Plant Archives Vol. 18 No. 2, 2018 pp. 2263-2269 **e-ISSN:2581-6063 (online), ISSN:0972-5210**

PHYTOCHEMICAL ANALYSIS OF *FRANKENIA AUCHERI* JAUB. ET SPACH (FRANKENIACEAE) BY GC-MS AND FT-IR TECHNIQUES

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Abstract

In this study, the phytochemical analysis of Halophyte plant *Frankenia aucheri* Jaub. et Spach (Frankeniaceae) in methanolic leaf extracts was taken up in Iraq and reported for the first time The GC-MS analytical studies revealed the presence of twenty-one composites of different types such as alkaloids, flavonoids, phenolics, terpenoids, esters, etc. Also, the study summarized the information concerning with various functional groups like alkenes, alkanes, and aliphatic fluoro compounds have been identified by FT-IR analysis compared with original and methanolic extract. These constituents may be responsible for pharmacological activities and may add new knowledge to the information in the traditional medical systems..

Key words : *Frankenia aucheri*, phytochemical compounds, methanolic extract, GC-MS and FT-IR.

Introduction

Frankenia aucheri Jaub. et Spach. is one of a halophyte species that belong to Frankeniaceae. It was described according to Al-Tameme's study in Iraq as a herb perennial with a woody base, procumbent, densely branched, puberulent or white-hirsute, usually leaves revolute margined in whorls of 3-4 leaves, leaf blade linear-oblong or linear, spreading white-ciliate hairs at base, Flowers corymbose terminal clusters (Al-Tameme, 2016).

Frankenia aucheri has a native name MILLAIH or SHUWAIWA (meaning "salt") and synonym named *Frankenia hirsute* L. var. *erecta* Boiss (Blakelock, 1955) or *Frankenia hirsuta* L. and *F. hispida* D. C. (Paulsen, 2013). It can grow in saline desert soils, sometimes with high water table or even waterlogged, on banks of a wadi, on the edge of an oasis, at altitude up to 150m, camels and sheep are only able to eat this when they can get fresh grass at the same time (Blakelock, 1955). Also, because *F. aucheri* has beautiful flowers, it considered as an ornamental plant which contains gummy resin, kaempferol, quercetin and tannin utilized for sticking blade cutting edges and to seal stoneware (Fegler and Mose, 1985). The last previous authors pointed the liquid extract of *F. aucheri* is used for catarrh infections, and mucous releases from the nose and genitourinary tracts additionally tea of roots employed as a cold therapy (Fegler and Mose, 1985), in another study used to enhance lactation in cows and camels especially in winter by mixing the rhizome of *F. aucheri* powder with milk (Alyemeni *et al*., 2010).

Despite the spread of *Frankenia* genus in many areas, particularly Iraq's neighboring regions such as Turkey (Webb, 1966), Lebanon, Jordan and Palestine (Townsend and Guest, 1980), Egypt (Salama *et al*., 1999), Syria (Al-Oudat and Qadir, 2011), Qatar (Abulfahij *et al*., 2002), Kuwait (Malallah *et al*., 2003) and Irano-Turanian and Mediterranean (Youcef *et al*., 2012), but no reports about phytochemicals are available on this genus expect phytochemical study on *Frankenia pulverulenta* by Altameme (2017). Therefore, the aim of this work was investigated substances have a biological effect and determined whether the GC-MS and FT-IR techniques can differentiate in other halophyte species of the same genus.

Materials and Methods

Collection plant and preparation of extraction

Clean and free of dust or fungi dry leaves of *F. aucheri* was collected randomly from various specimens were kept in Babylon University Herbarium. Took the

