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GCMS AND FTIR ANALYSIS ON THE METHANOLIC EXTRACT OF *CORIANDRUM SATIVUM* LEAVES

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ABSTRACT

Coriander leaves are one of most commonly used spices which has beneficial physiological effects on lipid metabolism, stimulate digestion, inhibit platelet aggregation, antioxidant, antilithogenic, anti-inflammatory potential, antibacterial, diuretic, expectorant, anti-pyretic, laxative etc. Our present work aimed to identify the possible phytochemical compounds using GCMS along with its functional groups using FTIR, present in the methanolic extract of *Coriandrum Sativum* leaves. The GC-MS analysis extract of *Coriandrum Sativum* leaves showed nearly 40 compounds and most of the compounds identified has proved for possessing potentially medicinal values. As per the data analysis of FTIR, it was found that the strong absorption bands at 3372.30 cm^{-1} which is representative for N-H stretching vibrations, characteristic of the presence of amino acids and also at 1253.54 cm^{-1} representing the stretching vibrations of C-O indicative of the acid.

KEYWORDS: GCMS, FTIR, *Coriandrum*, chromatogram, spectral analysis.

1. INTRODUCTION

Coriandrum Sativum L. (family Umbelliferae) belongs to ayurvedic medicinal herb known as the Dhanyaka. It was originated around the Mediterranean and is cultivated mainly in the tropical areas. It is small sized plant that grows throughout India and Italy. The leaves have a pleasant aroma and the plant (green coriander) is used in food industry for preparing sauces and for flavouring of curries and soups [1]. Coriander leaves majorly contains Kaempferol, quercetin, acacetin, as the major polyphenolic content and among the phenols it has vanillic acid, p-coumaric acid, Cis-ferulic acid, Trans-Ferulic acid.[2]

1.1. Taxonomic classification[3]

Kingdom	:	Plantae;
Subkingdom	:	Tracheobionta;
Super division	:	Spermatophyta;
Division	:	Magnoliophyta;
Class	:	Magnoliopsida;
Subclass	:	Rosidae
Order	:	Apiales
Family	:	Apiaceae
Genus	:	*Coriandrum L.*
Species	:	*Coriandrum Sativum L.*

1.2. Nutritional composition of *Coriandrum Sativum* leaves

Table – 1: Nutrient composition of coriander leaf as per USDA (National Nutrition Data base, 2013).[4]

Nutrient	Amount (per 100 g)
Water	7.30 g
Energy	279 kcal
Protein	21.93 g
Total lipid (fat)	4.78 g
Carbohydrates	52.10 g
Total dietary Fiber	10.40 g
Calcium, Ca	1246 mg
Iron, Fe	42.46 mg
Magnesium, Mg	694 mg
Phosphorus, P	481 mg
Potassium, K	4466 mg
Sodium, Na	211 mg
Zinc, Zn	4.72 mg
Vitamin C	566.7 mg
Thiamine	1.252 mg
Riboflavin	1.500 mg
Niacin	10.707 mg
Vitamin B-12	0.00 µg
Vitamin A,	293 µg
Fatty acids (MUFA)	2.232 g
Fatty acids (PUFA)	0.328 g

The protein is higher in leaves than in seeds (12.37g), possess high amount of calcium, iron, magnesium,

phosphorus, sodium, zinc, vitamin–C, thiamine, riboflavin, niacin than seeds.[4] The methanol extract of *Coriandrum Sativum* was found to be more effective against other solvents like acetone, benzene. The preliminary phytochemical test performed by using methanolic extracts contains Alkaloids, Flavanoids, Tannins, Saponin, Tarpenoids, Carbohydrates and Sterols. Hence, we can use *Coriandrum Sativum* as natural antimicrobial in industrial food & drugs.[5] Coriander plant has medicinal importance because of natural anti-oxidants acting as reducing agents, free radical scavenger etc. Anti-oxidant activity is due to presence of bioactive compounds i.e., flavones, anthocyanins, coumarins, lignans, catechins and isocatechins. Presently there is a greater interest to identify the anti-oxidants that possess lesser side effects or no side effects. Coriander is one of most commonly used spices which has beneficial physiological effects on lipid metabolism, stimulate digestion, inhibit platelet aggregation, antioxidant, antilithogenic, anti-inflammatory potential, antibacterial, diuretic, expectorant, anti-pyretic, laxative etc. It also known for medicinal and nutritional properties and also possess essential oils. Coriander is used in household medicines for stomach disorders, bed cold, seasonal fever etc. Hence coriander is considered to be store house for bioactive compounds.[6]

