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Research Article

Phytochemical Screening, GCMS and FTIR Profile of Bioactive Compounds in *Solanum lycopersicum* Wild Fruits collected from Palani Hill Ranges of the Western Ghats

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Abstract

Tomatoes are consumed worldwide as fresh vegetables because of their high contents of essential nutrients and antioxidant-rich phytochemicals. Tomatoes contain minerals, vitamins, proteins, essential amino acids (Leucine, Threonine, Valine, Histidine, Lysine, Arginine), monounsaturated fatty acids (Linoleic and Linolenic Acids), Carotenoids (Lycopene and β-Carotenoids) and Phytosterols (β-Sitosterol, Campesterol and Stigmasterol). GCMS analysis revealed the presence of 3-methylheptane, Ethylcyclohexane, 2-Methyl-4,6-octadiyn-3-one, 5,6-Dimethylundecane, (3E)3-Hexen-2-one, 2,2-Dimethylbutane, 1,2-Diphenyl-1-butanone, Isopropylbenzene (2-phenylpropane), 3,5-Dimethyloctane, 2-Phenyl-3-buten-1-ol, 2,4,4-Trimethylhexane, Benzoylcarboxaldehyde (Phenylglyoxal), Cis-3-Methyl-Endo-Tricyclo [5.2.1.0(2.6)] Decane, 2,4-Dimethylhexan-3-One, Benzene acetic acid,2-phenylethyl ester, Cyclopentacycloheptene, 2,3-Heptanedione, 1,6-Methano[10] annulene, 1-Naphthaleneacetic acid, methyl ester, N,N-Dimethylmethanesulfonamide, Methyl tridecanoate, Cis 9-Octadecanoic acid, Methyl 15-methylheptadecanoate, 9-Octadecenoic acid, Methyl (Z)-octadec-9-enoate, (Z)-octadec-9-enamide, Methyl 2-ethyl-2-methylicosanoate, 1,2,3,4,4a,5,6,7,8,9,10,11,12,12a-tetradecahydrobenzo [10] annulene Caffeic acid, Catechin, Chlorogenic acid, Chrysin, Cinnamic acid, Epicatechin, Ferulic acid, Kaempferol, Luteolin, Lycopene, Naringenin, P-coumaric acid, Phloretic acid, Quercetin, Resveratrol, Rutin, Sinapic acid, Vanillic acid. Lycopene, the main dietary carotenoid in tomato and tomato-based food products and lycopene consumption by humans has been reported to protect against Cancer, Cardiovascular Diseases, Cognitive function and Osteoporosis. Among phenolic compounds present in tomato, Quercetin, Kaempferol, Naringenin, Caffeic Acid and Lutein are the most common. These compounds have significant antioxidant properties and are effective in protecting human body against oxidative stress-related diseases.

Keywords: *Solanum lycopersicum*; Phytochemical Screening; GCMS; FTIR

INTRODUCTION

Tomatoes (*Solanum lycopersicum* L.; Family: Solanaceae), are frequently included in Mediterranean diet and are widely consumed as vegetables, play an important role in nutrition attributed to well-established health benefits[1]. Tomatoes are commonly used in processed food products[2]. Nutrients in tomatoes include vitamins, minerals, fiber, protein, amino acids, fatty acids, carotenoids and phytosterols[3-8]. These nutrients drive physiological, metabolic and biochemical processes associated with constipation high blood pressure, blood circulation, lipid profile, body fluids, detoxification and bone structure[9]. Tomatoes are excellent source of bioactive compounds involved in prevention of human chronic diseases - cardiovascular disease (CVD), cancer, and neurodegenerative diseases[10-11].

