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# Screening of phytoconstituents and antibacterial activity of leaves and bark of *Quercus leucotrichophora* A. Camus from Uttarakhand Himalaya

Prabhakar Semwal<sup>1\*</sup>, Sakshi Painuli<sup>1</sup>, Himani Badoni<sup>1</sup> and Rakesh K. Bacheti<sup>2</sup>

# Abstract

**Background:** *Quercus leucotrichophora* A. Camus (QL) belongs to the family Fagaceae, commonly known as Banj oak in the Garhwal region of Himalaya, where it is the principal source of fuel, fodder, and medicine.

**Methods:** In the present study, GC-MS analysis has been performed for profiling the chemical composition of methanolic extracts of leaves and bark of QL. The antibacterial activity was evaluated by using the disk diffusion method against five bacterial strains.

**Results:** Total 23 components in bark and 62 components in leaves extracts of QL were identified. The major components identified in the bark extracts were Linoleic acid (19.77%), Lupeol (17.91%), Epi-psi-Taraxastanonol (14.20), and cis-Vaccenic acid (13.10%), while others were present in relatively small amounts. For the leaves extract, the major components were Linoleic acid (17.09%), Simiarene (15.29%), Flavone 4'-oh, 5-oh,7-di-o-glucoside (15.26%), and D-Quinic acid (9.29%), respectively. As far as antibacterial assays are concerned, it was observed that both the extracts are active against most of the tested bacterial strains with the zone of inhibition ranging between  $8.53 \pm 0.50$  to  $19.07 \pm 0.31$  mm, respectively.

**Conclusion:** The GC-MS results revealed the presence of several phytochemical compounds in leaves and bark of QL extract and are recommended as a plant of pharmaceutical importance. The antibacterial analysis showed that both the extracts (leaves and bark) of QL have antibacterial activity against all gram positive (*S. aureus, B. subtilis* and *S. pyogenes*) and gram negative (*E. coli, P. aeruginosa*) bacterial strains.

Keywords: Antibacterial activity, Chemical composition, Himalaya, Quercus leucotrichophora

# Background

Use of plants and plant extracts as a source of medicine has been inherited and is an important component of the health care system in the world. India is the largest producer of medicinal herbs and is known as the botanical garden of the world [1]. The Himalayan region is well known for its huge diversity of flora with more than 10,000 natural plant species, especially medicinal plants. Banj oak (*Quercus leucotrichophora* A. Camus) belonging to the family Fagaceae is an evergreen tree of

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isolated from the ethanolic stem bark extract of OL, whereas the antimicrobial activity of the extract showed highest activity against E. coli followed by S. aureus, P. auroginosa and B. subtilis, respectively [13]. Further, the presence of twenty-three phytoconstituents (major phytocomponent: monoterpenoids) in the volatile extract of bark of QL were analyzed by GC-MS analysis [14]. The fruit extract of OL revealed the presence of higher amount of saturated fatty acid compared to unsaturated fatty acid. The bark and fruit extract of QL possess antimicrobial activity [14, 15]. The QL is used in traditional system of medicine, but still there are not many scientific reports to confirm its phytochemical activity and medicinal properties [16]. Thus, the present study was aimed to investigate the chemical composition and antibacterial activity of methanolic leaves and bark extracts of QL.

# Methods

## Plant collection and preparation of crude extracts

Leaves and bark of QL were collected from the Uttarakhand Himalaya (Tehri district), India and voucher specimens (BSI/NRC-115222) have been kept in the herbarium of Botanical Survey of India (BSI/NRC--Dehradun), Uttarakhand, India. Plant samples (leaves and bark) of QL were cleansed, shade dried and coarsely powdered. Crude powdered material (500 g) was extracted with methanol (80%) using a Soxhlet extractor. The extracts obtained were filtered and concentrated using a rotary vacuum evaporator (Strike-12, Steroglass, Italy) and used for further analysis (GC-MS and antibacterial analysis).

## **GC-MS** analysis

GC-MS analysis was performed at University Science Instrumentation Centre, Jawaharlal Nehru University (JNU), Delhi (India). The analyses of the methanolic extracts were carried out on a GCMS-QP2010 Plus (Shimadzu, Kyoto, Japan). The system was equipped with an auto injector (AOC-20i), head space sampler (AOC-20s), a mass selective detector with an ion source (220 °C) and an interface (260 °C). Rtx-5 MS capillary column (Restek Company, Bellefonte, USA) having 30 m  $(\text{length}) \times 0.25 \text{ mm}$  (diameter)  $\times 0.25 \mu \text{m}$  (film thickness) was used for GC-MS analyses. The mass range of 40-650 m/z with 1000 ev of threshold was used. The injector was set in the split injection mode having 250 °C of temperature. The starting temperature was adjusted to 80 °C (3 min), which afterwards increased to 280 °C with a ramp rate of 10 °C/min. Helium (> 99.99%) with 40.5 cm/s of linear velocity was employed as a carrier gas. The system was programmed with 16.3 ml/min of total flow rate and 1.21 ml/min of column flow according to stranded methods [17, 18]. The bark and leaves extract components were identified on the basis of retention time (RT) by gas chromatography and interpretation of mass spectrum was performed by comparing spectral fragmentation obtained, to the database provided by NIST11.LIB and Wiley8.LIB [17, 18].

## Antibacterial activity

Five pathogenic bacterial strains were used in this study for assessing the antibacterial activity of QL, including the Gram-negative and Gram-positive strains namely; *Escherichia coli* (MTCC-582); *Pseudomonas aeruginosa* (MTCC-2295); *Staphylococcus aureus* (MTCC-3160); *Bacillus subtilis* (MTCC-441); and *Streptococcus pyogenes* (MTCC-1924). The reference bacterial strains were obtained from the Institute of Microbial Technology (IMTECH), Chandigarh (India) and were maintained at 4 °C on slants of nutrient agar (NA) (Merck, Germany). The antibacterial activity of plant extracts was carried out using the disk diffusion method [19]. The methanolic bark and leaves extracts were dissolved in 10% of dimethyl sulfoxide (DMSO). The concentration and

