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FUNCTIONAL GROUP ANALYSIS OF SOME ETHNO-MEDICINAL PLANTS OF THE *KANIKKAR* TRIBE BY THE FOURIER TRANSFORM INFRARED (FTIR) SPECTROSCOPY

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ABSTRACT

The present study focuses on the phytochemical/functional group analysis by the Fourier Transform Infrared (FTIR) Spectroscopy in the fine powder of whole plant of *Ampelocissus indica*, *Aristolochia tagala*, *Diploclisia glaucescens*, *Leucas biflora*, *Piper mullesua*, *Polygala arvensis*, *Rivea hypocrateriformis*, *Solanum diphyllum*, *Thespesia lampas* and *Thottea siliquosa* used as medicine by the *Kanikkars* in various villages of Pechiparai panchayat (forest range) in Kalkulam taluk of Kanyakumari district in Tamil Nadu state and mainland India. The functional group analysis by the FTIR Spectroscopy confirms the presence of characteristic functional groups such as alkanes, aromatic compounds, carboxylic acids, ethers, halogen derivatives, ketones, nitro compounds, phenols and sulphur compounds. The results confirm the fact that the therapeutic benefits of these plants could be attributed to the presence of aforesaid functional groups.

KEYWORDS: Ethnobotany, Ethno-medicinal Plants, Traditional Plants, Traditional Medicine, Ethno-medicine, *Kanikkars*, Petchiparai, Kanyakumari, Fourier Transform Infrared (FTIR) Spectroscopy.

INTRODUCTION

There has been man's unending desire for good and healthy living since the time immemorial which has led to his curiosity to examine all aspects of his environment by trial and error (Dalziel, 1961). This gave rise to the traditional medicine practice which was the only way of saving life in the olden days before the advent of modern medicine as the earliest humans used various plants to treat illness (Ajiwe *et al*., 2008). Tribal medicine or traditional medicine plays a vital role in the primary healthcare of tribal as well as rural people (Rajiv, 1998; Patil, 2008). Traditional medicine which is widespread throughout the world has been recognized by World Health Organization (WHO) as an essential building block of primary healthcare (Bannerman, 1982). World Health Organization has stated that 80% of the world's population depends on traditional medicine for its primary healthcare and has become indispensable for its survival (Hiremath and Taranath, 2013). Today, these traditional medicines are increasingly popular as cost effective alternative to, or complementary to, orthodox medicine (Tang and Haliwel, 2010). Nowadays, traditional medicinal practices form an integral part of complementary or alternative medicine. Although their efficacy and mechanisms of action have not been tested scientifically in most cases, these simple medicinal preparations often mediate beneficial responses due to their active chemical constituents (Park and Pezzutto, 2002).

Traditional medicine is widely used and accounts for about 40% of all healthcare delivered (WHO, 2002-2005). About 85% of traditional medicines are plant derived (Fransworth, 1988), and about 80% of population in developing countries still use plant based traditional medicine for their healthcare (Silva, 1997). This reliance is much frequent in case of tribal communities as their livelihoods are mainly dependent on forests that have also built up their socioeconomic and cultural life (Shroff, 1997; Mukul *et al*., 2007). The plant based traditional knowledge has become a recognized tool in search for new sources of drugs (Bruce and Meeus, 2002). Traditional knowledge of medicinal plants has always guided the search for new cures. In spite of the advent of modern high throughput drug discovery and screening techniques, traditional knowledge systems have given clues to the discovery of valuable drugs (Buenz *et al*., 2004).

Tribals have deep belief in their native folklore medicine for remedies and they rely exclusively on their own herbal cure (Sajem and Gosai, 2006). Many tribal groups have been using several plant or animal products for medicinal preparations and these medicines are known as ethnomedicines (Pushpangadan and Atal, 1984). Ethnomedicine may be defined broadly as the use of plants by humans as medicines, but these uses could be called, more accurately, as ethnobotanic medicine (Fransworth, 1994). Ethnomedicine is one of the systems of medicine that is widely practiced among the tribal and aboriginal populations of our country for the treatment of ailments (Singh *et al*., 2013). Most ethnobotanical studies have been restricted to studies of tribal and rural people to record their knowledge and use of plants and to search for new sources of herbal drugs, edible plants and other properties of plants of value to man (Jain and Mudgal, 1999). Ethnomedical information/data are playing an important role for developing new scientifically validated and standardized drugs, i.e. both herbal and modern (Savnur, 1993).

Phytochemical/Functional Group Analysis by the FTIR Spectroscopy

The Fourier Transform Infrared (FTIR) spectroscopy allows the analysis of a relevant amount of compositional and structural information in plants Moreover, FTIR spectroscopy is an established timesaving method to characterize and identify functional groups (Grube *et al.*, 2008). FTIR is a rapid, noninvasive, high-resolution analytical tool for identifying types of chemical bonds in a molecule by producing an infrared absorption spectrum that is like a molecular fingerprint (Griffiths and de Haseth, 1986). It is more sensitive and selective than colorimetric methods and has played a vital role in pharmaceutical analysis in recent years (Ellis *et al.*, 2002). It measures predominantly the vibrations of bonds within chemical functional groups and generates a spectrum that can be regarded as a biochemical or metabolic "fingerprint" of the sample. The FTIR spectrum is used to identify the functional groups of the active components based on the peak value in the region of infrared radiation (Packialakshmi and Naziya, 2014). At present, particularly in phytochemistry, FTIR has been exercised to identify concrete structures of certain plant secondary metabolites and offers a rapid and non-destructive investigation to fingerprint herbal extracts or powders (Yang and Yen, 2002; Ivanova and Singh, 2003). It is one of the most widely used methods to identify the chemical constituents and elucidate the compound structure and has been used as a mandatory method to identify medicines in Pharmacopoeia of many countries (Liu *et al.*, 2006).