High concentrations of natural antioxidant, such as carotenoids (β-carotenoids and lycopene), ascorbic acid (vitamin C), tocopherol (vitamin E) and bioactive phenolic compounds (quercetin, kaempferol, naringenin and lutein,

caffeic, ferulic and chlorogenic acids), tomatoes ameliorate chronic several diseases[12,14]. Antioxidants inhibit reactive oxygen species (ROS) by scavenging free radicals, inhibiting cellular proliferation and damage, inhibit apoptosis, metal chelation, modulate enzymatic activity, cytokine expression and signal transduction[12-15]. Pharmacological activities of lycopene and other phenolic compounds include anticancer, anti-inflammatory, antidiabetic, anti-allergenic, anti-atherogenic, antithrombotic, antimicrobial, antioxidant, vasodilator and cardioprotection[12-18]. Polyphenolic compounds and carotenoids trigger sensory activities including maintaining good aroma, taste[19]. Tomato is a dietary source of soluble and insoluble dietary fibers - cellulose, hemicelluloses and pectin[20] reported to ameliorate bowel disorders, cancer, diabetes, CVDs, and obesity[21]. Proximate composition of tomatoes includes pH, energy, acidity and reducing sugars[22]. Combination of all - vitamins, minerals, amino acids, fatty acids makes tomato a balanced diet[23-25].

Nutritional composition of tomato is based on cultivar (genetic), soil conditions, extraction procedures, analytic methods and environmental/ climatic conditions. During processing of tomato products, up to 30% of original weight are turned into waste, which may be nutritive [26]. Peel and seeds are the main waste product of tomato, both are rich in protein, dietary fibers, bioactive compounds and lycopene[27]. By-products of SL are used as additives in meat processing industries. Further, by-products of SL contain multiple bioactive compounds that could serve as a renewable source for obtaining natural antioxidants and colorants (carotenoids)[28]. Although by products of SL is a rich source of nutrients, comprehensive research should be carried out before marketed for human consumption[29]. In spite of significant health benefits, tomatoes demonstrate few undesired effects when consumed in large amounts. Adverse effects of tomato excess-intake are associated with renal problems, allergies, arthritis, heartburn, and migraine[30]. With this background information the aim of this paper is to screen for bioactive secondary metabolites followed by GCMS and FTIR analysis of bioactive compounds in *Solanum lycopersicum* wild fruits.

MATERIALS AND METHODS

Phytochemical Screening

Fruits were collected from the wild in the Palani Hills region of the Western Ghats, TamilNadu, India. Type specimen was identified and authenticated by Prof. Dr. S. Sutha at Department of Medicinal Botany, Government Siddha Medical College, Palayamkottai, Tirunelveli District, Tamil Nadu, India. The methanolic extracts were subjected to chemical tests for the detection of different phytoconstituents using standard procedures[31-35].

Sample preparation

Using direct method of extraction, approximately 100 g of dried fruit powder was extracted with 500 ml of different solvents such as petroleum ether, chloroform and methanol to determine the profile of bioactive compounds in the test samples. The extract was transferred in to glass vials. The process was repeated 3 times with fresh solvent. The solvent was removed by Rotavapor® R-300 (BÜCHI-GmbH, Germany). The extracted residue was re-dissolved in the respective solvent to yield a final volume of 10mg/ml and the content was stored in cold (at 4°C) until further use. The extracts were subjected to chemical tests for the detection of phytoconstituents using standard procedures as described previously[31-40].

TEST FOR ALKALOIDS

Mayer's test: Few drops of Mayer's reagent was added to 1 mL of plant extract, appearance of a deep yellow or white precipitate indicated the presence of alkaloids in the solution. (Mayer's reagent was freshly prepared by dissolving mercuric chloride (1.36 g) and potassium iodide (5.00 g) in 100 ml water).

Dragendorff's test: To 2 mL of the extract added 1 mL of Dragendorff's reagent along the side of the test tube. Formation of orange or orange reddish brown precipitate indicated the presence of alkaloids. Dragendorff's reagent was prepared by Sol A: 0.85g bismuth subnitrate, 40mL water, and 10mL glacial acetic acid and Sol B: 8g potassium iodide and 20mL water. 5mL each of Sol A & B with 20mL of glacial acetic acid and 70-100 mL of water is mixed to prepare Dragendorff's reagent.

