Synthesis of 1,4-di-*t*-butyl-2,5-dimethoxybenzene
Supplementary Material

The synthesis of 1,4-di-*t*-butyl-2,5-dimethoxybenzene was performed and tested by more than 300 undergraduate second-year Chemistry students, to illustrate aromatic electrophilic substitution on an aromatic ring activated by metoxy groups. This experiment allows students to analyze the relative reactivity of the aromatic ring and the effect of the orientation of the substituents. Polyalkylation can be discussed once in this experiment only the dialkylated product is obtained because the two remaining positions on the ring are too sterically hindered for further reactions to occur. Starting from methoxybenzene, *ortho* and *para* alkylated products should be obtained. Friedel-Crafts alkylation is explored in this case as an alternative for the nitration of aromatic rings, usually studied during the first year. All unit operations are basic and this experiment may be performed by undergrad students from any course other than Chemistry, as long as it includes Experimental Organic Chemistry classes.

A microscale procedure for this experiment can be found in literature[1].

Additional notes for the preparation of 1,4-di-*t*-butyl-2,5-dimethoxybenzene:

Some changes were made on the procedure described in literature[2]. The reaction is performed using a three-neck round bottom flask instead of an Erlenmeyer flask, and manual stirring with a glass rod was replaced by magnetic stirring (figure **SM 5.2.1.1**). This minimizes the risks associated with the handling of concentrated and fuming sulfuric acids. The mixture of acids can be added in rapid drops over the course of 4-7 minutes, during which, precipitation of the product is observed. Figure **SM 5.2.1.2** shows the Vacuum filtration set-up. Yield of 1,4-di-*t*-butyl-2,5-dimethoxybenzene is 35-40%. Melting point is between 100 and 105ºC with a melting point range never superior to 2ºC (lit.: 104-105ºC[2] or 103-105ºC[3]).

Figure SM 5.2.1.1 – Reaction set apparatus for 1,4-di-*t*-butyl-2,5-dimethoxybenzene

Figure SM 5.2.1.2 – Vacuum filtration set-up.

IR spectrum:

IR spectrum of 1,4-di-*t*-butyl-2,5-dimethoxybenzene can be found online at the National Institute of Standards and Technology database[4]. Students easily identify a strong absorption band at 3000-2800 cm^{-1} due to the C-H stretching vibrations.
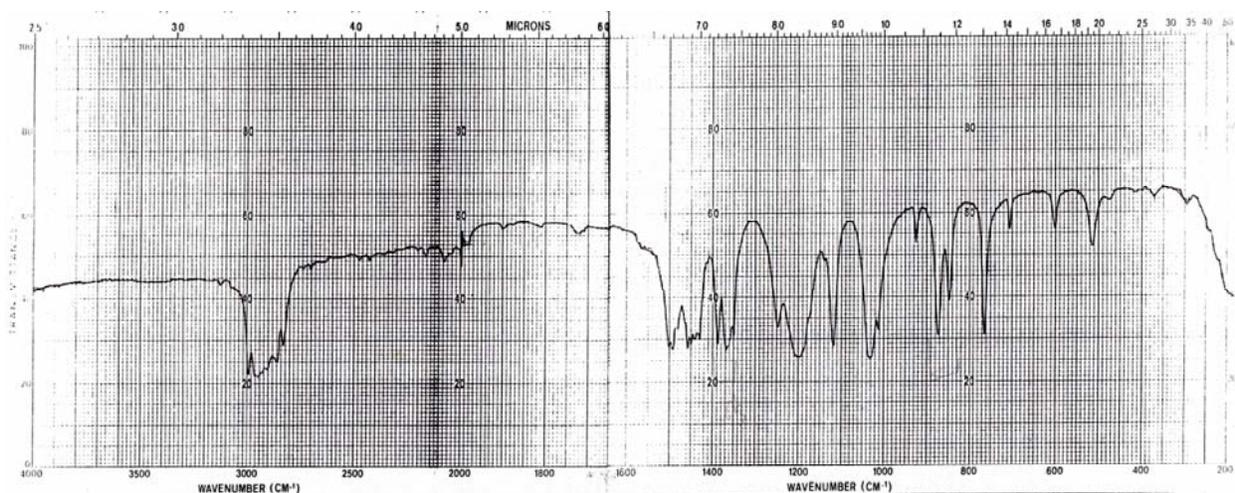
Figure SM 5.2.1.3: IR (KBr) of 1,4-di-*t*-butyl-2,5-dimethoxybenzene

^{1}H NMR spectrum:

The students easily assign the ^{1}H NMR spectra once only three singlets are present. The peak solvent (chloroform) at 7.26 ppm and a solvent impurity at 2.2 ppm can be observed (**Figure SM 5.2.1.4**).
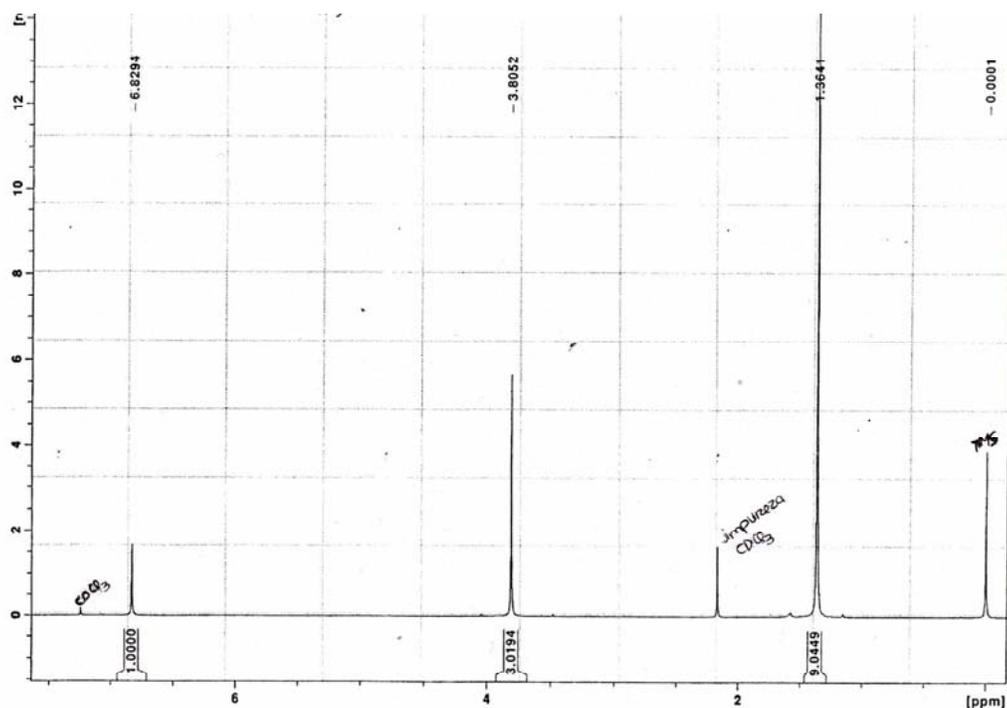
Figure SM 5.2.1.4: ^{1}H NMR (CDCl$_3$) of 1,4-di-*t*-butyl-2,5-dimethoxybenzene

[1] C. N. Hammond, M. J. Tremelling, *J. Chem. Ed.*, 1987, **64**, 440.
[2] L. Fieser, K. Williamson, *Organic Experiments*, 1998, 303.
[3] *Beilstein*, E IV 6, 6075.
[4] http://webbook.nist.gov/cgi/cbook.cgi?ID=C7323639&Units=SI&Mask=80, access in July 2015.

Synthesis of 4,4'-di-tert-butylbiphenyl
Supplementary Material

This experiment proposal was tested by students of elementary organic chemistry in which the concepts of electrophilic aromatic substitution were taught. This work is a great example to illustrate a Friedel-Crafts alkylation (namely the important role of Lewis acid catalyst in the electrophile formation), the orientation in electrophilic aromatic substitution and the increase of ring reactivity that lead to polyalkylation (one of Friedel-Crafts alkylation limitations). Experimental procedure can be easily performed by first and second-years undergraduate students and allow the use of gas trap to remove hydrogen chloride, a side product.

Additional notes on 4,4'-di-tert-butylbiphenyl

Dichloromethane was previously dried over anhydrous calcium chloride and was used without distillation. The reaction set-up apparatus including the gas trap is showed in **Figure SM 5.2.2.1**. Care should be taken with the gas trap. The funnel must be partially immersed in the water. Complete immersion could lead to water entry in the apparatus. Work in the fume hood could be avoided if leaks do not exist but is more safe use the fume hood. Three portions of aqueous HCl 10% solution are sufficient to liquid-liquid extraction. The dark grey solid obtained after removal of the solvent in the rotary evaporator turns white after addition of ethanol to proceed with purification. Usually c.a. 125 mL of ethanol are needed to recrystallize the product. If starting materials are dry, good yields are obtained around 70% and m.p between 123 and 128ºC (128.6-129ºC[1] or 127-128ºC[2]). The final product may be slightly yellow due to ferric chloride traces. According literature its can be removed by passing a hexane solution of the product in a column of basic alumina[2].

---

[1] *Dictionary of Organic Compounds*, Chapman and Hall, 5th ed., **1982**, D-02187.

[2] Deane A. Horne, *J. Chem. Educ.*, **1983**, *60* (3), 246.

Figure SM 5.2.2.1 – Reaction set-up apparatus for 4,4'-di-tert-butylbiphenyl

IR spectra:

IR spectrum for 4,4'-di-tert-butylbiphenyl is available in the literature (SDBS nº 8130)[3]

NMR spectra:

NMR spectra of starting compound were recorded in addition to compare with the 4,4'-di-tert-butylbiphenyl . ^{1}H and ^{13}C NMR spectra are also available in the literature (SDBS nº 1182 and 8130)[3].

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[3] http://sdbs.db.aist.go.jp/sdbs/cgi-bin/direct_frame_top.cgi
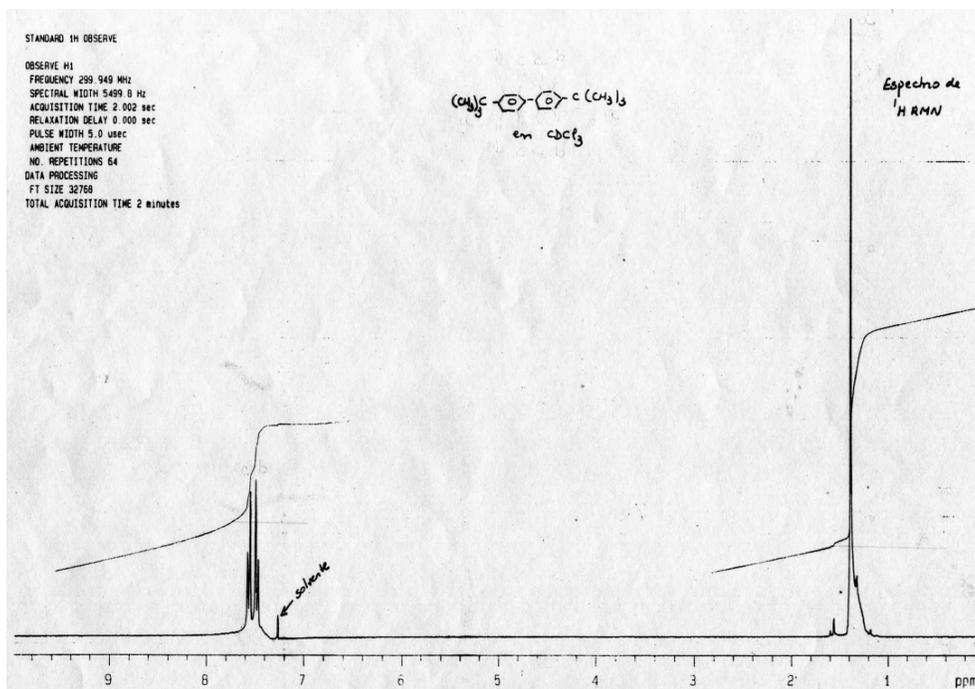
Figure SM 5.2.2.2 – ^{1}H NMR of 4,4'-di-tert-butylbiphenyl (300 MHz, CDCl$_3$)
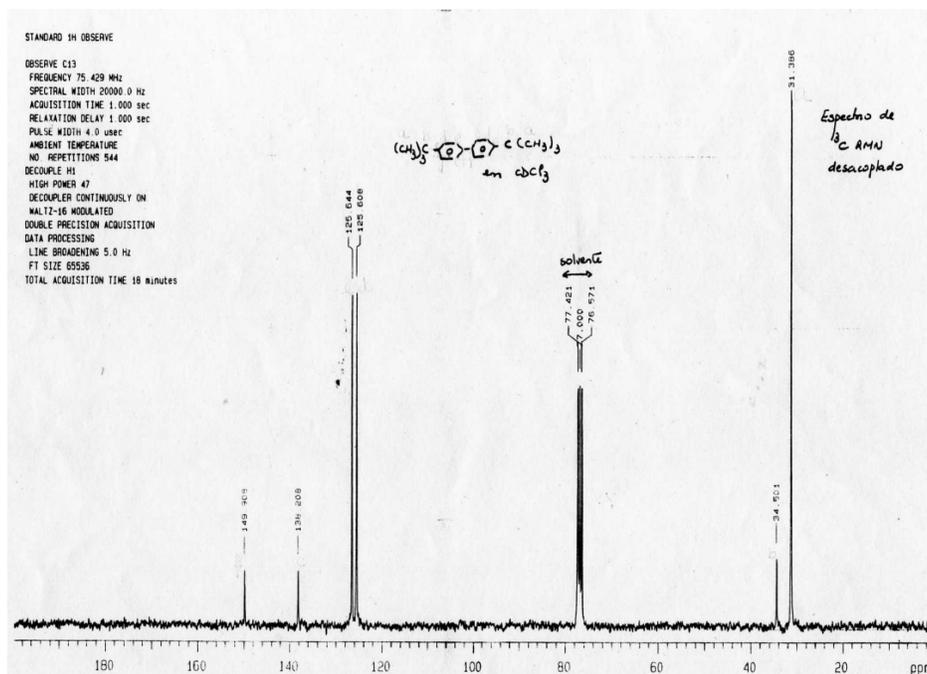
Figure SM 5.2.2.3 – ^{13}C NMR of 4,4'-di-tert-butylbiphenyl (300 MHz, CDCl$_3$)
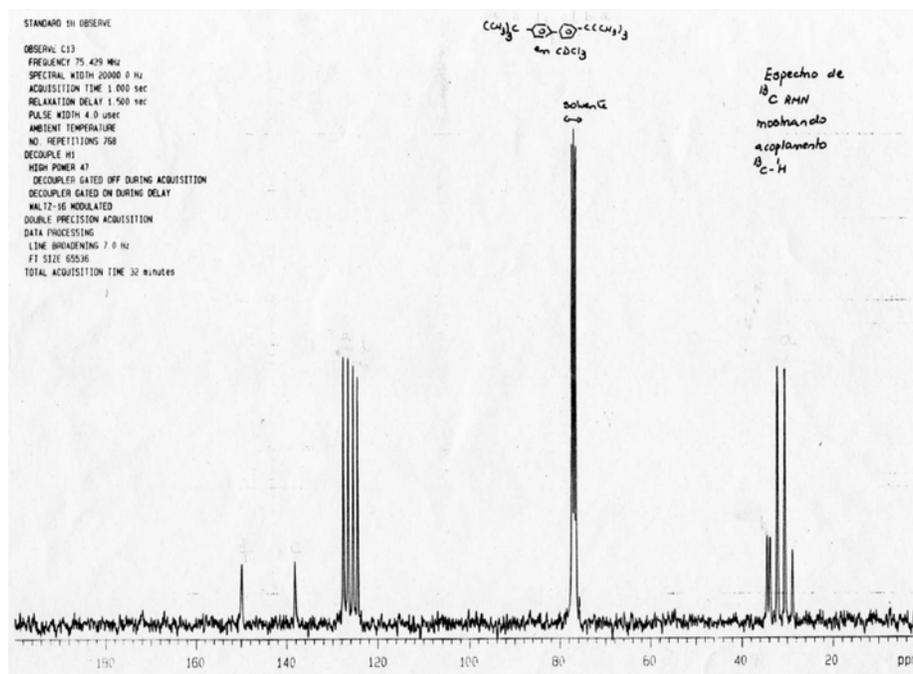
Figure SM 5.2.2.4 – Coupled ^{13}C NMR of 4,4'-di-tert-butylbiphenyl (300 MHz, CDCl$_3$)
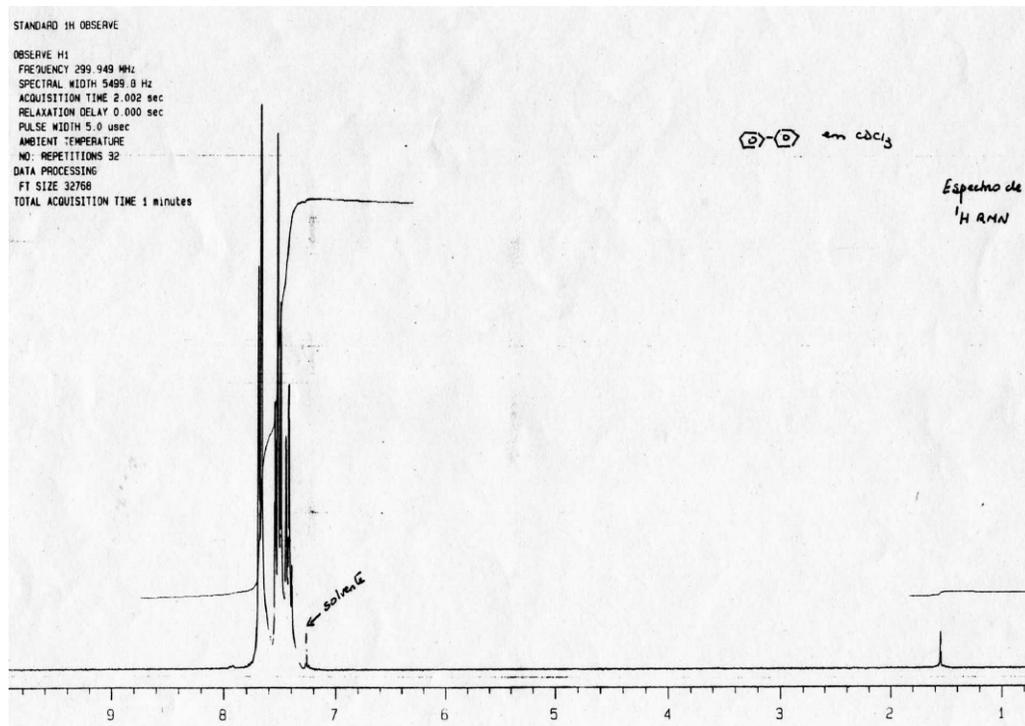
Figure SM 5.2.2.5 – ^{1}H NMR of biphenyl (300 MHz, CDCl$_3$)
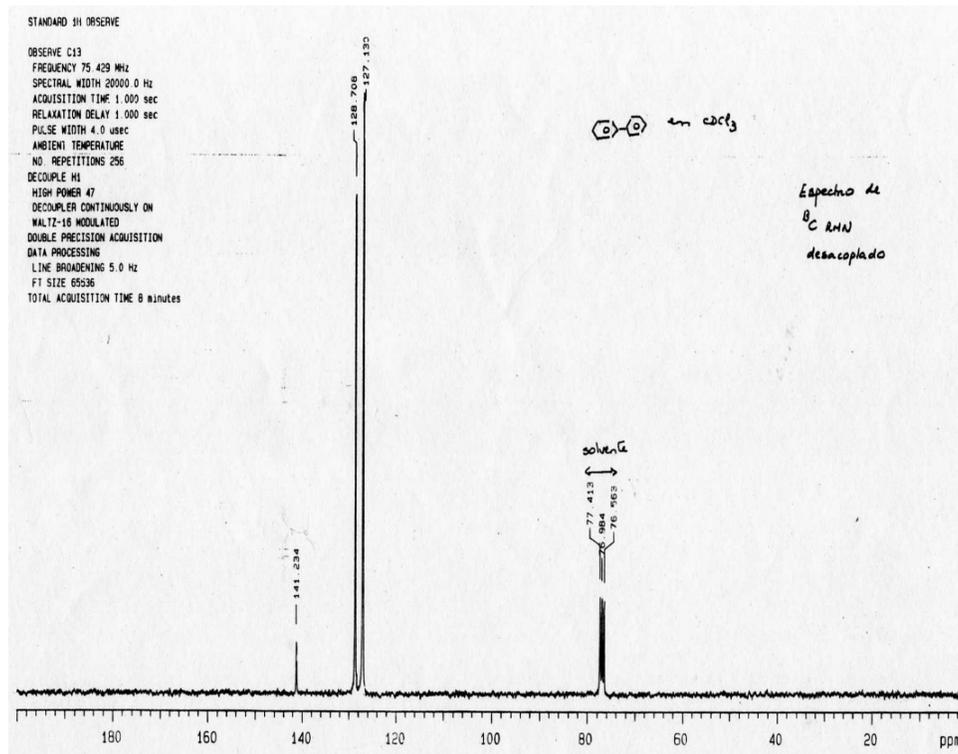
Figure SM 5.2.2.6 – ^{13}C NMR CDCl$_3$ of biphenyl

Synthesis of a macrocycle: *C*-Methyl[4]resorcinarene

Supplementary Material

Background

The synthesis of a macrocycle was developed, initially from Högberg's 1980 methods[1,2] and subsequently as part of a practical guide,[3] to support final year undergraduate lectures in supramolecular chemistry. Few macrocycles lend themselves to synthesis, purification and analysis within a two to three hour timeframe, however, *C*-methyl[4]resorcinarene can be synthesized in an hour and recrystallized with ease. For reasons outlined below there are few by-products and, most importantly, the macrocycle is isolated in only one of the four possible conformers.

Notes for the instructor

The experiment is one of three undertaken by final year undergraduates as part of a 12 week course covering areas of contemporary chemistry. It was designed to highlight a non-templated macrocycle synthesis through the reaction of reagents that would ordinarily be expected to form polymeric mixtures. The course is one of several options and so is taken by 10 to 15 students rather than the entire year group. Students usually work in pairs although individuals may conduct their own experiment. As detailed below, the practical comprises part of a larger exercise so student reports contain a significant proportion of individual work in addition to any collaborative synthetic work.

Students conducting this experiment will have had two years' of university level organic chemistry, both theory and practice, and will also be undertaking a final year laboratory-based research project. Consequently they should have a high degree of practical skill and be familiar with the techniques used. From material covered in lectures they should have the ability to interpret spectra and derive a plausible reaction mechanism.

The trick needed to successfully perform the experiment is to use 'fresh' reagents. We noticed that the yield of the product dropped significantly over three years and discovered that the key issue was the purity of the resorcinol. Older samples of resorcinol give less precipitate during the initial reaction. When recrystallization of the solid formed from the reaction of these older samples was attempted its greater solubility resulted in far less pure macrocycle than expected indicating that much of the precipitate had been a by-product. Once the reaction was repeated with a new bottle of resorcinol the yield was restored to its original value. Although we have not purified older samples of resorcinol they could be recrystallized from toluene and dried under vacuum to be suitable for later use. As noted in the experimental text, the acetaldehyde should be distilled prior to use to avoid traces of acetic acid. To save time this is done by technicians 24 hours in advance of the practical and stored below 5°C until required. The reaction is carried out in an open vessel and we have had no problems with spillage or splashing, however, if desired a water or air condenser could be fitted. It is important to carry out the reaction at 75°C as at lower temperatures the macrocyclic yield is poor and, at higher temperatures, polymers predominate. Occasionally students add the acetaldehyde too quickly which results in the immediate formation of an intractable cream solid, sometimes showing pink regions. This is mainly a polymer and, should it form, we recommend that the students simply repeat the experiment as very little laboratory time will have been wasted.

The experiment has been designed to work comfortably within a three hour laboratory session. The initial reaction takes up to one hour; the duration of the rest of the experiment depends upon the skill of the students to isolate and recrystallize the product. Time could be saved if all the equipment necessary for the reaction was already set up. It would also be possible to stop once the crude product had been isolated and recrystallize at another time, however, aerial oxidation may reduce the final yield. While it is considerably more expensive than deuterochloroform we have only been successful in obtaining good NMR spectra using acetone-d_6 due to the poor solubility of *C*-methyl[4]resorcinarene. Others may wish to experiment using other alternatives such as DMSO-d_6.

The reaction
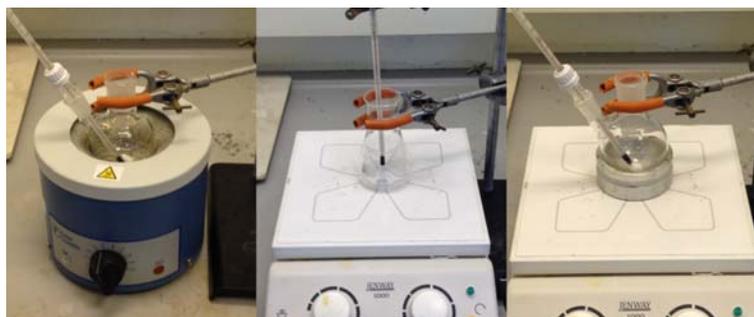
Figure SM 5.2.3.1. Three alternative setups depending on equipment availability: (left to right) heating

mantle, hotplate and hotplate with a heating block.

Figure SM 5.2.3.2. The reaction and product

Figure SM 5.2.3.3. Filtration and crude product

Figure SM 5.2.3.4. Recrystallization and product

Spectra
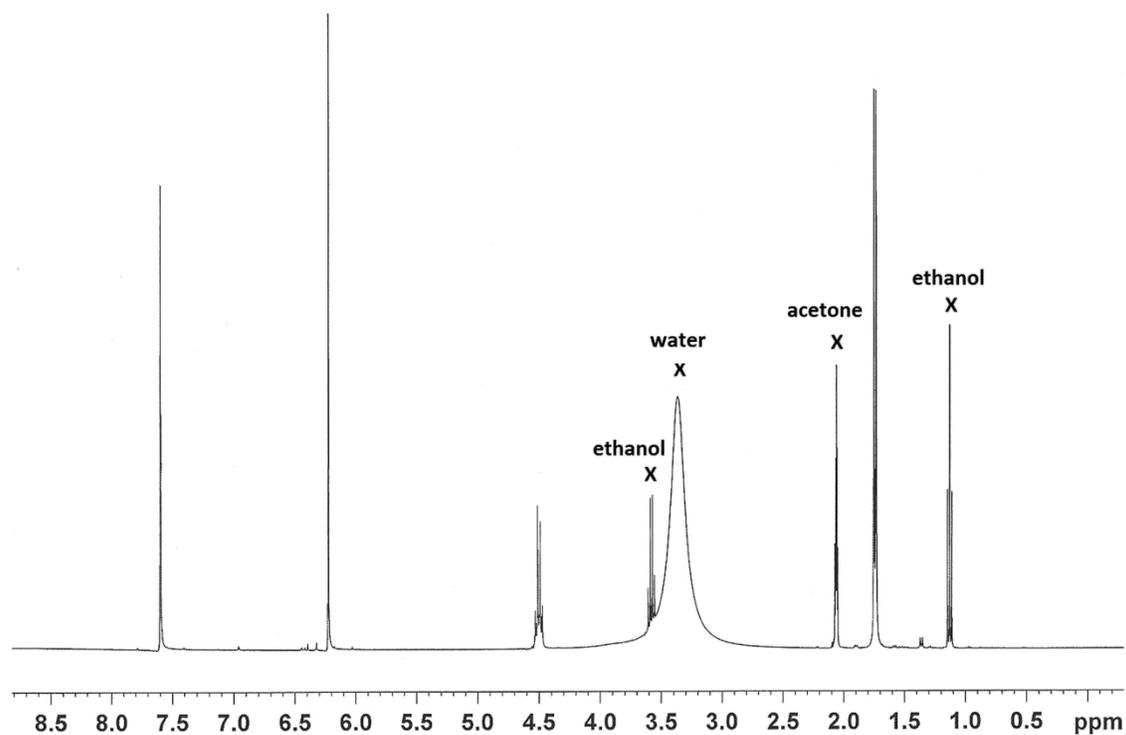
Figure SM 5.2.3.5. ¹H NMR spectrum of the recrystallized product containing excess solvent (student's sample)
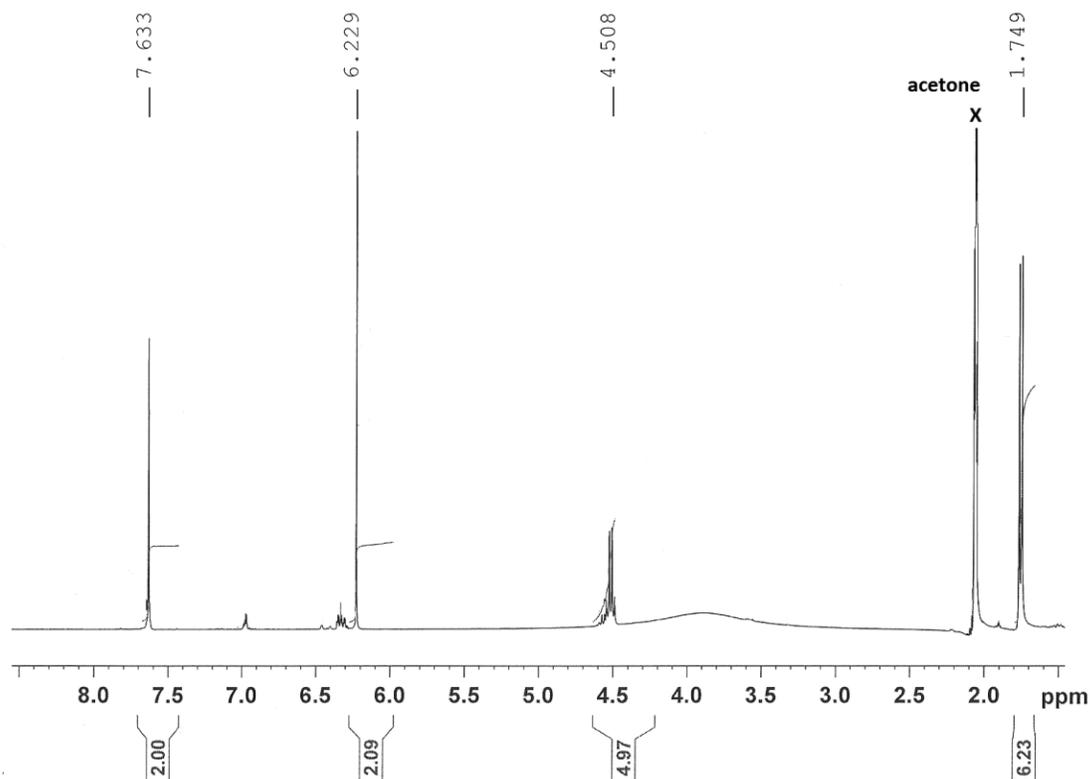
Figure SM 5.2.3.6. ¹H NMR spectrum of the recrystallized product (technician's sample)
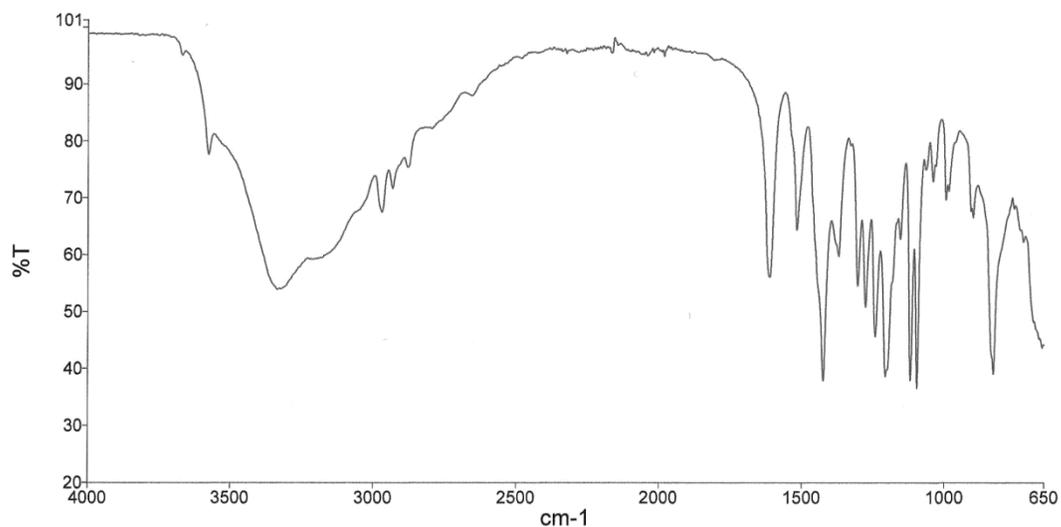
Figure SM 5.2.3.7. IR spectrum of recrystallized product

Onset	338.90 °C
Peak	349.94 °C
Endset	355.15 °C
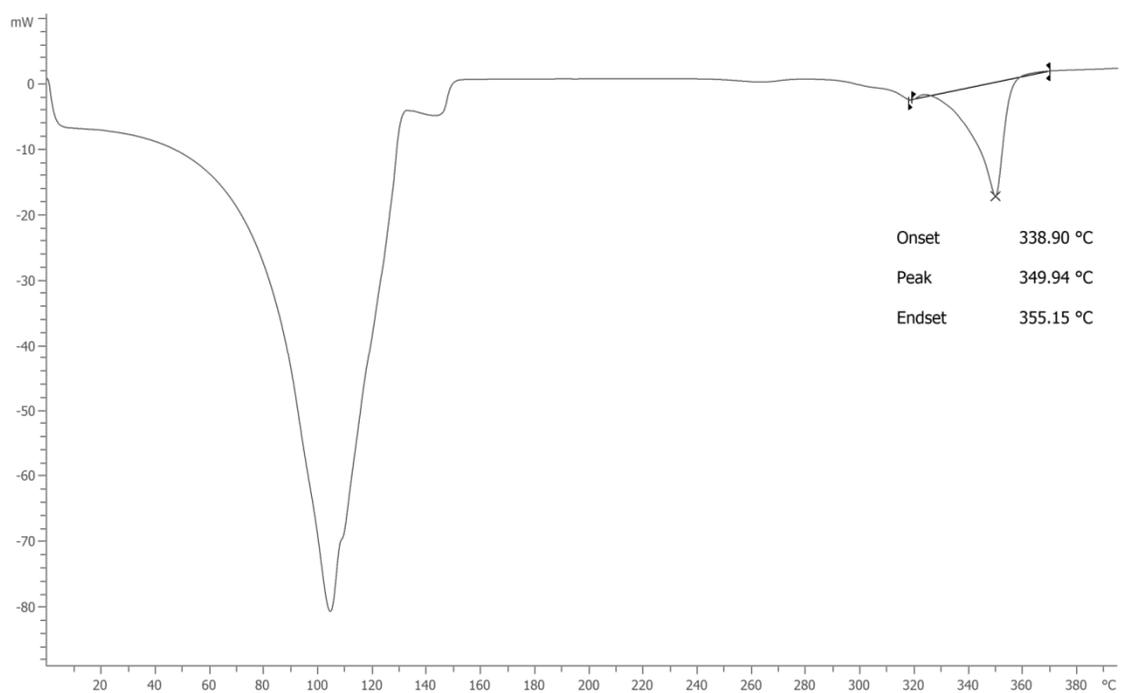
Figure SM 5.2.3.8. DSC trace for the recrystallized product. Most papers report melting points above 250°C or decomposition above 300°C – which the students will also find – but DSC shows a distinct melting point at 349°C

Answers to questions

1. Calculate the percentage yield of your recrystallized product.

The reactants have masses of 110.0 gmol^{-1} (resorcinol) and 44.0 gmol^{-1} (acetaldehyde); the product has a mass of 544.2 gmol^{-1}. Students often forget that the ratio of products to reactants is 1:4 and consequently underestimate their yields by a factor of four.

A typical calculation would be:

Resorcinol used =   5.5 g (50 mmol)   Acetaldehyde used =   2.2 g (50 mmol)

Ratio of resorcinol : acetaldehyde = 1:1   Ratio of resorcinol : *C*-methyl[n]resorcinarene = 4 : 1

Relative molecular mass of *C*-methyl[n]resorcinarene = 544.2 gmol^{-1}

Theoretical maximum yield of *C*-methyl[n]resorcinarene = 544.2 gmol^{-1} x 0.05 mol x 0.25 = 6.8 g

Experimental % yield = [actual yield (g)/6.8 g] x 100

Yields are typically in the region of 4-5 g (55-75%)

2. Many larger *C*-methyl[n]resorcinarene macrocycles could be formed in the reaction. Why do you think that *C*-methyl[4]resorcinarene is formed preferentially?

The cyclotetramer is the smallest macrocycle where the strain energy can be overcome by the benefits of cyclisation. Analysis of available conformers for a linear tetramer will show that a number of them will result in a close approach between termini thus predisposing the molecule to cyclisation. Larger linear species are progressively more flexible as the number of monomeric repeat units increases so the likelihood of both termini of the same molecule meeting becomes lower. In the absence of a template molecule or ion, as is sometime employed in macrocycle formation, further linear polymerization will be more likely to occur than cyclisation. A semi-empirical computational analysis indicates that when the linear tetramer folds up a hydrogen bond forms between one phenolic

hydrogen at one terminus and a methylol oxygen at the other terminus. The latter reacts with a proton

to give a water molecule and leaves a reactive carbocation which completes the macrocyclisation.
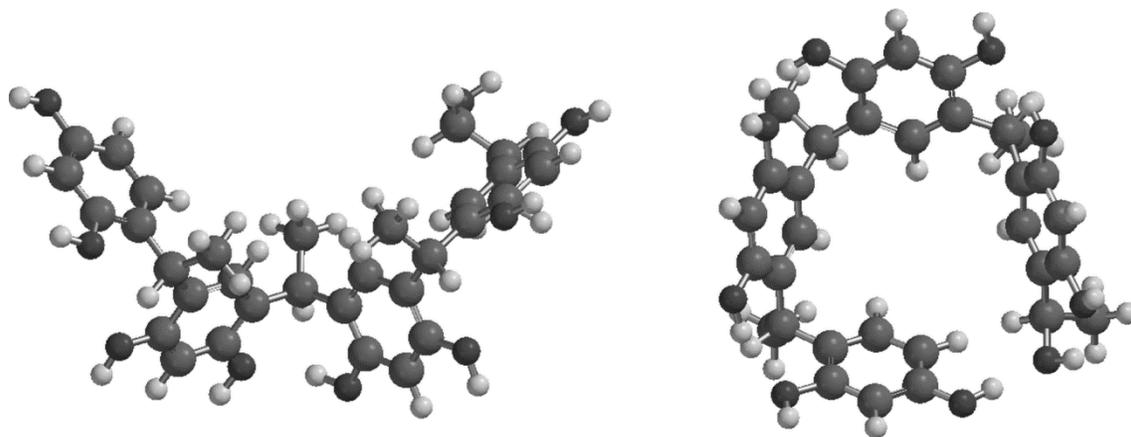
Figure SM 5.2.3.9. Linear and non-linear tetramers

3. *C*-Methyl[4]resorcinarene can exist in four conformers with C_{4v}, C_{2v}, C_{2h} or D_{2d} symmetry. Your

product is the C_{4v} form. Why is this conformer isolated?

The C_{4v} form is the least soluble of all the conformers and precipitates from the reaction mixture. The

other conformers remain in solution but, as the formation of the macrocycle is reversible in acidic

media, bonds in those conformers can be cleaved. Macrocycles that reform in the C_{4v} conformer will

then precipitate. This "thermodynamic sink"[2] drives the formation of the product in a single

conformation.

4. Interpret the ^{1}H NMR spectrum of your product.

The inner aromatic protons will appear as a singlet at about 7.6 ppm downfield of the other aromatic

protons which will also appear as a singlet but at about 6.2 ppm. The protons on the bridging carbons

will appear as a multiplet at about 4.5 ppm and the methyl groups as a singlet at about 1.7 ppm. The

phenolic protons may or may not be easily discernable depending on the presence of trace impurities

such as ethanol from the recrystallization.

5. Devise a mechanism for the formation of *C*-methyl[4]resorcinarene.
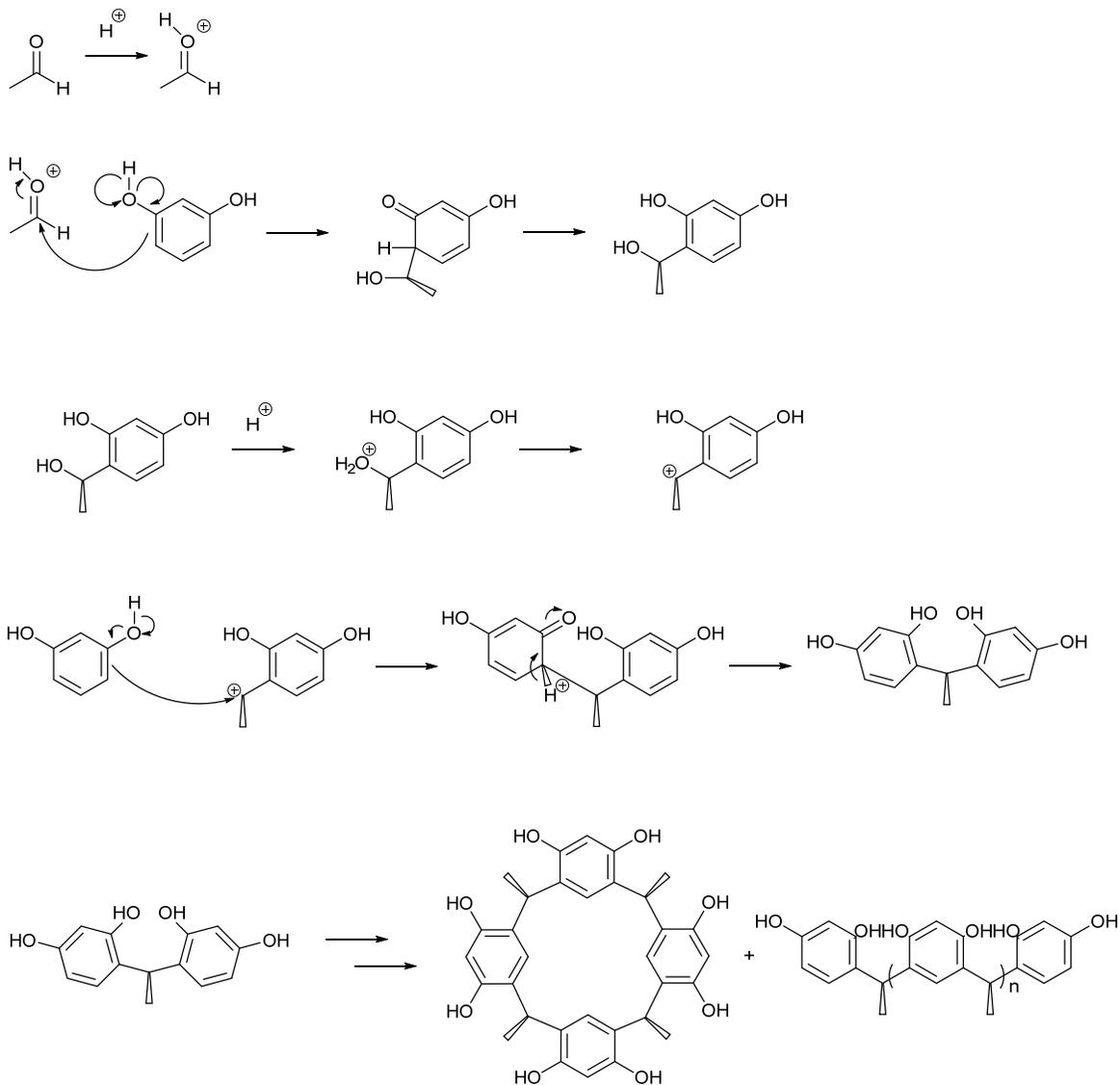
Further work

The practical is part of a broader investigation of macrocyclic chemistry which includes a computational exercise. Students use a program such as *ChemDraw* to generate *C*-methyl[4]resorcinarene and then optimize its geometry using a mechanics program available in *Chem3D* or *Spartan* software. From this structure they can measure the size of the macrocyclic cavity and estimate which small molecules or ions could become 'guests' for the macrocyclic 'host'. The students can also use semi-empirical methods, through *Spartan* or the *GAMESS* interface in *Chem3D*, to determine the molecule's heat of formation. The adventurous may wish to construct all four conformers and determine their relative steric energies to see if this correlates with the C_{4v} form being the most stable. They may also wish to undertake a conformational analysis of the linear tetramer precursor to show how the termini are quite likely to found within bonding distances of each other.

