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GCMS, FTIR AND HRLCMS - QTOF CHARACTERIZATION STUDY OF BROWN MACROALGA (*Sargassum linearifolium*)

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ABSTRACT

Nowadays, marine macroalgae are widely involved in various applications due to high richness of diversified bioactive constituents. Hence, the present study was carried out during March, 2021 to characterize the biochemical profile of marine macroalga *Sargassum linearifolium* by using different techniques such as Gas Chromatography Mass Spectrometry (GCMS), Fourier Transform Infrared Spectroscopy (FTIR), and High Resolution Liquid Chromatograph Mass Spectrometer (HRLCMS-QTOF) for revealing its chemical profile. First, an algal sample was collected during low tide conditions in January, 2021. Characterization of functional groups was done by FTIR technique. GCMS analysis was performed of *S. linearifolium*'s methanolic and ethanolic extract. Likewise, phycocompounds characterization was also accomplished by HRLCMS-QTOF analysis. In results, the FTIR study characterized the presence of different bioactive functional groups whereas 12 and 7 different phycocompounds were found in GCMS analysis of ethanolic and methanolic extract, respectively. In HRLCMS Q-TOF characterization, a total 63 different types of compounds belonging to different classes were found to be in *S. linearifolium*. This characterization study revealed *S. linearifolium* a good source of various bioactive constituents. The incorporation of seaweed-based bioactive components in different applications has been extensively growing due to its huge benefits.

(Key words : FTIR , GCMS, HRLCMS-QTOF, Macroalgae, *Sargassum linearifolium*)

INTRODUCTION

Marine macroalgae, also known as seaweed, are macroscopic, multicellular, eukaryotic photosynthetic organisms which are widely distributed along the sea coasts from tropical to polar regions. It is mainly found in floating form, sometimes attached with rock, sand or other substratum in intertidal or subtidal regions of coastal areas (García-Poza *et al*., 2020). Based on pigment composition, marine macroalgae mainly classified into three major classes, i.e. Phaeophyceae class (Brown algae), Rhodophyceae class (red algae) and Chlorophyceae class (green algae) are belong to *Phaeophyta phylum*, *Rhodophyta phylum*, and *Chlorophyta phylum*, respectively. Among these three marine algae, green and red algae belong to the Plantae kingdom, whereas brown algae belong to the Chromista kingdom (Wang *et al*., 2015). Species of seaweeds are rich in various structurally diversified biologically active constituents such carbohydrates, proteins, amino acids, lipids, fatty acids, phenolic compounds, pigments, vitamins, and minerals (Thiyagarasaiyar *et al*., 2020). Algal species contain a greater diversified chemical profile than terrestrial organisms. The bioactive compounds have multiple activities which can be incorporated in various benefits and applications.

Broad utilities of macroalgae are based on presence of valuable bioactive ingredients and potential benefits. Many previous research studies demonstrated the chemical profile of different macroalgae species, which were characterized by different techniques and experimentations. Hence, present study was undertaken to characterize brown alga *Sargassum linearifolium* derived bioactive compounds by using different characterization techniques, mainly using GCMS, FTIR and HRLCMS-QTOF techniques.

MATERIALS AND METHODS

Collection of *Sargassum linearifolium*

The fresh sample of *S. linearifolium* was collected by handpicking method in a sterile plastic bag having seawater in it from the sea coast of BeytDwarka, Western coast of Gujarat, India. (Atmospheric condition during collection: Precipitation: 0%, Humidity: 55%, Wind: 11 km/h, 27°C, February 2021). This collected sample was transported (by maintaining sample at 10°C) to the laboratory. The geographical location of the collection site

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(22°28'42.1"N 69°08'10.6"E) illustrated in figure 1, whereas the isolated sample of *S. linearifolium* depicted in figure 2. This collected sample was washed thrice with distilled water to remove adhered epiphytes, salts and marine debris in running tap water. Finally, after washing, it was allowed to dry under room temperature for 5 to 7 days. Then, this dried material was ground to fine powder using a mechanical grinder and kept in an air tight container until the next analysis.

Characterization of phycocompounds by GCMS analysis

Extract preparation and analysis

Extraction of *S. linearifolium* was carried out using 500 g of prepared powder in 80% ethanol at 70°C by continuous hot percolation using Soxhlet digestion. It was continued for 24 hrs. and then this extract was filtered and kept in an oven at 40°C for 24 hrs to evaporate the ethanol. The concentration of this extract was made to dry under reduced pressure at 20°C in the rotary evaporator. This obtained residue was used as a sample in GCMS characterization and injected into an EB-5 column in JMS-T100GCV (GC model) for chromatographic separation and AccuTof Mass from jeol for mass determination. In detail, GC and MS conditions used according to Kalasariya *et al.* (2021). In analysis, obtained data of chromatogram was compared with the available data of known compounds in the NIST (National Institute Standard and Technology) library ver. 2005.

