Electronic Supplementary Material (ESI) for RSC Advances.

Supplementary Information

Scaling the effect of hydrophobic chain length variations on gene transfer properties of di-alkyl, di-hydroxy ethylammonium chloride based cationic amphiphiles

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Contents **Page number**

Figure S1. Hydrodynamic diameters (top) and surface potentials (bottom) of liposomes of lipids, Lip**1818**-Lip**1810**

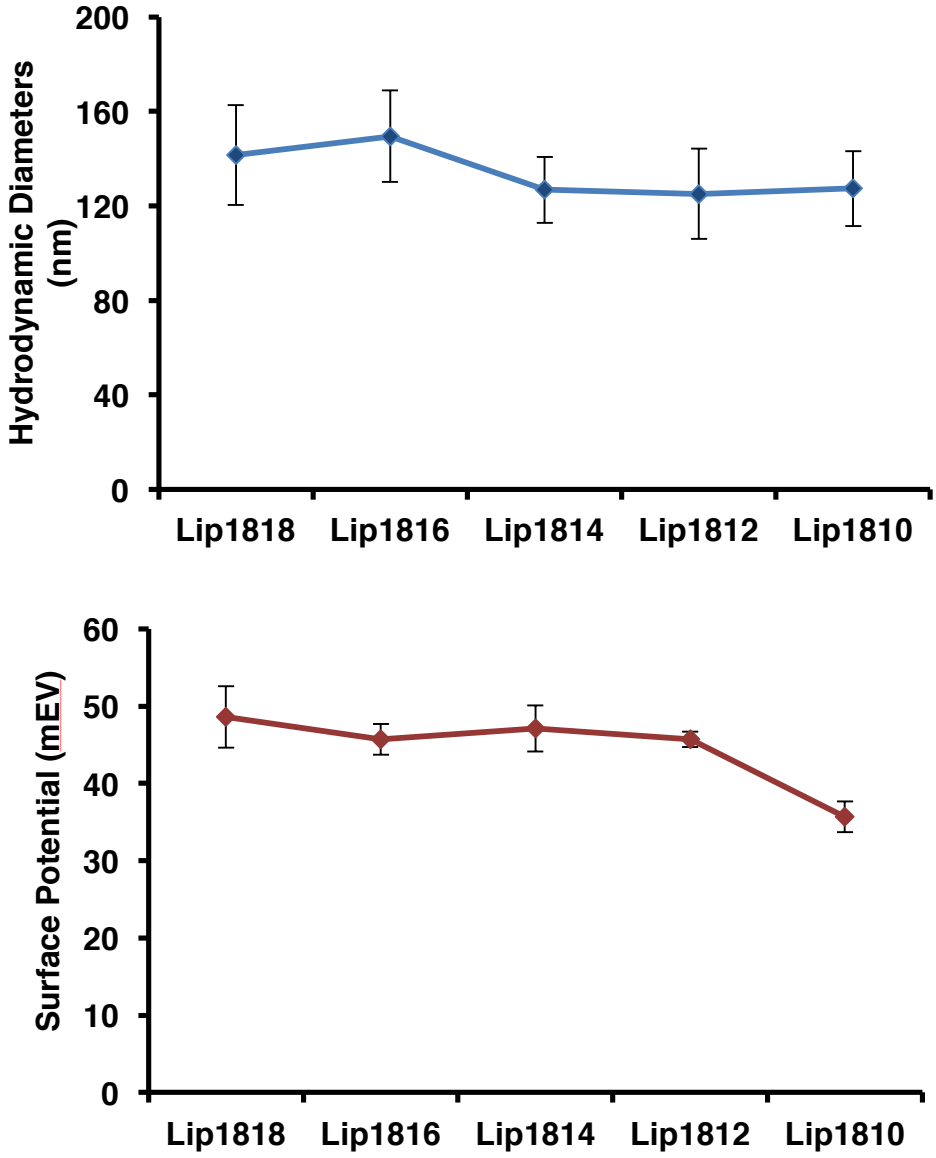


Figure S2. Sizes of the lipid-DNA complexes (lipoplexes) measured using dynamic light scattering.

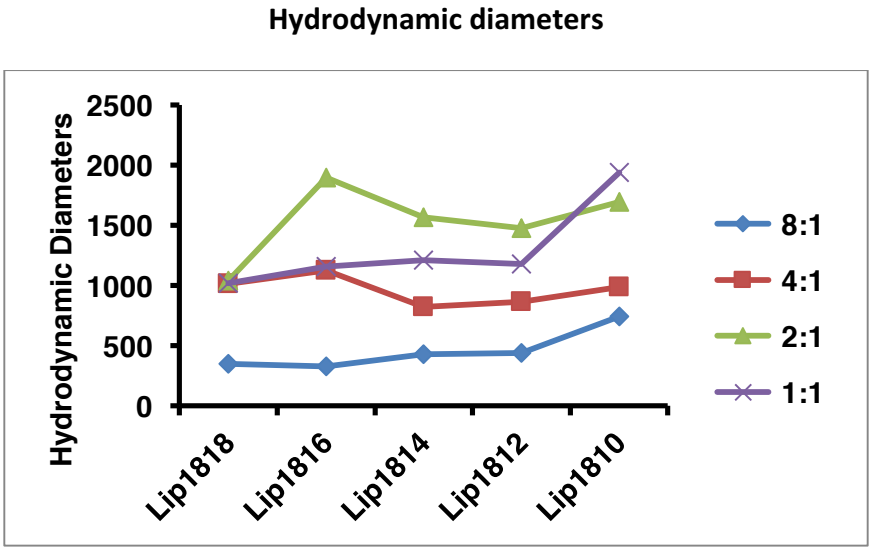
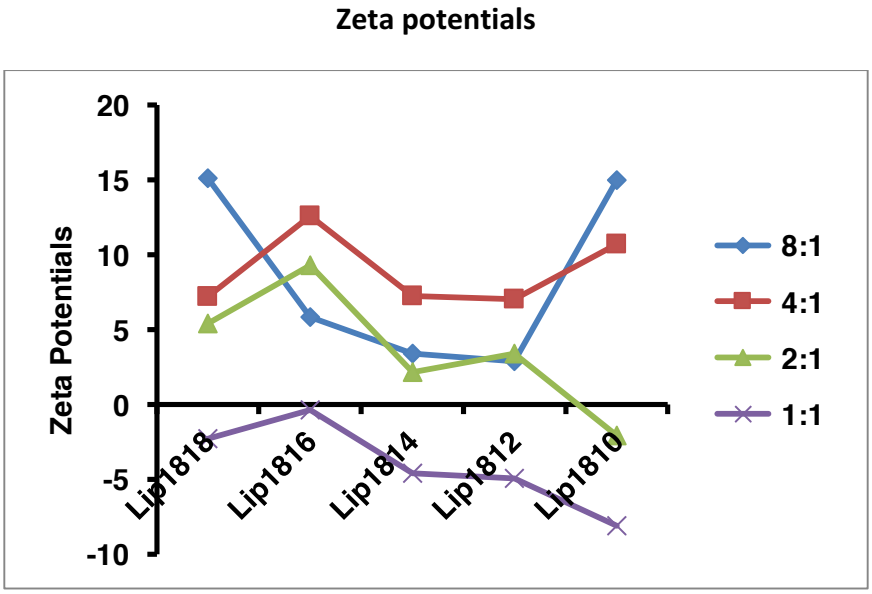


Figure S3. Surface charge of the lipid-DNA complexes (lipoplexes) measured using dynamic light scattering.



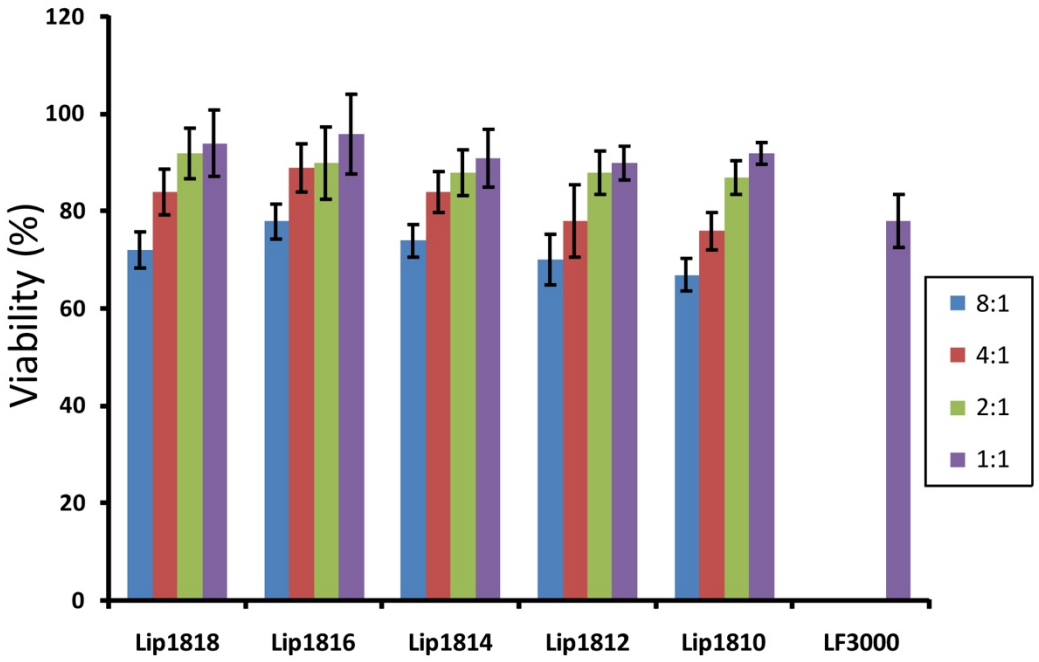


Figure S4. Cytotoxicity evaluation of Lipids (Lip1818-Lip1810) in HeLa cells for 24h incubation. HeLa cells were incubated with Lipids (Lip1818-Lip1810) for 24h at varying Lipid:DNA charge ratios along with Lipofectamine 3K. Control cells received DMEM complete media with 0.01% DMSO. MTT assay was performed as per the protocol described in Materials & Methods. Values shown are means ± SEM of three independent experiments performed in six to eight replicates.