The survey of literature revealed that the FTIR spectroscopic analysis was carried out by several researchers (Muruganantham *et al*., 2009; Kumar *et al*., 2010; Ragavendran *et al*., 2011; Anand and Gokulakrishnan, 2012; Janakiraman *et al*., 2012; Mariswamy *et al*., 2012; Raj *et al*., 2012; Sathish *et al*., 2012; Starlin *et al*., 2012; Vijayalakshmi and Ravindhran, 2012; Jothy *et al*., 2013; Maobe and Nyarango, 2013; Sahu and Saxena, 2013; Sandosh *et al*., 2013; Sathish *et al*., 2013; Abayomi *et al*., 2014; Asha *et al*., 2014; Ashokkumar and Ramaswamy, 2014; Kumar *et al*., 2014; Onyema and Ajiwe, 2014; Onyema *et al*., 2014; Pillai *et al*., 2014) to identify the various characteristic functional groups of the phytoactive compounds present in various solvent extracts/fine powder of whole plant or its different parts used as medicine.

MATERIALS AND METHODS

Study Area

Once known as Cape Comorin, Kanyakumari district is the southernmost district in Tamil Nadu state and mainland India. It is situated in between 77° 15' and 77° 36' east and 8° 03' and 8° 35' to the north longitudes (Map of the Study Area). It has a full-ranging surface features with ocean on three sides and the land mass that projects well above its surroundings of the Western Ghats abutting the northern face. It has its borders with Tirunelveli district, the Gulf of Mannar, the Indian Ocean, the Arabian Sea and the state of Kerala. Of the total district area of 1671.3 km^2, government forests occupy an area of 504.86 km^2 which comes to about 30.2 percent of the geographical area of the district. There are 14 types of forests from luxuriant tropical wet evergreen to tropical thorn forests. Rainfall varies from 103 cm to 310 cm elevation from sea level to 1829 m. An ethno-medicinal survey was conducted in various villages of Pechiparai panchayat (forest range) in Kalkulam taluk of Kanyakumari district.

Kanikkar

The *Kanikkar* is one of the important tribes living in several districts of Kerala and Tamil Nadu. The *Kanikkars* dwell in forests or near to forests in Thiruvananthapuram and Kollam in Kerala, and Kanyakumari and Tirunelveli in Tamil Nadu. The *Kanikkars* are also known as *Kanis*. Like the other aboriginal hunting and gathering tribes, *Kanis* also have the primitive history of hunting, gathering and shifting cultivation. Long back, the *Kanikkars* were employed by the Travancore Government to collect honey, wax, ginger, cardamom, dammar and elephant tusks (Thurston, 1909). The language they speak closely resembles Malayalam (Palanichamy, *et al*., 2010) with few Tamil words (Arun *et al*., 2007). *Kani* tribal medicinal experts are called as ‘‘*Philathies*” (Prasad *et al*., 1996).

During the field survey in the villages of Pechiparai panchayat (forest area) in Kalkulam taluk of Kanyakumari district (the study area), the *Kanikkar* informants were interviewed in their local languages Tamil and Malayalam to collect information about the traditional medicinal practices practiced by them for the treatment of various human ailments. The FTIR spectroscopic analysis was carried out to identify the various characteristic functional groups of the phytoactive compounds present in the fine powder of some whole plants used as ethnomedicine by the *Kanikkars.*

Phytochemical/Functional Group Analysis by the FTIR Spectroscopy

A little powder of plant sample was mixed with KBr salt and compressed into a thin pellet using a mortar and pestle. Infrared spectra were recorded as KBr pellets on a Thermoscientific Nicot iS5 iD1 transmission between 4000-400 cm^{-1}. Infrared absorptions were recorded in tables (Kareru *et al.*,2008).

RESULTS AND DISCUSSION

The *Kanikkar* informants in the study area disclosed ethno-medicinal data of 10 traditional plants distributed across 8 families as having medicinal properties against 10 human ailments including poisonous bites (Table 1).

The data on the characteristic peak values and probable functional groups present in the fine powder of whole plant of *Ampelocissus indica*, *Aristolochia tagala*, *Diploclisia glaucescens*, *Leucas biflora*, *Piper mullesua*, *Polygala arvensis*, *Rivea hypocrateriformis*, *Solanum diphyllum*, *Thespesia lampas* and *Thottea siliquosa* obtained by the FTIR spectroscopy are figured in Tables 2-11. The FTIR spectra profiles are illustrated in Figures 1-10.

The FTIR spectrum of the whole plant of *Ampelocissus indica* shows 11 major peaks at the range of 3447.13 to 535.70 cm^{-1}. The observed peaks correspond to the presence of various functional groups like phenols, nitro compounds (nitramine and lactams), ketones, ethers, aromatic compounds and halogen derivatives (chloro-compounds, bromo-compounds and iodo-compounds).

The FTIR spectrum of the whole plant of *Aristolochia tagala* shows 13 major peaks at the range of 3421.60 to 531.66 cm^{-1}. The obtained peaks confirm the presence of various functional groups such as phenols, alkanes, nitro compounds (nitrite), ketones, ethers, aromatic compounds and halogen derivatives (chloro-compounds and bromo-compounds).

The FTIR spectrum of the whole plant of *Diploclisia glaucescens* shows 9 major peaks at the range of 3734.41 to 608.42 cm^{-1}. The attained peaks justify the presence of various functional groups like phenols, carboxylic acids, nitro compounds (oxime), ketones, ethers, aromatic compounds and halogen derivatives (iodo-compounds).

The FTIR spectrum of the whole plant of *Leucas biflora* shows 15 major peaks at the range of 3431.16 to 471.74 cm^{-1}. The noted peaks prove the presence of various functional groups such as phenols, alkanes, carboxylic acids, nitro compounds (nitrite and lactams), ketones, ethers, aromatic compounds and halogen derivatives (chloro-compounds, iodo-compounds and bromo-compounds).