Table 1 GC-MS analysis of Quercus leucotrichophora (Bark) extract

SN	Retention time	Area per cent	Name of compounds
1	10.086	0.19	Beta-Himachalene
2	11.992	0.14	.AlphaEudesmol
3	12.152	0.62	Myristyl acrylate
4	13.239	0.08	1-Octadecene
5	14.150	0.10	Phthalychloride
6	14.643	0.14	Methyl Palmitate
7	15.067	4.53	Pentadecanoic acid
8	16.030	0.11	Heptadecanoic acid
9	16.318	0.24	Linoleic acid methyl ester
10	16.772	19.77	Linoleic acid
11	16.803	13.10	cis-Vaccenic acid
12	16.860	4.13	Ambrettolide
13	16.974	2.92	Octadecanoic acid
14	17.566	0.67	10,12-Hexadecadien-1-ol
15	20.814	0.41	Lignoceric alcohol
16	24.147	0.28	Nonadecyl pentafluoropropionate
17	26.641	0.31	2,3-Oxidosqualene
18	36.274	2.59	Taraxerone
19	36.631	1.45	Clionasterol
20	37.952	6.21	Simiarene
21	38.517	14.20	Epi-psi-Taraxastanonol
22	39.357	17.91	Lupeol
23	41.461	1.81	Sitostenone
Total		91.91	
Unidentified		0.34	

