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FOURIER TRANSFORM INFRARED SPECTROSCOPIC ANALYSIS OF ETHANOLIC LEAF EXTRACT OF *ALTERNANTHERA SESSILIS* (L.) AND *TRIGONELLA FOENUM-GRAECUM* (L.)

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ABSTRACT The present study was designed to investigate the phytochemical screening using FT-IR analyses for the qualitative identification of bioactive compounds in *Alternanthera sessilis* (L.) *Trigonella foenum-graecum* (L.) leaves extract. The qualitative FT-IR spectrum profile of ethanolic extract of *Alternanthera ssesilis* (L.) showed the peaks at 3294.42/ cm^{-1}, 2918.30 cm^{-1}, 2850.79/ cm^{-1}, 2362.80/ cm^{-1}, 1737.86/ cm^{-1}, 1624.06/ cm^{-1}. The FT-IR spectra analyses of *Trigonella foenum-graecum* (L.) confirmed the presence of different functional groups with a peak value of 537.11 cm^{-1}, 622.50 cm^{-1}, 834.43cm^{-1}, 1063.64 cm-1, 1102.83 cm-1,243.14 cm^{-1}, 1385.24 cm^{-1}, 1423.53 cm^{-1}, 1649.17cm^{-1},1736.40 cm^{-1}, 2144.79cm^{-1}, 2852.51 cm^{-1}, 2921.41 cm^{-1} and 3356.40 cm^{-1} in the ethanolic extract of leaves. These peak values confirmed the presence of characteristic functional groups of alkyl halides, alkynes, aromatics aliphatic amine, aromatic amines, amines, aromatics, nitriles, alkenes, alcohols and phenols in the leaf extract. They could be responsible for the various medicinal properties of *Alternanthera ssesilis* (L.) and *Trigonella foenum-graecum* (L.). The presence of various bioactive compounds justifies the use of the leaves for various ailments by traditional practitioners. The result of this study offer a platform of using *Alternanthera ssesilis* (L.) and *Trigonella foenum-graecum* (L.) leaves as herbal alternative for various diseases.

KEYWORDS : Ethanol extract, FT-IR, spectroscopy, functional group.

INTRODUCTION

Medicinal herbs are used in traditional medicine to cure various diseases. They consists of biologically active ingredients therefore they are used for the treatment of a large number of infectious diseases (Nisar *et al*., 2011; Sasidharan *et al*., 2011). They serve as source of medicine, ornamental purposes, flavouring, food additives and preservatives (Jeeva *et al*., 2006; Raja *et al*., 2011). The use of herbs in the treatment, management of diseases and disorders dates back to pre-historic days (Balakumar *et al*., 2011). The FT-IR has proven to be a valuable tool for the characterization and identification of compounds or functional groups (chemical bonds) present in an unknown mixture of plants extract (Hazra *et al*., 2007; Eberhardt *et al*., 2007). In addition, FT- IR spectra of pure compounds are usually so unique that they are like a molecular "fingerprint". For most common plant compounds, the spectrum of an unknown compound can be identified by comparison to a library of known compounds (Hazra *et al*., 2007).

Alternanthera sessilis commonly known as ponnanganni which belongs to botanical family amaranthaceae. In Ayurveda, this plant is considered bitter, astringent, sweet, cool and pungent. It is given to treat diarrhoea, skin diseases, night blindness, indigestion, and fever (Wild, 1995). Fenugreek is an important and one of the oldest medicinal plants. It is a self pollinating annual leguminous bean which belongs to Fabaceae family (Balch, 2003) commonly known as Indian methi. Historical and theoretical uses of fenugreek leaves includes abortifacient, appetite stimulant, baldness, boils, breast enhancement, bronchitis, cellulites, constipation, cough, diarrhea, eczema, flatulence, galactagogue, hepatitis disease, hernia, indigestion, leg ulcers, menopausal symptoms, myalgia, postmenopausal vaginal dryness, hyperglycemia, tuberculosis and wound healing. Therefore in the present study FT-IR techniques are employed to evaluate the IR finger print in the leaf extracts of ponnanganni and fenugreek and to analyse the functional groups of phytoactive compounds present in it.