Fatty acids characterization

Extract preparation and analysis

For characterization of fatty acids, the prepared fine powder of *S. linearifolium* was extracted in methanol in the ratio of 1:10 w/v in a flask for 72 hours. The mixture was filtered in a separate container. This process was carried out twice with the same residues using fresh methanol. After collection of supernatant, the excess solvent is removed by a rotary evaporator and used as a sample for further characterization by GCMS. This analysis was done by using the same model of GC and MS but for methanolic extract, HP-5 column was used. The other specifications of GC and MS as used in Kalasariya *et al.* (2021). The sample was injected in split mode at 1:10. In analysis, obtained data of chromatogram was compared with the available data of known compounds in the NIST library ver. 2005.

Characterization of functional groups

Functional groups characterization in *S. linearifolium* carried out by FTIR technique by KBr pellet method. The wavelength of light absorbed is characteristic of the bond that can be seen in the spectrum. This characterization was done by using 3000 Hyperion Microscope with Vertex 80 FTIR System (Bruker, Germany) model. In analysis, 5 mg of dried powder of *S. linearifolium* was added with potassium bromide (KBr) and evenly mixed to obtain a homogenized fine powder. Then, this mixture was placed in the mold and pressed using mechanical pressure for 30 seconds to prepare pellets, following this, put this pellet on a pan and proceeded for further analysis. The scanning range was 400–4000 cm^{-1} and scanning displayed in % transmission analysis.

Phycocompounds characterization by HRLCMS-QTOF

Acid hydrolysis and characterization

Acid digestion of *S. linearifolium*'s sample was performed by drying in an oven at 40°C for 48 hours, then a total 100 mg of the sample was placed in a screw cap tube and added 10 ml of 2M HCl containing 1% phenol. After that, this tube was closed under N_2 gas and kept in an electric oven at 80°C for 3 h and following this, vacuum filtered through Whatman no. 41 paper. The obtained filtrate was diluted to make total volume 25 using an ultrapure water which was again filtered to get the hydrolysate. This hydrolysate was considered as a sample and characterized by Hypersil GOLD C18 Column in 6550 iFunnel Q-TOFs, Agilent Technologies (USA) model and scanned in range from 150–1000 m/z for MS/MS Dual AJS ESI in the mode of ionization. Other specifications and scan source conditions are set as in Kalasariya *et al.* (2021). Resolved peaks obtained in liquid chromatogram were further identified with the help of reported values from the literature.

Figure 1. Geographical location of sample collection site in respect to Gujarat state, India.

Figure 2. Isolated sample of *Sargassum linearifolium* (Ochrophyta).

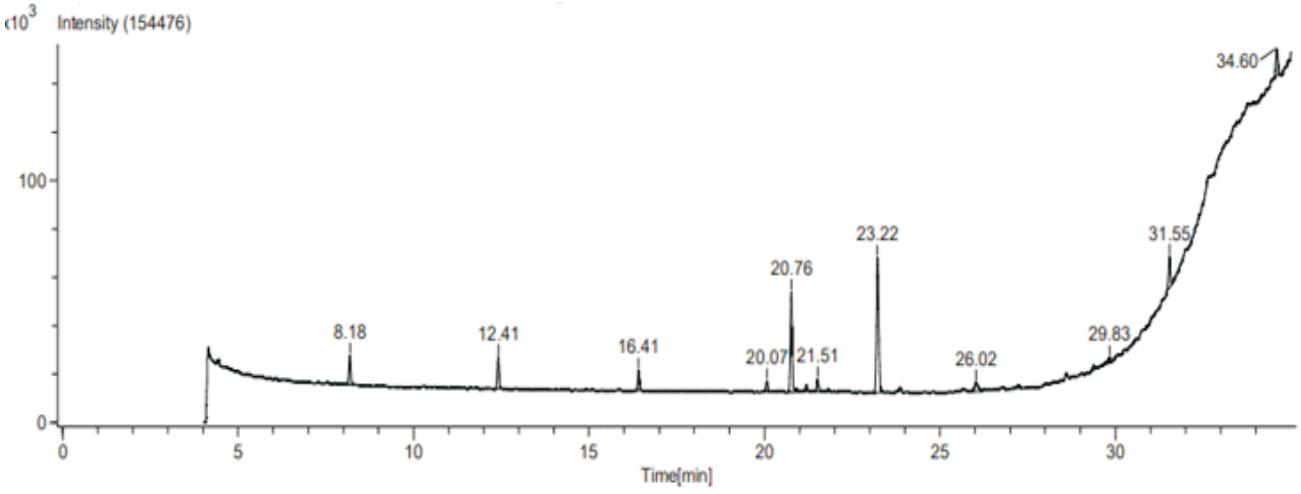

Figure 3. The gas chromatogram obtained in GC analysis of*S. linearifolium*ethanolic extract.

