

Copper-Catalyzed Multi-Component Synthesis and Biological Properties of Coumarin Derivatives

*A Thesis Submitted
in Partial Fulfilment of the Requirements
for the Degree of*

DOCTOR OF PHILOSOPHY

by

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The logo of the Indian Institute of Technology Guwahati is a circular emblem. It features a central stylized figure with three rounded, bulbous shapes, resembling a person or a deity. The figure is surrounded by a circular border containing text in both Hindi and English. The Hindi text at the top reads "भारतीय प्रौद्योगिकी संस्थान गुवाहाटी" and the English text at the bottom reads "Indian Institute of Technology Guwahati".

Dedicated To
My Family Members and
Teachers



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STATEMENT

I hereby declare that the matter embodied in this thesis is the result of investigations carried out by me in the Department of Chemistry, Indian Institute of Technology Guwahati, Guwahati, India under the supervision of Prof. Tharmalingam Punniyamurthy.

In keeping with the general practice of reporting scientific observations, due acknowledgement has been made wherever the work described is based on the findings of other investigators.

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CERTIFICATE

This is to certify that Mr. G. Murugavel has been working under my supervision since December 2010. I am forwarding his thesis entitled “*Copper-Catalyzed Multi-Component Synthesis and Biological Properties of Coumarin Derivatives*” for being submitted for the award of Ph.D. degree of this Institute. I certify that he has fulfilled all the requirements according to the rules of this institute, and regarding the investigations embodied in his thesis, this work has not been submitted elsewhere for a degree.

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Supervisor

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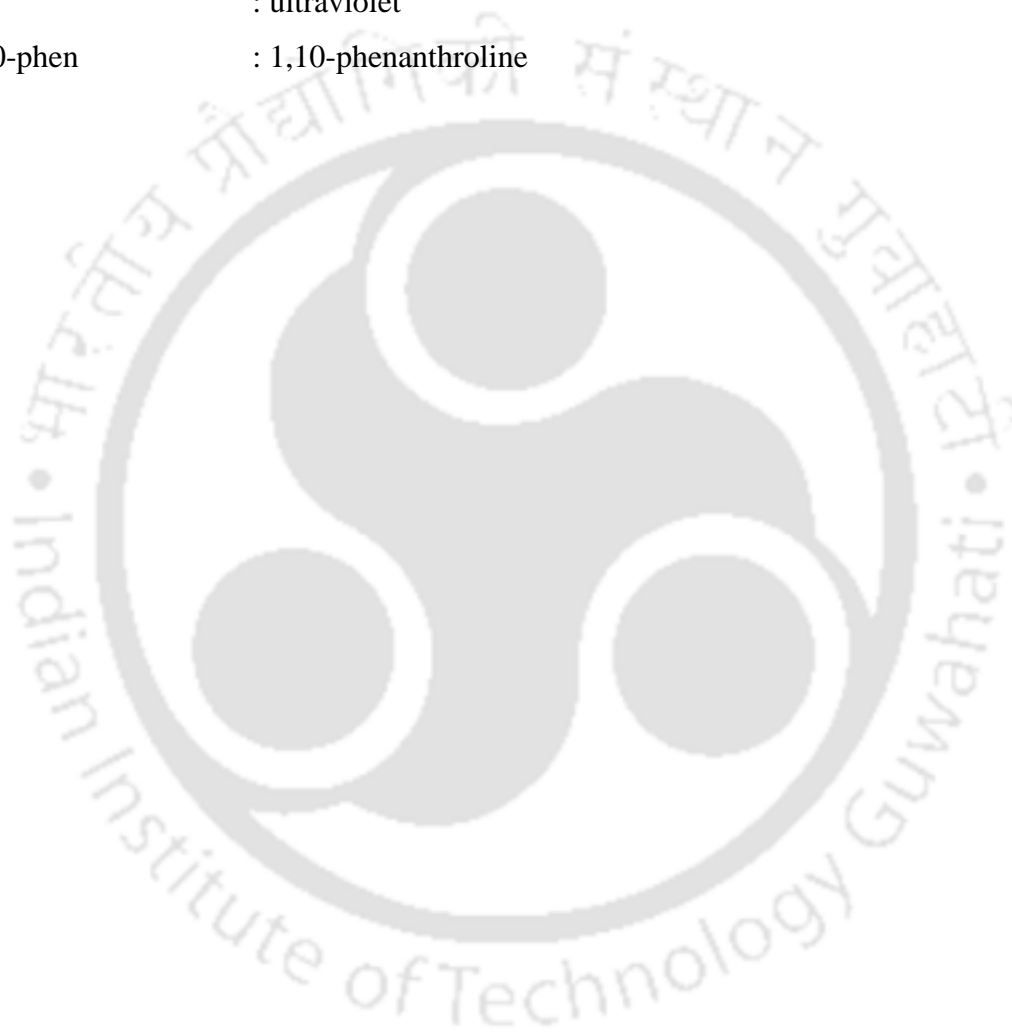
G. Murugavel

List of Abbreviations

Abs.	: absorption
Ac	: acetyl
acac	: acetylacetonate
AKR1B10	: aldo-keto reductase family 1 member b10
APCI	: atmospheric-pressure chemical ionization
Ar	: aryl
aq	: aqueous
BACE1	: beta-secretase 1
Bn	: benzyl
Bu	: butyl
Boc	: <i>t</i> -butoxycarbonyl
BSA	: bovine serum albumin
Bu	: butyl
calcd	: calculated
CBR1	: carbonyl reductase 1
CCDC	: cambridge crystallographic data centre
CK2	: casein kinase II
Conc.	: concentration
COSY	: correlation spectroscopy
COX2	: cyclooxygenase 2
DBU	: 1,8-diazabicycloundec-7-ene
dba	: dibenzylideneacetone
DCC	: <i>n,n'</i> -dicyclohexylcarbodiimide
DCE	: 1,2-dichloroethane
DCM	: dichloromethane
DEPT	: distortionless enhancement of polarisation transfer
DMEM F12	: dulbecco's modified eagle medium: nutrient mixture F-12
DMF	: dimethylformamide
DMSO	: dimethylsulfoxide
dr	: diastereomeric ratio
EDG	: electron donating group

eq	: equation
equiv	: equivalent
ESI	: electrospray ionization
Et	: ethyl
EWG	: electron withdrawing group
Fl.	: fluorescence
FT-IR	: fourier transform infrared spectroscopy
HER2	: human epidermal growth factor receptor 2
HIV	: human immunodeficiency virus
HRMS	: high-resolution Mass Spectrometry
HSA	: human serum albumin
HSQC	: heteronuclear single quantum coherence spectroscopy
MCRs	: multicomponent reactions
Me	: methyl
MOM	: methoxymethyl
mp	: melting point
MS	: molecular sieves
m/z	: mass to charge ration
MTT	: 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide
MW	: microwave
NMR	: nuclear magnetic resonance
ORTEP	: oak ridge thermal ellipsoid plot
PBS	: phosphate-buffered saline
PKC	: protein kinase C
Pr	: propyl
Q-TOF	: quadrupole-time-of-flight
R _f	: retention factor
ROS	: reactive oxygen species
rt	: room temperature
TBAB	: tetra-n-butylammonium bromide
TBACl	: tetra-n-butylammonium chloride
TBAI	: tetra-n-butylammonium iodide
TBS	: t-butyldimethylsilyl

Tf	: trifluoromethanesulfonyl
TFA	: trifluoroacetate
THF	: tetrahydrofuran
TLC	: thin-layer chromatography
TMS	: trimethylsilyl
TNBC	: triple negative breast cancer
Ts	: <i>p</i> -toluenesulfonyl
UV	: ultraviolet
1,10-phen	: 1,10-phenanthroline



List of Units

Å	: Angstrom
a	: Onsager radius
c	: velocity of light
cm	: centimeter
g	: gram
h	: Planck's constant
Hz	: hertz
IC ₅₀	: half maximal inhibitory concentration
K	: Kelvin
kcal	: kilocalories
K _i	: inhibitory constant
M	: molar
mg	: milligram
MHz	: mega hertz
min	: minutes
mol	: mole
mM	: millimolar
nm	: nanometer
SD	: standard deviation
λ	: wavelength
μ_e and μ_g	: excited state and ground-state dipole moments
μM	: micromolar
τ	: lifetime
$\bar{\nu}$: wavenumber
°C	: degree of Celsius

Abstract

The thesis is consisted of five chapters. The first chapter describes about the general introduction and enunciates the scope of copper-catalyzed multicomponent reactions for heterocycle synthesis *via in situ* generated highly reactive ketenimine intermediate. The second chapter describes the synthesis of highly functionalized iminocoumarin aryl methyl ether using Cu-catalyzed multicomponent reaction of ynals, sulfonyl azides and phenols. The third chapter is devoted to microwave-assisted Cu-catalyzed four component cascade reaction for the synthesis of substituted coumarin-3*N*-sulfonamide derivatives. The fourth chapter designates the studies on interaction of (*Z*)-7-(diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide as a new solvatochromic fluorescent probe with bovine serum albumin (BSA). The fifth chapter is solely dedicated to study the anticancer activity of iminocoumarin aryl methyl ethers against triple negative breast cancer cell line MDA- MB-231.

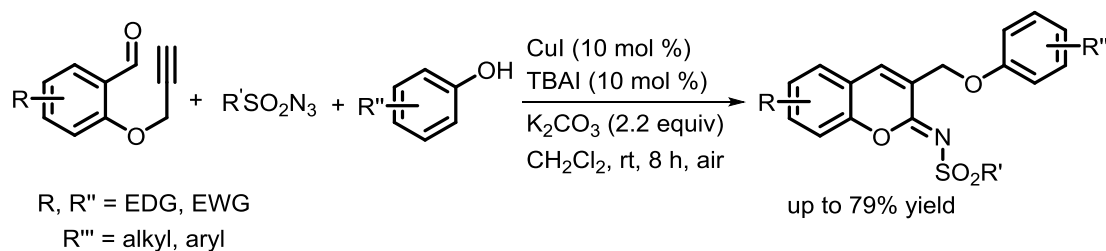
Chapter I. Copper-Catalyzed Multicomponent Reactions for Heterocycle Syntheses

Copper-catalyzed multi-component reaction (MCR) is one of the important synthetic tools for construction of highly functionalized heterocyclic compounds which access widespread application in medicinal and material sciences. In this chapter, a brief review on copper-catalyzed multicomponent reaction for the synthesis of biologically active and important heterocyclic motifs through *in situ* generated ketenimine intermediates has been studied. Moreover, this involves in various important transformations such as tandem nucleophilic addition and cyclization, cycloaddition reactions, intermolecular rearrangement, etc. with suitable coupling partners and their mechanistic pathway has been discussed.

Chapter II. Copper(I)-Catalyzed Three-Component Cascade Synthesis of Iminocoumarin Aryl Methyl Ethers

Iminocoumarins are privileged structural motifs, which are attributed with numerous applications in the field of material and biological sciences. This chapter describes the copper-catalyzed multicomponent reaction for the synthesis of iminocoumarin aryl methyl ethers from ynals, sulfonyl azides and phenols *via* copper-catalyzed azide alkyne cycloaddition (CuAAC), intramolecular ketenimine rearrangement, nucleophilic addition

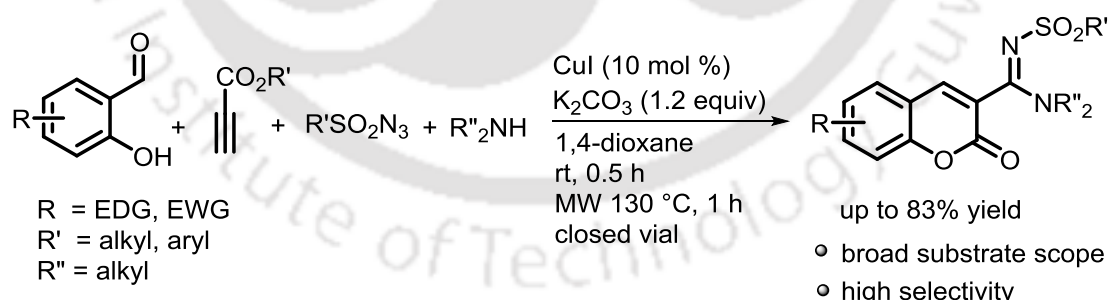
and tandem cyclization (Scheme 1). This protocol is general to access a wide variety of substrate scope and functional group tolerance in good yields at ambient conditions.



Scheme 1

Chapter III. Copper-Catalyzed Four-Component Cascade Synthesis of 3-*N*-Sulfonylamidine Coumarins

Coumarins are important classes of heterocyclic scaffolds which are widely exist in nature. This also exhibits several applications in biological, medicinal and material sciences. This chapter markedly focuses on the synthesis of 3-*N*-sulfonylamidine coumarins from salicylaldehydes, propiolates, sulfonyl azides and secondary amines (Scheme 2). The substrates undergo one-pot microwave-assisted copper-catalyzed four component coupling reaction using 10 mol % CuI in the presence of 1.2 equiv of K₂CO₃ in dioxane at 130 °C in closed vial to provide the corresponding coumarin derivatives. This protocol provides an effective route for the construction of highly functionalized coumarin structural frameworks in single operation with moderate to high yields.



Scheme 2

Chapter IV. Study on Interaction of Solvatochromic Fluorescent Probe (*Z*)-7-(Diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide with Bovine Serum Albumin

Solvatochromic fluorophores possesses emission properties which is sensitive to the nature of local micro-environments. These are widely used in the applications to study the

protein-binding interactions which can lead to design and development of drug discovery. This chapter demonstrates the synthesis of new coumarins based on solvatochromic fluorescence probe (*Z*)-7-(diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide from 4-(diethylamino)-2-hydroxybenzaldehyde, ethyl propiolate, tosyl azide and diisopropylamine. This was performed *via* microwave assisted copper catalyzed four component reaction and the photophysical studies on interaction of fluorophore with bovine serum albumin using UV-visible spectroscopy, fluorescence spectroscopy, time resolved fluorescence spectroscopy, fluorescence anisotropy and docking analysis (Figure 1).

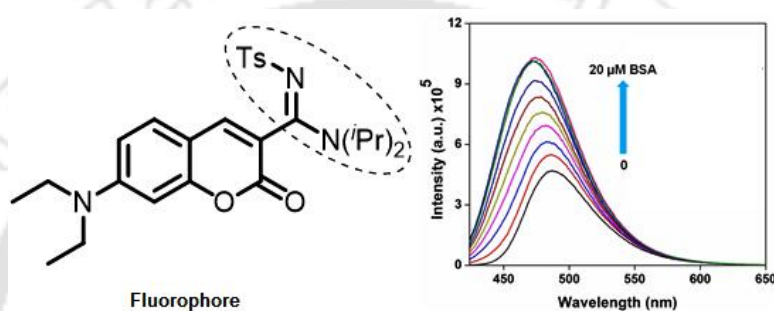


Figure 1. Interaction of new coumarin fluorophore with BSA

Chapter V. Anticancer Activity of Iminocoumarin Aryl Methyl Ethers Against Triple Negative Breast Cancer Cell (TNBC) Line MDA-MB-231

Triple negative breast cancers is one of the aggressive subtype of metastatic breast cancers, which does not express the gene such as estrogen receptors, progesterone receptors and human epidermal growth receptor 2 (HER2). Development of effective chemotherapeutic drug candidates with less toxicity for triple negative breast cancer cells are driving considerable attention among synthetic organic chemists and biologists, which relies on high efficiency, low cost and short synthetic routes. This chapter describes the study of anti-proliferative activity using series of iminocoumarin aryl methyl ethers against human metastatic triple negative breast cancer cell line MDA-MB-231 (Figure 2). The preliminary study provides a new iminocoumarin based drug candidates for the treatment of triple negative breast carcinoma.

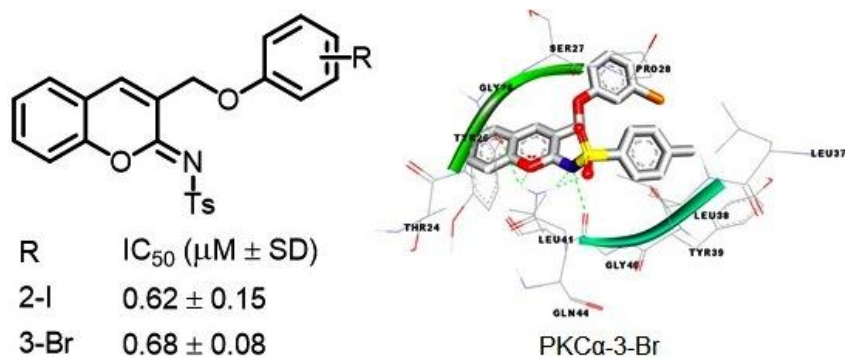


Figure 2. Anticancer activity against MDA-MB-231



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Copper-Catalyzed Multicomponent Reactions for Heterocycle Syntheses

In chemical syntheses, most of the important biological heterocyclic scaffolds are often synthesized divergently or sequentially in many steps, which vary on the complexity of target molecules rather than convergent methods. Optimization of each and every steps of divergent process leads to the lower yield of the target molecule, which associates costs, time, efforts, resources, selectivity and environmental impact. The “ideal synthesis” in organic chemistry should lead to the desired target molecule in a single step with high yield and 100% selectivity using the readily accessible starting materials (Figure 1). The synthetic variables towards ideal synthesis need to be optimized in terms of cost and time to improve the yield of the product.^{1,2} The preparative complexity in multistep synthesis increases proportionally to the number of steps involved in the reaction towards the target product. This reflected in product isolation and purification such as crystallization, extraction and distillation. One-pot convergent multicomponent reactions (MCRs) closely approached the concept of ideal synthesis by tackling the complexity of reaction.

