Photochemical Electrocyclic Ring Closure and Elimination Reactions of N-(Oxothioxanthenyl)Naphthothiophene Carboxamides

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PHOTOCHEMICAL ELECTROCYCLIC RING CLOSURE AND ELIMINATION REACTIONS OF N-(OXOTHIOXANTHENYL)NAPHTHOTHIOPHENE CARBOXAMIDES

by

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ABSTRACT

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Photochemical cleavage reactions have found a widespread used in biological applications that require the photorelease of biologically active molecules such as proteins, peptides, neurotransmitters, and nucleotide phosphates. This research focuses on the design of photoremovable protecting groups which can be utilized to release these biomolecules by photolysis. These biomolecules are attached to the photoremovable protecting group at the sites of functional groups that are present within these substrates. Such functional groups are carboxylates, phosphates, thiolates, or phenolates, which upon exposure to light, are released as anions of varying basicities. The photochemical reaction involved is an electrocyclic ring closure between aromatic groups that are bridged by a carboxamide linkage. The key intermediate produced by electrocyclization is thought to have zwitterionic character. This zwitterionic intermediate is believed to expel the leaving group as the anion.

One of the aromatic groups attached to the amide carbonyl carbon has been a benzothiophene ring system with leaving groups such as $LG^- = Cl^-$, PhS^-, HS^-, $PhCH_2S^-$ that are present at the C-3 position. The photochemical expulsion of these LG^-s is experimentally known to occur in triplet excited state. Furthermore, the initial step in the mechanism has been shown to involve the transfer of triplet excitation energy from the chromophore to the benzothiophene ring. When this energy transfer is unfavorable, energetically, the quantum yield is expected to be low. This is the case when thioxanthone is the chromophore and benzothiophene is the energy acceptor.

The projects described herein involve efforts to lower the triplet excited state energy of the energy acceptor to make the triplet excitation energy transfer step more efficient. With benzothiophene as energy acceptor this step is endothermic. This project replaces the benzothiophene ring system with a naphtho[1, 2-b]thiophene so that energy transfer will be somewhat exothermic. An additional replacement attempted to use phenyl-2-thienyl ketone as energy acceptor.

With the naphtho[1,2-b]thiophene energy acceptor the expulsion of the C-3 $LG^- = Cl^-$ is more efficient ($\Phi = 0.084$) than with the benzothiophene ring system under comparable aqueous buffered conditions using 385 nm light to generate the initial triplet

excited state of thioxanthone chromophore. With benzothiophene as energy acceptor Φ = 0.035. Quenching experiments show that the photocyclization occurs via a short-lived triplet excited state localized primarily on the naphthothiophene ring. The initial cyclization must therefore be a very fast reaction. The limiting step likely is the triplet energy transfer step, which is only 46% efficient. Another source of inefficiency is the intersystem crossing step of the thioxanthone, which is 67% efficient. Subsequent to energy transfer, the remaining reaction steps are 27% efficient. These efficiencies are obtainable through a series of sensitized photolyses using xanthone and thioxanthone and the anilide obtained by replacing the thioxanthone chromophoric group and are thought to be representative of those for the actual thioxanthone linked by carboxamide to naphthothiophene.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF SCHEMES

LIST OF NMR SPECTRA FOR APPENDIX-1

LIST OF ABBREVIATIONS

ATP	Adenosine triphosphate
aq	Aqueous
Bzt	Benzothiophene
cAMP	Cyclic adenosine monophosphate
CM	Coumarin-4-ylmethyl
CNB	Carboxy-2-methylnitrobenzyl
CNDO	Complete Neglect of Differential Overlap
COSY	Homonuclear Correlation Spectroscopy
Cys	Cysteine
DCE	Dichloroethane
DCM	Dichloromethane
DMAP	Dimethylaminopyridine
DMF	Dimethylformamid
DMSO	Dimethylsulfoxide
DFT	Density functional theory
E_T	Triplet Energy
Et	Ethyl

GABA	γ-Aminobutyric acid
Glu	Glutamic acid
GHS	Glutthione
HPLC	High Performance Liquid Chromatography
HRMS	High-resolution mass spectrometry
IR	Infrared
ISC	Intersystem crossing
LG	Leaving group
mp	Melting point
NMR	Nuclear Magnetic Resonance
NOESY	Nuclear Overhauser Effect spectroscopy
PCC	Pyridinium chlorochromate
pHP	*p*-hydroxyphenacyl
PPG	Photoremovable protecting group
p-TsOH	*p*-toluenesulpfonic acid
Pyr.	Pyridine
THF	Tetrahydrofuran
Thiox	Thioxanthone

TLC	Thin Layer Chromatography
TMS	Tetramethylsilane
TS.	Transition State
UV	Ultraviolet
Φ	Quantum yield
hν	Light energy

CHAPTER 1

INTRODUCTION

1.1. General Introduction

Our research program mainly focuses on development of photoremovable protecting groups (PPGs) which can be utilized for biological studies. The study of PPGs has received a fair share of attention since their first discussion by Kaplan[1] and Engles[2] in the late 1970s. They can release biomolecules by irradiation, which allows spatial and temporal control of the release.[3, 4] This ability allows them to be used widely in synthesis, physiology, molecular biology and medicine. Their applicability has led to considerable interest in designing new types of PPGs. A number of reviews and books on PPGs in synthesis[1-5], and mechanistic studies[1,6,7] have appeared in recent years.

Several different names can be found in the literature for PPGs. Most widely used names are "phototriggers", "caged compounds", and "photo-labile protecting groups". The cage compound is used to describe a biological molecule where activity or function is masked by chemical modification with a PPG. The time period for the release of the bioeffectors upon photolysis will need to be fast enough to allow a study of interest. As examples in the case of caged protein kinase A[8a], the bioeffectors can be released over minutes[8a], or seconds in the case of cage tyrosine Ca/calmodulin inhibitor[8b], or milliseconds as with cage ATP[8c], or microseconds as with caged neurotransmitters.[7,8d]

This chapter states the goal of the current project, provides some background information, summarizes mechanistic studies, and gives some advantages and disadvantages of currently available PPGs and applications. Our group focuses on the

caged compounds that expel leaving groups via zwitterionic intermediates produced through electrocyclic ring closure reactions of benzothiophene carboxamides derivatives. Some background information on the photorelease of leaving groups via zwiterionic intermediate also will be discussed. For most PPGs, the direct release of leaving groups from the protecting groups, or indirect release of leaving groups from sensitized photolysis has been reported. Finally, some background information about sensitized photolysis will be discussed.

1.2. The Goal of the Project and Problem Statement

Although a plethora of PPGs is currently available to release biologically important molecules (LG^-) (Figure 1.1), no universal PPG exists. The photolysis wavelength is the most addressable problem. The majority of cage compound use UV radiation for releasing biomolecules. The use of UV light can cause cell damage and mortality due to unintended side reactions of biomolecules[8b]. Many biological systems involve enzymes in an aqueous medium at high ionic strength. Under such physiological conditions the premature release of the bioeffectors can occur in the dark.[8] Another problem is the limited basicity of the releasable biological anion with most reported caged compounds. Almost all biological systems consist of biomolecules like proteins and peptides. The building blocks of those molecules are amino acids. These amino acids contain side chain functionality like phenolates and thiolates, which are basic and difficult to mask by the majority of PPGs. Few satisfactory compounds are currently available for thiols including the sulfhydryl group of cystein.[9] Those that have been reported have serious drawbacks, which will be discussed in a future section. Our

research focuses on developing new caged compounds that overcome the above mentioned problems.

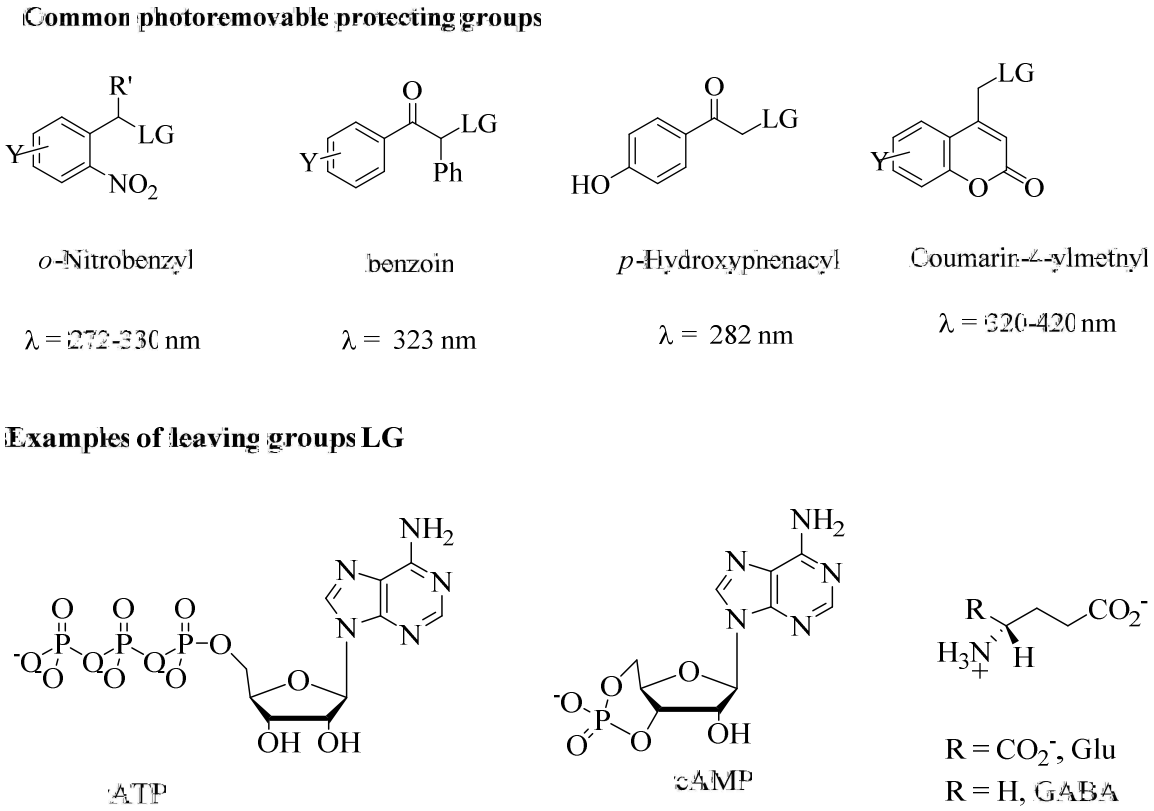


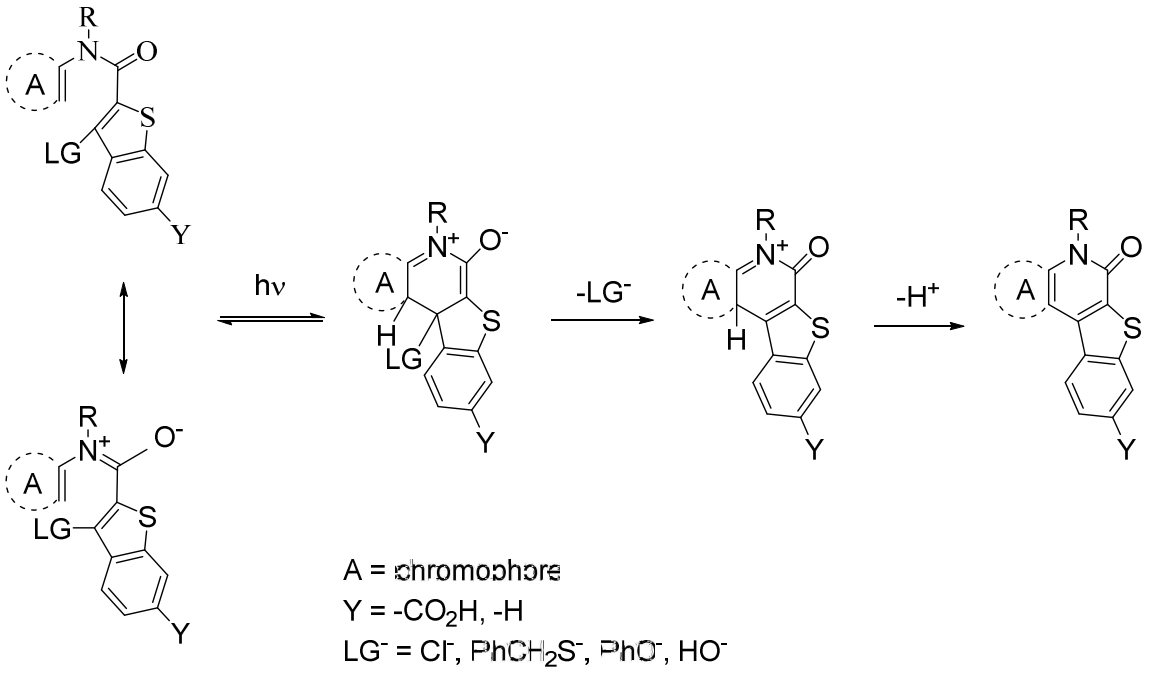
Figure 1.1. Common photoremovable protecting groups and biologically important leaving groups

Our group focuses on the caged compounds that expel leaving groups via zwitterionic intermediates produced through electrocyclic ring closure reactions of benzothiophene carboxamides[10] (Scheme 1.1). The above PPG allows the photolysis wavelength to be varied by attaching different chromophoric **A** structures to the nitrogen of the amide linker.

A problem found with this approach is that the attachment of chromophores absorbing light at longer wavelengths causes a decrease in the quantum yields for the

photochemical reaction. For example, with the *N*-phenyl group as **A** in compound **1**, Φ = 0.23 and the compound photolysis at 310 nm.[11] With the benzophenone moiety as **A** in compound **2**, Φ = 0.15 and the compound photolysis at 365 nm.[11] With the thioxanthone moiety as **A** in compound **3a**, photolysis occurs at 386 nm and Φ = 0.069.[10]

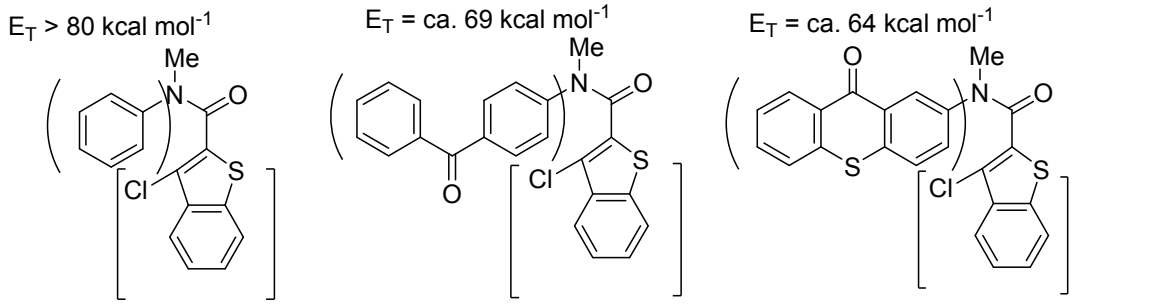
Scheme 1.1. Expulsion of Leaving Groups *via* Zwitterionic Intermediate Produced Through Electrocyclic Ring Closure Reaction of Benzothiophene Carboxamide[10]



Furthermore, quenching studies of the above compounds with 1,3-pentadiene indicate that these PPGs undergo photochemical electrocyclic ring closure reaction via the triplet excited state. Comparison of the triplet excited state energies of the chromophores to that of the benzothiophene ring suggests a solution to the above problem.

For N-phenyl compound **1**, the benzothiophene moiety absorbs the light. The initial singlet excited state intersystem crosses to generate the triplet benzothiophene, which releases the leaving group. The triplet benzothiophene has an energy E_T = ca. 69

kcal mol^{-1}.[12] With the para-benzoyl substituent attached to the N-phenyl group, the resulting benzophenone moiety absorbs the light at 365 nm. The benzothiophene absorbs at the shorter wavelength of 310 nm. So for compound **2** the lowest singlet excited state is localized on the benzophenone moiety. Intersystem crossing of this singlet excited state should be rapid and generates the triplet excited state, which has an energy E_T = ca. 69 kcal mol^{-1}. This energy is equal to the triplet energy of the benzothiophene (E_T = ca. 69 kcal mol^{-1}). However, when the benzophenone moiety is replaced by the thioxanthone group, which absorbs light at 386 nm, intersystem crossing produces a triplet excited state of E_T = ca. 64 kcal mol^{-1}.[13] This triplet excited state is expected to lie below that of the benzothiophene triplet excited state. Triplet energy transfer to the benzothiophene moiety would then be endothermic.



1

Φ = 0.23
hν 310 nm

2

Φ = 0.15
hν 365 nm

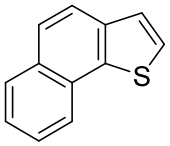
3a

Φ = 0.069
hν 386 nm

To summerize the above, the quantum yields decrease in the order 0.23, 0.15 and 0.069, when changing **A** from N-phenyl to benzophenone moiety to thioxanthone group. It could be postulated that the triplet excited state energy of the chromophore must be transferred to generate the triplet excited state of the benzothiophene group. For

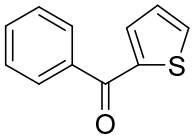
example, in case of thioxanthone compound whose quantum efficiency is the lowest, energy transfer is estimated to be endothermic by ca. 5 kcal mol^{-1}.

The goal of the current research is to facilitate the above triplet energy transfer from the chromophore to the thiophene ring system. In order to accomplish this; the triplet energy of the thiophene ring system should be lower. Some modifications will need to be done on the benzothiophene ring. For example, the benzothiophene ring in compound **3** could be replaced by the naphtho[1,2-b]thiophene **4** or phenyl-2-thienyl ketone **5** with E_T = 62 kcal mol^{-1}.[14,15]



naphtho[1,2-b]thiophene

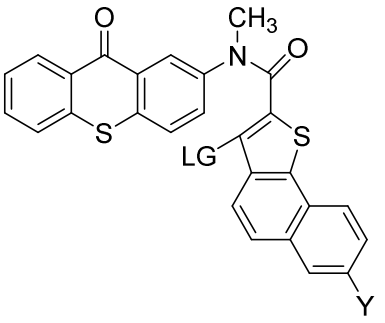
4



phenyl 2-thienyl ketone

5

The proposed compounds that need to be synthesized are compound **6**, and compound **7**, which bears a leaving group LG^- (Br^- and Cl^- respectively) at C-3 position and contains a carboxylic acid group at C-2 position to couple with the amine of the chromophore. The synthesis and photochemical studies will be discussed in Chapter 2,and 3.



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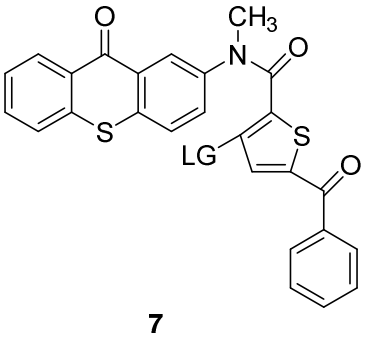

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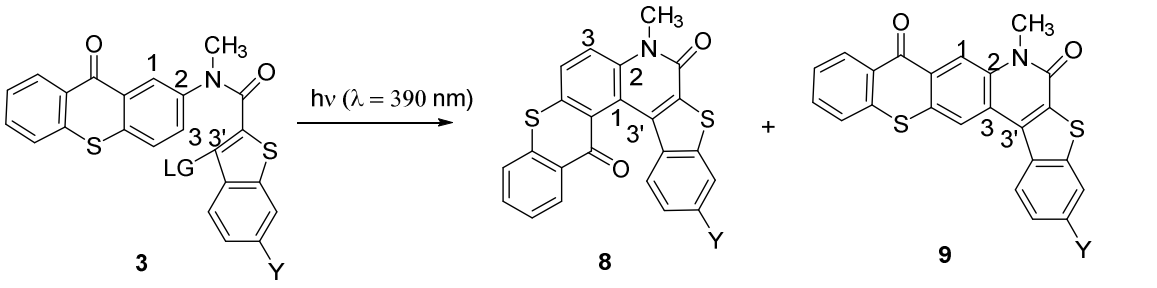
The rationale for proposing the above compounds, naphtho[1,2-b]thiophene **4** and phenyl-2-thienyl ketone **5** for studies are facile. First, as mentioned above, the quenching studies[10,11] using 1,3-pentadiene suggests that the photochemical reaction involves triplet excited states. DFT calculations[10] on N-(9-oxothioxanthenyl) benzothiophene compound **3** provides more information about the triplet excited state potential surfaces involved in the photoreaction of the PPG **3** with $LG^- = Cl^-$ (Scheme 1.2).

Scheme 1.2. Photoreaction of N-(9-oxothioxanthenyl)benzothiophene Compounds



3a (Y = -H, LG = -Cl)
3b (Y = $-CO_2CH_3$, LG = -Cl)
3c (Y = -COOH, LG = -Cl))

8a (Y = -H), 42%
8b (Y = $-CO_2CH_3$), 99%
8c (Y = -COOH), 98%

9a (Y = -H), 58%
9b (Y = $-CO_2CH_3$), not observed
9c (Y = -COOH), not observed

According to Figure 1.2, the initial lowest excited state, ^{3}**Thiox**, is located 59 kcal mol^{-1} in energy above the $\mathbf{S_0}$ state. Experimentally, the energy may be 64 kcal mol^{-1}.[13]. The ^{3}**Thiox** is not of much geometric difference from the $\mathbf{S_0}$ state. Another minimum with the triplet excitation localized mostly on the benzothiophene (^{3}**Bzt**), is located about 4 kcal mol^{-1} below the ^{3}**Thiox**. This latter energy may be as high as 69 kcal mol^{-1} for benzothiophenes, according to the literature[12]. Geometrically, ^{3}**Bzt** is different from ^{3}**Thiox** in that ^{3}**Bzt** C-3' carbon bearing the leaving group is pyramidal. The pyramidalized carbon attacks the C-1 carbon or the C-3 carbon to cyclize to form two isomers. These DFT calculations suggest that it may be reasonable to increase the quantum yields for reaction with benzothiophene replacements with other thiophene

groups that have lower triplet excited state energies than the thioxanthone triplet excited states.

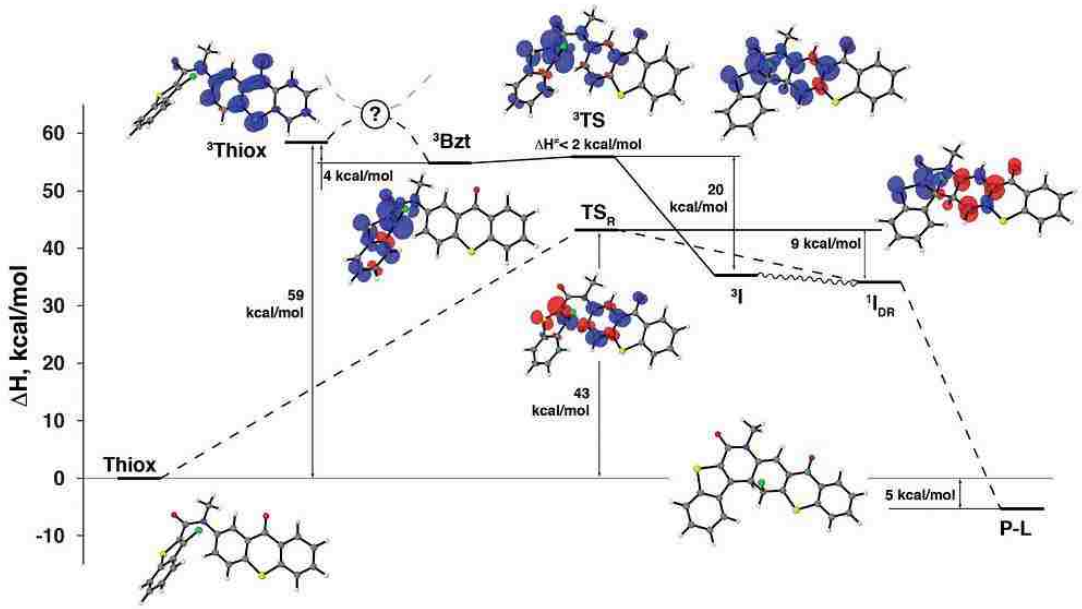


Figure 1.2. Relative enthalpies of the stationary points on the ground-state S_0 and the lowest triplet T_1 surfaces relevant for formation of the ring closure product from **3**. Unpaired spin density isosurfaces are shown for open-shell species.[10]

1.3. Background of PPGs

1.3.1. Criteria for the Successful PPG

The definition of the good PPG depends on the application. Criteria for the successful PPG for the release of common biological molecules have been proposed.[3,5] The desired properties in a PPG are:

1. The protected molecule should be stable in the absence of light.
2. The photoprotected substance must be soluble in aqueous buffered media.
3. The photoprotected substance must be stable to hydrolysis, especially at higher ionic strength.
4. The irradiation wavelength should be over 300 nm to avoid photolyzing.

biological media and should not be absorbed by the photoproduct, nor the biological media.

5. The photochemical reaction should be fast and have high quantum efficiency.
6. The cage compounds and the photoproduct must be biologically harmless.

Even though no PPG meets all the criteria, they can be considered to be excellent guidelines for designing and developing caged compounds. There are several PPGs reported which satisfy the criteria to different extent.

1.3.2. Introduction to Some PPGs, Mechanism of Photorelease, Advantages and Disadvantages

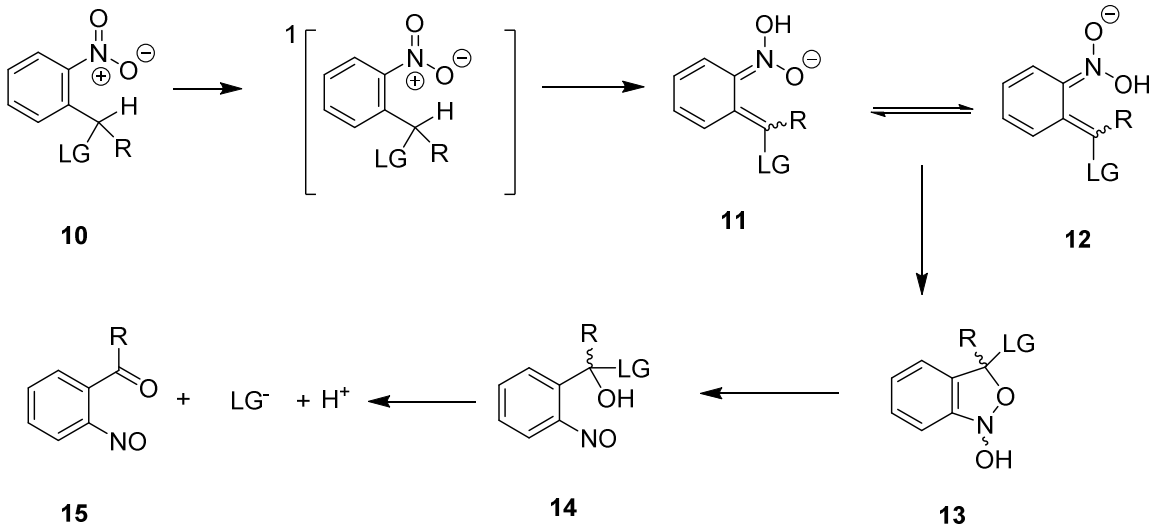
The four most widely used PPGs to date are based on o-nitrobenzyl derivatives, benzoin derivatives, phenacyl derivatives and coumarin derivatives. More details about each of these will be discussed in following sections.

1.3.2.1. The *o*-Nitrobenzyl Group

o-Nitrobenzyl groups are the most widely used in biological applications up to now. The use of o-nitrobenzyl group to release benzoic acid was first reported by Barltrop *et al*.[16] The uncaging of *o*-nitrobenzyl groups bearing a leaving groups at the α-position is thought to follow the following mechanism (Scheme 1.3).[17] Once compound **10** is irradiated by light the singlet aci-nitro intermediate **11** is formed by [1,5] hydrogen shift from *o*-methylene group. This short lived species can be detected by laser-flash photolysis. It isomerizes by hydrogen transfer to form another aci-nitro isomer **12,** which facilitates cyclization to generate benzoxazole intermediate **13**, which is a UV-silent. Its presence is established by time-resolved IR. The ring opening reaction takes place to

form hemiacetal **14**. Its presence is indicated by a new strong IR band for the nitroso group and the absence of a signal for a carbonyl group. Finally formation of nitrosobenzaldehyde **15** is established by appearance of the IR band for the carbonyl group.

Scheme 1.3. Mechanism of Photorelease of *o*-nitrobenzyl Group



The *o*-Nitrobenzyl group has some good absorption and photochemical properties. Quantum yields for release of leaving groups are considerably high, and absorption extends past 300 nm. However, it also has a number of drawbacks. A nitrosoarene is the byproduct of the release of the leaving groups (LG^-) from the o-nitrobenzyl protecting groups. Nitrosoarenes are toxic to cells. They also can oxidize newly released thiols or thiols already exists in living systems.[18] The *o*-nitrobenzyl group had been thought to be suitable for caging cystein (Cys) residue.[9] Unfortunately the nitrosoarene will undergo reduction in the ground state by released thiol which would be oxidized.[18] Indeed, one of the function of the glutathione (GHS) with its cystein sulfhydryl group is to reduce the toxicity of foreign substances[19] including nitrosocompounds by converting them to less toxic compounds like arylhydroxylamine , N-arylsulphenamides and anilines.[18] *o*-

Nitrobenzyl groups are therefore not suitable for caging biomolecule which contains sulfhydryl group like Cys and GHS.

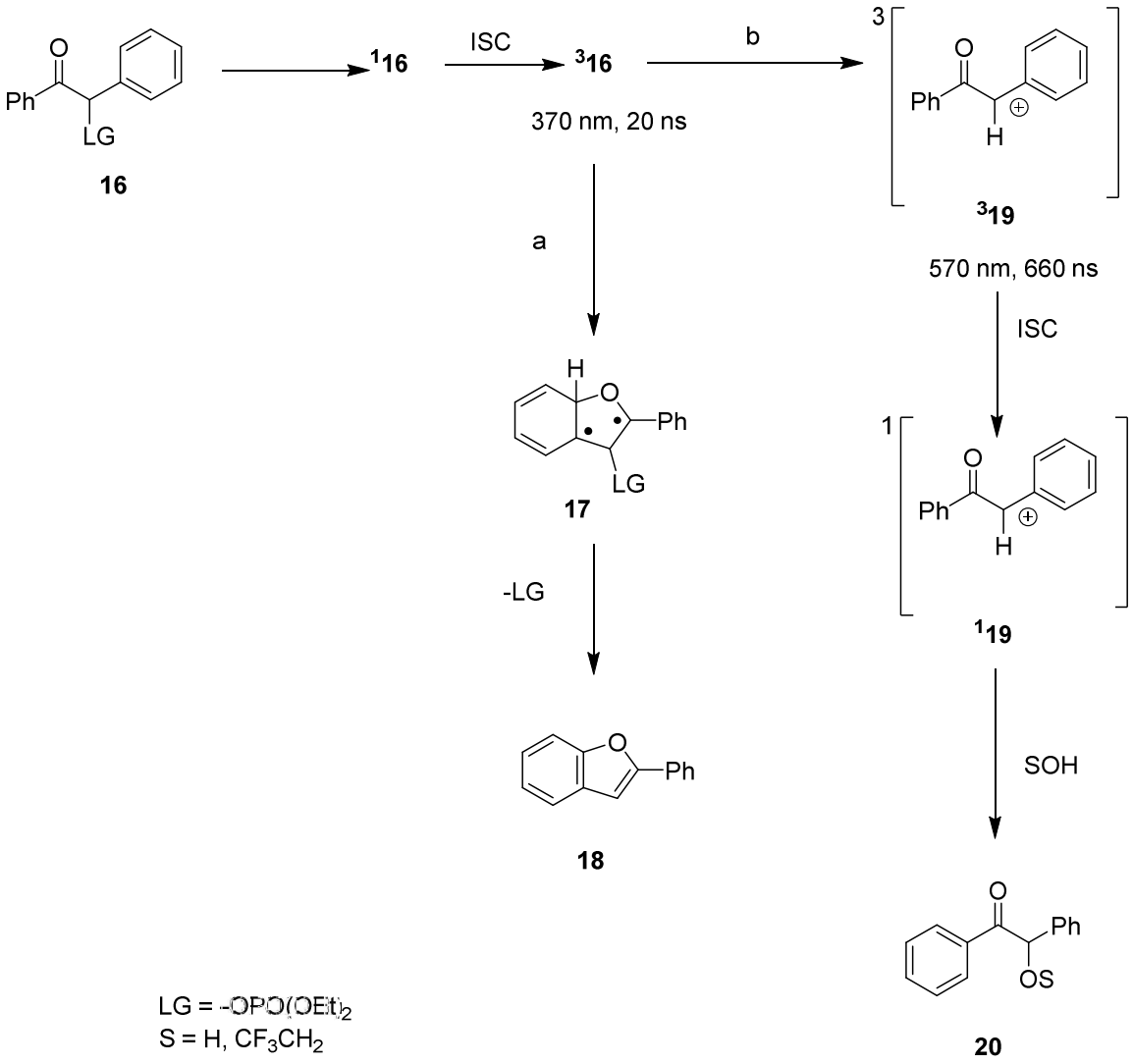
1.3.2.2. The Benzoin group (Desyl, Bnz)

Benzoin compounds were first studied by Sheehan and Wilson [20] and were found to expel acetate groups. Several different mechanisms of release have been proposed.[20,21] It was shown that substitution on the benzene ring, solvent, and the nature of the leaving group strongly influence mechanism. Wirz and Givens[21] reported the more recent mechanism for the non-substituted benzoin group to release phosphate (R =H and LG^- = OPO (OEt_2) (Scheme 1.4). The lowest excited triplet state ^{3}**16** is the reactive excited state of **16**. It is formed within few picoseconds by excitation through intersystem crossing (ISC) of ^{1}**16**. There are two solvent dependent competing pathways (**a**,**b**) to release LG^- that originate from this triplet. Path (**a)** dominates in most solvents except water and fluorinated alcohol to form benzofuran derivative **18** within 20 ns. It is assumed that this remarkably fast transformation is cyclization to form biradical **17**. In water and fluorinated alcohol, reaction path **(b)** dominates, and forms **20** as major product.

The photo reaction is relatively clean and uniform and leads to form a biologically inert byproduct. The byproduct benzofuran is nonpolar and can be easily separated from other polar or acidic components. The benzoin ester can be cleaved by relatively high absorption wavelength of 366 nm. The benzoin group also processes several drawbacks. The protection of chiral molecules with benzoin compounds can be problematic since the benzoin bears a chiral center too. Thus, incorporation of the chiral leaving group will

result in the formation of diastereomers. Another problem is the lower solubility of protected substrate and the main by-product in aqueous media.

Scheme 1.4. Mechanism of Photorelease of Benzoin Group

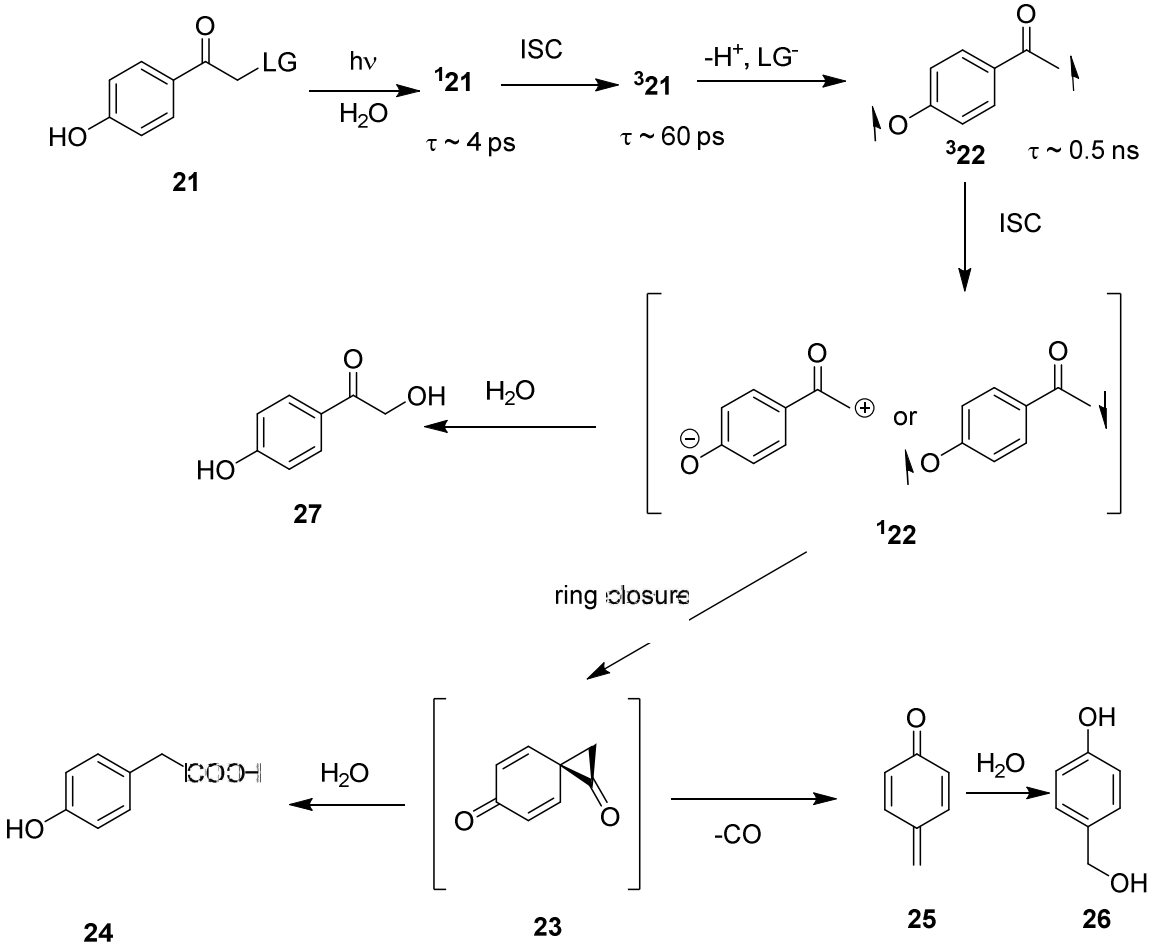


1.3.2.3. The *p*-Hydroxyphenacyl (pHP) Group

p-Hydroxyphenacyl group has been reported in synthetic organic chemistry [22], neurobiology[3,7a,23] enzyme catalysis.[7b,23b,24] This is an excellent alternative for o-

nitrobenzyl and benzoin group due to its remarkable properties. The mechanism of the photolysis is understood as Scheme 1.5 based on time resolved transient absorption analysis.[4,6,17a,25]

Scheme 1.5. Mechanism of Photorelease of *p*-hydroxyphenacyl Group



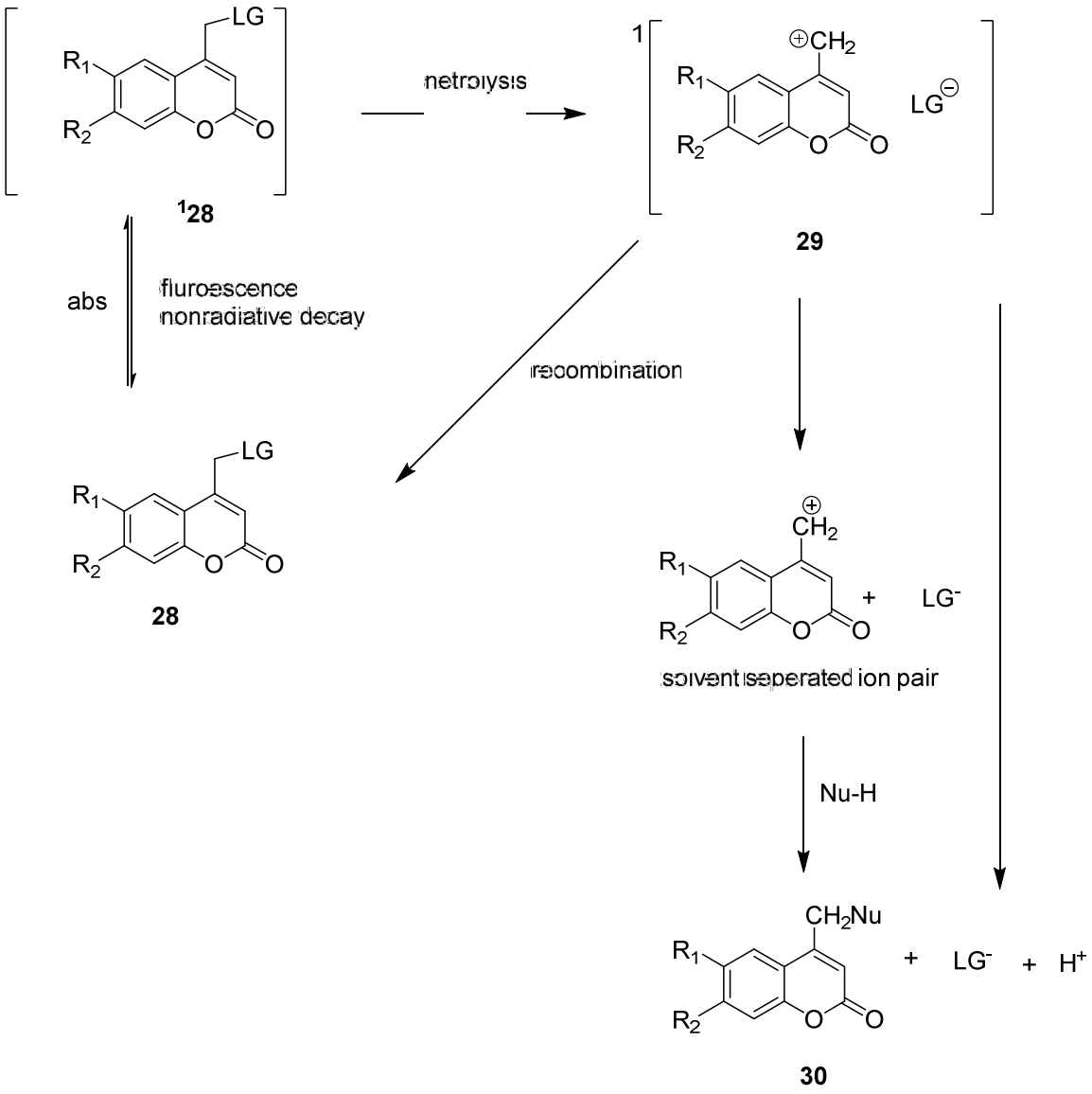
The triplet molecule **21** rearranges to generate the triplet biradical ^{3}**22** with a lifetime less than 1 ns by concerted expulsion of the leaving group and the phenolic proton. The ^{3}**22** undergoes intersystem crossing to give its ground state ^{1}**22** which cyclizes to form cyclopropanone **23**. Hydrolysis of the propanone **23** intermediate generates the carboxylic acid **24** while decarbonylation gives the alcohol **26.** This mechanism also provide pathway for the minor photolysis product **27** by hydrolysis of

^{1}22. This product is predominant for the ring-contraction photoreactions where ring strain discourages or prevent formation of the cyclopropanone intermediate **23**.[26] The photolysis reactions of *p*-Hydroxyphenacyl compounds often have high quantum yields and chemical yields. Other important features are good aqueous solubility and stability, and the lack of quenching by O_2 in aqueous media. But the disadvantage is that the photolysis wavelength lies deep in the UV, which limits their applications.

1.3.2.4. Coumarin-4-ylmethyl Groups (CM)

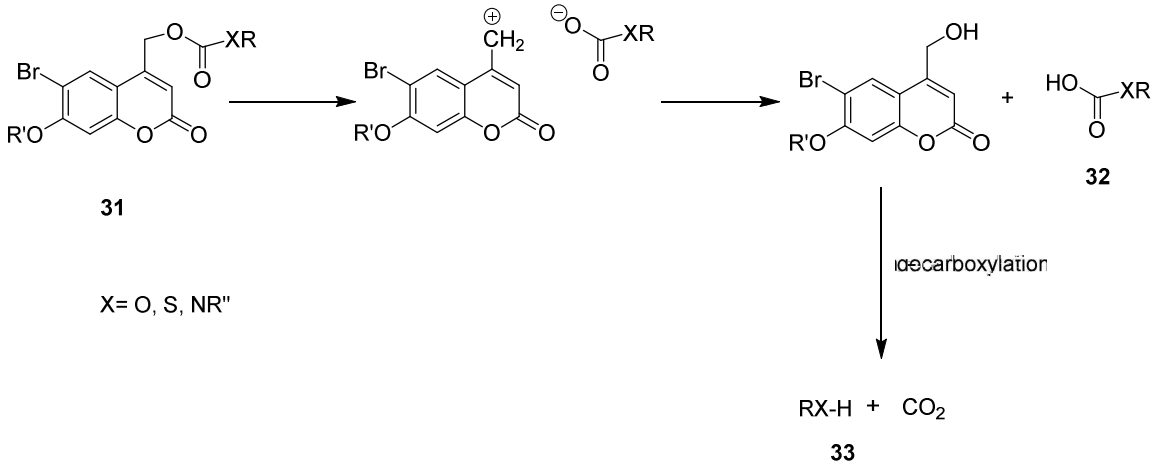
The coumarin groups have been attractive to the researchers because they absorb at very long wavelengths, even longer than 400 nm. Some also have the feature of being able to undergo two-photon excitation, so that photolysis can occur with near IR light of wavelengths 700-800 nm. However, the coumarin groups rely on an S_N1 reaction that occurs in the short lived singlet excited state.[27] With the increasing basicity of leaving group anion, the excited state S_N1 elimination of the leaving group slows, and the reaction becomes inefficient.[28] So the coumarin is limited to weakly basic leaving groups such as carboxylates and phosphates. The mechanism of photorelease of coumarin-caged esters is shown in Scheme 1.6.[29] Photolysis initially produces the lowest $^1(\pi, \pi^*)$ excited singlet state **^{1}28.** Deactivation occurs by fluorescence and non-radiative processes, respectively, and competes with the heterolytic cleavage of C-LG bond to form a singlet ion pair **29**. The initially formed ion pair **29** is the key intermediate. The coumarinylmethyl cation either reacts directly with nucleophiles or solvent to generate a new stable coumarylmethyl product **30** or recombines with LG^- to regenerate reactant.

Scheme 1.6. Mechanism of Photorelease of Coumarin-caged Compounds



Poor leaving groups such as alcohols, phenols, and thiols are resistant to heterolysis. Such groups can be released when caged through a carbonate linkage with the (6-bromo-7-hydroxyalkoxycoumarin-4-yl) methyl moiety **31** as given in Scheme 1.7. The initially formed carbonic or thiocarbonic acid **32** is unstable and undergoes decarboxylation to form alcohol or thiols **32**.

Scheme 1.7. Decarboxylative Photorelease of Alcohols, Thiols, and Amines[17a]



1.4. Applications

Photoremovable protecting groups have various uses in diverse field such as photolithograpy[30], peptide synthesis[31], medicine and drug discovery[32], studying biological process[6,32-35,20a] etc.. The following section will briefly discuss some of the applications where PPGs are useful.

1.4.1. Studying Neurotransmitters

Investigation of the kinetics of neurotransmitter mediated reactions on cell surfaces is a well known example.[6] This broad area of research has been covered in several reviews.[32-35] Neurotransmitters are responsible for important physiological functions. Glutamate, γ-aminobutyric acid (GABA), glycine, aspartate and kainic acid are the well known examples for neurotransmitters and neurotransmitter inhibitors. They bind to a receptor and cause the opening of ion channel resulting in a flow of current. Activation of synaptic transmission in the nervous system occurs on a sub-micron spatial scale and on a sub-millisecond time scale. So experimental procedures for studying those processes requires similar precision. Use of caged compounds to release

neurotransmitters is a useful strategy for that. Numerous photoactivable derivatives of neurotransmitter and neurotransmitter antagonists are available. When the neurotransmitters are released from their cages in a controlled manner, the resulting ion current can be monitored to provide information on the kinetics of the process. Grewer and co-workers[36] have reported the development of a nitrobenzyl protected caged compound, α-carboxy-2-methylnitrobenzyl (αCNB) ester of glycine with higher quantum efficiency (Φ= 0.38) and thermal stability at physiological pH. The decay of the aci-nitro intermediate occurs on the microsecond time scale. So this is a useful tool for investigation of the process involved in the opening of the glycine receptor channels and the effect of mutations of the glycine receptor. Kander, *et al.*[37] have reported pHP glutamate for studying long term potentiation and long term depression. They examined the role of postsynaptic cellular changes in CA1 hippocampal pyramidal cells which are thought to be involved in the mechanism of memory and learning.

1.4.2. Studying Photorelease of Protein and Peptides.

Proteins and peptides involve a wide range of biological activities and functions. Synthetic peptides can be used to selectively inhibit protein activity. Photoactivable peptides have the potential for such kinds of applications. Derivatizing an amino acid side chain in peptides can change their activity. Warker, *et al.*[8b] have described derivatization of tyrosine side chain in RS-20 by nitrobenzyl group. RS-20 is the target peptide for calmodulin, which binds to Ca^{2+} and is involved in a number of Ca^{2+} mediated reactions. They reported that caged peptides inhibit this calmodulin-dependent activity weakly, whereas uncaged version of same peptide inhibit nearly 2 orders of magnitude more.

The mechanism of protein folding is poorly understood and very difficult to study. Most of the methods applied, such as stopped-flow mixing or hydrogen-deuterium exchange, have time resolution of 0.1 ms. Hansen and co-workers[38] proposed a method to study these complex processes by using the benzoin group. They studied the time dependent changes of α-helix formation using benzoin protected villin. A small loop was formed by the N-terminus of the peptide with a cystein residue on the side chain of one internal amino acid using benzoin as the linker (Figure 1.3). This cross link prohibited the folding. When irradiation with light, cleavage of that link takes place and the protein forms an α-helical structure. These refolding processes were monitored by time-resolved photoacoustic calorimetry. The great advantage here is that no denaturant is used and protein folding can be observed in the natural environment.

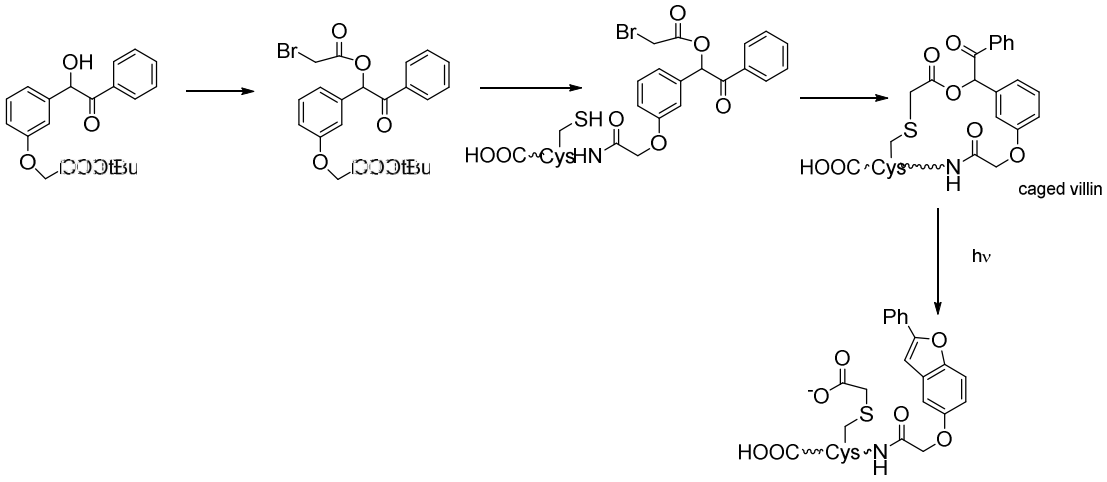


Figure 1.3. Strategy for synthesis of caged villin and photolysis[38]

1.4.3. Light-Directed Synthesis of High Density Arrays of Peptides and Oligoneucletides (Biochips)

This is one of the most interesting applications of photoremovable protecting groups. Application of this was first reported by Fodor and co-workers for combinatorial synthesis of peptides using 6-nitroveratryloxycarbonyl (NVOC) PPG.[31] This is a combination three areas, solid phase synthesis, photoremovable protecting groups, and photolithography. In this process a protected building block is attached to a solid support. As show in Figure 1.4, a substrate **S** bears amino groups that are blocked by PPG **X**. Irradiation of specific region with light through a mask $\mathbf{M_1}$ leads to deprotection of protecting groups X. Amino groups in those areas of the substrate are now free to couple with building blocks protected with PPG X as A-X**.** Different mask $\mathbf{M_2}$ is use to active different region of substrate. Then second building block B protected by **X** is added and attached to the newly exposed area. Repetition of irradiation and coupling steps using mask with variable pattern and different building blocks leads to the synthesis of desired polypeptide biochip.

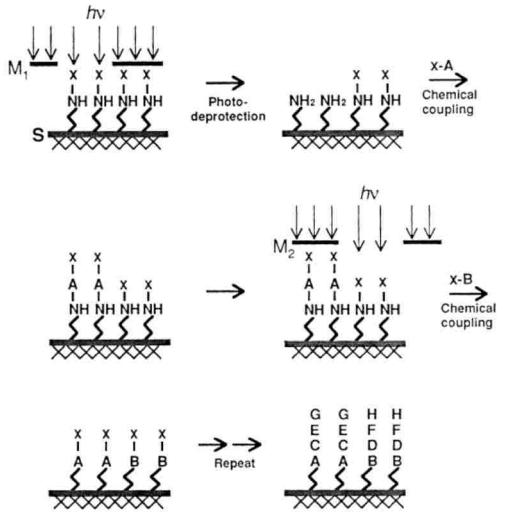


Figure 1.4. Light directed synthesis of high density array of peptide.