References

1. A. G. S. Högberg, *J. Am. Chem. Soc.*, 1980, **102**, 6046-6050.

2. A. G. S. Högberg, *J. Org. Chem.*, 1980, **45**, 4498-450.

3. *A Practical Guide to Supramolecular Chemistry*, P. J. Cragg, John Wiley & Sons, Ltd., Chichester, September 2005; ISBN: 978-0-470-86653-5/978-0-470-86654-2.

Synthesis of 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin
Supplementary Material

Experimental notes

Figures

Notes:

In this work, which is planned for two sessions of 3 hours, students (in groups of two) will synthesise a free-base porphyrin [5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin] (Figure SM 5.2.4.1 and SM 5.2.4.2). The 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin will be synthesised by a methodology based on the condensation of pyrrole with 2,6-dichlorobenzaldehyde, in a mixture of glacial acetic acid and nitrobenzene at 120 °C (Figure SM 5.2.4.3).

In the first session of three hours, the students will synthesise the crude 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin.

The reaction can be followed by thin layer chromatography (TLC). At the end of the reaction, a brown spot can be observed in the front of the solvent using CH_2Cl_2 as eluent (Figure SM 5.2.4.4).

The reaction can also be monitored by UV-Vis spectrophotometry. Figure SM 5.2.4.5 shows the typical UV-Vis spectrum of the 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin at the end of the reaction, where a strong Soret band can be observed at 417 nm, along with three less intense Q bands. The absorption spectrum of free-base porphyrins shows a typical absorption band of high intensity at around 420 nm, which is called the Soret band, and four bands (I, II, III, IV) between 500-650 nm, called Q bands.

The crude porphyrin is obtained directly from the reaction medium by the addition of methanol.

In the second session of three hours, the students will purify the 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin by recrystallization.

The crude porphyrin is recrystallized in a minimal amount of dichloromethane followed by the addition of methanol.

This synthesis has been performed in our laboratories during the last 20 years by dozens of students and the expected yields for the pure porphyrin should be around 4 - 6%.

The structure of the porphyrin can be confirmed by ^{1}H NMR (CDCl$_3$). The ^{1}H NMR spectrum of the 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin (Figure SM 5.2.4.6) presents the typical three signals of the symmetrical 5,10,15,20-tetraaryl substituted porphyrins:

- A singlet at $\delta - 2.54$ ppm, corresponding to the NH protons.
- A singlet at δ 8.67 ppm corresponding to the eight pyrrole protons.
- A multiplet at δ 7.67-7.81 ppm, corresponding to the twelve aromatic protons of the 5,10,15,20-tetraaryl substituents.

This experiment is intended for a fifth semester organic-inorganic-analytical-physical chemistry laboratory of the BSc Chemistry course. Normally, students who enrol in this laboratory have already attended two semesters of organic chemistry, one semester of a practical organic chemistry course, one semester of inorganic chemistry and one semester of a practical inorganic chemistry course.
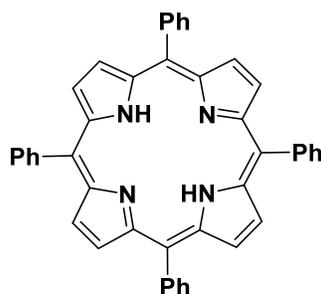
Figure SM 5.2.4.1 – Structure of 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin
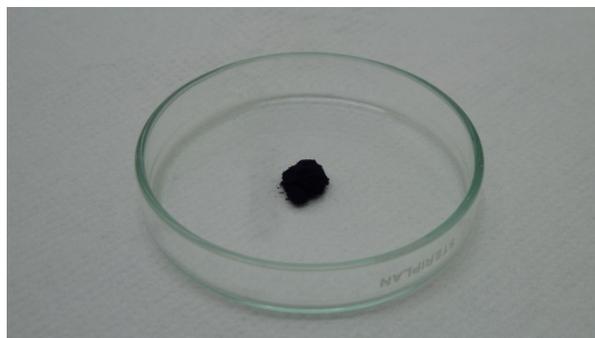
Figure SM 5.2.4.2 - Pure free-base 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin.
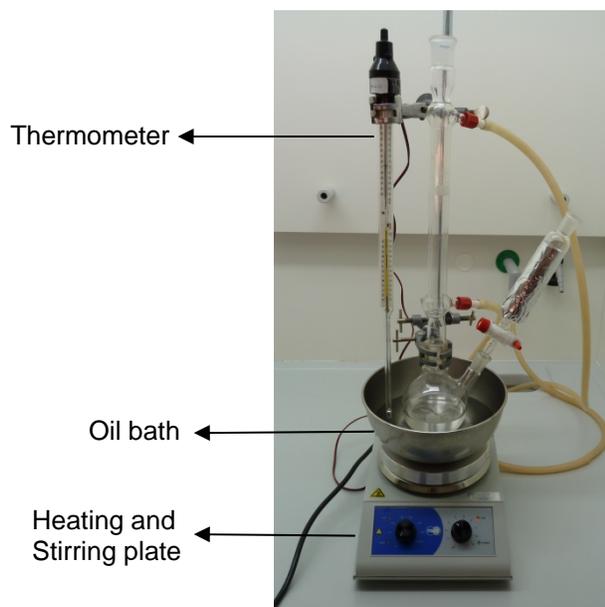
Figure SM 5.2.4.3 - Reaction apparatus.
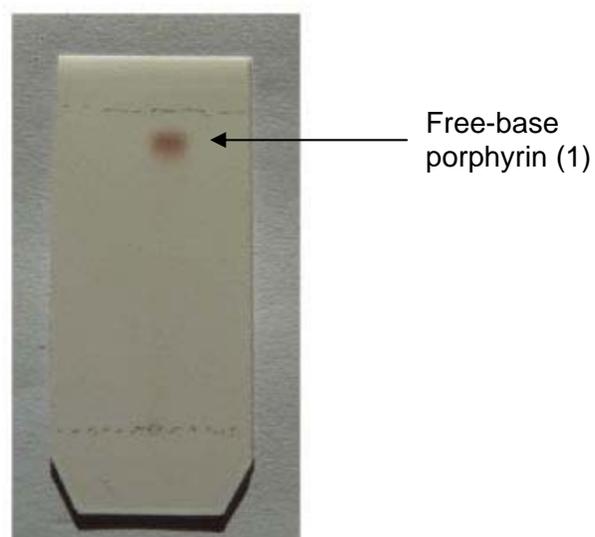
Figure SM 5.2.4.4 - TLC of the pure free-base 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin, using CH$_2$Cl$_2$ as eluent.
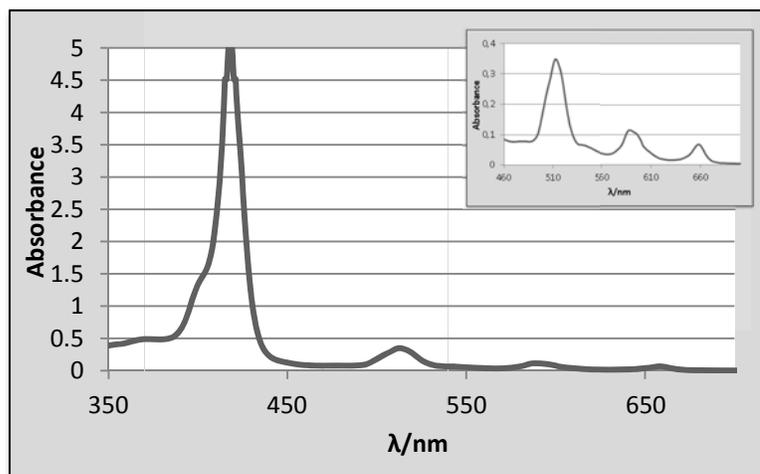
Figure SM 5.2.4.5 - Typical UV-Vis spectrum of the pure free-base 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin in CH_2Cl_2.
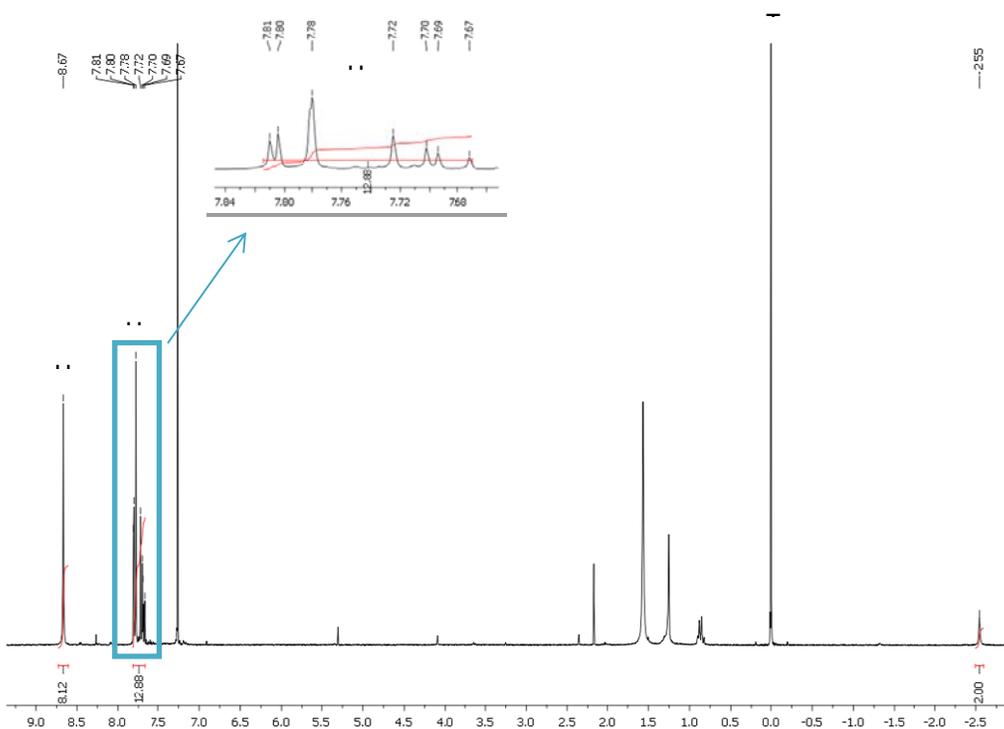
Figure SM 5.2.4.6 - ^{1}H NMR spectrum (300 MHz) of the pure free-base 5,10,15,20-tetrakis(2,6-dichlorophenyl)porphyrin, using $CDCl_3$ as solvent.

4

Solventless Synthesis, Separation and Characterization of Zinc and Free-Base Tetraphenylporphyrin

Experiment Notes

Figures

Introduction

The aim of this experiment is to synthesize both tetraphenylporphyrin (TPP) and zinc tetraphenylporphyrin (ZnTPP) via a solvent-free one-pot reaction. Because no solvent is required for the synthesis benzaldehyde and zinc acetate can be added directly to the reaction vessel. To avoid complete metalation of TPP, a small quantity of zinc acetate is used so that both products are formed. A ratio of approximately 1:16 (m/m) zinc acetate to pyrrole should be used. Benzoic acid is added to minimize the oxidation of benzaldehyde, and can be substituted for a slight molar excess of benzaldehyde.

Scheme SM 5.2.5.1 General synthetic mechanism.
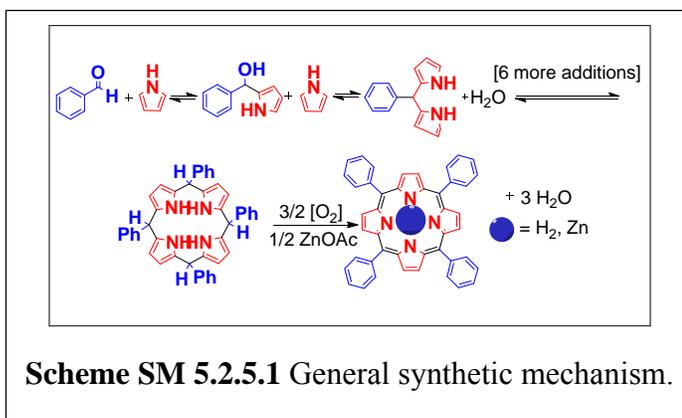
The reaction vessel is first heated to 140 °C in a stirring oil bath or sand bath on a hot plate. The reaction will proceed to completion within 30 minutes at ca. 30-40% yield. The product formed will condense on the sides of the reaction vessel as a dark purple solid. Both ZnTPP and TPP will dissolve easily in a minimum volume of chloroform directly from the reaction vessel. No washing is necessary. The primary reaction products are the porphyrins and largely insoluble polypyrrole compounds which adhere to silica (Figures SM 5.2.5.1- SM 5.2.5.3).

Thin-layer-chromatography (TLC) can be used to confirm the formation of both TPP and ZnTPP. Separation is visible between two purple bands, both emissive under a UV lamp, using hexanes:CH_2Cl_2 (2:1 v/v) as eluent. Products can be separated with a short silica column in the same eluent. TPP has a larger R_f, and will elute first (Figures SM 5.2.5.4 - SM 5.2.5.6). TLC and column separation are critical tools in synthetic organic chemistry, and this experiment provides

students with colorful examples of both techniques. Students will also be introduced to UV-visible spectroscopy as an analytical method for distinguishing between compounds of different symmetry.

Experiment reproducibility was evaluated by repeated trials by 3rd and 4th year Chemistry B.A. students at Hunter College of The City University of New York. The reaction reliably reaches completion in less than 30 minutes, and can be conducted on a small scale (as little as 0.5 mmol pyrrole was tested).

Product Yield

4 molar equivalents of pyrrole are required to synthesize a single equivalent of either TPP or ZnTPP. If the reaction is performed with 0.5 mmol pyrrole, the theoretical yield of total porphyrin should be 0.125 mmol. The reaction yielded 14 mg of TPP (0.023 mmol) and 16 mg of ZnTPP (0.024 mmol), a total of 0.047 mmol porphyrin (38%). The following general formula can be used to calculate the experimental yield:

$$\frac{(Moles_{TPP} + Moles_{ZnTPP})}{0.25\ Moles_{pyrrole}} \times 100\%$$

Characterization

UV-Vis absorption spectra distinguish the free-base and metallo-porphyrins. Traditionally, the intense absorption of the porphyrin at ca. 420 nm is called the Soret band, and the less intense absorptions observed between 450 nm and 700 nm are called the Q bands. The free base porphyrin has D_{2h} symmetry (Figures SM 5.2.5.7, SM 5.2.5.9) because of the protonated pyrroles on the interior of the macrocycle, and displays four distinct Q bands (Scheme SM 5.2.5.2). The ZnTPP molecule is of D_{4h} symmetry, which induces a degeneracy between the previously distinct molecular orbitals causing the four Q bands to become only two (Scheme SM 5.2.5.2, Figures SM 5.2.5.8, SM 5.2.5.11).
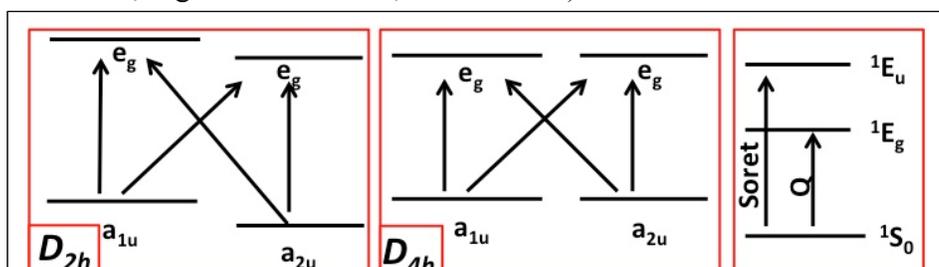
Scheme SM 5.2.5.2 The characteristic UV-visible spectrum is ascribed to the x,y polarized 4-orbital transitions $a_{1u}\ (\pi) \rightarrow e_{g}\ (\pi^*)$ and $a_{2u}\ (\pi) \rightarrow e_{g}\ (\pi^*)$ which accidently assume nearly degenerate energies in D_{4h} symmetry.

These changes are also observed in the ^{1}H NMR chemical shifts. In the free-base porphyrin, a ring current induced by the cyclized pyrroles shifts the peak corresponding to the interior N-H protons up field into the negative region of the spectra. This peak is not observed after metalation.

Additional Safety

   This reaction requires high temperatures in a sealed vessel. It is imperative that students be made aware of the dangers associated with oil baths and hot plates. Septa should be checked by the instructor to ensure a good seal. Make sure that the reaction is allowed to cool to room temperature before handling the glass reaction vessel or unsealing the stopper.

Figures and Spectra
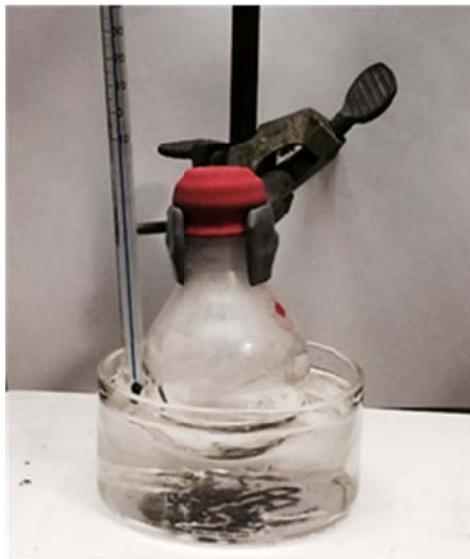
Figure SM 5.2.5.1 Reaction vessel with
rubber septum in oil bath.
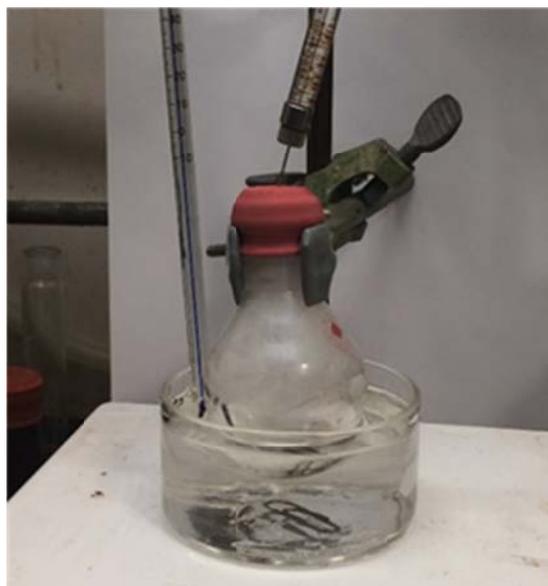
Figure SM 5.2.5.2 Reaction vessel after
addition of pyrrole.

Figure SM 5.2.5.3 Formation of TPP and ZnTPP via condensation

Figure SM 5.2.5.4 Thin-layer chromatography. ZnTPP(left), TPP (center), and crude reaction mixture (right). Eluent hexanes:CH_2Cl_2 (2:1 v/v)
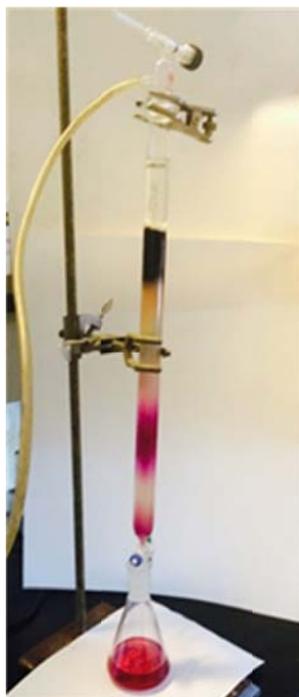
Figure SM 5.2.5.5 Silica column. TPP elutes first. Both bands are visible.

Figure SM 5.2.5.6 Products after column separation.
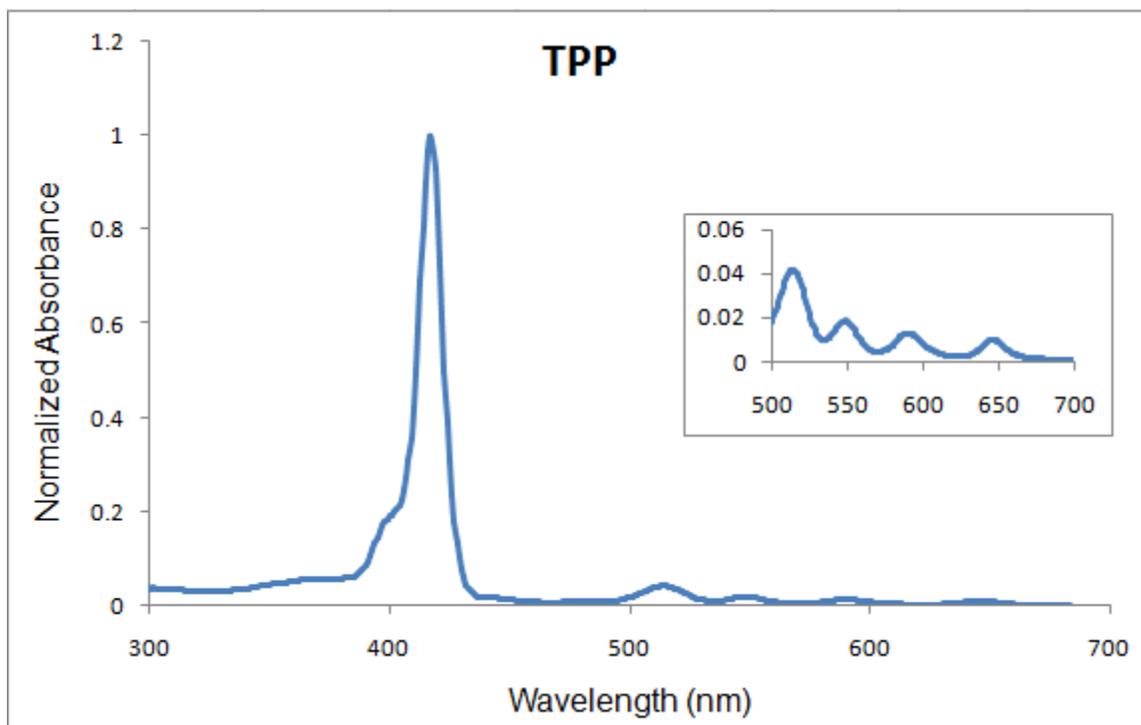
Figure SM 5.2.5.7 UV-Vis absorption spectrum of TPP. Q band region inset.
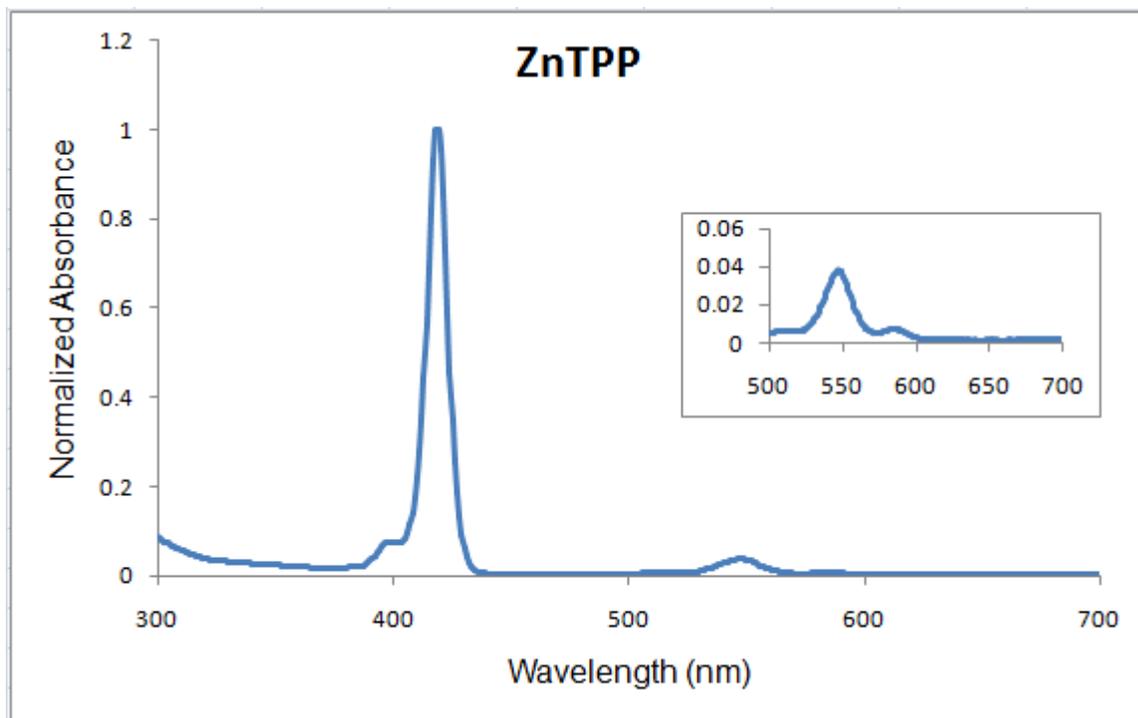
Figure SM 5.2.5.8 UV-Vis absorption spectrum of ZnTPP. Q band region inset.

TPP
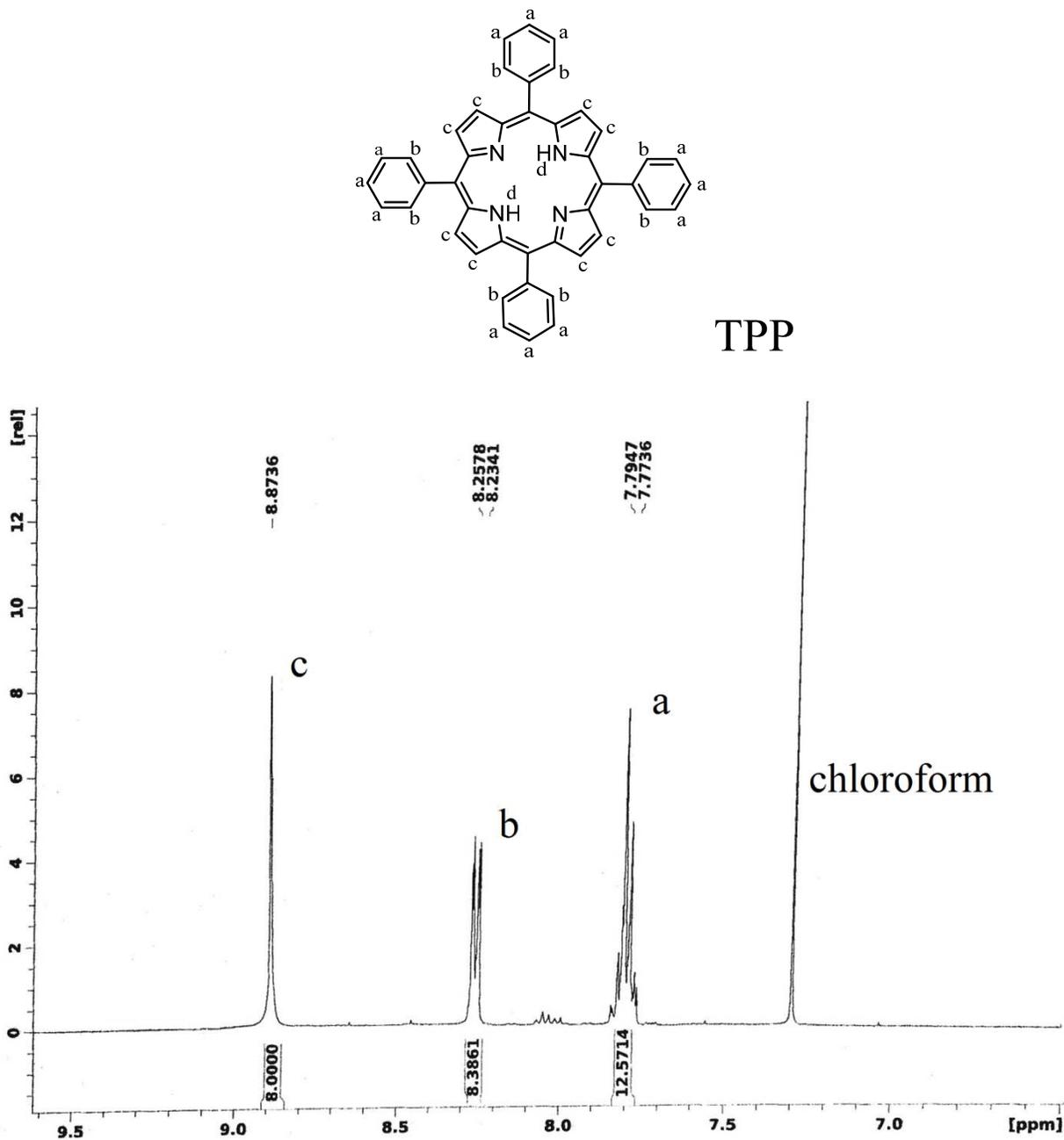
Figure SM 5.2.5.9 Structure of TPP and ^{1}H NMR Spectrum (400 MHz, CDCl$_3$) of aromatic region.
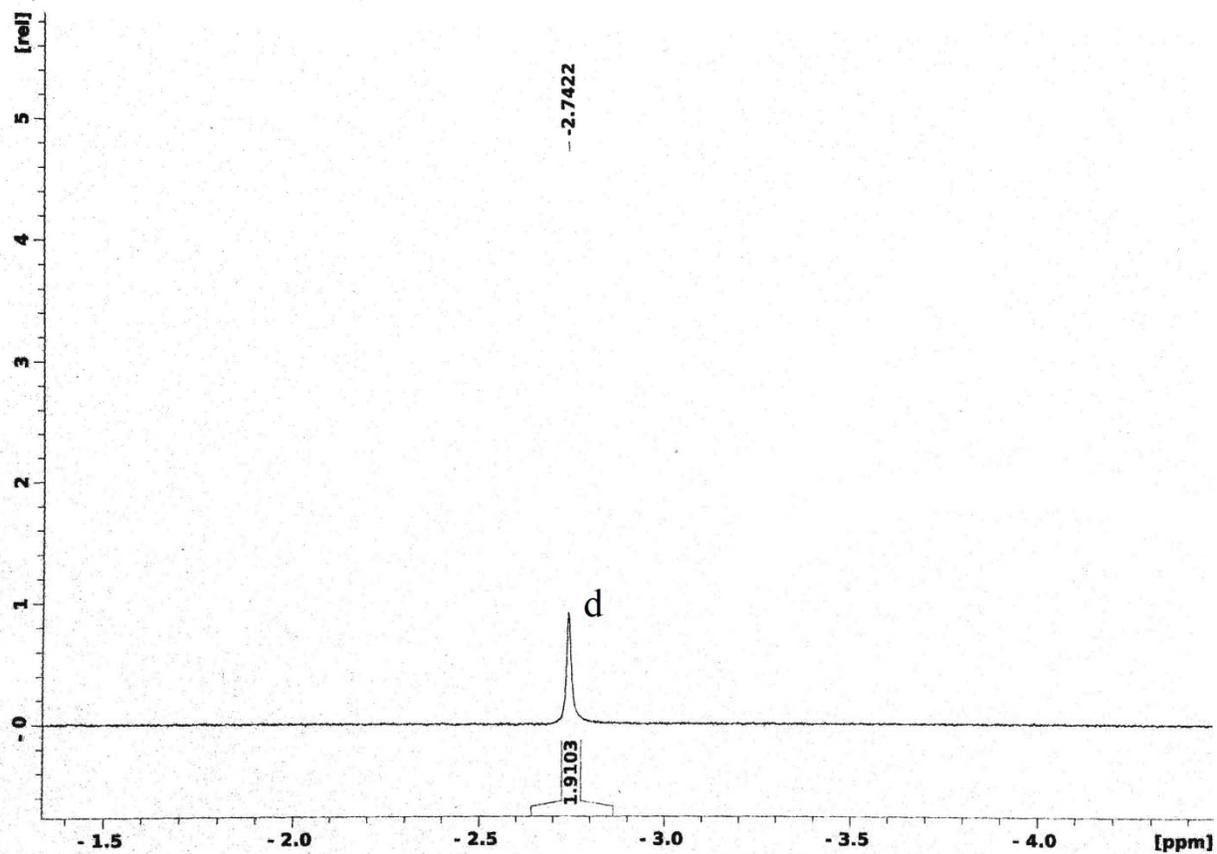
Figure SM 5.2.5.10 ^{1}H NMR Spectrum (400 MHz, CDCl$_3$) of TPP, negative region.
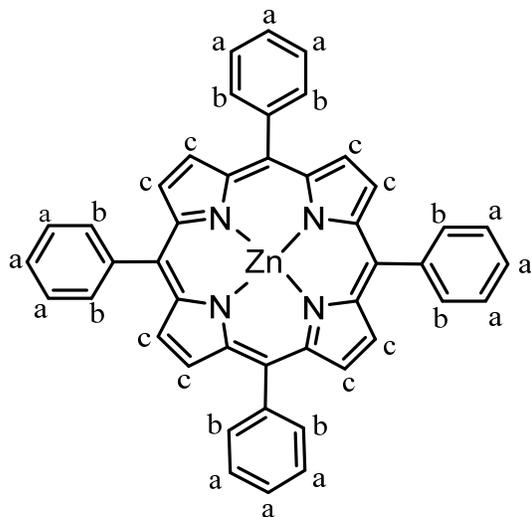
ZnTPP
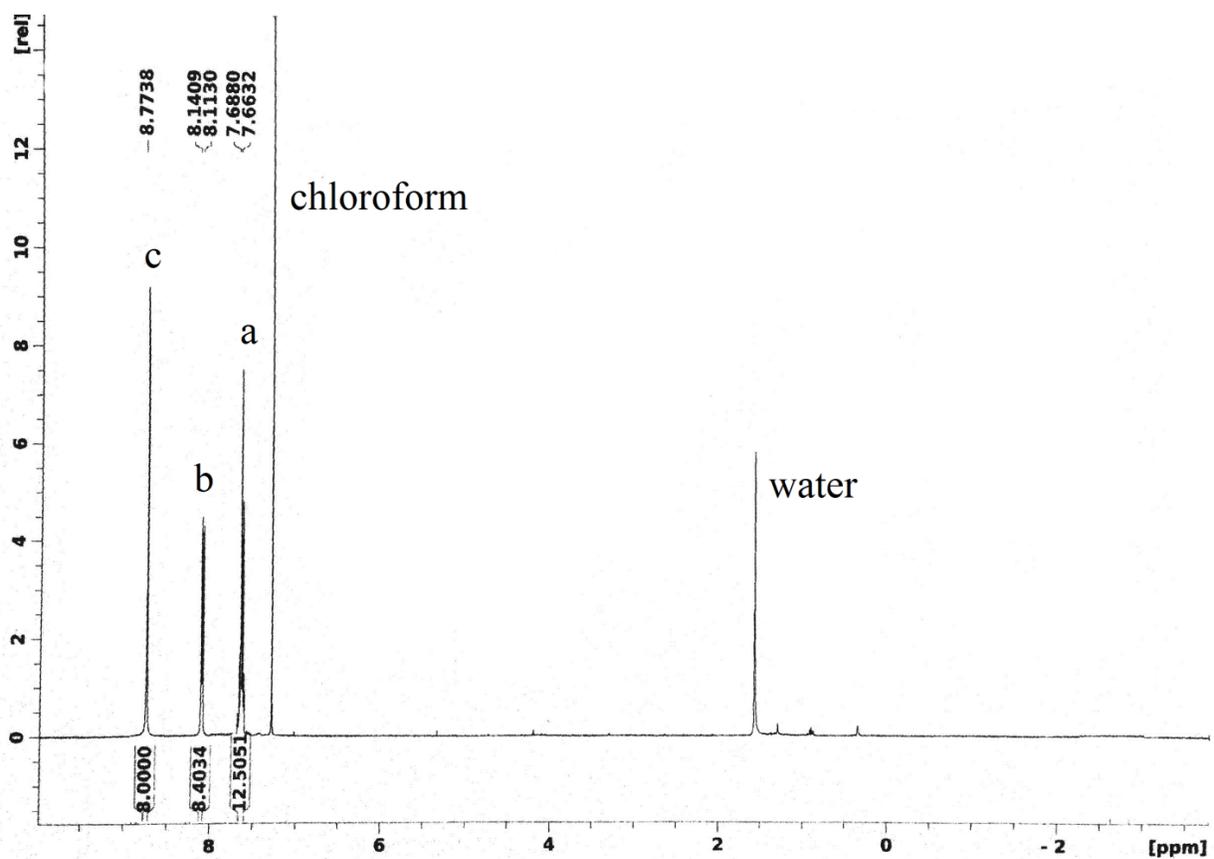
Figure SM 5.2.5.11 Structure of ZnTPP and ^{1}H NMR spectra (400 MHz, CDCl$_3$).
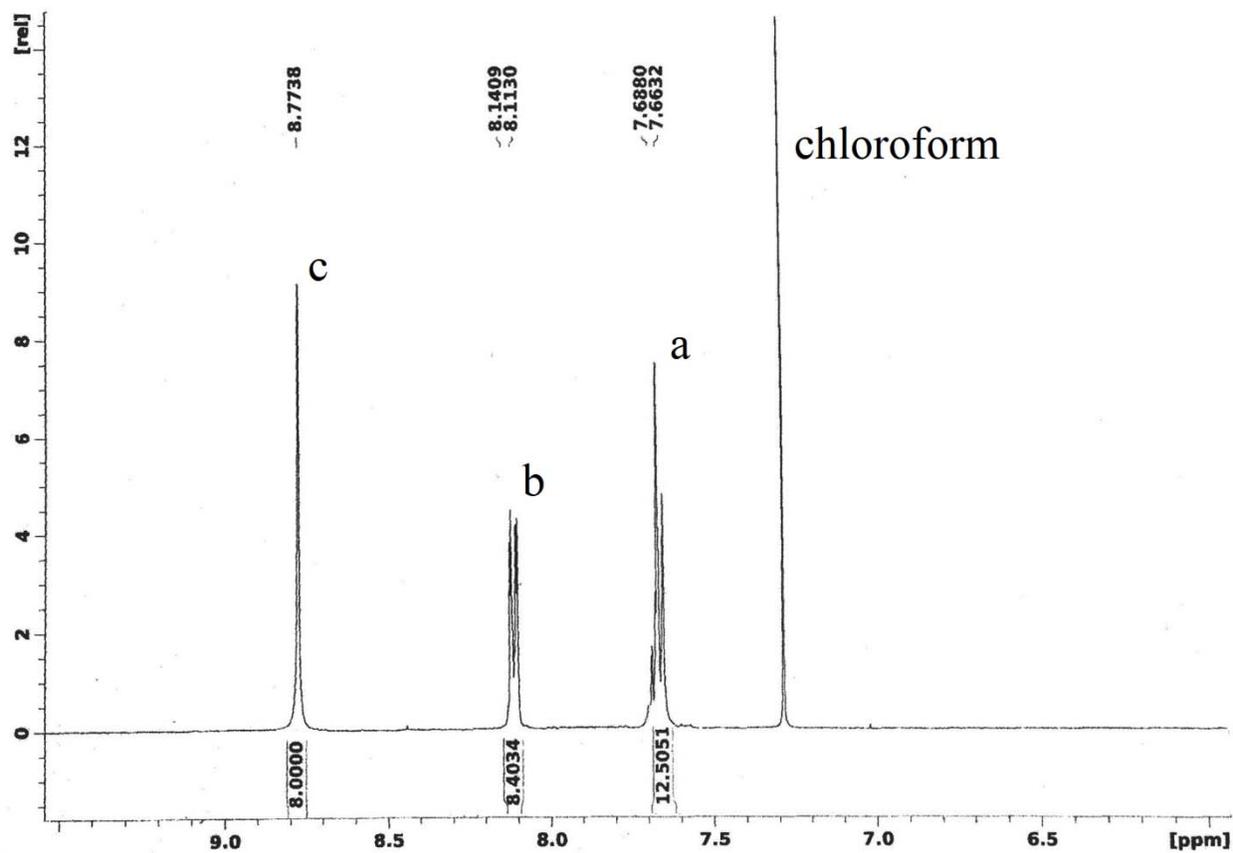
Figure SM 5.2.5.12 ^{1}H NMR spectra (400 MHz, CDCl$_3$) of ZnTPP, aromatic region.

Synthesis of 3-bromo-7-ethylcarbamate-4-methylcoumarin

Supplementary Material

The aim of this three steps synthesis is the preparation of 3-bromo-7-ethylcarbamate-4-methylcoumarin easily achieved from *m*-aminophenol. The reaction sequence starts with the protection of the amino group of *m*-aminophenol with ethoxycarbonyl chloride, followed by Pechmann condensation to the coumarin ring. Finally, bromination in position 3 can be attended in moderate conditions with bromine generated *in situ* from HBr and OXONE®. These reaction sets allow students to apply their knowledge in matters involving the reactivity of different functional groups as the nucleophilic duality of nitrogen *vs* oxygen, the chemistry of the carbonyl group and the electrophilic aromatic substitution.

The reproducibility of the three step reaction sequence was assessed by students from the 3rd year of the bachelor degree in Chemistry (a total of 8 groups of 2 students each) at the Faculty of Science and Technology at the Universidade Nova de Lisboa.. Yields of step 1 range from 80 to 91%, of step 2 from 70 to 83 and of step 3 from 55 to 63%.

The first synthetic step concerning the amine group protection is also feasible with methyl chloroformate and with similar income. As expected NaHCO$_3$ can also be used in the first step instead of KHCO$_3$. For the purpose of the experimental procedure both acylating reagents gave the 7-alkoxycarbonylamino-4-methylcoumarin in good yields.

3-Bromo-7-ethoxycarbonylamino-4-methylcoumarin is a very insoluble product whose extraction may be assisted with a chloroform:*n*-butanol (5%) mixture. Alternatively, after the terminus of the reaction the precipitated product can be filtered from the reaction mixture, washed with water and then ethanol.

Due to the insolubility of the compound the NMR spectra should be run in $CDCl_3$ with 2 to 3 drops of DMSO-d_6.

Results interpretation and additional questions

1. Propose a reaction mechanism for the Pechamnn condensation.
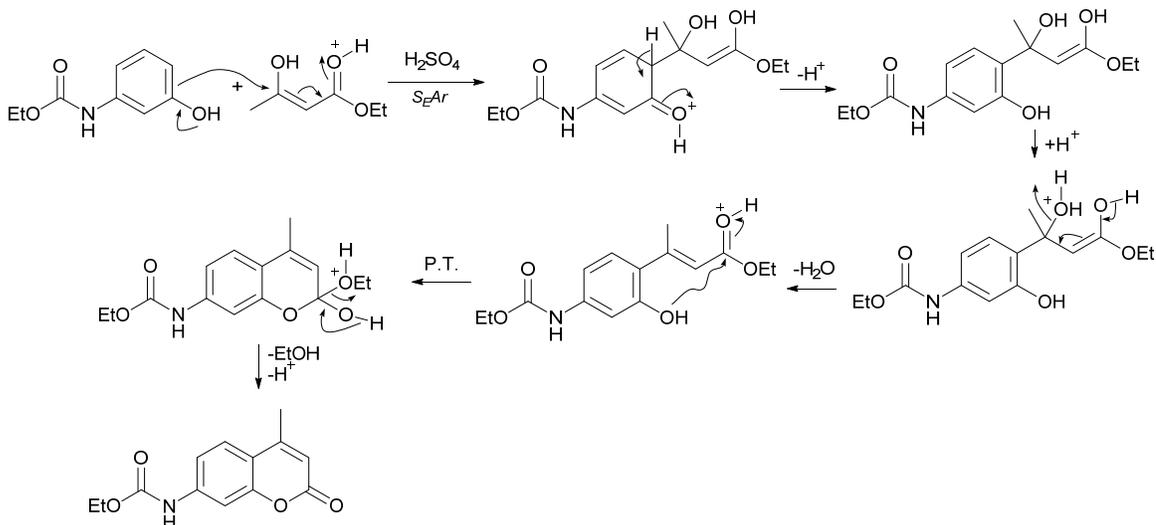
2. Can you provide an explanation for the experimental step of the first session where water was added and let to react for 2 to 3 hours? What is happening?

To perform the hydrolysis of the carbonate functionality formed on reaction of the phenol group with ethyl chloroformeate.

3. What is the role of OXONE®? Why the reaction did turn orange after the addition of OXONE?

Oxone does the oxidation of HBr to Br_2 which is responsible for the developed orange colour.

4. Justify why the bromination occurs on the 3-position and not on the phenyl ring (6 or 8 position).

The slightly activating carbamate group is an *orto*/*para* directing group but due to the steric demanding *orto* positions, the bromination is directed to the also activated but slightly farther away 3-position.

5. What is the principal characteristic on the ^{1}H and ^{13}C NMR spectra of 3-bromo-7-ethoxycarbonylamino-4-methylcoumarin that can justify the bromine insertion?

The absence of the doublet signal at 6.18 ppm that correspond to the hydrogen at the 3-position

and the change of the signal of the carbon at the same position from 112.95 ppm to 109.88 ppm.