Hager's test: Hager's test was done by adding a few drops of Hager's reagent to plant extracts and appearance of a yellow-color precipitate indicated the presence of alkaloids in the solution. Hager's reagent is saturated solution of picric acid.

Wagner's test: Approximately, 1 ml of crude extract was mixed with 2 ml of Wagner's reagent. Reddish brown colour precipitate indicates the presence of alkaloids. Wagner's Reagent was prepared by mixing 2.5 gm iodine in 12.5 gm of potassium iodide (KI 2); add 250 ml of water to produce solution.

TEST FOR GLYCOSIDES

Test For Anthraquinones Glycosides

Borntragers test: 0.5 g of extract was boiled with 10% hydrochloric acid for few minutes in water bath. It was filtered and allowed to cool. Equal volume of CHCl3 was added to the filtrate. Few drops of 10% ammonia was added to the mixture and heated. Formation of rose – pink color indicates of n-hexane, chloroform, ethyl acetate and methanol of the presence of the anthroquinones.

Baljet test: Part of plant containing cardiac glycoside is dipped in sodium picrate solution; formation of a yellow to orange colour indicates the presence of aglycones or glycosides in the plant tissues.

Legal's Test: To the concentrated ethanolic extract few drops of 10% NaOH were added, to make it alkaline. Then freshly prepared sodium nitroprusside was added to the solution. Presence of blue coloration indicated the presence of glycosides in the extract.

TEST FOR CARDIAC GLYCOSIDES

Keller-Kiliani test: 5 ml of extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underlayed with 1 ml of concentrated sulphuric acid. A browning of the interface indicates a deoxy-sugar characteristic of carotenoids. A violet ring may appear below the brown ring, while in the acetic acid layer, a greenish ring may form just gradually throughout thin layer.

TEST FOR CARBOHYDRATES

Molisch's test: Small portion of the plant extract was put in a test tube; 10 ml of distilled water was added and shaken vigorously and gently. The mixture was then filters and divided into two portions. To the first portion, two drops of Molish's reagent was added followed by few drops of concentrated sulphuric acid by the wall of the test tube.

Formation of brown or purple ring at the interphase indicated the presence of carbohydrates.

Fehling's test Equal volume of Fehling A and Fehling B reagents were mixed together and then add 2ml of crude extract in it and gently boiled. A brick red precipitate appeared at the bottom of the test-tube indicates the presence of reducing sugars.

Benedict's test 1 ml of crude extract was mixed with 2ml of Benedict's reagent and boiled. A reddish brown precipitate was formed which indicates the presence of the carbohydrates.

TEST FOR PHYTOSTEROLS

Libermann Burchard's Test: Dissolve one or two crystals of cholesterol in dry chloroform in a dry test tube. Add few drops of acetic anhydride and then 2 drops of concentrated H2SO4 and mix well. The formation of a green or green-blue colour after a few minutes indicates the presence of phytosterols. After the reaction, concentration of cholesterol can be measured spectrophotometry.

Salkowski's Test: On adding a few drops of conc. Sulphuric acid to the plant extract and allow the solution to stand for some time, formation of brown ring indicated the presence of phytosterols in the plant extract.

TEST FOR FLAVONOIDS

$FeCl_3$ Test: To 1 ml of the extract, 3 ml of distilled water followed by few drops of 10% aqueous Ferric chloride solution was added. Formation of blue or green colour indicates the presence of flavonoids. Shinoda Test: To 2 ml of the extract, 1 ml of 1% ammonia solution was added. Appearance of yellow colour indicates the presence of flavonoids.

Shinod's Test: In this test, four pieces of magnesium fillings (ribbon) are added to the ethanolic extract followed by a few drops of concentrated hydrochloric acid. A reddish colour indicates the presence of flavonoid.

TEST FOR FIXED OILS AND FATS

Spot test: Take the sample to be tested, press a little in the folds of the filter paper. On folding, if there is the appearance of greasy spot indicates the presence of oils or fats. The spot grows larger on heating and drying the filter paper.