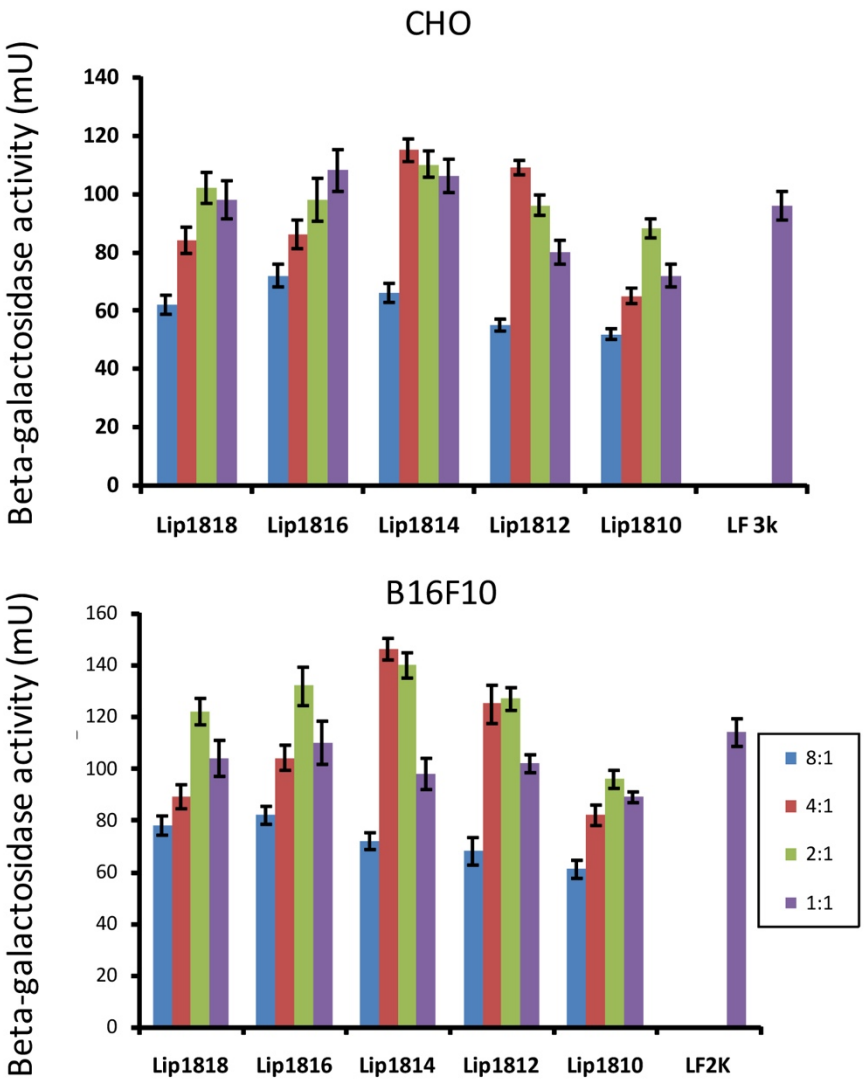


Figure S5. In vitro transfection efficiencies of Lipids (Lip1818-Lip1810) in CHO and B16F10 cell lines at lipid:DNA charge ratios of 8:1-1:1. Absorption at 405 nm was converted to β-galactosidase units using a calibration curve constructed with pure commercial β-galactosidase enzyme. Units of β-galactosidase activity were plotted against the varying lipid-to-DNA charge ratios (8:1-0.5:1). The transfection values shown are the average of triplicate experiments performed with 4-6 data points.