MATERIALS AND METHODS

Collection And Processing Of Plant Material:

The leaves of the plant ponnanganni and fenugreek were collected from the natural habitats of Thiruchirappalli district, Tamil Nadu, India. For the analysis, Fresh leaves of ponnanganni (*Alternanthera sessilis,* L.) fenugreek (*Trigonella foenum-graecum,* L.) were collected and washed properly to remove all the durt, debris and soil with double distilled water and dried under shade, dust-free condition for one week at room temperature. The leaves were then made powder in a mechanical grinder. The leaf powder was used for further study.

Preparation Of Leaf Extract

Five grams of leaf powder was weighed, 25ml of ethanol was added and kept for 3 days. The extract was filtered using Whatman No.1 filter paper and the supernatant was collected. The residue was again extracted two times (with 3 days of interval for each extraction) and supernatants were collected. The supernatants were pooled and evaporated (at room temperature, 28 ± 1° C) until the volume was reduced. Extracts of the leaf powder prepared was stored in air tight bottles for further analysis.

FT-IR Spectroscopic Analysis

A dried powder of solvent extracts of plant material was used for FT-IR analysis. 10mg of the dried extract powder was encapsulated in 100 mg of KBr pellet, in order to prepare translucent sample disc the powdered sample of each plant specimen was loaded in FT-IR Spectroscope (Perkin Elmer Spectrophotometer system), with a scan range from 400 to 4000 cm-1 with a resolution of 4cm-1. The peak values of the FT-IR were recorded. Each and every analysis was repeated twice for the spectrum confirmation.

RESULTS AND DISCUSSION

FT-IR spectroscopic analyses was reported on *Alternanthera sessilis* (L.) and *Trigonella foenum-graecum,* (L.) revealed the presence of different functional groups of the bioactive compounds present in the ethanolic leaves extract in the form of peaks (Figure -1 & 2). The absorption bands, the wave number (cm^{-1}) of prominent peaks obtained from absorption spectra were described (Table-1).

In the leaf extract of *Alternanthera sessilis*, L. the peak at 3294.42/ cm^{-1} was assigned to the functional group –OH stretching, presence of alcohols and phenols. The peak at 2918.30 cm^{-1} revealed the presence of the –CH-stretching vibration with functional group of alkenes. The functional group at 2850.79/ cm^{-1} was –C-H stretch with the presence of alkenes. 2362.80/ cm^{-1} and 1737.86/ cm^{-1} were found to be an unknown compounds. The peak at 1624.06/ cm^{-1} was assigned to the functional group N–H bend primary amines. The peak at 1315.45 cm^{-1}, 1238.30 cm^{-1}, 1101.35 cm^{-1} and 1018.41 cm^{-1} were due to the C-O stretching vibration, showed the presence of alcohols carboxylic acids, esters and ethers. C–Br stretch alkyl halides stretch was identified in

the peaks 659.66, 597.93 and 551.64 cm^{-1}. The peaks at 925.83 cm^{-1}, 474.49 cm^{-1}, 416.62 cm^{-1} and 405.05 cm^{-1} were unknown compound.

In the FT-IR spectrum of *Trigonella foenum-graecum,* L. the functional groups were separated based on its peak ratio. The peak at 537.11 cm^{-1} was assigned to the functional group C-Br stretch alkyl halides. The functional group at 622.50 cm^{-1} was –C(triple bond)C–H: C–H bend alkynes. The peak at 834.43cm^{-1} assigned to the C–H "oop" stretching vibration confirmed the functional group aromatics. The peak at 1063.64 and 1102.83 cm-1 assigned to the C-N stretching which means that some aliphatic amine compounds are present in *Trigonella foenum-graecum* ethanolic leaves extract. The functional group aromatic amines with the C-N stretching were revealed by the presence of peak at 1243.14 cm^{-1}. The peak at 1385.24 cm^{-1} was corresponding to N=O Bend suggests the presence of the functional group nitro in the leaves extract. C-C stretch (in-ring) aromatics compounds were recorded by the presence of peak at 1423.53 cm^{-1}. The presence of N-H Bend primary amines confirmed by the peak at 1649.17cm^{-1}. The leaves extract of *Trigonella foenum-graecum,* L. exhibited a characteristic band at 1736.40 cm^{-1} indicating the presence of carbonyl (C=O).

Multiple bonding Nitrile compounds were recorded by the presence of peak at 2144.79cm^{-1}. The peaks at 2852.51 cm^{-1} and 2921.41 cm^{-1} confirmed the presence of C-H stretch alkenes. The peak at 3356.40 cm^{-1} was assigned to the O-H stretch, H-bonded alcohols and phenols in the ethanolic extract of leaves of *Trigonella foenum-graecum* L.