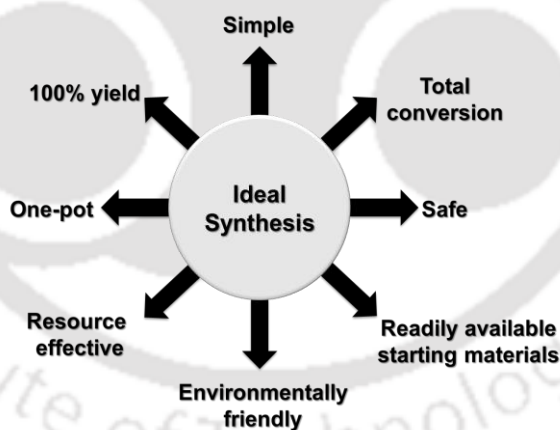


Figure 1. The ideal chemical synthesis^{4a}

MCRs afford powerful synthetic tool for the transformation of three or more readily available simple substrates into a library of highly functionalized molecules.³ The products are widely used in biological, medicinal and material sciences.⁴⁻⁶ In addition, MCRs are highly flexible, selective and atom efficient with high exploratory power (E_N)⁷, which is commonly identified as sub-class of domino⁸ or tandem⁹ reactions. MCRs can be broadly classified by the number of reaction component involved (Figure 2). They can

also be classified on the mechanistic pathway as three types:^{10,11} type I in which the reaction substeps are in equilibria (eg., Strecker and Mannich reactions), type II in which final step towards the product formation is irreversible (eg., Ugi and Passerini reactions) and type III in that the substeps of are irreversible (one-pot multicomponent synthesis of heterocycles).

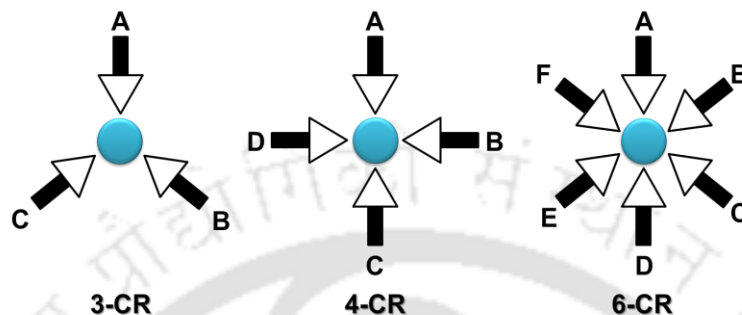
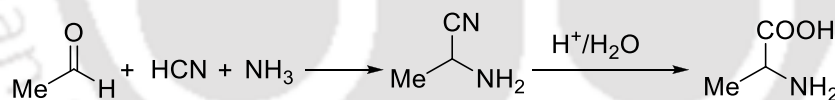


Figure 2. Multicomponent reactions.

1.1 History of MCRs for Heterocycle Synthesis

In 1850, Strecker synthesized α -amino nitrile via one-pot reaction of aldehyde, hydrogen cyanide and aqueous ammonia.¹³ This adduct is one of the important precursors for α -amino acids (Scheme 1).



Scheme 1

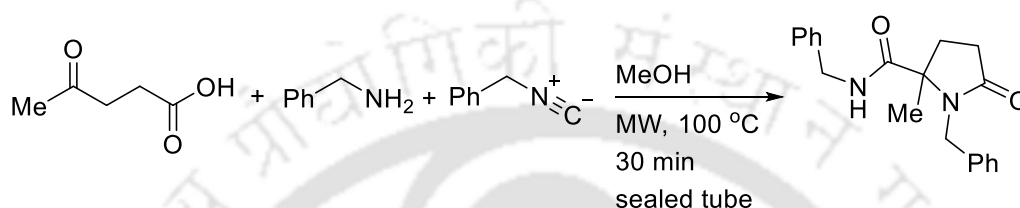
Table 1 summarizes some of the significant MCRs developed towards the synthesis of biologically important heterocycles. In 1882, Hantzsch reported the synthesis of symmetrical 1,4-dihydropyridines *via* the condensation of two equiv of β -ketoesters with one equiv of ammonia and aldehyde.¹⁴ Subsequently, Hantzsch showed the synthesis of pyrroles using condensation of α -halogenated ketones with β -ketoesters and primary amines.¹⁵ In 1891, Biginelli reported the synthesis of highly functionalized dihydropyrimidine using acid catalyzed condensation of β -ketoesters with aldehydes and urea.¹⁶ Later, Mannich invented the synthesis of β -aminoaldehydes or β -aminoketones *via* the condensation of enolizable carbonyl compounds with *in situ* generated iminium ions from aldehydes and amines.¹⁷ Robinson demonstrated the synthesis of tropinone using

Table 1. Some of the Classical and Most Significant MCRs

Name of the Reaction	Year	Heterocycle Synthesis
Hantzsch dihydropyridine synthesis ¹⁴	1882	
Hantzsch pyrrole synthesis ¹⁵	1890	
Biginelli reaction ¹⁶	1891	
Mannich reaction ¹⁷	1912	
Bucherer-Bergs reactions ¹⁸	1934	
Asinger reaction ¹⁹	1956	
Gewald reaction ²⁰	1965	

the condensation of methylamine, succinaldehyde and acetonedicarboxylic acid.^{17d} In 1934, Bergs and Bucherer reported independently the condensation of aldehydes or acetones with ammonium salts and cyanide sources such as HCN, KCN, *etc.*, to afford hydantoin, which can be transformed into α -amino acids by hydrolysis.¹⁸ In 1956, Asinger described the synthesis of thiozolines using the condensation of *in situ* generated 2-thioaldehydes or 2-thioketones with aldehydes and ammonia. Later, this reaction was successfully generalized to synthesize various five and six membered heterocycles such as imidazolines, oxazolines, oxazines, thiazines and pyrimidines.¹⁹ In 1965, Gewald reported the synthesis of functionalized thiophenes using the base catalyzed condensation of β -ketoesters, cyanoacetates and elemental sulphur.²⁰

In 1921, Passerini reported three component reaction of acids, carbonyl compounds and isocyanide to furnish α -acyloxycarboxamides.²¹ These reactions are highly versatile to synthesis of peptide-like linear compounds. By utilizing α -halogenated carbonyl compounds, cyclization can be achieved in one-pot process.²² In 1959, Ugi reported the four component reaction of amines, carbonyl compounds, acids and isonitriles to furnish α -acylamino amides.²³ Using bifunctional starting material as a multicomponent partner, the synthesis of heterocycles could be achieved (Scheme 2).^{4a,24}



Scheme 2

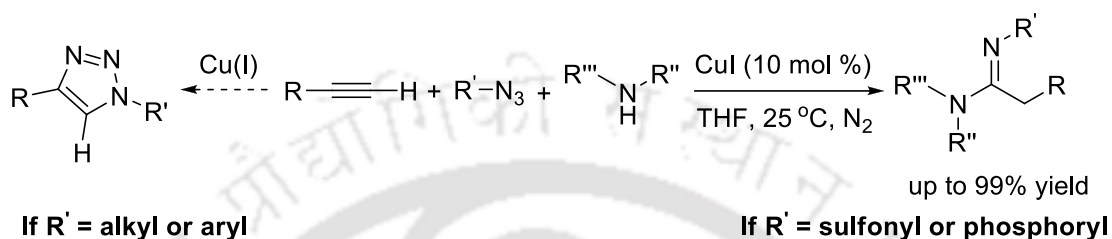
The use of transition metal (eg., Pd, Cu, Rh, Ru, *etc.*) based catalytic systems for MCRs facilitates the formation of highly functionalized heterocycles with high molecular diversity.²⁵⁻²⁸ Among them, Cu(I)-catalyzed MCRs is an efficient strategy for the synthesis of highly functionalized heterocyclic scaffolds with structural diversity using ketenimine intermediates.

1.2 Cu-Catalyzed MCRs for Heterocycle Synthesis

In 2002, Meldal and Sharpless independently reported Cu(I)-catalyzed cycloaddition of alkyl or aryl azides with terminal alkynes.²⁹ Since these prominent reports, versatility of these reactions are widely explored in the area of bioconjugation,³⁰ material science³¹ and drug discovery.³² Classically, thermally induced Huisgen cycloaddition usually afforded 1:1 mixture of 1,5-disubstituted and 1,4-disubstituted 1,2,3-triazoles.³³ On the other hand, Cu(I)-catalyzed cycloaddition of organic azides with terminal alkynes lead the formation of 1,4-disubstituted-1,2,3-triazoles.³⁴⁻³⁶ In 2006, Chang and co-workers reported an unprecedented Cu(I)-catalyzed three component synthesis of amidines with high yields via the coupling of sulfonyl azide and terminal alkyne (Scheme 3).³⁷

In case of phosphoryl and sulfonyl azides, the cycloaddition products are converted into ketenimine, which can be reacted in situ with variety of nucleophiles to furnish

diverse structural scaffolds (Figure 3).³⁸⁻⁴³ Cu(I) with alkyne produces copper(I)-acetylide copper acetylide **A**^{39c} that may undergo reaction with sulfonyl azides to produce *N*-sulfonyl (1,2,3-triazolyl) copper species **D** via intermediates **B** and **C**. Triazolyl copper species **D** may convert into the ketenimine **F** via the intermediate **E**. The active ketenimine may be reacted with nucleophiles or the combination of nucleophile and electrophile coupling partners.⁴⁴



Scheme 3

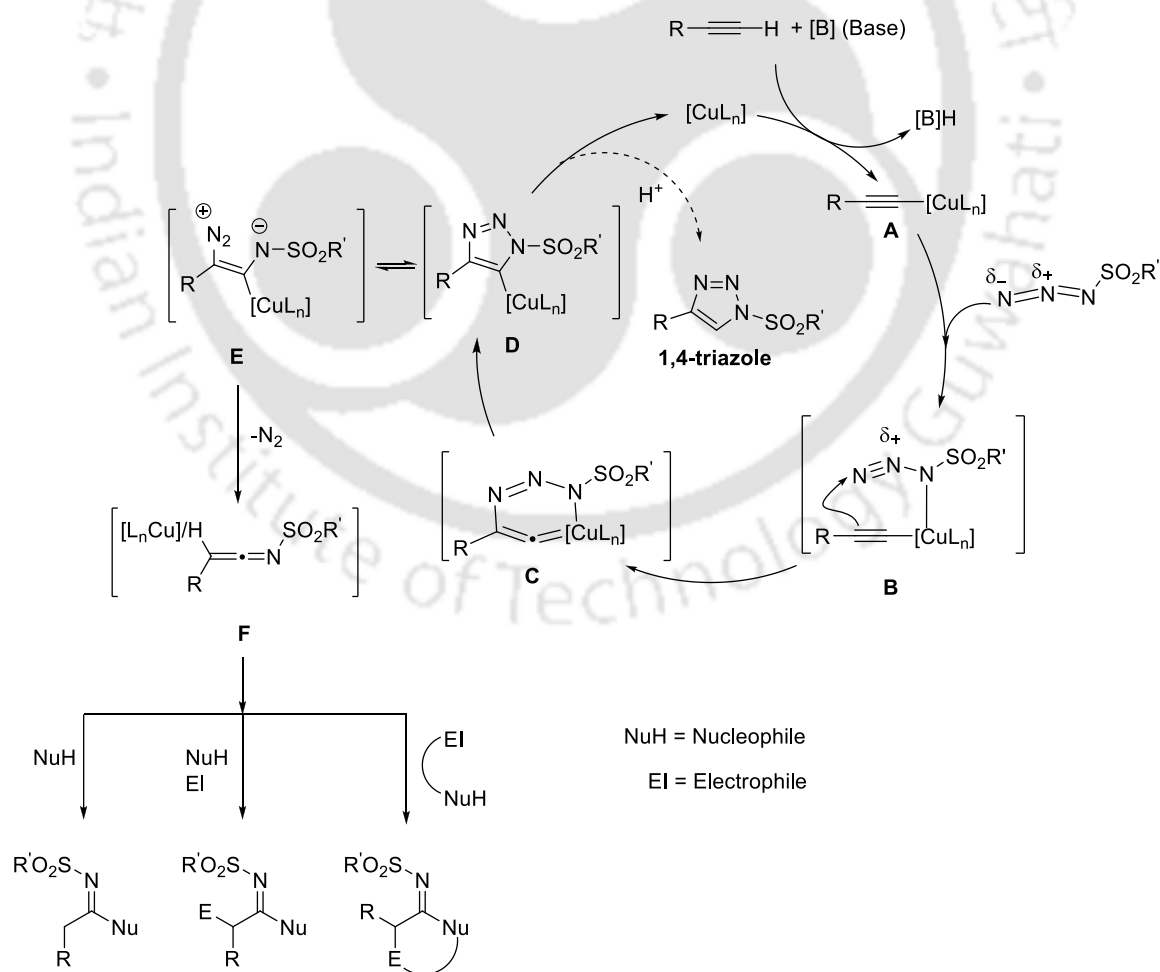
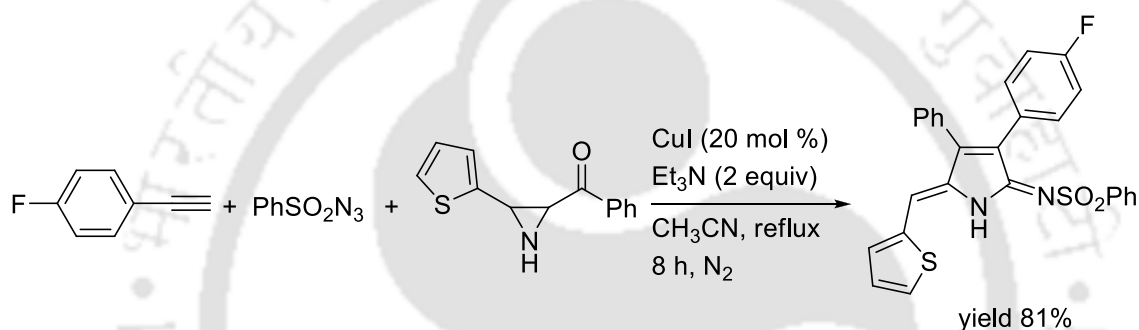


Figure 3. Plausible mechanistic pathways for the Cu(I)-catalyzed MCRs

1.2.1 Synthesis of Heterocycles Using Ketenimines

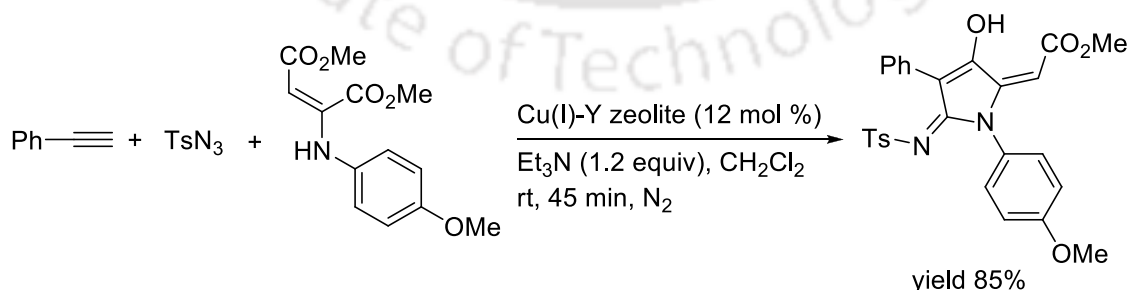
1.2.1.1 Reaction of Nitrogen Nucleophiles

Wang and co-workers reported the synthesis of 5-arylidene-2-imino-pyrrolines from 2-acylaziridines, sulfonyl azides and alkynes using 10 mol % CuI in the presence of 2 equiv Et₃N in CH₃CN at reflux under nitrogen atmosphere (Scheme 4).^{45a} The reaction proceeded *via* nucleophilic addition to *in situ* generated ketenimine followed by cleavage of C-N bond of aziridine to afford the target products. Under these conditions, aliphatic alkynes showed no product formation.



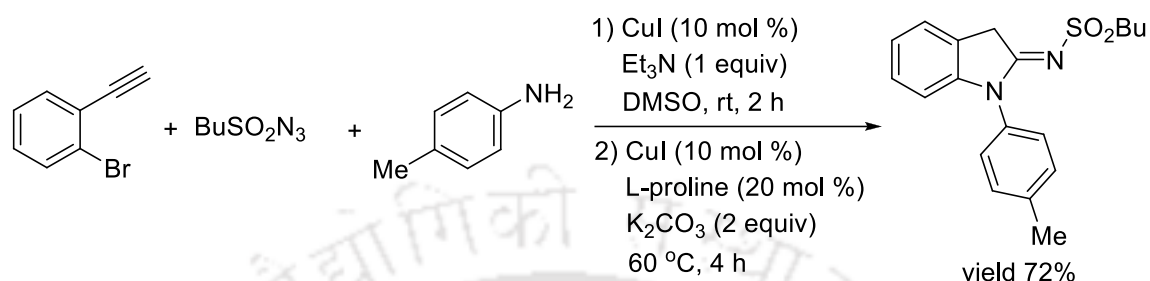
Scheme 4

Pitchumani and co-workers reported the synthesis of functionalized pyrrolidines from alkyne, sulfonyl azide and dimethyl 2-(phenylamino)maleate using zeolite-Cu(I)-catalysis (Scheme 5).^{45b} In this reaction, alkyne with sulfonyl azide generates ketenimine that undergoes reaction with dimethyl 2-(phenylamino)maleate followed by intramolecular [1,3]-H shift to produce the target products.