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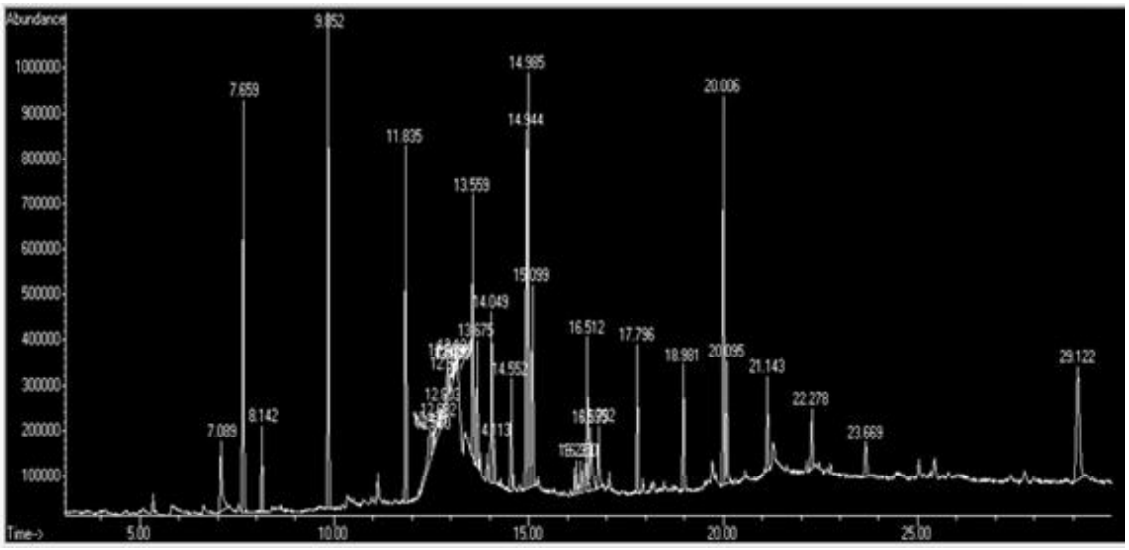

Fig. 1 : Chromatogram profile for GC-MS of *F. aucheri.*

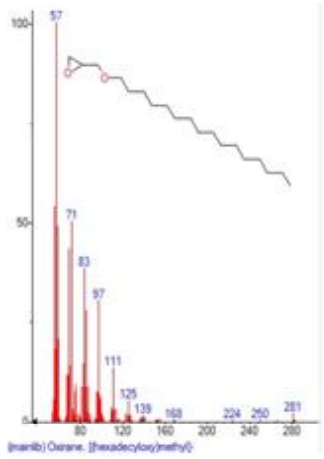

Fig. 2 : Chromatogram of Oxirane, [(hexadecyloxy)methyl]- in *F. aucheri.*

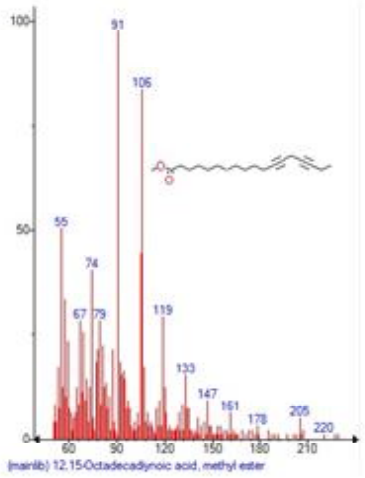

Fig. 3 : Chromatogram of 12,15-Octadecadiynoic acid, methyl ester in *F. aucheri.*

