Diagnosis of Some Active Organic Compounds and Study their Oxidation Reaction in Thyme Plant Leaves.

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Abstract

This study focused on diagnosis of volatile oil extracted from Breckland thyme plant by aqueous solvent by Reflux method. Thin layer chromatography was used to separate some active organic compounds from Breckland thyme leaves oil. (Toluene Ethyl acetate) (5:95) used in Thin layer chromatography as eluent. Results showed 6 spots, 5 spots were diagnosed by spectroscopic methods, qualitative and quantitative analytical methods gas chromatography (G.C). Results indicate that the 14 compound in A very small percentage 5 of that following compounds, Carvacrol, p-Cymene, Camphor, Thymol and α-Pinene are present in the following percent, 21.1, 10.1, 0.8, 24.77 and 19.92 respectively in Breckland thyme oil leaves and unknown compounds. Resulted showed oxalic acid is product of Breckland thyme leaves was study.
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Keywords: Oxalic, Breckland, oxidation, organic, plant.

Introduction

The Treatment in medicinal plants started since ancient times continued until the 19th century, when chemistry science started its progress and herbal time began, then chemists started to extract compounds from different plants [1]. Thyme has a big history of use in natural medicine with some respiratory problems contain approximately, bronchitis and chest congestion [2]. Recently, however, have researchers pinpointed of the components in Breckland thyme leaves that bring about its healing effects [3]. These plants and oils have been used for many years dating back to ancient civilizations that used them as sources of force heal [4]. Protect them from Microbial contamination; now research shows that Breckland thyme contains constituents that can prevent contamination [5].

The volatile oil of Breckland thyme is derived from thyme leaves [6]. The perennial herb, a member of the main family is used in aromatherapy, potpourri, mouth washes and elixirs, as well as, added to ointments [7]. The medical properties of Breckland thyme oils leave (which are extracted through steam distillation of fresh flowers and leaves) are due to their component Acne, Anticancer, Antispasmodic, Bactericidal, tonic, cordial, carminative, insecticide stimulant, and others [8]. The aim of the present paper was focused on isolation and characterization, active organic compounds and study the oxidation reaction of Breckland thyme leaves.

Experimental

Dry plant thyme leaves were obtained from the market, it is characterized by Education College for pure science, university of Anbar, it was air dried and packed in plastic containers.

Extraction of Breckland thyme leaves

In 1 litter a round bottomed flask, 200 cm^3 of distilled water was added to 40 gm. of plant. The extraction process was carried out for 3.5 hours, and then the volatile oil was separated from the aqueous phase with ethers and then dried with Na_2SO_4 anhydrous. Filtrate and evaporate in water bath at 100^oC [9].

(T.L.C) From different solvents, our choice the mixture of ethyl acetate and toluene in percent 5% and 95%, respectively, as mobile phase while suitable plastic paper coated with silica gel was chosen as a stationary phase. The chromatogram shows five zones, each zone have been scratched, isolate and dissolve in ether, than filtration. Removal of the solvent gave the desired compounds. The identification process depends on, IR spectra

and the value of R_f when compared with the standard. Compound under the same condition four zones were identified, while two zones are unknown. Gas chromatography also used to identify all components of oil [10].

Gas chromatography

Gas chromatography analysis was carried out in resources directorate by using a GC apparatus, Mustansiria, College of science using Shimadzu GCMS-QP2010Ultra with a flame ionization detector. A thermon-600T fused silica (50m X 0.25 mm ID) film thickness 0.25 µm to 60 µm long and capillary column coated with o. 3µm layer of macrogol 20,000 was used. The carrier gas has nitrogen at a flow rate of 2 ml/min.; On the Column Flow; both the injection and detector temperature were 240°C. The oven temperature column was kept at 70°C for 10 min., programmed to rise up to 180°C at a rate of 2°C/min., and then kept constant at 180°C for 30 min. Pressure: 100.0 KPa. Start Time: 4.50min End Time: 27.00min. The essential oil components were identified by comparing their retention times with those of authentic samples. Gas Chromatography was used for detection of the compounds quantitative and qualitative compounds in Breckland thyme. While mass analyzer detection about molecular weight of active compound in Breckland thyme.

