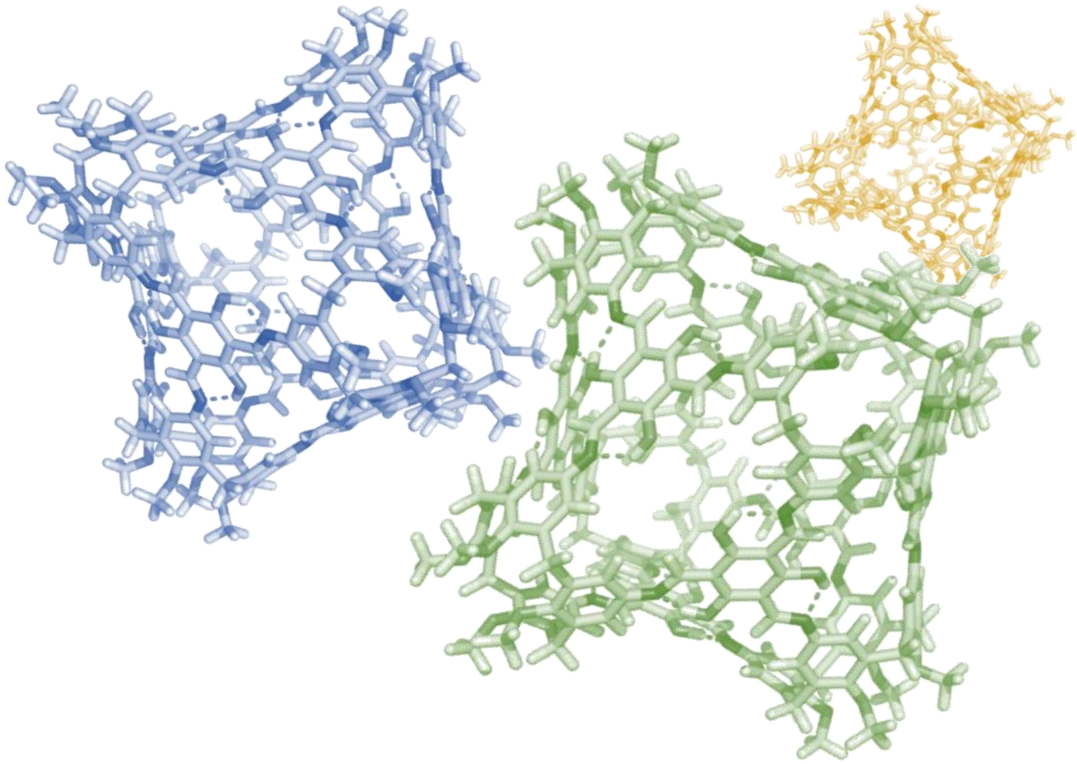
# SELF-IMMOLATIVE MOLECULAR CAPSULES



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# 1. Introduction

## 1.1 State of art: ¿Why is it necessary to encapsulate a molecule?

"*Supramolecular chemistry is the chemistry of intermolecular bonds, the one that implies the structures and functions of the formed compounds by association of two or more chemical specie*", Jean-Marie Lehn[1].

There is no doubt that defined cavities and confinement phenomena play a fundamental role in biology. For instance, liposomes, cellular organelles or virus capsids are examples of host systems, which are able to protect a guest inside; modify the reactivity or act as a molecular transporter. All of it drove chemists to emulate the incredible properties of the confined spaces for decades, finding the answer in systems called "molecular capsules" or "cages"[2].

Molecular capsules are a group of molecules that are assembled into a hollow system that can accommodate other compounds inside the cavity. They are usually classified according to the type of bond that keeps all the parts together: non-covalent, organometallic and covalent cages.

Molecular capsules displays a set of properties that make them really attractive for several reasons[3]:

- First of all, capsules allow interactions inside the cavity that hardly occurs by any other method.
- They increase the effective concentrations of the reactants, thus providing a higher rate of reaction.
- Guest is protected from the surrounding chemical environment.
- The cavity of capsules could be compared to the active site of an enzyme because of their structural and spatial characteristics.

Consequently, molecular capsules have been applied in many areas as containers/carriers[4], nanoreactors[3] or even molecular filters[5] since 90's, in competition with polymers and MOF's. Certainly, encapsulation could be the answer to actual challenges in different fields like stabilization of reactive/non stable compounds[5,6], selective recognition of guests[7,8], permanent porous materials[9,10] and catalysis of different types or reactions[11,12].

An interesting field where capsules could be used is in targeted drug delivery. There are clear advantages for this: protection of the drug, that can travel along the body until it is selectively released. In fact, other classes of carriers have been used with the same purpose in different cellular environments: liposomes, dendrimers, polymeric nanoparticles and even empty virus capsids[4,13]. A priori, molecular capsules display several advantages for their potential use as smart transporters in contrast with abovementioned carriers:

Note: Generally, "cage" is reserved for less closed structures, despite all of them are confined systems. Throughout the document the word "capsule" will be used to accurate a better understanding.

- They have a controlled size.
- Almost all chemical residues are controllable and modifiable.
- The exact number of drug molecules per carrier can be known by conventional NMR or mass spectrometry.
- The use of smaller carriers as delivery vehicles should help in tissue penetration and they are normally not immunogenic.
- Capsules are susceptible to be synthetized through classical organic chemistry, reducing manufacturing costs.

However, no examples of molecular capsules for targeted drug delivery have been described. The main reason is probably the nature of the bonds that keep the capsule together. Indeed, non-covalent structures are usually too labile, and therefore they are in continuous reversible equilibrium, encapsulating and releasing the host. Of course, the drug will be partially released before reaching its final destination. On the other hand, organometallic cages are much stronger, but the transition metals employed often display toxicity problems, as well as the required stimuli for opening the capsule are non-biocompatible. Covalent molecular capsules are obviously very strong and stable in plenty of biochemical environments. However, there are little precedents of a covalent cages able to respond to a stimulus.

An interesting approach and the objective of this work would be the design of an efficient method for building up a covalent capsule able to respond selectively to a stimulus which would degrade it, releasing the content. However, before continuing it is advisable to make a short review of molecular capsules, in order to understand them better.

## 1.2 Molecular capsules: definition and types.

### 1.2.1 Non-covalent capsules

*H-bond capsules:* Biological systems have used this type of bond in developing amazing structures, for examples the quaternary structure of proteins or the viral capsids consisting of linked protomers. The formation of H-bond-based non-covalent systems has been a remarkable hit in supramolecular chemistry, although still far away from the selectivity and robustness achieved by living organisms.

The dynamic nature of H bonds is well known. It depends on multiple factors such as pH, temperature and the solvent. The final outcome is a constant opening-closing equilibrium for the capsule. This ensures that there will always be some host inside the capsule,[3] but it is also a serious drawback. Indeed, such promiscuity in bond formation/breaking avoids applicability in targeted drug delivery, as well as any other application that requires the temporary or permanent confinement of a substance.

A brief example of these supramolecular systems can be the work done by Rebek et al[14], based on two identical units of diphenylglycolouryl which adopt spherical symmetry when they are oriented perpendicularly establishing 8 H bonds. This structure was dubbed "tennis ball" because it has two concave halves reminiscent of this figure. It was the first example of a non-covalent molecular capsule. Guests for such a system can be solvent molecules such as dichloromethane or gases like methane, ethane and ethylene. (*Figure 1*).

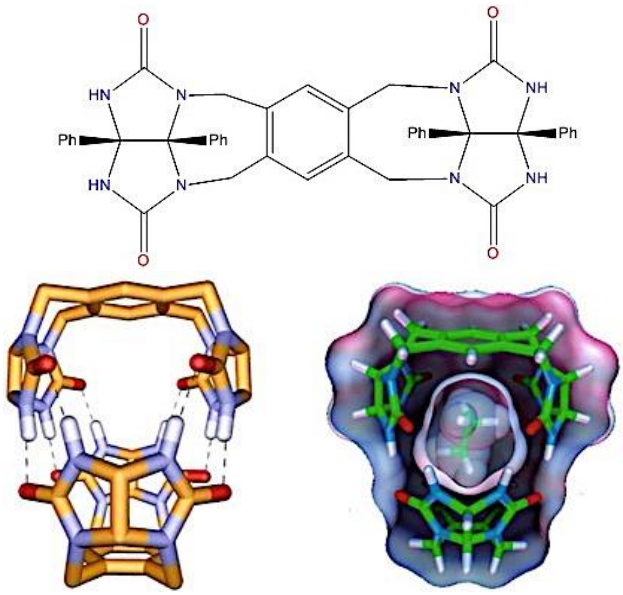



Figure 1. Upper: Structure of the basic unit. Lower: Non-covalent tennis ball capsule and the same with an ethane molecule inside.

Another example in this field and also classical structures in supramolecular chemistry are the resorcin[n]arenes. These structures are easily synthetized from resorcinol and any aldehyde in the presence of a strong acid. MacGillivray et al.[15] discovered how this compounds can self-assemble when they are solved in wet-benzene incorporating 6 molecules of water to build up a hexameric structure with a net of 60 H bonds. The cavity is able to accommodate up to 8 molecules of chloroform. (*Figure 2*).

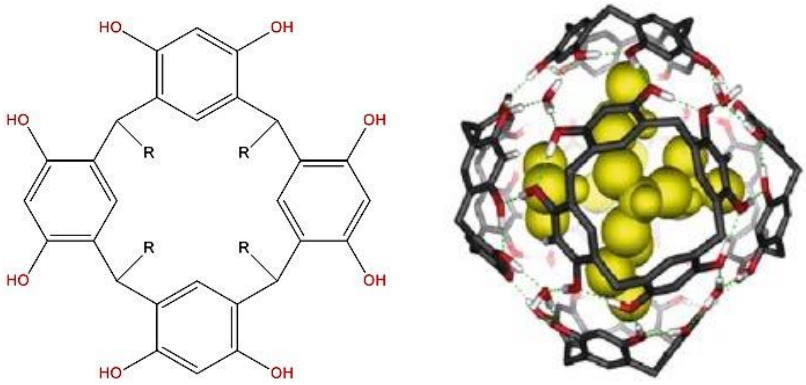



Figure 2. Left: Resorcin[4]arene structure. Right: Hexameric capsule containing 8 chloroform molecules.

An additional example of popular structures used in this area are the calix[n]arenes. These compounds were coined such a curious name because of their spatial geometry: the aromatic rings remind us a calix motive. They have been widely included in the synthesis of molecular capsules. Rebek's[16] work is a perfect example: a urea-derivative of calixarenes is able to dimerize into a capsule (*Figure 3*) where urea moiety acts simultaneously as H bond donor and acceptor.

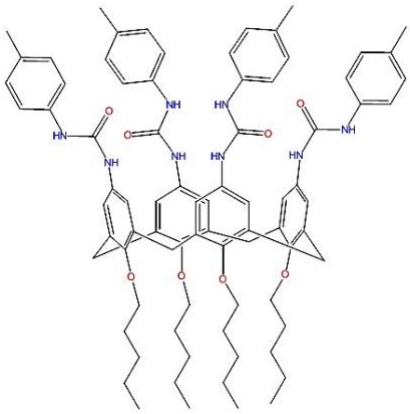


Figure 3. Calix[n]arene-amide used by Rebek[16].

*Ionic capsules:* Ionic supramolecular systems are still a developing field. Just a few examples can be founded in the literature. The vast majority are calix[n]arenes derivatives with polar groups[17,18]. The main advantage is the synthesis in mixtures of water with polar solvents like MeOH.

It is remarkable the formation of organometallic complexes inside the cavity of these structures[19] where applicability in catalysis is very likely; or the encapsulation of two partially polar molecules[20] in a hetero-capsule built up with an amine-calix[6]arene and a cyclotriveratrylene with acid groups (*Figure 4*).

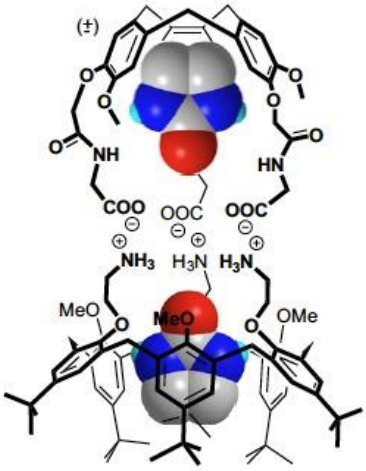



Figure 4. Heterodimeric ionic capsule containing 2 molecules of 2-imidazolidinone.

### 1.2.2 Organometallic capsules:

Most of the molecular capsules developed up till now are based on organometallic coordination complexes. These systems are generally build up with transition metals chelated by organic ligands, adopting different well-defined geometries. Probably, coordinative self-assembly is one of the most advanced methods for the facile preparation of large capsule-like supramolecules[21–23]. Indeed, one of the triumphs of coordination chemistry over the past twenty years has been the progression from simple transition-metal ligand complexes to increasingly complex functional supramolecular structures, which are suitable for a wide range of applications from sensing to catalysis.

One of the most cited authors in this area is Makoto Fujita[24]. This researcher and co-workers have developed organometallic supramolecules with the general formula $M_nL_{2n}$, where M is usually $Pt^{2+}$ or $Pd^{2+}$ and L is angular bis(pyridine) ligands which are able to make square planar metal complex, generating self-assembled structures displaying a huge variety of sizes and shapes. Some of the structures achieved are showed in *Figure 5*, where the metal ions are placed in the corners of the figures and the organic compounds in the edges:

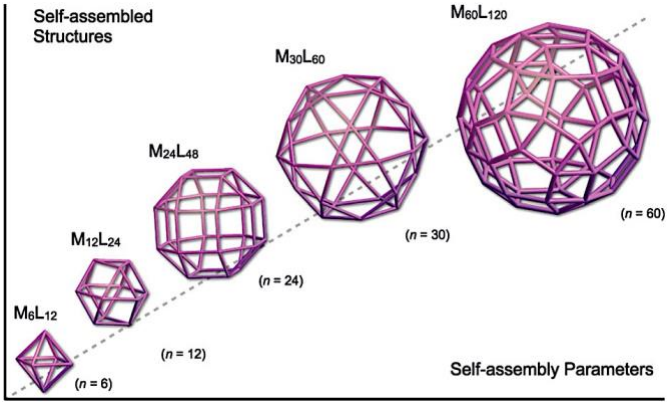



Figure 5. $M_nL_{2n}$ type organometallic capsules achieved by Fujita et al[24].

A few applications proposed by Fujita et al[24] range from encapsulation of small compounds like ions, fluoroalkanes or fullerenes, to host a protein in the cavity or the template effect in the synthesis of organic polymers and nanoparticles. Most of the mentioned works take advantage of a sensible functionalization the bis(pyridine) ligands (*Figure 6*).

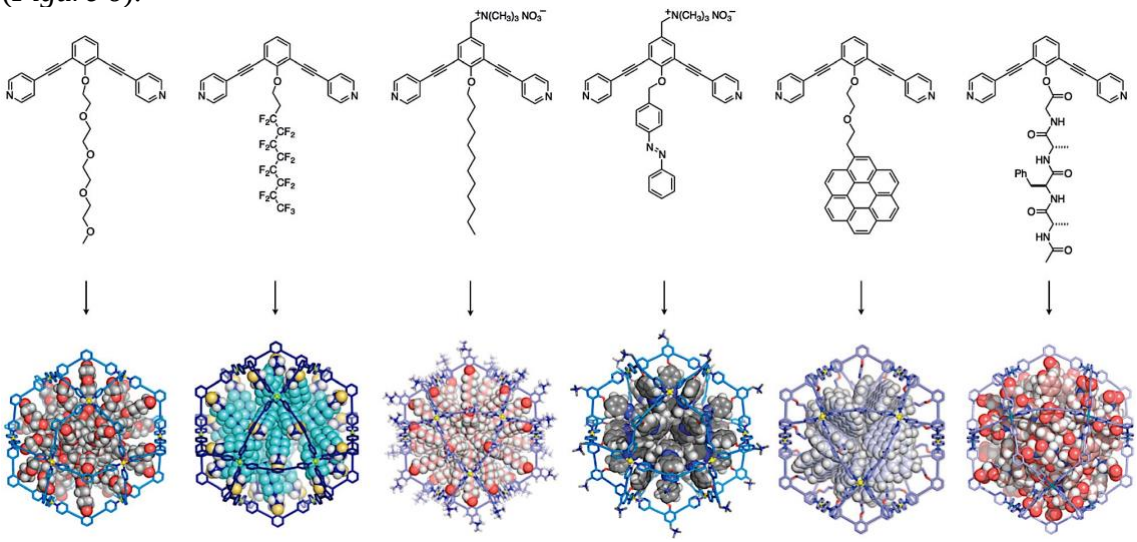


Figure 6. Organometallic capsules functionalized by Fujita et al.

Another benefit proposed for these capsules by Fujita is their industrial applications. However, it should be taken into account that their applicability in biological systems is restricted due to the toxicity of Pd ions.

### 1.2.3 Covalent capsules:

Covalent organic capsules are those whose linkages are formed by covalent bonds.[25] Even though their strength is an advantage for multiple applications, there are two main disadvantages for this type of cages: the first one is the lowest general yields obtained in

their synthesis, in comparison with their non-covalent analogues. Considering this drawback, two different approaches have been taken:[26]

- Dynamic covalent bond formation: these processes require the construction of reversible intermolecular linkages, which means that reactants interact establishing a forming-breaking equilibrium until the thermodynamically most-stable product is reached. Imines are the most representative functional group in this field and they are obtained after the condensation reaction between amines and aldehydes. As an excellent example it is worth mentioning the work developed by Cooper et al (*Figure 7*).[27]

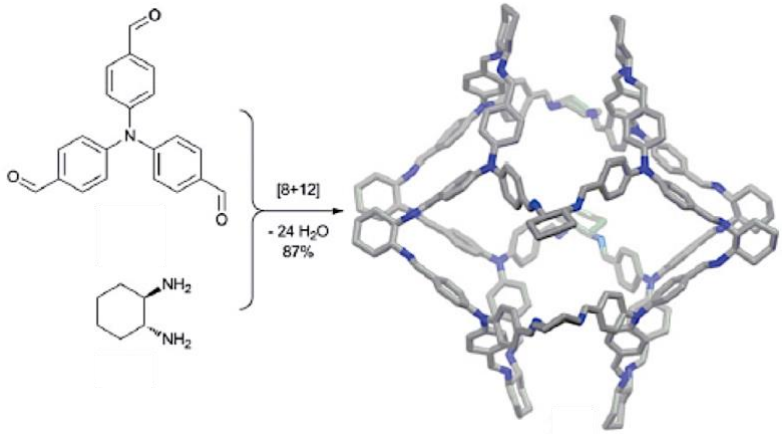



Figure 7. Covalent capsule designed by Cooper et al[27].

Possible alternatives to imines synthesis in dynamic covalent chemistry are combinations of the mentioned functional group with boronic esthers[28] or olefin metathesis[29].

- Irreversible bond formation: amide bonds are one of the options used, for example in the receptor synthetized by Davis et al. (*Figure 8*).[30] Aromatic nucleophilic substitution[31] and C-C bond formation[32] are also commonly used for this purpose.

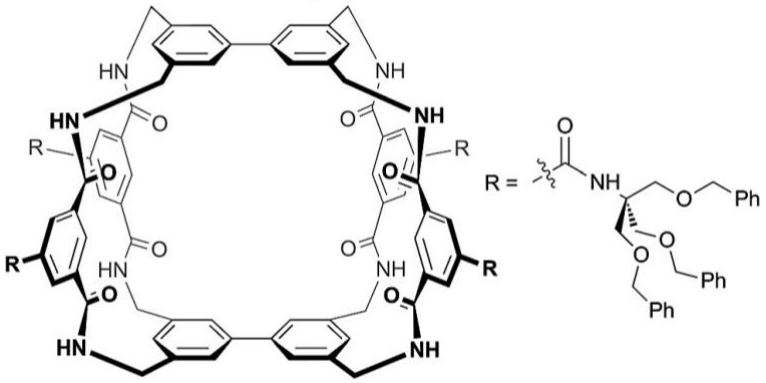



Figura 8.Davis et al[30] covalent capsule with amide bonds.

It should be also highlighted two recent methodologies in this area: firstly, Rivera and Wessjohann[33–35] developed a one-pot Ugi-4CR multicomponent reaction, that implies an amine, an ketone/aldehyde, a carboxylic acid and an isocyanide to obtain peptoids that recall carcerands structure. The second methodology has been developed in our group and it is based on the ionic thiol-ene reaction, from thiols and acrylates.[36]

The second problem of covalent capsules has to do with their potential use in biomedicine. Up till now, these capsules are considered as non-versatile structures because they are not able to be opened or closed. Introducing a suitable moiety for the selective response against a certain stimulus, covalent capsules could become into excellent carriers and they could applied in smart drug delivery. There are a few previous examples about, but in all cases the stimulation mechanism is not easily biocompatible: UV-radiation[37,38], high temperatures[39] or non-biological redox reactions[40,41].

For this reason, an interesting approach would be the combination of the robustness of covalent capsules with any efficient method for the degradation in the presence of a stimulus. In this regard, one of the most versatile methods for the controlled cleavage of covalent bonds is the self-immolative chemistry, which will be explained in the next section.

## 1.3 Self-immolative systems.

Self-immolative systems consists of a relatively recent high-efficiency methodology focused on the controlled cleavage of covalent bonds. In particular, once the correct stimulus is present (it could be chemical or biological), an electronic cascade will occur, that ends up with the degradation of the original compound and the release of multiple molecular fragments. The typical structure of a self-immolative moiety is a phenol (or an aniline) with a good leaving group in a methylene at ortho or para position. When such a phenol is protected, the system is stable, but once it is deprotected, an electronic rearrangement occurs that ends up with the release of the leaving group (*Figure 9*).

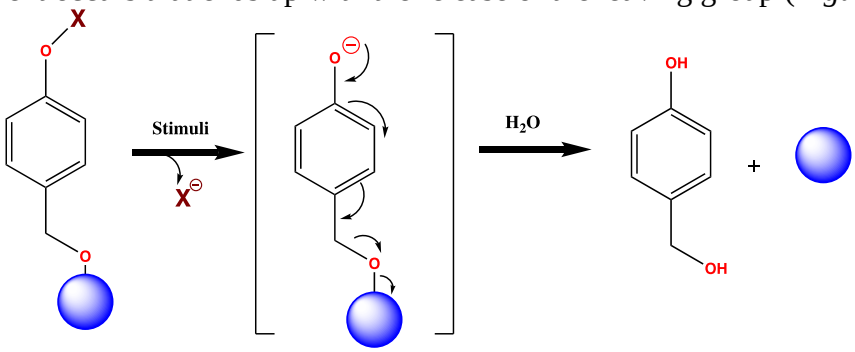



Figure 9. Example of self-immolative system.

This type of systems has been widely applied as prodrugs. Blencowe et al.,[42] propose a simple way to understand the concept of self-immolative chemistry, as well as the different mechanisms involved, by a classification in 3 main families:

- Self-immolative elimination: based on the spontaneous and irreversible decoupling of a multicomponent compound in its constituent fragments through a sequence of electronic movements. These phenomena its favoured by two reasons: an increasing of system's entropy coupled to the formation of thermodynamically stable products. An illustrative example is the lipase mediated 1,6-elimination carried out by Sauerbrei et al[43] (*Figure 10*):

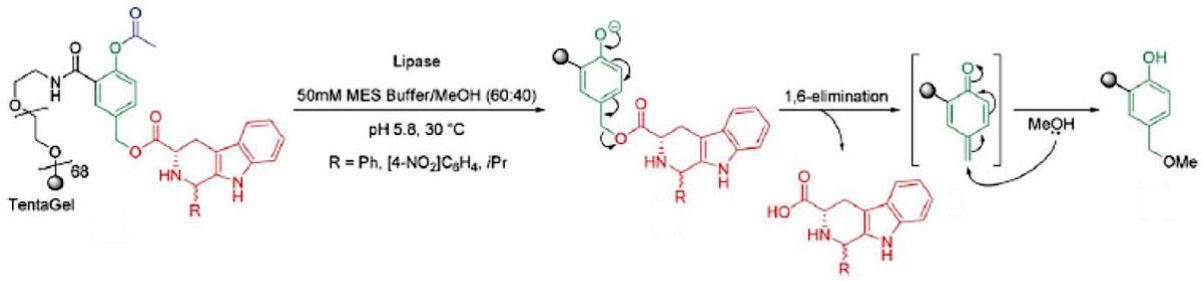



Figure 10. Example of lipase-mediated autoimmolative 1,6-elimination.

- Cyclisation elimination: a latent nucleophile present in the molecule produces its fragmentation once activated, followed by the displacement of a near carbonyl group. This process is again favored by an increase of entropy and the generation of thermodynamically stable products. An example is the p-nitrophenol releasing-system developed by Amir et al[44] (*Figure 11*).

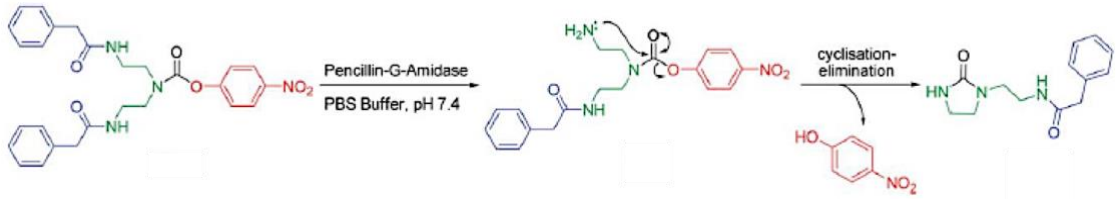



Figure 11. Example of elimination by cycling.

- Amplified self-immolative elimination: this class of mechanism reminds the first type mentioned but it implies a little difference: it only needs one activation event to trigger a multiple self-immolative event. A good example is the consecutive release of identical fragments of 2-phenylethanol triggered by the reduction of the nitro group, proposed by De Groot et al[45] (*Figure 12*).

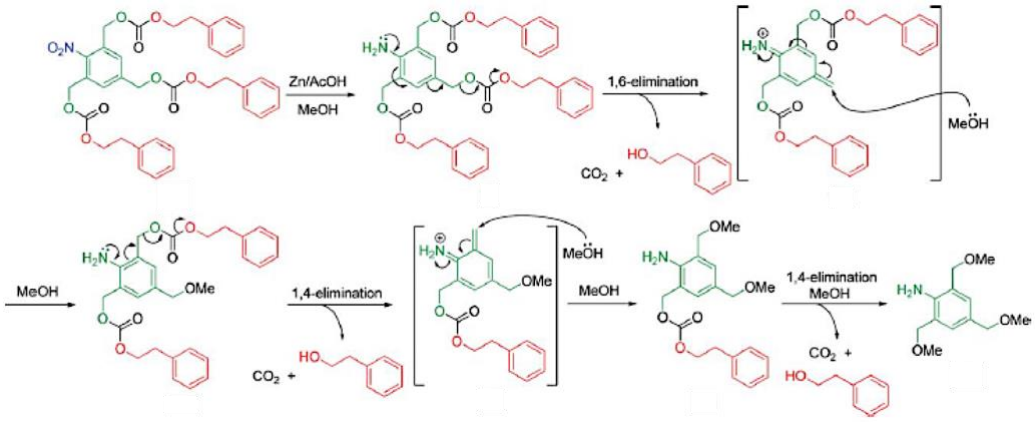



Figure 12. Example of amplified self-immolative elimination.

Considering all this, an interesting approach for the applications of molecular capsules on smart drug delivery (and several other applications) could be the synthesis of capsules incorporating self-immolative moieties. There are no precedents of such responsive systems, and therefore, this work represents a huge scientific challenge. However, the mentioned approach is very promising since this type of structures will combine the intrinsic robustness of covalent capsules with the selective degradation in response of a stimulus typical of self-immolative systems. This research line will be the main objective of this final master dissertation.

## 2. Objective.

The design of a self-immolative covalent molecular capsules and the spectroscopic studies involved: characterization and opening in the presence of an appropriate stimulus.

# 3. Developed work.

## 3.1 Click chemistry: The thiol-ene reaction.

Click reactions have been widely used since Sharpless et al[46] described it in 2001. The main characteristics for this type of reactions are: high yields with easily removable by-products (non-chromatographic processes) if any, regio- and stereospecificity, insensitivity to oxygen and water, mild solventness or aqueous reaction conditions, orthogonality with usual organic synthesis reactions and availability to a wide variety of starting compounds and complies the atom economy principle. Nowadays, the most extended sort of reaction is the Copper-Catalyzed Azide-Alkyne Cycloaddition (CuAAC) (*Figure 13*).

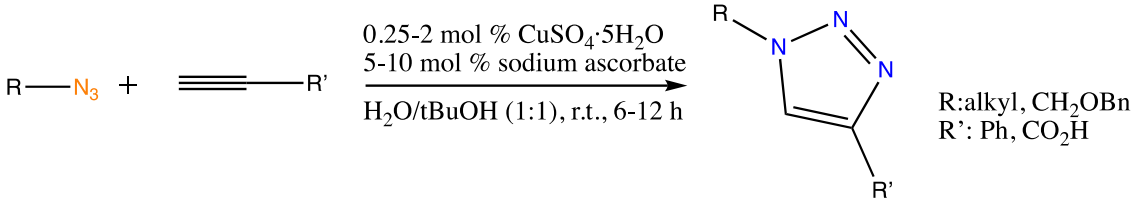



Figure 13. Copper-Catalyzed Azide-Alkyne Cycloaddition (CuAAC).

The thiol-ene reaction is also considered a click reaction due to the highly efficient reaction of thiols with reactive C-C double bonds or "enes". There are two types of thiol-ene reaction, although the mechanism of the reaction is not well defined yet (*Figure 14*):

1) Thiol-ene free radical addition to C-C double bonds.
2) Thiol Michael addition to electron-deficient alkenes.

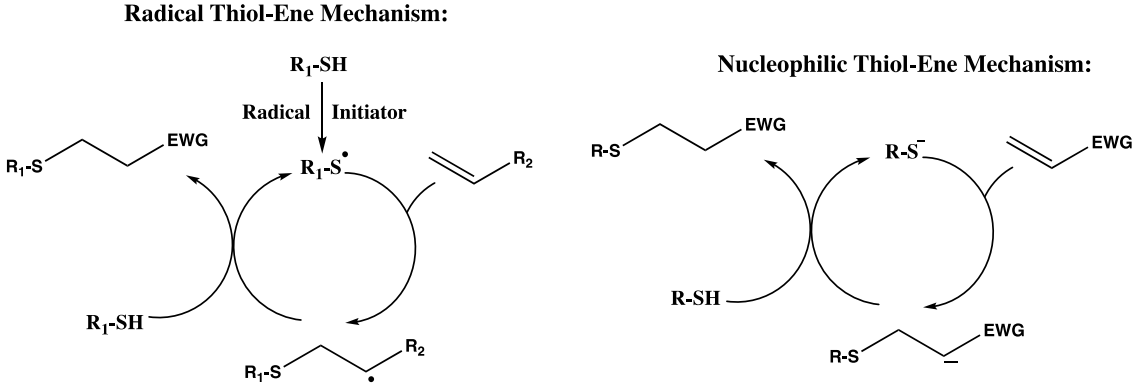



Figure 13. Mechanisms of the two thiol-ene reactions.

Both of them have been widely employed in organic synthesis, polymer chemistry and materials science[47–49]. In our cae, we have use the ionic thiol-ene reaction (also called thiol Michael addition click reaction) as a very powerful method to synthesize covalent capsules.

## 3.2 Self-immolative molecular capsules.

As it was above-mentioned, we have taken advantage of the ionic thiol-ene reaction to efficiently synthesize covalent molecular capsules.[36]

Specifically, this methodology will be applied to a triple thiol-ene reaction trying to assemble two different capsules, from a tri-thiol and a tri-acrylate units, incorporating a self-immolative moiety (*Figure14*).