GC/MS is a combination of two different analytical techniques Gas chromatography (GC) and Mass Spectrometry (MS), used to analyze biochemical and organic samples. GC can separate semi-volatile and volatile compounds present in sample with great resolution, but it cannot identify them. While the MS can provide detailed structural information so that they can be identified but cannot be quantified. Application of GCMS is to monitor and clean the environment, criminal forensics, law enforcement, security, food, beverage and perfume analysis. It can also be used in astro chemistry and medical field.[7]

FTIR is known as Fourier Transform Infrared Spectroscopy. It is a technique to identify the functional groups present in sample. It is also rapid method to characterize cell properties and identify their functional groups in organic molecules depending on their vibrating modes at different wave numbers. The bands correspond to protein, polysaccharide, polyphosphate groups, carbohydrates functional groups can be identified at different environments. The characteristic of chemical bond can be analyzed by wavelength of light absorbed. By interpreting the infrared absorption spectrum, the chemical molecule can be identified.[8] Our present work was aimed to identify the possible phytochemical compounds present in the methanolic extract of *Coriandrum Sativum L.* using GCMS along with its functional groups using FTIR.

2. MATERIALS AND METHODOLOGY

2.1. Preparation of plant materials and extract for In Vitro studies

The *Coriandrum Sativum L.* is collected from The Horticultural Department, University of Agricultural sciences, Gandhi Krishi Vignan Kendra, Bangalore. 10 grams of the dried leaves material was powdered and placed in Soxhlet extractor along with 150 ml of methanol and refluxed at 60°C for 8hrs. The methanolic extract was filtered through Whatmann No. 1 filter. The filtrate was evaporated to dryness at 80°C and stored until further analysis. For analysis, the dried material was reconstituted in 1 ml methanol, and was subjected for GCMS analysis.[9]

2.2. Gas Chromatography-Mass Spectrometry

The methanolic extract of the *Coriandrum Sativum L.* was subjected to GC- MS Clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: Restek Rtx^{R} – 5, (30 meter X 0.25 mm) (5% diphenyl / 95% dimethyl polysiloxane), running in electron impact mode at 70 eV; helium (99. 999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 1.0 µl was employed(split ratio of 10:1); injector temperature 280 ^{0}C. The oven temperature was programmed from 40°C (isothermal for 5 min.), with an increase of 6 ^{0}C / min to 280 ^{0}C, then ending with an isothermal for 15min at 280°C. Mass spectra were taken at 70 eV; 0.5 seconds of scan interval and fragments from 40 to 550 Da. Total GC running time was 60 minutes.

2.3. Identification of Compounds

Interpretation of mass spectrum GC-MS was done using the database of National Institute of Standard and technology (NIST). The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library.

2.4. Fourier Transform Infrared Spectrophotometer (FTIR) Analysis

Fourier Transform Infrared Spectrophotometer (FTIR) is the most important and powerful tool for identifying the functional groups present in the sample. The wavelength of light absorbed is the characteristic of the chemical bond. The chemical bonds in a molecule can be determined by interpreting the infrared absorption spectrum.

Reagents required: Potassium bromide (KBr).
Control: Pong oil.
Procedure: Dried powder of methanolic solvent extract of *Coriandrum Sativum* leaves was used for FTIR analysis. 10mg of the sample was encapsulated in 100mg of KBr pellet, to prepare translucent sample disc. The *Coriandrum Sativum* leaves methanolic extract was loaded in FTIR spectroscope (Burker make Tensor 27 model FT-IR, 64 scans at a spectral resolution of 4 cm^{-1}).