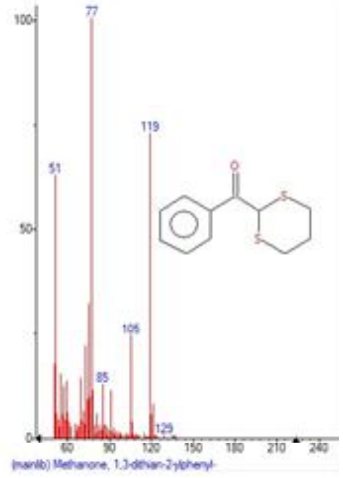

Fig. 4 : Chromatogram of Methanone, 1,3-dithian-2-ylphenyl- in *F.aucheri.*

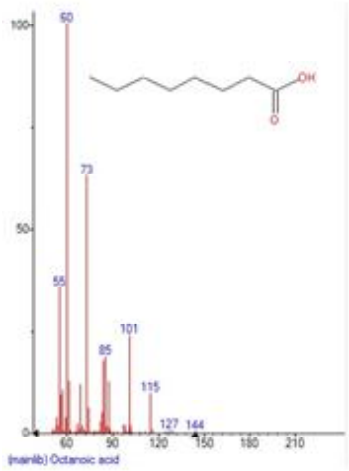

Fig. 5 : Chromatogram of Octanoic acid in *F.aucheri.*

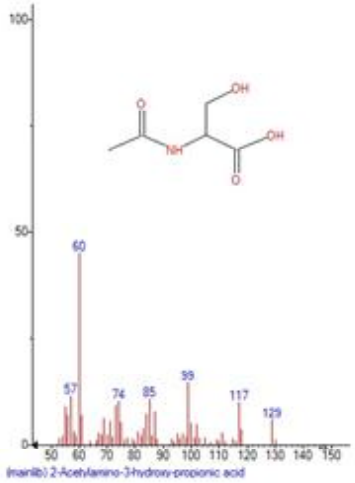

Fig. 6 : Chromatogram of 2-Acetylamino-3-hydroxy-propionic acid in *F.aucheri.*

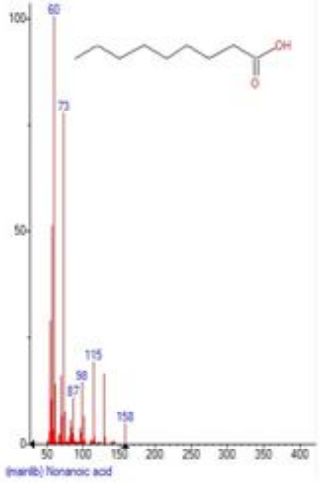

Fig. 7 : Chromatogram of Nonanoic acid in *F.aucheri.*

Table 1 : Components detected in the extract of *F. aucheri* leaves.

Retention time (min)	Name of the compound	Exact mass	Molecular formula	Structure
3.310	Oxirane , [(hexadecyloxy)methyl]-	298.28718	$C_{19}H_{38}O_2$	Figure 2
3.917	12,15-Octadecadiynoic acid, methyl ester	290.22458	$C_{19}H_{34}O_2$	Figure 3
5.124	Methanone ,1,3-dithian-2-ylphenyl-	224.032957	$C_{11}H_{12}OS_2$	Figure 4
5.891	Octanoic acid	144.115029	$C_8H_{16}O_2$	Figure 5
6.875	2-Acetylamino-3-hydroxy-propionic acid	147.053158	$C_5H_9NO_4$	Figure 6
7.098	Nonanoic acid	158.13068	$C_9H_{18}O_2$	Figure 7
7.973	6-Acetyl-â-d-mannose	222.073953	$C_8H_{14}O_7$	Figure 8
8.139	2,5-Dimethylhexane-2,5-dihydroperoxide	178.120509	$C_8H_{18}O_4$	Figure 9
8.443	Desulphosinigrin	279.077658	$C_{10}H_{17}NO_6S$	Figure 10
8.660	Pterin-6-carboxylic acid	207.039239	$C_7H_5N_5O_3$	Figure 11
10.222	2-Oxazolamine , 4,5-dihydro-5-(phenoxymethyl)-N-[(phenylamine	311.126991	$C_{17}H_{17}N_3O_3$	Figure 12
10.937	Dihydrotecomanine	181.146665	$C_{11}H_{19}NO$	Figure 13
11.264	N,N'-Bis(Carbobenzyloxy)-lysine methyl (ester)	428.194736	$C_{23}H_{28}N_2O_6$	Figure 14
12.883	3-O-Methyl-d-glucose	194.079039	$C_7H_{14}O_6$	Figure 15
14.062	Phthalic acid, butyl tetradecyl ester	418.30831	$C_{26}H_{42}O_4$	Figure 16
14.090	Oxiraneundecanoic acid , 3-pentyl-,methyl ester , cis-	312.266445	$C_{19}H_{36}O_3$	Figure 17
14.966	Phthalic acid, isobutyl octadecyl ester	474.37091	$C_{30}H_{50}O_4$	Figure 18
16.144	Corynan-17-ol,18,19-didehydro-10-methoxy-, acetate (ester)	368.209993	$C_{22}H_{28}N_2O_3$	Figure 19
16.362	6-epi-shyobunol	222.198365	$C_{15}H_{26}O$	Figure 20
16.831	Octadecanoic acid	284.27153	$C_{18}H_{36}O_2$	Figure 21
17.432	Ethyl iso-allocholate	436.318874	$C_{26}H_{44}O_5$	Figure 22