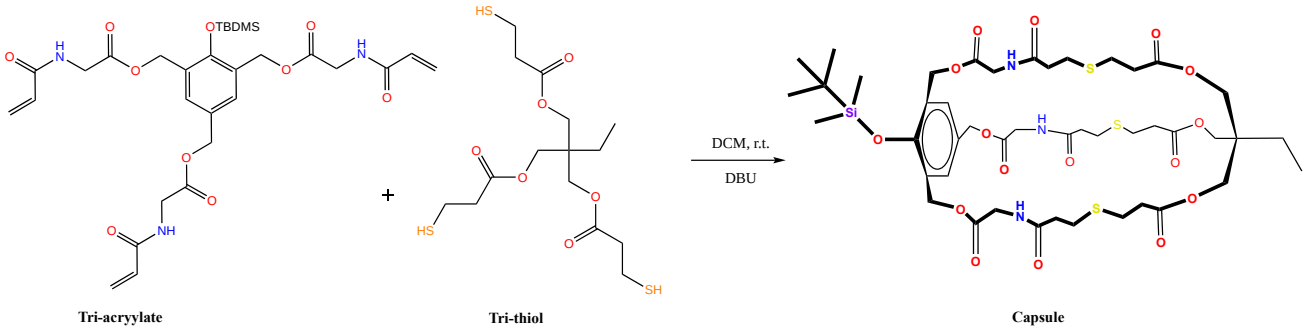



Figure 14. Synthesis pathway for a self-immolative molecular capsule.

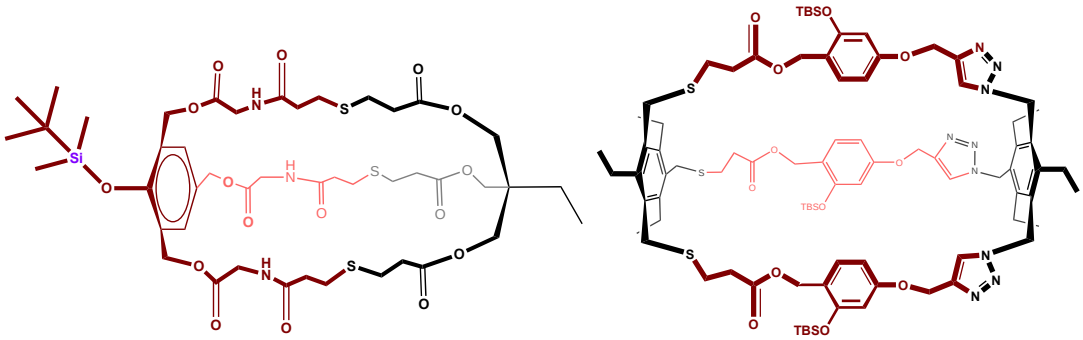



Figure 15. Proposed self-immolative molecular capsules: C-1 (left), C-2 (rigth).

The proposed capsules are shown in *Figure 15*. Due to the presence of a silyl ether protecting group in the phenol, the fluoride ion would act as the stimulus that triggers the self-immolative response. The driving force is the formation of F-Si bond, the strongest bond in chemistry, so, it should act as a thermodynamic irreversible process producing the opening of the molecular capsule.

The studies for checking that proposal will be carried out by NMR spectroscopy and they will be discussed in section 4.

## 4. Results and discussion.

The results related to the synthesis of compounds preceding the capsules were good in general terms, showing a variety of yields depending on the reactivity and nature of the reagents. Some of the reactions developed should be improved, for example, the formation of the acryl-amide precursor for the capsule in Figure 14 (only a 10% of yield).

Focusing on the capsule-formation reactions, they imply the triple specific formation of bonds between the two reagents (Figure 14), therefore, they should react one-to-one in an intramolecular way and avoiding intermolecular reactions once the first thiol-ene bond is formed. For that reason, obtaining the structure of a capsule is more difficult than their open analogues (polymerization). In some cases, the symmetry of the system and the spatial orientation of the implied functional groups could mean a huge difference.

The opening of the capsules C-1 (from now on, amide capsule) and C-2 (or symmetric capsule) was developed in MeOD due to the solubility of CsF, one of the most suitable

non-hygroscopic fluoride providing agents. In first case, amide capsule comparatives spectrums and the proposed mechanism of immolation are shown below:

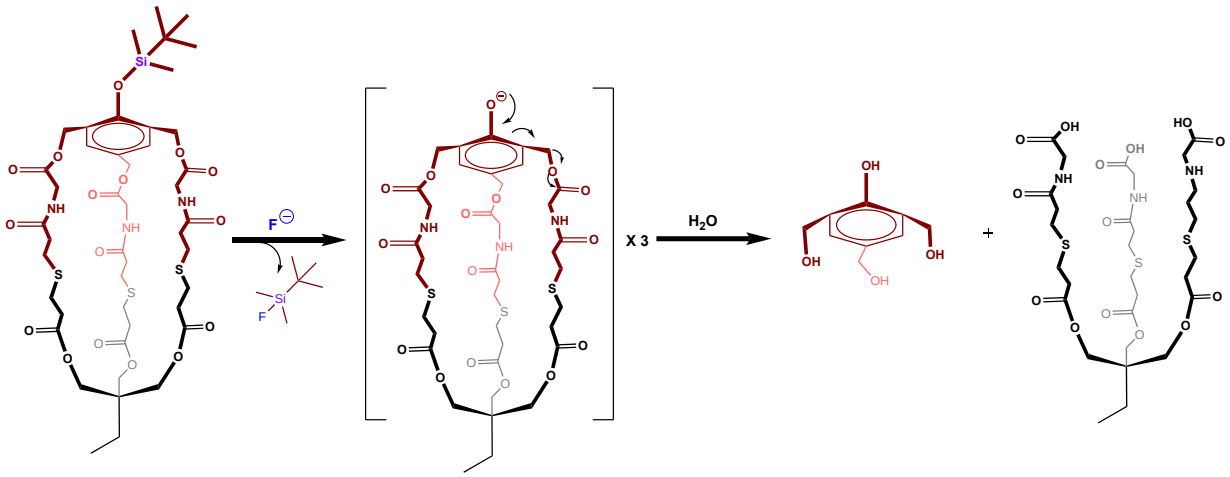



Figure 16. Proposed self-immolative response against stimulus for C-1.

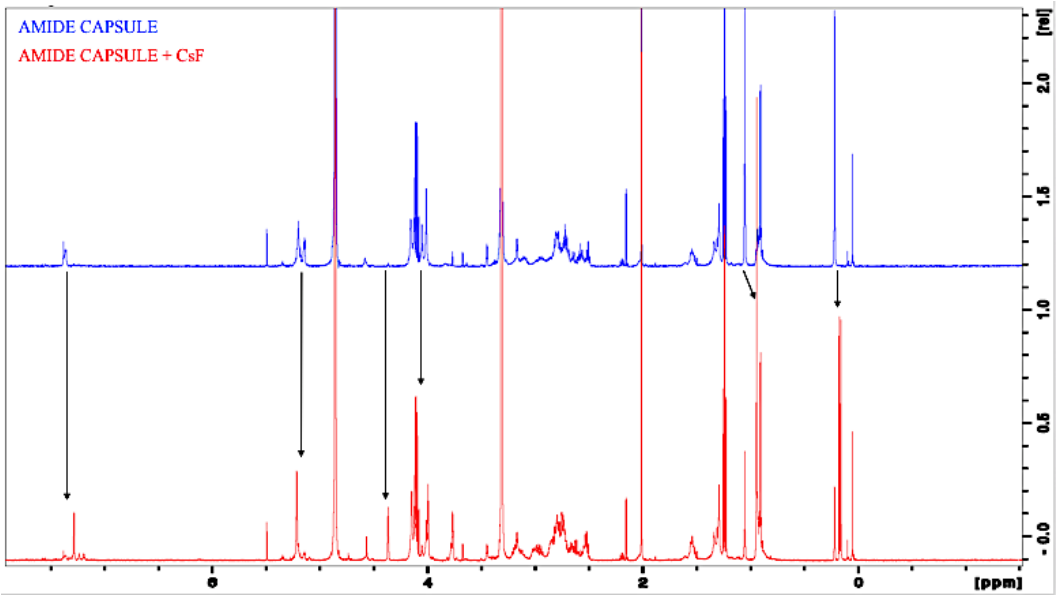



Figure 17. C-1 NMR comparative spectra 1, before and after stimulus.

As it can be seen, there was a shift of the peaks (changes in δ) from the capsule's spectrum (blue) to its broken homologue (red). The most relevant shifts are pointed out and they will be discussed below. It's included below a self-immolative mechanism scheme and the most relevant changes are pointed out to avoid confusion.

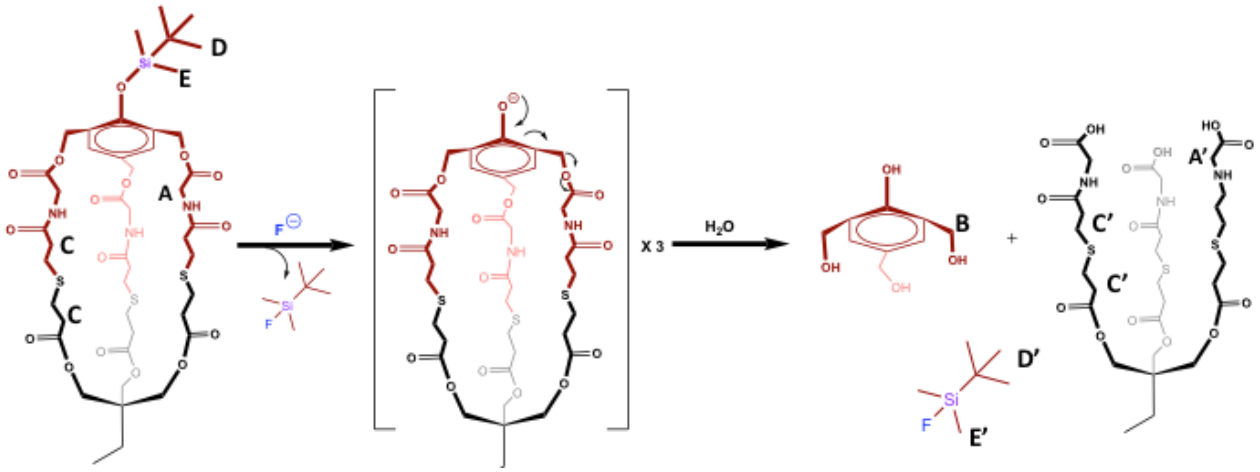



Figure 19. Self-immolative response against stimulus for C-1.

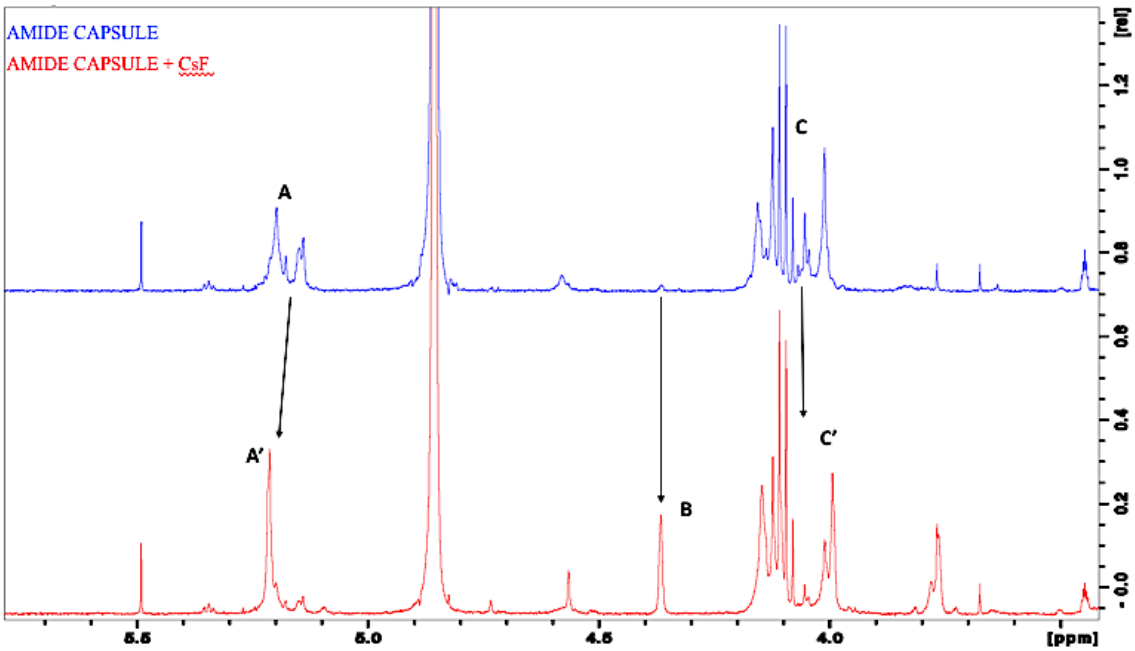



Figure 20. C-1 NMR comparative spectra 2, before and after stimulus.

The signals A and C shown a remarkably displacement and they appear in the second spectra as only 1 peak in the case of A due to the rupture of the capsule that was less symmetric (2 signals in NMR) than the open analogue A' (1 signal in NMR). 2 groups of signals that are separated in the case of C, appearing even a doublet where there was a singlet (signal C'). Indeed, one of the best evidences that C-1 was broken is the peak around 4.4 ppm, signal B, a new singlet appeared after the addition of CsF, probably due the formation of the benzylic alcohol (*Figure 20).*

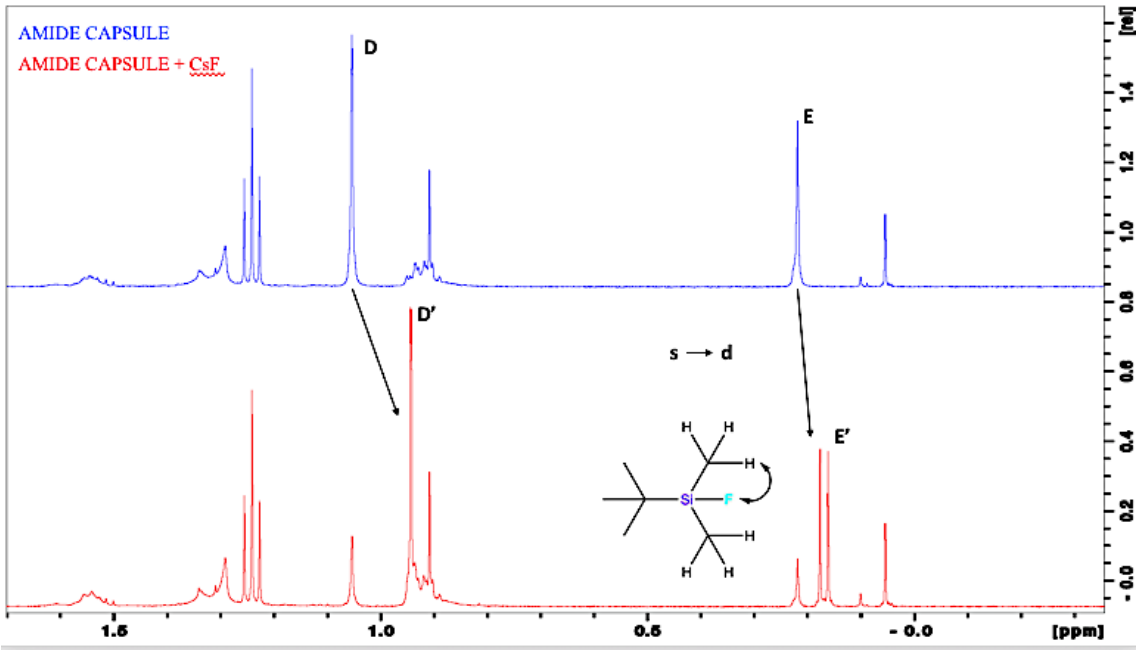



Figure 21. C-1 NMR comparative spectra 3, before and after stimulus.

Another obvious fact are the displacement of the signal D and E, that corresponds to the silyl ester group (TBDMS): *tert*-butyl and methyl groups respectively. In the first case we can appreciate a high field shift and in the case of E, a singlet becomes a doublet due to a new F-H coupling (signal E'), irrefutable proof that Si-F bond it is formed and the capsule was opened.

Related to the second example, C-2 or symmetric capsule, our proposed immolation mechanism is detailed in the next figure (*Figure 22*):

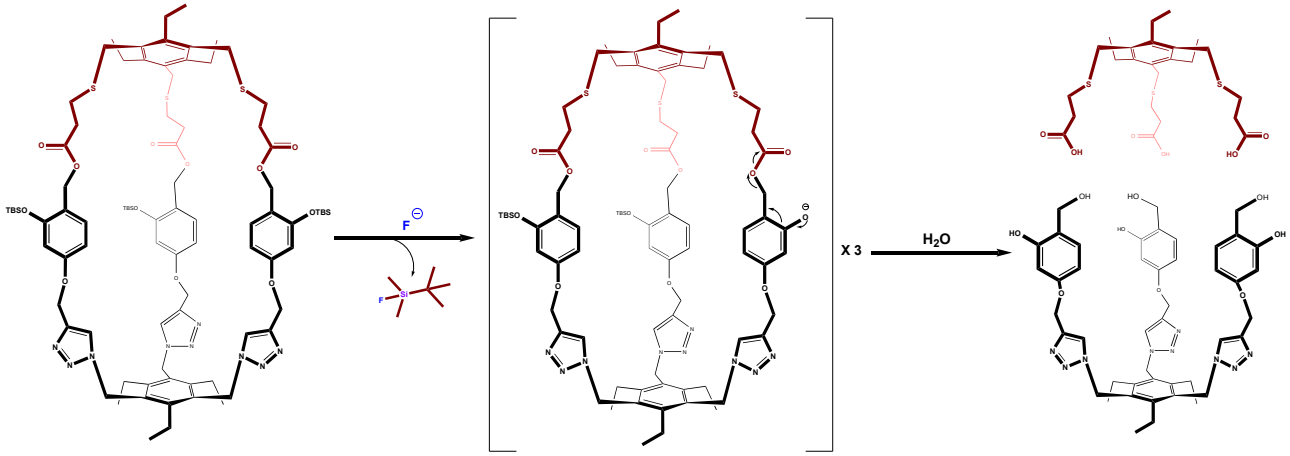



Figure 22. Proposed self-immolative response for C-2.

Again, several comparative NMR spectres will be shown with the aim of confirming our suggestion:

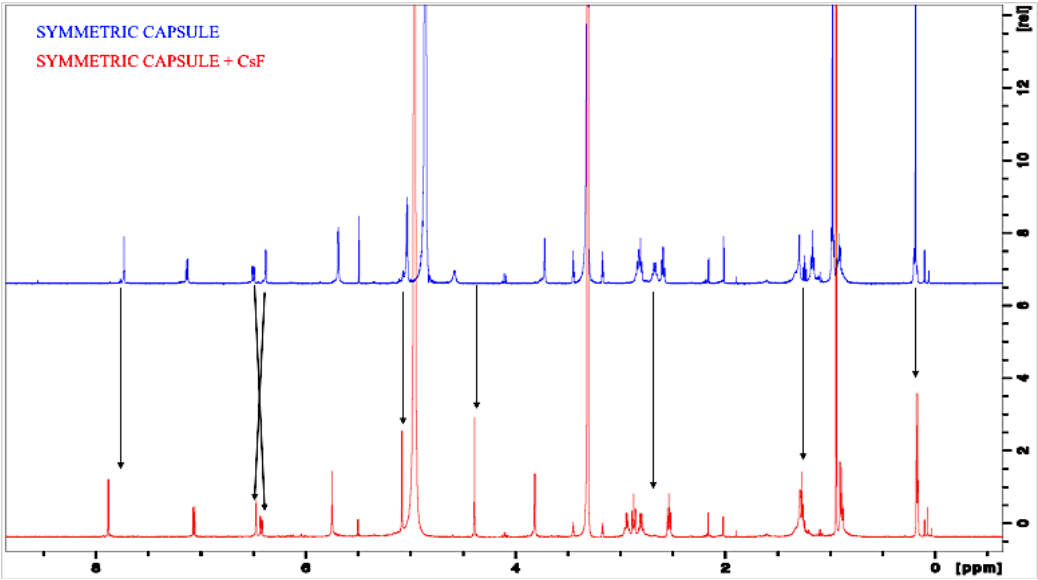


Figure 23. C-2 NMR comparative spectra 1, before and after stimulus.

Once more, there has been a displacement on the signals (changes in δ) from the capsule's spectra (blue) to the open system (red). The most relevant displacements are pointed out and they'll be commented below.

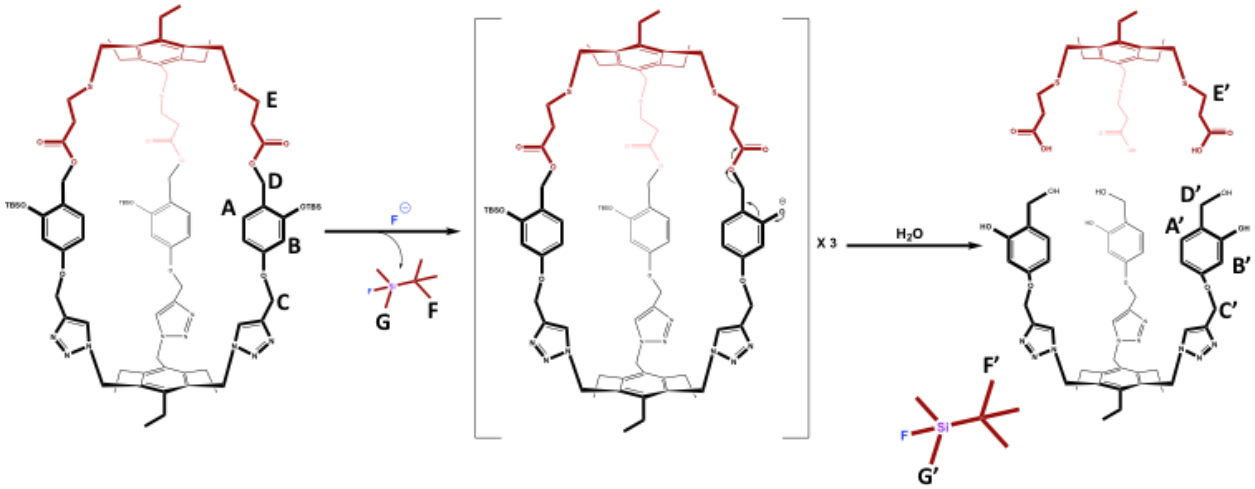


Figure 24. Self-immolative response against stimulus for C-2.

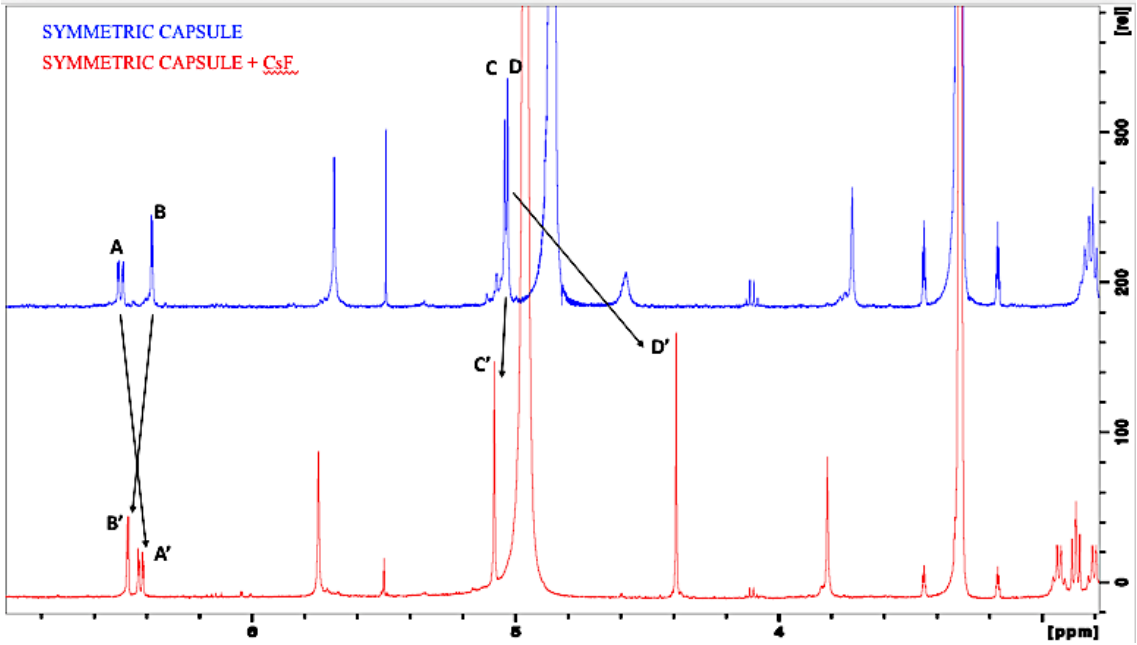


Figure 25. C-2 NMR comparative spectra 2, before and after stimulus.

A significant change can be observed in the aromatic signals A and B, as a result of the changed appearing order in the second spectra A' corresponds to the signal near the benzylic position and B' is the signal of the H near the OTBDMS group. Attending to C and D, they are 2 singlets together in the capsule's spectra, corresponding to benzylic ethers, and only one appears after the addition of CsF in the same position, and the new singlet D' is displaced due to opening of the capsule and the formation of benzylic alcohols. This is very important, because after self-immolation three benzylic alcohols are generated as it can be seen in *Figure 24*.

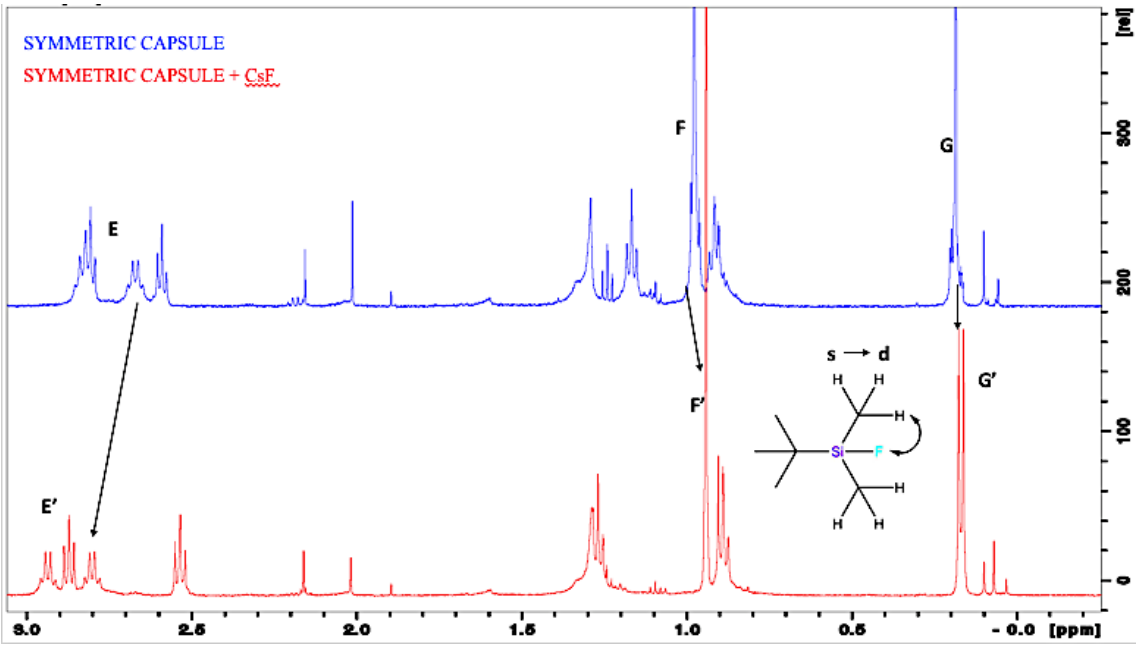


Figure 26. C-2 NMR comparative spectra 3, before and after stimulus.

Finally, the group of signals E have changed after the immolation event, becoming then into E' (signals of -$CH_2$- positions) but once more time, the important transformation is observed by the silyl ester high field shift of the peak F (to F') (*tert*-butyl signal) and the

appearing of a doublet (G to G') due to the F-H coupling. So, there's no doubt that our capsule has been opened.

All the self-immolative events are also currently being studied by mass spectrometry.

Finally, we can conclude that the goal of this work has been reached. Two new supramolecular capsules were synthetized and also their self-immolative response against a stimulus (fluoride anion) was checked via NMR spectroscopy.

## 5. Future work.

The synthesis of a self-immolative capsule that turns on fluorescent upon the application of the right stimulus is almost finished. The trigger in this case is the reduction of a nitro-group by $H_2S$. The structure and the proposed mechanism of self-immolation is showed in the figure below (*Figure 24*):

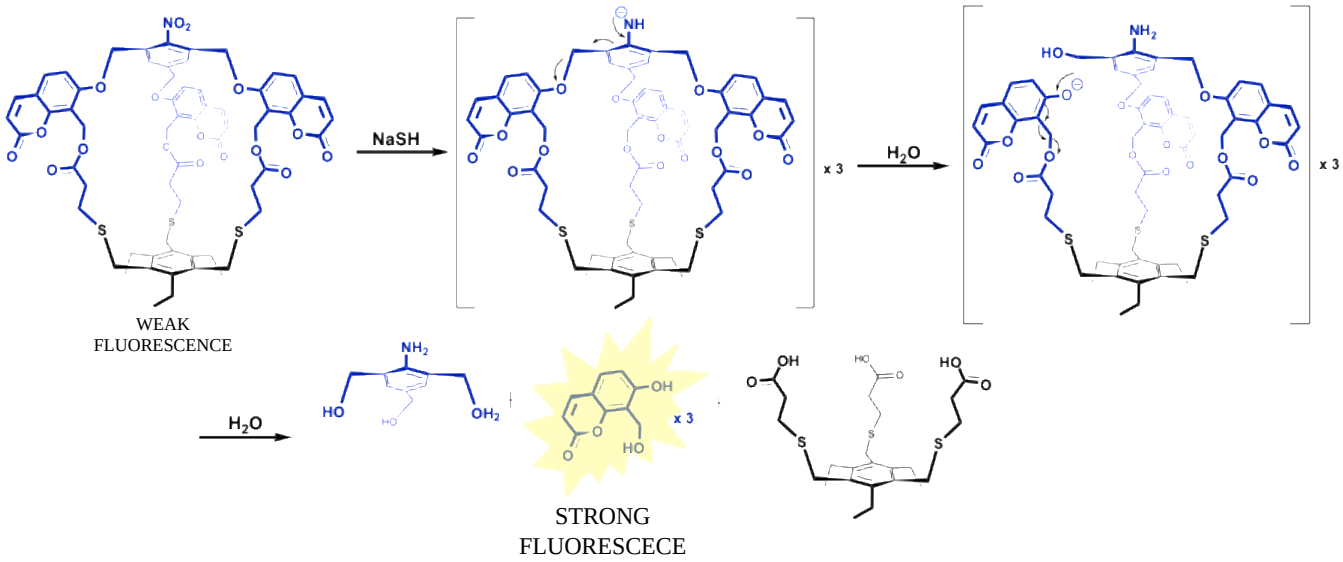



Figure 24. Fluorescent supramolecular self-immolative capsule

Our future goals will be to measure the association constant of these capsules with a suitable guest. We are also working on more complex capsules, including calixarenes moieties, CTV's and other biocompatible scaffolds that can display a high association constant with antioxidating agents against ROS species like taurine or GABA.