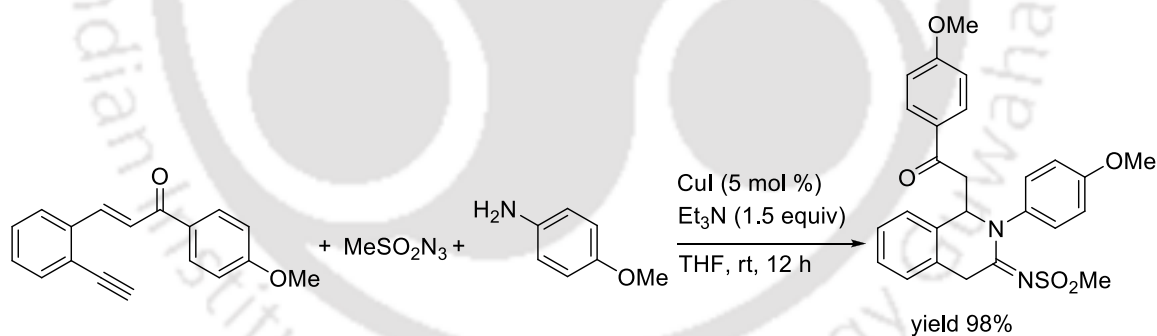
Scheme 5

Wang and co-workers showed Cu-catalyzed three-component synthesis of 2-(*N*-sulfonylimino)indolines from 2-bromo-phenylacetylene, sulfonyl azides and amines (Scheme 6).⁴⁶ The reaction proceeded *via* C-N bond cross-coupling reaction sequence to afford indoline derivatives with moderate to good yield.

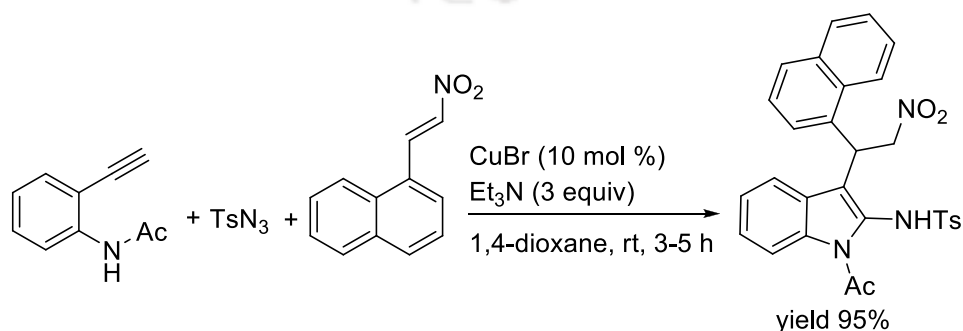


Scheme 6

Wu and co-workers reported the synthesis of 1,2-dihydroisoquinolin-3(4*H*)-imines from 2-ethynylphenylchalcone, sulfonyl azide and amine using 5 mol % CuCl in the presence of Et₃N in high yield (Scheme 7).⁴⁷ This reaction proceeded *via* double nucleophilic addition of amine nucleophile to both *in situ* generated ketenimine and Michael acceptor.



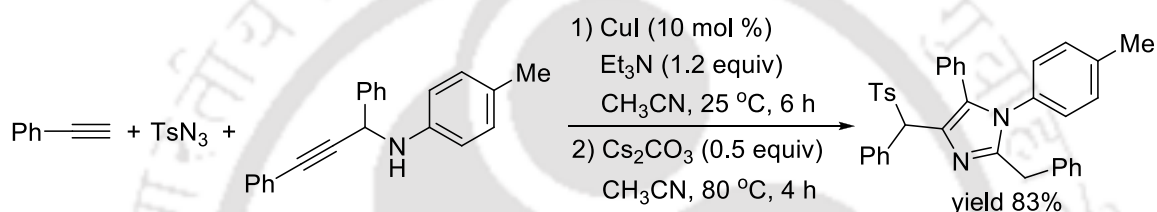
Scheme 7



Scheme 8

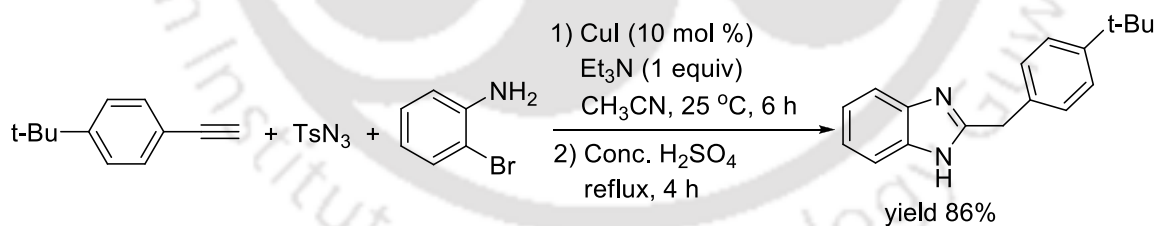
The authors also subsequently developed a new protocol for the synthesis of indole derivatives using copper catalyzed three component reaction of 2-ethynylaniline, sulfonyl azide and nitrostyrene *via* nucleophilic addition to *in situ* generated ketenimine followed by Michael addition to the styrene (Scheme 8).⁴⁸ Some of the derivatives are found to be HCT-116 inhibitor from the preliminary biological screening.

Wang and co-workers reported Cu(I)-catalyzed three component reaction alkyne, sulfonyl azide and propargylamine using 10 mol % CuI in the presence of Et₃N in CH₃CN (Scheme 9).⁴⁹ The reaction proceeded *via* cascade nucleophilic addition of amine to *in situ* generated ketenimine, 6 π -electrocyclization reaction and intramolecular [1,3] migration of sulfonyl substituents to furnish tetra-substituted imidazoles.



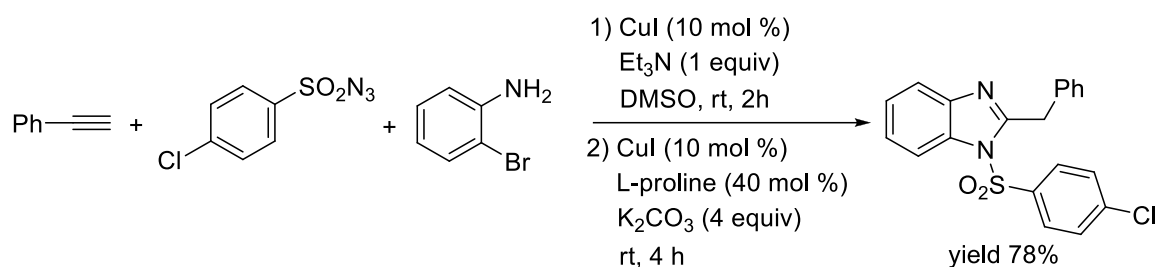
Scheme 9

The three-component reaction of 1,2-diamino benzene, alkyne and azide using 10 mol % CuI in the presence of Et₃N in CH₃CN followed by reflux with conc. H₂SO₄ afforded benzimidazoles (Scheme 10).^{50a}



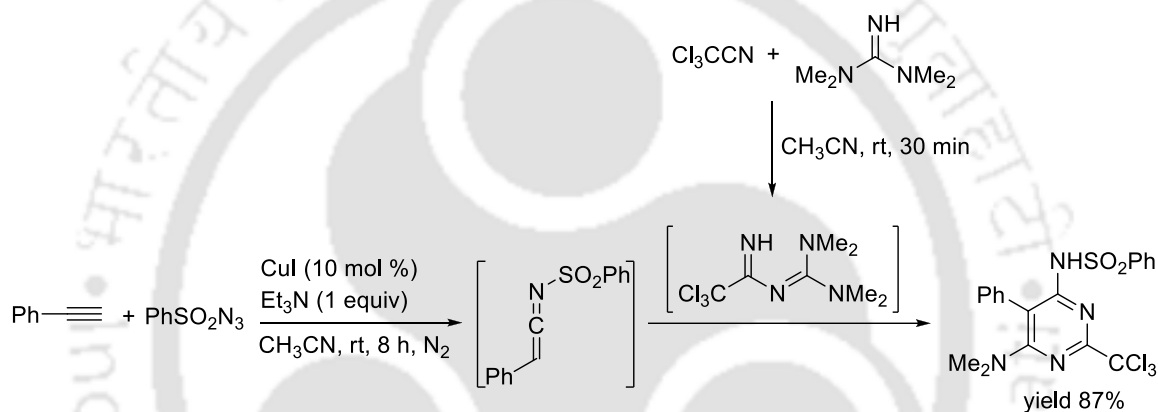
Scheme 10

Wang and co-workers reported the synthesis of functionalized *N*-sulfonyl substituted benzimidazole derivatives from 2-bromoaniline, sulfonyl azide and alkyne *via* one-pot Cu(I)-catalyzed three component reaction (Scheme 11).^{50b} The reaction proceeded *via* nucleophilic addition of aniline to *in situ* generated ketenimine followed by C-N cross-coupling reaction.



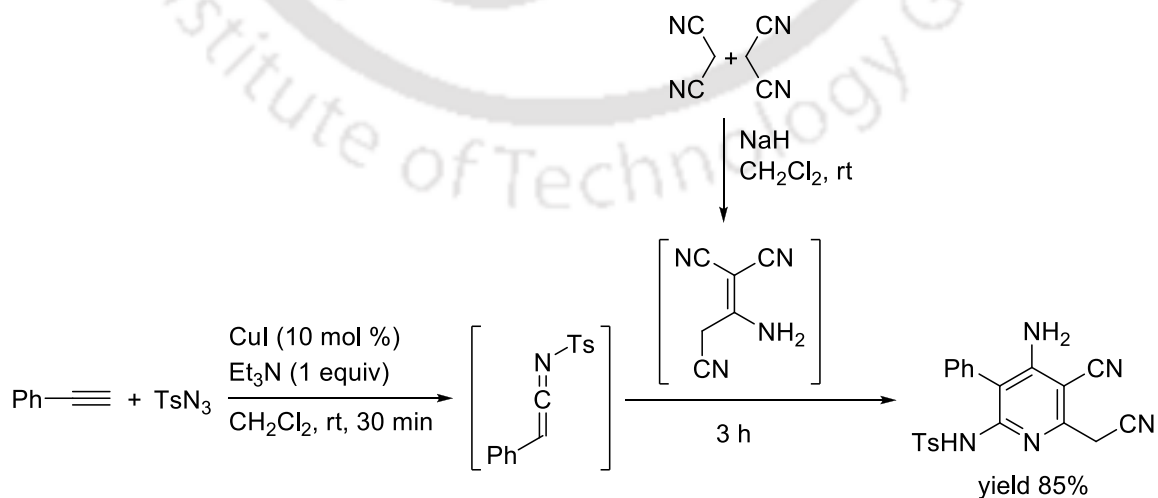
Scheme 11

Yavari and co-workers developed a new approach for the synthesis of polysubstituted pyrimidines using Cu(I)-catalyzed cycloaddition of alkyne with sulfonyl azide followed by the reaction of ketenimine with tetramethylguanidine derivative derived from trichloroacetonitrile and quinidine (Scheme 12).^{51a}



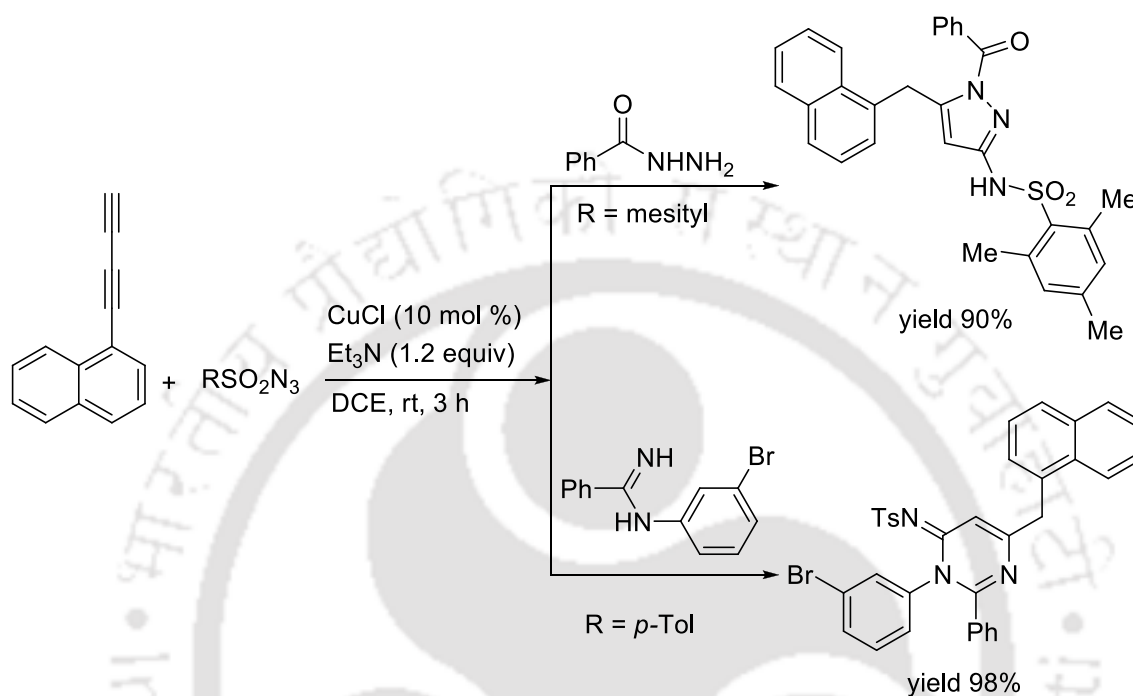
Scheme 12

The same group subsequently demonstrated the synthesis of pyridine derivatives from *in situ* generated ketenimine and 2-aminoprop-1-ene-1,1,3-tricarbonitrile (Scheme 13).^{51b}

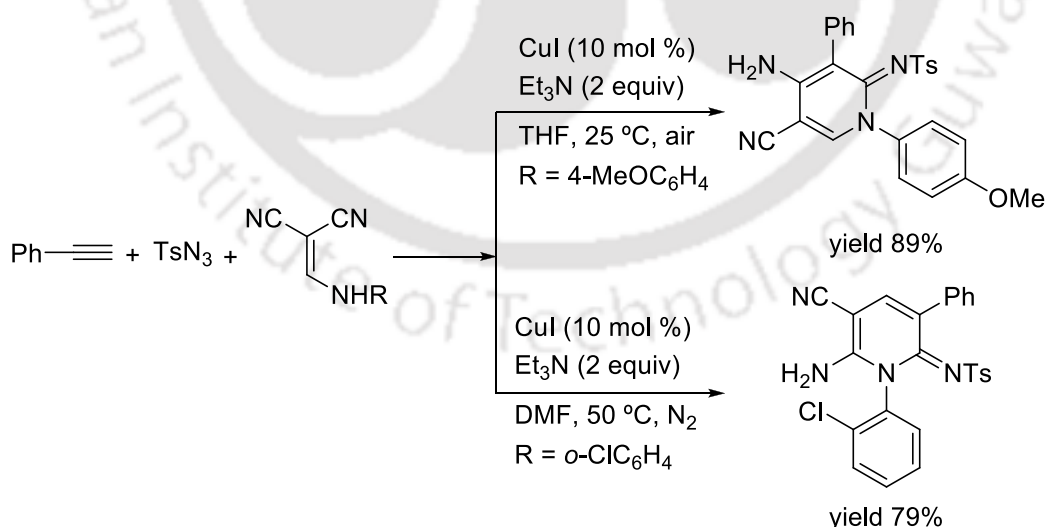


Scheme 13

The syntheses of 3-aminopyrazoles and 4-iminopyrimidine are demonstrated using 1,3-dinucleophilic addition of acyl hydrazine and *N*-aryl substituted imidamides to *in situ* generated ketenimine from conjugated alkyne using Cu(I)-catalyzed three-component reaction. In this reaction, β -alkynyl-*N*-sulfonyl ketenimine acts as 1,3-dielectrophilic equiv to furnish the corresponding heterocycles (Scheme 14).⁵²



Scheme 14

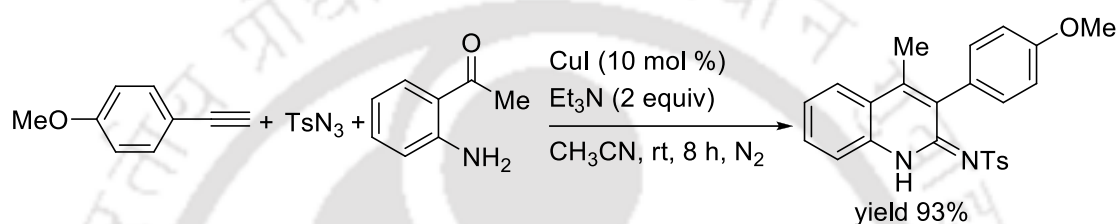


Scheme 15

Dong and co-workers reported the synthesis of functionalized 4-amino and 6-amino-2-iminopyridines from alkyne, sulfonyl azides and 2,2'-dicyanosubstituted enamine using 10 mol % CuI in the presence of 2 equiv Et_3N (Scheme 15).⁵³ When the reaction

performed in THF at 25 °C, 4-amino-2-iminopyridine derivatives is predominantly formed. On the other hand, 2-amino-2-iminopyridine is preferably formed in DMF at 50 °C under N₂ atmosphere *via* an intramolecular nucleophilic vinylic substitution (Scheme 15).

