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GC-MS analysis of ethanolic extract of *Ehretia laevis* Roxb

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Abstract
Ehretia laevis Roxb is one of such plants which being used in Indian traditional medicine for the treatment of liver ailments, belonging to the family Boraginaceous. Plant is a rich source of phytochemicals. These products are commonly termed "secondary metabolites" in contrast to the "primary metabolites" which are essential for plant growth and development. The aim of this study was to carry out to identify the Phytoconstituentspresent in the Ethanolic Extract of *Ehretia laevis* Roxb by Gas chromatography and Mass spectroscopy (GC-MS).

Keywords: Boraginaceous, GC-MS, phytoconstituents, ethanolic extract, *Ehretia laevis*

Introduction
Ehretia laevis Roxb is one of such plants which being used in Indian traditional medicine for the treatment of liver ailments. *Ehretia laevis* is an Indian medicinal plant. It is a deciduous shrub. It is considered as small tree due to its 12 m height belonging to the family Boraginaceae. *Ehretia laevis* a small tree. It is generally found in Asia and Australian tropics. Literature survey revealed wide biological activity of family Boraginaceae. The inner bark of *E. laevis* used as food. Leaves are applied to ulcers and in headache. Fruit is astringent, anthelmintic, diuretic, demulcent, expectorant and used in affections of urinary passages, diseases of lungs and spleen. Powdered kernel mixed with oil is a remedy in ringworm. Seeds are anthelmintic. This medicinal plant has an irregular trunk with a light grey or whitish bark. Leaves are variable in size and shape. They vary from 2 cm to 6.3 cm in length and 1.3 cm to 3.8 cm in width. Flowers of these plants are white in colour. The calyx of these flowers are 2.5 mm long, 3-lobed and the corolla are 6-8 mm long, in which 5 corolla are lobed. The tube and lobes of corolla are longer than the calyx. The present study deals with the GC MS analysis of phytocomponents in the ethanolic extract of *Ehretia laevis* Roxb.

Materials and methods
Plant Material
Flowers of *Ehretia laevis* plants, collected during the month of February 2018 from the Ambajogai, district Beed of Maharashtra state. Authenticated from Botanical Survey of India [Authentication number: No. BSI/WRC/100-2/Tech./2018/37], Minstry of Environment, Forest and Climate Change, Western regional center, Pune (Maharashtra) India.

Extraction procedure
The plant material of *Ehretia laevis* were dried at room temperature for fifteen days and then reduced to a coarse powder. This powder was used for the preparation of hydro alcoholic extract. The plant powder was extracted with 70% ethanol (500 ml) for 12 h at 50 ^{0}C. The obtained extract was concentrated under reduced pressure on rotary evaporator at 40 ^{0}C to obtain a brownish residue. The above extract (180 g) was dissolved in 700 ml of distilled water and partitioned sequentially with n-hexane, ethyl acetate and aqueous to obtain n-hexane, ethyl acetate and aqueous fractions. All these fractions were concentrated using rotary evaporator. The yield of hydro alcoholic extract obtained by reflux method was found to be 23.25% w/w. The yield of n-hexane, ethyl acetate and aqueous fractions obtained by successive solvent-solvent extraction of hydro alcoholic extract was found to be 0.75, 9.5 and 15.35% w/w, respectively.

GC-MS Analysis
GC MS analyses of these extracts were performed using a Mass Hunter GC MS system. Total run time for each sample cycle was 24.4 min. Gas chromatograph interfaced with a mass spectrometer equipped with a fused silica capillary column, composed of 100%

Dimethylpolysiloxane. For GCMS detection an electron ionization system with ionizing energy of 70eV was used. Helium gas was used as the carrier gas used at a flow rate of 1ml/min and an injection volume of 1 µL was employed with a split ratio of 1:10 was used. Injector temperature was kept at 280^0C. Total GC running time was 38 minutes. The relative percentage of each component was calculated by comparing its average peak to the total area. The column head pressure was adjusted to1.8804 psi. The MS was operated in the ACQ mode scanning from m/z 40 to 600.0. In the full scan mode, electronionization (EI) mass spectra in the range of 40–600 (m/z) wererecorded at electron energy of 70 eV. Compounds were identified by comparing mass spectra with library of the National Institute of Standard and Technology (NIST), USA/Wiley.

Result and Discussion

GC MS analysis was carried out in ethanolic extract of *Ehretia laevis*. Forty five compounds were detected. The Forty five compounds along with their retention times and peak area are given in the table 1. From the GCMS study the following compounds that are present in larger amounts.