# 6. Experimental.

## 6.1 General procedures.

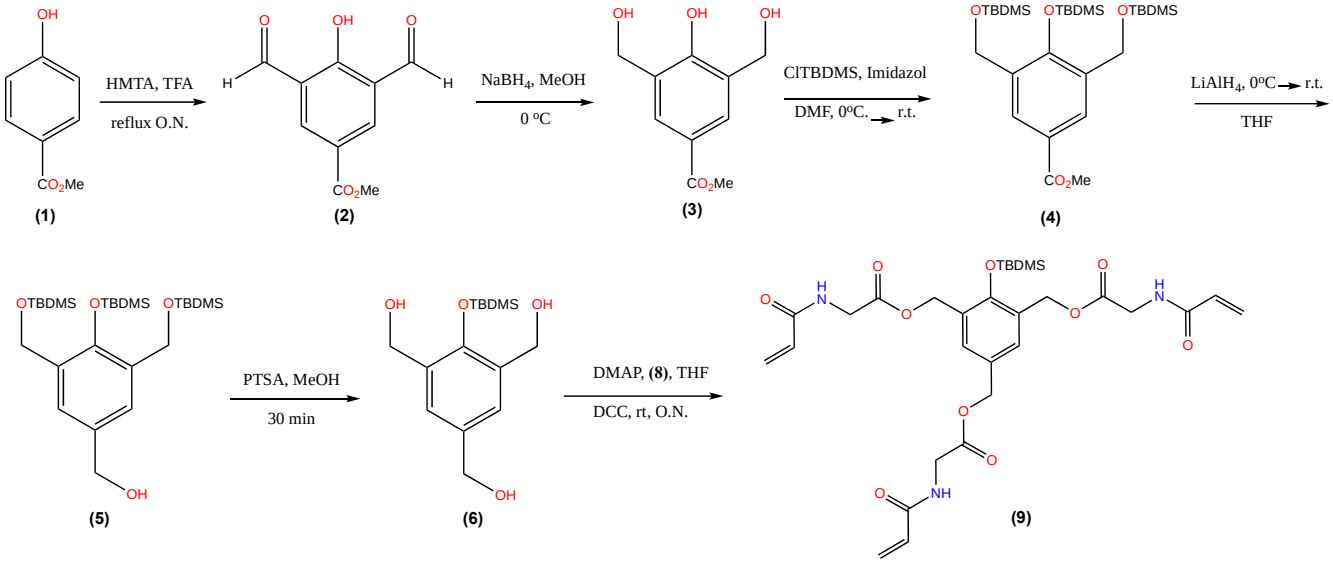
All reactions requiring anhydrous conditions were performed in oven-dried glassware under an Ar atmosphere unless otherwise noted. Chemical and solvents were either *puriss p.A.* or purified by standard techniques. Flash column chromatography was performed using silica gel, 60 Å and 0.2-0.5 mm with the indicated solvent system according to standard techniques. Compounds were visualized on TLC plates by use of UV light, or vanillin with acetic and sulfuric acid in ethanol with heating. Anhydrous magnesium sulphate was used for drying solutions.

$^{1}$H NMR spectra were recorded at 600 MHz and 500MHz, $^{13}$C NMR spectra were recorded at 125 MHz. Chemical shifts were reported in units (ppm) by assigning TMS resonance in the $^{1}$H NMR spectrum as 0.00 ppm ($CDCl_3$, 7.26 ppm; MeOD, 3.31 ppm; DMSO-$D_6$, 2.50 ppm and Acetone-$D_6$ , 2.05 ppm). The spectra were recorded at room temperature unless stated otherwise. Data were reported as follows: chemical shift, multiplicity (bs = broad singlet, s = singlet, d = doublet, dd = double doublet, t = triplet, m = multiplet, q = quartet and br = broad), coupling constant (J values) in Hz and integration. Chemical shifts for $^{13}$C NMR spectra were recorded in ppm from tetramethylsilane using the central peak of $CDCl_3$ (77.16 ppm), MeOD (49.00 ppm), Acetone-$D_6$ (29.84 ppm) and DMSO-$D_6$ (39.52 ppm). The experiments for checking the opening of the capsules were developed in MeOD at room temperature unless otherwise is noted. Accurate mass (HRMS) were determined by electrospray ionization (ESI-TOF) and electronic impact (EI-TOF).

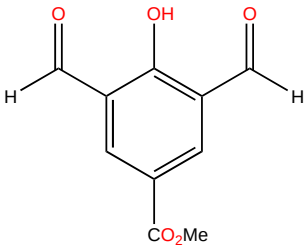
## 6.2 Abbreviations

| Abbreviation | Denotation |
|---|---|
| AcOEt | Ethyl acetate |
| HAc | Acetic acid |
| TBDMSCl | *tert*-Butyldimethylsilyl chloride |
| DBU | 1,8-Diazabicyclo[5.4.0]undec-7-ene |
| DCM | Dichloromethane |
| DCC | N,N′-Dicyclohexylcarbodiimide |
| DMAP | 4-(Dimethylamino)pyridine |
| DMF | Dimethylformamide |
| $Et_2O$ | Diethyl ether |
| Hex | Hexane |
| HMTA | Hexamethylenetetramine |
| NaAsc | Sodium ascorbate |
| NBS | N-Bromosuccinimide |
| O.N. | Overnight |
| PTSA | *p*-Toluenesulfonic acid |
| r.t. | Room temperature |
| sat | Saturated |
| TEA | Triethylamine |
| TFA | Trifluoroacetic acid |
| THF | Tetrahydrofuran |

## 6.3 Compounds Synthesis




**Compound (2): Methyl 3,5-diformyl-4-hydroxybenzoate.**



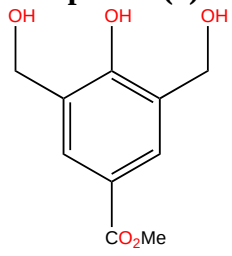
(2)

According to the procedure exposed by Routasalo et al[50], available commercial methyl ester **1** (3.10 g, 20.4 mmol) and HMTA (12.28 g, 87.6 mmol) were dissolved in 22 mL of TFA. The solution was refluxed at 120 ºC overnight. The orange-yellowish crude was cooled to 90 ºC and 120 mL of $H_2O$ were added appearing a precipitate. The mixture was refluxed again to 100 ºC for 30 extra minutes and cooled till room temperate. The yellow solid was filtered off and dried under vacuum to give compound **2**. No purification procedures were needed for our purposes and the product identity was checked according the bibliography. (Yield: 73 %).

**$^1$H NMR (500 MHz, CDCl3) δ (ppm):** 12.04 (bs, 1H), 10.28 (bs, 2H), 8.65 (s, 2H), 3.96 ppm (s, 3H).

**HRMS (EI-TOF) : m/z:** calc for $C_{10}H_8O_5$ [M+]: 208.0372, found: 208.0738.

**Compound (3): Methyl 4-hydroxy-3,5-bis(hydroxymethyl)benzoate.**



(3)

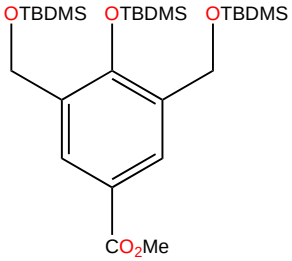
Following Routasalo et al[50] procedure, dialdehyde **2** (0.992 g, 4.76 mmol) was dissolved in 50 mL of MeOH at r.t. The solution was cooled down to 0ºC, $NaBH_4$ (0.24 g, 6.2 mmol) was slowly added while stirring and the mixture was allowed to reach r.t. The reaction was followed by TLC (AcOEt/Hex 50:50). Then 5 mL of of $H_2O$ were added and MeOH was removed on vacuum. A solution of HCl 1M was added over the crude until pH 2. The mixture was extracted with AcOEt (3x10 mL) and solvent was removed under reduced pressure to give compound **3** as an off white solid. No further purification process was required and the product was checked according the bibliography. (Yield: quantitative).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 8.67 (s, 1H), 7.79 (s, 2H), 4.86 (s, 4H), 3.89 (s, 3H), 3.49 ppm (s, 1H).

**$^{13}$C NMR (1205 MHz, $CDCl_3$) δ (ppm):** 166.86, 159.06, 129.66, 126.16, 121.85, 63.39, 53.11 ppm.

**HRMS (EI-TOF) : m/z:** calc for $C_{10}H_{12}O_5$ [M+]: 212.0685, found: 212.0690

**Compound (4): Methyl-4-((*tert*-butyldimethylsilyl)oxy)-3,5-bis(((*tert*-butyldimethylsilyl)oxy)methyl) benzoate.**




**(4)**

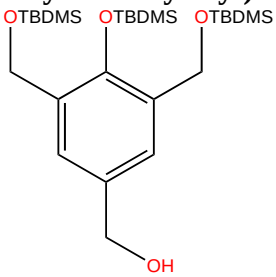
To a solution of triol **3** (1.04 g, 4.89 mmol) in 7 mL of DMF, were added TBDMSCl (5.16 g, 34.23 mmol) and imidazole (2.33 g, 34.23 mmol). The reaction was monitored by TLC (AcOEt/Hex 5:95). After completion of the process, the crude was diluted with 10 mL of $Et_2O$. The organic layer was washed with $NH_4Cl$ sat (3x10mL) followed by brine (1x10 mL) and dried over anhydrous $MgSO_4$. Compound **4** was purified by column chromatography on silica gel (AcOEt/Hex 5:95) to give the desire product as a colorless oil[51]. (Yield: quantitative).

**$^{1}$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 8.033 (s, 2H), 4.71 (s, 4H), 3.88 (s, 3H), 1.02 (s, 9H), 0.95 (s, 18 H), 0.19 (s, 6H), 0.09 ppm (s, 12H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ (ppm):** 152.52, 132.13, 128.14, 123.84, 60.63, 51.97, 26.15, 26.08, 19.00, 18.56, -3.11 ppm.

**HRMS (ESI-TOF) : m/z:** calc for $C_{28}H_{54}O_5Si_3Na$ [M+$Na^+$]: 577.3177, found: 577.3180.

**Compound (5): (4-((*tert*-butyldimethylsilyl)oxy)-3,5-bis(((*tert*-butyldimethylsilyl)oxy)methyl)phenyl)methanol.**




**(5)**

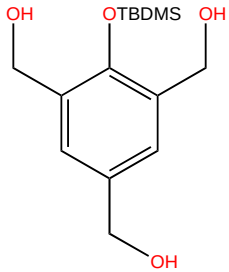
Trisilyl ester **4** (2.71 g, 4.89 mmol) was dissolved in 55 mL of THF at 0 $^{o}$C. $LiAlH_4$ was carefully added while stirring and the reaction was followed by TLC (AcOEt/Hex 20:80). 10 mL of $Et_2O$ were added and the crude was quenched with 15 mL of $NH_4Cl$ sat solution. The organic layer was collected and the other one was extracted twice with $Et_2O$. The organic phase was dried over anhydrous $MgSO_4$, the solvent was removed under reduced pressure to give compound **5** as a colorless oil. No further purification process was required for our purposes. (Yield: quantitative).

**$^{1}$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.37 (s, 2H), 4.71 (s, 4H), 4.64 (s, 2H), 1.02 (s, 9H), 0.94 (s, 18H), 0.16 (s, 6H), 0.09 ppm (s, 12H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 147.84, 134.26, 132.03, 65.88, 60.78, 26.18, 26.13, 25.80, 18.60, -3.19, -5.10 ppm.

**HRMS (ESI-TOF): m/z:** calc for $C_{27}H_{54}O_4Si_3Na$ [M+$Na^+$]: 549.3228, found: 549.3228.

**Compound (6): (2-((*tert*-butyldimethylsilyl)oxy)benzene-1,3,5-triyl)trimethanol.**



**(6)**

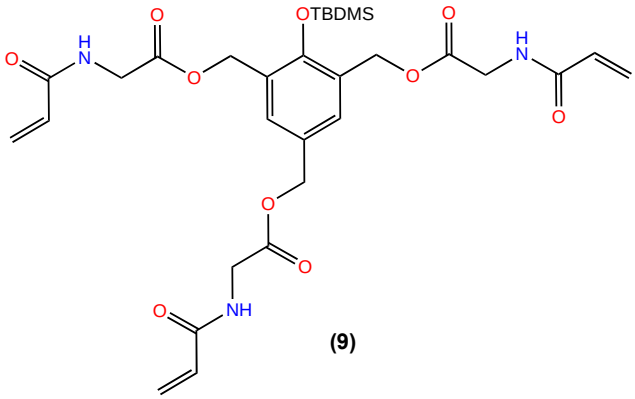
According to the experimental procedure[52], benzyl-alcohol **5** (2.58 g, 4.88 mmol) was dissolved in 50 mL of MeOH at r.t. Catalytic PTSA was added while stirring and the reaction was monitored by TLC (AcOEt) during 30 minutes. 10 mL of $NaHCO_3$ were added and the solution was extracted with AcOEt (3x10mL). The organic layer was washed with brine (1x10 mL) and compound **6** was purified by column chromatography on silica gel (AcOEt) to give the desire product as a white solid. (Yield: quantitative).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.21 (s, 2H), 4.61 (d, J= 5.9 Hz, 4H), 4.47 (d, J=5.5 Hz, 2H), 3.06-3.05 (m, 1H), 2.79 (t, J=6.0 Hz, 2H), 1.01 (s, 9H), 0.16 ppm (s, 6H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 149.50, 134.85, 132.25, 127.25, 64.92, 60.91, 26.13, 18.83, -3.47 ppm.

**HRMS (ESI-TOF): m/z:** calc for $C_{15}H_{26}O_4SiNa$ [M+$Na^+$]: 321.1489, found: 321.1501.

**Compound (9): (2-((tert-butyldimethylsilyl)oxy)benzene-1,3,5-triyl)tris(methylene) tris(2-acrylamidoacetate).**



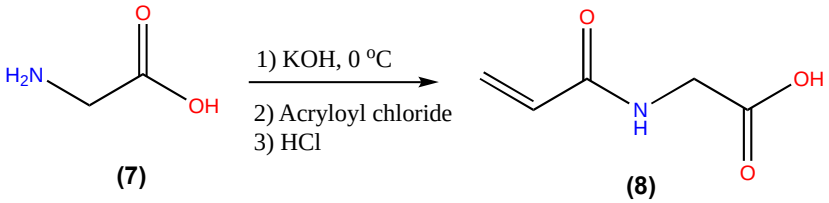
**(9)**

Triol **6** (50.01 mg, 0.17 mmol) was dissolved in THF (5 mL) with DMAP (82 .0 mg, 0.67 mmol) and acryloylglycine **8** (86.50 mg, 0.67 mmol). DCC (138 mg, 0,67 mmol) was added at r.t. and the mixture was stirred O.N. The reaction began as a colorless solution and finally end up as cloudy-yellow one. The reaction was followed by TLC (AcOEt) and was filtered through a celite layer once finished. The desire product was purified by column chromatography on silica gel (AcOEt) to give the desire product as yellow oil. (Yield: 10 %)

**$^1$H NMR (500 MHz, $CDCl_3$) δ:** 7.28 (s, 2H), 6.68 (t, J=5.50 Hz, 1H), 6.49 (t, J=5.56 Hz, 2H), 6.32-6.29 (dd, $J_1$= 17.00 Hz, $J_2$= 1.35 Hz, 2H), 6.31-6.27 (dd, $J_1$= 17.00 Hz, $J_2$=1.50 Hz, 1H), 6.22-6.15 (m, 3H), 5.69-5.67 (dd, $J_1$= 10.20 Hz, $J_2$=1.35 Hz, 2H), 5.68-5.66 (dd, $J_1$=10.10 Hz, $J_2$=1.55 Hz, 1H), 5.19 (s, 4H), 5.11 (s, 2H), 4.16 (d, J= 5.45 Hz, 4H), 4.13 (d, J=5.65 Hz, 2H), 1.02 (s, 9H), 0.19 ppm (s, 6H).

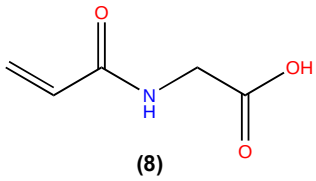
**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 169.87, 166.09, 165.90, 151.24, 130.31, 130.24, 130.02, 129.33, 127.50, 127.43, 127.12, 66.40, 62.31, 41.57, 25.97, 18.78, -3.51 ppm.

***HRMS: m/z:**




(7) (8)

**Compound (8): Acryloylglycine.**



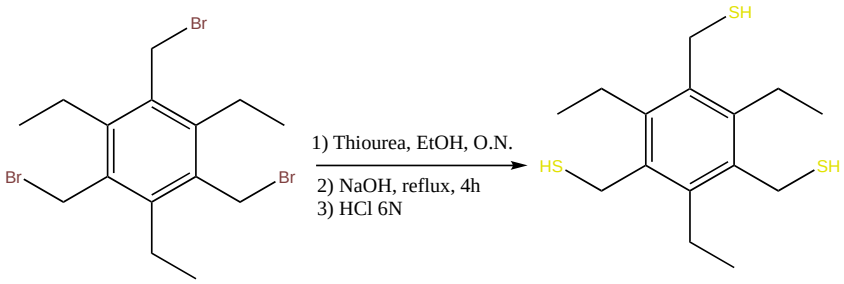

(8)

According to the protocol[53], glycine (4.5 g, 60 mmol) was dissolved in 60 mL of KOH (2M). The soution was cooled at 0 ºC using a ice-water bath and acryloyl chloride (6 mL, 73.6 mmol) was added dropwise using a dropping funnel. The mixture was stirred for 90 min at 0 ºC and other 90 min at r.t. The resulting solution was washed with $Et_2O$ (2 x 40 mL) and the aqueous phase was acidified to pH 2. The product was extracted with AcOEt (3x40 mL), dried over $MgSO_4$ and concentrated on the rotatory evaporator to provide a white solid. (Yield: 12 %).

**$^1$H NMR (500 MHz, DMSO-$D_6$) δ (ppm):** 12.57 (bs, 1H), 8.42 (s, 1H), 6.29 (dd, $J_1$=17.13 Hz, $J_2$=10.23 Hz, 1H), 6.10 (dd, $J_1$=17.13Hz, $J_2$=2.08 Hz, 1H), 5.62 (dd, $J_1$=10.23Hz, $J_2$=2.08 Hz, 1H), 3.83 ppm (d, J= 5.95 Hz, 2H).

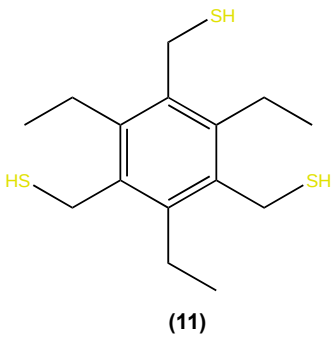
**$^{13}$C NMR (150 MHz, DMSO-$D_6$) δ:** 171.19, 164.88, 132.30, 125.64, 40.62 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_5H_7NO_3$ [M+]: 129.0426, found: 129.0432.




(10) (11)

**Compound (11): (2,4,6-triethylbenzene-1,3,5-triyl)trimethanethiol.**



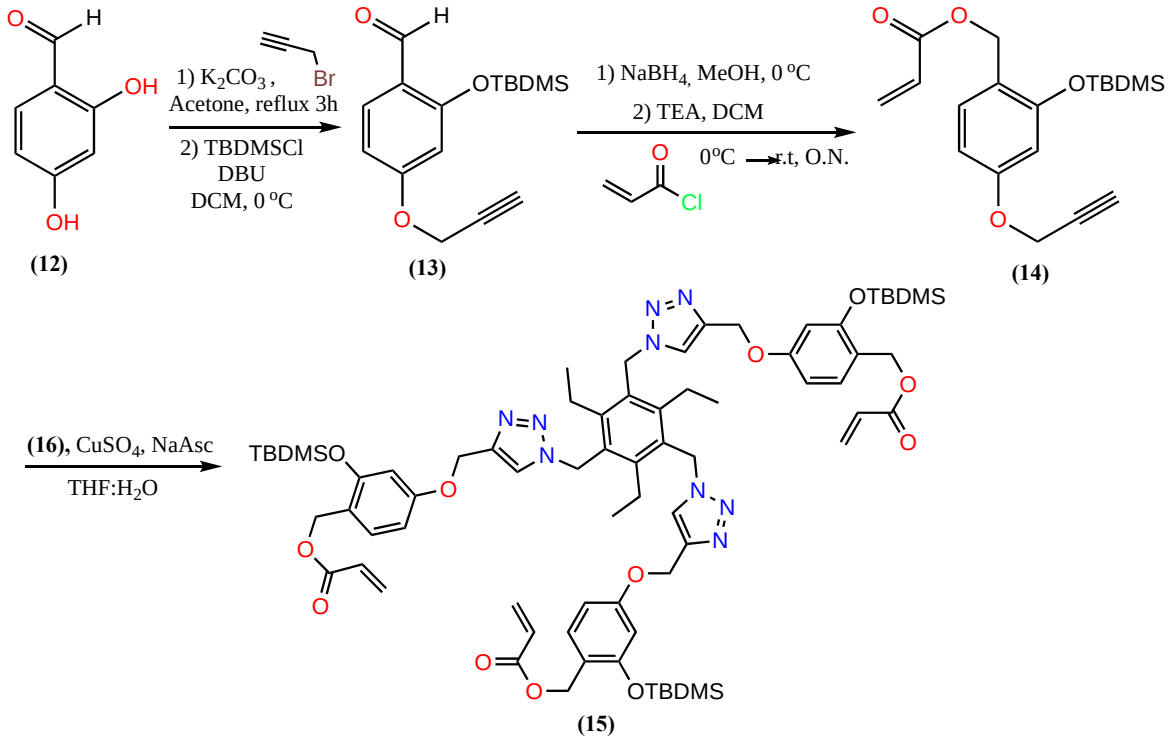

(11)

Commercially available 1,3,5-tris(bromomethyl)-2,4,6-trimethylbenzene **10** (0.2 g, 0.454 mmol) was dissolved in 2 mL of EtOH and thiourea (0.10 g, 1.36 mmol) was added. The solution was stirred O.N. at r.t. showing a white-cloudy aspect. The solvent was removed under vacuum to add a solution of NaOH (0.11 g, 2.72 mmol) in 2 mL of $H_2O$, and this mixture was refluxed 4 h. The solution was cooled down in an ice-bath and HCl (6N) was added up to pH 2. The water phase was extracted with DCM (3x10 mL), dried over $MgSO_4$ and concentrated on the rotatory evaporator to give trithiol **9** as a white solid[54]. (Yield: 82 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 3.76 (d, J=6.18 Hz, 6H), 2.86 (q, J=7.56 Hz, 6 H), 1.70 (t, J=6.24 Hz, 3H), 1.27 ppm (t, J=7.56 Hz, 9H).

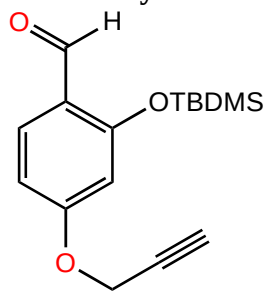
**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 140.41, 135.67, 22.97, 22.93, 16.26 ppm.

**HRMS (ESI-TOF): m/z:** calc for $C_{15}H_{24}NaS_3$ [M+$Na^+$]: 323.0938, found: 323.0942.




(12) (13) (14) (15)

**Compound (13): 2-((tert-butyldimethylsilyl)oxy)-4-(prop-2-yn-1-yloxy) benzaldehyde.**



(13)

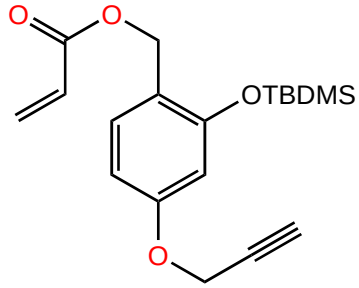
Following the experimental procedure[55,56], commercially available 2,4-dihydroxybenzaldehyde **12** (2.80 g, 20 mmol) was dissolved in acetone (30 mL). $K_2CO_3$ (2.80 g, 20 mmol) and propargyl bromide (2.4 g, 20 mmol) were added and the mixture was refluxed while stirring during 3 h. The reaction was cooled down to r.t. and the solvent was removed under vacuum and DCM was added (100 mL). The organic phase was washed with $H_2O$ (4x 20 mL), dried over $MgSO_4$, filtered and concentrated on the rotatory evaporator obtaining a pale-yellow solid. This crude material (97.2 mg, 0.55 mmol) was again dissolved in DCM (2 mL) and cooled using an ice-water bath. DBU (0.16 mL, 1.10 mmol) was added, followed by TBDMSCl (92,0 mg, 0,61 mmol). The reaction mixture was stirred for 2 h at r.t. and monitored by TLC (AcOEt/Hex 5:95). After complete reaction, it was quenched with $H_2O$, extracted with DCM (3x 20 mL), dried over $MgSO_4$, filtered and concentrated on the rotatory evaporator. Compound **13** was purified by column chromatography on silica gel (AcOEt/Hex 5:95) to give the desire product as a pale-yellow oil. (Yield: 90 %)

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 10.31 (s, 1H), 7.79 (d, J=8.75 Hz, 1H), 6.64 (dd, $J_1$=10.98 Hz, $J_2$=6.50 Hz, 1H), 6.46 (d, J=2.30 Hz, 1H), 4.72 (d, J=2.40 Hz, 2H), 2.57 (t, J=2.38 Hz, 1H), 1.02 (s, 9H), 0.29 ppm (s, 6H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 188.77, 163.61, 160.76, 130.17, 122.06, 108.90, 106.06, 77.78, 76.52, 56.07, 25.79, 18.48, -4.18 ppm.

***HRMS: m/z:**

**Compound (14): 2-((tert-butyldimethylsilyl)oxy)-4-(prop-2-yn-1-yloxy)benzyl acrylate.**




**(14)**

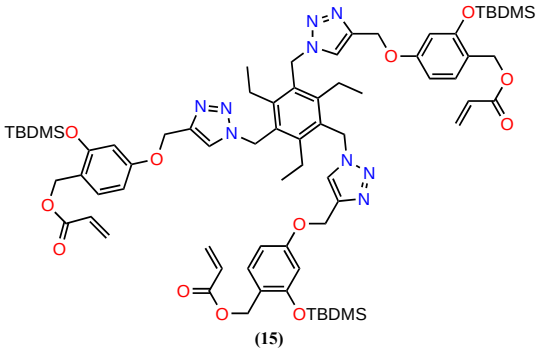
Aldehyde **13** (12.5 mg, 0,04 mmol) was dissolved in MeOH (4.3 mL) and cooled to 0ºC using an ice-water bath. After 10 min, $NaBH_4$ (1.9 mg, 0.052 mmol) was slowly added while stirring and the mixture was allowed to reach r.t. The reaction was followed by TLC (AcOEt/Hex 5:95) till consumption of the aldehyde, quenched with $H_2O$, extracted with AcOEt (3x15 mL), dried over $MgSO_4$, filtered through a silica gel layer and concentrated on the rotatory evaporator. The crude material obtained was dissolved again in DCM (5 mL) and cooled to 0ºC using an ice-water bath. TEA was added (8 µL, 0,06 mmol), stirred for 10 min and then acryloyl chloride (4 µL, 0,056 mmol) was slowly added. The reaction was allowed to reach r.t. and stirred O.N. The process was monitored by TLC (AcOEt/Hex 20:80), quenched with sat $NH_4Cl$, extracted with DCM (3x15 mL), dried over $MgSO_4$, filtered and concentrated on the rotatory evaporator. Product **14** was purified by column chromatography on silica gel (AcOEt/Hex 20:80) to give the desire product as a pale-yellow oil. (Yield: 40%)

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.26 (d, J=8.40 Hz, 1H), 6.57 (dd, $J_1$=8.43 Hz, $J_2$=2.48 Hz, 1H), 6.50 (d, J=2.50 Hz, 1H), 6.40 (dd, $J_1$=17.33 Hz, $J_2$=1.48 Hz, 1H), 6.12 (dd, $J_1$=17.35 Hz, $J_2$=10.45 Hz, 1H), 5.80 (dd, $J_1$=10.40 Hz, $J_2$=1.45 Hz, 1H), 5.14 (s, 2H), 4.66 (d, J=2.40 Hz, 2H), 2.53 (t, J=2.38 Hz, 1H), 0.99 (s, 9H), 0.25 ppm (s, 6H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 166.41, 158.89, 155.51, 131.77. 130.76, 128.71 119.94, 107.11, 106.27, 78.59, 75.83, 62.24, 56.01, 25.79, 18.35, -4.12 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_{19}H_{26}O_4Si$ [M+]: 346.1600, found: 346.1627

**Compound (15): ((((((5-(4-((4-((acryloyloxy)methyl)-3-((tert-butyldimethylsilyl)oxy)phenoxy)methyl)-1H-1,2,3-triazol-1-yl)-2,4,6-triethyl-1,3-phenylene)bis(methylene))bis(1H-1,2,3-triazole-1,4-diyl))bis(methylene))bis(oxy))bis(2-((tert-butyldimethylsilyl)oxy)-4,1-phenylene))bis(methylene) diacrylate**



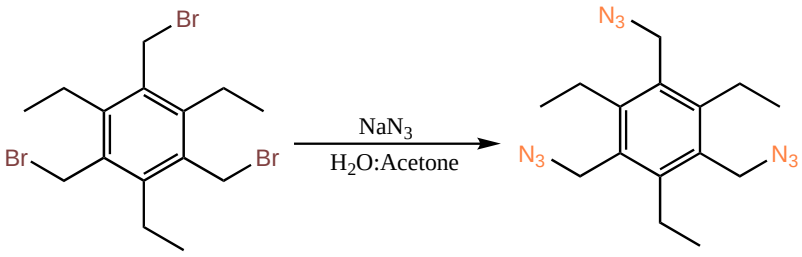

**(15)**

Triazide **16** (110 mg, 0.34 mmol) and compound **14** (595 mg, 1.7 mmol) were dissolve in THF (8 mL). $H_2O$ (8 mL) was added, followed by $CuSO_4$ (25.5 mg, 0.16 mmol) and NaAsc (61.4 mg, 0.31 mmol) were added and the reaction was stirred O.N. at r.t. The reaction was followed by TLC (AcOEt/Hex 40:60), quenched with DCM and $H_2O$, extracted with DCM (3x10 mL), dried over $MgSO_4$, filtered and concentrated on the rotatory evaporator. The desired compound was obtained after purification by column chromatography on silica gel (AcOEt/Hex 40:60). (Yield: 70 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.40 (s, 3H), 7.21 (d, J=8.45 Hz, 3H), 6.56 (dd, $J_1$=8.45 Hz, $J_2$=2.45 Hz, 3H), 6.45 (d, J=2.45 Hz, 3H), 6.39 (dd, $J_1$=17.33 Hz, $J_2$=1.43 Hz, 3H), 6.11 (dd, $J_1$=17.33 Hz, $J_2$=10.43 Hz, 3H), 5.80 (dd, $J_1$=10.43 Hz, $J_2$=1.43 Hz, 3H), 5.64 (s, 6H), 5.12 (s, 12H), 2.79 (q, J=7.49 Hz, 6H), 0.97 (s, 27H), 0.93 (t, J=7.53 Hz, 9H), 0.22 ppm (s, 18H).

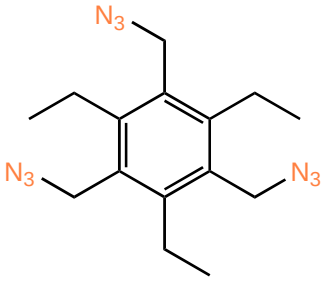
**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** ppm. 166.38, 159.48, 155.53, 146.66, 144.28, 131.81, 130.79, 129.73, 128.68, 122.48, 119.72, 107.18, 106.11, 62.23, 62.08, 48.14, 25.78, 23.80, 18.33, 15.39, -4.10 ppm.