sample to crush by kitchen grinder for 20 seconds, then prepared a powder from it by using blender machine (Al-Tameme, 2015). According to Harborne (1984), 2-3g of powder was soaked and extracted for 48-72 h. with 20-30 ml of methanol at room temperature with shaker during the initial six hours, Filtered the extracts by using filter paper (Whatman No. 1) for removing the residue.

GC-MS analysis and a definition of components

2µL of methanolic extract of leaves *F. aucheri* was performed on GC Clarus 500 Perkin Elmer interfaced to a mass spectrometer (GC-MS) and MS: Turbo mass gold – Perkin Elmer Turbomass 5.1 spectrometer and operating in EI mode at 70 eV, equipped with a splitless injector (250°C). The conditions were as follows: Column Elite-1ms fused silica capillary column (30×0.25mm ID x 0.25m film thickness, composed of 100% Dimethylpolysiloxane). Helium was a carrier gas at a continual flow of 1 ml/min at 280ºC and an injection volume of 2ml was used (10:1 split ratio), Temperature in ion-source was 200ºC. The temperature in the oven was programmed from 40ºC, with an increment of 5ºC min^{-1}, to 280ºC hold for 9 min. Mass spectra were taken at 70eV (electron ionization technique) at a scan interval of 0.5 seconds and fragments were scanned from 45 to 450 Da. Total running was 36min. Compounds in the chromatograms were determined by differentiation of their mass spectra with those in NIST library database, and through the comparison between their retention index with those reported in the literature (Altameme *et al*., 2015).

FTIR Spectroscopic analysis

10 mg of the dried extract powder was inflod in 100 mg of KBr pellet, in order to prepare to thin sample disc. The powder was loaded in FT-IR spectroscopy (Shimadzu, IR Affinity1, Japan), with a scan range from 400 to 4000 cm^{-1} with a resolution of 4cm^{-1} (Jasim *et al*., 2015).

Results and Discussion

As we know, plants are the abundant source of derivative metabolites with interesting biological effectiveness. Generally, these secondary metabolites are important provenance with an assortment of structural arrangements and properties (De-Fatima *et al*., 2006) and may be responsible for displaying different pharmacological activities (Gu *et al*., 2016). The identification of each compound was made through gas chromatography-mass spectrometry (GC-MS), which is a useful tool for quantitative and qualitative analysis of a

Table 2 : FT-IR peak values of solid analysis of *F.aucheri* leaves extract (original).

Group frequency	Functional group	Stretching	Intensity	Peak values (Wave number cm^{-1})	No.
675-995	Alkenes	C-H	74.338	692.44	1.
675-995	Alkenes	C-H	71.080	871.82	2.
675-995	Alkenes	C-H	72.401	916.19	3.
675-995	Alkenes	C-H	53.256	989.48	4.
1340-1470	Alkanes	C-H	79.893	1404.18	5.

Table 3 : FT-IR peak values of solid analysis methanolic extract of *F.aucheri* leaves.

Group frequency	Functional group	Stretching	Intensity	Peak values (Wave number cm^{-1})	No.
675-995	Alkenes	C-H	79.165	873.75	1.
675-995	Alkenes	C-H	65.187	989.48	2.
-	Unknown	-	64.751	999.13	3.
1000-1050	Aliphatic fluoro compounds	C-F	65.427	1014.56	4.

broad domain of compounds, in addition it is a cleaner, faster and less expensive than the traditional extraction methods also, it has been closely used in medical, biological, and nutrition survey (Harborne, 1984). For this reason adopted this method in this work and the results showed a distinction in the number of compounds within species under the study.