1     \$184     0.23     23-01hydro-35-othydroxy-6-methyl-4 hyspan-t-one       2     8.307     0.26     4-Props/phonel       3     10.055     0.07     Lauric and       4     11.354     0.36     beta-Methydpurcside       5     11.750     9.29     D-Quinic add       6     11.924     0.14     21-Indexn012-bif/prom 2-one, 3.34.45.6.7.8.8b octahydro-8.8 dimethyl       7     12111     0.87     Tecadeo) acrylate       8     12.37     0.12     Methosy-upnol       9     12.533     0.08     21-Hydroxy-5-sopropyl-24-6-cychothydrox-9.8 dimethyl       10     12.733     0.25     Methyl-4Hydroxy-5-sopropyl-24-6-cychothydrox-9.5 dvtimethyl-4-B-oxo-1-butenyl-12       11     12.856     1.44     Confical       12     13.283     0.05     Capylone       13     13.453     0.84     2-Cychothecane       14     13.453     0.84     2-Cychothecane       14     14.64     0.25     C-2-Tecadecan-1-01       15     Natopyhytakine     12     13.07 <t< th=""><th>SN</th><th>Retention time</th><th>Area percent</th><th>Name of compounds</th></t<>	SN	Retention time	Area percent	Name of compounds
2     8397     0.26     4Propylphenol       3     10.073     0.07     Lauric acid       3     10.075     0.07     Lauric acid       5     11.750     9.29     D.Quinic acid       6     11.944     0.14     21-finderol 1_2-Bifuon 2-one, 3.3.4.5.6.7.88b-octahydro-88-dimethyl       7     12.111     0.87     Teradaccyl acyl-tyrus-       8     12.367     0.12     Methodys-synchronyl-3.4.6.6.7.88b-octahydro-88-dimethyl-       9     12.333     0.85     2-Hydroxy-Swnchronyl-3.4.6.cylchinptatrienone       10     12.838     0.85     Cycloperadceane       11     12.856     1.44     Conferol       12     13.201     0.67     Cycloperadceane       13     13.203     0.84     Caplylone       14     13.453     0.84     Caplylone       14     13.453     0.84     Caplylone       14     13.456     0.20     Caplylone       14     14.464     0.25     E-2-Teradecen-1-ol       14     14.566     0.27	1	5.184	0.23	2,3-Dihydro-3,5-dihydroxy-6-methyl-4 h-pyran-4-one
3     10825     007     Lauric acid       4     11354     0.36     bata-Methylglucoidie       5     11324     0.14     Defaultic acid       6     11924     0.14     2141nden012-bifuan-2-one, 3,3a,45,6,788b-octahydo-88-dmethyl       7     12.111     0.87     Testadecyl acylune       7     12.331     0.68     214 bifuxy-3-socropyl 24.6-cycloheptatricnone       10     12.733     0.55     Methyle(4-bydroxy-3-methoxynbyneyl) acetat       11     12.856     0.76     Cyclopentadcane       12     13.000     0.07     Cyclopentadcane       13     13.783     0.56     Cyclopentadcane       14     13.453     0.64     Cyclopentadcane       15     13.711     0.67     Oldic acid       16     13.670     0.15     Neophytadiene       17     14.063     0.20     Captoine       18     14.164     0.25     Caltoracent-i-d       19     14.56     0.20     Restadoranic acid       19     14.56     0.20	2	8.397	0.26	4-Propylphenol
4     11.454     0.36     Jack-Methylglucoside       5     11.759     0.29     Quinti and       6     11.924     0.14     Hinderol (2-biftma-2one, 3.34,45,47,8,8-octalydes-8,8-dmethyl)       7     12.111     0.87     Terdaccy acylate       8     12.367     0.12     Methyl-Hydroxy-Stoprop/12.6-cyclohepatitionone       9     12.533     0.25     Methyl-Hydroxy-Stoprop/12.6-cyclohepatitionone       10     12.733     0.25     Methyl-Hydroxy-Stoprop/12.6-cyclohepatitionone       12     3.200     0.07     Cycloheam-1-one, 4-hydroxy-3,56-trimethyl-4-(3-oxel-buteryl)-       13     13.271     0.67     Oles acid       14     13.453     0.84     Cycloheam-1-one, 4-hydroxy-3,56-trimethyl-4-(3-oxel-buteryl)-       15     13.711     0.67     Oles acid       14     13.453     0.84     Capplone       18     14.164     0.25     E-Terdaccon-1-ol       19     14.459     0.20     SetTerdaccon-1-ol       19     14.596     0.27     Methyl finitare       21     15.05	3	10.625	0.07	Lauric acid
511.7599.790-Quine acid611.9240.142H-Indero[1,2-bi]tran-2-one, 3.3a, 5.0.78.8b-octahydro-8.8-dimethyl712.1110.87Teradecyl arylate812.3670.12Methoryeagool912.330.862Hydroxy-isscorop/24.6-cycloheptationene1012.7330.862Hydroxy-isscorop/24.6-cycloheptationene1112.7330.862Hydroxy-isscorop/24.6-cycloheptationene1213.0000.07Cycloherabecane1313.2830.842-Cycloherabecane-Ione, 4-hydroxy-3.5.6-trimethyl-4-3-oxo-1-butenyl-1413.4330.842-Cycloherabecane-Ione, 4-hydroxy-3.5.6-trimethyl-4-3-oxo-1-butenyl-1513.7110.67Oleic acid1613.9700.15Keophyladiene1714.0930.25E-2-Terradecen-1-ol1814.1610.25E-2-Terradecen-1-ol1914.4580.20Scomenthy-4-Hydroxybhorydodecane-1.4-dione2115.2640.16Scomenthy-4-Hydroxybhorydodecane-1.4-dione2215.2540.16Scomenthy-4-1-methyletylhoydodecane-1.4-dione2315.4630.17A-dxarolecadio acid2415.4900.20Berzeneycogonic acid, 3-5-dimethoy-12515.9960.37A-barolecadio-acid2616.2700.17Methyl lineatecadienoate2715.9400.12cadecanic-acid2816.3710.14Methyl casalecadienoate2915.940 <td>4</td> <td>11.354</td> <td>0.36</td> <td>.betaMethylglucoside</td>	4	11.354	0.36	.betaMethylglucoside
6     11.924     0.14     2H-Indexol Jacylatc       7     12.11     0.87     Tendaccyl acylatc       8     12.367     0.12     Methosyeagenol       9     12.333     0.08     2-Hydroxy-i-tograpyl-2/A6-cycloheptatienone       10     12.733     0.25     Methosyeagenol       11     12.856     1.44     Conferol       12.813     0.26     C-Hydroxy-3-methoxyphenyll acetat       13     13.283     0.26     C-Holoide       14     13.453     0.44     2-Cyclohexen-toore, 4-Hydroxy-3.56-trimethyl-4-(3-oxo-t-butenyl)-       15     13.711     0.67     Cyclohexen-toore, 4-Hydroxy-3.56-trimethyl-4-(3-oxo-t-butenyl)-       16     13.870     0.15     Neophytadiene     1       17     14.083     0.26     Capylone     2       18     14.164     0.25     Enethosy-4-Hydroxy-4.56-trimethyl-4-(4-oxo-t-butenyl)-       19     14.586     0.20     35 Dimethyl-2-Hydroxyhaphenthylamine       20     15.595     0.11     2.4-Trimethyla-1-Motoxyhaphenthylaxyhadexan=1.4-dione       21	5	11.750	9.29	D-Quinic acid
7121110.87Tetradecyl acylate812.370.12Methospeugenol912.5330.2824bytoxy-5-koproph/2,46-cycloheptatienone912.5330.25Methyl (4bytoxy-3-methosyphenyl) acetat1112.8561.44Conferol1213.2000.07Cyclopentadecane1313.2030.26(-1-clololde1413.4530.842-Cyclohezen-1-one, 4-hydroxy-3.5,6-trimethyl-4(3-oxo-1-butenyl)-1513.7110.67Oleic acid1613.9700.15Neophytadlene1714.0930.05Capolyton1814.1640.25E-2-Teitradecen-1-ol1914.4580.203.5/Dimethoy-4-hydroxy-bachytohexen-1.4-dione2015.0205.44Pertadecancia caid2115.0200.162-9-Dimethyl-2-(1-methylethylkyclobecane-1.4-dione2215.2540.185.9Dimethyl-2-(1-methylethylkyclobecane-1.4-dione2315.4050.112-4-Dixabouth/Scyclobes-2-enone2415.4090.20Barneenpropancia caid, 2.5-dimethoxy-2515.9560.374-Doazolecanolic acid, 4.5-dihydro-2-phenyl, 1-methylethyl ester2616.2700.17Methyl laneate2716.3170.14Methyl aleate2816.4910.21Methyl deale3916.9191.43Octadecanolic acid3118.5270.42Methyl deale3218.5190.52M	6	11.924	0.14	2H-Indeno[1,2-b]furan-2-one, 3,3a,4,5,6,7,8,8b-octahydro-8,8-dimethyl
812.870.12Methosyseygenol912.530.0824/pdroxy-5-isopropyl-2A/scycloheptatienone1012.7330.25Methyl-(4-hydroxy-3-methosyphenyl) aceta1112.8661.44Coniferoil1213.2000.07Cyclopentadecane1313.2830.26Cy-clopentadecane1413.4530.642.Cyclohexen-1-one, 4-hydroxy-3.56-trimethyl-4-3 oxor-1-butenyly-1513.7110.67Oleic acid1613.9700.15Rephyladene1714.0640.25E-2-Terdocen-1-ol1814.1640.25E-2-Terdocen-1-ol1914.580.203.5-Dimethosy-4-hydroxyphenethylamine2014.5960.27Methyl paimitate2115.025.04Pentadecanoic acid2215.4050.112.44-Trimethyl-3-(3-oxoburyl)cyclohex-2-enone2315.9060.374-Dazorearopanoic acid, 2.5-dimethosy-2415.4050.112.44-Trimethyl-3-(3-oxoburyl)cyclohex-2-enone2515.9060.374-Dazorearopanoic acid, 4.5-dimethosy-2616.700.14Methyl oleate2716.3170.14Methyl oleate2816.7201.79Unolete acid2916.7200.12Gadecanoic acid2018.970.42Methyl hexadecaniencate2118.570.42Methyl torahydronol2218.970.12Gadecanoic acid23	7	12.111	0.87	Tetradecyl acrylate