1.4.4. Applications of Caged Ca^{2+}

Ionized calcium Ca^{2+} is the single most important information carrier in cells. Changes in Ca^{2+} concentration control a variety of cellular functions including muscle contraction, secretion of neurotransmitters, and enzymes, gene transcription, synaptic plasticity, movement of cells, and wound healing. PPGs for Ca^{2+} allow the rapid concentration changes of Ca^{2+}. It cannot be caged by derivatization like other caged molecules. Usually Ca^{2+} is caged by complexation of chelator molecule that can change the affinity of Ca^{2+} upon irradiation. Some available Ca^{2+} chelators are given in Figure 1.5. Azid-1 is the novel caged calcium chelator developed by Adams *et al*[39]

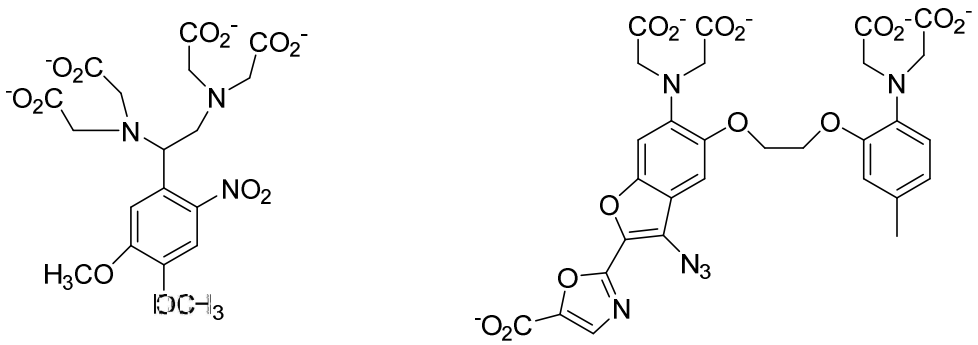
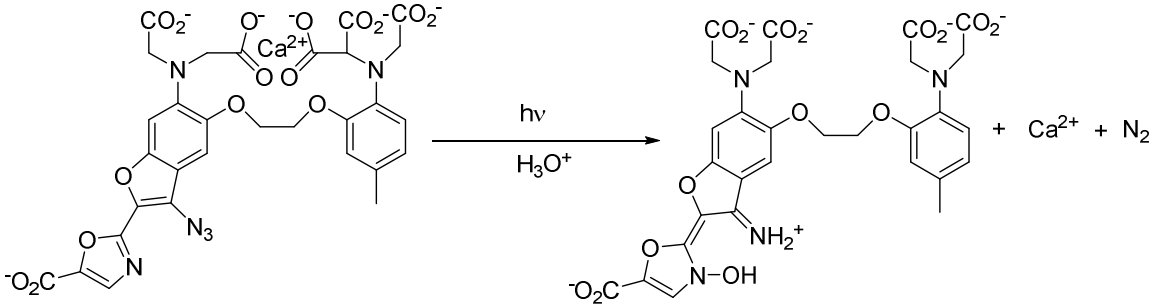


Figure 1.5. Calcium chelators

Scheme 1.8. Photorelease of Ca^{2+} from Azid-1



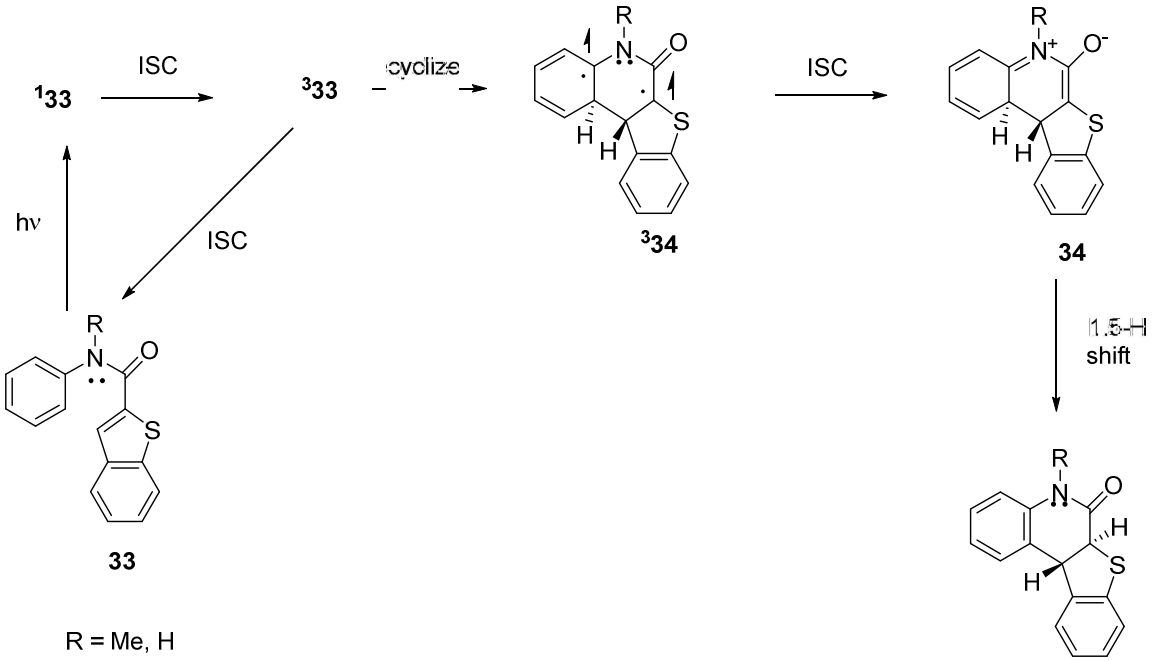
Azid-1 binds a calcium with a dissociation constant (Kd) of 230 nM which, changes to 120 μM by irradiation with UV light (330-380 nm). The photolysis of Azid-1 release Ca^{2+} with unit quantum yield (Scheme 1.8). The photolysis of azid-1 releases N_2 to form nitrinium ion, which reacts with water to form an amidoxime cation The electron withdrawing ability of this cation reduces the chelator's Ca^{2+} affinity. Long term changes in synaptic efficacy is thought to be the neuronal basis for learning and memory. They studied long term depression in rat cerebellar slices by monitoring synaptic current of the Purkinje cell while simultaneously introducing Azid-1.

1.5. Background on Zwitterionic Intermediates with the Expulsion of Leaving Groups

Numerous studies on photochemical electrocyclic ring closure reaction which generate zwitterionic intermediates have been reported over the past 40 years.[40] Most of them involve cyclization of a 6π electron system with an amide functional group. According to Scheme 1.1, N-aryl-substituted benzothiophene carboxamides can undergo electrocyclic ring closure to give zwitterionic intermediates. Various leaving group anions of biological importance can be released from the zwitterionic intermediates. The electrocyclic ring closure should be a photochemically allowed conrotatory process to form a six-membered ring. This conrotatory mode has been established by Witkop and coworkers[41] (Scheme 1.9). Because of the absence of a leaving group at the C-3 carbon, their study provides information on this aspect. For their example the initial cyclization is followed by suprafacial 1,5-H migration, to give a tetracyclic product. The conrotatory motion involved in the first step of the reaction is inferred from the stereochemistry at the ring junction in the photoproduct. Although Witkop's benzothiophene **33** does not have a leaving group at the C-3 position, Castle and coworkers[42] reported studies that have

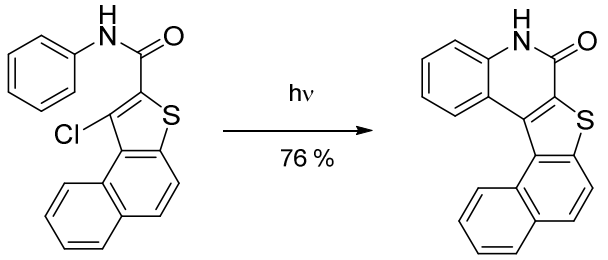
chloride as a leaving group at the C-3 position of the benzothiophene ring (Scheme 1.10). The focus of the research by Castle, however, was to take advantage of the photocyclization reaction for the synthesis of heterocyclic ring systems, which is useful for medicinal chemistry.

Scheme 1.9. Witkop's Mechanism of Conrotatory Electrocyclic Ring Closure Reaction of Benzothiophene Carboxanilide[41]



The photochemical reaction observed by Castle and co-workers is essentially the same as that studied in this project, although neither the mechanism nor quantum yields were studied. Furthermore, the triplet multiplicity was never specified or established experimentally, by either Witkop or Castle.

Scheme 1.10. Photocyclization of Naphthothiophene Anilide[42]



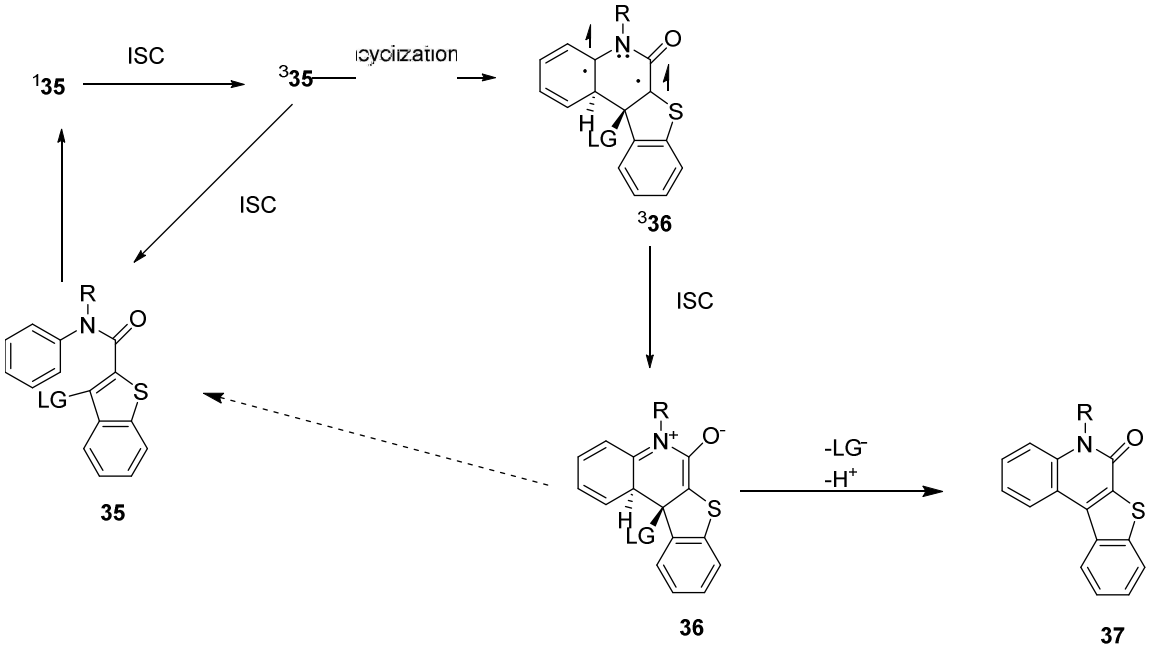
The dependency of Φ on the basicity of leaving group was studied by Sarker and co-workers.[11] A variety of leaving groups (Cl^-, $PhCH_2CO_2^-$, PhS^-, $PhCH_2S^-$, PhO^-) attached at the C-3 position in the benzothiophene ring could be released with various quantum yields, and in high chemical yields upon photolysis at λ = 310 nm. (Scheme 1.11). They reported that carboxanilide **35** could release various leaving groups at 310 nm that vary in basicity in essentially quantitative yields to form **37**. However**,** quantum yields decreased with increasing basicity of the leaving group (Figure 1.3). Quantum yields were over the range 0.23-0.007 (LG^- = Cl^-, $PhCH_2CO_2^-$, PhS^-, $PhCH_2S^-$, PhO^-) (Table 1.1). Dependence of Φ on LG^- basicity is consistent with the formation of ground state intermediate **36**, which expels leaving group in competition with ring opening to give starting material. Quenching studies and a heavy atom effect indicated that the reaction takes place through a triplet excited state. Even though the photolytic wavelength is low, the ability to release relatively basic leaving groups such as thiolates and phenolates is advantageous for biological studies.

Table 1.1. Quantum Yield for Releasing Different Leaving Groups (LG^-) from Benzothiophene Carboxanilide

LG^-	Cl^-	$PhCH_2CO_2^-$	PhS^-	$PhCH_2S^-$	PhO^-	HO^-
Φ	0.23	0.16	0.10	0.075	0.074	0.007

.

Scheme 1.11. Mechanism of Releasing Leaving Groups from Benzothiophene Carboxanilide[11]



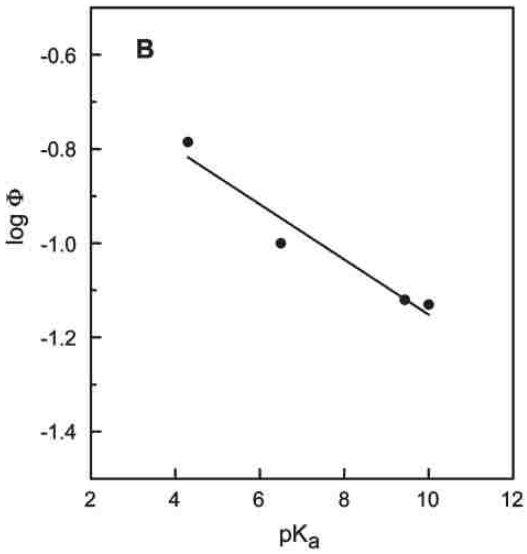
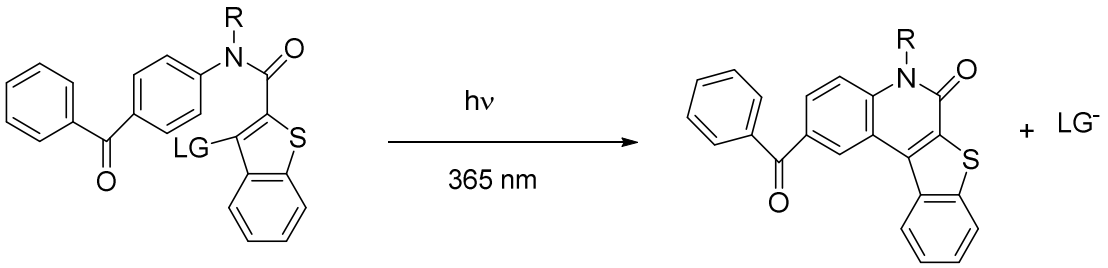


Figure 1.6. Dependence of Φ on LG^- basicity[11]

By incorporating a *p*-benzoyl group onto the benzene ring of anilide **35** (Scheme 1.12) the photolytic wavelength could be extended to 365 nm. The quantum yield was only somewhat lower compared to benzothiophene carboxanilide **35**.

Scheme 1.12. Photorelease of Leaving Group from Benzothiophene Carboxamide with Benzophenone Chromaphore



38

As given in Scheme 1.2, incorporating thioxanthone as the chromophoric group they could extend photolytic wavelength to 385 nm.[10] Without any substituent in compound **3a** in the benzothiophene moiety they reported the formation of two products by cyclization at the C-1 and C-3 positions (Scheme 1.2) of the thioxanthone ring system to form **8** and **9** in a 42:58 ratio in aqueous phosphate buffer in acetonitrile. When substituent Y was a carboxylic or methyl ester at the C-6 position of benzothiophene **3c** the only product formed was via cyclization at the C-1 position (**8c)**. By incorporating a carboxylic acid group at the C-6 position they also were able to increase the aqueous solubility by a considerable amount. Product quantum yields for this system are given in Table 1.2. Decreasing quantum yield in the presence of oxygen and piperyline indicated a triplet excited state reaction. Involvement of a triplet excited state was further supported by incorporating a heavy atom, bromine at C-7 position of thioxanthone ring system. When a heavy atom is in the molecule, it increases the quantum yield for the reaction. Similar to benzothiophene carboxanilide, this system also show decrease in

quantum efficiency when increasing leaving group basicity. However it could be increase the photolytic wavelength and solubility, the disadvantage of this system is the low quantum yield compare to benzothiophene carboxanilide **35** and compound with benzophenone chromophore **38**.

Table 1.2. Quantum Yields for Compound **3a-c**

Reactant	**LG^-**	**Solvent**	**Φ**
3a	Cl^-	N_2 saturated	0.069
3b	Cl^-	N_2 saturated	0.039
	Cl^-	O_2 saturated	0.019
3c	Cl^-	N_2 saturated	0.034
	PhS^-	N_2 saturated	0.017
	$PhCH_2^-$	N_2 saturated	0.011
	HS^-	N_2 saturated	0.008

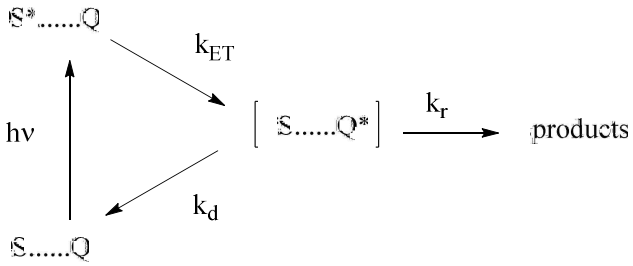
1.6. The Sensitized Release of Leaving Group

Energy transfer is the process by which the excitation energy of an excited state molecule (Sensitizer or Donor) is transferred to a neighboring molecule (Quencher or Acceptor). PPGs can be cleaved to release leaving groups by two different methods, direct irradiation and sensitized irradiation. Direct irradiation involve a PPG which is light sensitive and which upon irradiation, undergoes chemical change which releases the protected molecule. Here the light absorption and bond cleavage take place within the same molecule. Sensitized irradiation utilizes an external molecule, called a sensitizer, to absorb light and transfer its energy to generate the triplet excited state of the PPG

quencher which would then undergo chemical transformation and release a leaving group. The two components can be linked together or just separated in the media.

As noted above, to make the energy transfer efficient, the energy transfer process must be exergonic. In addition, the sensitizer should be able to undergo efficient intersystem crossing, in other words, the triplet quantum yield $^3\Phi$ should be high enough to populate enough triplet excited states and the triplet lifetime should be long enough to enable the triplet energy transfer.[44] The general process is shown in Scheme 1.13.

Scheme 1.13. General Scheme for Photosensitization



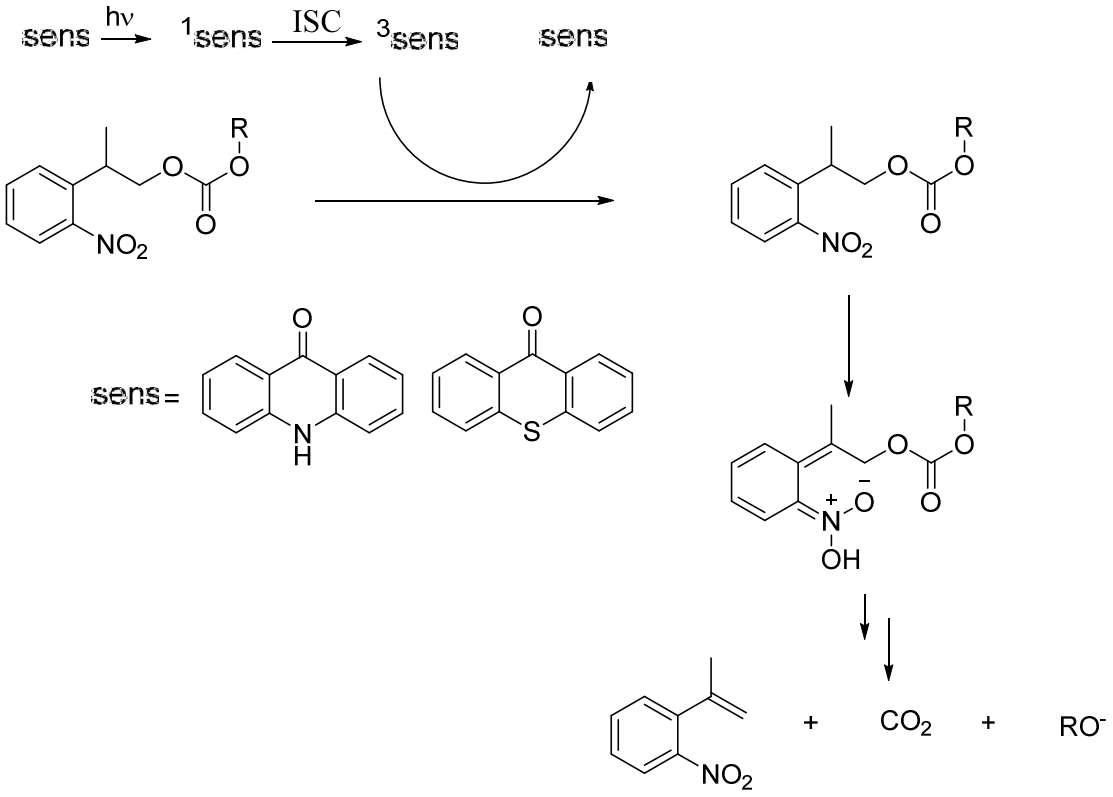
… = bond linker or separation of two components

Q = PPG, S = sensitizer

An example of intermolecular energy transfer has been provided by Steiner, Creen, and co-workers.[44] They reported that a sensitizer such as thioxanthone can increase the sensitivity of 2-(2-nitrophenyl)propyl group to two-photon excitation (Scheme 1.13).

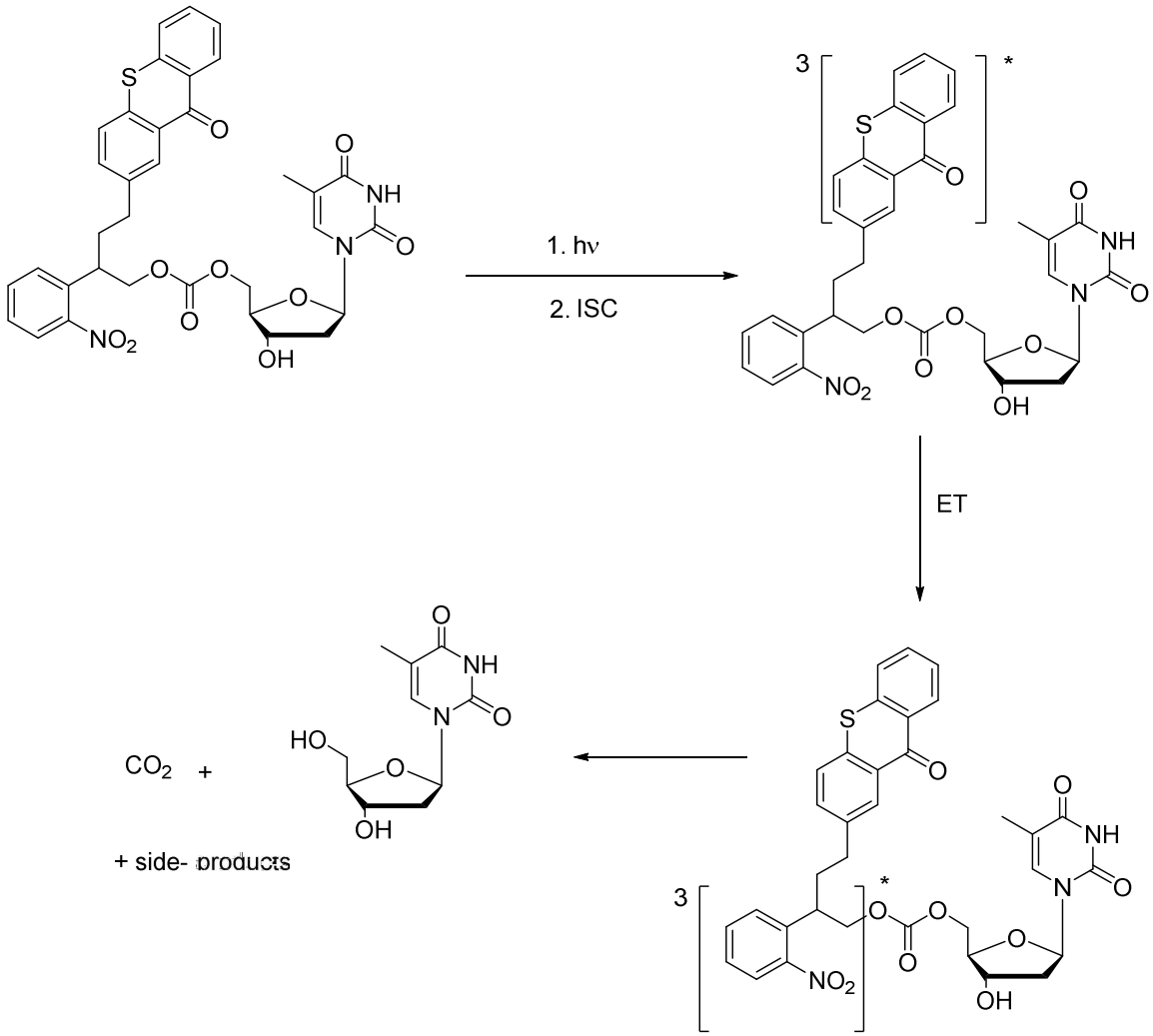
Wöll *et al*[45] reported intramolecular triplet sensitization of 2-(2-Nitrophenyl)propyl chromophore by thioxanthone.(Scheme 1.14).here the sensitizer is directly attached to the photoactive chromophore.

Scheme 1.14. Intermolecular Triplet Sensitization of the 2-(2-Nitrophenyl)propyl Chromophore[14,44]



In the current project, the chromophore, thioxanthone is attached to the nitrogen of an amide linker. The results of sensitized photolysis can complement those of direct photolysis, where the chromophore intramolecularly transfers its triplet energy to the reactive aromatic thiophene site that eventually releases the leaving group. The intermolecular energy transfers (sensitization) are also possible. Study on the efficiency of energy transfer between thioxanthone and naptho[1,2-b]thiophene also will be discussed in Chapter 2.

Scheme 1.15. Intramolecular Triplet Sensitization of the 2-(2-Nitrophenyl)propyl Chromophore[14,45]



CHAPTER 2

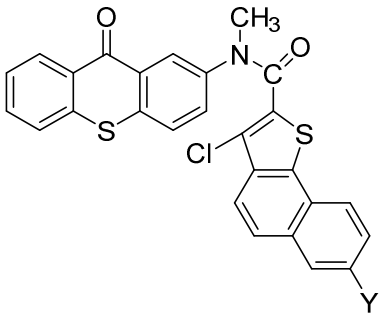
PHOTOCHEMICAL ELECTROCYCLIC RING CLOSURE AND LEAVING GROUP EXPULSION FROM N-(9-OXOTHIOXANTHENYL)NAPHTHO[1,2-b]THIOPHENE CARBOXAMIDES AND SENSITIZED RELEASE OF LEAVING GROUPS FROM NAPHTHO[1,2-b] THIOPHENE ANILIDE BY THIOXANTHONE SENSITIZER.

2.1 Introduction

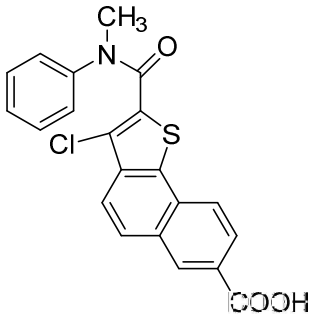
As mentioned in the Chapter 1, benzothiophene carboxamide, **3** which contains thioxanthone as a chromaphoric group expels leaving groups inefficiently by photolysis. Quenching studies indicate that the electrocyclization occurs in the triplet excited state to produce a triplet excited state diradical that intersystem crosses to give the zwitterionic intermediate. It is thought that energy transfer from triplet excited chromaphore to benzothiophene ring is required for electrocyclization reaction. This triplet excited state is expected to lie below that of the benzothiophene triplet excited state. According to DFT calculations triplet energy transfer to the benzothiophene moiety would then be endothermic by ca. 5k cal mol^{-1}.[1] So efforts have made here to lower the triplet excited state energy of the benzothiophene part of the molecule, so that energy transfer will be exothermic. Naphtho[1,2-b]thiophene has an approximate triplet excited state energy of 62 kcal mol^{-1}.[2] This triplet energy is lower than the benzothiophene triplet energy of ca. 69 kcal mol^{-1}. It was planned to replace the benzothiophene moiety with napthothiophene ring system in order to facilitate the energy transfer from the triplet excited state of thioxanthone of ca. 64-65 kcal mol^{-1}.

In this chapter we will discuss the synthesis of compound **6a, 6b** and photochemistry related to N-(9-oxothioxanthenyl)naphtho[1,2-b]thiophene carboxamide. The direct photolysis of this compound and the identity of photoproduct are reported. Quantum yields for direct photolysis are determined. Comparison is made to the analogues with a benzothiophene ring system in place of the naphthothiophene ring system. Quenching studies are performed to test the multiplicity of the photoreaction.

For comparison, the direct and triplet sensitized photochemistry of anilide **2-1** are studied. The triplet sensitized photolysis provides a means to estimate the efficiency of intramolecular triplet excitation from the thioxanthone to the naphthothiophene ring system in **2-1**.



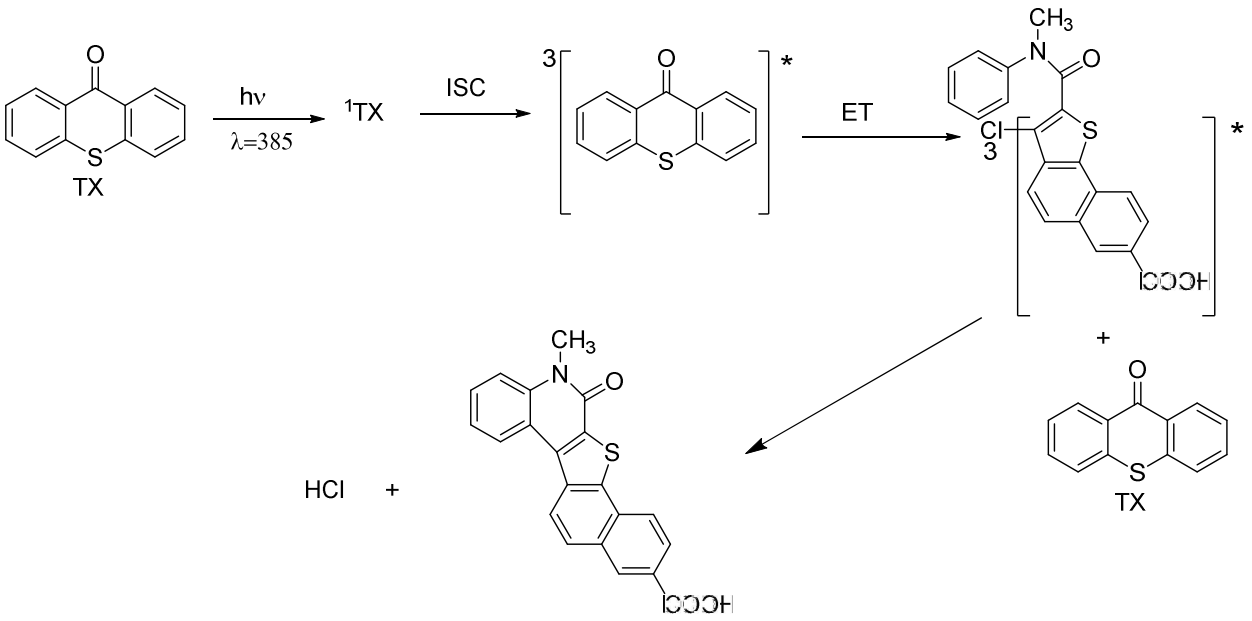
6 (a) (Y= -H)
6 (b) (Y = -COOH)



2-1

Photochemical experiments will be described that estimate the efficiency of intramolecular triplet excitation transfer from the thioxanthone to the naphthothiophene (Scheme 2.1).

Scheme 2.1 Sensitized Photolysis of Naphtho[1, 2-b] thiophene and Electrocyclic Ring Closure Reaction by Thioxanthone Sensitizer



2.2 Results and Discussion

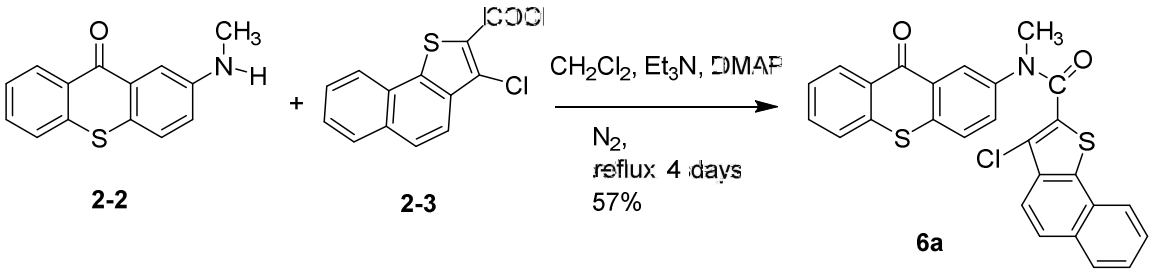
2.2.1 Synthesis of 3-chloronaptho[1,2-b]thiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide 6a

The napthothiophene carboxamide, **6a** was synthesized by coupling 2-methylaminothioxanthone **2-2** with acid chloride **2-3** (Scheme 2.2). Synthesis of amine **2-2 (**Scheme 2.3**)** involved six steps starting from thiophenol with 5-nitro-2-chlorobenzoic acid to form **2-4** which underwent cyclization reaction to give the nitro compound **2-5**. The nitro compound **2-5** was reduced to amine **2-6** with iron powder.[3] The amine was converted to the amide **2-7** by reaction with acetic anhydride. Then the amide **2-7** was alkylated to obtain the N-methyl amide **2-8**. Hydrolysis of **2-8** furnished the 2-methylaminoxanthone-9-one **2-2**.

Synthesis of the acid chloride **2-3** (Scheme 2.4) involved 2 steps. Commercially available naphthalene-2-carboxaldehyde gave 3-(2-naphthyl)propenoic acid **2-9** with the

malanoic acid[4], which was converted to 3-chloronaphtho[1,2-b]thiophene-2-carbonyl chloride **2-3** by refluxing with $SOCl_2$.

Scheme 2.2 Synthesis of 3-chloronaptho[1,2-b]thiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide **6a**



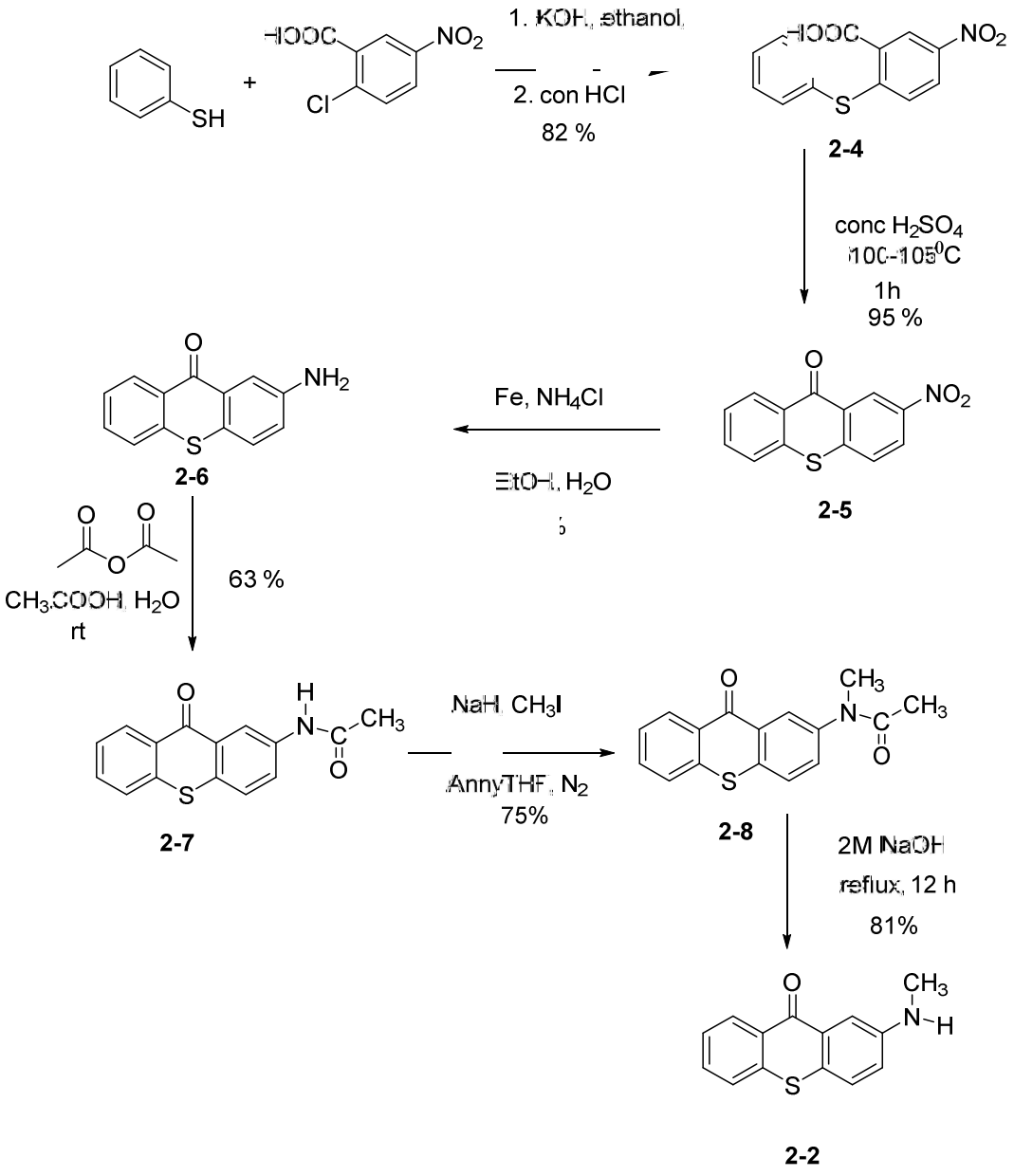
2-2 **2-3** **6a**

2.2.2. Photolysis of Compound 6a

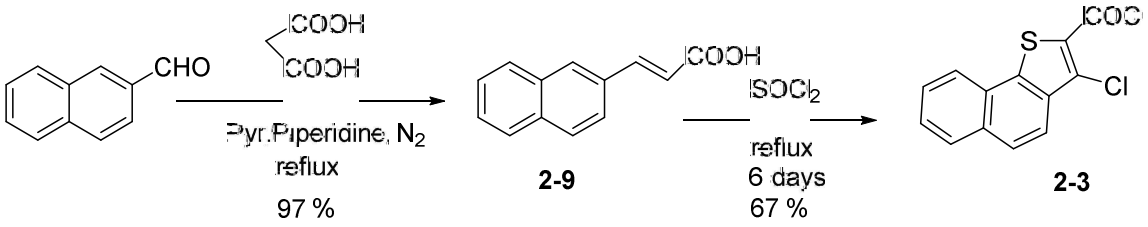
It is important for biological applications that caged biomolecules bearing a photoremovable protecting groups have appreciable solubility in aqueous buffered media.[5,6] Compound **6a** was sparingly soluble in DMSO. Attempts were made to dissolve compound **6a** in DMSO-d_6 and then filtered to obtain a clear solution. The resultant clear solution was treated with two drops of pH = 7 phosphate buffer. After adding the buffer a turbid solution was produced. A nitrogen saturated sample of 20 mg of **6a** in 2 mL DMSO-d_6, filtered through a syringe filter, was photolyzed using Pyrex-filtered light from a Hanovia 450 W medium pressure mercury lamp. A single photoproduct either **2-10** or **2-11,** (Scheme 2.4) was observed after 30 min. After 1.5 h 50% conversion was achieved. The product was distinguished from photoreactant by its N-methyl peak in the ^{1}H NMR spectrum which was shifted downfield from δ 3.64 to δ 3.98. Due to the above solubility problem, the quantum yield was not determined. But the high conversion of the photoreactant to the photoproduct in DMSO-d_6 provided the incentive to increase the solubility of compound **6a** in aqueous buffered media by

attaching the carboxylic acid group to the naphthothiophene ring at C-6 position, as with compound **6b**.

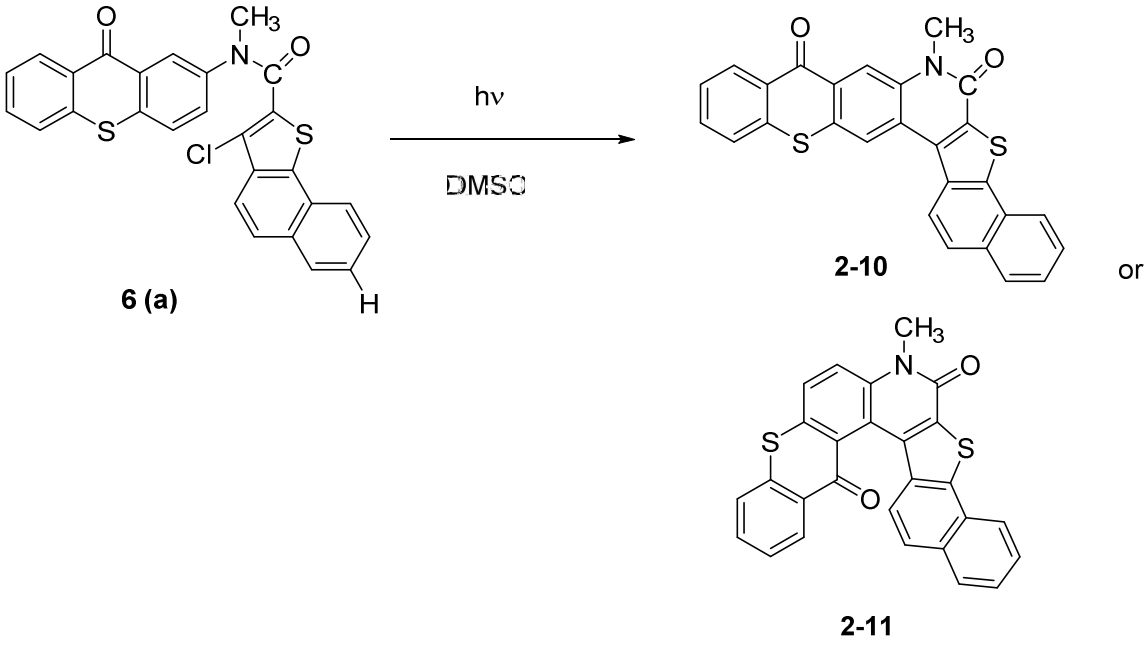
Scheme 2.3 Synthesis of Compound **2-2**



Scheme 2.4 Synthesis of Compound **2-3**



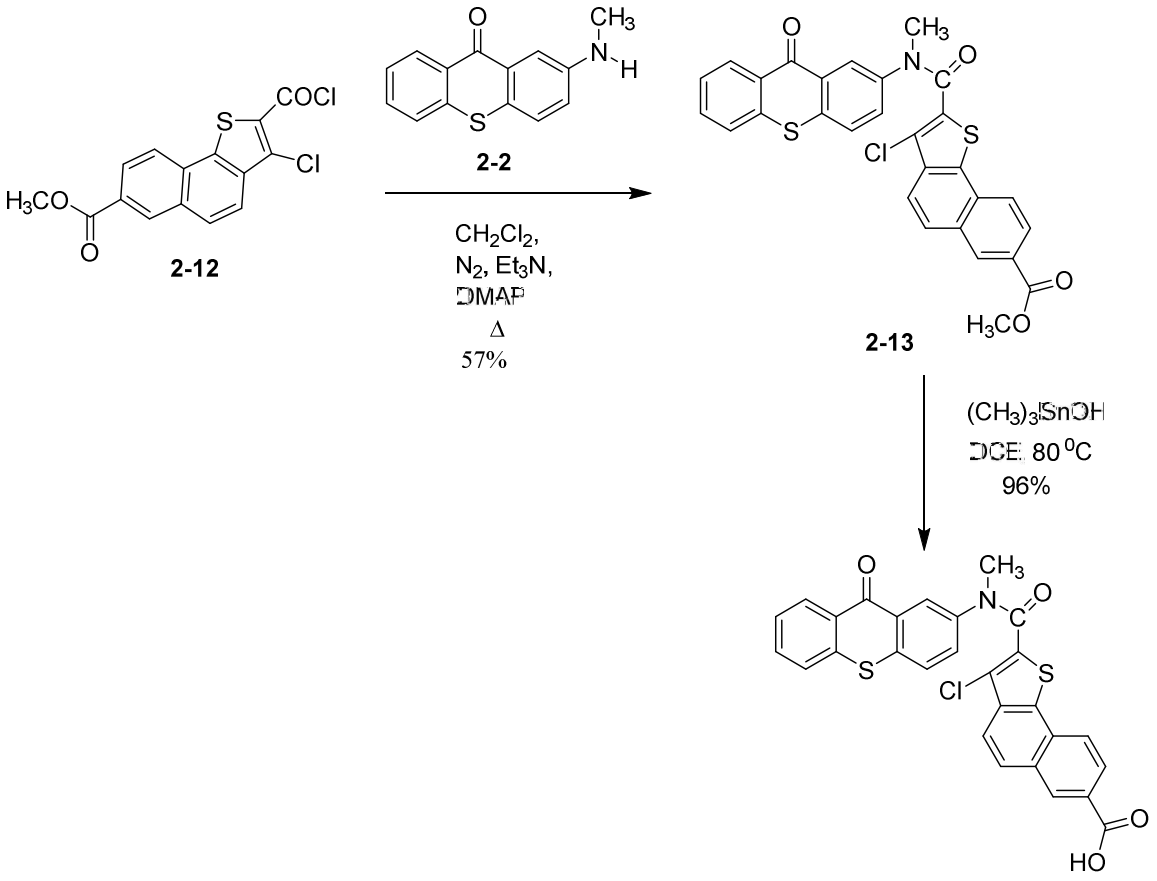
Scheme 2.5. Photolysis of Compound **6a**



2.2.3. Synthesis of Photoreactant 6b

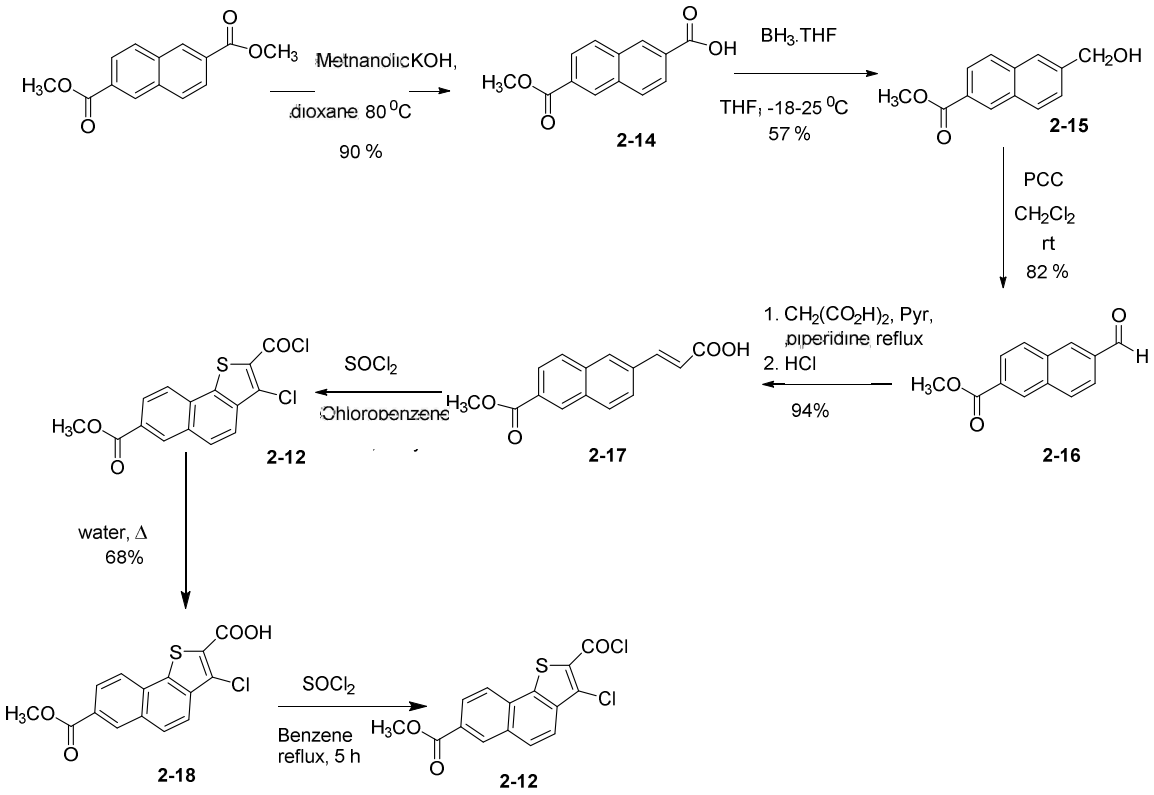
The synthesis of the compound **6b** is given in the Scheme 2.6. The compound **6b** was synthesized by a coupling reaction between the amine **2-2** and the acid chloride **2-12** followed by demethylation of the methyl ester **2-13** using trimethyltin hydroxide.

Scheme 2.6. Synthesis of Photoreactant **6b**



The synthesis of **2-12** involves 5 steps starting from commercially available dimethyl 2,6-naphthalenedicarboxylate (Scheme 2.7). Partially hydrolysis of dimethyl 2,6-naphthalenedicarboxylate with methanolic KOH gave 6-cabomethoxy-2-napthalenecarboxylic acid **2-14**. The carboxylic acid group of **2-14** was reduced to the alcohol **2-15** using BH_3.THF complex followed by PCC oxidation to produce aldehyde **2-16**. Condensation of **2-16** with malonoic acid gave 3-(2-napthyl) propenoic acid [4] **2-17**, which was converted to the acid chloride **2-12** by refluxing with $SOCl_2$. Hydrolysis of compound **2-12** gave the corresponding carboxylic acid **2-18** which was converted to the acid chloride **2-12** prior to the coupling reaction with amine **2-2**to form **6b.**

Scheme 2.7 Synthesis of Acid Chloride **2-12**



2.2.4. UV Spectra of Compound 6b

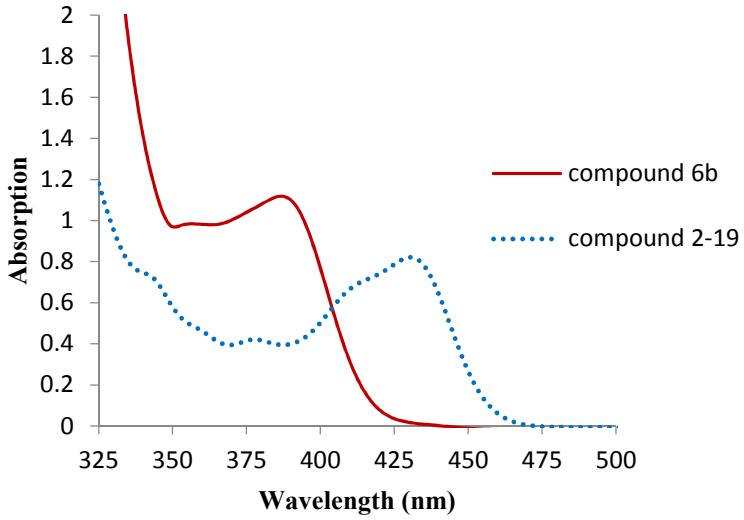


Figure 2.1. UV spectra of 2.45×10^{-4} M solution of compound **6b** and 2.02×10^{-4} M photoproduct **2-19** in 33% aq dioxane containing 100 mM pH 7 phosphate buffer.

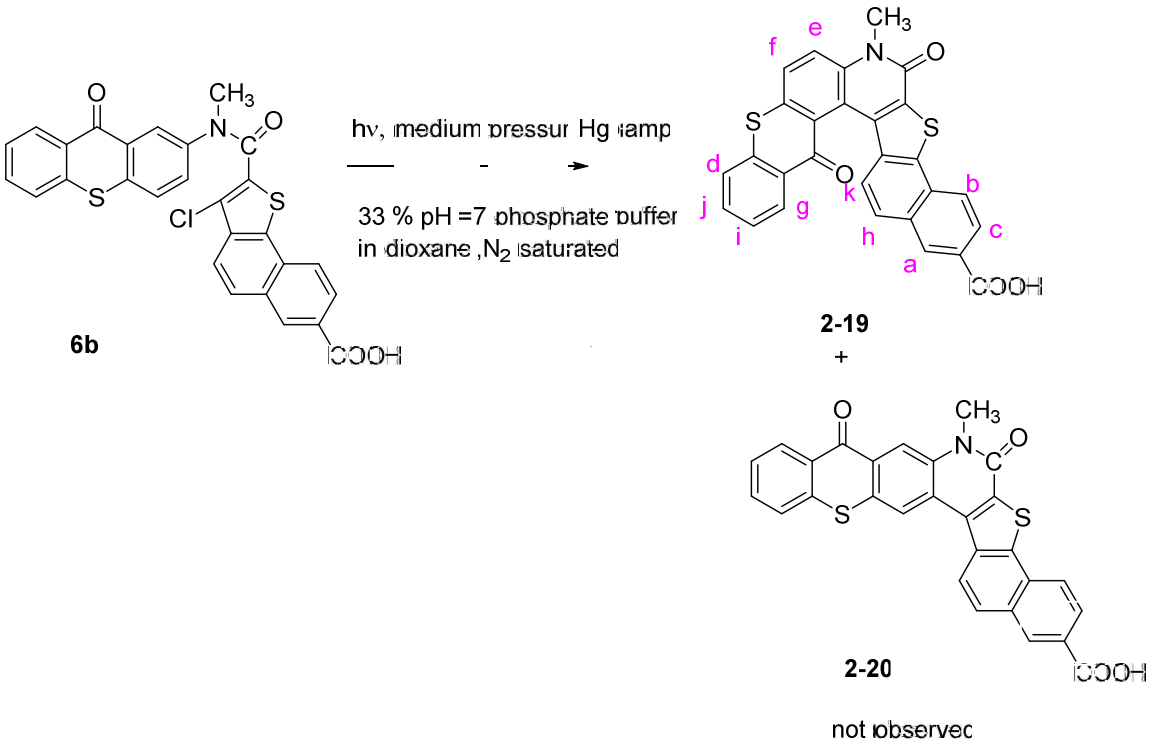
The absorption spectrum of the compound **6b** was recorded in 33% 100 mM pH 7 phosphate buffer in dioxane (Figure 2.1). The absorption maximum at 386 nm had ε = 4707 M^{-1} cm^{-1}. The compound was photolysed at this wave length for the subsequent photochemical studies.

2.2.5. Preparative Direct Photolysis of Compound 6b

Preparative direct photolysis of 50 mL of 9.40 x 10^{-3} M of **6b** in argon saturated, buffered aq dioxane with a 200 W medium pressure mercury lamp equipped with a Pyrex filter gave a single photoproduct in 64% yield after 3 h (Scheme 2.8). Solid photoproduct was collected by filtration at time intervals during the photolysis. Solid photoproduct adhered onto the vessel surface during photolysis ultimately limiting the conversion achieved in the photolysis. ^{1}H NMR spectroscopy of the precipitated product showed a single peak at δ 3.88 ppm corresponding to the *N*-methyl group. NMR analysis of the remaining solution after concentration to remove all solvent showed the same spectrum. The photolysis thus appeared to produce a single regioisomeric photoproduct. In this regard, **6b** showed a similar photochemical outcome as the previously studied benzothiophenes **3b** and **3c**, which gave a single regioisomer, while benzothiophene **3a** showed a pair of *N*-methyl signals in the ^{1}H NMR spectrum corresponding to two regioisomeric photoproducts produced upon direct photolysis[1]. It was initially believed that the structure of the photoproduct of **6b** was **2-19** rather than **2-20** (Scheme 2.8) on the basis of precedent.[1] Repeated attempts failed to produce satisfactory crystals for X-ray diffraction to elucidate the structure. However, good evidence for the structural assignment of the photoproduct as **2-19** was instead provided by the 400 MHz ^{1}H NMR COSY spectrum, which showed six vicinal couplings as cross peaks corresponding to

proton pairs b-c, h-k, g-i, i-j, j-d, e-f (Scheme 2.6) within the benzenoid rings of structure **2-19**. If the structure were **2-20**, only five vicinal couplings would have been observed, and such a structure would not account for the presence of the extra cross peaks observed in ^{1}H NMR COSY which are readily accommodated by vicinal protons e-f of structure **2-19 (**See Appendix-1 -Figure 33 for COSY).

Scheme 2.8 Photochemical Electrocyclization of Compound **6b**



2.2.6. Quantum Yield Determination for Compound 6b

Quantum yields for the formation of product were determined by direct photolysis of **6b** at 386 nm. ^{1}H NMR spectroscopy was initially used to quantify the photoproduct by integrating the *N*-CH_3 protons against an *N*-CH_3 signal of DMF added as standard. For three runs the average quantum yield was $\Phi = 0.083$ for 56% conversion. The quantum yields did not vary significantly with % conversion. Nor did the quantum yields

vary with the concentration of reactant. At the relatively high 1.7 – 3.3 x 10^{-3} M concentrations of **6b** used for the NMR determinations, most of the photochemistry would occur at the front face of the cell, possibly lowering the efficiencies due to competitive absorption of light by the photoproduct. However, the quantum yields were found to be essentially unchanged at > 10-fold lower concentrations of 1.0 x 10^{-4} M reactant. At the lower concentrations of **6b**, $\Phi = 0.084$ for duplicate measurements at 16.4% conversions. For these latter runs, the photoproduct was quantified by absorption spectroscopy with use of a calibration curve (Figure 2.2), constructed from known mixtures of reactant and product. The similar Φ values found for the two different concentrations suggest that internal filter effect by product formed at the front face of the cell was insignificant, because the photoproduct absorption is minimum at photoreactant's absorption wavelength (Figure 2.1).

