



Original Research Article

Studying the antioxidant and antibacterial properties of radish and carrot seed extracts and their effect on preserving river fish meat

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Abstract: Plant seeds are among the most important natural sources of bioactive compounds. It has become a biologically active chemical, containing a wide range of fatty acids, phenols, flavonoids, terpenes, and volatile compounds with high biological activity. Studies indicate that radish and carrot seeds contain varying proportions of proteins, fats, and carbohydrates, in addition to phenolic compounds that contribute to enhancing their properties as effective antioxidants and antimicrobials. Preliminary chemical analyses of radish and carrot seeds showed high content of fats, proteins, and total sugars. The fat content in radish seeds was 35.34%, and in carrot seeds, it was 33.66%, while protein content ranged from 14.93% to 18.37%. Gas chromatography-mass spectrometry analysis also showed... (GC-MS) radish and carrot extracts contain a number of bioactive compounds, most notably saturated and unsaturated fatty acids such as linoleic acid, oleic acid, and palmitic acid, in addition to phytosterols and terpenoids that contribute to antioxidant activity and resistance to pathogenic microorganisms. Technology GC-MS is one of the most important analytical tools used to determine the molecular identity of active compounds in plant extracts, due to its high accuracy in separating and identifying volatile and semi-volatile compounds, thus enabling a deeper understanding of the relationship between chemical structure and biological activity.

Keywords: Bioactive compounds, radish seeds (*Raphanus sativus* and carrot seeds (*Daucus carota*) chromatography-mass spectrometry analysis.

INTRODUCTION

Plant seeds are rich sources of bioactive chemical compounds, contributing to numerous food and pharmaceutical applications due to their wide range of fatty acids, phenols, flavonoids, and volatile compounds with antioxidant and antimicrobial activity. Carrot seeds (*Daucus carota*) and radish seeds (*Raphanus sativus*) are among the most important of these natural sources, as they contain high levels of unsaturated fatty acids and biogenic phenolic compounds that give them antioxidant properties and help in food preservation.(1) (2). Chemical analyses and chromatographic studies indicate that extracts of radish and carrot seeds contain several active compounds, such as linoleic fatty acids (Linoleic acid, oleic acid, and palmitic acid, in addition to phytosterols and terpenoids, play a role in enhancing bioavailability and antioxidant capacity.(3)(4)

Furthermore, studies using gas chromatography-mass spectrometry (GC-MS) have shown that these seeds contain a wide range of bioactive and semi-volatile compounds, which contribute to increased stability of food products when used as natural antioxidants.(5)(6). Accordingly, the rich chemical composition of radish and carrot seeds makes them promising sources in the food and biological industries, especially when their active compounds are extracted using aqueous and alcoholic solvents and their effectiveness in biopreservation applications is studied.

Materials and working methods

Chemical analysis of seed powder Chemical Content of Powder Seed

Humidity estimation Moisture

The percentage of moisture in the samples was estimated by weighing 3 grams of seed powder

samples, placing them in a pre-weighted container, and drying them in an electric oven at a temperature of (105m) for 16 hours until weight stabilizes (7).
Percentage of moisture = $\frac{\text{Weight of sample before drying} - \text{Weight of sample after drying}}{\text{Weight of sample before drying}} \times 100$.

Protein estimation

The total protein content in the seeds was estimated as stated in (7). Using the Micro-Keldal method Semi-microkeldahl.

fat estimation

The fat content is measured using the Soxhlet method described in (7) (And according to the percentage of fat, as follows)

% Fat content = $\frac{(\text{Weight of empty flask} + \text{Fatty substance}) - (\text{Weight of empty flask})}{\text{Sample weight}} \times 100$

Ash estimation

The ash was measured by burning the samples in an incinerator. Muffle Furnace at 550°C for 16 hours as

described in (7).