9     12533     0.08     2+lydroxy-3-septopyl-2/A5-cycloheptatrienone       10     12,233     0.25     Methyl-(A1-hydroxy-3-methoxyphenyl) actat       11     12,2856     1.44     Conferol       13     0.300     0.07     Cyclopentadcane       13     13,283     0.26     (-)-Loliolide       14     13,433     0.84     2/Cyclohexen-1-one, 4-hydroxy-3,5,6-trimethyl-4/G-oxo-1-butenyl)-       15     13,711     0.67     Oleic acid       14     14,53     0.51     Nephyladiene       17     14,093     0.05     Capylone       18     14,164     0.25     E-2-Tetriadecen-1-ol       19     14,586     0.20     E-2-Tetriadecen-1-ol       14     1502     5.04     Pertradecanola acid       21     15,254     0.18     S-9-Dimethyla-2-(Insethylethylicyclohexe-2-enone       23     15,495     0.17     Methyl planitate       24     15,490     0.20     Erezerenzpanoic acid, 2.5-dimethoxy-       25     16,276     0.17     Methyl histata	8	12.367	0.12	Methoxyeugenol
10     12.733     0.25     Methyl-(4-hydroxy-3-methoxyphenyl) acetat       11     12.856     1.44     Conflerol       12     13.200     0.07     Cyclopentadecane       13     13.233     0.26     (-1-kiolide)       14     13.453     0.84     2-Cyclohexen-1-one, 4-hydroxy-3.5.6-trimethyl-4-G-oxo-1-butenyl-       15     13.711     0.67     Oleic acid       16     13.970     0.15     Neophyradiene       17     14.083     0.05     Capylone       18     14.164     0.25     E-2-Tetradecen-1-ol       19     14.458     0.20     Methyl palmiate       20     14.986     0.27     Methyl palmiate       21     15.020     5.44     Pentadecanoic acid       22     15.455     0.11     2.44-Trimethyl-4-G-oxochyl-Cyclodecane-1.4-dione       23     15.495     0.37     4-Oazolecaboxylic acid, 4.5-dihydrox-2-phenyl, 1-methylethyl ester       24     15.996     0.37     4-Oazolecaboxylic acid, 2.5-dimethoxy-       25     15.926     0.37     4-Oazolecaboxylic ac	9	12.533	0.08	2-Hydroxy-5-isopropyl-2,4,6-cycloheptatrienone
11     12.856     1.44     Conferol       12     13.00     0.07     Cyclopertadecane       13     13.203     0.26     (-)-toliolide       13     13.453     0.26     (-)-toliolide       14     13.453     0.67     Oleic acid       15     13.711     0.67     Oleic acid       16     13.970     0.15     Neophytadene       17     14.983     0.05     Capylone       18     14.164     0.20     Splinethoxy4-hydroxyfienethylamine       20     14.596     0.27     Methyl palmitate       21     15.202     5.04     Pentadecanoic acid       22     15.254     0.18     Sy-Dimethyl-2(-)-methylethyltcylodecane-1,4-dione       24     15.490     0.20     Benzenepropanic acid, 2,5-dimethoxy-1       25     15.956     0.37     4-Xazolecathoxylic acid, 4,5-dhydro-2,phenyl, 1-methylethyl ester       26     16.270     0.14     Methyl lonleate       27     16.317     0.14     Methyl lonleate       28     16.456	10	12.733	0.25	Methyl-(4-hydroxy-3-methoxyphenyl) acetat
12     13.200     0.07     Cyclopentadecane       13     13.283     0.26     (-)Loliolide       14     13.453     0.84     2.Cyclobexen-1one, 4-hydroxy-3.5,6 trimethyl-4(3-oxo-1-butenyl).       15     13.711     0.67     Oleic acid       16     13.970     0.15     Neophytadiene       17     14.093     0.05     Capylone       18     14.164     0.25     E-2-Tettadecen-1-ol       19     14.368     0.20     2.5-Direthyl-2-(1-methylethylarnine       20     14.560     0.27     Methyl palmitate       21     15.020     5.04     Pettadecanoic acid       22     15.405     0.11     2.4/4-Tirmethyl-3-(1-methylethyllcyclodecane-1,4-dione       23     15.405     0.11     2.4/4-Tirmethyl-3-30-xobut/byclobec2-enone       24     15.490     0.20     Benzeneptopanic acid, 2.5-dimethox-       25     15.906     0.77     Methyl linoleate       26     16.270     0.17     Methyl lokate       27     16.317     0.14     Methyl tetanydroinola	11	12.856	1.44	Coniferol
13     13.283     0.26     (-)-Loliolide       14     13.453     0.84     2.Cyclohexen-1-one, 4-hydroxy-3.5.6-trimethyl-4-G-oxo-1-butenyl-       15     13.711     0.67     Oleic acid       15     13.970     0.15     Neophytadiene       16     13.970     0.05     Capylone       17     14.093     0.05     Capylone       18     14.164     0.25     E-2-Tetradecen-1-ol       19     14.58     0.20     3.5-Dimethoxy-4-hydroxybenethylamine       20     15.020     0.27     Methyl palmitate       21     15.020     0.10     2.4-Frimethyl-3-G-oxobutylcyclodecane-1,4-dione       22     15.020     0.11     2.4-Frimethyl-3-G-oxobutylcyclodecane-1,4-dione       23     15.405     0.11     2.4-Frimethyl-3-G-oxobutylcyclodecane-1,4-dione       24     15.490     0.20     Benzenepropanoic acid, 2,5-dimethoxy-       25     15.940     0.17     Methyl Incleate       26     16.270     0.17     Methyl Incleate       27     16.317     0.14     Catadecanoic acid<	12	13.200	0.07	Cyclopentadecane
14     13453     0.84     2-Cyclohexen-1-one, 4-hydroxy-3,5,6-trimethyl-4-(3-oxo-1-butenyl)-       15     13,711     0.67     Oleic acid       16     13,970     0.15     Nephytadiene       17     14,093     0.05     Capylone       18     11,164     0.25     2-21etradecen1-ol       19     14,458     0.20     3,5-Dimethoxy-4-hydroxyphenethylamine       20     14,596     0.27     Methyl palmitate       21     15,200     5,04     Pertadecanolic acid       22     15,254     0.18     5,9-Dimethyl-2-(1-methylethylloyclodecane-1,4-dione       23     15,405     0.11     2,4-Trimethyl-3-G-oxobutylloyclobex-2-enone       24     15,400     0.20     Benzenepropanoic acid, 2,5-dimethoxy-       25     15,996     0.37     4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester       26     16,270     0.17     Methyl Inoleate       27     16,317     0.14     Methyl Inoleate       28     16,276     17.09     Linoleic acid       31     18,572     0.42 <td>13</td> <td>13.283</td> <td>0.26</td> <td>(–)-Loliolide</td>	13	13.283	0.26	(–)-Loliolide
15137110.67Olcic add1613.9700.15Neophytadiene1714.0930.05Caprylone1814.1640.25E.2-Tetradecen-1-ol1914.4580.203.5-Dimethoxy-4-hydroxyphenethylamine2014.5960.27Methyl palmitate2115.0205.04Pentadecanoic acid2215.2540.185.9-Dimethyl-2-(1-methylethyl)cyclodecane-1,4-dione2315.4050.112.4/4-Trimethyl-3-G-oxobutyl)tyclohex-2-enone2415.4900.20Benzenepropanoic acid, 2,5-dihydro-2-phenyl, 1-methylethyl ester2515.9960.374-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl, 1-methylethyl ester2616.2700.14Methyl linoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3118.5270.42Methyl haxalecadienoate3218.9780.62Arachidic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadeceanal352.01330.13Methyl Inoleate (AN)362.8800.11Ethyl Inoleate (AN)372.8800.62Arachidic acid3818.9780.62Arachidic acid392.8910.50cis-9-Hexadeceanal3118.9780.50cis-9-Hexadeceanal322.8800.51Ci	14	13.453	0.84	2-Cyclohexen-1-one, 4-hydroxy-3,5,6-trimethyl-4-(3-oxo-1-butenyl)-
16     13,970     0.15     Neophytadiene       17     14,093     0.05     Caprylone       18     14,164     0.25     E-2-Tetradecen-1-ol       19     14,596     0.20     3,5-Dimethoxy-4-hydroxyphenthylamine       20     14,596     0.27     Methyl palmitate       21     15,020     5,04     Pentadecanoic acid       22     15,254     0.18     5,9-Dimethyl-2(1-methylethyllcyclodecane-1,4-dione       23     15,405     0.11     2,44-Trimethyl-3-(3-oxobutylcyclohex-2-enone       24     15,490     0.20     Benzenepropancic acid, 2,5-dimethoxy-       25     15,996     0.37     4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-1-methylethyl ester       26     16,270     0.17     Methyl Inoleate       27     16,317     0.14     Methyl Inoleate       28     16,456     1.21     Phytol       29     16,726     17.09     Linoleic acid       31     18,527     0.42     Methyl hexadecalienoate       32     18,978     0.62     Arachidic acid	15	13.711	0.67	Oleic acid