Saponification: Take approximately 100 mg of oil or fat in a test tube. Add 3 mL of alcoholic-KOH and mix well. Place the tube in a boiling water bath for 15-20 min. Saponification value represents mg of potassium hydroxide required to saponify one gram of fat under the conditions specified. It is a measure of the average molecular weight of all the fatty acids present in the sample as triglycerides.

TEST FOR FREE AMINO ACIDS

Millon's reagent test: Millon's test is specific to phenol containing structures (tyrosine is the only common phenolic amino acid). Millon's reagent is concentrated HNO3, in which mercury is dissolved. As a result of the reaction a red precipitate or a red solution is considered as positive test.

Ninhydrin reagent test: A 2% solution of ninhydrin is prepared by dissolving 0.2 grams of ninhydrin in 10ml of either ethanol or acetone. 1% solution of the amino acid (analyte) in distilled water is prepared, few drops of 2% ninhydrin solution is added to this solution. Test tube is kept in a warm water bath for 5 min; development of a deep blue/violet colour indicates presence of amino acids.

TESTS FOR FIXED OILS AND FATS

Spot test Take the sample and place it between the folds of filter paper and rub it lightly. Presence of translucent spots on the filter paper confirms the presence of fats in in the plant material.

Saponification Take a sample a test tube, add strong alkali NaOH, boil the solution in a water bath for 5 min, add ethanol. Observe for the appearance of froth, formation of forth in the test tube indicates the presence of fat in the sample.

TEST FOR FREE AMINO ACID

Millon's test 1 ml of crude extract was mixed with 2ml of Millon's reagent; white precipitate appeared which turned red upon gentle heating that confirmed the presence of protein.

Ninhydrin test 1 ml of crude extract was mixed with 2ml of 0.2% solution of Ninhydrin and boiled. A violet colour precipitate was appeared suggesting the presence of amino acids and proteins.

TEST FOR TANNINS

5% Ferric chloride test: 5 mg of extract was taken and 0.5 ml of 5% ferric chloride was added. The development of dark bluish black color indicates the presence of tannins.

10% Lead acetate test: 10 mg of extract was taken and 0.5 ml of 1% lead acetate solution was added and the formation of precipitate indicates the presence of tannins and phenolic compounds.

TEST FOR SAPONINS

Foam Test: 2 ml of crude extract was mixed with 5 ml of distilled water in a test tube and it was shaken vigorously. Add some drops of olive oil. The formation of stable foam was taken as an indication for the presence of saponins.

GUMS & MUCILAGE

Ruthenium red test: 50 mg of dried mucilage powder was dissolved in 2 mL of distilled water, mixed with a few drops of Ruthenium red solution. Observed for pink color indicates the presence of gums and mucilage.

GC-MS Analysis:

Phyto-components were identified using GC–MS detection system as described previously, however with modification, whereby portion of the extract was analysed directly by headspace sampling. GC–MS analysis was accomplished using an Agilent 7890A GC system set up with 5975C VL MSD (Agilent Technologies, CA, and USA). Capillary column used was DB-5MS (30 m × 0.25 mm, film thickness of 0.25 µm; J&W Scientific, CA, USA). Temperature program was set as: initial temperature 50°C held for 1 min, 5°C per min to 100°C, 9°C per min to 200°C held for 7.89 min, and the total run time was 30 min. The flow rate of helium as a carrier gas was 0.811851 mL/ min. MS system was performed in electron ionization (EI) mode with Selected Ion Monitoring (SIM). The ion source temperature and quadruple temperature were set at 230°C and 150°C, respectively. Identification of phyto-components was performed by comparison of their retention times and mass with those of authentic standards spectra using computer searches in NIST 08.L and Wiley 7n.l libraries[40,41]