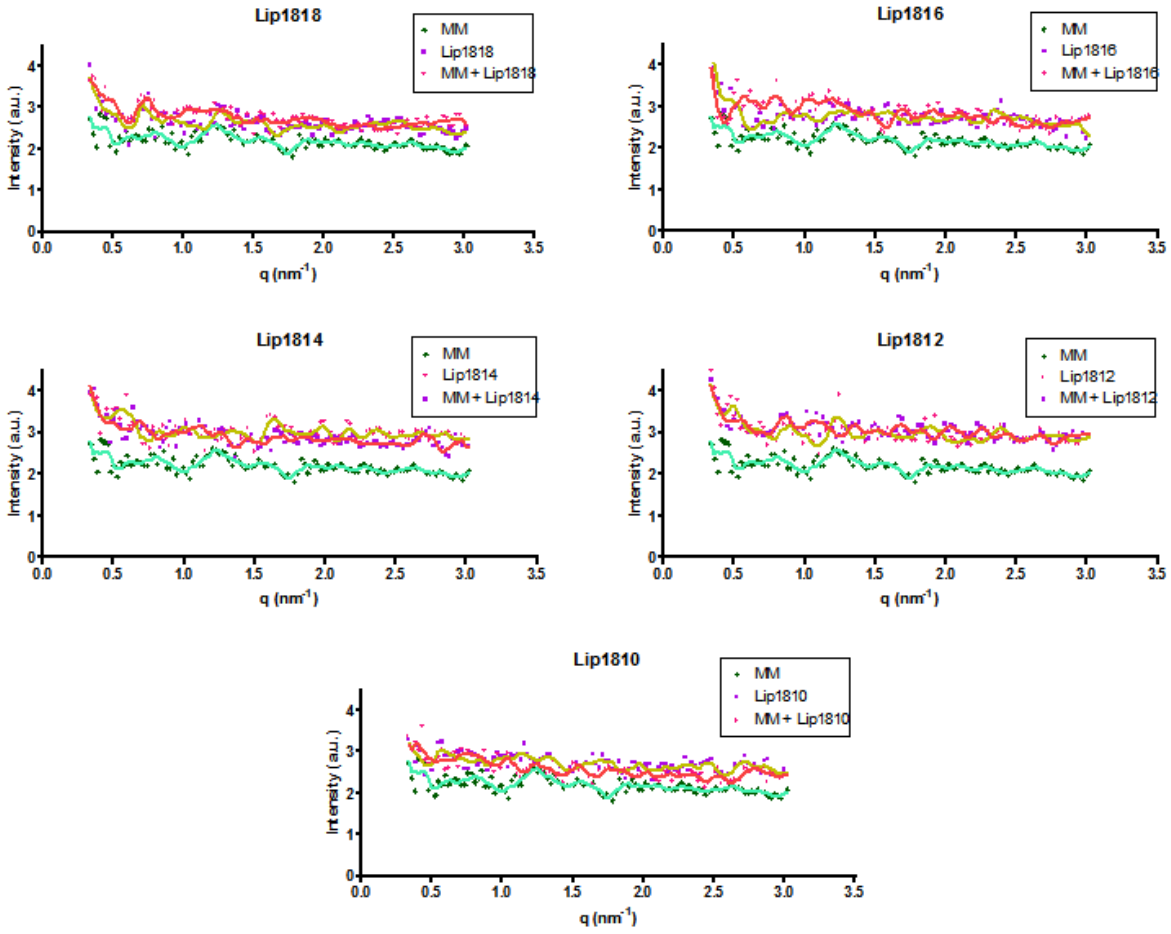
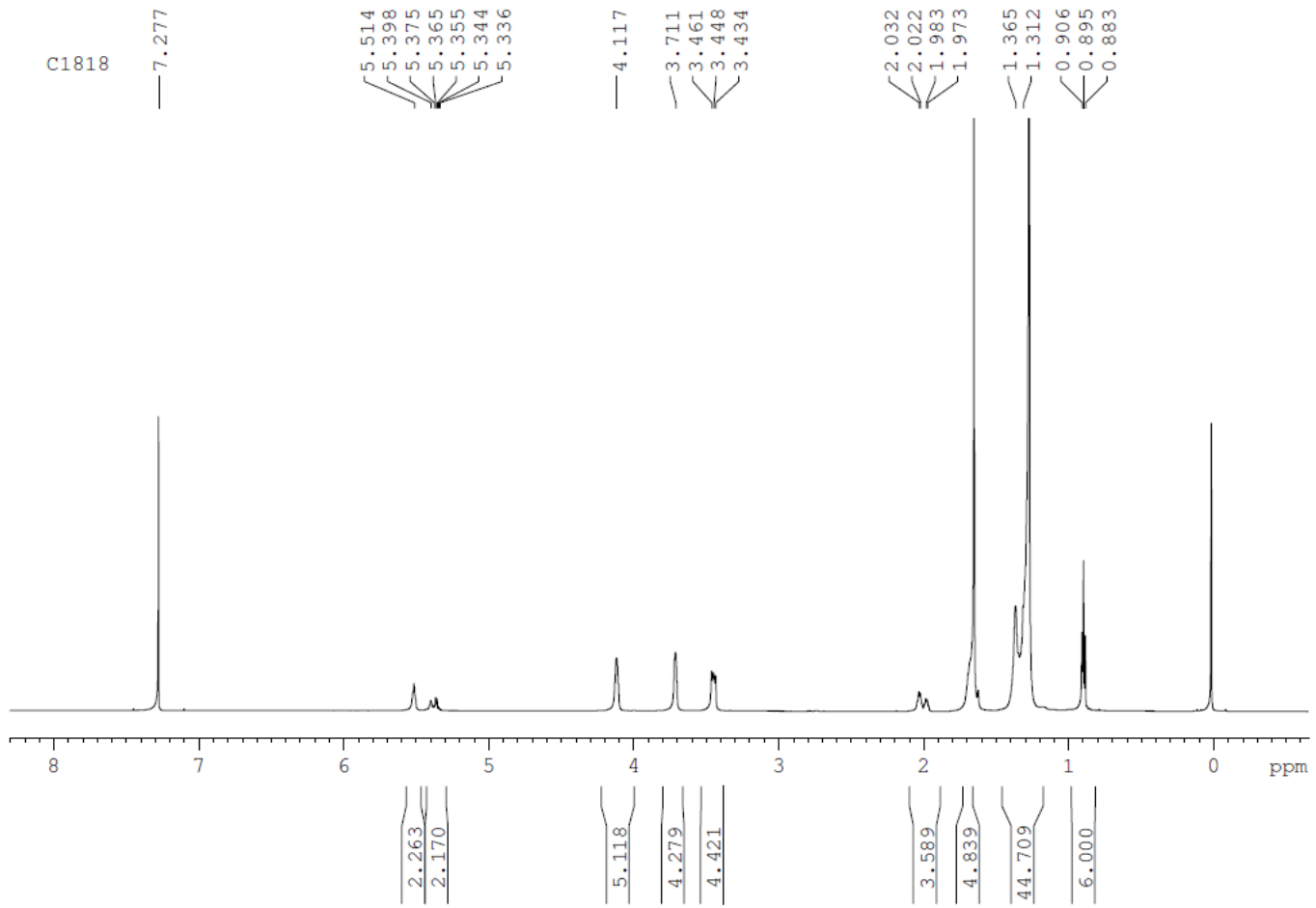
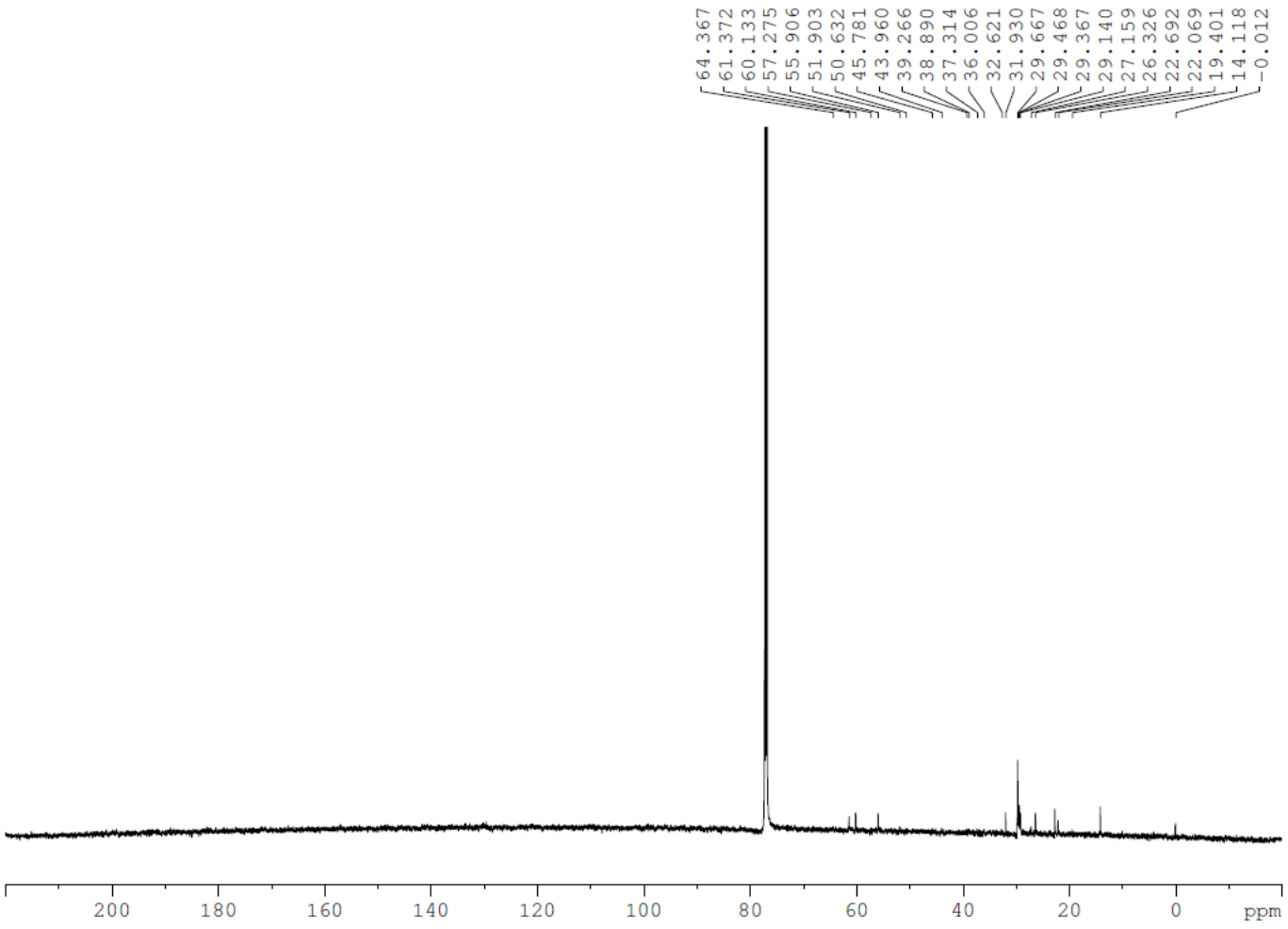


Figure S6. SAXS diagrams of Lipids (Lip1818-Lip1810). Radially integrated scattering intensities of the lipids, with and without the Model membrane (MM) at 1:1 molar ratio. The Model membrane is a phospholipid mixture of DOPC:DOPE:DOPS:Chol at 45:20:20:15, w/w content that closely mimics mammalian cell membrane. The symbols correspond to the measured data and the solid curves represent the smoothened curve with weighted average 9 neighbours to the data. Curve smoothing was done using GraphPad Prism software.