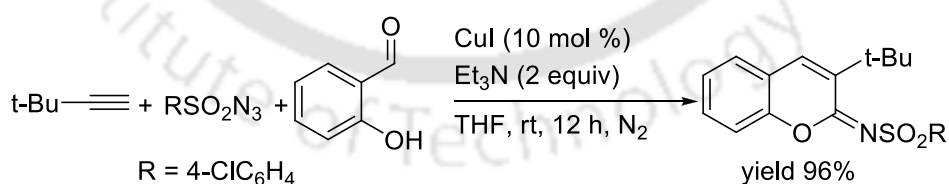
Wang and co-workers reported the synthesis of 2-imino-1,2-dihydroquinoline from terminal alkyne, sulfonyl azide and acetylene using 10 mol % CuI in the presence of 2 equiv Et₃N in CH₃CN under nitrogen atmosphere. This reaction proceeded *via* cascade nucleophilic addition to *in situ* generated ketenimine followed by cyclization (Scheme 16).⁵⁴



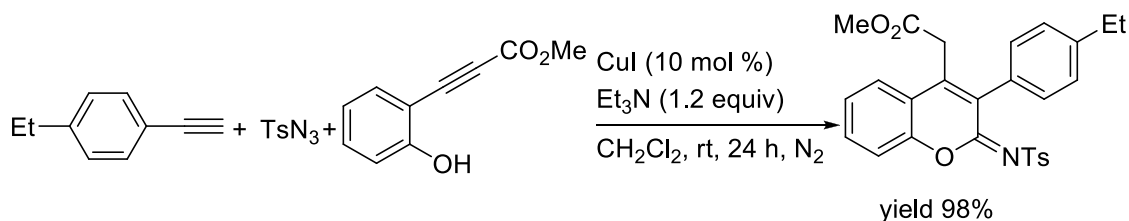
Scheme 16

1.2.2.2 Reaction of Oxygen Nucleophiles

Wang and co-workers developed Cu(I)-catalyzed three-component synthesis of functionalized iminocoumarin derivatives from terminal alkyne, sulfonyl azide and salicylaldehyde (Scheme 17).⁵⁵ The reaction proceeded *via* addition to *in situ* generated ketenimine with oxygen nucleophile followed by cyclization. Later, these authors showed the synthesis of 2-iminocoumarin from 2-hydroxyphenylpropiolates (Scheme 18).⁵⁶

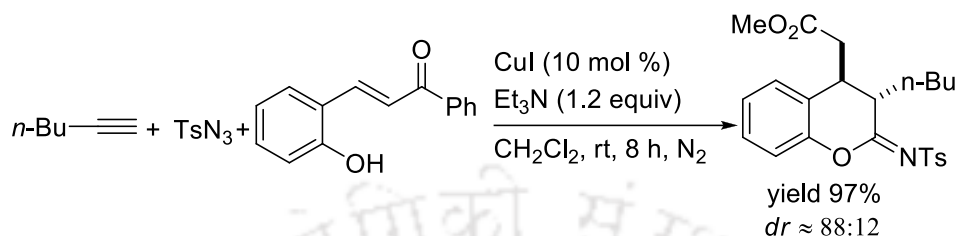


Scheme 17



Scheme 18

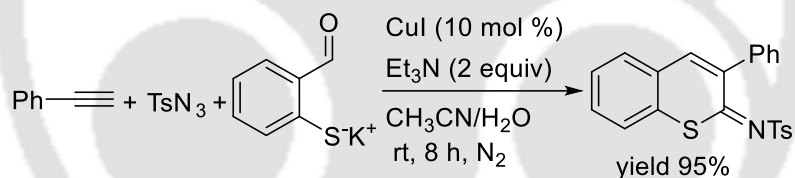
The synthesis of functionalized 2-iminodihydrocoumarin is demonstrated using Cu(I)-catalyzed three-component reaction of 2-hydroxychalcone, terminal alkyne and sulfonyl azide (Scheme 19).⁵⁶ The reaction proceeded *via* cascade trapping of *in situ* generated ketenimine by a nucleophilic addition followed by an intramolecular Michael addition.



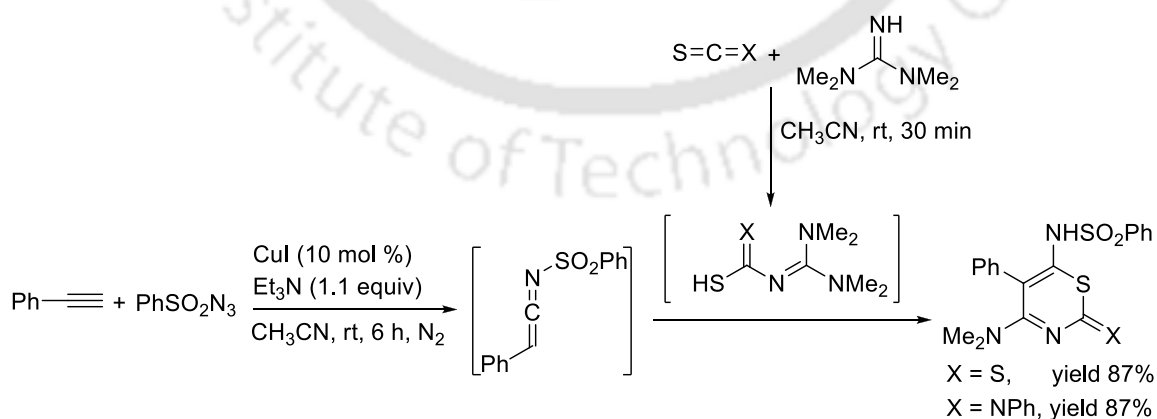
Scheme 19

1.2.1.3 Reaction of Sulphur Nucleophiles

Wang and coworkers reported that the synthesis of 2-iminothiocoumatins were achieved by Cu(I)-catalyzed three component reaction of alkynes, sulfonyl azides and potassium salt of 2-mercapto-benzaldehyde (Scheme 20).⁵⁴ The cascade process involves the trapping of *in situ* generate ketenimine by a thiol nucleophile and the cyclization.



Scheme 20

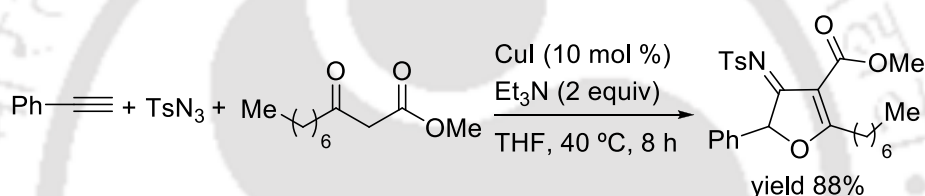


Scheme 21

Yuvari and co-workers reported the synthesis of 2-thioxo-2*H*-1,3-thiazines from alkyne and sulfonyl azide with *in situ* generated adduct of carbondisulphide with tetramethylquanidine using 10 mol % CuI in the presence of 1.1 equiv Et₃N in CH₃CN at room temperature (Scheme 21).⁵⁷ In addition, 2-arylimino-2*H*-1,3-thiazines were also synthesized using the same protocol from *in situ* generated adduct of arylthiocyanate with tetramethylquanidine.

1.2.1.4 Reaction of Carbon Nucleophiles

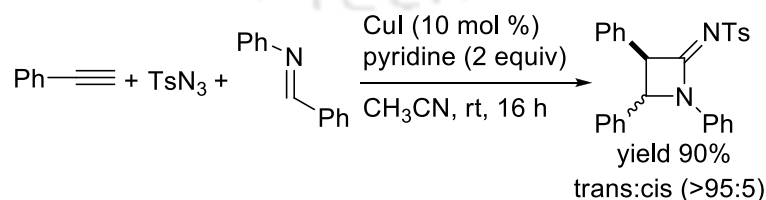
In 2010, Zhang and co-workers reported the synthesis of 4-arylsulfonylimido-4,5-dihydrofuran derivatives from terminal alkyne, sulfonyl azide and alkyl acetoacetate using 10 mol % CuI in the presence of 2 equiv Et₃N in THF (Scheme 22).⁵⁸ The reaction proceeded *via* carbon nucleophilic addition of *in situ* generated ketenimine.



Scheme 22

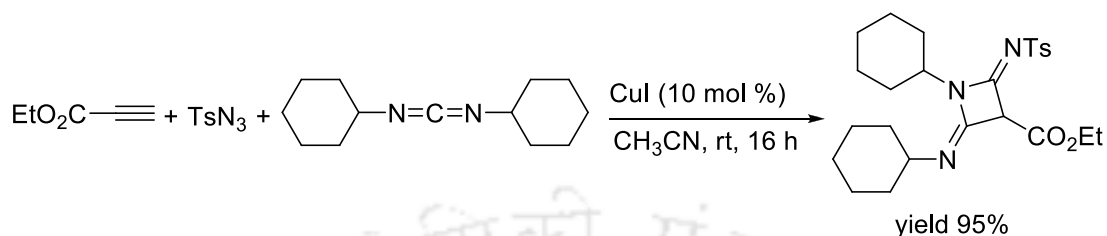
1.2.1.5 [2+2] Cycloaddition Reaction

In 2006, Fokin and co-workers showed the synthesis of diastereoselective azetidines from terminal alkyne, sulfonyl azide and imine using 10 mol % CuI in the presence of pyridine in CH₃CN at room temperature (Scheme 23).⁵⁹ These transformations take place *via* [2+2]-cycloaddition of *in situ* generated ketenimine with imine to afford azetidines with good diastereoselectivity



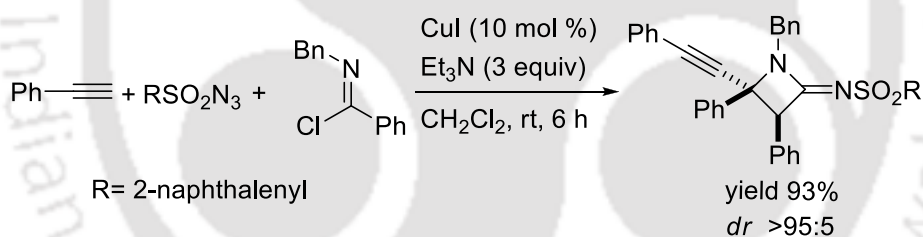
Scheme 23

Later, Xu and co-workers demonstrated the synthesis of 1,4-diimino-azetidines from alkyne, sulfonyl azide and dicyclohexylcarbodiimide using 10 mol % CuI in CH₃CN at room temperature (Scheme 24).⁶ This reaction proceeds *via* [2+2] cycloaddition of ketenimine with DCC to provide the corresponding heterocycles in good to high yields.



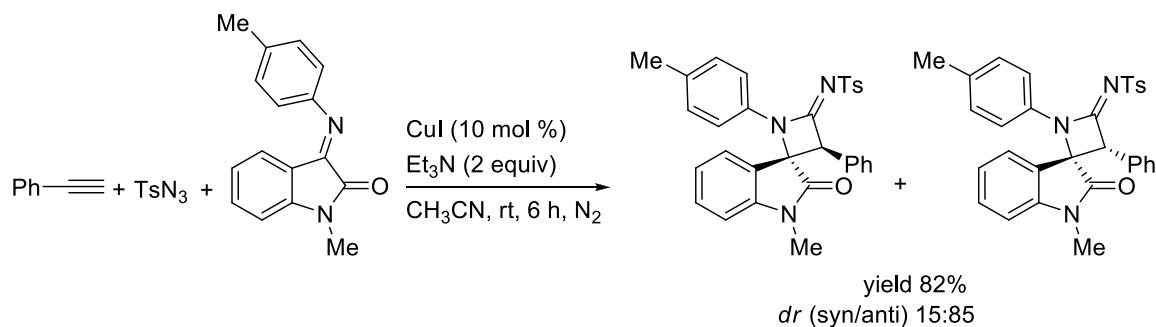
Scheme 24

Synthesis of highly substituted azetidine-2-imine derivatives from sulfonyl azide, alkyne and imidoyl chloride has been accomplished using 10 mol % CuI in the presence Et₃N in CH₂Cl₂ with good yields (Scheme 25).⁶¹ The transformation occurs *via* intermolecular [2+2] cycloaddition of *in situ* generated ketenimine with ynimine to afford the target with high diastereoselectivity



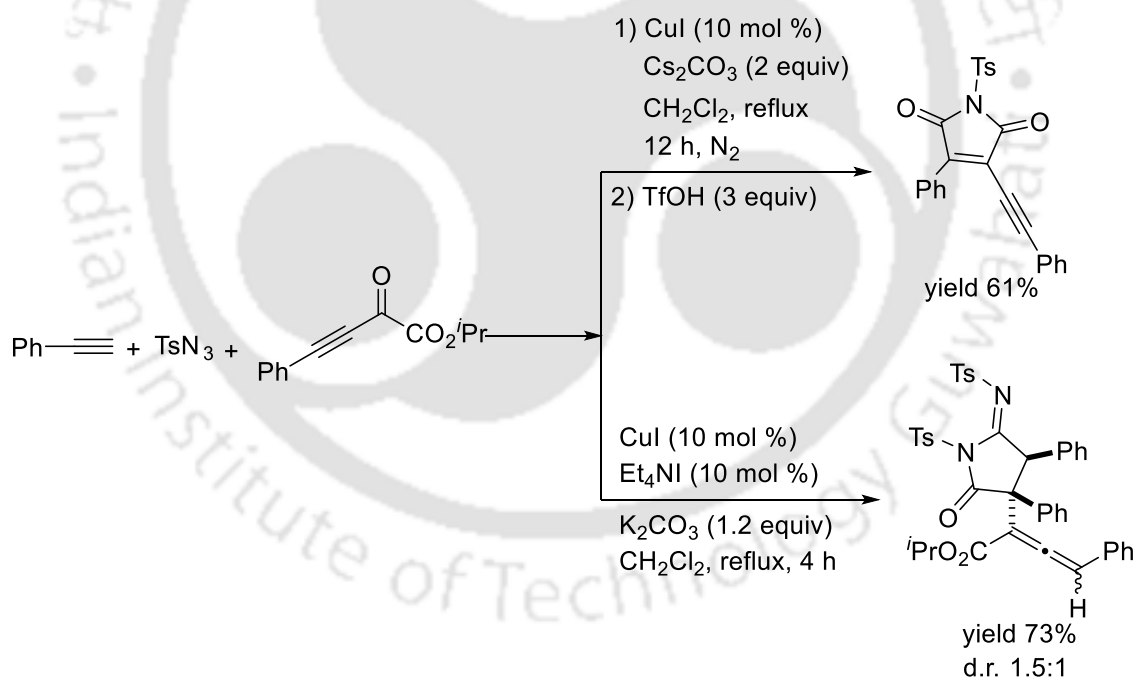
Scheme 25

Shanmugam and co-workers reported the synthesis of 3-spiroazetidinimine-2-oxindole from terminal alkyne, sulfonyl azide and 2-aryliminoindolin-2-one using 10 mol % CuI in the presence of 2 equiv Et₃N in CH₃CN under nitrogen atmosphere (Scheme 26).⁶² The reaction takes place *via* [2+2] cycloaddition of *in situ* generated ketenimine with 2-aryliminoindolin-2-one with high diastereoselectivity.



Scheme 26

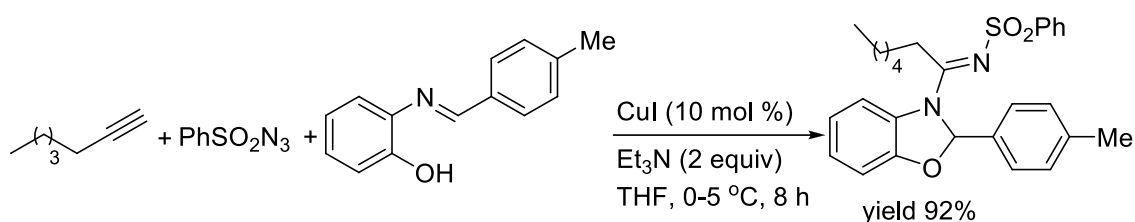
Ma and co-workers developed a new method for the functionalized 2-iminoxetanes from terminal alkynes, sulfonyl azides and aromatic 2-oxobut-3-ynoates using Cu(I)-catalyzed multicomponent *via* cascade [2+2] cycloaddition reaction of aromatic 2-oxobut-3-ynoates with *in situ* generated ketenimine (Scheme 27).⁶³ The skeletal rearrangements of 2-iminoxetane with appropriate reaction condition afford pyrrolidinones and maleimides derivatives.



Scheme 27

Zhang and co-workers reported the synthesis of functionalized bezaxozoline-amidine derivatives from terminal alkyne, sulfonyl azides and Schiff base of salicylaldehyde using 10 mol % CuI in the presence of Et₃N (Scheme 28).⁶⁴ The reaction proceeded *via* a

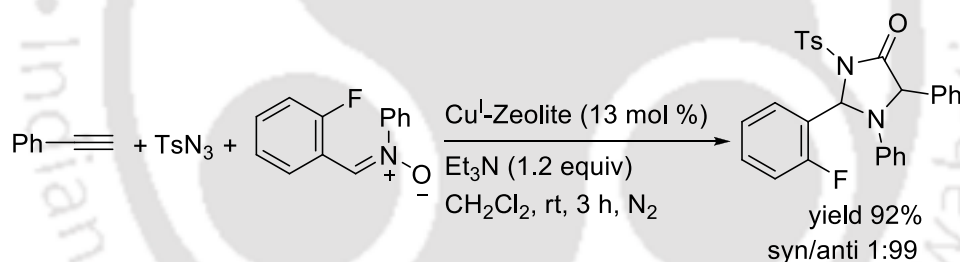
cascade [2+2]-cycloaddition reaction of *in situ* generated ketenimine with Schiff base followed by skeletal reorientation to afford the corresponding heterocycles in good yields.