6. What is the mechanism for the bromination reaction? How do you classify this reaction?
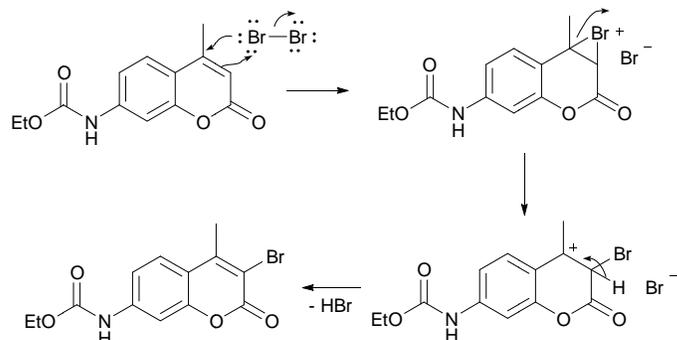
This is an aromatic substitution bromination although there is no prove of the non-brominium presence

IR, ¹H and ¹³C spectra
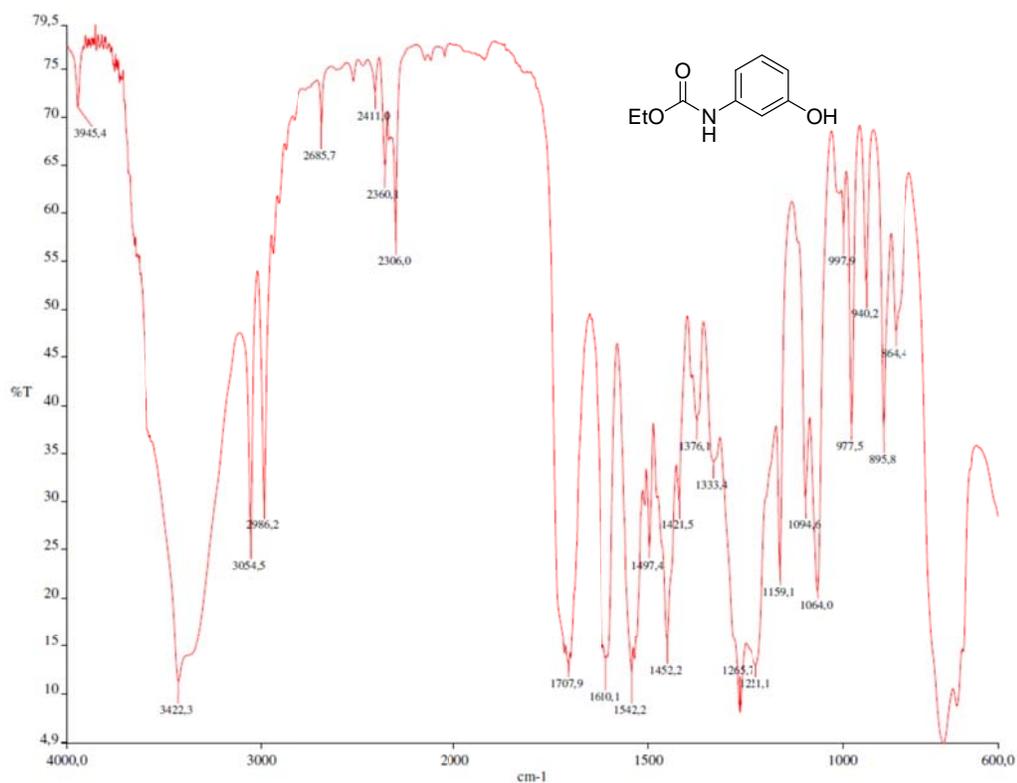
Figure SM 5.2.6.1. IR spectrum (film) of *m*-(*N*-ethoxycarbonylamino)phenol.
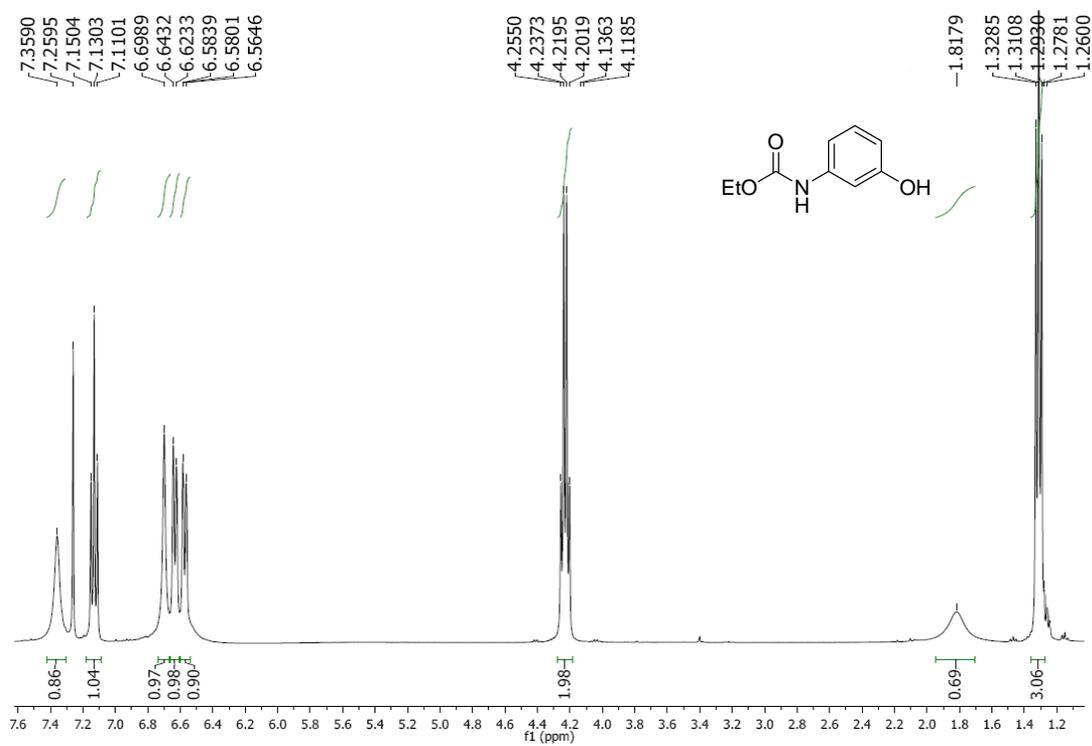
Figure SM 5.2.6.2. ^{1}H NMR spectrum (400 MHz, CDCl$_3$) of m-(*N*-ethoxycarbonylamino)phenol.
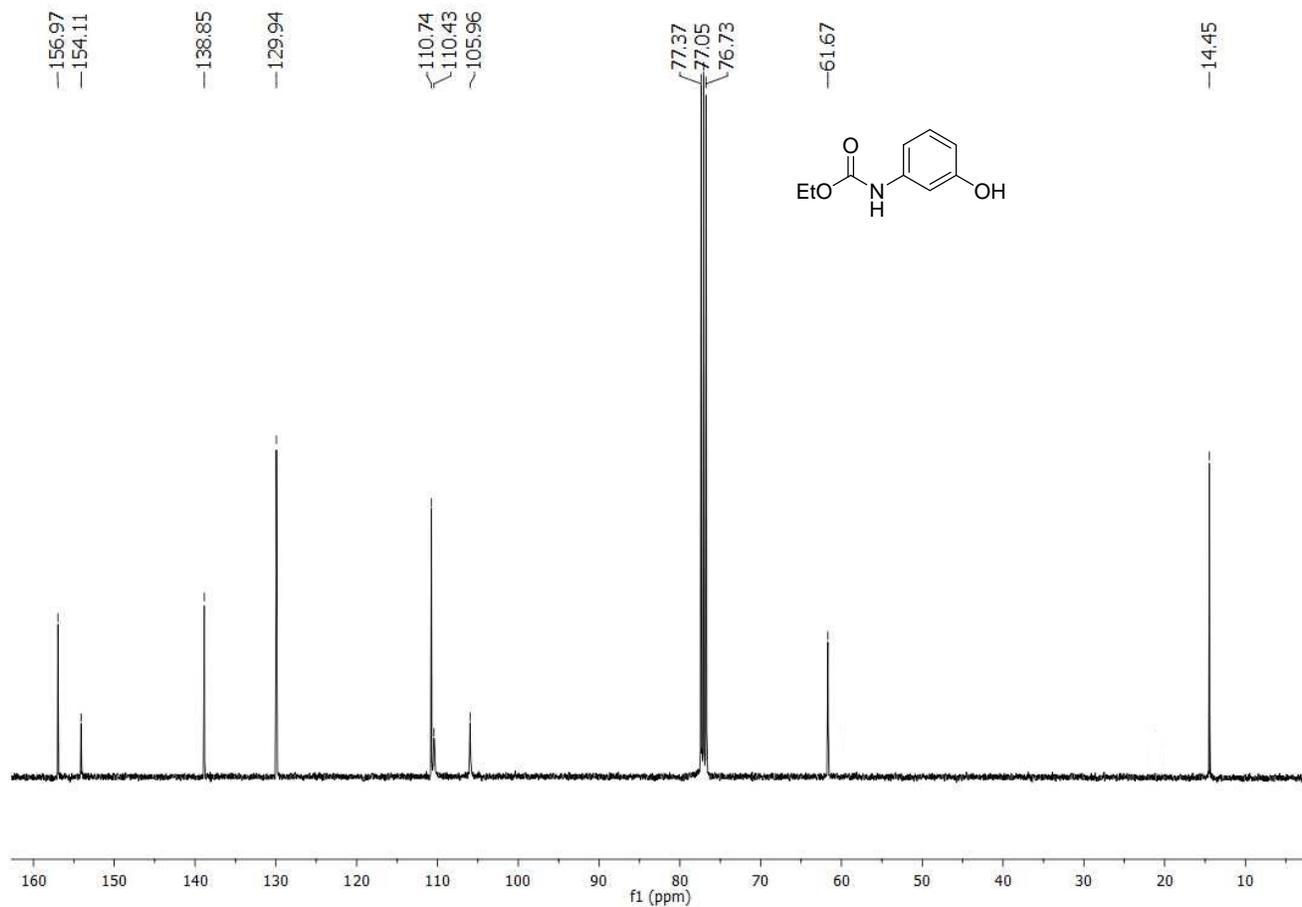
Figure SM 5.2.6.3. ^{13}C NMR spectrum (100.5 MHz, CDCl$_3$) of m-(*N*-ethoxycarbonylamino)phenol.
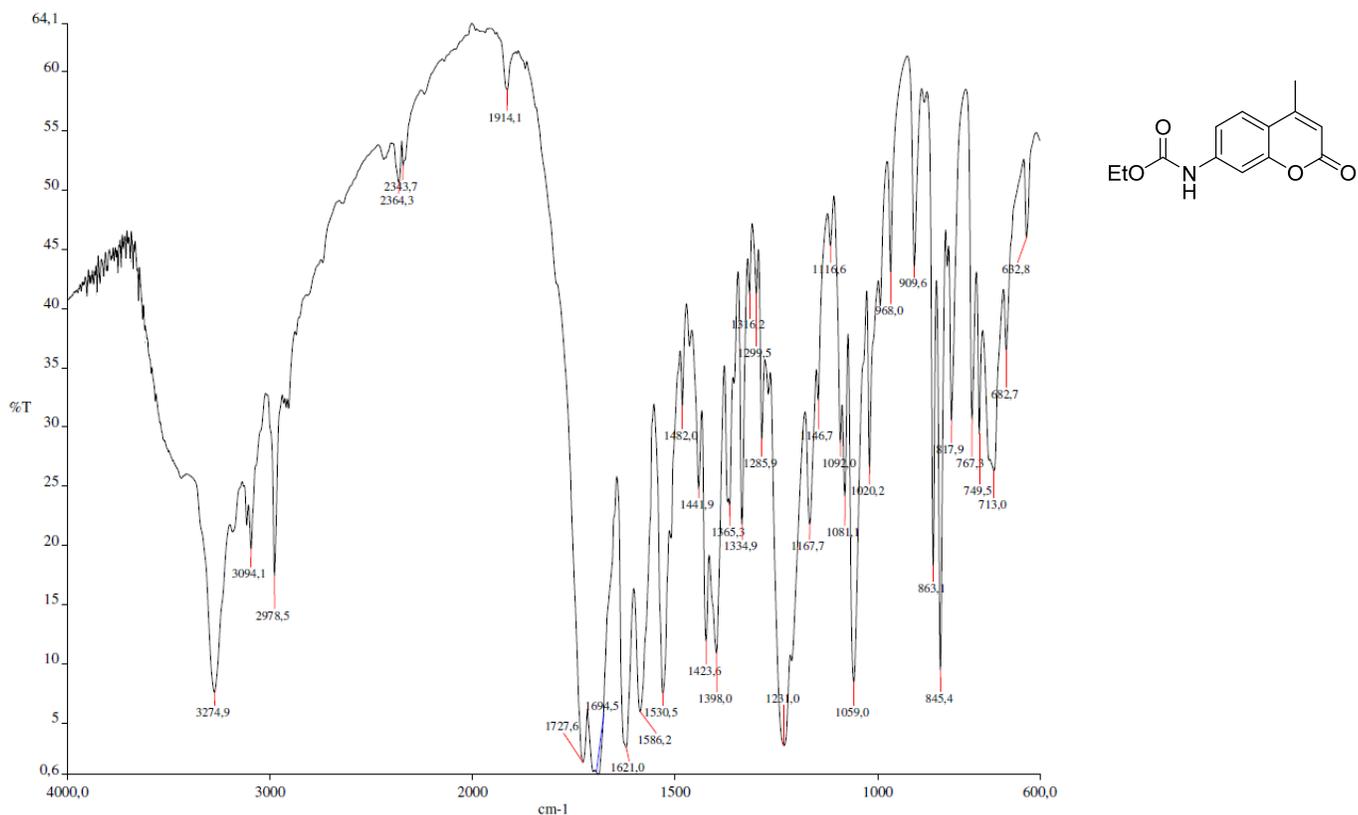
Figure SM 5.2.6.4. IR spectrum (KBr) of 7-Ethoxycarbonylamino-4-methylcoumarin.
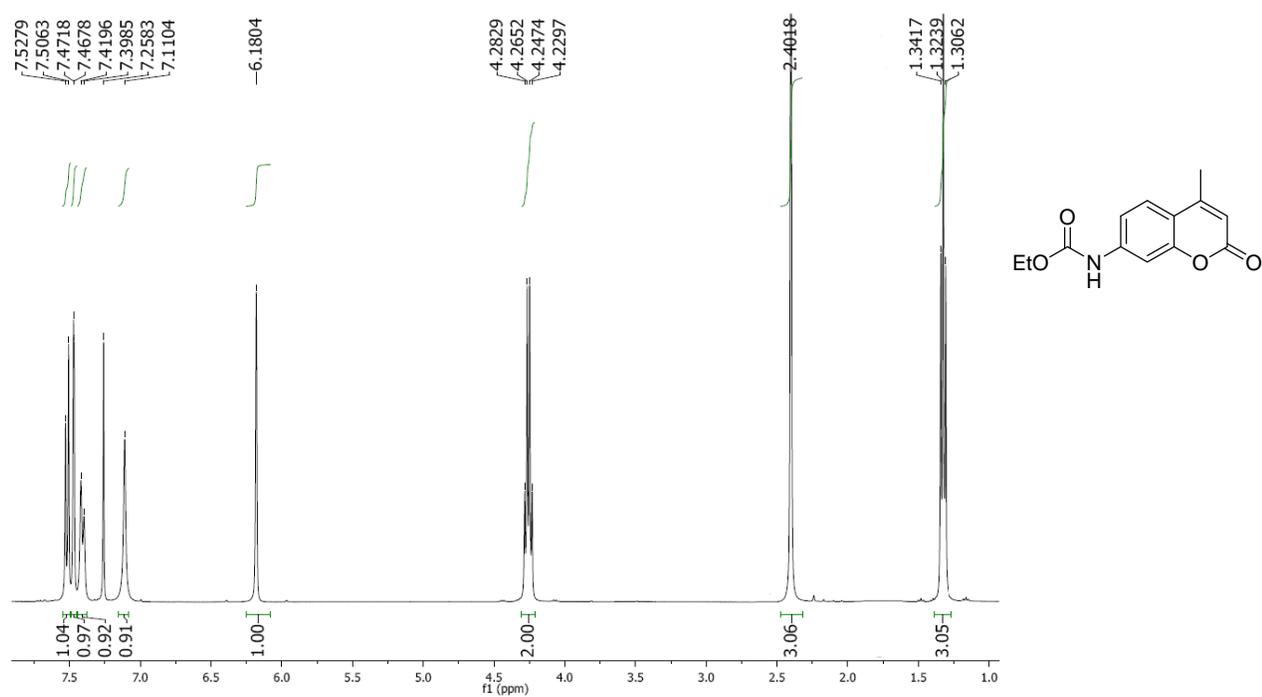
Figure SM 5.2.6.5. ^{1}H NMR spectrum (400 MHz, CDCl$_3$) of 7-ethoxycarbonylamino-4-methylcoumarin.

6
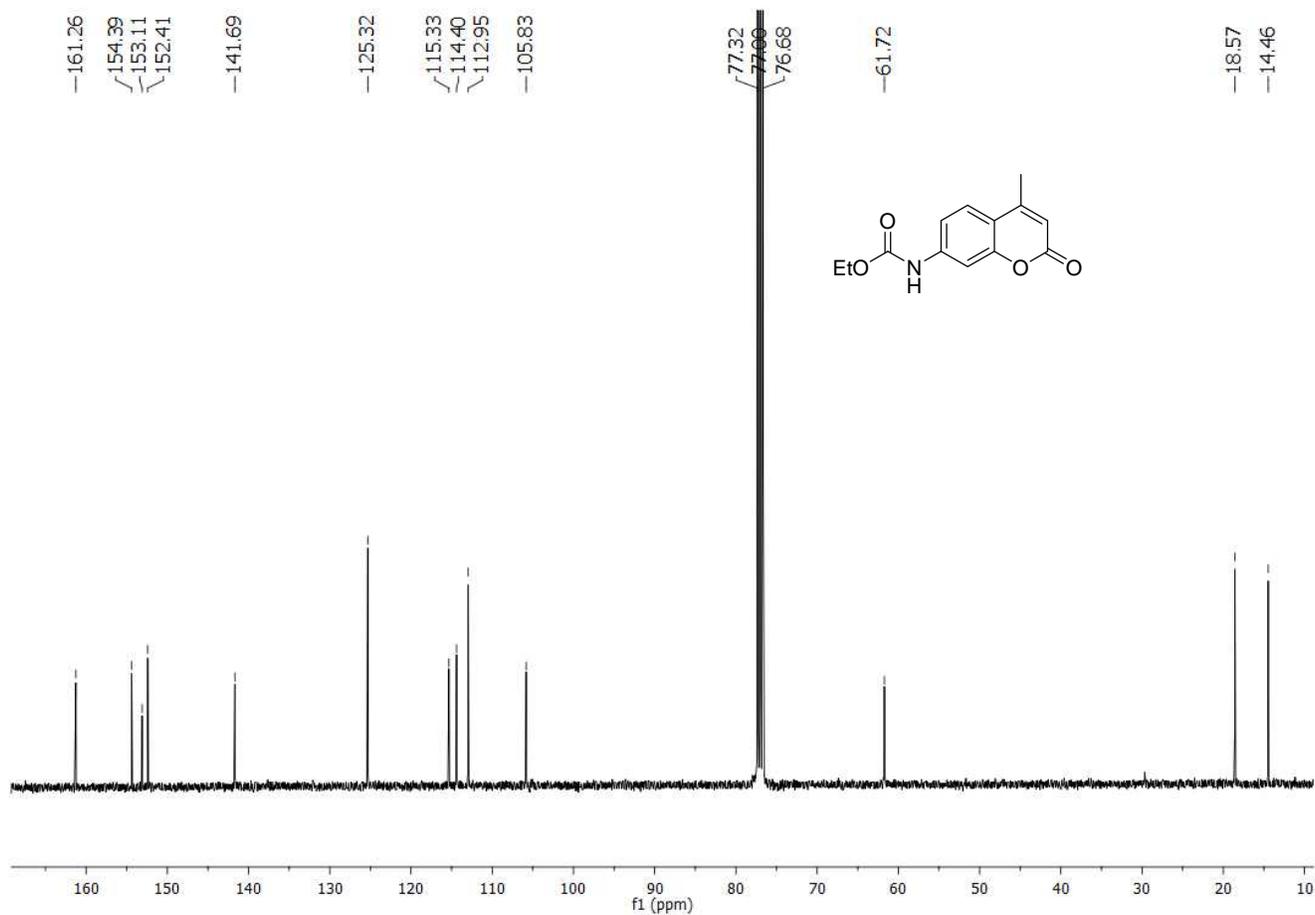
Figure SM 5.2.6.6. ^{13}C NMR spectrum (100.5 MHz, CDCl$_3$) of 7-ethoxycarbonylamino-4-methylcoumarin.
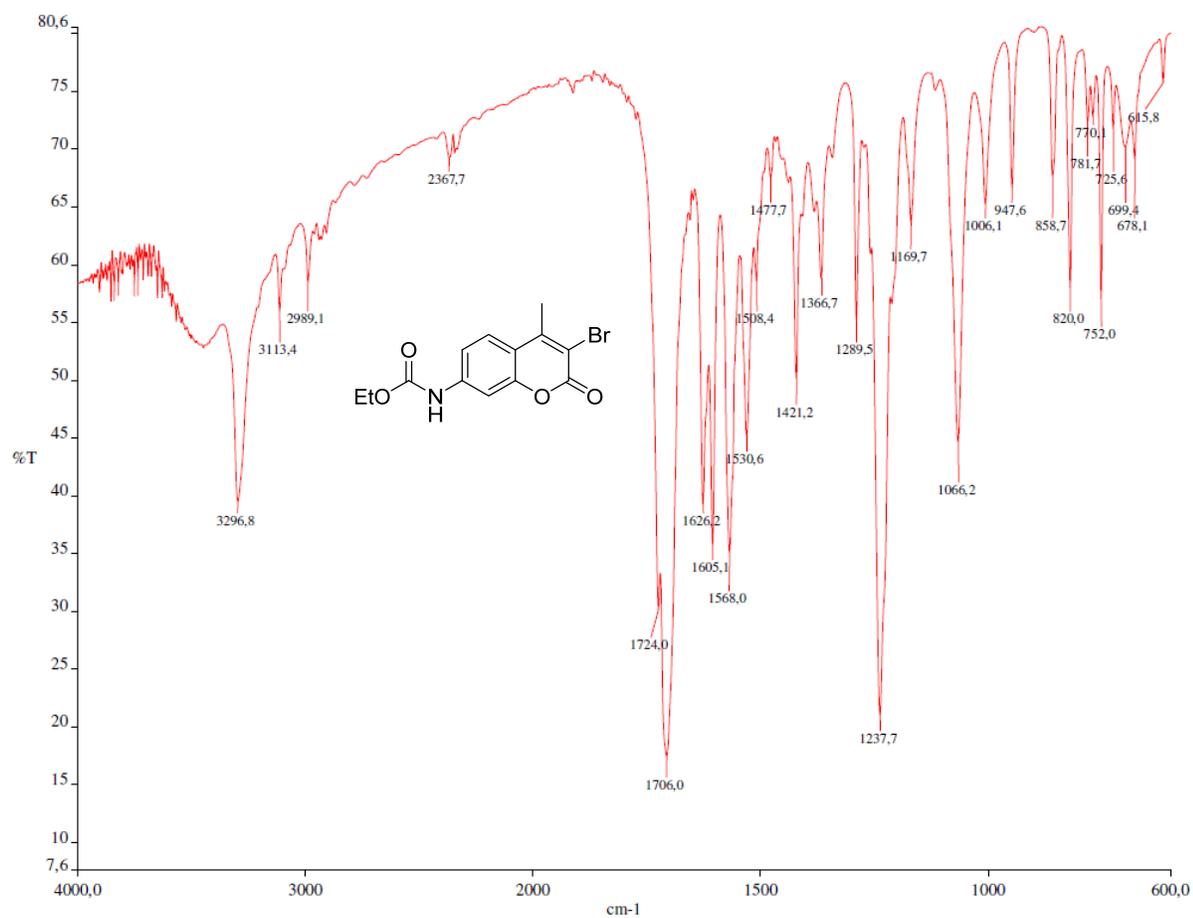
Figure SM 5.2.6.7. IR spectrum (KBr) of 3-bromo-7-ethoxycarbonylamino-4-methylcoumarin.
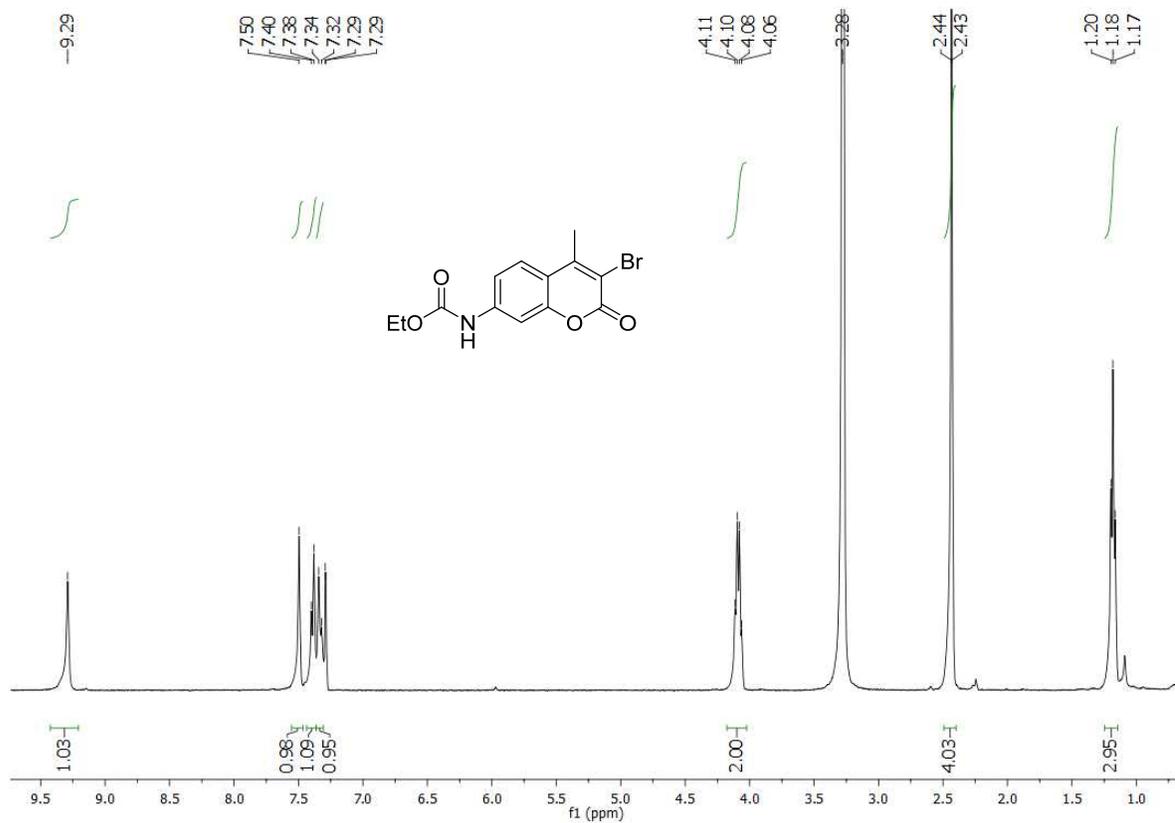
Figure SM 5.2.6.8. ^{1}H NMR spectrum ((400 MHz, CDCl$_3$ + DMSO-d$_6$) of 3-bromo-7-ethoxycarbonylamino-4-methylcoumarin.
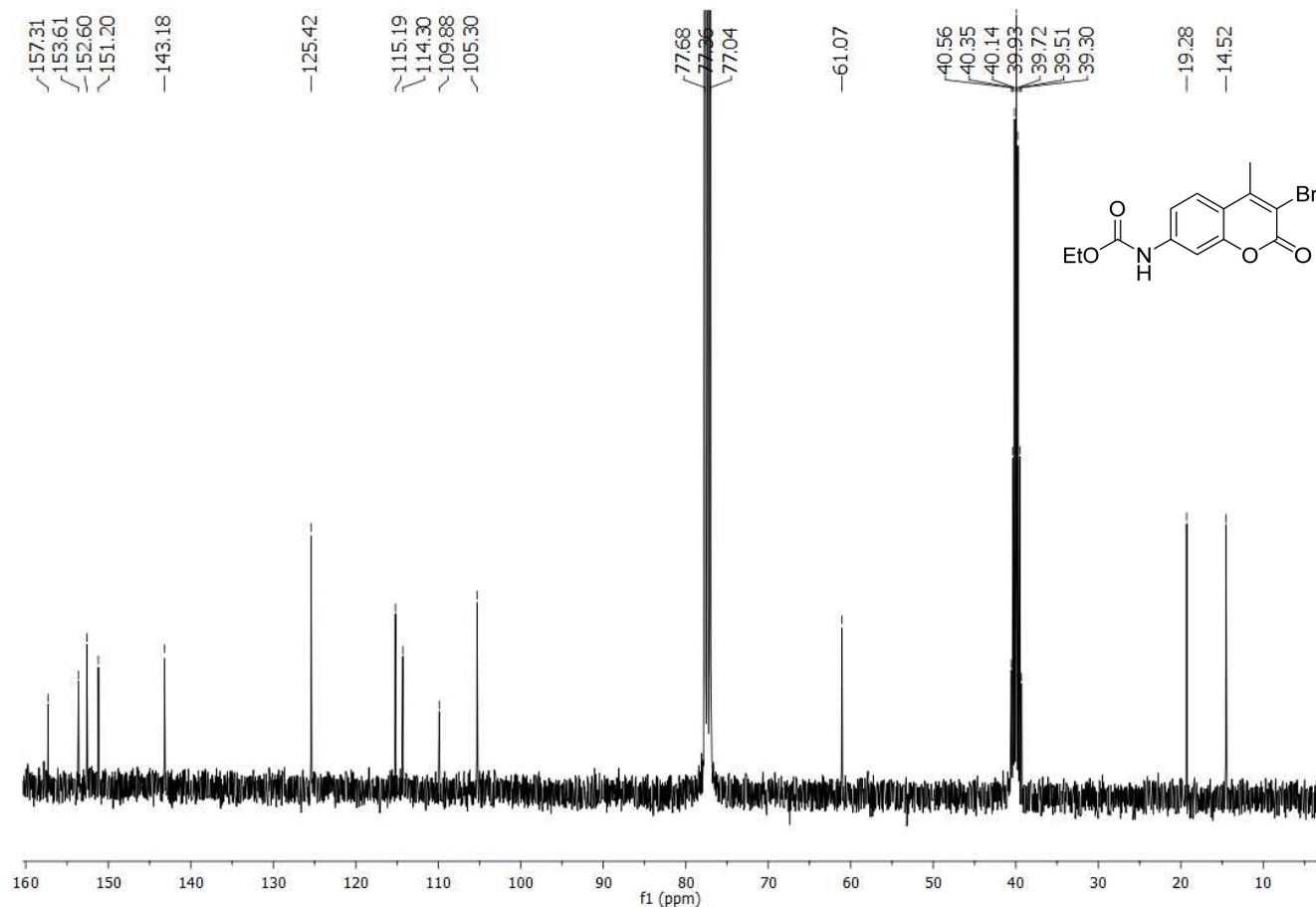
Figure SM 5.2.6.9. ^{13}C NMR spectrum (CDCl$_3$ + DMSO-d$_6$) of 3-bromo-7-ethoxycarbonylamino-4-methylcoumarin.

Synthesis of Musk Ketone

Supplementary Information

This synthesis of a common fragrance illustrates three Aromatic Electrophilic Substitutions (EArS). It has been performed by students of the advanced lab in organic chemistry. The students can discuss relative reactivity and orientation effects in the three reactions, which are important factors in EArS reactions. This work is base in literature work[1], but several important changes were made. The aluminium amalgam was replaced by the more common Lewis acid $FeCl_3$. Average yield is 40-43 g (80-82 %) of tert-butyl-3,5-dimethylbenzene (n_D=1.5038). The average yield in the acetilation is 19-20 g (46%, mp=44-46ºC). In the third step a lot of unchanged reactant was obtained when only fuming nitric acid was used. We followed the procedure of another paper that uses sulfuric acid as catalyst, which increased the yield.[2] Concentrated sulfuric acid also works, but we got higher yield using fuming sulfuric acid. Average yield is 1.0-1.2 g (35-41 %). NMR shows the product to be pure, and the formation of acid 4-t-butil-2,6-dimethyl-3,5-dinito benzoic was not observed. This oxidation was predicted to occur by some works when sulfuric acid is used at high temperatures. The t-butyl-3,5-dimethylbenzene has a nice "earthy" smell, reminiscent of t-butylbenzene, but surprisingly the final product doesn't have an intense smell.

Hints to the questions:

2) The methyl groups are *ortho/para* directors, but stereochemically position 5 is less hindered.

3) Same reason as above. Although the *tert*-butil group activates the product electronically, a second alkylation is prevented by stereochemical reasons.

4) Also due to stereochemical reasons.

Spectroscopy: spectra of pure compounds can be found in the SDBS database.[3]
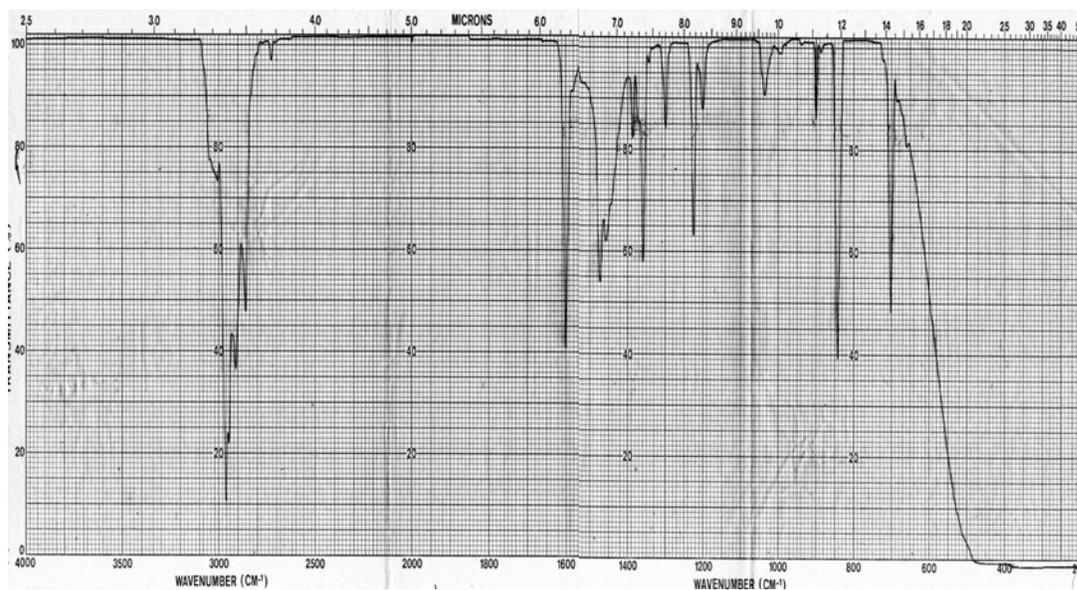
Figure SM 5.2.7.1: IR spectrum of a t-butyl-3,5-dimethylbenzene film (neat, NaCl windows)
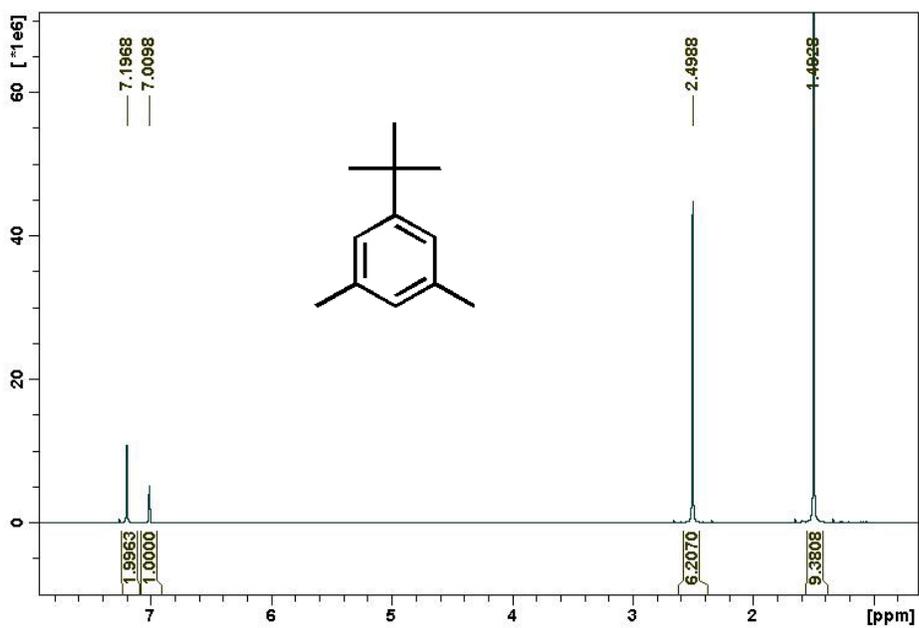
Figure SM 5.2.7.2: ^{1}H NMR spectrum of t-butyl-3,5-dimethylbenzene in CDCl$_3$
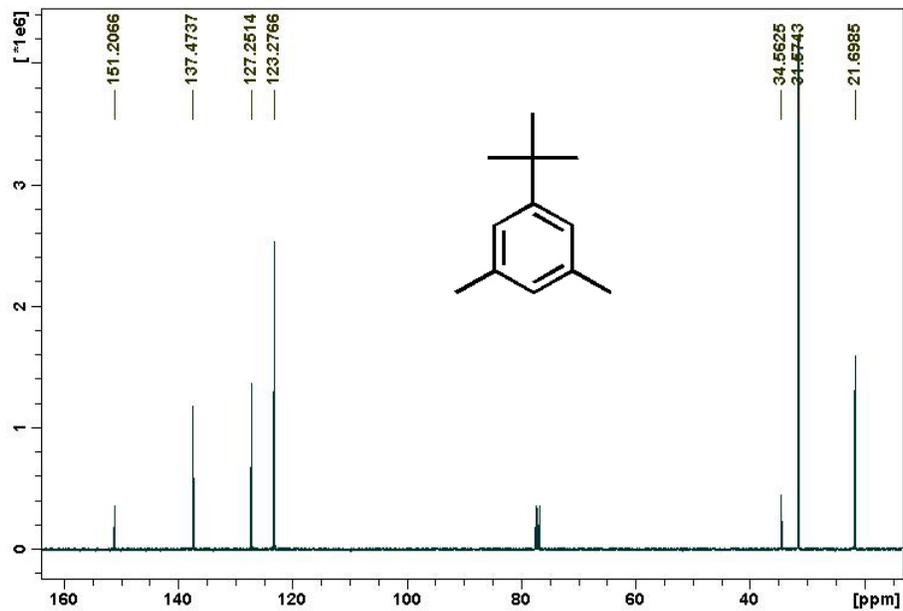
Figure SM 5.2.7.3: ^{13}C NMR spectrum of t-butyl-3,5-dimethylbenzene in CDCl$_3$
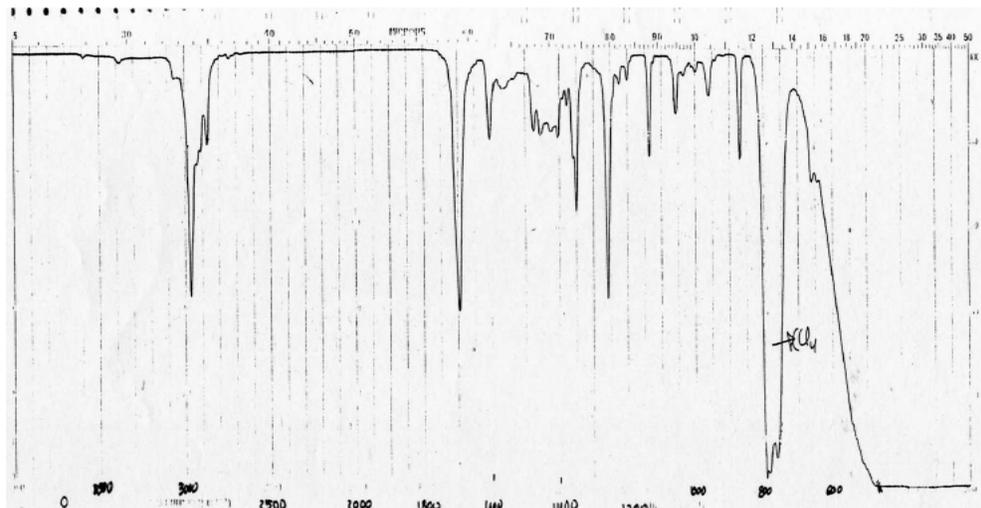
Figure SM 5.2.7.4: IR spectrum of a t-butyl-3,5-dimethylacetophenone film (in CCl$_4$, NaCl windows)
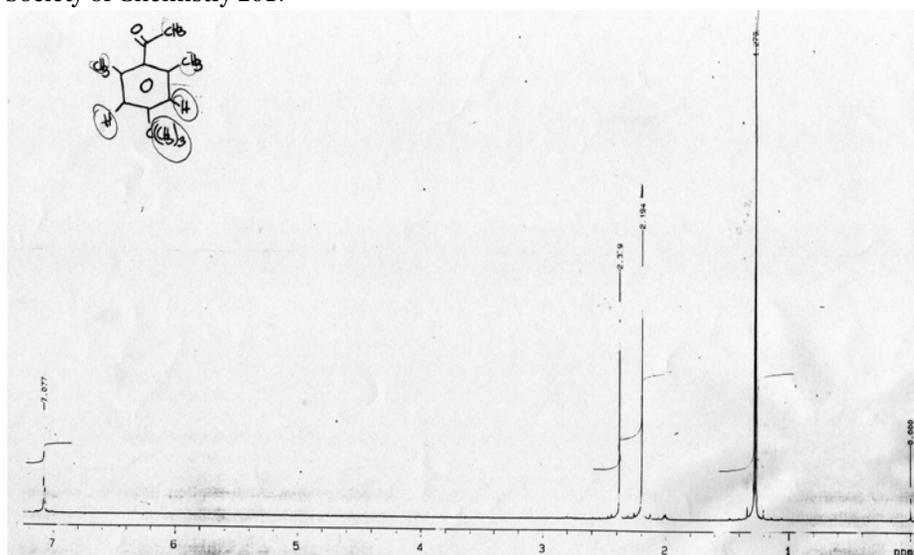
Figure SM 5.2.7.5: ^{1}H NMR spectrum of t-butyl-2,6-dimethylacetophenone in CDCl$_3$
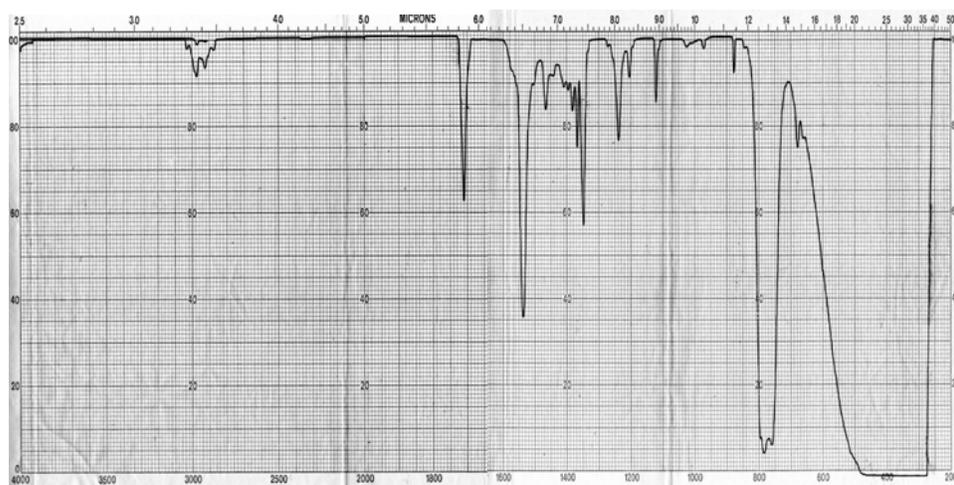
Figure SM 5.2.7.6: IR spectrum of musk ketone (in CCl$_4$, NaCl windows)
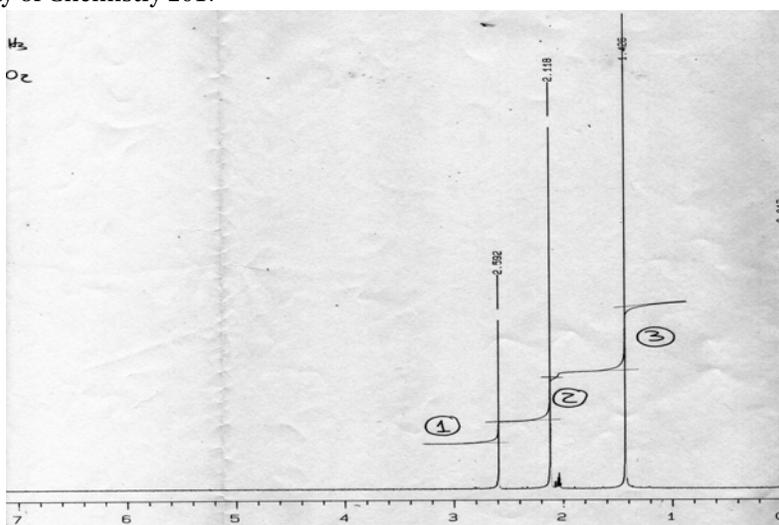
Figure SM 5.2.7.7: ^{1}H NMR spectrum of Musk Ketone in CDCl$_3$

[1] E. Nash, E. J. Nienhouse, T. A. Silhavy, D.E. Humbert and M. J. Mish, " Aromatic Nitro Musk Synthesis", *J. Chem. Educ.*, 1970, *47,* 10, 705.
[2] R.C. Fuson, J. Mills, T. G. Klose , M. S. Carpenter, *J. Org. Chem.*, 1947, **12**, 587.
[3] http://sdbs.db.aist.go.jp/sdbs/cgi-bin/cre_index.cgi

Synthesis of methyl 4-oxo-4-(thiophen-2-yl)butanoate
Supplementary Information

1. Experiment notes

2. Mechanisms

2.1- Conversion of an anhydride to an ester

2.2- Conversion of a carboxylic acid to an acyl chloride

2.3- Friedel-Crafts acylation of thiophene

3. Figures

3.1 Photos for the apparatus used in laboratory sessions 2 and 4.

3.2 ^{1}H NMR and IR spectra of the products

1. Experiment notes

The aim of this experiment is the synthesis of a γ-keto ester from the thiophene heterocycle using simple experimental techniques and cheap, commercially available reagents. The preparation of this compound involves several synthetic transformations, namely the preparation of the acylating reagent followed by the synthesis of a 1,4-dicarbonyl compound derivative of thiophene.