Oxidation Reaction

For examining the oxidation reactions of aqueous extract Breckland thyme plant leaves after separation oil using reagent KMnO4 in an acidic media, by titration process with aqueous extract of Breckland thyme until the occurrence of change the color of extracted fully. The solution cooled to a temperature 5°C for 12 hs. And filtrated the precipitate was kept to determine the effect of $KMnO_4$ by diagnosis in UV and FT IR Techniques by using distilled water as a solvent [11].

Results and Discussion:-

TLC test of Breckland thyme oil was revealed. This chromatogram showed six spots, then four spots were characterized by comparing with standard materials, and the results of these comparisons were predicted in Table (1) and Fig.(1).

Table (1)
Retention factor (Rf) of identification compound for volatile oil.

	Compound	Flow rate R_f
1	P-Cymene	0.91
2	Thymol	0.85
3	Carvacrol	0.64
4	α-Pinene	0.33
5	Camphor	0.20

Each component of this chromatogram was identified by spectroscopic methods. However Each zone was scratch was carefully isolated and dissolved in diethyl ether.

The spectral data of isolated compounds were shown in Table (2), and Figs. (2,3,4 and 5) were shown that compounds 1 and 2 exhibits a broad band appearing in 3267-3485cm^{-1} assigned to the stretching vibration of (OH) group. While compound (3) exhibits the band 1750 cm^{-1} was related to C=O stretching. Compound (4) exhibits the following bands, 3080 cm^{-1} was due to the aromatic C-H stretching band, 2982-2858 cm^{-1} was stretching due to the C-H methyl group, 1539 cm^{-1} was due to C=C stretching of ring besides other characteristic bands. Compound (5) exhibits the following bands, 2976 cm^{-1} was due to the alkan C-H stretching band, 2862-2911 cm^{-1} was stretching due to the C-H methyl group, 1444 cm^{-1} was due to C=C stretching and 1381 cm^{-1}.

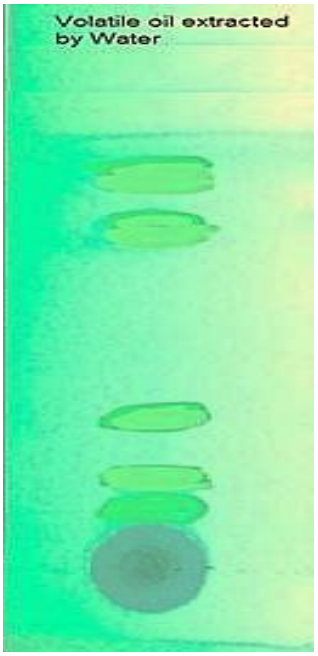


Fig.(1): TLC chromatogram.

Table (2)
Some characteristics bands of appeared compounds in the studied.

NO.	Compound	Assignment cm^{-1}				
		OH	CH Str.	C=C Str.	OH bending	Isopropyl group
1	Thymol	3485 broad	2962-2870	1570-1381	1350	1285
2	Carvacrol	3267	2926-2856	1521,1460,1458	1458-1241	1395
3	Camphor	-	2950	-	-	1390-1375
4	p-Cymene	CH aromatic str. 3080	2982-2858	1539		
5	α-Pinene	2976 CH ALK.	2862-2911	1444	-	CH3. 1381

C-O str. In phenol	CH aromatic bending	C=O
1246	792	1750
1251	802	-
-	750	-
-	794	-
-	-	-

Gas chromatography

The density and percentage of weight for active organic compound in Breckland thyme oil were found as follows: the percentage is (α-Pinene) 39.85 $*10^{-3}$ Intensity and 19.92w/w %, (*p*-cymene) is $20.9*10^{3}$ g.cm-3 and the percentage 10.1 %, (Camphor) is 1.9*103 g.cm-3 and the percentage is 0.8%, (Thymol) is 39.55*103 g.cm-3 and the percentage is 24.77%%. Carvacrol is 47.4*103 $g.cm^{-3}$ and the percentage is 21.1%. While the volume that is used in measurement is ($75*10^{-3}$ cm3) in Fig.(6).