Estimation of total fibers:

Fibers were estimated according to method (7) According to the following equation:

Fiber = $\frac{\text{Weight after drying} - \text{Weight after burning}}{\text{sample weight}} \times 100$

Carbohydrate estimation:

The carbohydrate content in the leaf powder was calculated according to the method described by (8) Using the following equation:

Carbohydrate % = $100 - (\text{Moisture} + \text{Ash} + \text{Protein} + \text{Fiber} + \text{Fat}) \%$

Estimation of active compounds using technology GC-MS for alcoholic extracts of radish and carrot seeds

Separating and identifying chemical compounds by comparing mass spectra According to method (9)).

RESULTS AND DISCUSSION

The chemical composition of plant seed powder:

Table (1) shows the results of the analysis, indicating that radish seeds contained a higher protein percentage of 18.37% compared to carrot seeds, which recorded 14.93%. This is consistent with what was indicated. (10) Radish seeds are rich in protein and can be used in food applications. Radish seeds are also characterized by a high fat content, reaching 35.34%, while carrot seeds have a fat content of 33.66%. This high fat content reflects the oily nature of oilseeds, as explained (11) In his study on radish seeds, he confirmed (12) Carrot seeds are also rich in oil and contain high amounts of unsaturated fatty acids. Ash content was higher in carrot seeds at 14.40% compared to radish seeds at 4.08%, indicating the richness of carrot seeds in minerals, a finding confirmed by further research. (13) This showed that carrot seeds are characterized by a high mineral content, while the carbohydrate content was low in both seeds, with carrot seeds recording 4.70% compared to 1.90% in radishes. This is consistent with the results (14) The study revealed that radish seeds are composed more of protein and fat than carbohydrates. The results also showed a lower moisture content in both seeds, reaching 1.2% in carrots and 1.4% in radishes. This contributes to a longer storage period and reduces the chances of chemical changes and microbial spoilage. ((15) Low moisture content is a key factor in the stability of oilseeds in general. Thus, it can be said that radish seeds are characterized by their high protein and fat content, while carrot seeds are characterized by their high ash and carbohydrate content. This gives both seeds complementary nutritional properties that can be utilized in the food industry and in the preservation of animal products..

Table (1) shows the chemical composition of radish and carrot seeds

plant seeds		the components %
Islands	Radish	
1.2	1.4	Humidity
14.93	18.37	protein
33.66	35.34	grease
14.4	4.08	Ashes
35.81	40.80	Carbohydrates

Estimation of active compounds using technology GC-MS for alcoholic extracts of radish and carrot seeds

Estimation of the active compounds in the extract of water radish seed powder

Analysis showed GC-MS of alcoholic extract of radish seeds in Table (2) contains a distinctive group of fatty acids and their ester derivatives (FAMEs), known for their biological activity as antioxidants and antimicrobials, suggest their potential use as a natural source of bioactive compounds in food and pharmaceutical applications. Methyl stearate (octadecanoic acid, methyl ester) and methyl palmitate (hexadecanoic acid, methyl ester) were found to constitute the largest proportions of the extract's components, at 27.36% and 23.66%, respectively. Studies have shown (16) (17) Their role is to inhibit oxidation and the growth of bacteria and fungi. The extract also contains other active compounds such as Methyl erucate (10.81%) with antitumor and antioxidant activity (18). and Oleic acid, methyl ester (10.33%) and Methyl oleate (2.43%), known for their antioxidant and anti-inflammatory properties. (16) These results indicate that the alcoholic extract of radish seeds is rich in bioactive fatty compounds with great potential to prolong food preservation and improve its quality thanks to its ability to resist oxidation and microbial activity,

making it a promising natural alternative to synthetic antibiotics.

Table (2) shows the active compounds in water radish seeds

No	RT (min)	Area%	Name	Quality	CAS Number
1	14.516	2.68	Cyclopentane, 1,2,4-trimethyl-, stereoisomer	38	016883-48-0
2	25.21	23.66	Methyl palmitate	99	000112-39-0
3	27.939	10.33	Oleic acid, methyl ester	98	013481-95-3
4	28.349	27.36	Octadecanoic acid, methyl ester	98	000112-61-8
5	30.86	2.43	Methyl oleate	74	000112-62-9
6	32.152	3.76	Hexanedioic acid, bis(2-ethylhexyl) ester	95	000103-23-1
7	33.558	10.81	Methyl erucate	97	001120-34-9