The FTIR spectrum of the whole plant of *Piper mullesua* shows 16 major peaks at the range of 3424.03 to 472.09 cm^{-1}. The recorded peaks validate the presence of various functional groups like phenols, alkanes, nitro compounds (nitrite and lactams), ketones, ethers, sulphur compounds, aromatic compounds and halogen derivatives (iodo-compounds and bromo-compounds).

The FTIR spectrum of whole plant of *Polygala arvensis* shows 10 major peaks at the range of 3421.78 to 468.93 cm^{-1}. The observed peaks correspond to the presence of various functional groups like phenols, alkanes, carboxylic acids, nitro compounds (oxime and nitramine), ethers, aromatic compounds and halogen derivatives (iodo-compounds).

The FTIR spectrum of the whole plant of *Rivea hypocrateriformis* shows 13 major peaks at the range of 3420.82 to 527.64 cm^{-1}. The obtained peaks confirm the presence of various functional groups such as phenols, alkanes, nitro compounds (oxime and lactams), ethers, aromatic compounds and halogen derivatives (chloro-compounds and bromo-compounds).

The FTIR spectrum of the whole plant of *Solanum diphyllum* shows 15 major peaks at the range of 3435.31 to 472.37 cm^{-1}. The attained peaks justify the presence of various functional groups such as phenols, alkanes, carboxylic acids, nitro compounds (nitrite and nitramine), ethers and halogen derivatives (chloro-compounds and bromo-compounds).

The FTIR spectrum of the whole plant of *Thespesia lampas* shows 13 major peaks at the range of 3455.92 to 607.31 cm^{-1}. The noted peaks prove the presence of various functional groups such as phenols, carboxylic acids, nitro compounds (oxime, nitramine, nitrosamine and lactams), ethers, aromatic compounds and halogen derivatives (chloro-compounds and iodo-compounds).

The FTIR spectrum of the whole plant of *Thottea siliquosa* shows 15 major peaks at the range of 3422.70 to 469.90 cm^{-1}. The recorded peaks validate the presence of various functional groups like phenols, alkanes, carboxylic acids, nitro compounds (oxime and lactams), ketones, ethers and halogen derivatives (chloro-compounds and bromo-compounds).

The functional group analysis by the FTIR Spectroscopy confirms the presence of characteristic functional groups such as alkanes, aromatic compounds, carboxylic acids, ethers, halogen derivatives, ketones, nitro compounds, phenols and sulphur compounds in the fine powder of whole plant of *Ampelocissus indica*, *Aristolochia tagala*, *Diploclisia glaucescens*, *Leucas biflora*, *Piper mullesua*, *Polygala arvensis*, *Rivea hypocrateriformis*, *Solanum diphyllum*, *Thespesia lampas* and *Thottea siliquosa*. The therapeutic benefits of these plants could be attributed to the presence of aforesaid functional groups. HPLC/MS and LC-MS studies are needed to identify the unknown functional groups.

The FTIR spectroscopic analysis was carried out by several researchers to pinpoint the various characteristic functional groups of the phytoactive compounds present in various solvent extracts/fine powder of whole plant or its different parts used as medicine. Carboxylic acids are often present as a functional group of effective medicines. They are involved in specific charge-charge interactions and are thus often critical for the binding of agents to their targets. The carboxylic acid functional group adds to the hydrophilicity of the drug as well as to its polarity and this may impede the bioavailability (Maag, 2007). In the present study, carboxylic acids as a functional group is present in *Diploclisia glaucescens*, *Leucas biflora*, *Polygala arvensis*, *Solanum diphyllum*, *Thespesia lampas* and *Thottea siliquosa*.