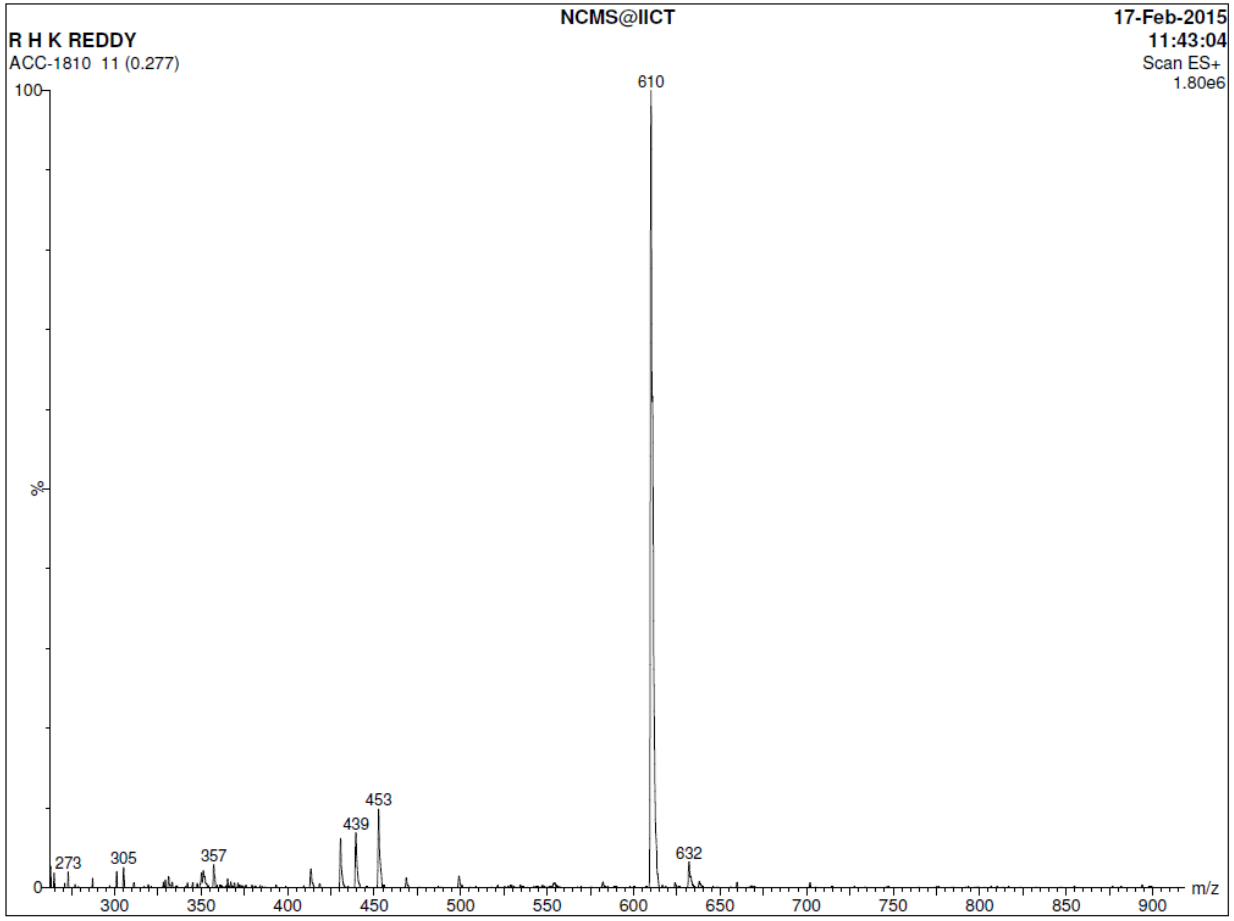
1H NMR of Lip1818



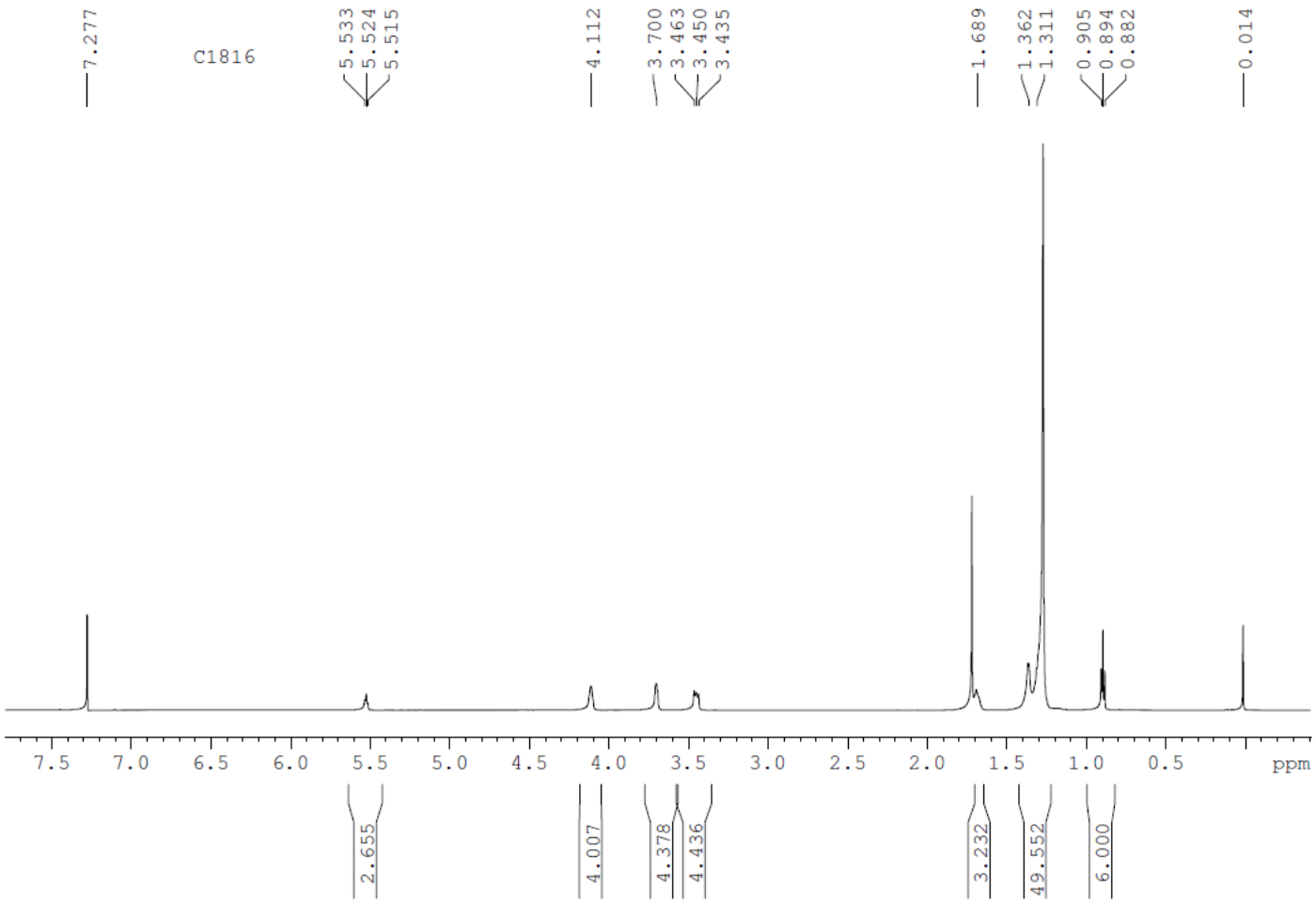
13C NMR of Lip1818



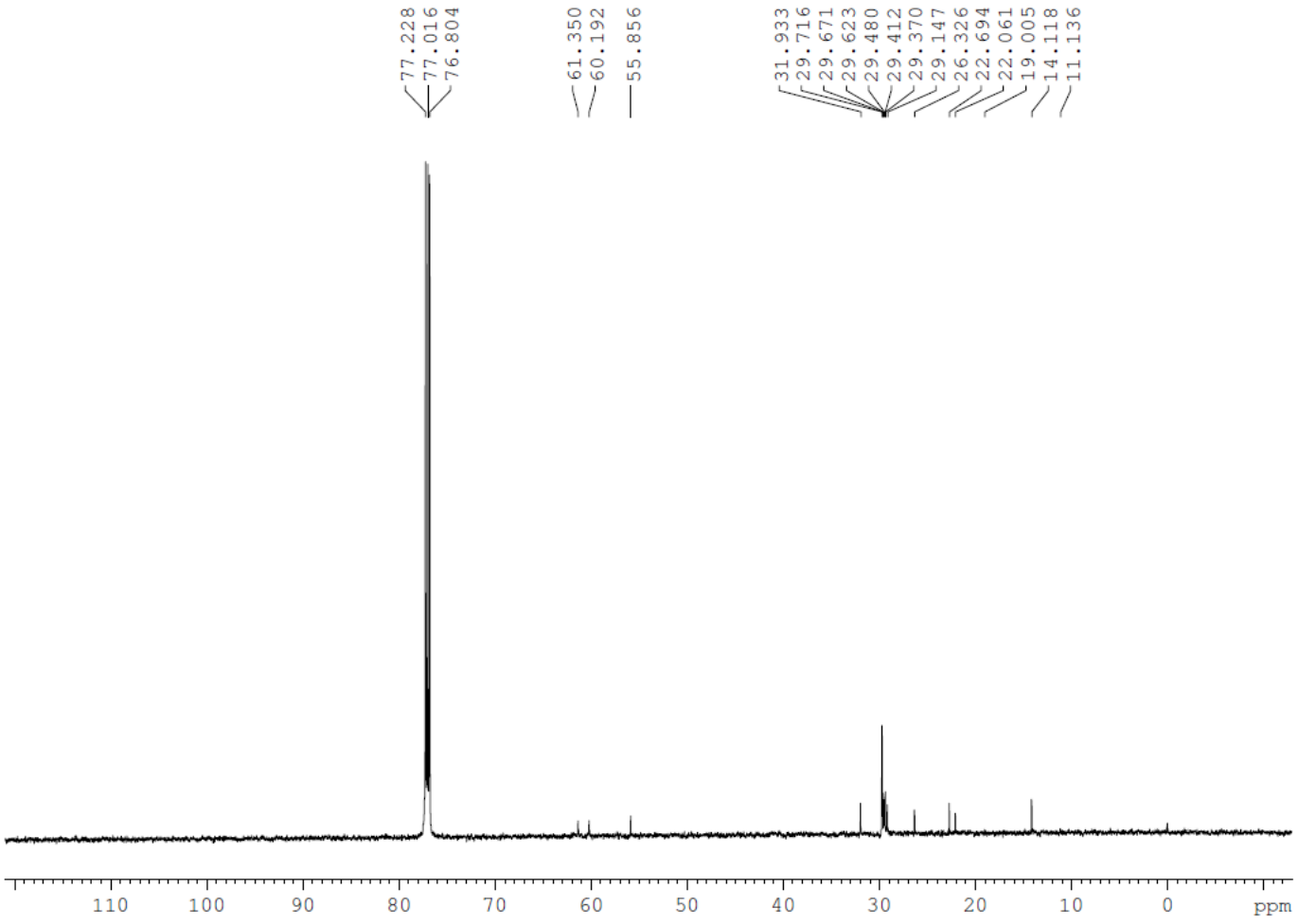
ESI-Mass of Lip1818



1H NMR of Lip1816