Estimation of the active compounds in the alcoholic extract of radish seed powder

Analysis showedGC–MS analysis of the alcoholic extract of radish seeds (Table 3) shows it contains a wide range of bioactive chemical compounds, especially fatty acids and their esters, which possess antioxidant and antimicrobial activity.(19)WhichmakeThe extract is a natural filter for preserving fish meat without the use of artificial substances. This includes fatty acids and their derivatives such as 9,12-Octadecadienoic acid, methyl ester (22.6%), oleic acid, methyl ester (12.2%), and methyl palmitate (11.6%) are compounds known for their vital role as natural antioxidants that inhibit auto-oxidation reactions in oils and fatty foods. These compounds are important factors in extending the shelf life of food products by reducing the formation of free radicals responsible for fat rancidity and loss of sensory and nutritional value.20The results also revealed the presence of ester compounds such asHexadecanoic acid, ethyl ester and 9,12-Octadecadienoic acid, ethyl ester are compounds that possess antibacterial and antifungal properties. Previous studies have shown their ability to inhibit the growth of food spoilage bacteria such as Staphylococcus aureus and Escherichia coli, making them important in developing natural food preservation systems as an alternative to chemical preservatives.21). presence of a compoundy-Sitosterol (13.47)% further enhances the functional value of the extract, as this plant sterol acts as a potent antioxidant and contributes to promoting the oxidative stability of fats and oils during storage or heat treatment. It also has protective effects in the human diet by reducing cholesterol absorption and supporting cardiovascular health (2).2As for secondary compounds such asThymol (2.43)% and Vitamin E (1.3%) are very effective food preservatives. Thymol has antibacterial and antifungal properties, which limit the growth of spoilage microorganisms and is used as a natural additive in food packaging and protection during storage. Vitamin E is one of the most important natural antioxidants that prevents fat oxidation and maintains the quality of food products for longer periods.23The presence of these compounds in alcoholic radish seed extract suggests its potential use as a natural source of preservatives and antioxidants in the food industry.(24).

No	RT (min)	Area%	Name	Quality	CAS Number
1	14.506	2.22	3-Undecene, 9-methyl-, (Z)-	62	074630-50-5
2	25.205	11.65	Methyl palmitate	99	000112-39-0
3	26.31	2.52	Hexadecanoic acid, ethyl ester	98	000628-97-7
4	27.82	22.67	9,12-Octadecadienoic acid, methyl ester, (E,E)-	99	002566-97-4
5	27.923	12.29	Oleic acid, methyl ester	99	000112-62-9
6	28.339	8.25	Methyl stearate	98	000112-61-8
7	28.831	6.87	9,12-Octadecadienoic acid, ethyl ester	99	007619-08-1
8	28.93	3.32	Methyl oleate	80	000112-62-9
9	33.538	7.78	Silane, methylenebis[dimethyl-]	43	018163-84-3
10	37.227	2.25	SEBACIC ACID, OCTYL ESTER	53	000000-00-0
11	40.428	0.65	Gibberellin A3	72	000077-06-5
12	40.843	1.30	Vitamin E	38	000059-02-9
13	42.327	1.43	N-ethyl-1,3-dithioisindoline	46	035373-06-9
14	42.436	2.43	Thymol	64	055012-80-1
15	43.251	13.47	.gamma.-Sitosterol	94	000083-47-6

Table (3) shows the active compounds in alcoholic radish seeds

Estimation of the active compounds in the aqueous extract of carrot seed powder

The results of the gas chromatography-mass spectrometry analysis (GC-MS) for aqueous carrot seed extract Table (4) shows that it contains a rich mixture of biologically active compounds, including terpene, phenolic, aromatic, and fatty acids with clear antioxidant and antimicrobial activity. The presence ofβ-Bisabolene, a monoterpene compound, exhibits antibacterial and antifungal properties, as confirmed by a study.(25), in addition toCarotol, a key component of carrot essential oil, contributes to inhibiting fungal growth. Results also showed the presence of Daucol, which possesses antioxidant and antibacterial properties. (26).andCIS-(β)-Asarone, which inhibits microbial

