Photochemical Elimination Reactions that Proceed via Triplet Excited State Electrocyclic Ring Closures

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PHOTOCHEMICAL ELIMINATION REACTIONS THAT PROCEED *via* TRIPLET EXCITED STATE ELECTROCYCLIC RING CLOSURES

by

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ABSTRACT

PHOTOCHEMICAL ELIMINATION REACTIONS THAT PROCEED *via* TRIPLET EXCITED STATE ELECTROCYCLIC RING CLOSURES

Himali Devika Jayasekara, B.Sc.(Hons.)

Marquette University, 2014

Cage compounds have become an important tool for the study of biological processes. The research focuses on new cage compounds that can unmask functional groups present in biologically important molecules such as proteins, peptides, and oligonucleosides. The project focuses on certain functional groups that are often difficult to release photochemically. These are the thiolate groups present in cysteine residues of proteins and peptides. Thiolate groups are fairly basic leaving groups, unlike the more labile groups such as the carboxylates that are present in proteins and peptides, or the phosphate groups present in nucleosides. The research takes advantage of the ability of zwitterionic intermediates to release basic leaving groups such as the thiolates. The zwitterionic intermediates are generated photochemically by electrocyclic ring closure of aromatic carboxamides that has the chromophore attached to the amide nitrogen. Most importantly, the research utilizes a chromophore that absorbs visible light, so as to minimize the damaging effects that short-wavelength light has on tissue and cells.

The research recognizes that triplet energy transfer from triplet excited state of the chromophore to the aromatic ring system attached to carboxamide carbonyl group must be exothermic in order for the electrocyclic ring closure to occur. For a thioxanthone chromophore (E_T = 64 kcal mol^{-1}), the aromatic ring system is a naphthothiophene ring system (E_T = 62 kcal mol^{-1}). The energy transfer would therefore be exothermic. This cage compound was synthesized with a 3-chloro leaving group. It undergoes photochemical electrocyclic ring closure and chloride expulsion in 50% yield after 1.5 h photolysis. The reaction qualitatively appears to be efficient. In comparison, a 5-benzoylthiophene aromatic ring system with 3-chloro group undergoes the same photoreaction in very low yields over 72 h, even though the triplet energy transfer is exothermic. In this case the photoproduct effectively competes for the incident light with the reactant.

The research shows that the naphthothiophene ring system is a viable solution to the triplet energy transfer problem. It also points to a need to improve the solubility of the cage compound by incorporating one or more carboxylate groups into the naphthothiophene ring or the thioxanthone ring.

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LIST OF ABBREVIATIONS

Anhyd.	Anhydrous
ATP	Adenosine triphosphate
cAMP	Cyclic adenosine monophosphate
Boc	*tert*-butoxycarbonyl
tBu	*tert*-butyl
CNDO	Complete Neglect of Differential Overlap
Cys	Cysteine
DBA	Dibenzylideneacetone
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DFT	Density functional theory
DMAP	Dimethylaminopyridine
DMF	Dimethylformamide
DMSO	Dimethylsulfoxide
DNA	Deoxyribonucleic acid
Et	Ethyl
GABA	γ-Aminobutyric acid
GHS	Glutthione

Glu	Glutamic acid
Gly	Glycine
HPLC	High Performance Liquid Chromatography
LG	Leaving Group
mp	Melting point
NMR	Nuclear Magnetic Resonance
NVOC	6-nitroveratroyloxycarbonyl
PPG	Photoremovable Protecting Group
PCC	Pyridinium chlorochromate
p-TsOH	*p*-toluene sulphonic acid
Pyr.	Pyridine
RNA	Ribonucleic acid
THF	Tetrahydrofuran
TMS	Tetramethylsilane
UV	Ultraviolet

CHAPTER 1. Introduction

1.1. General Introduction

Photoremovable protecting groups (PPGs) have been known to chemical society for a long time. They differ from classical protecting groups because they do not need cleaving reagents. This can offer major advantages in performing reactions with high selectivity when mild reaction conditions are needed.

PPGs were introduced to the World of life science by Kaplan[1] and Engels[2] in the late 1970s. Since this pioneering work many applications of PPGs in biochemistry, physiology, and medicine have emerged. Their usefulness has led to considerable interest in designing new types of PPGs. PPGs are not only important in biological studies, they have also been used in organic synthesis and in photolithography. A number of reviews and books on PPGs in synthesis[1-5] and mechanistic studies[1,6], have appeared in recent years.

Several synonyms can be found in the literature for PPGs. PPGs have been referred to as "phototiggers", "caged compounds" and "photolabile groups". The term "caged compound" is used to describe a biological molecule, whose activity or function is masked by chemical modification with a photoremovable protecting group. Typically biomolecules or bioeffectors are covalently bonded to the PPG. Excitation of a caged compound with light results in cleavage of that bond to more or less rapidly liberate the biomolecule of interest to trigger some biological processes. They are very useful in biological studies because irradiation with light can be used to control the release in terms of time and space.[7,8] The time period for the release of the bioeffector upon photolysis

will need to be fast enough to allow the study of interest. Such biological studies have used caged compounds where the bioeffector is released over minutes, as in the case of caged protein kinase A[8a], or seconds, as in the case of caged tyrosine Ca/calmodulin inhibitor[8b], or milliseconds as with caged ATP[8], or microsecond as with caged neurotransmitters.[7,8d]

1.2. Drawbacks of Common Photoremovable Protecting Groups and Objectives of Proposed Research

Although a number of caged compounds are currently used to release biological molecules (Figure 1.1), no universal photoremovable protecting group exists that is suitable for all applications. A major problem to be addressed is the photolysis wavelength. Most currently available caged compounds use UV light for releasing biomolecules.

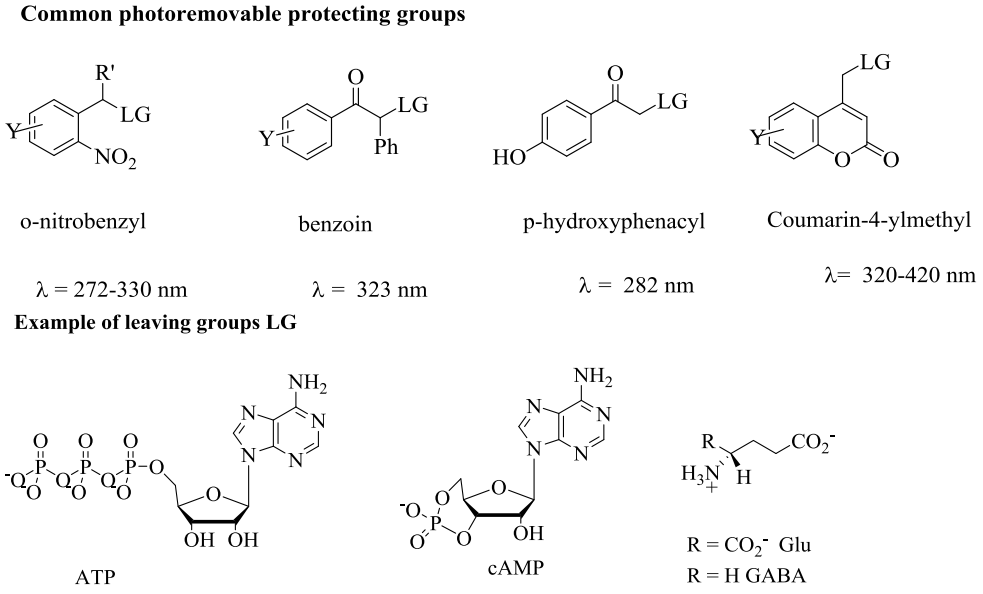


Figure 1.1. Common photoremovable protecting groups and biologically important leaving groups

The use of UV light can cause cell damage and mortality due to unintended side reactions of biomolecules.[8b] In addition, many biological systems involve enzymes in an aqueous medium. So the second problem is the premature release of the bio-effectors in the dark under such physiological conditions[8], as many caged compounds are not stable at high ionic strength under aqueous conditions. Another problem is the limited basicity of releasable biological anion with most caged compounds. Most biological systems involve biomolecule like, proteins and peptides and the building blocks of those compounds are amino acids. The amino acid residues contain side chain functionality like phenolates and thiolates, as for example cysteine and tyrosine, which are basic and difficult to mask. Few satisfactory caged compounds are currently available for thiols, including the sulfhydryl group of cysteine residue. The nitrobenzyl group had been thought to be suitable for caging cysteine residues.[9] In fact, o-nitrobenzyl protected cysteine was at one time commercially available from Molecular Probes (Invitrogen). However, nitrosoarene is a byproduct of the release of thiols from o-nitrobenzyl protected thiol compounds.[3] Unfortunately nitrosoarene will generally undergo reduction in the ground state by released thiol.[10] Indeed, one of the functions of glutathione(GHS) with it's cysteine sulfhydryl group is to reduce toxicity of foreign substances[11] including nitrosocompounds by converting them to less toxic compounds like arylhydroxylamines, N-arylsulfenamides, and anilines.[10] Therefore, o-nitrolbenzyl derivatives are not suitable for caging sulfhydryl groups of glutathione (GHS) or cysteine (Cys). *p*-hydroxyphenacyl-caged thiols have been reported by Goeldner and co-workers.[12] The disadvantage is that the photolytic wavelength lies deep in the UV. In addition 30% of the reaction produces a thioester byproduct due to nucleophlic attack of thiol on the cyclopropanone

intermediate formed in a step of the photoreaction. Therefore the ability to release more basic leaving group anions than carboxylates and phosphates is needed. Our research focuses on developing new caged compounds that overcome the above-mentioned problems. Our aim is to develop caged compounds with high absorption wavelength, high stability towards premature release, that are capable of releasing a wide range of leaving groups.

1.3. Necessary Criteria for Successful Caged Compound.

The criteria[3, 5] for successful caged compound for the release of common biological substrates has been proposed and included following: 1) The photoprotected substrate must be soluble in aqueous buffered media. 2) The caged compound must be stable to hydrolysis, especially at high ionic strength. 3) The absorption wave length should be >300 nm to avoid photolyzing the biological media. 4) The photochemical reaction should have high quantum efficiency, preferably with quantum efficiency > 0.1. 5) The caged compounds and the photolysis products must be biologically harmless. 6) The photoproduct should not absorb light at the same wave length where the caged compound absorbs.

Even though a caged compound doesn't meet all those criteria, it may still have utility. The proposed criteria can be considered as excellent guidelines for designing and developing new caged compounds.

1.4. Applications of Caged Compounds in Biological Studies

Caged compounds have become an important tool for studying biological processes. The photoreleased bio-effector leaving groups may be small molecules or they may be macromolecules such as polypeptides, proteins , DNA or RNA. There are many examples which illustrate the utility of caged compounds in biological studies.

A well known example is caged neurotransmitters, in investigations of the kinetics of neurotransmitter mediated reactions on cell surfaces.[6] This broad area of research has been covered in several reviews.[13-16] Activation of synaptic transmission in the nervous system occurs on a sub-micron spatial scale and a sub-millisecond time scale, so experimental approaches for studying synaptic function ideally require similar precision. A useful strategy is the use of a caged compounds to release the neurotransmitters. Numerous photoactivatable derivatives of neurotransmitters and neurotransmitter antagonists are available. Glutamate, γ-aminobutyric acid (GABA), glycine, aspartate, and kainic acid are the well known examples for the neurotransmitters and neurotransmitter inhibitors. The opening of ion channels takes place directly or indirectly when neurotransmitters are binding to their receptors. The use of caged neurotransmitters makes it possible to monitor the kinetics of this process. Grewer and co-workers[17] have reported the development of nitrobenzyl protected caged compound, α-carboxy-2-nitrobenzyl(αCNB) ester of glycine with high quantum efficiency ($\Phi = 0.38$) and thermal stability at physiological pH. The decay of the aci-nitro intermediate occurred with biexponential kinetics with lifetimes of 7 and 170 μs. So this is useful tool for investigation of the process associated with channel opening of glycine receptor channels and the effect of the mutations of glycine receptor and inhibition of these

processes. Kandler *et al.*[18] has reported on p-hydroxyphenacyl protected glutamate in the study of long term potentiation and depression, which are two neural processes that are thought to be involved in memory and learning.

Another well known area concerns studies of the photorelease of protein and peptides. Peptides have a wide range of biological activities and functions. Synthetic peptides can be used to selectively inhibit or activate protein activity. Photoactivable peptides have the potential for extensive applications. The design involves identification of amino acid substitution patterns which inhibit activity of peptide such as preventing the peptide from binding to a target protein. Walker *et al.*[8b] reported the study of nitrobenzyltyrosine in RS-20, which is target peptide for calmodulin, which binds Ca^{2+} and is involved in a number of Ca^{2+} mediated reactions. Caged RS-20 shows greater affinity for calcium-calmodulin than RS-20. This system was used for studing Ca^{2+} calmodulin binding activity and Calmodulin dependent Myosin Light-Chain Kinase(MLCK) activity.

Chan and co-workers[19] reported utilization of a benzoin protected protein to investigate the kinetics of protein folding. A subdomain of the protein villin, which folds to an α-helical structure in aqueous solution was used. A small loop was formed by the N-terminus of the peptide with cysteine residue on the side chain of one internal amino acid using benzoin as the linker (Figure 1.2). This cross link prohibited folding. When irradiated with light, cleavage of the linker takes place and the protein forms an α-helical structure.

Another interesting application of caged compounds is light directed synthesis of high density arrays of peptides and oligonucleotides(biochips)[20]. Solid phase synthesis,

photoremovable protecting groups, and photolithography have been combined to achieve this. This allows the preparation of thousands of different sequences at specific locations on a surface. Application of this was first reported by Fodor and co-workers[20a] for synthesizing an array of 1024 peptides using the 6-nitroveratryloxycarbonyl (NVOC) photoremovable protecting group. In this technique (Figure 1.3), a substrate **S** bears amino groups that are blocked with photoremovable protcting group **X**. Irradiation of specific regions with UV light through a mask M_1leads to deprotection of photolabile protecting group X. Amino group in the exposed area of the substrate are now free to couple with building block A containing protecting group X. The different mask M_2 is used to photoactivate a different region of substrate. Then second building block B containing protecting group X is added and joined to the newly exposed amino groups

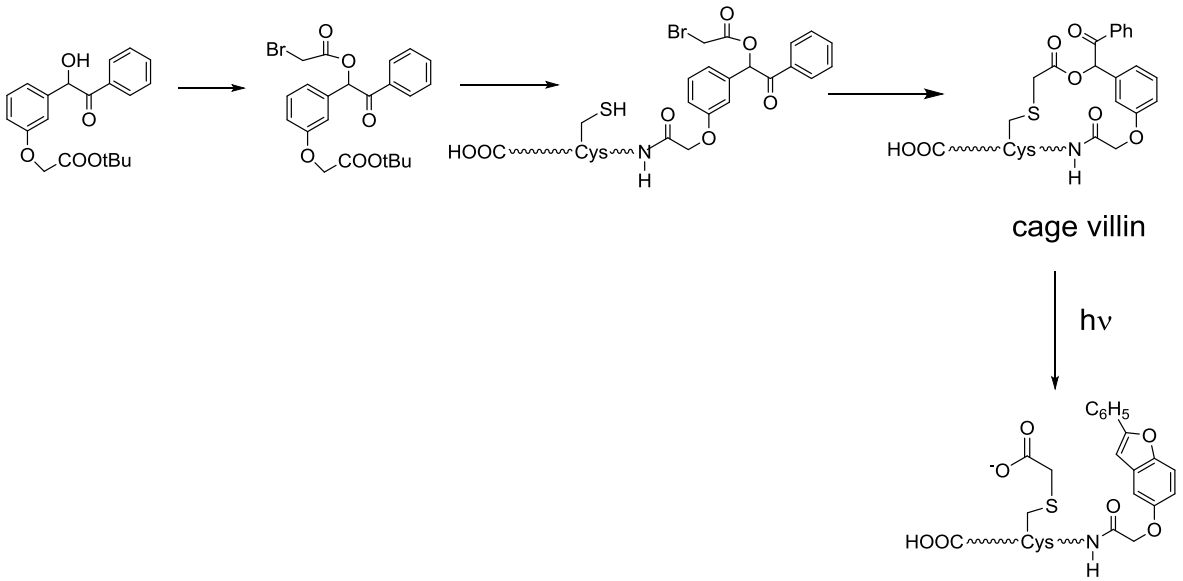


Figure 1.2. General strategy for peptide cyclization and photolysis

Repetition of irradiation and coupling steps using a mask with variable patterns and different compounds leads to the synthesis of a desired biochip.