13C NMR of Lip1816



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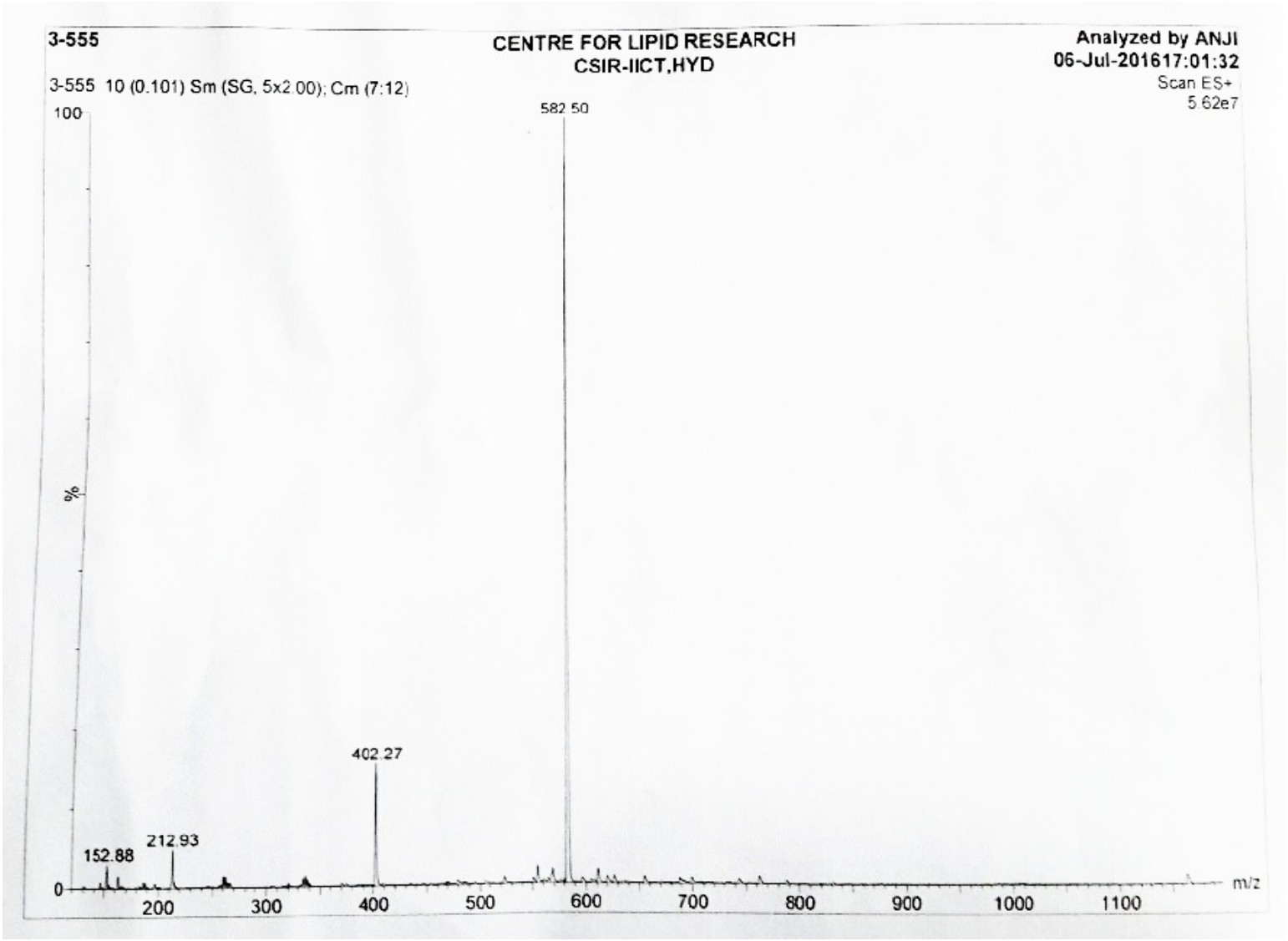
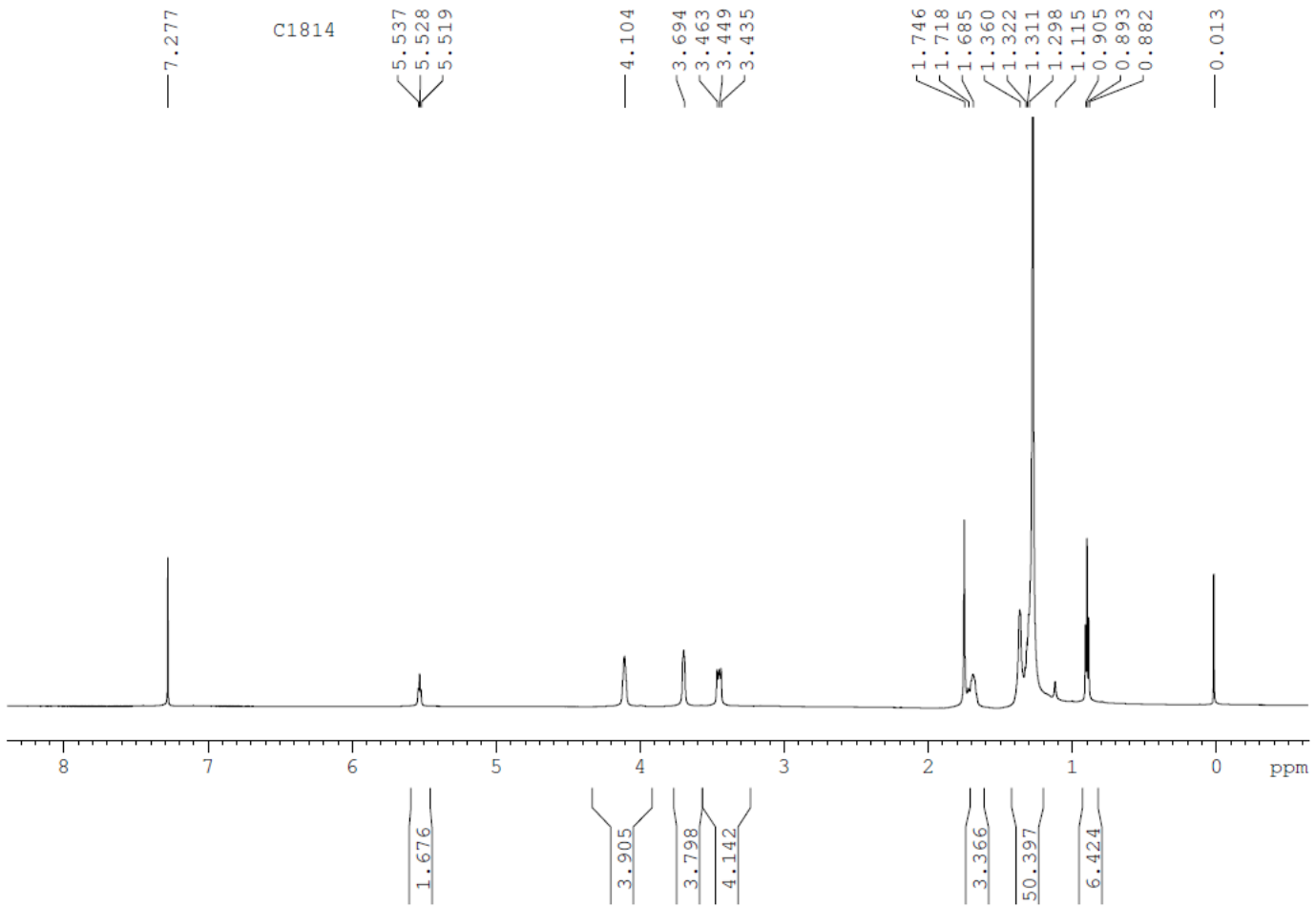
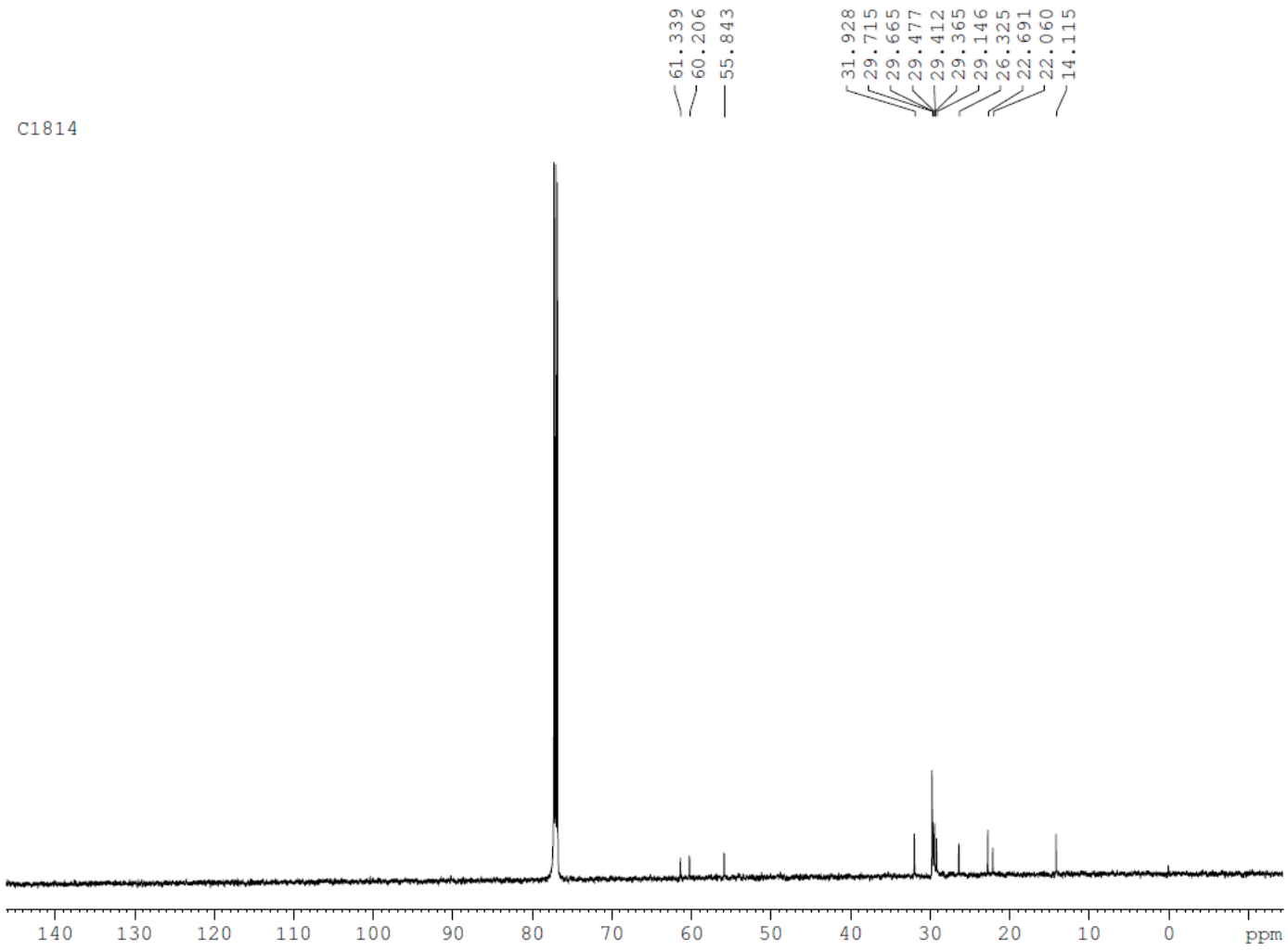