Profile chromatography of leaves extract *F.aucheri* in table 1 was able to identify the presence of twenty-one main peaks fixed as follows. Oxirane, [(hexadecyloxy) methyl]- was represented as the first peak; secondly peak was seemed to be 12,15-Octadecadiynoic acid, methyl ester; The rest of peaks deliberated on Methanone,1,3-dithian-2-ylphenyl-; Octanoic acid; 2-Acetylamino-3-hydroxy-propionic acid; Nonanoic acid; 6-Acetyl-β-d-mannose; 2,5-Dimethylhexane-2, 5-dihydroperoxide; Desulphosinigrin; Pterin-6-carboxylic acid; 2-Oxazolamine, 4,5-dihydro-5-(phenoxymethyl)-N-[(phenylamine; Dihydrotecomanine; N,N'-Bis (Carbobenzyloxy)-lysine methyl (ester); 3-O-Methyl-d-glucose; Phthalic acid, butyl tetradecyl ester; Oxiraneundecanoic acid, 3-pentyl-, methyl ester, cis-; Phthalic acid, isobutyl octadecyl ester; Corynan-17-ol, 18, 19-didehydro-10-methoxy-, acetate (ester); 6-epi-shyobunol; Octadecanoic acid and Ethyl iso-allocholate.

The FTIR spectrum of crude and leaf extracts prepared in methanol of *F.aucheri* are given in figs. 23 and 24. The data on the peak values and the probable functional groups present in tables 2 and 3.

Results of FTIR analysis for original powder of *F.aucheri* leaves are demonstrated the existence of alkenes and alkanes compounds which appeared the main peaks at 692.44, 871.82, 916.19, 989.48 and 1404.18 cm^{-1} vibration C-H stretching (table 2 and fig. 23), in constant the methanolic extract *F.aucheri* leaves (table 3 and fig. 24) are explained the band at 873.75 and 989.48 cm^{-1} showed alkenes as C-H stretching; the band at 1014.56 cm^{-1} showed Aliphatic fluoro compounds and the band at 999.13 cm^{-1} showed unknown compound. In this study, results demonstrated the similarity between two species of *Frankenia* (*F. pulverulenta* and *F. aucheri*) by possessing four common chemical compounds that were Dihydrotecomanine; 6-Acetyl-β-d-mannose; 3-O-Methyl-d-glucose and Ethyl iso-allocholate (Altameme, 2017).

So the outcome of this study united in opinion with the past observations which found by numerous plant biologist and taxonomist. The results of the spectrometry have confirmed the presence of effective constituents in the crude powder and methanolic extracts of *Frankenia* species which are responsible for many biological activities such as a presence of Ethyl iso-allocholate in this plant makes it suitable for anti-inflammatory activity and anti-infective (Hussein *et al*., 2016) and proved having *Frankenia thymifolia* Desf. antioxidant and antimicrobial properties (Wided *et al*., 2011), while a presence of Octadecanoic acid and n-Hexadecanoic acid compounds in halophyte plant could be the useful candidates to serve as a genetic source for this reason (Abdel-Hamid, 2014).

Thus in future, we need extra progressive spectroscopic studies are required for the structural illustration and recognition of bioactive principles present in the leaves *Frankenia* species could be used in practical research in medicine, agriculture and industry.

Conclusion

Frankenia aucheri is one of the species of *Frankenia* genus, which distributed in Iraq. Thus the