Table 1. Details of phycocompounds identified through GCMS analysis of *S. linearifolium* ethanolic extract

No.	Name	Pubchem ID	Mol. Formula	Mol. Weight (g/mol)	Retention time (min)	Retention index (iu)	% Peak area
1	Butanal, 3-methyl-	11552	$C_5H_{10}O$	86.13	8.18	643	6.21
2	Oxalic acid, allyl octyl ester	6420247	$C_{13}H_{22}O_4$	242.31	12.41	1638	6.64
3	2,2-Dimethyl-propyl 2,2-dimethyl-propane-thiosulfinate	551400	$C_{10}H_{22}OS_2$	222.4	20.07	1568	2.00
4	2H-pyran-2-dodecanoic acid,6-(17,19-dimethylheneicosyl)tetrahydro,- methyl ester	545770	$C_{41}H_{80}O_3$	621.1	20.76	4249	21.94
5	[s]-{+}-1-cyclohexylethylamine	5325951	$C_8H_{17}N$	127.23	21.19	1059	1.28
6	3-methyl-3,5--(cyanoethyl)tetrahydro-4-thiopyranone	541466	$C_{12}H_{16}N_2OS$	236.34	21.51	2116	2.62
7	Dibutyl phthalate	3026	$C_{16}H_{22}O_4$	278.34	23.22	2037	35.49
8	1,3-Propanediamine, N-methyl-	80511	$C_4H_{12}N_2$	88.15	26.02	860	3.57
9	Dihydrooxostephamiersine	621048	$C_{21}H_{27}NO_7$	405.4	29.38	2896	1.34
10	[2S,3S]-[-]-3-propyloxiranemethanol	10313120	$C_6H_{12}O_2$	116.16	29.82	912	1.47
11	6,9,12-Octadecatrienoic acid, phenylmethyl ester,[Z,Z,Z]-	5368209	$C_{25}H_{36}O_2$	368.6	31.55	2774	6.68
12	d-Mannitol, 1-decylsulfonyl-	568528	$C_{16}H_{34}O_7S$	370.5	34.60	3010	6.67

RESULTS AND DISCUSSION

Phycocompounds characterization in ethanolic extract

The GCMS characterization of *S. linearifolium*'s ethanolic extract showed a total 12 different compounds that were identified based on its retention time, retention index and % peak area. A gas chromatogram with different peaks is illustrated in figure 3. Chemical information of these compounds such as its PubChem ID, molecular weight, molecular formula, retention time (min), retention index (iu), % peak area and SMILE structure indicated in Table 1. These compounds were Butanal, 3-methyl-; Oxalic acid, allyl octyl ester; 2,2-Dimethyl-propyl 2,2-dimethyl-propane-thiosulfinate; 2H-pyran-2-dodecanoic acid,6-(17,19-dimethylheneicosyl) tetrahydro, - methyl ester; [s]-{+}-1-cyclohexylethylamine; 3-methyl-3,5—(cyanoethyl) tetrahydro-4-thiopyranone; Dibutyl phthalate; 1,3-Propanediamine, N-methyl-; Dihydrooxostephamiersine; [2S,3S]-[-]-3-propyloxiranemethanol; 6,9,12-Octadecatrienoic acid, phenylmethyl ester,[Z,Z,Z]- and d-Mannitol, 1-decylsulfonyl-. Among the idetified compoounds, Dibutyl phthalate was found to be the major compound which revealed the largest peak area (35.49 %) with the retention time (23.22 min) whereas [s]-{+}-1-cyclohexylethylamine compound showed the smallest peak area (1.28 %) with the retention time (21.19 min).

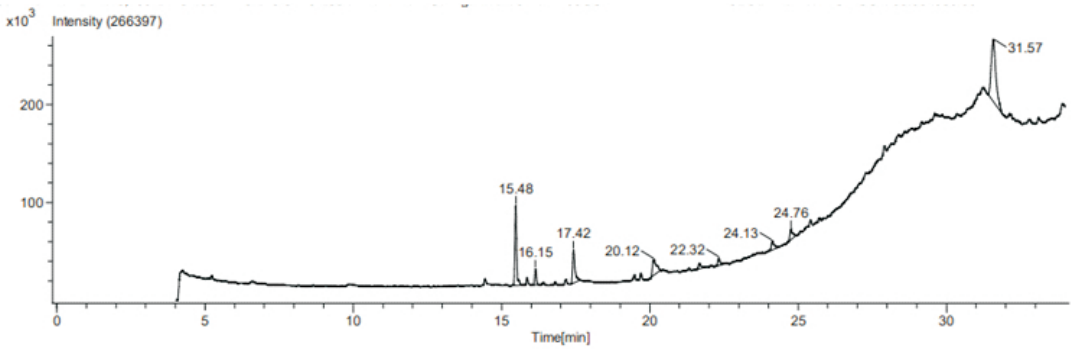

Figure 4. The gas chromatogram obtained in GC analysis of *S. linearifolium* methanolic extract.