1H NMR of Lip1814



13C NMR of Lip1814



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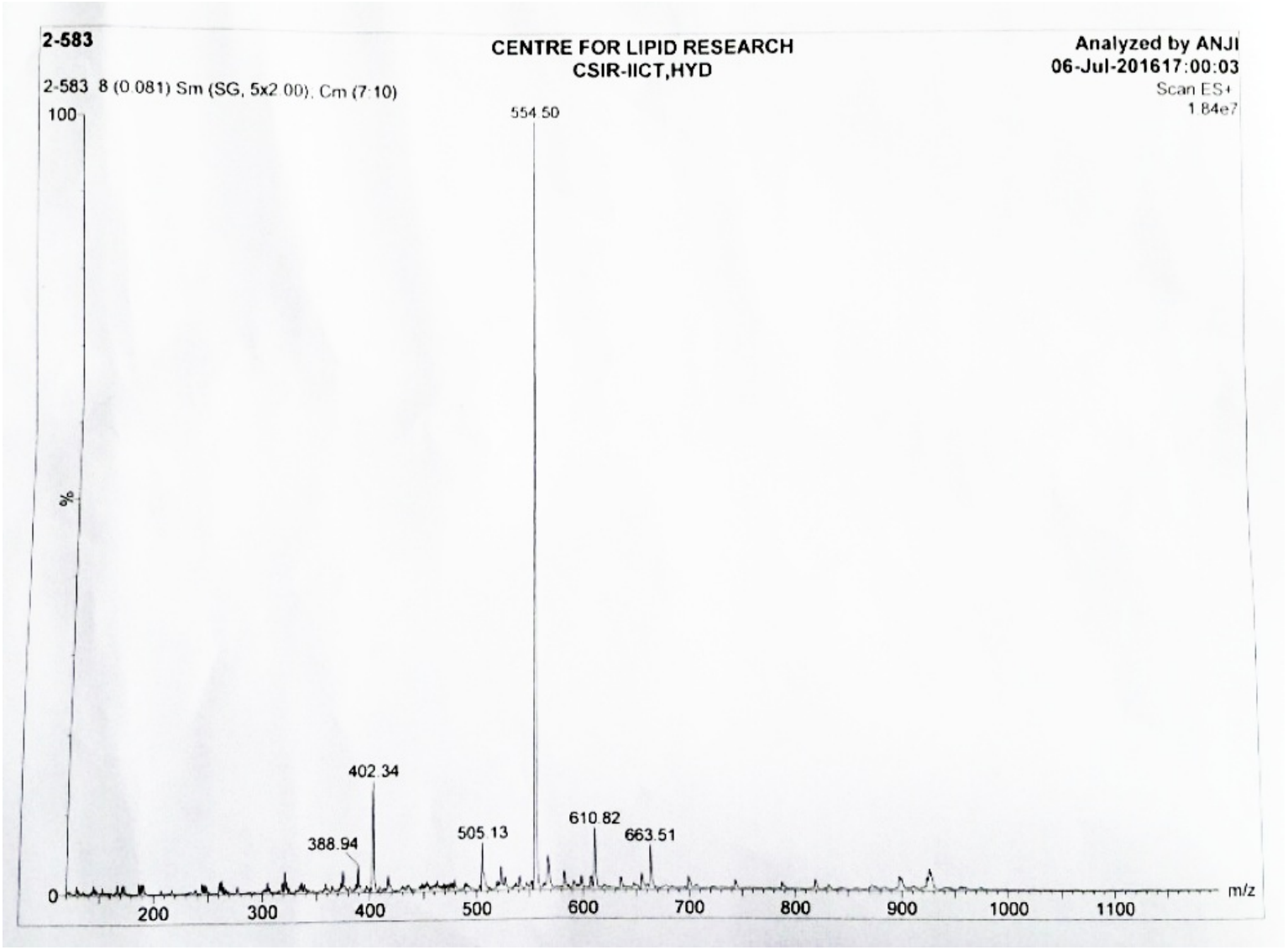
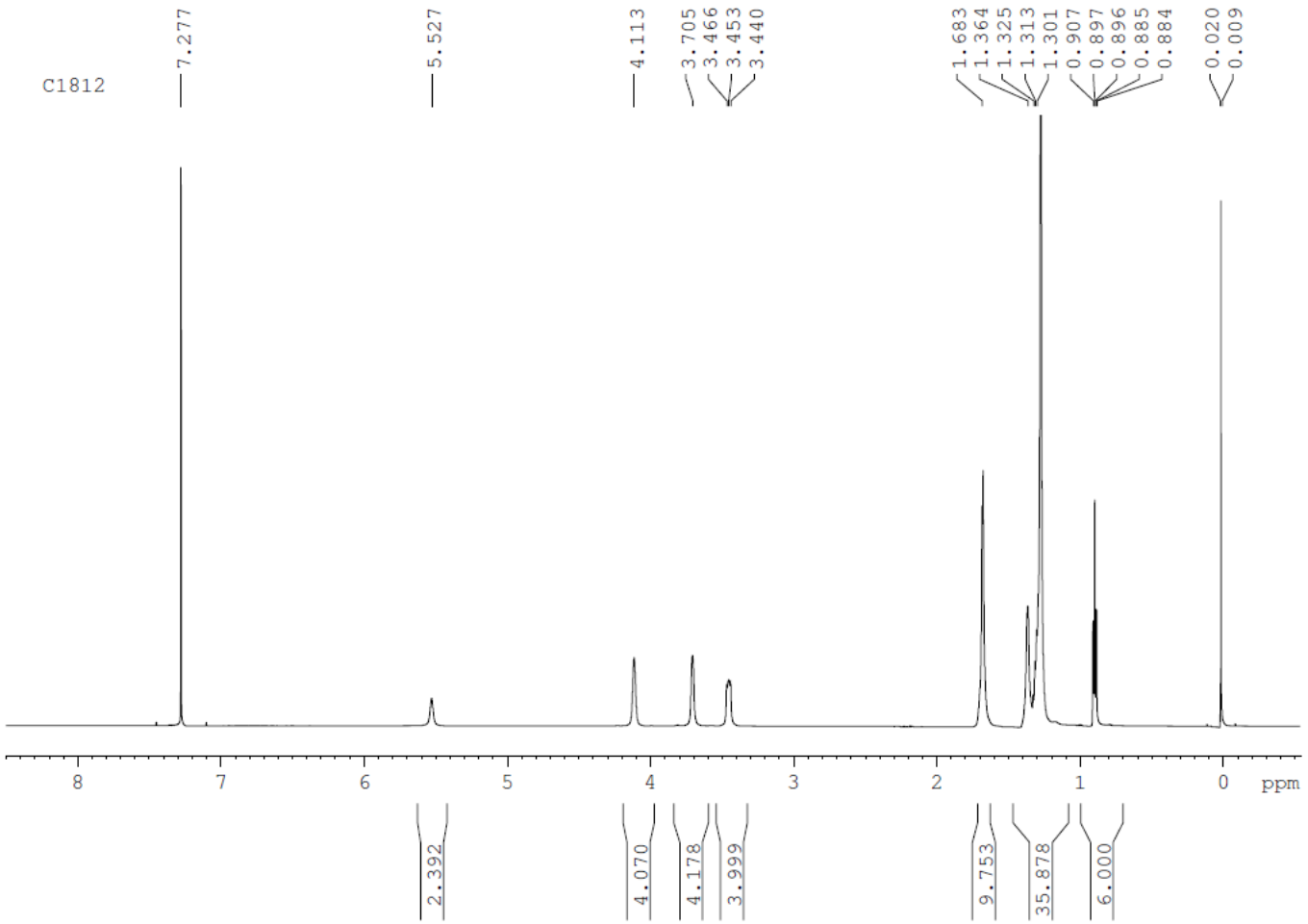
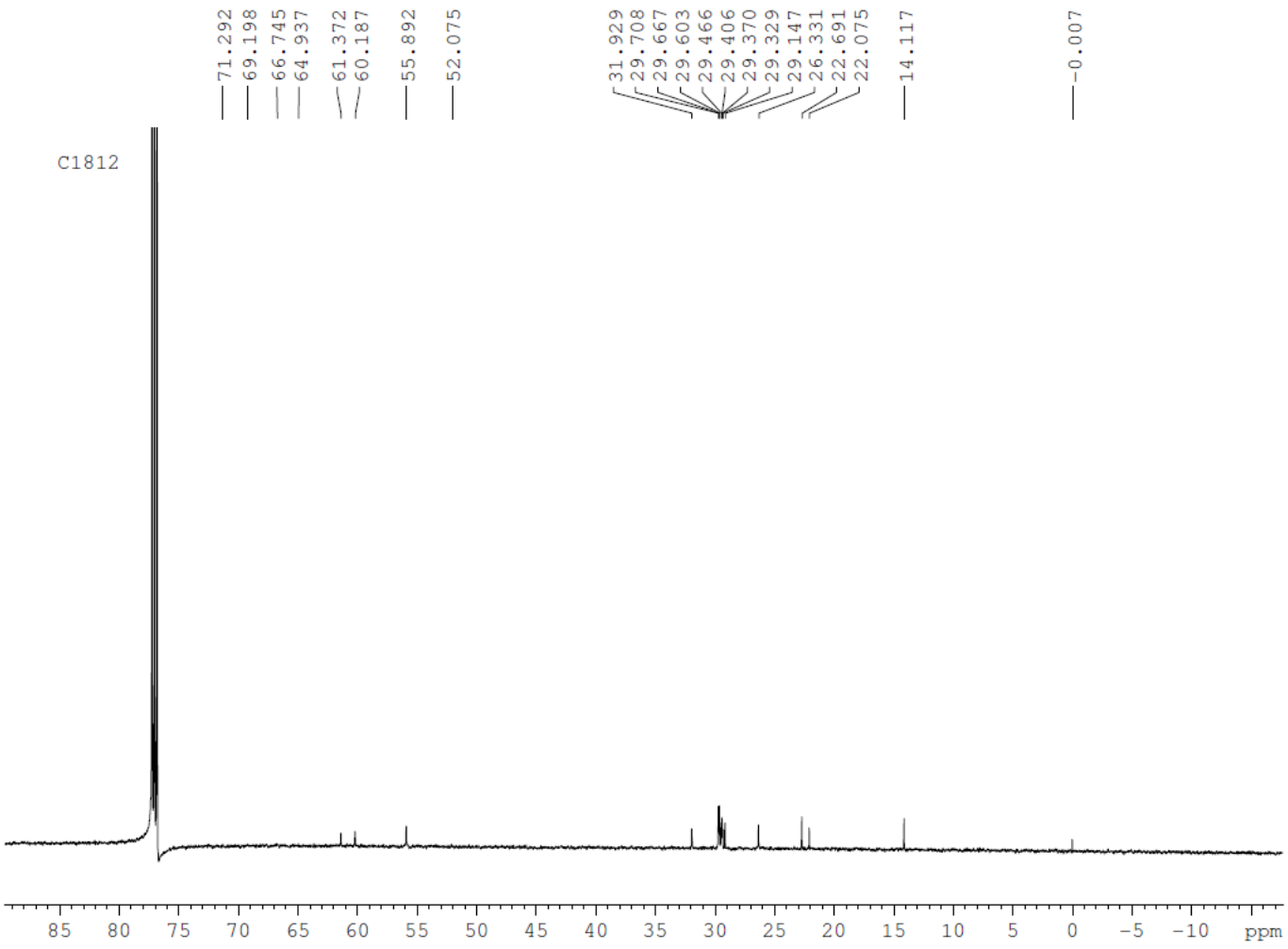