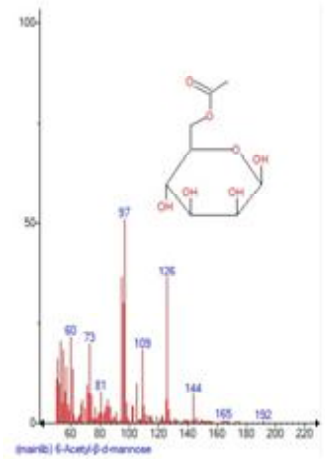

Fig. 8 : Chromatogram of 6-

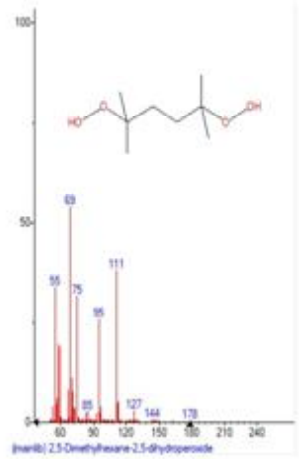

Fig. 9 : Chromatogram of 2,5-

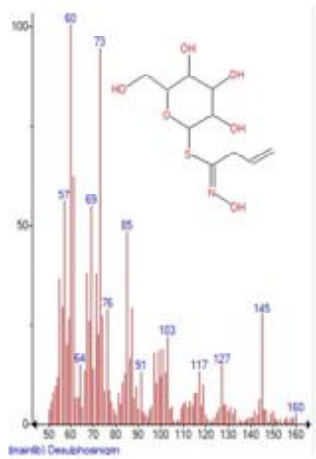

Fig. 10 : Chromatogram of

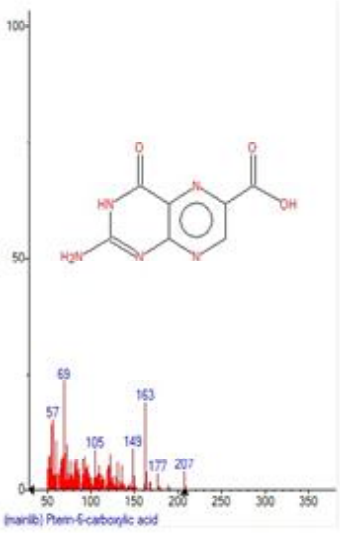

Fig. 11 : Chromatogram of Pterin-6-carboxylic acid in *F.aucheri*.

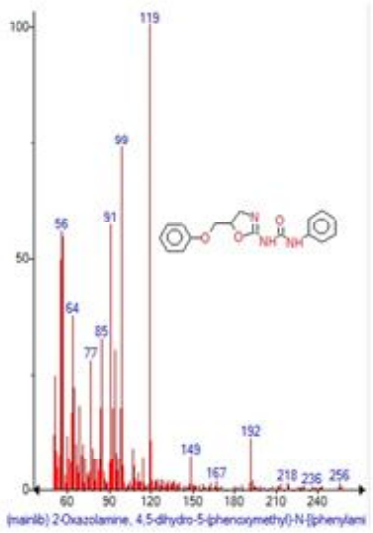

Fig. 12 : Chromatogram of 2-Oxazolamine, 4, 5-dihydro-5-(phenoxymethyl)-N- [(phenylamine in *F. aucheri*.

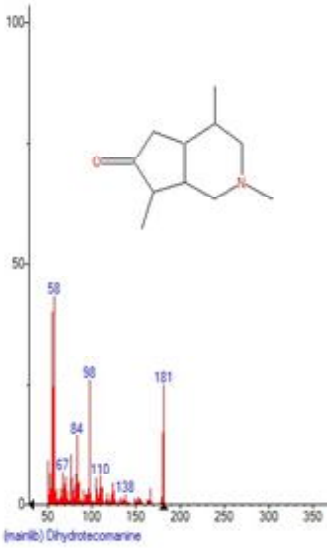

Fig. 13 : Chromatogram of Dihydrotecomanine in *F.aucheri*.

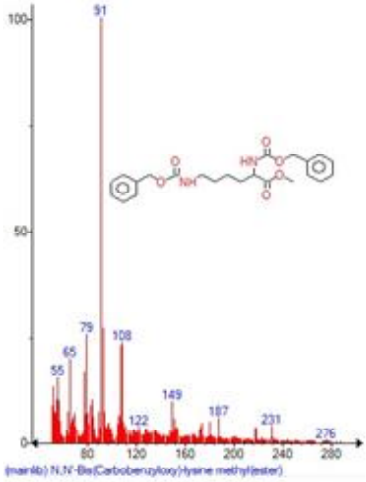

Fig. 14 : Chromatogram of N, N'-Bis(carbobenzyloxy)-lysine methyl (ester) in *F.aucheri*.