17     14.093     0.05     Capylone       18     14.164     0.25     E-2-Tetradecen-1-ol       19     14.458     0.20     3.5-Dimethoxy-4-hydroxyphenethylamine       20     14.596     0.27     Methyl palmitate       21     15.020     5.04     Pentadecanoic acid       22     15.254     0.18     5.9-Dimethyl-2-(1-methylethylkyc)dolecxa-e-1/4-dione       23     15.405     0.11     2.4-Trimethyl-3-(3-oxobuth/byc)dolex-2-enone       24     15.490     0.20     Benzenepropanoic acid, 2.5-dimethoxy-       25     15.996     0.37     4-Oxazolecarboxylic acid, 4.5-dihydro-2-phenyl-, 1-methylethyl ester       26     16.270     0.17     Methyl linoleate       27     16.317     0.14     Methyl oteate       28     16.456     1.21     Phytol       29     16.726     17.09     Linoleic acid       30     18.978     0.62     Arachidic acid       31     18.527     0.42     Methyl thexadecadienoate       32     18.718     0.11     1-Eicosenoic acid	16	13.970	0.15	Neophytadiene
1814.1640.25E-2-Tetradecen-1-011914.4580.203,5-Dimethoxy-4-hydroxyphenethylamine2014.5960.27Methyl palmitate2115.0205.04Pentadecanoic acid2215.2540.185,9-Dimethyl-2-(1-methylethyltyc)clobecane-1,4-dione2315.4050.112,44-Trimethyl-3-(3-oxobutyl)cyclobex-2-enone2415.4900.20Benzenepropanoic acid, 2,5-dimethoxy-2515.9960.374-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester2616.2700.17Methyl linoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.9191.43Octadecanoic acid3016.9191.43Octadecanoic acid3118.9780.42Methyl hexadecalienoate3218.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3521.6180.50cis-Vaccenic acid3621.6180.50cis-Vaccenic acid372.8000.11Ethyl linoleate (AN)382.8070.991.1*Biphenyl, 2-formyl-4/5/5-trimethory-392.6180.50cis-Vaccenic acid402.56310.105+Hydroxymethyl-1,14-trimethyl-6-methylenedecahydronaphthalen-2-ol412.8890.56Octadecanal422.65670.442.8-Oimethyl-2-(4.8.12-trimethyl-6-chromanol432.6915 </td <td>17</td> <td>14.093</td> <td>0.05</td> <td>Caprylone</td>	17	14.093	0.05	Caprylone
19144580.203.5-Dimethoxy-4-hydroxyphenethylamine2014.5960.27Methyl palmitate2115.0205.04Pentadecanoic acid2215.2540.18S.9-Dimethyl-2-(1-methylethyl)cyclodecane-1.4-dione2315.4050.112.4.4-Trimethyl-3-(3-coxbutyl)cyclohex-2-enone2415.4050.20Benzenepropanoic acid, 2.5-dimethoxy-2515.9960.374-Oxazolecarboxylic acid, 4.5-dihydro-2-phenyl-, 1-methylethyl ester2616.2700.17Methyl inoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3118.5270.42Methyl hexadecadienoate3218.9180.62Arachidic acid3419.2790.12cis-9-Hexadecenal352.1330.50cis-Vacenic acid362.8600.11Ethyl linoleate (AN)382.8070.091,1-Biphenyl, 2-formyl-47.5f-trimethoxy-392.6612.931-Heptacosanol412.8890.60Ctadecanal422.6570.412.8-Dirmethyl-2-(4.8,12-trimethyl-6-methylenedecahydronaphthalen-2-ol432.6510.442.8-Dirmethyl-2-(4.8,12-trimethyl-6-chromanol	18	14.164	0.25	E-2-Tetradecen-1-ol
20     14596     0.27     Methyl palmitate       21     15.020     5.04     Pentadecanoic acid       22     15.254     0.18     5.9-Dimethyl-2-(1-methylethyl)cyclodecane-1,4-dione       23     15.405     0.11     2.4.4-Trimethyl-3-(3-oxobutyl)cyclobex-2-enone       24     15.409     0.20     Benzenepropanoic acid, 2,5-dimethoxy-       25     15.996     0.37     4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester       26     16.270     0.14     Methyl loleate       27     16.317     0.14     Methyl exate       28     16.456     1.21     Phytol       29     16.726     17.09     Linoleic acid       31     18.527     0.42     Methyl thexadecadienoate       32     18.718     0.11     11-Eicosenoic acid       34     19.279     0.12     cis-9-Hexadecenal       35     2.0133     0.13     Methyl tethydroionol       36     2.3807     0.09     1,1'Biphenyl, 2-formyl-4/5/6'trimethoxy-       37     2.3680     0.10     Ethyl linole	19	14.458	0.20	3,5-Dimethoxy-4-hydroxyphenethylamine
1     15.020     5.04     Pentadecanoic acid       22     15.254     0.18     5.9-Dimethyl-2(1-methylethyl)cyclodecane-1,4-dione       23     15.405     0.11     2,4,4-Trimethyl-3-(3-oxobutyl)cyclohex-2-enone       24     15.490     0.20     Benzenepropanoic acid, 2,5-dimethoxy-       25     15.996     0.37     4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester       26     16.270     0.17     Methyl linoleate       27     16.317     0.14     Methyl oleat       28     16.456     1.21     Phytol       29     16.726     17.09     Citadecanoic acid       31     18.527     0.42     Methyl hexadecadienoate       32     18.78     0.11     1-Eicosenoic acid       34     19.279     0.12     cis-9-Hexadecanal       35     2.133     0.13     Methyl tertahydroionol       36     1.2     cis-9-Hexadecanal     cis-9-Gazonic acid       37     2.860     0.11     Ehyl linoleate (JAN)       38     2.937     0.09     1/1-Biphenyl, 2-for	20	14.596	0.27	Methyl palmitate
2215.2540.185.9-Dimethyl-2-(1-methylethyl)cyclodecane-1,4-dione2315.4050.112,4,4-Trimethyl-3-(3-oxobutyl)cyclohex-2-enone2415.4900.20Benzenepropanoic acid, 2,5-dimethoxy-2515.9960.374-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester2616.2700.17Methyl linoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl ternhydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091,1'Biphenyl, 2-formyl-4'5'6-trimethoxy-3924.0612.931-Heptacosanol4025.6310.105-Hydroxymethyl-1,1.4-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.890.56Octadecanal4226.5670.4128-Dimethyl-2'(48.12-trimethyltridecyl)-6-chromanol447.3720.4926.67 frimethylcyclohex-1-enylmethanesulfonylblenzene	21	15.020	5.04	Pentadecanoic acid
2315.4050.112.4.4-Trimethyl-3-(3-oxobutyl)cyclohex-2-enone2415.4900.20Benzenepropanoic acid, 2,5-dimethoxy-2515.9960.374-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester2616.2700.17Methyl linoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3419.2790.12cis-9-Hexadecenal3520.1330.50cis-Vaccenic acid3621.6180.50cis-Vaccenic acid372.8600.11Ethyl linoleate (JAN)382.8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-392.6610.105-Hydroxymethyl-1,1,4-trimethyl-6-methylenedecahydronaphthalen-2-ol412.8890.56Octadecanal422.6570.412&-Dimethyl-2(4,8,12-trimethyltridecyl)-6-chromanol432.9150.498.404442.3720.492.66.Trimethylcyclohex-1-enylmethansulfonylbenzene	22	15.254	0.18	5,9-Dimethyl-2-(1-methylethyl)cyclodecane-1,4-dione
24     15.490     0.20     Benzenepropanoic acid, 2,5-dimethoxy-       25     15.996     0.37     4-0xazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester       26     16.270     0.17     Methyl linoleate       27     16.317     0.14     Methyl oleate       28     16.456     1.21     Phytol       29     16.726     17.09     Linoleic acid       30     16.919     1.43     Octadecanoic acid       31     18.527     0.42     Methyl hexadecadienoate       32     18.718     0.11     11-Eicosenoic acid       34     19.279     0.12     cis-9-Hexadecenal       35     20.133     0.13     Methyl tetrahydroionol       36     21.618     0.50     cis-Vaccenic acid       37     2.680     0.11     Ethyl linoleate (JAN)       38     2.807     0.99     1.1'Biphenyl 2-formyl-4',5',6'-trimethyl-9-methylendecahydronaphthalen-2-of       40     2.657     0.10     5-Hydroxymethyl-1,1,4-trimethyl-6-methylendecahydronaphthalen-2-of       41     2.589     0.56 <td>23</td> <td>15.405</td> <td>0.11</td> <td>2,4,4-Trimethyl-3-(3-oxobutyl)cyclohex-2-enone</td>	23	15.405	0.11	2,4,4-Trimethyl-3-(3-oxobutyl)cyclohex-2-enone