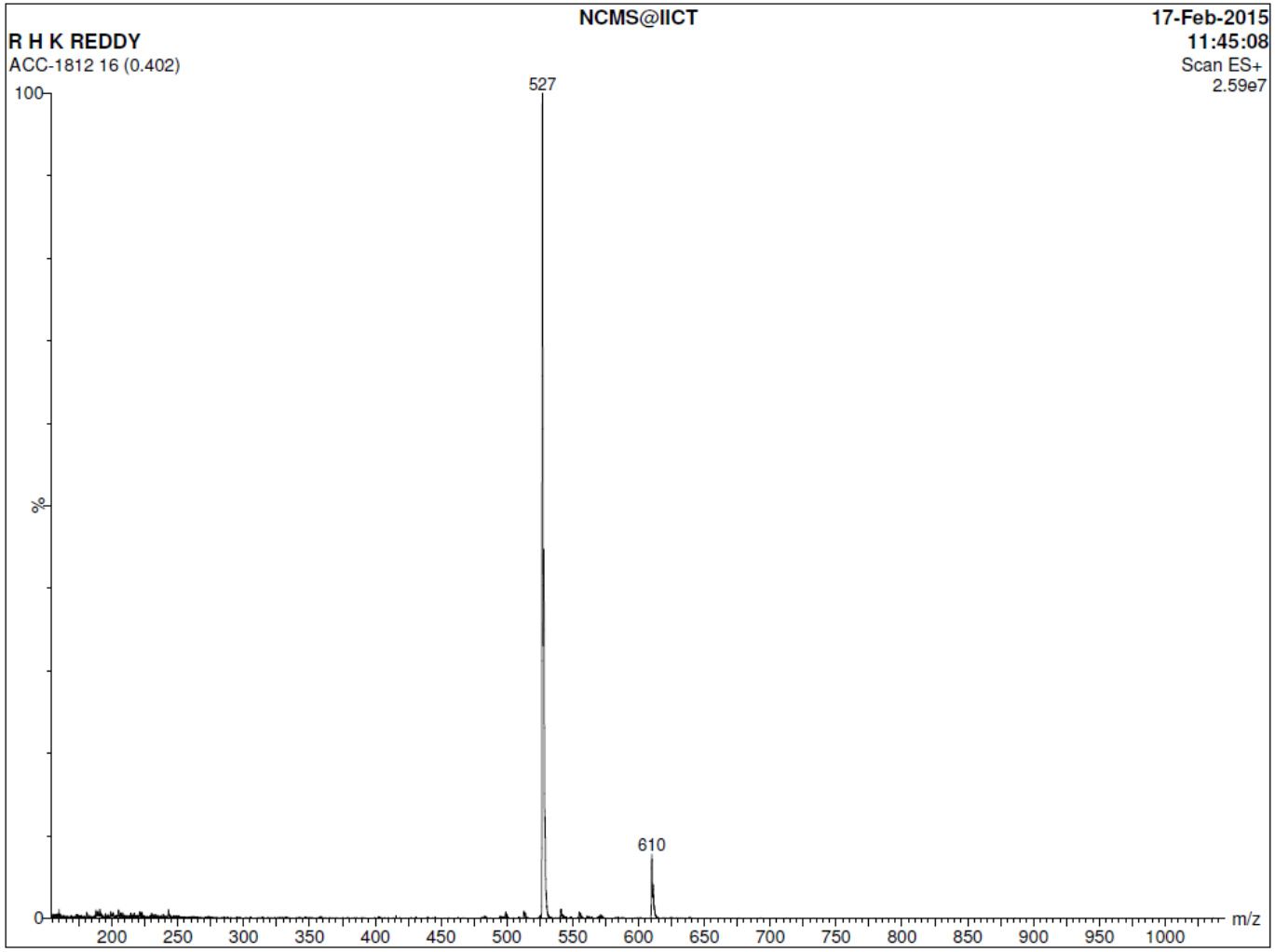
1H NMR of Lip1812



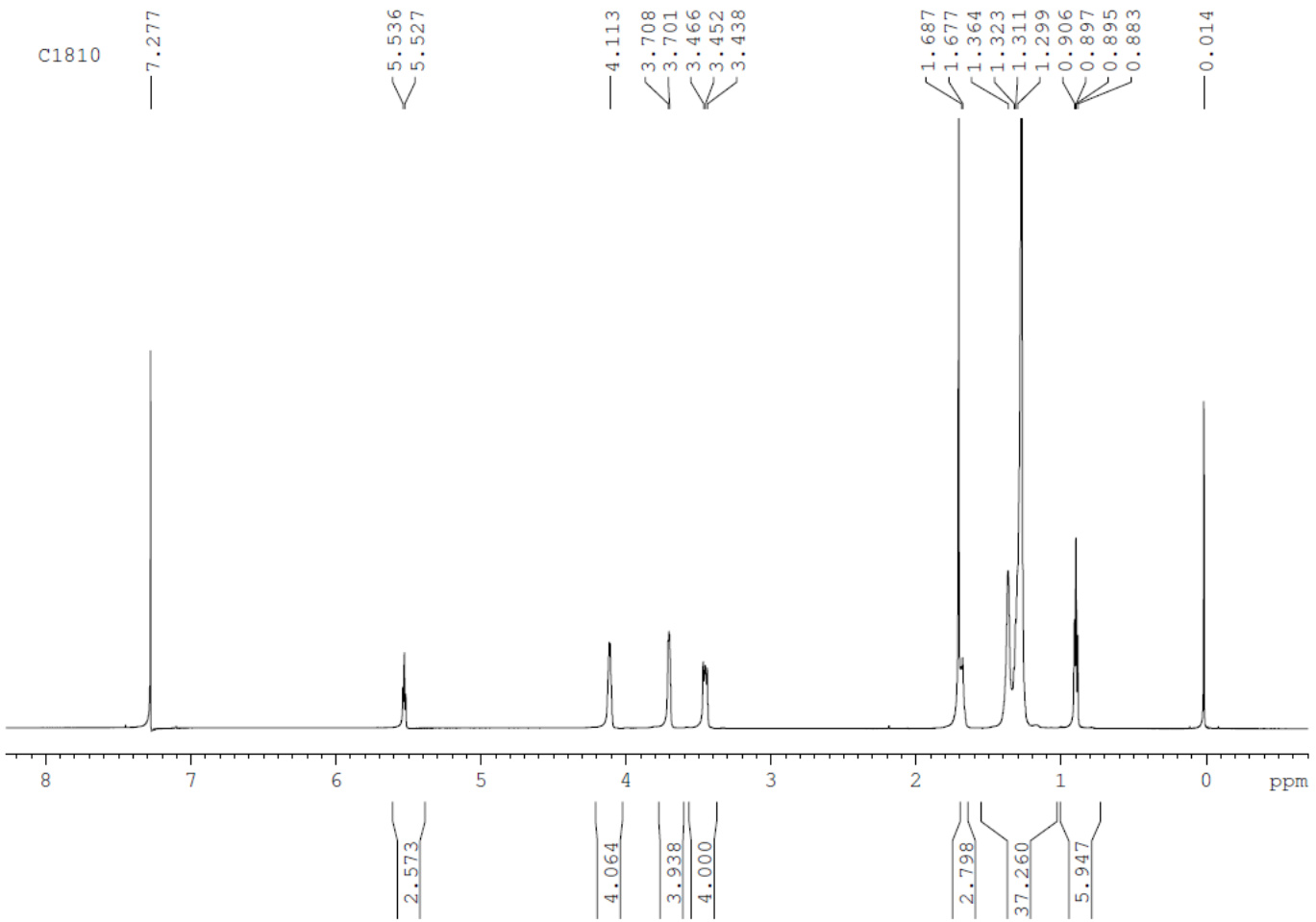
13C NMR of Lip1812



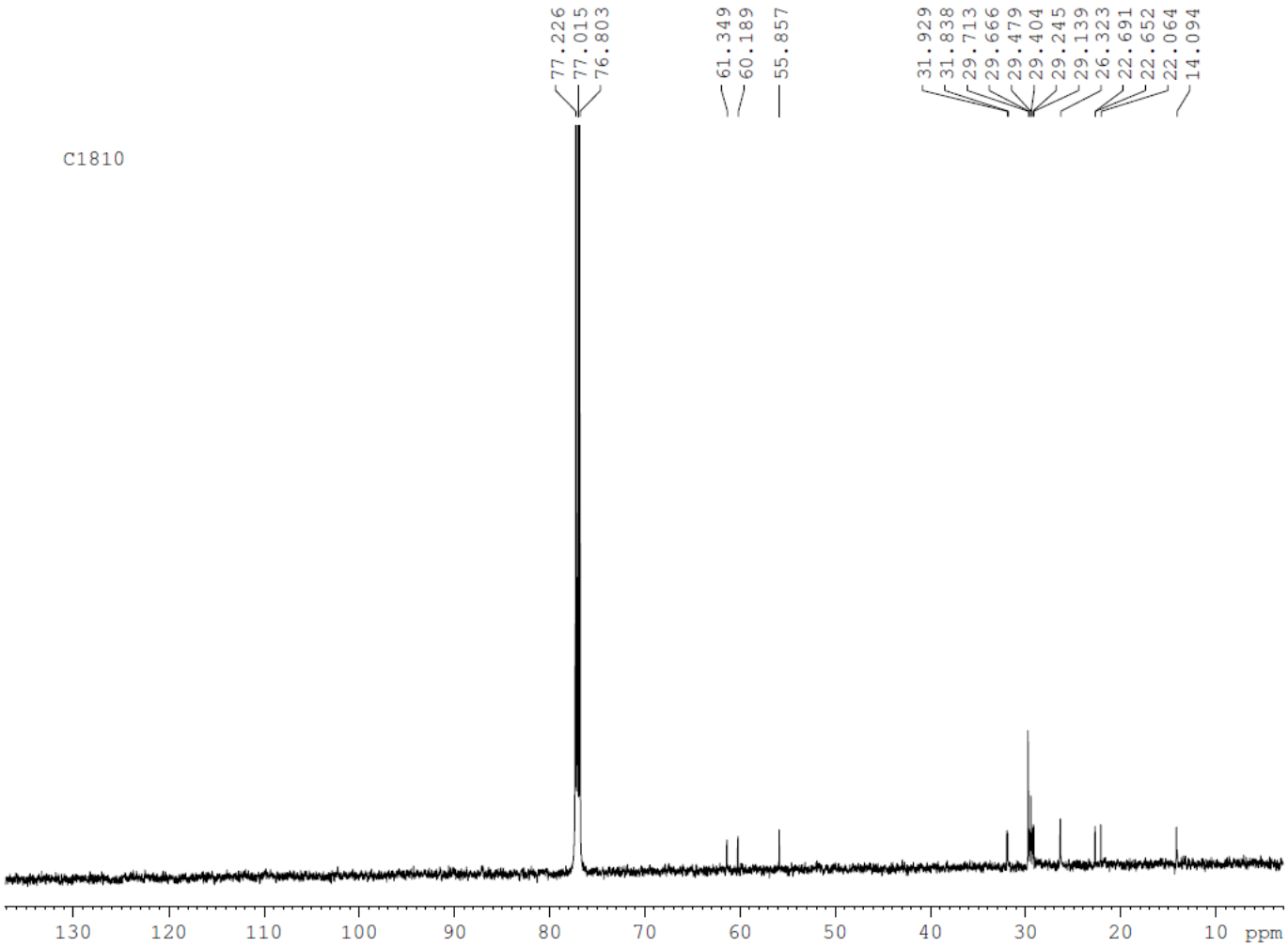
ESI-Mass of Lip1812



1H NMR of Lip1810

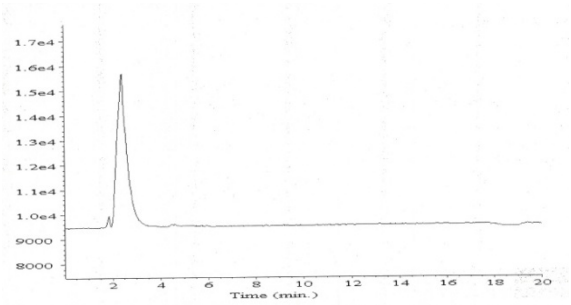


13C NMR of Lip1810

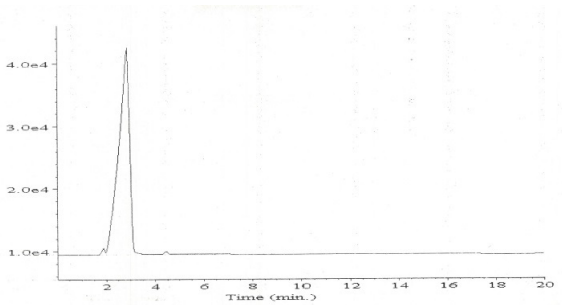


HPLC Profiles of lipids, Lip1818-Lip1810

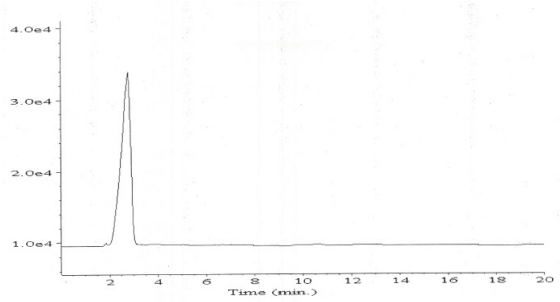