cell growth by disrupting the cell membrane (27) Other aromatic and fatty compounds were also discovered, such as Benzaldehyde, 2,4,5-trimethoxy- and 5-aminoisophthalic acid, are effective in inhibiting microbes, as are cyclic compounds such as 1,7,7-trimethyl-2-vinylbicyclo[2,2,1]hept-2-ene, which inhibit free radical formation. Several fatty acid esters, such as methyl palmitate, methyl stearate, and methyl 6-octadecenoate, have also been found to improve sensory stability and reduce rancidity in meat (28). Compounds such as hexadecanoic acid bis(2-ethylhexyl) ester and 9,12-octadecadien-1-ol have also demonstrated dual antioxidant and antimicrobial properties (29). These results support the possibility of using aqueous carrot seed extract as a natural preservative as an alternative to chemicals, as multiple studies such as (25) (23) have shown that terpene and phenolic plant compounds contribute effectively to food preservation and improving microbial safety, especially in animal products such as river flank meat.

Table (4) shows Estimation of the active compounds in the aqueous extract of carrot seed powder

Estimation of the active compounds in the alcoholic extract of carrot seed powder

The results of the gas chromatography-mass spectrometry analysis (The GC-MS analysis shown in Table (5) indicates

No	RT (min)	Area%	Name	Quality	CAS Number
1	17.707	1.32	β -Bisabolene	95	000495-61-4
2	19.44	18.87	Carotol	94	000465-28-1
3	20.229	3.00	Daucol	97	000887-08-1
4	20.898	2.19	CIS(.BETA.)-ASARONE	90	000000-00-0
5	21.697	1.18	Benzaldehyde, 2,4,5-trimethoxy-	90	004460-86-0
6	22.076	2.12	5-Aminoisophthalic acid	49	000099-31-0
7	22.2	3.35	1,7,7-Trimethyl-2-vinylbicyclo[2.2.1]hept-2-ene	25	130930-56-2
8	23.197	1.47	4-(3,3-Dimethyl-2-exo-norbornyl)-2-methylbut-2-en	44	113104-69-1
9	25.199	7.94	Methyl palmitate	99	000112-39-0
10	27.825	2.54	9,12-Octadecadienoic acid, methyl ester, (E,E)-	99	002566-97-4
11	27.944	27.83	Methyl 6-octadecenoate	99	052355-31-4
12	28.339	8.90	Methyl stearate	98	000112-61-8
13	32.142	4.37	Hexanedioic acid, bis(2-ethylhexyl) ester	95	000103-23-1
14	32.49	5.90	9,12-Octadecadien-1-ol	56	001577-52-2
15	33.548	1.38	1,5-Cyclodecadiyne	43	118620-70-5

that the alcoholic extract of carrot seeds contains a group of bioactive compounds that play an important role in enhancing meat stability and reducing spoilage rates resulting from oxidation or microbial contamination. Oleic acid, a monounsaturated fatty acid with antibacterial and anti-inflammatory effects, was identified, in addition to its role in protecting food from oxidative degradation and inhibiting the growth of both Gram-positive and Gram-negative bacteria, as demonstrated in the study.(25) N-hexadecanoic acid was also identified (n-Hexadecanoic acid (or palmitic acid), a saturated acid, has shown clear antimicrobial activity by affecting bacterial cell membrane permeability, as demonstrated in a study.(30) The results also showed the presence of 8.11-Octadecadienoic acid methyl ester, a derivative of linoleic acid known for its effectiveness as an antioxidant and lipid stabilizer, was confirmed by a study.(31) Its role in reducing rancidity in meat and fish. It was also revealed that Methyl oleate, used as a natural solvent, has a high capacity for stabilizing aromatic compounds and fatty acids in animal products (32). Additionally, oleamide, derived from oleic acid, possesses antimicrobial and antifungal effects and contributes to inhibiting microbial spoilage in animal products. Several terpene compounds, such as β -bisabolene, trans-Z- α -bisabolene epoxide, carotol, and daucol, have been identified as having high antioxidant and antimicrobial properties, as confirmed by a study (33) on aromatic compounds extracted from plants. Furthermore, plant sterol compounds, including stigmasterol and ethylcholest-5-en-3-beta-ol, have been identified as contributing to enhanced oxidative stability and the preservation of protein and lipid quality during storage.(34) In addition, phenolic and aromatic compounds such as 4- were observed. Dimethoxycinnamic acid 2.4 and Asarone, which is used as a natural antioxidant in the food industry, should be used with caution in high concentrations due to its toxic effects, as reported by the European Chemicals Agency (ECHA). Therefore, the presence of these fatty, terpene, phenolic, and sterol compounds in alcoholic carrot seed extract makes it a promising candidate as an effective and safe natural preservative for fish meat (such as flank meat), as it plays a role in reducing spoilage rates and improving microbial and chemical stability during storage.