Table 1: GC-MS spectral analysis of ethanolic extract of *Ehretia laevis*

Peak no.	Area	Compound	Retention time
1	15324060	1,4-Benzenediol,2,5-bis(1,1-dimethylethyl)-	4.4146
2	23535803	3-methoxy-4,5-methylenedioxyphenyl-2-propanone	4.5679
3	4309587	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	4.7417
4	10437101	Malonic acid, 2-(4-ethoxyphenyl)-2-hydroxy-,diisopropyl ester	5.0276
5	18396848	Benzofuran-5,6-diol-3-one, 2-benzylidene-	5.3278
6	12276458	1,3-Benzodioxole-5-(4-keto-butyric acid)	5.3387
7	11300102	Dimethyl hydrastate	5.7849
8	3984851	2,4,6-Trihydroxybenzaldehyde	5.8445
9	94326381	o-Diacetylbenzene	6.1606
10	5958179	2H-1-Benzopyran-2-one, 4-hydroxy-7-methoxy-3-phenyl-	6.3022
11	5614448	p-Ethoxyphenyl p-(pentyloxycarbonyloxy)benzoate	6.5477
12	55911455	Propiophenone, 2,2',4',6'-tetramethyl-	6.6013
13	9740178	Terephthalic acid, tridec-2-yn-1-yl ethyl ester	6.7713
14	18850625	Glycine, N-methyl-N-methoxycarbonyl-, ethyl ester	7.3130
15	20401832	2-(3-methyl-2-cyclopenten-1-yl)-2-methylpropionaldehyde	7.4069
16	239899886	Benzofuran-5,6-diol-3-one, 2-benzylidene-	7.5955
17	10656446	7H-Benzo[c]furo[2,3-f][1]benzopyran, 2,7,7,10-tetramethyl-4-pentyl-	7.6118
18	3319082	Diphenyl isophthalate	8.1176
19	5288394	Phthalic acid, methyl phenyl ester	8.1839
20	57470594	1,2-Benzenediol, O,O'-di(propargyloxycarbonyl)-	8.2386
21	22354050	2-Phenylaminobenzoic acid, ethyl ester	8.4005
22	28810247	2-Phenylaminobenzoic acid, ethyl ester	8.4026
23	21522590	Isophthalic acid, hexadecyl propyl ester	8.4491
24	18583192	Benzene acetic acid, 2-propenyl ester	8.5300
25	5839697	4-Nitrobenzoic acid, 4-isopropylphenyl ester	8.5801
26	19369377	3-Cyclohexylidene-5-(4-pentanoyloxyphenyl)-furan-2(3H)-one	8.6177
27	200602418	Benzofuran-5,6-diol-3-one, 2-benzylidene-	8.7300
28	74022332	4-n-Propylbenzoic acid	8.8028
29	9461180	2-Oxo-2H-chromene-4-carboxylic acid	9.0121
30	13735890	2-Benzylidene-coumaran-3-one	9.1644
31	50045518	Pentatonic acid, 5-hydroxy-, 2,4-di-t-butylphenyl esters	9.1709
32	10957893	Aspirin methyl ester	9.4035
33	26238127	4-Phenyl-2,3-dihydro-1H-1,5-benzodiazepin-2-one	9.8042
34	36495855	3-Nitro-9-isopropyl-carbazole	9.9196
35	7943864	2-Butoxy-7-propoxy-fluoren-9-one	10.2986
36	21842586	Isophthalic acid, 2,7-dimethyloct-7-en-5-yn-4-yl ethyl ester	10.3720
37	17928193	Benzoic acid, 3,4-methylenedioxy-, 3-formylphenyl ester	11.5899
38	199588023	Cyclobuta[1,2-d:3,4-d']bis[1,3]dioxole,tetrahydro-,(3a.alpha.,3b.alpha.,6a.alpha.,6b.alpha.)-	15.1673
39	247012959	2,2'-Bi-1,3-dioxolane	16.0049
40	298164191	5-Acetoxymethyl-2-furaldehyde	16.3176
41	279486139	Ethane, 1,1-dimethoxy-	16.6866
42	324835463	Dimethyl methoxymalonate	17.2372
43	187822531	Anisindione	17.4267
44	190427607	2,2'-Trimethylenebis-1,3-dioxolane	17.6066
45	203661537	Leucodrin	17.9381

Conclusion

The presence of various bioactive compounds in the *Ehretia laevis* justifies the use of whole plant for various ailments by traditional practitioners. However, isolation of individual phytochemical constituents and subjecting it to the biological activity will definitely give fruitful results. From the results, *Ehretia laevis* contains various bioactive compounds. Therefore, it is recommended as a plant of phytopharmaceutical importance. From this study it can be concluded that the *Ehretia laevis* may serve as a new potential source of medicines due to the presence of these phytochemicals and bioactive compounds.

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