Scheme 28

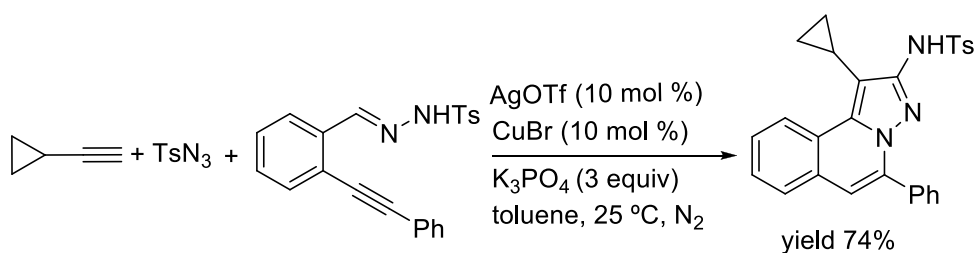
1.2.1.6 [3+2] Cycloaddition Reaction

Pitchumani and co-workers reported the synthesis of functionalized diastereoselective imidazoline-4-ones from alkyne, sulfonyl azide and nitron using 13 mol % Cu(I)-zeolite in the presence of 1.2 equiv Et_3N in CH_2Cl_2 under nitrogen atmosphere (Scheme 29).⁶⁵ This transformation takes place *via* a cascade [3+2] cycloaddition of *in situ* generated ketenimine with nitron followed by skeletal rearrangement.



Scheme 29

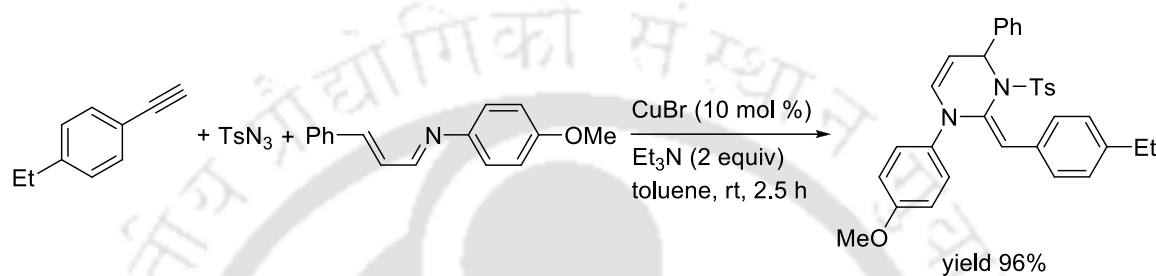
The synthesis of functionalized 2-amino-*H*-pyrazolo[5,1-*a*]-isoquinolines is reported through an intermolecular [3+2]-cycloaddition of *in situ* generated ketenimine with isoquinolinium-2-yl-amide which was obtained in good yields (Scheme 30).⁶⁶



Scheme 30

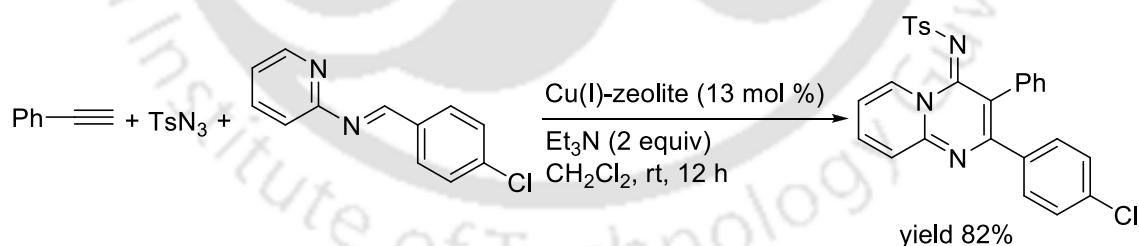
1.2.1.7 [4+2] Cycloaddition Reaction

In 2009, Wang and co-workers reported Cu(I)-catalyzed three-component synthesis of tetrahydropyrimidines from terminal alkynes, sulfonyl azides and α,β -unsaturated imine (Scheme 31).⁶⁷ At room temperature, tetrahydropyrimidine is selectively formed *via* formal [4+2] cycloaddition of *in situ* generated ketenimine with α,β -unsaturated imine. On the other hand, under reflux, [2+2] product is obtained.



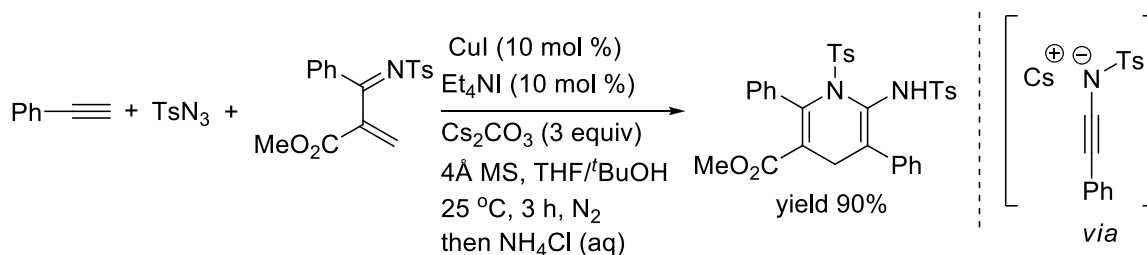
Scheme 31

Pitchumani and co-workers accomplished chemo- and regioselective synthesis of pyrido[1,2-*a*]pyrimidin-4-imines from sulfonyl azide, alkyne and *N*-arylidene-pyridine-2-amine using 13 mol % Cu(I)-Zeolite in the presence of 2 equiv Et₃N in CH₂Cl₂ at room temperature (Scheme 32).⁶⁸ This transformation proceeds *via* formal [4+2] cycloaddition of *in situ* generated ketenimine with *N*-arylidene-pyridine-2-amine.



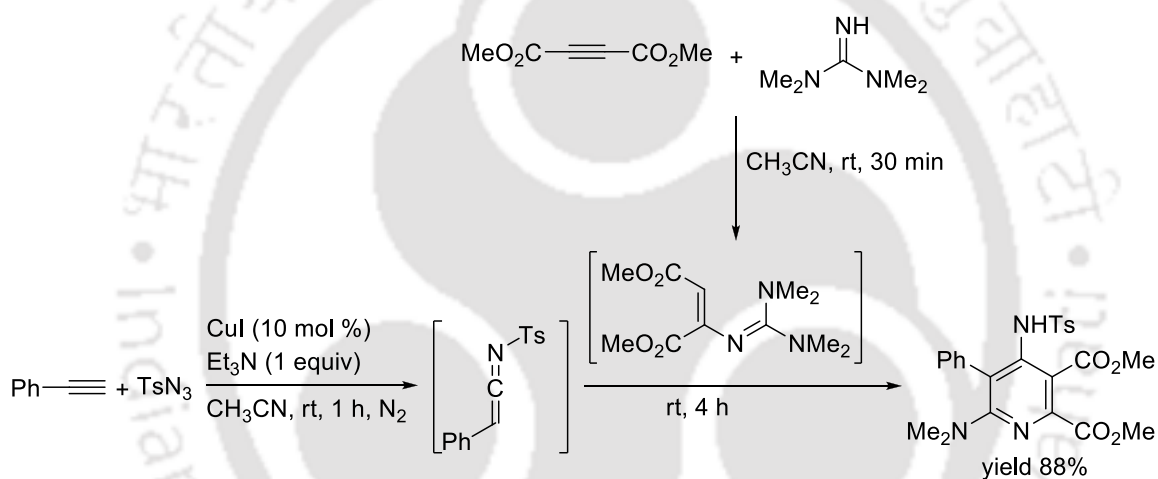
Scheme 32

Ma and co-workers reported a new protocol for the synthesis of dihydropyridones *via* copper catalyzed three-component inverse electron-demand hetero Diels Alder reaction of *in situ* generated cesium stabilized ketenimine intermediate with electron demand conjugate α,β -unsaturated tosylimine (Scheme 33).⁶⁹ This reaction has broad substrate scope with good yields



Scheme 33

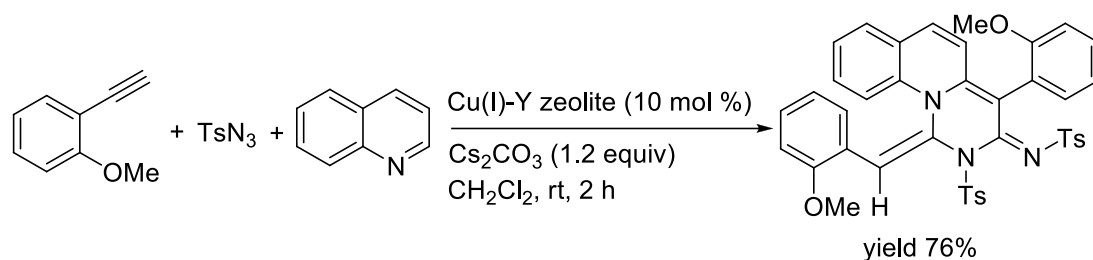
Yuvari and co-workers developed new method to access polysubstituted pyridine from *in situ* generated ketenimine with *in situ* formed stabilized adduct between dialkyl propiolate and quinidine *via* one-pot Cu-catalyzed three-component reaction (Scheme 34).⁷⁰



Scheme 34

1.2.1.8 [2+2+2] Cyclotrimerization Reaction

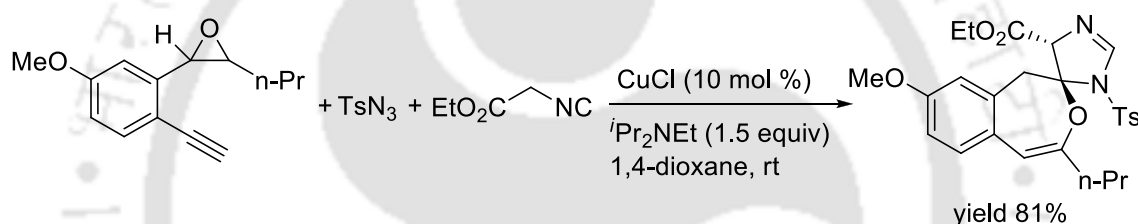
In 2015, Pitchumani and co-workers reported Cu^I-zeolite catalyzed three-component synthesis of pyrimido-[1,6-*a*]quinolone derivatives from terminal alkynes, sulfonyl azides and quinolone using 10 mol % Cu(I)-Y zeolite in the presence of 1.2 equiv Cs₂CO₃ in CH₂Cl₂ at room temperature (Scheme 35).⁷¹ This reaction proceeds *via* tandem [2+2+2] trimerization of two equiv of *in situ* generated ketenimine with one equiv of quinolone to give the corresponding heterocycles with good yields.



Scheme 35

1.2.1.9 Miscellaneous Reactions

Wu and co-workers reported the synthesis of highly functionalized fused spirocycles from 2-(2-ethynylphenyl)oxirane and sulfonyl azide using 10 mol % CuCl in the presence of 1.5 equiv $i\text{Pr}_2\text{NEt}$ in 1,4-dioxane at ambient temperature (Scheme 36).⁷² The protocol has broad substrate scope with good yields.



Scheme 36

In summary, Cu-catalyzed cycloaddition of sulfonyl azides with terminal alkynes furnish reactive ketenimine that can be readily reacted with diverse nucleophiles to afford highly functionalized structural scaffolds. This strategy provides a potential route for the construction of highly functionalized heterocyclic compounds with utilizing suitable coupling partners.

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Copper(I)-Catalyzed Three-Component Cascade Synthesis of Iminocoumarin Aryl Methyl Ethers

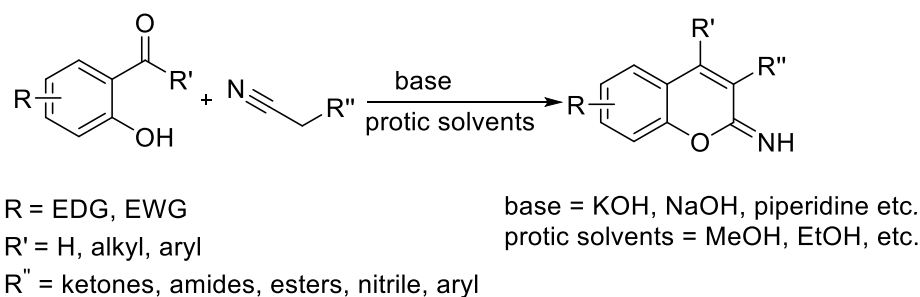
Iminocoumarins are important heterocyclic scaffolds and find broad applications in biological and material sciences. For examples, the compounds having iminocoumarin structural motif exhibit anticoagulant,¹ anticonvulsant,² anti-inflammatory,³ antimicrobial,⁴ antioxidant⁵ and antitumor properties.⁶ In addition, they are found to be inhibitors of lymphocyte protein-tyrosine kinase p56^{lck},⁷ tyrosine kinase p60^{c-src},⁸ dynamin GTPase,⁹ tumour maker AKR1B10,¹⁰ β -secretase (BACE1)¹¹ and human carbonyl reductase 1 (CBR1).¹² Furthermore, iminocoumarins are utilized as dyes¹³ and fluorescent probes for metal sensors,¹⁴ anionic sensors¹⁵ and biological probes.¹⁶ As a result, development of effective methods for the construction of iminocoumarin derivatives is highly desirable.

2.1 Strategies for Synthesis of Iminocoumarins

Iminocoumarins are prepared by various methods such as Knoevenagel condensation, annelation of ketenimine with salicylaldehydes, cascade condensation and copper-catalyzed azide-alkyne cycloaddition reactions.

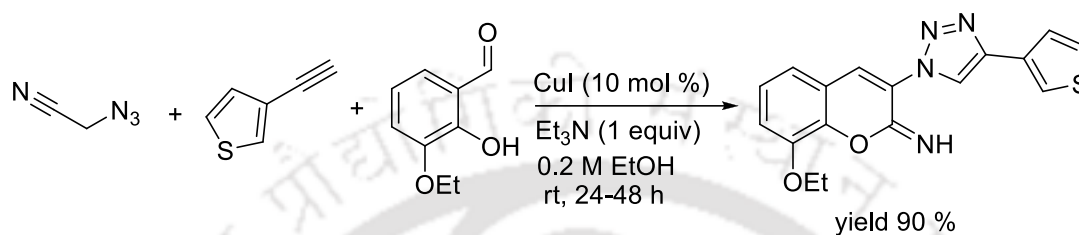
2.1.1 Knoevenagel Condensation

The classical methods for synthesis of iminocoumarin take an advantage of Knoevenagel condensation reaction. In the presence of base such as metal hydroxides, piperidine *etc.*, condensation of salicylaldehydes with active methylene nitrile compounds in protic solvents afford iminocoumarin derivatives (Scheme 1).¹⁷ However, this reaction has limited substrate scope and less functional groups tolerance



Scheme 1

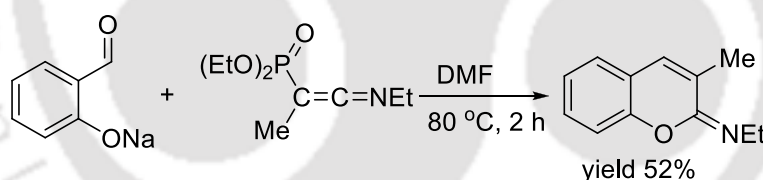
Qian and co-workers described copper-catalyzed three-component synthesis of 3-triazolyl-2-iminocoumarins (Scheme 2).^{18a} In this reaction, Cu(I)-catalyzed azide-alkyne cycloaddition between 2-azidoacetonitrile and acetylene produces a triazole. The activated methylene group of triazole then undergoes an aldol-cyclization-dehydration with salicylaldehyde to furnish iminocoumarin. This reaction has been subsequently extended to the synthesis of glycosylated iminocoumarin derivatives.^{18b}



Scheme 2

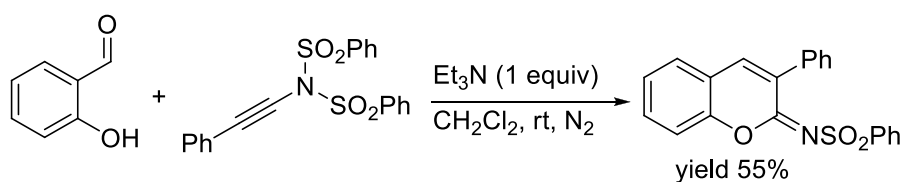
2.1.2 Annulation of Ketenimine

In 1979, Motoyoshiya and co-workers reported the synthesis of iminocoumarin *via* annulation of stable C-phosphono-ketenimine with sodium salt of salicylaldehyde (Scheme 3).¹⁹



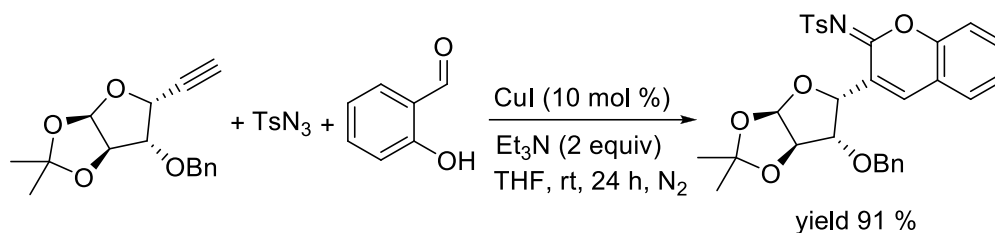
Scheme 3

Cao and co-workers reported Et₃N-mediated synthesis of iminocoumarin from *N,N*-disulfonylaminides and salicylaldehydes (Scheme 4).²⁰ In this reaction, the annulation of reactive ketenimine intermediate with salicylaldehyde affords the target heterocycles.