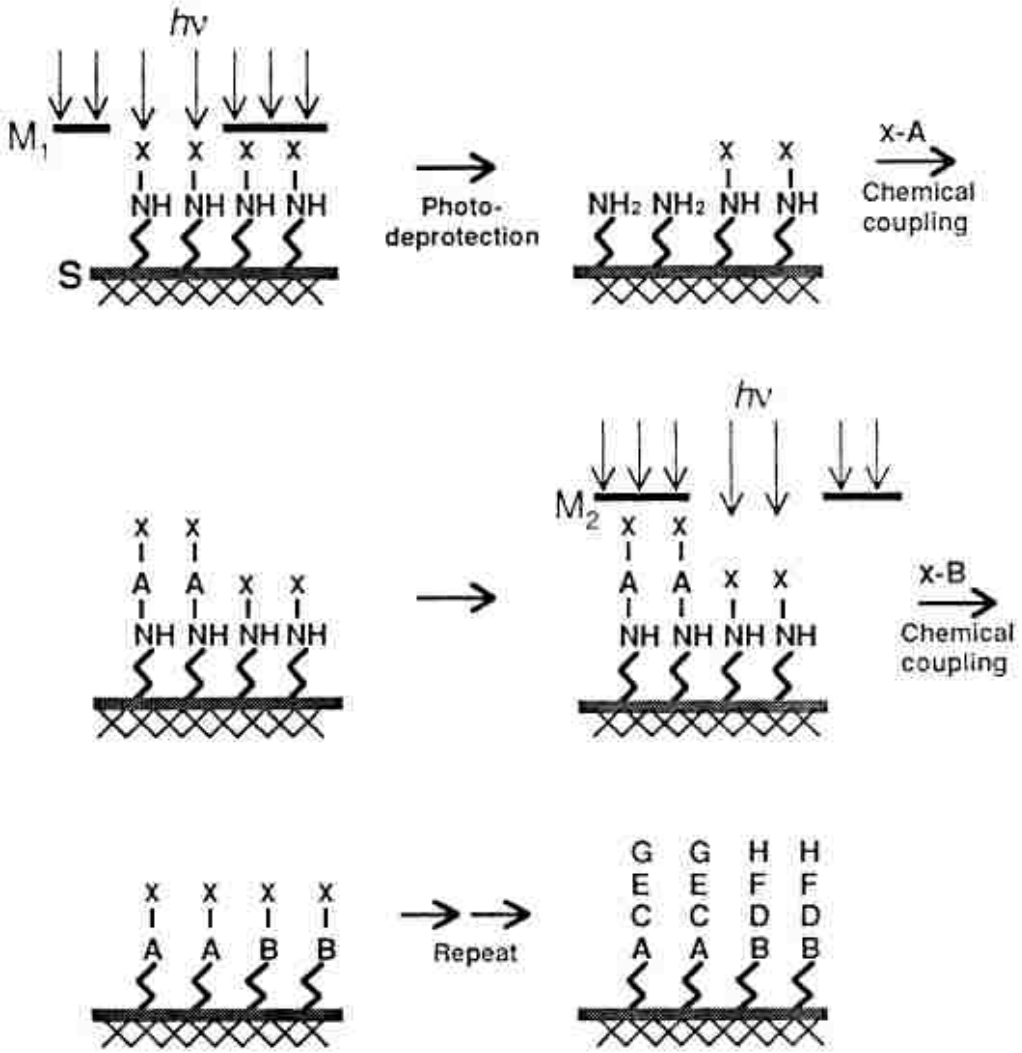
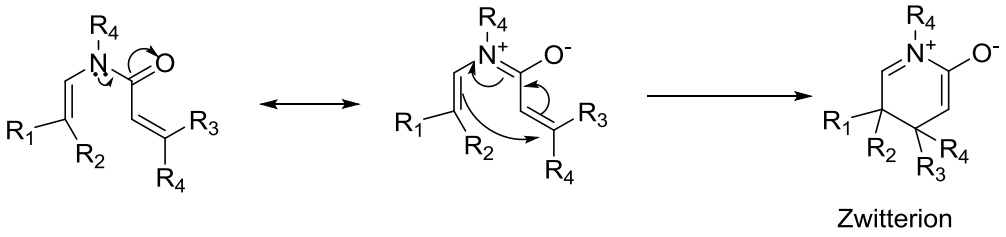


Figure 1. 3. Light directed synthesis of high density array of peptide

1.5. Previous Studies of Electrocyclic Ring Closure Reactions via Zwitterionic Intermediates with the Expulsion of Leaving Groups

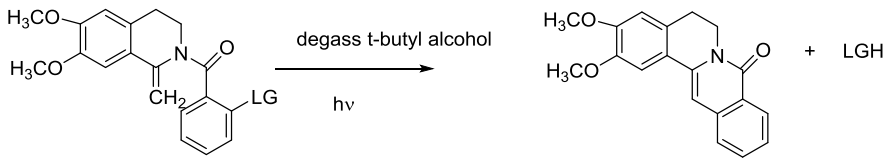
Photochemical electrocyclic ring closure reactions which involve zwitterionic intermediate have long history, and numerous studies have been reported over the past 40 years.[21] Most of them involve cyclization of a 6π electron system with an amide functional group. The C-N bond of amide has double bond character. So it can act as two double bond of the 6π electron system, and the electrocyclic ring closure reaction takes place to form a zwitterion (Scheme 1.1). Our research group has been adopting these electrocyclization of amide for the purpose of releasing leaving groups (LG^-) *via* the zwitterionic intermediate generated by photolysis. The zwitterionic intermediate possesses a basic site that, in principle, can be utilized to effect the elimination of leaving group anions. One of the major advantages of such type of intermediates is that they are capable of eliminating a wide range of leaving anions.

Scheme 1.1



George Lenz and co-workers[22] reported the utilization of electrocyclic ring closure and leaving group expulsion reaction in the synthesis of alkaloids (Scheme 1.2 and Table 1.1).

Scheme 1.2



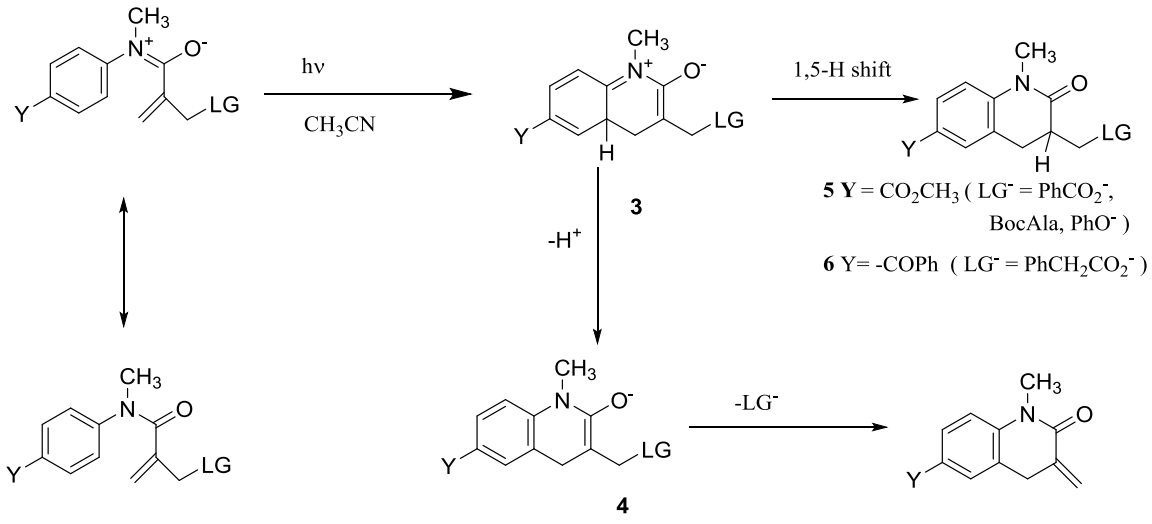
Enamide, (LG =)	**Product (% yield)**
F	85
Cl	50
Br	50
O_2CCH_3	76
SCH_3	55

Table 1.1. Chemical yields for the reaction in Scheme 1.2

Photochemical electrocyclic ring closure has been previously used in this laboratory to generate zwitterionic intermediates that would be capable of expelling leaving groups such as carboxylates and phenolates, which are functionality present in many biologically important molecules. Their study used a photochemical electrocyclic ring closure to generate zwitterionic intermediates from α, β-unsaturated anilide **1**,**2** bearing leaving groups. The anilides **1**,**2** undergo photochemical conrotatory electrocylic ring closure reaction with leaving group release from α-methylene lactam occurring in **3** or **4** to give **7**,**8** (Scheme 1.3).[23] Leaving groups with wide range of basicities from carboxylate to phenolate could be expelled under aqueous conditions and the efficiency is insensitive to leaving group basicity. The electrocyclization was 8-10 %

efficient with respect to light utilization. Leaving group expulsion probably doesn't occur directly from zwitterionic intermediate **3**, but were instead eliminated from enolate **4** produced by deprotonation of zwitterion **3**. This could account for why the efficiencies were insensitive to leaving group basicity. Formation of minor photoproduct **5**, **6** also reported by thermally allowed suprafacial sigmatropic 1, 5- H shift. So leaving group expulsion does not represent 100% reaction. The quantum efficiency for leaving group release appeared to be controlled by the competition between deprotonation and 1, 5- H shift. The quenching studies indicate that the photochemistry derived from the singlet excited state.

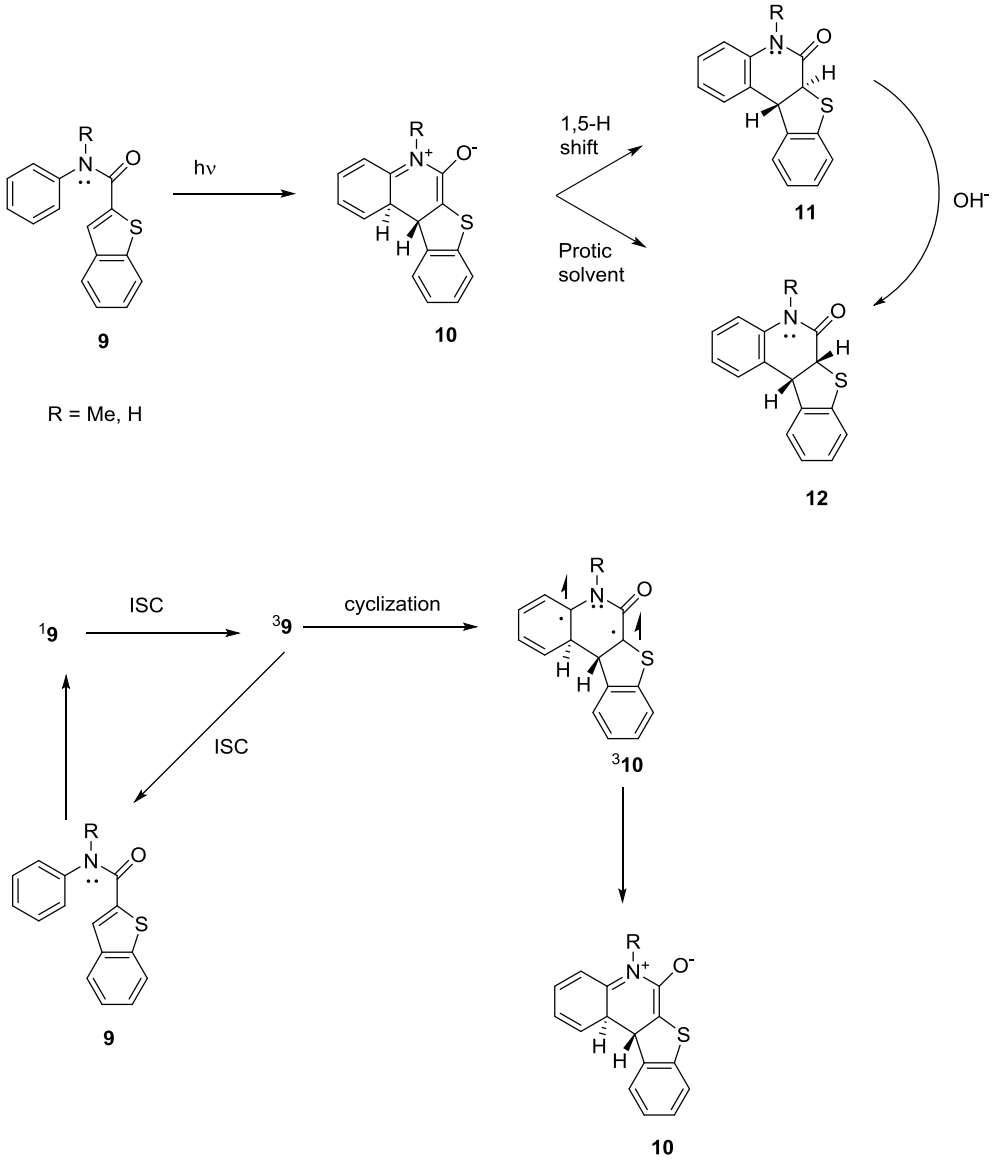
Scheme 1.3



In order to increase the competition for the deprotonation pathway they have planned to replace acryl amide moiety with aromatic ring system. Previous study by Witkop and co-workers[24] have reported that incorporation of the benzothiophene ring system instead of

the acrylic group led to high yield of electrocyclic ring closure reactions (Scheme 1.4) but they have not reported the quantum efficiency. Photochemistry of this reaction has been reported tentatively as proceeding *via* triplet excited state.

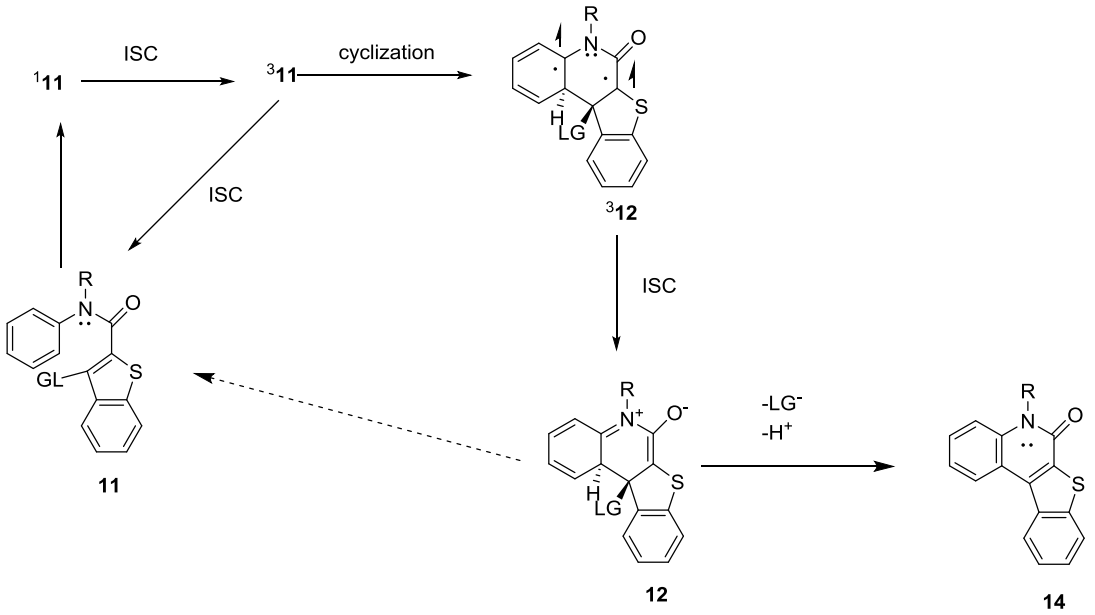
Scheme 1.4



So our research group adapted to use the benzothiophene carboxanilide system which incorporates various leaving groups (LG^-) at the C-3 position of benzothiophene ring

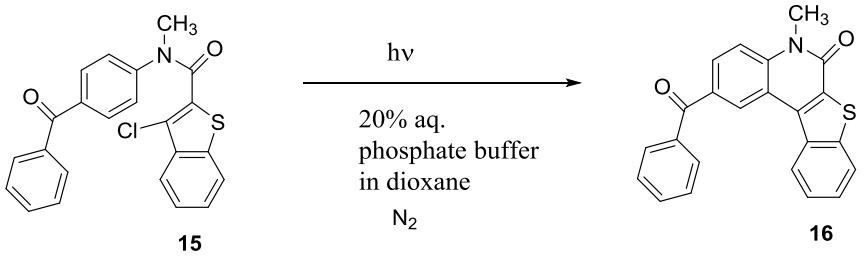
system (Scheme 1.5).[25] They reported that carboxanilide **11** could release various leaving groups (LG^-) at 310 nm that vary in basicity in essentially quantitative yield to form **14** and quantum yields decreased with increasing basicity of the leaving group (LG^-). Quantum yield were over the range 0.23-0.007 (LG^- = Cl^-, $PhCH_2CO_2^-$, PhS^-, $PhCH_2S^-$, PhO^-). Dependence of Φ on LG^- basicity is consistent with the formation of ground state intermediate **12**, which expel leaving group or ring opening to give starting material. Quenching studies and heavy atom effect indicated that the reaction takes place through triplet excited state. Even though the photolytic wave length is low, the ability to release relatively basic leaving groups such as thiolates and phenolates is advantageous for biological studies.

Scheme 1.5



By incorporating a *p*-benzoyl group onto the benzene ring of anilide **15** (Scheme 1.6) the photolytic wave length could be extended to 365 nm. The quantum yield was only somewhat lower compared to benzothiophene caboxanilide **11**.

Scheme 1.6



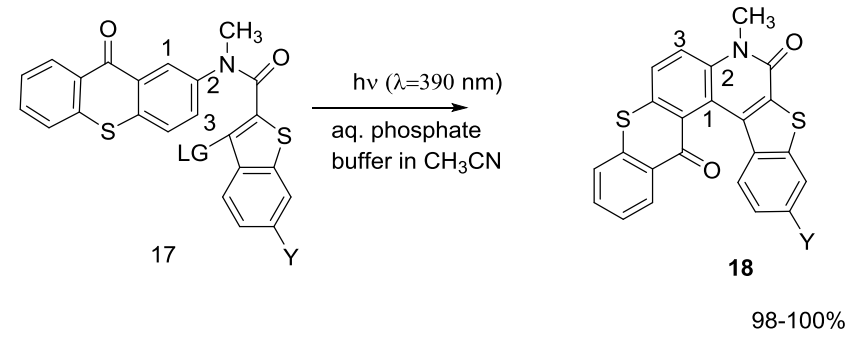
λ = 365 nm

ϕ = 0.15

As given in Scheme 1.7, incorporating thioxanthone as chromophoric group they could extend photolytic wave length to 385 nm.[26] Without any substituent Y (**19**) in the benzothiophene moiety they reported the formation of two products by cyclization at the C-1 (**20**) and C-3 positions (**21**) of the thioxanthone ring system in a 42:58 ratio in aq. phosphate buffer in acetonitrile. When substituent Y was carboxylic or methyl ester at C-6 position of benzothiophene (**17**) system the only product formed was via cyclization at the C-1 position (**18**). By incorporating a carboxylic acid group at the C-6 position they also were able to increase the aqueous solubility by considerable amount. Product quantum yields for this system are given in Table 1.2. Decreasing quantum yield with oxygen and piperyline indicated a triplet excited state reaction. Involvement of a triplet excited state was further supported by incorporating heavy atom, bromine at C-7 position of thioxanthone ring system (Scheme 1.8). 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, the disadvantage of this system is the low quantum yield compare to benzothiophene carboxanilide **11** and compound with benzophenone chromophore **15**.