2515.9960.374-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester2616.2700.17Methyl linoleate2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3521.6180.50cis-Vaccenic acid3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.991.1'-Biphenyl, 2-formyl-4',5',5'-trimethoxy-3924.0612.931-Heptacosanol4125.8390.56Octadecanal426.5670.4128-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol4326.9150.44Butanedioic acid, di-9-dodecyn-1-yl ester442.73720.492,66-Trimethylcyclohex-1-enylmethanesulfonylbenzene	24	15.490	0.20	Benzenepropanoic acid, 2,5-dimethoxy-
26162700.17Methyl linoleate27163170.14Methyl oleate28164561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3926.6310.10S-Hydroxymethyl-1,1,4-a-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.8890.56Octadecanal426.5670.4128-Dimethyl-2-(48,12-trimethyltridecyl)-6-chromanol4427.3720.492,66-Trimethylcyclohex-1-enylmethanesulfonylbenzene	25	15.996	0.37	4-Oxazolecarboxylic acid, 4,5-dihydro-2-phenyl-, 1-methylethyl ester
2716.3170.14Methyl oleate2816.4561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl hetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091.1'Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924.0612.931-Heptacosanol4025.6310.105-Hydroxymethyl-1,1,4-a-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.8890.56Octadecanal426.5670.412.8-Dimethyl-2-(48,12-trimethyltridecyl)-6-chromanol432.9150.44Butanedioic acid, di-9-dodecyn-1-yl ester447.3720.492,66-Trimethylcyclohex-1-enylmethanesulfonylbenzene	26	16.270	0.17	Methyl linoleate
28164561.21Phytol2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.070.931.1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-392.6310.105-Hydroxanol402.6310.56Octadecanal412.5890.56Octadecanal426.5670.412,8-Dimethyl-2-(4,8,12-trimethyl-fi-chromanol432.9.3720.492,6-Frimethylcyclohex-1-enylmethanesulfonyl)benzene	27	16.317	0.14	Methyl oleate
2916.72617.09Linoleic acid3016.9191.43Octadecanoic acid3118.5270.42Methyl hexadecadienoate3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroinol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.991.1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924.0612.931-Heptacosanol4125.8890.56Octadecanal426.5670.412,8-Dimethyl-2-(4,8,12-trimethyltidecyl)-6-chromanol4326.9150.44Butanedioic acid, di-9-dodecyn-1-yl ester4427.3720.492,6-Frimethylcyclohex-1-enylmethanesulfonyl)benzene	28	16.456	1.21	Phytol
3016,9191.43Octadecanoic acid3118,5270.42Methyl hexadecadienoate3218,7180.1111-Eicosenoic acid3318,9780.62Arachidic acid3419,2790.12cis-9-Hexadecenal3520,1330.13Methyl tetrahydroionol3621,6180.50cis-Vaccenic acid3723,6800.11Ethyl linoleate (JAN)3823,8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924,0612,931-Heptacosanol4025,6310.105-Hydroxymethyl-1,1,4-trimethyl-6-methylenedecahydronaphthalen-2-ol4125,8990,56Octadecanal4226,6770,412,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol4326,9150,44Butanedioic acid, di-9-dodecyn-1-yl ester4427,3720,492,6-Trimethylcolohex-1-enylmethanesulfonyl)benzene	29	16.726	17.09	Linoleic acid
31   18.527   0.42   Methyl hexadecadienoate     32   18.718   0.11   11-Eicosenoic acid     33   18.978   0.62   Arachidic acid     34   19.279   0.12   cis-9-Hexadecenal     35   20.133   0.13   Methyl tetrahydroionol     36   21.618   0.50   cis-Vaccenic acid     37   23.680   0.11   Ethyl linoleate (JAN)     38   23.807   0.09   1,1'-Biphenyl,2-formyl-4',5',6'-trimethoxy-     39   24.061   2.93   1-Heptacosanol     41   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     42   26.567   0.41   2,8-Dimethyl-2-(48,12-trimethyltridecyl)-6-chromanol     43   26,915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethylcyclohex-1-enylmethanesulfonylbenzene	30	16.919	1.43	Octadecanoic acid
3218.7180.1111-Eicosenoic acid3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924.0612.931-Heptacosanol4025.6310.105-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.8890.56Octadecanal4226.5670.412,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol4326.9150.49Butanedioic acid, di-9-dodecyn-1-yl ester4427.3720.492,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	31	18.527	0.42	Methyl hexadecadienoate
3318.9780.62Arachidic acid3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924.0612.931-Heptacosanol4025.6310.105-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.8890.56Octadecanal4226.5670.412,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol4326.9150.49Butanedioic acid, di-9-dodecyn-1-yl ester4427.3720.492,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	32	18.718	0.11	11-Eicosenoic acid
3419.2790.12cis-9-Hexadecenal3520.1330.13Methyl tetrahydroionol3621.6180.50cis-Vaccenic acid3723.6800.11Ethyl linoleate (JAN)3823.8070.091,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-3924.0612.931-Heptacosanol4025.6310.105-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol4125.8890.56Octadecanal426.5670.412,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol4326.9150.498utanedioic acid, di-9-dodecyn-1-yl ester4427.3720.492,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	33	18.978	0.62	Arachidic acid
35   20.133   0.13   Methyl tetrahydroionol     36   21.618   0.50   cis-Vaccenic acid     37   23.680   0.11   Ethyl linoleate (JAN)     38   23.807   0.09   1,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-     39   24.061   2.93   1-Heptacosanol     40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6-6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	34	19.279	0.12	cis-9-Hexadecenal
36   21.618   0.50   cis-Vaccenic acid     37   23.680   0.11   Ethyl linoleate (JAN)     38   23.807   0.09   1,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-     39   24.061   2.93   1-Heptacosanol     40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	35	20.133	0.13	Methyl tetrahydroionol
37   23.680   0.11   Ethyl linoleate (JAN)     38   23.807   0.09   1,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-     39   24.061   2.93   1-Heptacosanol     40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6-6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	36	21.618	0.50	cis-Vaccenic acid
38   23.807   0.09   1,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-     39   24.061   2.93   1-Heptacosanol     40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.49   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	37	23.680	0.11	Ethyl linoleate (JAN)
39   24.061   2.93   1-Heptacosanol     40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyl/tridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	38	23.807	0.09	1,1'-Biphenyl, 2-formyl-4',5',6'-trimethoxy-
40   25.631   0.10   5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol     41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,66-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	39	24.061	2.93	1-Heptacosanol
41   25.889   0.56   Octadecanal     42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	40	25.631	0.10	5-Hydroxymethyl-1,1,4a-trimethyl-6-methylenedecahydronaphthalen-2-ol
42   26.567   0.41   2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol     43   26.915   0.44   Butanedioic acid, di-9-dodecyn-1-yl ester     44   27.372   0.49   2,6,6-Trimethyltrylcyclohex-1-enylmethanesulfonyl)benzene	41	25.889	0.56	Octadecanal
43 26.915 0.44 Butanedioic acid, di-9-dodecyn-1-yl ester   44 27.372 0.49 2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	42	26.567	0.41	2,8-Dimethyl-2-(4,8,12-trimethyltridecyl)-6-chromanol
44 27.372 0.49 2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene	43	26.915	0.44	Butanedioic acid, di-9-dodecyn-1-yl ester
	44	27.372	0.49	2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene

