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Chemical study of pumpkin seed oil (*Cucurbita sp*.) using gas chromatography-mass spectrometry (GC-MS)

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I. Abstract:

Pumpkin seed oil was extracted using a saxophone device in the laboratories of the Medicinal and Aromatic Plants Unit, Faculty of Agriculture, University of Basrah, during the 2024 season. The quantity and quality of fatty acids present in the extracted oil were determined using gas-liquid chromatography-mass spectrometry (GC-MS) in the Food Engineering Laboratory of the Department of Food Sciences, University of Basrah. The results revealed the detection of 12 compounds, with saturated fatty acid stearic acid accounting for the highest percentage, at 54.3%, followed by cyclopropane octanic acid (2-(2-ethylcyclopropyl)methyl) at 21.56%. These compounds are the main compounds of pumpkin seed oil.

Keywords: Pumpkin, GC-MS, fatty acids

II. Introduction:

Pumpkin (*Cucurbita sp*.) is an important member of the Cucurbitaceae family. It is a creeping or climbing herbaceous plant that grows to about 4 m tall. It has broad leaves and yellow, angiosperm-shaped flowers. Its fruits are large and vary in shape and size, the most famous being the long, cylindrical, orange-colored ones. The sweet-tasting fruits are eaten cooked, used in jams, and are also used in ice cream. The plant's native habitat is the Mediterranean basin, the Arabian Peninsula, and Iraq. It is used as a laxative, tonic, tapeworm repellent and treats many ailments, such as headaches, colds, intestinal colic, emaciation and weakness, and increases sexual potency. (Al-Sayed and Abdel Tawab, 2020). Every 100 g of dry weight of pumpkin seeds contains 6.75 g of water, 40.1 g of protein, 28.3 g of crude fat, 16.0 g of fiber, 4.5 g of carbohydrates, 4.3 1 gm ash, 608.5 mg potassium, 328.2 mg magnesium, 276.8 mg sodium, 98.2 mg calcium and 14.3 mg iron (Shalaby *et al*., 2020). The oil consists of saturated fatty acids (Total saturated) at a rate of 27.73%, consisting of Palmitic acid 16.41% and Stearic acid 11.14%, and unsaturated fatty acids (Total unsaturated) at a rate of 73.03%, most of which consists of Oleic acid 18.14% and Linoleic acid 52.69% (Alfawaz, 2004). Ali *et al*. (1999) mentioned the importance of pumpkin seed oil in containing seven types of Phytosterols that are used in the manufacture of medicines used to treat prostate tumors and its dual effectiveness in dealing with types of beneficial cholesterol (HDL) High Density Liprotein and reduces harmful cholesterol Low Density Liprotein, pumpkin oil also increases the softness and hydration of the skin due to the fatty acids in its composition and some of the physical properties of oils, in addition to being a skin disinfectant from microbes (Al-Shahwani *et al*., 2008) and inhibiting prostate cancer cells, treating some diseases such as heart and arteries, asthma , cancer and as an immune stimulant in humans, in addition to its beneficial role in benign prostatic hyperplasia (BPH) due to its high content of B-Sitosterol ((Ambavade *et al*., 2014 ; Iwo *et al*., 2014; Mando *et al*., 2023) . Badr *et al*. (2011) showed the presence of six organic compounds that were isolated from pumpkin seeds, namely Linoleic acid,