Scheme 1.7



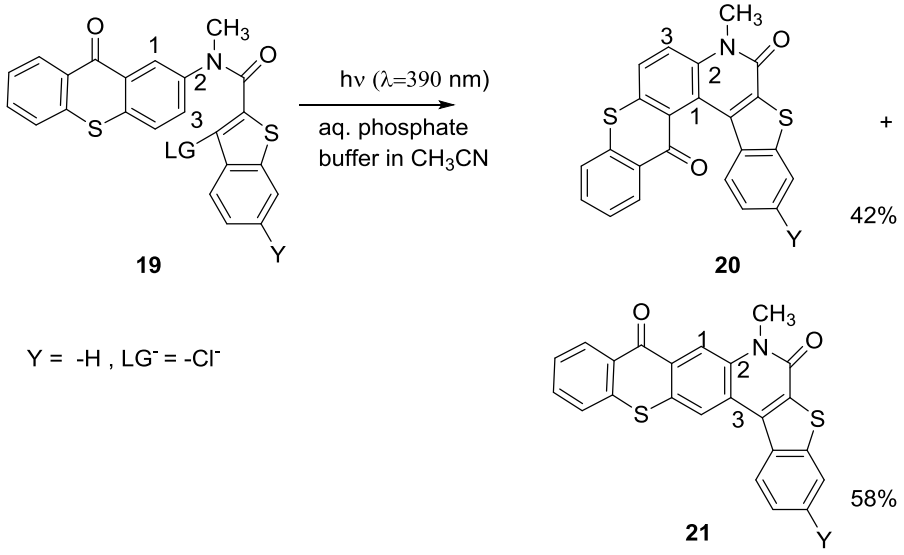
17

18

98-100%

Y = $-CO_2CH_3$, LG^- = $-Cl^-$

Y= -COOH, LG^- = Cl^- , PhS^- , HS^-, $PhCH_2S^-$



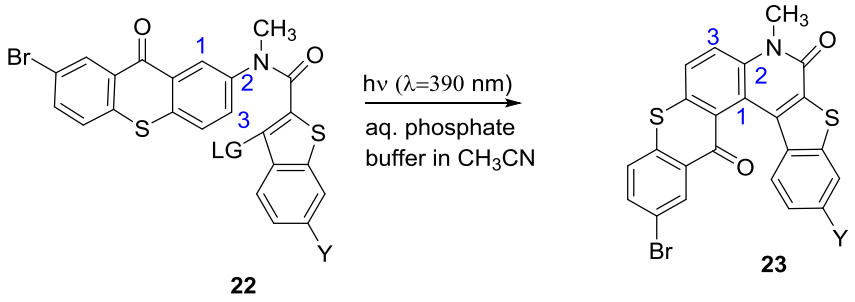
19

20

21

Y = -H , LG^- = $-Cl^-$

Scheme 1.8



22 **23**

99%

$LG^- = Cl^-$

$Y = -CO_2CH_3$

Reactant (Y=)	**LG^-**	**Reaction Conditions**	**Product %**	**Φ**
$-CO_2CH_3$	Cl^-	N_2 saturated	18(99%)	0.039
	Cl^-	O_2 saturated	-	0.019
$-CO_2CH_3$ (Br at C-7 position of thioxanthone)	Cl^-	N_2 saturated	23(99%)	0.053
-COOH	Cl^-	N_2 saturated	18(98%)	0.034
	PhS^-	N_2 saturated	18(99%)	0.017
	$PhCH_2^-$	N_2 saturated	18(99%)	0.011
	HS^-	N_2 saturated	(98%)	0.008
-H	Cl^-	N_2 saturated	20(42%)+21(58%)	0.069

Table 1.2. Quantum yield for the photoproduct **18-23**

1.6. Current Approach and Future Studies

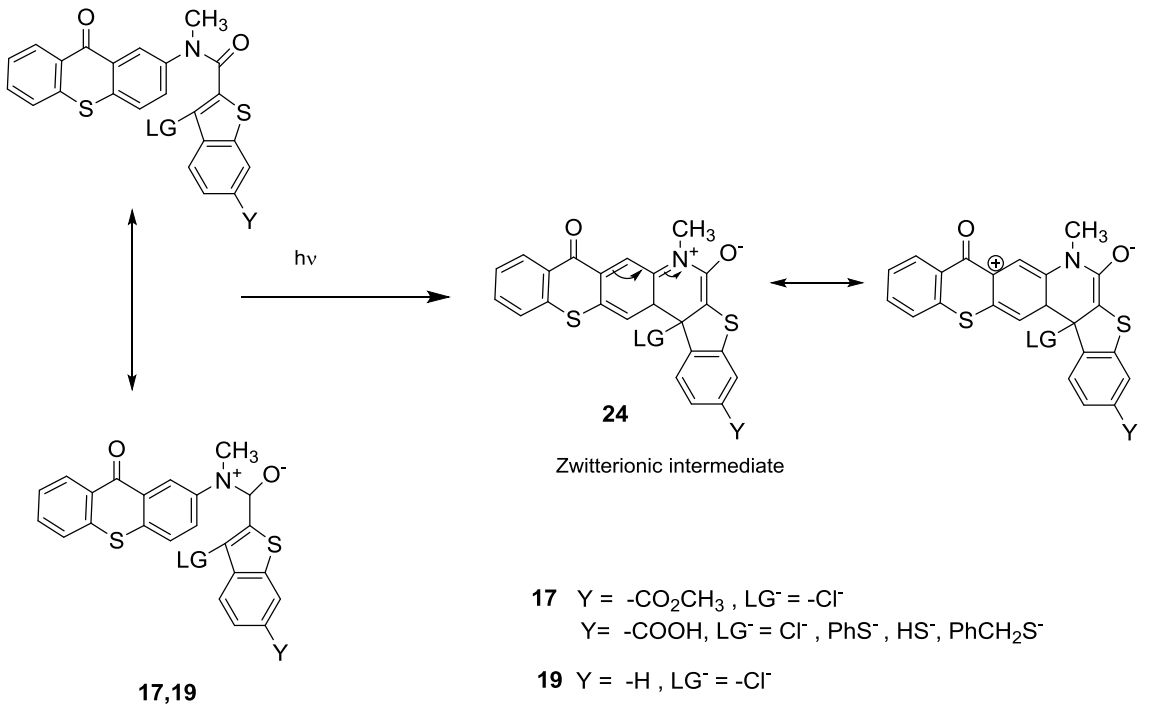
As mentioned in the above section N-(9-oxothioxanthenyl) benzothiophene carboxamide (**17** and **19**) bearing leaving groups ($LG^- = Cl^-$, PhS^-, HS^-, $PhCH_2S^-$) at the C-3 position of the benzothiophene ring system photochemically cyclizes with 98-99% yield.[26] This compound shows low quantum efficiencies for leaving group expulsions. Here they studied the molecule where the amide N is attached to the C-2 position of the thioxanthone moiety. When N is at the C-2 position of thioxanthone moiety the zwitterion **24** formed by electrocyclic ring closure reaction might be unstable (Scheme 1.9). Initially we thought that this could be the reason for the low quantum efficiency for the above compound. Therefore, we initially planned to synthesize N-(9-oxothioxanthenyl) benzothiophene carboxamide **25**, where the amide nitrogen is at the C-3 position of thioxanthone moiety. This will be discussed in Chapter 4.

Plans changed when DFT calculations [26] (Figure 1.1) became available for N-(9-oxothioxanthenyl) benzothiophene compound **17**, **19** which contain N-methyl amide at C-2 position of thioxanthone moiety. The calculations suggest that the reaction takes place via the initial triplet excited state where the excitation is localized on the thioxanthone ring system. Energy transfer then takes place to give a triplet excited benzothiophene moiety. Therefore the lower reactivity of the thioxanthone compound may be due to the fact that E_T =64 kcal mol^{-1} for thioxanthone.[27] The energy transfer to give the triplet benzothiophene[28] (E_T =69 kcal mol^{-1}) is endothermic by 5 kcal mol^{-1}. This less favorable energy transfer might be account for the lower quantum yield. Although it was thought that even though the photolytic wave length could be extended to 390 nm, combining thioxanthone and benzothiophene through the amide bond is

unsuitable for a caged compound with high quantum efficiency. This point it was planned to modify or replace the benzothiophene ring to facilitate the energy transfer from the triplet thioxanthone.

Phenyl-2-thienyl ketone, dithienyl ketone[29] and naphtho[1,2-b]thiophene[30] have triplet exited state with energies of 62 kcal mol^{-1} (Scheme 1.10). Those triplets are lower in energy than the benzothiophene triplet (64 kcal mol^{-1}). It was planned to replace the benzothiophene moiety with those molecules.

Scheme 1.9



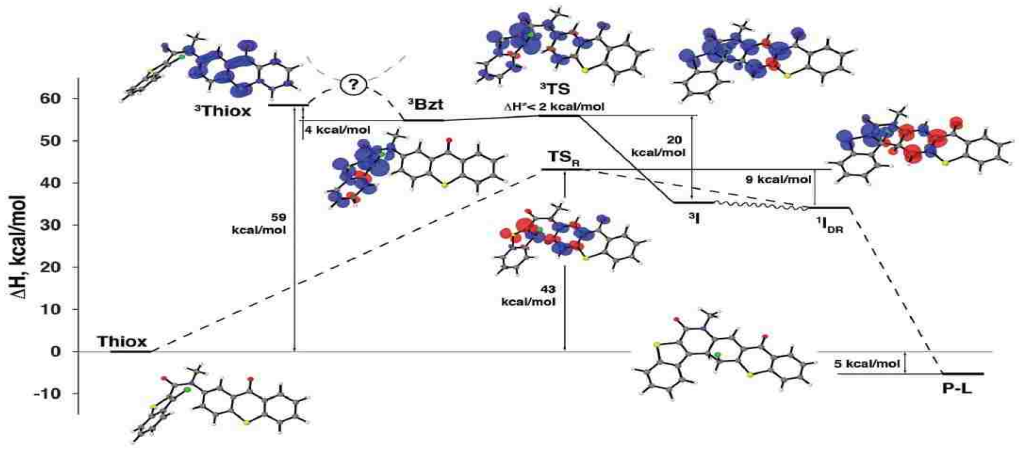
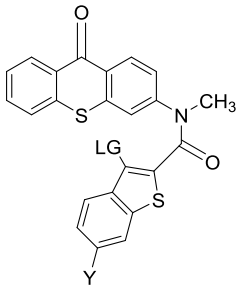


Figure 1.4. Relative enthalpies of the stationary points on the ground-state S_0 and the lowest triplet T_1 surfaces relevant for formation of the ring closure product from **17**, **19**. Unpaired spin density isosurfaces are shown for open-shell species.



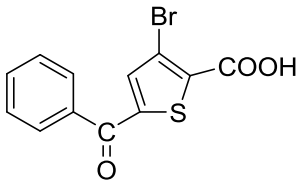
25

Y = -COCH3, -COOH

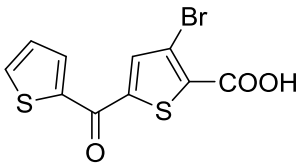
LG- = Cl-, and S containing leaving groups

Therefore, the part of the research studied the expulsion of a leaving group ($LG^- = Br^-$) from 5-benzoyl-3-bromothophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide **26** (Scheme 1.11). The results are given in the Chapter 2.

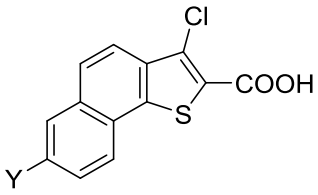
Scheme 1.10



phenyl 2-thieneyl ketone



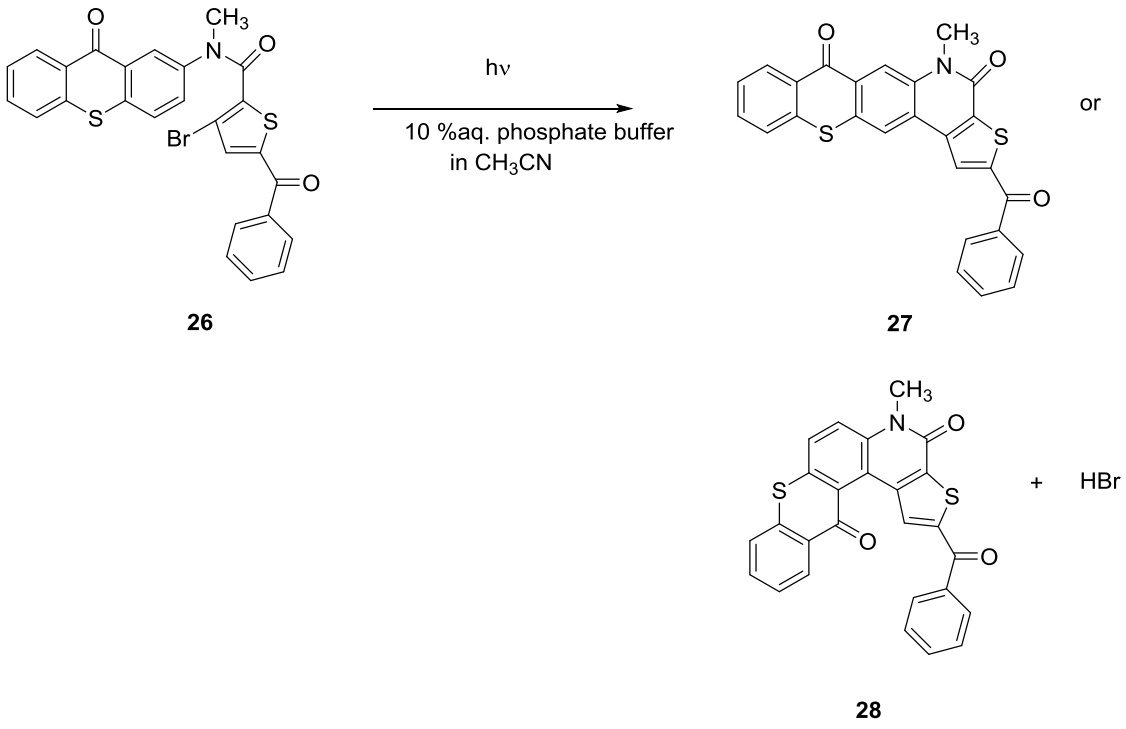
dithienyl ketone



naphtho[1,2-b]thiophene

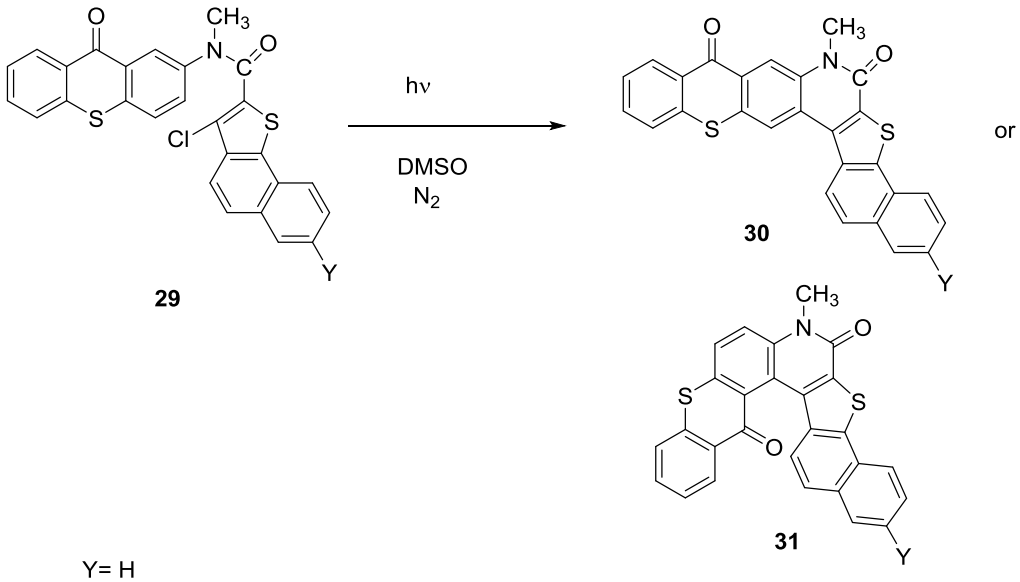
The expulsion of a chloride leaving group from 3-chloro-naphtho[1,2-b]thiophene-2-carboxylic amide 29 is also being studied (Scheme 1.12). The compound with Y = H has been synthesized. To improve aqueous solubility, a derivative with Y = -$COCH_3$ and -COOH (**32** and **33**) are being synthesized. Preliminary results suggest that **29** (Y = H) undergoes efficient photoreaction in DMSO. Therefore, the work on **29** (Y = -COOH) seems warranted. So our future plan is to synthesis compound **32** and **33** and study their photochemistry. Future studies of compound **32** and **33** will initially be focused on yield and efficiencies with less basic leaving groups and will be extended to attach more basic leaving groups like PhO^- and RS^- and study their photochemistry. Further studies will incorporated caged cysteine and tyrosine residues. This would allow further elaboration into other peptides by peptide synthesis.[69, 70] Conditions will also be determined to cage commercially available glutathione (GHS) at the central cysteine residue under aqueous conditions.

Scheme 1.11



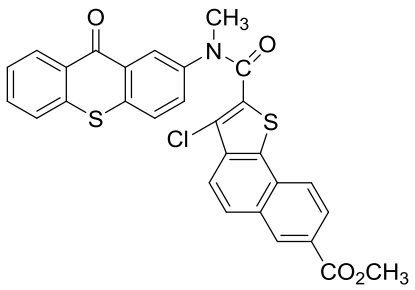
26 **27** **28**

Scheme 1.12

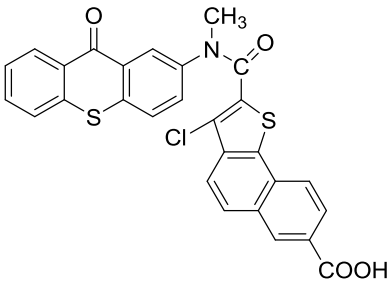


29 **30** **31**

Y= H



32



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CHAPTER 2. 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

2.1. Introduction

From DFT calculations [26] (Figure 1.1) for N-(9-oxothioxanthenyl) benzothiophene compound **17**, **19** which contain N-methyl amide at C-2 position of thioxanthone moiety suggest that the reaction takes place via the initial triplet excited state where the excitation is localized on the thioxanthone ring system. Energy transfer then takes place to give a triplet excited benzothiophene moiety. Therefore the lower reactivity of the thioxanthone compound with the benzothiophene moiety may be due to the fact that E_T = 64 kcal mol^{-1} for thioxanthone.[27] The energy transfer to give the triplet benzothiophene[28] (E_T =69 kcal mol^{-1}) is endothermic by 5 kcal mol^{-1}. This less favorable energy transfer might account for the lower quantum yield. Although it was thought that even though the photolytic wave length could be extended to 390 nm, combining thioxanthone and benzothiophene through the amide bond is unsuitable for a caged compound with high quantum efficiency. At this point it was planned to modify or replace the benzothiophene ring to facilitate the energy transfer from the triplet thioxanthone.

Phenyl-2-thienyl ketone, dithienyl ketone[29] and naphtho[1,2-b]thiophene[30] have triplet exited state with energies of 62 kcal mol^{-1} (Scheme 1.13). Those triplets are lower in energy than benzothiophene triplet (64 kcal mol^{-1}). It was planned to replace the benzothiophene moiety with those molecules.

Therefore, this part of our research studied the expulsion of a leaving group ($LG^- = Br^-$) from 5-benzoyl-3-bromothophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide **26** (Scheme 1.11).