***HRMS: m/z:**




**(10)** **(16)**

**Compound (16): 1,3,5-tris(azidomethyl)-2,4,6-triethylbenzene**



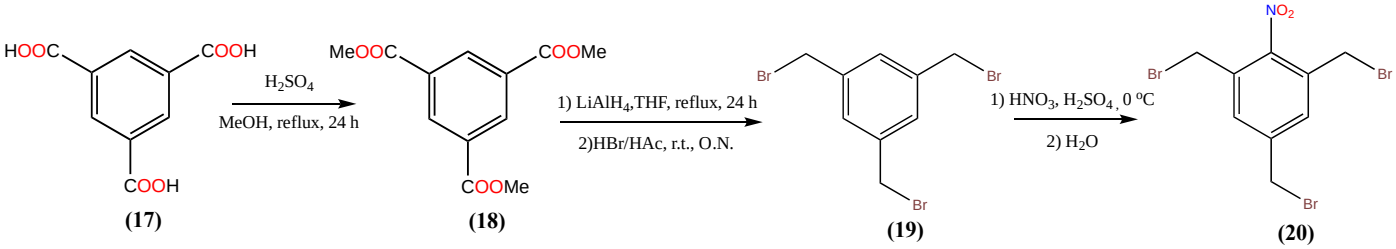

**(16)**

Compound **10** (0.5g, 1.13 mmol) was dissolved in a 22,5 mL mixture of $H_2O$/Acetone (1:4). $NaN_3$ was slowly added (110 mg, 1.7 mmol) and the reaction was followed by TLC until compound **10** ran out. The aqueous phase was extracted wit DCM (3x10 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure. Compound **16** was obtained as a white solid after purification by column chromatography on silica gel (AcOEt/Hex 50:50). (Yield: 85 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 4.49 (s, 6 H), 2.85 (q, J= 7.60 Hz, 6H), 1.24 ppm (t, J=7.59 Hz, 9 H).

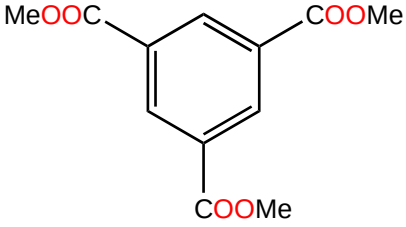
**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 145.13, 130.15, 48.09, 23.31, 15.90 ppm.

**HRMS (ESI-TOF): m/z:** calc for $C_{15}H_{21}NaN_3$ [M+$Na^+$]: 350.1818, found: 350.1823.




**(17)** **(18)** **(19)** **(20)**

**Compound (18): trimethyl benzene-1,3,5-tricarboxylate.**




**(18)**

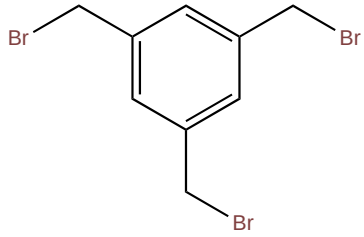
Trimesic acid, **17**, (3.00 g, 14.3 mmol) was dissolved in 60 mL of MeOH. $H_2SO_4$ was slowly added (0.75 mL) and the reaction was refluxed during 24 h before being allowed to cool to r.t. $NaHCO_3$ sat solution was slowly added to neutralize the mixture. The aqueous phase was extracted wit $Et_2O$ (3x20 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure. Compound **18** was obtained as a white solid[57]. (Yield: quantitative).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 8.86 (s, 3H), 3.98 ppm (s, 9H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 165.58, 134.75, 131.37, 52.78 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_{12}H_{12}O_6$ [M+]: 252.0634, found: 252.0639

**Compound (19): 1,3,5-tris(bromomethyl)benzene**




**(19)**

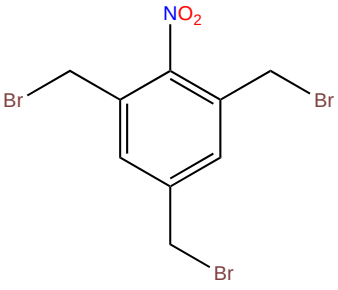
According to the literature[57], triester **18**, (3.6 g, 14.3 mmol) was dissolved in THF (75 mL) and was added dropwise over a solution of $LiAlH_4$ (2.56 g, 64.4 mmol) in THF at 0 ºC. The reaction was refluxed during 24 h before being allowed to cool. Upon cooling, $H_2O$ was carefully added (50 mL) and the mixture was filtered through Celite pad, and the filter cake was washed with DCM. The volatiles were concentrated under reduced pressure. After that, the crude was mixed with 20,9 mL of HBr in HAc solution (33% w/v) and stirred O.N. to obtain a needle-like off-white solid that was filtered, generously washed with water on the filter cake and dried under reduced pressure[58]. (Yield: 79%).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.34 (s, 3H), 4.46 ppm (s, 6H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 139.21, 129.72, 32.31 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_9H_9{}^{81}Br_3$ [M+]: 359.8193, found: 359.8217; calc for $C_9H_9{}^{79}Br_3$ [M+]: 353.8254, found: 353.8262

**Compound (20): 1,3,5-tris(bromomethyl)-2-nitrobenzene**



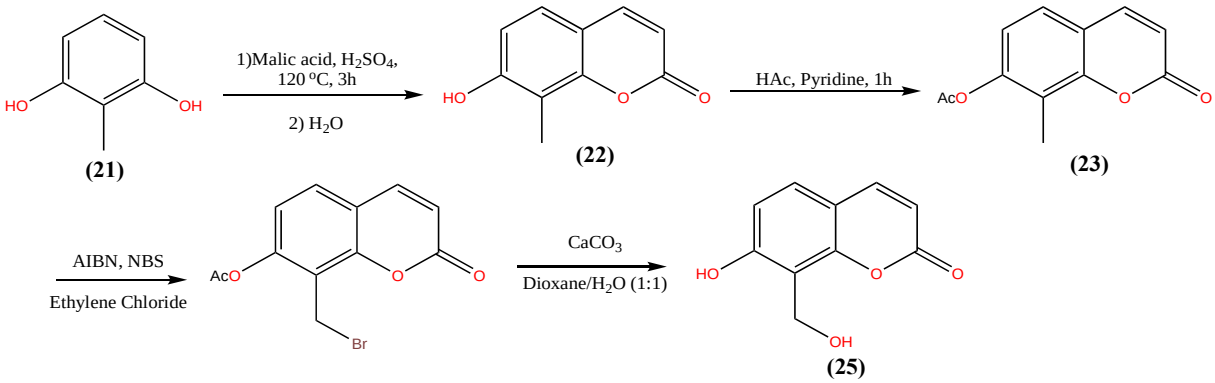

**(20)**

A mixture of $HNO_3$ (1.4 mL) and $H_2SO_4$ (1.6 mL) in a round bottom flask was cooled down in an ice-water bath. Compound **19** was carefully added in a period of 10 min keeping the temperature at 0 ºC for 1h after the complete addition. The mixture was allowed to reach r.t. and an ice-water mixture was added. The aqueous phase was extracted with $Et_2O$ (3x15 mL), and the organic layer was washed with $H_2O$ (2x10 mL), dried over $MgSO_4$, filtered and concentrated under reduce pressure. Compound **20** was obtained as a white solid after purification by column chromatography on silica gel (AcOEt/Hex 5:95). (Yield: 76 %).

**$^{1}$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.52 (s, 2H), 4.48 (s, 4H), 4.45 ppm (s, 2H).

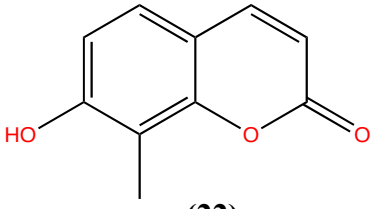
**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 141.75, 132.28, 131.8, 30.33, 26.27 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_9H_8NO_2{}^{81}Br_3$ [M+]: 404.8044, found: 404.8058; calc for $C_9H_8NO_2{}^{79}Br_3$ [M+]: 398.8105, found: 398.8110




**(21)** **(22)** **(23)** **(25)**

**Compound (20): 7-hydroxy-8-methyl-2H-chromen-2-one**




**(22)**

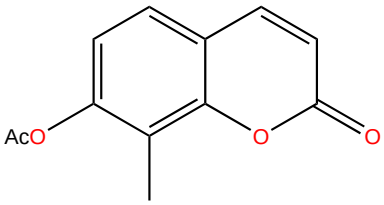
Malic acid (2.65 g, 19.8 mmol) and 2-methylresorcinol (2.50g, 20.0 mmol) were mixed with 10 mL $H_2SO_4$ and the reaction was heated to 120 ºC for 3h. Upon cooling to r.t. $H_2O$ was slowly added and a precipitate appeared. The solid was filtered and washed with water to give compound **22** as a dark-pink solid[59]. (Yield: 47%).

**$^{1}$H NMR (500 MHz, MeOD) δ (ppm):** 7.83 (d, J=9.40 Hz, 1H), 7.29 (d, J=8.50 Hz, 1H), 6.80 (d, J=8.45 Hz, 1H), 6.17 (d, J=9.40 Hz, 1H), 2.25 ppm (s, 3H).

**$^{13}$C NMR (150 MHz, MeOD) δ:** 164.09, 161.01, 155.12, 146.59, 127.43, 113.31, 113.16, 112.86, 111.69, 7.90 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_{10}H_8O_3$ [M+]: 176.0473, found: 176.0491

**Compound (23): 8-methyl-2-oxo-2H-chromen-7-yl acetate**




**(23)**

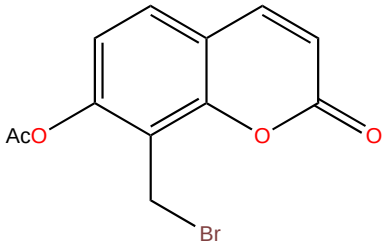
Cumarin **22** (1.23 g, 6.98 mmol) was dissolved in acetic anhydride (13 mL) and inmersed in an ice-water bath before the addition of pyridine (0.67 mL, 8.32 mmol). the mixture was stirred 12h at r.t. Excess acetic anhydride was removed under reduced pressure. The residue was dissolved in DCM (15 mL), washed with saturated $NaHCO_3$ (2x10 mL) , water (2x10 mL), dried over $MgSO_4$ and concentrated on the rotatory evaporator. Compound **23** was purified by column chromatography on silica gel (AcOEt/Hex 50:50) to give the desired coumarin as a white solid[59]. (Yield: 97 %).

**$^{1}$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.68 (d, J=9.55 Hz, 1H), 7.34 (d, J=8.40 Hz, 1H), 7.00 (d, J=8.45 Hz, 1H), 6.39 (d, J=9.50 Hz, 1H), 2.37 (s, 3H), 2.28 ppm (s, 3H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 168.85, 160.70, 153.28, 151.89, 143.51, 125.68, 119.58, 118.67, 116.80, 115.81, 20.88, 9.12 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_{12}H_{10}O_4$ [M+]: 218.0579, found: 218.0591

**Compound (24): 8-(bromomethyl)-2-oxo-2H-chromen-7-yl acetate**




**(24)**

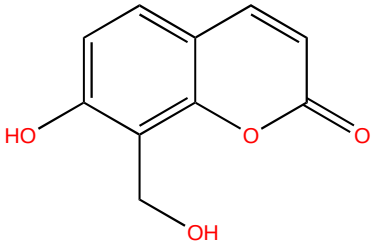
Compound **23** (1.43 g, 6.55 mmol) was dissolved in 10 mL of ethylene chloride, then NBS (1.49 g, 8.40 mmol) was added to the mixture, fllowed by catalytic AIBN. The reaction was refluxed for 6h and the solvent was removed under reduced pressure upon cooling. The desired product, **24**, was purified by column chromatography on silica gel (AcOEt/Hex 40:60) to obtain a yellow solid[59]. (Yield: 65 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 7.68 (d, J=9.60 Hz, 1H), 7.46 (d, J=8.50 Hz, 1H), 7.10 (d, J=8.50 Hz, 1H), 6.41 (d, J=9.60 Hz, 1H), 4.65 (s, 2H), 2.41 ppm (s, 3H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 168.41, 159.65, 152.45, 151.71, 143.14, 128.41, 119.50, 118.90, 116.93, 116.26, 21.06, 19.19 ppm.

**HRMS (EI-TOF): m/z:** calc for $C_{12}H_9O_4{}^{81}Br$ [M+]: 297.9664, found: 297.9674; calc for $C_{12}H_9O_4{}^{79}Br$ [M+]: 295.9684, found: 295.9669

**Compound (25): 7-hydroxy-8-(hydroxymethyl)-2H-chromen-2-one**




**(25)**

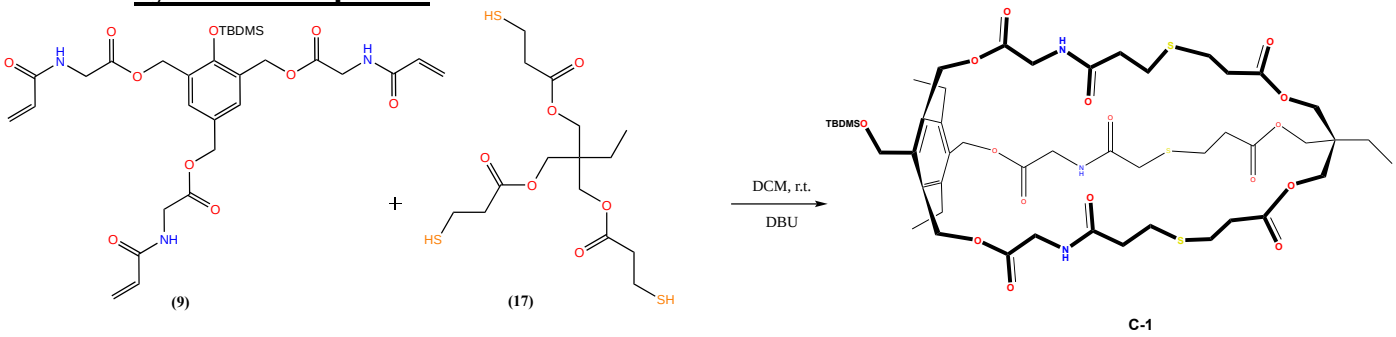
A mixture of $CaCO_3$ (2.56 g, 25.73 mmol) in 12 mL of $H_2O$ was added to a solution of cumarine **24** (1.47g, 4.95 mmol) in 12 mL of dioxane. The mixture was heated to 50 ºc for 24h. The solvent was removed under reduced pressure to give a white solid which was redissolved on HCl 2M and extracted with AcOEt (3x15 mL), dried over $MgSO_4$, filtered and concentrated on the rotovap. The product was purified by column chromatography on silica gel (AcOEt/Hex 60:40) to give diol **25** as a white solid[59]. (Yield: 80 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ (ppm):** 9.25 (bs, 1H), 7.23 (d, J=9.50 Hz, 1H), 6.82 (d, J=8.50 Hz, 1H), 6.20 (d, J=8.50 Hz, 1H), 5.54 (d, J=9.50 Hz, 1H), 4.40 (s, 2H).

**$^{13}$C NMR (150 MHz, $CDCl_3$) δ:** 178.96, 161.14, 160.89, 153.91, 145.13, 129.17, 114.01, 113.97, 112.65, 112.59, 56.04 ppm.

***HRMS: m/z:**

**Synthesis of capsules:**

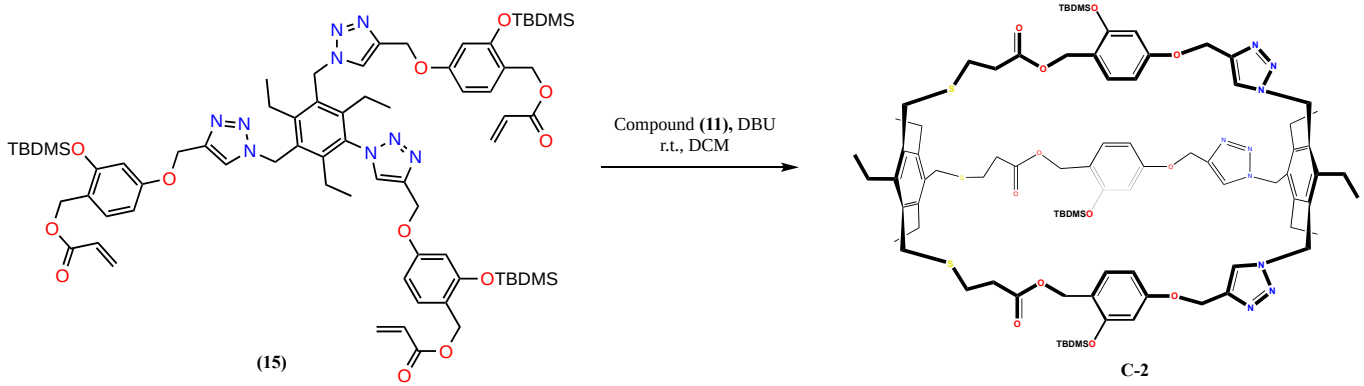



**Capsule 1 (C-1):** according to the protocol,[36] triacrylate **9** (12 mg, 0.019 mmol) and trithiol **17** (6.25 µL, 0.019 mmol) were dissolved in DCM (2 mL) at r.t. DBU (4.3 µL, 0.029 mmol) was added while stirring and the reaction was monitored by TLC (AcOEt/Hex 40:60). 10 ml of $NH_4Cl$ sat solution were added, and the aqueous layer was extracted with DCM (3x10 mL). The gathered organic phases were dried over anhydrous $MgSO_4$ and the solvent was evaporated under reducer pressure. Compound **(10)** was purified by column chromatography on silica gel (AcOEt/Hex 40:60→60:40) to give the desire capsule. (Yield: 40 %).

**$^1$H NMR (500 MHz, $CDCl_3$) δ:** 7.27 (s, 2H), 6.84 (t, J=5.13 Hz, 1 H), 6.58 (t, J=5.25 Hz, 2H), 5.19 (s, 4H), 5.11 (s, 2H), 4.11 (s, 4H), 4.10 (s, 2H), 4.08 (s, 6H), 2.81-2.69 (m, 12 H), 2.61-2-50 (m, 12H), 1.49 (q, J=7.55 Hz, 2H), 1.03 (s, 9H), 0.89 (t, J=7.58 Hz, 3H), 0.19 ppm (s, 6H).

**$^{13}$C NMR (125 MHz, $CDCl_3$) δ:** 172.06, 172.00, 171.96, 171.66, 169.80, 169.47, 151.19, 129.80, 129.50, 127.45, 66.25, 64.74, 64.54, 62.17, 41.77, 41.67, 40.95, 36.80, 36.73, 34.93, 34.77, 27.97, 27.78, 27.38, 27.22, 25.98, 23.56, 18.78, 7.58, -3.45 ppm.

***HRMS: m/z:**




**Capsule 2 (C-2):** according to the protocol,[36] triacrylate **15** (191.0 mg, 0,14 mmol) and trithiol **11** (42,1 mg, 0.14 mmol) were dissolved in DCM (28 mL) at r.t. DBU (31 µL, 0,21 mmol) was added while stirring and the reaction was monitored by TLC (AcOEt/Hex 50:50). 10 ml of $NH_4Cl$ sat solution were added, and the aqueous layer was extracted with DCM (3x10 mL). The gathered organic phases were dried over anhydrous $MgSO_4$ and the solvent was evaporated under reducer pressure. Capsule **C-2** was purified by preparative chromatography on silica gel (AcOEt/Hex 50:50) to give the desire product. (Yield: 16 %)

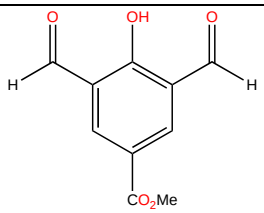
**$^{1}$H NMR (500 MHz, $CDCl_3$) δ:** 7.33 (s, 3H), 7.14 (d, J=8.50 Hz, 3H), 6,48 (dd, $J_1$=10.80 Hz, $J_2$=6.10 Hz, 3H), 6.39 (d, J=2.35 Hz, 3H) 5.60 (s, 6 H), 5.05 (d, J=6.90 Hz, 12 H) 3.73 (s, 6H), 2.86-2.79 (m, 12H), 2.72 (q, J=7.40 Hz, 6H), 2.56 (t, J=7.00 Hz, 6H), 1.20 (t, J=7.50 Hz, 9H), 1.09 (t, J=7.48 Hz, 9H), 0.97 (s, 27H), 0.19 ppm (s, 18 H).

**$^{13}$C NMR (125 MHz, $CDCl_3$) δ:** 171.92, 159.21, 155.06, 146.73, 144.03, 142.52, 131.49, 131.41, 129.57, 122.49, 119.76, 107.15, 106.22, 62.09, 61.95, 47.94, 35.15, 31.27, 28.26, 25.80, 23.60, 23.03, 18.33, 16.28, 15.66, -4.11 ppm.

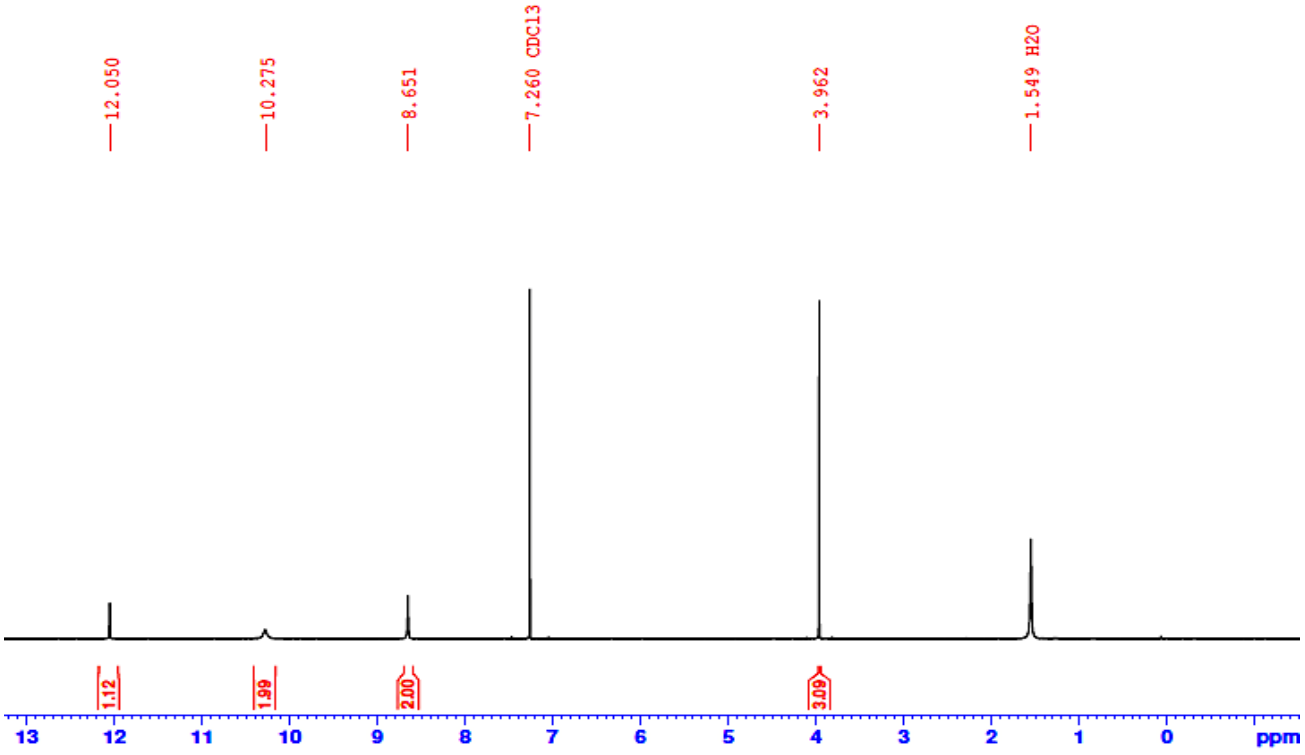
***HRMS: m/z:**

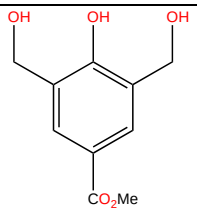
**Note: The HRMS experiments marked with an "*" weren't performed due to technical problems of the ESI-TOF equipment at IPNA-CSIC. The EI-TOF facility at ULL was also unable to perform the experiments, so, the products were sent to Universidad Complutense to obtains the results as soon as possible.**

## 6.4 Appendix: NMR spectra.

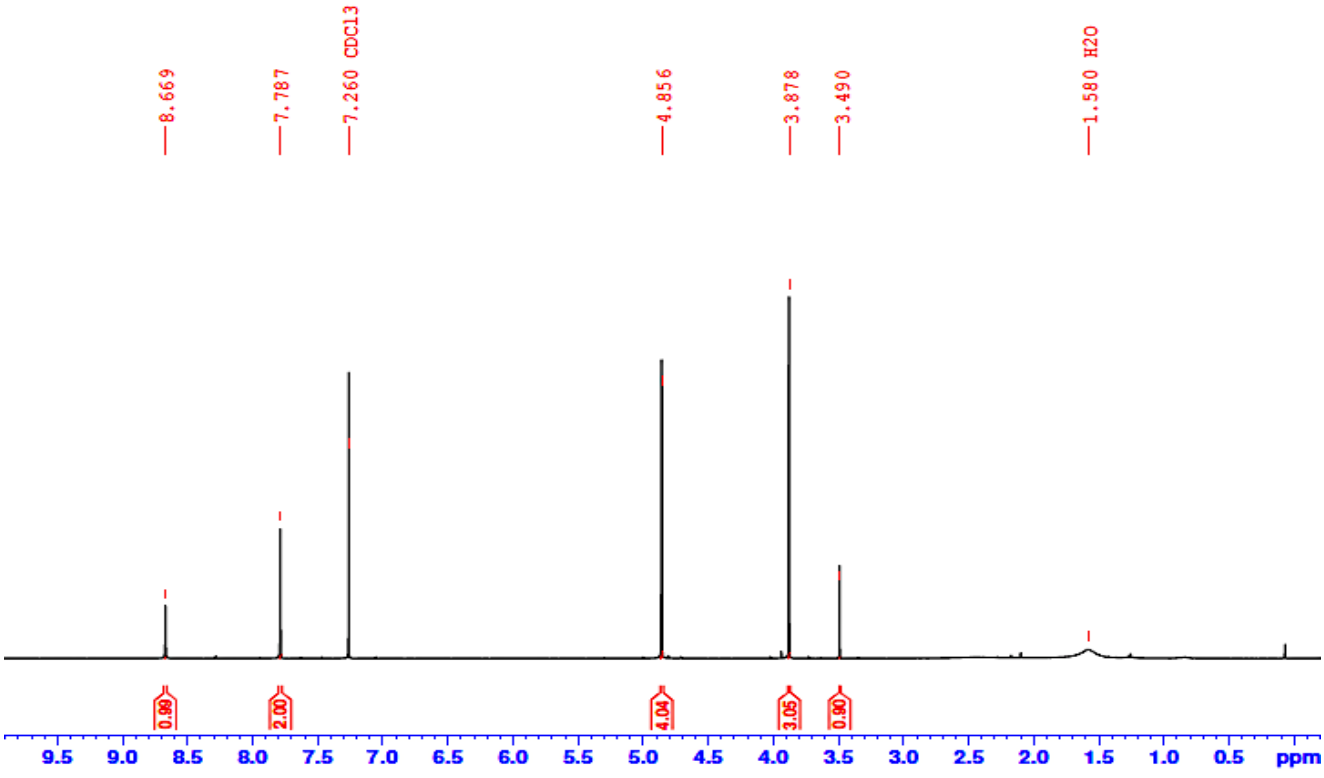



**$^{1}$H NMR (500 MHz, $CDCl_3$):**

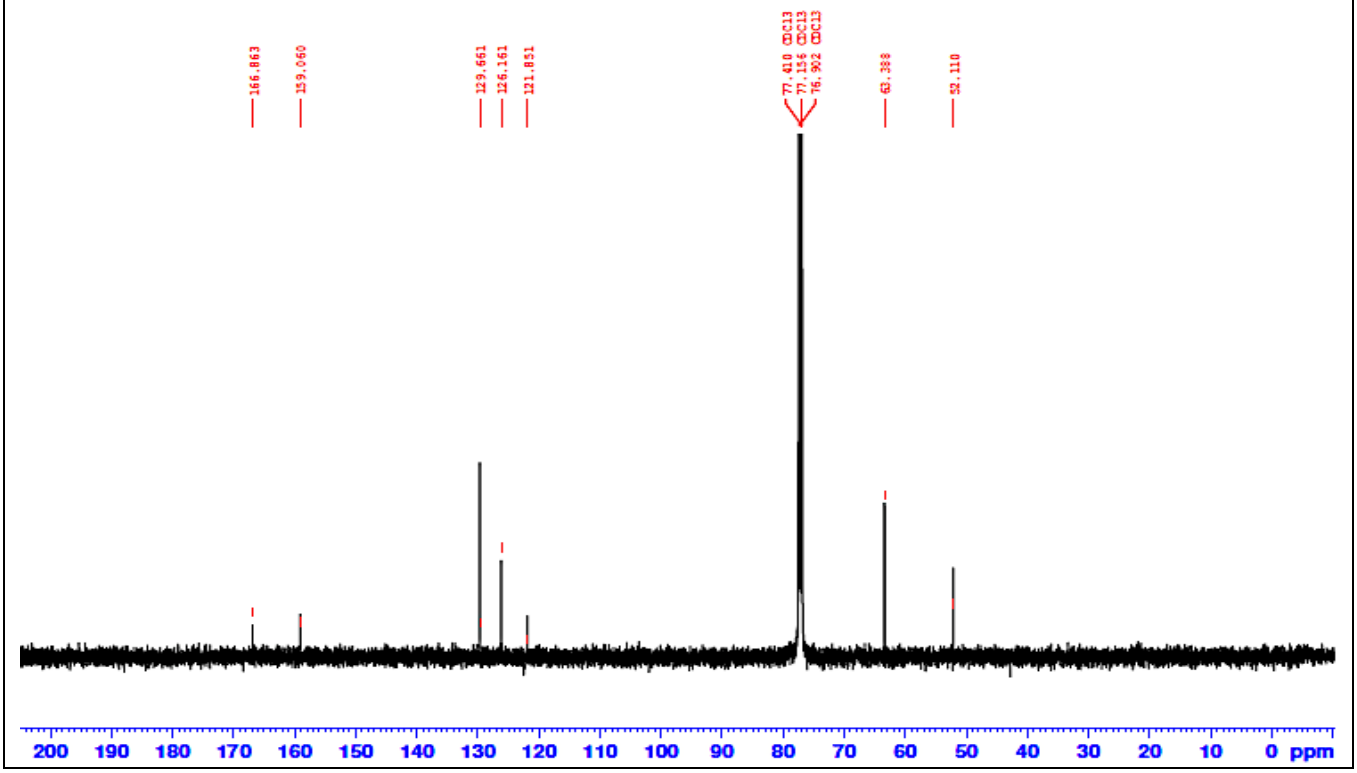


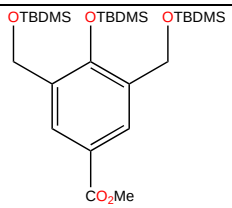



**$^{1}$H NMR (500 MHz, $CDCl_3$):**




**$^{13}$C NMR (125 MHz, $CDCl_3$):**



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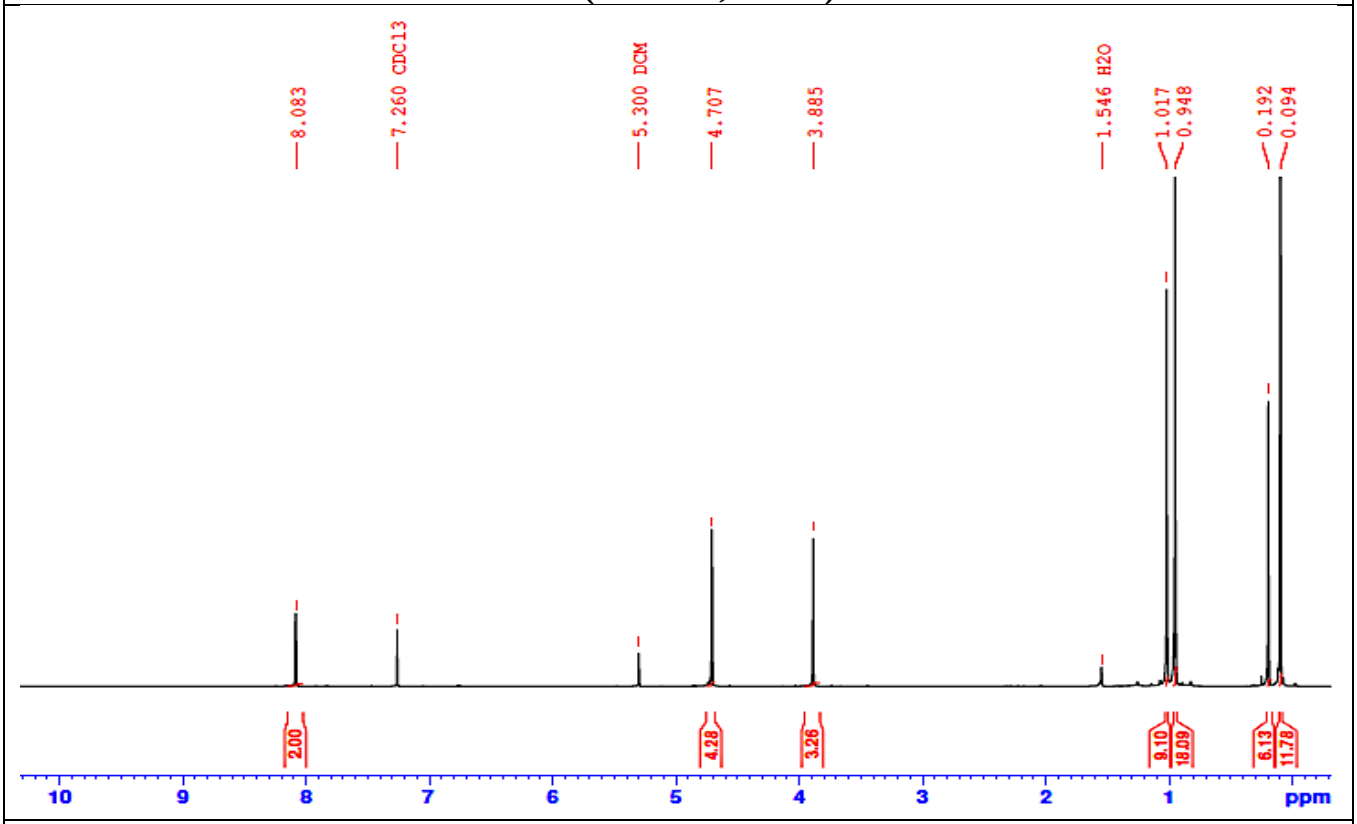