The theoretical concepts associated with the work are vast and have an intermediate degree of difficulty. The students will have contact with several classical reactions in organic chemistry: preparation of an ester using an anhydride as precursor (alcoholysis), nucleophilic substitution (conversion of a carboxylic acid to an acid chloride) and electrophilic aromatic substitution of an heterocycle (Friedel-Crafts acylation).[1]

Several experimental techniques will be used such as heating at reflux under anhydrous conditions, thin layer chromatography (TLC), gravity filtration, liquid-liquid extraction and evaporation of an organic solvent under reduced pressure.[2] The determination of the structure of all compounds synthesized will be also performed through ^{1}H NMR and IR spectroscopic techniques.[3]

Within the scope of this experiment other synthetic methodologies for the synthesis of 1,4-dicarbonyl compounds could be also discussed. In particular the possibility of using several acylating reagents as well as different Lewis acids that could be used as catalysts are interesting topics to explore.

The students should interpreted [1]NMR and IR spectroscopic data of all synthesized products in order to identify the compounds obtained as well to check their purity.

This experiment was developed in the research group of the author and was later performed by students of the 4th year of the undergraduate course in Chemistry of University of Minho as well as by Erasmus students in Chemistry from the University of Metz, France. This experiment could be perform by any undergraduate Chemistry students that already have already some skills concerning the experimental techniques used as well as the knowledge about the theoretical concepts presented (synthesis, reactivity and spectroscopic data interpretation).

Session 1: Synthesis of 4-methoxy-4-oxobutanoic acid
 It is crucial that after the synthesis, the 4-methoxy-4-oxobutanoic acid acid becomes very dry (without methanol) in order to be used in the next reaction step. The presence of residues of methanol can be detected by acquiring an ^{1}H NMR spectrum.

4-Methoxy-4-oxobutanoic acid: colourless solid (97%). ^{1}H NMR (CDCl$_3$) δ 2.60-2.80 (4H, m, 2xCH$_2$), 3,68 (3H, s, OCH$_3$), 9.89 (1H, broad s , OH).
The range of yields obtained earlier by students of the degree course in Chemistry and by Erasmus students was 90-97%.

The reactions of sessions 2 and 3 were performed in dry dichloromethane in order to prevent the hydrolysis of methyl-4-chloro-4-oxobutanoate. When dichloromethane of analytical grade was used the yields of methyl-4-chloro-4-oxobutanoate were 5-10% lower.

Session 2: Synthesis of methyl-4-chloro-4-oxobutanoate
The thionyl chloride used must be previously distilled or obtained from a newly opened bottle, in order to avoid the presence of carboxylic acid which will lower the yields. The dichloromethane used must be previously dry[2] or obtained from a new open bottle. In order to prevent its hydrolysis the methyl-4-chloro-4-oxobutanoate should be stored away from moisture in a closed container until used in the next session.

Methyl-4-chloro-4-oxobutanoate: colourless oil (100%). ^{1}H NMR (CDCl$_3$) δ 2.80 (2H, t, J = 6.6 Hz, CH$_2$), 3.20 (2H, t, J = 6.6 Hz, CH$_2$), 3.71 (3H, s, O CH$_3$).
The yields obtained earlier by students of the degree course in Chemistry and by Erasmus students were quantitative.

Section 3: Synthesis of methyl 4-oxo-4-(thiophen-2-yl)butanoate

The reaction mixture should be stirred at room temperature for 12 h. Alternatively, the reaction mixture can be left stirring for more than 24 h at room temperature or left in the fridge during a week till their purification.Therefore this session could be separated into two sessions. The fourth session will be used for the isolation of the synthesized compound.

Methyl 4-oxo-4-(thiophen-2-yl)butanoate[4]: yellow oil (83%). ^{1}H NMR (CDCl$_3$) δ 2.77 (2H, t, J= 6.6 Hz, CH$_2$), 3.27 (2H, t, J = 6,6 Hz, CH$_2$), 3.71 (3H, s, CH$_3$), 7.12-7.16 (1H, m, 4´-H), 7.65 (1H, dd, J = 5.2 and 1.3 Hz, 5´-H), 7.78 (1H, dd, J = 3.9 and 1.3 Hz, 3´-H). IR (liquid film): ν 1739, 1666 cm^{-1}.
The range of yields obtained earlier by students of the degree course in Chemistry and by Erasmus students was 75-83%.

References

[1] J. March, "Advanced organic chemistry: reactions, mechanisms and structure, 4th Ed.; John Wiley and Sons, New-York, 1992, p. 539.

[2] W.-L.F. Armarego and D. D. Perrin, "Purification of laboratory chemicals", 4th Ed.; Butterworth Heinemann, Oxford, 2000.

[3] (a) R. M. Silverstein, F. X. Webster and D. J. Kiemle, "Spectrometric Identification of Organic Compounds", 7th Edition, John Wiley and Sons: New York, 2005. (b) E. Pretsch, P. Buhlmann and C. Affolter, "Structure Determination of Organic Compounds" 3rd Edition, Springer-Verlag: Berlin, 2000.

[4] M. M. M. Raposo, M. M. M. and G. Kirsch *Heterocycles*, 2001, **55**, 1487.

2. Mechanisms

2.1- Conversion of an anhydride to an ester (nucleophilic substitution on the carbonyl group) alcoholysis

The alcoholysis of a carboxylic anhydride is a nucleophilic substitution on the carbonyl group, using an alcohol as nucleophile that leads to an ester through ring opening (Scheme SM 5.2.8.1.).
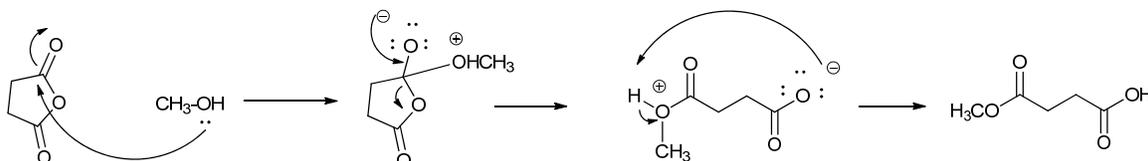
Scheme SM 5.2.8.1. Mechanism of conversion of an anhydride to an ester.

2.2- Conversion of a carboxylic acid to an acyl chloride (nucleophilic substitution on the carbonyl group)

The conversion of a carboxylic acid into an acyl chloride can be performed using $SOCl_2$ or PCl_3. The problem to be solved is the changing of a poor leaving group (OH) into a good one. The transformation begin with the conversion of a carboxylic acid into an inorganic derivative in which the substituent on the carbonyl carbon is a good leaving group. This reaction proceed through an addition-elimination mechanism (Scheme SM 5.2.8.2.).
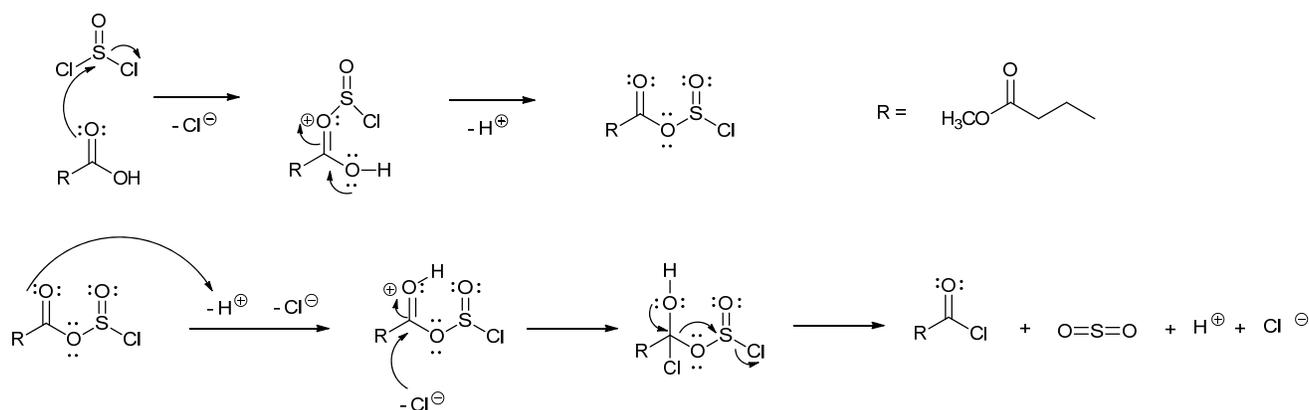
Scheme SM 5.2.8.2. Mechanism of conversion of a carboxylic acid into an acyl chloride.

2.3- Friedel-Crafts acylation of thiophene (eletrophilic aromatic substitution)

The Friedel-Crafts acylation is an electrophilic aromatic substitution which proceeds through the intermediacy of an acylium cation with the general structure: $R-C\equiv O^{+}$. This species can be form by the reaction of acid chlorides with aluminum chloride, stannic chloride, etc. The Lewis acid initially coordinates to the carbonyl oxygen. This complex is in equilibrium with an isomer in which the Lewis acid is bound to the halogen. Dissociation then produces the acylium ion, which is stabilized by resonance and, unlike alkyl reactions, it is not prone to rearrangements (Scheme SM 5.2.8.3.).

Step 1: Formation of acylium ion
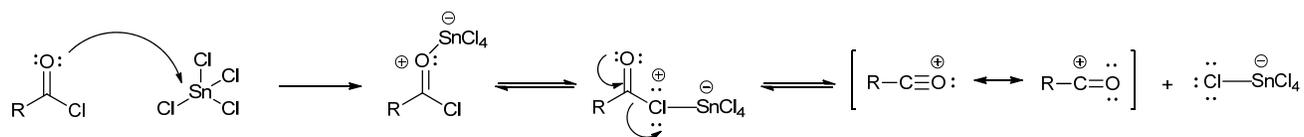
Step 2: Eletrophilic acylation of thiophene
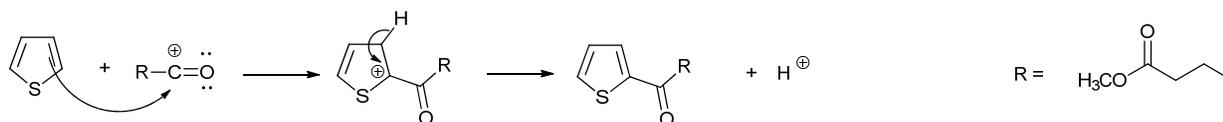
Scheme SM 5.2.8.3. Mechanism of the Friedel- Crafts acylation of thiophene.

3. Figures

3.1 Photos for the apparatus used in laboratory sessions 2 and 4.

Figure SM 5.2.8.1 Left - Photo of the apparatus for heating at reflux in dry conditions (Laboratory session 2); Right - Photo of the apparatus for the Friedel-Crafts acylation of thiophene (Laboratory session 4).

3.2 ^{1}H NMR and IR spectra of the products
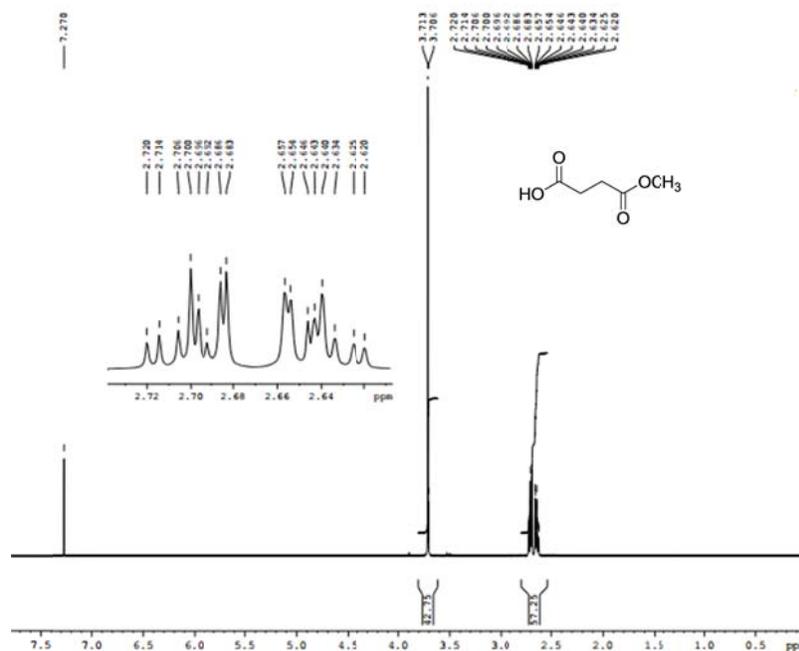
Figure SM 5.2.8.2. ^{1}H NMR Spectrum of 4-methoxy-4-oxobutanoic acid in CDCl$_3$ obtained using a Bruker Avance III 400 spectrometer operating at 400 MHz at 25 $^{\circ}$C.
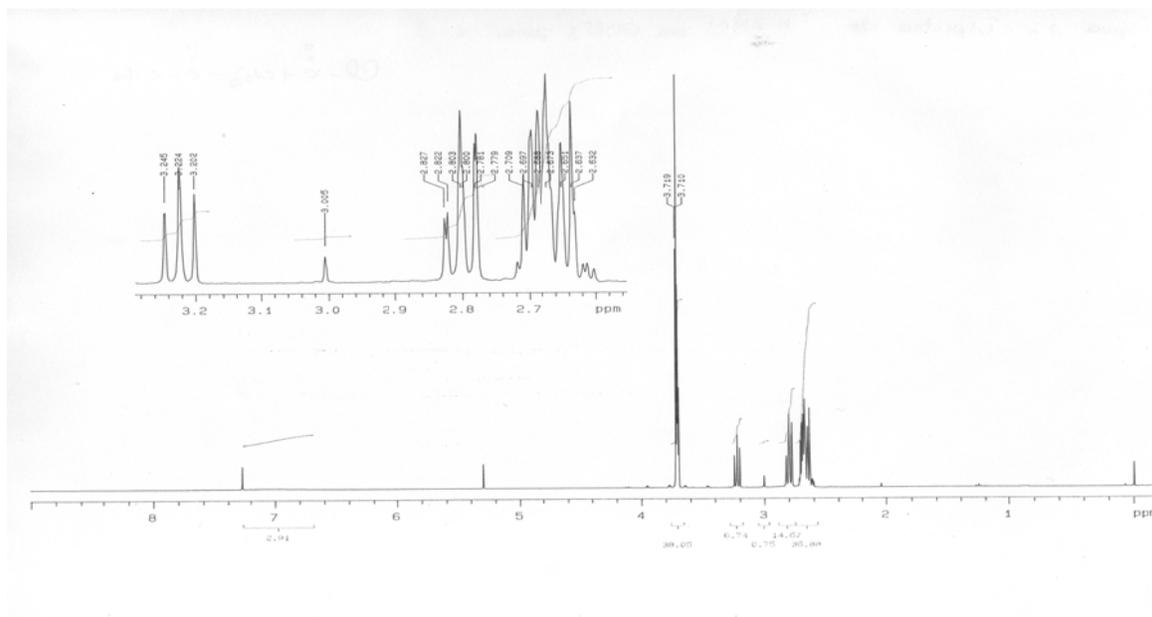
Figure SM 5.2.8.3. ^{1}H NMR Spectrum of methyl-4-chloro-4-oxobutanoate in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 300 MHz at 25 $^{\circ}$C.
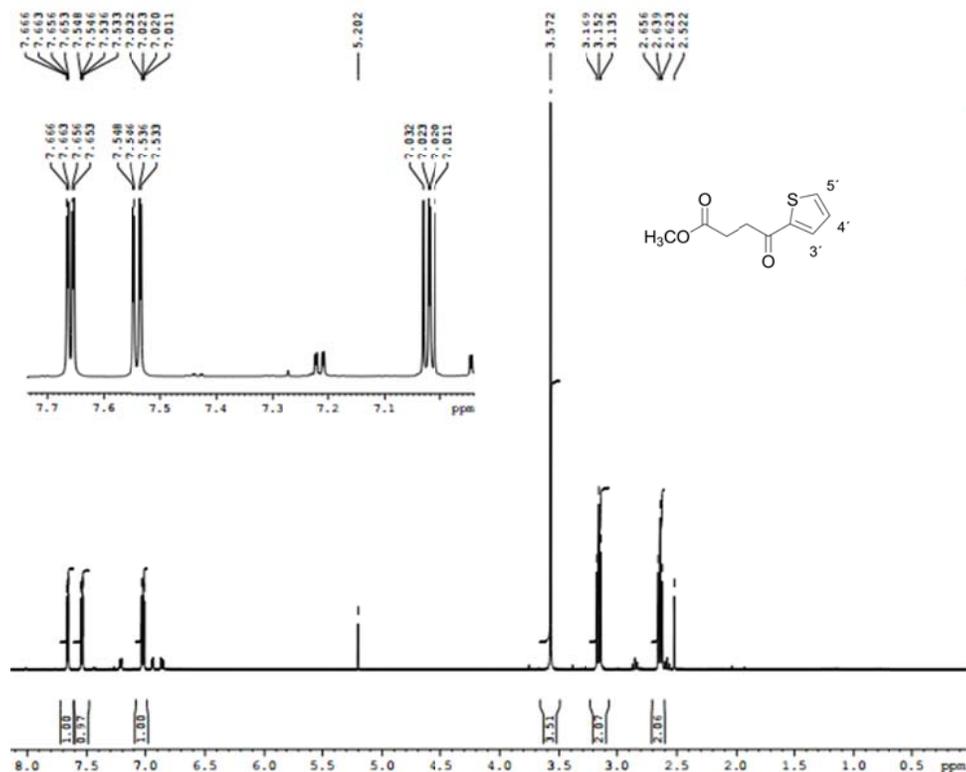
Figure SM 5.2.8.4. ^{1}H NMR Spectrum of methyl 4-oxo-4-(thiophen-2-yl)butanoate in CDCl$_3$ obtained using a Bruker Avance III 400 spectrometer operating at 400 MHz at 25 °C.
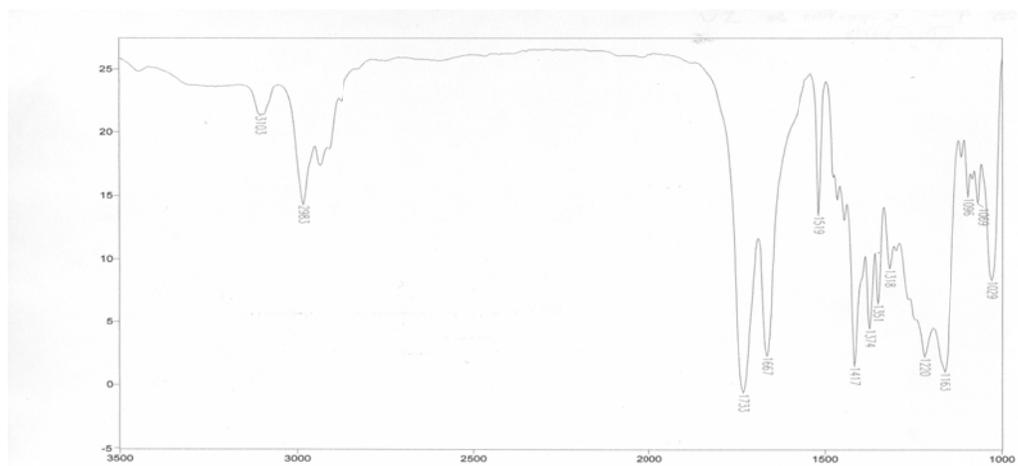
Figure SM 5.2.8.5. IR spectrum of methyl 4-oxo-4-(thiophen-2-yl)butanoate in liquid film obtained in a Perkin Elmer FTIR-1600 spectrophotometer.

Synthesis of eosin
Supplementary Material

This experiment proposal was tested by students of advanced organic chemistry[1] in which the concepts of Friedel-Crafts type reactions and electrophilic aromatic substitution were taught. These reactions take place on strongly activated aromatic rings due to the presence of OH groups. It's a good example to explain both reactivity and orientation effect of substituents of aromatic rings. In the Friedel–Crafts reactions, the new C–C bonds are only formed at *ortho* and *para* positions to the OH group and fluorescein bromination only occurs *ortho* to the OH groups. Fluorescein synthesis was performed with zinc chloride as Lewis acid, but in literature a suitable procedure using sulphuric acid is described[2]. The experimental procedure is easy but the use of bromine is not advisable to less skilled students. This work became a success among students due to fluorescence of synthesised products, the chemiluminescent reaction and the dyeing process with eosin.

Additional notes on the preparation of fluorescein

Some changes were made to fluorescein preparation[1,3] in order to optimize the reaction. This reaction was performed in a 3 necked flask fitted with reflux condenser and thermometer instead reaction tube due to the excessive moisture liberation. It was not tried but the sand bath probably could be substituted for oil bath. Melting point of phthalic anhydride is 131ºC and 110ºC for resorcinol. The product was isolated by vacuum filtration instead liquid removal with Pasteur pipette[3]. The yield is around 70-75 % (80%) and melting point 312.3-313.1ºC (314-316ºC[4]). When dissolved in alkali, a splendid green fluorescence appears even in the sun light. This effect is stronger under UV light. With acidification this fluorescence disappears (**Figure SM 5.2.9.1**).

Figure SM 5.2.9.1 - Fluorescein basic solution under sun light and UV light

Additional notes on the preparation of eosin

This reaction was also performed with a 3 necked flask fitted with reflux condenser, thermometer and dropping funnel. Carbon tetrachloride is the usual solvent for this kind of reaction but is highly toxic and causes environmental problems since it is involved in stratospheric ozone depletion. Suggested solvents substitutes are normally dichloromethane or chloroform. Trifluorotoluene can be a suitable alternative due to its lower toxicity and higher boiling point. CF_3 group is relatively stable and deactivates the aromatic ring in electrophilic aromatic substitutions[5], being much less reactive than fluorescein. The vacuum filtration has to be also performed in the fume hood due to excess of bromine. The yield is around 65-75 % (77%) and melting point 294.5-296.3ºC (295-296ºC[4]). When a few milligrams are dissolved in ethanol, an orange fluorescence can be observed under a UV light (**Figure SM 5.2.9.2**).
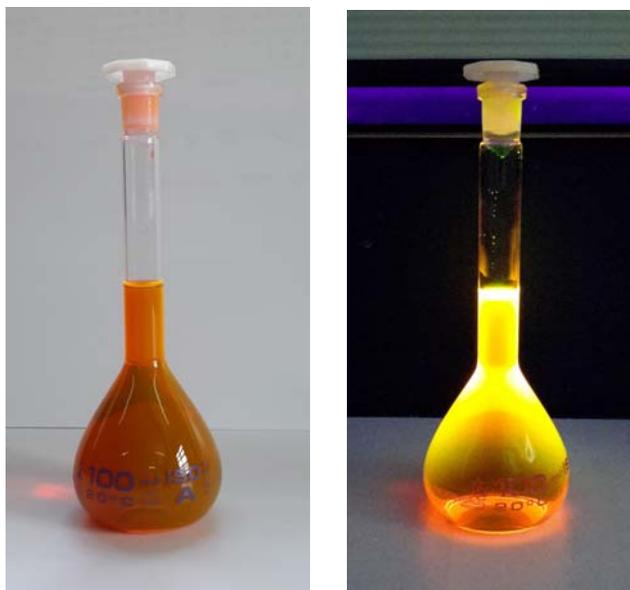
Figure SM 5.2.9.2 – Eosin ethanolic solution under sun light and UV light

Additional notes on dyeing of cotton

Scheme SM 5.2.9.3 shows the eosin ammonium salt formation suitable to dyeing.
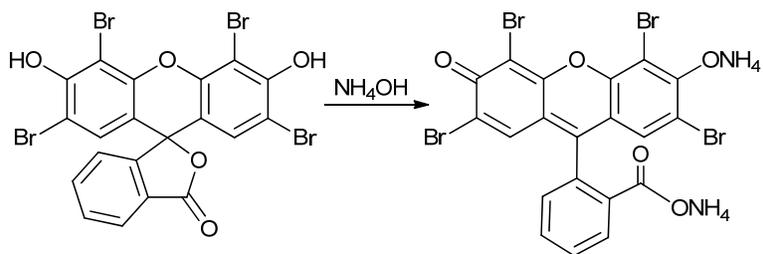
Scheme SM 5.2.9.3 – Scheme of eosin ammonium salt formation

The **Figure SM 5.2.9.4** shows the preparation of eosin ammonium salt by exposition to ammonia vapors.
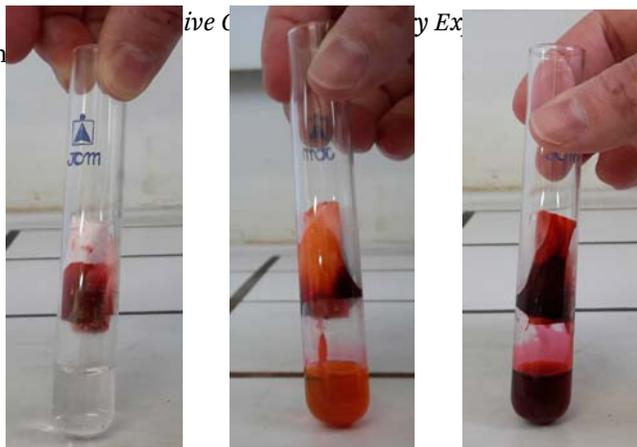
Figure SM 5.2.9.4 - Preparation of eosin ammonium salt

The **Figure SM 5.2.9.5** shows the dyeing process and a piece of cotton before and after dyeing with eosin.

Figure SM 5.2.9.5 – Dyeing process of a piece of cotton with eosin

Quemiluminescence[6] reaction was performed using a peroxyoxalate system[7] with bis(2,4,6-trichlorophenyl oxalate) and sodium acetate[8] in the presence of eosin. The addition of hydrogen peroxide results in a strong light-orange color emission that lasts for several minutes (**Figure SM 5.2.9.6**).

Figure SM 5.2.9.6 – Quemiluminescence reaction using bis(2,4,6-trichlorophenyl oxalate).

IR spectra:
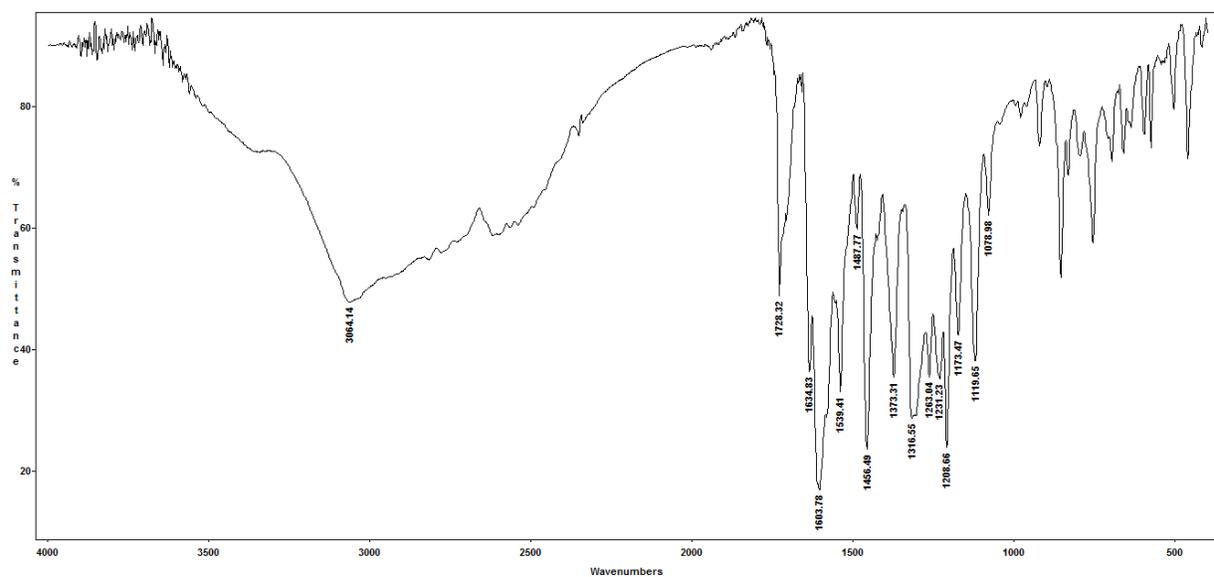
Figure SM 5.2.9.7 - IR (KBr) of fluorescein (lactonic form)
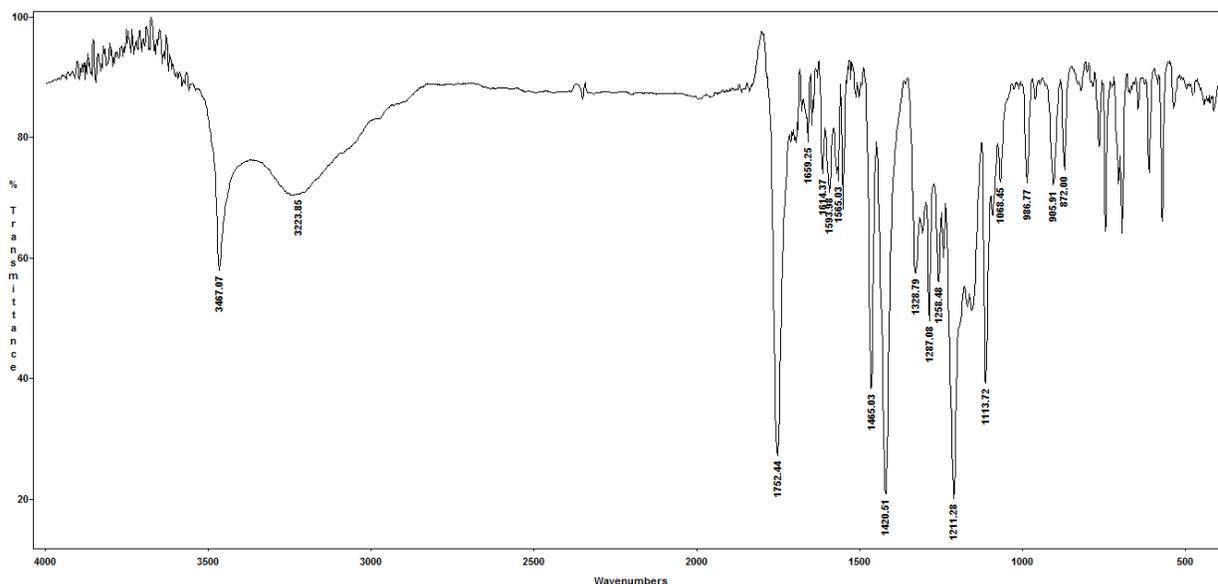
Figure SM 5.2.9.8 - IR (KBr) of eosin

IR spectra of fluorescein (lactonic form) and eosin can be found in SDBS (nº 6347 and 2299)[9].

NMR spectra:
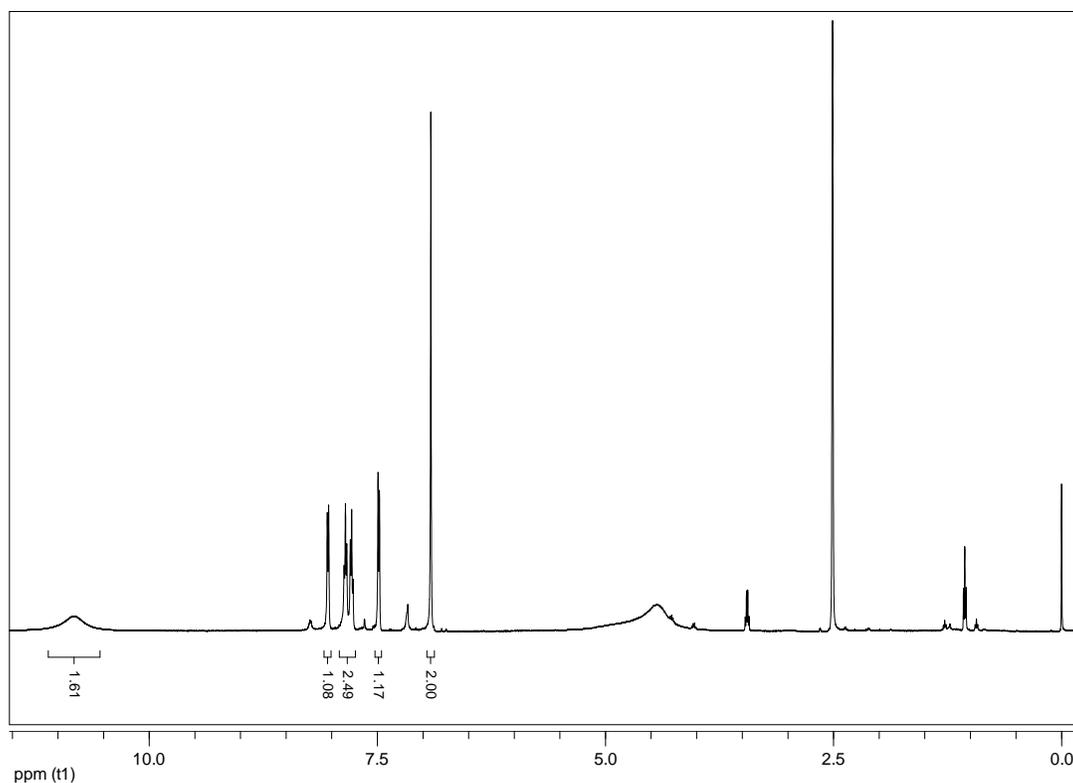
Figure SM 5.2.9.9 - ^{1}H RMN of eosin (300 MHz, DMSO-d6)

^{1}H and ^{13}C RMN spectra of fluorescein (lactonic form) and eosin can be found in SDBS (nº 6347 and 2299)[9].

[1] C. A. M. Afonso, D. P. Simão, L. P. Ferreira, M. S. Serra, M. M. M. Raposo, *100 Experiências de Química Orgânica*, IST Press, 2011, 381.

[2] J. V. Mc. Cullagh and K. A. Dagget, *J. Chem. Ed.,* 2007, 84 (11), 1799.

[3] F. Fieser; K. L. Williamson, *Organic Experiments*, Houghton Mifflin Company, 8th ed, 1998, 451-453.

[4] W. R. Orndorff; A.J.Hemmer, *J. Am. Chem. Soc.*, 1927, **49**, 1272-1280.

[5] J. L. O' Connell, J. S. Simpson, P. G. Dumanski, G. W. Simpson and C. J. Easton*, Org. Biomol. Chem.,* 2006, **4***,* 2716-2723.

[6] http://www.scienceinschool.org/2011/issue19/chemiluminescence, access in May 2015.

[7] R. Albertin, M. Arribas, E. Bastos, S. Röpke, P. Sakai, A. Sanches, C. Stevani, I. Umezu, J.Yu e W. Baader, *Química Nova*, 1998, **21**, 6, 772.

[8] A. G. Hadd, D. W. Lehmpuhl, L. R. Kuck and Galen P. Mell, *J. Chem. Educ.*, 1999, **76**, 1237-1240.

[9] http://sdbs.db.aist.go.jp/sdbs/cgi-bin/direct_frame_top.cgi, access in May 2015.

Synthesis and formylation of 5-piperidino-2,2´-bithiophene
Supplementary Material

1. Experiment notes

2. Mechanism

3. Figures

3.1 Photo of the TLC plate with all products

3.2 ^{1}H, ^{13}C NMR and IR spectra of the products

1. Experiment notes

The aim of this experiment is the synthesis of a 5-*N,N*-dialkylamino-2,2´-bithiophene as well as its associated 4- and 5´-formyl derivatives through several synthetic transformations, namely: i) thionation of carbonyl groups[1] of a tertiary γ-ketoamide derived from thiophene using Lawesson´s reagent followed by cyclization of the corresponding thioxoamide through intramolecular nucleophilic attack[2] yielding the 5-*N,N*-dialkylamino-2-2´-bithiophene derivative; formylation of the bithiophene derivative using two different synthetic methodologies: ii) Vilsmeier-Haack reaction (electrophilic aromatic substitution)[3] or iii) lithiation followed by electrophilic aromatic substitution.[4] Information about the mechanisms can be found in references 3c,d.

The formylation of 5-piperidino-2,2'- bithiophene *via* two different methods of synthesis selectively yields two different products (4- or 5´-formyl derivatives), this promotes a discussion concerning the selectivity of each reaction having in mind the reagents used in each method as well as the mechanism of these transformations.

Realizing that Vilsmeier formylation is an aromatic electrophilic reaction the electrophile (the Vilsmeier reagent ClCH=NMe$_2^+$) will react at the most electron rich part (nucleophilic) of the 5-piperidino-2,2'-bithiophene that is the adjacent position to the *N,N*-dialkylamino group to produce 4-formyl-5-piperidino-2,2´-bithiophene. On the other hand, the reaction of 5-piperidino-2,2'-bithiophene with *n*-BuLi will selectively yield the organolithium derivative by deprotonation of the adjacent proton to the sulphur atom due its higher acidity (5´-position of the 5-piperidino-2,2'-bithiophene). Subsequent reaction of the organolithium derivative with DMF will give the 5´-formyl-5-piperidino-2,2´-bithiophene. A correlation between the selectivity in this reaction can be found through the analysis of the ^{1}H NMR spectra. The most acidic proton 7.05-7.11 (1H, m, 5´-H) will be selectively metalated by *n*-BuLi.

Other methods for the synthesis of bithiophene derivatives such as Stille and Suzuki cross coupling reactions could also be discuss in order to broaden the approach to this subject.[5]

The students should interpret ^{1}H NMR, ^{13}C NMR and IR spectroscopic data of all synthesized products in order to identify the compounds obtained as well to check their purity. Additionally the characterization of the compounds through the usual spectroscopic techniques (^{1}H NMR, ^{13} NMR, IR) is extremely interesting and could be used in order to compare the spectra of the precursors with the spectra of the final products having in mind the confirmation of the functional group transformations as well as the positions for which the functionalization at the thiophene ring occurs by considering the ^{1}H NMR coupling constants and multiplicity of signals.[6]

The students will use several experimental techniques such as heating at reflux under anhydrous conditions with an inert atmosphere, liquid-liquid extraction, evaporation of organic solvents with a rotary evaporator, gravity and vacuum filtration, thin layer chromatography (TLC) and column chromatography on silica gel and melting point.[7]

This experiment was previously developed in the research group of the author and was performed later by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students from the undergraduate Chemistry degree course at the University of Metz, France. This experiment is appropriate for undergraduate Chemistry students who have previously acquired some skills with the experimental techniques involved ~~used~~ as well some knowledge regarding the theoretical concepts presented (synthesis, reactivity and spectroscopic data interpretation). Therefore this experiment may be appropriate for the last year project in Chemistry degree or as the practical component of advanced chemistry subjects at Master level.

The synthetic methodology described in this experiment could also be used in general for the preparation of 4-*N,N*-dialkylamino-2,2-bithiophenes[8] as well as the corresponding formylated derivatives[9]

Notes for the instructor

Session 1: Synthesis of 5-piperidino -2,2'- bithiophene

The reaction mixture should be heated at gentle reflux during 30 minutes (check the conversion of the amide by TLC). Prolonged heating of the reaction mixture may lead to decomposition products of the Lawesson reagent.

Session 2: Purification of 5-piperidino-2,2'-bithiophene

The purification of the crude bithiophene obtained in session 1 should be performed by column chromatography with a silica gel (60Ä; 200-300 mesh): column height and diameter: 50 and 3,5 cm; silica weight: 90 g; silica height: ≈ 40 cm; column packing solvent: petroleum ether (40-60 °C).

The dissolution of the residue obtained in session 1 should be done using a minimum amount of chloroform and the elution should start with petroleum ether (40-60 °C) followed by elution with a mixture of petroleum ether (40-60 °C)-diethyl ether (9:1). The contents of each collected fraction should be check by TLC (eluent: petroleum ether (40-60 °C)-diethyl ether (9:1).

The preparation of the silica gel column and the separation of the components of the mixture (session 2, 4 and 6) should be run in a well-ventilated fume hood in order to avoid the contact with the silica gel as well as the inhalation of volatile solvents such as chloroform, ethyl ether and petroleum ether. Silica gel may cause respiratory irritation, do not breathe the associate dust. After being used the silica gel must be stored in a closed container in the fume hood for subsequent disposal.

5-Piperidino-2,2'-bithiophene:[8] pale yellow solid (79%). Mp: 59-60 °C. ^{1}H NMR (CDCl$_3$) δ 1.50-1.64 (2H, m, CH$_2$), 1.68-1.80 (4H, m, 2xCH$_2$), 3.10-3.20 (4H, m, 2xCH$_2$), 6.00 (1H, d, J = 4.0 Hz, 4-H), 6.89 (1H, d, J = 4.0 Hz, 3-H), 6.95-7.00 (2H, m, 3′ and 4′-H), 7.05-7.11 (1H, m, 5′-H). ^{13}C NMR (CDCl$_3$) δ 23.60, 25.09, 52.58, 105.48, 121.39, 122.43, 123.04, 127.51, 138.48, 158.44. IR (liquid film): v 2939, 2826, 1556, 1514, 1488, 1446, 1301, 1276, 1242, 1192, 1121, 1068, 1013, 894, 826, 810, 769, 682 cm^{-1}. UV (EtOH): λ$_{max}$ nm (ε/M^{-1} cm^{-1}) 343.0 (18,039), 241.5 (4914), 214.0 (6102).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 70-75%.

Sessions 3: Synthesis of 4-formyl-5-piperidino-2,2′-bithiophene

The drying agent used (anhydrous Na$_2$SO$_4$) should be washed several times with diethyl ether in order to recover the maximum amount of compound.

The reactions performed in sessions 3 and 5 should be carried out in anhydrous conditions and under an inert atmosphere (session 5) due to the higher reactivity/decomposition of POCl$_3$, DMF and *n*-BuLi in the presence of water or oxygen.

The function of POCl$_3$ and DMF reagents in session 3 is to produce the reactive species (the Vielsmeier reagent: ClCH=NMe$_2$$^+$).

Session 4: Purification of 4-formyl-5-piperidino-2,2'-bithiophene

The purification of the crude bithiophene derivative obtained in session 3 should be performed by column chromatography using a silica gel (60Ä; 200-300 mesh): column height and diameter: 50 and 2,5 cm; silica weight: 50 g; silica height: $\approx$ 40 cm; column packing solvent: petroleum ether (40-60 °C).

The dissolution of the residue obtained in session 3 should be done using a minimum amount of chloroform and the elution should start with petroleum ether (40-60 °C)- diethyl ether (9:1) followed by elution with a mixture of petroleum ether (40-60 °C)-diethyl ether (1:1). The contents of each collected fraction should be check by TLC (eluent: petroleum ether (40-60 °C)-diethyl ether (1:1).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 65-75%.