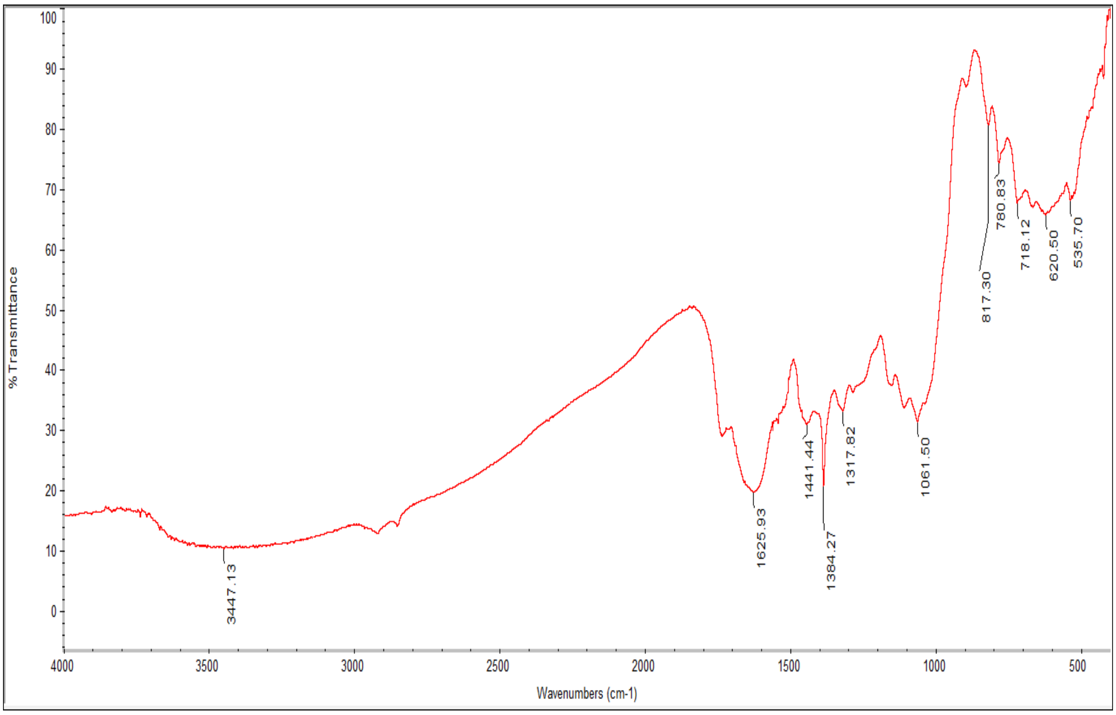

Figure 1: FTIR Spectrum Profile of *Ampelocissus indica.*

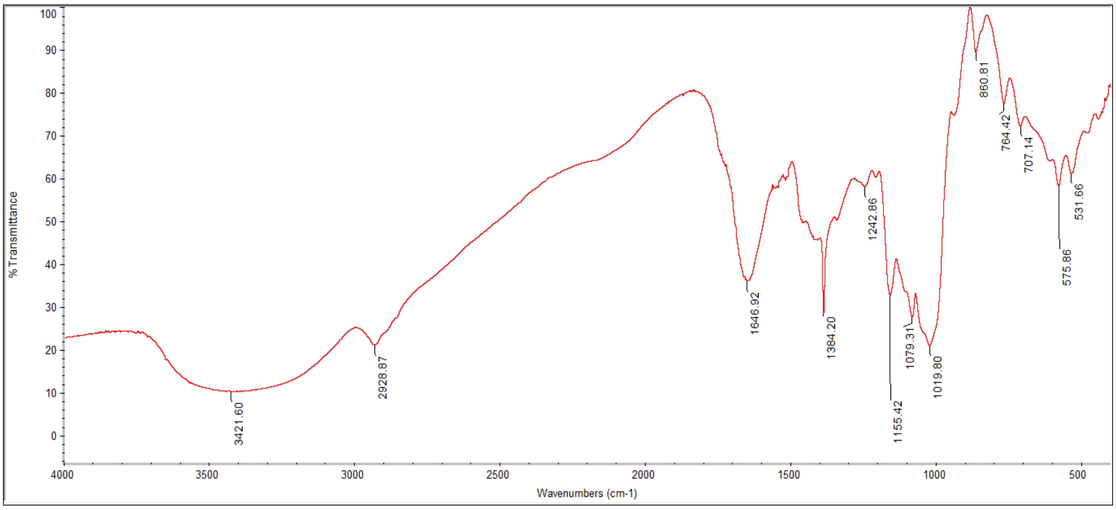

Figure 2: FTIR Spectrum Profile of *Aristolochia tagala.*

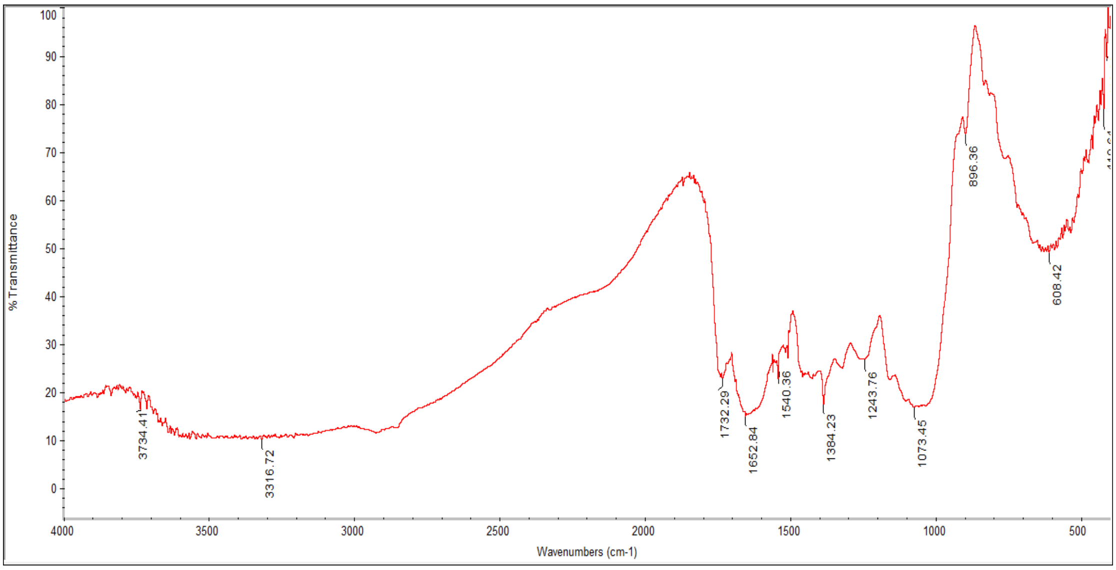

Figure 3: FTIR Spectrum Profile of *Diploclisia glaucescens.*

