

Study of the active compounds and antioxidant capacity of *Origanum majorana* Extracts

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Abstract

The aqueous and alcoholic extracts of the marjoram plant were prepared, and the percentages of moisture, protein, fat, ash, and carbohydrate contents were found to be 5.6%, 5.5%, 7.3%, 4.17%, and 77.43%, respectively. The marjoram extract was used as a preservative for minced meat at a concentration of 1% for 0, 3, 7, and 10 days. The DPPH scavenging activity of the aqueous and alcoholic extracts was also monitored. The antioxidant activity increased with increasing concentration, with the lowest value recorded at 0.12 mg, reaching 62.613% and 62.687% for the aqueous and alcoholic extracts, respectively. The highest antioxidant activities were 72.437% and 74.584% for the aqueous and alcoholic extracts, respectively, compared to the control treatment, which was 1.0739% .

Keywords: marjoram, GC-MS, antioxidant , DPPH ,active compound.

Introduction :

Marjoram, also known as dosh, contains many vitamins, minerals, essential oils, flavonoids, hydroquinone glycosides, water-soluble sugars, and triterpenes. These components collectively endow marjoram with significant health properties, including antimicrobial, anti-inflammatory, analgesic, antidepressant, and antiviral activities.

The plant also contains volatile oils, with thymol and carvacrol being the major constituents. Its leaves and flowers are commonly used as spices or brewed into tea, while its oil is traditionally used for treating colds [1.]

Moreover, marjoram has demonstrated strong anti-inflammatory and antimicrobial properties. It has been used medicinally to help treat a variety of conditions, including digestive disorders, inflammation, and painful menstruation [2.]

Additionally, [3,4].emphasized the important role of plant extracts in food preservation due to their high content of natural antioxidants.

Research objectives

-1Aqueous and alcoholic extraction of active substances and antagonists in marjoram.

-2Estimation of the concentration of active compounds from marjoram powder extracts using GC-MS technique

Active compounds extracted from plants have been used since ancient times in many industries. Among the most widely used compounds are tannins, which have been used and coloring materials. [5].Terpenes Carbohydrate organic compounds produced by a group of plants, often possessing a strong odor that protects them from parasites .

Terpenes are terpenes but contain an additional oxygen atom and consist of a main unit called Isoprene, which is anticancer and antioxidant [6]. Alkaloids Alkaloids contain branched nitrogenous bases with a heterogeneous ring containing a nitrogen atom, and they arise in dicotyledonous plants and increase in a number of plant families, including legumes, and each family is characterized by the presence of special alkaloid substances that characterize it, and alkaloids vary in their distribution in plant tissues according to the type and age of the plant tissue, and Alkaloids in plants are usually found in a free state or in the form of salts of some acids and the majority of them are crystalline solids, except for some alkaloids in which there is no oxygen element, they are liquid and most alkaloids are colorless and odorless and bitter in taste, few of them are colored, they are not soluble in water while their salts dissolve in it Harborne . [5]. Phenolic compounds : They are considered aromatic compounds and consist of a benzene ring attached to one or more hydroxyl (OH) side groups and have the ability to dissolve in water and organic solvents [7]. It also contains essential oil, a natural extract characterized by its powerful antioxidant and antimicrobial properties. It is widely used in food preservation and health applications to combat pathogens and oxidative spoilage[8].

Phenolic compounds have the ability to combine with cell protein and precipitate it [9].

The perennial herb *Origanum majorana* is native to Mediterranean regions such as Cyprus, Algeria, and Egypt, and its leaves and flower oil are utilized as flavoring and seasoning in food[10]. The essential oil derived from oregano is a botanical extract known to be devoid of adverse effects. This botanical substance has the potential to serve as a sustainable and ecologically sound substitute for synthetic food additives, chemical preservatives, and animal feed supplements. This has been evidenced by

several investigations, such as those carried out by Carvalho [11].

Material and Methods:

The herb was obtained from the local market in Najaf and was in the form of dried leaves and then milled in a French-made Moulinex electrostatic mill and kept in a bottle at laboratory temperature until the study.