3. RESULTS

3.1. Gas Chromatography Mass Spectrometry (GCMS) Analysis

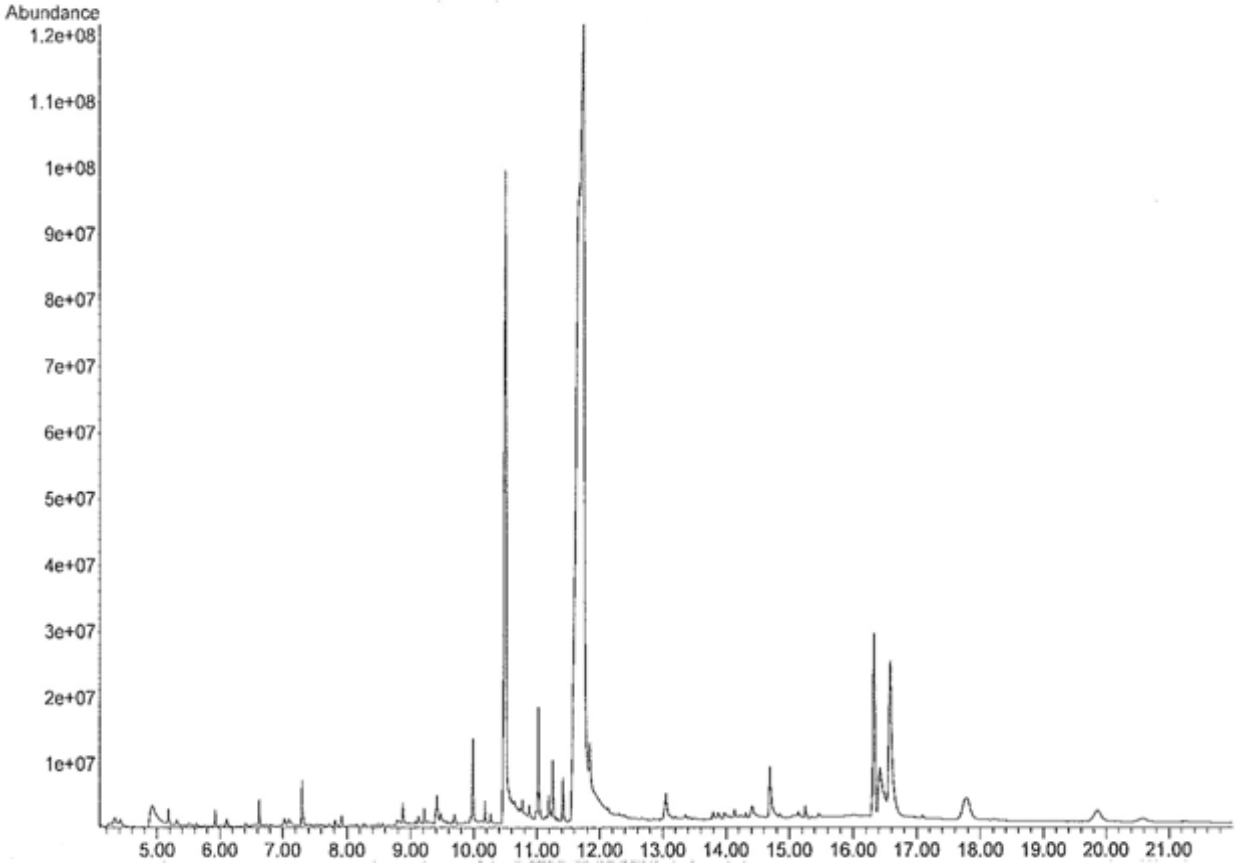

Figure – 1: GC- MS chromatogram of methanolic extract of *Coriandrum Sativum* leaves.

Table – 2: GC- MS chromatogram of methanolic extract of *Coriandrum Sativum* leaves.