4-Formyl-5-piperidino-2,2′-bithiophene:[9] beige solid (80%). Mp: 71-72 °C. ^{1}H NMR (CDCl$_3$) δ 1.60-1.70 (m, 2H, CH$_2$), 1.70-1.90 (m, 4H, 2xCH$_2$), 3.30-3.40 (m, 4H, 2xCH$_2$), 6.98-7.06 (m, 1H, 4′-H), 7.17 (dd, 1H, J=3.6 and 1.2 Hz, 3′-H), 7.30 (s, 1H, 3-H), 7,39 (dd, 1 H, J=5.1 and 1.2 Hz, 5′-H), 9,83 (s, 1H, CHO). ^{13}C NMR (CDCl$_3$) δ 23.55, 25.43, 56.25, 122.66, 123.00, 123.94 (2 overlapped signals), 124.18, 127.64, 136.97, 168.56, 182.57. IR (liquid film): ν 2936, 2920, 1660 (CHO), 1562, 1511, 1495, 1461, 1442, 1381, 1333, 1246, 1165, 1127, 1074, 1040, 991, 910, 826, 752, 694 cm^{-1}. UV λ$_{max}$ nm (etanol, ε/M^{-1} cm^{-1}) 332.0 (14,794), 253.0 (17,719), 214.0 (6102).

Session 5: Synthesis of 5′-formyl-5-piperidino-2,2′-bithiophene

Particular attention should be taken during the manipulation of the *n*-Butyllithium due to its inflammability, especially when concentrated solutions (2.5 M or higher) are exposed to air. The drying agent used (anhydrous MgSO$_4$) should be washed several times with diethyl ether in order to recover the maximum amount of compound.

Session 6: Purification of 5′-formyl-5-piperidino-2,2'-bithiophene

The purification of the crude bithiophene derivative obtained in session 5 should be performed by column chromatography using a silica gel (60Ä; 200-300 mesh): column height and diameter: 30 and 2,5 cm; silica weight: 15 g; silica height: $\approx$ 20 cm; column packing solvent: petroleum ether (40-60 °C).

The dissolution of the residue obtained in session 5 should be done using a minimum amount of chloroform and the elution should start with petroleum ether (40-60 °C)- diethyl ether (8:2) followed by elution with a mixture of petroleum ether (40-60 °C)-diethyl ether (1:1). The contents of each collected fraction should be check by TLC (eluent: petroleum ether (40-60 °C)-diethyl ether (1:1).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 71-76%.

5´-Formyl-5-piperidino-2,2´- bithiophene:[9] orange solid (84%). Mp: 145-146 °C, (143-145 °C).[10] ^{1}H RMN (CDCl$_3$) δ 1.50-1.85 (m, 6H, 3xCH$_2$), 3.15-3.30 (m, 4H, 2xCH$_2$), 6.00 (d, 1H, J=4.0 Hz, 4´-H), 6.97 (d, 1H, J=4.0 Hz, 3´-H), 7.12 (d, 1 H, J=4.0 Hz, 3-H), 7.59 (d, 1H, J=4.0 Hz, 4-H), 9.77 (s, 1H, CHO). ^{13}C RMN (CDCl$_3$) δ 23.60, 25.02, 51.65, 104.47, 120.25, 120.90, 127.05, 138.02, 138.84, 149.44, 161.74, 181.94. IR (KBr): ν 2950, 2840, 2780, 1650 (C=O), 1507, 1490, 1420, 1380, 1250, 1230, 1120, 1080, 1050, 1000, 900, 860, 820, 800, 760, 750, 660 cm^{-1}. λ_{max} nm (ethanol, ε/M^{-1} cm^{-1}) 442.0 (22,133), 287.0 inf. (3809), 268.0 (5790), 217.0 (5095).

2. Mechanism

A plausible mechanism for the formation of the thiophene heterocycle from secondary amides in the presence of LR, involves an initial thionation of the amides to the corresponding 4-thioxo thioamides followed by a ring closure of the imidothiol form of 4-thioxo thioamide to give 5-amino-2,2´-bithiophenes (Scheme SM 5.2.10.1.) A similar mechanism was proposed earlier by Nisho for the formation of pyrroles and thiophenes from diphenyl-4-oxobutanamides.[2]
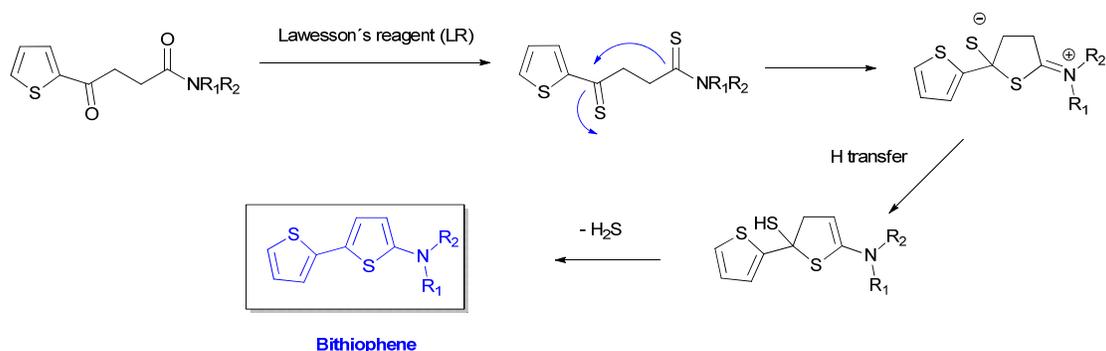
Scheme SM 5.2.10.1. Mechanism for the formation of thiophene heterocycle from tertiary γ–keto amides by reaction with Lawesson reagent.

References

[1] (a) M. P. Cava and M. I. Levinson, *Tetrahedron*, 1985, **41**, 5061. (b) R. A. Cherkasov, G. A. Kutyrev and A. N. Pudovick, *Tetrahedron*, 1985, **41**, 2567. (c) T. Ozturk, E. Ertas and O. Mert, *Chem Rev.*, 2007, **107**, 5210.

[2] T. Nisho, T. *Helv. Chim. Acta*, 1998, **81**, 1207.

[3] (a) C. W. Bird, Ed.; *Comprehensive Heterocyclic Chemistry II*; Pergamon: Oxford, 1984, Vol. 4, p 751-754 and p. 771. (b) G. Jones and S. P. Stanforth, In: The Vilsmeier Reaction of Fully Conjugated Carbocycles and Heterocycles", L. A. Paquete Ed.; Organic Reactions, Wiley: New York, 1997, Vol. 49, p. 15-17. (c) J. March, "Advanced organic chemistry: reactions, mechanisms and structure, 4th Ed., John Wiley and Sons, New-York, 1992, p. 525-526 and 542-543.(d) L. Kurti, B. Czakó, Strategic applications in named reactions in organic synthesis, Elsevier, S. Diego, USA, 2005, p 468.

[4] (a) M. Schlosser, *Angew. Chem. Int. Ed.*, 2005, **44**, 376. (e) R. Chinchilla, C. Nájera and M. Yus, *Chem. Rev.*, 2004, **104**, 2667.

[5] (a) P. Bäuerle In: The Synthesis of Oligothiophenes, D. Fichou Ed. Handbook of Oligo- and Polythiophenes, Wiley-VCH: Weinheim, **1999**; Cap. 3, p 89-173. (b) M. M. M. Raposo, A. M. C. Fonseca and G. Kirsch, *Tetrahedron*, 2004, **60**, 4071. (c) C. Herbivo, A. Comel, G. Kirsch and M. M. M. Raposo, *Tetrahedron*, 2009, **65**, 2079.

[6] (a) R. M. Silverstein, F. X. Webster and D. J. Kiemle, "Spectrometric Identification of Organic Compounds", 7th Edition, John Wiley and Sons: New York, 2005. (b) E. Pretsch, P. Buhlmann and C. Affolter, "Structure Determination of Organic Compounds" 3rd Edition, Springer-Verlag: Berlin, 2000.

[7] W.-L.F. Armarego and D. D. Perrin, "Purification of laboratory chemicals", 4th Ed.; Butterworth Heinemann, Oxford, 2000.

[8] M. M. M. Raposo and G. Kirsch, *Heterocycles*, 2001, **55**, 1487.

[9] M. M. M. Raposo and G. Kirsch, *Tetrahedron,* 2003, **59**, 4891.

[10] P. V. Bedworth, Y. Cai, A. Jen and S. R. Marder, *J. Org. Chem.* 1996, **61**, 2242.

3. Figures

3.1 Photo of the TLC plate with all products

A= 5-piperidino-2,2´-bithiophene; R_f=0.91

B=4-formyl-5-piperidino-2,2´-bithiophene; R_f=0.86

C=5´-formyl-5-piperidino-2,2´-bithiophene; R_f=0.24

Figure SM 5.2.10.1. Photo of the TLC plate with all products obtained (eluente: petroleum ether (40-60 °C)-diethyl ether (7:3).

3.2. ^{1}H, ^{13}C NMR and IR spectra of the products
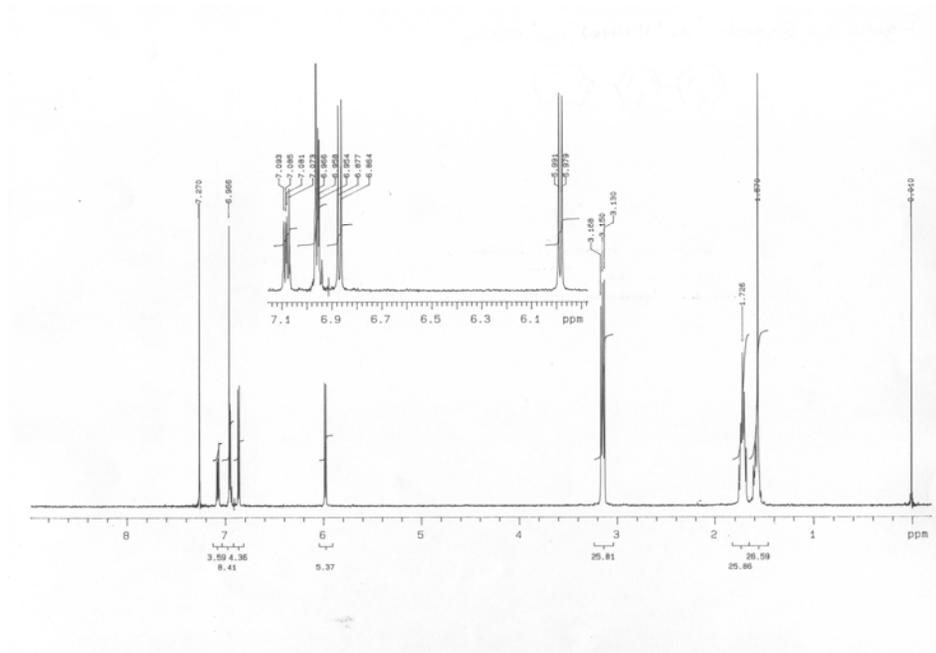
Figure SM 5.2.10.2. ^{1}H NMR Spectrum of 5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
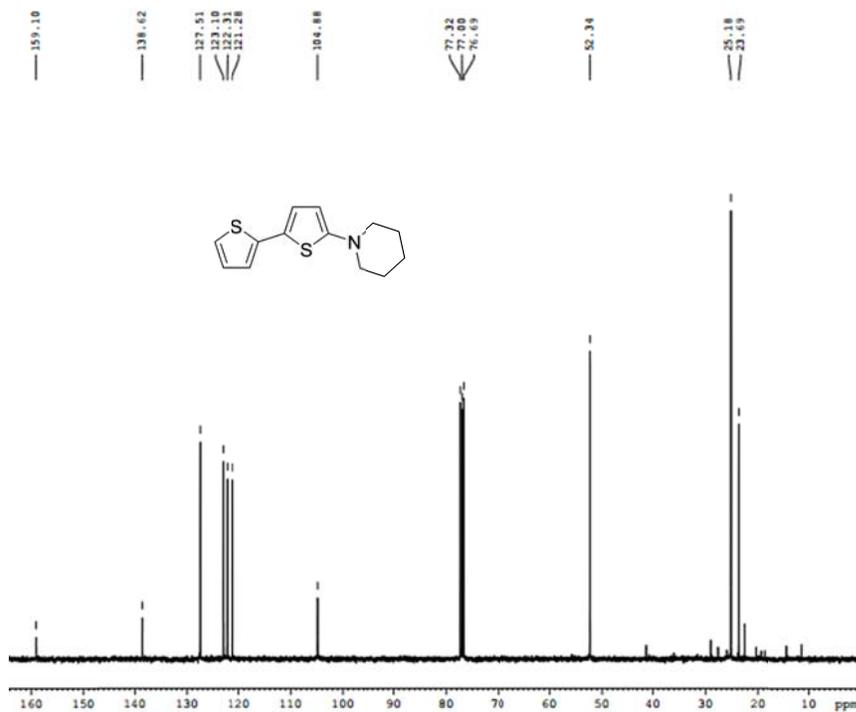
Figure SM 5.2.10.3. ^{13}C NMR Spectrum of 5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 100 MHz at 25 $^{\circ}$C.
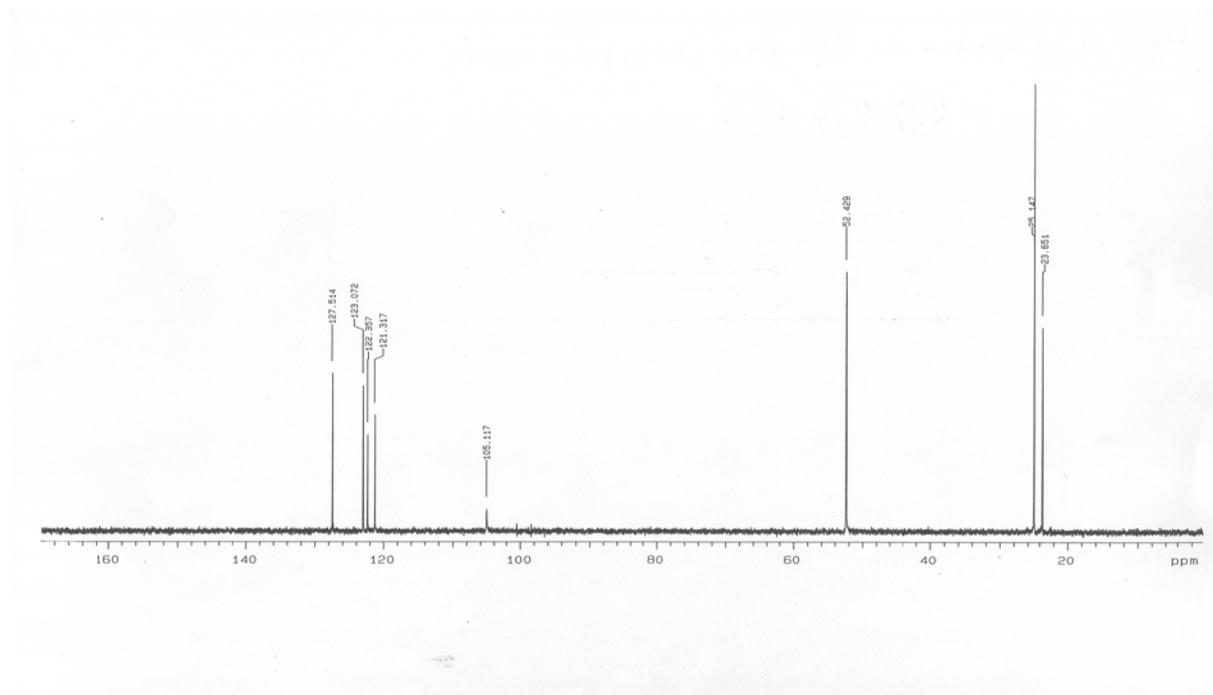
Figure SM 5.2.10.4. ^{13}C NMR (DEPT) Spectrum of 5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 $^{\circ}$C.
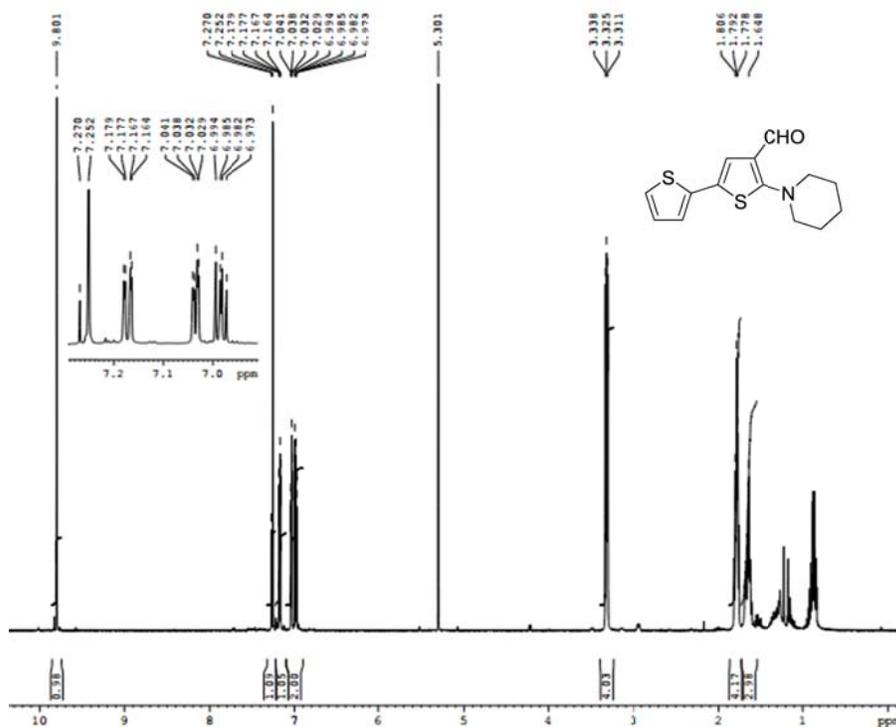
Figure SM 5.2.10.5. ^{1}H NMR Spectrum of 4-formyl-5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 400 MHz at 25 ^{o}C.
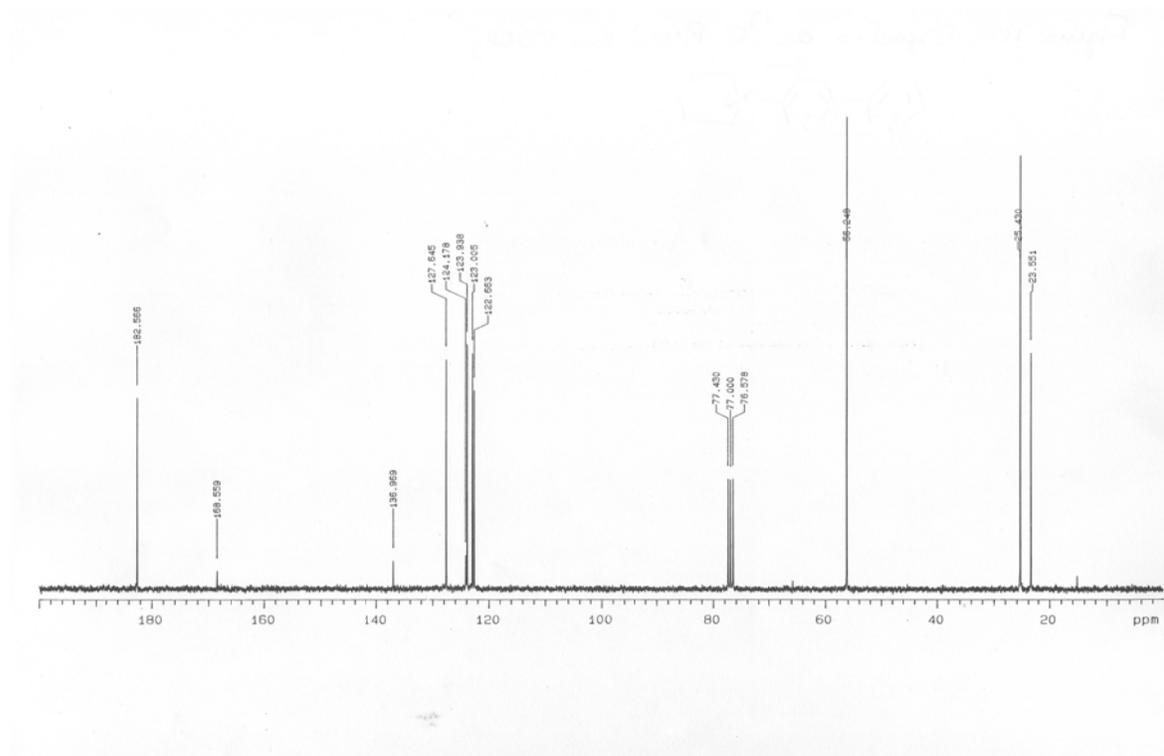
Figure SM 5.2.10.6. ^{13}C NMR Spectrum 4-formyl-5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 °C.
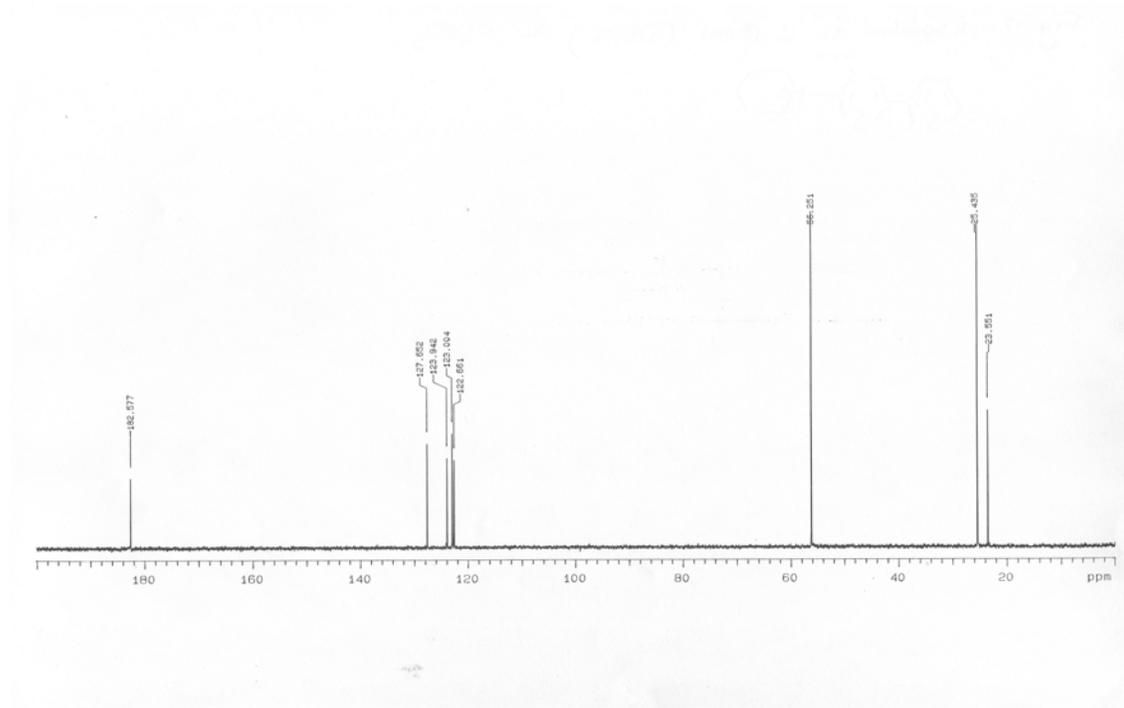
Figure SM 5.2.10.7. ^{13}C NMR (DEPT) Spectrum of 4-formyl-5-piperidino-2,2'- bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 °C.
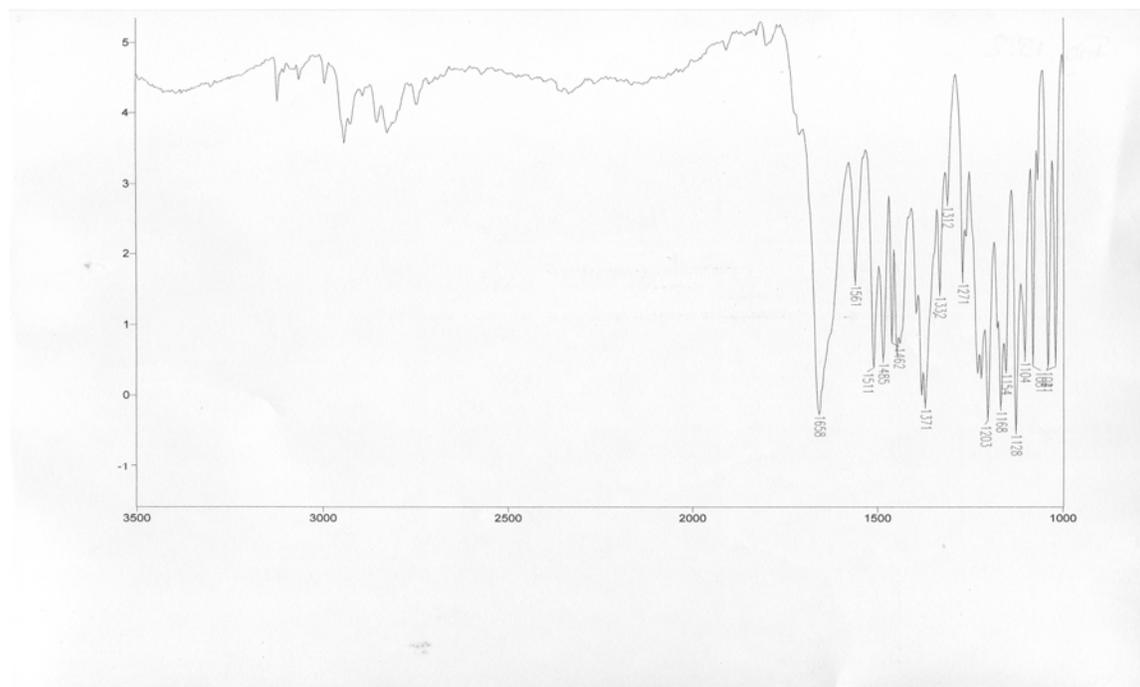
Figure SM 5.2.10.8. IR spectrum of 4-formyl-5-piperidino-2,2'-bithiophene in liquid film obtained using a Perkin Elmer FTIR-1600 spectrophotometer.
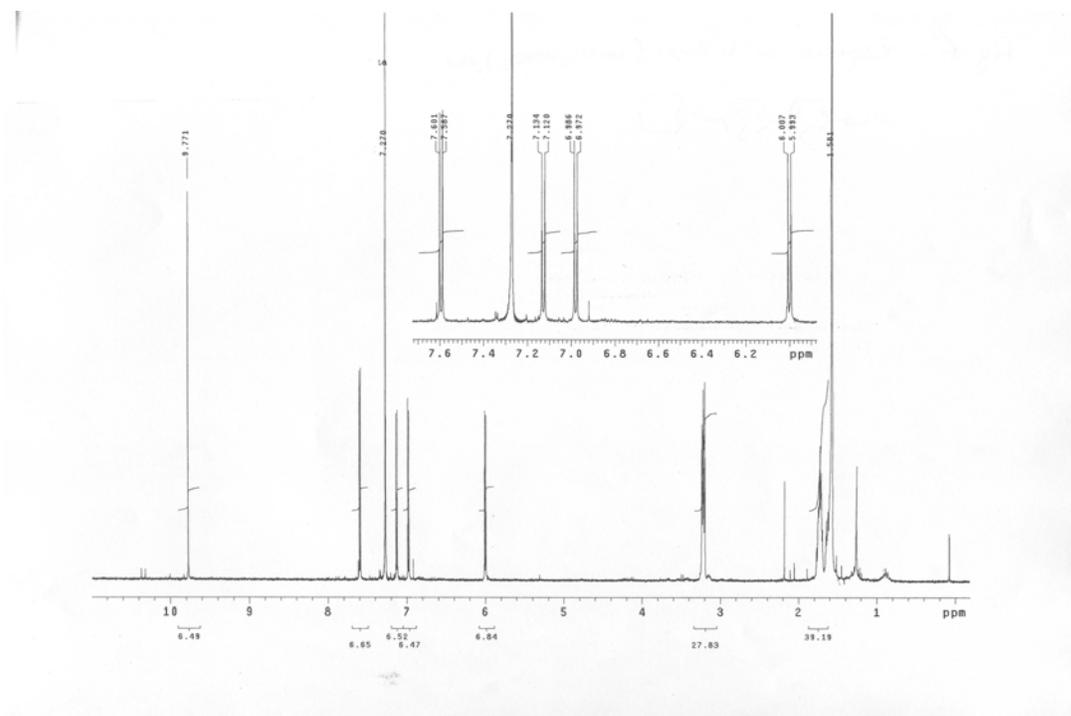
Figure SM 5.2.10.9. ^{1}H NMR Spectrum of 5′-formyl-5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
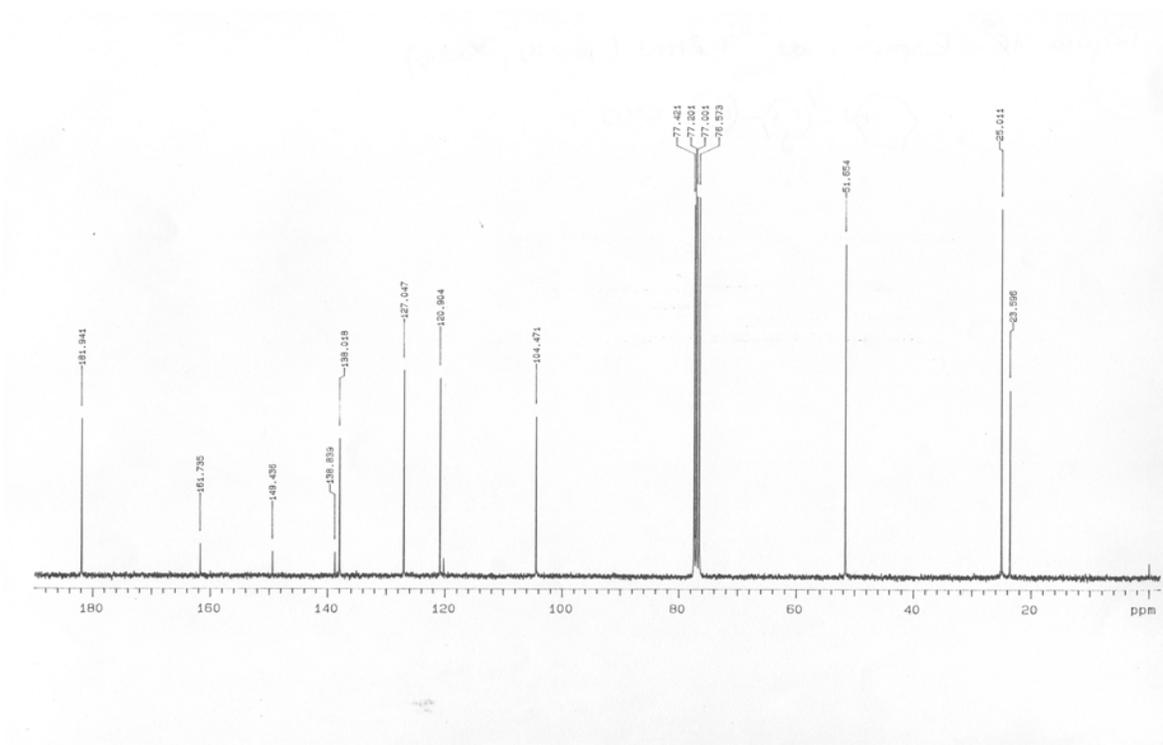
Figure SM 5.2.10.10. ^{13}C NMR Spectrum of 5´-formyl-5-piperidino-2,2'-bithiophene in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 ^{o}C.
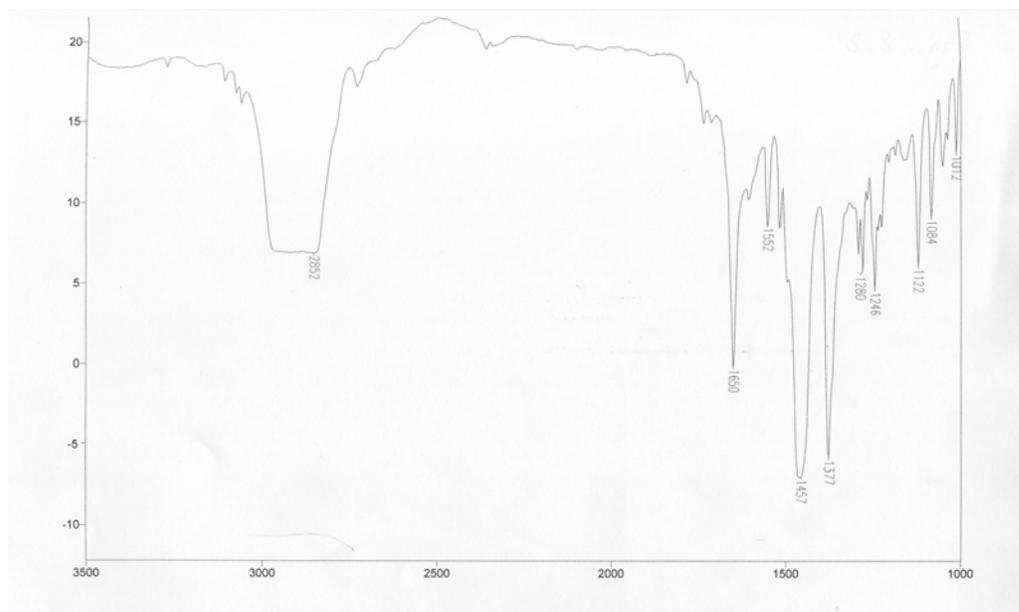
Figure SM 5.2.10.11. IR spectrum of 5´-formyl-5-piperidino-2,2'-bithiophene in nujol obtained using a Perkin Elmer FTIR-1600 spectrophotometer.

Synthesis of 3-bromosalicylaldehyde by *Ortho*-formylation of 2-bromophenol

Supplementary Material

Experimental Notes

Physical and spectroscopic data

Copies ^{1}H and ^{13}C NMR spectra

References

Introduction

Pharmacy students have conducted the experiment in the last year of their bachelor program over the last ten years, with several different phenols. The experiment aims at the preparation of 3-bromosalicylaldehyde (3-bromo-2-hydroxybenzaldehyde). 2-Bromophenol is reacted with the magnesium chloride ($MgCl_2$)/triethylamine (Et_3N) base system, affording the magnesium phenoxide, that reacts with the monomeric form of paraformaldehyde. As a solid, paraformaldehyde is a convenient starting material for the introduction of the formyl group. The monomeric form is liberated upon heating. TLC is easy to use for following the progress of the reaction. It is of pivotal importance that stirring is maintained.

This experiment is intended for students who previously have performed experiments in organic chemistry earlier and with some experience with organic synthesis experiments. The students should be familiar with ordinary glassware and heating techniques, but who also have gained experience with extractive work-up protocols and recrystallization techniques. We have demonstrated the experiment with success on students who have finished an introductory laboratory class in organic chemistry. It is an advantage if the students are familiar with electrophilic aromatic substation and the basic mechanisms and principles applied in electrophilic aromatic substation theory. The experiment itself is rather easy to execute and the product is easy to work with, since it is a solid. The time spent on reflux may be reduced if the molar equivalents of magnesium chloride, triethylamine and paraformaldehyde are increased. It is an advantage if magnesium chloride and paraformaldehyde are dried beforehand. Also, it is an advantage if tetrahydrofuran and triethylamine is distilled, as outlined in the notes below. Otherwise, commercially available dry reagents and solvents may also be employed. Magnesium chloride as beads provides the

highest yields and with the highest conversion rates. In the absence of these precautions, the reaction still works, but the yield is diminished.

Experimental Notes

1. Dry magnesium chloride (MgCl$_2$) as beads was used. Magnesium chloride was dried over phosphorus pentoxide under reduced pressure for 24 hours prior to use.

2. The reaction requires two equivalents of paraformaldehyde. An excess of this reagent results in a faster reaction, but results in the formation of some byproducts.

3. Paraformaldehyde was dried over phosphorus pentoxide under reduced pressure for 24 hours prior to use.

4. Triethylamine was distilled from calcium hydride and stored over 4Å molecular sieves prior to use. This step may be omitted if the triethylamine purchased is of high quality and without water.

5. Equimolar amounts of triethylamine and magnesium chloride is an advantage for the reaction.

6. 2-Bromophenol was used as received from vendor.

7. Tetrahydrofuran was distilled from sodium benzophenone ketyl prior to use. This step may be omitted if the solvent purchased is of high quality and without water.

8. The reaction mixture was heated with an oil bath.

9. Progress of the reaction was followed by thin layer chromatography using Merck silica gel 60 F$_{254}$ aluminum-backed plates, eluted with 30 % EtOAc in hexane visualized with a 254 nm UV lamp or by KMnO$_4$ stain. For the product R$_f$ = 0.45 and for 2-bromophenol R$_f$ = 0.40.

10. Analytical GLC was performed on a Varian GC 3300 instrument equipped with a 25 m SP2100 capillary column. This step may be omitted.

11. This product is in most cases sufficiently pure for further synthetic use. The product obtained is a pale yellow solid with mp 52-56 ^{o}C. However, the aldehyde obtained may be purified by recrystallization from n-hexane (a minimum amount, usually 3-5 mL) to give the aldehyde as colorless needles with melting point 55-56 ^{o}C (lit.[5] 52 ^{o}C).

12. Take 50 mg of the product to an NMR tube and add 600 uL of CDCl$_3$ to the NMR tube. Acquire the ^{1}H and ^{13}C NMR spectra. This sample may also be used for recording the FTIR spectrum.