Manoj and Ragothaman (1999) suggested that the more intense bands occurring at 3419 cm-1 2924, 2854, 1635, 1406, 1242, 1070, and 617 cm-1 corresponding to O-H/N-H, C-H, C-O and C-CI/C-CS stretching/bending vibrations respectively suggest the presence of amino acids, alkenes, nitrates, ethers, organic halogen compounds and carbohydrates in plants. Muruganantham *et al*. (2009) carried out the FT-IR and EDS spectral analysis of plant parts like leaf, stem and root of the medicinal plants, *Eclipta alba* and *Eclipta prostrata* and reported the presence of characteristic functional groups of carboxylic acids, amines, amides, sulphur derivatives, polysaccharides, nitrates, chlorates, and carbohydrate that are responsible for various medicinal properties of both herbal plants.

Ragavendran *et al*. (2011) screened the functional groups of carboxylic acids, amines, amides, sulphur derivatives, polysaccharides, organic hydrocarbons, halogens that are responsible for various medicinal properties of *Aerva lanata*. Starlin Parag and Bhanu Raman (2013) analyzed the methanolic leaf extract of *Ampelocissus latifolia* by FT-IR and reported that the transition metal carbonyl compounds and aliphatic fluoro compounds were only present in the extract.

CONCLUSION

The presence of characteristic functional groups of alkyl halides, alkynes, aromatics aliphatic amine, aromatic amines, amines, aromatics, nitriles, alkenes, alcohols and phenols could be responsible for the various medicinal properties of *Alternanthera sessilis,* L. and *Trigonella foenum-graecum,* L. The presence of various bioactive compounds justifies the use of the leaves for various ailments by traditional practitioners. It could be concluded that *Alternanthera sessilis,* L and *Trigonella foenum-graecum,* L. contains various bioactive compounds. Further studies are needed with this herb to identify the unknown functional groups, isolate, characterize and elucidate the structure of the bioactive compounds which are responsible for the antimicrobial activity and other medicinal values.

Figure -1 The FT-IR Spectrum Of Ethanolic Leaf Extract Of Alternanthera Sessilis (L.)

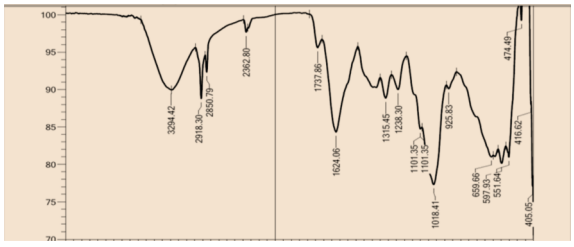

Figure -2
The FT-IR spectrum of ethanolic leaf extract of T*rigonella foenum-graecum* (L.)

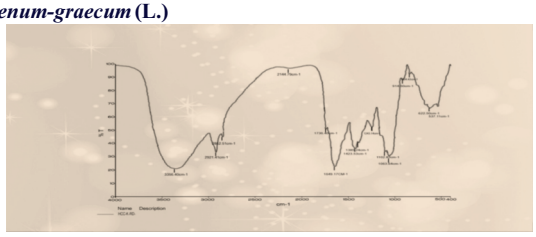

Table- 1
The FT-IR fingerprint of ethanolic extract of A*lternanthera sessilis* (L.) and T*rigonella foenum-graecum* (L.)

S. No	Frequency ranges(cm^{-1})	Functional groups	Frequency ranges(cm^{-1}) in *Alternanthera sessilis* (L.)	Frequency ranges(cm^{-1}) in *Trigonella foenum-graecum* (L.)
1.	690-515	C-Br stretch alkyl halides	659.66, 597.93	537.11
2.	700-610	–C(triple bond)C–H: C–H bend alkynes	-	622.50
4.	1250–1020	C–N stretch aliphatic amines	1018.41	1063.64
5.	1250–1020	C–N stretch aliphatic amines	1101.35	1102.83
6.	1335–1250	C–N stretch aromatic amines	1238.30	1243.14
7.	1400-1300	N=O Bond nitro	1315.45	1385.24
8.	1500-1400	C-C stretch(in ring) aromatics	-	1423.53
9.	1650-1580	N-H bond primary amines	1624.06	1649.17
10.	1850-1700	C=O carbonyl group	-	1736.40

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