Retention Time	Name of the Compounds	Peak Area	Biological Activity
4.332	4- pyranone, 2,3- dihydro-1,3-cyclopentanedione 4,5- dihydro-2-methylimidazole-4-on	0.47	Antibacterial, Anticancer agent. Anticancer activity.
4.925	Glycerin	2.49	Used as Solvent, emollient, pharmaceutical agent or sweetening agent.
7.303	2- Methoxy-4-vinylphenol 3- Methoxyacetophenone	0.78	Aromatic Substance used as a Flavoring agent. Responsible for Natural Aroma of Buckwheat. Activity not found
9.412	Tetradecanoic acid	0.70	Anti-constipation, Protein kinase inhibitor, Used in the treatment of mycosis, neoplastic diseases, inflammatory, immune diseases
9.981	Cyclohexadecane Cyclotetradecane Cetene	1.45	Antioxidant and Antibacterial agent, Used for drug formulation. Antimicrobial agent, Drug formulation and Antioxidant activity. Antitumor and Antioxidant activity.
10.508	n- Hexadecanoic acid	20.85	Anti – inflammatory and Antimicrobial activity
11.029	E-15-Heptadecenal Cyclohexadecane n-Nonadecanol-1	1.79	Invitro hypoglycemic and Anti-microbial activity. Hepatitis b Antiviral agent, Inhibitors of viral replication, inhibitor of beta secretase. Cell parameters, temperatures and enthalpies of transition can be detected.
11.257	9,12-Octadecadienoic acid (Z, Z)-methyl ester	0.97	Hepatoprotective, Antihistaminic, hypocholesterolemic, Anti-eczemic

	11,14-Octadecadienoic acid, methyl ester 10,13-Octadecadienoic acid, methyl ester		No activity reported. No activity reported
11.418	trans-Geranylgeraniol 5,9,13-Pentadecatrien-2-one,6,10,14-trimethyl-, (E, E)-2,2-dimethyl-3-(3,7,16,20- tetramethyl-heneicosa-3,7,11,15,19-pentaenyl)-oxirane	0.76	For treating Neovascular diseases, Inflammatory diseases and Cancer No activity reported.
11.688	9,12-octadecadienoic acid (Z, Z)-	44.64	Hepatoprotective, Antihistaminic, hypocholesterolemic, Anti-eczemic
11.844	9,12-octadecadienoic acid (Z, Z)- 9,12,15-octadecatrienoic acid (Z, Z, Z)-	3.40	Hepatoprotective, Antihistaminic, hypocholesterolemic, Anti-eczemic Antimicrobial, Anticancer, Hepatoprotective, Anti-arthritic, Anti-asthma, diuretic.
13.048	Cyclohexane,1- (1,5- dimethylhexyl)-4- (4-methylpentyl)- 1-Eicosene 2-piperidinone, N-[4-bromo-n-butyl]-	1.03	Antibacterial, Anticancer activity Immunosuppressant, Anticancer agent, Suitable for radical scavenging. myelostimulatory effect, Antibacterial and Antifungal activity.
14.413	Octadecane, 1-(ethenyloxy)- 3-Eicosene- (E)-1-tridecene	0.57	Antimicrobial, Antioxidant, Anti – inflammatory effect, Immunosuppressant, Anticancer agent, Suitable for radical scavenging. Treating Alzheimer's disease, Protease inhibitor
14.695	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester Glycerol 1-palmitate Palmitoyl chloride	1.56	Anti – inflammatory and Antimicrobial activity Antibacterial, Antimicrobial agent, Used to increase blood stability, Treating Secondary Hyperparathyroidism. Antimicrobial, Antibacterial agent and it is used as Antibiotic
16.324	Squalene	5.23	Antimicrobial, Antioxidant, Antitumor agent, Potential uses in cosmetic dermatology.
16.420	Oleic acid,3-hydroxypropyl ester oleic anhydride 7,12a-dimethyl-1,2,3,4,4a,11,12,12a-octahydrochrysene	3.28	Antibacterial, Antifungal and Antioxidant activity. No activity reported
16.581	9,12-octadecadienoic acid (Z, Z)- 2-hydroxy-1-(hydroxymethyl) ethyl ester 2,3-dihydroxy propyl ester	7.65	Hepatoprotective, Antihistaminic, Hypocholesterolemic, Anti-eczemic. Antibacterial, Antimicrobial agent Antimicrobial activity and used as Antibiotic.
17.779	Benzenesulfonyl chloride, 4-fluoro Benzenesulfonyl chloride, 4-fluoro 3-azabicyclo[3.3.0] octane- 2,4- dione, 7-isopropylidene-3- phenyl-	2.38	Antiviral, Proteosome inhibitor and Antiinfective agent. Proteasome inhibitor, Kinase modulator, Used to treat retroviral infections, Sulfonamide inhibitor. Antibacterial, Antimicrobial agent. Anticancer, Antibacterial agent and Herbicidal activity.