Table (5) shows the estimation of the active compounds in the alcoholic extract of carrot seed powder.

No	RT (min)	Area%	Name	Quality	CAS Number
1	9.395	0.28	Benzeneacetonitrile	91	000140-29-4
2	17.691	0.90	β -Bisabolene	98	000495-61-4
3	18.724	0.75	4-Nonylphenol	35	000104-40-5
4	19.16	0.57	(-)-Caryophyllene oxide	98	001139-30-6
5	19.523	36.31	Carotol	91	000465-28-1
6	20.239	6.02	Daucol	99	000887-08-1
7	20.742	0.27	trans-Z-.alpha.-Bisabolene epoxide	44	000000-00-0
8	20.877	4.80	Asarone	99	002883-98-9
9	21.567	2.05	Gazarin	90	004460-86-0
10	22.065	1.65	Ethyl 2,2,5-trimethyl-3,4-nonadienoate	64	143237-43-8
11	22.231	4.20	Ylangenol	50	055683-15-3
12	22.704	0.39	3-Acetamidocyclohexene	53	000000-00-0
13	23.202	0.36	DIEPI-.ALPHA.-CEDRENEPOXIDE	55	000000-00-0
14	23.414	1.03	Alloaromadendrene oxide-(1)	47	000000-00-0
15	23.881	0.25	6-Aminobenzo(G)quinoxaline-5,10-dione	58	072225-24-2
16	24.426	0.78	Alloaromadendrene oxide-(1)	59	000000-00-0
17	24.613	0.83	2,3-Dimethyl-8-oxo-non-2-enal	38	000000-00-0
18	25.962	2.09	n-Hexadecanoic acid	97	000057-10-3
19	27.82	0.37	8,11-Octadecadienoic acid, methyl ester	99	056599-58-7
20	27.939	0.81	Methyl oleate	99	000112-62-9
21	28.816	21.79	Oleic acid	99	000112-80-1
22	31.353	0.31	Benzene, [(1-ethenyl-1-propenyl)thio]-, (Z)-	58	084132-32-1
23	31.597	1.36	Methyl 1,2-Dimethyl-2-propenyl Ether	12	000000-00-0
24	31.706	2.01	Oleamide	87	000301-02-0
25	32.116	0.55	2-t-Butylprop-2-en-1-ol	38	039497-64-8
26	32.609	0.60	Methyl 3-methyl-2-butenolate	38	000924-50-5
27	35.68	1.15	.beta.-Monoolein	56	003443-84-3
28	35.846	0.99	(R)-(-)-14-Methyl-8-hexadecyn-1-ol	95	064566-18-3
29	36.988	0.31	CIS(.BETA.)-ASARONE	87	000000-00-0
30	37.232	0.95	Bis(2-ethylhexyl) sebacate	58	000122-62-3
31	40.002	0.69	Acetylsarcoglaucol	55	068299-88-7
32	40.661	0.43	2,4-Dimethoxycinnamic acid	76	006972-61-8
33	42.42	0.55	Stigmasterol	94	000083-48-7
34	43.235	0.84	(23S)-ethylcholest-5-en-3.beta.-ol	98	113845-28-6

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