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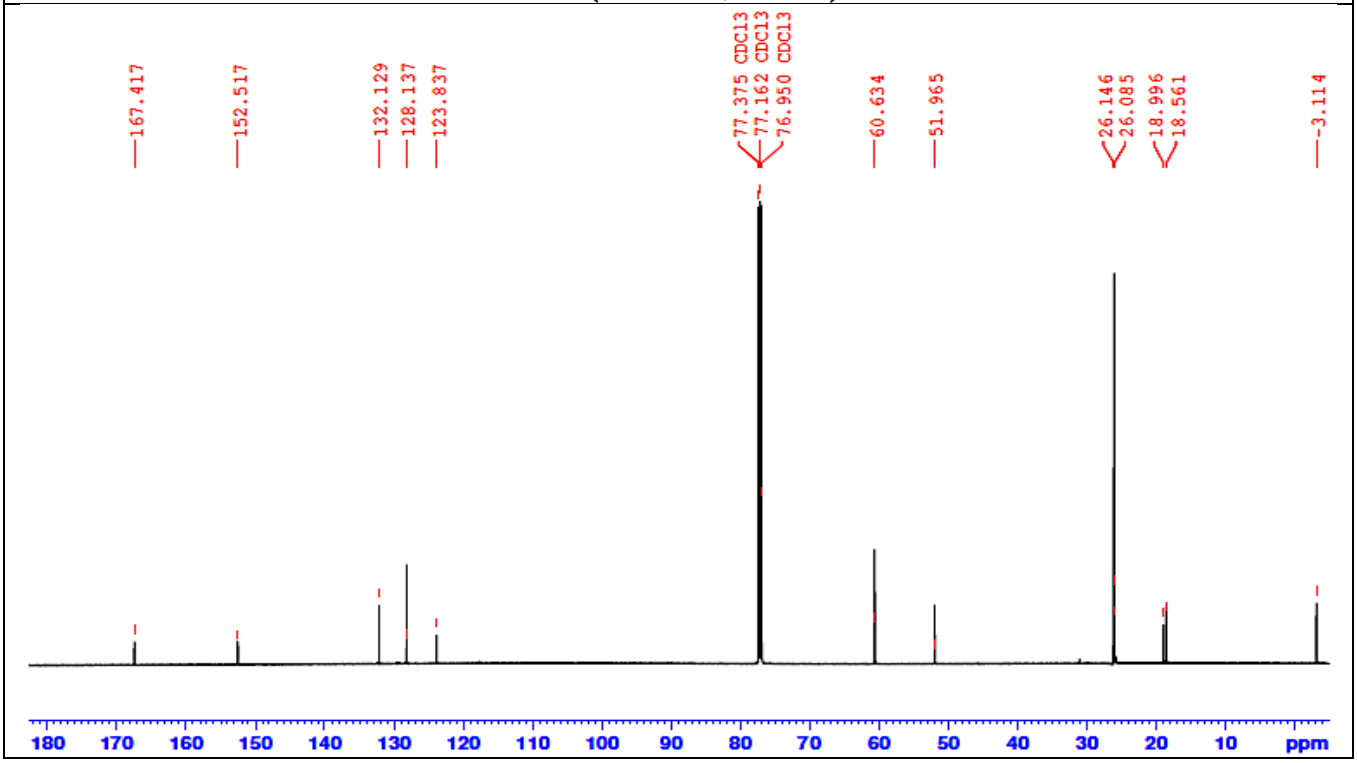

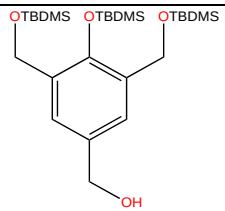


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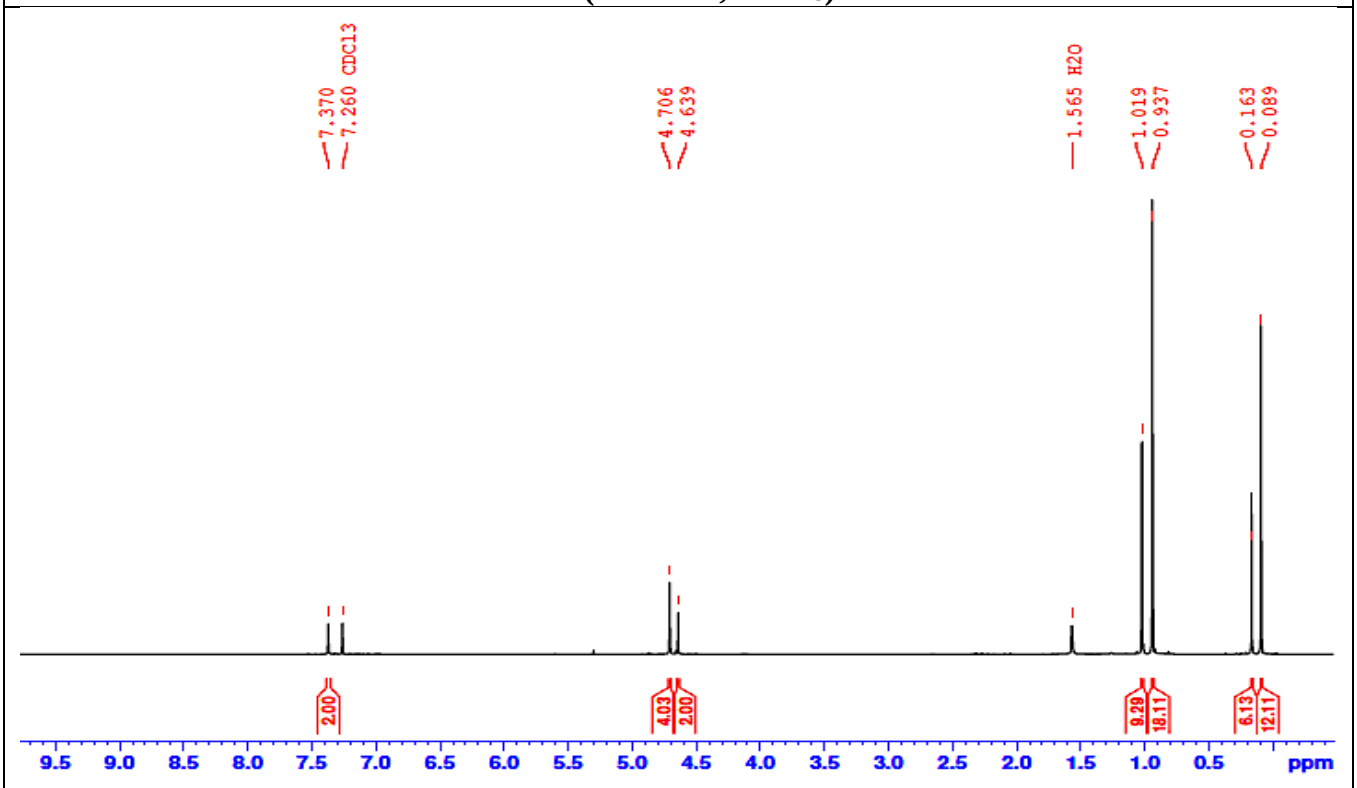
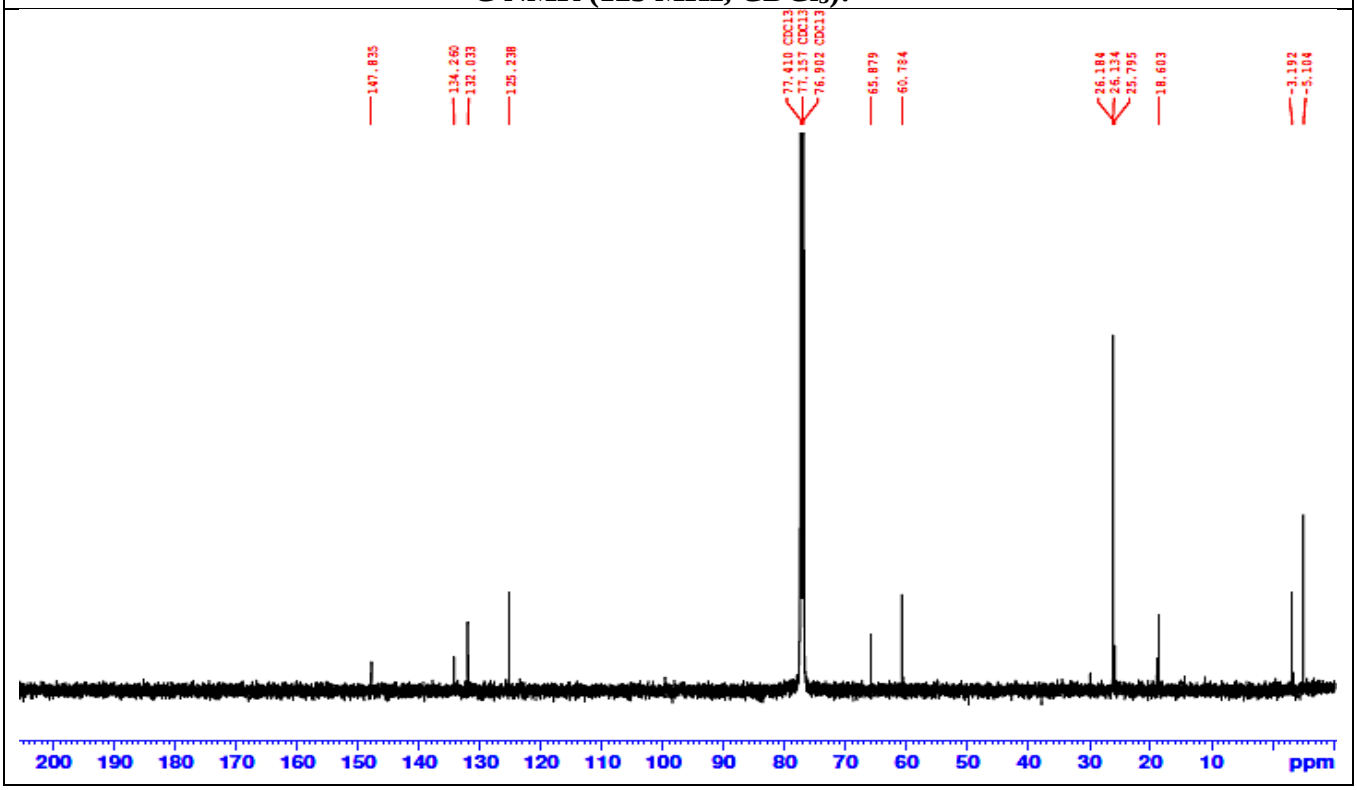
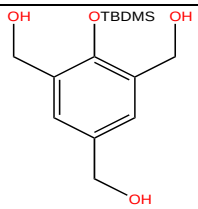



**$^{13}$C NMR (125 MHz, $CDCl_3$):**



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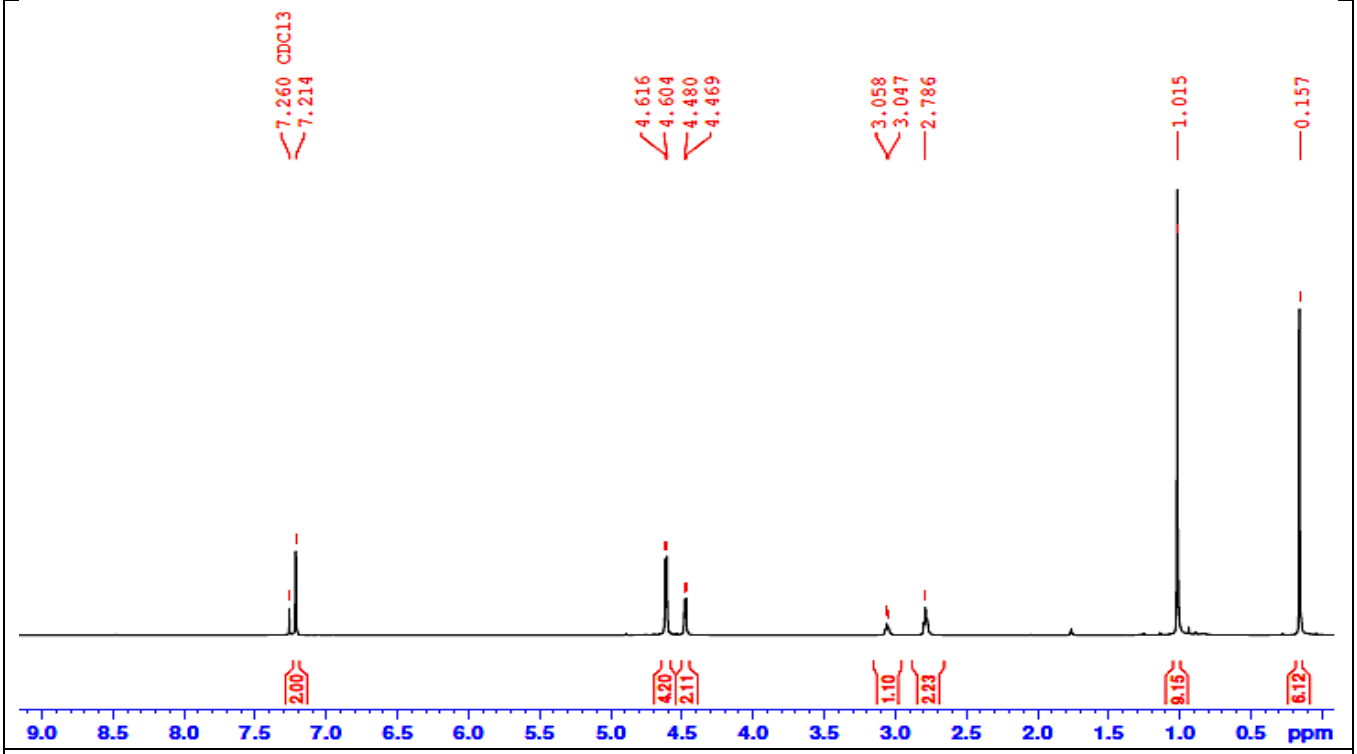
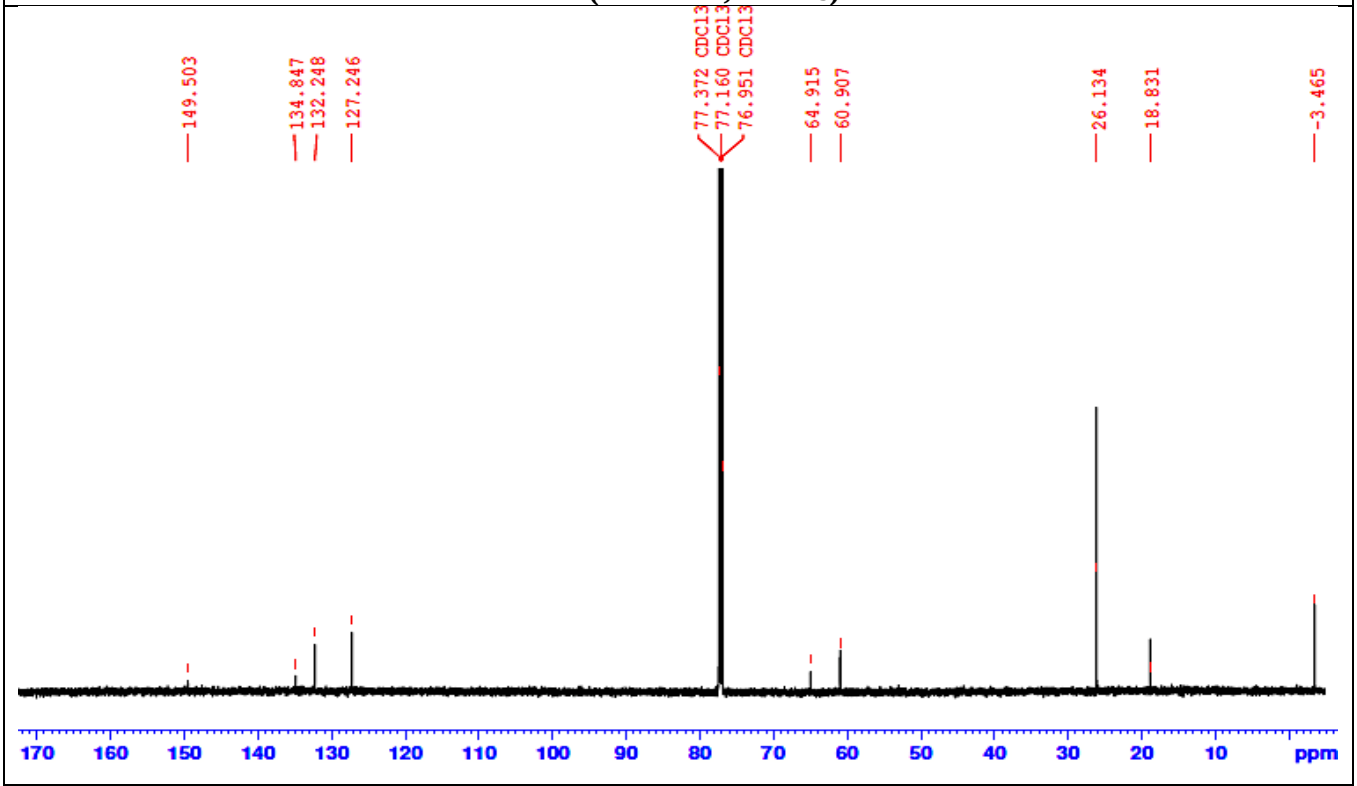
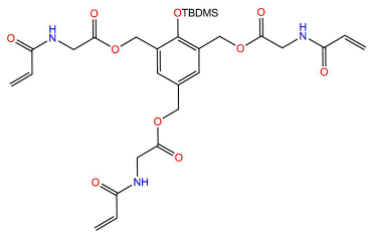



**$^{13}$C NMR (125 MHz, $CDCl_3$):**



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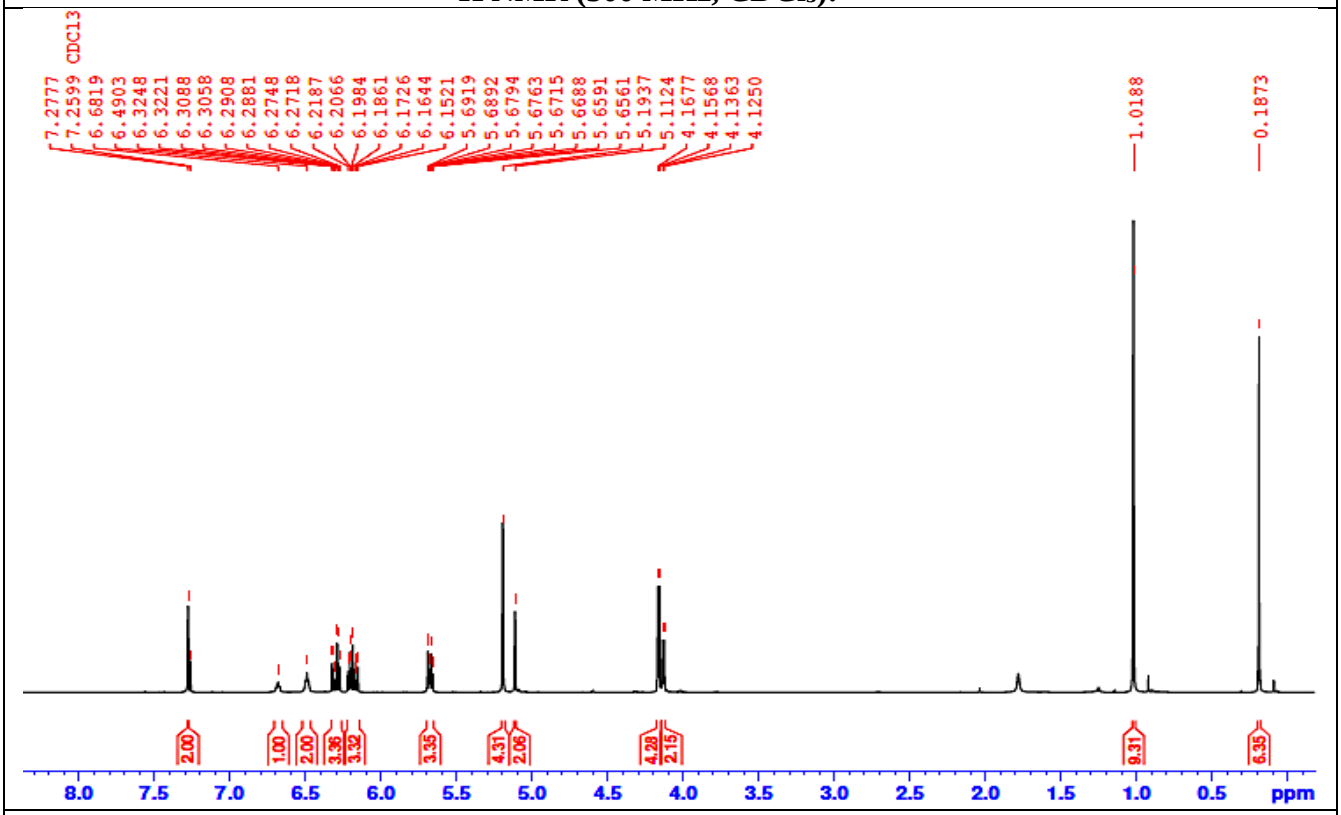
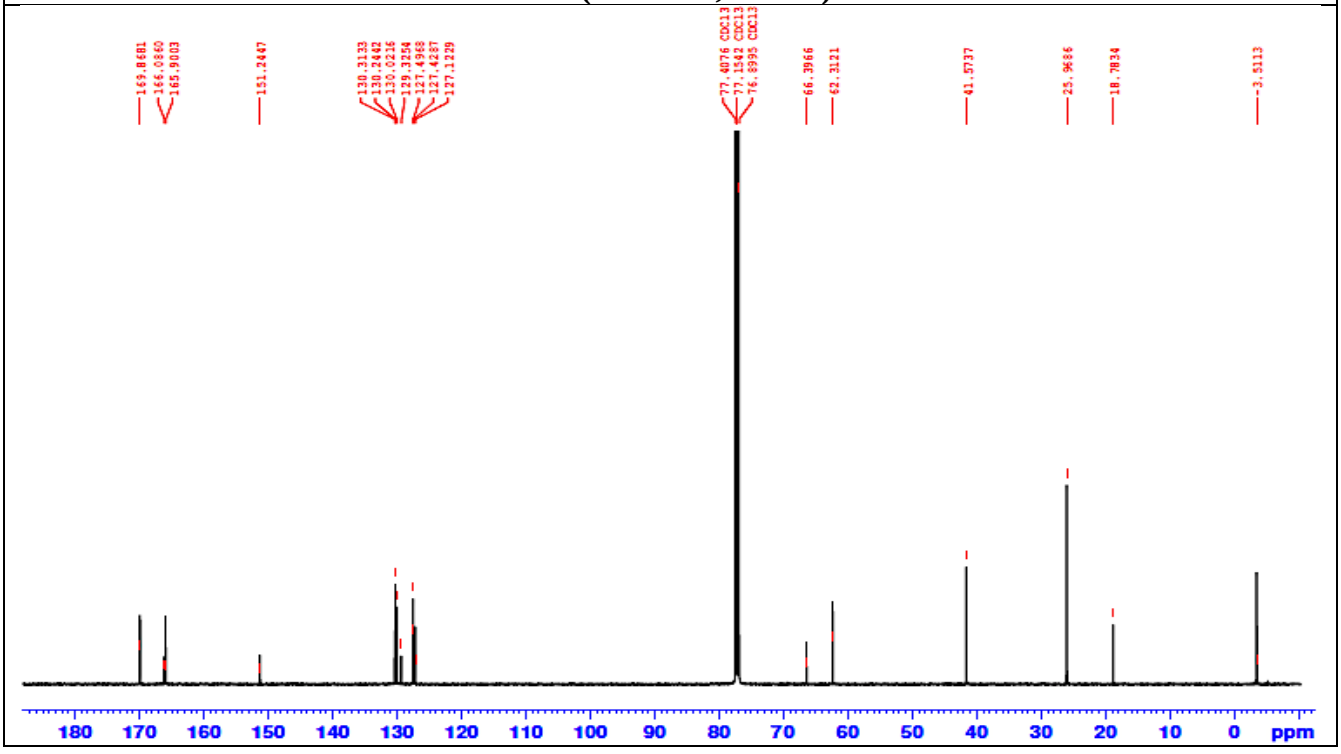
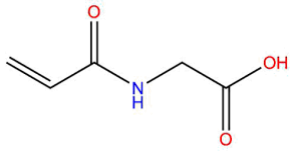



**$^{13}$C NMR (125 MHz, $CDCl_3$):**



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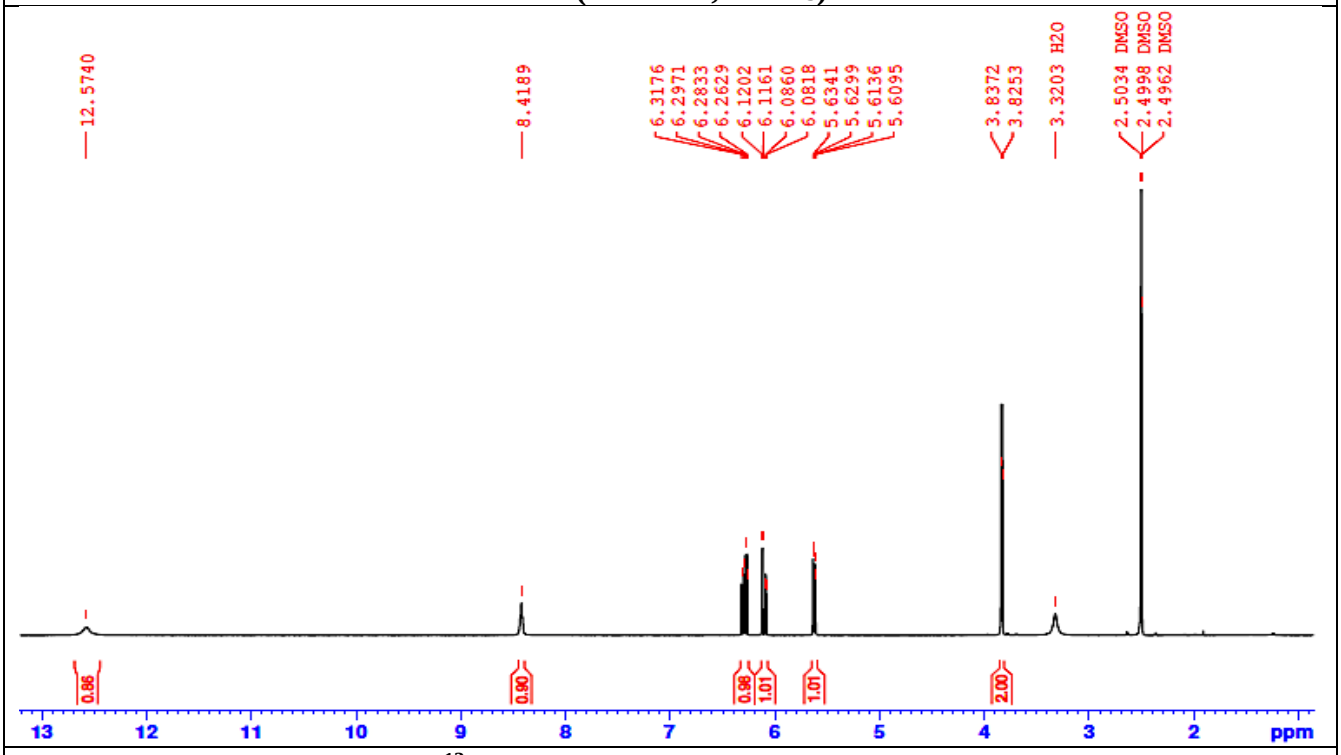