FTIR Analysis

Some of the sample fractions absorb light when infrared light passes through a sample. However, some frequencies are transferred through the sample without any absorption occurring. Infrared absorption is related to the vibrational changes that occur inside a molecule when it is exposed to IR. Infrared spectroscopy is a vibrational spectroscopy that can detect the variations in the vibrations in the range. Different bonds (C–C, C=C, C≡C, C–O, C=O, O–H, N–H) have altered vibrational frequencies. If these chemical bonds are present in an organic molecule, they can be identified by characteristic frequency absorption band in IR spectrum. Fourier Transform Infrared Spectroscopy is a high resolution analytical tool to identify the chemical bonds in a compound. FTIR offers a rapid and non-destructive investigation to fingerprint herbal extracts. FTIR were studied using Shimadzu spectrometer (Shimadzu, Japan). For this, 3.0 mg of sample was dispersed in 300 mg of spectroscopic grade KBr and subsequently pressed into disk at 10 MPa for 3 min. The spectra obtained were recorded with an average of 25 scans and a resolution of 4 cm^{-1} in the range of 4000–400 cm^{-1}.

RESULTS AND DISCUSSION

Tomato, a widely consumed staple vegetable crop, offers a real potential to combat human nutritional deficiencies. Tomatoes are rich in micronutrients and other bioactive compounds (including vitamins, carotenoids, and minerals) known to be essential or beneficial for human health[41]. Preliminary phytochemical screening of SL wild fruit extracts revealed the presence of alkaloids phenols, flavonoids, tannins, steroids, and resins, in Petroleum Ether Extract (SLFPEE) Chloroform Extract (SLFCE) Methanol Extract (SLFME) (Table 1) which could be responsible for wide range of biological properties. GCMS analysis of the wild fruit juice of *Solanum lycopersicum* revealed the presence of following compounds (CID number – Molecular Formula given in parenthesis) 3-methylheptane (11519 - C_8H_{18}); Ethylcyclohexane (1550 - C_8H_{16}); 2-Methyl-4,6-octadiyn-3-one (562128 - $C_9H_{10}O$); 5,6-Dimethylundecane (519454 - $C_{13}H_{28}$); (3E)3-Hexen-2-one (5367744 - $C_6H_{10}O$); 2,2-Dimethylbutane (6403 - C_6H_{14}); 1,2-Diphenyl-1-butanone (297636 - $C_{16}H_{16}O$); Isopropylbenzene (2-phenylpropane) (7406 - C_9H_{12}); 3,5-Dimethyloctane (139989 - $C_{10}H_{22}$); 2-Phenyl-3-buten-1-ol (250465 - $C_{10}H_{12}O$); 2,4,4-Trimethylhexane (28024 - C_9H_{20}); Benzoylcarboxaldehyde (Phenylglyoxal) (14090 - $C_8H_6O_2$); Cis-3-Methyl-Endo-Tricyclo[5.2.1.0(2.6)]Decane (557024 - $C_{11}H_{18}$); 2,4-Dimethylhexan-3-One (85770 - $C_8H_{16}O$); Benzene acetic acid,2-phenylethyl ester (7601 - $C_{16}H_{16}O_2$); Cyclopentacycloheptene (9231 - $C_{10}H_8$); 2,3-Heptanedione (60983 - $C_7H_{12}O_2$); 1,6-Methano[10] annulene (139979 - $C_{11}H_8Br_2$); 1-Naphthaleneacetic acid, methyl ester (17891 - $C_{13}H_{12}O_2$); N,N-Dimethylmethanesulfonamide (70191 - $C_3H_9NO_2S$); Methyl tridecanoate (15608 - $C_{14}H_{28}O_2$); Cis 9-Octadecanoic acid (445639 - $C_{18}H_{34}O_2$); Methyl 15-methylheptadecanoate (554152 - $C_{19}H_{38}O_2$); 9-Octadecenoic acid (637517 - $C_{18}H_{34}O_2$); Methyl (Z)-octadec-9-enoate (5364509 - $C_{19}H_{36}O_2$); (Z)-octadec-9-enamide (5283387 - $C_{18}H_{35}NO$); Methyl 2-ethyl-2-methylicosanoate (575666 - $C_{24}H_{48}O_2$); 1,2,3,4,4a,5,6,7,8,9,10,11,12,12a-tetradecahydrobenzo[10]annulene (565944 - $C_{14}H_{26}$) (Table 2).