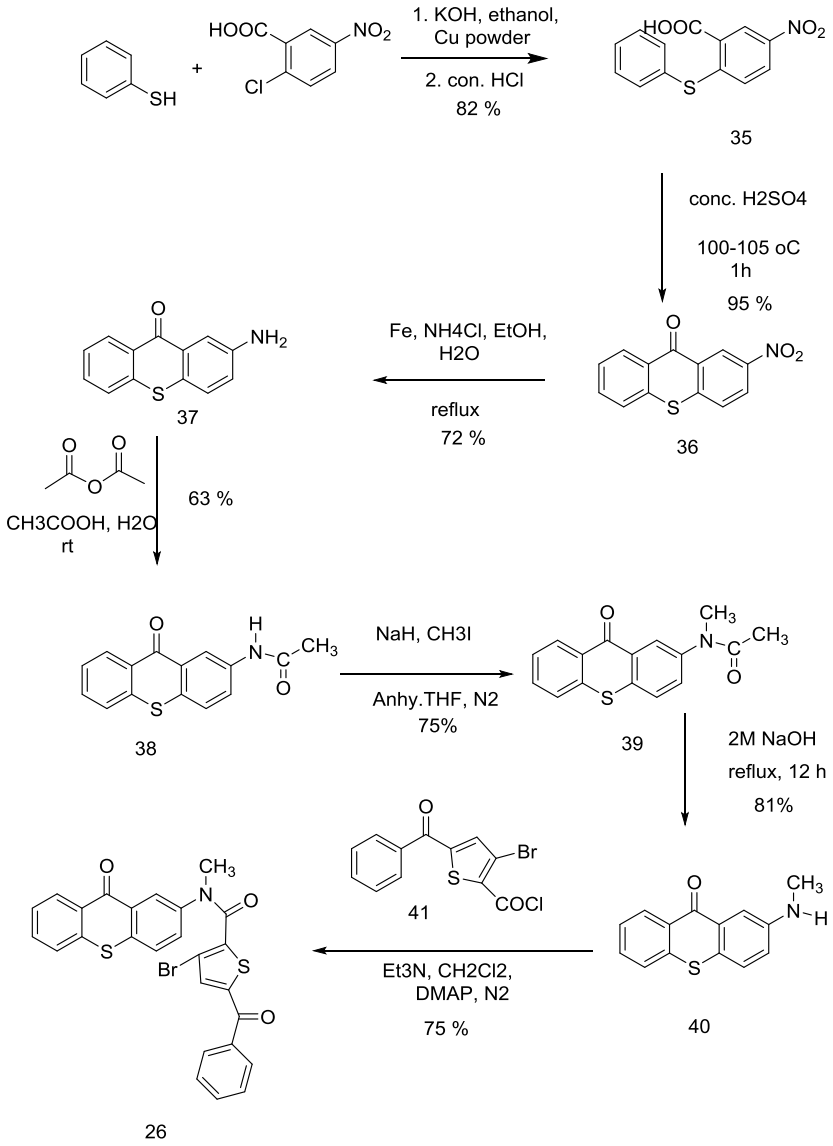
2.2. Results

2.2.1. Synthesis of Photochemical Reactant (**26**)

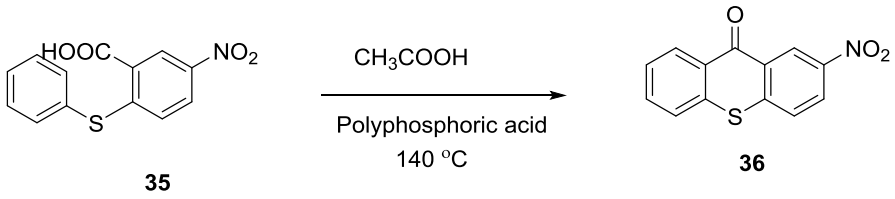
The photoreactant 5-benzoyl-3-bromothophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide **26** was synthesized by reacting the acid chloride **41** with 2-methylaminothioxanthen-9-one **40** (Scheme 2.1). The synthesis of **40** involves 6 steps starting with the reaction of thiophenol and **35** to obtain the nitro compound **36**. Initially cyclization step was carried out using polyphosphoric acid as reported in literature.[31] (Scheme 2.2). Problems associated with this route are the difficulty in scale up and seperation problems when doing workup. Compared to cyclization with polyphosphoric acid, the H_2SO_4 method has following advantages, such as less time consuming, high yield, and ability of scaling up the reaction. The nitro compound **36** was reduced to the amine **37** with iron powder.[31] Initially we use magnetic stirrer for the reaction. So we couldn't observe complete reduction because most of the ion powder was stick to stirrer bar and it prevented stirring. By using a mechanical stirrer we could see complete conversion of the nitro compound to amine. Then the amine **37** was converted to amide **38** by reaction with acetic anhydride. Then the amide **38** was alkylated using NaH and CH_3I to obtain **39**.[26] Base hydrolysis of the N-methyl acetamide **39**, furnished the 2-methylaminothioxanthen-9-one **40**. Synthesis of acid chloride **41** involved 6 steps starting from commercially available 4-bromothiophene carboxyaldehyde (Scheme 2.3).

The Grignard reaction of aldehyde with phenyl magnesium bromide formed secondary alcohol **42** which was converted to ketone **43** by oxidation with Jone's reagent. The ketone group was protected by making the acetal **44** before α lithiation. Then the α lithiated compound was treated with dry ice to obtain caboxylic acid **45.** Deprotection of the acetal with glacial acetic acid gave the carboxylic acid **46** which was converted to compound **41** by refluxing with $SOCl_2$

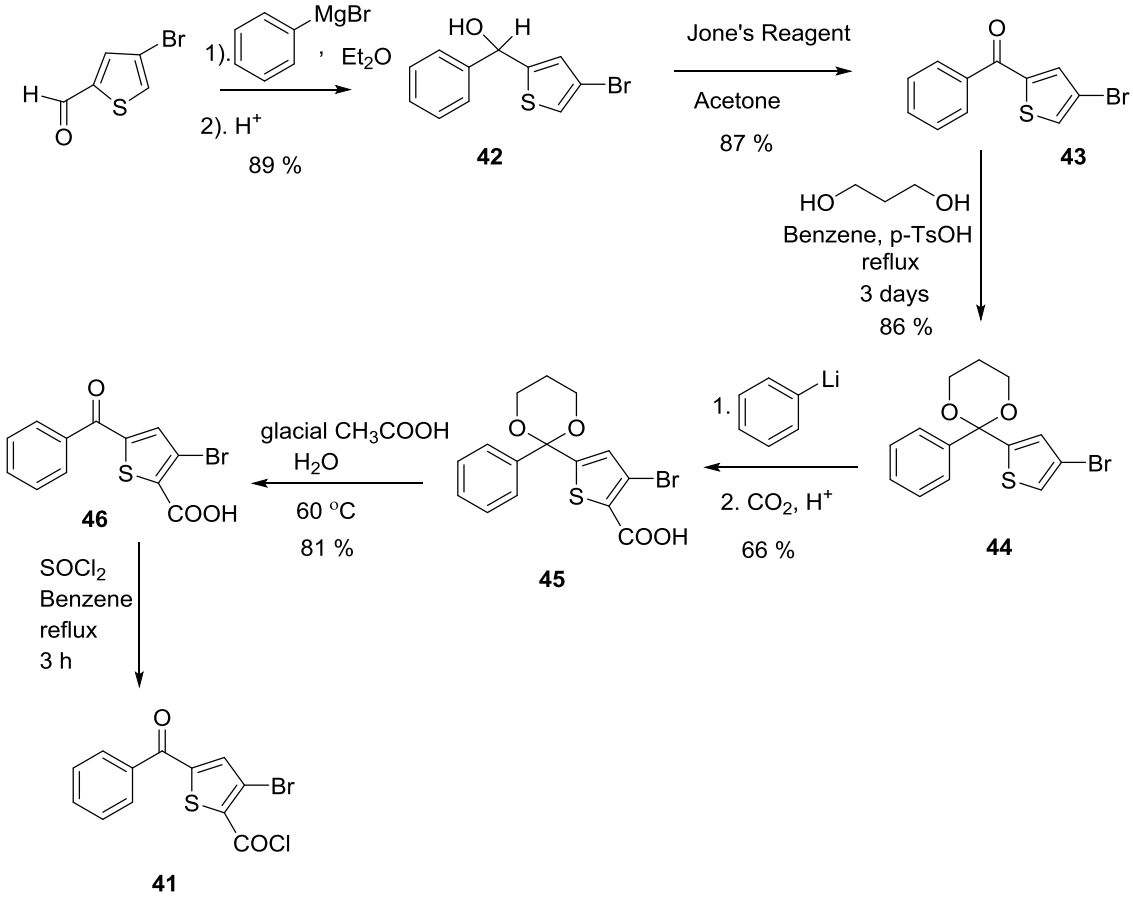
Scheme 2.1



Scheme 2.2



Scheme 2.3



2.2.2. Crystal Structure of Photochemical Reactant (26)

According to the X-ray crystallographic analysis by Oxford Supernova diffractometer using Cu(Kα) radiation, the compound **26** exhibits folded shape with amide group I a *cis*-configuration. 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.

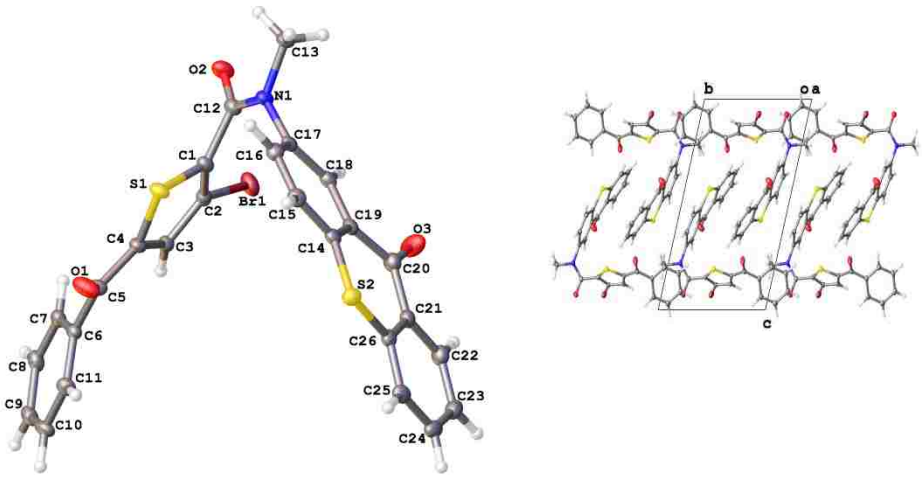


Figure 2.1. Crystal structure of photoreactant **26**

2.2.3. UV spectra for the Photoreactant 26

The compound **26** exhibits absorption maxima at 388 nm in aqueous acetonitrile (Figure 2.2) with $\varepsilon = 6270\ M^{-1}\ cm^{-1}$.

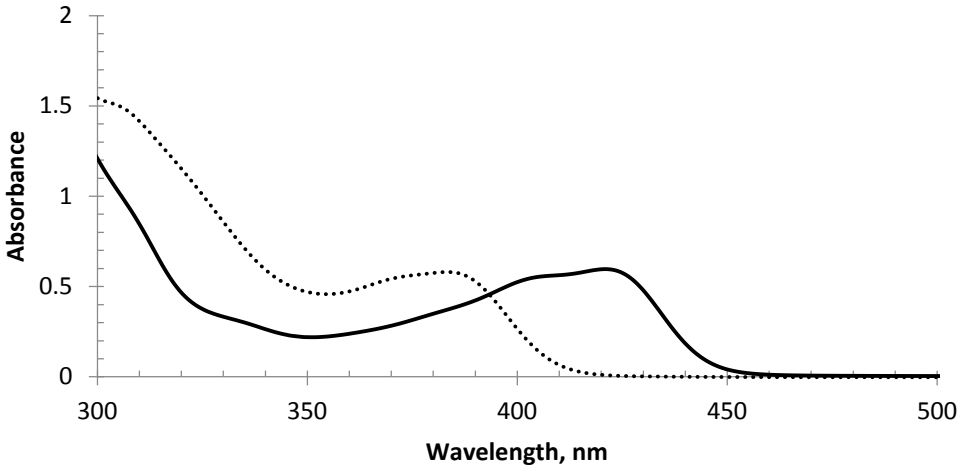
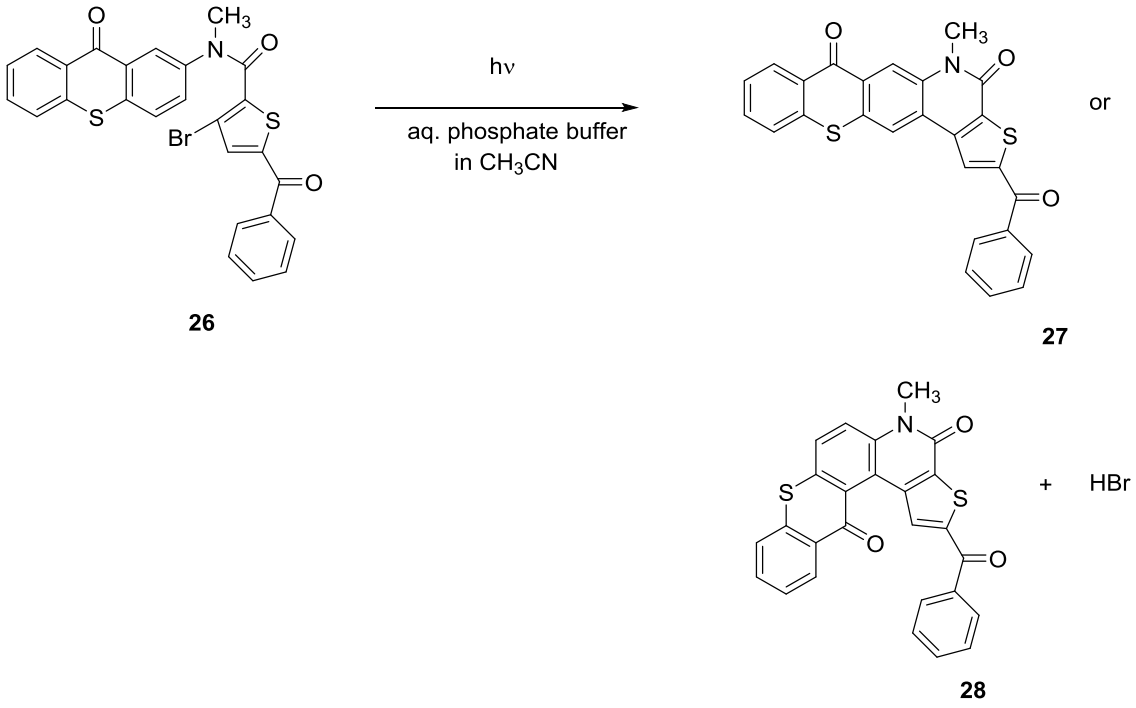


Figure 2.2. Absorption spectra of 1.0 x 10^{-4} M of compound **26** (.....) and 1.0 x 10^{-4} photoproduct produced from **26** (——) in 10 % aq. phosphate buffer (pH = 7) in CH_3CN.

2.2.3. 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 **26** 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 (Scheme 2.3). However, the cyclization and expulsion of leaving group from compound **2**6 was very slow. After photolysis for three days solid product formed was filtered washed with water and dried to obtained 30 mg of photoproduct. The product was identified and distinguished from photoreactant by ^{1}H 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.

Scheme 2.4



26 **27** **28**

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 showed a long wave length maximum at 410 nm with $\varepsilon = 6990\ M^{-1}\ cm^{-1}$ in aqueous acetonitrile containing 10%, pH 7 phosphate buffer (Figure 2.2).

2.2.4. Quantum Yield

The quantum yield for the electrocyclic ring closure reaction of amide **26** 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 6.3×10^{-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 ^1H

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.

2.3. 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 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 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 **26** (Scheme 2.3). In structure **26** 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.[29] 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 **26** 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, **17**, the reactant absorbed at 385 nm, while the photoproduct absorbed at 432 nm. In the case of **26** 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. Another photolysis for a quantum yield is planned. 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. This is planned for a future experiment.

Although the quantum yield may be low due to the aforementioned experimental flaws, it is thought that more likely the compound **26** is inherently and unexpectedly less reactive than the benzothiophene system **17**. 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 **26**. 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 **26** 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.

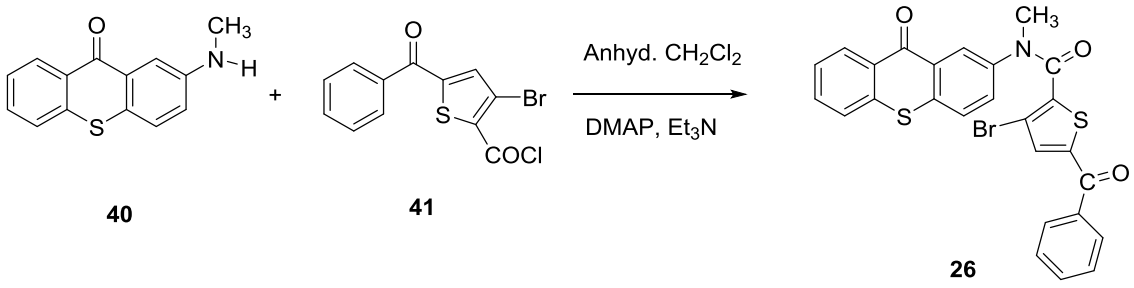
In the meantime, it was urgent to ascertain whether another choice would be more appropriate as a replacement for the benzothiophene ring system. Efforts therefore focused upon testing a naphthothiophene ring system in place of benzothiophene. In the naphthothiophene the triplet excited state is estimated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

2.4. 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[32].

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

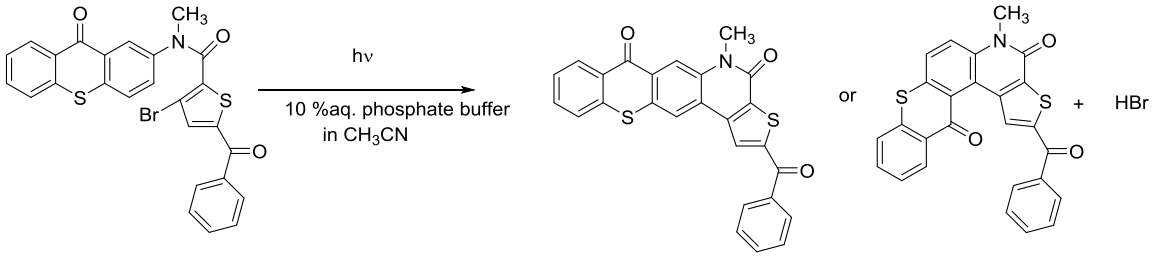


40 **41** **26**

To a solution of 1.2 g (5.0 mmol) of 2-methylaminoxanthen-9-one **40** 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 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, H_2O, with 2 M HCl, H_2O, 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 **26**. Crystallization of the solid material with ethanol gave 2.00 g (74.9 %) of yellow coloured 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), 7.55 (t, *J* = 5.2, 2H), 7.64 (dt, *J* = 8.0, 1.75, 1H), 7.71 (s, 1H), 7.74 (s,1H), 8.52 (d, *J* = 2.5, 1H), 8.60 (d, *J* = 8.3, 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, 141.0, 144.1, 162.4, 179.35, 186.9.

Photolysis of compound (**26**)

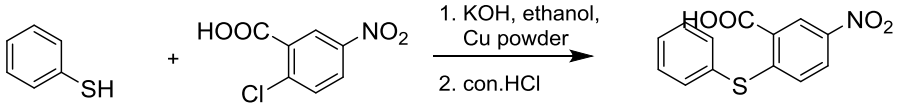


26 **27** **28**

A solution of 50 mL of 6.3 x 10^{-3} M **26** 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 **27** or **28** 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), 7.53 (d, *J* = 7.2, 1H), 7.64-7.70 (m, 6H), 7.75 (t, *J* = 9.3, 1H), 8.08 (t, *J* = 9.5 Hz, 1H), 8.10 (s,1H), 8.13 (d, *J* = 8.1, 1H), 8.28 (d, *J* = 8.1, 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, 138.8, 139.0, 145.7, 157.9, 183.4, 188.5.

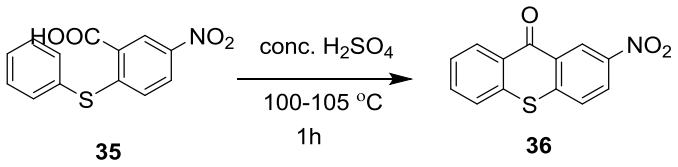
Preparation of 4-Nitrophenyl Sulphide-2-carboxylic acid (**35**)[33]



35

To a solution of 21.4 g (105 mmol) of 5-nitro-2-chlorobenzoic acid in 300 mL absolute ethanol was added 12.4 g (119 mmol) of thiophenol and 15.7 g (280 mmol) of potasium hydroxide dissolved in 300 mL ethanol while stirring. Then trace amount of copper powder was added and refluxed overnight under N_2. After two third 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 was washed with water. The crude product was recrystallized with 80% aqueous ethanol to obtain 24.1 g (82% yield) of compound **35** as dark yellow crystals, mp. 233-234 °C. The spectral data were as follows: ^1H 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, 3.41 Hz), 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, 130.4, 131.4, 131.0, 136.2.