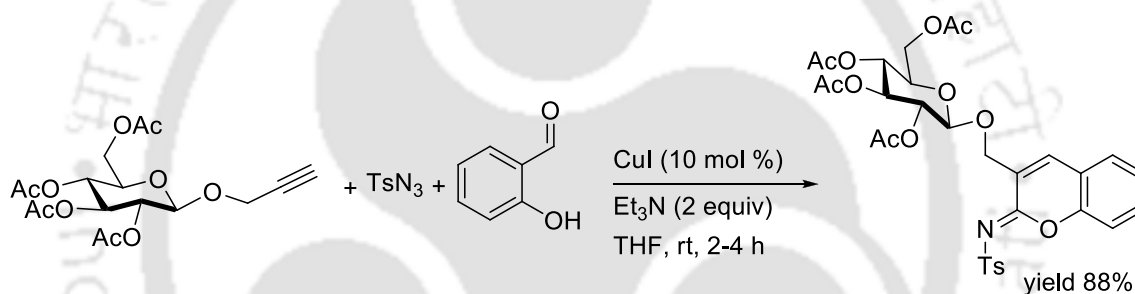
Scheme 4

Kaliappan and co-workers reported Cu(I)-catalyzed MCR of sugar-derived alkynes, tosyl azide and salicylaldehydes to provide 3-C-linked glycosyl iminocoumarin (Scheme 5).^{21a-b}



Scheme 5

In 2014, Mandal extended strategy for the synthesis of glycosylated iminocoumarins *via* Cu(I)-catalyzed MCR of sugar alkynes, sulfonyl azides and salicylaldehydes (Scheme 6).^{18b,22}



Scheme 6

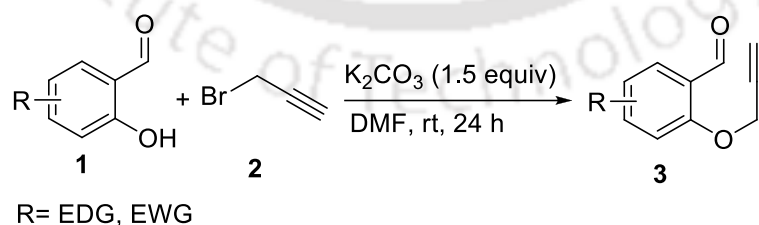
2.2 Present Study

Development of an effective method for synthesis of functionalized iminocoumarins is important in synthetic chemistry. The present study describes the synthesis of iminocoumarins using Cu(I)-catalyzed MCR of ynals, sulfonyl azides and phenols *via* cycloaddition, intramolecular ketenimine rearrangement and cyclization reactions. The ynals **3** were prepared from salicylaldehydes **1** and propargyl bromide **2** (Scheme 7),²³ while sulfonyl azides **5** were synthesized from sulfonyl chlorides **4** and sodium azide (Scheme 8).²⁴

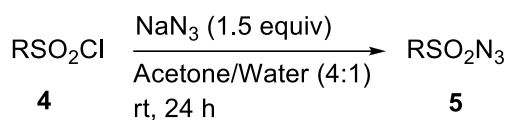
First, the reaction was optimized using ynal **3a**, *p*-toluenesulfonyl azide (TsN₃) **5a** and phenol **6a** as the model substrates in the presence of different Cu(I) sources, bases, additives and solvents at room temperature (Table 1). Gratifyingly, the reaction occurred to provide the iminocoumarin aryl methyl ether **7a** in 30 % yield when the substrates **3a**, **5a** and **6a** stirred with 10 mol % CuI in the presence of 2.2 equiv of K₂CO₃ in CH₂Cl₂ at

ambient conditions (entry 2). In a set of additives screened, tetrabutylammonium iodide (TBAI), tetrabutylammonium bromide (TBAB) and tetrabutylammonium chloride (TBACl), the former afforded the best results (Table 1, entry 3-5).²⁵ Among the copper sources studied, CuI, CuBr, CuCl and Cu₂O, CuI afforded the superior results (Table 1, entries 5, 13-15). In case of solvent, CH₂Cl₂ was found to be the solvent of choice, whereas toluene, THF, CH₃CN and 1,2-dichloroethane afforded inferior results. The control experiment without copper source showed no product formation (Table 1, entry 16).

Next, the substrate scope has been explored towards the iminocoumarin aryl methyl ether libraries using a series of substituted phenols **6a-n** with ynal **3a** and tosyl azide **5a** as standard substrates (Table 2). The target heterocycles were obtained in good yields. For example, when the reactions were performed with the substituted phenols **6a-e** having phenyl, 2-iodo, 3-bromo, 2-methyl and 3-methyl groups, the iminocoumarin aryl methyl ethers **7a-e** were obtained with good yields. The phenols **6f-h** having 4-fluoro, 4-bromo and 4-chloro substituents afforded **7f-h** in 60-63% yields. Similarly, the phenols **6i-k** having 4-formyl, 4-methoxy and 4-methyl substituents produced the target heterocycles **7i-k** in 49-66% yields, whereas the disubstituted phenols **6l-n** with dimethyl groups gave iminocoumarins **7l-n** in 72-77% yields. In addition, the use of methanol instead of phenol produced the iminocoumarin methyl methyl ether **7o** in 10% yield. The reactivity of phenols having electron donating substituents showed greater reactivity than electron withdrawing groups. Recrystallization of **7j** in a 1:1 mixture of CH₂Cl₂ and MeOH, produced single crystals whose structure was confirmed by single crystal X-ray analysis (Figure 1).



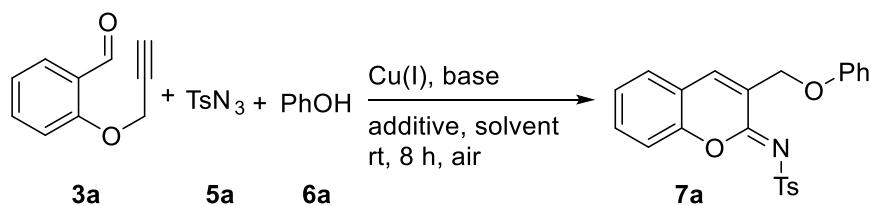
Scheme 7



R = alkyl and aryl

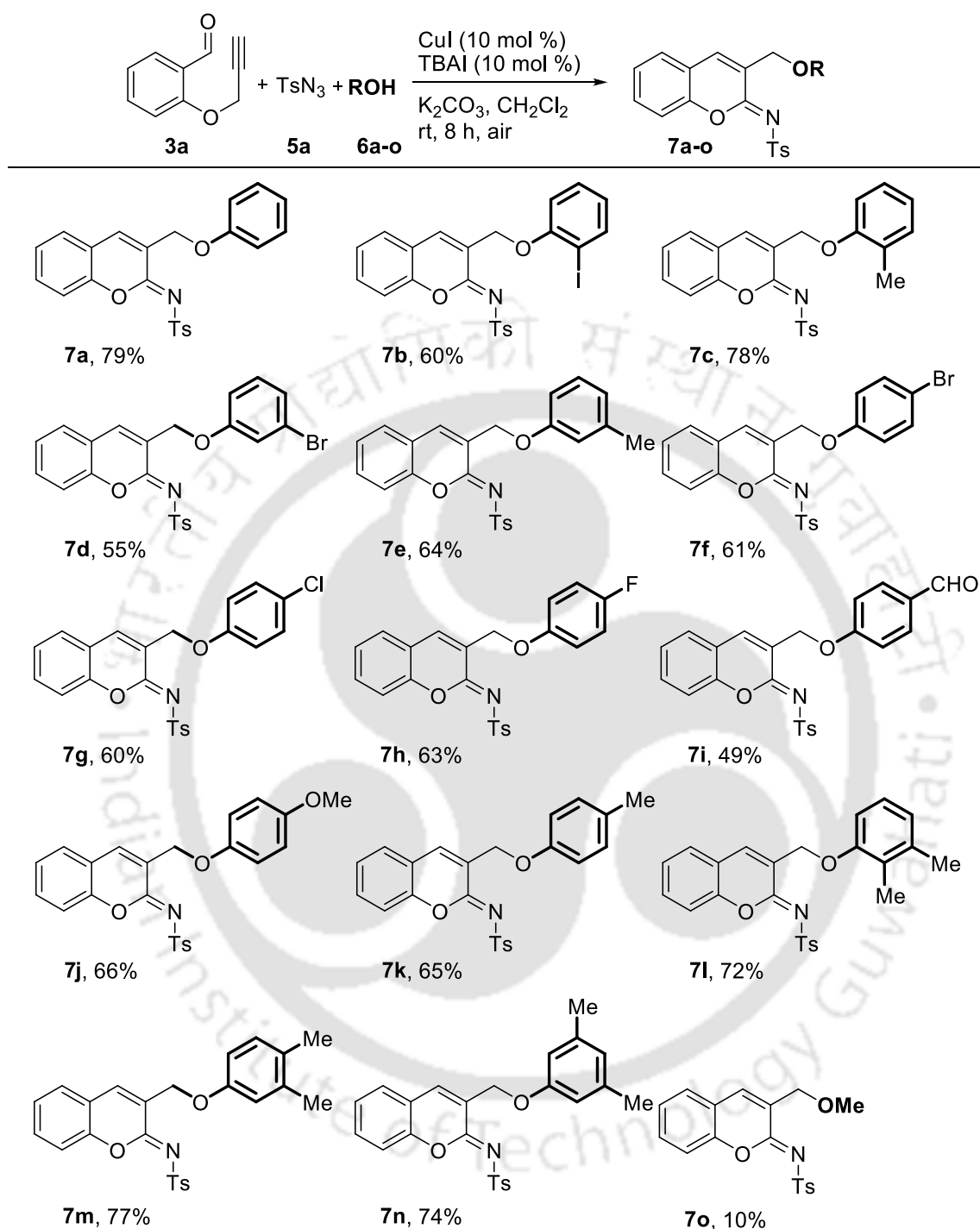
Scheme 8

Table 1. Optimization of the Reaction Conditions^a



Entry	Catalyst	Base	Additive	Solvent	Yield (%) ^b
1	CuI	Et ₃ N	-	CH ₂ Cl ₂	n.d.
2	CuI	K ₂ CO ₃	-	CH ₂ Cl ₂	30
3	CuI	K ₂ CO ₃	TBAB	CH ₂ Cl ₂	76
4	CuI	K ₂ CO ₃	TBACl	CH ₂ Cl ₂	71
5	CuI	K₂CO₃	TBAI	CH₂Cl₂	83
6	CuI	K ₂ CO ₃	TBAI	CH ₃ CN	42
7	CuI	K ₂ CO ₃	TBAI	(CH ₂ Cl) ₂	74
8	CuI	K ₂ CO ₃	TBAI	THF	28
9	CuI	K ₂ CO ₃	TBAI	toluene	60
10	CuI	Na ₂ CO ₃	TBAI	CH ₂ Cl ₂	35
11	CuI	Cs ₂ CO ₃	TBAI	CH ₂ Cl ₂	n.d.
12	CuI	K ₃ PO ₄	TBAI	CH ₂ Cl ₂	n.d.
13	CuBr	K ₂ CO ₃	TBAI	CH ₂ Cl ₂	67
14	CuCl	K ₂ CO ₃	TBAI	CH ₂ Cl ₂	71
15	Cu ₂ O	K ₂ CO ₃	TBAI	CH ₂ Cl ₂	41
16	-	K ₂ CO ₃	TBAI	CH ₂ Cl ₂	n.d.

^aYnal **3a** (0.5 mmol), tosyl azide **5a** (0.6 mmol), phenol **6a** (0.6 mmol), catalyst (10 mol %), base (1.1 mmol), additive (10 mol %), solvent (3.0 mL), 8 h, air. ^bDetermined by 400 MHz ¹H NMR. n.d. = not detected.

Table 2. Substrate scope of the Reaction with Substituted Phenols^{a,b}


^aYnal **3a** (0.5 mmol), substrate **6** (0.6 mmol), tosyl azide **5a** (0.6 mmol), CuI (10 mol %), K₂CO₃ (1.1 mmol), TBAI (10 mol %), CH₂Cl₂ (3.0 mL), 8 h, air. ^bIsolated yield.

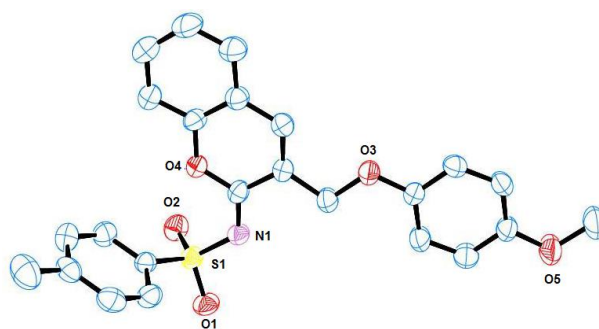
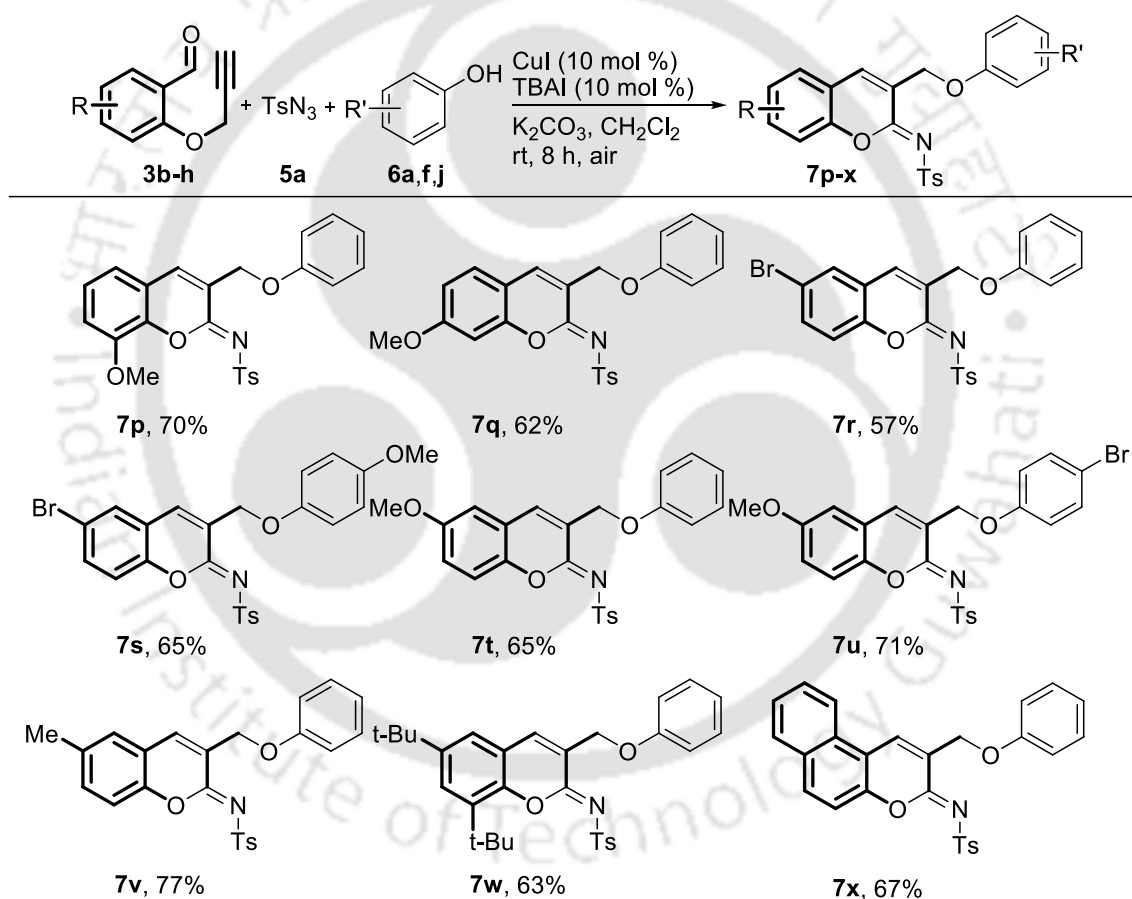


Figure 1. ORTEP diagram of **7j**. Thermal ellipsoids are drawn at a 50% probability level. Hydrogen atoms have omitted for clarity.²⁶

Table 2.3. Reaction of Tosyl azide with different substituted Ynals and Phenols^a

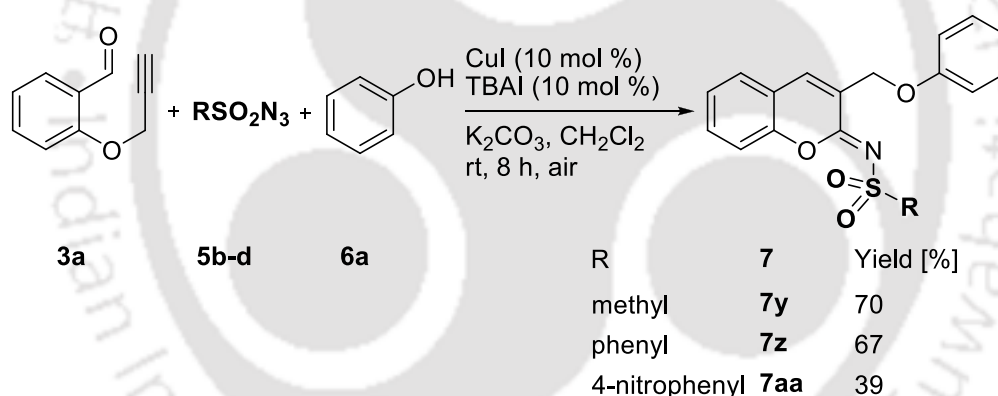


^aYnal **3b-h** (0.5 mmol), phenol **6a,f,j** (0.6 mmol), tosyl azide **5a** (0.6 mmol), CuI (10 mol %), K_2CO_3 (1.1 mmol), TBAI (10 mol %), CH_2Cl_2 (3.0 mL), 8 h, air. ^bIsolated yield.

The substrate scope of reaction was further studied with various substituted ynals **3b-g** and phenols **6a**, **6f** and **6j** and *p*-toluenesulfonyl azide **5a** (Table 3). For example, the ynals **3b-c** containing 3-methoxy and 4-methoxy substituents underwent reactions with phenol **6a** to afford **7p** and **7q** in 70% and 62% yields, respectively. Likewise, the ynal

3d with 4-bromo substituent proceeded reaction with phenol **6a** and 4-methoxy phenol **6j** to furnish the iminocoumarins **7r** and **7s** in 57% and 65 % yields, respectively. The 5-methoxy substituted propargyl ether **3e** reacted with both phenol **6a** and 4-bromophenol **6f** to produce **7t** and **7u** in 65% and 71% yields, respectively. Similarly, the ynals **3f-g** with 5-methyl and 3,4-di-*tert*-butyl groups afforded the corresponding iminocoumarin aryl ethers **7v-w** in 63-77% yields. The reaction of ynal **3h** bearing fused aromatic ring with phenol provided the fused heterocycle **7x** in 67% yield.