Fruit juice of *Solanum lycopersicum* showed FTIR peaks at 2991.05, 2921.63, (N-H stretching) 2873.42, 2856.06, (CH3, CH2 & CH 2 or 3 bands) between the reference ranges 2850-3000 and 2695-2830 indicated the presence of alkanes. The peaks at 2770.24 (2695-2830), 2141.56 (2140-2175), 1716.34 (1706-1720), 1646.91, 1665.45 ((1640-1690), 1530.24, 1505.17 (1500-1550), 1454.06 (1400-1500), 1436.71 (1395-1440), 1408.75 (1380-1410), 1322.93 (1210-1390), 1246.75, 1194.69 ((1020-1250), 1070.3,1042.34 (1030-1070), 939.163, 921.807 (910-959) and 897.701 (675-900) specifies the presence of aldehydes & ketones, thiocyanate, carboxylic acid, imine/oxime, nitro compound, aromatic, carboxylic acid, sulfonyl chloride, phenol, alkyl aryl ether, sulfoxide, carboxylic acids and aromatics respectively. The peaks at 828.882, 821.527 (550-850), 700.998 (610-700) and 590.111 (515-690) represents the existence of alkyl halides and alkynes (Table 3; Fig. 1). Presence of these functional groups in the sample analysed signifies the presence of phytocompounds and nutrients such as potassium, folic acids, neoxanthin, lutein, α-cryptoxanthin, α-carotene, β-carotene, cyclolycopene, and β-carotene 5, 6-epoxide as indicated by 42. Faria-Silva et al.[42], flavonoids, vitamin C, hydroxycinnamic acid derivatives and anti-oxidative and anti-cancer phyto compound lycopene's[43]. ADMET prospecting these bioactive molecules in SL could pave way for the successful exploitation of the PBNPs in the days to come[44-49].

CONCLUSION

Tomato has high global demand and hence widely cultivated fruit world-over. It has a unique savoury flavour, rich nutrient content and health-promoting properties. Tomato fruit juice is a rich source of natural products with bioactives which are ultimately responsible for health promoting activities. In vitro and in vivo studies with animal models along with human cell lines have demonstrate that tomato fruit has compounds with antiplatelet, antioxidant, anticancer, antimutagenic, antimicrobial, and neuroprotective activities. However, further research is warranted to unravel in-depth molecular mechanism underlying with bioactivities of PBNPs in tomato. Furthermore, information on bioavailability and bioaccessibility of compounds from tomato in human body is far lacking. Therefore, ADMET evaluation with regard to the safety and efficacy of bioactive compounds is expected expand the capabilities of PBNPs in tomato for its utilization in the functional food, cosmeceutical and health-care (pharmaceutical) industries.

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Table 1: Qualitative Phytochemical Examination with solvent plant extracts

S. No	Plant constituents tested & Reagent used	*Solanum lycopersicum* Wild Fruit Sample		
		Petroleum Ether Extract (SLFPEE)	Chloroform Extract (SLFCE)	Methanol Extract (SLFME)
1	**TEST FOR ALKALOIDS**			
1.1	*Mayer's test*	-	-	+
1.2	*Dragendorff's test*	-	-	+
1.3	*Hager's test*	-	-	+
1.4	*Wagner's test*	-	-	+
2.1	**TEST FOR GLYCOSIDES – Anthroquinone**			
2.1.1	*Borntrager's test*	-	-	-
2.1.2	*Baljet test*	-	-	-
2.1.3	*Legal's test*	-	-	-
2.2	**TEST FOR GLYCOSIDES - Cardiac**			
2.2.1	*Keller-Killani test*	-	-	-
3	**TEST FOR CARBOHYDRATES**			
3.1	*Molish's test*	+	-	+
3.2	*Fehling's solution test*	+	-	+
3.3	*Bendict's reagent test*	+	-	+
4	**TEST FOR PHYTOSTEROLS**			
4.1	*Libermann Burchard's*	+	-	+
4.2	*Salkowski's test*	+	-	+
5	**TEST FOR FLAVONOIDS**			
5.1	*Ferric chloride test*	-	-	+
5.2	*Shinod's test*	-	-	+
6	**TEST FOR FIXED OILS AND FATS**			
6.1	*Spot test*	-	-	-
6.2	*Saponification*	-	-	-
7	**TEST FOR FREE AMINO ACIDS**			
7.1	*Million's reagent*	+	+	+
7.2	*Ninhydrin reagent*	+	+	+
8	**TEST FOR TANNINS**			
8.1	*5% Ferric chloride*	+	+	+
8.2	*10% Lead acetate*	+	+	+
9	**TEST FOR SAPONINS**			
9.1	*Foam test*	-	+	-
10	**GUMS & MUCILAGE**	-	-	-