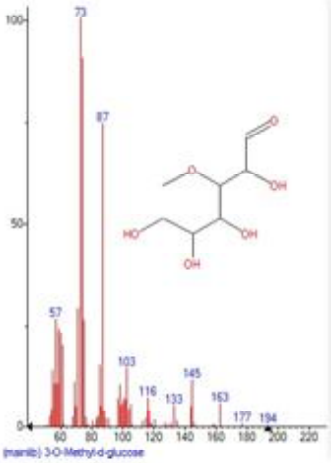

Fig. 15 : Chromatogram of 3-O-Methyl-d-glucose in *F.aucheri*.

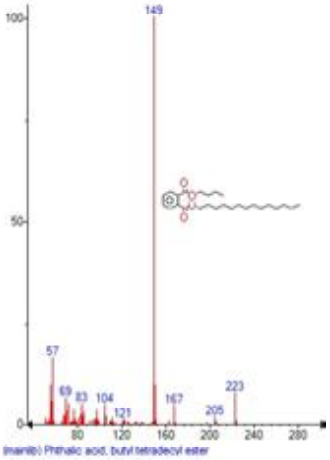

Fig. 16 : Chromatogram of Phthalic acid, butyl tetradecyl ester in *F.aucheri*.

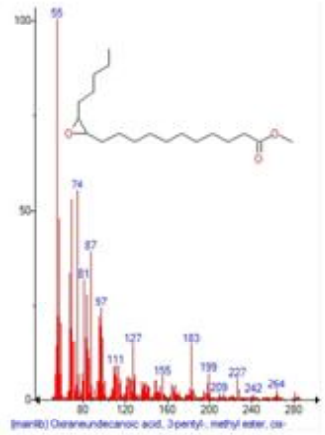

Fig. 17 : Chromatogram of Oxiraneundecanoic acid, 3-pentyl-, methyl ester, cis- in *F.aucheri*.

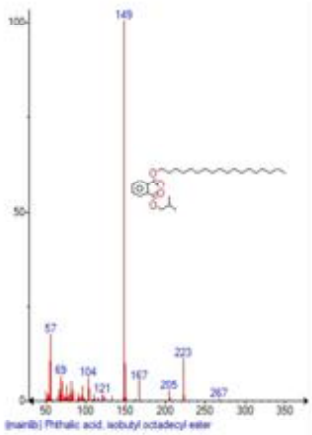

Fig. 18 : Chromatogram of Phthalic acid, isobutyl octadecyl ester in *F.aucheri*.

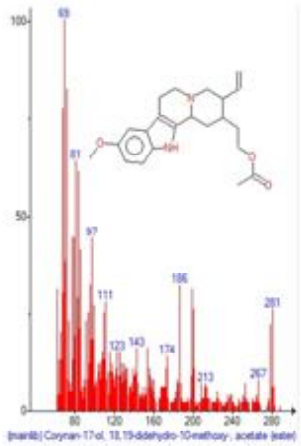

Fig. 19 : Chromatogram of Corynan-17-ol,18,19-didehydro-10-methoxy-, acetate (ester) in *F. aucheri*.

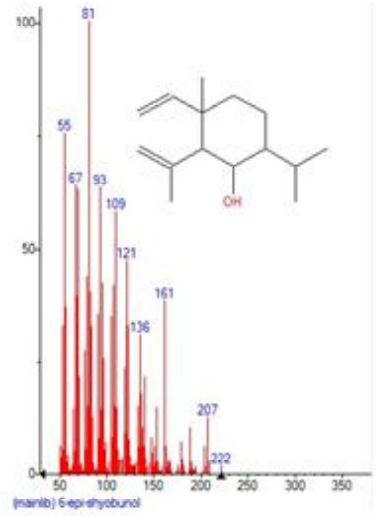

Fig. 20 : Chromatogram of 6-epi-shyobunol in *F.aucheri*.