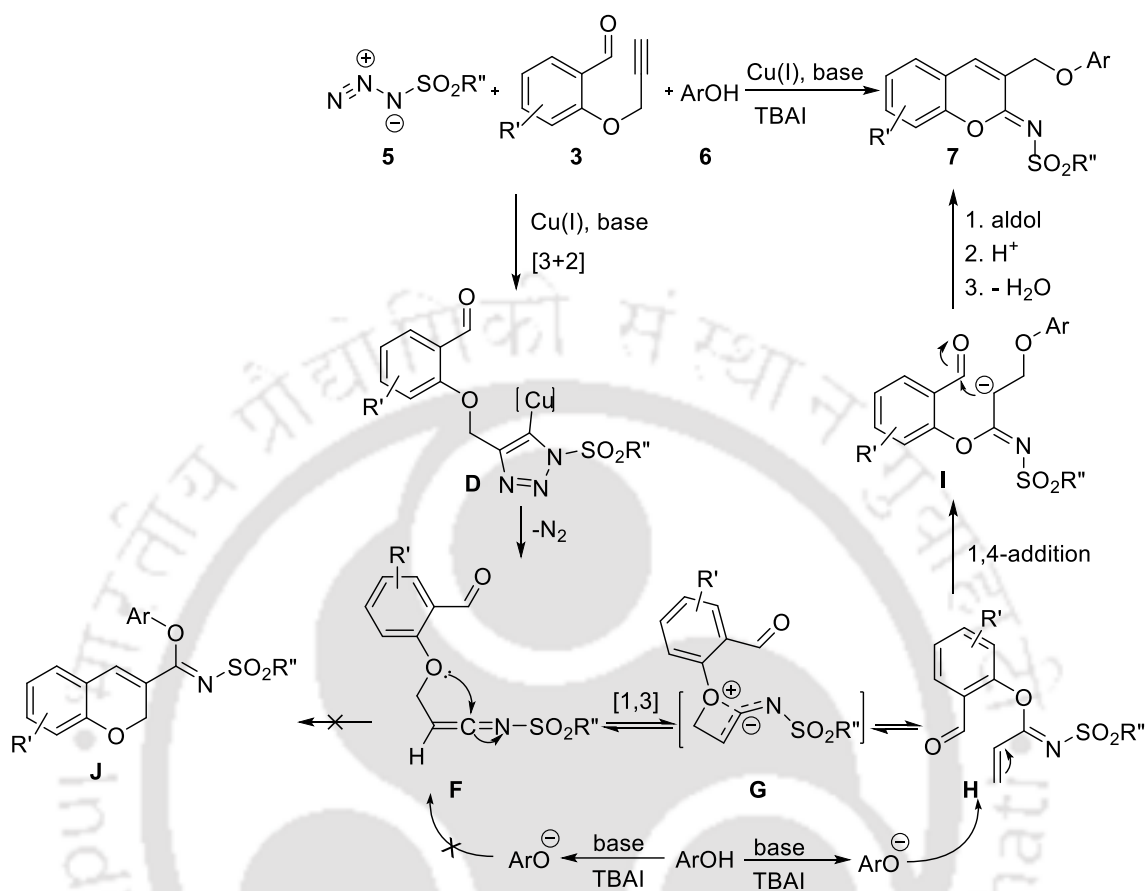
Finally, the reaction of different sulfonyl azides **5b-d** bearing methyl, phenyl and 4-nitro-phenyl functionalities was studied with ynal **3a** and phenol **6a** (Scheme 9). The reaction readily took place to afford the target products in moderate to good yield. The methanesulfonyl azide **5b** and phenylsulfonyl azide **5c** afforded the targets **7y-z** with 70% and 67% yields, respectively. Subsequently, the arylsulfonyl azide bearing 4-nitro substituents showed moderate reactivity furnishing the iminocoumarin aryl methyl ether **7aa** in 39% yield.



Scheme 9

The proposed catalytic cycle is shown in Scheme 10. The Cu(I)-catalyzed azide-alkyne cycloaddition (CuAAC) of alkyne **3** with azide **5** may generate ketenimine⁵ **F** via intermediate **D**, which may undergo the pseudopericyclic [1,3]-migration²⁷ of 2-formylaryloate through the four membered cyclic zwitterionic transition state **G** to give an intermediate **H**. The 1,4-conjugate addition of the phenoxide ion with **H** may lead to the formation of the intermediate **I**. The aldol-type condensation followed by dehydration of **I** could yield the target product **7**. The absence of formation of **J** suggests that this protocol involves the rearrangement of ketenimine **F** to afford **H** which was

compared to the direct intermolecular reaction of ketenimine **F** with the phenoxide ion that could lead to **J**



Scheme 10

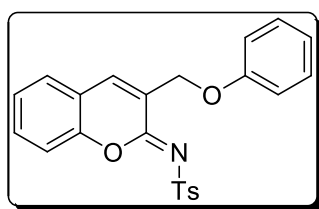
In summary, the synthesis of highly functionalized iminocoumarin aryl methyl ethers has been accomplished using copper-catalyzed three-component reaction of ynal, phenols and sulfonyl azide at ambient conditions *via* a cascade [3+2]-cycloaddition, 1,3-pseudopericyclic ketenimine rearrangement, 1,4-conjugate addition and aldol-type condensation. This protocol is simple and has wide substrate scopes for the construction of a library of iminocoumarin aryl methyl ethers in good yields, which may find broad applications in medicinal and material sciences.

2.3 Experimental Section

General Information. 1H and ^{13}C NMR spectra were recorded on DRX-400 Varian and Bruker UltrashieldTM 300 spectrometers using $CDCl_3$ as solvent and the data are accounted as follows: chemical shifts (δ ppm) (multiplicity, coupling constant (Hz),

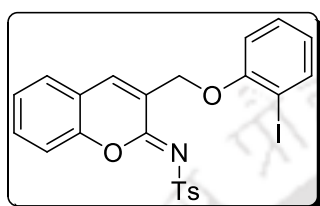
integration). The abbreviations for multiplicity are as follows: s = singlet, d = doublet, t = triplet, m = multiplet and dd = doublet of doublets. Infrared (IR) spectra recorded on a Perkin Elmer Spectrum one FT-IR spectrometer. Melting points were determined with a Büchi B-540 apparatus and are uncorrected. HRMS mass were analyzed with Agilent Q-TOF 6500. For single crystal X-ray analysis, the intensity data were collected using Bruker SMART APEX-II CCD diffractometer, equipped with 1.75 kW sealed-tube Mo-K α irradiation ($\lambda = 0.71073 \text{ \AA}$) at 298(2) K and the structures were solved by direct methods using SHELXL-97 (Göttingen, Germany) and refined with full-matrix least squares on F² using SHELXL-97. Phenols, CuI (98%), CuBr (98%), Cu₂O (97%), Na₂CO₃, K₂CO₃, Cs₂CO₃, and K₃PO₄ were purchased from Aldrich and used without further purification. Propargyl bromide 80 wt % in toluene was purchased from ACROS Organics and used as received. TBAI, TBAB and TBACl were purchased from Spectrochem and used without further purification. Solvents were purchased from Merck and distilled by the standard protocol and stored over molecular sieves under nitrogen atmosphere prior to use.²⁸ The reactions were monitored by analytical TLC on Merck silica gel G/GF 254 plates. The column chromatography was performed with Rankem silica gel (60-120 mesh).

General Procedure for the Synthesis of Iminocoumarin Methyl Aryl Ethers. To a stirred solution of ynal **3** (0.5 mmol), sulfonyl azide **5** (0.6 mmol), phenol **6** (0.6 mmol) and CuI (9.5 mg, 10 mol %, 0.05 mmol) in CH₂Cl₂ (3 mL) using CaCl₂ guard tube under air, K₂CO₃ (152 mg, 1.1 mmol) and TBAI (18.5 mg, 10 mol %, 0.05 mmol) were added. The reaction was stirred at room temperature for the appropriate time. Progress of the reaction was monitored by TLC using ethyl acetate and hexane as eluent. The reaction mixture was then diluted using CH₂Cl₂ (30 mL) and washed with brine (1 x 10 mL) and water (2 x 10 mL). Drying (Na₂SO₄) and evaporation of the solvent gave a residue that was purified by silica gel column chromatography using hexane and ethyl acetate as eluent.

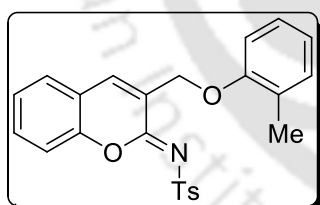


(Z)-4-Methyl-N-(3-(phenoxy)methyl)-2H-chromen-2-ylidenebenzenesulfonamide (7a): colourless solid; yield: 79%; mp 192-193 °C. ¹H

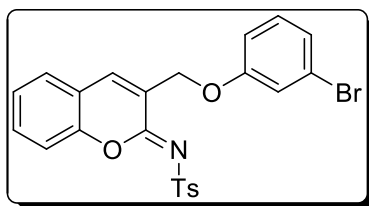
NMR (400 MHz, CDCl₃) δ 8.01 (d, J = 7.6 Hz, 2H), 7.89 (s, 1H), 7.55-7.49 (m, 2H), 7.42 (d, J = 8 Hz, 1H), 7.34-7.27 (m, 5H), 6.98 (d, J = 7.2, 3H), 4.93 (s, 2H), 2.39 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 157.8, 156.0, 151.9, 143.5, 138.8, 137.5, 131.9, 129.7, 129.4, 128.4, 127.6, 125.9, 125.8, 121.6, 119.1, 116.3, 114.8, 64.1, 21.6; FT-IR (KBr) 3053, 2923, 2857, 1632, 1558, 1497, 1459, 1410, 1316, 1303, 1290, 1246, 1209, 1158, 1085, 1046, 824 cm⁻¹. HRMS (ESI): Calcd for C₂₃H₁₉NO₄S [M+H]⁺: 406.1108, found: 406.1109.



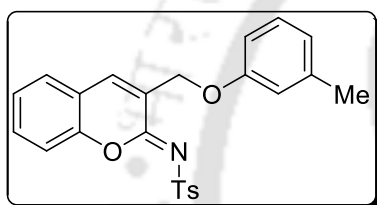
(Z)-N-(3-((2-Iodophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7b): colourless solid; yield: 60%; mp 207-208 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.18 (s, 1H), 8.02 (d, J = 7.6 Hz, 2H), 7.76 (d, J = 7.6 Hz, 1H), 7.56-7.51 (m, 2H), 7.40 (d, J = 8.0 Hz, 1H), 4.93 (s, 2H), 2.39 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 156.3, 155.9, 152.1, 143.7, 139.6, 138.7, 138.4, 132.1, 129.9, 129.5, 128.7, 127.8, 126.0, 125.4, 123.6, 119.3, 116.5, 112.8, 86.4, 65.3, 21.7; FT-IR (KBr) 3056, 2919, 1631, 1554, 1448, 1314, 1289, 1214, 1156, 1085, 836 cm⁻¹. HRMS (ESI): Calcd for C₂₃H₁₈NO₄SI [M+H]⁺: 532.0074, found: 532.0082.



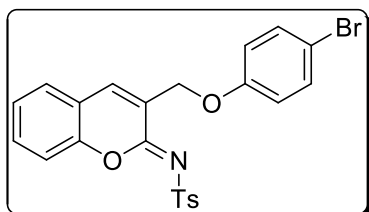
(Z)-4-Methyl-N-(3-(o-tolyloxymethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7c): brown solid; yield: 78%; mp 210-211 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.00 (d, J = 7.6 Hz, 2H), 7.88 (s, 1H), 7.54-7.50 (m, 2H), 7.44 (d, J = 8.0 Hz, 1H), 7.31-7.30 (m, 3H), 7.16-7.11 (m, 2H), 6.91-6.84 (m, 2H), 4.93 (s, 2H), 2.40 (s, 3H), 2.32 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 156.1, 156.0, 152.1, 143.6, 138.9, 137.3, 132.0, 131.0, 129.4, 128.4, 127.7, 127.2, 126.8, 126.4, 126.0, 121.4, 119.3, 116.5, 111.8, 64.4, 21.7, 16.5; FT-IR (KBr) 3060, 2922, 1633, 1559, 1497, 1447, 1320, 1289, 1248, 1157, 1086, 830 cm⁻¹. HRMS (ESI): Calcd for C₂₄H₂₁NO₄S [M+H]⁺: 420.1264, found; 420.1269.



(Z)-N-(3-((3-Bromophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7d): colourless solid; yield 55%; mp 183-184 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.00 (d, $J = 7.6$ Hz, 2H), 7.86 (s, 1H), 7.56-7.50 (m, 2H), 7.42(d, $J = 8.0$ Hz, 1H), 7.35-7.30 (m, 3H), 7.15 (d, $J = 11.2$ Hz, 2H), 6.91 (d, $J = 6.8$ Hz, 1H), 4.90 (s, 2H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 158.6, 156.0, 152.1, 143.7, 138.7, 137.7, 132.2, 130.9, 129.5, 128.5, 127.7, 126.0, 125.4, 124.9, 123.1, 119.1, 118.4, 116.5, 113.6, 64.4, 21.7; FT-IR (KBr) 3065, 2962, 1631, 1556, 1476, 1453, 1409, 1309, 1287, 1232, 1210, 1155, 1085, 1041, 829 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{23}\text{H}_{18}\text{BrNO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 484.0213, found: 484.0221.

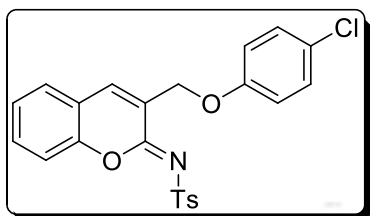


(Z)-4-Methyl-N-(3-(*m*-tolylloxymethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7e): colourless solid; yield: 64%; mp 198-199 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.01 (d, $J = 8.0$ Hz, 2H), 7.89 (s, 1H), 7.55-7.49 (m, 2H), 7.42 (d, $J = 8.0$ Hz, 1H), 7.34-7.30 (m, 3H), 7.18 -7.14 (m, 1H), 6.81 (t, $J = 8.8$ Hz, 3H), 4.92 (s, 2H), 2.40 (s, 3H), 2.32 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 156.1, 152.0, 143.6, 139.9, 138.8, 137.4, 132.0, 129.5, 129.4, 128.4, 127.7, 126.2, 126.0, 122.6, 119.2, 116.4, 115.6, 111.9, 64.1, 21.7, 21.6; FT-IR (KBr) 3054, 2923, 1631, 1614, 1557, 1488, 1450, 1406, 1316, 1288, 1258, 1207, 1155, 1085, 1057, 829 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 420.1264, found: 420.1278.

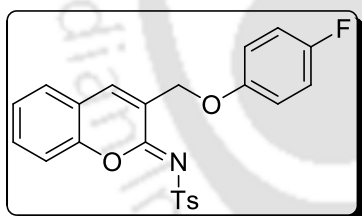


(Z)-N-(3-((4-Bromophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7f): colourless solid; yield: 61%; mp 231-232 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.00 (d, $J = 8.0$ Hz, 2H), 7.85 (s, 1H), 7.56-7.49 (m, 2H), 7.43-7.27 (m, 6H), 6.86 (d, $J = 8.4$ Hz, 2H), 4.90 (s, 2H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$

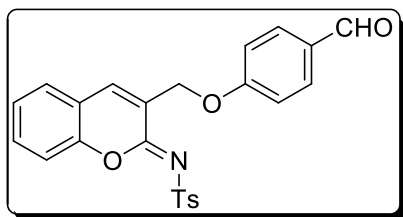
NMR (100 MHz, CDCl₃) δ 157.0, 156.1, 152.2, 143.8, 138.8, 137.6, 132.7, 132.2, 129.5, 128.5, 127.8, 126.1, 125.6, 119.2, 116.8, 116.7, 114.1, 64.6, 22.1; FT-IR (KBr) 3056, 2962, 2919, 1632, 1550, 1488, 1457, 1319, 1290, 1247, 1208, 1159, 1082, 1040, 826 cm⁻¹. HRMS (ESI): Calcd for C₂₃H₁₈BrNO₄S [M+H]⁺: 484.0213, found: 484.0235.



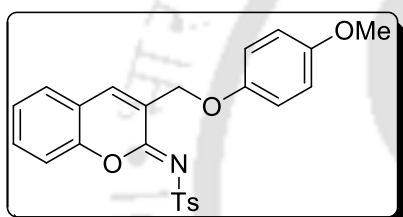
(Z)-N-(3-((4-Chlorophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7g): pale yellow solid; yield: 60%; mp 230-231 °C. ¹H NMR (400 MHz, CDCl₃) δ 7.99 (d, *J*=8.4 Hz, 2H), 7.85 (s, 1H), 7.55-7.49 (m, 2H), 7.42 (d, *J*=8.4 Hz, 1H), 7.34-7.29 (m, 3H), 7.23 (d, *J*=6.4 Hz, 2H), 6.90 (d, *J*=8.4 Hz, 2H), 4.89 (s, 2H), 2.39 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) 156.5, 156.1, 152.1, 143.7, 138.7, 137.6, 132.2, 129.7, 129.5, 128.4, 127.8, 126.7, 126.1, 125.7, 119.2, 116.6, 116.2, 64.6, 21.8; FT-IR (KBr) 3082, 3057, 2962, 2923, 1632, 1551, 1491, 1457, 1318, 1288, 1246, 1208, 1159, 1083, 1041, 827 cm⁻¹. HRMS (ESI): Calcd for C₂₃H₁₈ClNO₄S [M+H]⁺: 440.0718, found: 440.0721.