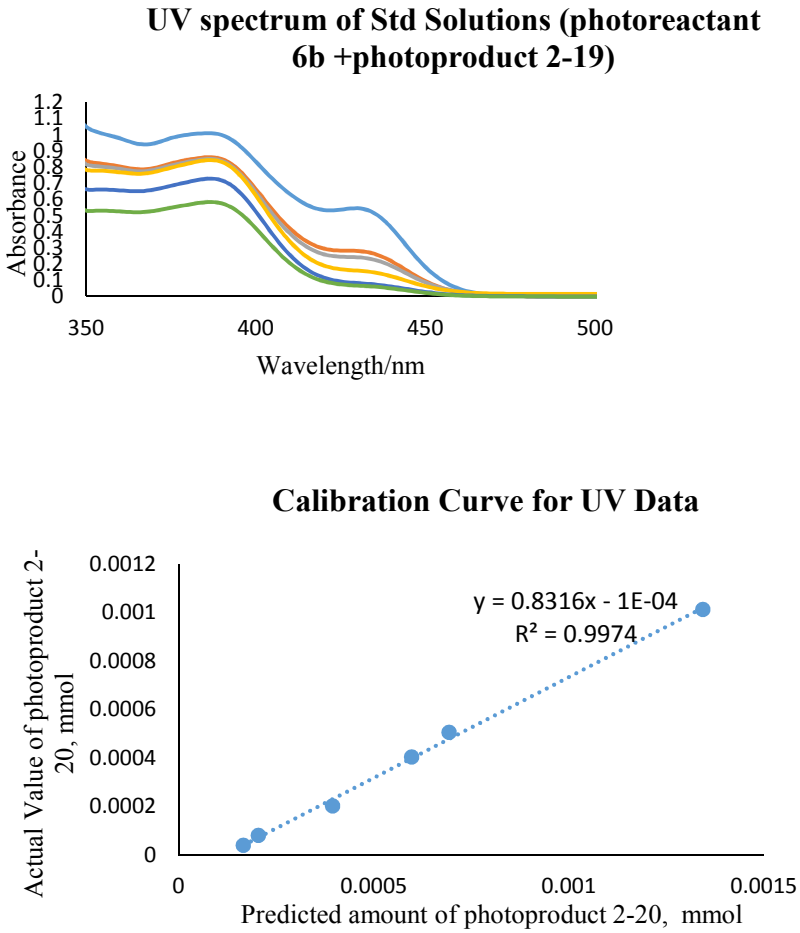


Figure 2.2. Calibration curve for compound **2-19** using absorption spectroscopy

2.2.7. Quenching Studies of Photoreactant 6b

For the benzothiophene **1** and **3** the photochemistry was shown to occur in the triplet excited state, as evidenced by efficient quenching of the reaction by the triplet quencher 1,3-pentadiene.[2,7] This conclusion was additionally supported by our computational study, which delineated the progress of the triplet excited state reaction on the the potential energy surfaces[2]. In the case of napthothiophene **6b** repeated attempts failed to quench the triplet excited state by 1,3-pentadiene. However, naphthothiophene **6 b** photoreaction is quenched inefficiently at high concentrations of the triplet quencher,

cyclohexadiene. Thus, the quenching evidence for a triplet excited state is less definitive than was the case for **1** and **3**. The data points show a poor fit to straight line and show significant curvature at low concentrations of quencher, and inefficient quenching is observed at high concentrations. This quenching profile (Figure 2.3) also is suggestive of the involvement of two triplet excited states in the reaction. A long-lived triplet excited state appears to be quenched at very low concentrations of cyclohexadiene. A very short-lived triplet (ca. 2 ns) excited state which would account for the inefficient quenching observed at the higher concentrations of cyclohexadiene. Whereas the longer-lived triplet excited state may reside on the thioxanthone, which would normally have a long microsecond triplet lifetime in the case of the isolated molecule in solution (13.3 μs in methanol and 6.7 μs in CH_3CN)[8], the short-lived triplet excited would likely reflect the occurrence of a facile reaction that deactivates the triplet excited state. Such a facile reaction could be the cyclization of the thiophene ring with the thioxanthone benzo group. Inefficient quenching behavior is observed with anilide **2-1** at all concentrations of the quencher. From the Stern-Volmer slope ($k_q\tau$) of 70.7.M^{-1}, $^3\tau$. = ca. 7 ns.

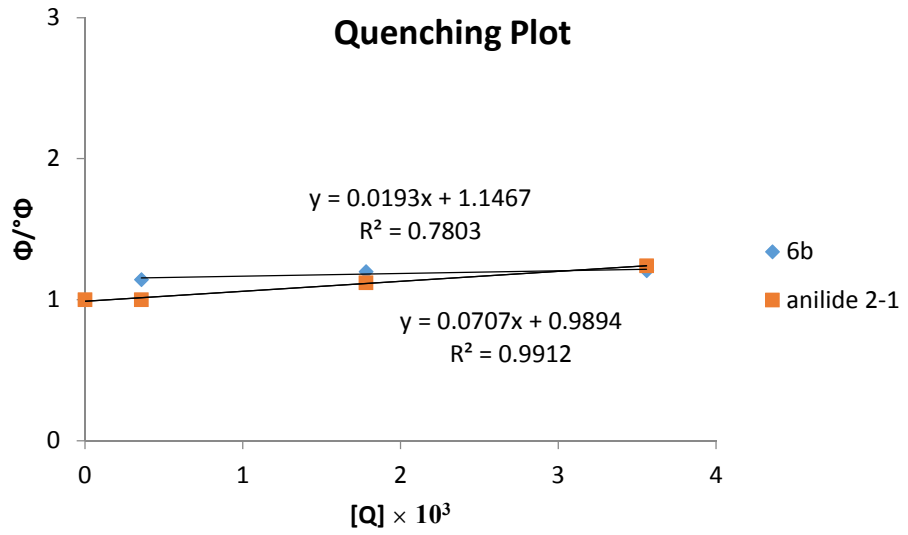


Figure 2.3. Quenching of amide **6b** by 1,3-cyclohexadiene, and quenching of anilide **2-1** by 1,3-cyclohexadiene. **Note**: Benzothiophene systems (anilide and thioxanthone) readily quenched with pyperylene. Life times of triplet excited states are 932 ns and 7µs respectively.[2,7]

2.2.8. Synthesis of Anilide 2-1

The anilide derivative **2-1** of naphthothiophene-2-carboxylic acid **2-18** was synthesized for use as a triplet energy acceptor in sensitized photolysis that were performed with xanthone and thioxanthone to estimate the efficiency of intramolecular triplet energy transfer from the thioxanthone moiety to the naphthothiophene-2-carboxamide ring in reactant **6b**.

The anilide was also need for comparison of quenching to compound **6b** by cyclohexadiene. Synthesis of photoreactant **2-1** involves coupling of acid chloride **2-12** and N-methylaniline followed by demethylation of the ester **2-21** with trimethyltin hydroxide (Scheme 2.9). Characterization of compound **2-1** was done by ^{1}H NMR, ^{13}C NMR, HRMS and elemental analysis. The absorption spectrum of the acid **2-1** (Figure

2.4) showed a shoulder at 352 nm (ε= 1730 M^{-1} cm^{-1}) and an intense, lower wavelength absorption in aq.in 25% aq. dioxane containing 100 mM phosphate buffer.

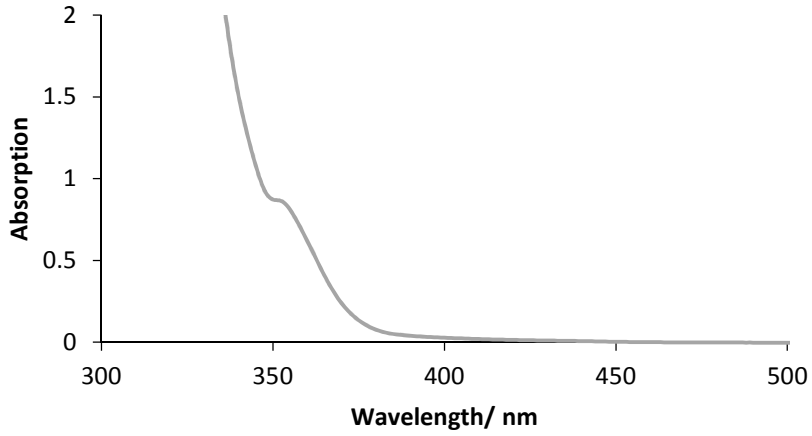
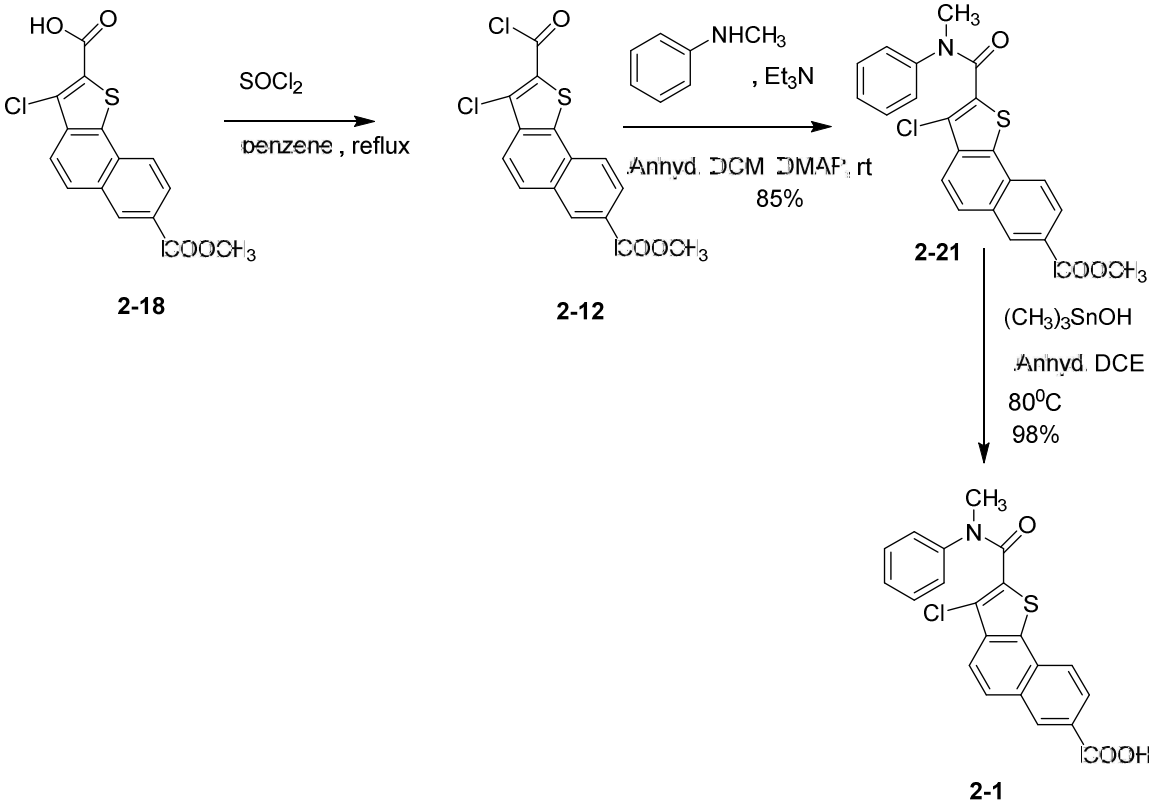


Figure 2.4. UV spectra of 5.00×10^{-4} M solution of compound **2-1** in 25% 100mM pH 7 phosphate buffer in dioxane.

Scheme 2.9. Synthesis of Anilide **2-1**



2.2.9. Preparative Direct Photolysis of Photoreactant 2-1.

For preparative photolysis, Pyrex-filtered light from a Hanovia medium pressure mercury lamp was used. Preparative photolysis of 10^{-2} M solution of the acid **2-1** in nitrogen saturated 25%dioxane containing 100 mM phosphate buffer (pH =7) resulted in nearly quantitative expulsion of the chloride leaving group and formation of the photoproduct **2-22** as quantified by ^{1}H NMR integration against DMF as an internal standard (Scheme 2.10). The photoproduct **2-22** was identified by ^{1}H NMR spectroscopy ^{13}C NMR, HRMS and elemental analysis. The absorption spectrum of 2-**22** (Figure 2.5) showed a long wavelength maximum at 373 nm (ε = 9000 M^{-1} cm^{-1}) in aq. dioxane containing phosphate buffer.

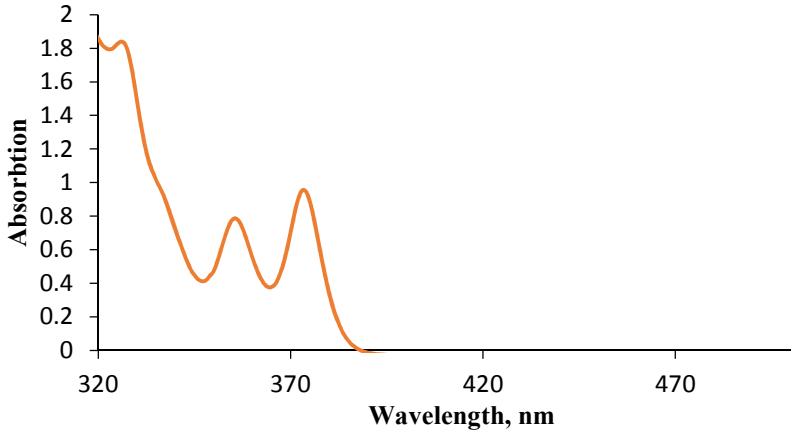
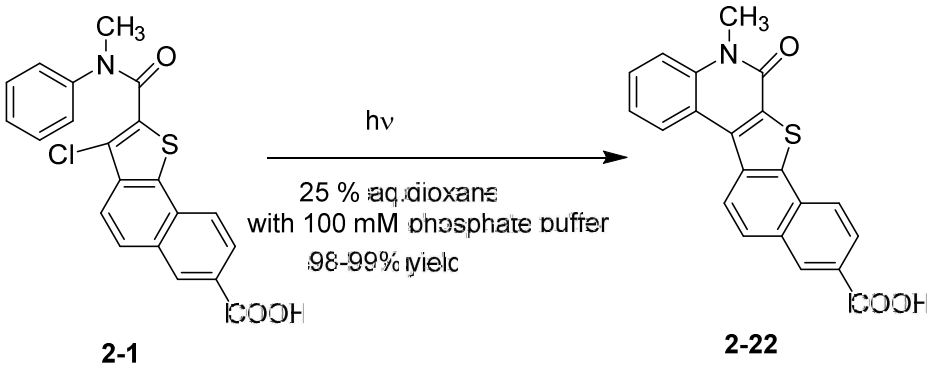


Figure 2.5 UV spectra of 1.00×10^{-4} M solution of photoproduct **2-22** in 25% 100 mM, pH 7 phosphates buffer in dioxane.

Scheme 2.10 Photolysis of Photoreactant **2-1** in 25% Phosphate Buffer in Dioxane



2-1 **2-22**

2.2.10. QuantumYields for Direct and Sensitized Photolysis of compound 2-1

The quantum yield for direct photolysis was determine at 365 nm using a high pressure mercury lamp as the light source. The quantum yield for the formation of **2-22** in nitrogen saturated 25% aq. dioxane containing100 mM phosphate buffer was found to be 0.17 according to ^{1}H NMR spectroscopy using DMF as standard.

To estimate the efficiency for triplet excitation transfer from the thioxanthone chromophore to the naphthothiophene ring system in **6**, triplet sensitized photolyses of anilide **2-1** were conducted using xanthone and thioxanthone as triplet energy donors.

The sensitized photolyses of anilide **2-1** give **2-22** as the photoproduct in 4:1 dioxane: water (v/v) containing 100 mM phosphate buffer at pH = 7. The chemical yields were not determined.

The quantum yields for the direct and the xanthone and thioxanthone triplet sensitized photolyses of anilide **2-1** are summarized in Table 2.1. For xanthone as the triplet sensitizer, concentrations of anilide **2-1** were kept sufficiently low so that singlet sensitization would be negligible[9]. Concentrations of xanthone sensitizer and anilide **2-1** also had to be chosen to minimize direct absorption of light by the anilide at the

photolysis wavelength, 365 nm. In addition, a range of photochemical conversions were examined to determine whether possible triplet energy transfers from sensitizer to photoproduct **2-22** formed would lower the observed quantum yield Φ. In the case of thioxanthone a much longer photolysis wavelength of 386 nm could be used, which lessened the latter concern.

From the xanthone and thioxanthone sensitized quantum yields, the efficiency of intermolecular triplet energy transfer from thioxanthone as the sensitizer to the naphthothiophene ring system of anilide **2-1** as the quencher can be estimated.

First, in the case of xanthone as sensitizer the triplet excited state cyclization efficiency, Φ_r, can be calculated using eq. 2.1 with Φ_{isc} determined experimentally (vide infra) and assuming $\Phi_{et} = 1$ for exothermic energy transfer.

$$\Phi = \phi_{isc}\,\phi_{et}\,\phi_r \quad \text{.....eq. 2.1}$$

Table 2. 1. Quantum Yield Data for Direct and Sensitized Photolyses of Anilide **2-1.**

2-1, M	**Sensitizer, M**	**Wavelength, nm**	**Φ_{isc}**	**Φ_{obs}**
2.56×10^{-3}	None	385	n/a[a]	0.17
2.33×10^{-3}[b]	Xanthone, 4.00×10^{-2} [c]	365	0.98	0.27[d]
2.41×0^{-3}	Thioxanthone, 2.50×10^{-3} [e]	386	0.67	0.081

[a]Not applicable. [b]Concentrations of quencher were kept low to avoid singlet energy transfer[9] [c]Xanthone absorbed 85.9% of the light. [d]Correction applied for direct absorption by compound **2-1** up to 14% of incident light. [e]Thioxanthone absorbed 99.8% of the light

This is a reasonable assumption, in light of the high triplet excited state energy of xanthone (E_T = ca. 74 kcal mol^{-1})[10] and the much lower triplet energy estimated for an unsubstituted [1,2-b]naphthothiophene as a model for the energy accepter in compound **6(b) (**the theoretical value is E_T = ca. 62 kcal mol^{-1}). From Φ_{isc} for thioxanthone and Φ_r the intermolecular energy transfer efficiency, Φ_{et} = 0.46 is obtained as an estimate for the donor acceptor pair, thioxanthone and anilide **2-1**.

2.2.11. Triplet Yields for Xanthone and Thioxanthone

The values for Φ_{isc} for xanthone and thioxanthone in aq dioxane containing buffer were determined experimentally by the method of Lamola and Hammond[11] for use in Eq. 2.1 to calculate the Φ_r and Φ_{et} values (Table 1). Their method for obtaining Φ_{isc}, as implemented here, involved the sensitized *E* to *Z* isomerization of (*E*)-1,2-diphenylpropene (Scheme 2.11).

The compound to be studied (xanthone and thioxanthon) was used as a sensitizer for (*E*) to (*Z*) isomerization of 1,2-diphenylpropene and the triplet quantum yields were calculated. To quantfication of the (*E)* and the (*Z)* isomers could be done by GC-MS analysis. The amount of the light absorbed was measured by ferroxilate actinometry using the splitting ratio method. The following equation (eq.2.2) described by Lamola and Hammond[10b] was used to calculate the triplet quantum yield of the thioxanthone and the xanthone.

$$\Phi_{isc} = \frac{\beta_{t\to c}}{I}(1 + x) \quad \text{.....eq 2.2}$$

where, $\beta_{t\to c} = 2.303\alpha \log\left[\frac{\alpha}{(\alpha-\beta')}\right]$; $\beta_{t\to c}$ is conversion of *trans* to *cis* without back reaction; α, the conversion at the stationary state; and β', the conversion

measured experimentally.

$x = \frac{[trans]_s}{[cis]_s}$; $[trans]_s$ and $[cis]_s$ are the concentrations of *trans* and *cis* isomer at photostationary state.

I = the light absorbed for the photoreaction.

The reported value for the $\frac{[trans]_s}{[cis]_s} = 0.81$ for 1,2-diphenylpropene sensitized by a high energy sensitizer.

The following example describes the calculation of the triplet quantum yield.

Example: One of the experiments performed gave the following GC data after irradiation of 1.473 mmol of *trans*-1,2-diphenylpropene in aqueous dioxane in the presence of xanthone as the high energy sensitizer for 4 hours . The intensity of light absorbed was 0.068 mE h^{-1}.

GC data: Area (trans) = 477683122

Area (cis) = 47463272

Calculations: $\frac{[trans]_s}{[cis]_s} = 0.81$

$$\frac{1.4754mmol - \alpha}{\alpha} = 0.81$$

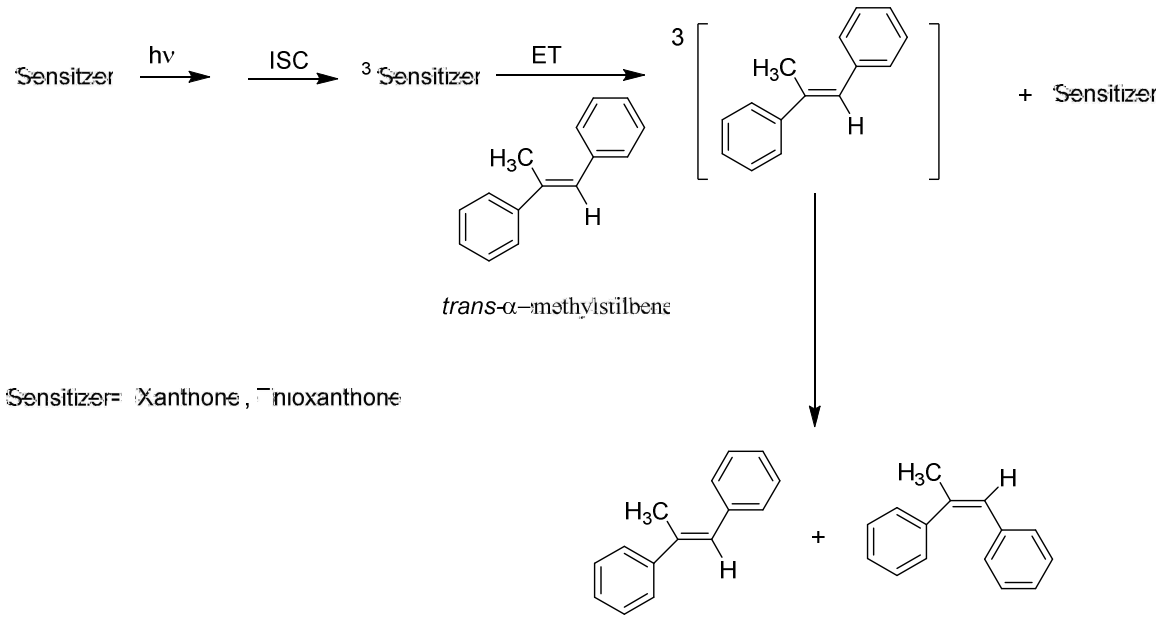
so α =0.814 mmol

$\beta' = 1.475mmol \times \left(\frac{47463272}{(247463272+477683122)}\right)$=0.133 mmol

then $\beta = 2.303 \times 0.814 \log \frac{0.814\ mmol}{(0.814\ mmol - 0.133mmol)}$=0.145

so that $\Phi_{isc} = \frac{0.145\ mmol \times (1+0.81)}{0.068\ mEh^{-1} \times 4\ h} = 0.98$

Scheme 2.11. *E* to *Z* isomerization of (*E*)-1,2-diphenylpropene in the presence of sensitizer



The two experiments, the average was found to be $\Phi_{isc} = 0.67$ for thioxanthone 25% aqueous 100 mM phosphate buffer in dioxane. The average for xanthone was $\Phi_{isc} = 0.98$ in the same aq solvent. The litrature values for xanthone and thioxanthone in non polar solvent are 0.97[11] and 0.85[12] respectively. In ethyl acetate reported values are 0.99 and 0.90[13] In methanol the literature value for thioxanthone is 0.56[12]. No values have been previously reported for aq dioxane. The Φ_{isc} for this solvent had to be determined, because recent paper showed on the basis of DFT calculations(B3LYP/TZVP or TZVPP and COSMO moldel for solvent environment)[14] that Φ_{isc} could change significantly when the solvent contained substantial amount of water. To check our implementation, we determined $\Phi_{isc} = 1.05$ for benzophenone in benzene, which is within experimental error of the known unit efficiency in this case. For xanthone the Φ_{isc} in aq dioxane with

buffer is not significantly different from the literature value of 0.97 in in carbontetrachloride.[11] With thioxanthone the presence of water with the dioxane substantially lowers Φ_{isc}, and our value is comparable to that reported for methanol.

2.3. Conclusions

Although the photochemistry upon direct photolysis of [1,2-b]naphthothiophene **6b** is significantly more efficient ($\Phi = 0.084$) than the corresponding benzothiophene **3** ($\Phi = 0.035$) [2], the observed quantum yield is still substantially lower than the values obtained for direct or xanthone sensitized photolysis of anilide **2-1**, where $\Phi = 0.17$ or $\Phi = 0.27$, respectively. Evidently, the triplet excited state energy transfer from the thioxanthone to the naphthothiophene ring system is still a somewhat inefficient process with an efficiency of 46%. This is in the context of thioxanthone E_T = 64-65 kcal mol^{-1} and naphthothiophene E_T = 62 kcal mol^{-1}, keeping in mind that these triplet energies are only approximate. The observed $\Phi = 0.084$ is fully accounted for by eq. 2.1 using $\phi_{isc} = 0.67$, $\phi_{et} = 0.46$, $\phi_r = 0.27$ obtained the determination of xanthone and thioxanthone triplet sensitized quantum yields.

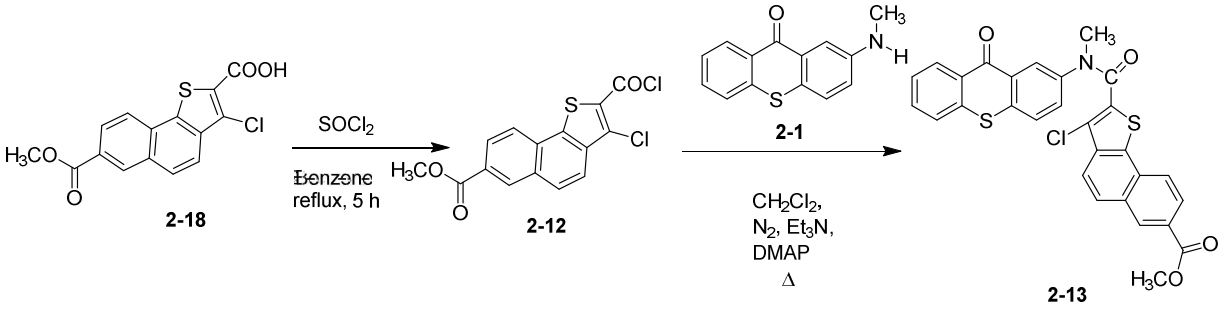
2.4. Experimental

Chemicals and General Methods

All the chemicals were purchased from Sigma-Aldrich, VWR, or TCI America and used as received unless otherwise mentioned. The ^1H, and ^{13}C NMR spectra were recorded on a Varian 300 or 400 spectrometer. Solutions required for the actinometry was prepared using the procedure reported by Zimmerman.[12] HRMS spectra were

collected using a Shimadzu LCMS-IT-TOF instrument at the Department of Chemistry and Biochemistry UW-Milwaukee, Wisconsin. Midwest Micro Lab, LLC, Indianapolis, Indiana 45250, performed all elemental analyses. All melting point determinations were made on Fisher-Johnes melting point apparatus. UV absorption was measured by a Cary 5000 UV spectrophotometer. GC-MS analysis was done using an Agilent 6850 GC-MS spectrometer with a HP-5 (5% phenylmethylpolysiloxane) column (30 m×0.32 mm×0.25 μm).

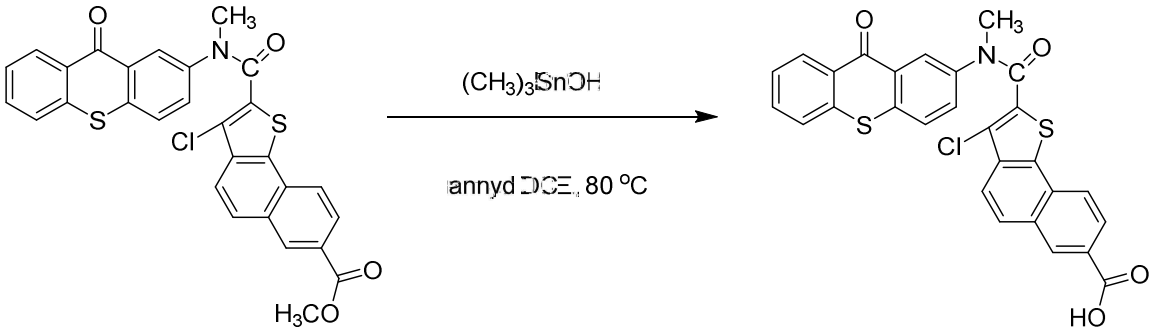
Preparation of N-(9-oxothioxanthenyl)naphtho[1,2-b]thiophene carboxylic acid methyl ester 2-13



The procedure was adapted from the procedure reported by Sarker and coworkers.[2] 1.47 g (4.58 mmol) of **2-18** was refluxed with 50 mL benzene and 20 mL of $SOCl_2$ for 5 h. The benzene and $SOCl_2$ were removed in vacuo. The remaining $SOCl_2$ was co-distilled with anhydrous DCM twice and the yellow residue of **2-12** was used for next step. A solution of 2-methylaminoxanthen-9-one(**2-2**) (0.920g, 3.82 mmol) and 11 mL of triethylamine in 10 mL of anhydrous CH_2Cl_2 was added to acidchloride **2-12** dissolved in 30 mL of anhydrous CH_2Cl_2 at room temperature. A catalytic amount of DMAP was added. The reaction mixture was warmed at temperatures below 40 ^oC for 96 h under nitrogen while stirring. The reaction mixture was filtered to remove triethylamine hydrochloride,

washed with saturated aqueous $NaHCO_3$, water, 2 M HCl, water brine. Then extract was dried over anhydrous Na_2SO_4 and concentrated *in vacuo* to obtain a golden yellow solid containing **2-13**. The crude compound was purified by silica gel flash column chromatography eluting with 30% ethyl acetate in hexane initially and gradually increasing polarity to 60% ethyl acetate in hexane to obtain 1.18 g (57% yield) of product as yellow colored crystals, mp. 228-230 ^oC. The spectral data: ^1H NMR (400 MHz, $CDCl_3$) δ 3.62 (s, 3H), 3.95(s, 3H),7.40-7.54 (m, 4H), 7.60 (t, J = 8.4 Hz, 1H), 7.73 (d, J = 10.1 Hz, 1H), 7.82 (d, J = 8.4 Hz, 1H), 7.99 (d, J = 8.4 Hz, 1H), 8.15 (d , J = 8.9 Hz, 1H), 8.55-8.59 (m, 2H), 8.62(s, 1H); LCMS (APCI-IT-TOF) m/z: $[M+H]^+$ Calcd for $C_{29}H_{18}ClNO_4S_2$ 544.0439; Found 544.0423; Anal. Calcd. for $C_{29}H_{18}ClNO_4S_2$: C, 64.04; H, 3.33; N, 2.58. Found: C, 64.24; H, 3.41; N, 2.58.

Preparation of N-(9-oxothioxanthenyl)naphtho[1,2-b]thiophene carboxylic acid 6b

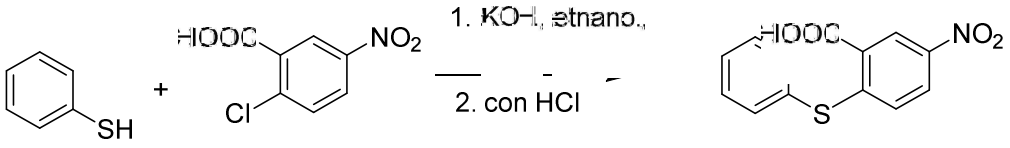


2-13 **6b**

To stirred solution of 1.000g(1.840 mmol) of N-(9-oxothioxanthenyl)naptho[1,2-b]thiophene carboxylic acid methyl ester **2-13** in DCE at 80 °C under argon was added 5 equivalent 0.3300g (9.200.mmol) of the weight of ester **2-13** of trimethyltin hydroxide. The reaction mixture was refluxed under nitrogen.[13] The reaction was monitored by

TLC. When the reaction was complete, the solvent was removed in *vacuo* and ethyl acetate was added. Dilute HCl was added to the ethyl acetate and after extraction; the organic layer was washed several several times with water, brine, and dried over anhydrous sodium sulfate. Concentrtion in *vacuo* gave a yellow solid residue. The yellow residue was washed with water to remove remaining trimethyltin hydroxide. The product was filtered and dried under vaccum to give 0.9360g (96 % yield) of compound **6** (Y = -COOH, LG = -Cl) as yellow powder, m.p.294-296 ºC. .The spectral data : ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.56 (s, 3H), 7.46 (t, J = 8.5 Hz, 1H), 7.76 (m, 3H), 7.80 (s, 2H), 8.10 (m, 3H), 8.37 (d, J = 8.5 Hz, 1H), 8.45 (s, H), 8.60 (d, J = 14.1, 1H), 13.2 (s,br,1H). ^{13}C NMR (100 MHz, DMSO-d_6): δ 38.4, 120.1, 120.7,127.1, 127.5, 127.9, 128.2, 128.3, 128.6, 129.3, 129.6, 129.9, 130.0, 131.1, 131.8,132.8, 133.7, 134.5, 135.5, 135.9, 136.7, 161.9,167.7, 178.8; LCMS (APCI-IT-TOF) m/z: $[M-H]^+$ Calcd for $C_{28}H_{16}ClNO_4S_2$ 528.0137; Found 528.0130; Anal. Calcd. for $C_{28}H_{16}ClNO_4S_2$: C, 63.45; H, 3.04; N, 2.64. Found: C, 63.73; H, 3.34; N, 2.58.

Preparation of 4-Nitrophenyl Sulphide-2-carboxylic acid (2-4)

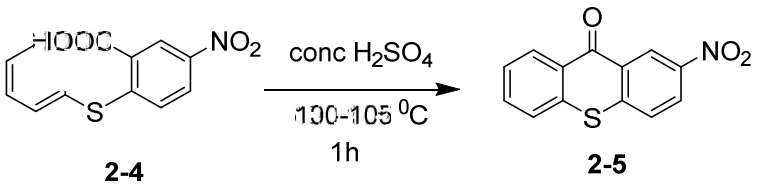


2-4

The procedure was adapted from the procedure reported by Amstutz and co-workers.[15] To a solution of 21.4g. (105 mmol) of 5-nitro-2-chlorobenzoic acid in 300 mL absolute ethanol , 12.4g (119 mmol) of thiophenol, 15.7 g (280 mmol) of potassium hydroxide dissolved in 300 mL ethanol while stirring. Then reaction mixture was refluxed overnight under nitrogen. After two thirds of the alcohol had been removed in

vacuo, the residue was diluted with water, acidified with conc HCl to pH=2, filtered and the solid precipitate was washed with water. The crude product was crystallized with 80% aqueous ethanol to obtain 24.1g (82%) of compound **2-4** as dark yellow crystals, mp 233-234 °C (lit. mp.232.2-234.5)[14]. ^{1}H-NMR (400 MHz, DMSO-d_6): δ 6.87 (d, J = 10.0 Hz, 1H), 7.54-7.68 (m, 5H), 8.19 (dd, J = 9.1, 3.4 Hz, 1H), 8.66 (d, J = 9.5 Hz, 1H); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 126.2, 127.0, 127.2, 127.5, 130.5, 131.5, 131.0, 136.2, 144.3, 152.4, 166.2.

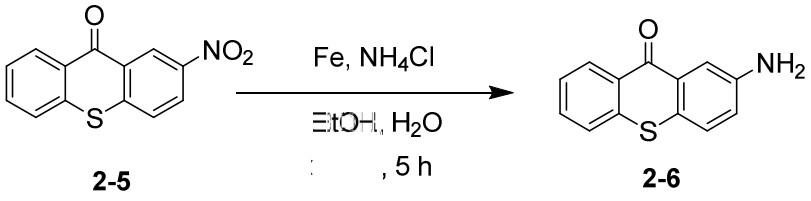
Preparation of 2-nitroxanthone (2-5)[15]



2-4 **2-5**

To 185 mL of concentrated sulphuric acid at 100 °C was added 21.2 g (77.0 mmol) of 4-nitrophenylsulphide-2-carboxylic acid **2-4.** The temperature of the mixture was maintained at 100-105 °C for 1 h. The reaction mixture was cooled to room temperature and poured onto 100 g of ice. The resultant precipitate was filtered, washed with water, sodium bicarbonate solution, and water. After air drying , 18.9 g (95%) of **2-5** was obtained as NMR pure yellow-green solid, mp 225-228 °C(lit. mp 226.8-226.9 crystallized sample from nitrobenzene).[15] This compound was used in the next step without further purification. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.67 (t, J = 7.7 Hz, 1H), 7.85 (t, J = 7.5 Hz, 1H), 7.94 (d, J = 8.1 Hz, 1H), 8.15 (d, J = 9.5 Hz, 1H), 8.46 -8.52 (m, 2H), 9.08 (d, J = 2.5 Hz, 1H).

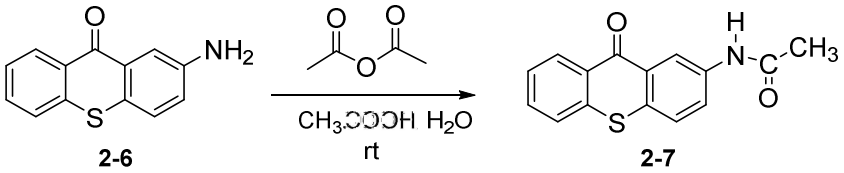
Preparation of 2-Amino-thioxanthen-9-one (2-6)



2-5 **2-6**

The procedure was adapted from a procedure reported by Sarker[2] and Moon.[3] A mixture of 18.8 g (73.4 mmol) of 2-nitroxanthone **2-5**, 800 mL of ethanol, 200 mL of water, 23.6 g (440 mmol) ammonium chloride, and 16.4g (294 mmol) iron powder was refluxed for 5 h while mechanically stirring. After hot vacuum filtration through silica gel, the silica gel was washed with ethanol and the wash combined with the original filtrate. The combined filtrate was concentrated in *vacuo*. The product was extracted into $CHCl_3$. The $CHCl_3$ extract was dried over anhydrous sodium sulfate and concentrated in vacuo to give 12.0 g (72% yield) of **2-6** as a dark yellow powder, mp 226-228 °C (lit. mp 231-233°C).[6] The spectral data were similar to that reported previously[2]: ^{1}H NMR (400 MHz, DMSO-d_6) δ 5.66 (2H, br), 7.06 (d, J = 8.5 Hz, 1H), 7.45-7.51 (m, 2H), 7.61-7.77 (3H, m), 8.41 (d, J = 8.5 Hz, 1H) .

Preparation of N-(9-oxo-9H-thio-xanthen-2-yl) acetamide (2-7)

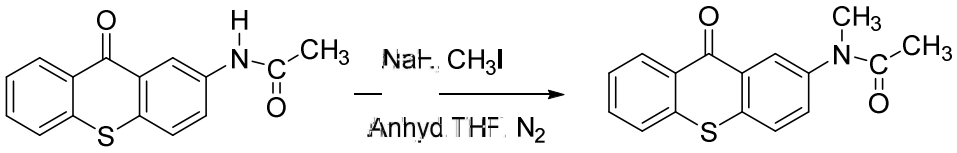


2-6 **2-7**

Procedure was adapted from the procedure reported by Sarker and coworkers.[16] A mixture of 11.0 g (48.2 mmol) of amino ketone **2-6,** 200 mL glacial acetic acid, and 81.1 mL (859 mmol) of acetic anhydride was stirred for 5 h at room temperature. After

adding 300 g of ice water with stirring, the resultant precipitate was filtered, washed with water and 50 mL methanol. The precipitate was washed with $CHCl_3$ and dried under vacuum to give 8.12 g (63% yield) of acetamide derivative **2-7** as a light yellow crystalline powder, mp 241-242 ^oC. The 1HNMR spectral data were similar to that previously reported.[16]: ^1H NMR (400 MHz, DMSO-d_6) δ 2.11(s, 3H), 7.59 (t, J = 7.6 Hz, 1H), 7.71-7.84 (m, 3H), 8.05 (d, J = 8.5 Hz, 1H), 8.46 (d, J = 7.96 Hz, 1H), 8.71 (s, 1H), 10.35 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 24.6, 118.5, 125.2,127.2, 127.8, 128.58, 129.42, 129.79, 130. 87, 133.56, 133.59, 137.38, 138.95, 148.48, 169.46, 179.29.

Preparation of N-methyl-N-(9-oxo-9H-thioxanthen-2-yl)-acetamide (2-8)[2]

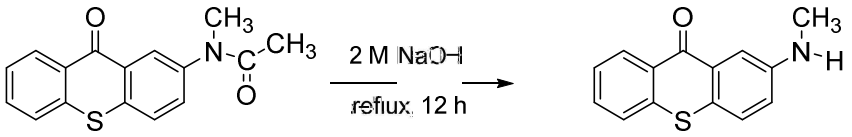


2-7 **2-8**

The procedure was similar to Sarker and coworkers[2]. To a stirred solution of 9.50 g (35.3 mmol) of N-(9-oxo-9H-thio-xanthen-2-yl)acetamide **2-7** in 170 mL of anhydrous THF was added 1.81 g (45.2 mmol) of NaH (60%) under N_2. The mixture was stirred for 15 min followed by drop wise addition of 3.32 mL (53.3 mmol) of methyl iodide. The reaction mixture was stirred at room temperature for 48 h and then concentrated in vacuo to obtain the crude solid residue. To the residue was added $CHCl_3$, followed by filtration and concentration in *vacuo* to obtain 7.5 g (75% yields) of methyl amide **2-8** as a yellow powder. Crystallization with ethanol gave dark yellow needles mp 246-248 °C (lit. 246-248)[2]. The ^{1} HNMR spectral data were similar to that previously reported[2]. The spectral

data as follows: ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.92 (s, 3H), 3.33 (s, 3H), 7.41-7.74 (m, 5H), 8.44 (s, 1H)), 8.62 (d, J = 9.8 Hz, 1H).

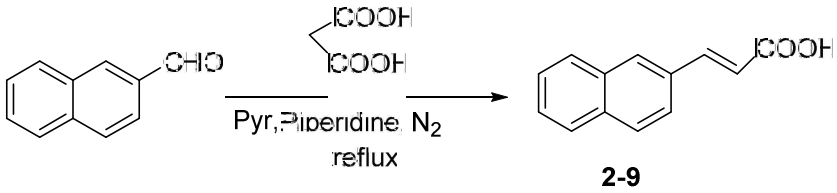
Preparation of 2-methylaminothioxanthen-9-one (2-2)



2-8 **2-2**

Procedure was similar to that of Sarker and coworkes.[2] A mixture of 7.00g (24.7 mmol) of amide **2-8** and 250 mL of aqueous 2 M NaOH was refluxed for 12 h. The reaction mixture was cooled to room temperature and the precipitate was filtered, washed with water, and dried to obtain 4.8 g (81 % yield) of **2-2** as a yellow powder, mp 173-174 °C (lit mp 172-175 °C)[2]. The ^{1} H NMR spectral data were similar to that previously reported.[2] : ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.95 (s,3H), 3.98 (bs, 1H), 6.98 (d, J = 8.7 Hz, 1H), 7.36-7.48 (m, 2H), 7.53-7.60 (m, 2H), 7.77 (s, 1H), 8.64 (d, J = 8.1 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 30.7., 109.5, 120.1, 124.9, 125.6, 125.9, 126.7, 128.7, 129.8, 130.20, 131.6, 137.8, 148.0, 180.0.

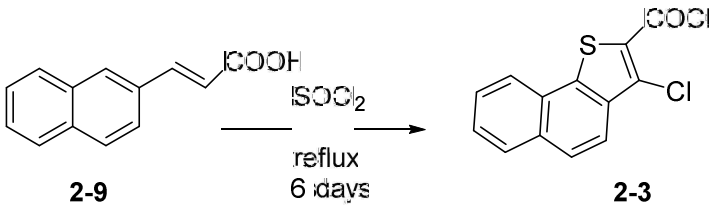
Preparation of 3-(2-naphthyl)propenoic acid (2-9)[17]



2-9

To a solution of 50 mL pyridine (621 mmol) and 11.4 g of malonic acid (110 mmol) was added 14.2 g (90.1 mmol) of naphthalene-2-carboxaldehyde in increments followed by1 mL(100 mmol) of of piperdine at room temperature. The reaction mixture was refluxed until evolution of CO_2 ceased (1.5 h). Afterwards, it was cooled to room temperature. The solution was then poured into 50 mL of ice and conc HCl to give a precipitate. The precipitate was filtered, washed with water and dried. The solid was crystallized from ethanol to obtained 17.5 g (97% yield) of product **2-9** as an off white crystalline solid mp 206-208 °C (lit. mp 208-209°C).[15] ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.70 (d, J = 16.1 Hz, 1H), 7.54-7.60 (m, 2H) 7.78 (d, J = 16.1 Hz, 1H), 7.88-8.00 (m, 4H), 8.20 (s, 1H), 12.51(s, 1H). (100 MHz, DMSO-d_6) δ 119.9, 124.3, 127.2, 127.7, 128.1, 128.9, 130.1, 132.4, 133.3, 134.2, 144.4, 168.1.

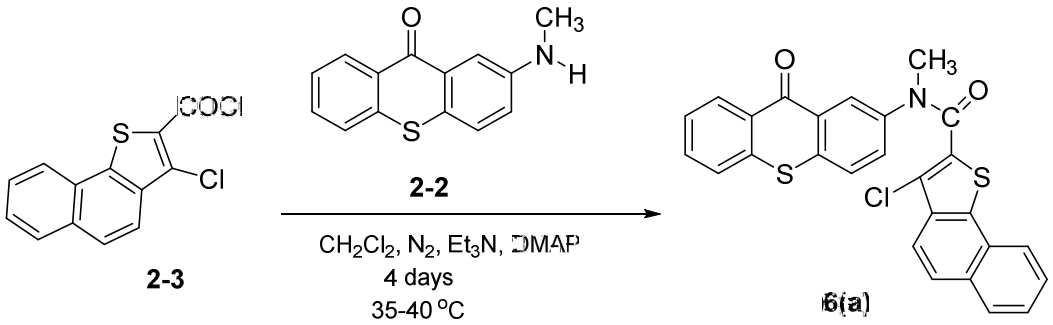
Preparation of 3-chloronaphthol[1,2-b]thiophene-2-carbonyl chloride (2-3)[18]



2-9 **2-3**

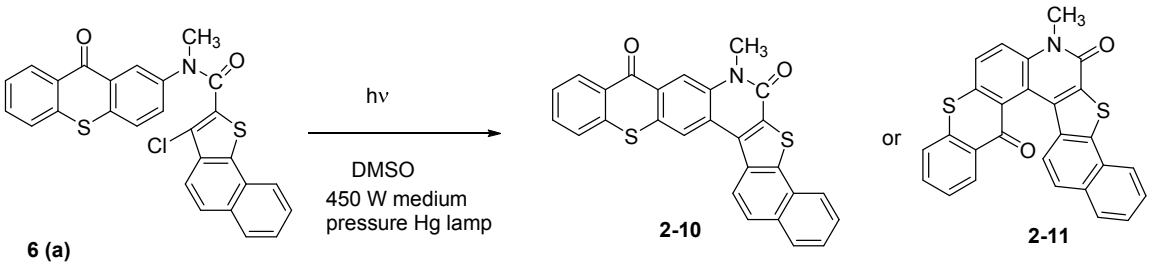
A mixture of 17.5 g (88.0 mmol) of **2-9**, 100 mL of chlorobenzene, 1.6 mL (20 mmol) of pyridine and 36.4 mL (500 mmol) of thionyl chloride was refluxed for 72 h. After cooling to room temperature suction filtration gave 16.5 g (67% yield) of product **2-3** as yellow needles, mp 191-193 °C (lit mp 191-192 °C).[16] ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.73 (m, 2H), 7.90 (dd, J = 8.7, 1.1 Hz, 1H) 8.13 (d, J = 8.7 Hz, 1H), 8.12-8.15 (m, H), 7.25-8.28 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 120.4, 123.8, 126.5, 126.5, 127.8, 128.4, 128.6, 129.7, 132.3, 134.8, 137.2, 162.1.

Preparation of 3-chloronaphthol[1,2-b]thiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide 6(a)



To a solution of 1.2 g (5.0 mmol) of 2-methylaminoxanthen-9-one **2-2** and 15 mL of triethylamine in 30 mL of anhydrous CH_2Cl_2 was added 1.7 g (6.1 mmol) of 3-chloronaphthol[1,2-b]thiophene-2-carbonyl chloride **2-3** suspension in 10 mL of anhydrous CH_2Cl_2 at room temperature. A catalytic amount of DMAP was added. The reaction mixture was heated to temperature below 40 ^oC for 96 h under nitrogen while stirring. The reaction mixture was filtered to remove triethylamine hydrochloride, washed several times with saturated aqueous $NaHCO_3$, water and then with 2 M HCl, water and brine. After drying over anhydrous Na_2SO_4, the solution was concentrated *in vacuo* to obtain a golden yellow crystalline solid containing **6 (a)**. Crystallization from ethanol gave 1.36 g (57% yield) of product as dark yellow colour powder, mp 210-212 ^oC. The spectral data were as follows: ^1H NMR (400 MHz, $CDCl_3$) δ 3.64 (s, 3H), 7.35-7.62 (m, 7H), 7.65 (d, J = 8.5 Hz, 1H), 7.72 (d, J = 8.5 Hz, 1H), 7.87 (d, J = 8.5 Hz, 1H), 7.96 (d, J = 8.5 Hz, 1H), 8.56 (s, 1H), 8.58 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.5, 111.4, 122.2, 124.4, 126.3, 126.9, 127.4, 127.6, 128.9, 129.0, 129.4, 129.6, 130.1, 130.2, 130.9, 132.9, 133.2, 136.5, 136.9, 136.8, 137.1, 139.9, 142.0, 162.9, 171.3, 179.5.

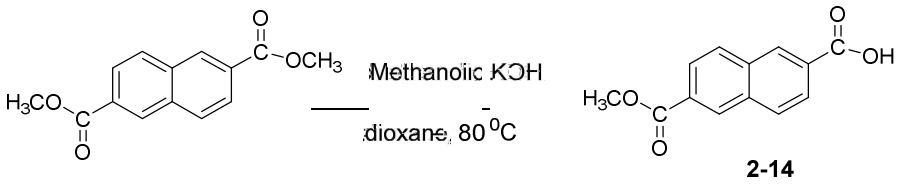
Direct Photolysis of Compound 6(a) to give 2-10 or 2-11



6 (a) **2-10** **2-11**

A solution was obtained by dissolving 150 mg of compound **6 (a)** in 50 mL DMSO followed by syringe filtration to remove a small amount of suspended reactant. Then the clear solution was saturated with nitrogen gas for 30 min and photolyzed by a 450 W medium pressure Hg lamp with a Pyrex filter. After 4 h the precipitate was , washed with water, and dried in *vacuo* to obtain 85 mg of a yellow crystalline solid of **2-10** or **2-11** mp > 300 ºC. ^{1}H NMR (400 MHz, $CDCl_3$): 3.98 (s, 3H), 7.60-7.75 (m, 8H), 7.83 (d, J = 9.1 Hz, 1H), 7.94 (d, J = 9.6 Hz, 1H), 8.29 (d, J = 8.7 Hz, 1H), 8.35 (d, J = 7.7 Hz, 1H) The ^{13}C NMR could not be obtained due to poor solubility.

Preparation of 6-cabomethoxy-2-naphthalenecarboxylic acid (2-14)

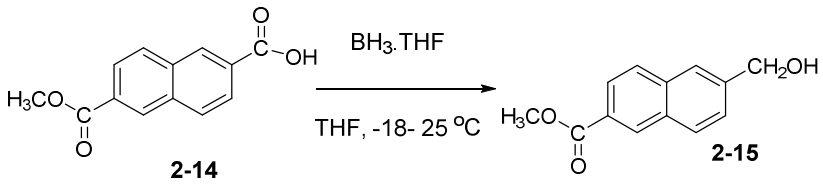


2-14

The procedure is similar to that reported by Wendt and coworkers.[19] A suspension of 10.0 g (40.9 mmol) of dimethyl 2,6-naphthalenedicarboxylate in 60 mL dioxane was heated at 80 ºC until all solid dissolved. The solution of 2.6 g (42 mmol) of KOH in 2 mL of MeOH was slowly added and, the reaction mixture was stirred for 2 h at 80 ºC. The reaction mixture was cooled to room temperature, filtered and the precipitate

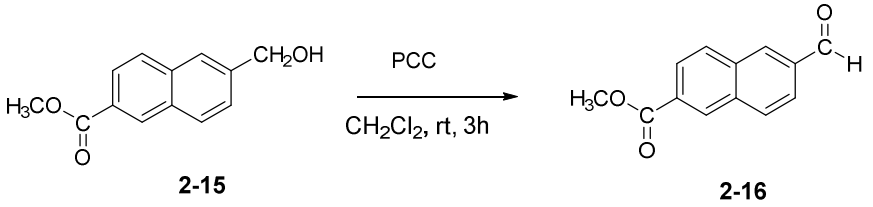
was washed with diethyl ether. The precipitate was dissolve in water and the solution acidified to pH 2 with 2 M HCl. The resultant precipitate was filtered, washed with water, and dried to obtain 8.5 g (90% yield) of **2-14** as a colorless powder, mp 249-251^{o}C. The spectral data were similar to that reported previously.[18] ^{1}H NMR (400 MHz, DMSO-d_6) 3.95 (s, 3H), 8.06 (d, J = 8.6 Hz, 2H), 8.24 (d, J = 8.6 Hz, 2H), 8.69 (s, 1H), 8.68 (s, 1H), 13.34 (br, 1H).

Preparation of 6-hydroxymethyl-naphthalene-2-carboxylic acid methyl ester (2-15)



This method was adapted from Phillippe and coworkers[20] and Krisnamurthy and coworkers.[21] To a suspension of 1.5 g (6.5 mmol) of **2-14** in 30 mL of anhydrous THF at -15 ^{o}C was added 13 mL of BH_3.THF complex slowly. The reaction mixture was stirred overnight at room temperature under nitrogen. Water was added and the mixture was concentrated in *vacuo*. Saturated $NaHCO_3$ was added and the mixture extracted by ethyl acetate. The ethyl acetate extract was washed several times with water, brine, dried over anhydrous Na_2SO_4, and concentrated in *vacuo* to give 1.1g of crude product as a colorless powder. The product was purified by silica gel chromatography eluting with 50% ethyl acetate in hexane to obtain 0.8g (57 %) of colourless product **2-15** mp 125-127 ^{o}C. The spectral data were similar to that reported previously[19]: ^{1}H NMR (400 MHz, DMSO-d_6) 3.91 (s, 3H), 4.84 (s, 2H), 7.54 (d, J = 9.4 Hz, 1H), 7.85 (d, J = 7.7 Hz, 2H), 7.93 (d, J = 9.4 Hz, 1H), 8.06 (d, J = 8.5 Hz, 1H), 8.61 (s, 1H).