Estimation of the chemical composition of the marjoram plant: Chemical estimations were carried out at the rate of three replicates and were calculated on the basis of dry weight and included moisture, protein and ash according to [12]. and the carbohydrate ratio was calculated by the difference between the mentioned ingredients as described [13].

Preparation of extracts:

Preparation of aqueous extract: The aqueous extract was prepared according to [14]. by soaking 20 g of dried plant powder in 100 ml of water for 24 hours at laboratory temperature and then concentrating the extract using a rotary evaporator .

Preparation of the alcoholic extract: The alcoholic extract was prepared by weighing 20 g of plant powder in 100 ml of ethanol alcohol with stirring for 24 hours, then filtered and concentrated using a rotary evaporator [15].

DPPH test: A solution of 0.1 mM DPPH (hydrazyl picryl-2-diphenyl-1,1-) with 0.0039 mM DPPH in 95% ethanol was prepared and

the volume was adjusted to 100 mL. The method of [16]. was followed to inhibit the DPPH free radical by mixing 1 mL mM 0.1 mL of DPPH solution dissolved in 95% ethanol was mixed with 1 mL of the extract, mixed well and left in the dark at room temperature for 60 minutes until the purple color of DPPH disappeared and a violet or

light pink color appeared, which was determined by measuring the decrease in absorbance at a wavelength of nm517 DPPH as a percentage based on the following equation: DPPH free radical inhibition %] = absorbance reading of control treatment - absorbance reading of mixture / absorbance reading of control treatment] × 100.

Estimation of active compounds using GC-MS of the alcoholic extract GC-MS analysis of active compounds:

Gas Chromatography Mass Spectrophotometer Anlysis(GCMS)for active compounds

Detection of active compounds in the alcoholic plant extract of marjoram plant) Using GC-MS/MS analysis, the mass spectra of the active compounds present in alcoholic extracts were measured at Al-Nahrain University/Baghdad at the Biotechnology Research Center The active compounds in the plant extract were estimated using GC-MS/MS analysis, a 50 µm thick film capillary column was used for separation, the initial Column Oven Temp. 40 °C and the final temperature was 280 °C. To maintain the program temperature, it was held for 1 minute at 120 °C and the rate of temperature rise was 8 °C/min until it reached 210 °C and then held for 45 minutes at 210 °C. The sample volume required for glaucoma is 1 microliter, and the temperature of the glaucoma area. Injection Temp 280) °C and the detector temperature is 280 °C. The carrier gas is helium at a constant Pressure Pressure of 96.1 kPa, and the carrier gas flow rate in the Column Flow is 1.71 mL/min .

1g of the extract was taken and placed in a test tube and 1 mL of methanol, 1 mL (0.2N) of potassium hydroxide, and 10 mL of heptane were added to the tube and shaken for 20 seconds, after which the mixture was allowed to separate into two aliquots .

Results and discussion

Chemical composition of marjoram :

The results shown in Table (1) indicate the percentage of the basic chemical components of marjoram leaves, which were represented by moisture, protein, fat, ash and carbohydrates on a dry weight basis, and the percentage of fat in marjoram amounted to 3%. 7 The result was more than what[17]. found, as he found that the percentage of fat was 4.7%, and the percentage of protein was 5%. The result was lower than that found by [18].while the moisture content was 5.6%, which is close to the moisture content measured by [18].who found a moisture content of 5.6%, while the ash content of the leaves amounted to 4%. 17 The ash is an indicator of the plant's mineral content, where the higher the percentage of ash, the higher the percentage of elements, and the result was more than what was found [17].as the result was 14%.

Table 1: Chemical composition of marjoram

Compon ents	Fat	Protei n	As h	Moist ure	Carboh ydrates
Percenta ge %	7.3	5.5	4.1 7	5.6	77.43

As for the carbohydrate, the result of the table showed that marjoram contained 77.43% carbohydrate which is the highest percentage compared with the other compound.

important role in plant life, which provides plants with the necessary energy for growth, which is close to what was found [17.]