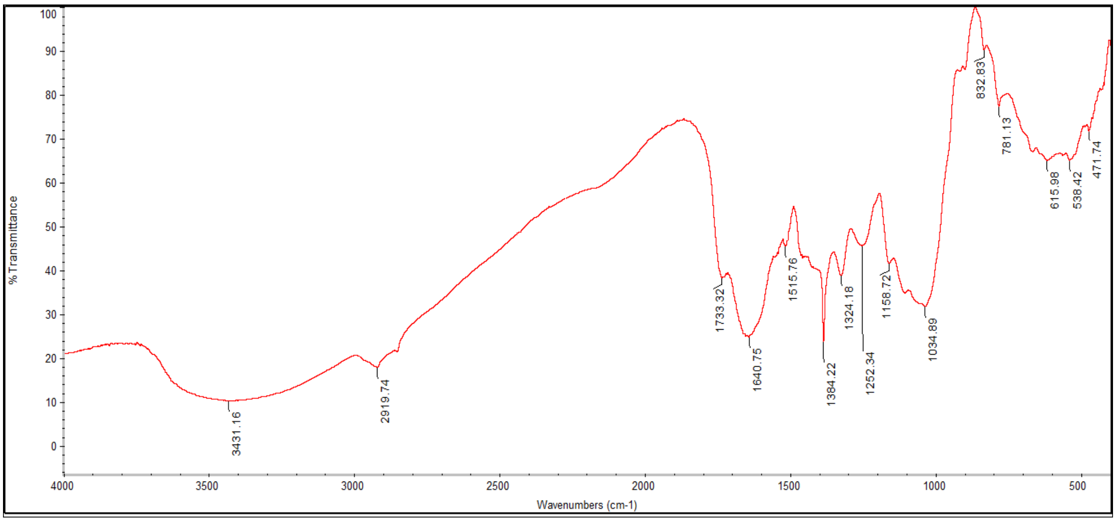

Figure 4: FTIR Spectrum Profile of *Leucas biflora.*

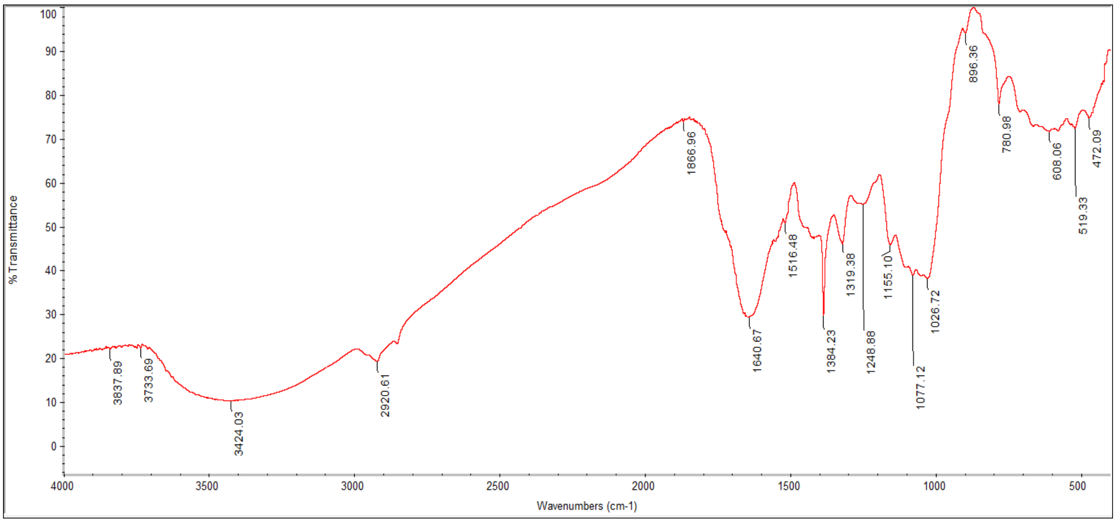

Figure 5: FTIR Spectrum Profile of *Piper mullesua.*

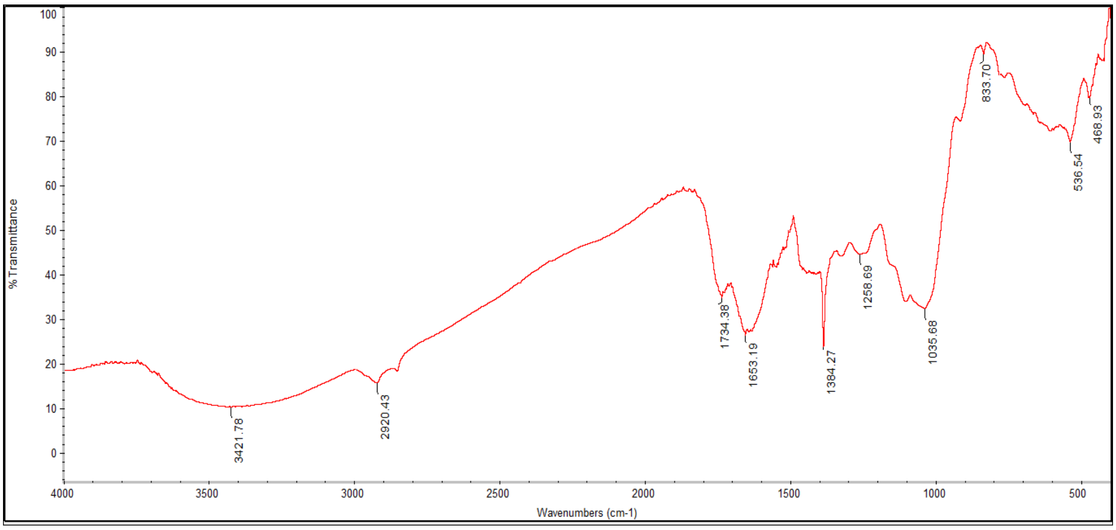

Figure 6: FTIR Spectrum Profile of *Polygala arvensis.*