Note: + indicates positive result; - indicates negative result

Table 2: GCMS profile of *Solanum lycopersicum* Fruit Methanol Extract

RT-1 (min)	Name of the Compounds	CID	MF	MW	PA (%)
3.83	3-methylheptane	11519	C_8H_{18}	11.23	5.81
4.46	Ethylcyclohexane	1550	C_8H_{16}	112.21	1.75
5.11	2-Methyl-4,6-octadiyn-3-one	562128	$C_9H_{10}O$	134.17	3.01
6.34	5,6-Dimethylundecane	519454	$C_{13}H_{28}$	184.36	6.09
7.17	(3E)3-Hexen-2-one	5367744	$C_6H_{10}O$	98.14	1.35
7.46	2,2-Dimethylbutane	6403	C_6H_{14}	86.18	1.51
7.87	1,2-Diphenyl-1-butanone	297636	$C_{16}H_{16}O$	224.3	1.59
8.94	Isopropylbenzene (2-phenylpropane)	7406	C_9H_{12}	120.19	5.76
9.64	3,5-Dimethyloctane	139989	$C_{10}H_{22}$	142.28	6.98
10.16	2-Phenyl-3-buten-1-ol	250465	$C_{10}H_{12}O$	148.21	2.29
10.44	2,4,4-Trimethylhexane	28024	C_9H_{20}	128.25	1.09
10.87	Benzoylcarboxaldehyde (Phenylglyoxal)	14090	$C_8H_6O_2$	134.13	0.89
12.33	Cis-3-Methyl-Endo-Tricyclo[5.2.1.0(2.6)]Decane	557024	$C_{11}H_{18}$	150.26	2.19
12.95	2,4-Dimethylhexan-3-One	85770	$C_8H_{16}O$	128.21	1.51
14.16	Benzene acetic acid,2-phenylethyl ester	7601	$C_{16}H_{16}O_2$	240.31	1.73
14.72	Cyclopentacycloheptene	9231	$C_{10}H_8$	128.16	1.61
16.05	2,3-Heptanedione	60983	$C_7H_{12}O_2$	128.16	0.81
18.02	1,6-Methano[10] annulene	139979	$C_{11}H_8Br_2$	299.99	4.79
18.44	1-Naphthaleneacetic acid, methyl ester	17891	$C_{13}H_{12}O_2$	200.23	4.11
21.38	N,N-Dimethylmethanesulfonamide	70191	$C_3H_9NO_2S$	123.18	0.59
26.69	Methyl tridecanoate	15608	$C_{14}H_{28}O_2$	228.37	3.12
27.09	Cis 9-Octadecanoic acid	445639	$C_{18}H_{34}O_2$	282.5	18.63
27.33	Methyl 15-methylheptadecanoate	554152	$C_{19}H_{38}O_2$	298.5	3.64
27.29	9-Octadecenoic acid	637517	$C_{18}H_{34}O_2$	282.5	19.53
28.53	Methyl (Z)-octadec-9-enoate	5364509	$C_{19}H_{36}O_2$	296.5	9.41
28.75	(Z)-octadec-9-enamide	5283387	$C_{18}H_{35}NO$	281.5	10.82
29.07	Methyl 2-ethyl-2-methylicosanoate	575666	$C_{24}H_{48}O_2$	368.6	2.47
30.65	1,2,3,4,4a,5,6,7,8,9,10,11,12,12a-tetradecahydrobenzo[10]annulene	565944	$C_{14}H_{26}$	194.36	3.56