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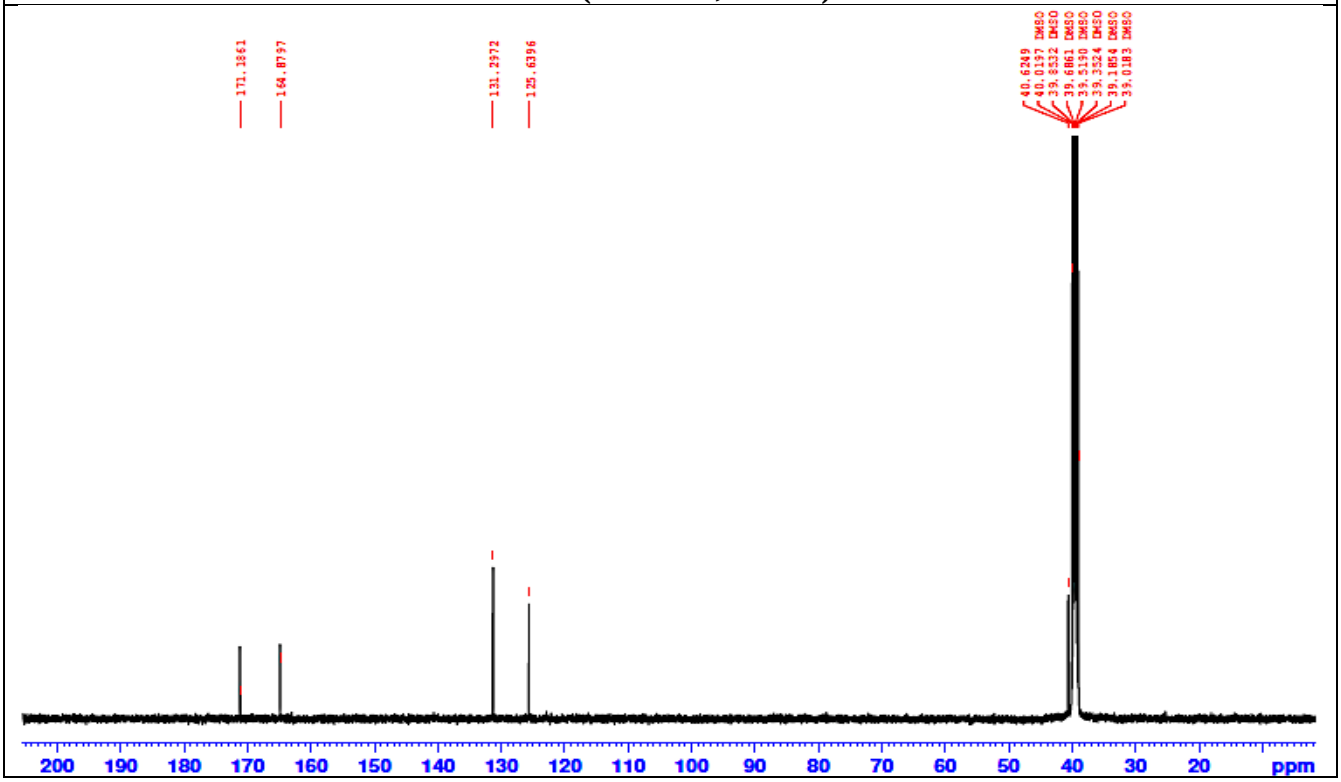

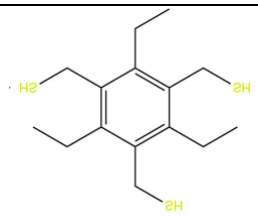

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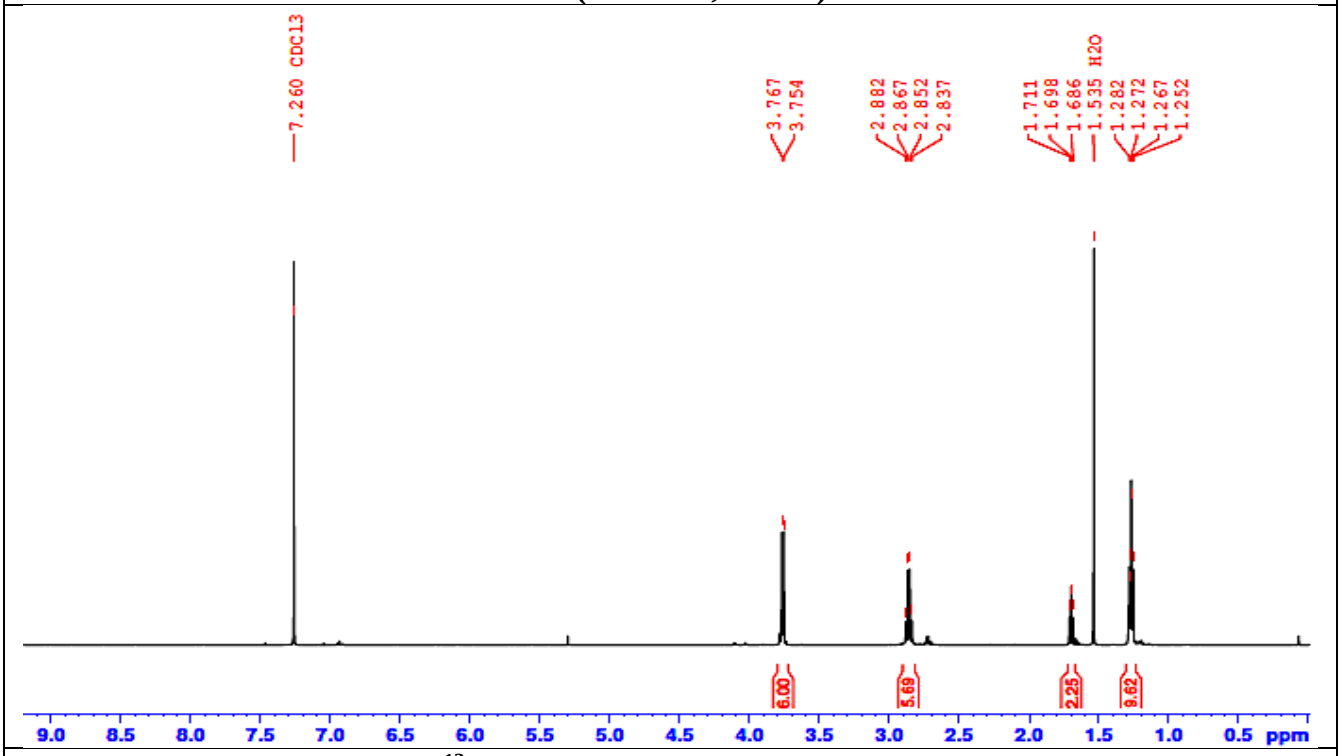


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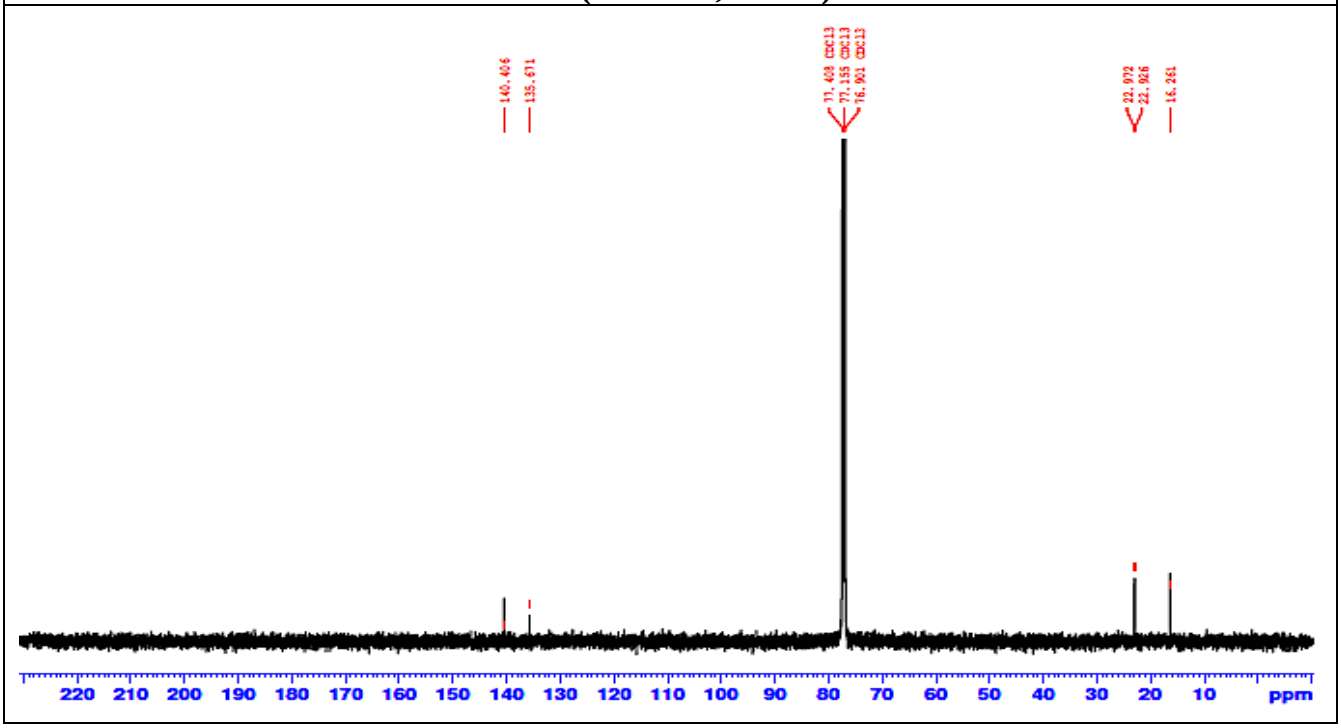

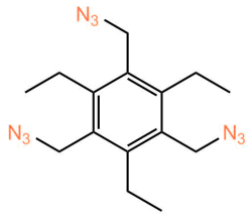


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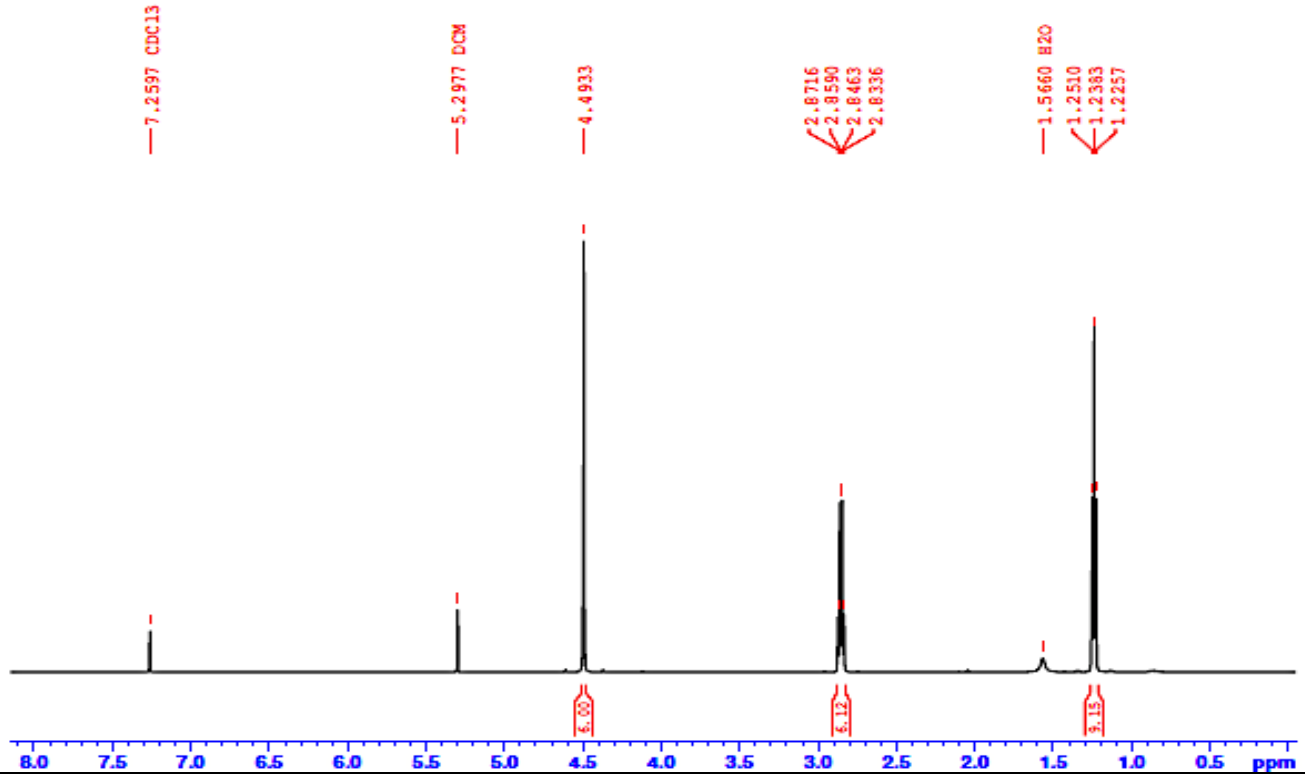


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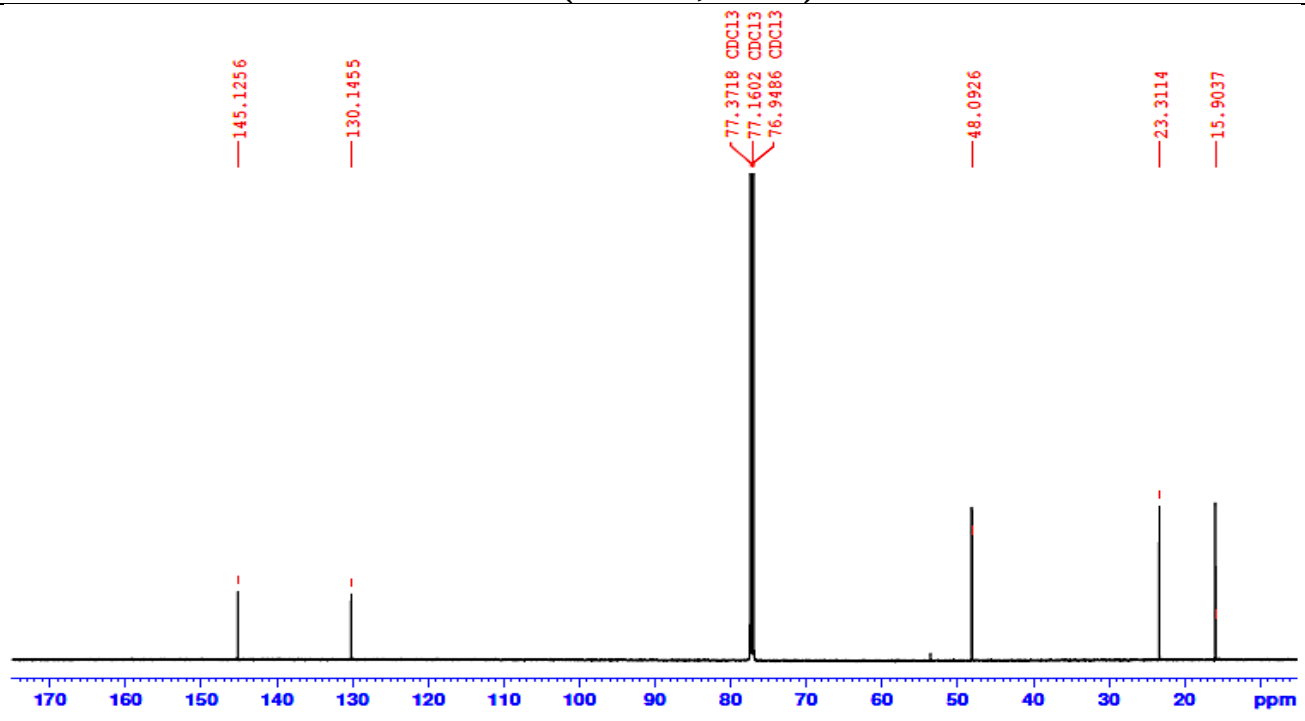

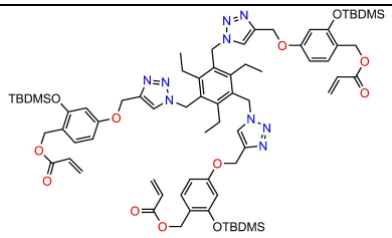


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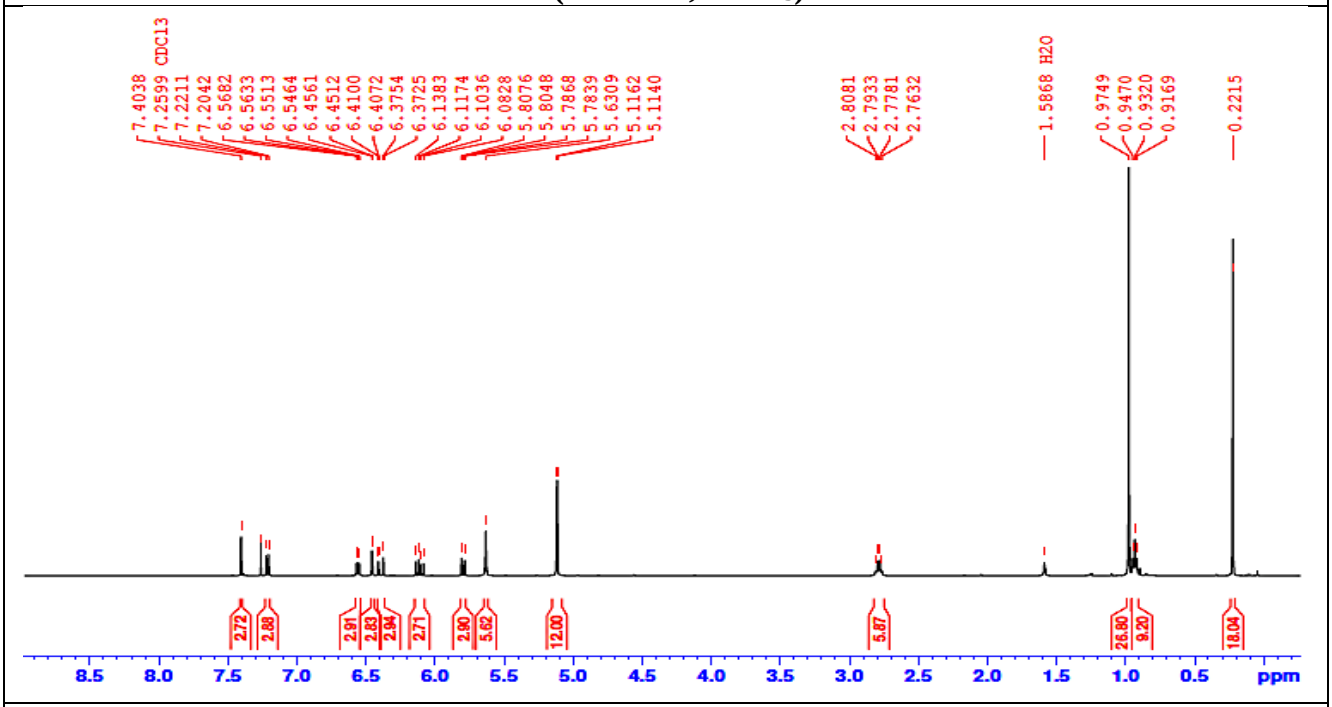


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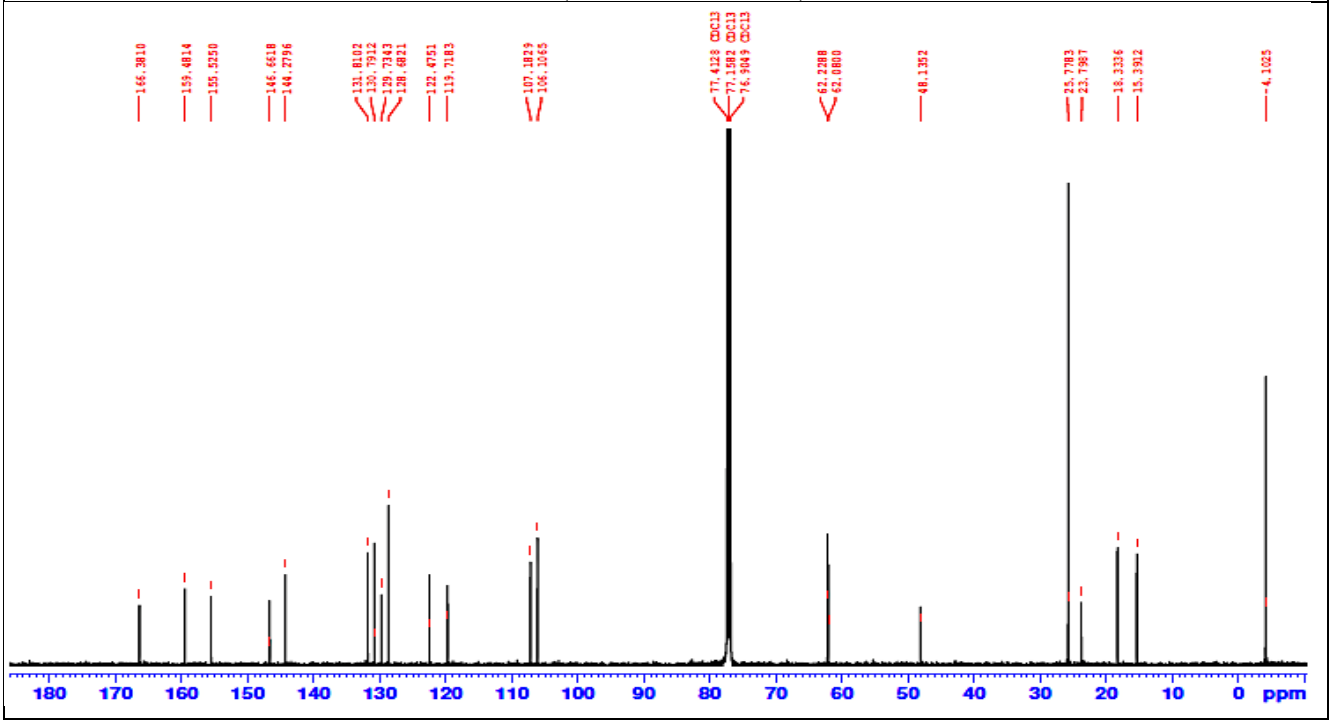

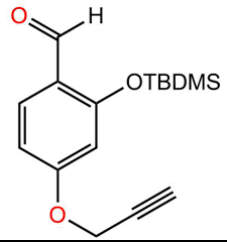


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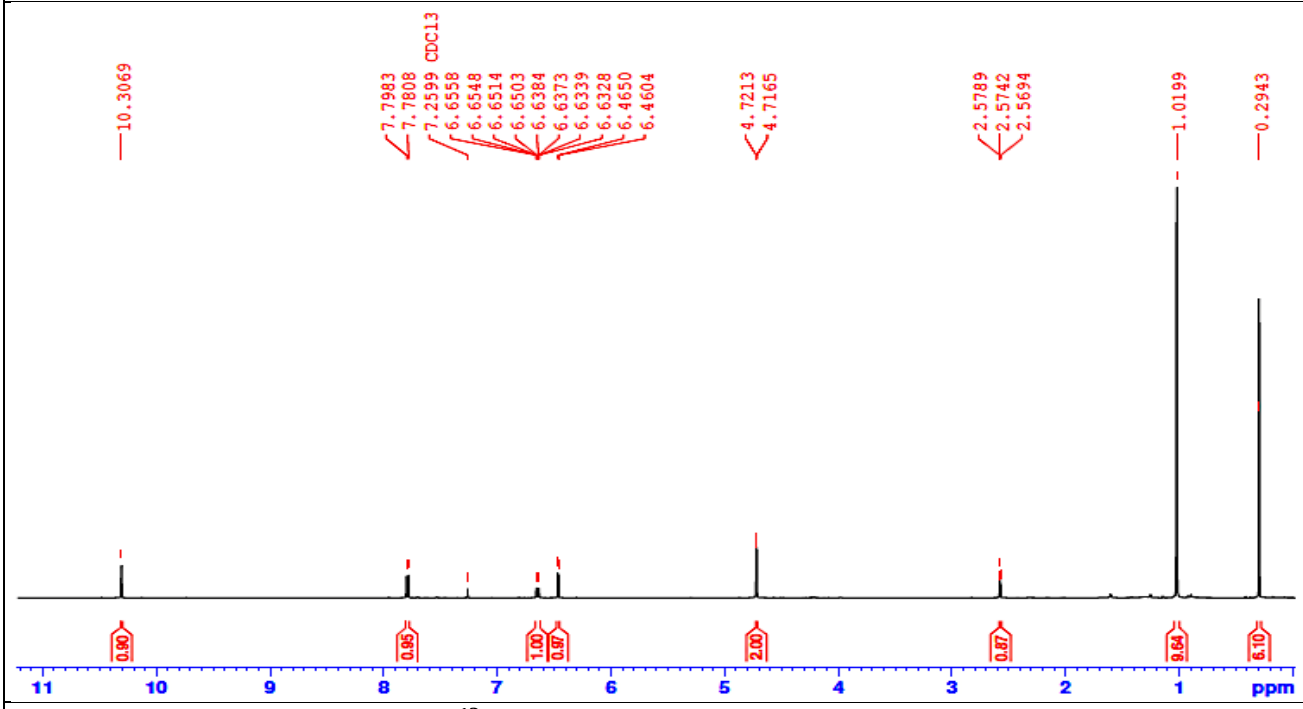


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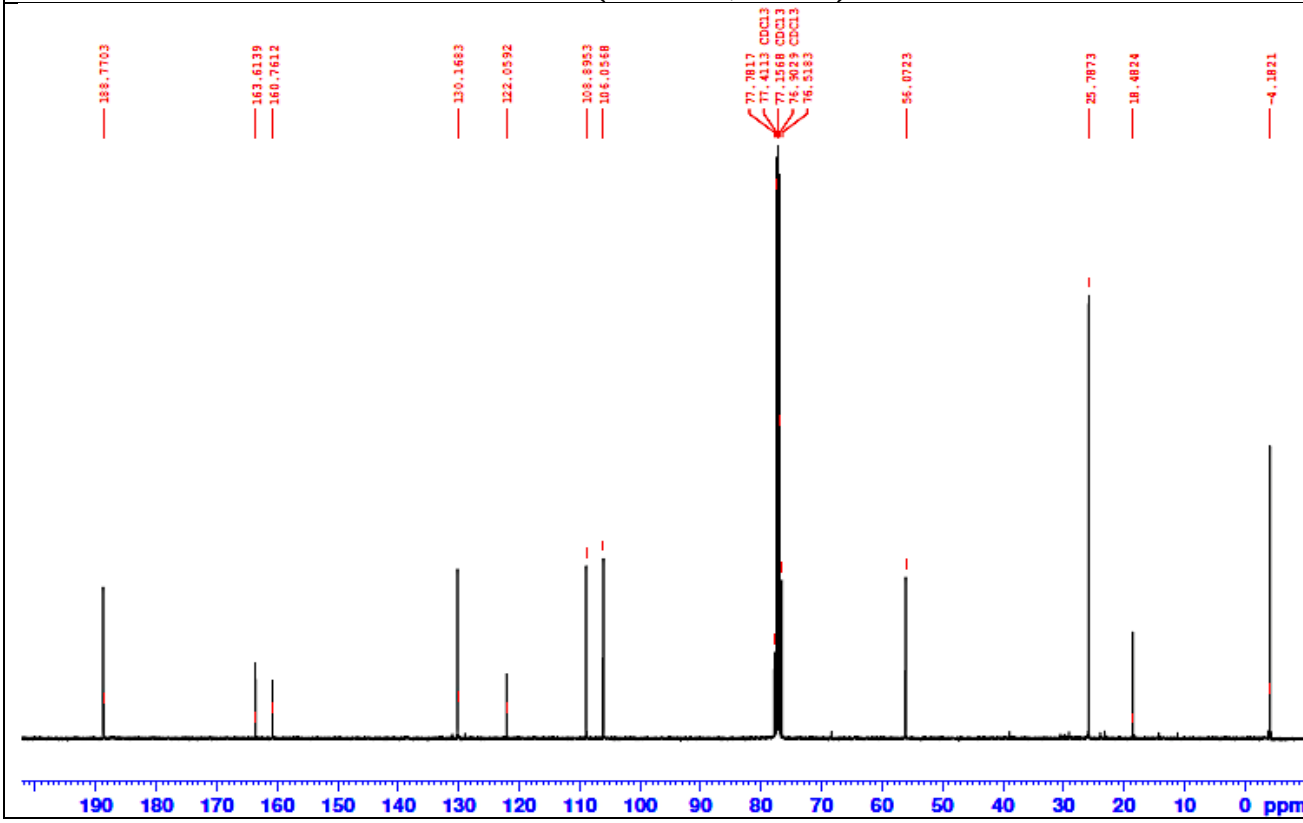

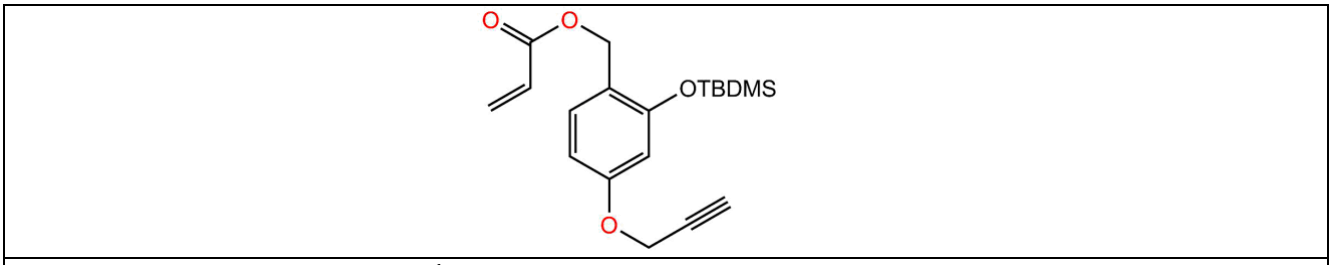


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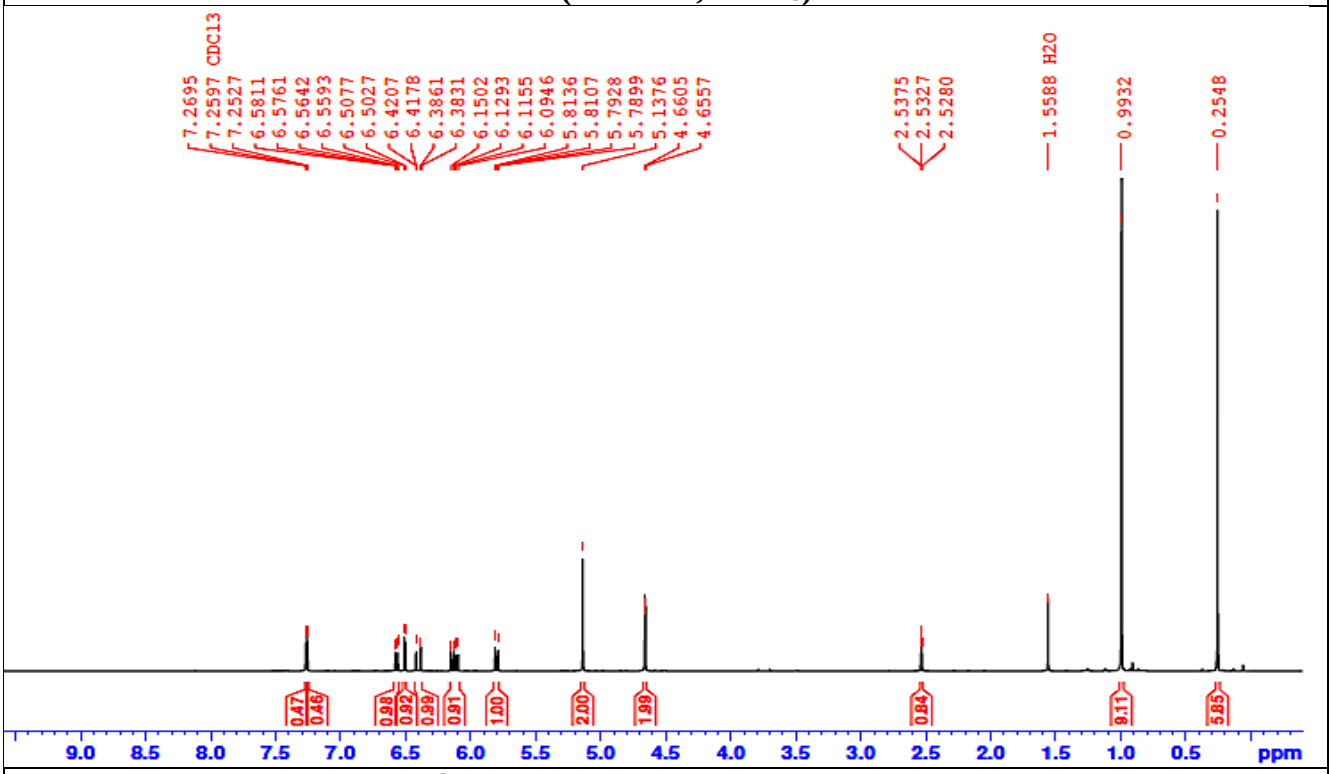