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Triglyceride fatty acid mixture, Calotropoleanyly ester, Tetrahydro-thiophene and Oleanen -3-0l.-(18)13 and Chelesterol. These compounds have been shown to have anti-cancer activity in the colon, intestine, and cervix. Prommaben *et al*. (2021) analyzed pumpkin seed oil (Cucurbita moschata Duch) using GC-MS to detect 13 active compounds. Cis-linoleic acid (C18:2 omega-6) ranked first, accounting for 39.09%, followed by Oleic acid (C18:1 omega-9) at 31.22%, Palmitic acid (16:0) at 19.08%, and Stearic acid (18:0) at 9.37%. Anyanwu *et al*. (2022) reported the presence of 5 active compounds when analyzing pumpkin oil using GC-MS. The n-Hexadecanoic acid (Palmitic acid) ranked highest at 40.01%, followed by Cis-Linoleic acid. Octadecadinoic acid (Z,Z)—- 9,12 at 39.11% and Hexadeanoic acid, ethyl ester (Ethyl- palmitat) tetraderanoic acid, ethyl ester (Ethyl- Myristic) at 9.7%. Aboseea *et al*. (2023) observed the appearance of 47 compounds when analyzing pumpkin seed oil using GC-MS, with Hexadecanoic acid, trimethylsilyl ester occupying the highest percentage at 42.06%, followed by Decadion-1-OI-2,4 at 15.85% and Squalene at 11.16%. Mulu hagos *et al*. (2023) observed that when analyzing pumpkin seed oil using GC-MS, 47 compounds appeared, with Hexadecanoic acid, trimethylsilyl ester occupying the highest percentage at 42.06%, followed by Decadion-1-OI-2,4 at 15.85% and Squalene at 11.16%. The presence of four fatty acids when analyzing pumpkin seed oil using GC-MS technology, the acid Cis-9- octadecenoic acid (Linoleic acid) 18:2 occupied the highest percentage of 50.7%, followed by Cis-9- octadecenoic acid (oleic acid) 18:1, at a percentage of 18.8%, the compound Hexadecanoic acid (Palmitic acid 16:0), at a percentage of 17.9%, and the compound Ladeecanoic acid (Steraic acid 18:0), at a percentage of 12.4%. Al-Turki and Ibrahim (2022) confirmed that the analysis of the oil of two species of Syrian pumpkin, *C. moschata* and *C. maxima*, using GC-MS technology, showed that the oil extracted from the seeds contained a high percentage of saturated fatty acid Palmitic acid (C 16:0 18.40, 14.01)%, respectively, and the fatty acid Stearic acid (C18:0) at a rate of (6.52, 7.68)%, respectively, and the total saturated fatty acids reached (24.92, 21.69)%, respectively, while it contained a high percentage of unsaturated fatty acids composed of Oleic acid (18:1) at a rate of (18.72, 30.05)% and Linoleic acid (18:2) at a rate of (56.34, 48.24)%, respectively, so that the total unsaturated fatty acids were (75.06, 78.29)%, respectively. The study aims to identify the fatty acids present in pumpkin seed oil using GC-MS technology and their percentages.

III. Materials and Methods:

The biologically active compounds in pumpkin seed oil were identified by extracting the oil from the seeds using hexane in the laboratories of the College of Agriculture, University of Basrah, Iraq. Gas chromatography-mass spectrometry (GC-MS) model QP210 Ultra. Shamadzu.APAN was performed, equipped with a DB-MAS5 capillary column (95% methylpolysiloxane, 5% vinyl) as the stationary phase, in addition to the use of helium gas (99.9%). The following conditions were used: injection mode: split, column oven temperature: 40°C, injection temperature: 280°C, column flow: 1.71 ml/min, pressure: 96.1 kPa, purge flow: 3.0 ml/min, split ratio: 46.9, total flow: 79.2 ml/min, linear velocity: 47.2 cm/s, interface temperature: 280°C, mass spectrometer: ion source temperature: 200 °C, Solvent cut-off time: 3.00 min, Detector gain: 1.24 kV + 0.10 kV, Start time: 3.00 min, End time: 35.00 min, ACQ mode: Scan, Event time: 0.50 s, Scan speed: 2000, Start speed: 50.00, End speed: 800.

IV. Results and Discussion:

Table 1 shows the GC-MS chromatograms resulting from the analysis of pumpkin seed oil, where 12 compounds appeared. The saturated fatty compound Octadecadienoic acid, methyl ester (E,E)-9,12

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(chemical name for Stearic acid, chemical formula CH3-(CH2)16-COOH, consisting of 18 carbon atoms) occupied the highest percentage, at 54.30%. This was followed by the fatty acid Cyclopropane octanoic acid, 2((2-(2-ethyl cyclopropyl)methyl. which is a fatty acid containing a cyclopropane ring, chemical formula C22H38O2, containing 22 carbon atoms, representing 21.61%. This was followed by the saturated fatty acid Hexadecanoic acid methyl ester (chemical name for Palmitic acid, chemical formula CH3(CH2)14-). COOH contains 16 carbon atoms and its percentage is 21.54%. This result is consistent with what was obtained by (prommaben et al., 2021, Anyanwn et al., 2022, Al-Turki and Ibrahim, 2022, Abusrea *et al*., 2023, and Muluttagos *et al*., 2023).

