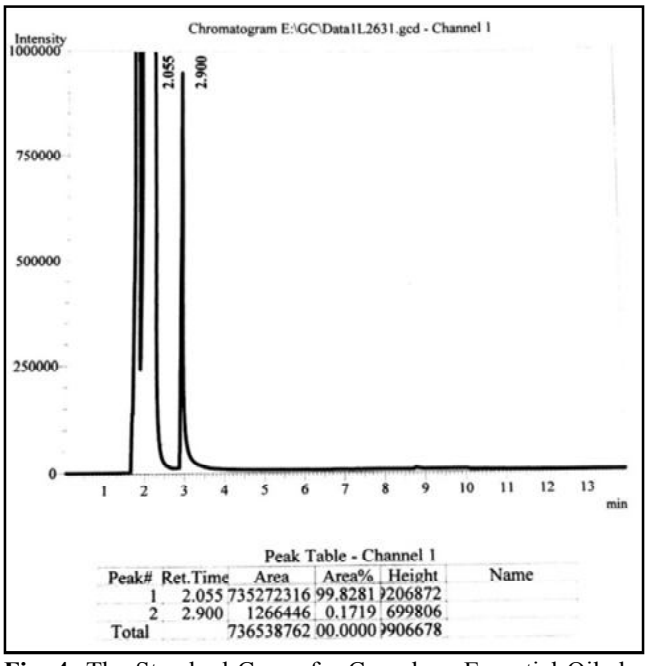

Fig. 4: The Standard Curve for Comphor Essential Oils by GLC Technology.

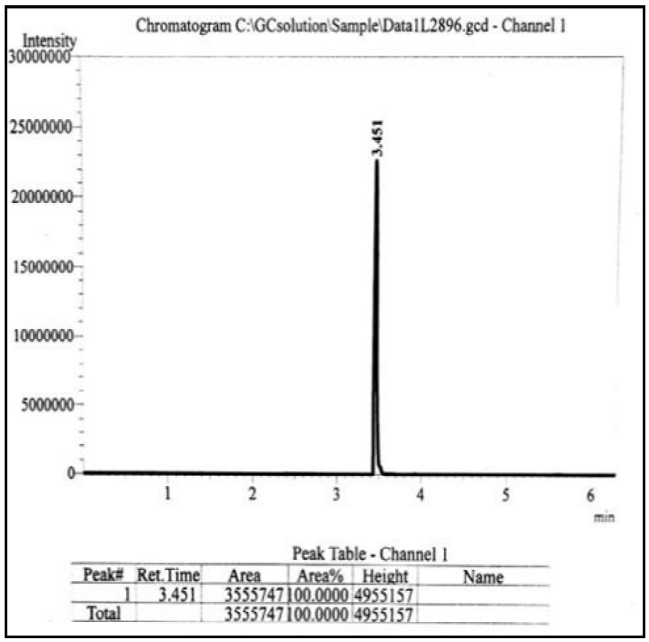

Fig. 5: The Standard Curve for Camphene Essential Oils by GLC Technology.

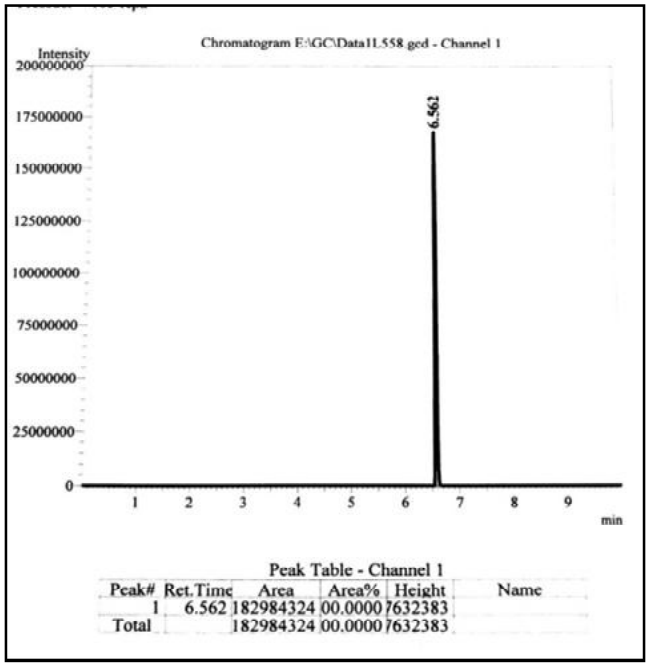

Fig. 6: The Standard Curve for Linalool Essential Oils by GLC Technology.