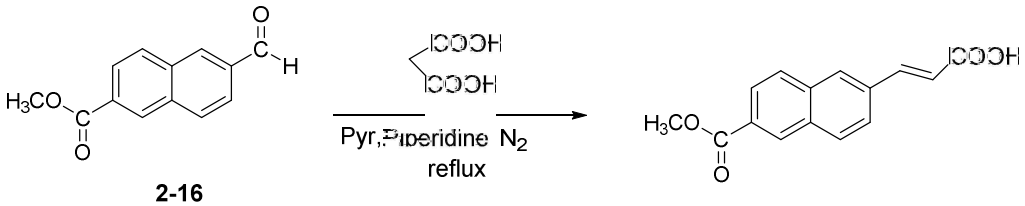
Preparation of methyl -6-formylnaphthalene-2-carboxyllate (2-16)



2-15 **2-16**

The procedure for the PCC oxidation was adopted from the procedure reported by Corey and coworkers [22] with some modifications. To a mixture of 3.51 g (16.3 mmol) of PCC in 60 mL of anhydrous CH_2Cl_2 was added 1.50 g of celite and a solution of 1.60 g (7.40 mmol) of alcohol **2-15** in 10 mL of anhydrous CH_2Cl_2 and the reaction mixture was stirred for 3 h at room temperature. The solution was decanted into another flask and the remaining solid material was washed with CH_2Cl_2 several times and combined to the decanted solution. The combined supernatant liquid was filtered through Florisil (60-100 mesh) column until the orange colour of the solution disappeared. The filtrate was evaporated under *vacuo* to obtain 1.31 g (82 % yields) of pure product **2-16** mp 134-136 °C. The spectral data were as follows: ^{1}HNMR (400 MHz, DMSO-d_6) δ 3.95 (s, 3H), 7.98 (d, J = 9.0 Hz, 1H) 8.10 (d, J = 9.1, 1H), 8.30 (t, J =7.8 Hz, 2H), 8.66 (s, 1H), 8.72(s, 1H), 10.20(s, 1H). ^{13}C NMR (100 MHz, DMSO-d_{63}) δ 52.8, 123.7, 126.4, 129.9, 130.9, 130.82, 130.65, 134.4, 135.4, 135.9, 166.4, 139.5,

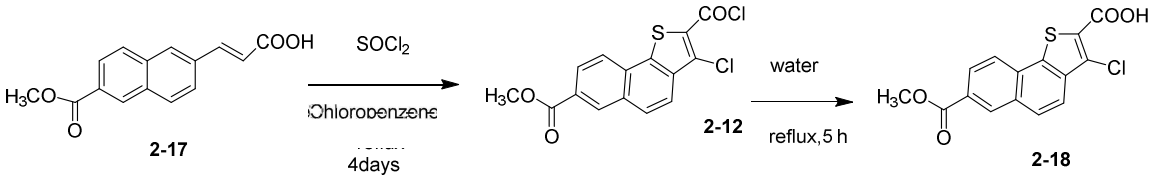
Preparation of 2-17



2-16 **2-17**

The procedure adapted from that reported by Summers and coworkers.[17] To a solution of 3.49 g (33.6 mmol) of malonic acid in 16 mL pyridine at room temperature was added 6.00(28.0 mmol) of methyl 6-formylnaphthalene-2-carboxylate **2-16** in and 1 mL piperidine. The reaction mixture was refluxed 1.5 h until the evolution of CO_2 ceased. The reaction mixture was cooled and poured in to 20 mL conc HCl in ice to produce precipitate. The precipitate was filtered, washed with water, and dried. The precipitate was crystallized from 95% ethanol to obtained 6.72 g (93.7% yield) of product **2-17** as a colorless crystalline product, mp 206-208 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.92(s, 3H), 6.76(d, 1H), 7.77 (d, J = 16.1 Hz, 1H), 8.04(m, 3H), 8.15 (d, 1H), 8.29(s, 1H), 8.65(s, 1H), 12.61(s, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 52.6, 121.4, 125.4, 125.9, 128.2, 129.2, 129.4, 129.6, 130.4, 130.7, 133.2, 134.7, 135.4, 143.8, 166.6, 167.9. (APCI-IT-TOF) m/z: $[M-H]^-$ Calcd for255.0663; Found 255.0660.

Preparation of 2-18



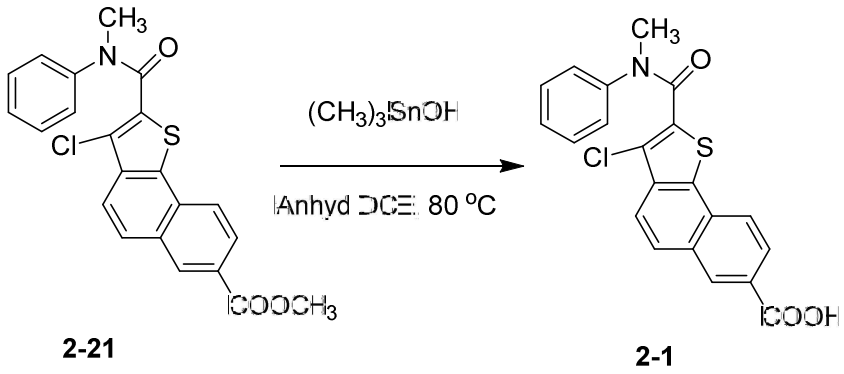
The procedure was adapted from the procedure reported by Castle and coworkers[18] with modifications. A mixture of 6.70 g (26.2 mmol) of **2-17** in 40 mL of chlorobenzene, with 2 mL of pyridine and 11.4 mL (157 mmol) of thionyl chloride was refluxed for 96 h. After cooling to room temperature, suction filtration gave a yellow crude product of **2-12**. The crude sample of **2-12** was refluxed for 5 h in water, cooled to room temperature and suction filtered to obtain 5.68 g (68% yield) of product **2-18**as yellow needles, mp 204-205 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.95

(s,3H), 7.96 (d, J = 9.0 Hz,1H), 8.16 (dd, J = 8.4 Hz, J = 2.0 Hz,1H), 8.21 (d,J = 9.1Hz, 1H), 8.35 (d, J = 9.1 Hz, 1H), 8.77 (d, J = 1.5 Hz, 1H); LCMS (APCI-IT-TOF) m/z: $[M-H]^-$ Calcd for $C_{15}H_9ClO_4S_3$ 318.9837; Found 318.9833. Anal. Calcd. for $C_{15}H_9ClO_4S$: C, 56.17; H, 2.83. Found: C, 53.36; H, 2.45.

Photolysis of compound 6(b) to form 2-19

A 1.0×10^{-2} M solution of **6(b)** in N_2 saturated 33% 100 mM aqueous phosphate buffer in dioxane was irradiated with a 450 W Hanovia medium pressure mercury lamp with a Pyrex filter for 2 h. The product was isolated by filtration, washed with water, $CHCl_3$ and dried under vacuum. The product was a yellow crystalline solid, mp >300°C. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.88 (s, 3H), 7.54 (t, J = 9.2 Hz, 1H), 7.67 (t, J = 10.5 Hz, 1H), 7.81 (t, J = 7.9 Hz, 1H), 7.89 (t, J = 8.6 Hz, 2H), 7.98 (d, J = 7.9 Hz, 1H), 8.10 (d , J = 9.9 Hz, 1H), 8.15 (d , J = 7.9 Hz, 1H) 8.35 (d , J = 7.9 Hz, 1H), 8.60(s,1H). The ^{13}C NMR could not be obtained due to poor solubility in DMSO-d_6. The ^{1}HNMR was taken at 40°C and a COSY spectrum in DMSO-d_6 (Appendix-1) was obtained at 60°C. LCMS (APCI-IT-TOF) m/z: $[M+H]^+$ Calcd for $C_{28}H_{15}NO_4S_2$ 492.0370; Found 492.0360. Anal. Calcd. for $C_{28}H_{15}NO_4S_2$: C, 66.14; H, 3.06; N, 2.84. Found: C, 66.29; H, 3.02; N, 2.83

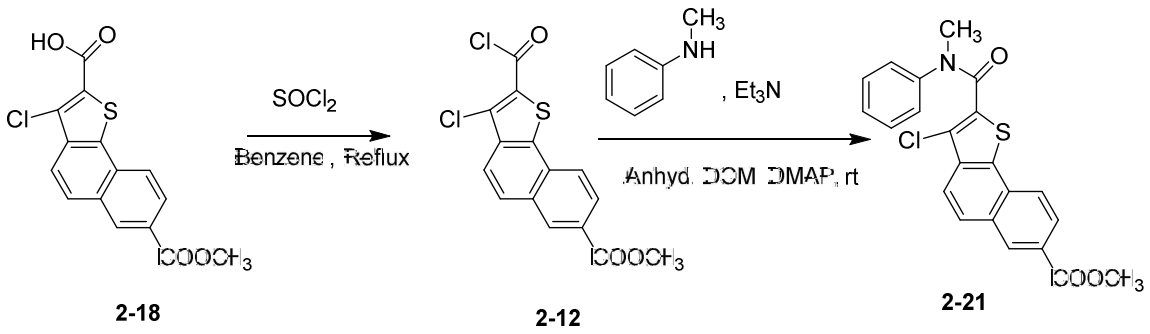
Preparation of photoreactant 2-1



2-21 **2-1**

To stirred solution of 3.00 g (7.32 mmol)of compound ester **2-21** in dichloromethane at 80 °C under Ar was added 5(6.59, 36.6mmol) equivalents of weight of ester **2-21** of trimethyltin hydroxide. The mixture was refluxed and monitored by TLC. After the reaction was complete 50 mL of ethyl acetate was added, followed by aq HCl. After extraction, the organic layer was collected and washed several times with water, brine, dried over anhydrous sodium sulfate and concentrated in *vacuo* to obtain pale yellow crystalline solid. The solid product was washed with water to remove remaining trimethyltin hydroxide. Suction filtration and drying gave 2.47g (96% yield)of **2-1** as pale yellow crystalline solid mp280-282 °CThe spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.47(s, 3H), 7.16 (d, J = 8.3, 1H), 7.27 (t, J = 7.7, 2H), 7.34(s, 1H), 7.36(s, 1H), 7.75(d, J = 8.3 Hz, 1H),8.11 (s, 3H), 8.14(m, 3H), 8.71 (s, 1H), 13.3(s, 1H) . ^{13}CNMR (100 MHz, DMSO-d_6) δ 38.37, 120.7, 120.9, 123.9, 127.3, 127.9, 128. 2, 128.7, 129.7, 129.8, 130.2, 130.1, 131.2, 132.1, 132.5, 134.6, 143.2,161.8,167.8. LCMS (APCI-IT-TOF) m/z: $[M-H]^-$ Calcd for $C_{21}H_{14}ClNO_3S$ 394.0310; Found 394.0304.

Preparation of Photoreactant 2-21

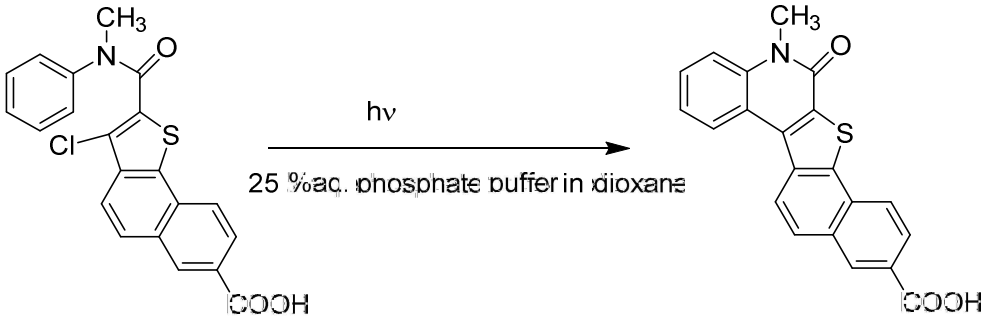


2-18 **2-12** **2-21**

A solution of 4.10 g (12.9 mmol) of **2-18** was refluxed with 50 mL benzene and, 20 mL (283mmol) of $SOCl_2$ was refluxed for 5 h. The reaction mixture was concentrated in *vacuo* dicholoromethane was added followed by concentration in vacuo to give a dry solid residue acid chloride that was used without further purification. To the acid chloride **2-12** in 30 mL oh anhydrous CH_2Cl_2 added 1.15 mL (1.13 g, 10.6 mmol) of N-methylaniline and 31.5 mL of triethylamine in 10 mL of anhydrous CH_2Cl_2. A catalytic amount of DMAP was added. The reaction mixture was heated at temperatures below 40 ^oC for 96 h while stirring under nitrogen. The reaction mixture was filtered to remove trimethylamine hydrochloride, washed several times with saturated aqueous $NaHCO_3$, water, 2 M HCl, water and brine and dried over anhydrous Na_2SO_4 and concentration *in vacuo* gave 4.49.g of a pale yellow crystalline solid (85 % yield) of **2-21,** mp 158-160 $^\circ C$. The spectral data were as follows: ^1H NMR (400 MHz, DMSO-d_6) δ 3.55 (3H,s), 3.99 (3H,s), 7.16-7.27 (m, 4H), 7.76 (d, J = 9.1 Hz, 1H), 7.86 (d, J = 8.9 Hz, 1H), 7.98 (d, J = 8.8 Hz, 1H), 8.15(dd, J = 8.8 Hz,1.53, 1H), 8.66 (d, J = 2.0 Hz, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.2, 52.4, 120.9, 123.2, 126.6, 126.8, 127.3, 127.5, 128.2, 129.2, 130.4, 130.9, 131.3, 134.9, 135.8, 142.9, 162.5, 166.7. LCMS (APCI-IT-TOF) m/z: $[M+H]^+$

Calcd for $C_{22}H_{16}ClNO_3S$ 410.0612; Found 410.0602. Anal. Calcd. for $C_{22}H_{16}ClNO_3S$: C, 64.47; H, 3.93; N, 3.42. Found: C, 64.84; H, 4.01; N, 3.37.

Preparation of compound 2-24 by direct photolysis of 2-22



2-1 **2-22**

A 1.0×10^{-2} M (200mg in 50 mL) solution of **2-1** in nitrogen saturated 25% 100 mM aqueous phosphate buffer in dioxane was irradiated with a 450 W Hanovia medium pressure mercury lamp with a Pyrex filter for 90 min. The photoproduct was isolated by filtration. The photoproduct was washed with water and a small amount of $CHCl_3$ and dried under vacuum to give 178m.g (98%yield) of **2-22** as a crystalline solid, mp > 300°C. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.73 (3H, s), 7.45 (t, J =7.3 Hz, 1H), 7.62 (m), 8.03 (d, J = 9.4 Hz, 1H), 8.14 (d, J = 10.3Hz, 1H), 8.21(d, J =10.4 Hz, 1H), 8.64(s,1H), 8.75(s,1H), 8.76(s, 1H). The ^{13}C NMR could not be obtained due to poor solubility in DMSO-d_6. LCMS (APCI-IT-TOF) m/z: $[M+H]^+$ Calcd for $C_{21}H_{13}NO_3S$ 360.0689; Found 360.0680.

Sensitized Photolysis of Compound 2-1 by Xanthone and Thioxanthone.

A solution containing 0.0274g (0.0692 mmol)of compound **2-1**(2.33×10^{-3} M) and 0.2318 g(1.181mmol) of xanthone (4×10^{-2} M) in 29.5mL of nitrogen saturated 25% 100 mM aqueous phosphate buffer in dioxane was irradiate at 365 nm with a 450W Hanovia

medium pressure mercury lamp with a Pyrex filter for 2-3 hours. Photolysate was evaporated and freeze dried. To the residue was added a standard DMF solution and 2mL DMSO-d_6.The solution was filtered to remove salt and ^{1}H NMR was taken to quantify the photoproduct and quantum yield was determined by using the general procedure for quantum yield determination described below. Similar procedure was followed for a solution containing 0.0282g (0.0713 mmol) of compound **2-1**(2.41×10^{-3} M) and 0.0159 g (0.0749 mmol) of thioxanthone (2.50×10^{-3} M) at wavelength 395 in 29.5 mL of the same solvent system.

Triplet Yield Determination for Xanthone and Thioxanthone.

A solution containing of 0.2894g (1.475 mmol) of pure (E)-1,2-diphenylpropene (0.05 M) and 0.3131 g (1.475 mmol).of thioxanthone (0.05 M) in 29.5 mL of nitrogen saturated 25% 100mM phosphate buffer in dioxane was irradiate at 390 nm with a 450W Hanovia medium pressure mercury lamp with a Pyrex filter for 2-3 hours solution of in photolysate was evaporated and freeze dried. The residue was dissolved in anhydrous benzene and dried over anhydrous Na_2SO_4.The syringe filtered sample of the solution was injected to GC-MS with a HP-5 (5% phenylmethylpolysiloxane) column (30 m×0.32 mm×0.25 μm) to find the ratio of two isomers of 1,2-diphenypropene. Similar procedure was followed with a solution containing 0.2894g (1.475 mmol) of pure (E)- 1,2-diphenylpropene (0.05 M) and 0.3131 g (1.475 mmol).of thioxanthone (0.05 M) in 29.5 mL of the same solvent system.

Quantum Yield Determination.

General Procedure for Product Quantum yield Determination

A semi-micro optical bench was used for quantum yield determinations using ferroxilate actinometry[23], similar to the apparatus described by Zimmerman.[24] A 200 W high pressure mercury lamp was used as light source. Light from the UV lamp was passed through the Oriel monochromator, which was set to desired wavelengths. The light was collimated through a lens. A fraction of light was split 90° degrees by a beam splitter to a 10×3.6 cm side cylindrical quartz cell containing and actinometer solution. The photolysate was contained in 10×1.8 cm quartz cylindrical cell of 29.5 mL volume. Behind the photolysate was mounted a quartz cell containing 29.5 mL of actinometer. The light absorbed quantified by ferroxiolate actinometry using the splitting ratio technique. All quantum yields reported herein where the average of two or more independent runs.

2. 5. Supporting Information -Appendix-1

CHAPTER 3

PHOTOCHEMICAL ELECTROCYCLIC RING CLOSURE AND LEAVING GROUP EXPULSION FROM 5-BENZOYL-3-BROMOTHIOPHENE-2-CARBOXYLIC ACID N-METHYL-(9-OXO-9H-THIOXANTHEN-2-YL) AMIDE

3.1. Introduction

As discussed in the chapter 1, phenyl-2-thienyl ketone **5** has triplet exited state with energies of 62 kcal mol^{-1}.[1] Those triplets are lower in energy than benzothiophene triplet (69 kcal mol^{-1}). So it was planned to replace the benzothiophene moiety with phenyl-2-thienyl ketone in order to facilitate the energy transfer from the triplet excited state of thioxanthone In this chapter we will discuss the synthesis of 5-benzoyl-3-bromothiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide 7 and some photochemistry related to that. The direct photolysis of this compound and the identity of photoproduct are reported. Quantum yields for direct photolysis are determined.

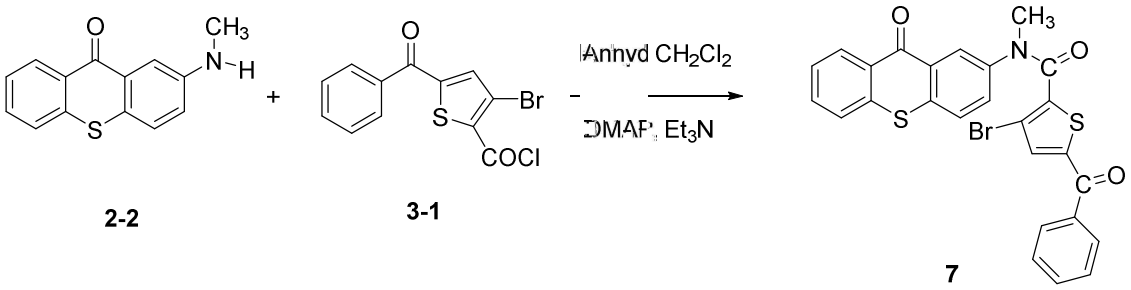
3.2. Results

3.2.1. Synthesis of Photochemical Reactant 7.

The photoreactant 5-benzoyl-3-bromothophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide **7** was synthesized by reacting the acid chloride **3-1** with 2-methylaminothioxanthen-9-one **2-2** (Scheme 3.1). Synthesis of acid chloride **3-1** involved 6 steps starting from 4-bromothiophene carboxyaldehyde. The Grignard reaction of aldehyde with phenyl magnesium bromide formed secondary alcohol **3-2**

which was converted to ketone **3-3** by oxidation with Jone's reagent. The ketone group was protected by making the acetal **3-4** before α lithiation. Then the α lithiated compound was treated with dry ice to obtain caboxylic acid **3-5**. Deprotection of the acetal with glacial acetic acid gave the carboxylic acid **3-6** which was converted to compound **3-1** by refluxing with $SOCl_2$(Scheme 3.2)

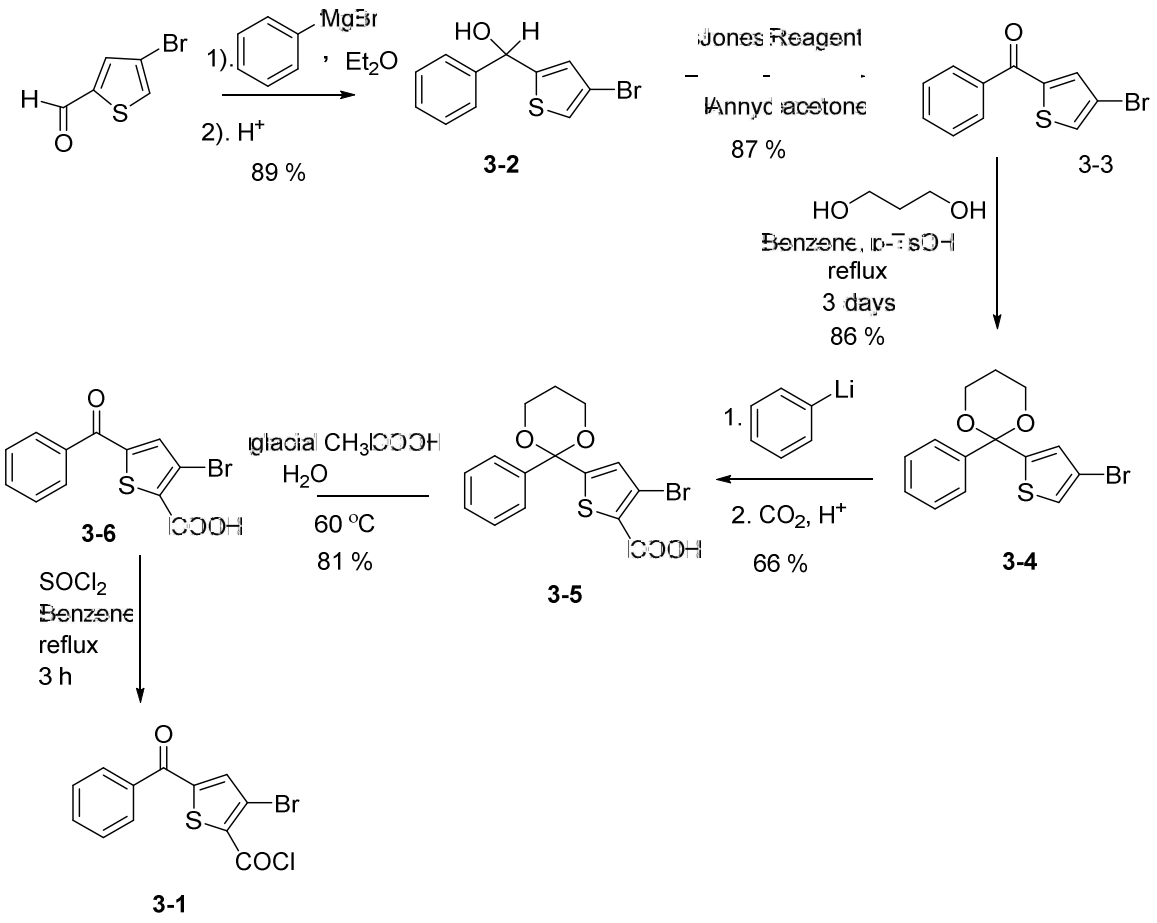
Scheme 3.1 Synthesis of Compound 7



3.2.2. Crystal Structure of Photochemical Reactant 7

According to the X-ray crystallographic analysis (by Oxford Supernova diffractometer using Cu(Kα) radiation, the compound **7** exhibits folded shape with amide group I a *cis*-configuration(Figure 3.1). Both thioxanthone and thiophene moieties are rotated out of conjugation with the amide group because of steric hindrances. There are some deviations from planarity for atom C1 of thiophene ring and some folding of thioxanthone along S…O line. There is some stacking interaction between C21…C26 benzene rings related by inversion center. The bond distances from the carbon (C2) occupied by the bromide leaving group to two ortho positions of the thioxanthone ring system to the amide group are 3.88 Å and 4.90 Å.

Scheme 3.2 Synthesis of compound **3-1**



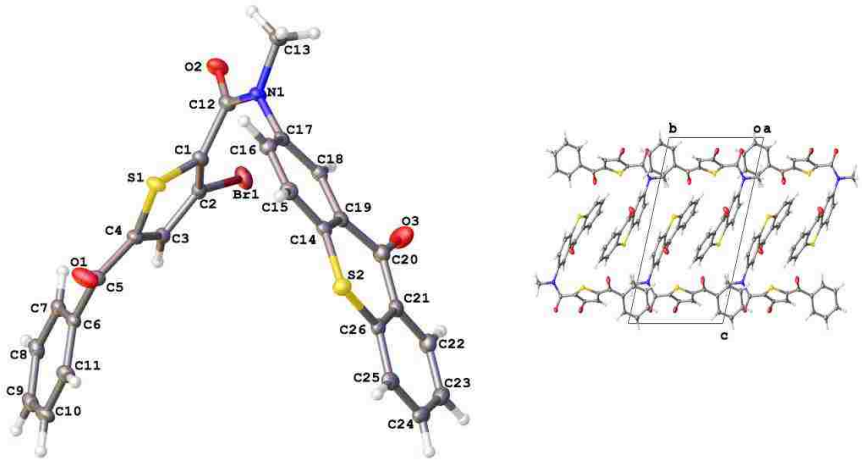


Figure 3.1. Crystal structure of photoreactant **7**

3.2.3. UV Spectra for the Photoreactant 7

The compound **7** exhibits absorption maxima at 388 nm in aqueous acetonitrile (Figure 3.2) with ε = 6270 M^{-1} cm^{-1}.

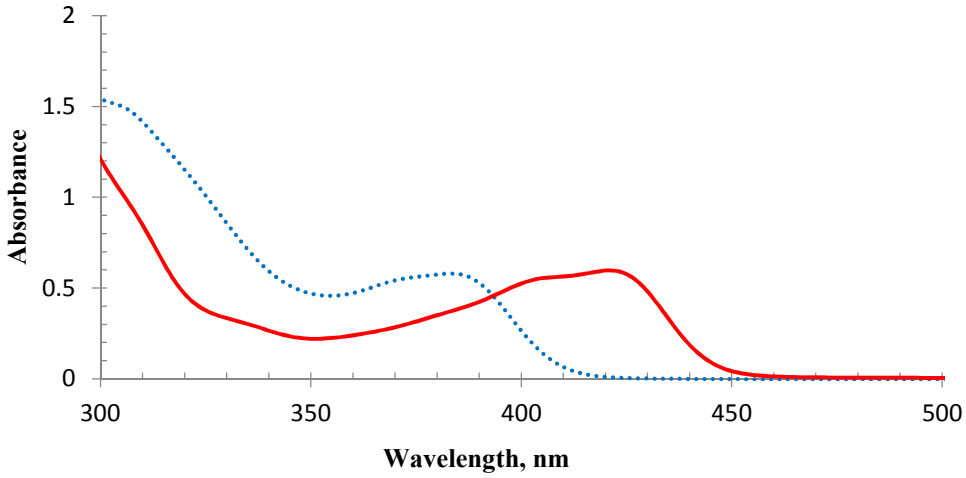


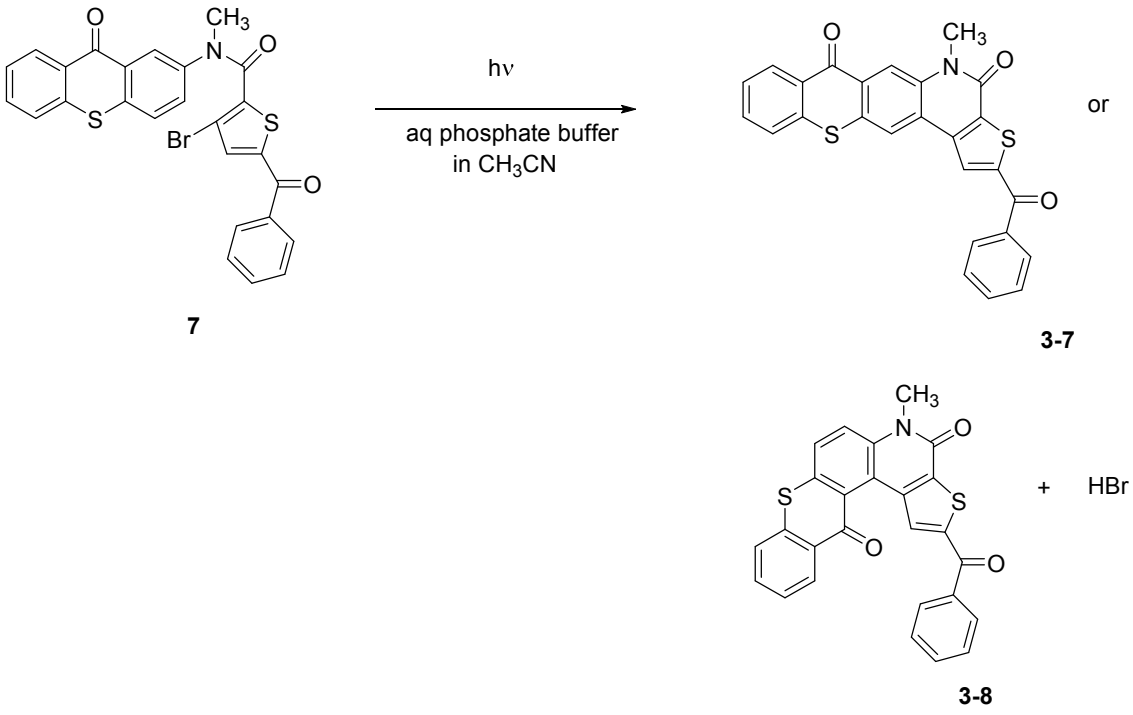
Figure 3.2. Absorption spectra of 1.0 x 10^{-4} M of compound **7** (**(.....**)and 1.0 x 10^{-4} photoproduct produced from **3-7**(—) in 10 % aq. phosphate buffer (pH= 7) in CH_3CN.

3.2.4. Preparative Direct Photolysis

For preparative photolysis, Pyrex-filtered light from a Hanovia 450 W medium pressure mercury lamp was used. Photolysis of 10^{-2} M sample of **7** in N_2 saturated 10% H_2O containing 100 mM phosphate buffer at pH 7 in CH_3CN resulted in expulsion of the bromide leaving group and formation of single regioisomeric photoproduct (Scheme3. 2). However, the cyclization and expulsion of leaving group from compound 7 was very slow. After photolysis for three days solid product formed was filtered washed with water and dried to obtain 30 mg of photoproduct. The product was identified and distinguished from photoreactant by ^1H NMR as N-methyl peak shifted downfield from δ

3.55 to 3.93 ppm, and also aromatic region counts for 12 protons instead of 13 protons which were counted for photoreactant. The melting point was found to be 244-245 ^{o}C for the photoproduct, whereas, 132-133 ^{o}C for the photoreactant **7**.Repeated attempts were done to make crystals for determination of structure using x-ray diffraction. But the compound was obtainable only as a powder. The absorption spectra of the photoproduct **3-7 or 3-8** showed a long wave length maximum at 410 nm with ε = 6990 M^{-1} cm^{-1} in aqueous acetonitrile containing 10% pH 7 phosphate buffer (Figure 3.2).

Scheme 3.3 Photolysis of Compound **7**



3.2.5. Quantum Yield

The quantum yield for the electrocyclic ring closure reaction of amide 7 was determined at 388 nm in N_2 saturated 10% phosphate buffer at pH 7 in CH_3CN. The light output for the photochemical reaction was 0.034 mE/h. After 21.5 h photolysis of the 6.3

x 10^{-3} M solution, the quantum yield of the reaction was found to be 0.004. The quantum yield determination was carried out using ferrioxalate actinometry. The quantum yield determinations involved quantifying the photoproduct formed by using ^{1}H NMR spectroscopy with DMF as an internal standard. In addition, attempts were made to obtain the quantum yield for a 1.0 x 10^{-4} M solution (A = 0.627), but the photoproduct could not be quantified by absorption spectroscopy due to overlap with the photochemical reactant absorption spectrum, which could not be adequately deconvoluted. As found, this particular photoreaction is inefficient.

3.3. Discussion and Conclusion

DFT calculations showed that the crucial step in the electrocyclization of benzothiophene amides with attached thioxanthone chromophores involves excitation transfer from the initial thioxanthone triplet excited state to the benzothiophene ring. An important structural change that accompanies excitation transfer is the pyramidylization of the C-3 carbon bearing the leaving group in the benzothiophene ring. This structural change seems to be important in the conrotatory electrocyclic ring closure step to form the triplet excited state of the putative zwitterionic intermediate. In the case of the previously studied benzothiophene amide with attached thioxanthone chromophore, the energy transfer from the thioxanthone to the benzothiophene is ca. 5 kcal mol^{-1} endothermic in the triplet excited state. The initial triplet excited state energy of thioxanthone is 64-65 kcal mol^{-1}, whereas the triplet excited state energy of the benzothiophene acceptor is 69 kcal mol^{-1}. Therefore, for the current project the objective was to replace the benzothiophene ring with a conjugated thiophene that had a triplet

energy that would be lower than that of the thioxanthone, in order to facilitate the triplet excitation transfer in the critical step of the electrocyclization.

Two choices were thought to be appropriate for substituting the benzothiophene ring system with a thiophene that would have a triplet excitation energy below 64-65 kcal mol^{-1}, which would be favorable for energy transfer from thioxanthone. The first choice which was implemented was to introduce a 5-benzoylthiophene ring system in place of the benzothiophene, e.g. see structure **7** (Scheme 3.2). In structure **7** the 5-benzoylthiophene ring is estimated to have a triplet energy of 62 kcal mol^{-1} on the basis of unsubstituted 2-benzoylthiophene as the model compound.[1] This choice was also considered appropriate, because the electronic configuration of the triplet excited state would be $^3\pi,\pi^*$, which would be essentially unchanged from that of the benzothiophene system that is being replaced.

Photophysical and theoretical studies of 2-benzoylthiophene indicate that the lowest energy singlet excited state is n,π^*, whereas S_2 is π,π^*. These assignments can be made on the basis of solvent effects on the energies of the corresponding bands in the absorption spectrum. As expected, the π,π^* bands appear at 256 nm and 284 nm and are red shifted with increasing polarity of the solvent. On the other hand, the longer wavelength n,π^* band at >350 nm is blue shifted with increase in solvent polarity.

The relative energies of the two singlet excited states are also supported by theoretical calculations. In the ground state, *ab initio* calculations show the thienyl ring to be almost coplanar with the C=O, such that transfer of charge occurs from sulfur to oxygen. S is +0.273 and O is -0.223. The C=O and the thiophene S are cisoid in the ground state, due to the favorable electrostatic interaction. Thus, sulfur strongly interacts,

conjugatively, with the C=O in 2-benzoylthiophene. Moreover, sulfur should have a stabilizing effect on the π,π* excited state of the compound, but not to the extent that this configuration would lie below the n,π* state in the singlet excited state manifold.

The phosphorescence spectrum is consistent with a lowest π,π* triplet configuration, in contrast with the singlet excited states. This assignment is based on the fact that the emission does not show the vibronic progression typical of the carbonyl group, which would be the case, if the emissive triplet was n,π* in character. Moreover, the lifetime of the phosphorescence is quite long, $\tau > 100$ ms at 77 K, which is a characteristic of π,π* triplets. The π,π* assignment for the configuration of the lowest energy triplet excited state is further consistent with the CNDO/S calculation. Note that qualitatively, one would expect that the π,π* configuration should be stabilized relative to the n,π* configuration by the thienyl sulfur, as shown for the singlet excited state. Moreover, the large triplet singlet splitting typical of π,π* excited states (25-30 kcal mol^{-1}) vs. the small 5-6 kcal mol^{-1} S-T splitting typical for n,π* singlet could be responsible for the inversion of the π,π* and n,π* configurations in the triplet excited state manifold.

The above electronic disposition of n,π* and π,π* singlet and triplet excited states is favorable for spin orbital coupling and intersystem crossing. The quantum yield for intersystem crossing in 2-benzoylthiophene is 0.9, which reflects that the change in multiplicity is accompanied by a change in orbital angular momentum in going from the n,π* singlet to the π,π* triplet.

The above photophysical properties are also manifested in reduced reactivity of 2-benzoylthiophene towards hydrogen abstraction from 2-propanol. It is well-known that n,π* triplet excited states of ketones such as benzophenone undergo efficient

photoreduction by hydrogen atom abstraction from 2-propanol to produce ketyl radicals that recombine to give pinacols.

The photolysis of **7** was very slow. In part, this was seen as due to the strong overlap between the absorption spectrum of the product and the photochemical starting material. Whereas in the previously studied benzothiophene amide with thioxanthone chromphore, **3**, the reactant absorbed at 385 nm, while the photoproduct absorbed at 432 nm. In the case of 7 the starting material absorbs at 388 nm and the photoproduct absorbs at 410 nm and 425 nm in 10% aqueous phosphate buffer in CH_3CN. Three days photolysis gave only a 4% yield of photoproduct with a 450 W Hanovia medium pressure mercury lamp.

The quantum yield of photoproduct was estimated as $\Phi = 0.004$ for a single photolysis. The quantitative determination of the photoproduct was done by NMR spectroscopy using DMF as a standard. The experimental conditions are not optimal for the quantum yield determinations, because the absorbance in the 1 cm path cell was A = 39 for the 0.0063 M solution. Attempts were made to obtain the quantum yield for a 1.0×10^{-4} M solution (A = 0.627), but the photoproduct could not be quantified by absorption spectroscopy due to overlap with the photochemical reactant absorption spectrum, which could not be adequately deconvoluted. The issue here is that it is potentially important for the incident light to penetrate some distance into the sample, so as to avoid forming photoproduct within a thin layer at the front face of the cell. This would lower the observed quantum yield due to the internal filter effect of the photoproduct formed early in the photolysis. To avoid the effect, the more dilute solution is desirable, but would require an alternate analytical method to quantify the photoproduct than NMR

spectroscopy. HPLC analysis might be a suitable alternate method for product quantification.

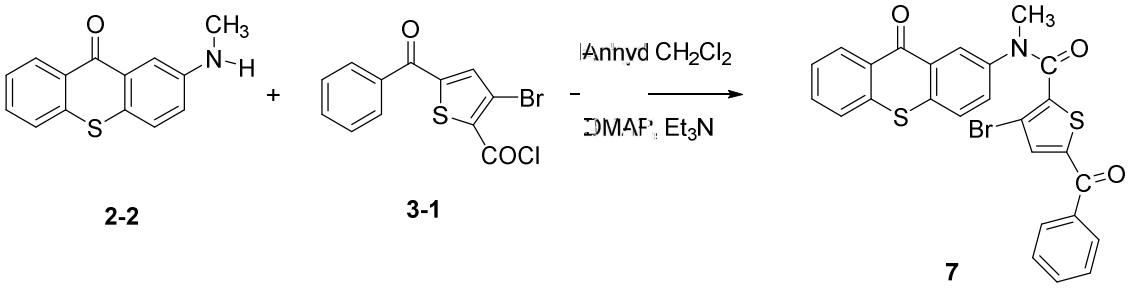
Although the quantum yield may be low due to the aforementioned experimental flaws, it is thought that more likely the compound 7 is inherently and unexpectedly less reactive than the benzothiophene system **3**. The initial concern might be the electronic configuration might not be similar to that of the benzothiophene, which is expected to be π,π^* in both. Theoretical calculations will be needed to ascertain details of the electronic configuration of the triplet excited state of the 5-benzoylthiophene in**7.** Such calculations would reveal whether the C-3 position of the thiophene ring is indeed pyramidalized, as is the case for the benzothiophene ring system. One concern with **7** is that the triplet excitation is localized in the S-conjugated carbonyl group. Whether such localization of excitation elsewhere in the benzoylthiophene moiety suppresses pyramidalization is the question. The low quantum yield has nothing to do with the low chemical yield, which should be 100% regardless, so long as the photoproduct does not compete for light with the reactant.

In the meantime, it was urgent to ascertain whether another choice would be more appropriate as a replacement for the benzothiophene ring system. Efforts therefore focused upon testing a naphthothiophene ring system in place of benzothiophene. In the naphthothiophene the triplet excited state is estimated to be a 62 kcal mol^{-1}, which would lie below the triplet energy of the thioxanthone.

3.4. Experimental

The NMR spectra were recorded at 400 MHz or 300 MHz for ^{1}H and 100 MHz or 75 MHz for ^{13}C with TMS as the standard. Oxford Supernova diffractometer using Cu(Kα) radiation was used to X-ray crystallographic analysis. All melting points were determined using Fischer-Jones melting point apparatus. Absorption measurements were recorded on an Agilent 8453 UVspectrometer. All commercial reagents were used without further purification unless otherwise noted.The solvent used for photolysis were CH_3CN (99.3+%, HPLC grade, Sigma-Aldrich), deionized water, CD_3CN (99.8% d, Cambridge), and D_2O (99.9% d, Cambridge). Solutions required for the actinometry was prepared using the procedure reported by Zimmeman.[2]

Preparation of 5-benzoyl-3-bromothiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide (26)

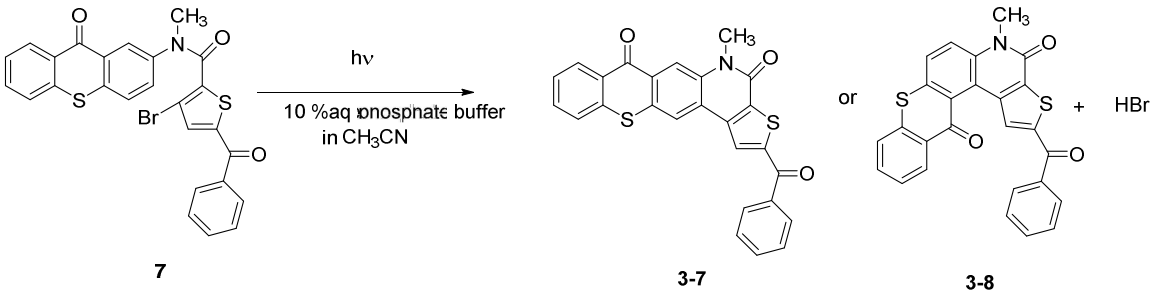


2-2 **3-1** **7**

To a solution of 1.2 g (5.0 mmol) of 2-methylaminoxanthen-9-one **2-2** and 15 mL of triethylamine in 30 mL of anhydrous CH_2Cl_2 was added 2.0 g (6.1 mmol) of 3-bromo-4-benzoylthiophene-2-carbonyl chloride **3-1** dissolved in 10 mL of anhydrous CH_2Cl_2 at 5-8 ^{o}C in an ice bath. A catalytic amount of DMAP was added. The reaction mixture was warmed to room temperature and stirred for 72 h under nitrogen. The reaction mixture was filtered to remove trimethylamine hydrochloride, washed several times with

saturated aqueous $NaHCO_3$ solution, water, with 2 M HCl, water and brine. The CH_2Cl_2 solution was then dried over anhydrous Na_2SO_4 and concentrated *in vacuo* to obtain 2.30 g (86 % yield) of a golden yellow solid of **7**. Crystallization of the solid material with ethanol gave 2.00 g (74.9 %) of yellow colored powder, mp 158-159 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.58 (s, 3H), 7.29 (s, 1H), 7.44 (t, *J* = 7.8 Hz, 3H), 7.50 (t, *J* = 8.0 Hz, 2H), 7.57 (t, *J* = 5.2 Hz, 2H), 7.64 (dt, *J* = 8.0, 1.75 Hz, 1H), 7.72 (s, 1H), 7.74 (s,1H), 8.52 (d, *J* = 2.5 Hz, 1H), 8.60 (d, *J* = 8.3 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.5, 111.4, 126.3, 126.9, 127.4, 127.5, 129.0, 129.3, 129.5, 130.0, 130.1, 130.9, 132.9, 133.2, 136.4, 136.8, 136.7, 137.0, 140.1, 141.1, 144.1, 162.4, 179.35, 186.9.

Photolysis of compound 7

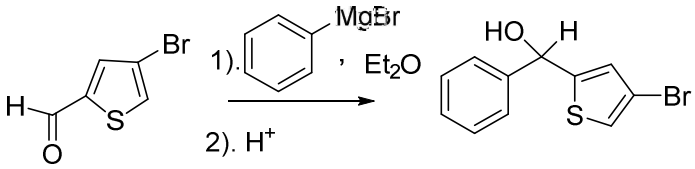


7 **3-7** **3-8**

A solution of 50 mL of 6.3 x 10^{-3} M 7 in 10% aqueous phosphate buffer in acetonitrile was flushed with N_2 for 30 min. Then it was photolyzed using a 450 W medium pressure mercury lamp with a Pyrex filter for 3 days. Resultant precipitate was filtered, washed with acetonitrile and water, and dried to obtain 30 mg of product **3-7** or **3-8** as a yellow powder, mp 244-245 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.93 (s, 3H), 7.52 (t, *J* = 7.2 Hz, 1H), 7.60-7.77 (m, 7H), 7.75 (t, *J* = 9.3 Hz, 1H), 8.08 (s, 1H), 8.09 (s,1H), 8.11 (d, *J* = 8.1 Hz, 1H), 8.26 (d, *J* = 8.1 Hz,

1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 30.8, 118.2, 120.2, 125.5, 126.1, 126.7, 127.0, 128.7, 129.12, 130.1, 132.2, 132.7, 133.3, 135. 7, 135.5, 137.2, 138.0, 138.9, 139.1, 145.7, 157.8, 183.5, 188.7.

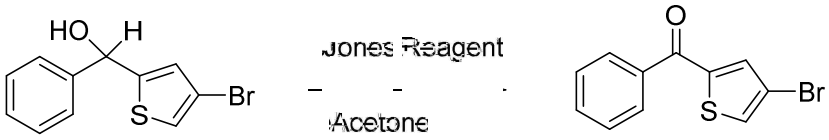
Preparation of 3-2



3-2

The procedure was adapted from a procedure reported by Alexander.[3] To 8.17 g of (336 mmol) of Mg was added to 300 mL anhydrous diethyl ether in dry three neck flask attached to a condenser. Using an addition funnel solution of bromobenzene 49.3 g (314 mmol) in 15 mL of anhydrous diethyl ether was added slowly. Reaction mixture was stirred until all Mg has dissolved. Then 30.0 g (157 mmol) of 4-bromobenzene carboxyaldehyde in anhydrous ether was added into it slowly. Reaction mixture was refluxed for 3 h. The reaction mixture was added into cold solution of 3M HCl to quench the reaction. Ether was added to dissolve all compound. The ether layer was separated, dried over anhydrous Na_2SO_4 and evaporated. The crude solid material was recrystallized from hexane to obtained 37.6 g (89%) of compound 3-2 as a white solid, mp. 81-82 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.47 (s, 1H), 5.97 (d, J = 3.9 Hz, 1H), 6.75 (t, J = 1.5 Hz, 1H), 7.14-7.46 (m, 5H); ^{13}C NMR (100 MHz, $CDCl_3$): δ 72.1, 109.1, 122.5, 126.2, 127.3, 128.4, 128.7, 142.1, and 149.3.

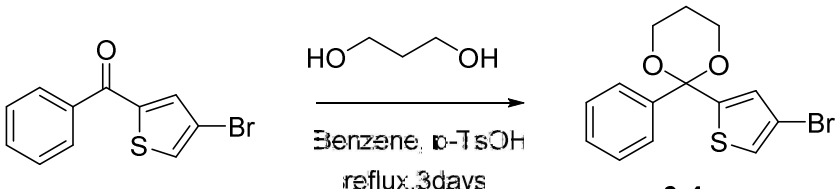
Preparation of 2-benzoyl- 4-bromothiophene (3-3).



3-2 **3-3**

To prepare Jone's reagent 26.7 g (268 mmol) of chromic oxide was dissolved in 23 mL of conc. H_2SO_4, diluted with water to 100 mL at 0 °C. To a solution of 13.5 g (50.0 mmol) of **3-2** in 100 mL acetone was added 14 mL of Jone's reagent previously prepared portionwise while maintaining the temperature below 20 °C. The reaction mixture was stirred further for 3 h. The liquid in the flask was decanted into another flask. The solid material remaining in the flask was washed with ether and combined to the above liquid. To remove excess Cr(VI) ion, sodium bisulphite was added until orange color dissapeared, then washed with water, saturated $NaHCO_3$ and brine. Then it was filtered through Florosil, dried over anhydrous Na_2SO_4 and evaporated *in vacuo* to obtain 10.7 g (87% yield) of **3-3** as off white crystals, mp 84-86 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.5 (m, 3H), 7.6 (t, J = 9.8 Hz, 2H), 7.84 (m, 1H), 7.85 (m, 1H); 13C NMR (75 MHz, $CDCl_3$) δ 110.8, 128.6, 129.2, 131.5, 132.7, 136.5, 137.2, 144.4, 187.9.

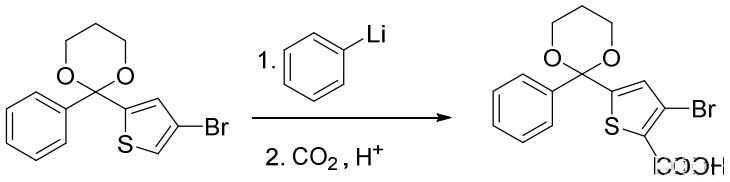
Preparation of 1,3-Dioxane, 2-(3-bromothienyl)-2-phenyl (3-4)



3-3 **3-4**

The procedure was adapted from the procedure reported by Angibaud.[4] To a solution of 21.0 g (78.6 mmol) of 2-benzoyl-4-bromothiophene **3-3** in 120 mL of anhydrous benzene was added 14.4 g (188 mmol) of 1,3-propanediol and 10 mg of *p*-toluene sulfonic acid**.** The reaction mixture was refluxed for 3 days using Dean-Stark apparatus to remove water, cooled to room temperature, washed with saturated $NaHCO_3$, water and brine, dried over anhydrous Na_2SO_4 and evaporated to obtain colorless oil. Upon standing for two days it gave 21.9 g (86 % yield) of acetal **3-4** as colorless crystals, mp. 61-63 ^oC. The spectral data were as follows: ^1H NMR (400 MHz, $CDCl_3$) δ 1.63 (m, 1H), 1.92 (m, 1H), 4.02 (m, 4H), 6.75 (s, 1H), 7.12 (s, 1H), 7.13-7.53 (m, 3H), 7.53 (t, J = 7.5 Hz, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 25.3, 62.1, 99.3, 108.9, 123.5, 126.6, 128.3, 128.4, 128.6, 129.1, 140.5, 149.1.

Preparation of 3-5

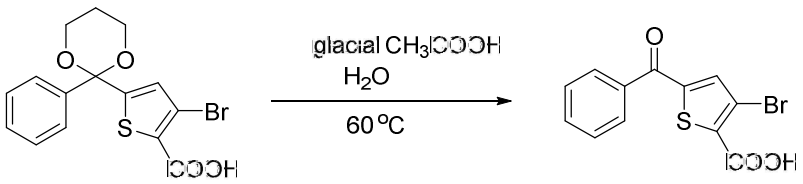


3-4 **3-5**

Procedure was adapted from the procedure reported by Katritzky.[5] 12.0 g (36.8 mmol) of **3-4** in 60 mL anhydrous diethyl ether was treated with 50 mL of 1M PhLi (Prepared by dissolving 0.82 g (118 mmol) Li and 9.24 g (58.8mmol) of bromobenzene in 600 mL of anhydrous diethyl ether. The dark brown mixture was stirred for 5 h at room temperature and it was slowly added to the flask which contained dry ice and kept for an overnight at room temperature. Then the compound was extracted with water and washed with ether. On acidification with conc. HCl (to pH= 2) the acid was obtain as

brown oil. The oil formed was extracted into diethyl ether, dried with anhydrous Na_2SO_4 and evaporated under *vacuo* to obtain 8.62 g (66% yield) of **3-5** as brown solid, mp.149-151 ^oC. The spectral data were as follows: ^1H NMR (400 MHz, $CDCl_3$) δ 1.64 (m, 1H), 1.91 (m, 1H), 4.02 (m, 4H), 6.9 (s, 1H), 7.34 (t, J = 7.6 Hz, 1H), 7.41 (t, J = 9.8 Hz ,2H), 7.55 (d, J = 7.6 Hz, 2H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 25.1, 62.0, 99.0, 117.5, 126.2, 127.2, 129.1, 130.9, 139.6, 155.1, 166.2.

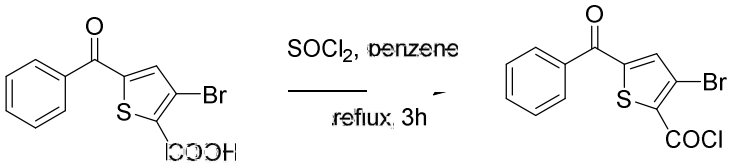
Preparation of 5-benzoyl-3-bromo-2-thiophenecaboxylic acid (3-6)



3-5 **3-6**

Procedure was adapted from procedure reported by Babler.[6] To 10.0 g (28.4 mmol) of **3-5** was added 100 mL of glacial acetic acid and 25 mL of water. The reaction mixture was heated at 65 ^oC for overnight while stirring. Then water was added to dilute the reaction mixture, and extracted with ether. The ether layer was dried over anhydrous Na_2SO_4 and evaporated under *vacuo*. Remaining CH_3COOH was evaporated by vacuum distillation to give 7.92 g of brown color solid. Recrystallization of brown color solid with aqueous ethanol formed 7.16 g (81% yield) of **3-6** as off white color crystals, mp176-178^oC. The spectral data were as follows: ^1H NMR (400 MHz, $CDCl_3$ 7.55 (t, J = 7.8 Hz, 2H), 7.62 (s, 1H), 7.68 (t, J = 7.4 Hz, 1H), 7.89 (d, J = 7.0 Hz, 2H); 13C NMR (100 MHz, DMSO-d_6) δ 115.8, 129.3, 133.9, 135.4, 136.3, 138.7, 145.0, 161.5, 186.9.

Preparation of 5-benzoyl-3-bromo-thiophenecarbonyl chloride 3-7



3-6 **3-1**

To a solution of 4.00 g (12.9 mmol) of **3-6** in 100 mL benzene was added 6.12 g (51.4 mmol) of $SOCl_2$ and refluxed for 3 h. Then the reaction mixture was evaporated under vacuum. Remaining $SOCl_2$ was co-evaporated with CH_2Cl_2 to obtained **3-1** and it was used to next step without further purification.

Procedure for Product Quantum Yield Determination

A semi-micro optical bench was used for quantum yield determinations, similar to the method described by Zimmerman.[2] A 200 W high pressure mercury lamp was used as a light source. Light from the UV lamp was pased through the Oriel monochromator, which was set to 388 nm wavelength. The light was collimated through a lens. A fraction of the light was diverted 90° by a beam splitter to a 10 cm x 3.6 cm side quartz cell containing 41.5 mL of an actinometry solution. The photolysate was contained in 27 mL volume quartz cylindrical cell with 10 cm x 1.8 cm dimensions. Behind the cell containing photolysate was mounted a quartz cylindrical cell 10 cm x 1.8 cm containing 27 mL of actinometry solution. The light absorbed was quantified the ferrioxalate actinometry using the splitting ratio method.

For compound **7**, photolysate was evaporated to remove CH_3CN. Then anhydrous benzene was added. Anhydrous sodium sulphate was added to remove water and

evaporated again under vacuum to remove remaining water and benzene. The residue was dissolved in $CDCl_3$. DMF was added as a standard for NMR analysis. Product was analyzed by ^1H NMR spectroscopy using DMF as the internal standard and conversion was 4.3%.