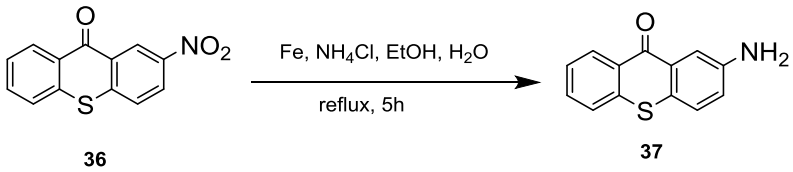
Preparation of 2-nitroxanthone (**36**)[33]



35 **36**

To 185 mL of concentrated sulphuric acid at 100 ^{o}C was added 21.2 g (77.0 mmol) of 4-nitrophenylsulphide-2-carboxylic acid **35**. The temperature of the mixture was maintained at 100-105 °C for one hour. 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 drying in air, gave 18.9 g (95% yield) of **36** was obtained as NMR pure yellow-green solid, mp. 225-228 ^{o}C. This compound was used in the next step without further purification. The spectral data were as follows: ^{1}HNMR (400 MHz, DMSO-d_6) δ 7.65 (d, *J* = 7.6 Hz, 1H), 7.84 (t, *J* = 7.4, 1H), 7.91 (d, *J* = 8.0 Hz, 1H), 8.12 (d, *J* = 9.5 Hz, 1H), 8.42 -8.51 (m, 2H), 9.05 (d, *J* = 2.4 Hz, 1H).

Preparation of 2-Amino-thioxanthen-9-one (**37**)

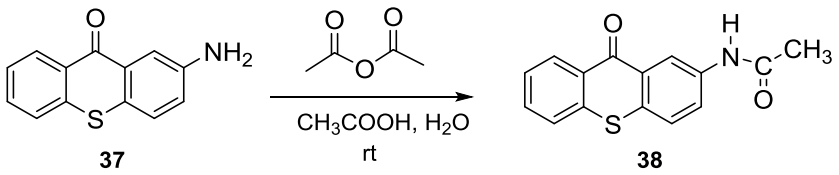


36 **37**

The procedure was adapted from a procedure reported by Steinmetz[26] and Moon.[31] A mixture of 18.8 g (73.4 mmol) of 2-nitroxanthone **36**, 800 mL of ethanol, 200 mL of water, ammonium chloride 23.6 g (440 mmol), and iron 16.4 g (294 mmol) was refluxed 5 h while mechanically stirring . After hot vacuum filtration through silica gel, the silica gel was washed with ethanol and combined with the filtrate. The combined filtrate was concentrated *in vacuo*. The product was extracted into $CHCl_3$. The $CHCl_3$ solution was

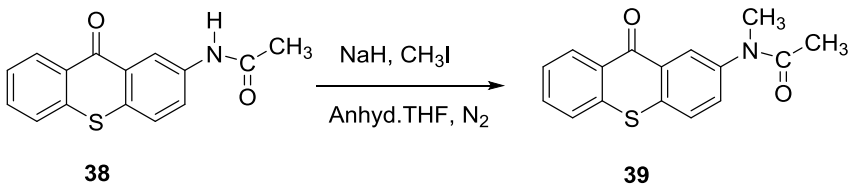
dried over anhydrous sodium sulphate and concentrated *in vacuo* to give 12.0 g (72% yield) of **37** as a dark yellow powder, mp. 226-228 ^{o}C. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) δ 5.66 (br, 2H), 7.06 (d, *J*=8.5, 1H), 7.45-7.51 (m, 2H), 7.61-7.77 (3H, m), 8.41 (d, *J* = 8.5 Hz, 1H).

Preparation of N-(9-oxo-9H-thio-xanthen-2-yl)-acetamide (**38**)



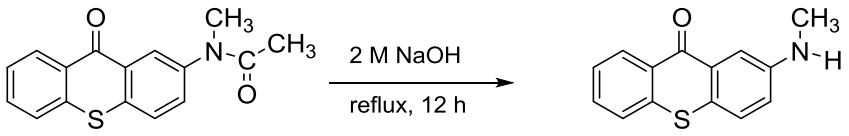
The procedure was adapted from a procedure reported by Steinmetz.[26] A mixture of 11.0 g (48.2 mmol) of amino ketone **37**, 200 mL glacial acetic acid, 81.1mL (859 mmol) of acetic anhydride was stirred for 5 h at room temperature. After adding 300 g of ice water with stirring, the resultant precipitate 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 **38** as a light yellow powder, mp. 241-242 ^{o}C. The spectral data were as follows: ^{1}H 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* = 7.9 Hz, 1H), 8.71 (s, 1H), 10.35 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 24.6, 118.5, 125.1, 127.2, 127.7, 128.5, 129.4, 129.7, 130.8, 133.5, 133.5, 137.3, 138.9, 148.5, 169.4, 179.3.

Preparation of N-methyl-N-(9-oxo-9H-thioxanthen-2-yl)-acetamide (**39**)[26]



The procedure was adapted from a procedure reported by Steinmetz.[26] To a stirred solution of 9.50 g (35.3 mmol) of N-(9-oxo-9H-thio-xanthen-2-yl)-acetamide **38** 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 7.56 g (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. The residue was added $CHCl_3$, followed by filtration and concentration *in vacuo* to obtain 7.5 g (75% yield) of methyl amide **39** as a yellow powder, mp 246-248 °C. 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* = 8.5 Hz, 1H).

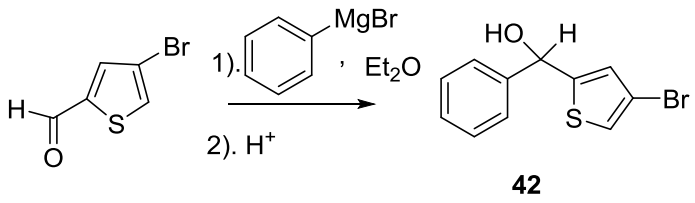
Preparation of 2-methylaminothioxanthen-9-one (**40**)



39 **40**

A mixture of 7.00 g (24.7 mmol) of amide **39** and 250 mL of aqueous 2 M NaOH was refluxed for 12 h. The reaction mixture was cooled to room temperature and solid material was filtered, washed with water and dried to obtain 4.8 g (81 % yield) of **40** as a yellow colour powder, mp. 173-174 °C. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.95 (s, 3H), 3.98 (br, 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 30.5, 109.2, 119.9, 124.8, 15.6, 125.9, 126.9, 128.7, 129.7, 130.2, 131.8, 137.7, 147.9, 180.1.

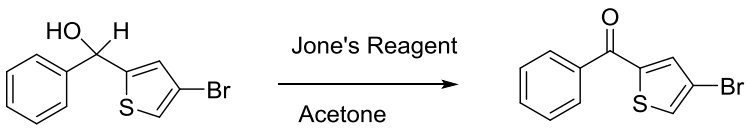
Preparation of 2-thiophenemethanol, 4-bromo-α-phenyl (**41**)



42

The procedure was adapted from a procedure reported by Alexander.[34] 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 **42** 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* = 1.46 Hz, 1H), 7.14-7.46 (m, 5H); ^{13}C NMR (100 MHz, $CDCl_3$): δ 72.3, 109.2, 122.4, 126.3, 127.2, 128.2, 128.7, 142.1, 149.3.

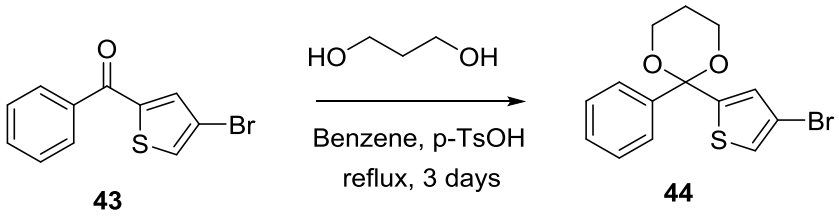
Preparation of 2-benzoyl- 4-bromothiophene (**43**)



42 **43**

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 2-thiophenemethanol, 4-bromo-α-phenyl **42** 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, 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 **43** as an 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); ^{13}C NMR (75 MHz, $CDCl_3$) δ 110.8, 128.9, 129.4, 131.6, 133.0, 136.7, 137.4, 144.2, 187.9.

Preparation of 1,3-Dioxane, 2-(3-bromothienyl)-2-phenyl (**44**)

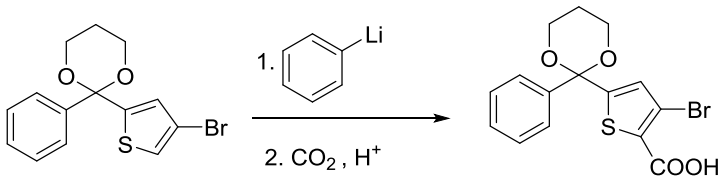


43 **44**

The procedure was adapted from the procedure reported by Angibaud.[35] To a solution of 21.0 g (78.6 mmol) of 2-benzoyl-4-bromothiophene **43** in 120 mL of anhydrous benzene was added 14.4 g (188 mmol) of 1,3-propanediol and catalytic amount of *p*-toluene sulphonic 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 colourless oil. Upon standing for two days it gave 21.9 g (86 % yield) of acetal **44** as colourless crystals, mp. 61-63 ^oC. The spectral data were as follows: ^1H NMR (400 MHz, $CDCl_3$) δ 1.63 (m , 1H), 1.91 (m, 1H), 4.02 (m ,4H), 6.75 (s, 1H), 7.12 (s, 1H), 7.27-7.43 (m, 3H), 7.59 (t, *J* = 7.5 Hz, 2H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 25.3, 62.1, 99.4, 108.9, 123.5, 126.8, 128.4, 128.7, 128.9, 140.5, 149.1.

Preparation of (**45**)

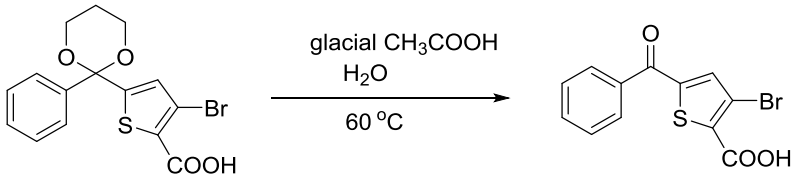


44 **45**

Procedure was adapted from the procedure reported by Katritzky.[36] 12.0 g (36.8 mmol) of **44** 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 **45** 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 25.3, 62.0, 99.0, 117.2, 126.6, 127.5, 129.1, 130.9, 139.8, 154.4, 166.4.

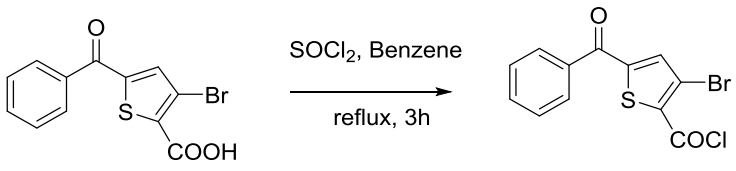
Preparation of 5-benzoyl-3-bromo-2-thiophenecaboxylic acid (**46**)



45 **46**

Procedure was adapted from procedure reported by Babler.[37] To 10.0 g (28.4 mmol) of **45** was added 100 mL of glacial acetic acid and 25 mL of water. The reaction mixture was heated at 65 °C 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 colour solid. Recrystallization of brown colour solid with aqueous ethanol formed 7.16 g (81% yield) of **46** as off white colour crystals, mp176-178°C. The spectral data were as follows: ^{1}H 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); ^{13}C NMR (100 MHz, $CDCl_3$) δ 115.8, 129.7, 133.9, 135.4, 136.4, 138.7, 145.0, 161.7, 186.9.

Preparation of 5-benzoyl-3-bromo-thiophenecarbonyl chloride (**41**)



46 **41**

To a solution of 4.00 g (12.9 mmol) of **46** 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 **41**and it was used to next step without further purification.

General Procedure for Product Quantum Yield Determination

A semi-micro optical bench was used for quantum yield determinations, similar to the method described by Zimmerman.[32] Light from 200 W high pressure mercury lamp was set to 388 nm wavelength and was collimated through a lens. A fraction of the light was diverted 90° by a beam splitter to a 10 x 3.6 cm side quartz cell containing 41 mL of an actinometry solution. The photolysate was contained in 27 mL volume quartz cylindrical cell with 10 x 1.8 cm dimensions. Behind the cell containing photolysate was mounted a quartz cylindrical cell 10 x 1.8 cm containing 27 mL of actinometry solution. Light output was measured by the ferrioxalate actinometry using the splitting ratio method.

For compound **26**, 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%.

CHAPTER 3. Photochemical Electrocyclic Ring Closure and Leaving Group Expulsion from 3-chloronaptho[1,2-b]thiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide

3.1. Introduction

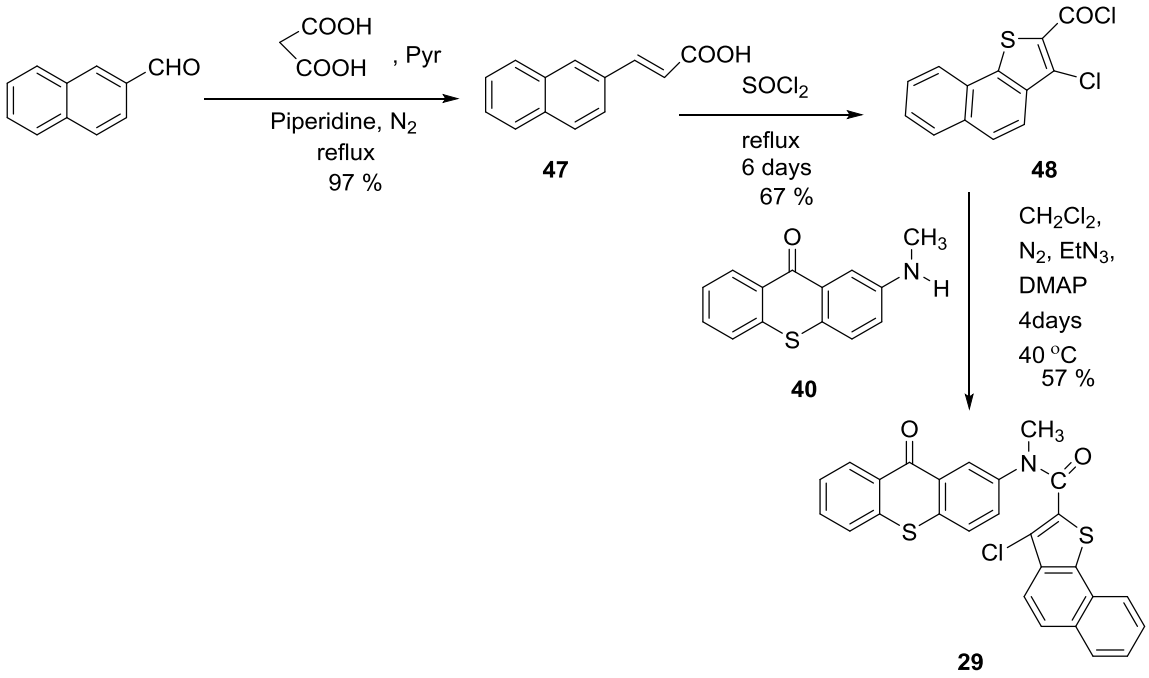
As mentioned in the Chapter 2, naphtho[1,2-b]thiophene[30] has triplet excited state with energies of 62 kcal mol^{-1} (Scheme 1.13). This triplet is lower in energy than benzothiophene triplet (64 kcal mol^{-1}). It was planned to replace the benzothiophene moiety with napthothiophene ring system in order to facilitate the energy transfer from the triplet thioxanthone.

3.2. Results and Discussion

3.2.1. Synthesis of Photochemical Reactant (**29**)

The synthesis of photoreactant **29** (Scheme 3.1) involved a coupling reaction between acid chloride **48** and amine **40**. Reaction of commercially available napthalene-2-carboxaldehyde with malonic acid gave 3-(2-napthyl)propenoic acid[38] **47**, which was converted to 3-chloronaptho[1,2-b]thiophene-2-carbonyl chloride **48** by refluxing for six days with $SOCl_2$.[39]

Scheme 3.1

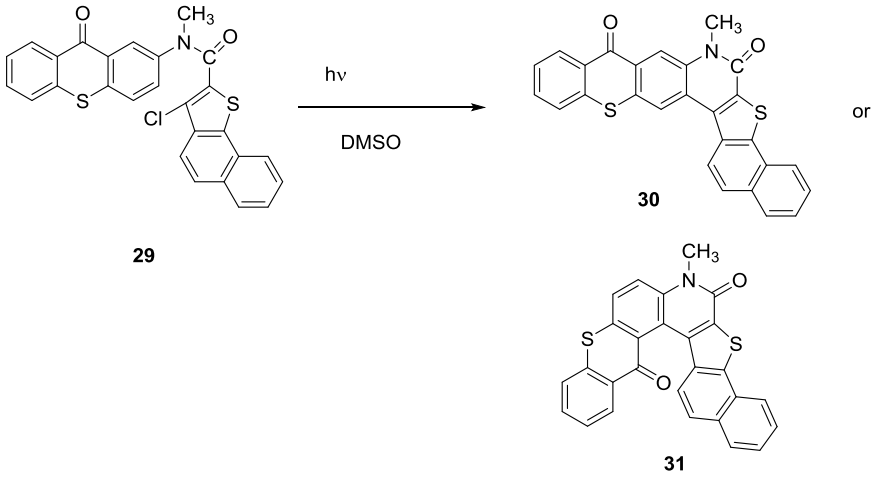


3.2.2. Photolysis of Compound 29

One criteria[3, 5] for the designing of biological important photoremovable protecting groups is the solubility in aqueous buffered medium. Compound **29** is insoluble in aqueous buffered media. We made attempt to dissolve this compound in CH_3CN. It was unsuccessful, but it was sparingly soluble in DMSO. Attempts were made to dissolve compound **29** in DMSO-d_6 and then filter 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. So it was difficult to photolyse **29** compound in aqueous buffered media due to the solubility problem. Photolysis of a nitrogen saturated sample of 20 mg in 2 mL DMSO-d_6 , filtered through a syringe filter using Pyrex-filtered light from a Hanovia 450 W medium pressure mercury lamp, without added buffer, showed a formation of single photoproduct of **30** or **31** (Scheme

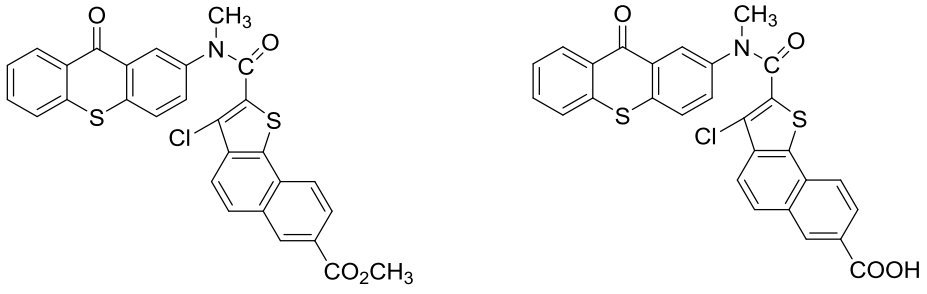
3.2) after 30 min. After 1.5 h it showed about 50% conversion. The product was identified and distinguished from photoreactant by ^{1}H NMR as N-methyl peak shifted downfield from δ 3.64 to 3.98 ppm. Due to the above solubility problem, the quantum yield was not determined. But the high conversion of reactant to photoproduct in DMSO-d_6 provided the incentive to use this system with modifications to increase the solubility in aqueous buffered media.