Volatile oils extracted by Clevenger pivot steam distillation machine

Volatile oil was extracted from the seeds of the study plant using a specialized Clevenger device to extract the light oil and connected with a volumetric flask with a capacity of 500ml, as 15g of vegetable seed powder was mixed with 200ml of distilled water, and then the distillation process was carried out using a tissue fabric and at a boiling point 100°C and the process of distillation lasted between 1-2 hours. Distilled water containing the volatile

oil was collected, and put in the separating funnel 100 ml of it and 50 ml of Di ethyl ether were added to it and for two stages, shake the mixture well and then left to settle, so two layers were produced: an upper layer containing the ether with the oil and a bottom water layer, So I took the upper layer and neglected the lower layer. After collecting the samples, anhydrous magnesium sulfate $MgSO_4$ was added at 3gm to dry the remaining water in the ether layer. The samples were then concentrated using the Rotary vacuum evaporator at a temperature of 25-30°C. The crude oil was placed in sealed bottles and kept in the refrigerator until identified (British,1985).

Saponification

Take 5 ml of the crude extract of the petroleum ether and added 100 mL of 1N (KOH), Heating the solution for 90 minutes at 100°C, Then, added 100 ml of distilled water and 50 ml ether solvent and put in the separating funnel, and tooked the aqueous layer and added the concentrated sulfuric acid H_2SO_4 until PH=2. In the end add 50 ml of ether and put again in the separating funnel and take the organic layer and retain well (Arthur,1972).

Identification of fatty acids and volatile oil by GLC technique

The separated fatty acids volatile oil were diagnosed in the laboratories of the Ministry of Science and Technology / Department of Environment and Water by GLC model (Shimanezo) Japanese (2010) using ionized flame detector and Using the poetic column type (SE-30) wavelengths (0.25mm 0.5um, 30m) The temperature was in the injection area and the detector (330 and 280) While the column temperature gradually starts from (120-280) m At a rate of 8°/ min using passive nitrogen gas as a carrier gas at a rate of 100 KP.

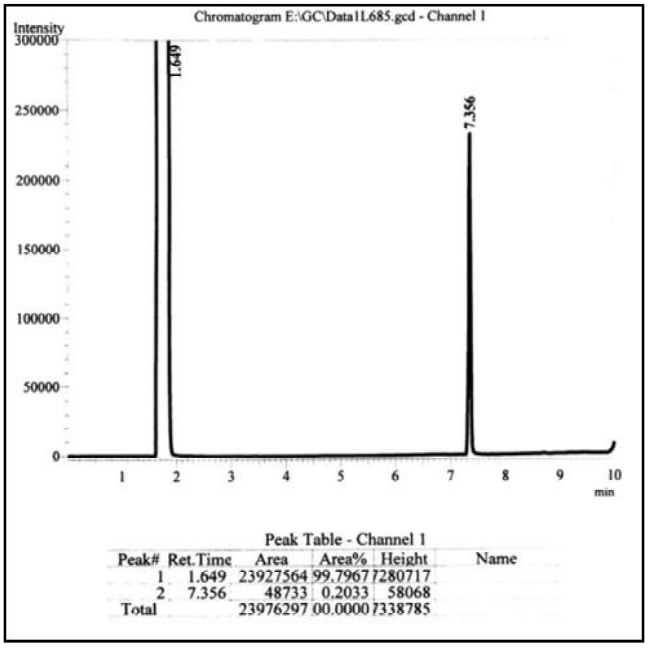

Fig. 7: The Standard Curve for Myrcine Essential Oils by GLC Technology.

Results and Discussion

The identification of fatty acid compounds of *Carum carvi* by GLC technique

The Identification of the petroleum ether extract after saponification by GLC showed the presence of the following fatty acids Fig. 1 and 2, table 1: Butyric acid at