Table 2. Details of phycocompounds identified through GCMS analysis of *S. linearifolium* methanolic extract

No.	Name	Pubchem ID	Mol. Formula	Mol. Weight (g mol^{-1})	Retention time (min)	Retention index (iu)	% Peak area
1.	Glycine, N-allyloxy carbonyl-, Penta decyl ester	6420282	$C_{21}H_{39}NO_4$	369.5	15.48	2632	19.53
2	Pentadecanal-	17697	$C_{15}H_{30}O$	226.4	16.14	1701	3.77
3	Hexadecanoic acid, tetradecyl ester	78294	$C_{30}H_{60}O_2$	452.8	17.43	3171	10.07
4	2-Decenal, [E]-	5283345	$C_{10}H_{18}O$	154.25	20.13	1212	13.14
					21.67	1647	1.89
5	Cyclooctaneacetic acid,2-oxo-	536995	$C_{10}H_{16}O_3$	184.23	22.32	1647	2.29
					24.13	1647	4.54
6	2-Undecanethiol,2-methyl-	82330	$C_{12}H_{26}S$	202.4	24.76	1433	5.48
7	Cholest-5-en-3-ol,24-propylidene-,[3β]-	6443745	$C_{30}He2_{50}O$	426.7	31.58	2880	39.30

Phycocompounds characterization in ethanolic extract

In analysis, total 7 different compounds were detected in *S. linearifolium*'s methanolic extract such as Glycine, N-allyloxycarbonyl-,pentadecyl ester; Pentadecanal-; Hexadecanoicacid,tetradecyl ester; 2-Decenal,[E]-; Cyclooctane Acetic acid,2-oxo-; 2-Undecanethiol,2-methyl- and Cholest-5-en-3-ol,24-propylidene-,[3â]-. Data regarding gas chromatogram with different peaks for methanolic extract are presented in figure 4, whereas chemical information such as its retention time, retention index, % peak area, PubChem ID and SMILE structure are reported in Table 2. Among the identified compounds, Cholest-5-en-3-ol,24-propylidene-,[3â]- was found to be the major compound that attained the largest peak (39.30%) with a retention time (31.58 min), whereas Cyclooctane Acetic acid,2-oxo- showed the lowest peak area of 1.89% with a retention time of 21.67 min.

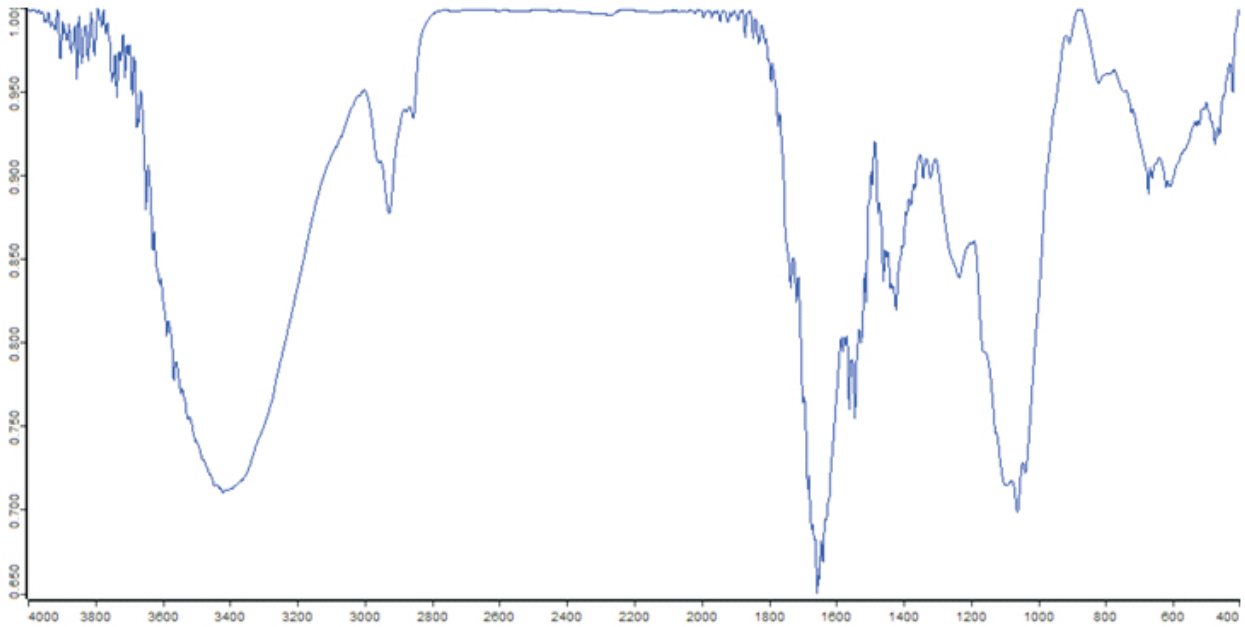

Figure 5. FTIR spectrum of *S. linearifolium* powder by KBr pellet method

Table 3. Characterization of functional groups by FTIR analysis of *S. linearifolium*'s powder