Physical and spectroscopic data

3-Bromosalicylaldehyde (3-bromo-2-hydroxybenzaldehyde) exhibited the following physical and spectral properties: mp (uncorr.) 55-56 °C (lit.[3] 52 °C); ^{1}H NMR (300 MHz, CDCl$_3$) δ: 6.95 (t, J = 7.8 Hz), 7.55 (dd, J = 1.3Hz, 7.7Hz), 7.79 (dd, J = 1.3Hz, 7.7Hz), 9.87 (s, 1H), 11.61 (s, 1H); ^{13}C NMR (75 MHz, CDCl$_3$) δ: 111.26, 120.90, 121.42, 133.04, 140.07, 158.15, 196.12; FTIR spectrum (CHCl$_3$) cm^{-1}: 3152, 2851, 1672, 909.
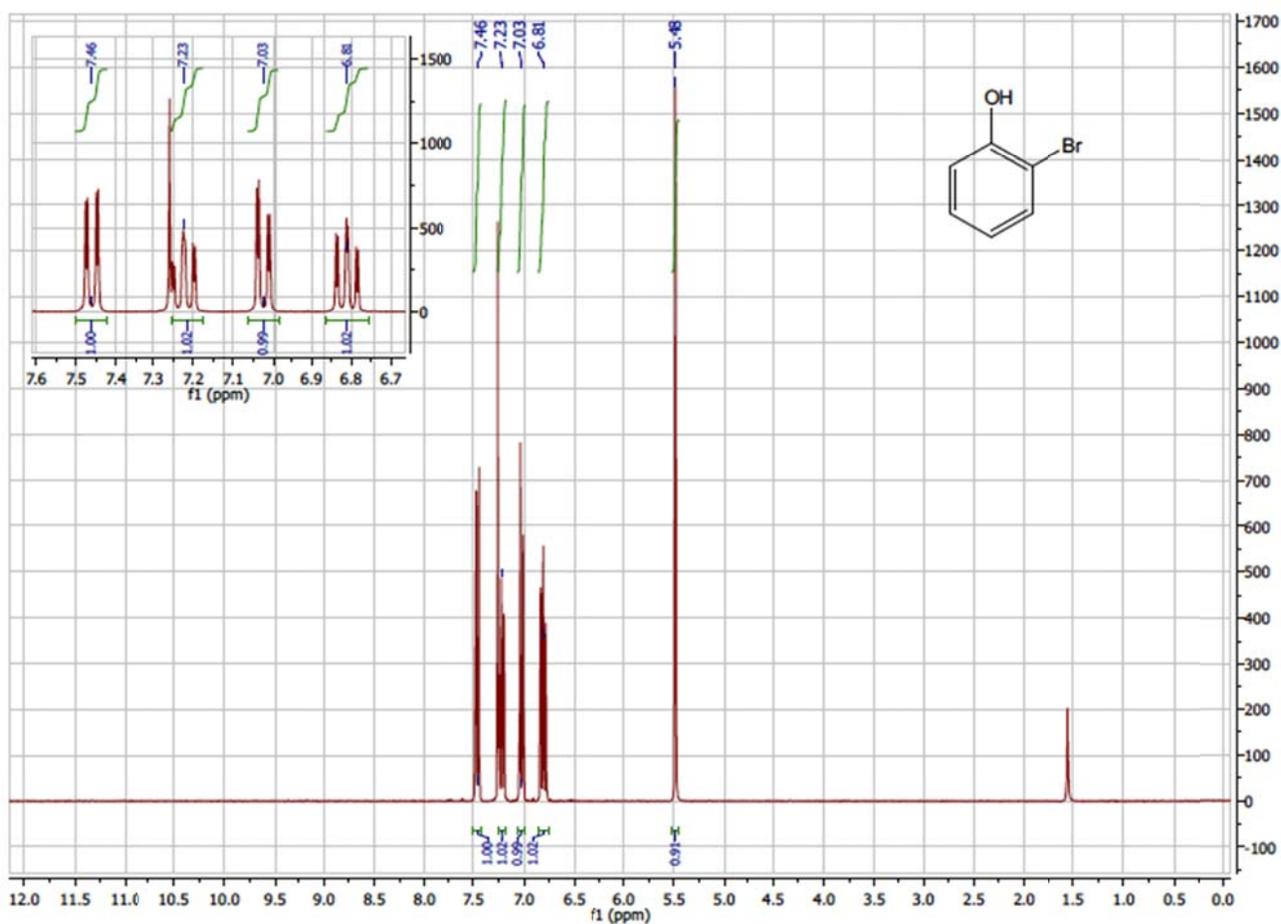
Figure SM 5.2.11.1 - ^{1}H NMR spectrum (300 MHz, CDCl$_3$) of 2-bromophenol. The peak at 1.52 ppm is residual water from CDCl$_3$.
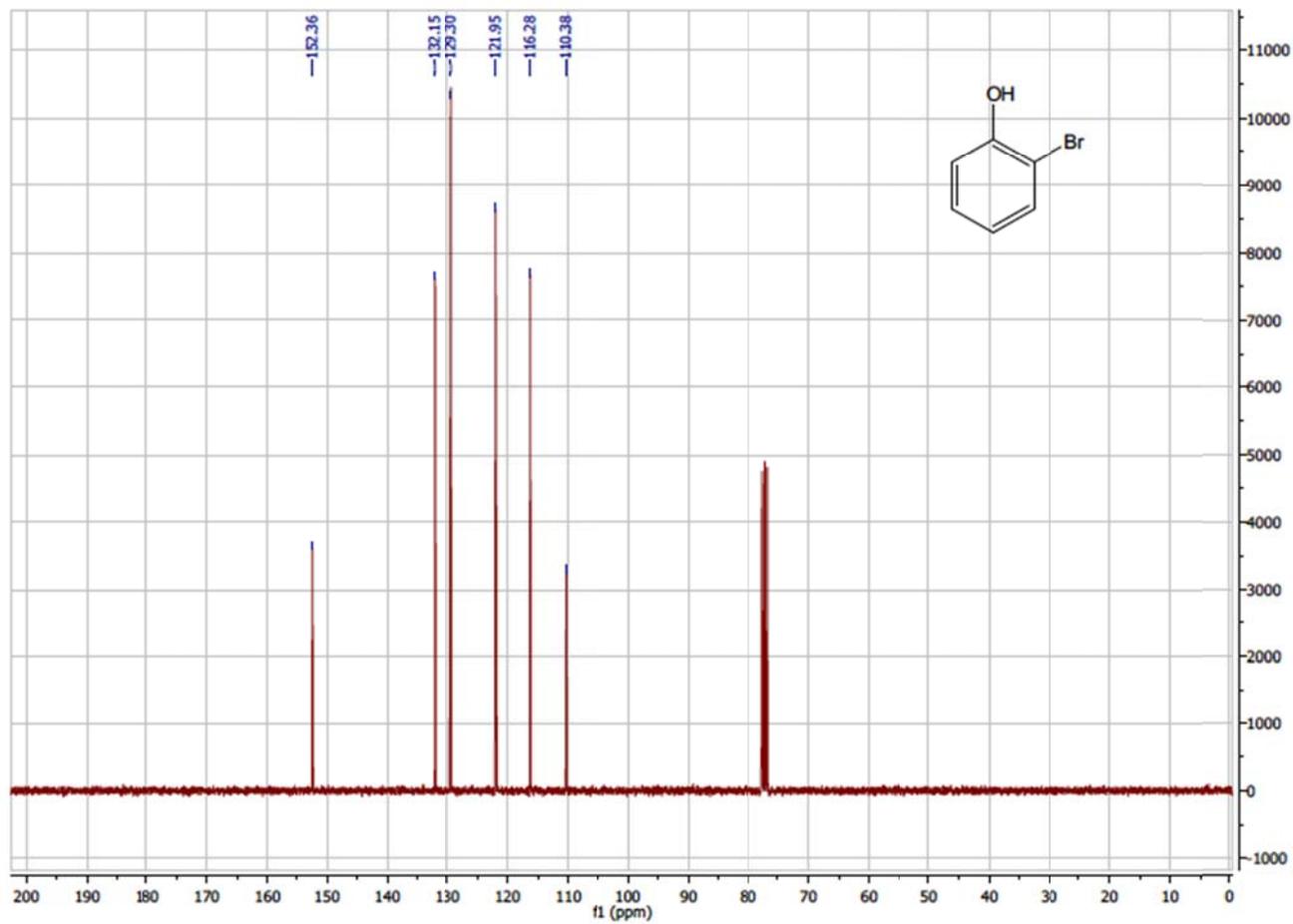
Figure SM 5.2.11.2 - ^{13}C NMR spectrum (75 MHz, CDCl$_3$) of 2-bromophenol.
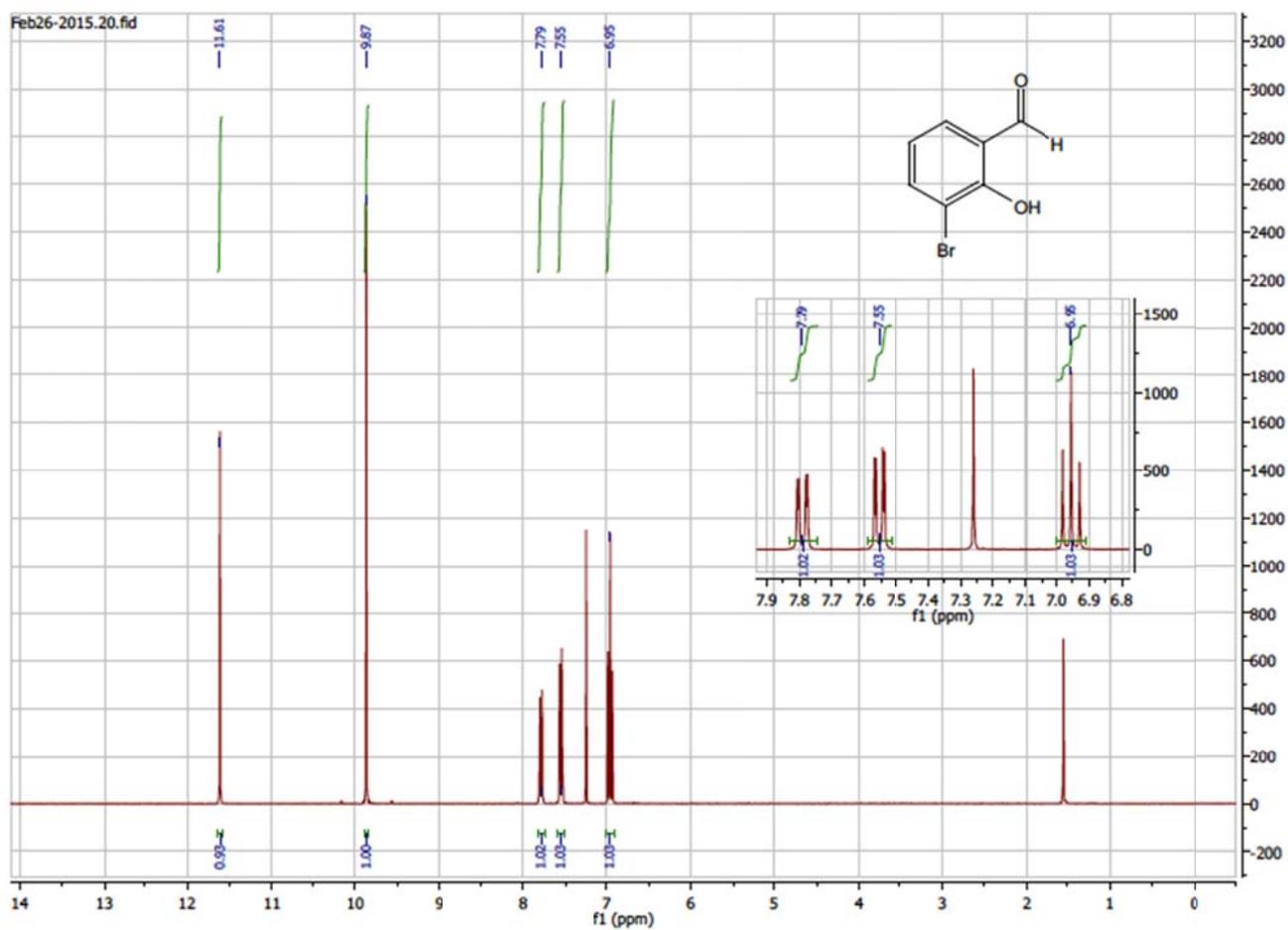
Figure SM 5.2.11.3 - ^{1}H NMR spectrum (300 MHz, CDCl$_3$3) of 3-bromosalicylaldehyde. The peak at 1.52 ppm is residual water from CDCl$_3$.
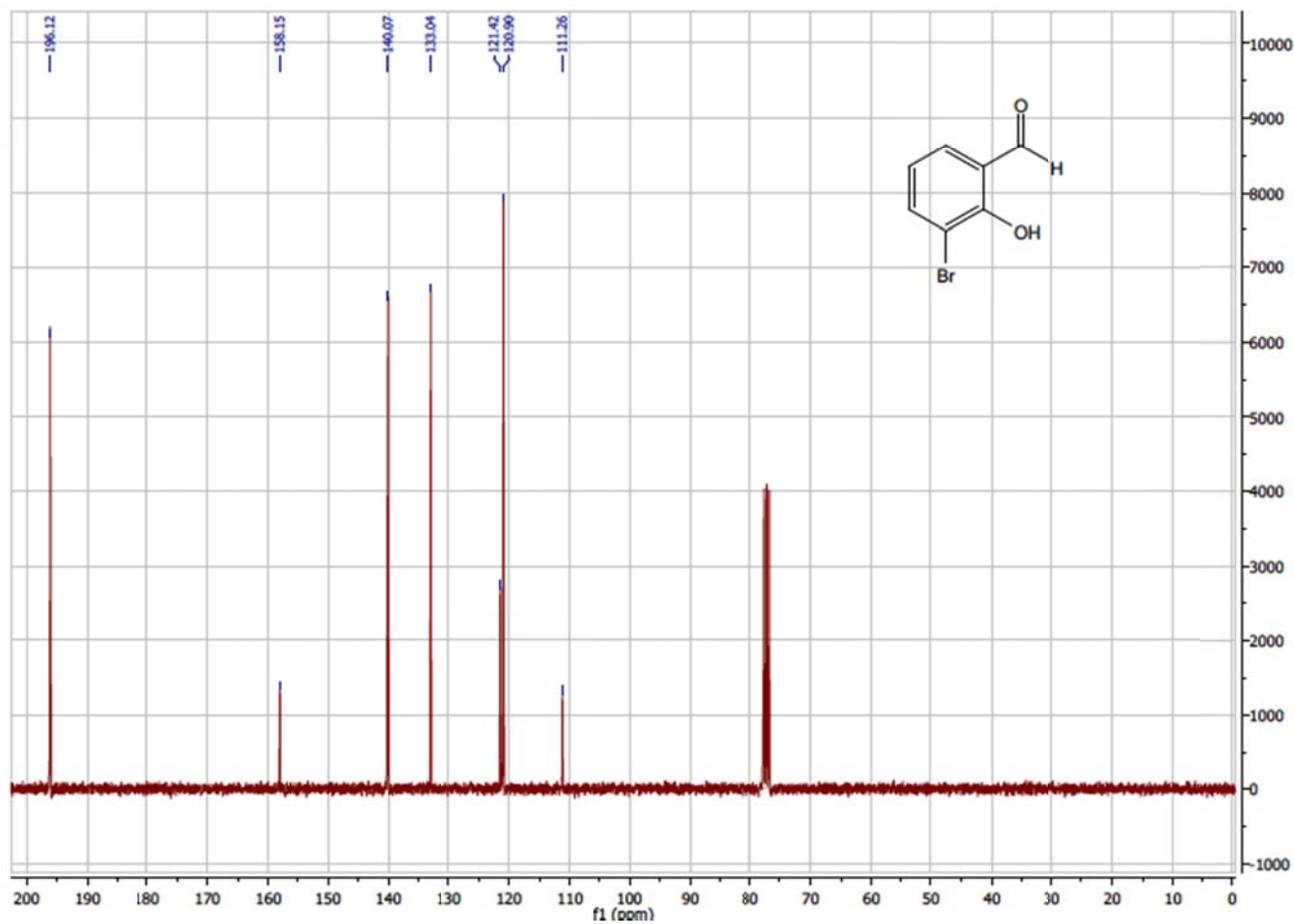
Figure SM 5.2.11.4 - ^{13}C NMR spectrum (75 MHz, CDCl$_3$) of 3-bromosalicylaldehyde.
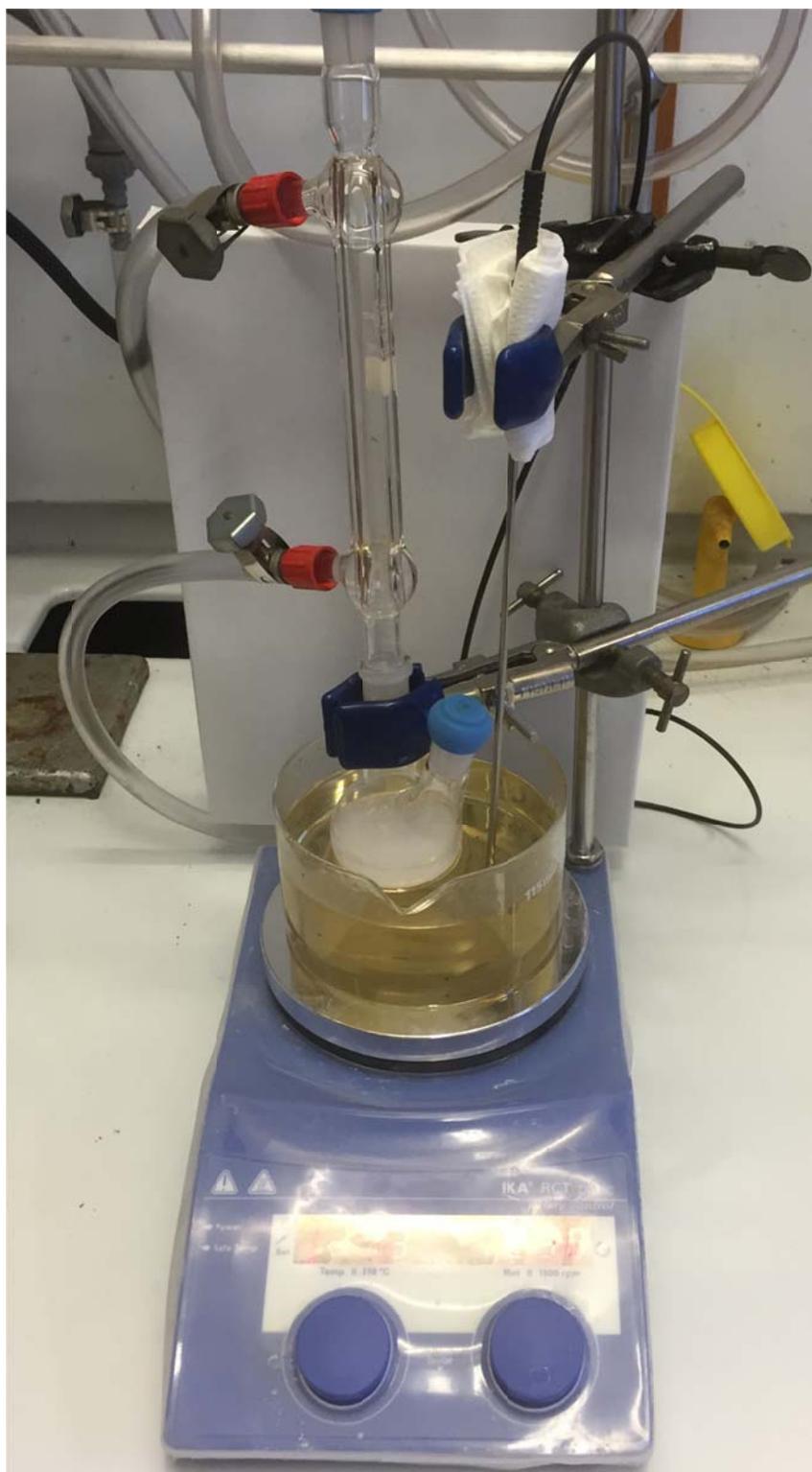
Figure SM 5.2.11.5 - Picture of the reaction mixture before 2-bromophenol is added.

Figure SM 5.2.11.6 - Picture of the reaction mixture after reflux for three hours. The color of the reaction mixture changes to pale yellow.

References:
The articles associated with this protocol are available free of charge, see below each reference.

1) N. U. Hofsløkken, L. Skattebøl, *Acta Chem. Scand.* 1999, **53**, 252.

See http://actachemscand.org/doi/10.3891/acta.chem.scand.53-0258

2) Hansen, T. V., Skattebøl, L., *Org. Synth.* 2005, **82**, 64.

See http://www.orgsyn.org/demo.aspx?prep=v82p0064

3) Hansen, T. V.; Skattebøl, L. *Discussion Addendum*:*Organic Syntheses* 2012*, **89***, 220*.
See http://www.orgsyn.org/demo.aspx?prep=v89p0220

Synthesis of indolo[3,2-*b*]quinolin-11-one by acid-catalysed intramolecular double cyclisation

Supplementary Material

1. Experiment Notes

The synthesis of indolo[3,2-*b*]quinolin-11-ones catalysed by polyphosphoric acid (PPA) has been routinely performed at Faculty of Pharmacy (University of Lisbon) in the past years by several students of different levels of study, namely 3rd year Bachelors in Pharmaceutical Sciences and MSc students of Pharmaceutical and Medicinal Chemistry courses. The reaction is successfully achieved with different substituents (Cl, CH_3, OCH_3, COOR) in positions 3, 7 or 8[1-5] but not with NO_2 or CF_3 substituents in C7.[4] Laboratory experience of the student performing the reaction is crucial for product purity and yield, but in all cases best results were obtained from starting material with no substitutions, as in the case of the proposed laboratory experiment. Even so, for better results, teacher must alert students for the following aspects:

o For completion of the reaction in the proposed time, starting material homogenization with PPA is crucial. Due to its high density/viscosity, this reagent should be put into the flask after the solid starting material. Altering the order of reagents addition is not advised since in this case the starting material will initially be floating on PPA. For the same reason, a strong stirring of the reaction mixture is advised. Reaction mixture viscosity will decrease on heating and color will be change to dark green (almost black);

o Due to the high boiling point of PPA, an air condenser attached to the flask is sufficient;

o After reaction, the mixture must be cooled to a warm temperature, so it can be safely handled but viscosity of the mixture has not yet increased too much. This will allow transferring the mixture into the ice water without leaving to much product in the reaction vessel. In any case, an additional small portion of cold water can be used to rinse the reaction vessel.

o Liquid-liquid extraction is another determinant step for product final yield and purity. To avoid formation of emulsion a gentle shaking of the two phases is advisable, but to ensure best extraction each shaking step must be done for 1-2 minutes. After 5-10 minutes, clear separation of both phases must be observed. First extraction is the most difficult to perform due to possible formation of emulsion and dark colors of both organic and aqueous layers (see photo).

This laboratory experiment can also be used to revise heterocycles nomenclature using as example the indoloquinoline isomeric structures in Figure SM 5.2.12.1.

Figure SM 5.2.12.1 – Structures of isomeric indoloquinoline nucleus with an example to help revising the IUPAC nomenclature of fused heteroaromatic rings.

2. Photos of experiment

Erlenmeyer with starting material
and PPA before microwave irradiation

Microwave set-up

Reaction mixture in an ice bath after neutralization

Liquid-liquid extraction. From left to right: first extraction, second extraction and organic layers wash with water.
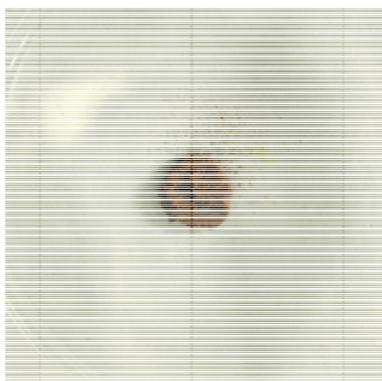
Product before crystallization

Product after crystallization

3. Results obtained by students of different levels of study

Table SM 5.2.12.1. – Percentage yields of isolated products: 2-(2-Bromoacetamido)benzoic acid (entries 1-4); 2-[2-(phenylamino)acetamido]benzoic acid (entries 5-8) and 5*H*-indolo[3,2-*b*]quinolin-11(10*H*)-one after purification by recrystallization (entries 9-12).

Entry	Level of study	Yield, (%)
1	MSc	75
2	MSc	72
3	MSc	72
4	3rd year BSc	70
5	MSc	65
6	MSc	53
7	MSc	55
8	3rd year BSc	52
9	MSc	57
10	MSc	37
11	MSc	51
12	3rd year BSc	50

4. NMR spectra
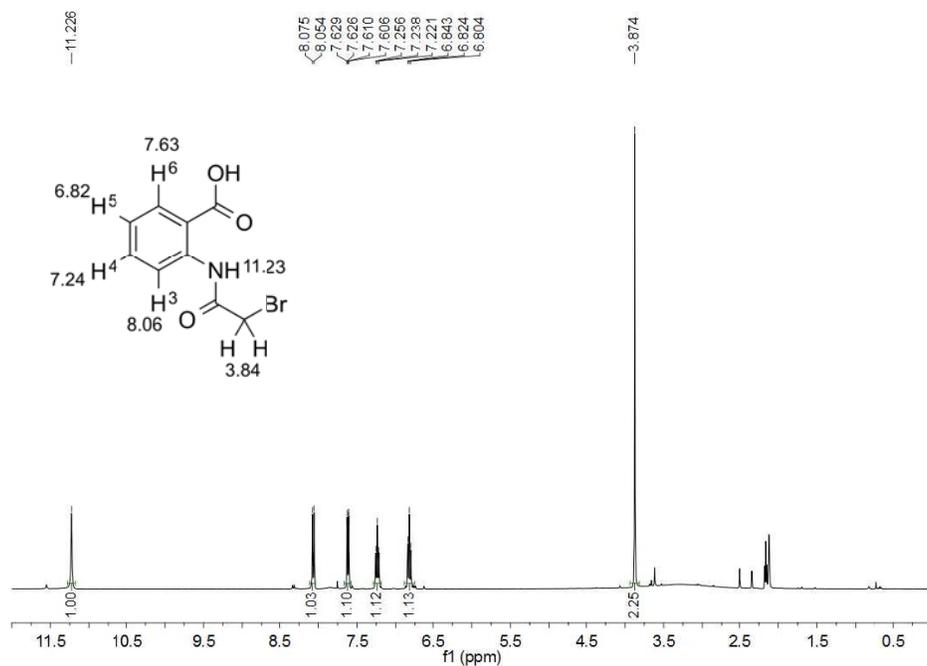
Figure SM 5.2.12.2. ^{1}H NMR spectra of 2-(2-Bromoacetamido)benzoic acid in DMSO-d$_6$.
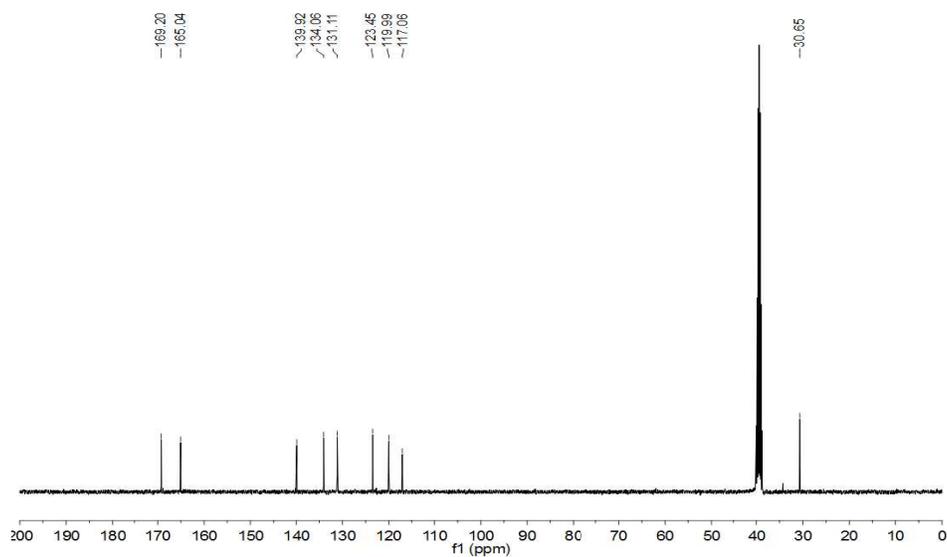
Figure SM 5.2.12.3. ^{13}C NMR spectra of 2-(2-Bromoacetamido)benzoic acid in DMSO-d$_6$.
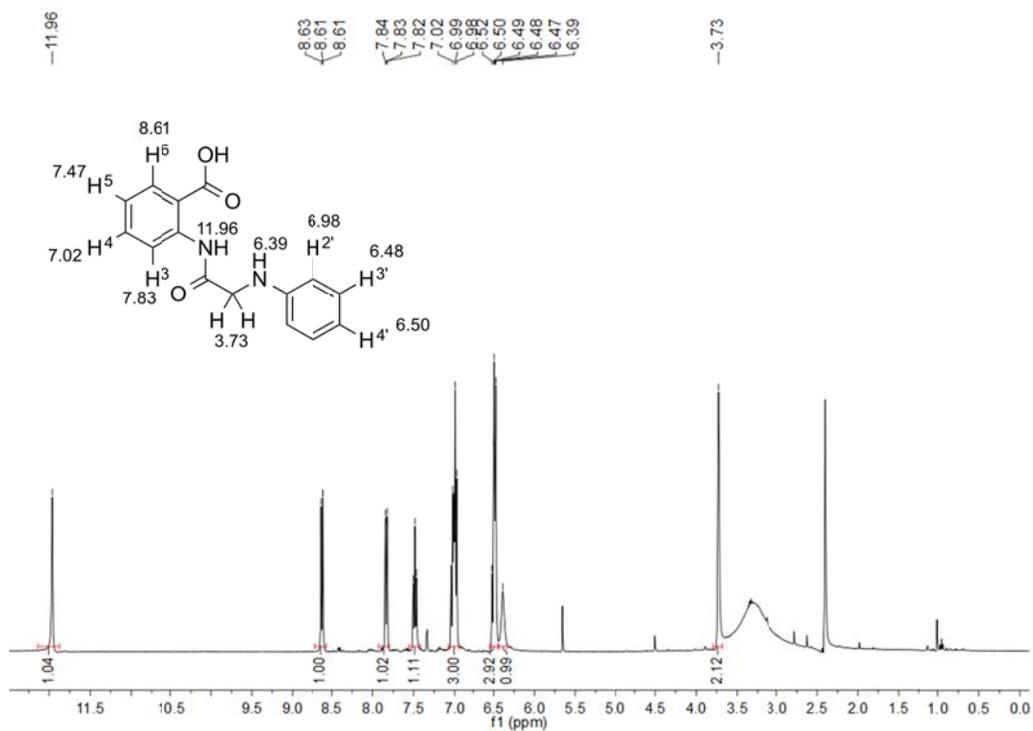
Figure SM 5.2.12.4. ^{1}H spectra of **2-[2-(phenylamino)acetamido]benzoic acid** in DMSO-d$_6$.
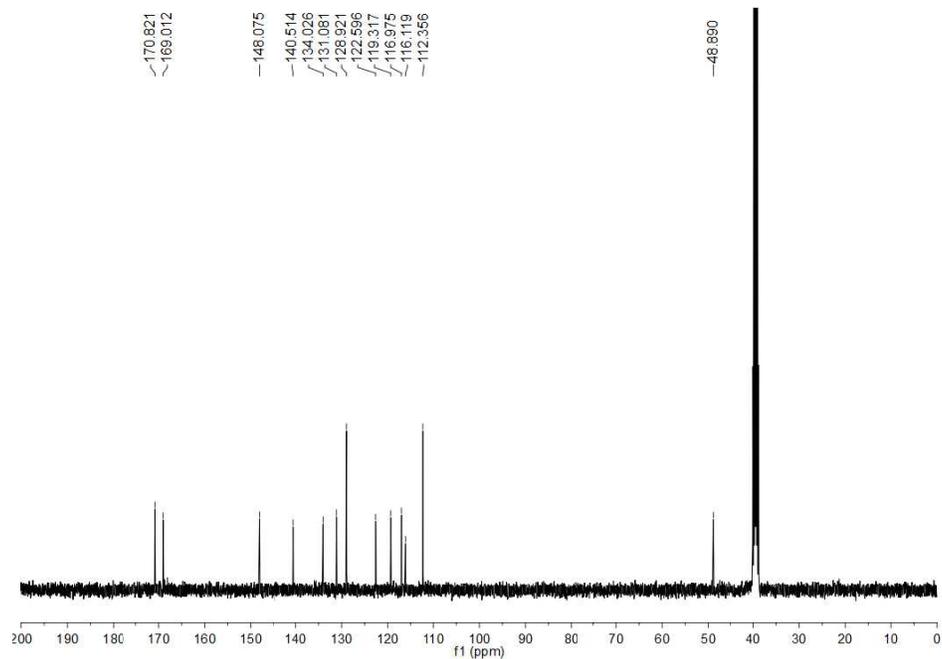
Figure SM 5.2.12.5. ^{13}C NMR spectra of **2-[2-(phenylamino)acetamido]benzoic acid** in DMSO-d$_6$.
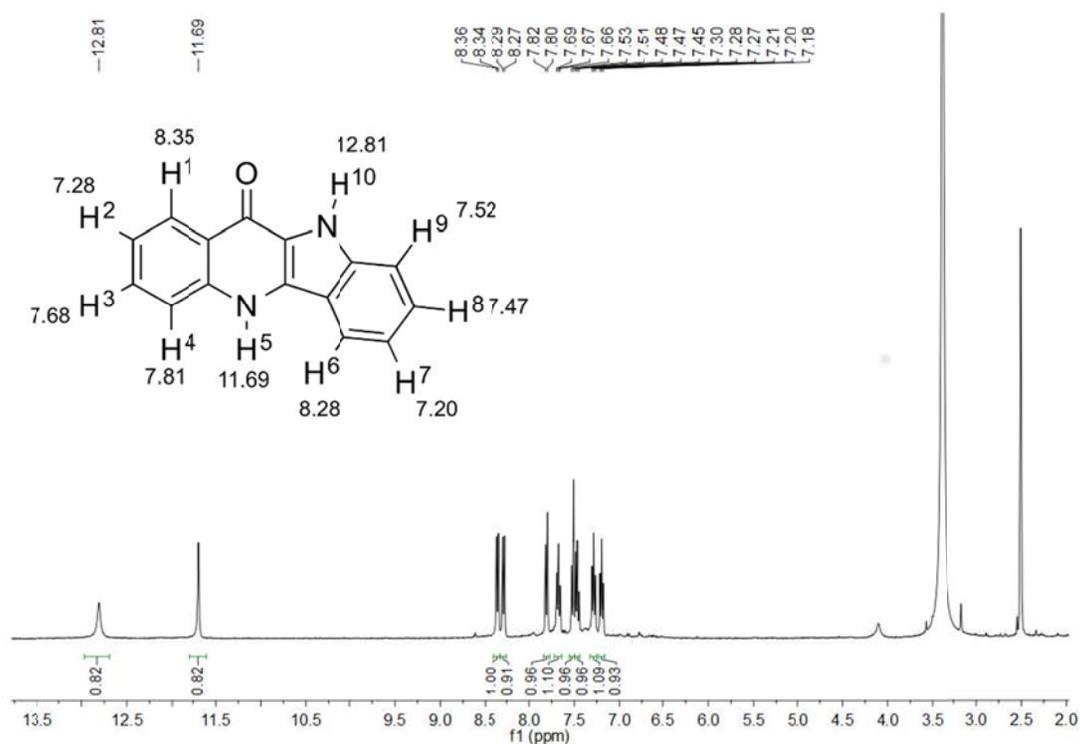
Figure SM 5.2.12.6 – ¹H NMR spectrum of 5*H*-indolo[3,2-*b*]quinolin-11(10*H*)-one after crystallization in DMSO-d₆.
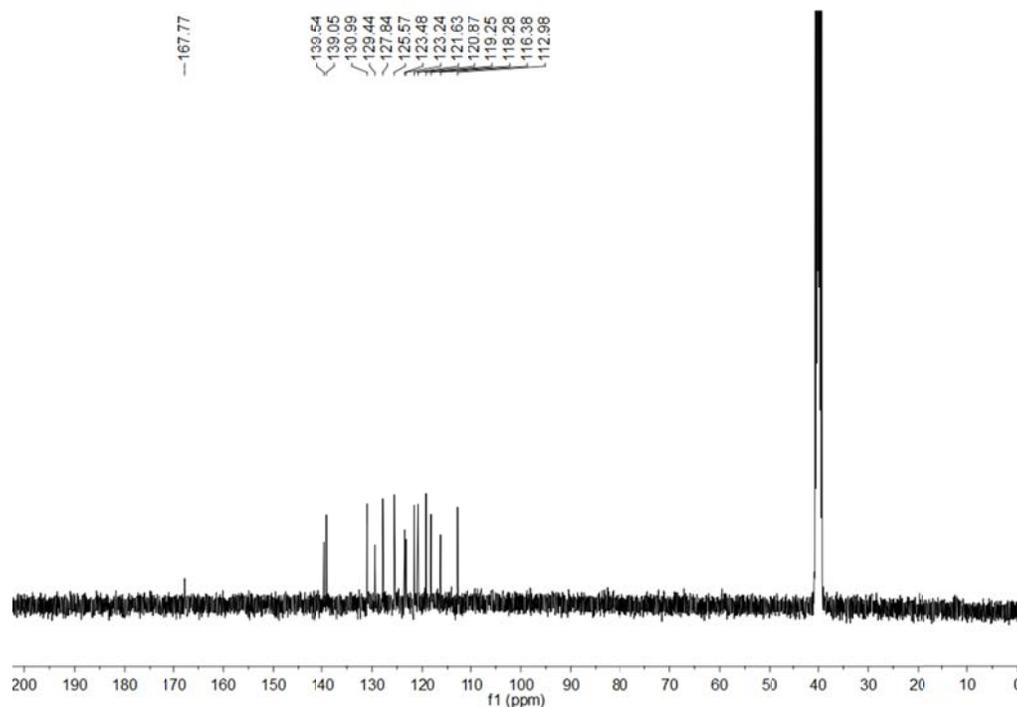
Figure SM 5.2.12.7 – ¹³C NMR spectrum of 5*H*-indolo[3,2-*b*]quinolin-11(10*H*)-one after crystallization in DMSO-d₆.
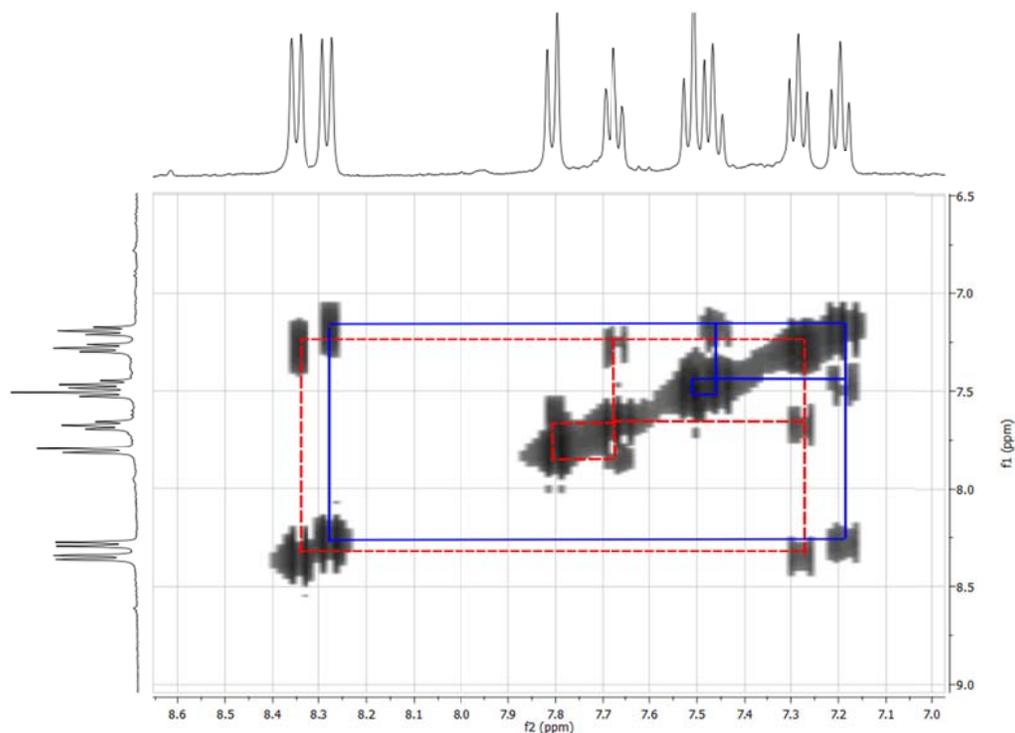
Figure SM 5.2.12.8 – 2D ^{1}H-^{1}H NMR spectrum of pure 5*H*-indolo[3,2-*b*]quinolin-11 (10*H*)-one in DMSO-d$_6$.

5. Proposed mechanism of double cyclisation
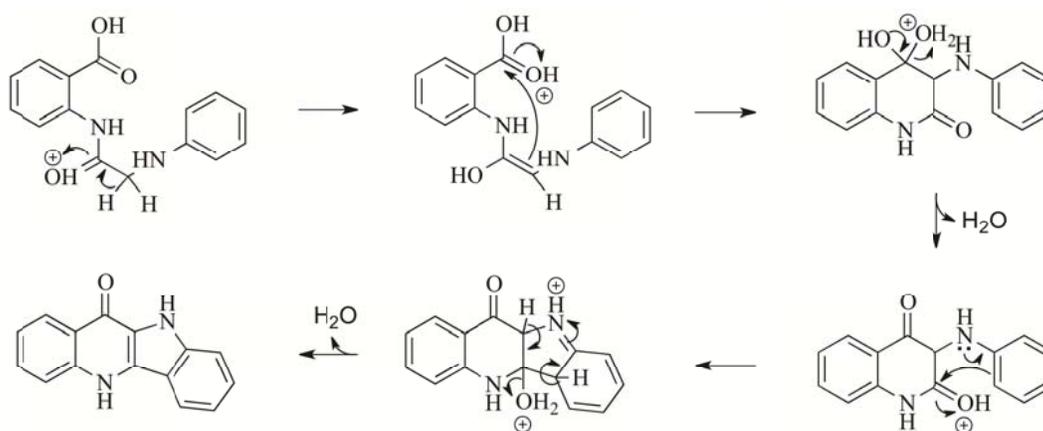
Scheme SM 5.2.12.1. Acid-catalysed bi-ciclisation of 2-[2-(phenylamino)acetamino]benzoic acid.[1,4]

7. References

[1] Lavrado, J., PhD thesis, Faculty of Pharmacy, University of Lisbon, Lisbon, **2010**. (http://repositorio.ul.pt/handle/10451/11987)

[2] Lavrado, J.; Gani, K.; Nobre, P. A.; Santos, S. A.; Figueiredo, P.; Lopes, D.; Rosario, V.; Gut, J.; Rosenthal, P. J.; Moreira, R.; Paulo, A. *Bioorg. Med. Chem. Lett.* **2010,** 20, 5634-5637.

[3] Lavrado, J.; Cabal, G. G.; Prudencio, M.; Mota, M. M.; Gut, J.; Rosenthal, P. J.; Diaz, C.; Guedes, R. C.; Dos Santos, D. J.; Bichenkova, E.; Douglas, K. T.; Moreira, R.; Paulo, A. *J. Med. Chem.* **2011,** 54, 734–750.

[4] Lavrado, J.; Borralho, P. M.; Ohnmacht, S. A.; Castro, R. E.; Rodrigues, C. M.; Moreira, R.; Santos, D. J.; Neidle, S.; Paulo, A. *ChemMedChem* **2013,** 8, 1648-1661.

[5] Paulo, A.; Figueiras, M.; Machado, M.; Charneira, C.; Lavrado, J.; Santos, S. A.; Lopes, D.; Gut, J.; Rosenthal, P. J.; Nogueira, F.; Moreira, R. *J. Med. Chem.* **2014,** 57, 3295-3313.

Mild and fast Friedel-Crafts acylation over zeolites
Supplementary Material

1. Experiment Notes

The purpose of this experiment is the use of commercial zeolites as heterogeneous catalysts for Friedel-Crafts acylation of thiophene with acetic anhydride.

The advantage of using zeolites as an alternative to other classic catalysts, such as $AlCl_3$ is the possibility to work under mild reaction conditions along with easy recovery and possible reutilization of the catalysts. It can thus be faced as a step forward towards greener chemistry procedures.

Zeolites are crystalline aluminosilicates, exhibiting a regular pore structure with dimensions comparable those of organic molecules, with pore openings ranging from 3 to 10 Å, according to the type of structure. All zeolite structures are designated by a 3 letters code, attribute by IZA (International Zeolite Association). Table SM 5.2.13.1 displays the main properties of some of the most common commercial zeolites.

Table SM 5.2.13.1 – Characteristics of some of the most common commercial zeolites.

Common designation	IZA code	Type of structure	Pore opening dimensions (Å)	Typical Si/Al ratio[a]	Typical surface area (m^2 g^{-1})[a]
Beta	BEA	Tridimensional structure with channels	6.6 × 6.7 and 5.6 × 5.6	12.5	680
Faujasite or Y	FAU	Tridimensional structure with channels and cages.	Supercages 13 diameter access through 7.4 × 7.4 openings.	2.5	730
ZSM-5	MFI	Tridimensional structure with channels	5.2 × 5.8 and 5.4 × 5.6	15	405
Mordenite	MOR	Monodimensional structure with channels	6.5 × 7.0 and 2.6 × 5.7 (the last one is not accessible by organic molecules)	10	500

[a] Data supplied by Zeolyst International Inc.

Zeolites are accessible and cheap materials. They should preferably be acquired in the protonic form (H-zeolite) to be easily used in the context of the present work. In case of acquiring materials in the ammonium form (NH$_4$-Zeolite), they should be submitted to a thermal treatment under air flow at 500 ºC to convert them into the H-zeolite. If several zeolite structures are available, students can use distinct structures such as monodimensional MOR and tridimensional MFI and compare the effect of the zeolite structure on the catalytic results.

As regards the aromatic substrate, although thiophene is used in the example shown, other substrates can also be utilised. In any case, the experimental procedure is the same but the amounts of each reactant should be adapted in order to preserve the substrate to acetic anhydride ratio (1:5 molar ratio).

The apparatus used to perform the reaction can be adapted according to the material available in the laboratory. For instance, if the heating plate has no temperature control, a simple thermometer can be

adapted and manual temperature adjustments can be made. The separation of the reaction mixture is attained by using a 25 mm EMD Millipore Swinnex Filter Holder with Millipore Durapore membrane filter (0.45 μm pore size) adapted to a syringe. However, other separation methods can be used, including filtration with a filter paper or silica porous plates adapted to vacuum filtration.

The GC capillary column that was used is a HP-5-MS from Agilent. Compatible alternatives are: CP-Sil 8 (Varian); BP-5 (SGE); ZB-5-MS (Phenomex) or SPB-5 (Supelco).

Quantification of the reaction mixture by GC can be more rigorously calculated using the internal standard method in order to account for differences in the detector response. We have successfully used dodecane or hexamethylbenzene as internal standards. In this method, a calibration factor (Z) was determined from at least 5 consecutive chromatograms of a given calibration solution, containing both the reaction product and the internal standard, according to Eq.1:

$$Z = \left(\frac{A_p}{A_{is}}\right) \cdot \left(\frac{W_{is}}{W_p}\right)$$

Eq.1

where A_{is} and A_p are the integrated areas of the peaks corresponding to internal standard and to product, respectively, and W_{is} and W_p are their known weights in the calibration solution.

The yields of products (in the reaction mixture) were determined according to Eq. 2:

$$Yield = \left(\frac{1}{Z}\right) \cdot \left(\frac{A'_p}{A'_{is}}\right) \cdot \left(\frac{W_{is}}{W_r}\right) \cdot \left(\frac{MW_r}{MW_p}\right) \cdot 100$$

Eq.2

where Z is the calibration factor determined as mentioned above, A'_{is} and A'_{ip} are the average areas of the peaks corresponding to the internal standard and product, MW_r and MW_p the molecular weights of reactant and product, and W_{is} and W_r are the weights of internal standard and reactant, respectively.
In Eq. 2 thiophene was the only reactant considered and 2-acetylthiophene the only product, since acetic anhydride was in great excess and acetic acid is a secondary product, formed in 1:1 proportion, being irrelevant for the calculation of the yield.

This experiment was already implemented at an advanced level course, in a class of master degree students, but it can also be carried out at an intermediate level, with appropriate adjustments. It is recommended that only one group of two or three students perform the experiment at a time. A 3 h laboratory session is more than enough to perform the work envisaged, comprising: a) weighting of reactants and catalyst and set up of the experimental apparatus for the reaction (about 30 min);

b) withdrawal of aliquots of the reaction mixture (1 h) and c) analysis of the reaction mixture by GC (1.5 h). To save time, tasks b) and c) can run in parallel. Post laboratory students' work is anticipated in order to fulfil a thorough analysis of the spectra, as well as a complete kinetic treatment. The inclusion of kinetic results should be emphasized because it is very uncommon in Organic Chemistry courses of all levels. With this approach we aim to make students go beyond strict Organic Chemistry onto Physical Organic Chemistry and to stimulate the integration of knowledge from different scientific areas.

All suggested materials and techniques should be available to advanced Organic Chemistry students which are expected to work at a medium difficulty level.

2. Figures

2.1 Scanning electron microscopy (SEM) of zeolites structures
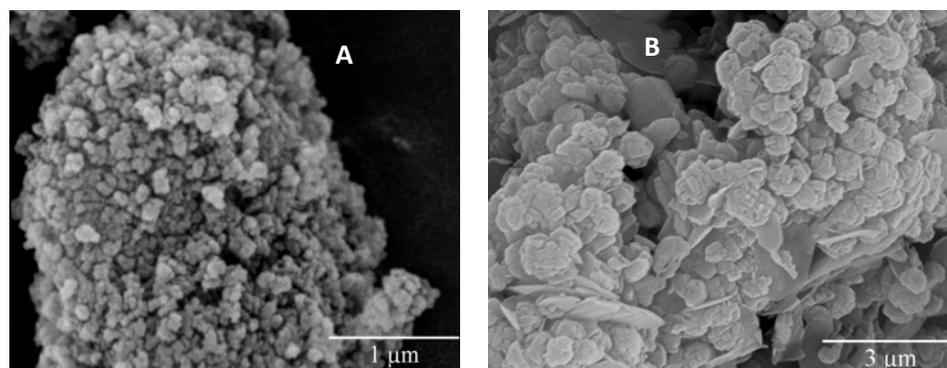
Figure SM 5.2.13.1 - SEM images for BEA (A) and MOR (B) zeolite structures.