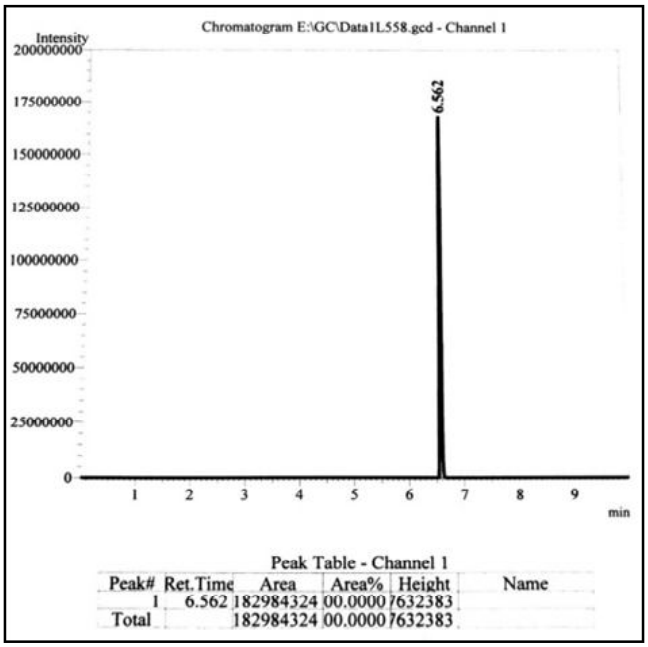

Fig. 6: The Standard Curve for Linalool Essential Oils by GLC Technology.

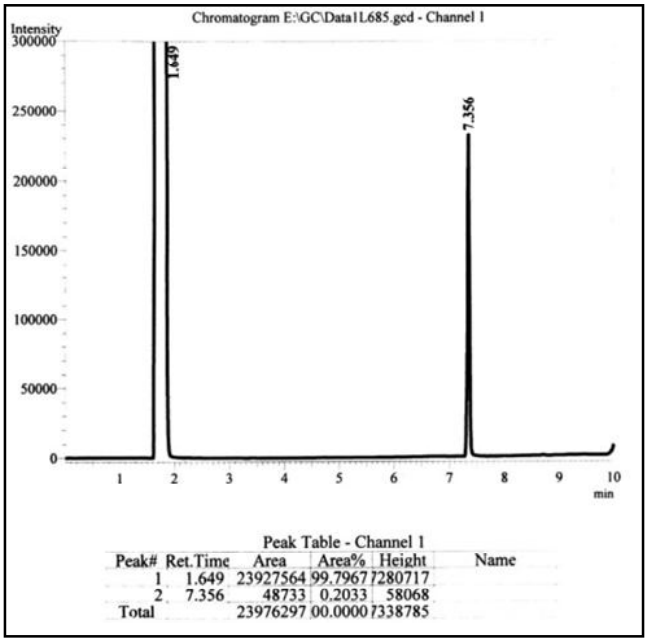

Fig. 7: The Standard Curve for Myrcine Essential Oils by GLC Technology.

a time of retention (3.030) minutes and corresponds to the standard compound at a time of retention (3.485) minutes and concentration (0.0015). Undecanoic acid acid at a time of retention (4.860) minutes and corresponds to the standard compound at a time of retention (4.786) minutes and concentration (0.0007). Elaidic acid at a time of retention (9.430) minutes and corresponds to the standard compound at a time of retention (9.904) minutes and concentration (0.0002). Oleic acid at a time of retention (10.942) minutes and corresponds to the standard compound at a time of retention (10.731) minutes and concentration (0.0001). Lenolic acid at a time of retention (12.824) minutes and corresponds to the standard compound at a time of retention (12.550) minutes and concentration (0.0002). Arachidic acid at a time of retention (14.198) minutes and corresponds to the standard compound at a time of retention (14.184) minutes and concentration (0.0002). Linolenic acid at a time of retention (16.132) minutes and corresponds to the standard compound at a time of retention (16.308) minutes and concentration (0.0002). Erucic acid at a time of retention (17.361) minutes and corresponds to the standard compound at a time of retention (17.041) minutes and concentration (0.002). Tricosanoic acid at a time of retention (18.095) minutes and corresponds to the standard compound at a time of retention (18.294) minutes and concentration (0.0001).