Table 3: FTIR spectra of functional groups in SLFME sample

PEAK VALUE (cm^{-1})	REFERENCE RANGE	BOND NATURE	FUNCTIONAL GROUP
3700.73	3584-3700	O-H Stretching, Medium, Sharp	Alcohol
3395.07	3200-3550	N-H Stretching (Medium)	Aliphatic Primary Amine
2995.87	2800-3000	C-H Stretching (Medium)	Amines
2930.31		C-H Stretching (Medium)	
2356.59	-	-	Unknown
2112.64	2100-2140	-C≡C- Stretching (Weak)	Alkynes
1827.22	1650-2000	C=C=C Stretching	Allene
1617.02	1610-1620	C=C Stretching,	A,B –Unsaturated Ketone
1504.2	1500-1550	N-O Stretching, Strong	Nitro Compound
1403.92	1380-1410	S=O Stretching, Strong	Sulfonyl Chloride
1294	1266-1342	C-N Stretching, Strong	Aromatic
1239.04	1200-1275	C-O Stretching, Strong	Alkyl Aryl Ether
1197.58	1124-1205	C-O Stretching, Strong	Tertiary Alcohol
1067.41	1050-1085	C-O Stretching, Strong	Primary Alcohol
940.128	910-950	O-H Bend (Medium)	Carboxylic Acid
916.022			
863.953	675-900	C-H 'Opp'' (Strong)	Aromatics
819.598	790-840	C=C, Bending Medium	Alkenes
778.136	550-850	C-Cl Stretching, Medium	Alkyl Halides
751.138			
626.752	610-700	-C≡C-H:C-H Bend (Broad, Strong)	Alkynes
516.829	515-690	C-Br Stretching (Medium)	Alkyl Halides

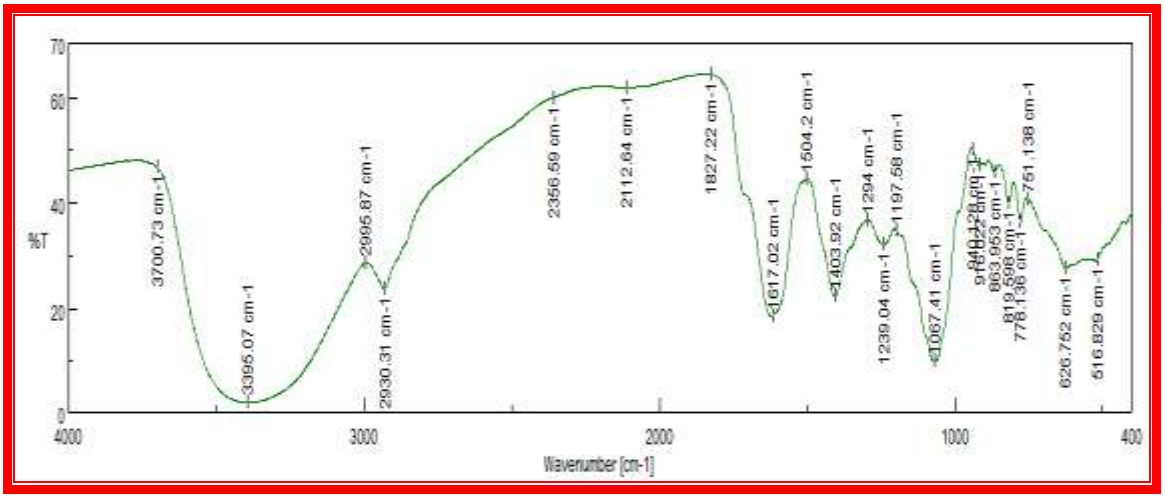

Figure 1: FTIR spectra of functional groups in SLFME sample