Scheme 3.2



29 **30** **31**

Attention thus was focused upon compounds **32** and **33** Scheme (3.3) by attaching an ester group and carboxylic acid at the C-6 position of the naphthalene moiety to increase the solubility in aqueous buffered media.

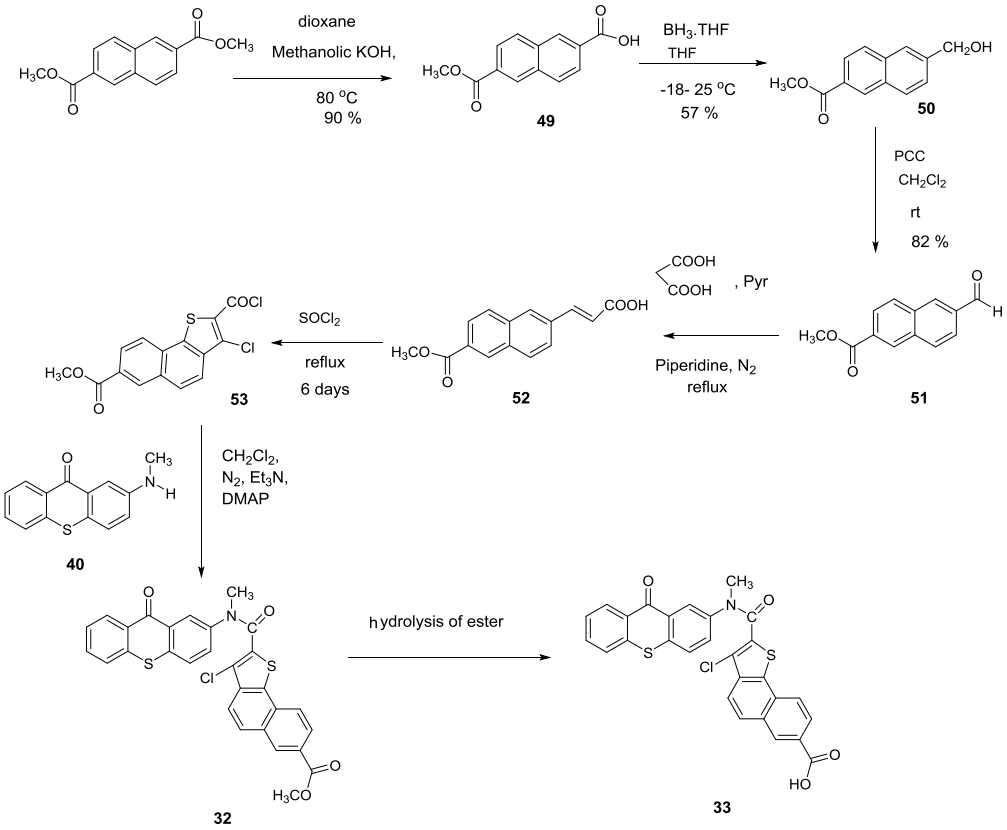


32 **33**

3.3. Synthesis of Photoreactant 32 and 33

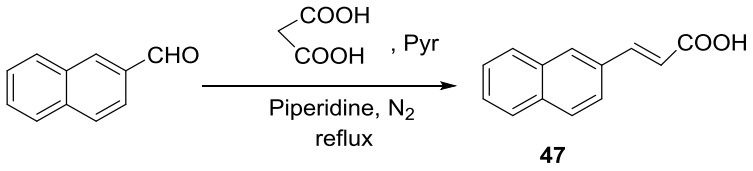
To improve aqueous solubility, a derivative with Y = -$COCH_3$ and -COOH (**32**, **33**) are being synthesized. The plan for the synthesis of **32** and **33** is given in the Scheme 3.3. The compound **32** will be synthesized by a coupling reaction between amine **40** (synthetic routes given in Scheme 2.1) and acid chloride **53**. Synthesis of **53** involves 5 steps starting commercially available dimethyl 2,6-naphthalenedicarboxylate. It was found that dimethyl 2'6-naphthalenedicarboxylate would be partially hydrolyzed with methanolic KOH to form 6-cabomethoxy-2-napthalenecarboxylic acid **49**. The carboxylic acid group of **49** was reduced to the alcohol **50** using BH_3.THF complex followed by PCC oxidation to form aldehyde **51**. This part of the synthesis was successful**.** The remaining steps to obtain **53** should be routine. It is therefore expected that **32** and **33** will be obtained in the very near future.

Scheme 3.3



3.2.4. Experimental

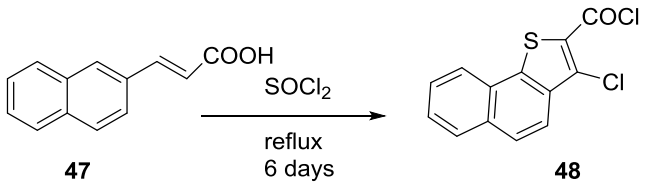
Preparation of 3-(2-naphthyl)propenoic acid (47)[38]



To a solution of 50 mL pyridine (621 mmol) 11.4 g of malonic acid (110 mmol) was added. 14.21 g of naphthalene-2-carboxaldehyde (91 mmol) in little increments. Then 1 mL (101 mmol) of piperdine was added at room temperature. The reaction mixture was heated to reflux 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 form a

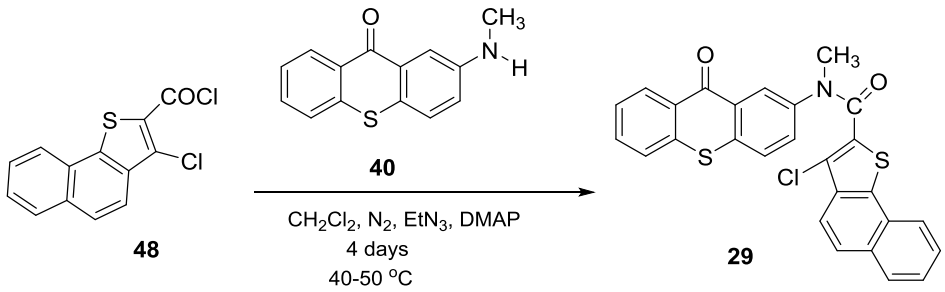
precipitate. The precipitate was filtered, washed with water and dried. The solid was recrystallized from ethanol to obtained 17.5 g (97% yield) of product **47** as an off white solid, mp. 206-208 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.64 (d, *J* = 16.1, 1H), 7.55-7.59 (m, 2H) 7.72 (d, *J* = 16.1 Hz, 1H), 7.86-7.96 (m, 4H), 8.19 (s, 1H), 12.42(s, 1H).

Preparation of 3-chloronaphthol[1,2-b]thiophene-2-carbonyl chloride (**48**)[39]



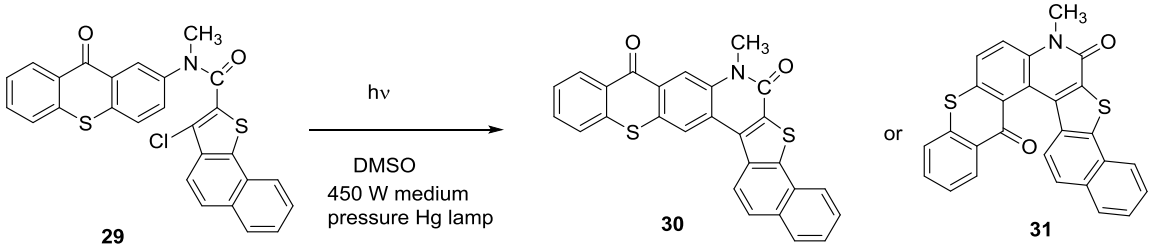
A mixture of 17.5 g (88 mmol) of **47**, 100 mL of chlorobenzene, 1.6 mL (19.8 mmol) of pyridine and 36.4 mL (500 mmol) of thionyl chloride was refluxed for 72 h. After cooling to room temperature and suction filtration gave 16.5 g (67% yield) of product **48** as yellow needles, mp. 191-193 ^{o}C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.73 (m, 2H), 7.90 (dd, *J* = 8.7 Hz, 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, 126.8, 127.0, 128.4, 28.0, 129.7, 132.3, 134.8, 137.2, 162.2.

Preparation of 3-chloronaphthol[1,2-b]thiophene-2-carboxylic acid N-methyl-(9-oxo-9H-thioxanthen-2-yl) amide (**29**)



The procedure was adapted from the procedure reported by Steinmetz.[26] 1.2 g (5.0 mmol) of 2-methylaminoxanthen-9-one **40** 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 **48** dissolved in 10 mL of anhydrous CH_2Cl_2 at room temperature. A catalytic amount of DMAP was added. Then reaction mixture was heated at temperature between 40-50 ^oC for 96 h under nitrogen while stirring. The reaction mixture was filtered to remove trimethylamine hydrochloride, washed several times with saturated aqueous $NaHCO_3$ solution, H_2O and then with 2 M HCl, H_2O and brine. Then dried over anhydrous Na_2SO_4 and concentrated *in vacuo* to obtain a golden yellow solid containing (**29**). Recrystallization of solid material from ethanol gave 1.36 g (57% yield) of product **29** 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), 7.3-7.59 (m, 7H), 7.62 (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 (d , J = 8.5 Hz, 2H); ^{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.

Photolysis of compound 29 to form 30 or 31

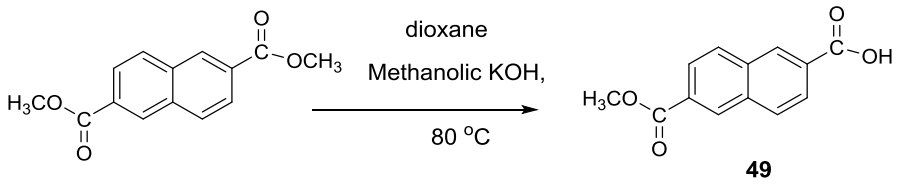


29 **30** **31**

To 50 mL of DMSO 150 mg of compound **29** was added and stirred to dissolve compound. As this compound is slightly soluble in DMSO before photolysis particles

were filtered through syringe filter. Then the clear solution was flushed with N_2 for 30 min and photolyzed by 450 W medium pressure Hg lamp with Pyrex filter. After 4 h the compound which was precipitated inside the reaction tube was filtered and dried under *vacuo* to obtain 85 mg of a yellow colour product of **30** or **31** mp. > 300 ^{o}C. ^{1}HNMR (400 MHz, $CDCl_3$): 3.98 (s, 3H), 7.60-7.75 (m, 7H), 7.83(d, *J* = 9.1 Hz, 1H), 7.94 (d, *J* = 9.6 Hz, 1H), 8.30 (d, *J* = 8.7 Hz, 1H), 8.35 (d, *J* = 7.7 Hz, 1H).

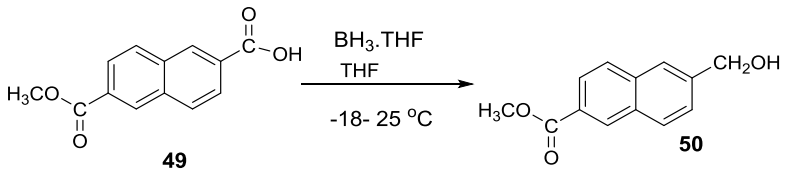
Preparation of 6-cabomethoxy-2-naphthalenecarboxylic acid (**49**)[40]



49

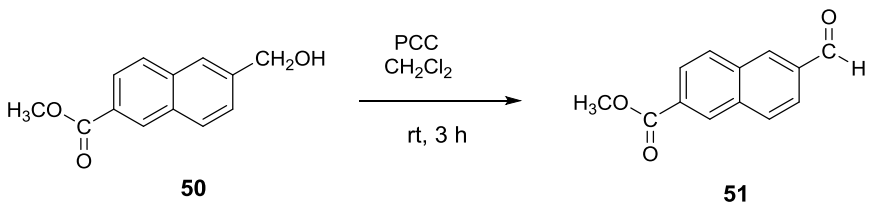
A suspension of 10.0 g (40.9 mmol) of commercially available dimethyl 2,6-naphthalenedicarboxylate in 60 mL dioxane was heated at 80 ^{o}C until all solid dissolved. The solution of 2.6 g (42 mmol) in 2 mL MeOH was slowly added and stirred for 2 h at 80 ^{o}C. The reaction mixture was cooled to room temperature, filtered and solid residue was washed with diethyl ether. The solid was dissolved in water and treated with 2 M HCl to pH =3. The resultant precipitate was filtered, washed with water, and dried to obtain 8.5 g (90% yield) of **49** as a white powder, mp. 248-251^{o}C. The spectral data were as follows: ^{1}H NMR (400 MHz, DMSO-d_6) 3.91 (s, 3H), 8.04 (d, J = 8.6 Hz, 2H), 8.22 (d, J = 8.6 Hz, 2H), 8.66 (s, 1H), 8.68 (s, 1H), 13.25 (br, 1H).

Preparation of 6-hydroxymethyl-naphthalene-2-carboxylic acid methyl ester (**50**)



To a suspension of 1.5 g (6.5 mmol) of **49** in 30 mL of anhydrous THF at -15 ^{o}C was added 13 mL of BH_3.THF complex slowly. The reaction mixture was allowed to become room temperature while stirring under N_2 and further stirred for overnight. The saturated $NaHCO_3$ was added and extracted into ethyl acetate. The ethyl acetate layer was washed several times with water, brine, dried over anhydrous Na_2SO_4, and evaporated under *vacuo* to give 1.1 g of crude product as a white colour powder. The mixture was purified on a silica gel column eluted with ethyl acetate/hexane (50%) to obtain 0.8 g (57 % yield) of pure product **50**, mp. 125-127 ^{o}C. The spectral data were as follows: ^{1}HNMR (400 MHz, $CDCl_3$) 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 6-formyl-naphthalene-2-carboxylic acid methyl ester (**51**)



To a mixture of 3.51 g (16.3 mmol) of PCC in 60 mL CH_2Cl_2 was added a solution of 1.6 g (7.4 mmol) of compound **50** in 10 mL of CH_2Cl_2 and stirred for 3h at room temperature. The solution was decanted into another flask and remaining solid material was washed with CH_2Cl_2 several times and combined to the liquid. The combined liquid was filtered through Florisil until the orange colour of the solution disappeared. The

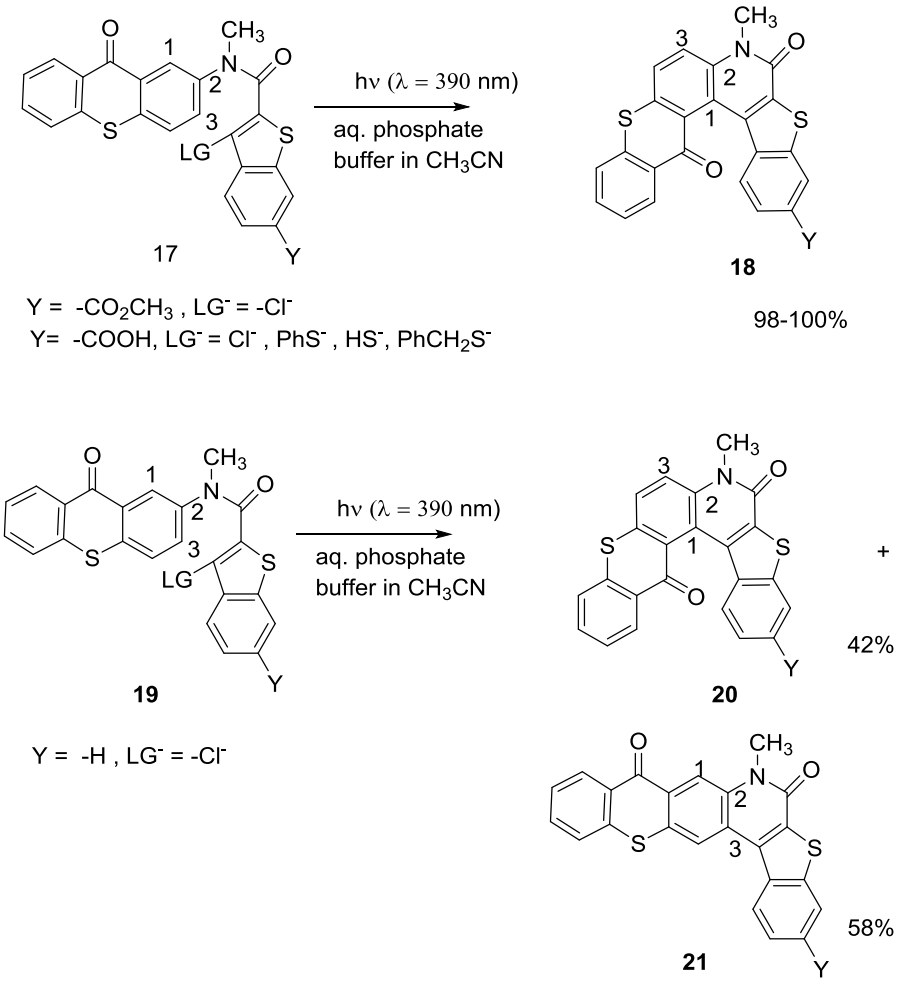
filtrate was evaporated under *vacuo* to obtain 1.3 g (82 % yield) of crude product. ^{1}H NMR gave a peak around 10.18 which corresponds to the aldehyde. Further purification is needed to collect pure product.

CHAPTER 4. Photochemical Electrocyclic Ring Closure and Leaving Group Expulsion from N-(9-oxothioxanthenyl)benzothiophene carboxanilide

4.1. Introduction

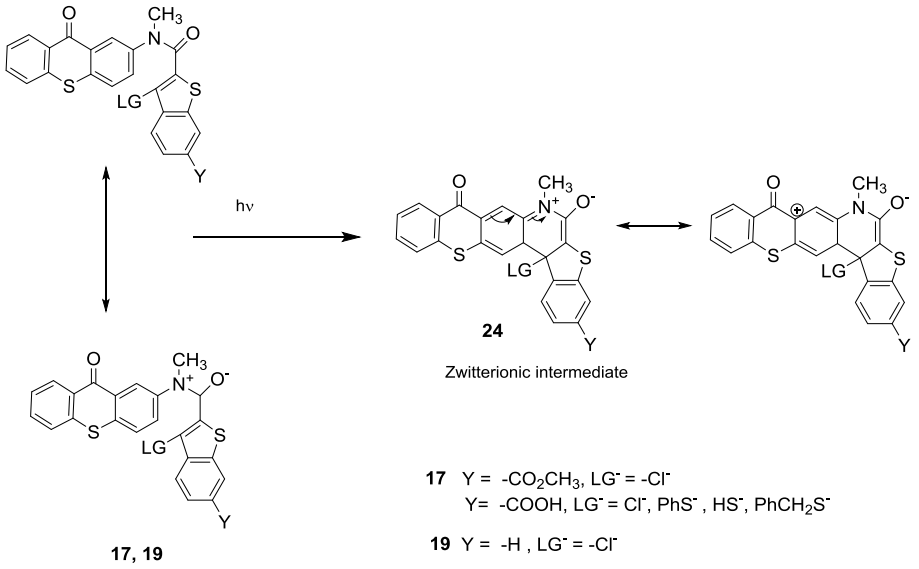
This was the first research objective at the beginning of my graduate studies. At that time our research group have been studying photochemical electrocyclic ring closure reaction and leaving group expulsion from the **17** and **19** N-(9-oxothioxanthenyl)benzothiophene compound (Scheme 4.1).