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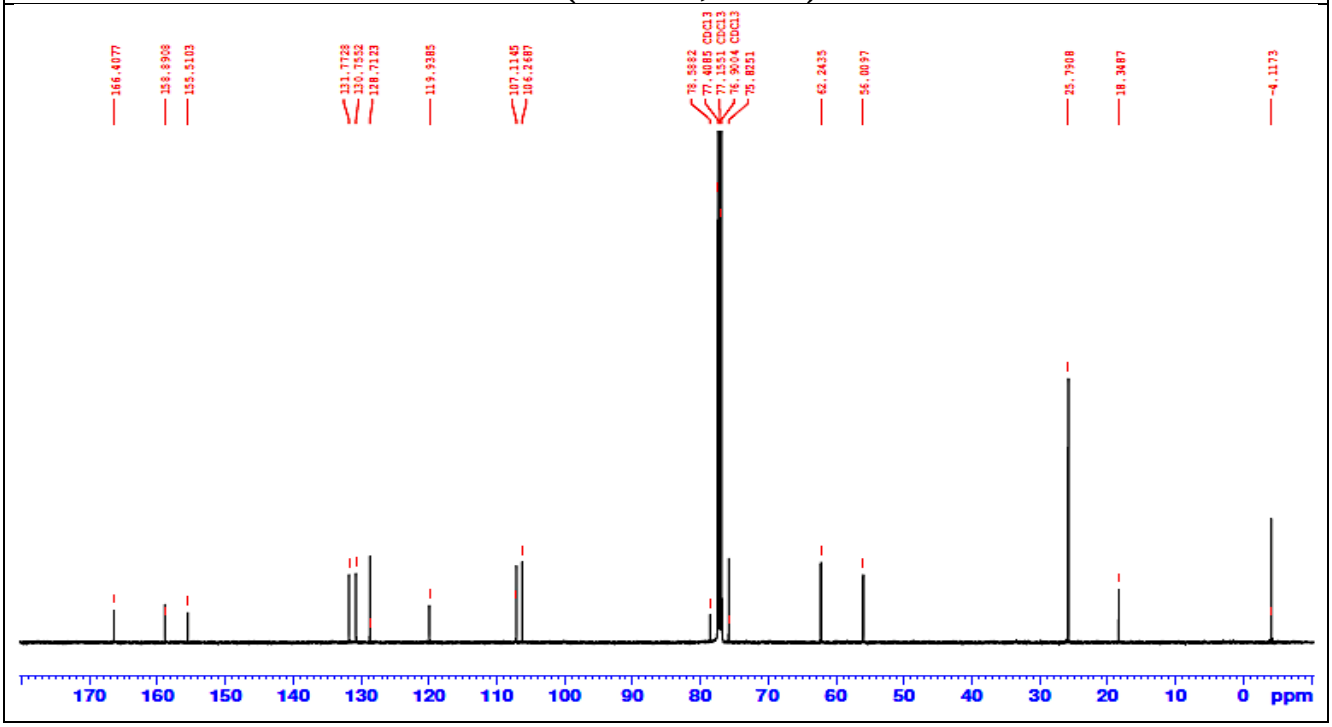

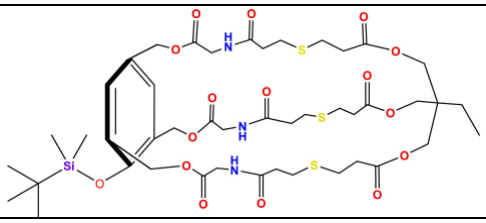

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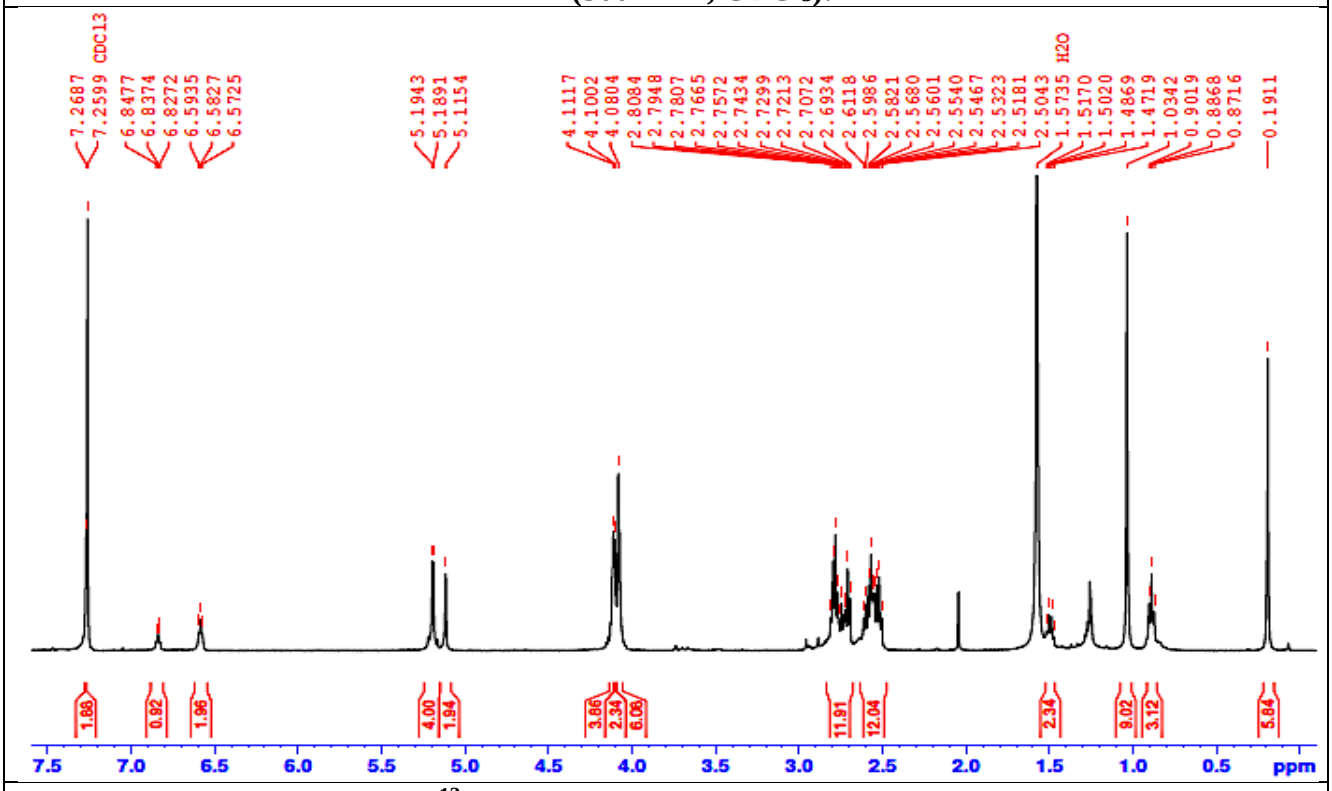


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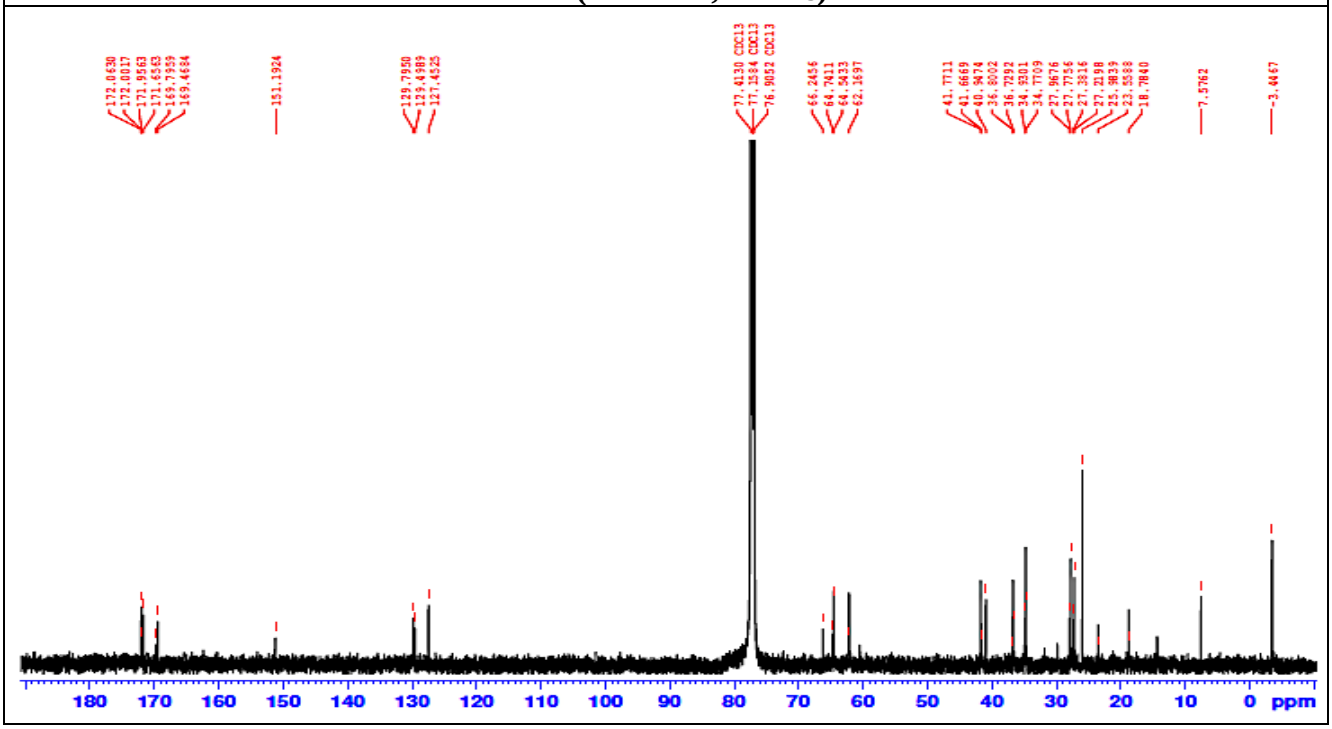

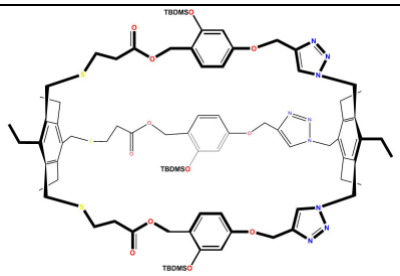

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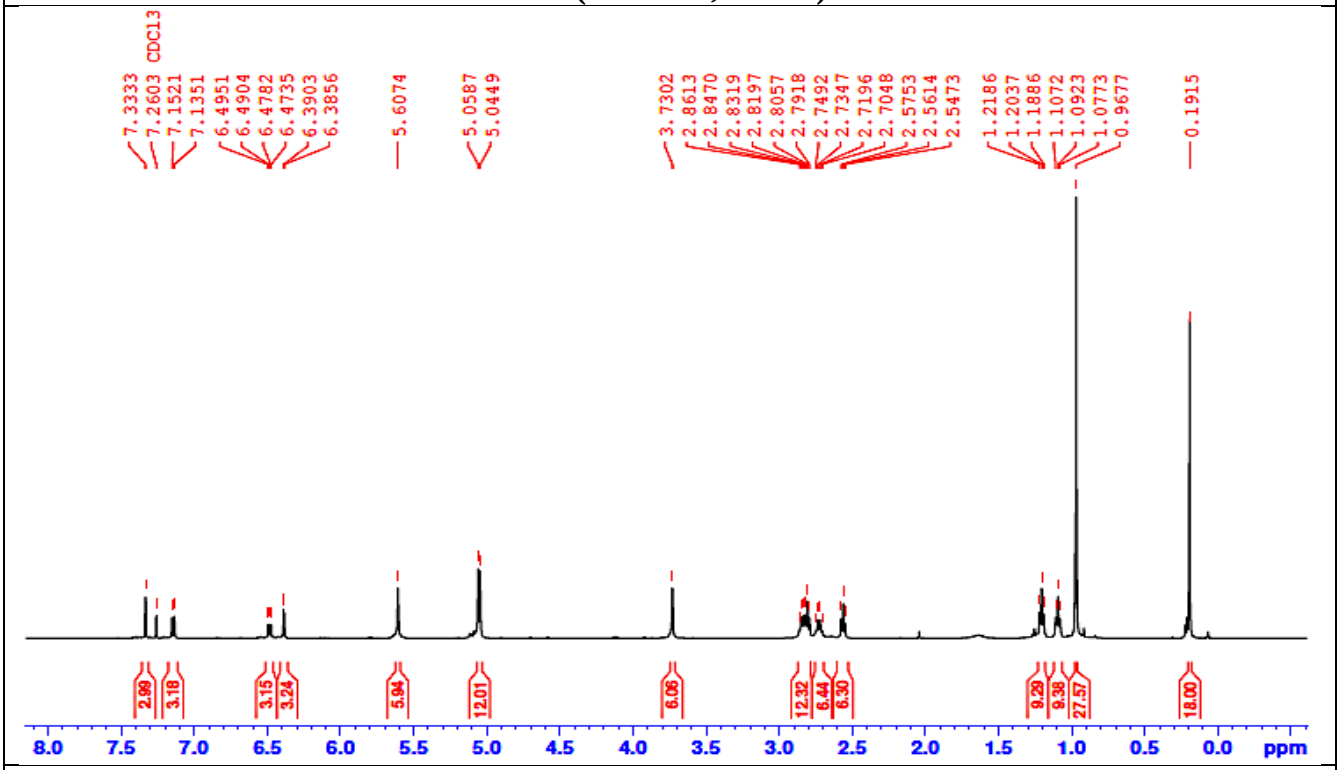


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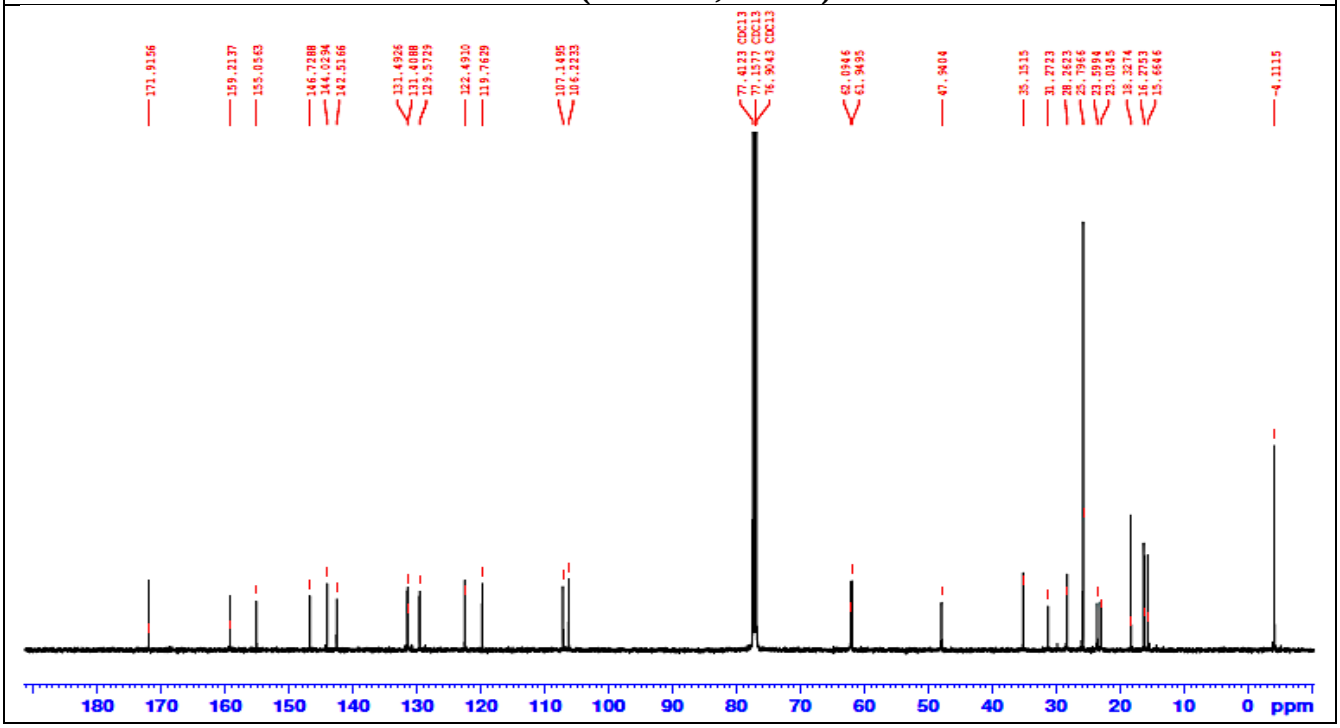

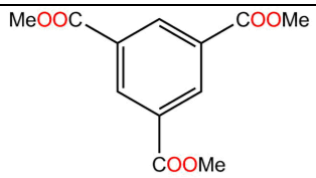


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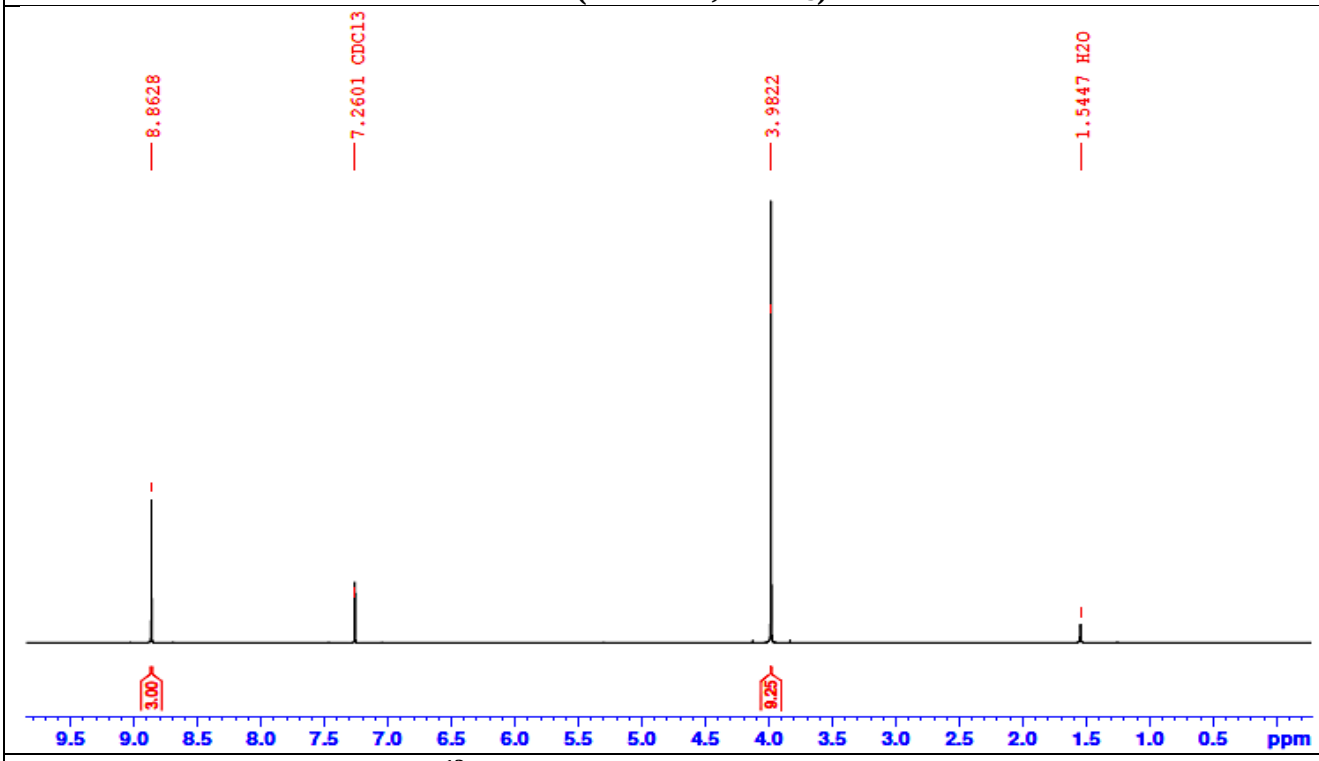


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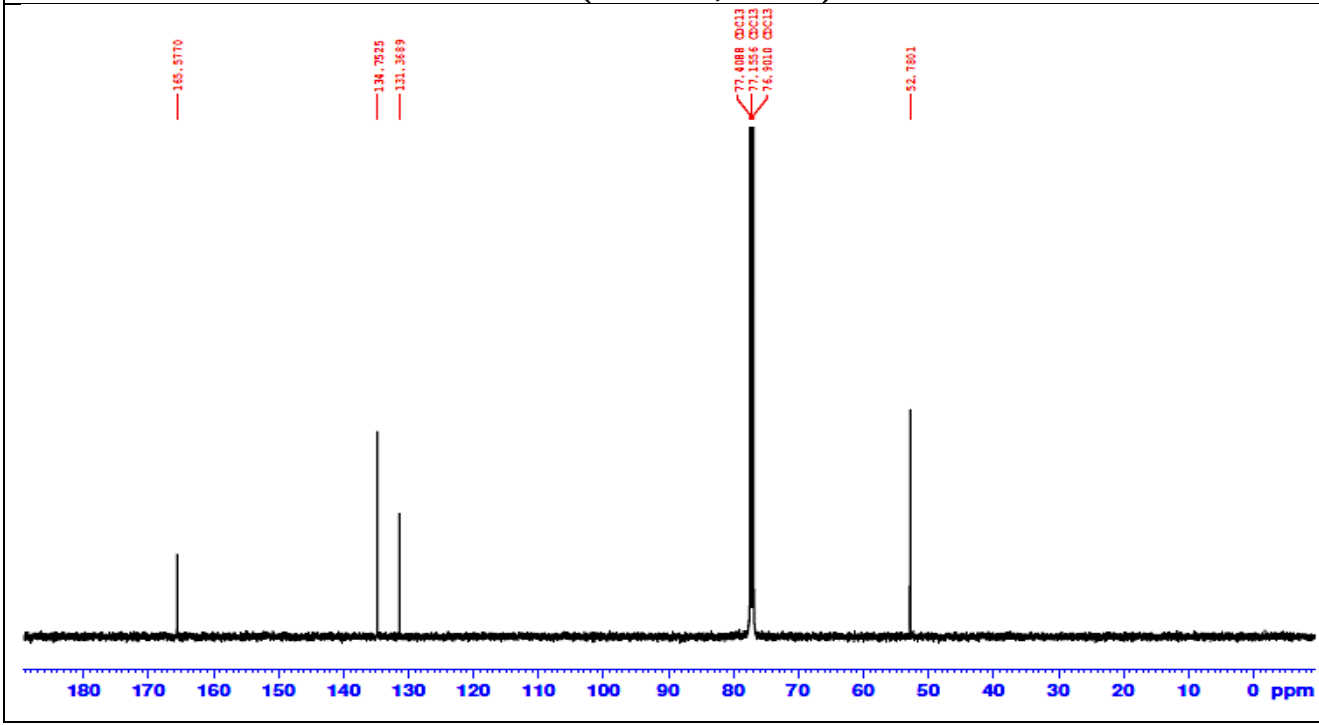

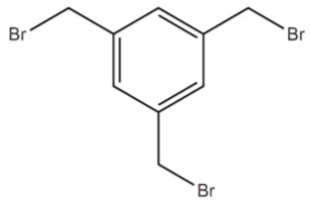


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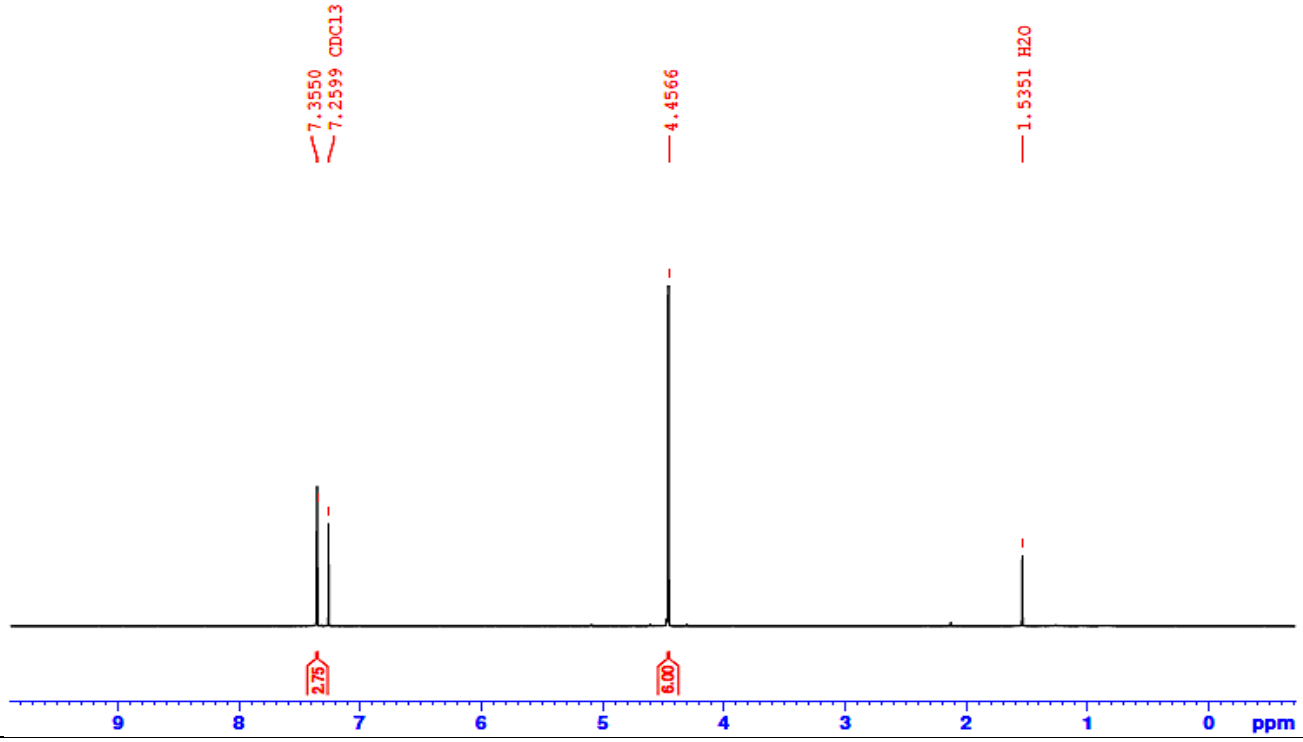


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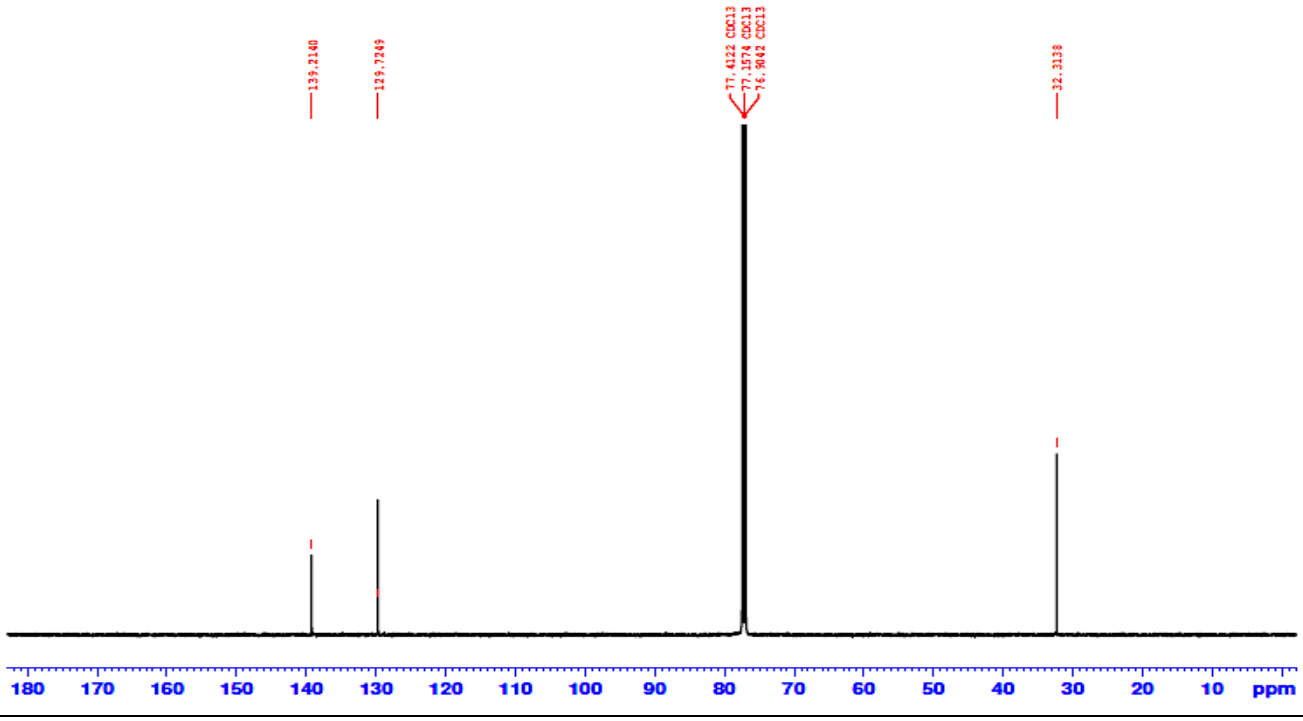