Table 1.Components Indentified in *Cucurbita* sp.

Peak Area %	Name of the compound	RT	No.
0.09	4-Estren-4,17.beta.-diol-3-one, tri-trimethylsilyl	11.067	1
0.46	Methyl tetradecanoate	14.350	2
0.57	(Z)-Methyl hexadec-11-enoate	17.679	3
21.56	Hexadecanoic acid, methyl ester	18.183	4
0.08	Voacamine	18225	5
0.24	5-(4-Methoxymethylphenyl)-10,15,20-triphenyl-21H,23H-porphine zinc	18.359	6
0.13	9,12-Octadecadienoic acid, methyl ester, (E,E)-	19.438	7
0.19	Methyl 9-heptadecenoate or 9-17:1	19.525	8
0.61	Heptadecanoic acid, methyl ester	20.035	9
54.30	9,12-Octadecadienoic acid, methyl ester	21.257	10
0.17	benzenamine, 4,4'-[(4-bromophenyl)methylene]bis[N,N-bis(4-methylphenyl)-	21.300	11
21.61	Cyclopropaneoctanoic acid, 2-[[2-[(2-ethylcyclopropyl)methyl]cyclopropyl]methyl]-, meth	21.450	12

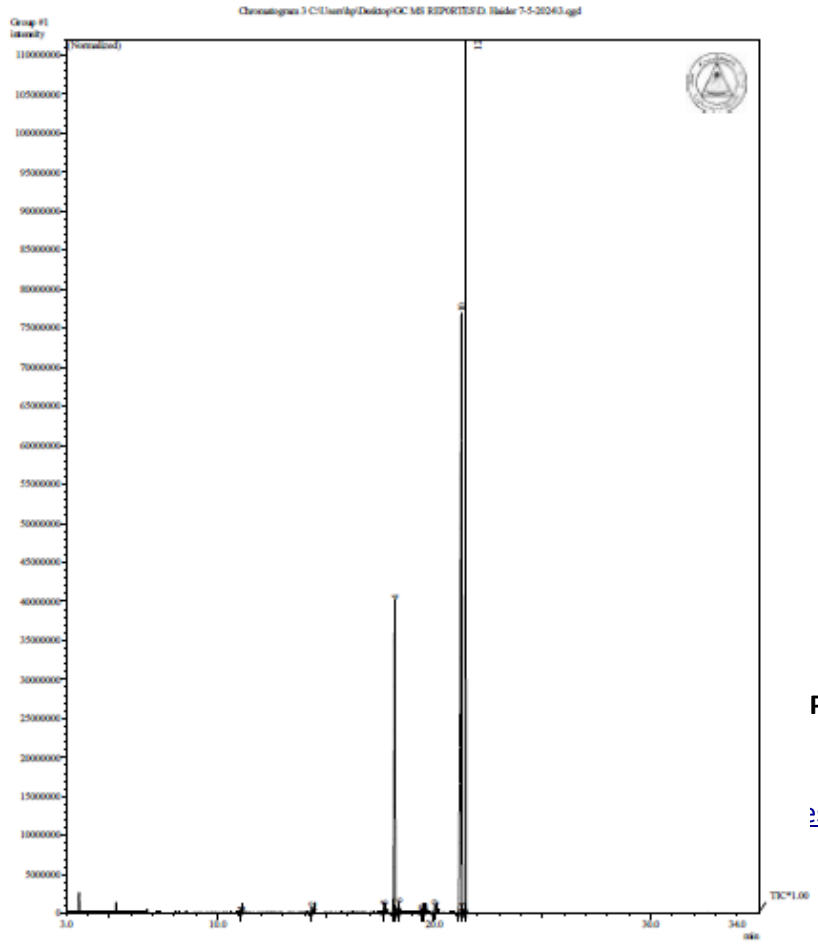

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Fig.1: A bioactive compound in Pumpkin seeds that indentified by GC-Mass device

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