Table 2 GC-MS analysis of Quercus leucotrichophora (Leaves) extract

volume of the extracts used for the analysis of antibacterial activity were 5 mg/ml and 20  $\mu$ l (extract soaked by each disc), respectively. The antibacterial activity was assessed by measuring the zone of inhibition surrounding the disks and each experiment was carried out in triplicate. In the present study, DMSO (10%) and ampicillin (1 mg/ml) were used as negative and positive controls, respectively.

# **Results and discussion**

Bacterial strains

Table 3 Antibacterial profile of QL extracts

QLB (ZOI)

OLB (Ave ± SD) mm

This study focused on the chemical composition and antibacterial screening of QL extracts. The yield of bark and leaves extracts were found to be 9.7% and 13.6%, respectively. A range of volatile phytoconstituents have been identified by GC-MS in different Quercus species other than QL [20, 21]. In the present study, the

percentages (area per cent) and the retention time (RT) of the components are listed in Tables 1 and 2. In leaves extract of QL, 62 components were identified, representing 94.54% of the total plant extract, in which Linoleic acid (17.09%), Simiarene (15.29%), and Flavone 4'-OH,5-OH,7-di-O-glucoside (15.26%) were the major components, however, in bark extract of QL, 23 components were identified, representing 91.91% of the total plant extract, in which Linoleic acid (19.77%), Lupeol (17.91%), Epi-psi-Taraxastanonol (14.20%), and cis-Vaccenic acid (13.00%) were the major compounds. Linoleic acid is an omega-6-fatty acid and is enormously used in cosmetic industries, whereas the conjugated linoleic acid was accounted to have anticarcinogenic, fat reducing, antiatherogenic and immune enhancing activity [22]. Lupeol is a triterpenoid which possess