Scheme 4.1



Using this system they were able to release variety of leaving groups by photolyzing at λ = 390 nm. However, they observed that the quantum yields for those reactions were low (Table 1.2). Here they studied the molecule where the amide N is attached to the C-2 position of the thioxanthone moiety. When N is at the C-2 position of thioxanthone moiety the zwitterion **24** formed by electrocyclic ring closure reaction might be unstable (Scheme 4.2). Initially it was thought that this could be the reason for the low quantum efficiency for the above compound. Therefore, the initial plan was to synthesize N-(9-oxothioxanthenyl) benzothiophene carboxamide **25**, where the amide nitrogen is at the C-3 position of thioxanthone moiety (Scheme 4.3).

Scheme 4.2



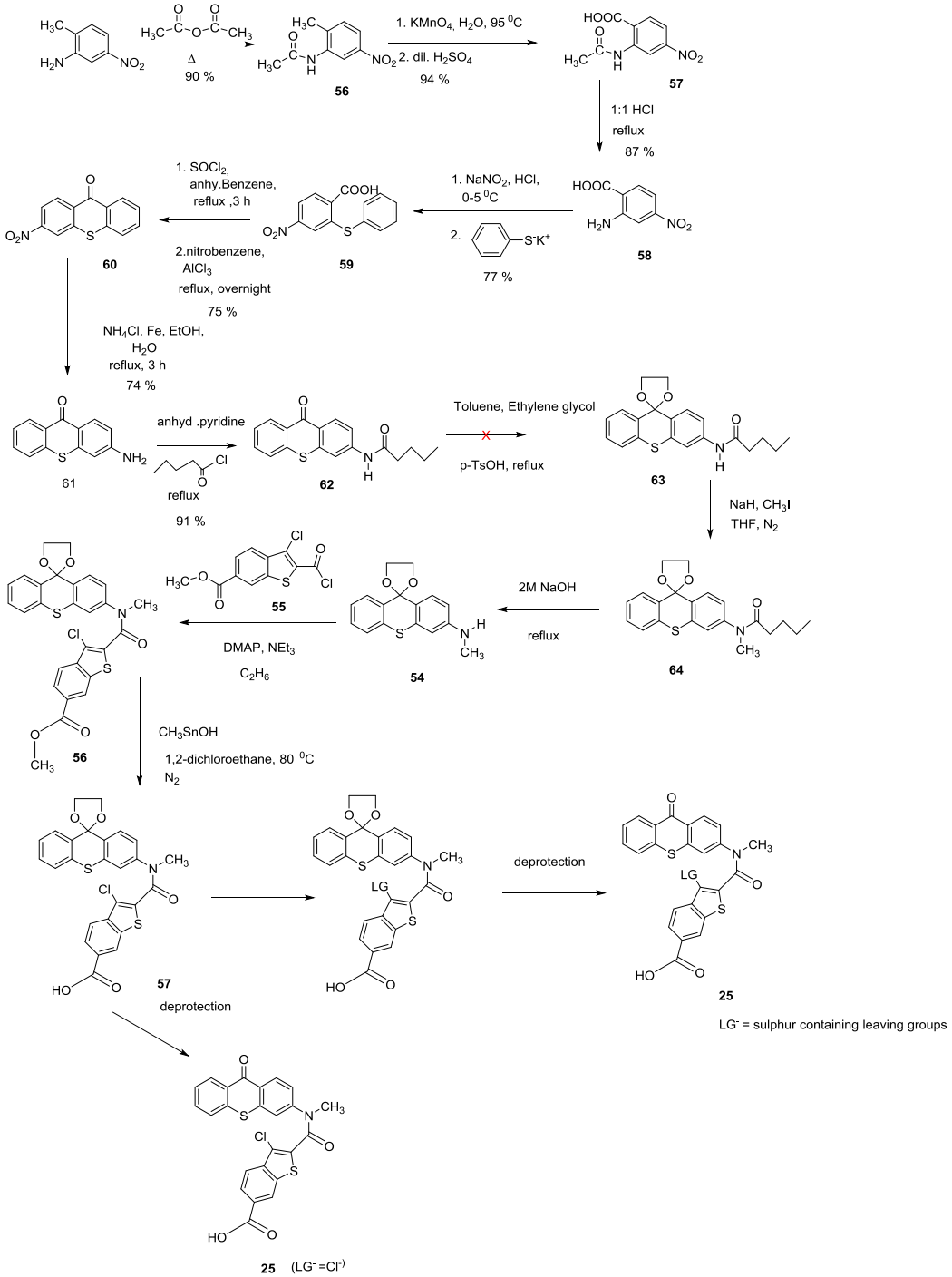
4.2. Results and Discussion

4.2.1. Synthesis of Photochemical Reactant (**25**)

The plan for the synthesis of N-(9-oxothioxanthenyl) benzothiophene (**25**)which has

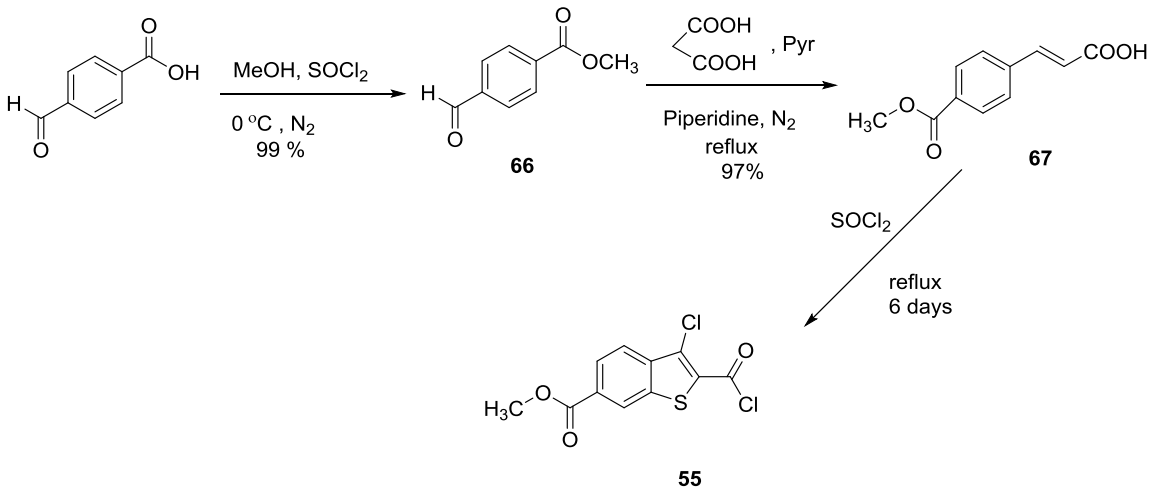
has the N-methyl amide at C-3 position of thioxanthone ring system would use amine **54** and acid chloride **55** (Scheme 4.3).

Scheme 4.3



The compound **55** (Scheme 4.4) was prepared by refluxing methyl ester of *trans*-cinnamic acid **67** with thionylchloride for 6 days.[42] Acid 67 was produced by a two steps process starting form 4-formylbenzoic acid (see Experimental).[43, 44]

Scheme 4.4



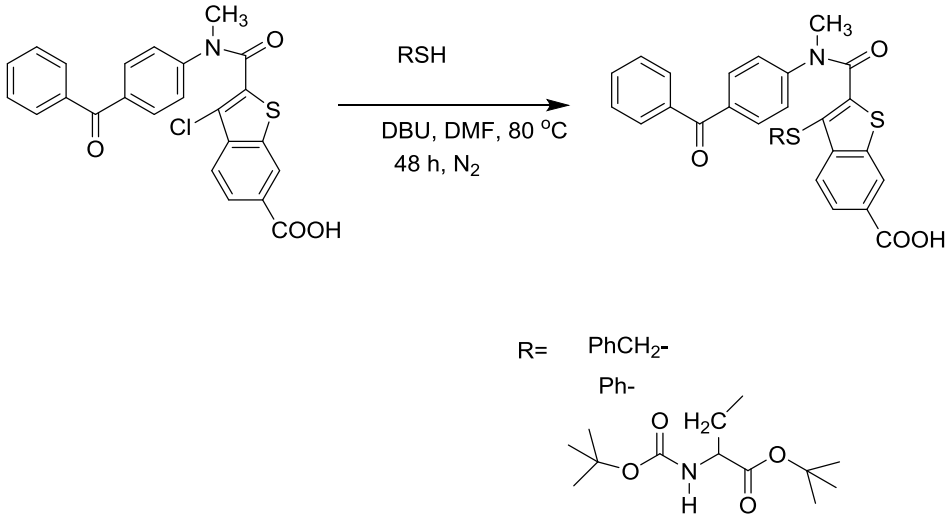
Reaction of commercially available 4-methyl-5-nitroaniline with acetic anhydride produced 2-methyl-5-nitro-acetanilide **56**, which was oxidized by $KMnO_4$ under neutral conditions to form carboxylic acid **57**.[45] Hydrolysis of compound **57** by 1:1 HCl gave 2-amino-4-nitrobenzoic acid **58**[45] which was diazotized and reacted with thiophenolate to form 4-nitro-2(phenylthio) benzoic acid **59**.[46] Initially this reaction was carried out without any catalyst and gave a low yield.[46b] By using silica gel as the catalyst the yield could be increased for that reaction. Reaction of the acid **59** with $SOCl_2$ followed by Friedel-crafts acylation reaction produced 3-nitroxanthone **60**.[46a] Reduction of the nitro compound with iron gave 3-aminoxanthone **61** which was converted to amide **62** with the reaction of pentanoyl chloride. Formation of the amide was first carried out using triethylamine in the presence of DMAP as catalyst.[26] But this reaction did not give a

good yield. In the presence of anhydrous pyridine it gave a 91% yield. In the previous study for synthesizing photoreactant which contain sulphur containing leaving groups with a benzophenone chromophore **26**[47] (Scheme 4.5) they observed that incorporation of thiolate LG^- in place of Cl^- in compound **25** encountered a problem due to the instability of amide regarding cleavage. Problem was solved by protecting the carbonyl group before coupling the acid chloride with amine. The compound here has a similar type of conjugated π electron system. Therefore, the previous method was adapted involving protection of the carbonyl group of the thioxanthone ring system proceeding with the coupling reaction. Protection of carbonyl group of compound **62** used ethylene glycol in the presence of toluene and catalytic amount of p-TsOH.[48] However, acetal formation was unsuccessful due to very low yield. Then the plan was changed to reduce carbonyl group before coupling with acid chloride and oxidize again after coupling and introducing sulphur containing leaving groups. The compound **62** was planned to reduce using ZnI_2 and $NaCNBH_3$ to obtain compound **65**. This reaction was unsuccessful.

At this point plans were changed when DFT calculations[26] (Figure 1.4) became available for N-(9-oxothioxanthenyl) benzothiophene compound **17**, **19** which contain N-methyl amide at C-2 position of thioxanthone moiety. The calculations showed that the reaction takes place via the initial triplet excited state where the excitation is localized on the thioxanthone ring system. Energy transfer then took place to give a triplet excited state of the benzothiophene moiety. Therefore, the lower reactivity of the thioxanthone compound may be due to the fact that E_T =64 kcal mol^{-1} for thioxanthone.[27] The energy transfer to give the triplet benzothiophene[28] (E_T =69 kcal mol^{-1}) is endothermic by 5 kcal mol^{-1}.[26] This unfavorable energy transfer might be account for the lower quantum

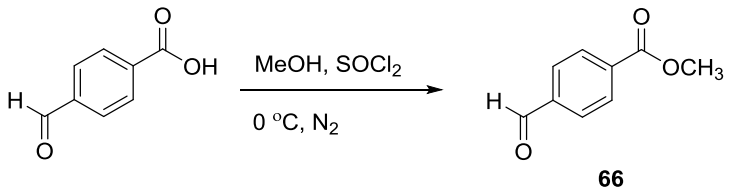
yield. At this point it was planned to modify or replace the benzothiophene ring to facilitate the energy transfer from the triplet thioxanthone.

Scheme 4.5



4.3. Experimental

Preparation of Methyl 4-formylbenzoate (66)[43]

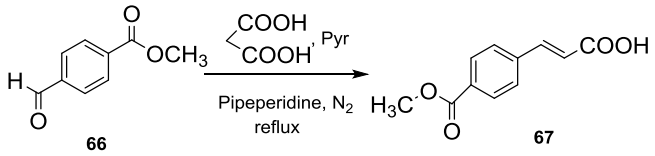


66

To commercially available 5.0 g (33 mmol) of 4-formylbenzoic acid dissolve in 75 mL of anhydrous MeOH was added 5 mL (68 mmol) of thionyl chloride dropwise at 0 °C under N_2. The reaction mixture was brought to room temperature and stirred for overnight. The solvent was removed *in vacuo*. Excess $SOCl_2$ was co-evaporated with dichloromethane (2x100 mL) to give 5.4 g (99% yield) of **58** as a pale brown solid, mp. 52-53 °C. The

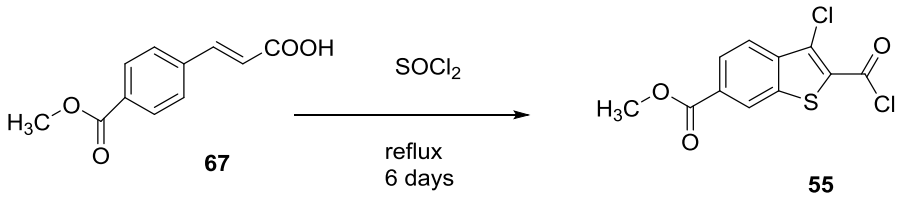
spectral data were as follows: ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.96 (s, 3H), 7.95 (d, *J* = 6.7 Hz, 2H), 8.20 (d, *J* = 6.7 Hz, 2H), 10.11 (s, 1H).

Preparation of 4-Methoxycarbonylcinnamic acid (67)[44]



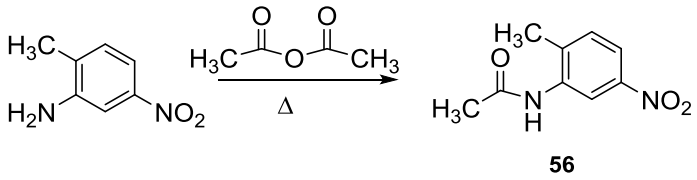
To a solution of commercially available malonic acid 3.5 g (33 mmol) dissolved in 20 mL of anhydrous pyridine was added at room temperature 5.4 g (33 mmol) of methyl-4-formylbenzoate **66** dissolved in another 75 mL anhydrous pyridine followed by 1 mL piperidine (cat. amount) dropwise under nitrogen. The mixture was then placed in a sand bath and slowly stirred while the temperature was increased to 80 -90 ^{o}C while CO_2 evolved, and then refluxed until CO_2 evolution ceased. After cooling to room temperature, the reaction mixture was poured onto ice and conc. HCl to form a precipitate. The acid **67** was filtered off, washed with water several times, dried to give 6.68 g (97% yield) of **67** as a creamy white solid with, mp. 234.5 –236.2 ºC. The spectral data were as follows: ^{1}H NMR (400 MHz, $CDCl_3$): δ 3.84 (s, 3H), 6.67 (d, *J* =16.10 Hz, 1H), 7.66 (d, *J* = 16.1 Hz, 1H), 7.83 (d, *J* = 8.3 Hz, 2H), 7.97 (d, *J* = 8.2 Hz, 2H), 12.59 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 52.7, 122.2, 128.7, 130.0, 131.2, 139.5, 142.9, 166.2, 167.9.

Preparation of 6-Methoxycarbonyl-3-chlorobenzo[b]thiophene-2-carbonyl chloride (55)[42]



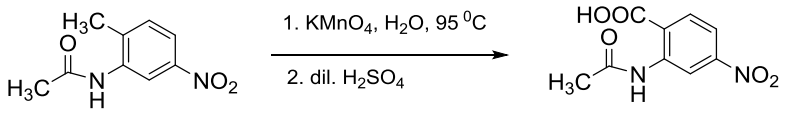
To a stirred solution of 10.0 g (50 mmol) of 4-carbomethoxy cinnamic acid **67** in 52.0 mL (720 mmol) of thionylchloride was added dropwise 0.8 mL of pyridine. The reaction mixture was refluxed for 6 days. The hot reaction mixture was filtered to remove pyridiniumhydrochloride salt. The filtrate concentrated *in vacuo*. The residue was triturated with hexane several times, filtered, and dried to give an off white solid. The solid was then used for the next step without further purification.

Preparation of 2-methyl-5-nitro-acetanilide (**56**)



The procedure was adapted from the procedure reported by Steiner.[45] To commercially available 1.60 g (10.5 mmol) of 4-methyl-5-nitroaniline was added 3 mL (30 mmol) of acetic anhydride and gently boiled for short time with stirring. Then reaction mixture was poured in to ice water while stirring. The solid product formed was filtered under suction and recrystallized with methanol to obtained 1.75 g (90 % yield) of product **56** as white crystals, mp. 151-152 °C. The spectral data were as follows: ^{1}H NMR (300 MHz, DMSO-d_6) δ 2.09 (s, 3H), 2.32 (s, 3H), 7.46 (d, J = 8.7 Hz, 1H), 7.90 (d, J = 9.3 Hz, 1H), 8.47 (s, 1H), 9.54 (s, 1H).

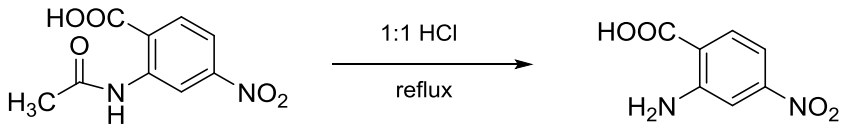
Preparation of 2-acetamino-4-nitrobenzene-1-carboxylic acid (**57**)[45]



56 **57**

To 1.5 g (7.7 mmol) of 2-methyl-5-nitro-acetanilide **56** was added 75 mL water and heated to 80 °C. Finely powdered 3.44 g (21.8 mmol) of $KMnO_4$ was added, the temperature was increased to 95 °C and stirred for 3 h. A 5 mL portion of ethanol was added to the reaction to remove excess $KMnO_4$, and the hot solution was filtered. The filtrate was acidified with dil. H_2SO_4 to make precipitate. The resultant precipitate was filtered under suction and dried to obtain 1.62 g (94% yield) of compound **57** as a pale yellow colour crystals, mp 215-216 °C. The spectral data were as follows: ^{1}H NMR (300 MHz, DMSO-d_6) δ 2.16 (s, 3H), 7.91 (d, *J* = 9.39 Hz, 1H), 8.15 (d, *J* = 8.83 Hz, 1H), 9.25 (s, 1H), 11.31 (s, 1H).