Table (3)
Quantitative and qualitative composition (w/w %) of the Breckland thyme essential oils studied.

No.	Components	Retention time min	Density $g.cm^{-3}$	w/w%
1	α-Pinene	5.390	39.85 $*10^{-3}$	19.92%
2	P-Cymene	6.235	$20.9*10^{-3}$	10.44 %
3	Camphor	12.820	$1.9*10^{-3}$	0.945 %
4	Thymol	17.420	$39.55*10^{-3}$	19.77%
5	Carvacrol	17.865	$47.4*10^{-3}$	23.7 %

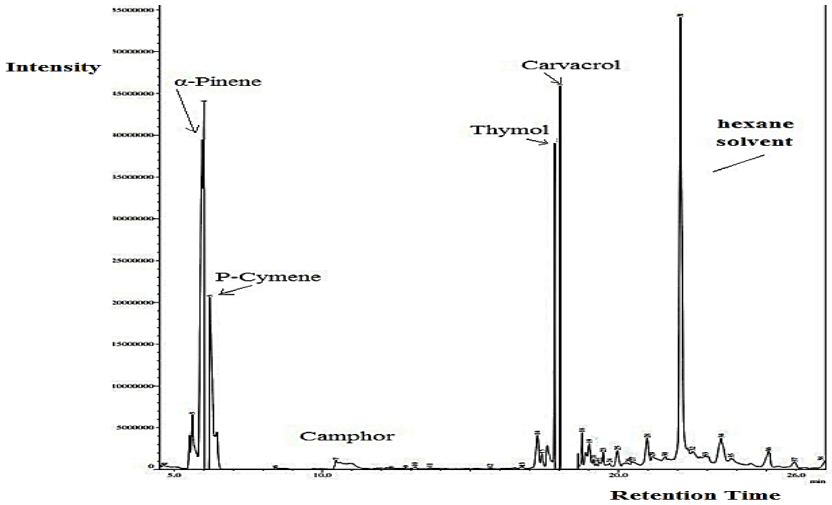
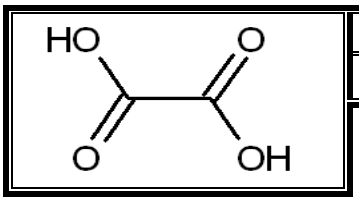


Fig.(6): Gas chromatography of active organic compound in the volatile oil of Breckland thyme by using solvent Hexane.

Oxidation reaction

The reaction was observed which formed precipitate to be built from manganese dioxide and the completion of the reaction was obtained from colorless crystalline precipitate. The precipitate was diagnosed by using (UV-visible) and FT.IR techniques. The results of oxidative carbonyls to oxalic acid $H_2C_2O_4.2H_2O$ as by-products main and using an aqueous solvent.

As a result, the oxidation react of water extract containing sugars. It was shown in Fig.(7) which showed λmax at 320 nm. This is due to π -π*, and it showed λmax at 370 nm and this is due to n-π* by UV-vis. It Makes Breckland thyme more stable towards the air [12]. FT.IR spectrum of oxidation reacts in aqueous extract of oxalic acid as shown in Fig.(8), and Table (3-13). FT.IR Data of oxalic acid, exhibits a band appearing at region 3415 cm^{-1} st. OH, 1741 cm^{-1} st. C=O. 1234 cm^{-1} st. C-O. 729 cm^{-1} bend OH.



3415 cm^{-1} st. OH	1234 cm^{-1} st. C-O
1741 cm^{-1} st. C=O	729 cm^{-1} bend OH

IR spectrum of Oxalic acid as solvent at room temperature.

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