3.2. Fourier Transform Infrared Spectrophotometer (FTIR) Analysis

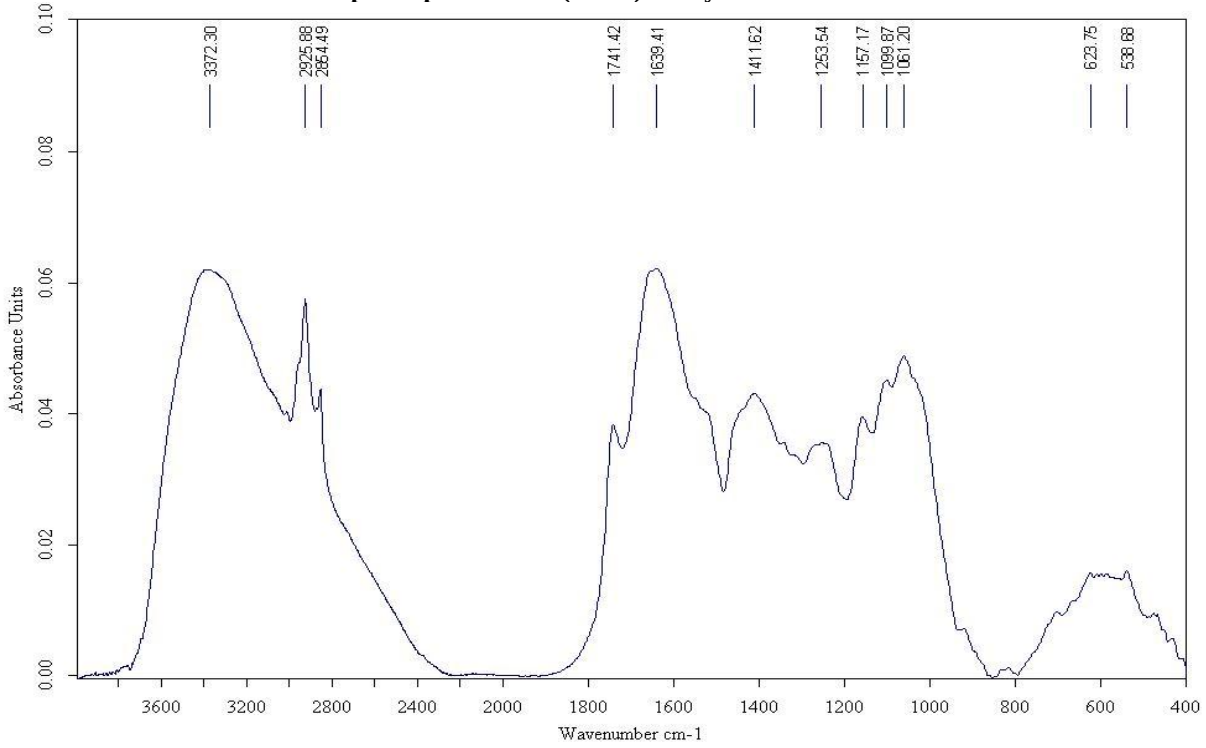

Figure – 2: Infrared spectroscopy spectrum for methanolic extract of *Coriandrum Sativum* leaves.

Table – 3: Infrared spectroscopy spectrum for methanolic extract of *Coriandrum Sativum* leaves.