3.5. Supportive Information-Appendix-1

CHAPTER 4

CONCLUSIONS

Photoremovable protecting groups (PPGs) are widely used in applications in biochemistry, cellular biology, and physiology because they allow light-controlled release of bioeffectors with microscopic spatial resolution. Various PPGs have been developed for various functionalities and applications. Although a number of such PPGs are currently in use, no universal photochemically removable protecting group exists that are suitable for all applications. Some of the common protecting groups were discussed in Chapter 1. While these groups have several advantages, they also have drawbacks, such as requiring UV light, slow release rates, harmful or reactive byproduct formation, premature release of the bioeffector leaving group under aqueous conditions at high ionic strength, and limited basicity of the releasable leaving group.

Thus the major goals for this project were the following: (1) to extend the photolysis wavelength for achieving release of bioeffectors from the 350 nm region into the visible wavelength region and (2) determine the efficiency for triplet excited state energy transfer from the chromophore to the thienyl aromatic ring system that is linked to the chomophore via a carboxamide linker.

In this project a thioxanthone serves as the chromophore and is linked to a naphtho[1,2-b]thiophene via a carboxamide linker. Direct photolyses at 386 nm generates the excited singlet state, which intersystem crosses to the triplet excited state manifold. The lowest energy triplet excited state must then transfer excitation energy to the naphthothiophene. This would be followed by electrocyclization to form the

zwitterionic intermediate, which would expel the leaving group anions. Importantly, the energy transfer step would be slightly exothermic with the thioxanthone triplet excitation donor and the naphthothiophene energy acceptor. By making this energy transfer step more efficient, it was thought that the quantum yield for leaving group expulsion would increase over and above that found for a previously studied PPG, which had the thioxanthone linked to a benzothiophene ring system. In that case, $\Phi = 0.035$ for release of $LG^- = Cl^-$. The energy transfer step was considered unfavorable, energetically, comparing the energy of the chromophore (E_T = ca. 64-65 kcal mol^{-1}) to that of the benzothiophene (E_T = ca. 69 kcal mol^{-1}).

The naphthothiophene system was successfully synthesized by a multistep route and coupled to the thioxanthone to give compound **6b**. Photolysis did result in release of $LG^- = Cl^-$. However the quantum yield was somewhat disappointing. Observed quantum yield was $\Phi = 0.081$. The possibility was considered that the relatively inefficient photoreaction was due to the an inefficient energy transfer step involving the thioxanthone and naphtho[1,2-b] thiophene.

To estimate the efficiency for triplet excitation transfer from the thioxanthone chromophore to the naphthothiophene ring system in **6b**, triplet sensitized photolyses of the corresponding anilide **2-1** was studied. In anilide **2-**1 the naphtho[1,2-b]thiophene (E_T = ca.62 kcal mol^{-1}) ring is the triplet energy acceptor, and the triplet energy donors (sensitizers) xanthone (E_T = ca. 74 kcal mol^{-1}) and thioxanthone (E_T = ca. 64-65 kcal mol^{-1}). In this case we have kept the leaving group (LG^- = Cl) as chloride ion. The quantum yields obtained for direct and xanthone sensitized photolysis of napthothiophene anilide **2-1** where $\Phi = 0.17$ or $\Phi = 0.27$, respectively. Evidently, the triplet excited state

energy transfer from the thioxanthone to naphthothiophene ring system is still a somewhat inefficient process with an efficiency of 46%. This is in the context of thioxanthone E_T = ca. 64-65 kcal mol^{-1} and napthothiophene E_T = ca. 62 kcal mol^{-1}, keeping in mind that these triplet energies are only approximate. The values for Φ_{isc} for xanthone and thioxanthone was required for the calculation of energy transfer efficiency. The values for Φ_{isc} are already known for both xanthone and thioxanthone in certain nonpolar and polar solvents. But such values for 2: 1 dioxane : water containing 100 mM phosphate buffer at pH = 7 are unavailable. So value for Φ_{isc} for xanthone and thioxanthone in aq dioxane containing buffer were determined. For xanthone the Φ_{isc} in aq dioxane with buffer was found to be 0.98 and is not significantly different from the literature value of 0.97 in carbon tetrachloride. With thioxanthone the presence of a protic solvent substantially lowers Φ_{isc}. In aq dioxane Φ_{isc} was found to be 0.67.

Quenching experiments were performed to find the multiplicity of reactive excited state and the triplet lifetime. The photoreaction is quenched inefficiently at high concentrations of the triplet quencher, cyclohexadiene (E_T = ca.53 kcal mol^{-1}). For the benzothiophene linked energy acceptor **6b** the photochemistry was shown to occur in the triplet excited state, as evidenced by efficient quenching of the reaction by the triplet quencher 1,3-pentadiene and by computational studies. The lifetime of the triplet excited state in this case was 7 μs, which would be consistent with triplet energy primarily localized on the thioxanthone chromophore. The long lifetime suggests that energy transfer from the thioxanthone to the benzothiophene was rather inefficient, such that the chromophore lifetime had not been shortened appreciably by energy transfer. In the case of the naphthothiophene linked thioxanthone,**6b** the inefficient quenching observed

suggests that the lifetime of the triplet excited state is very short (ca. 2 ns) and therefore the triplet excited state is difficult to quench. This suggests that much of the triplet excitation energy is transferred to the naphthothiophene ring, which has a short lifetime due to the rapid electrocyclization and further reaction that ensues. This latter type of quenching behavior was also observed for anilide **2-1**, which was also difficult to quench with cyclohexadiene. It is noteworthy that the Stern-Volmer quenching plot for **6b** does not intersect 1 on the origin. The plot would have to curve downward to intersect unity, suggesting that a longer lived triplet excited state is being quenched at low quencher concentrations. The curved nature of the plot suggests that there are two triplet excited states involved in the reactivity of **6b**.

The photolysis of **6b** produced a single regioisomeric photoproduct. In this regard, **6b** showed a similar photochemical outcome as the previously studied benzothiophenes **3b** and **3c**, which gave a single regioisomer, whereas benzothiophene **3a** showed a pair of *N*-methyl signals in the ^{1}H NMR spectrum corresponding to two regioisomeric photoproducts produced upon direct photolysis. It was initially believed that the structure of the photoproduct of **6b** was **2-19** rather than **2-20** on the basis of precedent. Good evidence for the structural assignment of the photoproduct as **2-19** was provided by the 400 MHz ^{1}H NMR COSY spectrum, which showed six vicinal couplings as cross peaks corresponding to proton pairs b-c, h-k, g-i, i-j, j-d, e-f (Scheme 2.8) within the benzenoid rings of structure **2-19.** If the structure were **2-20**, only five vicinal couplings would have been observed, and such a structure would not account for the presence of the extra cross peaks observed in structure **2-19** in Scheme 2.8.

In addition to naptho[1,2-b]thiophene carboxamide with thioxanthone we synthesized carboxamide **7** with benzoylthiophene, which has a triplet excited state that is of lower energy than the thioxanthone chromophore. If the energy transfer is favorable, energetically, one would expect that the the photoreactivity would be more efficient than observed for the benzothiophene system **3a-c**. Very slow photolyis was actually observed 388 nm. The photoproduct absorbs at 410 nm and 425 nm. This slow reaction was seen as due to strong overlap between the absorption spectrum of the photoproduct and the photochemical starting material. The quantum yield was estimated as $\Phi = 0.004$. Although the quantum yield may be low due to aforementioned absorption of light by photoproduct in competition with the reactant, it is also thought that the compound might be inherently and unexpectedly less reactive than the benzothiophene system. The initial concern might be the electronic configuration of benzoylthiophene might not be similar to that of benzothiophene, which is expected to have a π,π^* triplet excited state. Theoretical calculation will be needed to ascertain details of the electronic configuration of the triplet excited state of the benzoylthiophene.

BIBLIOGRAPHY

CHAPTER 1

1. Kaplan, J. H.; Forbush, B. I.; Hofmann, J. F. *Biochemistry* **1978**, *17*, 1929-1935.

2. Engels, J.; Chlaeger, E. J. *J. Med. Chem*. **1977**, *20*, 907-911.

3. Pelliccioli, A. P.; Wirz, J. *Photochem. Photobiol. Sci*., **2002**, *1*, 441-458.

4. Goeldner, M.; Givens, R. (ed.), Dynamic Studies in Biology, Wiley-VCH; 2005.

5. Marriott, G., Ed., Methods in Enzymology; Academic Press: San Diego, 1998; vol, 291.

6. Zou, K.; Cheley, S.; Givens, R.S.; Bayley, H. *J. Am. Chem. Soc*. **2002**, *124*, 8220-8229.

7. (a). Marriott, G., Ed., Methods in Enzymology; Academic Press: San Diego, 1998; vol, 291. (b). Goeldner, M.; Givens, R. (ed.), Dynamic Studies in Biology, Wiley-VCH; 2005. (c). Mayer, G.; Heckel, A. *Angew. Chem. Int. Edu*. **2006**, *45*, 4900-4921.

8. (a). Zou, K.; Cheley, S.; Givens, R.S.; Bayley, H. *J. Am. Chem. Soc*. **2002**, *124*, 8220-229. (b). Walker , J.W.; Gilbert, S.H.; Drummond, R.M.; Yamada, M.; Sreekumar, R.; Carraway, R.E.; Ikebe, M.; Fay, F.S. *Proc. Natl. Acad. Sci USA*,**1998**, *98*, 1568-1573. (c). Kaplan, J. H.; Forbush, B. I.; Hofmann, J. F. *Biochemistry* **1978**, *17*, 1929-1935. (d). Canepari, M.; Nelson, L.; Papageorgiou, G.; Corrie, J. E.T.; Ogden, D. *J. Neurosci. Methods*, **2001**, *112*, 29-42.

9. (a). Chang, C. Y.; Fernandez, T.; Panchal, R.; Bayley, H. *J. Am. Chem. Soc*. **1998,** *120*, 7661. (b). Marriott, G., Ed., Methods in Enzymology; Academic Press: San Diego, 1998; *291*, 117-135.

10. Sarker, M.; Shahrin, T.; Steinmetz, M. G.*;* Timerghazin, Q. K. *Photochem.Photobiol. Sci.* **2013**, *12*, 309-322.

11. Sarker, M.; Shahrin, T.; Steinmetz, M. G. *Org. Lett*. **2011**, *13*, 872-875.

12. (a). Seixas De Melo, J.; Rodrigues, L. M.; Serpa, C.; Arnaut, L. G.; Ferreira, I. C. F.; Queiroz, M. J. R. P. *Photchem. Photobiol*. **2003**, *77*, 121-128. (b). Wex, B.; Kaafarani, B. R.; Danilov, E. O. ; Neckers, D. C. *J. Phys. Chem. A*. **2006**, *110*, 13754 -13758.

13. Allonas, X.; Ley, C.; Bibaut, C.; Jacqes, P.; Fouassier, P. *Chem. Phys. Lett*. **2000**, *322*, 483-490.

14. Zander, M.; Jacob, J.; Lee, M. L. *Z.Naturforsch*. **1987**, *42A*, 735-738.

15. Becker, R.S.; Favaro, G.; Poggi, G.; Romani, A. *J. Phys. Chem*. **1995**, *99,* 1410-1417.

16. Barltrop, J.A.; Plant, P. J.; Schofield, P. *J.Chem. Soc. Chem. Comm* **1966**, 822.

17. (a). Klan, P. *et al*. *Chem. Rev.* **2013**, *113*, 119-191. (b). Kohl-Landgraf, J.; Buhr, F.; Lefrancois, D.; Mewes, J.; Schwalbe, H.; Dreuw, A.; Wachtveitl, J. *J. Am. Chem. Soc.* **2014**, *136*, 3430-3438. (c). II'ichev, Y. V.; Wirz, J. *J. Phys. Chem*. A **2000**, *104*, 7856-7870.

18. Montanari, S.; Paradisi, C.; Scorrano, G. *J. Org. Chem*. **1999**, *64*, 3422.

19. (a). Hwang, C.; Sinskey, A. J.; Lodish, H.F. *Science*, **1992**, *257*, 1496. (b). Shaw, C. A. Ed. Tayler & Francis, Washington, D.C. Ch1, pp3-23.

20. Sheehan, J. C.; Wilson, R. M. *J. Am. Chem. Soc*. **1964**, *86*, 5277–5281.

21. Givens, R. S.; Matuszewski, B. *J. Am. Chem. Soc.* **1984**, *106*, 6860.

22. Greene, T. W.; Wutts, P. G. M. Protective Groups in Organic Synthesis; Wiley-Interscience: New York, 2006.

23. Givens, R. S.; Yousef, A. l. p-hydroxyphenacyl: A photoremovable protecting group forcaging bioactive substrates. in dynamic Studies in biology; Goelender, M., Givens, R. S., Eds.; Wiley VCH: Weinheim, germany, 2006: p 55. (b). Givens, R. S.; Lee, J. L. *J. Photosci* **2003**, 10, 37. (c) Givens, R. S.; Weber, J. F. W.; Jung, A. H.; Park, C. H. New photoprotecting groups: Desyl and p-hydroxyphenacyl phosphate and carboxylate esters. In Methods Enzymol.; Marriot, G., Ed.; Academic Press: New York, 1998; *Vol.291*,p1

24. Givens, R.S.; Rubina, M.; Wirz, *J. Photochem. Photobiol. Sci*. **2012**, *11*, 472.

25. Givens, R. S.; Heger, D.; Hellrung, B.; Kamdzhilov, Y.; Mac, M.; Conrad, P. G.; Cope, E.; Lee, J. I.; Mata-segreda, J. F.; Schowen, R. L.; Wirz, J. *J. Am. Chem. Soc.* **2008**, *130*, 3307-3309.

26. Balachandran Kamath, V.; Solomek, T.; Ngoy, b. P.; Heger, D.; Klan, P.; Rubina, M.;Givens, R. S. *J. Org. Chem*. 2013.

27. Hagen, V.; Bendig, J.; Frings, S.; Eckardt, T.; Helm, S.; Reuter, D.; Kaupp, U. B. *Angew, Chem. Int. Ed*. **2001**, *40*, 1046-1048.

28. Suzuki, A. Z.; Watanabe, T.; Kawamoto, M.; Nishiyama, K.; Yamashita, H.; Ishii, M.; Iwamura, M.; Furuta, T. *Org. Lett.* **2003**, *5*, 4867-4870.

29. (a). Schade, B.; Hagen, V.; Schmidt, R.; Herbrich, R.; Krause, E.; Eckardt, T.; Bendig, J. *J. Org. Chem*. **1999**, *64*, 9109. (b). Eckardt, T.; Hegan, V.; Schade, b,; Schmidt, R.; Schweitzer, C.; Bending, J. *J. Org. Chem.* **2002**, *67*, 703-710. (c). Schmidt, R.; Gessler, D,; Hagen, V.; Bendig, J. *J. Phys. Chem. A* **2007**, *111*, 5768-5774.

30. Pirrung, M. C.; Fallon, L.; McGall, G. *J. Org. Chem*. **1998**, *63*, 241-246.

31. Fodor, S. A. P.; Read, J. L.; Pirrung, M. C.; Stryer, L.; Tsai Lu, A; Solas, D. *Science*, **1991**, *251*, 767-773.

32. Corrie, J. E. T.; Trentham, D. R. Biological Application of Photochemical Switches. Vol2, ed. H.Morrison, Wiley, **1993**, pp. 243-305.

33. Adams, S. R.; Tsien, R. Y. *Annu. Rev. Physiol*. **1993**, *55*, 755-784.

34. Mayer, G.; Heckel, A. *Angew. Chem. Int. Ed*. **2006**, *45*, 4900-4921.

35. Gee, K. R.; Carpenter, B. K.; Hess, G. P. *Methods. Enzymol*. **1998**, *291*, 30-50.

36. Grewer, C.; Jager, J.; Carpenter, B. K.; Hess, G. P. *Biochemistry*, **2000**, *39*, 2063.

37. Kandler, K.; Katz, L. C.; Kauer, J. A. *Nat. Neurosci.* **1998**, *1*, 119-123.

38. Hansen, K.C.; Rock, R. S.; Larsen, R. W.; Chan, S. I. *J. Am. Chem. Soc*. **2000**, *122*, 11567-11568.

39. Adams, S.R.; Lev-Ram, V.; Tsien, R. Y. *Chem. Biol*. **1997**, *4* , 867-878.

40. (a) Ninomiya, I.; Naito, T. H*eterocycles*, **1981**, *15*, 1433-1462. (b) Lenz, G. R. *Synthesis*, **1978**, 489-518. (c) Campbell, A. L.; Lenz, G. R. *Synthesis*, **1987**, 421-452.

41. Kanaoka, Y.; Itoh, K.; Hatanaka, Y.; Flippen, J. L.; Karle, I. L.; Witkop, B. *J. Org Chem*. 1**975**, *40*, 3001–3003.

42. Kudo, H.; Castle, R. N.; Lee, M. L. *J. Heterocyclic Chem.* **1985**, *22*, 211-214.

43. Klan, P.; Wirz, J. *Photochemistry of organic compounds: From concepts to practice* ; Wiley: Chichester, U.K., 2009.

44. Wöll, D.; Walbert, S.; Stengele, K. P.; Albert, T. J.; Richmond, T.; Norton, J.; Singer, M.; Green, R. D.; Pfleiderer, W.; Steiner, U. E. *Helv. Chim. Act.* **2004**, *87*, 28.

45. Wöll, D.; Laimgruber, S.; Galetskaya, M.; Smirnova, J.; Pfeiderer, W.; Heinz, B.; Gilch, P.; Steiner, U. E. *J. Am. Chem. Soc*. **2007,** *129*, 12148.

CHAPTER 2

1. Sarker, M.; Shahrin, T.; Steinmetz, M. G.*;* Timerghazin, Q. K. *Photochem. Photobiol. Sci.* **2013**, *12*, 309-322.

2. Zander, M.; Jacob, J.; Lee, M. L. *Z.Naturforsch*. **1987**, *42A*, 735-738.

3. Moon, J. K.; Park, J. W.; Lee, W. S.; Kang, Y. J.; Chung, H. A.; Shin, M. S. ; Yoon, Y. J. *J. Heterocyclic. Chem*. **1999**, *36*, 793-798.

4. Summers, J.B.; Hormoz, M.; Holms, J.H.; Ratajczyk, J.D., Dyer, R.D., Carter, G.W. *J. Med. Chem.* **1987**, *30*, 574-580.

5. Pelliccioli, A. P.; Wirz, *J. Photochem. Photobiol. Sci*., **2002**, *1*, 441-458.

6. Goeldner , M.; Givens, R. (ed.), Dynamic Studies in Biology, Wiley-VCH; 2005.

7. Sarker, M.; Shahrin, T.; Steinmetz, M. G. *Org. Lett*. **2011**, *13*, 872-875.

8. Neumann, M. G.; Gehlen, M. H.; Encinas, M. V.; Allen, N. S.; Corrales, T.; Peinado, C.; Catalina, F. *J. Chem. Soc., Faraday Transactions*, **1997**, *93*, 1517-1521

9. Zimmermann, H. E.; Jian-hau, X.; King, R. K.; Caufield, C. E. *J. Am. Chem. Soc*.

1985,*107*, 724-7732.

10 . (a).Hammond, G. S.; Saltiel, J.; Lamola, A. A.; Turro, N. J.; Bradshaw, J. S.; Cowan, D. O.; Counsell, R. C.; Vogt, V.; Dalton, C. J. *Am. Chem. Soc*. **1964**, *86*, 3197-3217. (b). Lamola, A. A.;Hammond, G. S. *J. Chem. Phys*. **1965**, *43*, 2129-21135.

11. Scaiano, J. C. *J. Am. Chem. Soc*. **1980**, *102*, 7747-7753.

12. Rubio-Pons, O.; Serrano-Andres, A.; Burget, D.; Jacques, A. *J. Photchem. Photobiol.*

A: Chem. **2006**, 179,298-304.

13. Merkel, P. B.; Dinnocenzo, J. P. *J. Photochem. Photobiol.,A.* **2008**, *193*, 110-121.

14. Rai-Constapel, V.; Etinski, M.; Marian, C. M. *J. Phys. Chem. A* **2013**,*117*, 3935-3944.

15. Amstutz, E. D.; Neumoyer, C. R. *J. Am. Chem. Soc*. **1947**, *69*, 1925-1929.

16. 47. Sarker, M. "Photochemical Eilmination Reactions via Zwitterionic Intermediates Generated byElectrocyclic Ring Closures" (2012). *Dissertations (2009).* Paper 239 http://epublications.marquette.edu/dissertations_mu/239.

17. Summers, J.B.; Hormoz, M.; Holms, J.H.; Ratajczyk, J.D., Dyer, R.D., Carter, G.W. *J. Med. Chem.* **1987**, *30*, 574-580.

18. Kudo, H.; Castle, R.N.; Lee, M. L. *J. Heterocyclic Chem.* **1985**, *22*, 211-214.

19. Wendt, M. D.; Rockway, T. W.; Geyer, A.; McClellan, W.; Weitzberg, M.; Zhao, X.; Mantei, R.; Nienaber, V. L.; Stewart, K.; Klinghofer, V.; Giranda, L. *J. Med. Chem*. **2004,** *47*, 3030-324.

20. Diaz, Phillippe *et al.* PCTInt. Apll., 2004046096, 2004. (b). Livingstone *et al.*, U.S. Pat. Appl. Publ., 2004204455**, 2004**.

21. Herbert, C. B.; Krishnamurthy, S.; Thomas, P. S. *J. Org. Chem*. **1973**, *73*, 2786-2792.

22. Corey, E. J.; Suggs, J. W. *Tetrahedron Lett.* **1975**, *31*, 2647-2650.

23. Hatchard, C. G.; Parker, C. A. *Proc. Roy. Soc (London),* **1965**, *A253*, 518.

24. Zimmermann, H. E.; Hancock, G.; K. *J. Am. Chem. Soc*. **1968**, *90*, 3749-3758.

CHAPTER 3

1. Becker, R.S.; Favaro, G.; Poggi, G.; Romani, A. *J. Phys. Chem*. **1995**, *99,* 1410-1417.

2. (a) Zimmermann, H. E., *Mol. Photochem*. **1971**, *3*, 281–292; (b) Nicolauo, K.C.; *Angew. Chem. Int. Ed*. **2005**, *44*, 1378 – 1382.

3. Alexander, O.; Hinsberger, S.; Werth, R.; Sandrine, M. O.; Martin, F.; Rolf, W. H. *J. Med. Chem.* **2010**, *53*, 8176-8186.

4. Angibaud, P. R.; Venet, M. G.; Filliers, W.; Broecks, R.; Lingy, Y. AA.; Muller, P.; Poncelet, V. S.; End, D. W. Eur. *J. Org. Chem.* **2004**, *3*, 479-486.