2.2 Experimental setup

Cooling water out

Cooling water in

Rubber septum cap

Two-necked flask (to allow aliquots' collection)

Oil bath

Magnetic stir bar

Magnetic hot plate
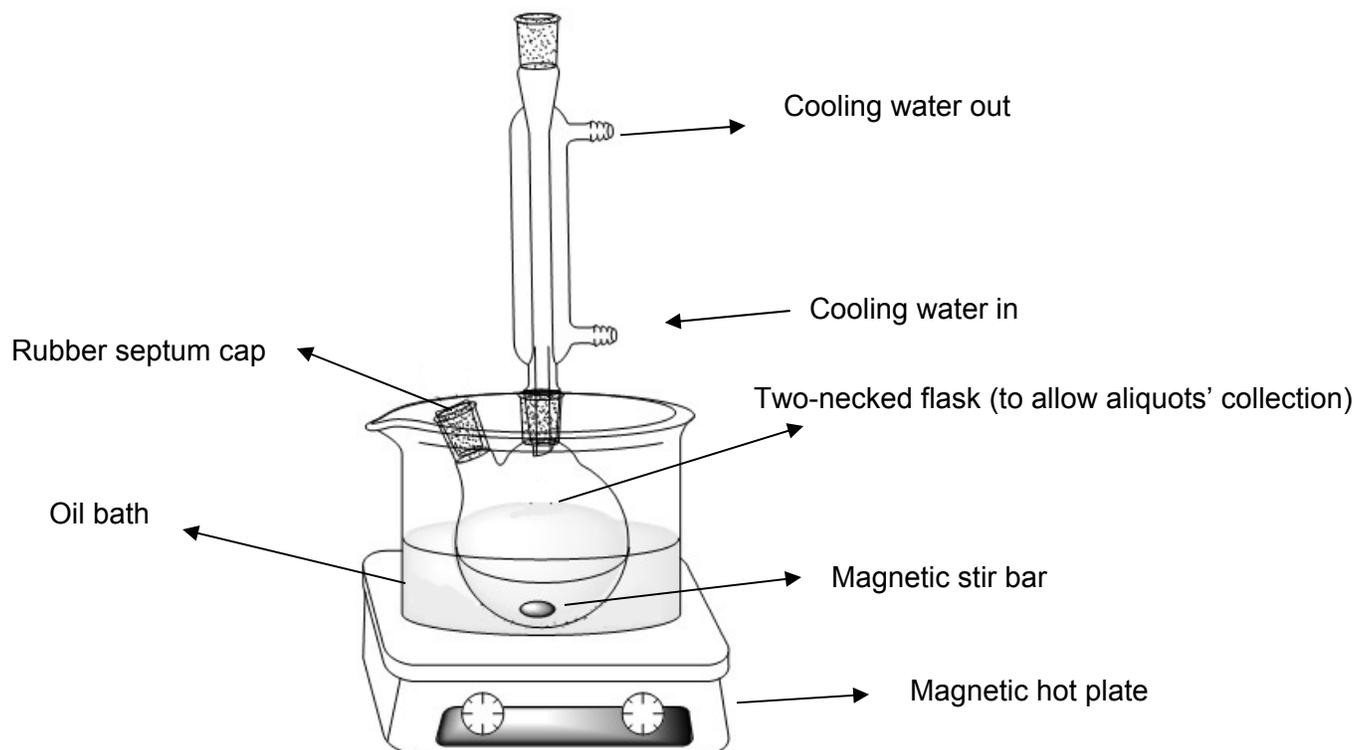
Figure SM 5.2.13.2 – Experimental setup for the Friedel-Crafts acylation.

2.3 GC chromatograms and MS spectra

Figures SM 5.2.13.3-5 show typical GC chromatograms of aliquots collected at different reactions times. The peaks identification is done in the chromatogram.

Note: acetone was used as solvent to dilute the samples.
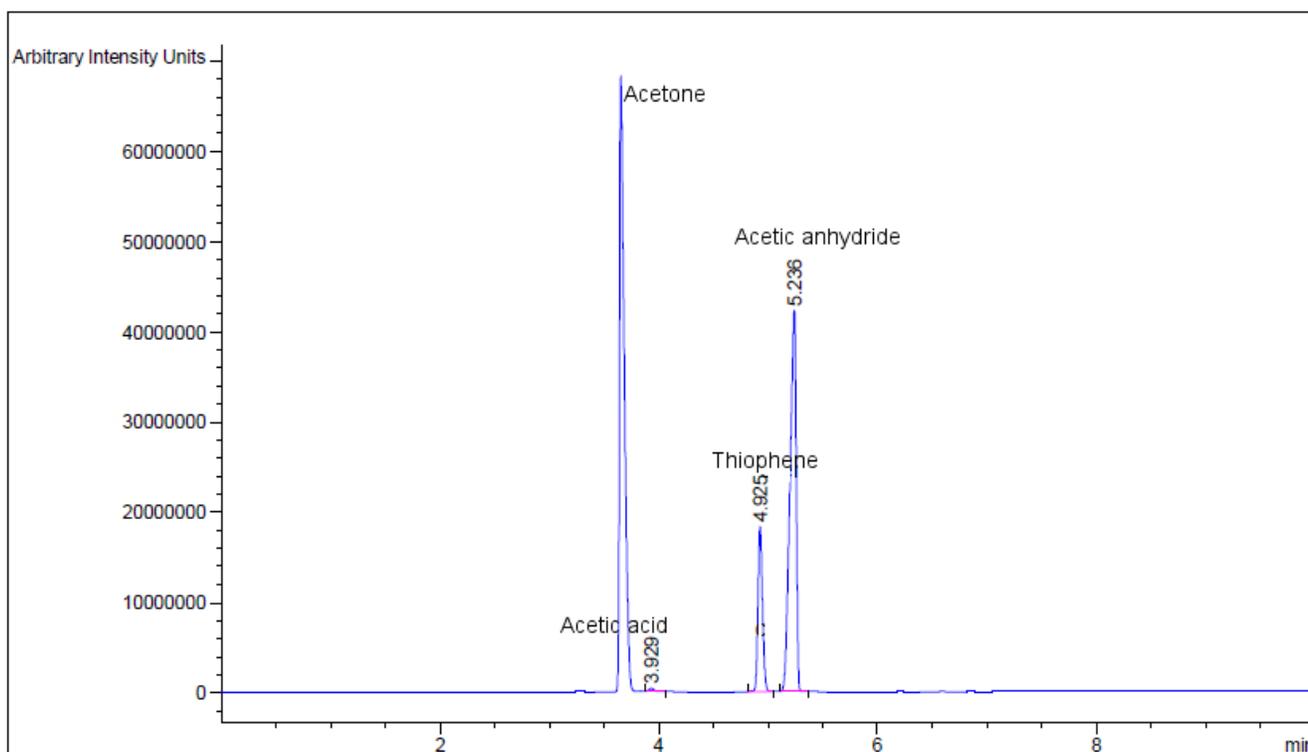
Figure SM 5.2.13.3 - Chromatogram for the reaction of thiophene over HBEA Zeolite at 0 min.
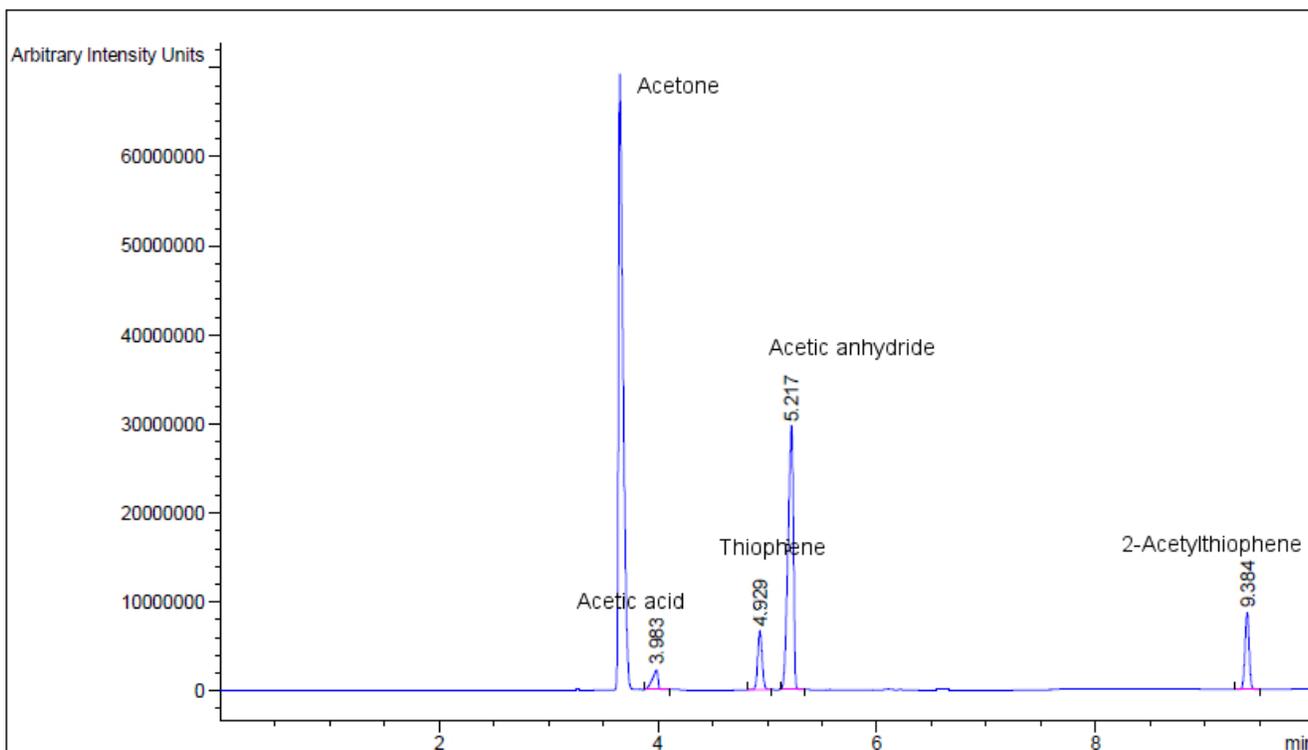
Figure SM 5.2.13.4 - Chromatogram for the reaction of thiophene over HBEA Zeolite at 2 min.
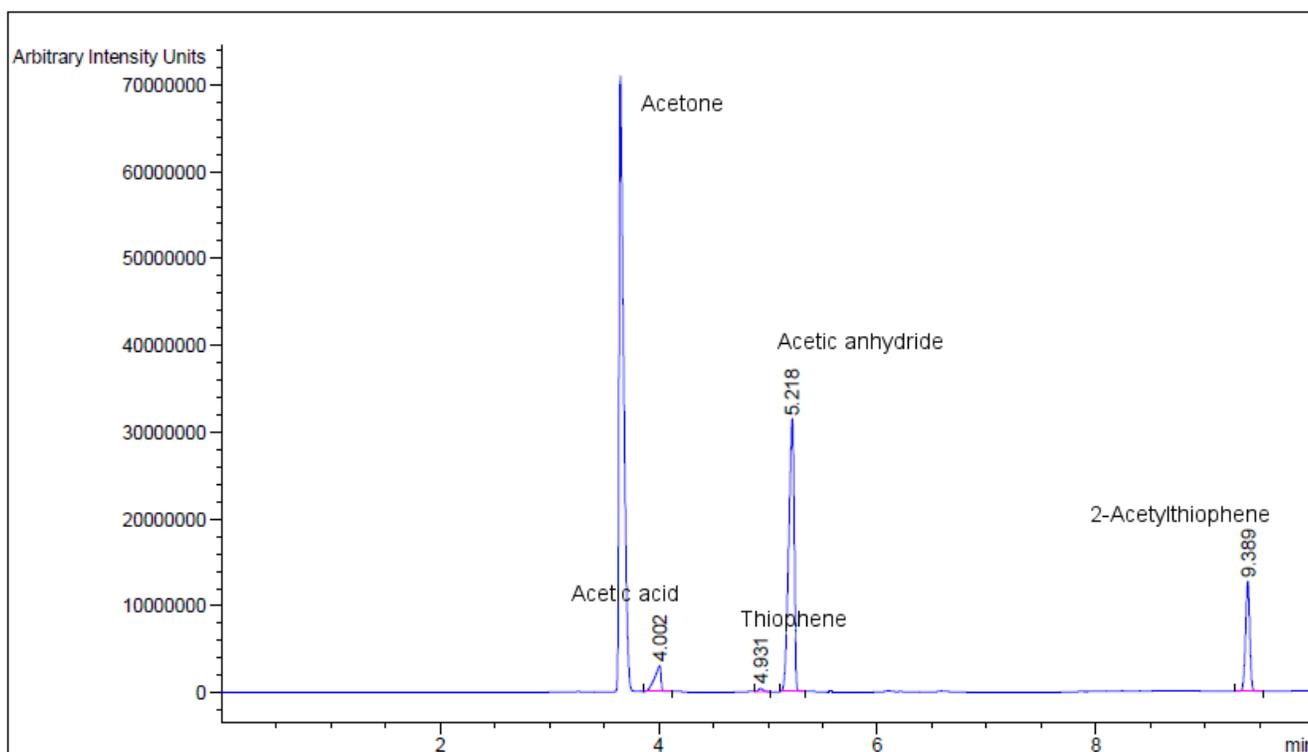
Figure SM 5.2.13.5 - Chromatogram for the reaction of thiophene over HBEA Zeolite at 30 min.

Figures SM 5.2.13.6-7 show the mass spectra of GC peaks and their correspondent identification by library software.
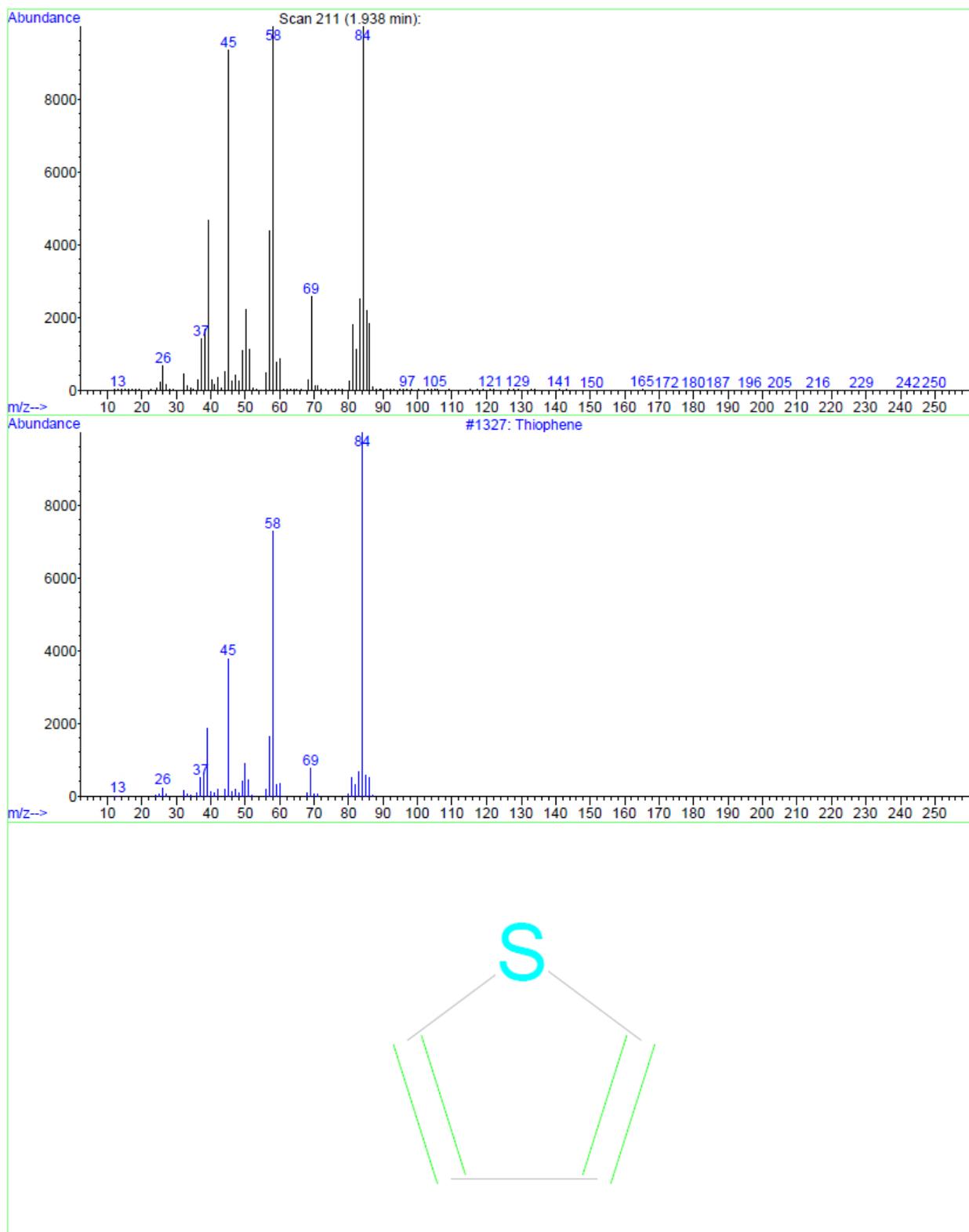
Figure SM 5.2.13.6 - Mass spectrum of the 1.938 min GC peak and its attribution to thiophene.
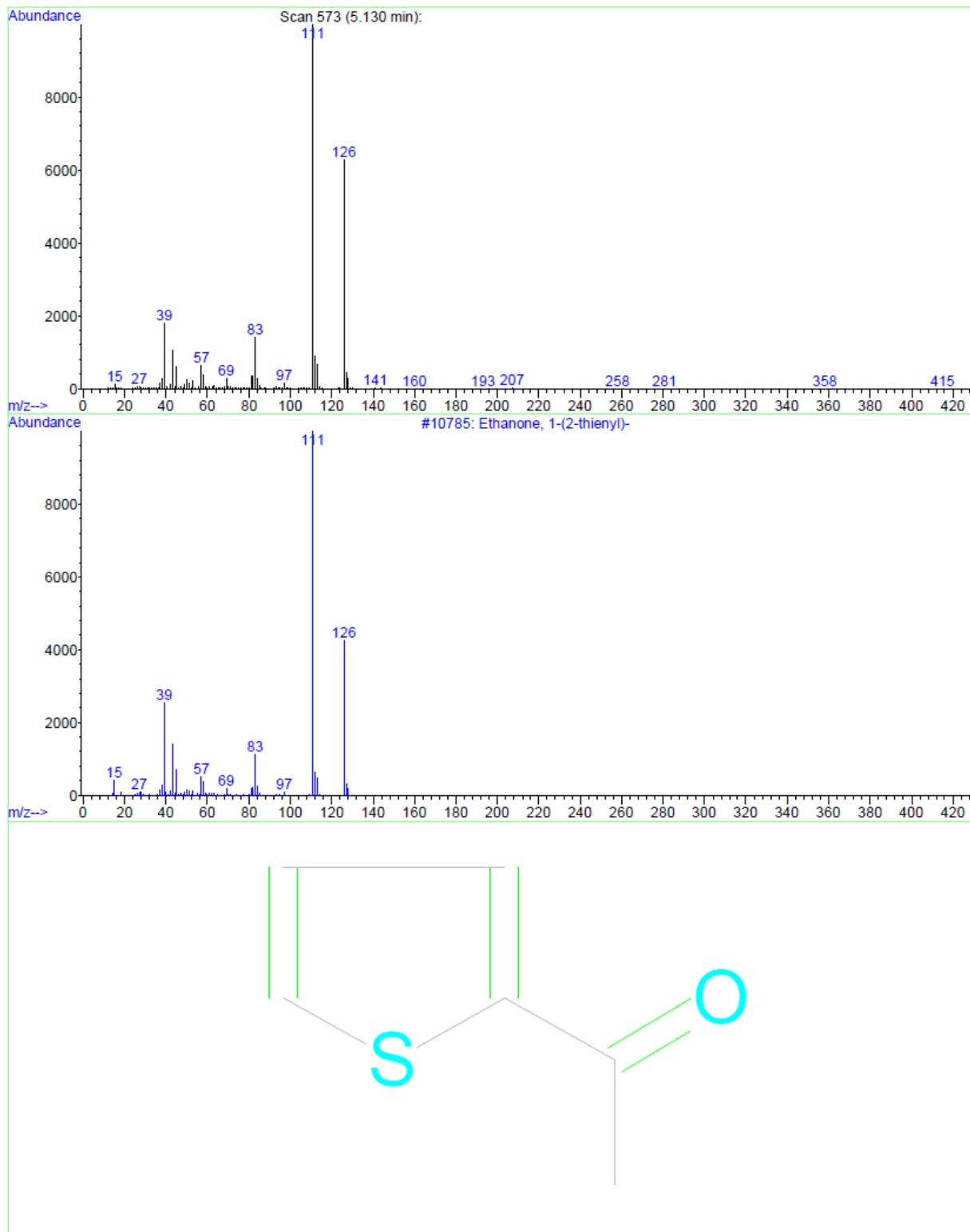
Figure SM 5.2.13.7 - Mass spectrum of the 5.130 min GC peak and its attribution to 2-acetyl-thiophene.

3. Kinetic results: Step-by-step tutorial for kinetic analysis

Kinetic analysis can be performed quite conveniently using a Workbook given the nature of data involved. Although this brief tutorial is intended to be self-explanatory, the reader can find a downloadable worked example along the same guidelines at

http://structreact.fc.ul.pt/Friedel_Crafts.xls

Consider:

A(=Thiophene) + B(=Acetic anhydride) → C(=2-acetylthiophene)

In previous works (*e.g.* ref. 9 in main document), it was assumed that the indicated Friedel-Crafts (FC) reaction was irreversible and first order in relation to A, as long as B (acetic anhydride) was in large excess, as is the case in the present example, since A:B ratio is 1:5, and therefore $C_B \simeq$ constant during the course of the reaction.

The differential rate equation for the irreversible reaction is given by:

$$-\frac{dC_A}{dt} = kC_A^\alpha C_B^\beta \qquad\qquad [1]$$

Since $C_B \gg C_A$, then $k\, C_B^{(t=0)} = $ cte. $= k_1$ and [1] simplifies to:

$$-\frac{dC_A}{dt} = k_1 C_A \qquad\qquad [2]$$

where k_1 is called a pseudo-first order rate constant. Integrating eq. [2] one obtains,

$$C_A = C_A^{(t=0)} \times e^{-k_1 t} \qquad\qquad [3]$$

Equation [3] is usually linearized to give:

$$\ln(C_A) = \ln(C_A^{(t=0)}) - k_1 t \Leftrightarrow \ln\left(\frac{C_A}{C_A^{(t=0)}}\right) = -k_1 t \qquad\qquad [4]$$

which is an equation of the form $\quad y = -a_1 x \qquad\qquad$ [5]

Therefore a plot of *y vs. x* passing through the origin easily leads to the value of k_1.

To do so, start MSOffice Excel's® and either open a new blank workbook by clicking the Office button (top left) and then 'New' followed by 'Blank document', with a structure similar to the one on Figures SM 5.2.13.8-9 or, having downloaded a workbook named "Friedel_Crafts" from the address given above, open it with the same programme.

Reaction:	Acetylation of thiophene with acetic anhydride (molar ratio of 1:5) using zeolite BEA as catalyst

Reactants	Mr /g mol^{-1}	d /kg L^{-1}	v /mL	m_0 /g	n /mol
Thiophene	84.14	1.051	0.8	0.88	0.011
Acetic Anhydride	102.09	1.08	5.0	5.40	0.053

Product		Mr /g mol^{-1}	d /kg L^{-1}
P1	2-acetylthiophene	126.18	1.168
P2	Acetic acid	60.05	1.049

Time / min	Area				
	Total	Acetic anhydride	Thiophene	P1	P2
0	17701064	10962868	3843111	0	276174
1	20350272	12983311	2713524	854172	1007564
2	24563052	13824825	3441775	2094354	1289862
3	22780380	12408798	2933307	2382709	1328264
4	29021248	16295720	2990218	3746815	2025870
6	22298108	15038743	1455385	2069765	1079865
8	26453948	16664519	1462658	3663421	1563237
10	25485520	16449274	1236163	3526537	1506853
15	17629926	12762058	401793	1813042	1225778
20	21845244	16160273	371964	2008656	1678840
25	24938432	15994193	454380	3969053	1621451
30	22825936	14631133	379565	3515700	1379267
40	19595924	13058297	227768	2959621	1204546
50	27205708	16726473	241872	4827746	1876080
60	27928104	18068494	175671	4544839	1757166

Figure SM 5.2.13.8 – Data input from GC chromatograms and physical/chemical properties.
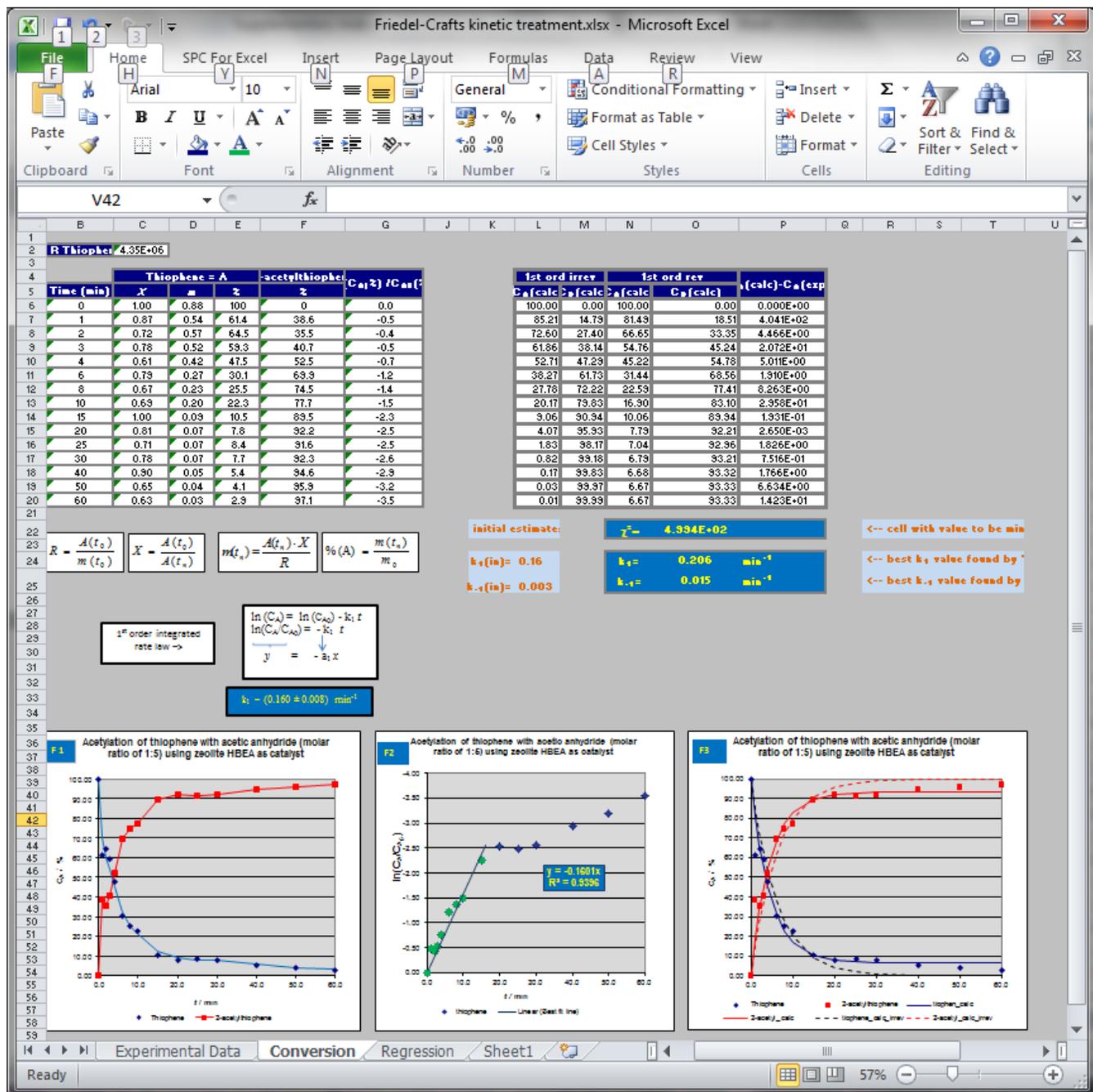
Figure SM 5.2.13.9 – Data processing and computation of kinetic parameters.

If 'Blank document' is chosen, create a workbook with a similar structure to that of the given example, if you need to follow this step-by-step tutorial.

Figure SM 5.2.13.9 shows the evolution of thiophene (A) and 2-acetylthiophene (C) over time on the left most graph at the bottom (F1). A line connecting dots was used in both cases to facilitate the plot's visualization. Column B (cells B6:B20) contains the different times at which each aliquot was taken from the reactor, column E (cells E6:E20) the weight percentage of reagent A and column F (cells F6:F20) the same quantity for product C. Column G (cells G6:G20) contains values of $\ln(C_A/C_{A_0})$.

On the bottom of the same figure, the graph at the centre (F2) contains two data series: series 1 includes all data values obtained, represented by blue squares; cells G6:G20 were used for the '*y*' axis, and cells B2:B20 for the '*x*' axis values.

As can be easily spotted, there is a linear relation only up to $t =15$ min. We have, therefore, added a second series, series 2, using this time only values between 0 and 15 min for the x-axis and the corresponding column G values for the y-axis. Clicking on 'Format data series', green squares were chosen (overlapping the previous blue ones) and a linear trendline, through the origin, was added passing through these data points, as follows: clicking any data point of series 2 with the mouse's right button, choose 'Add trendline' on the open box; a new box appears and you should tick 'Set intercept =0.0', 'Display equation on chart' and 'Display R-squared value on chart', and then click 'Close'.

For other experimental conditions (different temperatures, other zeolites, etc.) linearity may be better (or worse) and it may end before or after the limit indicated above. In fact, the reaction is not entirely irreversible and thus the influence of the backward reaction becomes more and more apparent in the plot as time grows and will influence the extent of reaction in which linearity holds.

The above approach only retrieves basic information about the regression. Students should complement this with a more complete data analysis using Office Excel's® 'Data analysis' add-in (*vd.* ref. 1 for further information on this subject). To use this add-in, start by choosing 'Data' on the main menu and then you will find 'Data Analysis' at the far end of the data ribbon. Pressing 'Data Analysis', a pop up window will open with all sort of options. Choose 'Regression' and a new window opens where you should choose the Input Y Range (in the worked example: $G$6:$G$14) and then the Input X-Range (in the worked example: $B$6:$B$14), and then click 'Constant is zero' since this regression is supposed to go through the origin (in our example we also chose 'New Worksheet Ply' with the name "Regression" so, if you create a new worksheet, you must give it another name or delete the one already created with the same name). At the end click OK. A new worksheet appears with all

relevant data as can be seen in Figure SM 5.2.13.10. In the example given, cells with green background contain the most important parameters of the regression for the envisaged purpose, namely the determination coefficient, 'R Square' (which should be close to 1), the standard error of the fit, 'Standard Error' (which should be small), the Fisher-Snedecor parameter, F (which should be high), the a_1 coefficient (in this case k_1) and the corresponding standard error, and the t-Student's statistics ($|t|$ should be > 1, for a coefficient to be statistically significant). Column J contents were added in order to obtain directly the confidence interval on k_1, by calculating '1-P').
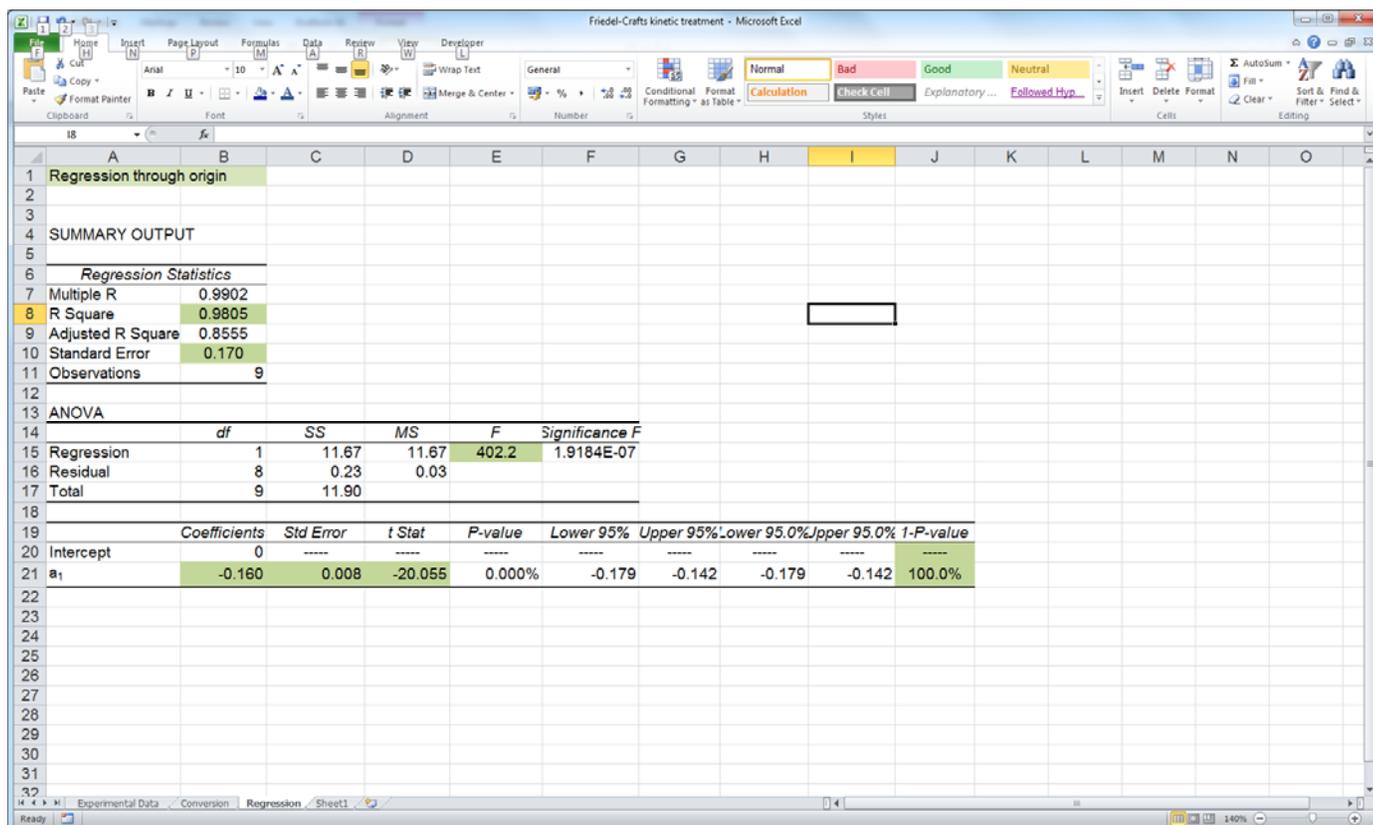
Figure SM 5.2.13.10 – Statistical analysis of linear regression.

In the presented example, the k_1 value and its standard deviation were copied onto a blue text box on row 33 of 'Conversion' worksheet (Figure SM 5.2.13.9).

If a more ambitious approach is sought, or for more interested students, one can also explore the case of the reaction being partially reversible.[2] In this instance, since $C_B \simeq$ constant, one can write,

$$C_A^{(t=0)} = C_A + C_C \iff C_C = C_A^{(t=0)} - C_A \qquad [6]$$

$$-\frac{dC_A}{dt} = k_1 C_A - k_{-1} C_C = k_1 C_A - k_{-1}\left(C_A^{(t=0)} - C_A\right) = (k_1 + k_{-1})C_A - k_{-1}C_A^{(t=0)} \qquad [7]$$

where k_{-1} is a true first order constant for the backward reaction. Integration of the above differential equation gives,

$$\ln\frac{(k_1 + k_{-1})C_A - k_{-1}C_A^{(t=0)}}{k_{-1}C_A^{(t=0)}} = -(k_1 + k_{-1})t \qquad [8]$$

that can be rewritten as,

$$C_A = \left(\frac{C_A^{(t=0)}}{k_1 + k_{-1}}\right)\left[k_{-1} + k_1 e^{-(k_1 + k_{-1})t}\right] \qquad [9]$$

and, of course, $C_C = C_A^{(t=0)} - C_A$, as stated above.

In the presented example, cells L6:L20 and M6:M20 in Figure SM 5.2.13.9 contain the % of A and C, respectively, calculated from the irreversible 1st order rate law, whereas cells N6:N20 and O6:O20 contain the % of A and C, respectively, calculated from the reversible 1st order rate law. It can easily be seen, at the right end of Figure SM 5.2.13.9 (F3), that the lines calculated using the latter rate law follow the observed values much better than the ones estimated from the former one.

k_1 and k_{-1} values used (blue box on rows 24 and 25) are optimized values that were obtained from another very useful MS Excel® feature, the Solver, an add-in that uses numerical methods to solve all kinds of equations. There is no need to deepen any further into the involved mathematics, but the interested reader can find all information needed on Excel's help menu on this add-in and on ref. 1. It is enough to say that cells P6:P20 compute $[C_A(calc)-C_A(exp)]^2$ using k_1 and k_{-1} values contained in O24 and O25. Cell O22 calculates

$$\chi^2 = \sum_{i=1}^{\substack{nr\ of \\ data\ points}} [C_A, i(calc) - C_A, i(exp)]^2 \qquad [10]$$

the sum of squared errors, χ^2 (or SSE), and this value is to be minimized by 'Solver' routines starting from initial estimates of k_1 and k_{-1} (in the example given, these values are on cells L24 and L25, respectively) and, of course, the logical choice for the initial estimate of k_1 is the value obtained

considering irreversibility. The choice of the initial value of k_{-1} is less obvious, but it must be significantly smaller than k_1.

To use solver, go to 'Data' and then, on the right end of the ribbon you will find 'Solver'. After clicking on it, a pop-up window entitled 'Solver Parameters' appears. You must then choose (set) the Target cell ('Set Objective') which, in our example is cell O22. This cell contains χ^2 (or SSE) value and since we must have it minimized (ideally it should be nil) we choose on 'Equal to', option 'Min', to find the "best" pair of k_1 and k_{-1} values resulting from this minimization procedure (in the example shown, we introduce in the field 'By Changing Variable Cells' the values to be optimized at cells \$O\$24:\$O\$25). This search cannot use the 'Subject to the Constraints' option (*e.g.*, force rate constants to be positive) since it can lead it away from the global minimum. Proceed to ´solver options' and choose a 'max time' of at least 500s, in the worked example 1000, 10000 iterations, 1e-6 for precision and 0.5% for tolerance. Leave the remaining options unchanged, click OK and then click 'Solve'. If you decide to keep the solver solution found, you should click OK one last time.

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[1] E.J. Billo, Excel for Chemists - A comprehensive guide, 3rd. ed., Wiley, Hoboken, 2011.

[2] D.G. Cox, Modern Liquid Phase Kinetics, Oxford Science Publications, Oxford, 1994.

Reactivity studies of 1-propyl-2-(thiophen-2-yl)-1*H*-pyrrole

Supplementary Information

1. Experiment notes

2. Figures

2.1 Photos of the apparatus used in experimental session 5

2.2 ^{1}H, ^{13}C NMR, IR and UV-visible spectra of the products

1. Experiment notes

The aim of this work is to study the reactivity of a thienylpyrrole heterocyclic system, specifically the selective functionalization of the pyrrole ring through electrophilic aromatic substitution (Vilsmeier-Haack formylation[1-2] and azo coupling[3]), or the selective functionalization of the thiophene heterocycle through metalation followed by electrophilic aromatic substitution.[4-5]

Thiophene and pyrrole are electron-rich heteroaromatic compounds. They are especially susceptible to an attack by electrophiles followed by a substitution reaction. According to earlier reports, the pyrrole nitrogen atom has a greater ability to delocalize the positive charge of σ-complexes than the sulfur atom in thiophene; pyrrole is therefore considerably more reactive towards electrophilic substitution than thiophene. Even when both α-positions (C-2 and C-5) of the pyrrole ring are occupied, electrophilic substitution will preferentially occur in the β-position (C-3) of the pyrrole ring rather than the α-position of the thiophene ring.[1-2, 3a] As a consequence, electrophilic substitution reactions of thienylpyrroles were found to be very selective. Having in mind that Vilsmeier formylation and azo coupling are aromatic electrophilic reactions the electrophiles (the Vielsmeier reagent ClCH=NMe$_2$$^+$) and the diazonium salt will react at the most electron rich part (nucleophilic) of the 1-(*n*-propyl)-2-(2′-thienyl)pyrrole that is the alpha position of the pyrrole heterocycle of the thienylpyrrole molecule to produce respectively 1-propyl-5-(thiophen-2-yl)-1H-pyrrole-2-carbaldehyde or 2-(4-nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1H-pyrrol-2-yl)diazene. On the other hand, the reaction of 1-(*n*-propyl)-2-(2′-thienyl)pyrrole with *n*-BuLi will selectively yield the organolithium derivative by deprotonation of the proton adjacent to the sulphur atom of the thiophene ring due its higher acidity. Subsequent reaction of the organolithium derivative with DMF will give the 5-(1-propyl-1H-pyrrol-2-yl)thiophene-2-carbaldehyde. A correlation between the selectivities in this reaction can be found through the analysis of the ^{1}H NMR spectrum of 1-propyl-2-(thiophen-2-yl)-1H-pyrrole (supplementary information of experiment

133). In fact, the most acidic proton 7.29 ppm (dd, 1H, J=5.1 and 1.2 Hz, 5′-H) ppm will be selectively metalated by *n*-BuLi.[6,7a,7b,7d]

Complete information about the mechanisms can be found in references 2 and 3.

Other methods for the synthesis of 2-thienylpyrroles derivatives could also be discuss in order to give a broad approach to this subject.[8]

The students should interpret ^{1}H NMR, ^{13}C NMR and IR spectroscopic data of all synthesized products in order to identify the compounds obtained as well to check their purity. Additionally the characterization of the compounds through the usual spectroscopic techniques (^{1}H NMR, ^{13}C NMR, IR) is extremely interesting and could be used in order to compare the spectra of the precursors with the spectra of the final products to confirm the functional group transformations. In addition an examination of ^{1}H NMR coupling constants and multiplicity of signals allows one to verify the positions at which functionalization occurs in the thiophene or pyrrole rings[9].

The students will use several different experimental techniques such as heating at reflux in anhydrous conditions and under an inert atmosphere, liquid-liquid extraction, evaporation of organic solvents with a rotary evaporator, gravity and vacuum filtration, thin layer chromatography (TLC) and column chromatography using silica gel and melting point identification.[10]

This experiment was previously developed in the research group of the author and was performed later by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students from the undergraduate Chemistry degree course at the University of Metz, France. Therefore, this experiment is appropriate for undergraduate Chemistry students who have previously acquired some skills with the experimental techniques involved ~~used~~ as well some knowledge regarding the theoretical concepts presented (synthesis, reactivity and spectroscopic data interpretation).

Synthesis and purification of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde (sessions 1-2)

In session 1 the DMF used should be previously dried[10] or this reagent should have a superior analytical quality in order to contain the least amount of water. After drying the organic phase over anhydrous Na_2SO_4 or $MgSO_4$, the drying agent should be very well washed with several portions of diethyl in order to recuperate the greatest possible amount of compound.

The preparation of the silica gel column and the separation of the components of the mixture (session 2, 4 and 6) should be run in a well-ventilated fume hood in order to avoid the contact with the silica gel as well as inhalation of volatile solvents such as chloroform, ethyl ether and petroleum ether. Silica gel may cause respiratory irritation, do not breathe the associated dust. After being used the silica gel must be stored in a closed container in the fume hood for subsequent disposal.

1-Propyl-5-(thiophen-2-yl)-1H-pyrrole-2-carbaldehyde:[7d] green oil (63%). ^{1}H NMR (CDCl$_3$) δ 0.90 (t, 3H, J=7.5 Hz, (CH$_2$)$_2$CH$_3$), 1.70-1.82 (m, 2H, CH$_2$CH$_2$CH$_3$), 4.41 (t, 2H, J=7.8 Hz, NCH$_2$), 6.40 (d, 1H, J=4.2 Hz, 3-H), 6.96 (d, 1H, J=4.2 Hz, 4-H), 7.12-7.16 (m, 1H, 4'-H), 7.18 (dd, 1H, J=3.6 and 1.2 Hz, 3'-H), 7.44 (dd, 1H, J=5.1 and 1.2 Hz, 5'-H), 9.54 (s, 1H, CHO). ^{13}C NMR (CDCl$_3$) δ 10.80, 24.68, 47.50, 111.89, 124.73 (two overlapped signals), 127.00, 127.42, 127.59, 132.17, 132.51, 136.36. IR (liquid film) ν 2964, 1658 (C=O), 1509, 1473, 1428, 1396, 1314, 1294, 1251, 1225, 1198, 1154, 1042, 847, 776, 702 cm^{-1}. UV-visible (EtOH): λ$_{max}$ nm (ε/M^{-1} cm^{-1}), 321.5 (24650).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 55-60%.

Synthesis and purification of 5-(1-propyl-1*H*-pyrrol-2-yl)thiophene-2-carbaldehyde (sessions 3-4)
In session 3 the DMF used should be previously dried[10] or this reagent should have a superior analytical quality in order to contain the least amount of water.
The diethyl ether used in session 3 should also be dry, however the use of a superior analytical grade reagent is preferred instead to of drying it in the laboratory due to security reasons. After drying the organic phase over anhydrous Na$_2$SO$_4$ or MgSO$_4$, the drying agent should be very well washed with several portions of diethyl in order to recuperate the greatest possible amount of compound.

5-(1-Propyl-1H-pyrrol-2-yl)thiophene-2-carbaldehyde:[7d] orange oil (68%). ^{1}H NMR (CDCl$_3$) δ 0.93 (t, 3H, J=7.5Hz, (CH$_2$)$_2$CH$_3$), 1.77-1.90 (m, 2H, CH$_2$CH$_2$CH$_3$), 4.08 (t, 2H, J=7.5 Hz, NCH$_2$), 6.20-6.22 (m, 1H, 4-H), 6.53 (dd, 1H, J=3.9 and 1.8 Hz, 3-H), 6.82-6.86 (m, 1H, 5-H), 7.12 (d, 1H, J=3.9 Hz, 3'-H), 7.71 (d, 1H, J=3.9 Hz, 4'-H), 9.87 (s, 1H, CHO). ^{13}C NMR (CDCl$_3$) δ 11.06, 24.61, 49.64, 108.70, 112.38, 112.44, 125.56, 125.70, 137.22, 140.94, 145.32, 182.54. IR (liquid film): ν 1659 (C=O), 1554, 1513, 1475, 1436, 1381, 1283, 1229,

1061, 941, 808, 724, 668, 611, 506 cm⁻¹. UV-visible (EtOH): λ_{max} nm (ε/M⁻¹ cm⁻¹) 374.0 (9474).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 52-58%.