Absorption	Appearance	Group	Compound Class
524	Strong	C-Br stretching	halo compound
	strong	C-I Stretching	halo compound
617	Strong	C-Br stretching	halo compound
	strong	C-Cl stretching	halo compound
667	Strong	C-Cl stretching	halo compound
	Strong	C-Br stretching	halo compound
	Strong	C=C bending	alkene
			alkene
818	Medium	C=C bending	halo compound
	Strong	C-Cl stretching	1,4-disubstituted or 1,2,3,4 tetra
	strong	C-H bending	substituted
1061	Strong	C-F stretching	Fluoro compound
	Strong	C-O stretching	Primary alcohol
	strong	S=O stretching	Sulfoxide
	medium	C-N stretching	Amine
1235	Strong	C-F stretching	Fluoro compound
	Medium	C-N stretching	amine
	Strong	C-O stretching	alky aryl either
1421	medium	O-H bending	Carboxylic acid
1545	strong	N-O stretching	Nitro compound
1637	Medium	C=C stretching	alkene
	Medium	C=C stretching	Conjugated alkene
	Medium	C=C stretching	Cyclic alkene
	medium	N-H bending	Amine
1653	Medium	C=C stretching	alkene (disubstituted)
	Medium	C=N stretching	Imine/oxime
	Medium	C=C stretching	alkene(vinylidene)
	weak	C-H bending	aromatic compound
1734	Weak	C-H bending	aromatic compound
	Strong	C=O stretching	aldehyde
1869	Weak	C-H bending	aromatic compound
2855	Strong broad	O-H stretching	Carboxylic acid
	Week broad	O-H stretching	alcohol
	Strong broad	N-H stretching	amine salt
	medium	C-H stretching	alkane
2929	Strong broad	O-H stretching	Carboxylic acid
	Week broad	O-H stretching	alcohol
	Strong broad	N-H stretching	amine salt
	Medium	C-H stretching	alkane
3424	Strong broad	O-H stretching	alcohol
3586	Medium sharp	O-H stretching	alcohol
3629	Medium sharp	Medium sharp	alcohol
3675	Medium sharp	Medium sharp	alcohol
3586	Medium sharp	O-H stretching	alcohol
3629	Medium sharp	Medium sharp	alcohol
3675	Medium sharp	Medium sharp	alcohol

Functional groups characterization study

FTIR spectrum analysis was used to characterize the functional groups of bioactive constituents based on the peak values at different intensities in the infrared region. The IR spectrum illustrated in figure 5, whereas absorption, appearance, groups and compound class tabulated in Table 3. The more intense band occurring at 524 cm^{-1}, 617 cm^{-1}, 667 cm^{-1}, 818 cm^{-1}, 1061 cm^{-1}, 1235 cm^{-1}, 1421 cm^{-1}, 1545 cm^{-1}, 1637 cm^{-1}, 1653 cm^{-1}, 1734 cm^{-1}, 1869 cm^{-1}, 2855 cm^{-1}, 2929 cm^{-1}, 3424 cm^{-1}, 3586 cm^{-1}, 3629 cm^{-1}, and 3675 cm^{-1} corresponding to C-Br/C-I/C-Cl/C=C/C-H/C-F/C-O/S=O/C-N/O-H/N-O/C=C/N-H/C=N/C=O. This result of FTIR

spectral study revealed the presence of halo compound, alkene, Fluoro compound, Primary alcohol, Sulfoxide, Amine, alkyl aryl ether, Carboxylic acid, Nitro compound, Cyclic alkene, Imine/oxime, aromatic compound, Carboxylic acid, alcohol, amine salt, and alkane in output.