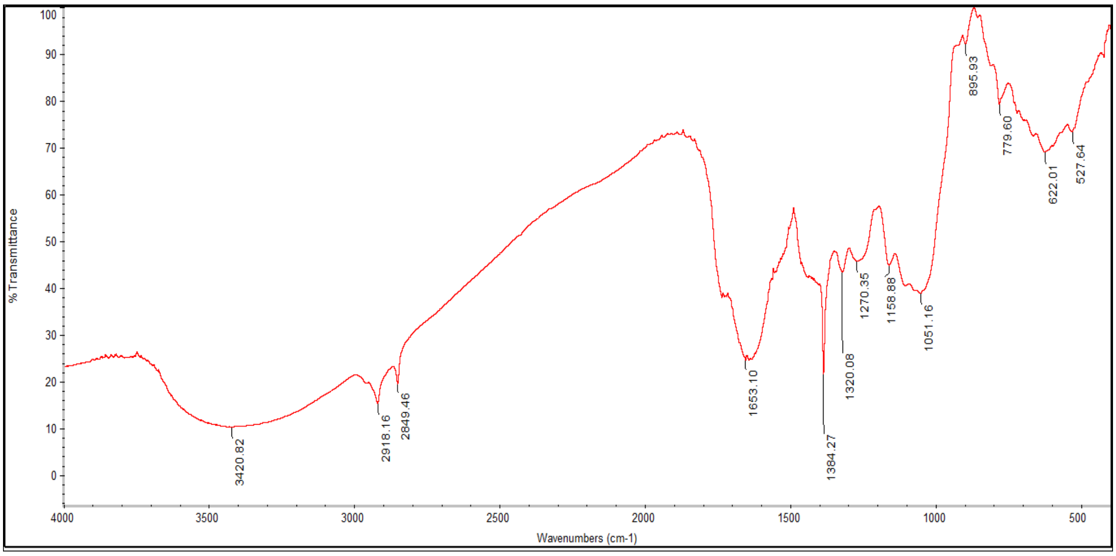

Figure 7: FTIR Spectrum Profile of *Rivea hypocrateriformis.*

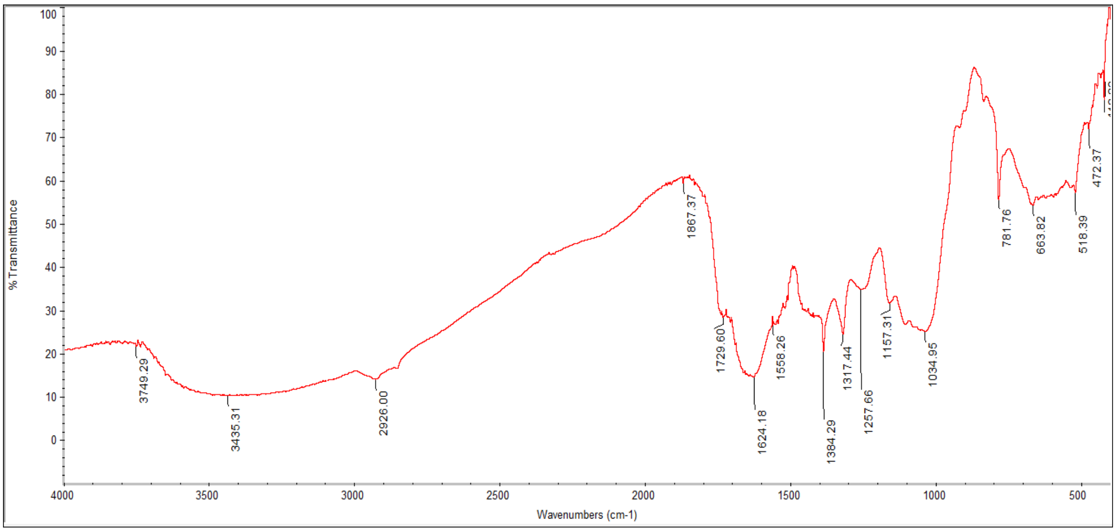

Figure 8: FTIR Spectrum Profile of *Solanum diphyllum*.

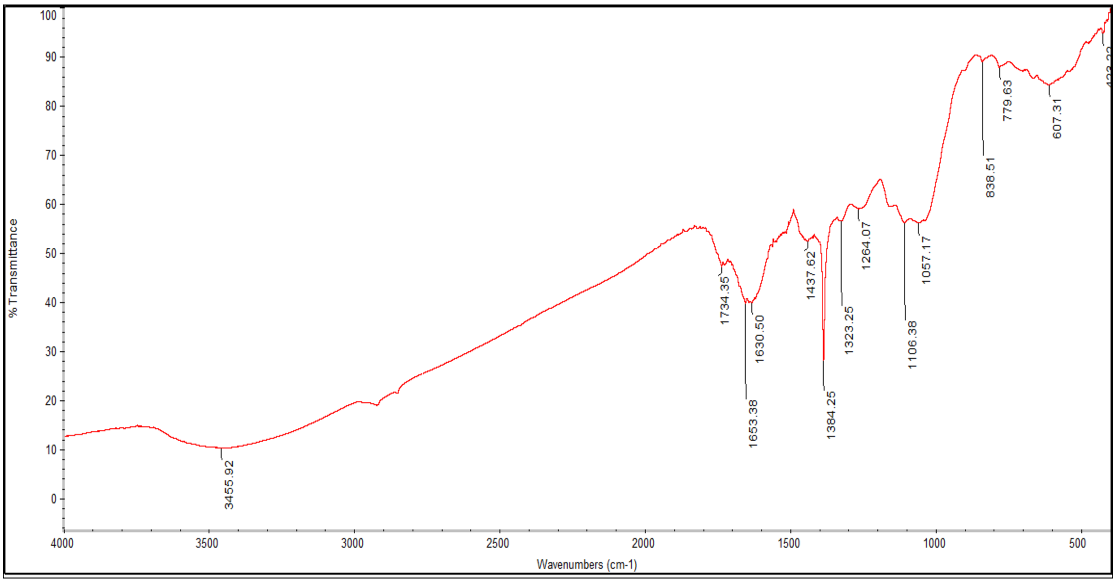

Figure 9: FTIR Spectrum Profile of *Thespesia lampas*.