S.No	Frequency	Group	Intensity
1	3372.30	Amines (N-H)	Medium
2	2925.88	Alkyl (C-H[stretching])	Medium-Strong
3	2854.49	Alkane (C-H)	Medium
4	1741.42	Ester (C=O)	Strong
5	1639.41	Alkene(C=C[stretching])	Variable
6	1411.62	Aromatic (C=C[stretching)	Variable
7	1253.54	Acid (C-O[stretching])	Strong
8	1157.17	Amine (C-N)	Medium
9	1099.87	Secondary Alcohol(C-O [stretching])	Strong
10	1061.20	Primary Alcohol (C-O[stretching])	Strong
11	623.75	Alkyl Halide (C-Cl)	Strong
12	538.68	Alkyl Halide(C-Br)	Strong

4. DISCUSSION

4.1. Gas Chromatography Mass Spectrometry (GCMS) Analysis

From the Figure – 1 and Table – 2, the GC-MS chromatogram of a methanolic extract of *Coriandrum Sativum* leaves showed nearly 40 compounds. Most of the compounds which were reported from leaves were found to be rich in 4- pyranone, 2,3- dihydro-1,3 cyclopentanedione, 4,5- dihydro-2-methylimidazole-4-on, Glycerin, 2- Methoxy-4-vinylphenol,3-Methoxyacetophenone, Tetradecanoic acid, Cyclohexadecane, Cyclotetradecane, Cetene, n-Hexadecanoic acid, E-15-Heptadecenal, Cyclohexadecane, n-Nonadecanol-1, 9,12-Octadecadienoic acid (Z, Z)-methyl ester,11,14-Octadecadienoic acid, methyl ester,10,13-Octadecadienoic acid, methyl ester, trans-Geranylgeraniol, 9,12,15-octadecatrienoic acid (Z, Z, Z)-, Cyclohexane,1- (1,5- dimethylhexyl)-4- (4-methylpentyl)-,1-Eicosene, 2-piperidinone, N-[4-bromo-n-butyl]-, Octadecane,1-(ethenyloxy)- 3-Eicosene-, (E)-1-tridecene, Glycerol 1-palmitate, Palmitoyl chloride, Squalene, Oleic acid,3-hydroxypropyl ester oleic anhydride, 7,12a-dimethyl-1,2,3,4,4a,11,12,12a-octahydrochrysene, 2-hydroxy-1-(hydroxymethyl) ethyl ester, 2,3-dihydroxypropyl ester, Benzenesulfonyl chloride, 4-fluoro Benzenesulfonyl chloride, 4-fluoro 3-azabicyclo[3.3.0] octane- 2,4- dione, 7-isopropylidene-3-phenyl- at retention time 4.33, 4.92, 7.30, 9.41, 9.98, 10.50, 11.02, 11.25, 11.41, 11.68, 11.84, 13.04, 14.41, 14.69, 16.32, 16.42, 16.58, 17.77 respectively. Glycerin has the retention time at 4.92 and peak area of 2.49 is used as solvent, emollient, pharmaceutical agent and as a sweetener.[10]

Cyclohexadecane retention time at 9.98 and peak area of 1.45 is used as Antioxidant and Antibacterial agent and for drug formulation.[11] Cyclotetradecane and Cetene having the same retention time and peak area is used as Antimicrobial agent, Drug formulation, Antioxidant activity and Antitumor activity respectively. n-Hexadecanoic acid having the retention time at 10.50 and peak area of 20.85 is used as Anti inflammatory and antimicrobial agent.[12] E-15-Heptadecenal has the retention time at 11.02 and peak area of 1.79 is used as hypoglycemic agent, anti-microbial agent and Hepatitis B antiviral agent.[13] Cyclohexadecane, n-Nonadecanol-1 with the same retention time and peak area is used as inhibitors of viral replication, beta secretase and measuring the cell parameters, temperatures and enthalpies of transition respectively. 9, 12-octadecadienoic acid (Z, Z) - with the retention time at 11.68 and peak area of 44.64 is used as Hepatoprotective, antihistaminic, hypocholesterolemic and anti-eczemic.[14]