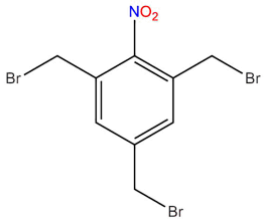


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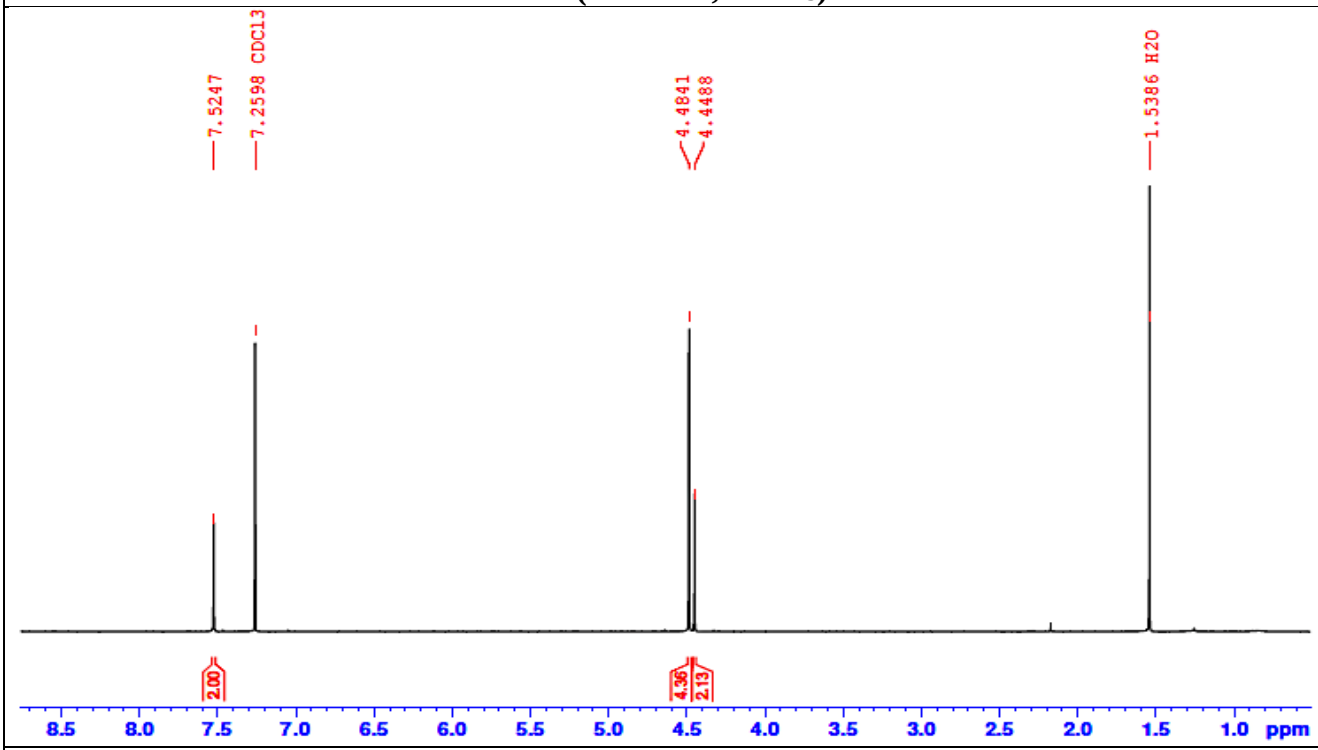


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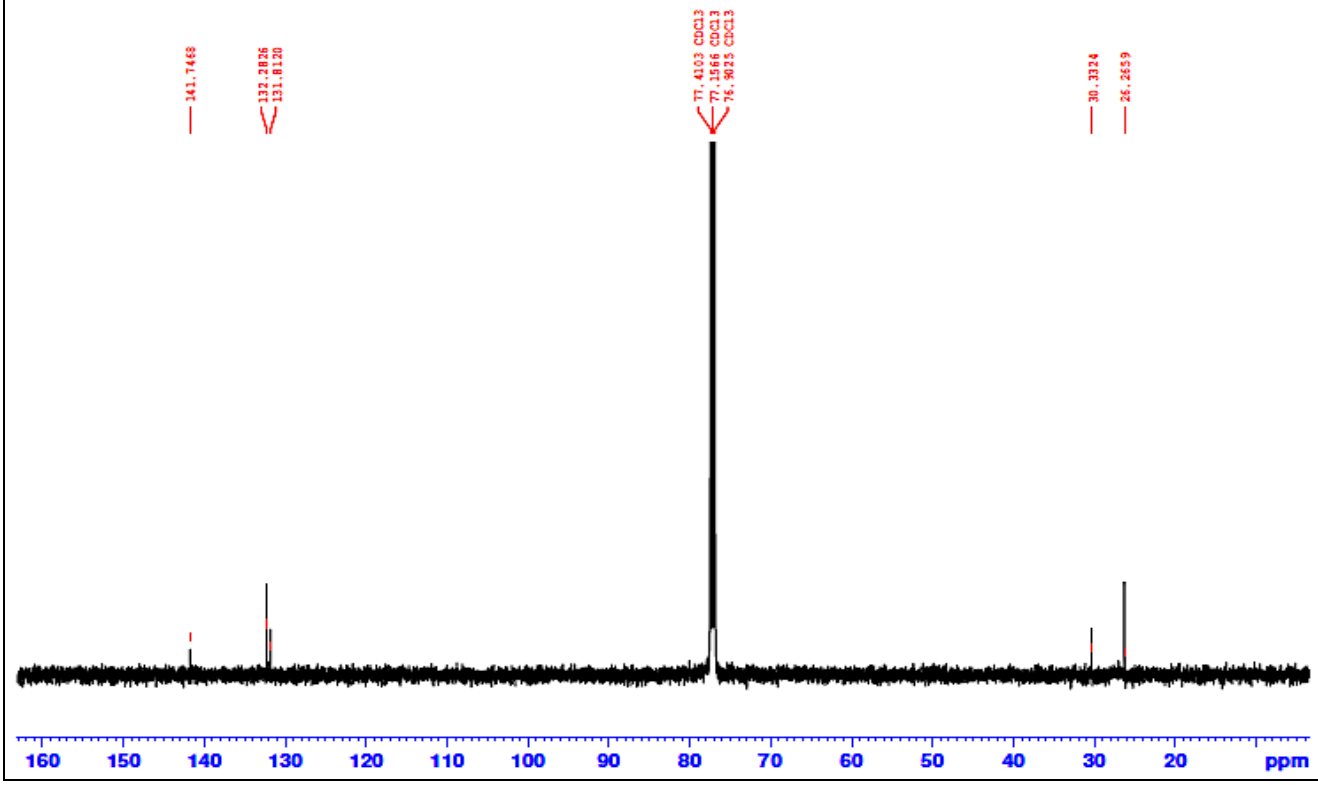

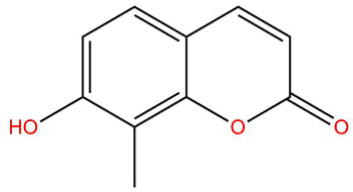


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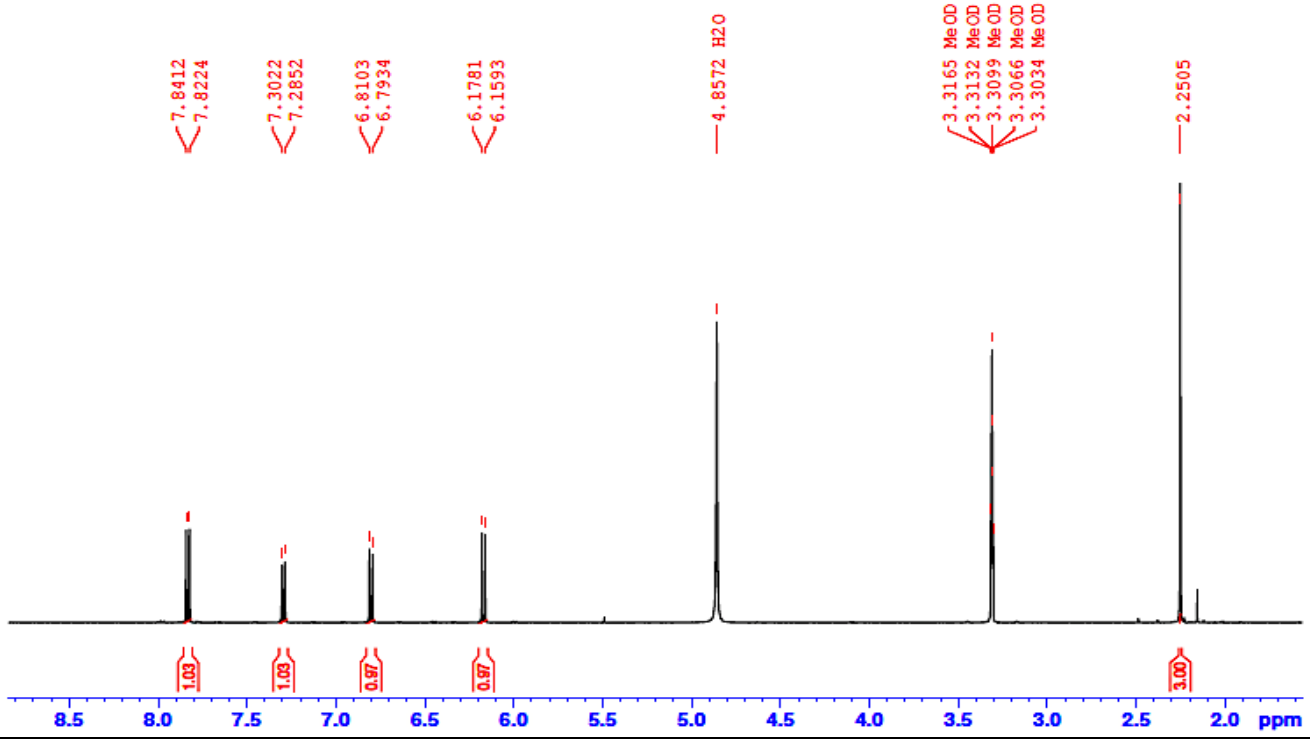


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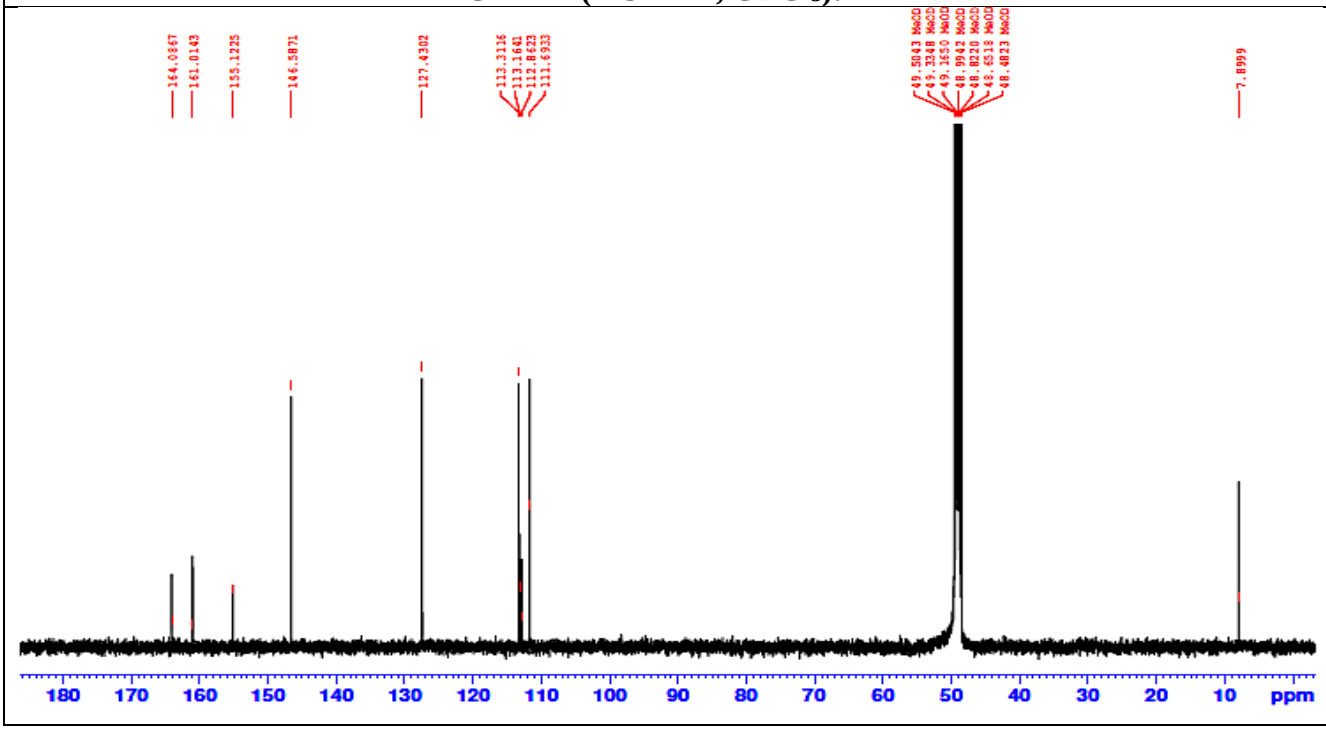
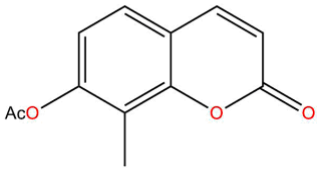
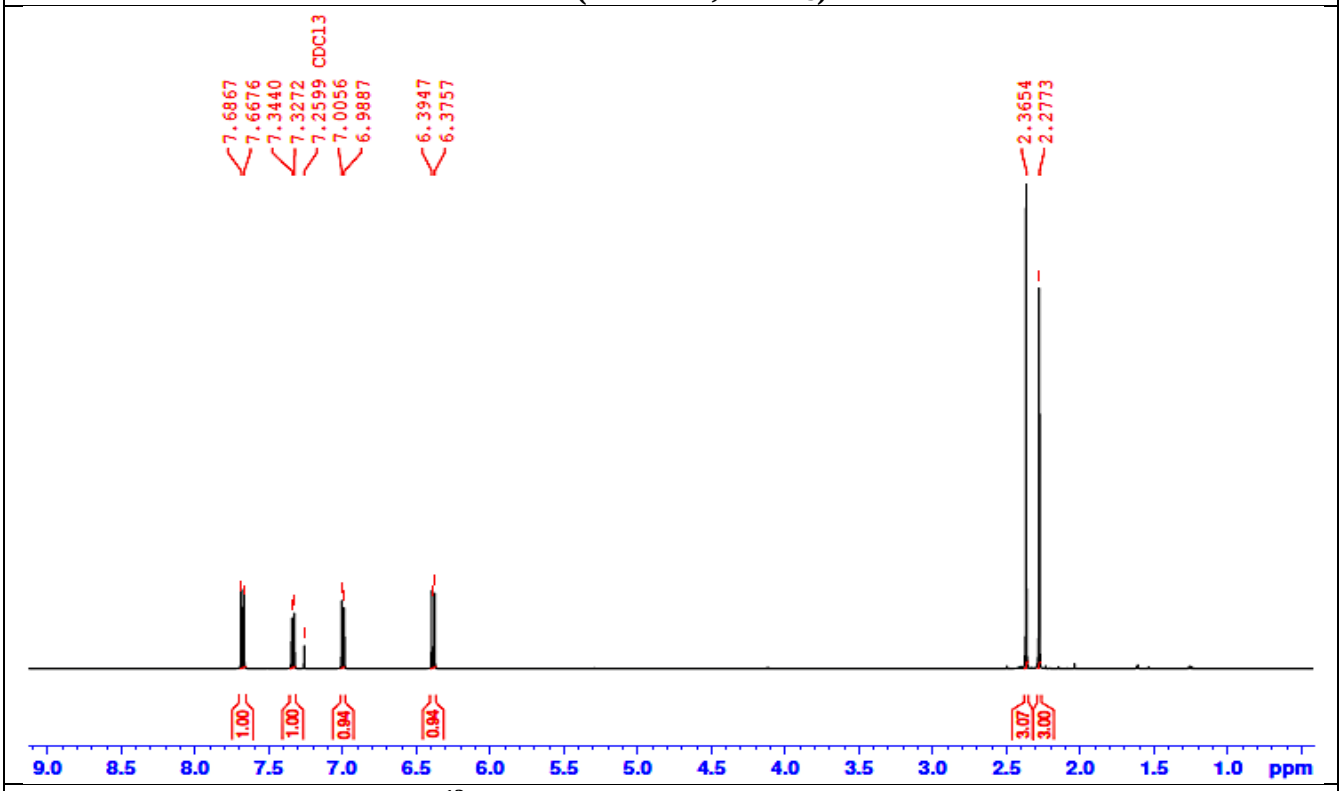
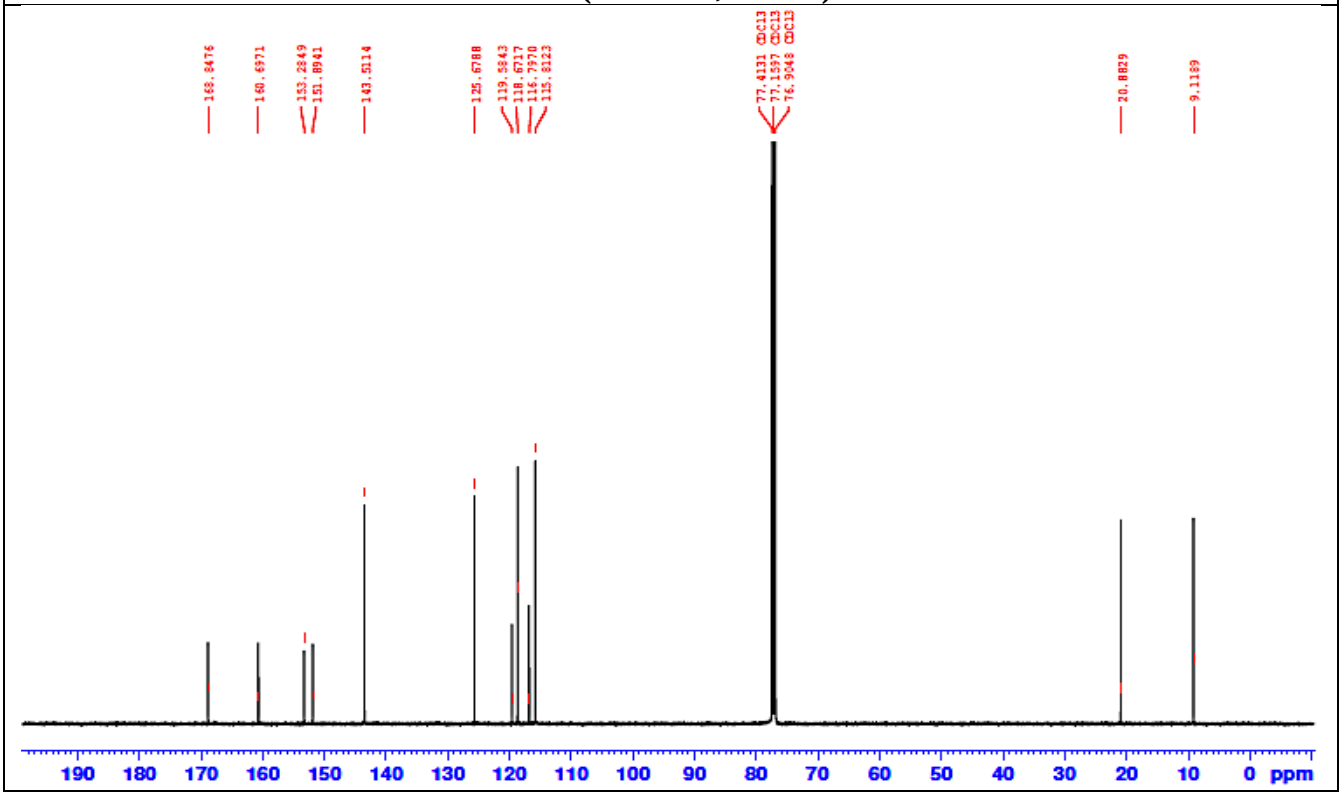


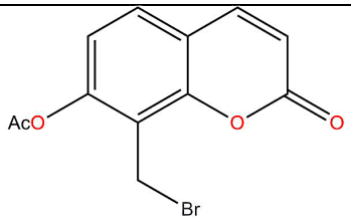



**$^{1}$H NMR (500 MHz, $CDCl_3$):**

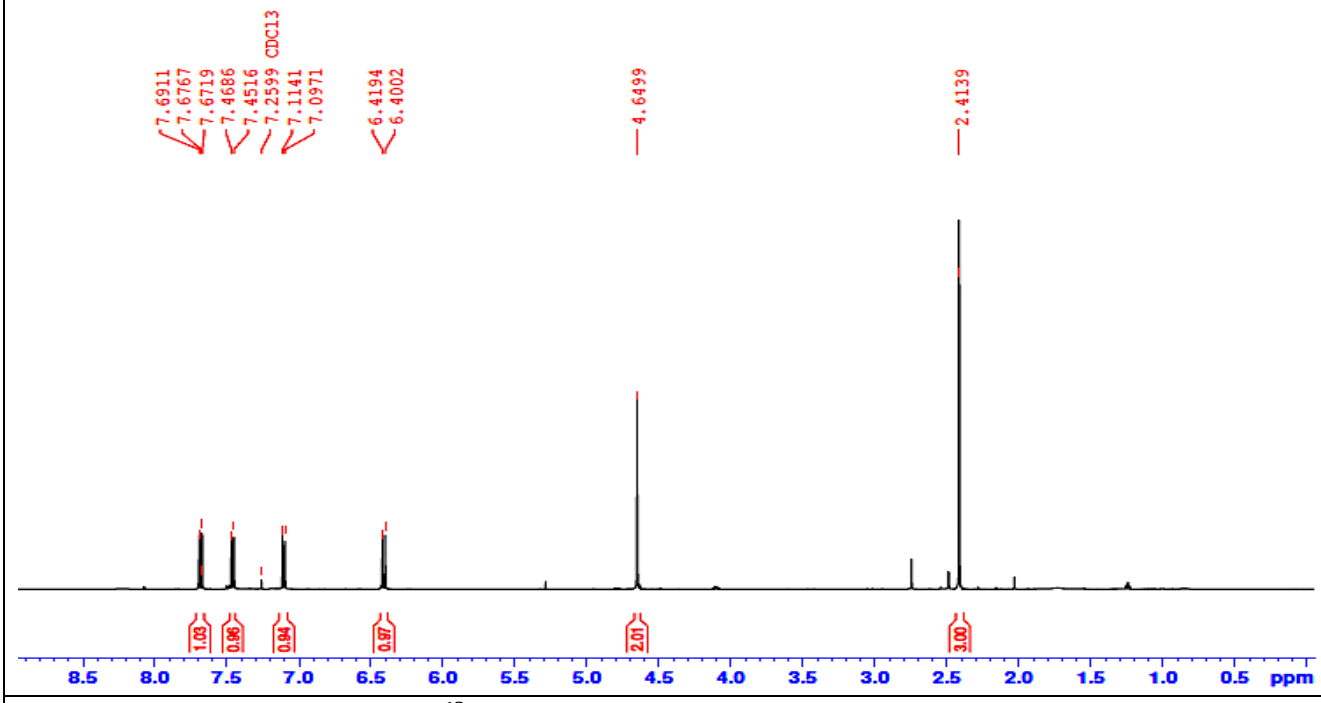



**$^{13}$C NMR (125 MHz, $CDCl_3$):**

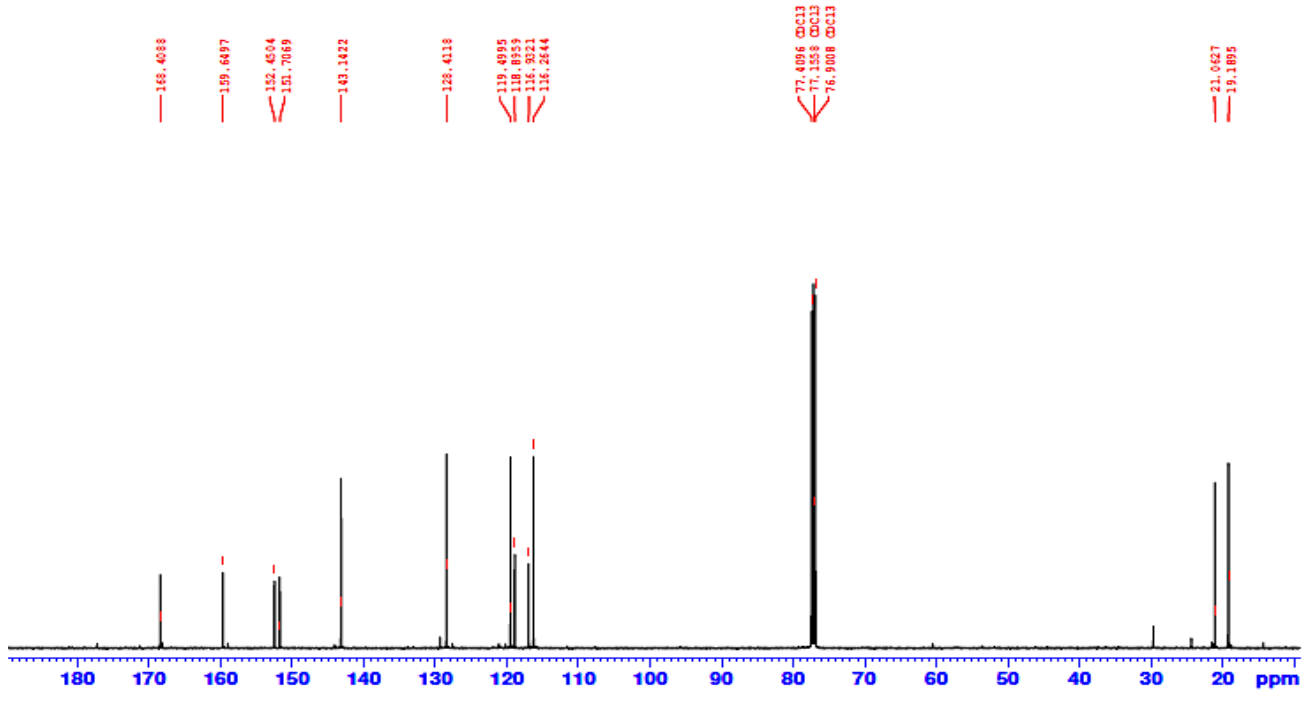


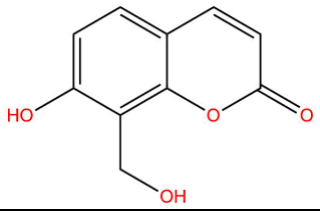



**$^{1}$H NMR (500 MHz, $CDCl_3$):**

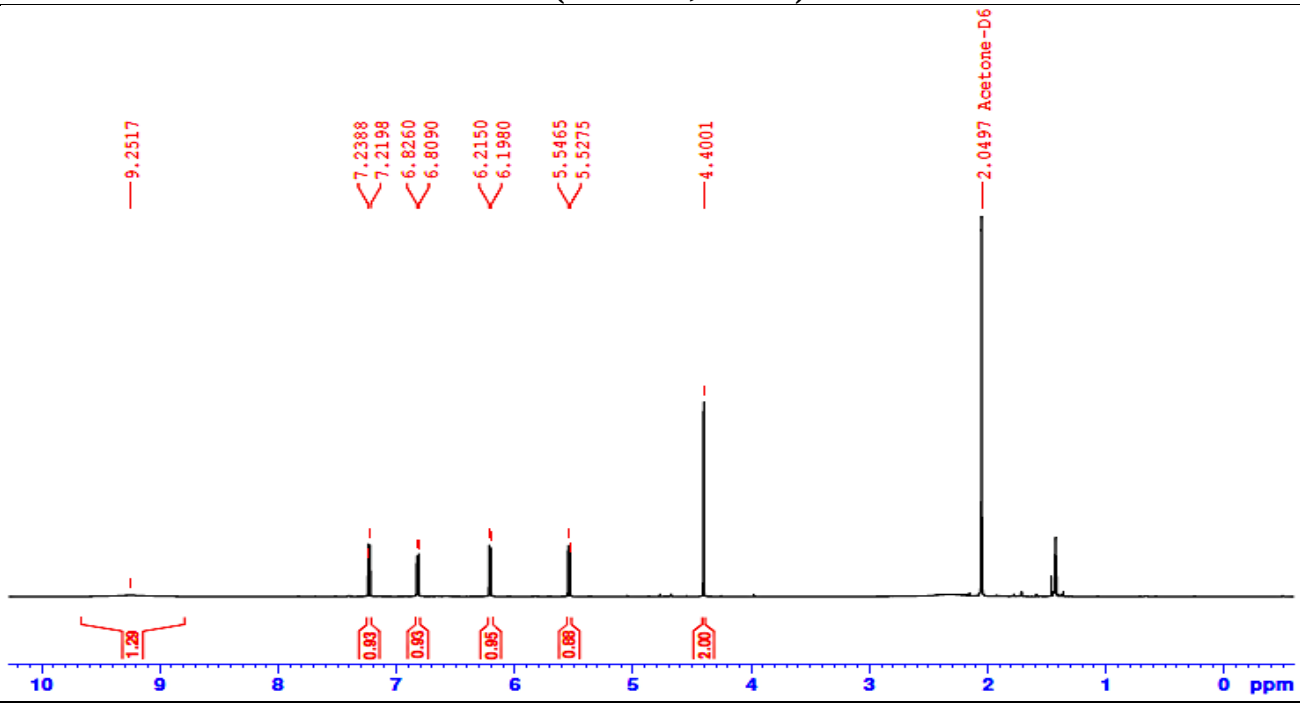



**$^{13}$C NMR (125 MHz, $CDCl_3$):**

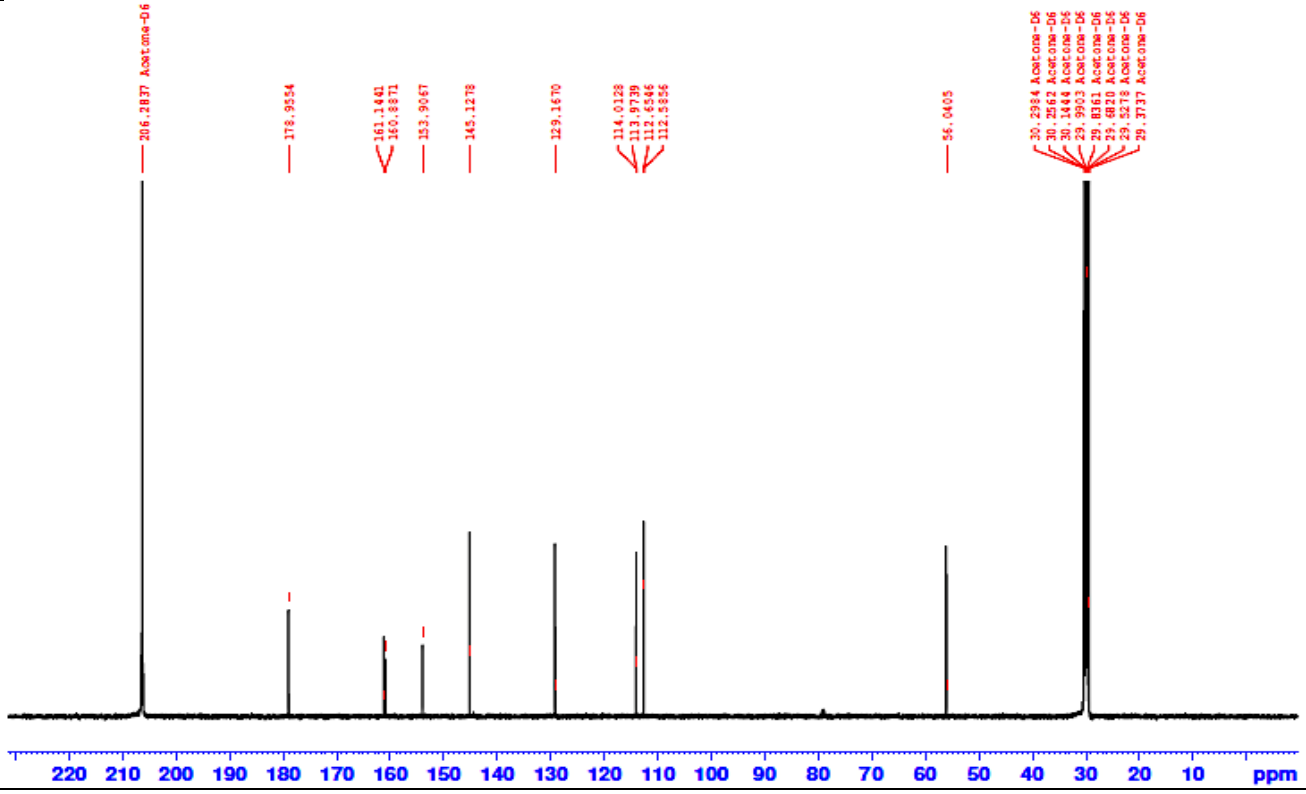





**$^{1}$H NMR (500 MHz, $CDCl_3$):**




**$^{13}$C NMR (125 MHz, $CDCl_3$):**



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