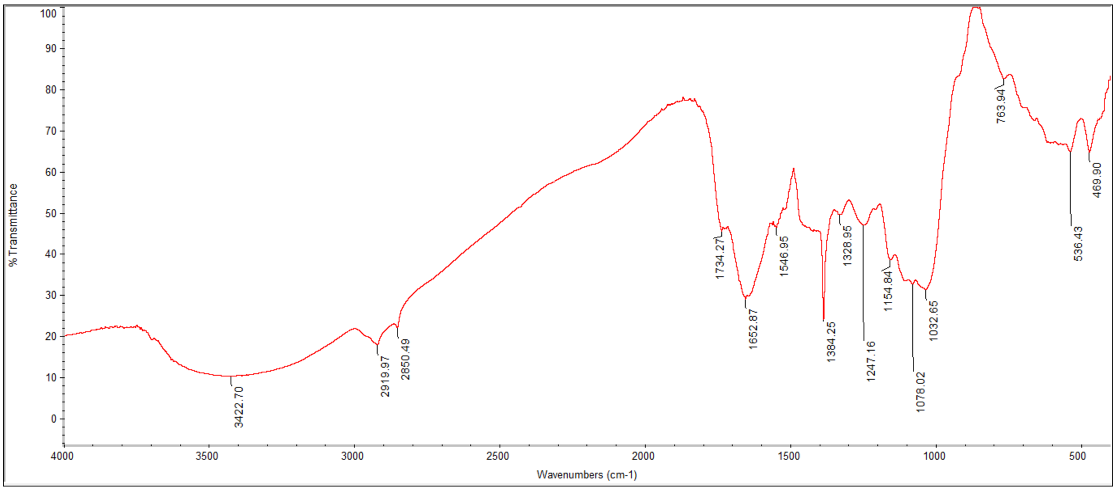

Figure 10: FTIR Spectrum Profile of *Thottea siliquosa*.

Map of the Study Area

Kanyakumari District in Tamil Nadu

Villages in Pechiparai Panchayat in Kalkulam Taluk of Kanyakumari District

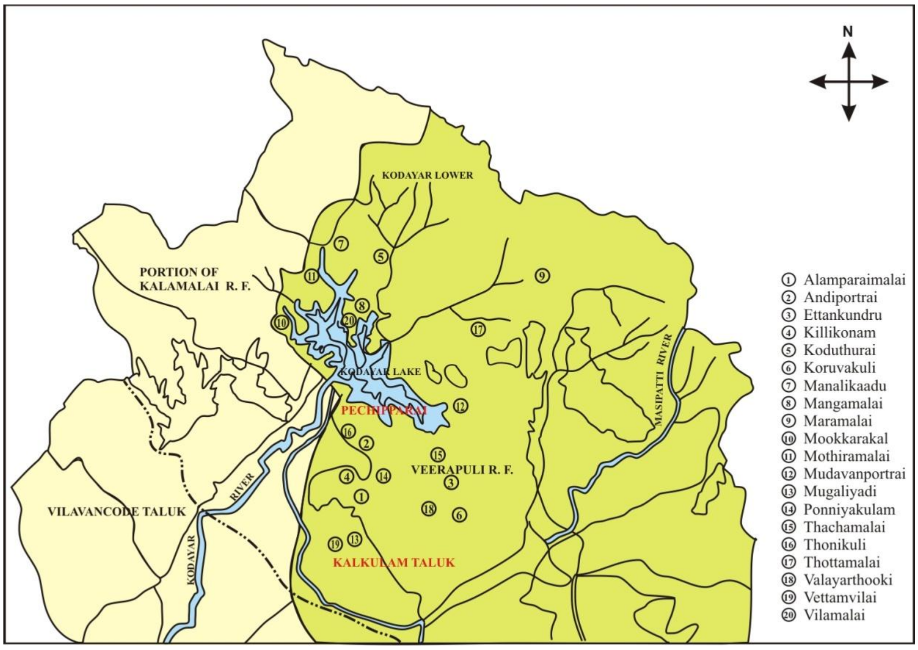

Table 1: List of Ethno-medicinal Plants Used by the *Kanikkars* in the Study Area.

S/N	Botanical Name	Family	Part Used	Ailments Treated	Ailment Categories
1	*Ampelocissus indica* (L.) Planch.	Vitaceae	Root	Jaundice	Liver Problems
2	*Aristolochia tagala* Cham.	Aristolochiaceae	Root	Snake Bite	Others
3	*Diploclisia glaucescens* (Blume) Diels	Menispermaceae	Leaf	Scabies	Dermatological Problems
4	*Leucas biflora* (Vahl) R. Br. ex Sm.	Lamiaceae	Leaf	Skin Infections	Dermatological Problems
5	*Piper mullesua* Buch.- Ham. ex D. Don	Piperaceae	Climbing branch	Cold, Cough and Flu	Respiratory Problems and Fever
6	*Polygala arvensis* Willd.	Polygalaceae	Leaf	Snake Bite	Others
7	*Rivea hypocrateriformis* (Desr.) Choisy	Convolvulaceae	Leaf	Hematuria	Urogenital Problems
8	*Solanum diphyllum* L.	Solanaceae	Seed	Intestinal Worm Infection	Gastro Intestinal Problems
9	*Thespesia lampas* (Cav.) Dalzell & A. Gibson	Malvaceae	Leaf	Dermatophytosis	Dermatological Problems
10	*Thottea siliquosa* (Lam.) Ding Hou	Aristolochiaceae	Root	Centipede Bite and Snake Bite	Others

Table 2: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Ampelocissus indica.*

S/N	Peak Values cm^{-1}	Functional Groups
1	3447.13	Phenols
2	1625.93	Nitro compounds (nitramine)
3	1441.44	Nitro compounds (lactams)
4	1384.27	Nitro compounds
5	1317.82	Ketones
6	1061.50	Ethers
7	817.30	Aromatic compounds
8	780.83	Halogen derivatives (chloro-compound)
9	718.12	Halogen derivatives (bromo-compound)
10	620.50	Halogen derivatives (iodo-compound)
11	535.70	Halogen derivatives (iodo-compound)