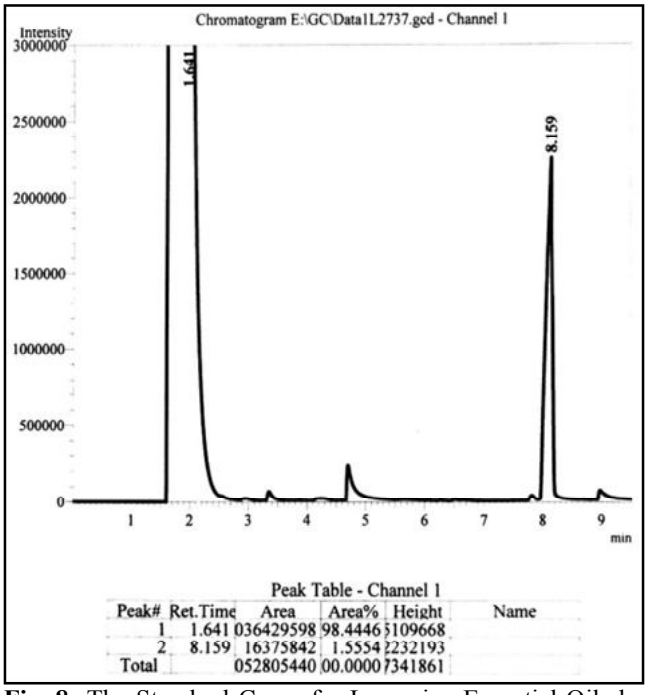

Fig. 8: The Standard Curve for Lemonine Essential Oils by GLC Technology.

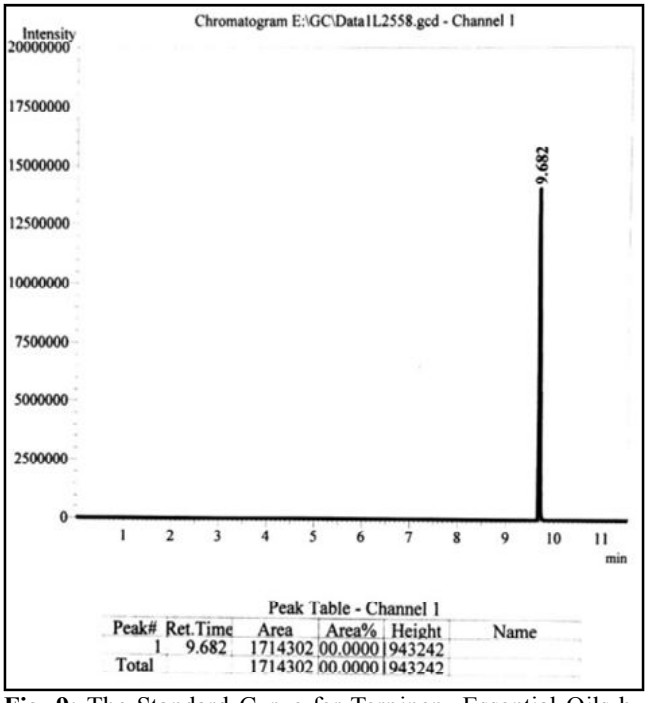

Fig. 9: The Standard Curve for Terpinen Essential Oils by GLC Technology.

Qualitative identification of volatile oils using GLC technology for *Carum carvi* seeds:

Chromatographic analysis charts were obtained in which the retention time of each compound was determined for study samples compared to the standard sample retention time of Camphor (2.900) minutes, Camphene (3.451) minutes, Linalool (6.562) minutes, Myrcine (7.356) minutes, Limonine (8.159) minutes, Terpinen (9.682) minutes and Sabinen 11.126 minutes, Figures. The identification showed the approval of the

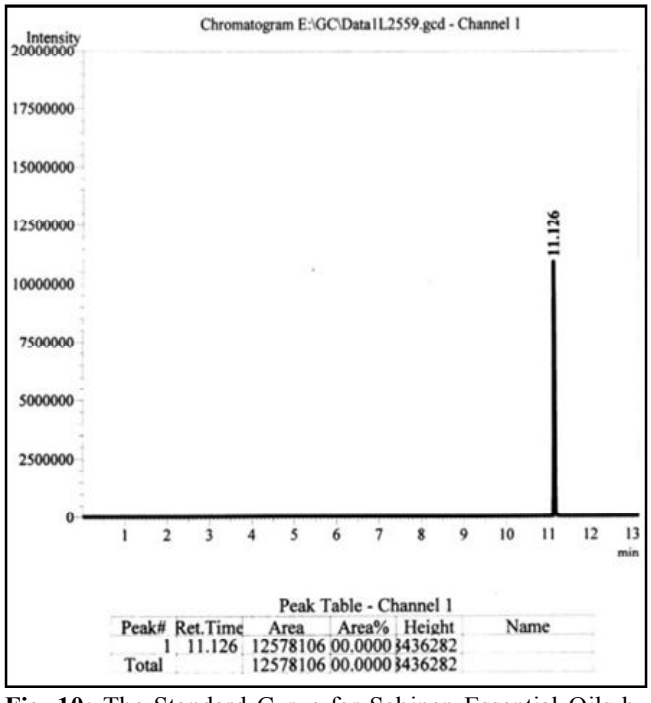

Fig. 10: The Standard Curve for Sabinen Essential Oils by GLC Technology.