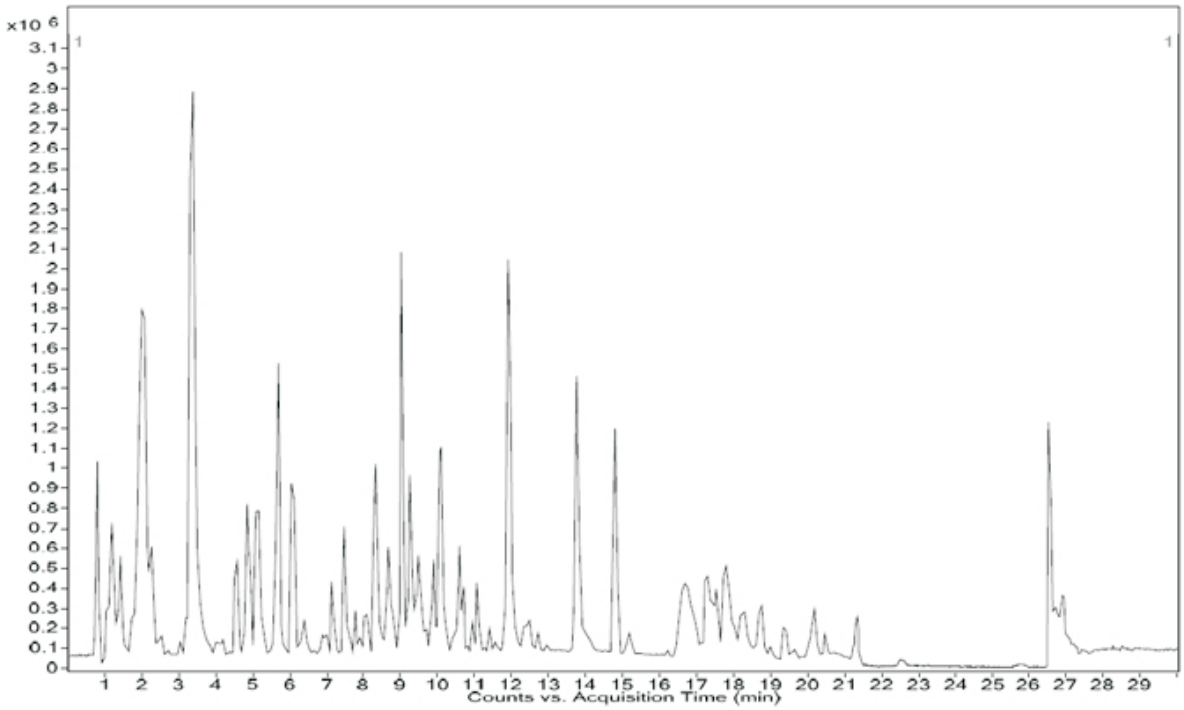

Figure 7. Liquid chromatogram obtained by HRLCMS-QTOF characterization of *S. linearifolium*

Table 6. Characterization of phycochemicalsobtained in *S. linearifolium* by HRLCMS-QTOF

Retention time (min.)	Mass (Da)	Abundance	Name	Formula	Hits (DB)
0.824	218.0138	-	8-Hydroxy-2chlorodibenzofuran	$C_{12}H_7ClO_2$	2
0.825	276.0554	-	Sulfabenzamide	$C_{13}H_{12}N_2O_3S$	3
1.149	171.1246	183304	Gabapentin	$C_9H_{17}NO_2$	1
1.151	203.1148	214034	L-Glutamic acid n-butyl ester	$C_9H_{17}NO_4$	3
1.218	222.1704	277267	(1S)-2,2-Dimethyltetrahydrospiro[cyclo pentane-1,1'-pyrrolizin]7'(7a'H)-one oxime	$C_{13}H_{22}N_2O$	10
1.364	267.0955	210011	Miserotoxin	$C_9H_{17}NO_8$	10
1.476	207.1249	94663	Salsolidine	$C_{12}H_{17}NO_2$	10
1.537	240.1466	297693	Pirbuterol	$C_{12}H_{20}N_2O_3$	10
1.929	134.1072	985884	1,4-Diethylbenzene	$C_{10}H_{14}$	10
2.1	207.1248	-	Salsolidine	$C_{12}H_{17}NO_2$	10
2.357	207.1002	262857	5-(alpha-Phenylethyl)semioxamazide	$C_{10}H_{13}N_3O_2$	6
3.21	198.1724	146016	Cycluron	$C_{11}H_{22}N_2O$	1
3.394	149.1198	-	2,6-Diethylaniline	$C_{10}H_{15}N$	9
3.41	236.1516	-	Procaine	$C_{13}H_{20}N_2O_2$	3
4.023	245.1594	130217	Pinacidil	$C_{13}H_{19}N_5$	1
4.23	343.2191	144312	Isopiperolein B	$C_{21}H_{29}NO_3$	5
4.54	387.245	-	N-linoleoyl taurine	$C_{20}H_{37}NO_4S$	10
4.604	376.1369	201319	Loganic acid	$C_{16}H_{24}O_{10}$	10