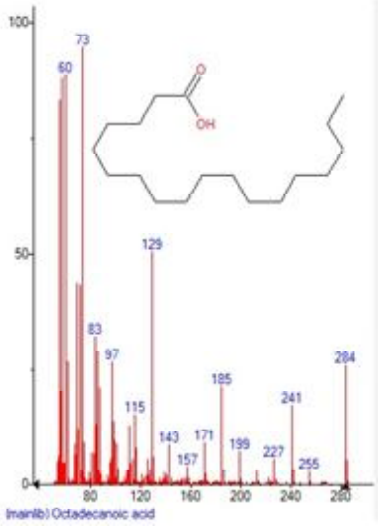

Fig. 21 : Chromatogram of Octadecanoic acid in *F.aucheri*.

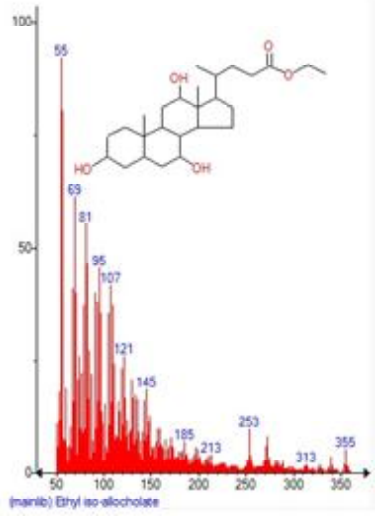

Fig. 22 : Chromatogram of Ethyl iso-allocholate in *F.aucheri*.

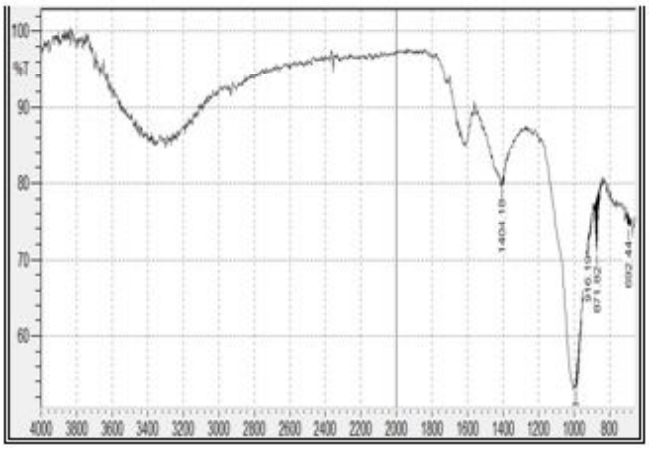

Fig. 23 : FT-IR profile solid analysis of *F.aucheri* leaves extract (original).

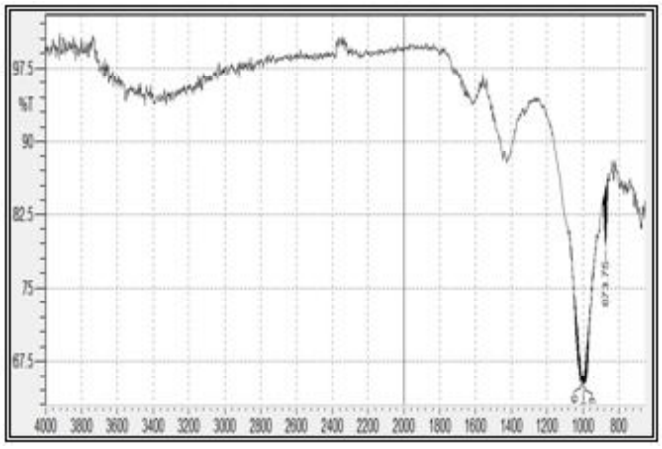

Fig. 24 : FT-IR profile solid analysis of *F.aucheri* leaves methanolic extract.

phytochemical screening that carried by GC-MS and FT-IR techniques have led to the isolation and identification twenty-one of structurally chemical compounds and presence of various functional groups in the crude and methanolic extract, which are responsible for many biological activities.

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