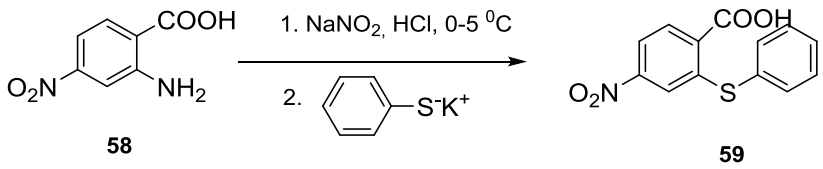
Preparation of 2-amino-4-nitrobenzoic acid (**58**)[45]



57 **58**

To 1.6 g (8.4 mmol) of 2-acetamino-4-nitrobenzene-1-carboxylic acid **57** was added 1:1 HCl and refluxed for 4 h. The solution was adjusted to pH 4-5 using 50% NaOH to form an orange precipitate. The precipitate was filtered under vacuum and recrystallized from aqueous ethanol to obtain 1.32 g (87% yield) of an orange colour needle like crystals, mp. 253-254 ^{o}C. The spectral data were as follows: ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.22 (dd, *J* = 1.97Hz, 8.79 Hz, 1H), 7.61 (d, *J* = 1.97 Hz, 1H), 7.89 (d, *J* = 8.79 Hz, 1H).

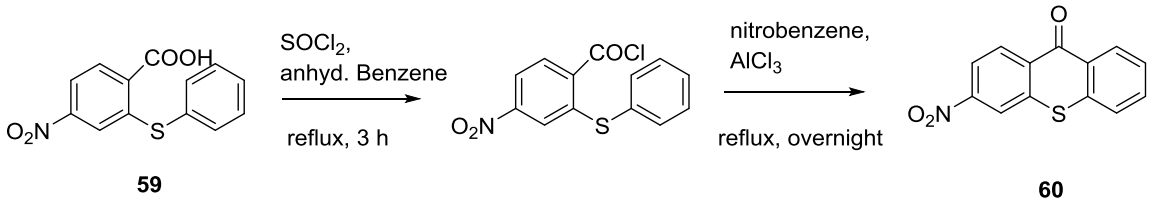
Preparation of 4-nitro-2(phenylthio) benzoic acid (**59**)



58 **59**

The procedure was adapted by the procedure reported by Turnbull.[46] 1.91 g (10.5 mmol) of 2-amino-4-nitrobenzoic acid **58** was dissolved in 15 mL of water and 5 mL conc. HCl and cooled in an ice bath. 0.75 g (10.8 mmol) of $NaNO_2$ was dissolved in 2 mL of water and kept in an ice bath. To cold solution of 58 in HCl was added the above $NaNO_2$ solution gradually. The reaction mixture was stirred for 30 minutes in an ice bath. Sulphamic acid was added to remove excess HNO_2 while testing with KI/Starch paper. The diazonium salt solution was allowed to reach 3-8 ^oC and it was added to well stirred solution of 1.1 g (10.0 mmol) thiophenol, 0.35 g silica gel, and 2 mL of 10 M NaOH in 50 mL water. The reaction mixture was stirred at 3-8 ^oC for 2 h and another 1 h at 20 ^oC. Silica gel was filtered off and pH of the solution was adjusted to pH 1 using 10 M HCl. Resultant yellowish brown colour precipitate was filtered off, washed until it was neutral and recrystallized from acetic acid to obtain 2.25 g (77 % yield) of a yellow colour powder, mp. 213-215 ^oC. The spectral data were as follows: ^1H NMR (300 MHz, DMSO-d_6): δ 7.40 (d, J = 1.91, 1H), 7.58-7.77 (m, 5H), 7.99 (d, J = 7.90 Hz, 1H), 8.13 (d, J = 9.07 Hz, 1H); ^{13}C NMR (75 MHz, DMSO-d_6): δ 119.9, 121.3, 127.8, 129.9, 131.3, 131.9, 133.5,136.5, 145.5, 150.0, 167.1.

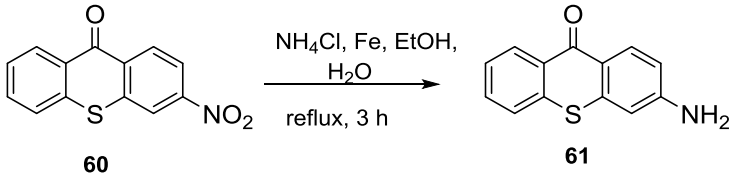
Preparation of 3-nitroxanthone (60)[46a]



59 **60**

A solution of 1.65 g (5.60 mmol) of 4-nitro-2(phenylthio) benzoic acid **59** in 20 mL anhydrous benzene with 1.6 mL (20 mmol) of $SOCl_2$ was refluxed for 3 h. The benzene was evaporated under *vacuo* and the residue was dissolved in warm nitrobenzene. A cold solution of 1.60 g (11.9 mmol) anhydrous $AlCl_3$ in 20 mL nitrobenzene was added dropwise to the above suspension at 0 ^oC. The reaction mixture was stirred at 45-55 ^oC for two days, decomposed by the addition of ice, removed nitrobenzene by vacuum distillation. The residual solution was cooled and the tarry material was collected, dried and boiled with 15 mL glacial acetic acid for an hour. The solution was cooled and the yellow colour product was collected by filtration, washed with glacial acetic acid, water , and dil. NaOH solution, and then dried to obtain 1.10 g (75 % yield) of a yellow colour powder. Recrystallization with glacial acetic acid formed 0.9 g (62%) of golden yellow plates, mp. 252-253 ^oC. The spectral data were as follows: 1H NMR (300 MHz, DMSO-d_6) δ 7.63 (t, J = 7.4 Hz, 1H), 7.83 (t, J =7.4 Hz, 1H), 7.93 (d, J = 9.0 Hz, 1H), 8.28 (dd, J = 2.0, 9.0 Hz, 1H), 8.47 (d, J = 9.0 Hz, 1H), 8.85 (s, 1H); ^{13}C NMR (75 MHz, DMSO-d_6) δ 121.2, 123.1, 127.4, 128.1, 128.8, 129.9, 131.7, 132.6, 134.3, 137.2, 138.6, 149.8, 178.7.

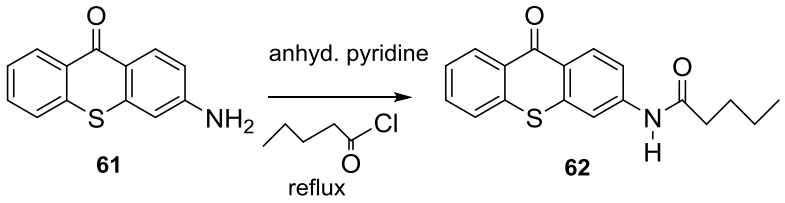
Preparation of 3-aminothioxanthone (**61**)



60 **61**

The procedure was adapted from the procedure reported by Steinmetz.[26] A mixture of 1.3 g (5.0 mmol) of 3-nitroxanthone **60**, 200 mL ethanol, 60 mL of water, 1.6 g (31 mmol) of NH_4Cl and 0.9 g (16 mmol) iron powder was refluxed for 3 h. The reaction mixture was filtered over silica gel to remove inorganic materials and residue was washed with 30 mL of ethanol. The combined filtrate was concentrated under *vacuo*. The product was extracted with $CHCl_3$ (50 x 3 mL). The $CHCl_3$ layer was dried over anhydrous Na_2SO_4 and evaporated under vacuo to obtain 0.84 g (74%) of **61** as an orange colour powder**.** It was crystallized from ethanol to give pale-yellow needles, mp. 253-255 ^oC. The spectral data were as follows: ^1H NMR (300 MHz, DMSO-d_6) δ 6.41 (bs, 1H), 6.67 (dt, *J* =1.8 Hz, 1H), 6.74 (d, *J* = 8.3 Hz, 1H); ^{13}C NMR (75 MHz, DMSO-d_6) δ 106.8, 115.1, 118.0, 126.7, 126.9, 129.3, 129.6, 131.7, 132.1, 136.6, 139.4, 153.7, 177.5.

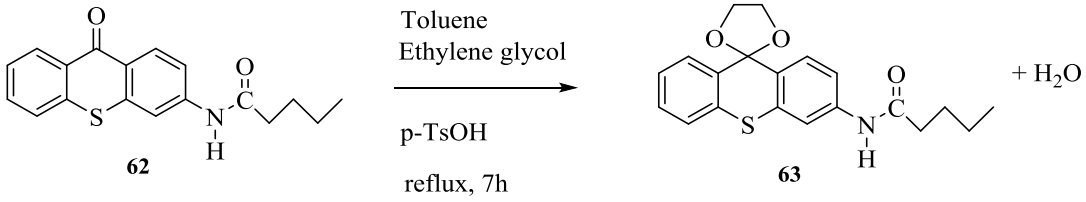
Preparation of 3-aminothioxanthone (**62**)



A mixture of 0.62 g (2.7 mmol) of **61** and 0.35 mL (2.8 mmol) of pentanoil chloride was refluxed in pyridine for 8 h. After completion of the reaction, the reaction mixture was evaporated. 40 mL water and 100 mL ethyl acetate was added. The organic layer was washed with 5% Na_2CO_3, water, saturated NaCl solution, dried over anhydrous Na_2SO_4. and evaporated under *vacuo* to give 0.78 g (91% yield) of product **62** as an orange pink colour solid, mp. 201-203 ^oC. The spectral data were as follows: ^1H NMR (300 MHz, DMSO-d_6) δ 0.91 (t, *J* = 8.7 Hz, 3H), 1.32 (t, *J* =8.7 Hz, 2H), 1.59 (qn, *J* =8.7 Hz, 2H), 2.39 (t, *J* = 8.0 Hz, 2H), 7.57 (s, 1H), 7.95 (s, 1H), 7.21-7.86 (m, 2H), 8.23 (d , *J*= 1.3 Hz,

1H), 8.40 (d, *J* = 8.8 Hz, 1H), 8.44 (d, *J* = 8.2 Hz, 1H), 10.48 (br, 1H); ^{13}C NMR (75 MHz, DMSO-d_6) δ 13.9, 22.6, 27.8, 37.7, 114.9, 118.1, 125.3, 126.5, 126.2, 129.4, 129.9, 131.2, 132.4, 137.6, 139.7, 141.6, 172.3, 179.0.

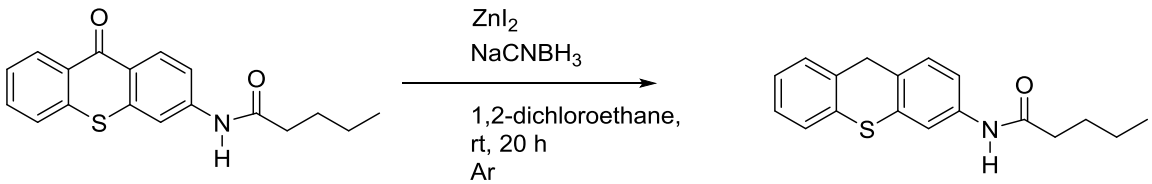
Preparation of 63



62 63

The procedure was adapted from the procedure reported by Shibasaki.[48] To a stirred solution of 1.56 g (5.00 mmol) of **62** in 30 mL of toluene, 0.85 mL (15 mmol) of ethylene glycol and catalytic amount of *p*-TsOH. The reaction mixture was refluxed for 7 hours by using Dean-Stark apparatus. The reaction mixture was cooled to room temperature and saturated $NaHCO_3$ was added to quench the reaction. The product was extracted into $CHCl_3$, washed with water and brine, dried over anhydrous Na_2SO_4 and evaporated to obtain a little amount of product **63** with some product by hydrolysis of **62**.

Synthesis of compound 65



62 65

To a solution of 0.20 g (0.65 mmol) of **62** in 5 mL of CH_2Cl_2 at room temperature was added 0.31 g (0.97 mmol) of solid ZnI_2 and 0.31 g (4.8 mmol) of $NaCNBH_3$. The

reaction mixture was stirred at room temperature for 20 h at argon environment. It was then cooled and poured into an iced-cold mixture of saturated NH_4Cl contain 10 % 6N HCl. The mixture was extracted with ethyl acetate, dried over anhydrous $MgSO_4$, filtered and evaporated to dryness. The pale pink product converted to dark red colour when exposure to air. This reaction was unsuccessful to obtain compound **65**.

CHAPTER 5. Future Plans

1. For compound **26** another photolysis for quantum yield determination will be planned with a more diluted (10^{-4} M) solution using HPLC to quantify the photoproduct in order to get more reliable results.

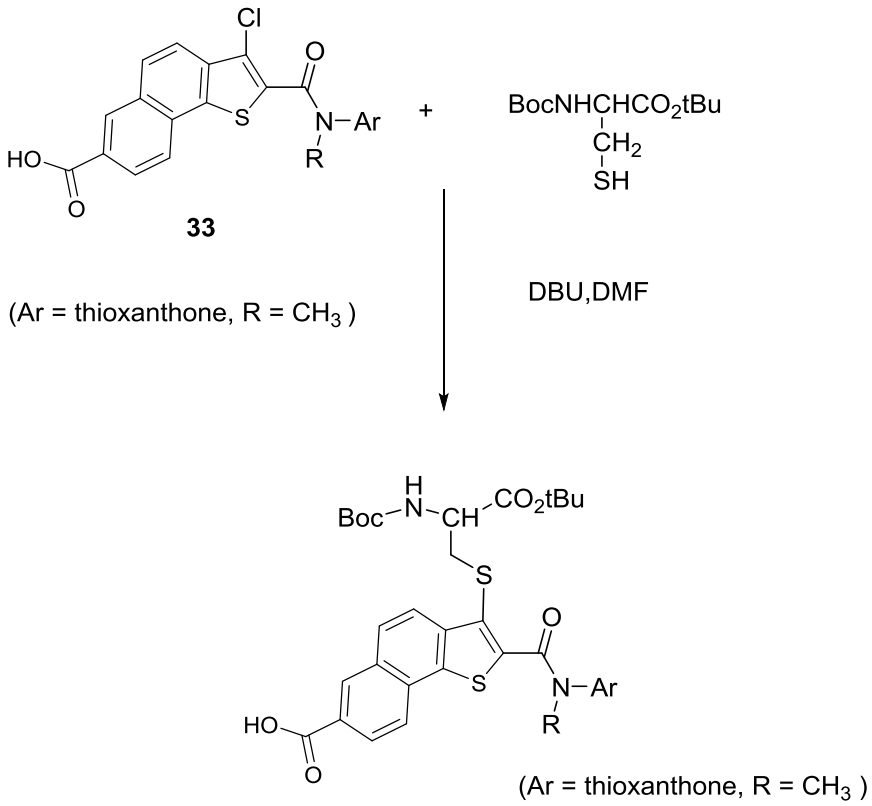
2. Theoretical calculations will be planned to ascertain details of the electronic configuration of the triplet excited state of the 5-benzoylthiophene in **26**. Such calculations would reveal whether the C-3 position of the thiophene ring is indeed pyramidalized, as is the case for the benzothiophene ring system.[26] One concern with **26** 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.

3. Synthesis of compounds **32** and **33** is in progress. It is therefore expected that they will be obtained near future. Further studies of **33** will initially focus on products yields and efficiencies. Future plans will also include the finding the multiplicity of the reactive excited state through quenching studies of **33** with the piperylene and oxygen. Quenching studies with piperylene also helpful to find the triplet excited state life time. Moreover, studies of **33** will be extended to release more basic leaving groups PhO^-, PhS^-, $PhCH_2S^-$. 3-chloro group can be readily substituted by variety of thiols using DBU as a base.[48] The preliminary studies with **33** containing leaving groups Cl^-, PhO^-, PhS^- and $PhCH_2S^-$ will be completed for publication.

4. Incorporating cysteine and glutathione into compound **33** and photochemical studies will also be planned. This would involve displacing the C-3 chloride in the molecule. The

literature route for displacing C-3 chloride by thiols entails stirring both in DMF and DBU.[46] The caging of simple thiol groups using 33 is straightforward. For example compound **33** is stirred with thiol and DBU in DMF. Similarly, Boc-γ-Glu-Cys-Gly-OtBu could be introduced. Cysteine could be attached *via* its thiol group using Boc-Cys-OtBu (Scheme 5.1).

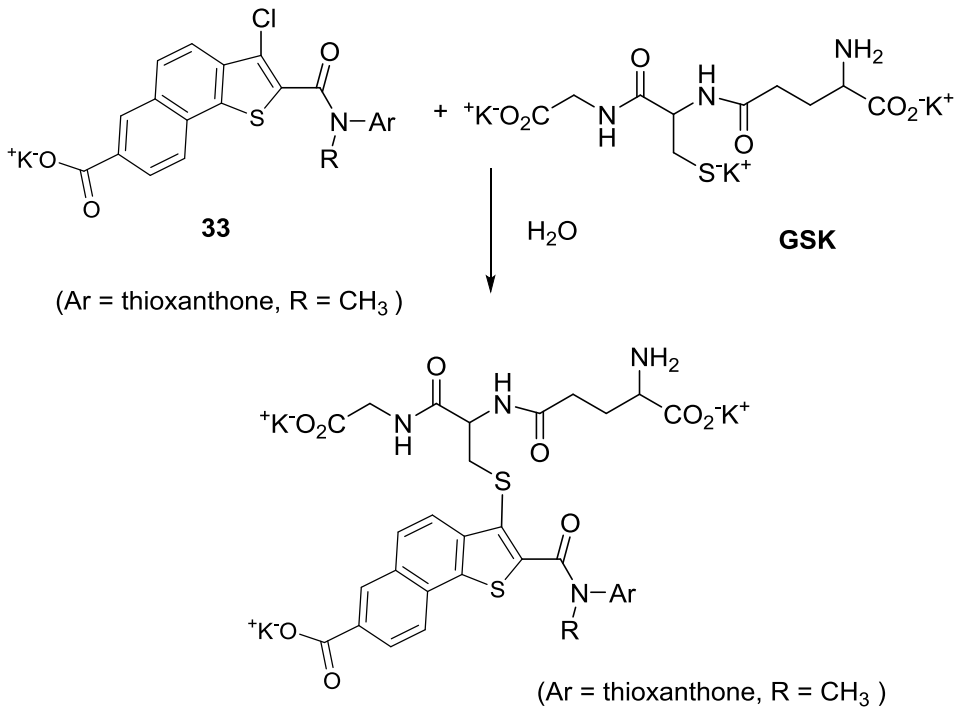
Scheme 5.1



Another approach to attach Cys and GHS would use aqueous basic conditions with water soluble caging group **33** (Scheme 5.2). Thiolate should readily substitute the C-3 chloro group of **33**, while the amino and carboxylate groups should not react.

The product isolation would entail a Sephadex chromatography.

Scheme 5.2



5. Future work will incorporate protected cysteine caged by compound **33**. This would allow further elaboration into other peptides synthesis.[49, 50]

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