5. Kartitzky, A. R.; Jayaram C.; Vassilatos, S. N. *Tetrahedon,* **1983**, *39*, 2023-2029.

6. Babler, J. H.; Malek, N. C.; Coghlan, M. J. *J. Org. Chem*. **1978**, *43*, 1821-1823.

Appendix 1 : NMR Spectra

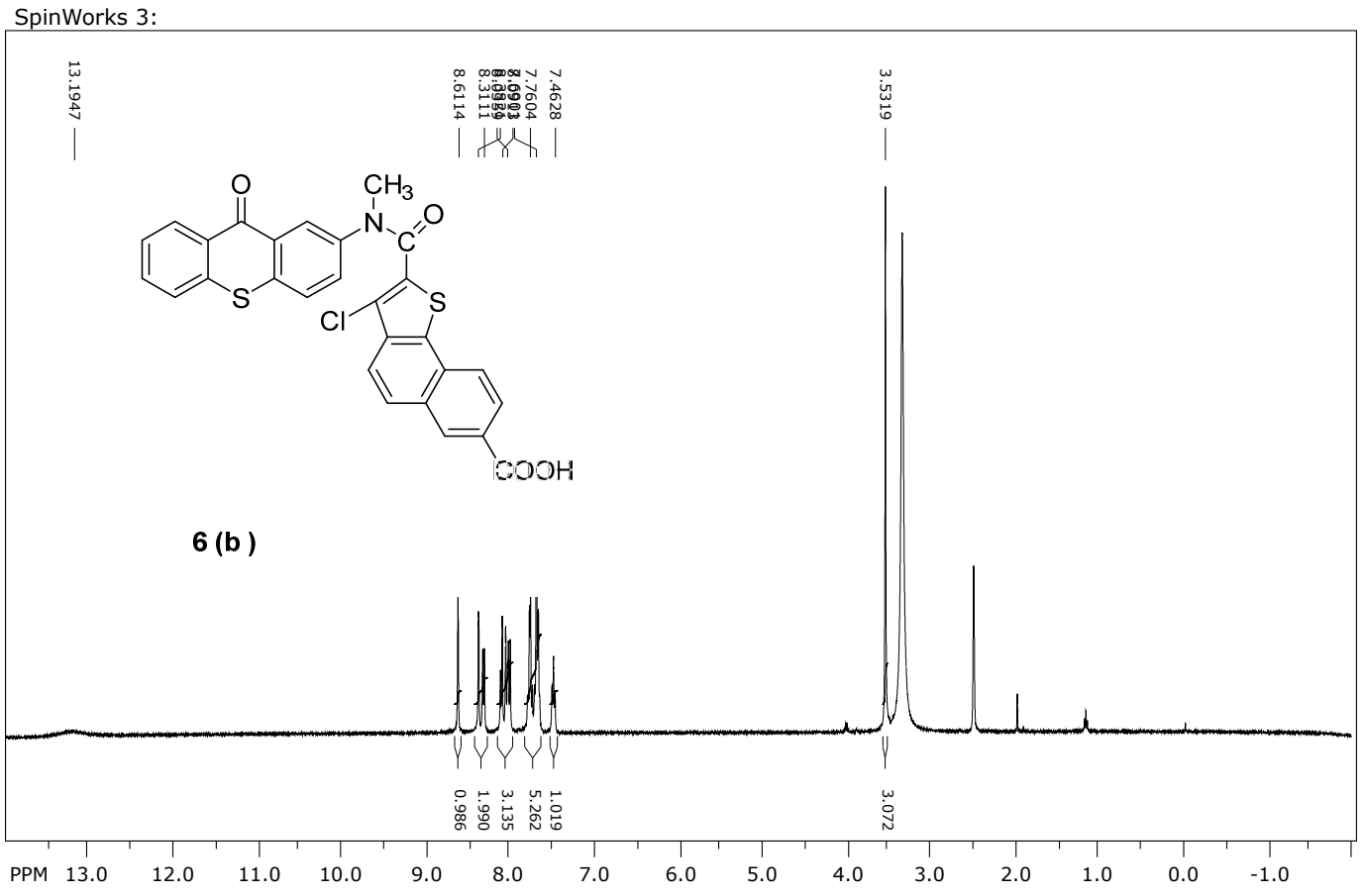


Figure 1. ^{1}HNMR spectrum of compound **6 (b)** in DMSO-d_6

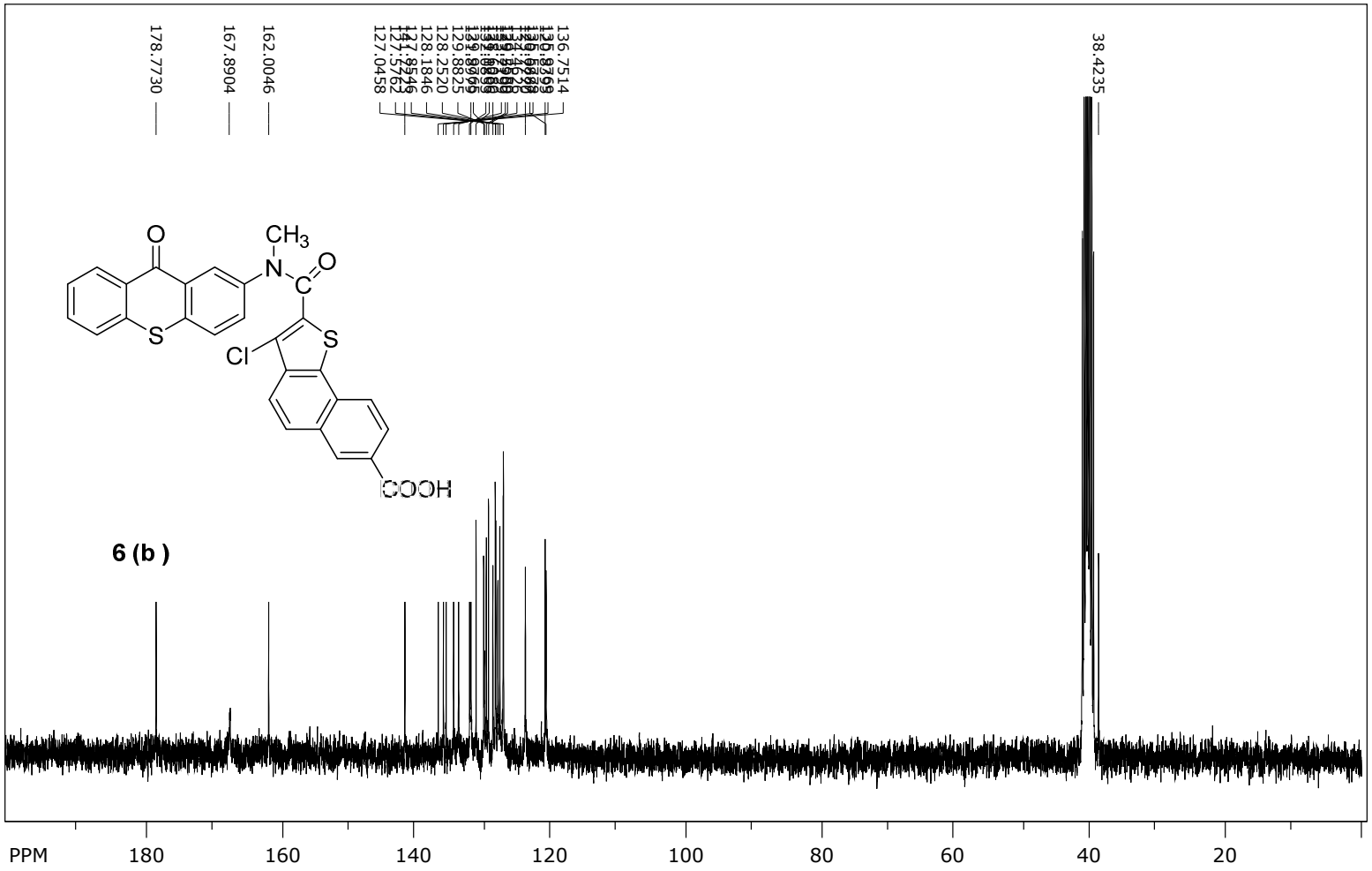


Figure 2. ^{13}C NMR spectrum of compound **6 (b)** in DMSO-d_6

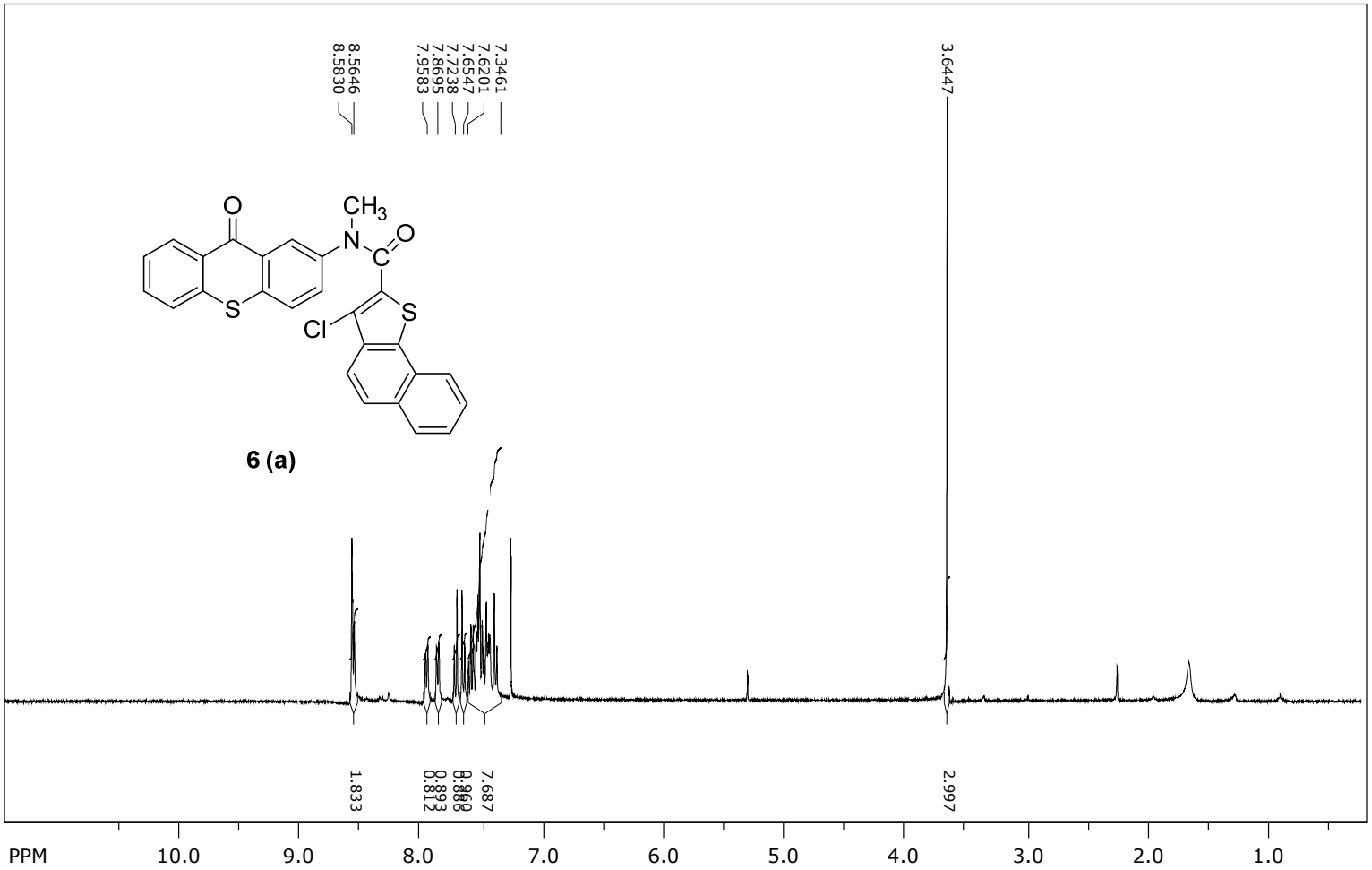


Figure 3. ^{1}H NMR spectrum of compound **6 (a)** in $CDCl_3$

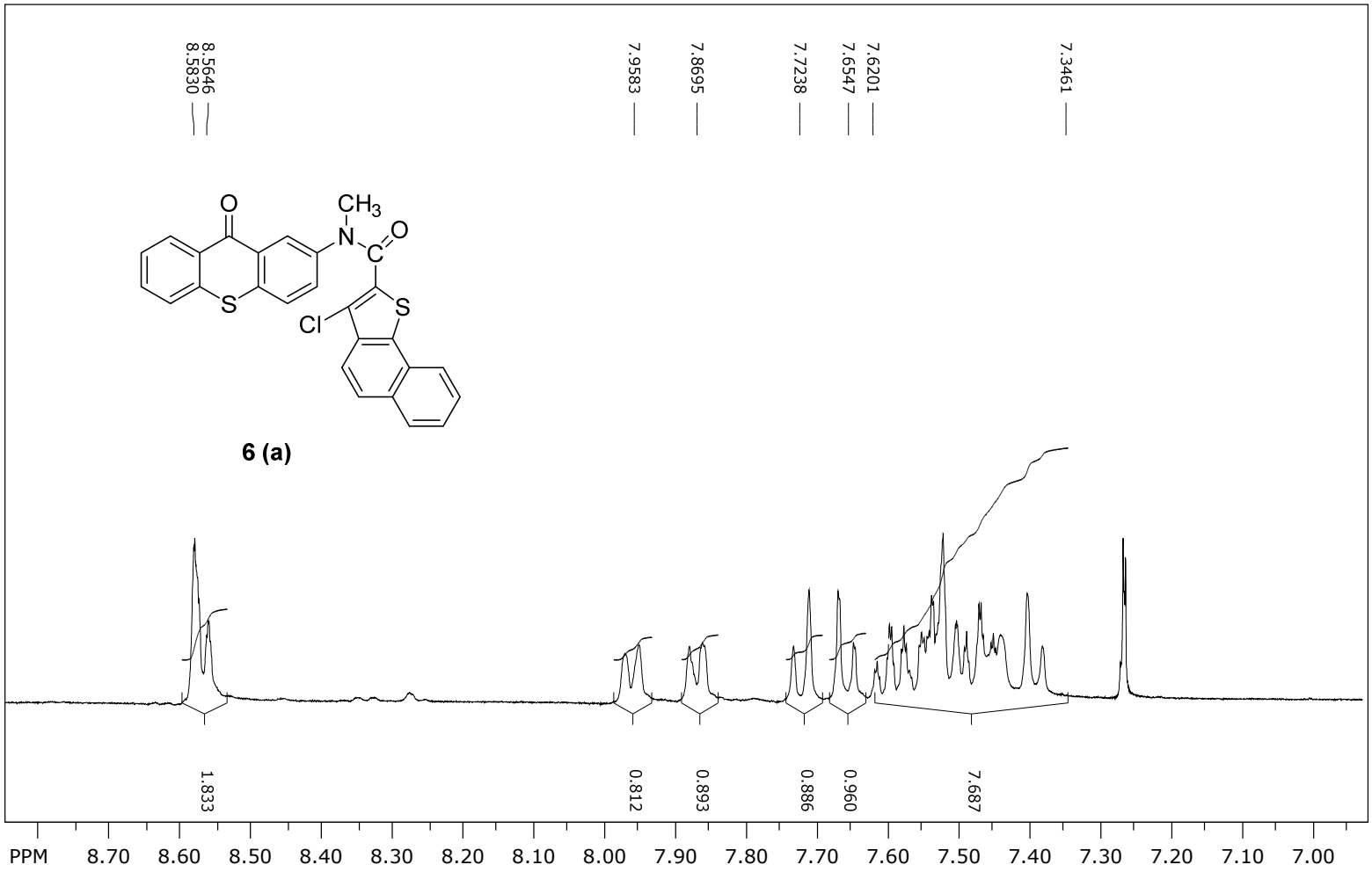


Figure 4. Expansion of the aromatic region of **Figure 3**

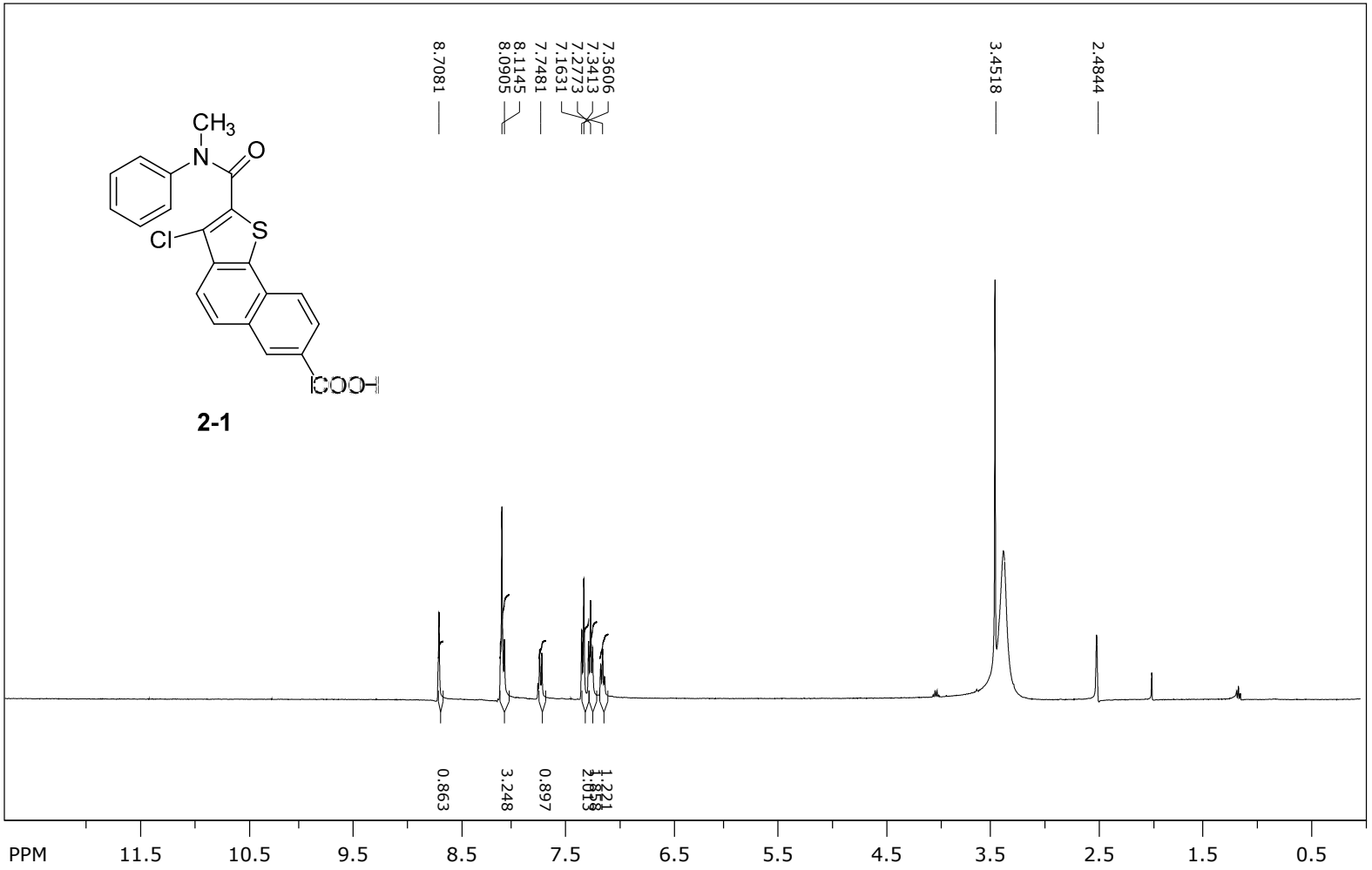


Figure 5. ^{1}H NMR spectrum of compound **2-1** in DMSO-d_6

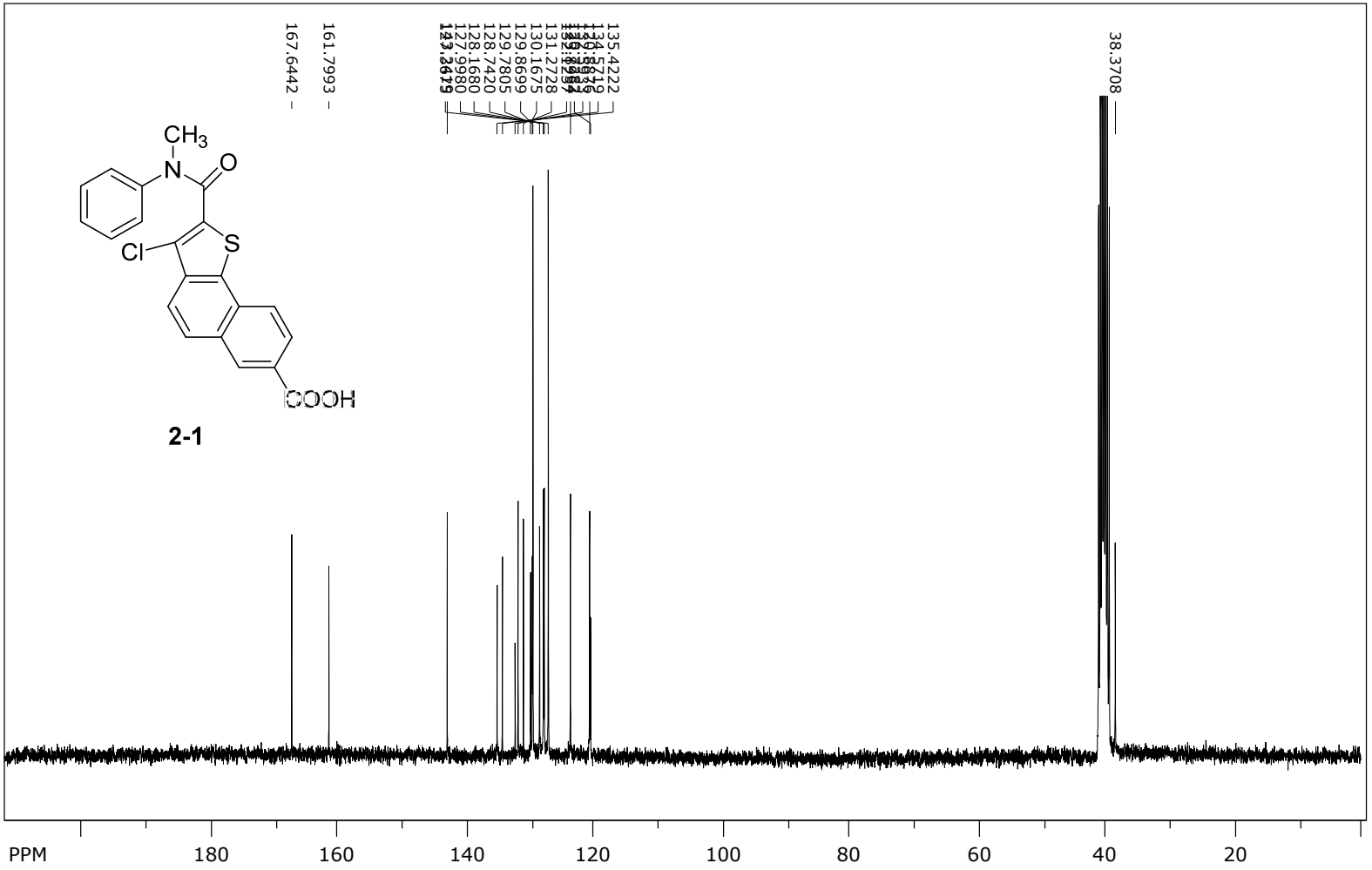


Figure 6. ^{13}C NMR spectrum of compound **2-1** in DMSO-d_6

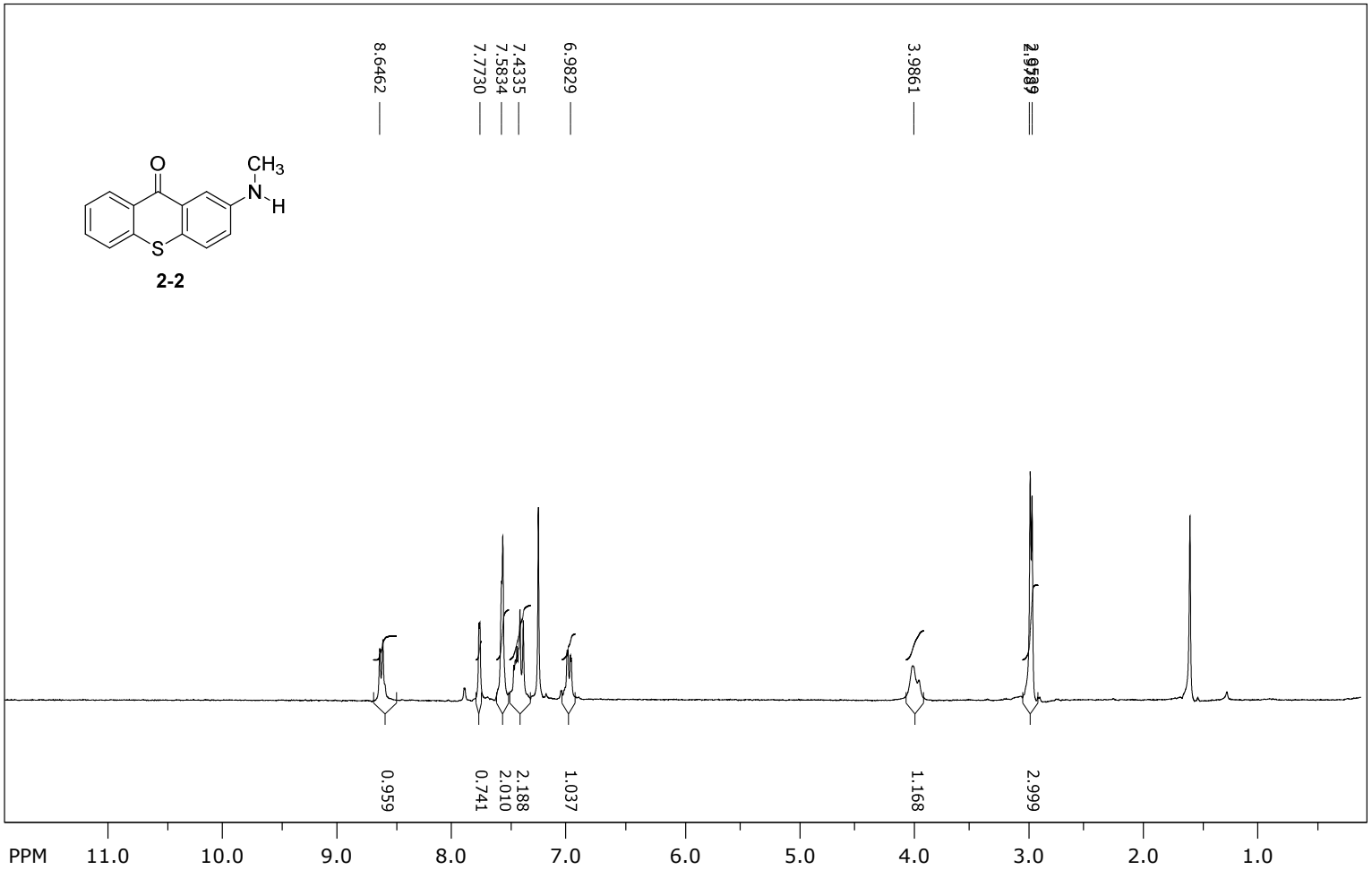


Figure 7. ^{1}H NMR spectrum of compound **2-2 in $CDCl_3$**

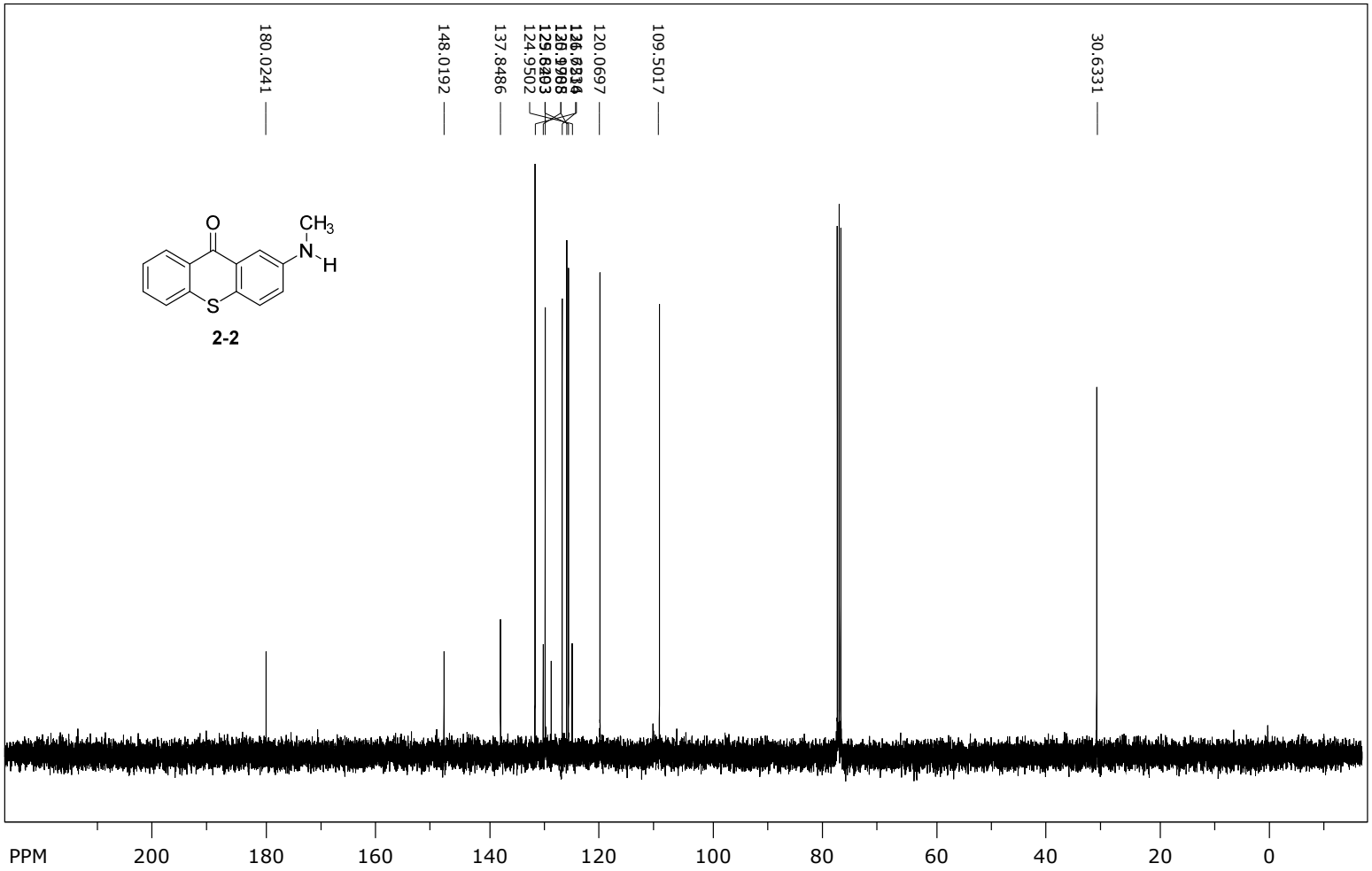


Figure 8. ^{13}C NMR spectrum of compound **2-2** in $CDCl_3$

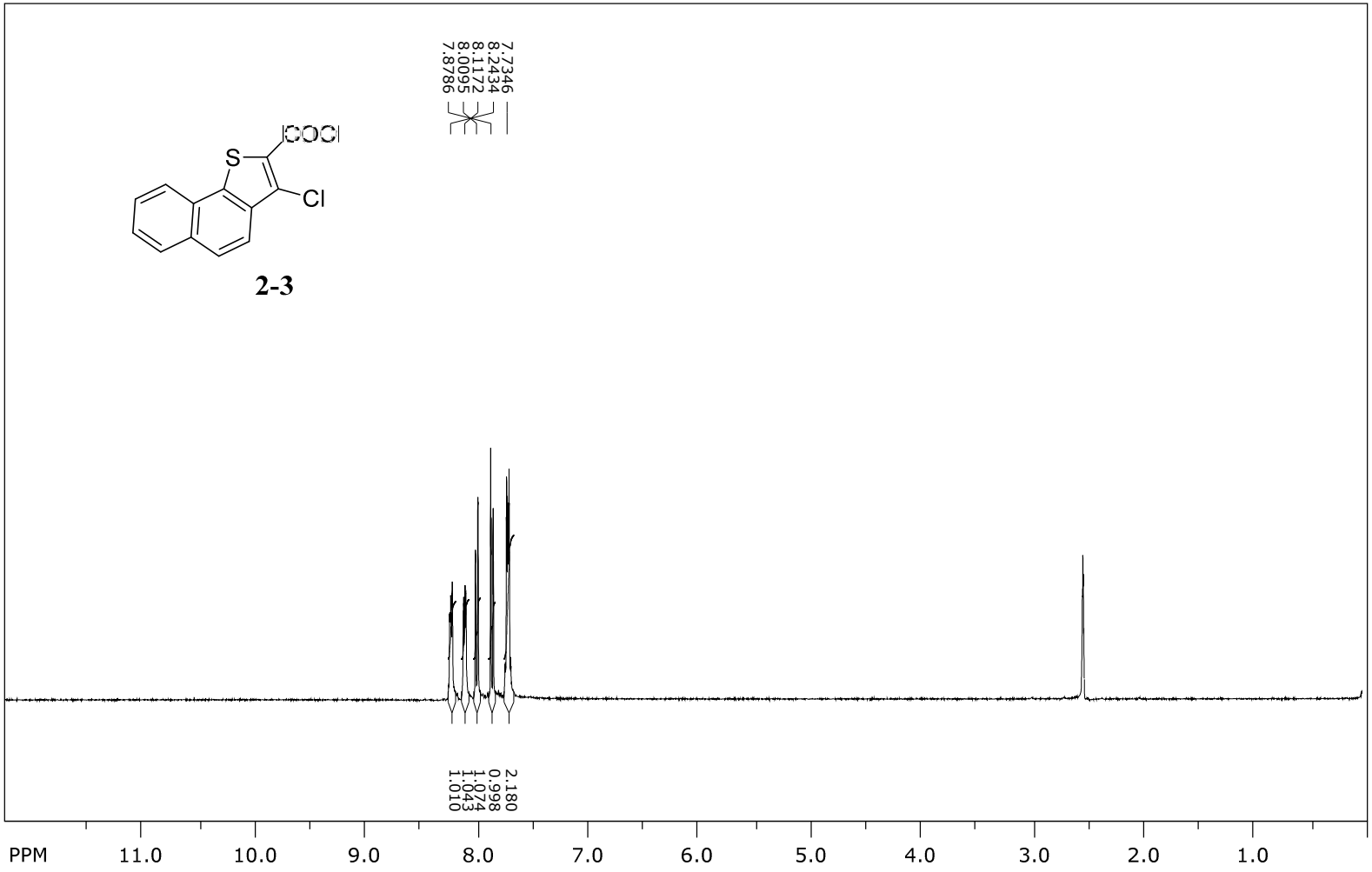


Figure 9. ^{1}H NMR spectrum of compound **2-3** in DMSO-d_6

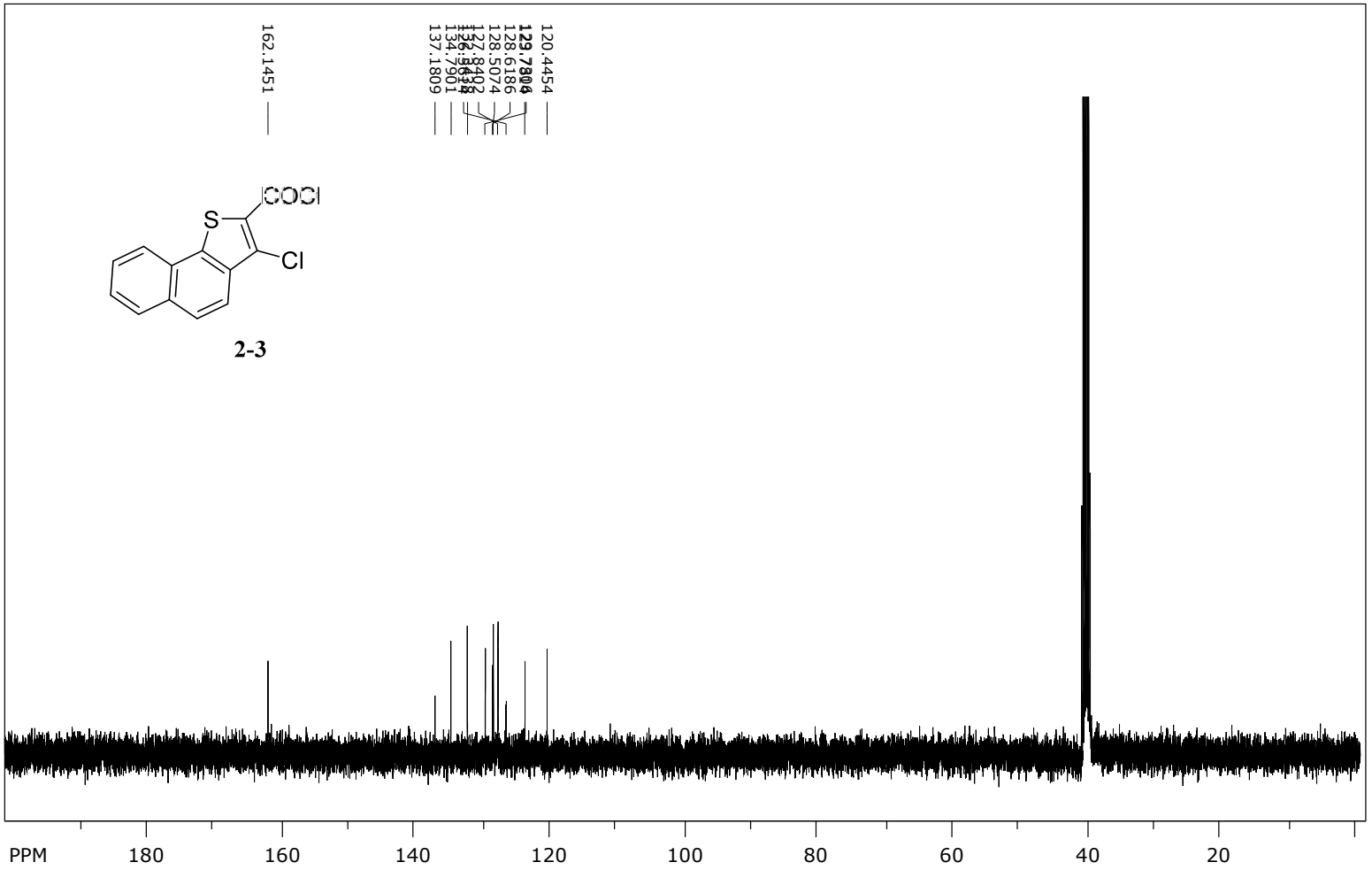


Figure 10. ^{1}C NMR spectrum of compound **2-3** in DMSO-d_6

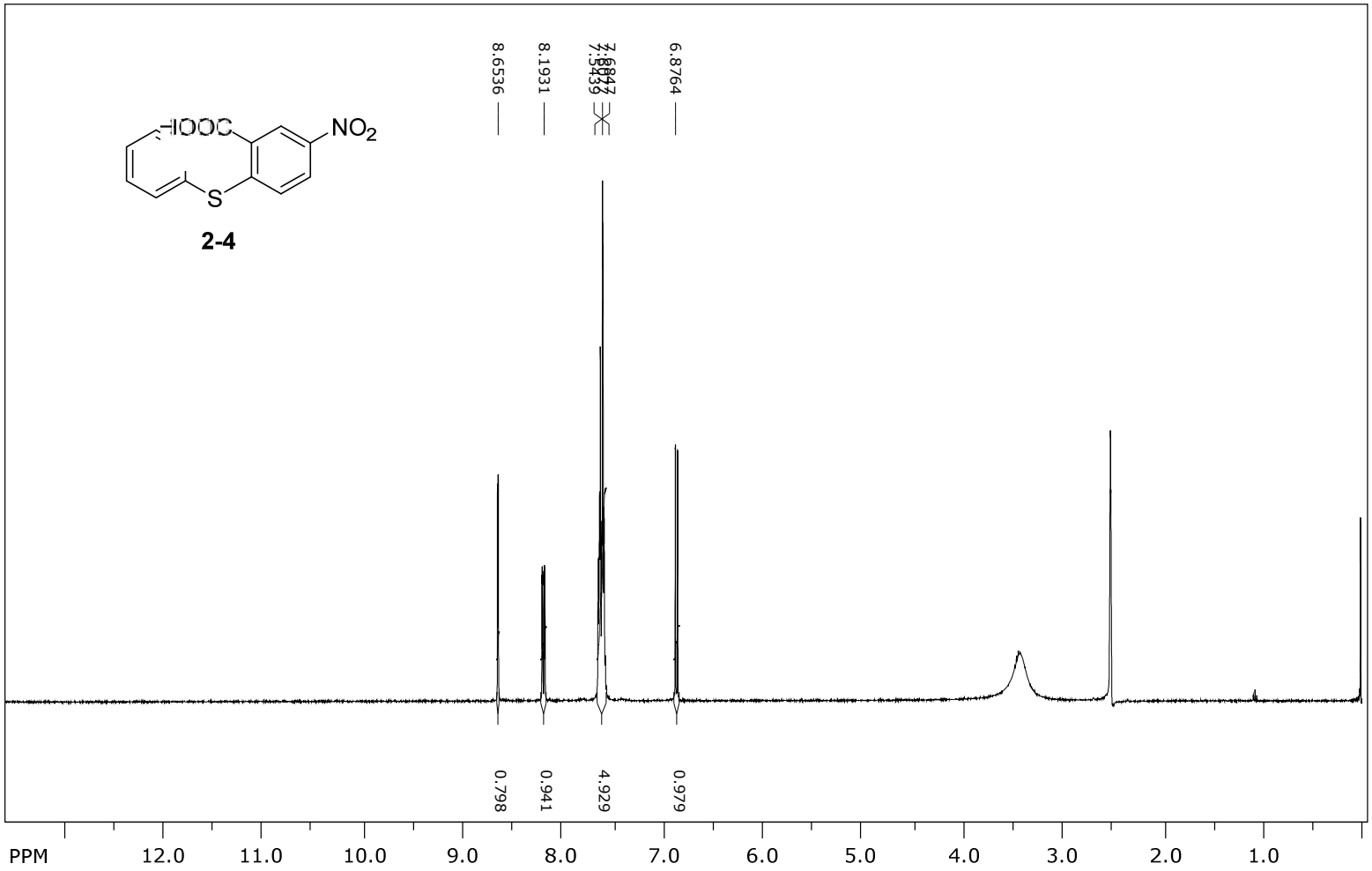


Figure 11. ^{1}H NMR spectrum of compound **2-4** in DMSO-d_6

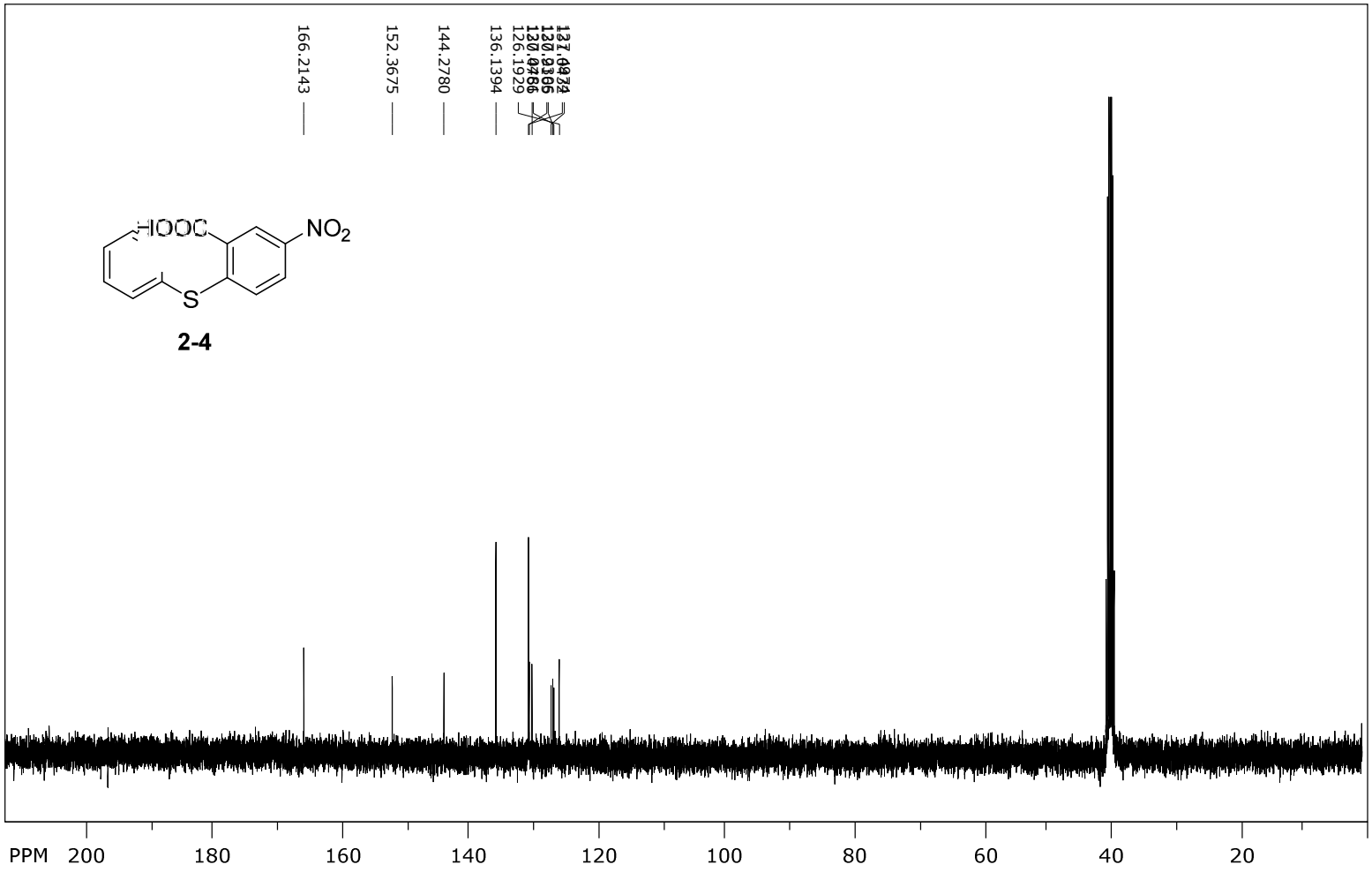


Figure 12. ^{1}C NMR spectrum of compound **2-4** in DMSO-d_6

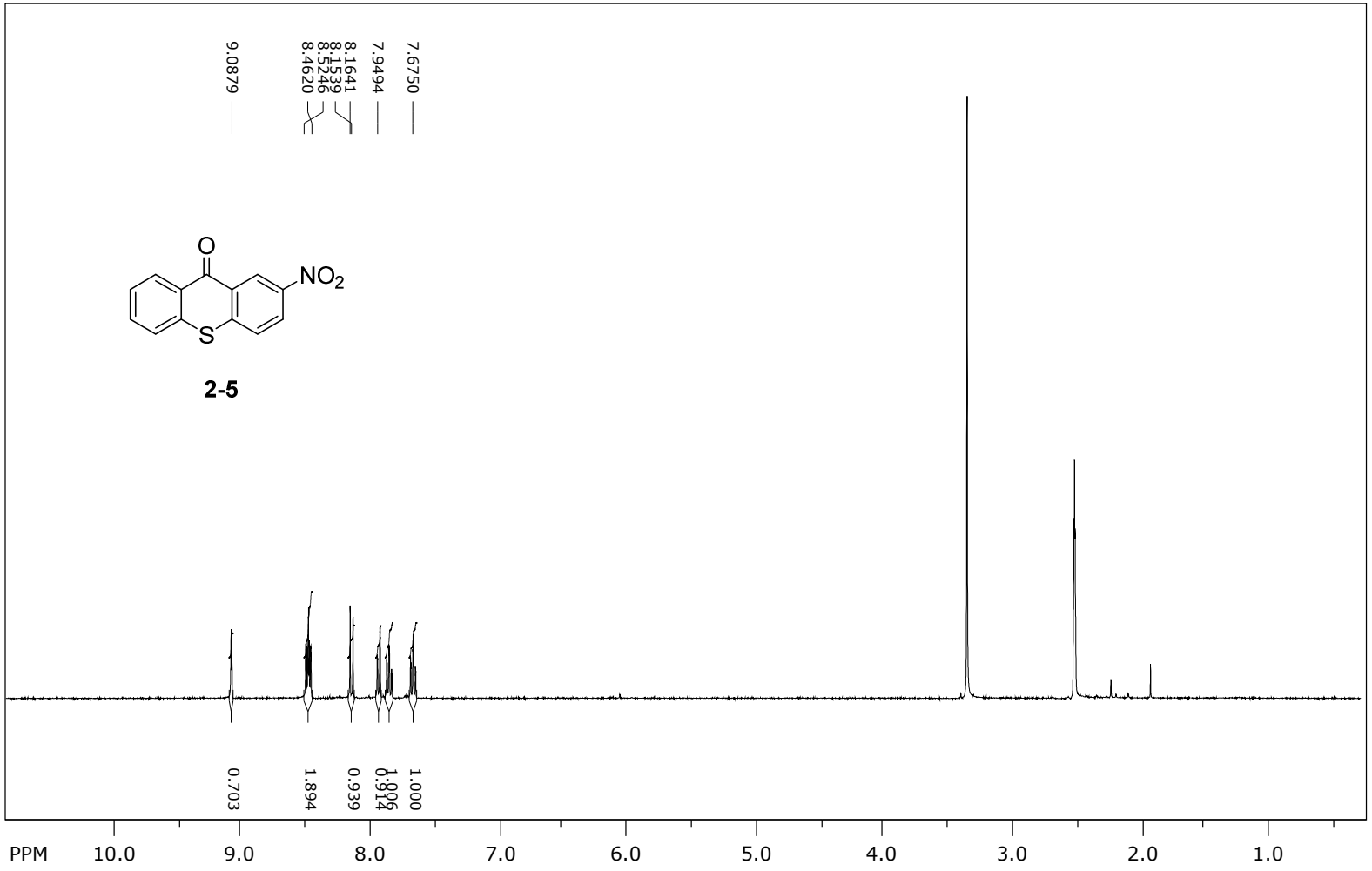


Figure 13. [1] H NMR spectrum of compound **2-5** in DMSO-d_6

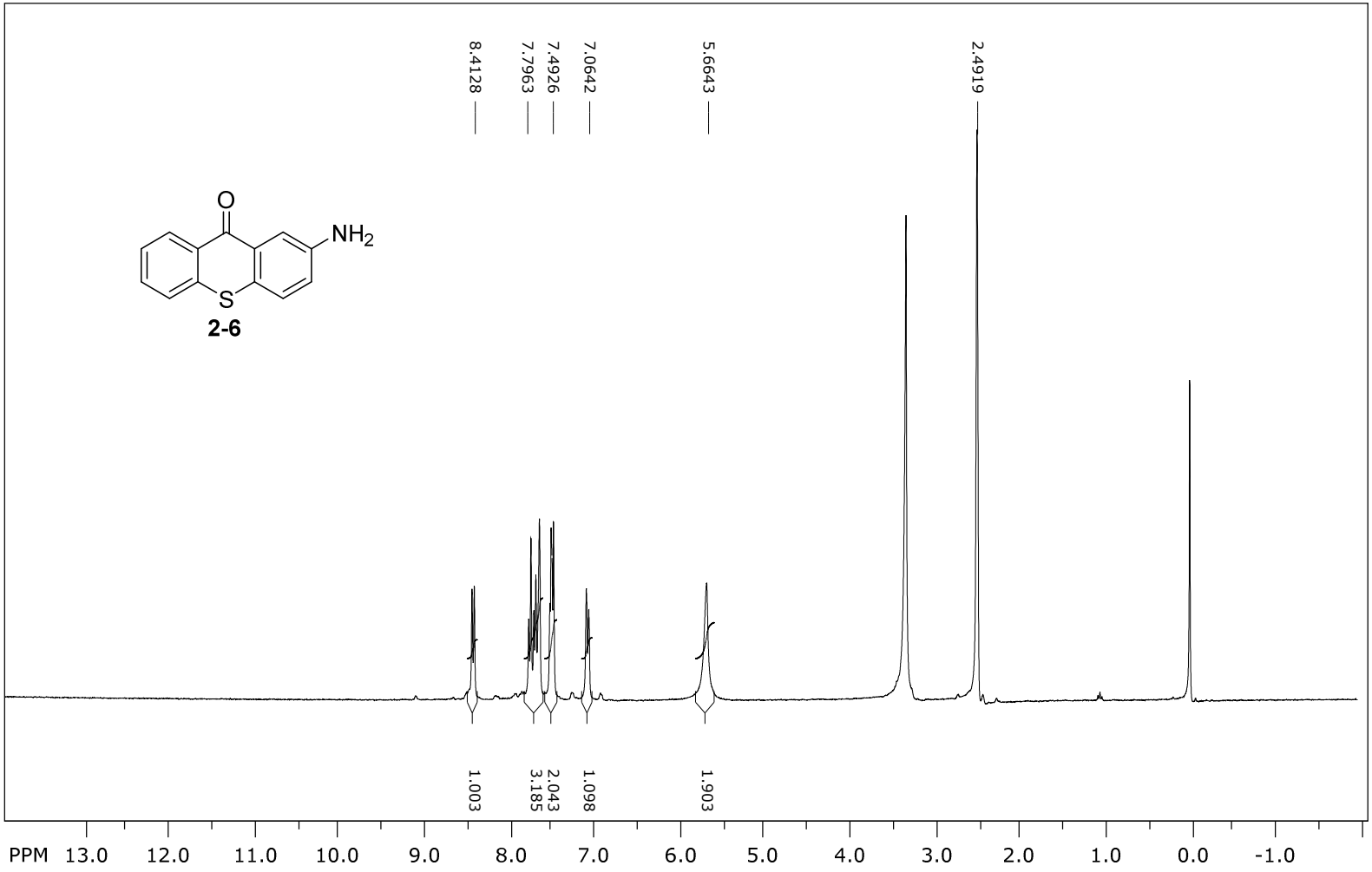


Figure 14. [1] H NMR spectrum of compound **2-6** in DMSO-d_6

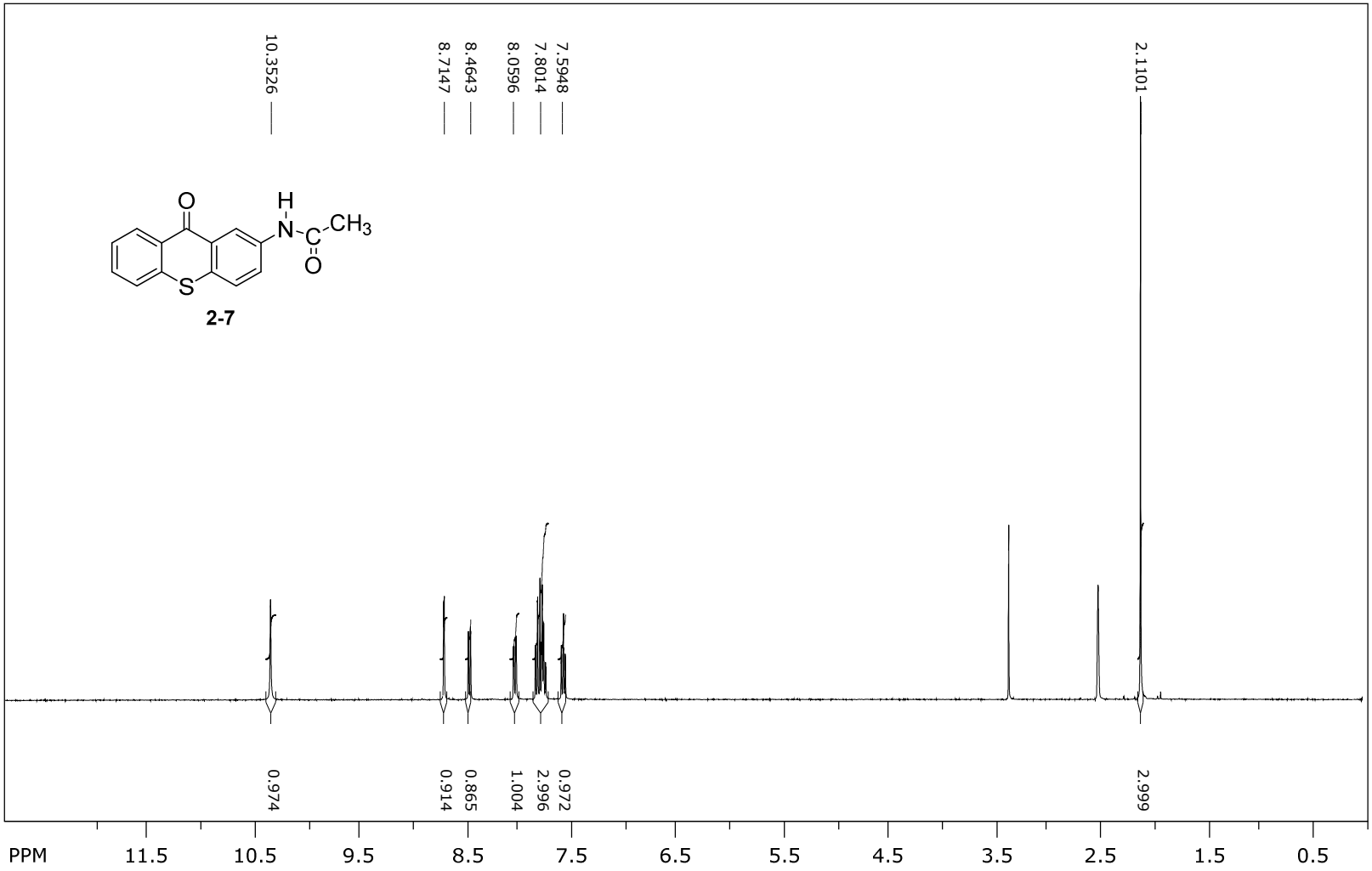


Figure 15. ^{1}H NMR spectrum of compound **2-7** in DMSO-d_6

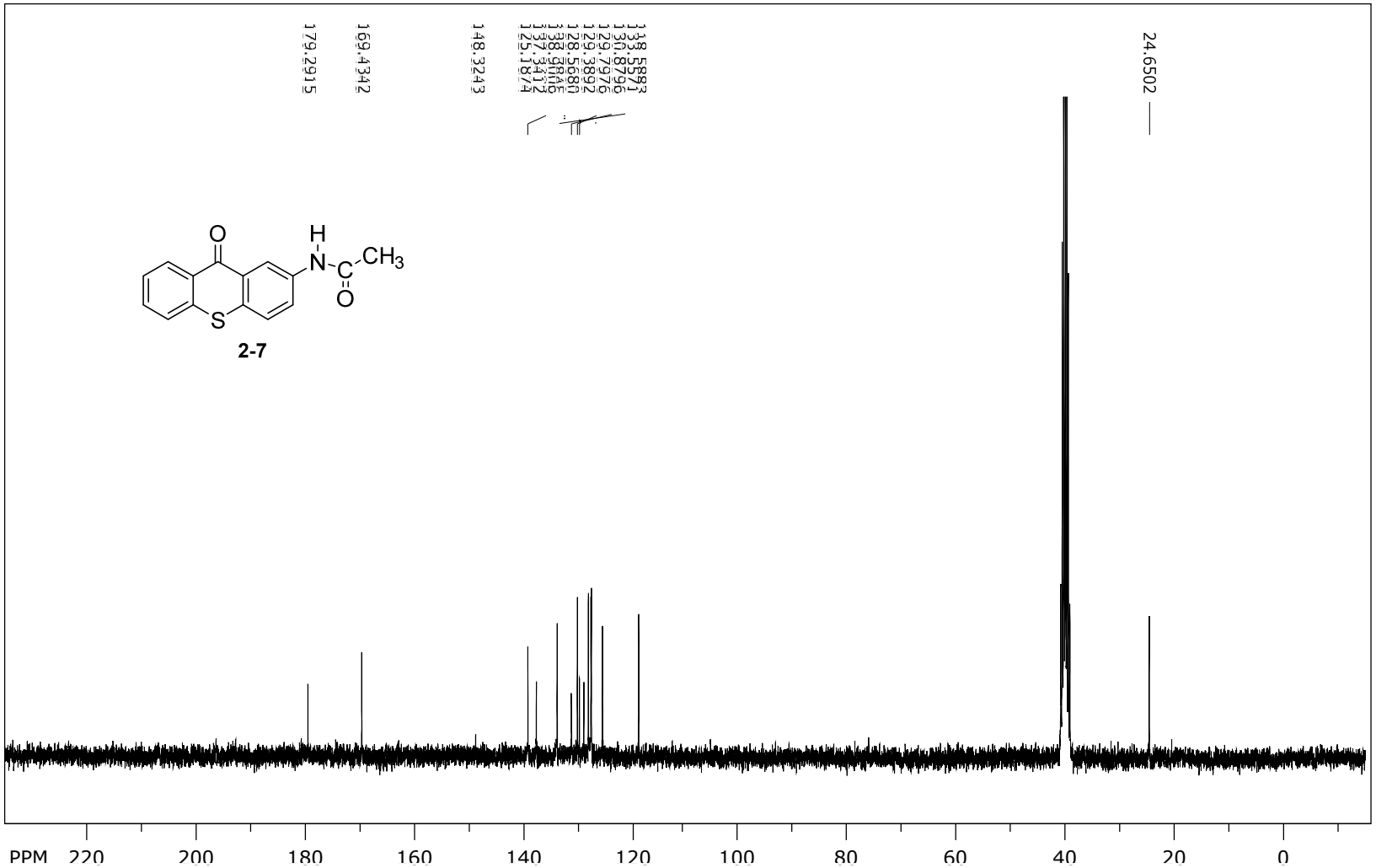


Figure 16 ^{13}C NMR spectrum of compound **2-7** in DMSO-d_6

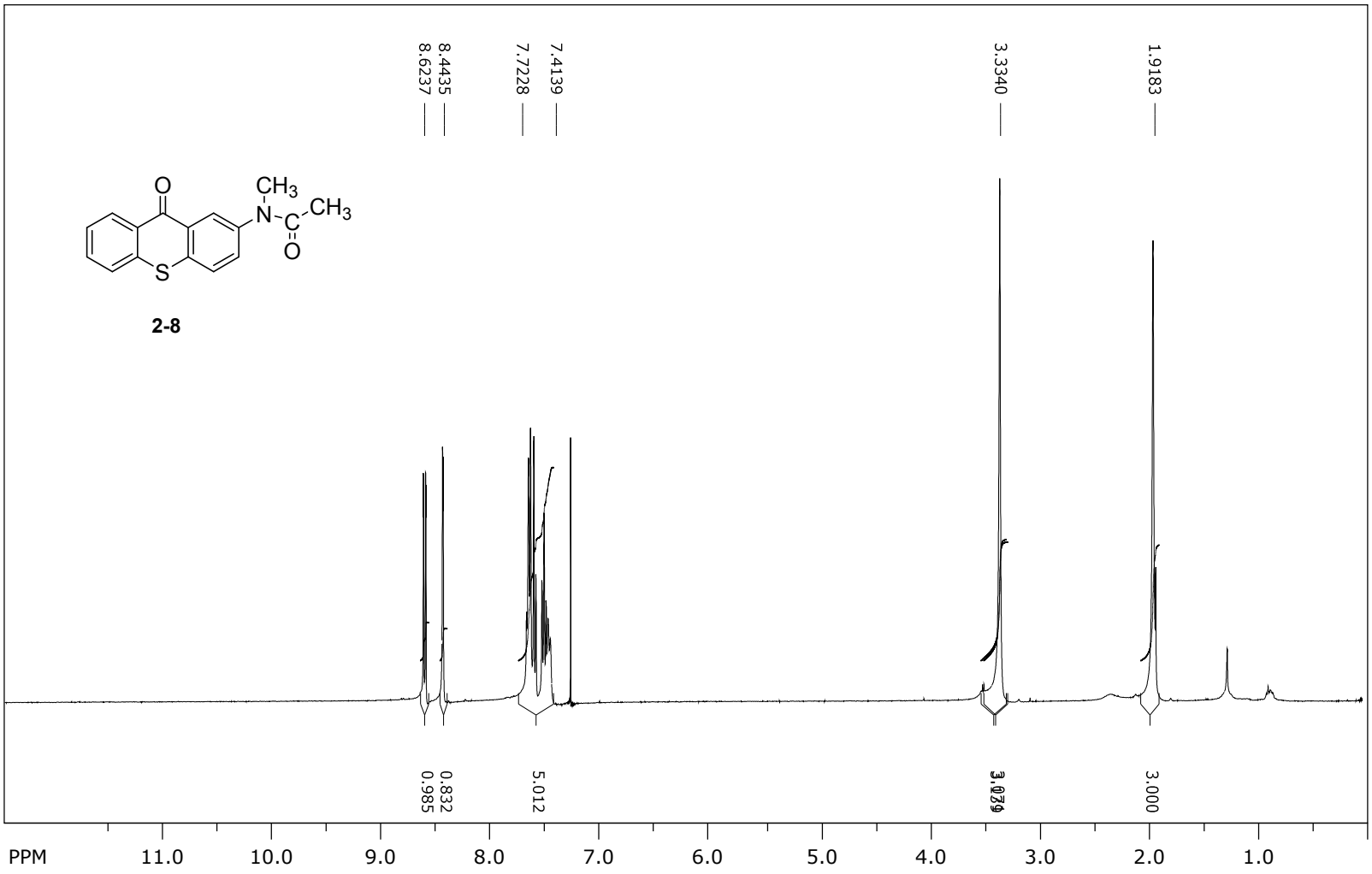


Figure 17 ^{1}H NMR spectrum of compound **2-8** in $CDCl_3$

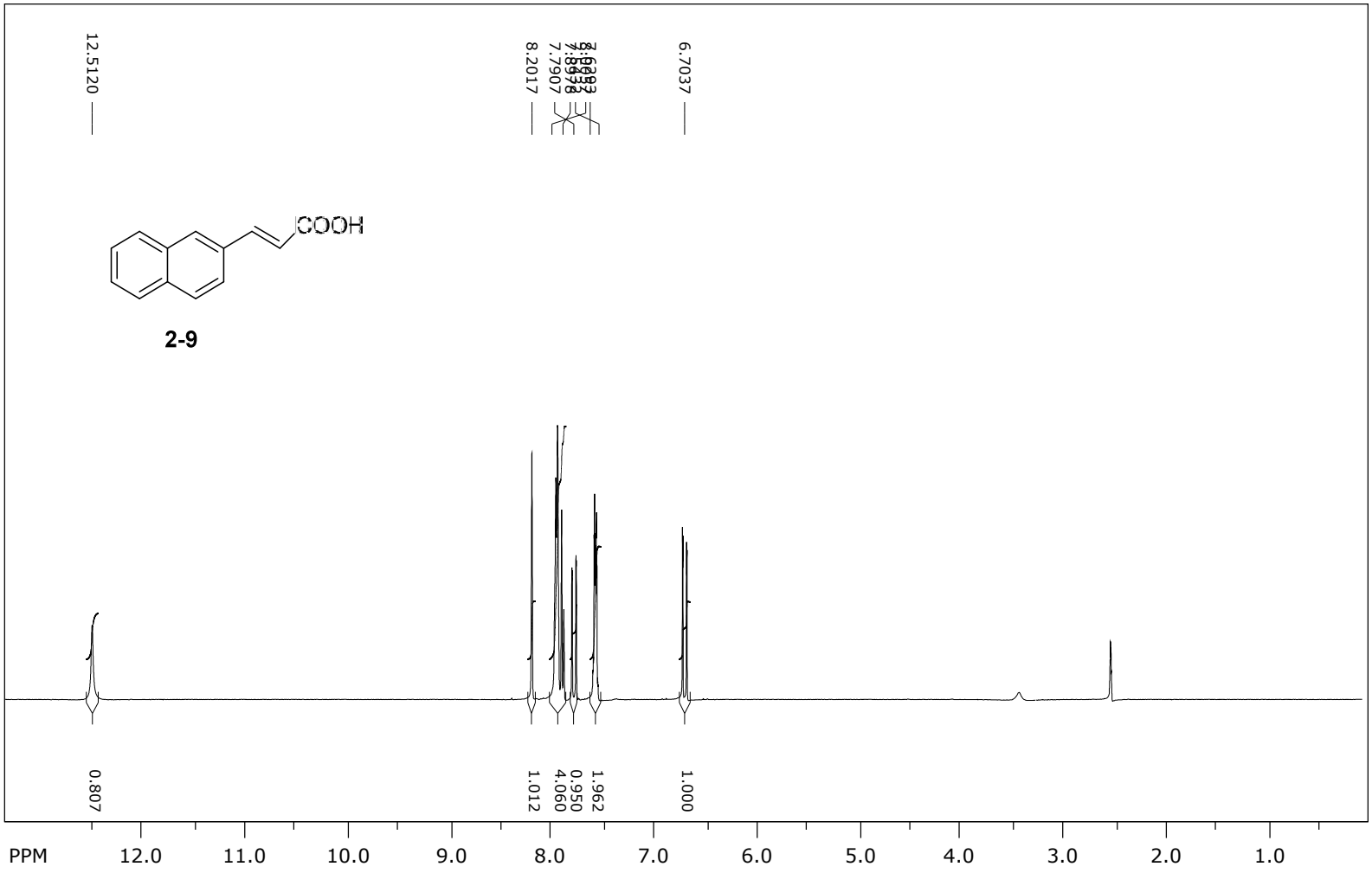


Figure 18. ^{1}H NMR spectrum of compound **2-9** in DMSO-d_6

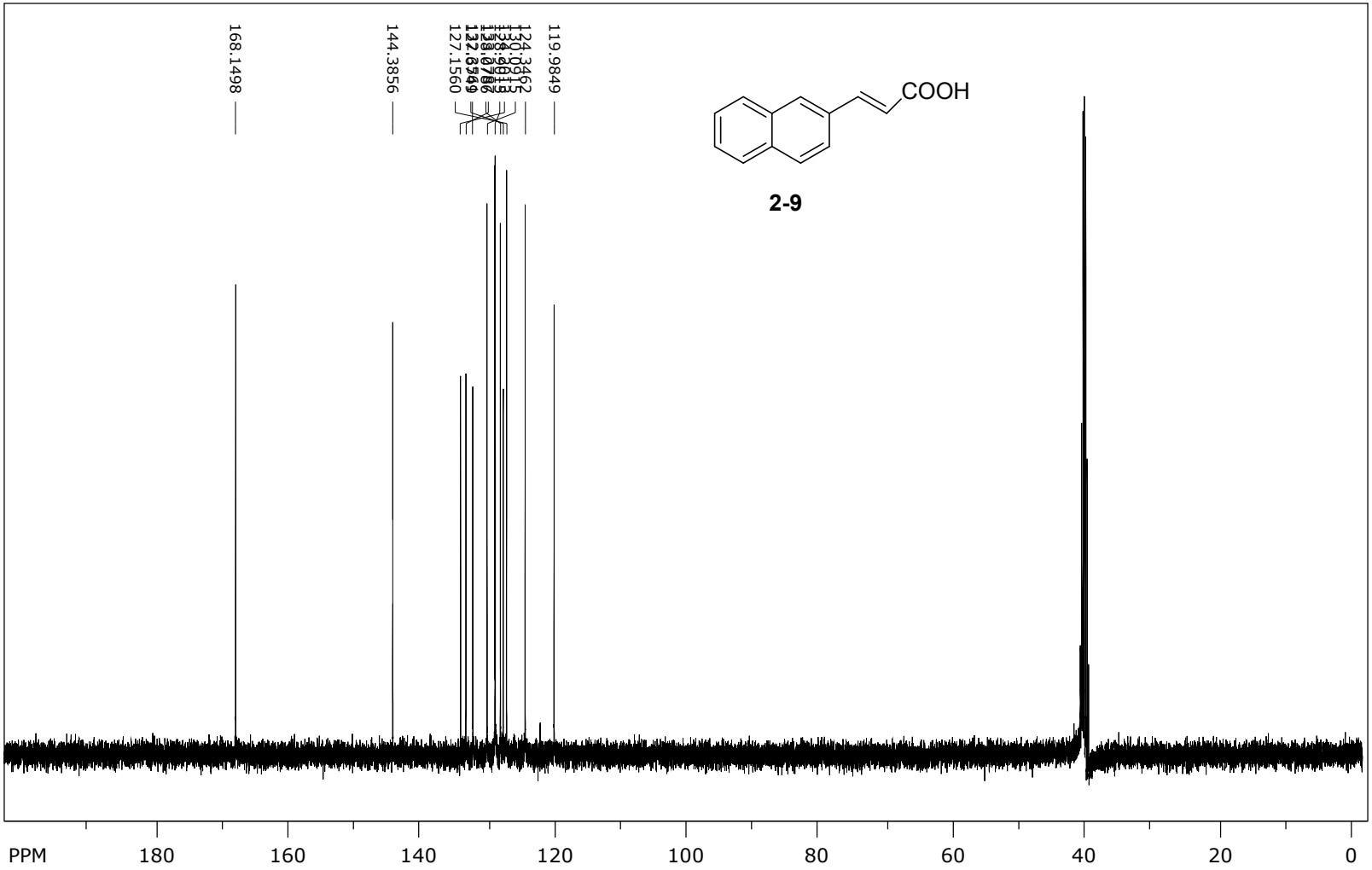


Figure 19. ^{13}C NMR spectrum of compound **2-9** in DMSO-d_6

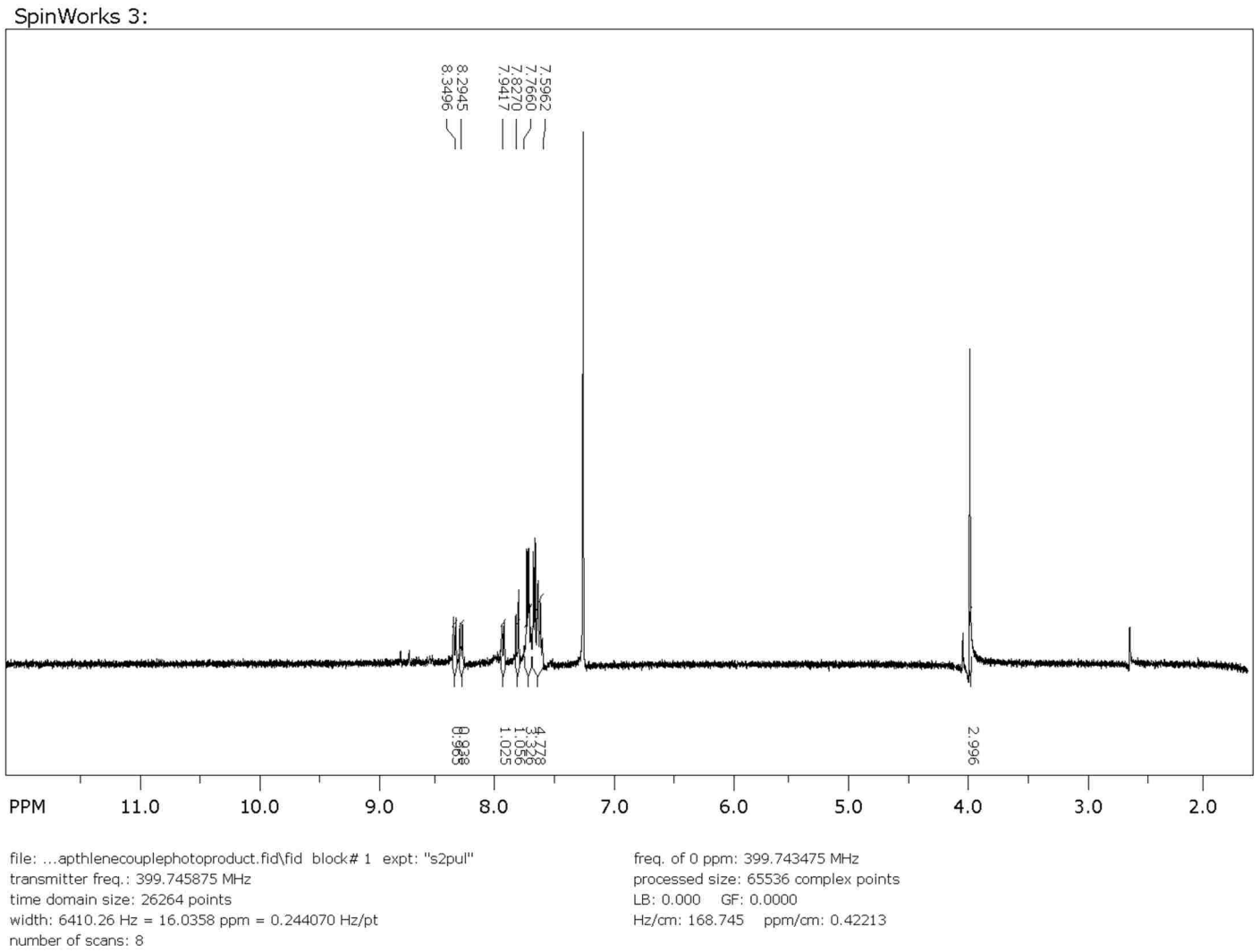


Figure 20. ^{1}H NMR spectrum of compound **2-10 or 2-11** in $CDCl_3$. Solubility is poor in most of the solvent.

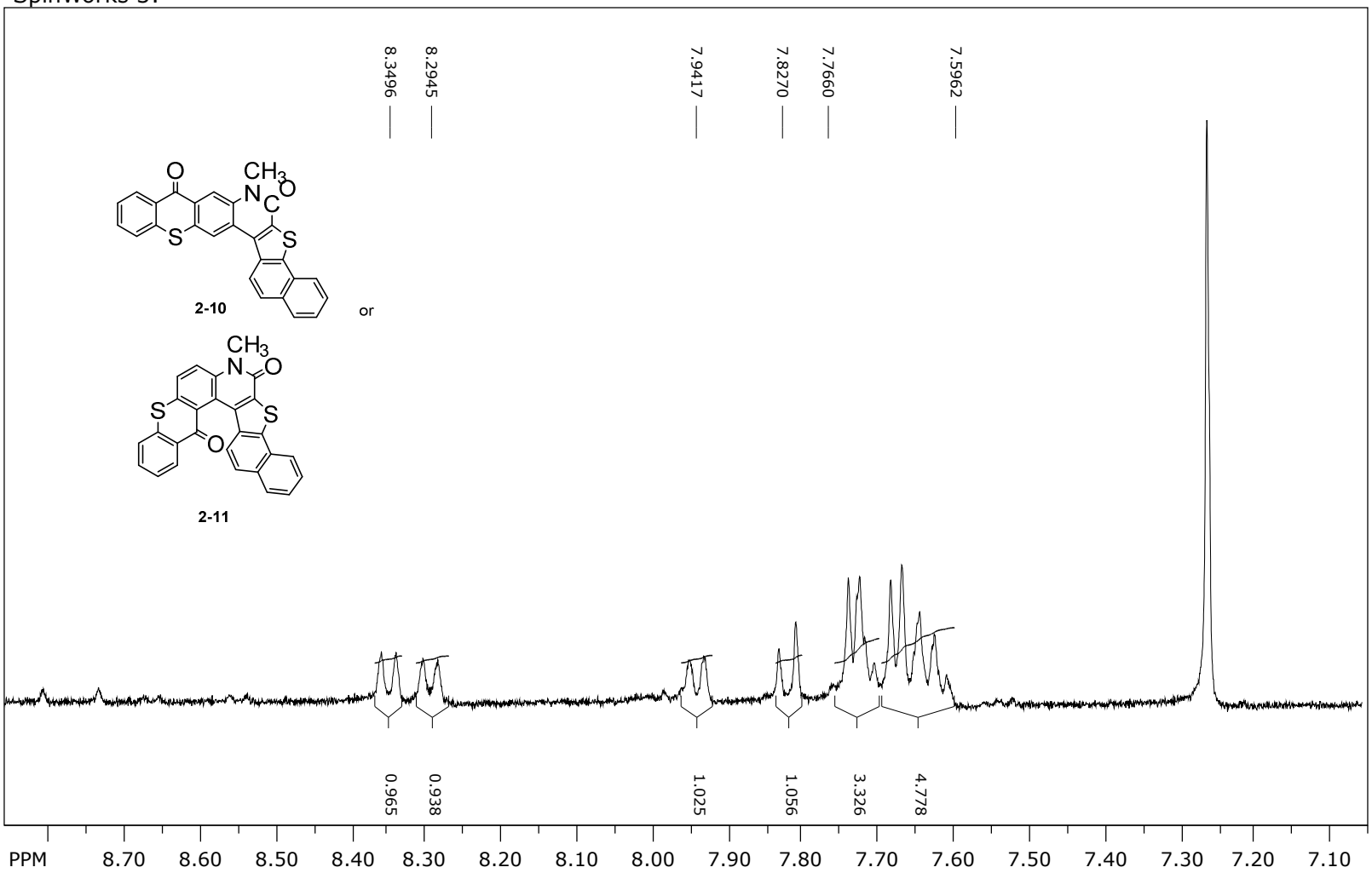
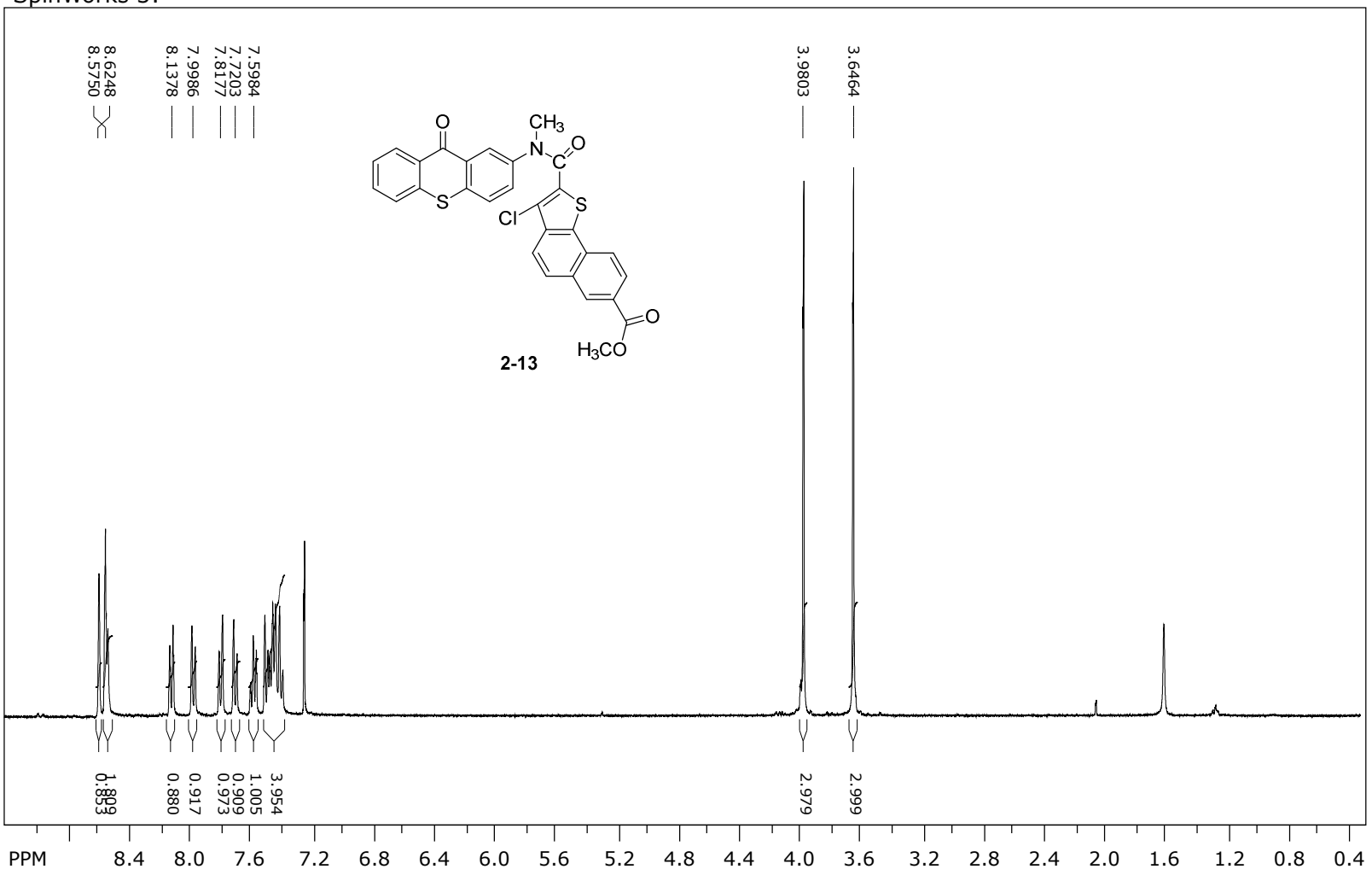


Figure 21. ^{1}H NMR spectrum of compound **2-10 or 2-11** in $CDCl_3$ (Expansion of aromatic region)

SpinWorks 3:



file: methylestercouplepre1HNMR.fid\fid block# 1 ext: "s2.ul"
transmitter freq.: 399.736103 MHz
time domain size: 26264 points
width: 6410.26 Hz = 16.0362 ppm = 0.244070 Hz/pt
number of scans: 8

freq. of 0 ppm: 399.733702 MHz
processed size: 65536 complex points
LB: 0.000 GF: 0.0000
Hz/cm: 142.845 ppm/cm: 0.35735

Figure 22. ^{1}H NMR spectrum of compound **2-13** in $CDCl_3$

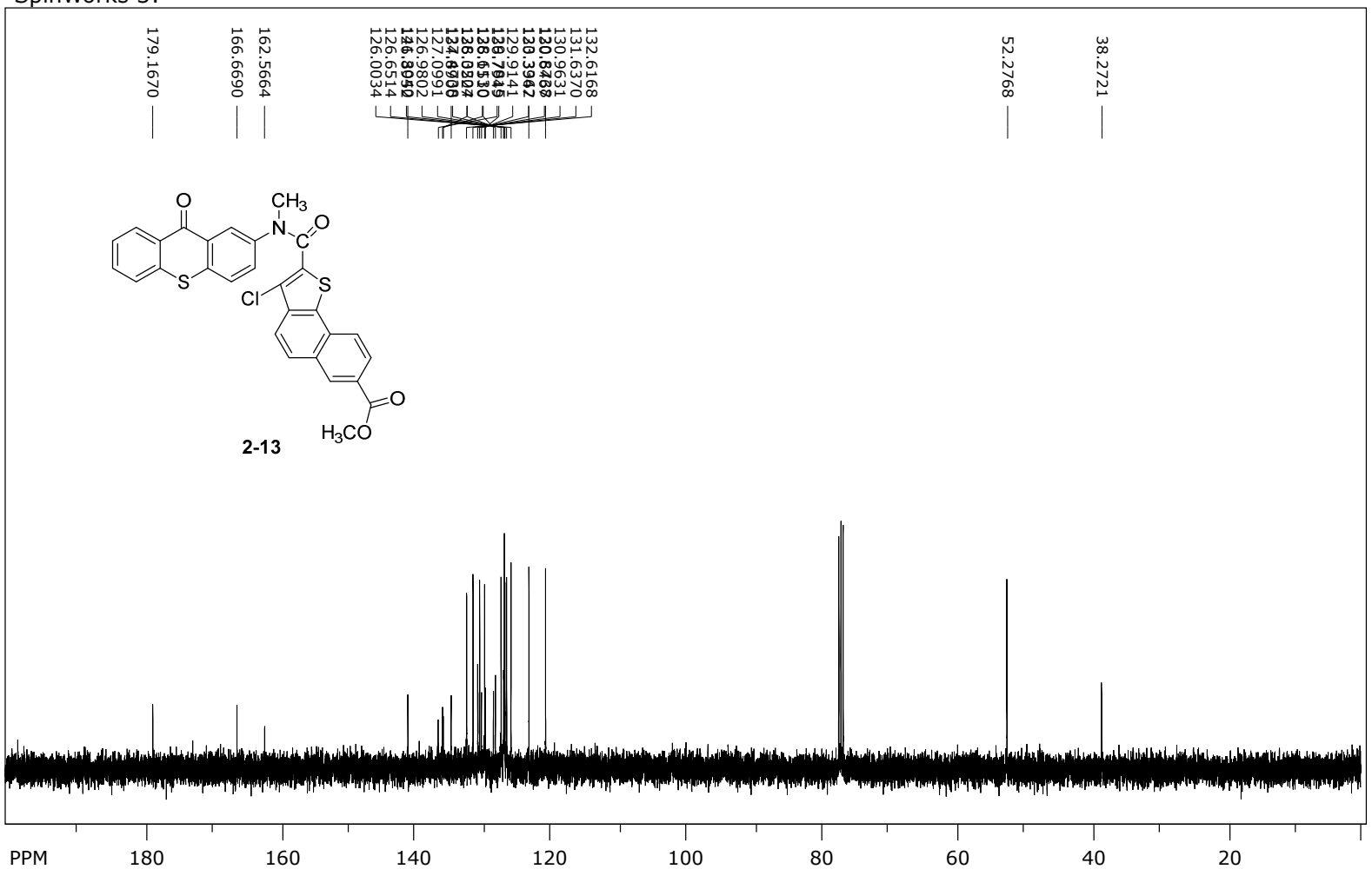


Figure 23. ^{13}C NMR spectrum of compound **2-13** in $CDCl_3$

SpinWorks 3:

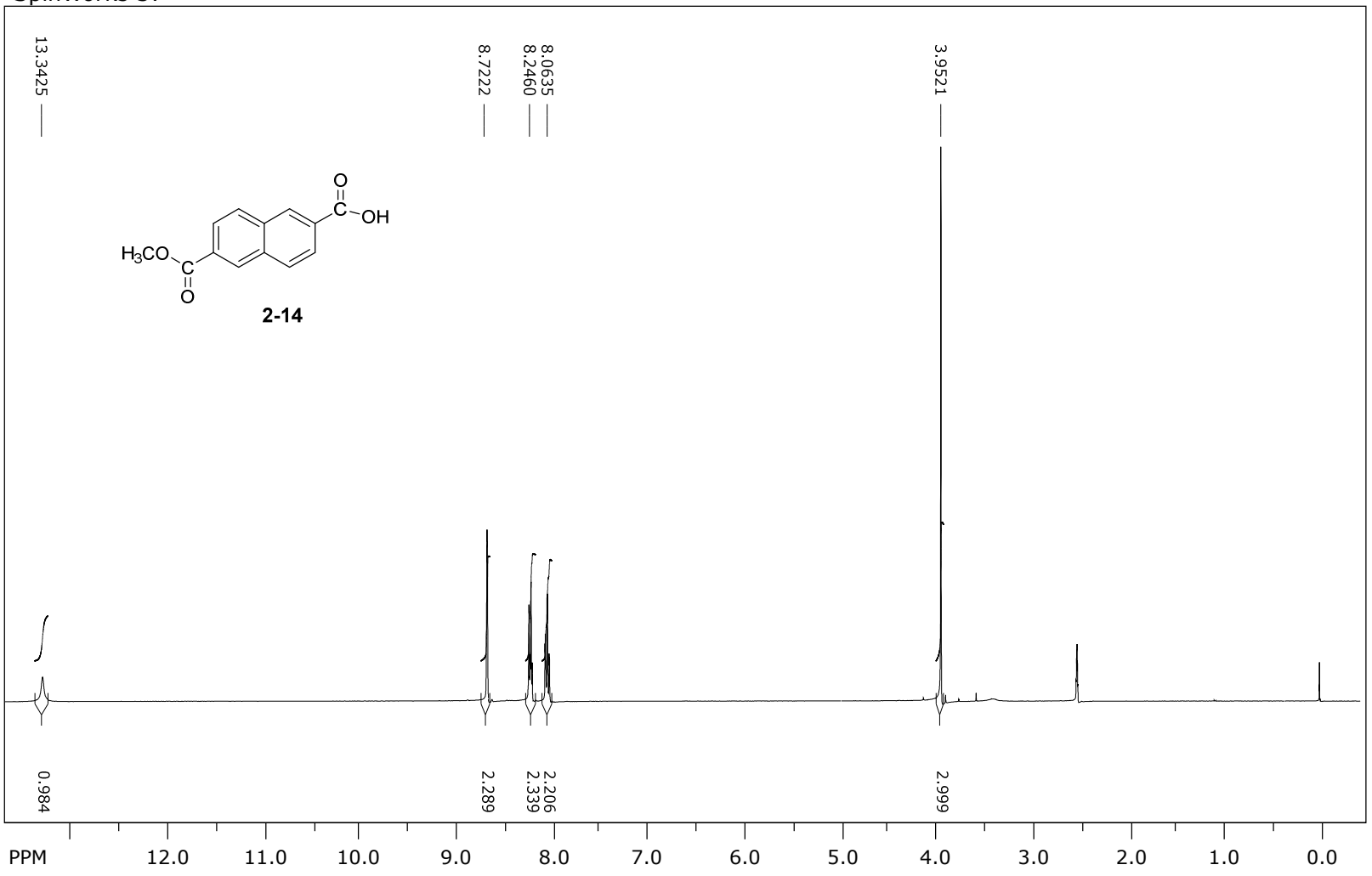


Figure 24. ^{1}H NMR spectrum of compound **2-14** in DMSO-d$_6$

...aphthalenemethylesterstep11.fid\fid" block# 1 expt: "s2pul
transmitter freq.: 399.747774 MHz
time domain size: 26264 points
width: 6410.26 Hz = 16.0358 ppm = 0.244070 Hz/pt
number of scans: 8

freq. of 0 ppm: 399.745357 MHz
processed size: 65536 complex points
LB: 0.000 GF: 0.0000
Hz/cm: 226.365 ppm/cm: 0.56627

SpinWorks 3:

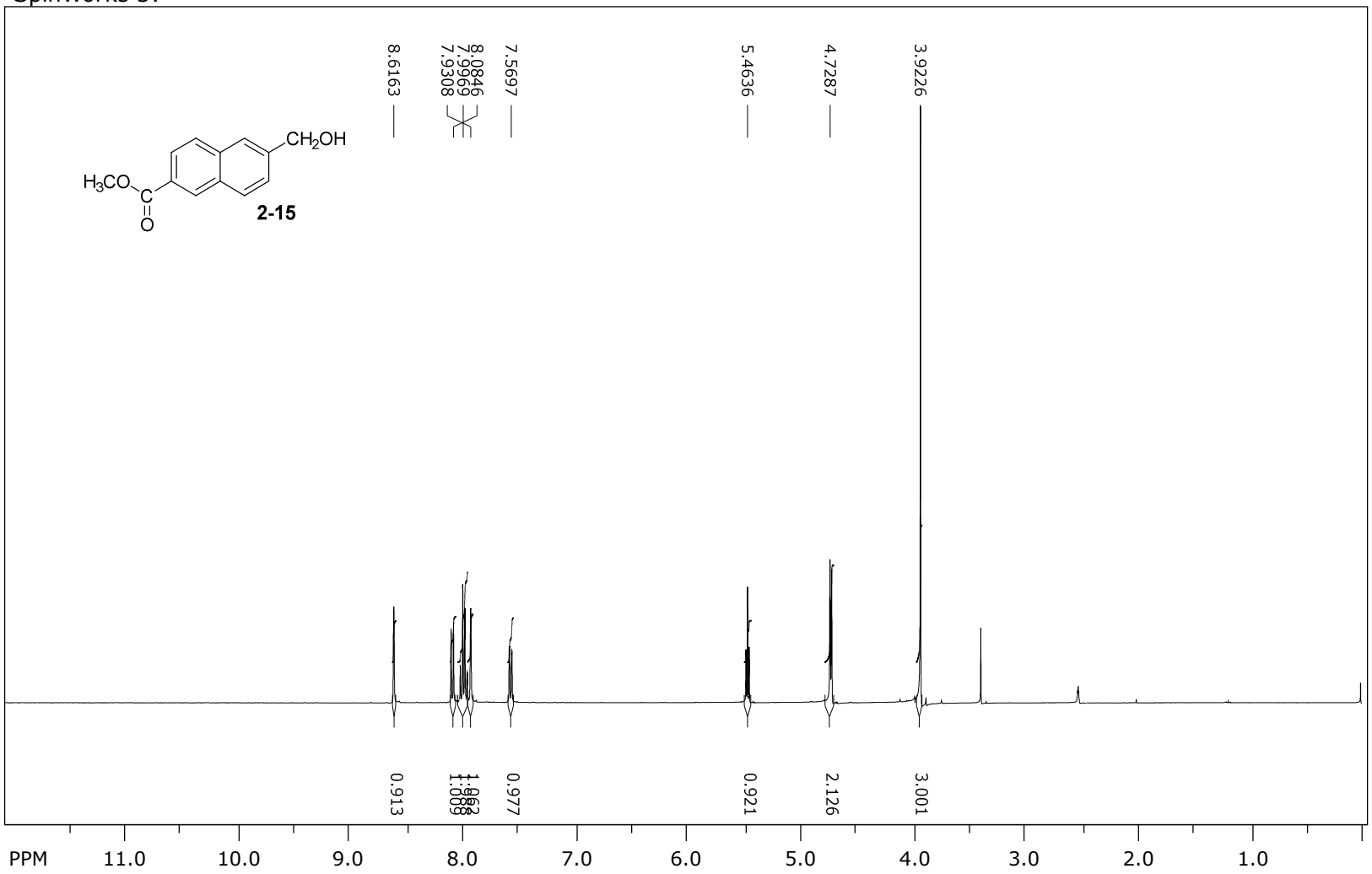


Figure 25. ^{1}H NMR spectrum of compound **2-15** in DMSO-d6

file:S:..ethylesterstep2columnpure2.fid\fid block# 1 expt: s2pul
transmitter freq.: 399.738002 MHz
time domain size: 26264 points
width: 6410.26 Hz = 16.0361 ppm = 0.244070 Hz/pt
number of scans: 8

freq. of 0 ppm: 399.735590 MHz
processed size: 65536 complex points
LB: 0.000 GF: 0.0000
Hz/cm: 193.440 ppm/cm: 0.48392

SpinWorks 3:

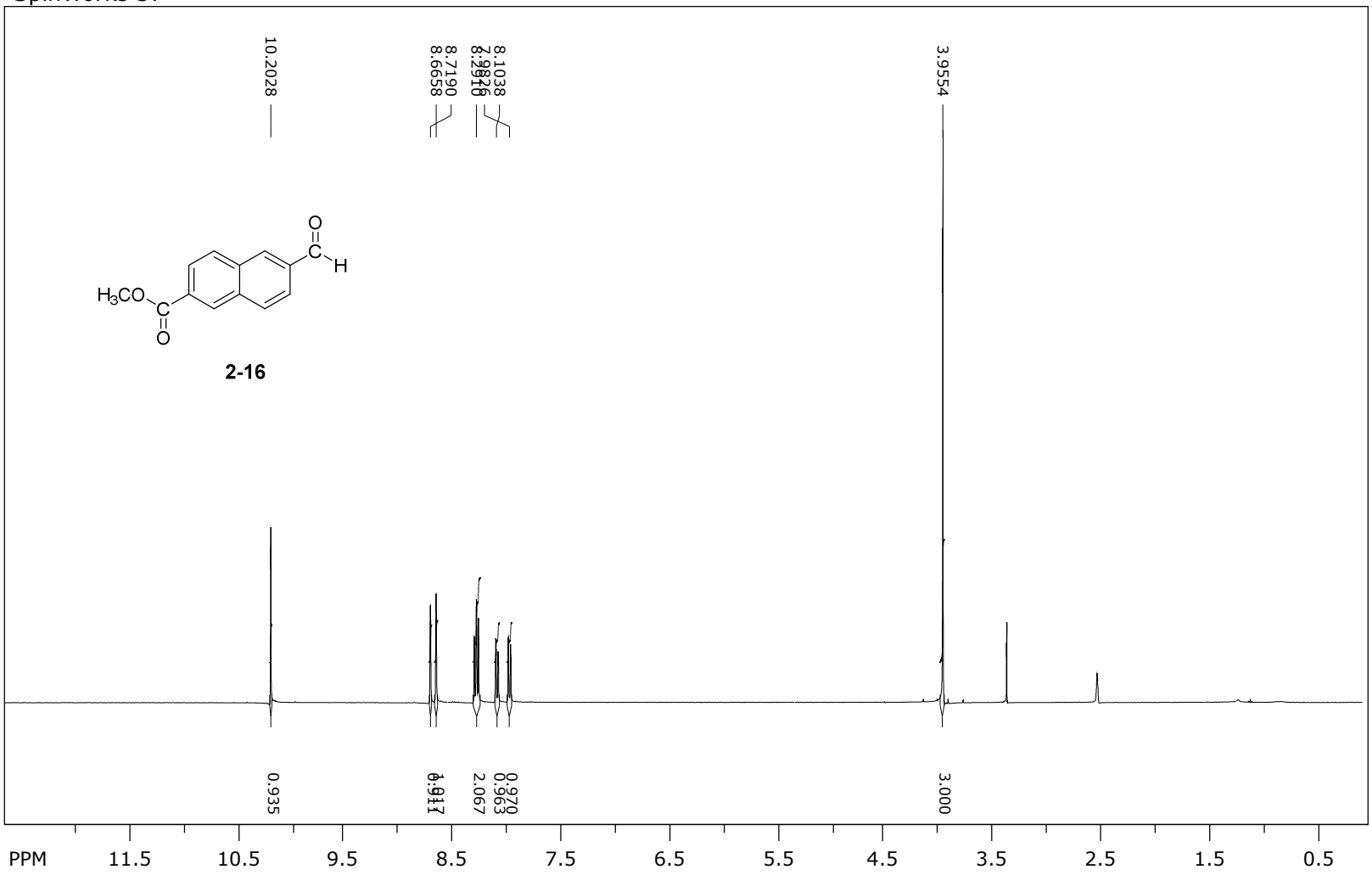
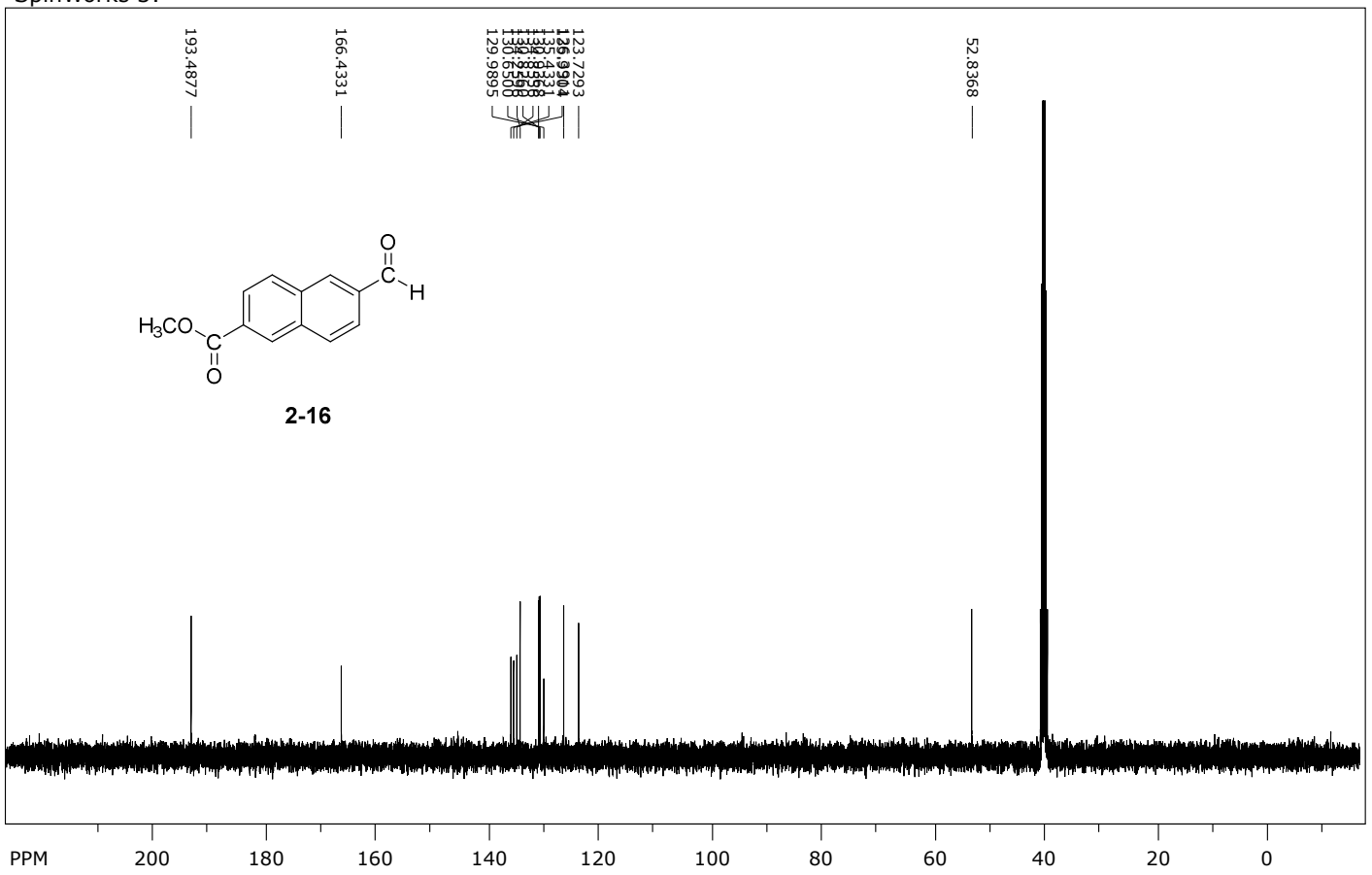


Figure 26. [1]HNMR spectrum of compound **2-16** in DMSO-d6

SpinWorks 3:



file: ...enemethylesterstep3pureC13.fid\fid block# 1 expt: "s2pul"
transmitter freq.: 100.524151 MHz
time domain size: 63750 points
width: 24509.80 Hz = 243.8201 ppm = 0.384468 Hz/pt
number of scans: 256

freq. of 0 ppm: 100.513598 MHz
processed size: 65536 complex points
LB: 0.000 GF: 0.0000
Hz/cm: 980.392 ppm/cm: 9.75280

Figure 27. ^{13}C NMR spectrum of compound **2-16** in DMSO-d_6

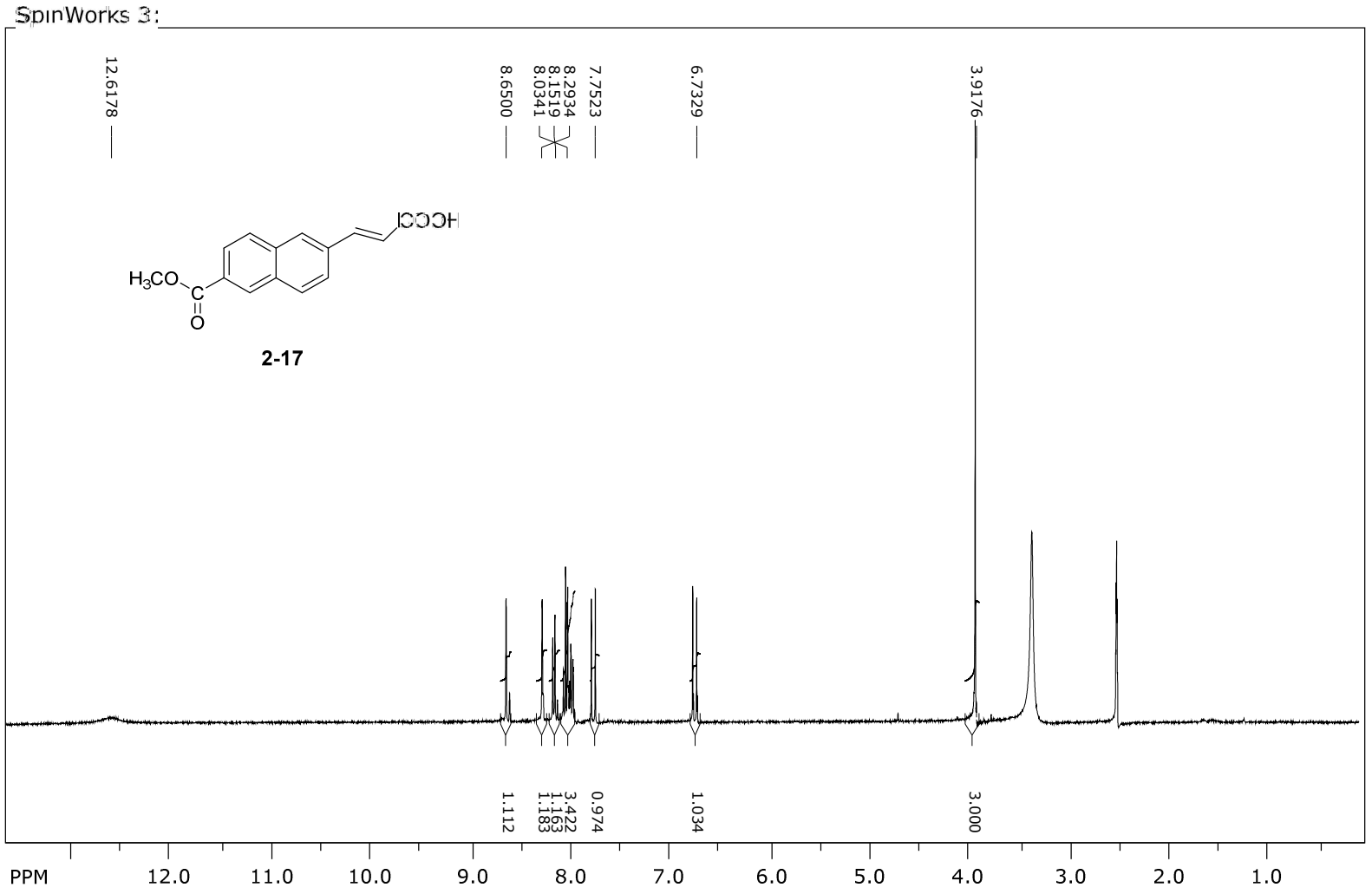


Figure 28. ^{1}H NMR spectrum of compound **2-17** in DMSO-d_6

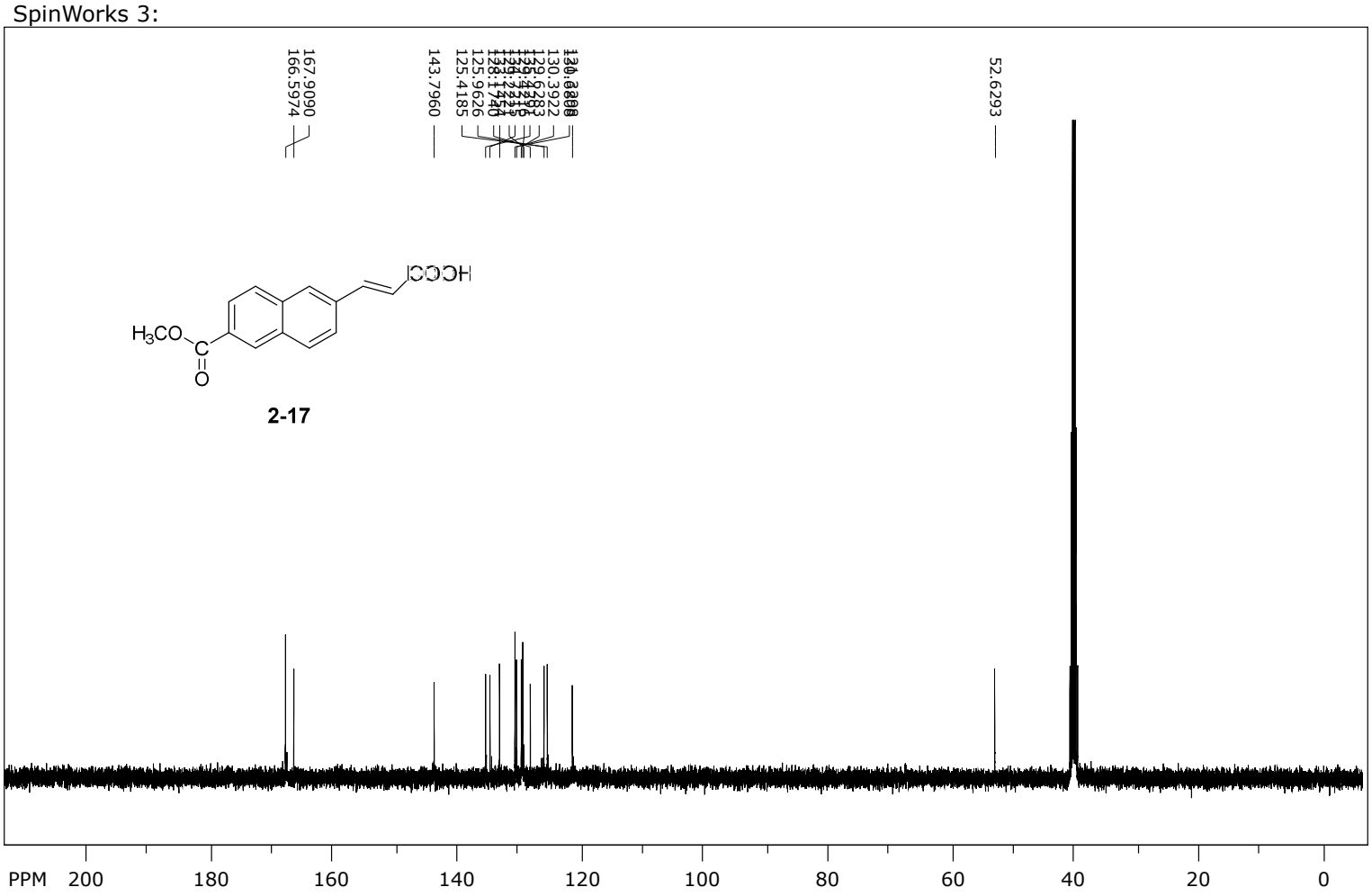


Figure 29. ^{13}C NMR spectrum of compound **2-17** in DMSO-d_6

SpinWorks 3:

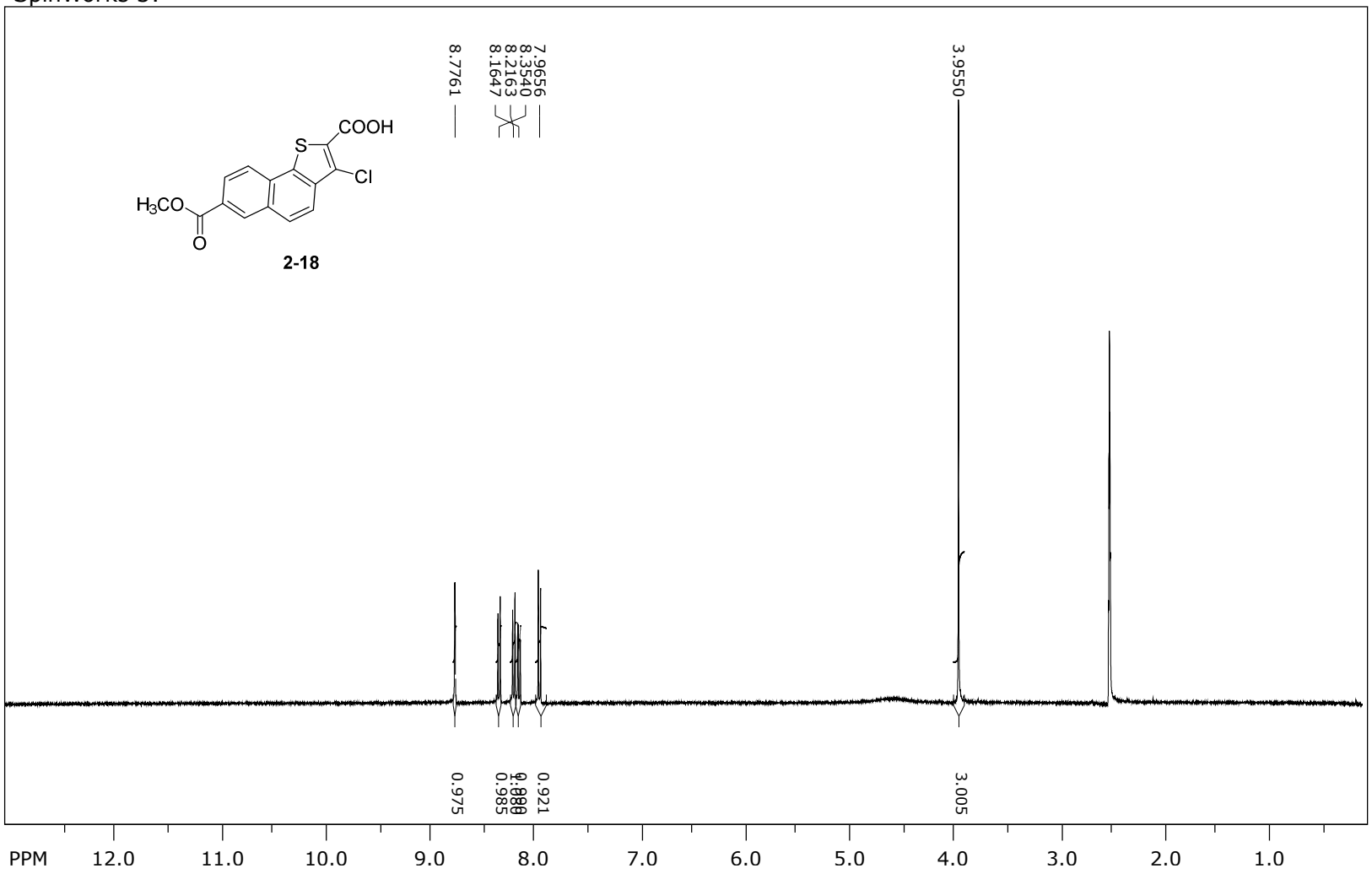


Figure 30. ^{1}H NMR spectrum of compound **2-18** in DMSO-d_6

file: ...methylestercaboxilicacidbc.fid\fid block# 1 expt: "s2pul"
transmitter freq.: 399.738002 MHz
time domain size: 26264 points
width: 6410.26 Hz = 16.0361 ppm = 0.244070 Hz/pt
number of scans: 8

freq. of 0 ppm: 399.735595 MHz
processed size: 65536 complex points
LB: 0.000 GF: 0.0000
Hz/cm: 207.845 ppm/cm: 0.51995

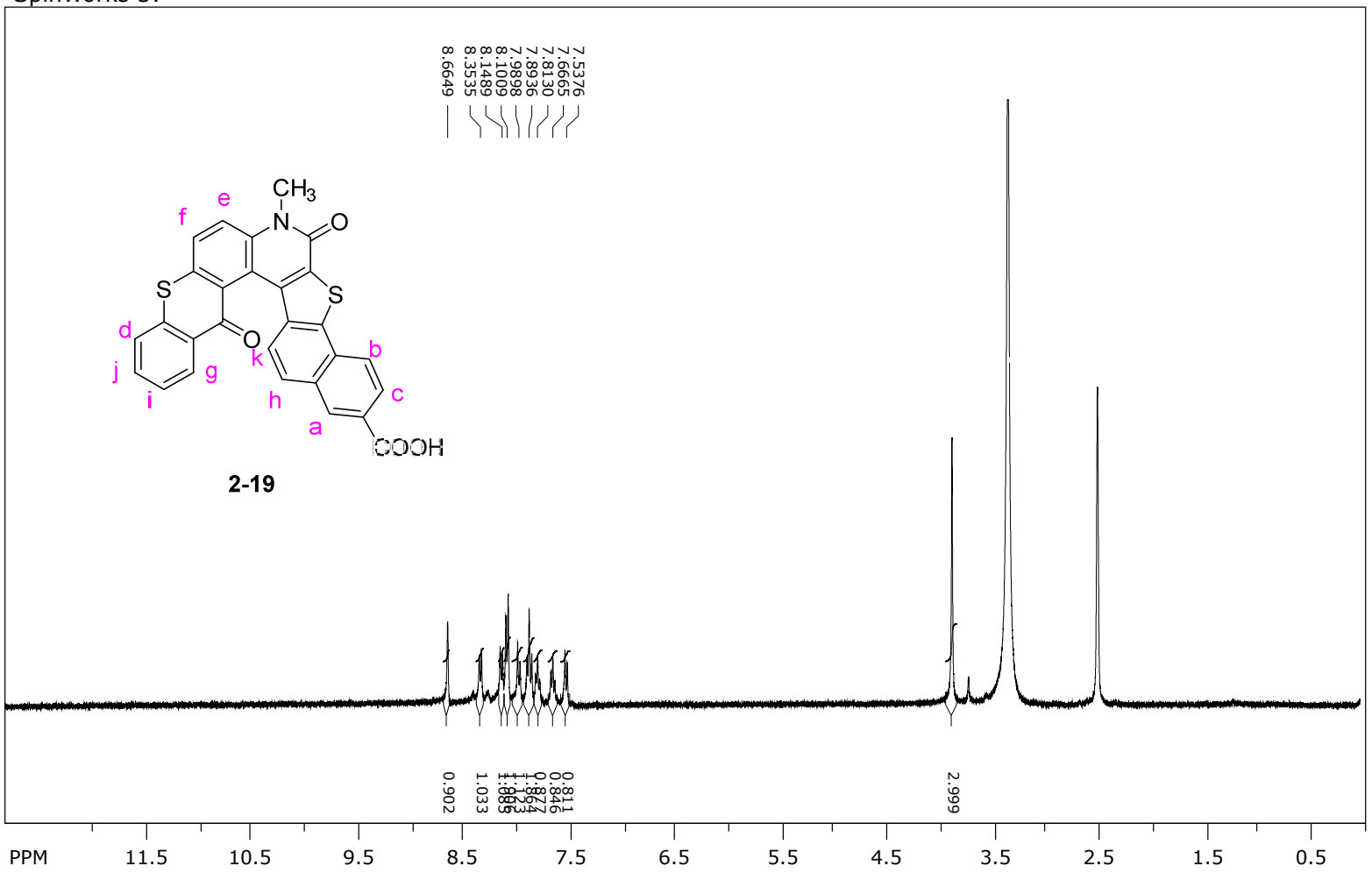


Figure 31. ^{1}H NMR spectrum of compound **2-19** in DMSO-d_6

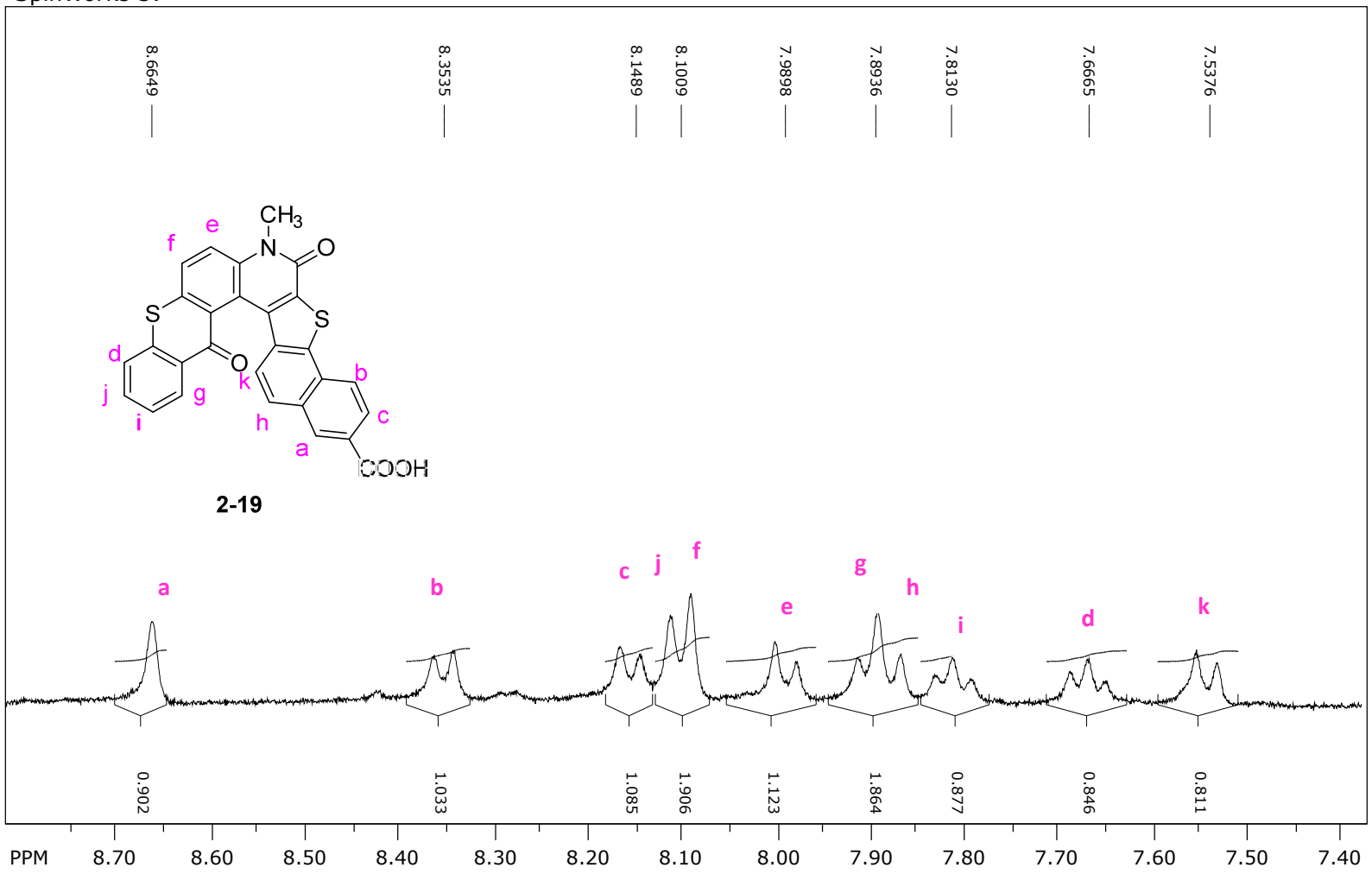


Figure 32. Expansion of aromatic region of ^{1}H NMR spectrum of **2-19** in DMSO-d_6

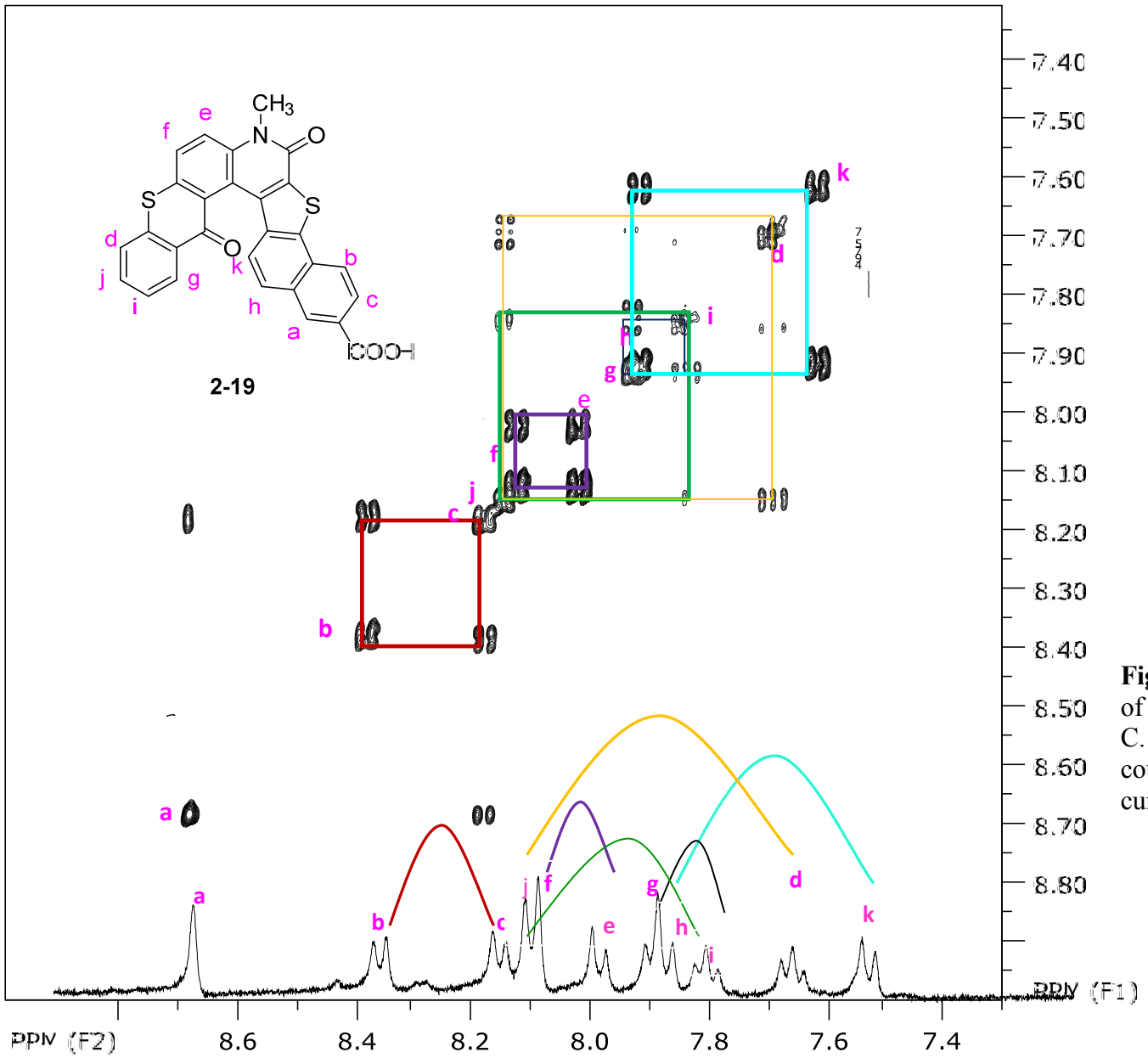


Figure 33. ^{1}H NMR COSY spectrum of compound **2-19** in DMSO-d_6 at 60° C. Protons which shows vicinal coupling are connected by each curved line in 1D ^{1}H NMR.