Ampi (Ave ± SD) mm

Table 2 GC-MS analysis of Quercus leucotrichophora (Leaves) extr	act (Continued)
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Area percent

E. coli	9.37 ± 0.65	22.7 ± 0.65	8.53 ± 0.50	21.5 ± 0.62	0.00
P. aeruginosa	13.97 ± 0.42	23.2 ± 0.74	13.27 ± 0.25	23.3 ± 0.70	0.00
S. aureus	16.97 ± 0.25	22.6 ± 0.62	13.80 ± 1.51	20.7 ± 0.56	0.00
S. pyogenes	15.97 ± 0.65	21.2 ± 0.46	15.83 ± 0.29	22.4 ± 0.52	0.00
B. subtilis	19.07 ± 0.31	22.2 ± 0.51	17.03 ± 0.55	20.6 ± 0.57	0.00

QLL (ZOI)

OLL (Ave ± SD) mm

Note: QLB Quercus leucotrichophora bark, QLL Quercus leucotrichophora Leaves, ZOI Zone of inhibition, Ampi Ampicillin, DMSO Dimethyl sulfoxide

Ampi (Ave ± SD) mm

Retention time

SN

		1	
45	27.673	0.45	Tocopherol
46	28.406	0.56	3-Hydroxycholest-4-en-6-one
47	29.332	0.47	.betaTocopherol
48	29.583	0.24	.gammaTocopherol
49	30.461	1.17	Baccharane
50	31.294	2.78	Vitamin E
51	32.319	1.25	(+)-γ-Tocopherol, O-methyl-
52	33.809	0.47	1H-Indole
53	36.231	15.29	Simiarene
54	36.396	0.74	Clionasterol
55	36.793	0.82	Verticiol
56	37.508	0.49	.betaAmyrin
57	37.714	0.45	Methyl ursolate
58	39.351	0.67	D:C-Friedo-B':A'-neogammacer-9(11)-en-3-one
59	40.473	0.62	9,19-Cyclolanost-23-en-3-ol, 25-methoxy-, acetate, (3.beta.,23e)-
60	40.807	0.16	-Heptadecyloxirane
61	41.130	3.81	Stigmast-4-en-3-one
62	44.451	15.26	Flavone 4'-OH,5-OH,7-di-o-glucoside
Total		94.54	
Unidentified		1.10	

Name of compounds

DMSO (ZOI)

anticancer and anti-inflammatory activities [23]. Flavone 4'-OH,5-OH,7-di-O-glucoside is a isoflavonoid and possess antioxidant activity [24]. Cis-vaccenic acid is a omega-7 fatty acid is known for its antibacterial activity and hypolipidemic effect in rats [24]. Epi-psi-Taraxastanonol is a terpenoid and is known for its therapeutic activity against cardiovascular diseases [25]. A total of seven components were found to be the common for both extracts of QL. Previous studies on Quercus genus suggested that the species are rich in monounsaturated fatty acids, mostly oleic acid and also essential fatty acids such as linoleic ( $\omega$ -6) and linolenic ( $\omega$ -3) fatty acids, sesquiterpenes, terpenoids, flavonoids and phenolic acid [20, 21, 26] and in the present study same pattern of phytoconstituents were observed in the leaves and bark extracts of QL. Differences in quantity and quality of chemical components of any plant extract are highly influenced by several genetic and environmental factors, such as the genetic and seasonal variation, geographical origin, and the part of the plant used for the study, even agronomic conditions, developmental stage, time of collection, extraction method and solvent system [27].

The quantification of antibacterial activity for methanolic extracts of QL has been evaluated against five bacterial species by means of the agar disk diffusion method. The results of antibacterial activity of QL extracts are expressed as the diameter of the inhibition zone in millimetre (shown in Table 3). OLB and OLL extracts showed zone of inhibition (ZOI) from a range of  $9.37 \pm 0.65$  to  $19.07 \pm 0.31$  mm and  $8.53 \pm 0.50$  to  $17.03 \pm 0.55$  mm, respectively. Both the extracts showed the maximum and minimum zone of inhibition (ZOI) against B. subtilis and E. coli, respectively. Ampicillin showed ZOI from a range of  $21.2 \pm 0.46$  to  $23.3 \pm$ 0.70 mm for all the bacterial strains, and DMSO was used as a negative control, which showed no zone of inhibition. Previously, the antimicrobial profile of the volatile extract of QLB was recorded against three microbial cultures, namely; Streptococcus pyogenes, Streptococcus aureus, and Escherichia coli. The volatile extract of QLB exhibited a potential antimicrobial activity against Streptococcus pyogenes, compared to Streptococcus aureus, and Escherichia coli [14]. The antibacterial activity of the fatty acid methyl ester (FAME) extract of QL fruits was recorded against four bacterial stains namely; Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa and Escherichia coli from a range of 7.8 to 15.9 mm [15]. The extract of FAME showed dissimilar activity against different bacterial strains due to the chemical nature, antimicrobial agents, and their mode of action on different microorganism [28]. In the present study, both the extracts of QL demonstrated better antibacterial activity compared to previous studies.

# Conclusion

The GC-MS analysis of methanolic extract of bark and leaves of QL revealed the presence of highly composite profiles of medicinally important bioactive components. This study also revealed the antibacterial activity of QLB and QLL against pathogenic microbes. Therefore, it can be concluded that the methanolic leaf and bark extracts of QL have shown the presence of active compounds having pharmacological and industrial importance.

#### Abbreviations

Ampi: Ampicillin; *B. subtilis: Bacillus subtilis*; DMSO: Dimethyl sulfoxide; *E. coli: Escherichia coli*; GC-MS: Gas Chromatography- Mass spectrometry; *P.aeruginosa: Pseudomonas aeruginosa*; QL: *Quercus leucotrichophora*; QLB: *Quercus leucotrichophora* bark; QLL: *Quercus leucotrichophora* leaves; RT: Retention time; *S. aureus: Staphylococcus aureus*; *S. pyogenes: Streptococcus pyogenes*; ZOI: Zone of inhibition

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#### Availability of data and materials

All data generated or analysed during this study are included in this article.

#### Authors' contributions

PS, SP and HB reviewed the literature, collected the samples, performed all the experiments, and drafting the manuscript with RKB. All authors read and approved the final manuscript.

### Ethics approval and consent to participate

Not applicable.

#### Consent for publication

Not applicable.

#### **Competing interests**

The authors declare that they have no competing interests.

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