Lip1818



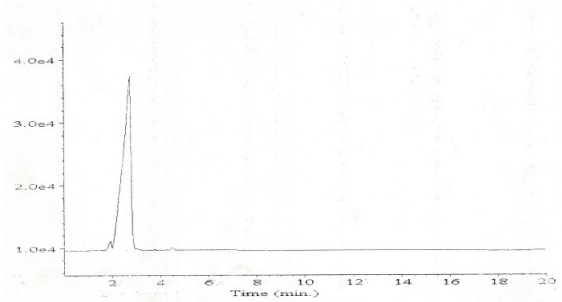
Lip1816



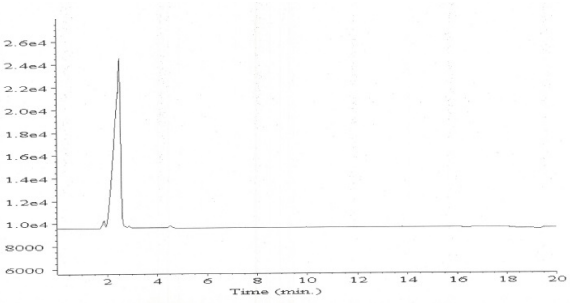
Lip1814



Lip1812



Lip1810



HPLC Conditions:

System:	Agilent 1100 Series HPLC Value System
Column:	LiChrospher® 100A, RP-18e (5µm), 125 x 4 mm
Mobile Phase:	100% Methanol.
Flow Rate:	2.0 mL/min
Column Pressure:	60-65 Bars
Detection:	Evaporative Light Scattering Detector (ELSD)