Table 3: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Aristolochia tagala*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3421.60	Phenols
2	2928.87	Alkanes
3	1646.92	Nitro compounds (nitrite)
4	1384.20	Nitro compounds
5	1242.86	Ketones
6	1155.42	Ethers
7	1079.31	Ethers
8	1019.80	Ketones
9	860.81	Aromatic compounds
10	764.42	Halogen derivatives (chloro-compound)
11	707.14	Halogen derivatives (bromo-compound)
12	575.86	Halogen derivatives (bromo-compound)
13	531.66	Halogen derivatives (bromo-compound)

Table 4: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Diploclisia glaucescens*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3734.41	Phenols
2	1732.29	Carboxylic acids
3	1652.84	Nitro compounds (oxime)
4	1540.36	Nitro compounds
5	1384.23	Nitro compounds
6	1243.76	Ketones
7	1073.45	Ethers
8	896.36	Aromatic compounds
9	608.42	Halogen derivatives (iodo-compound)

Table 5: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Leucas biflora*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3431.16	Phenols
2	2914.74	Alkanes
3	1733.32	Carboxylic acids
4	1640.75	Nitro compounds (nitrite)
5	1515.76	Nitro compounds
6	1384.22	Nitro compounds
7	1324.18	Nitro compounds (lactams)
8	1252.34	Ketones
9	1158.72	Ethers
10	1034.89	Ethers
11	832.83	Aromatic compounds
12	781.13	Halogen derivatives (chloro-compound)
13	615.98	Halogen derivatives (iodo-compound)
14	538.42	Halogen derivatives (bromo-compound)
15	471.74	Unknown

Table 6: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Piper mollesua.*

S/N	Peak Values cm^{-1}	Functional Groups
1	3424.03	Phenols
2	2920.61	Alkanes
3	1866.96	Unknown
4	1640.67	Nitro compounds (nitrite)
5	1516.48	Nitro compounds
6	1384.23	Nitro compounds
7	1319.38	Nitro compounds (lactams)
8	1248.88	Ketones
9	1155.10	Ethers
10	1077.12	Ethers
11	1026.72	Sulphur compounds
12	896.36	Aromatic compounds
13	780.98	Aromatic compounds
14	608.06	Halogen derivatives (iodo-compound)
15	519.33	Halogen derivatives (bromo-compound)
16	472.09	Unknown

Table 7: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Polygala arvensis.*

S/N	Peak Values cm^{-1}	Functional Groups
1	3421.78	Phenols
2	2920.43	Alkanes
3	1734.38	Carboxylic acids
4	1653.19	Nitro compounds (oxime)
5	1384.27	Nitro compounds
6	1258.69	Nitro compounds (nitramine)
7	1035.68	Ethers
8	833.70	Aromatic compounds
9	536.54	Halogen derivatives (iodo-compound)
10	468.93	Unknown

Table 8: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Rivea hypocrateriformis.*

S/N	Peak Values cm^{-1}	Functional Groups
1	3420.82	Phenols
2	2918.16	Alkanes
3	2849.46	Alkanes
4	1653.10	Nitro compounds (oxime)
5	1384.27	Nitro compounds
6	1320.08	Nitro compounds (lactams)
7	1270.35	Ethers
8	1158.88	Ethers
9	1051.16	Ethers
10	895.93	Aromatic compounds
11	779.60	Halogen derivatives (chloro-compound)
12	622.01	Halogen derivatives (bromo-compound)
13	527.64	Halogen derivatives (bromo-compound)

Table 9: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Solanum diphyllum*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3435.31	Phenols
2	2926.00	Alkanes
3	1867.37	Unknown
4	1729.60	Carboxylic acids
5	1624.18	Nitro compounds (nitrite)
6	1558.26	Nitro compounds
7	1384.29	Nitro compounds
8	1317.44	Nitro compounds
9	1257.66	Nitro compounds (nitramine)
10	1157.31	Ethers
11	1034.95	Ethers
12	781.76	Halogen derivatives (chloro-compound)
13	663.82	Halogen derivatives (bromo-compound)
14	518.39	Halogen derivatives (bromo-compound)
15	472.37	Unknown

Table 10: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Thespesia lampas*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3455.92	Phenols
2	1734.35	Carboxylic acids
3	1653.38	Nitro compounds (oxime)
4	1630.50	Nitro compounds (nitramine)
5	1437.62	Nitro compounds (nitrasamine)
6	1384.25	Nitro compounds
7	1323.25	Nitro compounds (lactams)
8	1264.07	Nitro compounds (nitramine)
9	1106.38	Ethers
10	1057.17	Ethers
11	838.51	Aromatic compounds
12	779.63	Halogen derivatives (chloro-compound)
13	607.31	Halogen derivatives (iodo-compound)

Table 11: FTIR Peak Values and Functional Groups in the Powder of Whole Plant of *Thottia siliquosa*.

S/N	Peak Values cm^{-1}	Functional Groups
1	3422.70	Phenols
2	2919.97	Alkanes
3	2850.49	Alkanes
4	1734.27	Carboxylic acids
5	1652.87	Nitro compounds (oxime)
6	1546.95	Nitro compounds
7	1384.25	Nitro compounds
8	1328.95	Nitro compounds (lactams)
9	1247.16	Ketones
10	1154.84	Ethers
11	1078.02	Ethers

12	1032.65	Ethers
13	763.94	Halogen derivatives (chloro-compound)
14	536.43	Halogen derivatives (bromo-compound)
15	469.90	Unknown

CONCLUSION

The presence of various functional groups in these plants justifies the use of them as the traditional medicine by the *Kanikkars* in the study area. Further studies are needed with these plants to identify the unknown functional groups, isolate, characterize and elucidate the structure of the phytoactive compounds which are responsible for other therapeutic benefits.

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