Estimation of active compounds in marjoram extract:

The results in Figure (1) and Table (2) indicated that marjoram extract contained seventeen active compounds that were estimated by gas chromatography-mass spectrometry. The results of the chromatographic analysis showed the presence

of 1,4-Benzenediol (25.58%), ethyl phthalate(23.54%),Hexadecane (1.53%), Eicosane (0.88%). Ethylene glycol monolauryl ether (0.62%). Neophytadienewith three percentages ranging from (3.70%) to (1.24%). Nonadecane by (0.87)% and Hexadecanoic acid, methyl ester by (2.30)%. 9,12-Octadecadienoic acid, methyl ester at (2.28)% and 9,12-Octadecadienoic acid, methyl ester at (2.28)%. The analysis showed the presence of Ethyl Linoleolate (7.27%). 9-Octadecenoic acid, (E)-from (1.59)% and Phytol from (1.37)%. Podocarpa-8,11,13-trien-16-ol, 13-isopropyl- and Ethyl Linoleolate (7.27%). Stearic acid, methyl ester (0.9% (

Table (2) Estimation of active compounds in marjoram by GC/MS

N O	Name	Area%	RT(min)
1	1,4-Benzenediol	25.58	13.66
2	Ethyl phthalate	23.54	19.694
3	Hexadecane	3.70	24.882
4	Eicosane	0.88	28.692
5	Ethylene glycol monolauryl ether	0.62	32.141
6	Neophytadiene	4.70	33.803
7	Neophytadiene	2.37	44.285
8	Neophytadiene	3.24	53.0
9	Nonadecane	0.87	69.0
10	Hexadecanoic acid, methyl ester	4.34	81.0
11	9,12-Octadecadienoic acid, methyl ester	2.28	110.0
12	Ethyl Linoleolate	7.27	39.0
13	9-Octadecenoic acid, (E)-	1.59	53.0
14	Phytol	3.37	69.0
15	Stearic acid, methyl ester	0.99	81.0
16	Podocarpa-8,11,13-trien-16-ol, 13-isopropyl-	7.39	95.0
17	Ethyl Linoleolate	7.27	110.0

Estimating antioxidant activity using the DPPH:

The DPPH scavenging ability of the aqueous and alcoholic extracts was estimated by increasing the concentration, as shown in Tables (3) and (4). The lowest values were recorded at a concentration of (2.0) mg, reaching (62.613% and 62.687%) for the

aqueous and alcoholic extracts, respectively. The highest values were (72.437% and 75.564%) for the aqueous and alcoholic extracts at a concentration of 1 mg. In comparison, the control sample showed a value of (1.0739%).

Table 3: DPPH antioxidant capacity values of aqueous extract of parsley.

sample name	Concentration	Absorbency	anti oxidant %
C1	0.12 mg	0.402	62.613
D1	0.25 mg	0.364	66.133
E1	0.5 mg	0.330	69.271
F1	1 mg	0.296	72.437
	Control	1.0739	

C1: aqueous extract=0.12mg

D1: aqueous extract=0.25mg

E1: aqueous extract=0.5mg

F1: aqueous extract=1mg

Table 4:- DPPH antioxidant capacity values of parsley alcoholic extract.

sample name	Concentration	Absorbency	anti oxidant %
C1	0.12 mg	0.401	62.687
D1	0.25 mg	0.301	71.999
E1	0.5 mg	0.295	72.558
F1	1 mg	0.274	74.564
	Control	1.0739	

C1: alcoholic extract=0.12mg

D1: alcoholic extract=0.25mg

E1: alcoholic extract=0.5mg

F1 alcoholic extract=1mg.

Conclusion

From the results we can conclude the following:

GC-MS results confirmed that marjoram seed powder extract contains active compounds and

the possibility of using marjoram extracts in extending the shelf life of meat tablets due to the antimicrobial and antioxidant compounds they contain as explained by (3)

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