5.183	220.0663		Brassitin	$C_{11}H_{12}N_2OS$	10
5.346	188.1196	163103	2-Butyl-3-phenyl-2-propen-1al	$C_{13}H_{16}O$	10
5.569	405.2348	125756	Cypridina luciferin	$C_{22}H_{27}N_7O$	7
6.006	196.1091	478569	Benzenemethanol, 2-(2hydroxypropoxy)-3-methyl-	$C_{11}H_{16}O_3$	10
6.081	178.0988	138435	2-Phenylethyl propanoate	$C_{11}H_{14}O_2$	10
6.765	223.1207	84202	beta-Butoxyethyl nicotinate	$C_{12}H_{17}NO_3$	7
6.944	308.1975	77718	Methylgingerol	$C_{18}H_{28}O_4$	10
7.522	268.2049	111977	(1R,2R)-3-oxo-2-pentylcyclopentanehexanoic acid	$C_{16}H_{28}O_3$	10
7.797	438.1171	290409	Loquatoside	$C_{20}H_{22}O_{11}$	10
8.193	274.1922	237865	13-Octadecene-9,11-diynoic acid, (Z)-	$C_{18}H_{26}O_2$	10
8.347	343.2337		Dibucaine	$C_{20}H_{29}N_3O_2$	1
8.469	272.1767	218890	5,8,11-Octadecatriynoic acid	$C_{18}H_{24}O_2$	10
8.56	274.1925	223080	13-Octadecene-9,11-diynoic acid, (Z)-	$C_{18}H_{26}O_2$	10
8.608	290.1871	122661	Ecklonialactone A	$C_{18}H_{26}O_3$	10
8.611	328.2244	166152	9,12,13-trihydroxy-10,15octadecadienoic acid	$C_{18}H_{32}O_5$	10
8.668	292.2032	144819	17beta-Hydroxy-2-oxa-5alphaandrostan-3-one	$C_{18}H_{28}O_3$	10
8.901	274.1924	261639	13-Octadecene-9,11-diynoic acid, (Z)-	$C_{18}H_{26}O_2$	10
8.96	350.2063		4,4-Difluoropregn-5-ene-3,20dione	$C_{21}H_{28}F_2O_2$	10
9.169	262.1563	208711	Acutifolane A	$C_{16}H_{22}O_3$	8
9.305	205.1461	275349	3-Ethylethcathinone	$C_{13}H_{19}NO$	8
9.36	292.2026	146163	17beta-Hydroxy-2-oxa-5alphaandrostan-3-one	$C_{18}H_{28}O_3$	10
9.481	158.1318	166243	3-methyl-octanoic acid	$C_9H_{18}O_2$	10
9.594	639.3091		34a-Deoxy-rifamycin W	$C_{35}H_{45}NO_{10}$	4
9.644	310.2136	210591	2(R)-HPOT	$C_{18}H_{30}O_4$	10
9.698	270.2206	160558	9-keto palmitic acid	$C_{16}H_{30}O_3$	10
9.841	273.2661	260667	C16 Sphinganine	$C_{16}H_{35}NO_2$	1
10.015	278.085	206049	Triphenylphosphine oxide	$C_{18}H_{15}OP$	10
10.045	292.2027	204033	17beta-Hydroxy-2-oxa-5alphaandrostan-3-one	$C_{18}H_{28}O_3$	10
10.108	224.1879	164102	Cuscohygrine	$C_{13}H_{24}N_2O$	3
10.157	312.2283	118460	9(S)-HpODE	$C_{18}H_{32}O_4$	10
10.16	294.2183	99470	10-Oxo-11-octadecen-13-olide	$C_{18}H_{30}O_3$	10
10.342	308.1976	99461	Corchorifatty acid A	$C_{18}H_{28}O_4$	10
10.453	274.1922	135396	13-Octadecene-9,11-diynoic acid, (Z)-	$C_{18}H_{26}O_2$	10
10.676	254.2077	55612	18-Nor-4(19),8,11,13abietatetraene	$C_{19}H_{26}$	7
11.101	292.2027	157274	17beta-Hydroxy-2-oxa-5alphaandrostan-3-one	$C_{18}H_{28}O_3$	10
11.932	431.2295		Trp Arg Ala	$C_{20}H_{29}N_7O_4$	6
12.031	392.2205		Nigakilactone B	$C_{22}H_{32}O_6$	10
14.797	362.2111		Hydrocortisone	$C_{21}H_{30}O_5$	10
17.266	527.2351		Hygromycin B	$C_{20}H_{37}N_3O_{13}$	6
17.434	269.2707		Capsi-amide	$C_{17}H_{35}NO$	1
17.746	519.2681	320124	PS(18:3(6Z,9Z,12Z)/0:0)	$C_{24}H_{42}NO_9P$	2
18.872	283.2864	172057	Stearamide	$C_{18}H_{37}NO$	1
19.3	337.3332	105321	N-Cyclohexanecarbonylpentadec ylamine	$C_{22}H_{43}NO$	3
26.549	478.0858		CCG-100602	$C_{21}H_{17}ClF_6N_2O_2$	10
26.711	224.1757		7Z,9E-Dodecadienyl acetate	$C_{14}H_{24}O_2$	10
26.76	212.1049		MeIQ	$C_{12}H_{12}N_4$	3