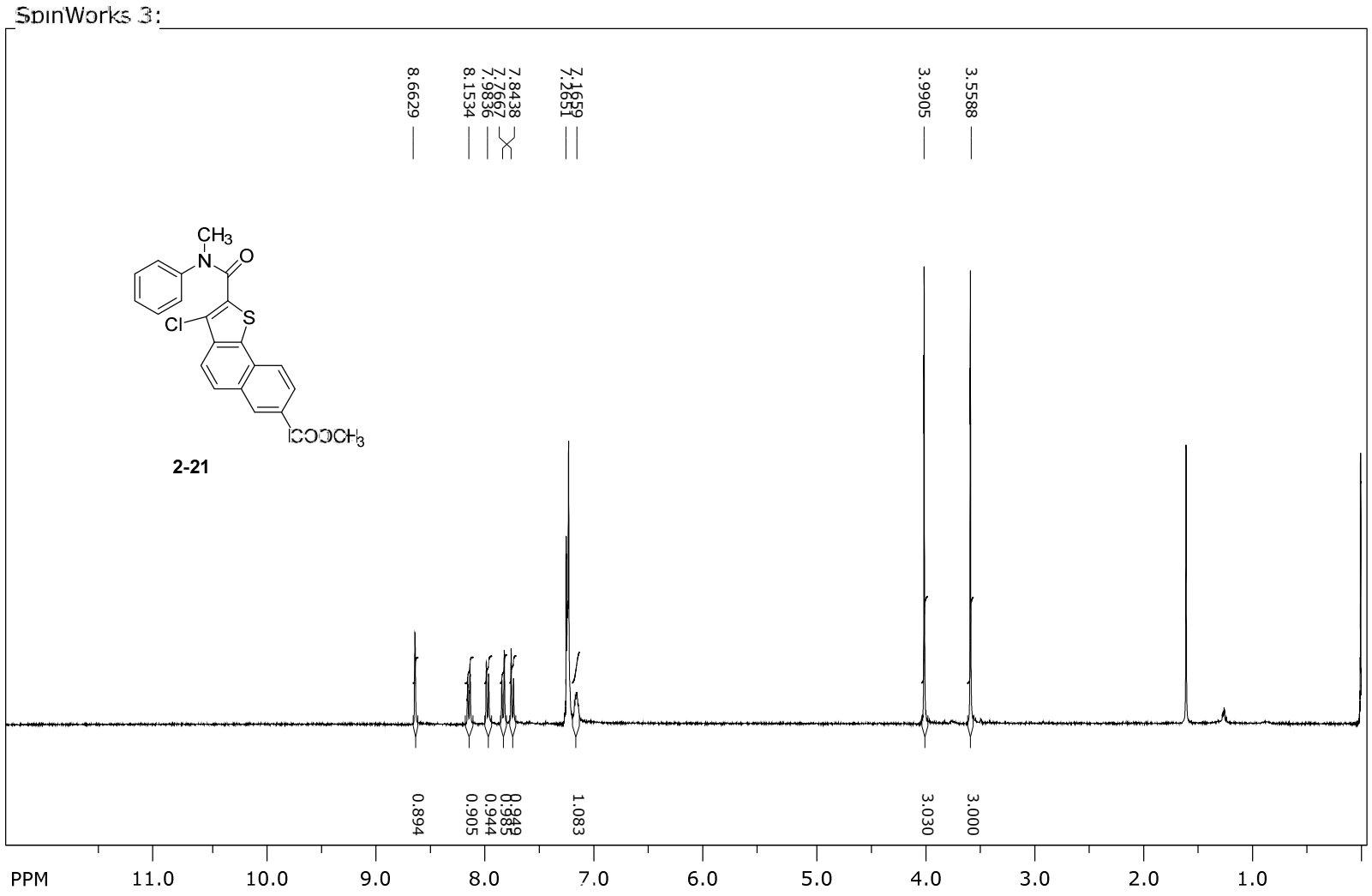


Figure 34. ^{1}H NMR spectrum of compound **2-21** in $CDCl_3$

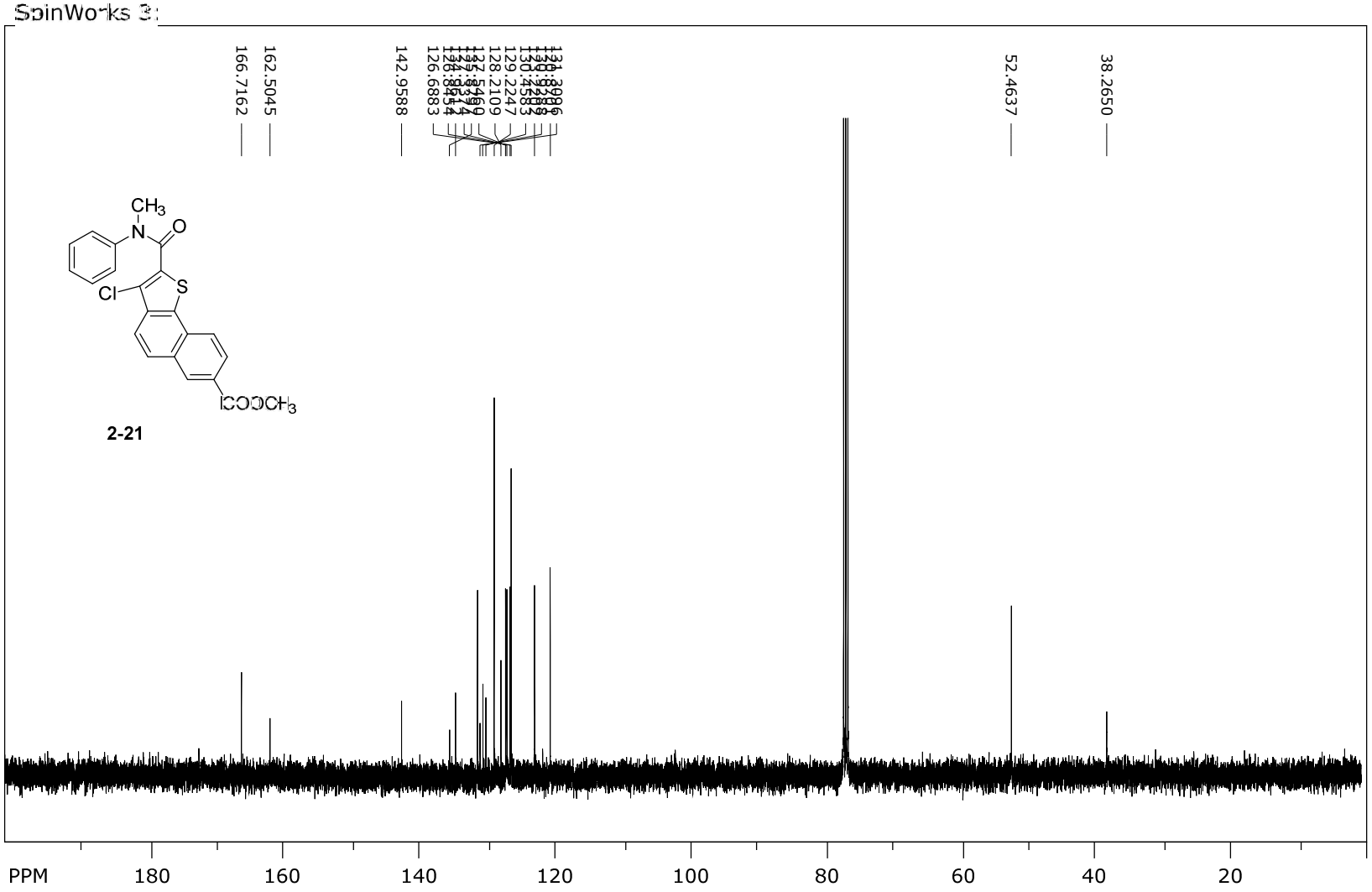


Figure 35. ^{13}C NMR spectrum of compound **2-21** in $CDCl_3$

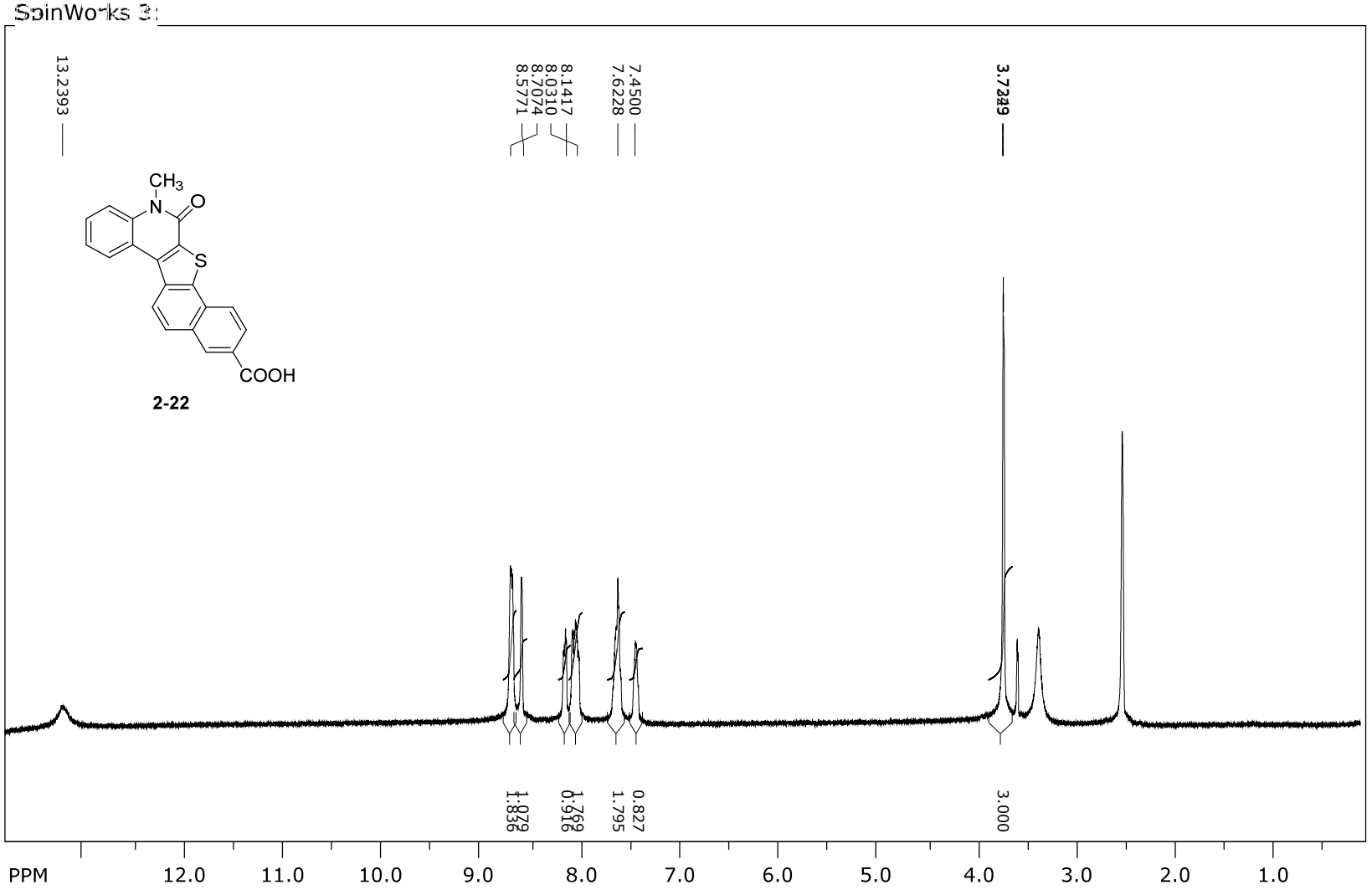


Figure 36. ^{1}H NMR spectrum of compound **2-22** in DMSO-d_6

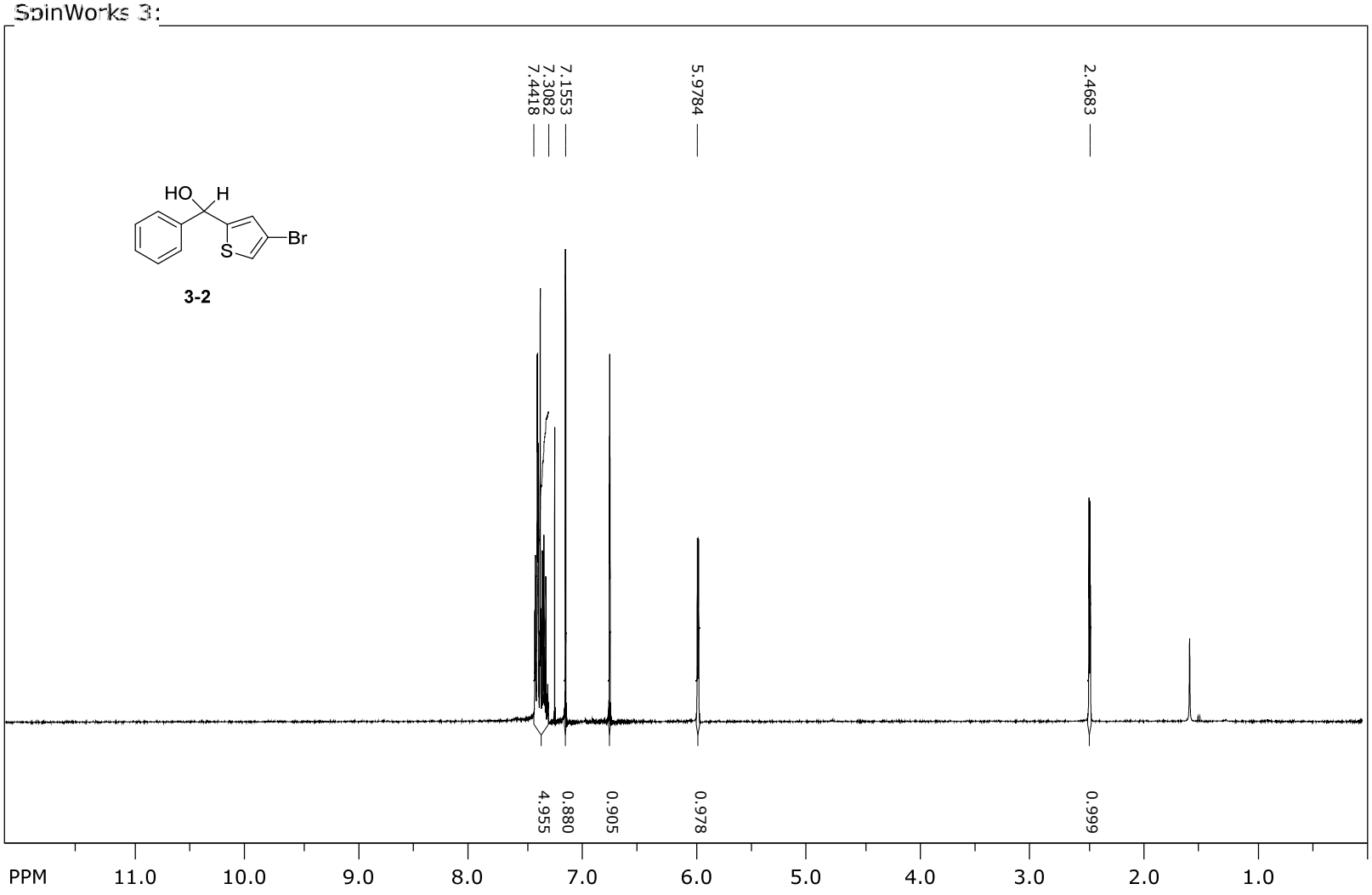


Figure 37. ^{1}H NMR spectrum of compound **3-2** in $CDCl_3$

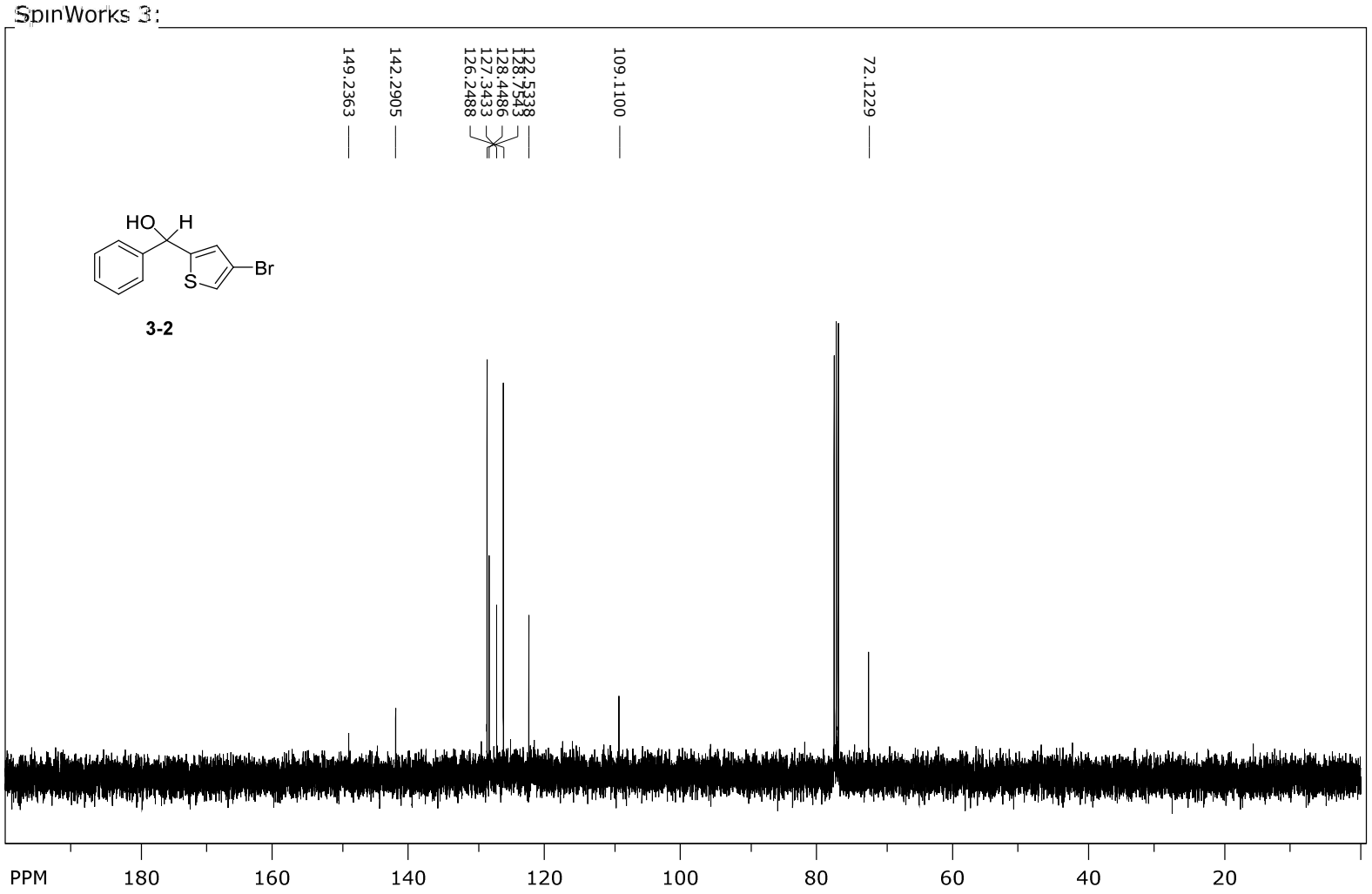


Figure 38. ^{13}C NMR spectrum of compound **3-2** in $CDCl_3$

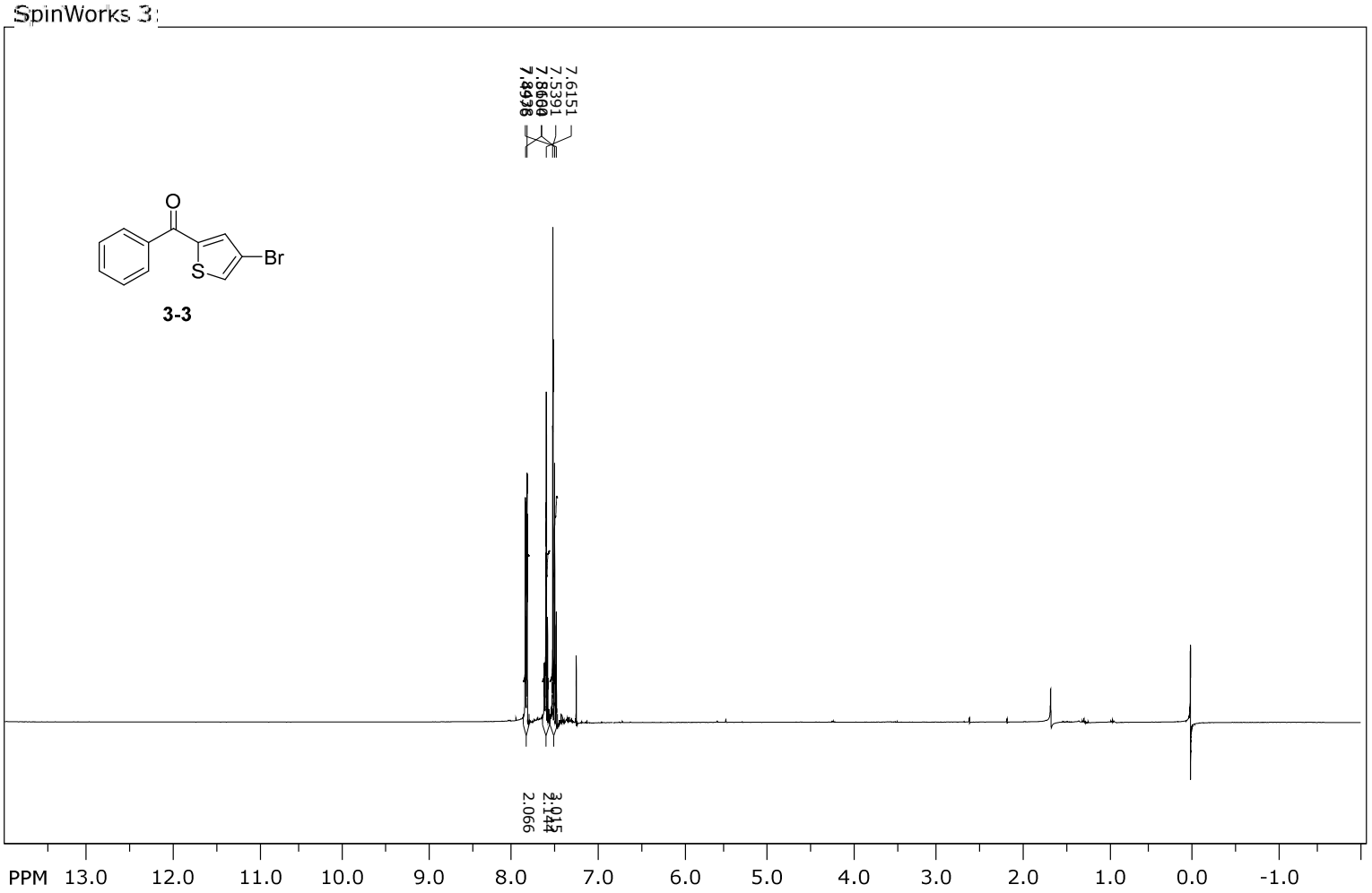


Figure 39. ^{1}H NMR spectrum of compound **3-3** in $CDCl_3$

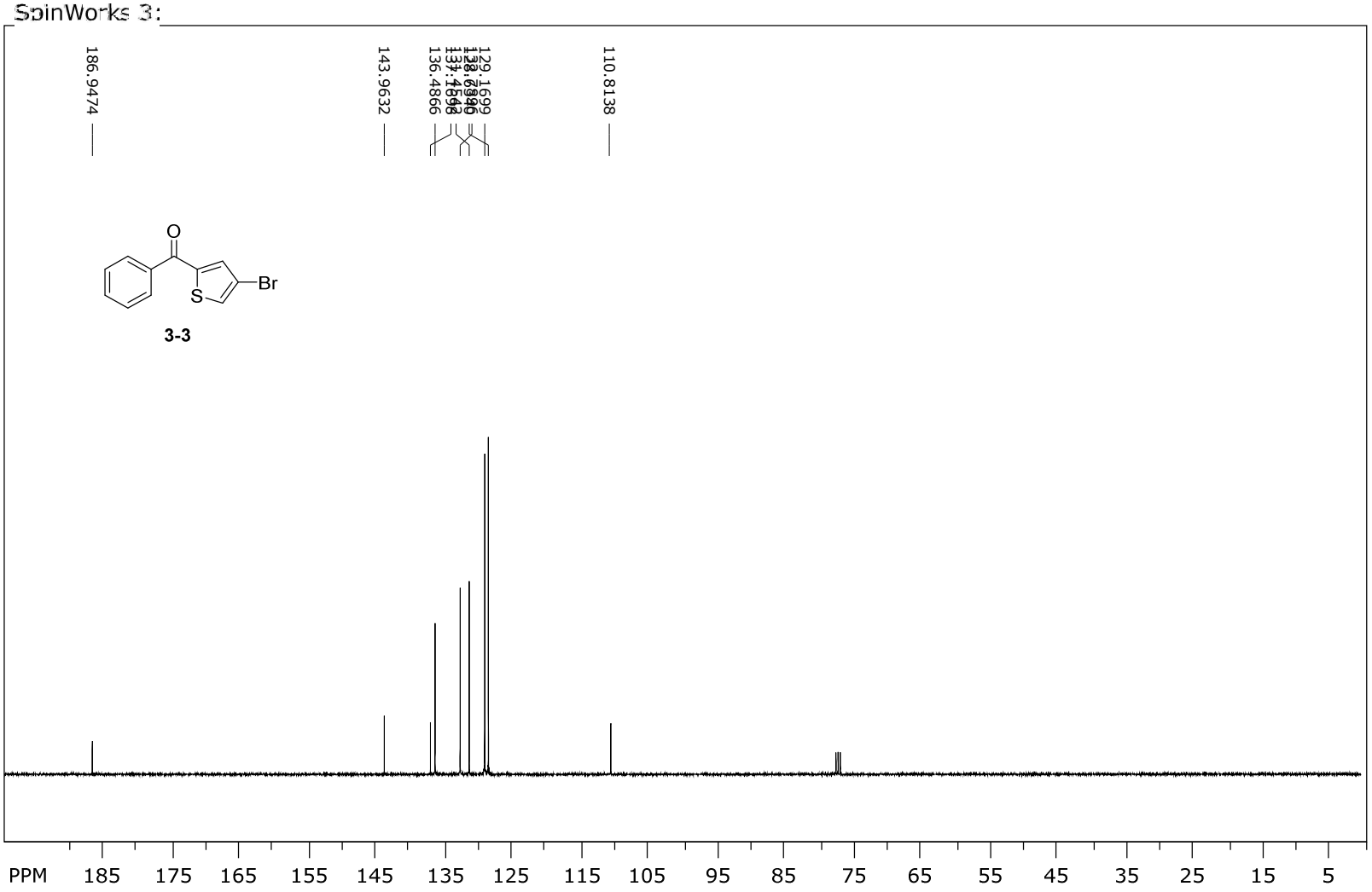


Figure 40. ^{13}C NMR spectrum of compound **3-3** in $CDCl_3$

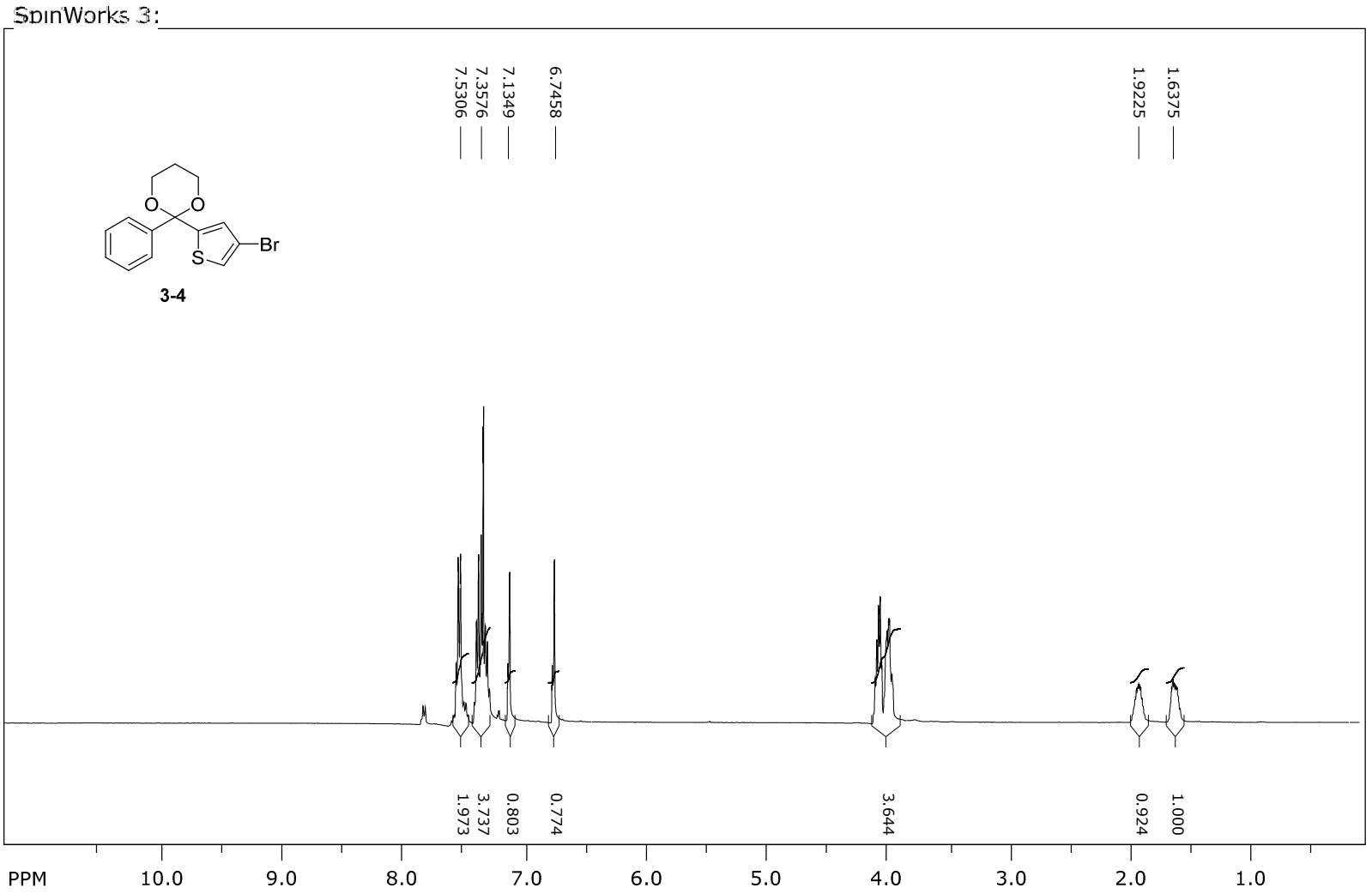


Figure 41. ^{1}H NMR spectrum of compound **3-4** in $CDCl_3$

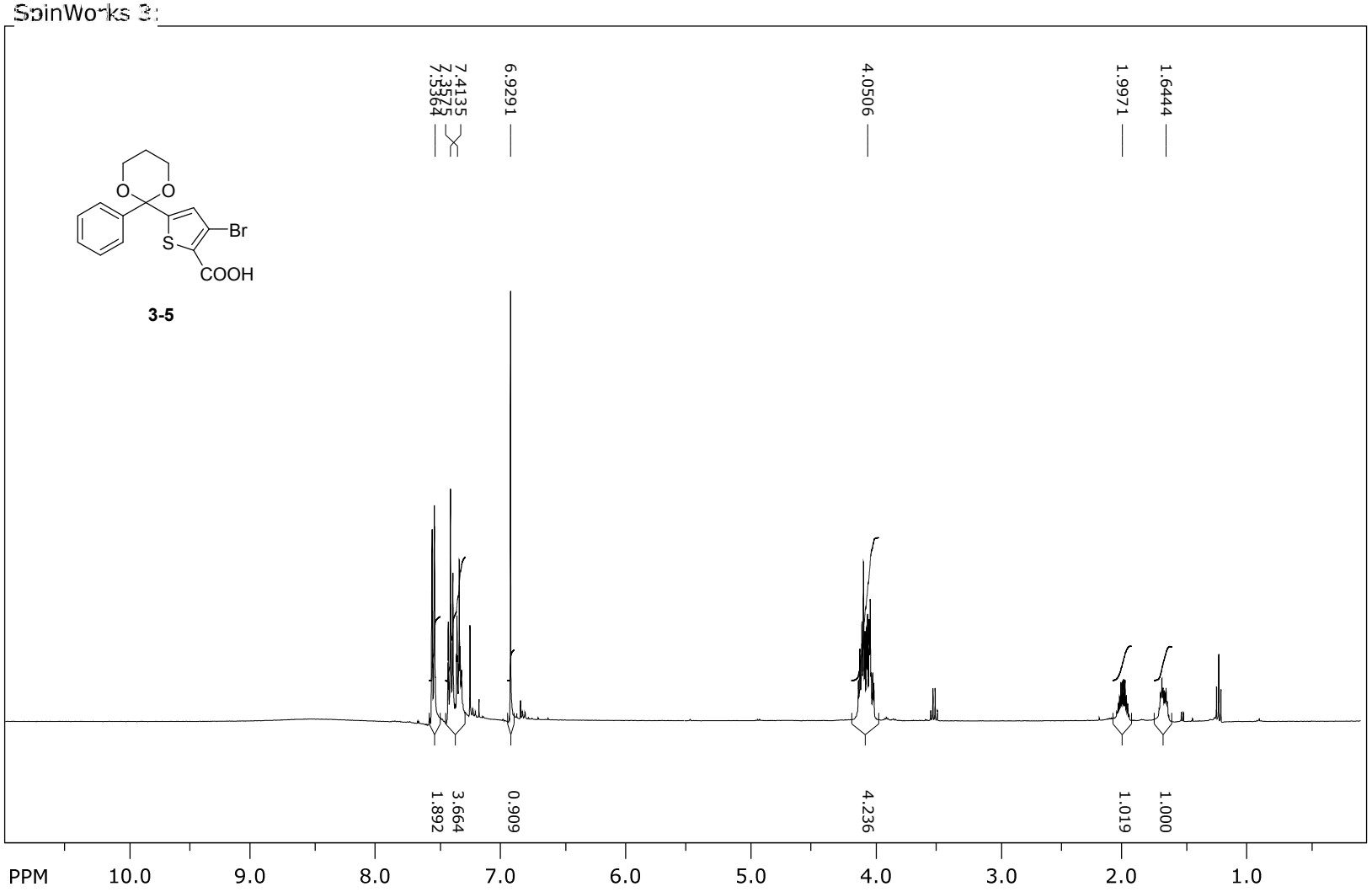


Figure 42. ^{1}H NMR spectrum of compound **3-5** in $CDCl_3$

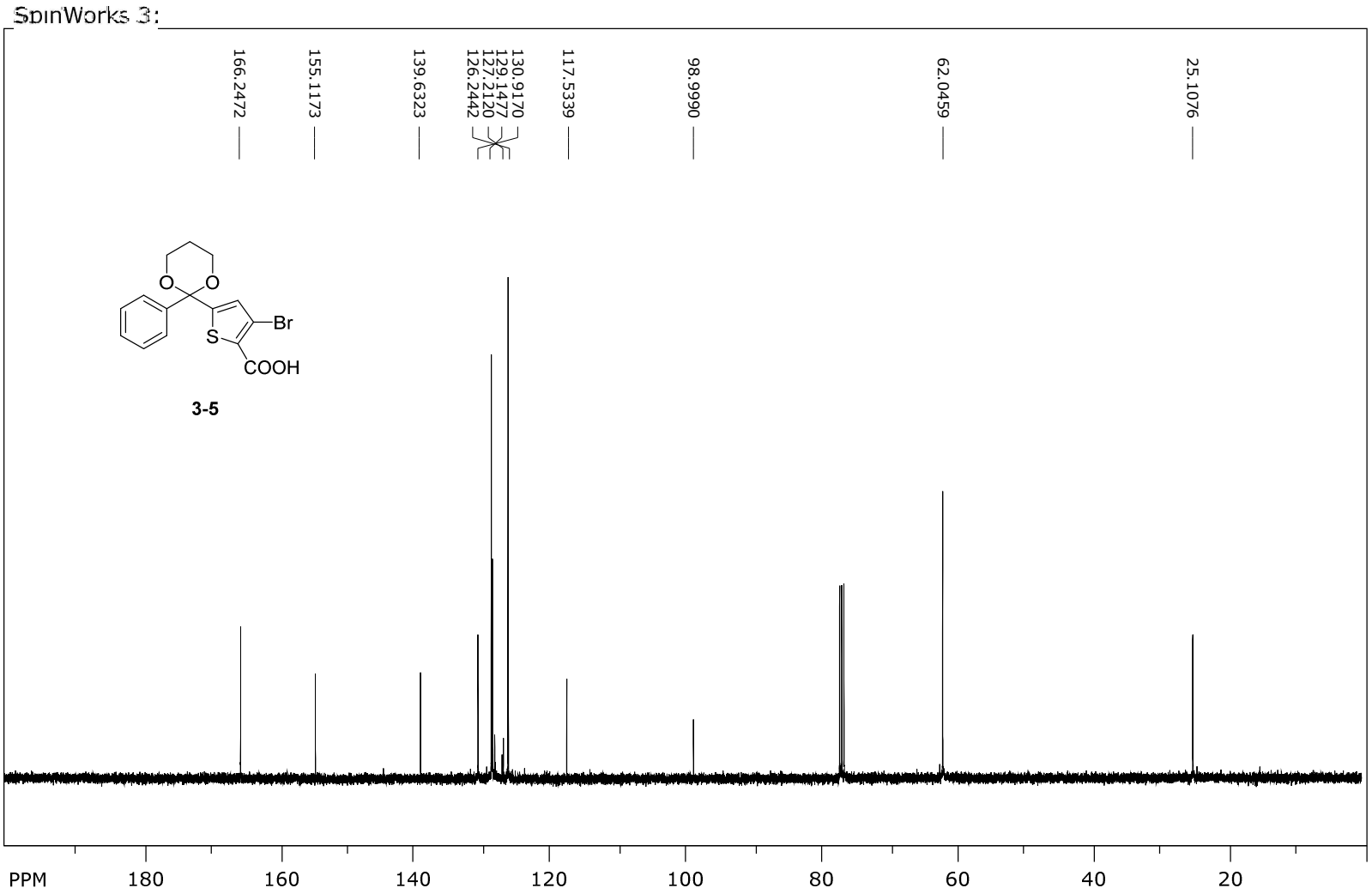


Figure 43. ^{13}C NMR spectrum of compound **3-5** in $CDCl_3$

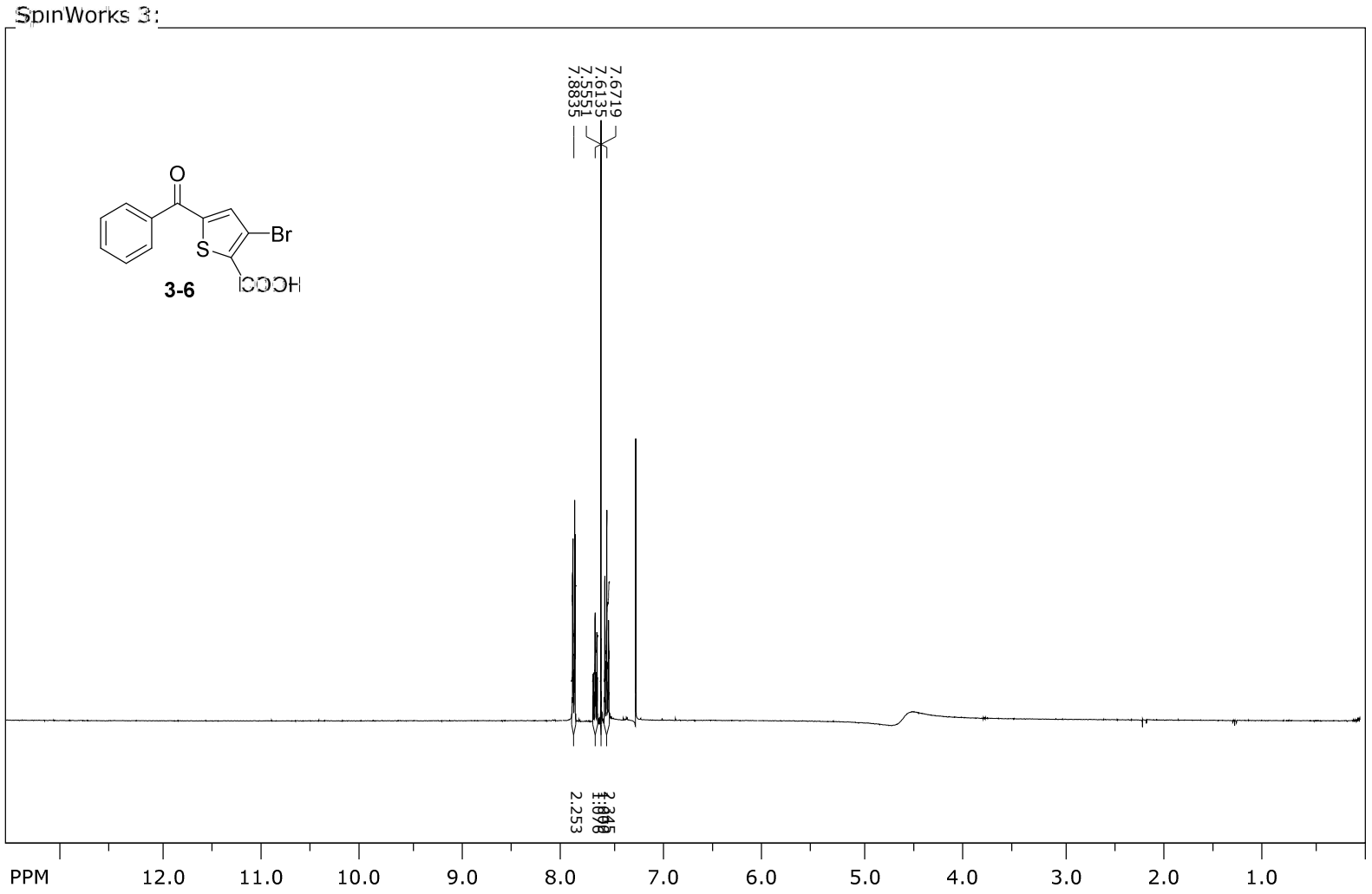


Figure 44. ^{1}H NMR spectrum of compound **3-6** in $CDCl_3$

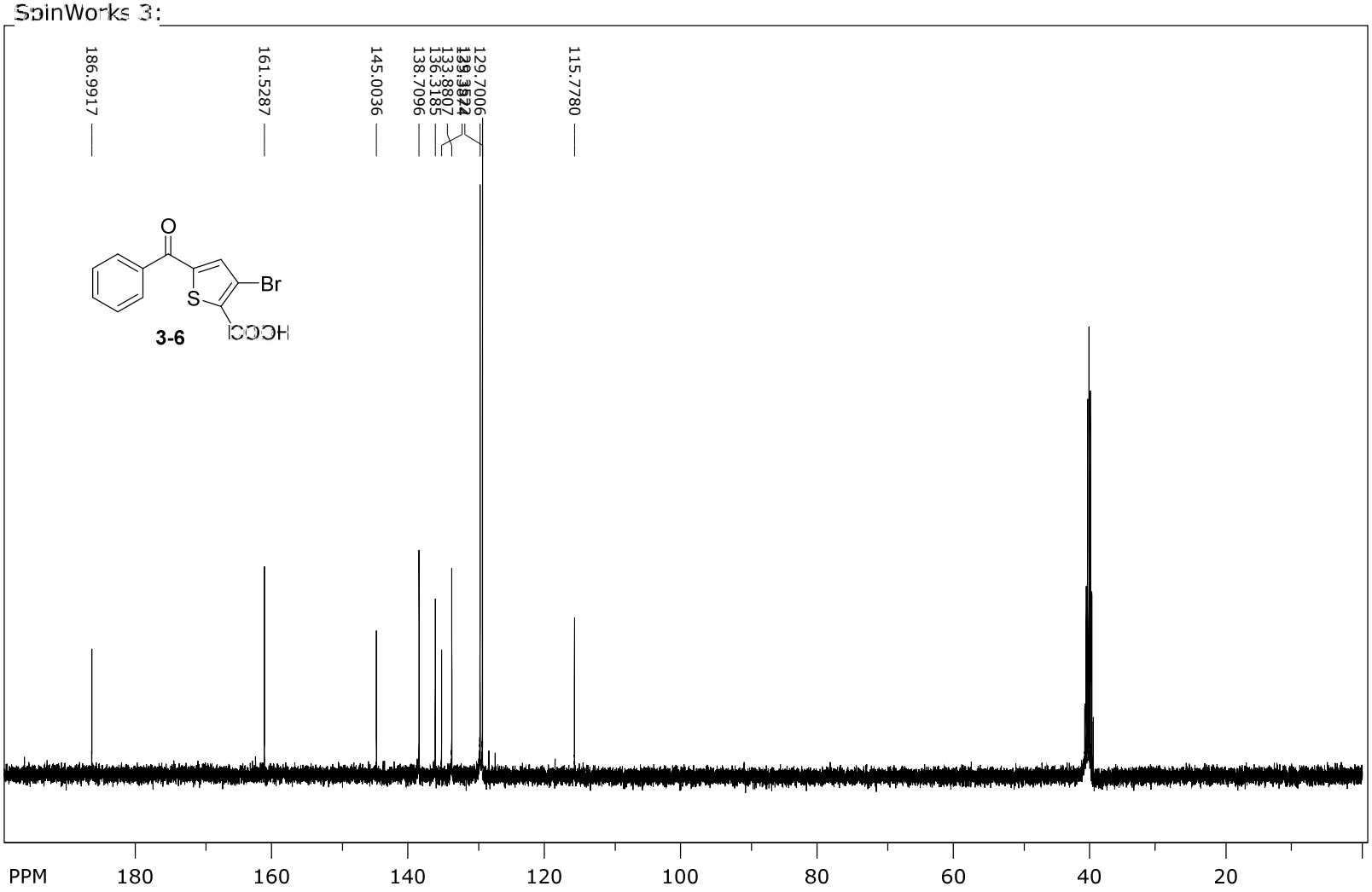


Figure 45. ^{13}C NMR spectrum of compound **3-6** in DMSO-d_6

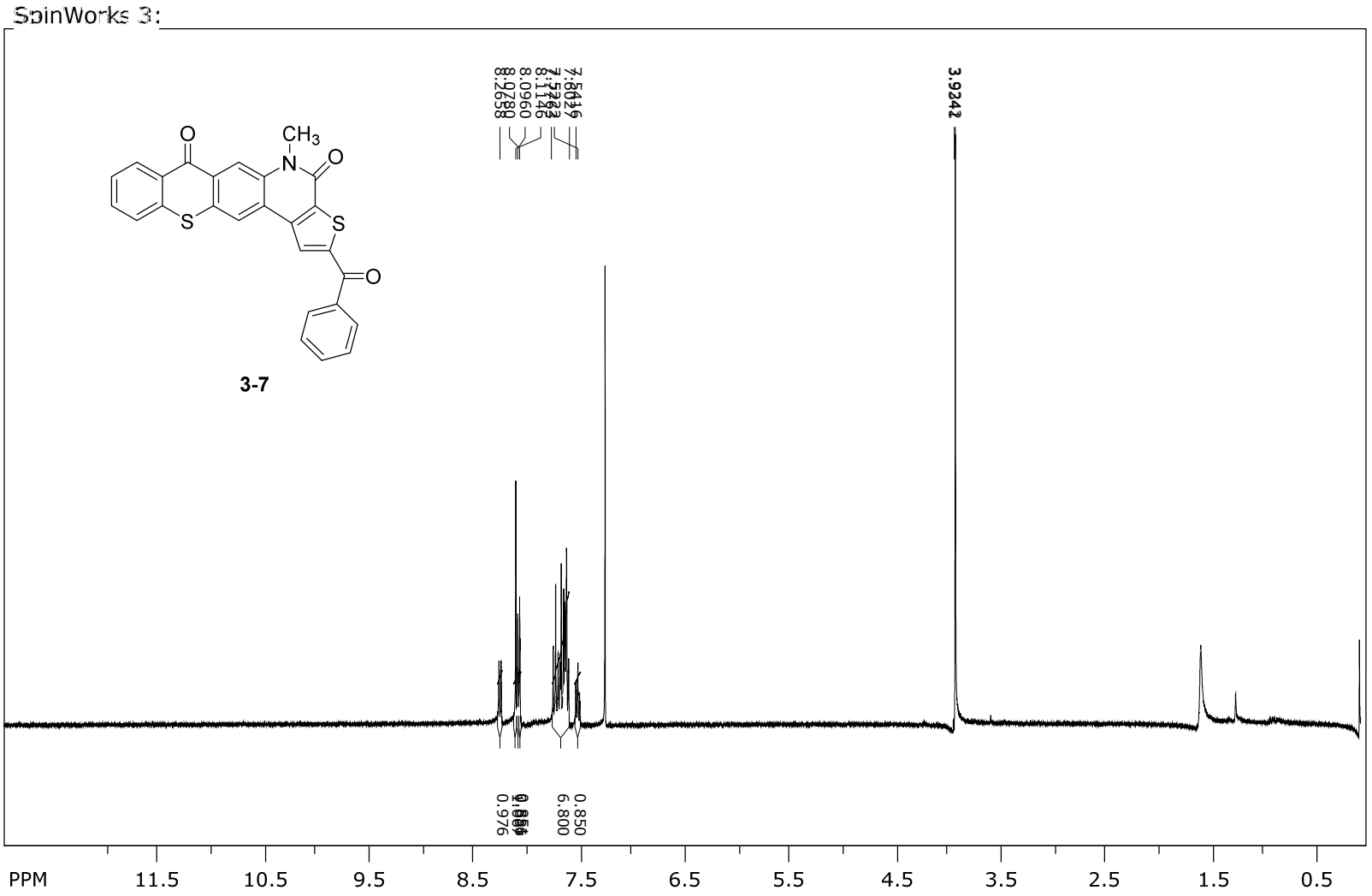


Figure 46. ^{1}H NMR spectrum of compound **3-7** in $CDCl_3$

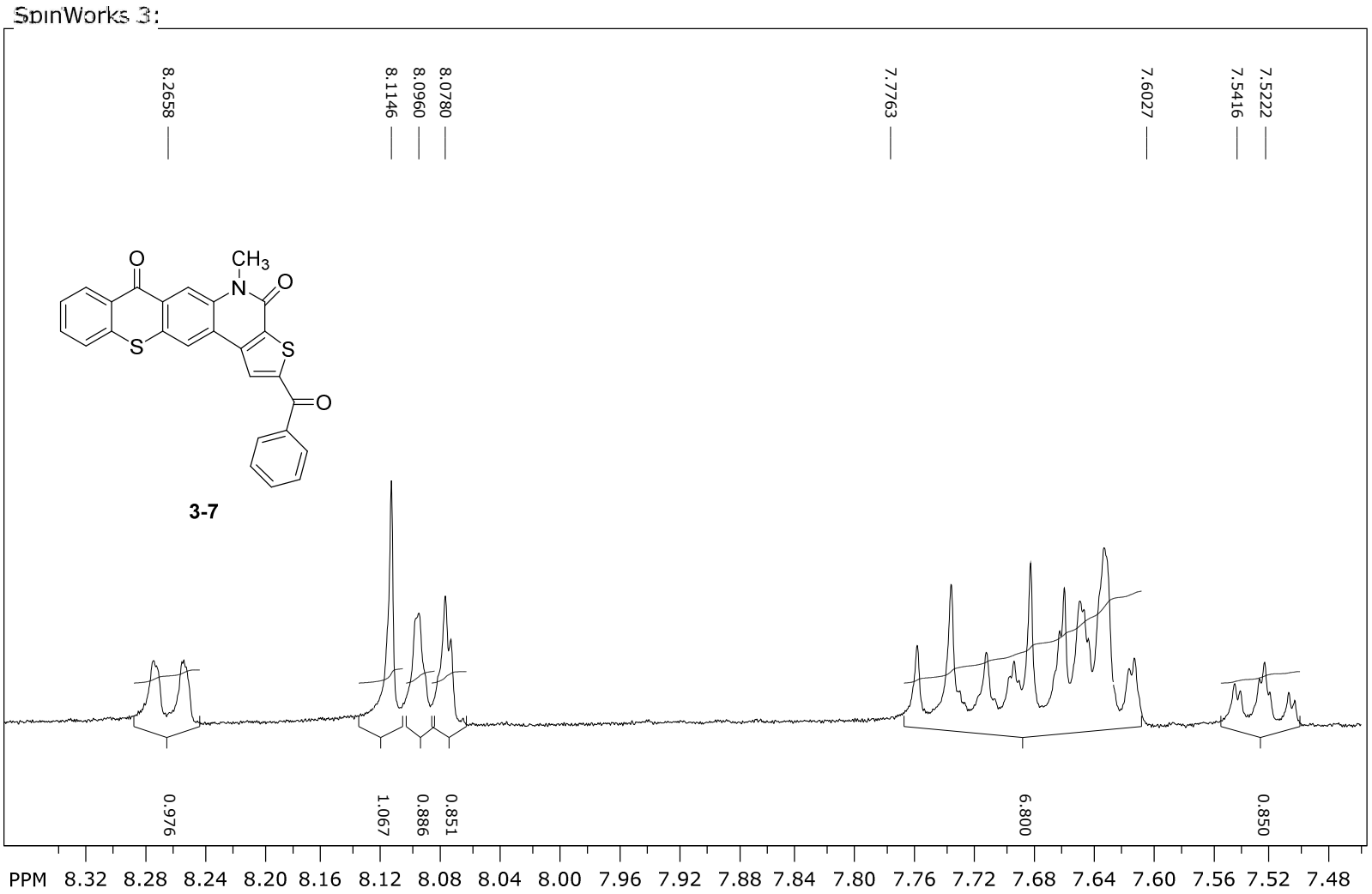


Figure 47. Expansion of the aromatic region of the ^{1}H NMR spectrum of compound **3-7** in $CDCl_3$

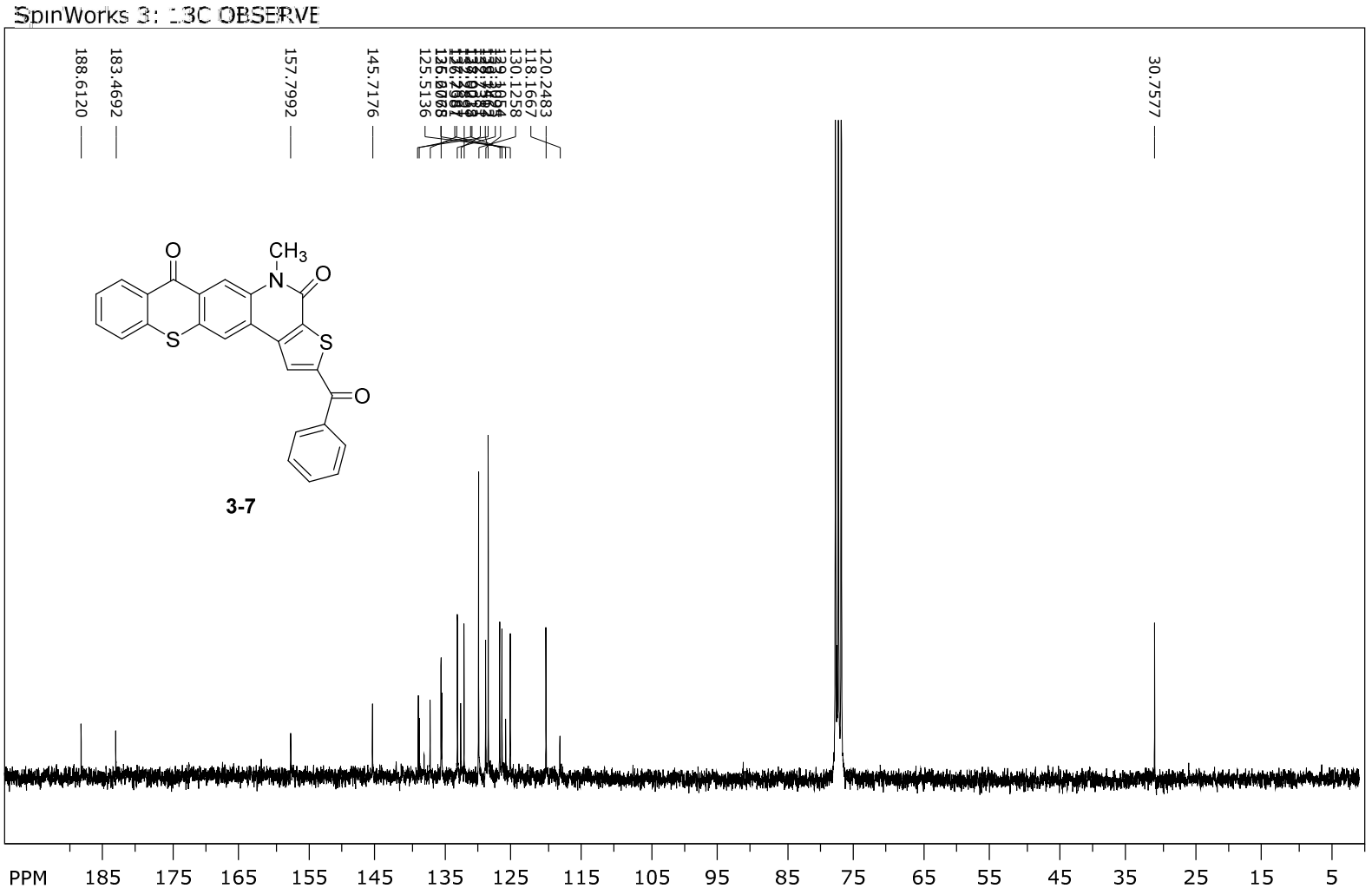


Figure 48. ^{13} C NMR spectrum of compound **3-7** in $CDCl_3$

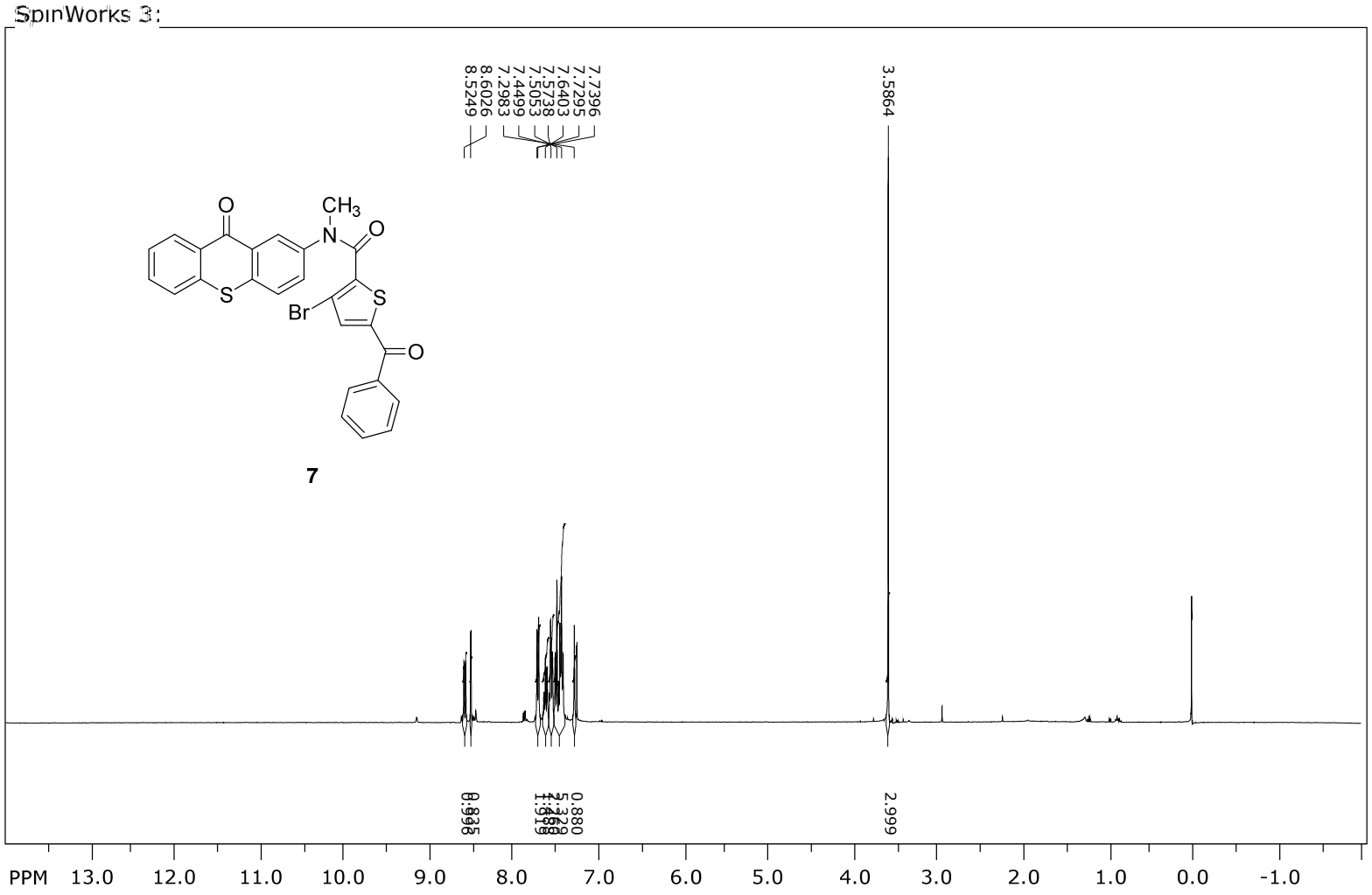


Figure 49. [1] H NMR spectrum of compound **7** in $CDCl_3$

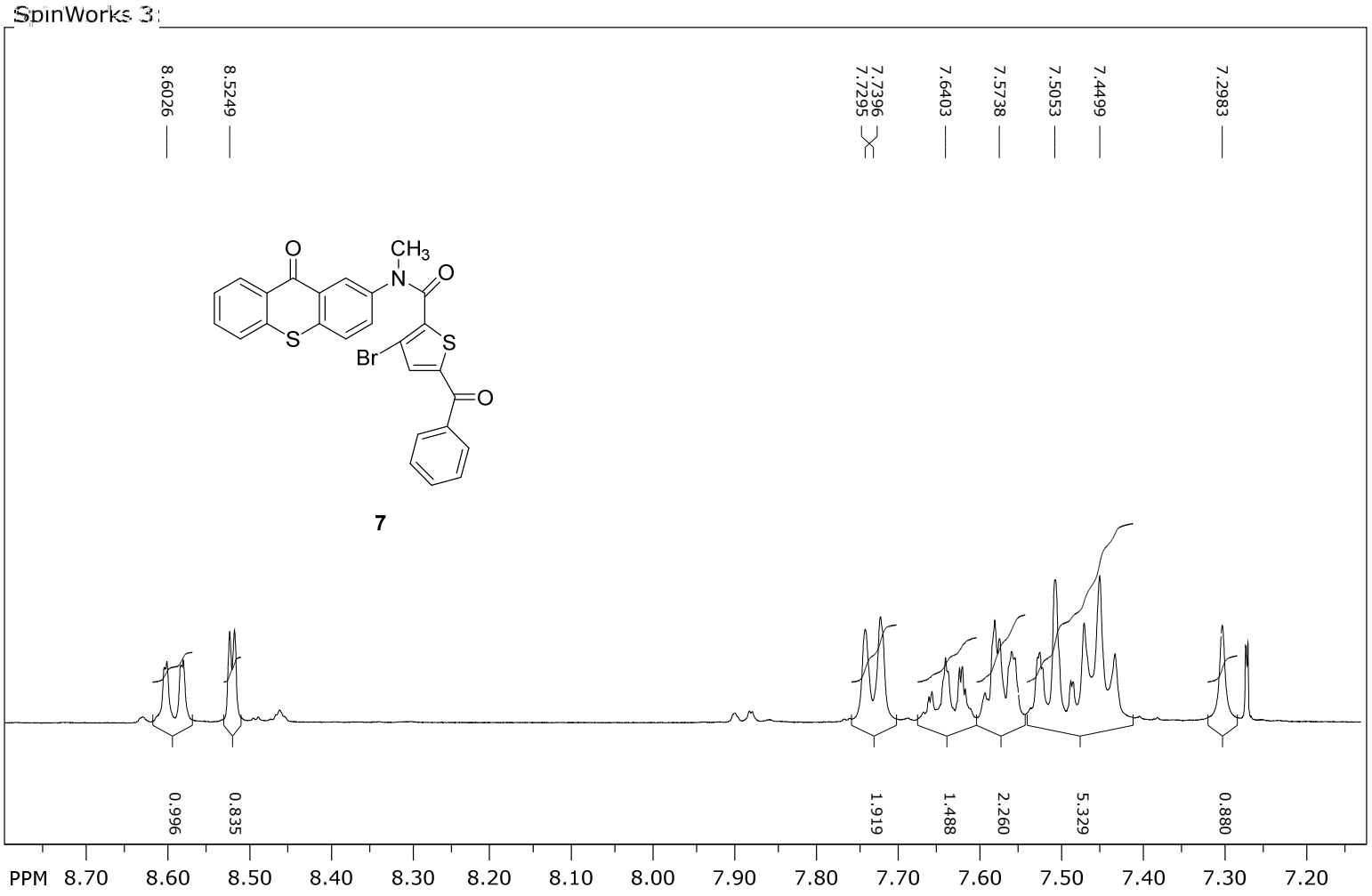


Figure 50. Expansion of the aromatic region of the ^{1}H NMR spectrum of **3-7** in $CDCl_3$

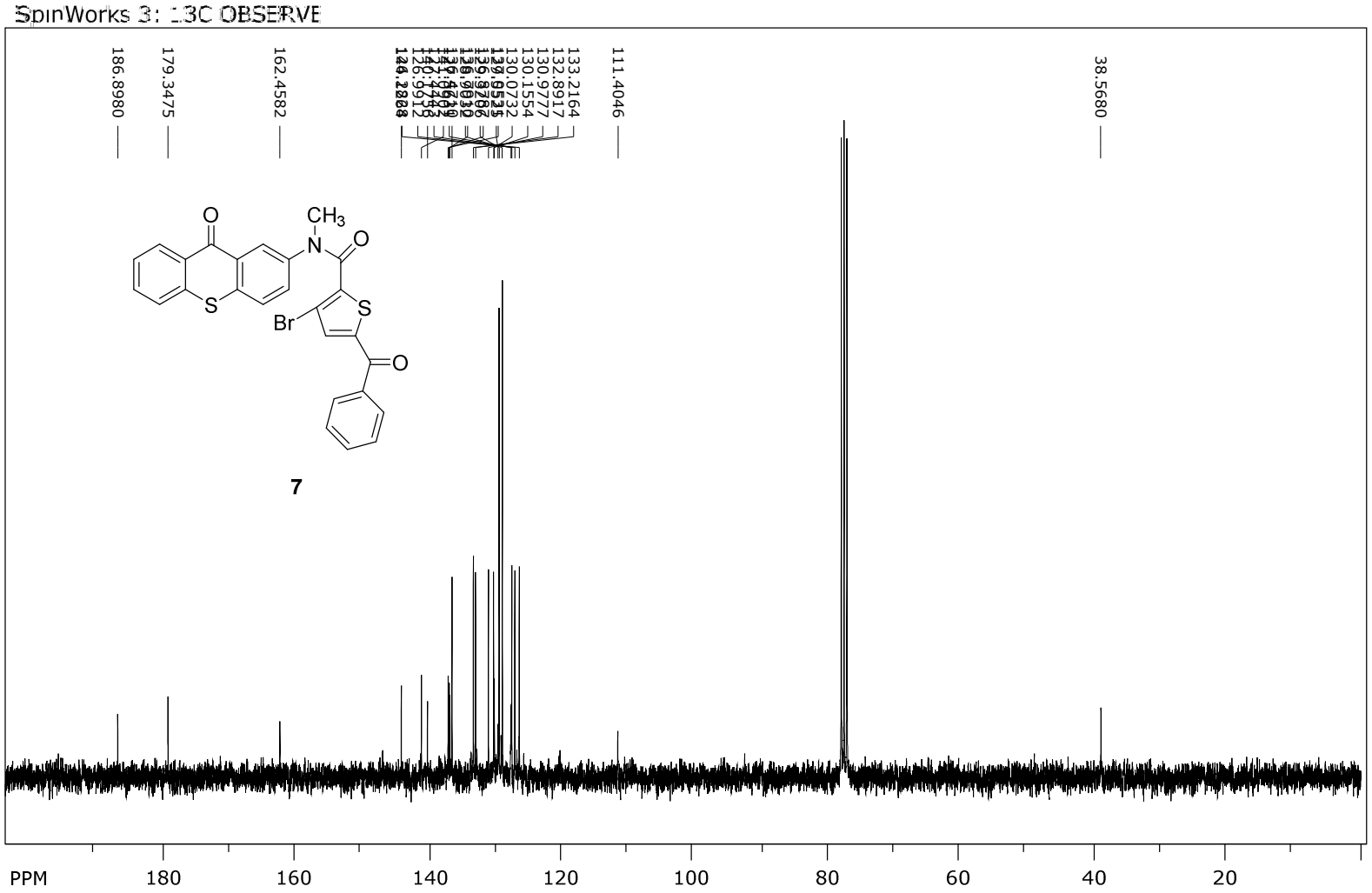


Figure 51. ^{13}C NMR spectrum of compound **7** in $CDCl_3$