Synthesis and purification of (*E*)-2-(4-nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1*H*-pyrrol-2-yl)diazene (sessions 5-6)

In session 5, when preparing the diazonium salt from 4-nitroaniline, KI indicator paper should be used to confirm the total conversion of the aniline to the diazonium salt (the colour of indicator paper changes from white to dark blue).

(*E*)-2-(4-Nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1H-pyrrol-2-yl)diazene:[7b] green solid with metallic luster (63%). Mp: 150.0-151.0 °C (acetone). ¹H NMR (DMSO-d₆) δ 0.87 (t, 3H, J=7.2, CH₂CH₂CH₃), 1.77-1.83 (m, 2H, CH₂CH₂CH₃), 4.50-4.60 (m, 2H, CH₂CH₂CH₃), 6.80 (d, 1H, J=4.5 Hz, 3-H), 6.96 (d, 1H, J=4.5 Hz, 4-H), 7.24-7.28 (m, 1H, 4′-H), 7.54 (dd, 1H, J=3.6 and 1.2 Hz, 3′-H), 7.80 (dd, 1H, J=5.1 and 1.2 Hz, 5′-H), 7.94 (d, 2H, J=9.3 Hz, 2′′ and 6′′-H), 8.34 (d, 2H, J=9.3 Hz, 3′′ and 5′′-H). ¹³C NMR (DMSO-d₆) δ 10.99, 24.26, 45.18, 103.35, 114.35, 122.13, 125.14, 127.83, 128.38, 128.57, 131.94, 135.38, 146.56, 147.65, 156.87. IR (Nujol) ν 1615, 1550, 1488, 1417, 1330, 1283, 1260, 1137, 851, 748, 533, 509 cm⁻¹. UV-visible (EtOH): λ_{max} nm (ε /M⁻¹ cm⁻¹) 488.0 (25,100), 289.0 (7900), 218.0 inf. (11,600).

The range of yields obtained earlier by students of the 4th year of the undergraduate Chemistry degree course at the University of Minho as well as by Erasmus students was 53-60%.

References

[1] A. H. Jackson, M. Artico, H. J. Anderson, C. E. Loader, A. Gossauer, P. Nesvadba, N. Dennis, In *The Chemistry of Heterocyclic Compounds*, R. A. Jones, Ed., Wiley: New York, 1990, Vol. 48, Part 1; p. 295.

[2] (a) C. W. Bird, Ed.; *Comprehensive Heterocyclic Chemistry II*; Pergamon: Oxford, 1984, Vol. 4, p 751-754 and p. 771-773. (b) G. Jones, S. P. Stanforth, In *The Vilsmeier Reaction of Fully Conjugated Carbocycles and Heterocycles*, L. A. Paquete Ed.; Organic Reactions, Wiley: New York, 1997, Vol. 49, p. 15-17. (c) L. March, *Advanced organic chemistry: reactions, mechanisms and structure*, 4th Edition, John Wiley and Sons, New-York, 1992, p.

542-543.(d) L. Kurti, B. Czakó, Strategic applications in named reactions in organic synthesis, Elsevier, S. Diego, USA, 2005, p 468.

[3] (a) J. March, *Advanced organic chemistry: reactions, mechanisms and estructure*, 4th Edition, John Wiley and Sons, New-York, 1992, p. 525-526. (b) K. Peter, C. Vollhardt, N. E. Schore, Organic Chemistry: structure and function, 5th Edition, W. H. Freeman, New-York, 2007, p. 979-981 and p. 1041.

[4] M. Schlosser, *Angew. Chem. Int. Ed.*, 2005, **44**, 376.

[5] R. Chinchilla, C. Nájera, M. Yus, *Chem. Rev.*, 2004, **104**, 2667.

[6] (a) R. E. Niziursky-Mann, M. P. Cava, *Heterocycles*, 1992, **34**, 2003. (b) L. I. Belen´kii, *Heterocycles*, 1994, **37**, 2029.

[7] (a) M. M. M. Raposo, "Recent developments in the chemistry of 2-thienylpyrroles: synthesis, reactivity and applications" In *Targets in Heterocyclic Systems*: *Chemistry and Properties*, , O. A. Attanasi, D. Spinelli, Eds, Italian Society of Chemistry, 2007, Vol. 11, 122-154. (b) M. M. M. Raposo, A. M. R. C. Sousa, A. M. C. Fonseca, G. Kirsch, *Tetrahedron*, 2005, **61**, 8249. (c) M. M. M. Raposo, A. M. R. C. Sousa, G. Kirsch, F. Ferreira, M. Belsley, E. Matos Gomes, A. M. C. Fonseca, *Tetrahedron*, 2005, **61**, 11991. (d) M. M. M. Raposo, A. M. R. C. Sousa, A. M. C. Fonseca, G. Kirsch, *Tetrahedron*, 2006, **62**, 3493. (e) M. M. M. Raposo, M. C. R. Castro, A. M. C. Fonseca, M. F. S. Cardoso, M. Belsley, L. M. Carvalho, P. Coelho, *Dyes Pigments*, 2011, **91**, 62.

[8] (a) G. P. Bean, "Pyrroles" In *The Chemistry of Heterocyclic Compounds*, R. A. Jones, Ed., Wiley: New York, 1990; Vol. 48, Part 1; p. 105. (b) T. L. J. Gilchrist, *Chem. Soc. Perkin Trans. 1*, 1999, **1**, 2849. (c) C. Schmuch, D. Rupprecht, *Synthesis*, 2007, 3095.

[9] (a) R. M. Silverstein, F. X. Webster, D. J. Kiemle, *Spectrometric Identification of Organic Compounds*, 7th Edition, John Wiley and Sons: New York, 2005. (b) E. Pretsch, P. Buhlmann, C. Affolter, *Structure Determination of Organic Compounds*, 3rd Edition, Springer-Verlag: Berlin, 2000.

[10] W.-L.F. Armarego and D. D. Perrin, "Purification of laboratory chemicals", 4th Ed.; Butterworth Heinemann, Oxford, 2000.

2. Figures

2.1 Photos of the apparatus used in experimental session 5

Figure SM 5.2.14.1 Left - Photo of the apparatus for the preparation of the diazonium salt (laboratory session 5i); Right - Photo of the apparatus for the coupling of the diazonium salt with the thienylpyrrole derivative (laboratory session 5ii).

2.2 ^{1}H, ^{13}C NMR, IR and UV-visible spectra of the products
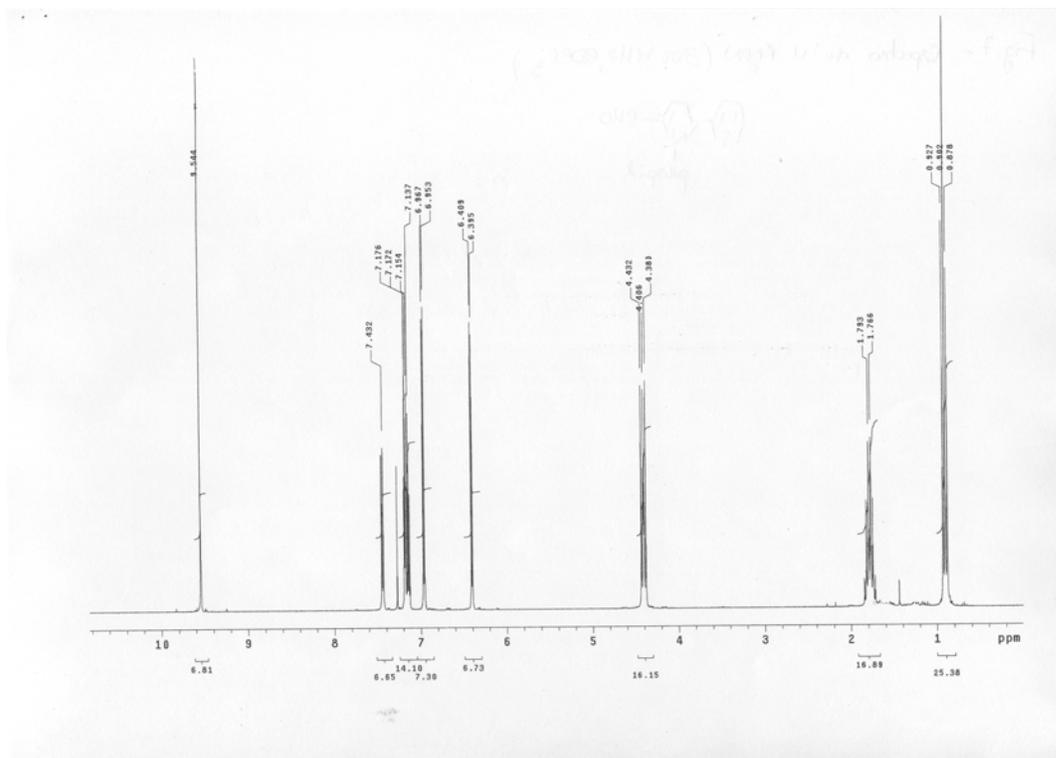
Figure SM 5.2.14.2. ^{1}H NMR Spectrum of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in CDCl$_3$ using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
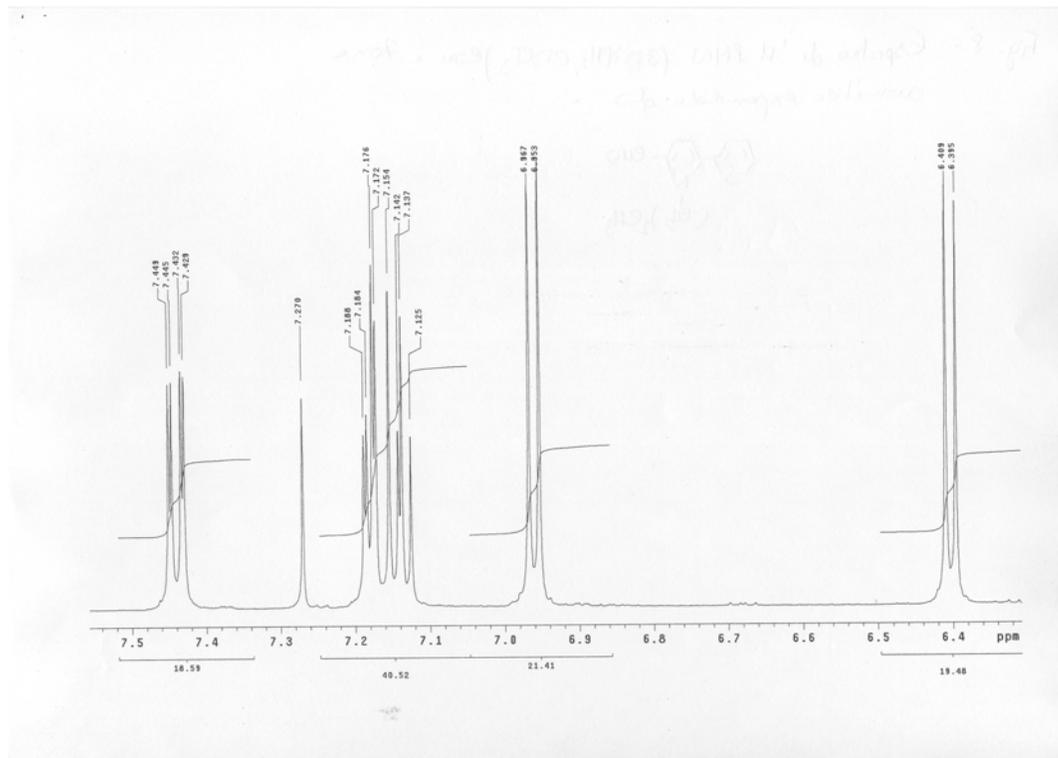
Figure SM 5.2.14.3. ^{1}H NMR Spectrum, with expanded aromatic zone, of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in CDCl$_3$ using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
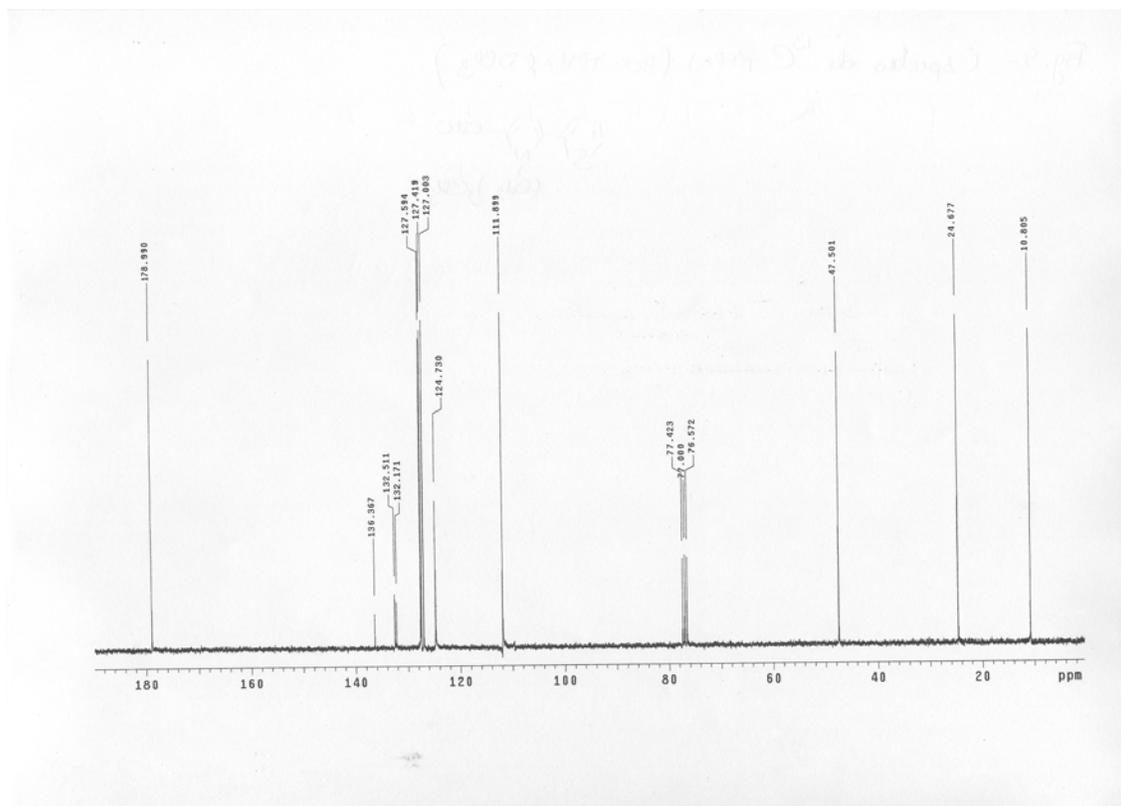
Figure SM 5.2.14.4. ^{13}C NMR Spectrum of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in CDCl$_3$ using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 °C.
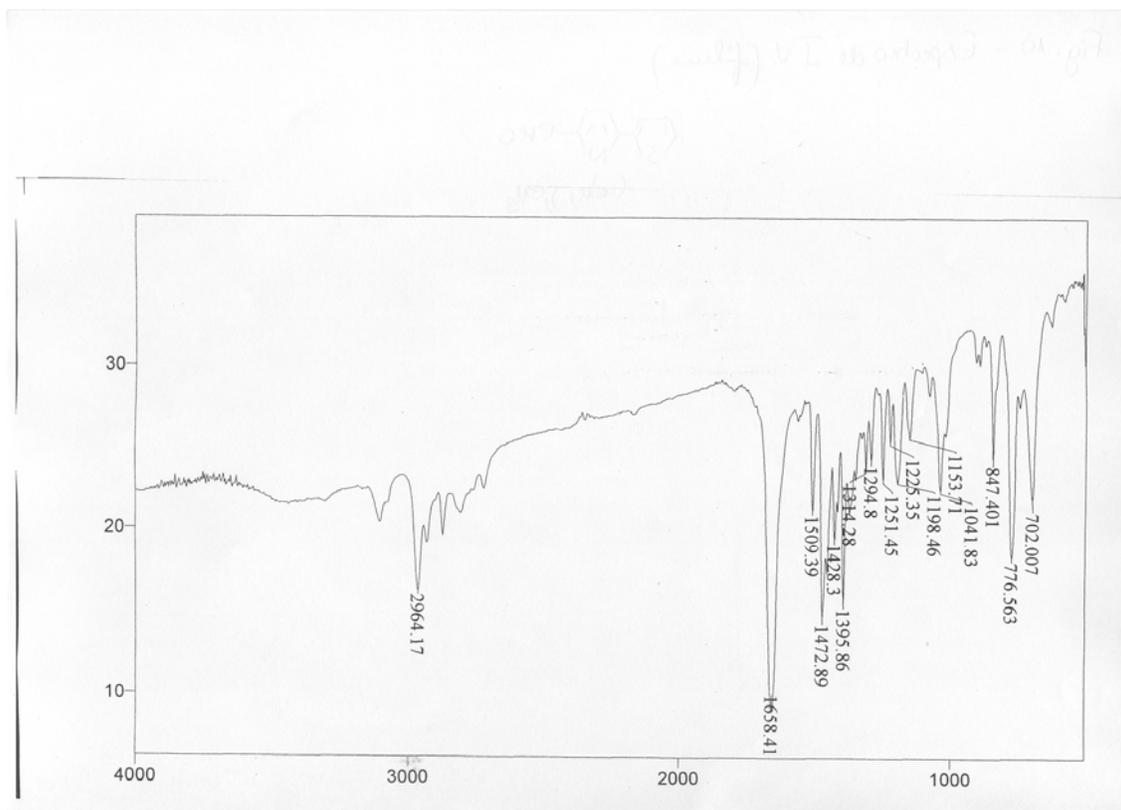
Figure SM 5.2.14.5. IR Spectrum of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in liquid film using a Perkin Elmer FTIR-1600 spectrophotometer.

9
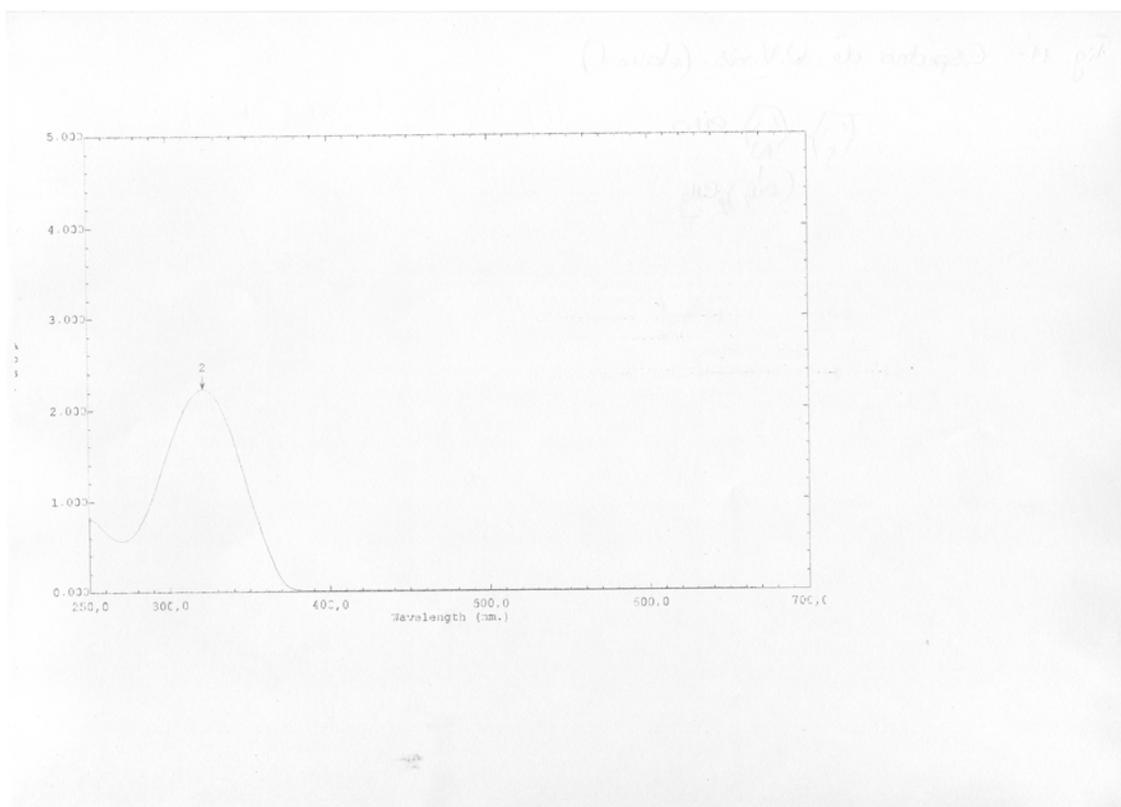
Figure SM 5.2.14.6. UV-visible Spectrum of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in ethanol using a Shimadzu UV/2501PC spectrometer at 25 °C.
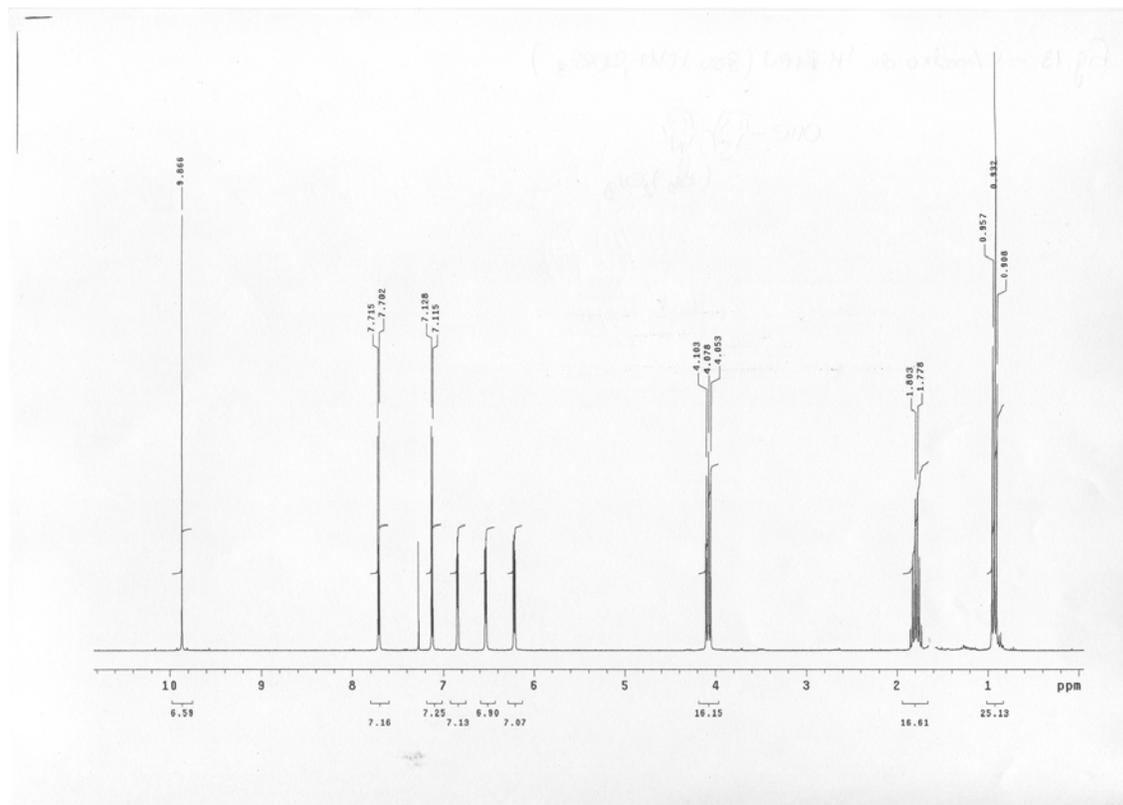
Figure SM 5.2.14.7. ^{1}H NMR Spectrum of 5-(1-propyl-1*H*-pyrrol-2-yl)thiophene-2-carbaldehyde in CDCl$_3$ obtained using a Varian Unity Plus spectrometer operating at 300 MHz at 25 °C.
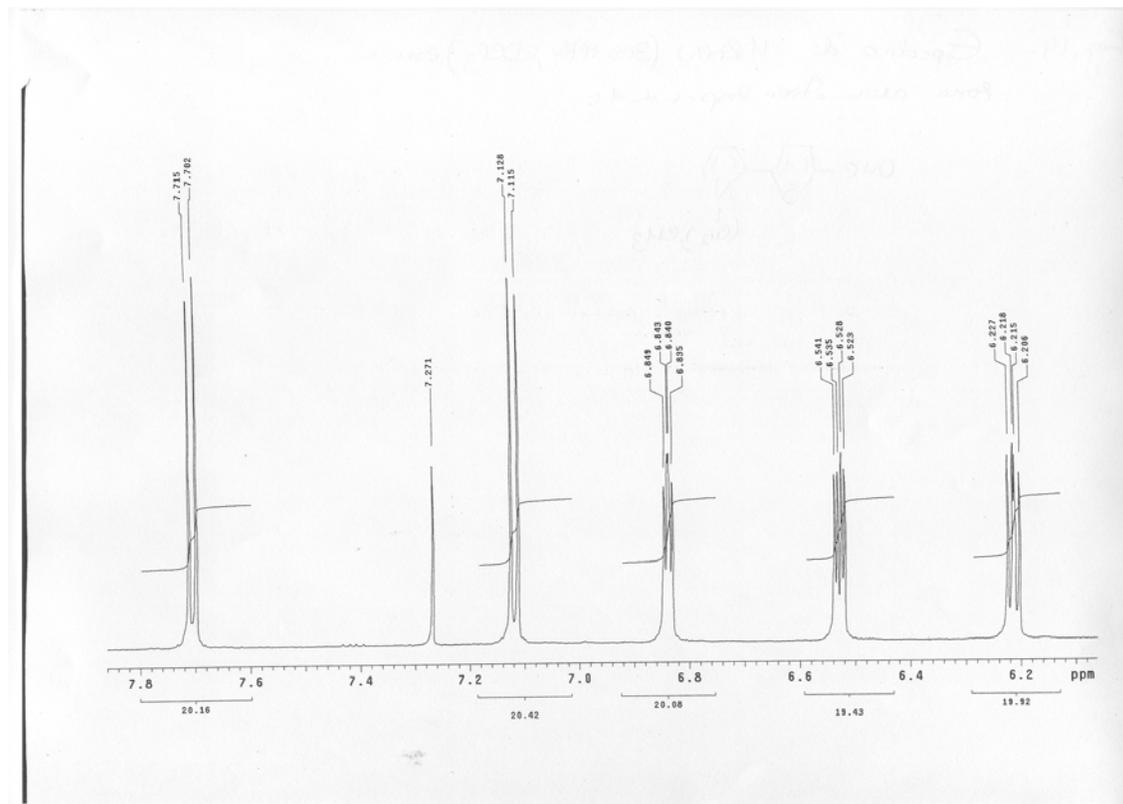
Figure SM 5.2.14.8. ^{1}H NMR Spectrum, with expanded aromatic area, of 1-propyl-5-(thiophen-2-yl)-1*H*-pyrrole-2-carbaldehyde in CDCl$_3$ using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
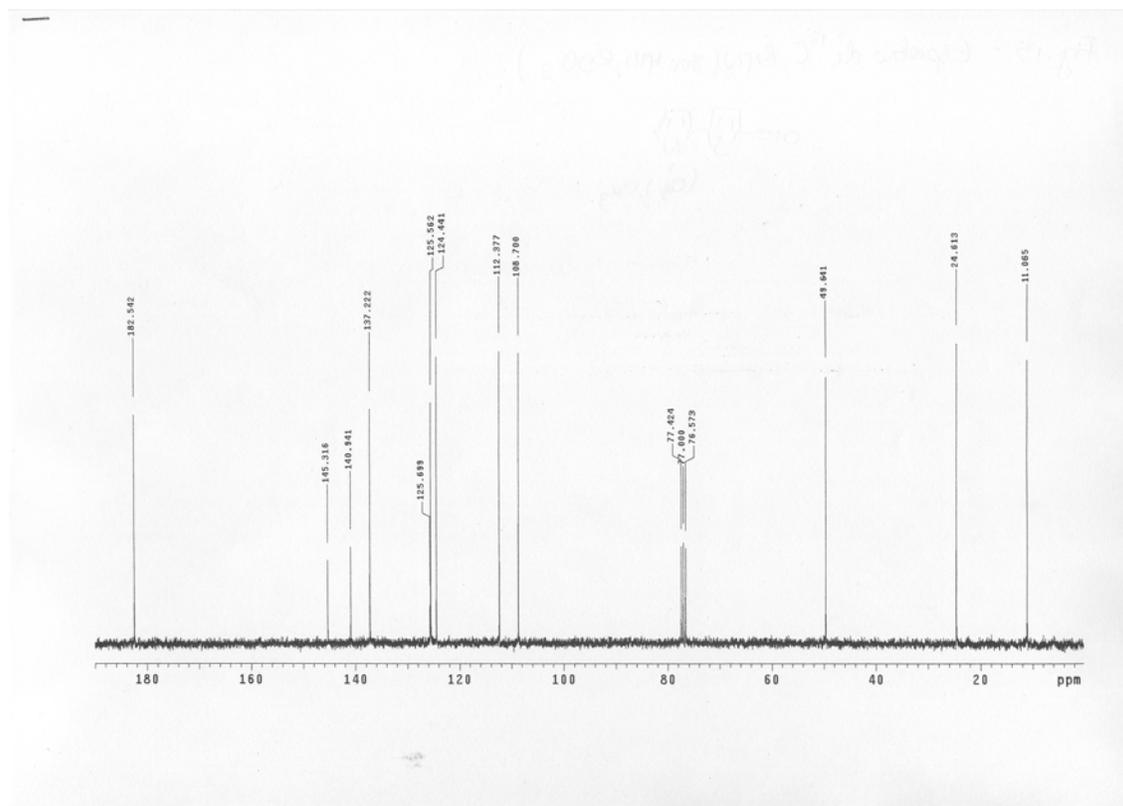
Figure SM 5.2.14.9. ^{13}C NMR Spectrum of 5-(1-propyl-1*H*-pyrrol-2-yl)thiophene-2-carbaldehyde in CDCl$_3$ using a Varian Unity Plus spectrometer operating at 75.4 MHz at 25 °C.
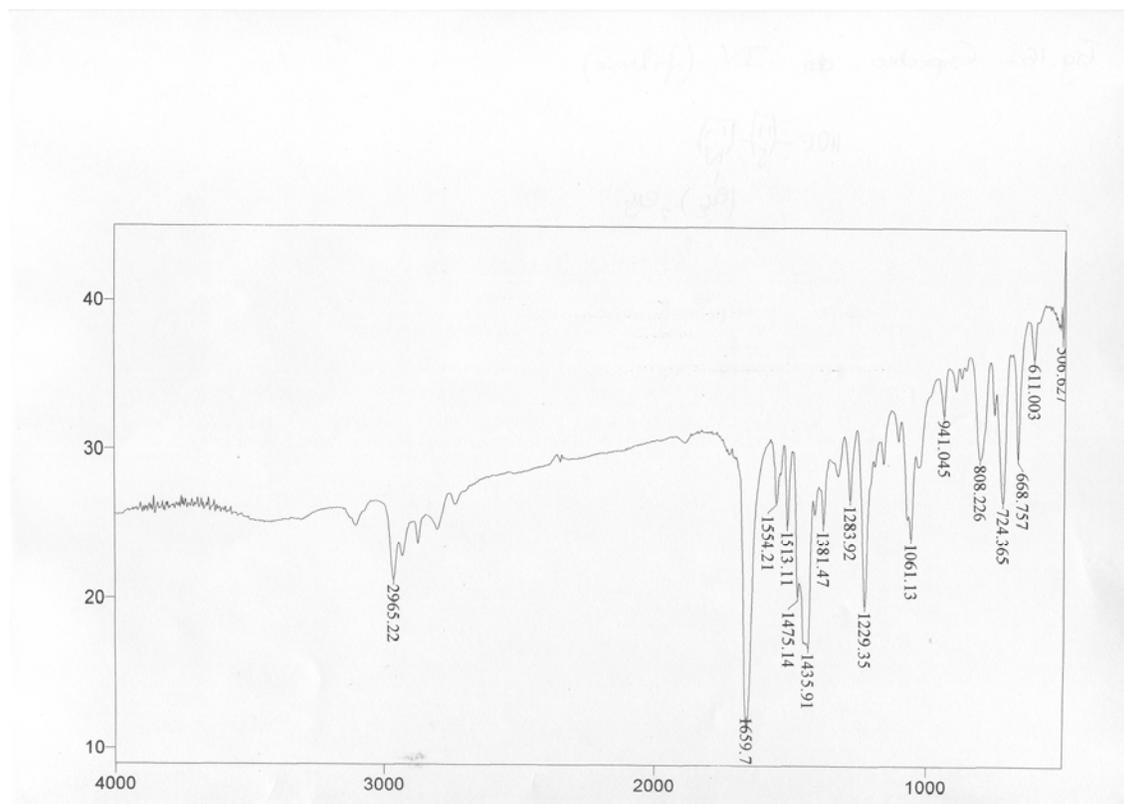
Figure SM 5.2.14.10. IR Spectrum of 5-(1-propyl-1*H*-pyrrol-2-yl)thiophene-2-carbaldehyde in liquid film using a Perkin Elmer FTIR-1600 spectrophotometer.
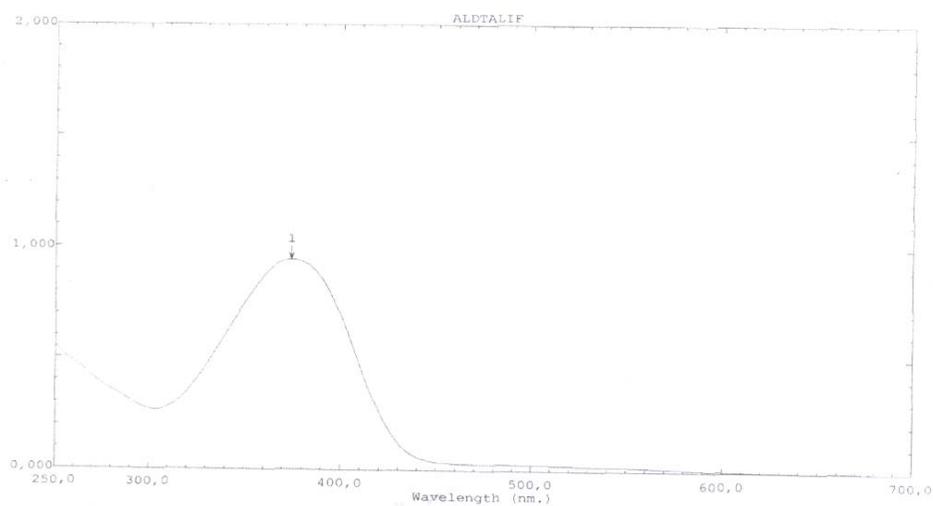
Figure SM 5.2.14.11. UV-visible Spectrum of 5-(1-propyl-1*H*-pyrrol-2-yl)thiophene-2-carbaldehyde in ethanol using a Shimadzu UV/2501PC spectrometer at 25ºC.
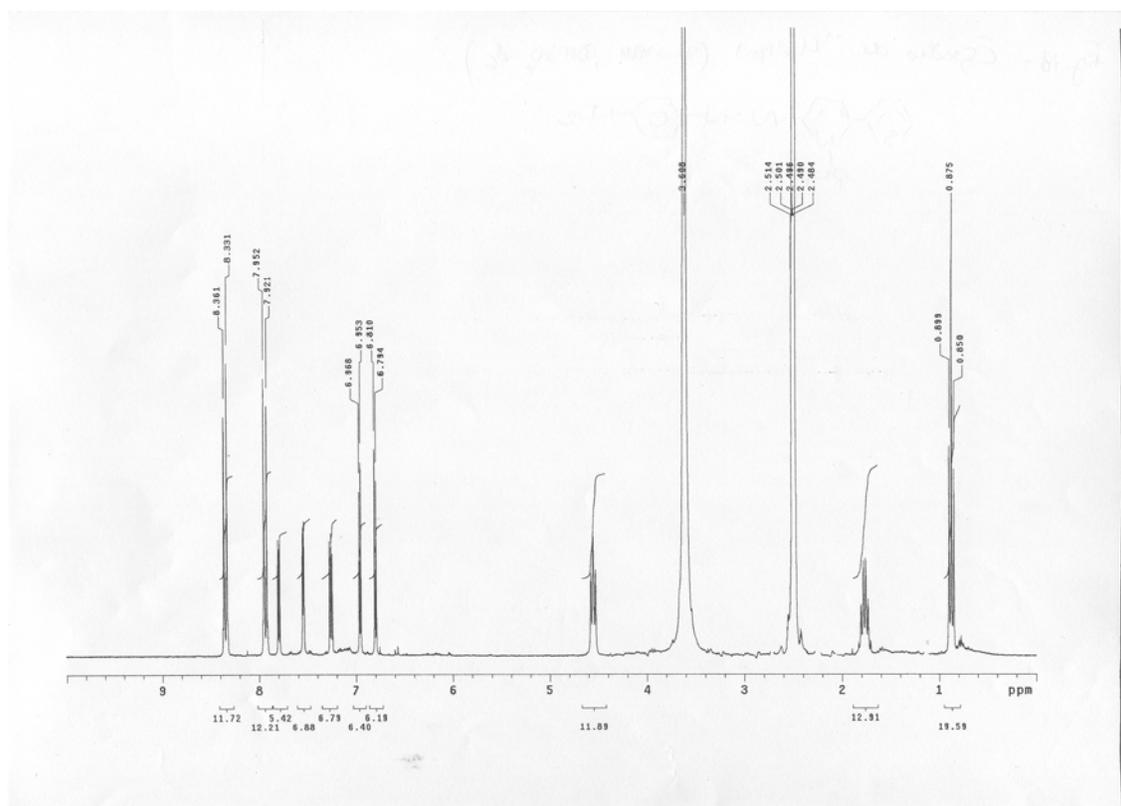
Figure SM 5.2.14.12. ^{1}H NMR Spectrum of 2-(4-nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1H-pyrrol-2-yl)diazene in DMSO-d$_6$ using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
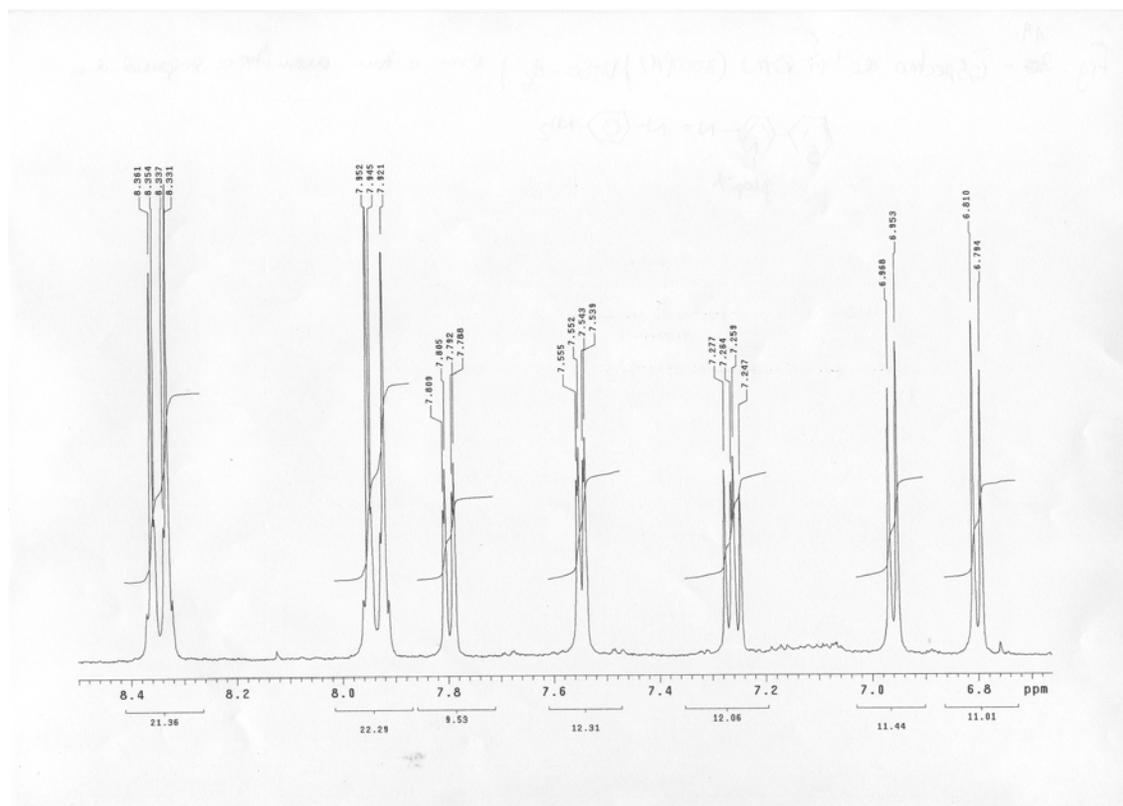
Figure SM 5.2.14.13. ^{1}H NMR Spectrum, with expanded aromatic area, of 2-(4-nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1H-pyrrol-2-yl)diazene in DMSO-d$_6$ using a Varian Unity Plus spectrometer operating at 300 MHz at 25 ^{o}C.
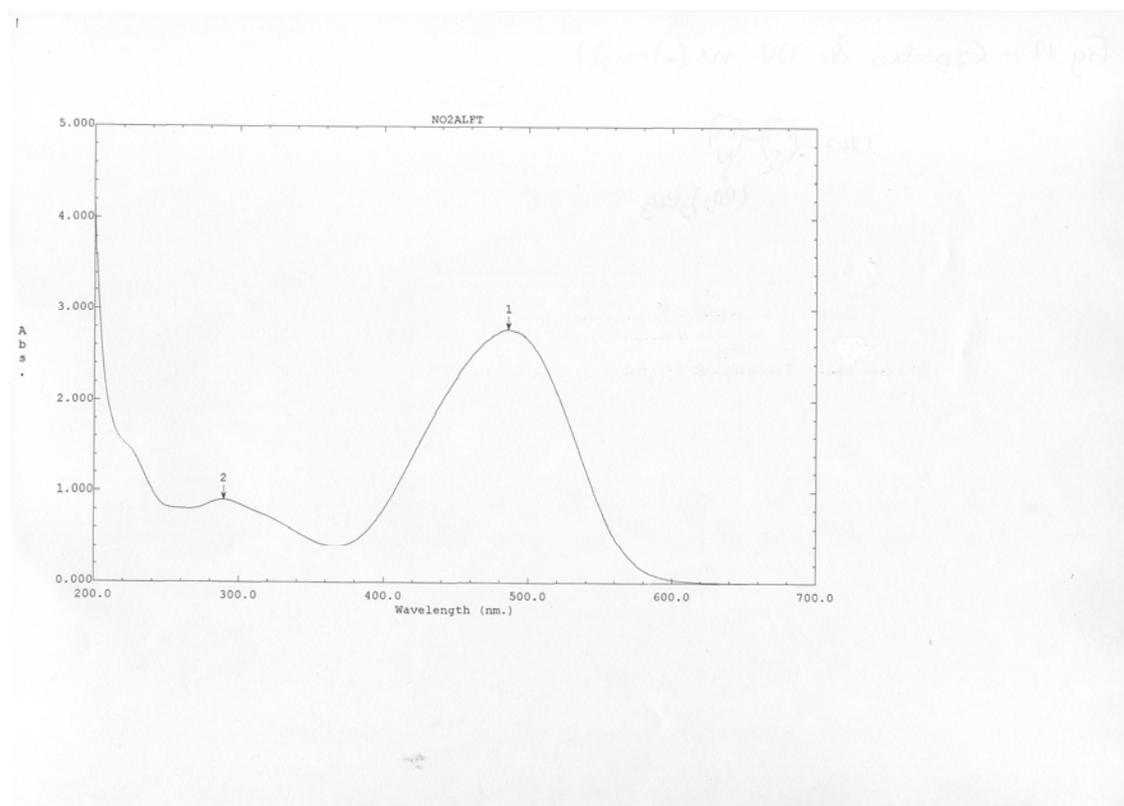
Figure SM 5.2.14.14. UV-visible Spectrum of 2-(4-nitrophenyl)-1-(1-propyl-5-(thiophen-2-yl)-1H-pyrrol-2-yl)diazene in ethanol using a Shimadzu UV/2501PC spectrometer at 25ºC.