Phycocompounds characterization by HRLCMS-QTOF

High Resolution Liquid Chromatography Mass Spectrometry Quadrupole Time of Flight (HR-LCMS Q-TOF) analysis of *S. linearifolium* showed a total of sixty-three different compounds at different retention times. Liquid chromatogram of *S. linearifolium* with different peaks is illustrated in figure 7, whereas retention time, mass, abundance, molecular formula, and hits of each compound tabulated in Table 6. Moreover, among identified compounds, Lipid like compounds, Acutifolane A; 3-methyl-octanoic acid; 2(R)-HPOT; 9-keto palmitic acid; 9(S)-HpODE; Corchorifatty acid A; C16 Sphinganine; 13-Octadecene-9,11-diynoic acid, (Z)-; Hydrocortisone; PS(18:3(6Z,9Z,12Z)/0:0); 7Z,9E-Dodecadienyl acetate; Stearamide; 4,4-Difluoropregn-5-ene-3,20dione; 13-Octadecene-9,11-diynoic acid, (Z)-; 9,12,13-trihydroxy-10,15octadecadienoic acid; 13-Octadecene-9,11-diynoic acid, (Z)-; 13-Octadecene-9,11-diynoic acid, (Z)-; 5,8,11-Octadecatriynoic acid; (1R,2R)-3-oxo-2-pentylcyclopentanehexanoic acid; and N-linoleoyltaurine were found. Besides, some organic molecules were identified such as Triphenylphosphine oxide; Methylgingerol; Ecklonialactone A; 34a-Deoxy-rifamycin W; Nigakilactone B; Dibucaine; Cypridinaluciferin; Pinacidil; (1S)-2,2-Dimethyltetrahydrospiro [cyclo pentane-1,1'-pyrrolizin]7'(7a'H)-one oxime and Sulfabenzamide. Other class of compounds such as Alkaloids (3-Ethylethcathinone); Gonadal Steroid Hormones (17beta-Hydroxy-2-oxa-5alphaandrostan-3-one), Carbohydrates (Hygromycin B; Miserotoxin; Loquatoside), Amino acid derivative (Gabapentin; L-Glutamic acid n-butyl ester), Heterocyclic Compounds (Cuscohygrine), peptide (TrpArgAla), Carboxylic Acids (Capsi-amide), and Heterocyclic Compounds (CCG-100602). In addition, Benzyl Alcohols, Benzenemethanol and 2-(2hydroxypropoxy)-3-methyl-; Heterocyclic Compounds (Dibenzofurans and 8-Hydroxy-2chlorodibenzofuran), Amino Alcohol (Pirbuterol), Benzene Derivatives (1,4-Diethylbenzene), amino acid amide (5-(alpha-Phenylethyl)semioxamazide), Hydrocarbons (Cycluron), Amines (2,6-Diethylaniline),heterocyclic Compounds (Pyrrolidines and Isopiperolein B),Terpenes (Monoterpenes and Loganic acid), Aldehydes (2-Butyl-3-phenyl-2-propen-1al), Heterocyclic Compounds (2-Ring Indoles), Brassitin whereas carboxylic Acid derivative (Procaine), Vitamin B Complex(beta-Butoxyethylnicotinate) and carbocyclic compound(2-Phenylethyl propanoate) were identified. Moreover, Alkaloids(Salsolidine), Organooxygen compounds (N-Cyclohexanecarbonylpentadecylamine; MeIQ) and Hydrocarbon derivatives (10-Oxo-11-octadecen-13-olide; 18-Nor-4(19),8,11,13abietatetraene) were also found in *S. linearifolium*.

Many previous research studies characterized different marine macroalgae species by using different characterization techniques to reveal its chemical profile. Sumayya *et al.* (2020) characterized phytochemicals, HPLC and FTIR analysis of red alga *Gracilaria dura*'s methanolic extract that suggested the presence of reducing sugar, flavonoids, glycosides, terpenoids and tannins. They also identified a good fraction of phenolic compounds such as gallic acid, vanillic acid, sinapic acid, p-coumaric acid, hydroxybenzoic, phloroglucinol, catechol and cinnamic acid in HPLC analysis, whereas found the presence of active constituents containing alkanes, ketones, alkyl halides, hydroxyl groups etc. Likewise, Deepika (2019) found significant metabolites in macoalga *Sargassum wightii* collected from Vedalai, Gulf of Mannar, Rameswaram, Tamil Nadu by FTIR, EDS and GCMS characterization study. She found specific compounds such as 8,10-Dodecadien-1-ol acetate and 18-oxo-methyl ester, 13-Docosenoic acid methyl ester in GCMS analysis, whereas the frequency of functional groups by FTIR analysis. Likewise, EDS analysis revealed a good number of elements in *S. wightii*. Moreover, Majumder *et al.* (2020) explored the potential of green macroalga *Chaetomorpha brachygona* collected from Indian Sundarbans derived chemicals against the proliferation of cervical cancer cells *in vitro* to check its anticancer property. In this study, chloroform extract used on SiHa cell lines showed significant cytostatic and cytotoxic activity. This alga derived phytochemical identified by GCMS, LCMS and column chromatography study and some of the identified anticancer compounds have shown strong Bcl-2 binding capacity, as analysed through molecular docking study. In addition, Goulitquer *et al.* (2012) reviewed Mass Spectrometry-Based (MS based) Metabolomics to explore functions in marine organisms and ecosystems. They also pointed out some technical challenges mainly extraction of chemical components from different matrices and data management that need to be overcome in order to improve its metabolites applications in marine systems. In this study, researchers also integrated data of metabolite profiles with layers of omics as well as their importance for the development of system biology approach to study several biological processes and to analyse interaction between organisms of community as well as their importance for the development of systems biology approaches in marine systems to study several biological processes, and to analyse interactions between organisms within communities. In conclusion, the present characterization study of brown alga *S. linearifolium* by different characterization techniques such as GCMS, FTIR and HRLCMS-QTOF technique that revealed that this alga contained potential bioactive constituents which can be useful for various applications after *in vitro* and *in vivo* pharmacological evaluation and safety analysis.

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