9, 12, 15-octadecatrienoic acid (Z, Z, Z) - with the retention time at 11.84 and peak area of 3.40 has Antimicrobial, Anticancer, Hepatoprotective, Anti-arthritic, anti-asthama, diuretic activity.[15] 1-Eicosene has the retention time at 13.04 and peak area of 1.03 is used as Immunosuppressant, Anticancer agent, Suitable for radical scavenging.[16] Glycerol 1-palmitate with the retention time at 14.69 and peak area of 1.56 is used to increase blood stability and treating Secondary Hyperparathyroidism.[17] Squalene with the retention time at 16.32 and peak area of 5.23 is used as Antimicrobial; Antioxidant and Antitumor agent has potential uses in cosmetic dermatology. 4-fluoro Benzenesulfonyl chloride has retentiom time at 17.77 and peak area of 2.38 is used as Proteosome inhibitor, Kinase modulator, Used to treat retroviral infections and sulfonamide inhibitor.[18]

4.2. Fourier Transform Infrared Spectrophotometer (FTIR) Analysis

FTIR spectroscopy is used to identify some qualitative aspects regarding the organic compounds in methanolic extract of *Coriandrum Sativum* leaves. Several indicator bands that are pertained to functional groups represent chemical components or metabolic products. The FT-IR spectrum exhibits the characteristic fingerprint band features. The infrared spectrum can identify not only the major components inorganic materials but also to find some differences among them. From the Figure – 2 and Table – 3, the very strong absorption bands at 3372.30 cm^{-1} which is the representative for N-H stretching vibrations may be due to the presence of amino acids. The bands at 2925.88 cm^{-1} is due to the stretching vibration of $–CH_3$ and $–CH_2$ groups which indicates the presence of chlorophyll groups.[19] The 1741.42 cm^{-1} corresponds to ester group (C=O) indicative of lactone group. The strong bands at 1253.54 cm^{-1} represent the stretching vibrations of C-O due to the presence of acid. The 1061.20 cm^{-1} band, predict the presence of alcohol. The band at 538-623 cm^{-1} represents alkyl halide compounds.[20]

5. CONCLUSION

The GC-MS chromatogram of the methanolic extract of *Coriandrum Sativum* leaves showed nearly 40 compounds. Most of the compounds which were reported were found to be rich in 4- pyranone, 2,3-dihydro-1,3 cyclopentanedione, 4,5- dihydro-2-methylimidazole-4-on, Glycerin, 2- Methoxy-4-vinylphenol,3- Methoxyacetophenone, Tetradecanoic acid, Cyclohexadecane, Cyclotetradecane, Cetene, n-Hexadecanoic acid, E-15-Heptadecenal, Cyclohexadecane, n-Nonadecanol-1, 9,12-Octadecadienoic acid (Z, Z)-methyl ester,11,14-Octadecadienoic acid, methyl ester,10,13-Octadecadienoic acid, methyl ester, trans-Geranylgeraniol, 9,12,15-octadecatrienoic acid (Z, Z, Z)-, Cyclohexane,1- (1,5- dimethylhexyl)-4- (4-methylpentyl)-,1-Eicosene, 2-piperidinone, N-[4-bromo-n-butyl]-, Octadecane,1-(ethenyloxy)- 3-Eicosene-, (E)-1-tridecene, Glycerol 1-palmitate, Palmitoyl chloride, Squalene, Oleic acid,3-hydroxypropyl ester oleic anhydride, 7,12a-dimethyl-1,2,3,4,4a,11,12,12a-octahydrochrysene, 2-hydroxy-1-(hydroxymethyl) ethyl ester, 2,3-dihydroxypropyl ester, Benzenesulfonyl chloride, 4-fluoro Benzenesulfonyl chloride, 4-fluoro 3-azabicyclo[3.3.0] octane- 2,4- dione, 7-isopropylidene-3-phenyl- respectively. As per the data from FTIR analysis using infrared spectroscopy correlation table, it was found that the very strong absorption bands at 3372.30 cm^{-1} which is the representative for N-H stretching vibrations may be due to presence of amino acids and also at 1253.54 cm^{-1} represent the stretching vibrations of C-O may be an indicative of the acid. Hence, we can conclude that the methanolic extract of *Coriandrum Sativum* leaves is rich in the presence of amino acids and acid groups.

6. ACKNOWLEDGEMENT

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