Table 5: Volatile oils identified using the GCL technique.

No.	Standard volatile oils	Standard retention time(minute)	The retention time for the study sample is minute
1	Camphor	2.900	2.589
2	Camphene	3.451	3.880
3	Linalool	6.562	6.100
4	Myrcine	7.356	7.505
5	Limonine	8.159	8.701
6	Terpinen	9.682	9.961
7	Sabinen	11.126	11.221

separated essential oils of the study plant for a number of standard aromatic compounds, which included the table 2.

The results indicated the presence of Camphor essential oil in the seeds of *Carum carvi* plant with a retention time of (2.589) minutes, the aromatic compound Camphene with a retention time of (3.880) minutes, the aromatic compound Linalool with a retention time of (6.100) minutes, the aromatic compound Myrcine with a retention time of (7.505) minutes, the aromatic compound Limonine with a retention time of (8.701) minutes, the aromatic compound Terpinen With a retention time of (9.961) minutes, and aromatic compound Sabinen with a retention time of (11.221) minutes.

Conclusion

From the results it is confirmed that the *Carum carvi* seeds are among the plants rich in fatty acids and volatile oils, because the seeds are a primary source of general metabolism secondary compounds, including fatty acids and volatile oils.

References

Al-Daody, A. Ch. (1998). Chemical study on some Iraq plants. Ph. D. thesis, College of Science, University of Mosul, 112-113.

Alessio, P.A. (2009). Molecular evidence of polyphyletism in the plant genus *Carum* L. (Apiaceae), New work, p. 560.

Arthur, I.V. (1972). "Practical organic chemistry including qualitative organic analysis, 3rd edition, 445.

Atal, C.K. and N.M. Sood (2015). Study of Indian caraway and its substitutes. I. Essential oil from *Carum carvi*. *Indian J. Pharm.,* **29:** pp.42-44.

Bhuiyan, M.A.R., M.Z. Hoque and S.J. Hossain (2009). Free Radical Scavenging Activities of *Zizyphus mauritiana*, *World J. Agric. Sci.,* **5(3):** 318-322.

BOWN, D. Encyclopaedia of Herbs and their Uses. Dorling Kindersley, London. 1995.

British, P. (1958). "The pharmaceulical press". London App. XI.1273.

Eddouks, M., A. Lemhadri and J. Michel (2004). Caraway and caper: Potential antihyperglycaemic plants in diabetes rats. *J. Ethnopharm.,* **94:** 143-148.

Ghourchian, A., H. Homa, A. Leila, Ch. Rasool, K. Mohammad, S. Ahmad, G. Latif and M. Mahnaz (2016). Essential Oil and Fixed Oil Content of Nigella sativa after A Traditional Medicine Processing-A Comparative Study. *Biological Forum – An International Journal,* **8(2):** 120-125.

Iacobellis, N.S., P.L. Cantore, F. Capasso and F. Senatore (2005). Antibacterial activity of *Cuminum cyminum* L. and *Carum carvi* L. essential oil. *J. Agric. Food. Chem.,* **53:** 57-61.13.

Kamenik, J. (2001). The basics of caraway crop management (in Czech). *Urda,* **3:** 1–3.

Karaman, I., P. Sahin, M. Gul-luce, H. Oguten, M. Son-gul and A. Adiguzed (2003). Antimicrobial activity of aqueous and methanol extracts of *Juniperus oxycedrus* L. *Journal of Ethnophar-macolology*, **85(2-3):** Pp. 231-235.

Laribi, B., K. Kouk, A. Mougou and B. Marzok (2010). Fatty acid and essential oil composition of three Tunisian caraway (*Carum carvi* L.) seed ecotypes. *J. Sci. Food Agric.,* **1:** 1-6.

Lidefelt, J.O. (2014). A review on the pharmacological aspects of *Carum carvi*, *Journal of Biology and sciences,* **(13):** pp. 68-78.

Nascimento, C.F., J. Locatelli, P.C. Freitas and G.L. Silva (2000). Antibacterial activity of plant extracts and phytochemicals on antibiotic resistance bacteria. *J. Microbiol.,* **31:** 247-256.

Sachan, Kr. Anupam, Doli R. Das and Mukesh Kumar (2016). Carum carvi-An important medicinal plant. *Journal of Chemical and Pharmaceutical Research,* **8(3):** 529-533.