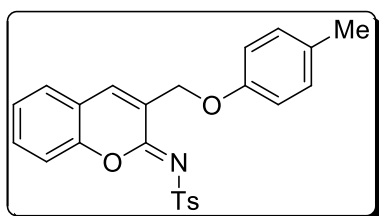
(Z)-N-(3-((4-Fluorophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7h): colourless solid; yield 63%; mp 218-219 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.00 (d, *J*=8 Hz, 2H), 7.87 (s, 1H), 7.55-7.49 (m, 2H), 7.41 (d, *J*=8.0 Hz, 1H), 7.34-7.29 (m, 3H), 6.98-6.89 (m, 4H), 4.88 (s, 2H), 2.39 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 159.1, 156.1, 154.1, 152.2, 143.7, 138.9, 137.5, 132.1, 129.5, 128.4, 127.8, 126.0, 119.2, 116.6, 116.3, 116.1 (d, *J*_{C-F}=3.8 Hz), 65.1, 21.7; FT-IR (KBr) 3049, 2978, 2923, 1633, 1601, 1550, 1507, 1451, 1412, 1317, 1306, 1288, 1249, 1209, 1160, 1085, 1043, 828 cm⁻¹. HRMS (ESI): Calcd for C₂₃H₁₈FNO₄S [M+H]⁺: 424.1013, found: 424.1021.



(Z)-N-(3-((4-Formylphenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7i): colourless solid; yield: 49%; mp 224-225 °C. ¹H NMR (400 MHz, CDCl₃) δ 9.89 (s, 1H), 8.00 (d, *J* = 8.0 Hz, 2H), 7.87 (s, 1H), 7.86 (d, *J* = 8.8 Hz, 2H), 7.58-7.51 (m, 2H), 7.44 (d, *J* = 8.0 Hz, 1H), 7.36-7.30 (m, 3H), 7.11 (d, *J* = 8.0 Hz, 2H), 5.02 (s, 2H), 2.40 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 191.0, 162.7, 156.1, 152.3, 143.9, 138.7, 137.8, 132.4, 130.9, 129.5, 128.6, 127.9, 126.1, 125.1, 119.1, 116.7, 115.4, 64.2, 21.8; FT-IR (KBr) 2956, 2921, 1688, 1632, 1600, 1549, 1458, 1415, 1318, 1252, 1211, 1157, 1083, 1035, 825 cm⁻¹. HRMS (ESI): Calcd for C₂₄H₁₉NO₅S [M+H]⁺: 434.1057, found: 434.1059.

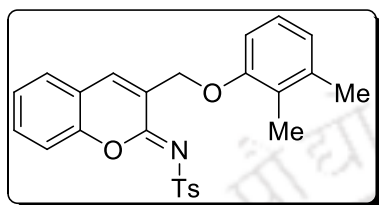


(Z)-N-(3-((4-Methoxyphenoxy)methyl)-2H-chromen-2-ylidene)-4-ethylbenzenesulfonamide (7j): colourless solid; yield: 66%. mp 209-210 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.00 (d, *J* = 8.4 Hz, 2H), 7.89 (s, 1H), 7.52-7.48 (m, 2H), 7.41 (d, *J* = 8.0 Hz, 1H), 7.33-7.29 (m, 3H), 6.91 (d, *J* = 9.2 Hz, 2H), 6.82 (d, *J* = 9.2 Hz, 2H), 4.87 (s, 2H), 3.7 (s, 3H), 2.39 (s, 3H). ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 156.2, 154.6, 152.2, 143.6, 139.0, 137.5, 132.0, 129.5, 128.4, 127.7, 126.4, 126.0, 119.4, 116.6, 116.0, 115.0, 65.1, 55.9, 21.7. FT-IR (KBr) 3057, 2958, 2923, 1637, 1556, 1453, 1316, 1288, 1232, 1208, 1159, 1082, 1047, 821 cm⁻¹. HRMS (ESI): Calcd for C₂₄H₂₁NO₅S [M+H]⁺: 436.1213, found: 436.1204.



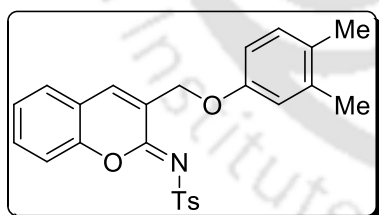
(Z)-4-Methyl-N-(3-(p-tolyloxymethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7k): colourless solid; yield: 65%; mp 217-218 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.01 (d, *J* = 7.2 Hz, 2H), 7.88 (s, 1H), 7.51-7.47 (m, 2H),

7.40 (d, $J = 8.0$ Hz, 1H), 7.31 (d, $J = 7.6$ Hz, 3H), 7.07 (d, $J = 7.6$ Hz, 2H), 6.86 (d, $J = 7.6$ Hz, 2H), 4.89 (s, 2H), 2.39 (s, 3H), 2.26 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.0, 155.7, 151.9, 143.5, 138.7, 131.5, 130.9, 130.0, 129.3, 128.3, 127.5, 126.0, 125.9, 119.1, 116.3, 114.6, 64.2, 21.5, 20.4; FT-IR (KBr) 3075, 2919, 2899, 2858, 1630, 1548, 1450, 1321, 1292, 1244, 1209, 1155, 1145, 1082, 1050, 826 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 420.1264, found: 420.1264.



(Z)-N-(3-((2,3-Dimethylphenoxy)methyl)-2H-chromen-

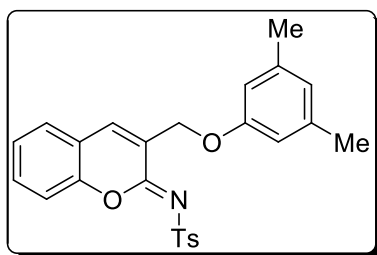
2-ylidene)-4-methylbenzenesulfonamide (7l): colourless solid; yield: 72%; mp 217-218 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.01 (d, $J = 8.0$ Hz, 2H), 7.89 (s, 1H), 7.55-7.51 (m, 2H), 7.43 (d, $J = 8.4$ Hz, 1H), 7.35-7.30 (m, 3H), 7.04 (t, $J = 8.0$ Hz, 1H), 6.82 (d, $J = 7.6$ Hz, 1H), 6.73 (d, $J = 8.4$ Hz, 1H), 4.91 (s, 2H), 2.40 (s, 3H), 2.27 (s, 3H), 2.23 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.2, 155.8, 152.1, 143.6, 138.9, 138.4, 137.2, 132.0, 129.4, 128.4, 127.7, 126.6, 126.2, 125.9, 125.3, 123.3, 119.3, 116.6, 109.8, 64.7, 21.7, 20.2, 12.1; FT-IR (KBr) 2922, 1634, 1559, 1450, 1410, 1315, 1263, 1207, 1156, 1089, 835 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{25}\text{H}_{23}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 434.1421, found: 434.1427.



(Z)-N-(3-((3,4-Dimethylphenoxy)methyl)-2H-chromen-

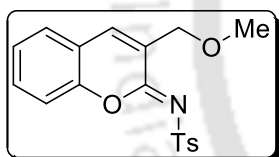
2-ylidene)-4-methylbenzenesulfonamide (7m): pale pink solid; yield: 77%; mp 202-203 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.02 (d, $J = 8.0$ Hz, 2H), 7.89 (s, 1H), 7.53-7.48 (m, 2H), 7.41 (d, $J = 8.0$ Hz, 1H), 7.32 (d, $J = 7.6$ Hz, 2H), 7.03 (d, $J = 8.4$ Hz, 1H), 6.79 (s, 1H), 6.71 (d, $J = 8.0$ Hz, 1H), 4.90 (s, 2H), 2.40 (s, 3H), 2.22 (s, 3H), 2.17 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.2, 156.0, 152.0, 143.6, 138.8, 138.1, 137.4, 131.9, 130.6, 129.7, 129.4, 128.4, 127.7, 126.3, 125.9, 119.3, 116.4, 116.2, 111.8, 64.2, 21.7, 20.1, 18.9; FT-IR (KBr) 2919, 2857, 2120, 1639, 1602, 1557, 1506, 1458, 1322,

1251, 1211, 1156, 1084, 829 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{25}\text{H}_{23}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 434.1421, found: 434.1415.



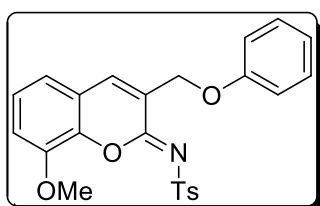
(Z)-N-(3-((3,5-Dimethylphenoxy)methyl)-2H-chromen-

2-ylidene)-4-methylbenzenesulfonamide (7n): mellow solid; yield: 74%; mp 177-178 $^{\circ}\text{C}$. ^1H NMR (400 MHz, CDCl_3) δ 8.02 (d, $J = 8.0$ Hz, 2H), 7.89 (s, 1H), 7.53-7.49 (m, 2H), 7.41 (d, $J = 8.4$ Hz, 1H), 7.34 (t, $J = 8.0$ Hz, 4H), 6.63 (d, $J = 5.2$ Hz, 2H), 4.91 (s, 2H), 2.40 (s, 3H), 2.28 (s, 6H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.8, 156.0, 151.8, 143.5, 139.4, 138.7, 137.4, 131.8, 129.3, 128.3, 127.6, 126.0, 125.8, 123.3, 119.1, 116.1, 112.4, 63.8, 21.5, 21.3; FT-IR (KBr) 2917, 2857, 1643, 1588, 1561, 1456, 1320, 1204, 1151, 1088, 816 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{25}\text{H}_{23}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 434.1421, found: 434.1409.



(Z)-N-(3-(Methoxymethyl)-2H-chromen-2-ylidene)-4-

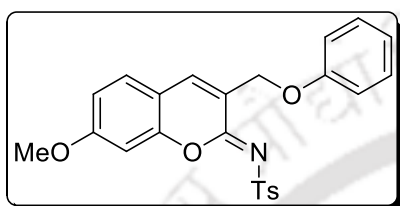
methylbenzenesulfonamide (7o): yellow viscous liquid; yield: 10%. ^1H NMR (400 MHz, CDCl_3) δ 7.84 (d, $J = 8.0$ Hz, 2H), 7.71 (s, 1H), 7.28-7.20 (m, 4H), 6.95 (t, $J = 7.6$ Hz, 1H), 6.85 (d, $J = 8.0$ Hz, 1H), 4.96 (s, 2H), 3.75 (s, 3H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 166.0, 155.4, 143.2, 139.8, 136.8, 132.5, 129.7, 129.5, 126.7, 122.2, 121.9, 121.3, 116.3, 65.1, 55.8, 21.7; FT-IR (KBr) 2945, 2922, 2851, 1692, 1583, 1481, 1456, 1380, 1344, 1292, 1252, 1227, 1153, 1086, 1035, 813 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{18}\text{H}_{17}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 344.0951, found: 344.0957.



(Z)-N-(8-Methoxy-3-(phenoxy)methyl)-2H-chromen-2-

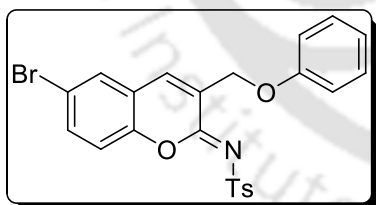
ylidene)-4-methylbenzenesulfonamide (7o): colourless solid; yield 70%; mp 178-179

°C. ^1H NMR (400 MHz, CDCl_3) δ 8.19 (d, $J = 8.0$ Hz, 2H), 7.79 (s, 1H), 7.32-7.21 (m, 5H), 7.21 (t, $J = 7.6$ Hz, 1H), 7.10 (t, $J = 8.0$ Hz, 2H), 7.00 (m, 2H), 6.89 (m, 1H), 4.96 (s, 2H), 4.06 (s, 3H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 155.9, 147.6, 143.6, 142.0, 138.0, 137.6, 129.8, 129.1, 128.6, 126.5, 126.0, 121.8, 120.2, 119.7, 115.4, 114.9, 114.0, 64.2, 56.6, 21.8; FT-IR (KBr) 3038, 2918, 2846, 1633, 1580, 1552, 1480, 1314, 1242, 1208, 1159, 1084, 1045, 833 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 436.1213, found: 436.1216.



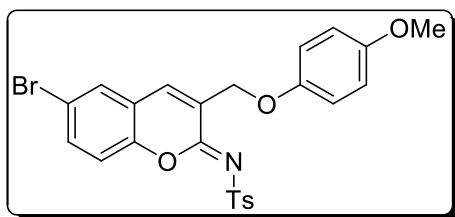
(Z)-N-(7-Methoxy-3-(phenoxyethyl)-2H-chromen-

2-ylidene)-4-methylbenzenesulfonamide (7q): colourless solid; yield 62%; mp 193-194 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.99 (d, $J=8.0$ Hz, 2H), 7.83 (s, 1H), 7.40 (d, $J=8.8$ Hz, 1H), 7.31-7.26 (m, 4H), 6.98-6.89 (m, 5H), 4.90 (s, 2H), 3.88 (s, 3H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 163.0, 157.9, 156.7, 153.8, 143.4, 139.1, 137.9, 129.8, 129.4, 129.3, 127.5, 122.3, 121.6, 114.9, 114.6, 112.9, 100.4, 64.1, 56.2, 21.7; FT-IR (KBr) 3054, 2923, 2846, 1635, 1618, 1599, 1538, 1500, 1412, 1314, 1240, 1158, 1085, 864 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 436.1213, found: 436.1221.

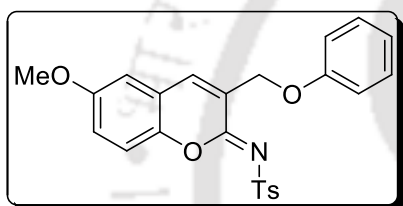


(Z)-N-(6-Bromo-3-(phenoxyethyl)-2H-chromen-2-

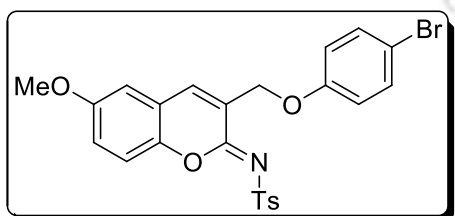
ylidene)-4-methylbenzenesulfonamide (7r): yellow solid; yield 57%; mp 208-209 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.98 (d, $J=8.0$ Hz, 2H), 7.80 (s, 1H), 7.65-7.60(m, 2H), 7.32-7.27 (m, 5H), 7.00-6.94 (m, 3H), 4.91 (s, 2H), 2.41 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 155.6, 151.0, 143.9, 138.7, 135.9, 134.7, 130.7, 129.9, 129.6, 127.7, 122.0, 120.8, 118.3, 114.9, 114.8, 64.2, 21.8; FT-IR (KBr) 3098, 3065, 2922, 2857, 1629, 1585, 1557, 1498, 1412, 1326, 1240, 1163, 1085, 823 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{18}\text{BrNO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 484.0213, found: 484.0217.



(Z)-N-(6-Bromo-3-((4-methoxyphenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7s): pale yellow solid; yield 65%; mp 207-208 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.98 (d, $J=8.0$ Hz, 2H), 7.80 (s, 1H), 7.64-7.59 (m, 2H), 7.32-7.27 (m, 4H), 7.00-6.93 (m, 3H), 5.27 (s, 2H), 4.91 (s, 3H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) 157.7, 155.5, 150.9, 143.9, 138.6, 136.0, 134.8, 130.6, 129.9, 129.6, 127.7, 121.9, 120.8, 118.7, 115.5, 114.9, 64.1, 53.6, 21.8; FT-IR (KBr) 3098, 3067, 2956, 2923, 2120, 1629, 1586, 1556, 1498, 1326, 1240, 1163, 1084, 823 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{20}\text{BrNO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 514.0319, found: 514.0325.

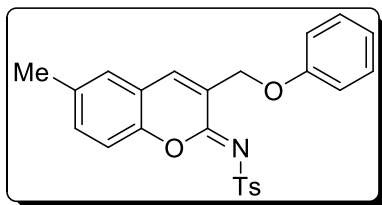


(Z)-N-(6-Methoxy-3-(phenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7t): yellow solid; yield 65%; mp 176-177 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.00 (d, $J=7.6$ Hz, 2H), 7.85 (s, 1H), 7.35-7.27 (m, 5H), 7.10 (d, $J=9.2$ Hz, 1H), 6.99-6.91 (m, 4H), 4.93 (s, 2H), 3.81 (s, 3H), 2.39 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 157.1, 156.3, 146.7, 143.5, 138.9, 137.3, 129.8, 129.4, 127.7, 126.4, 121.8, 120.0, 119.8, 117.7, 114.9, 110.0, 64.2, 56.0, 21.7; FT-IR (KBr) 2961, 2917, 1630, 1585, 1548, 1496, 1315, 1281, 1230, 1153, 1086, 1051, 823 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 436.1213, found: 436.1223.

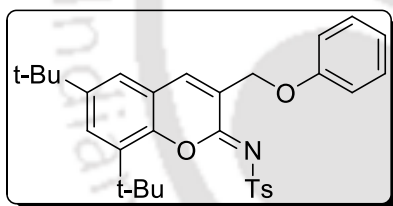


(Z)-N-(3-((4-Bromophenoxy)methyl)-6-methoxy-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7u): yellow solid; yield: 71%; mp 197-198 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.99 (d, $J=8.0$ Hz, 2H), 7.81 (s, 1H), 7.39-7.27 (m, 5H), 7.11-7.09 (m, 1H), 6.91-6.84 (m, 3H), 4.90 (s, 2H), 3.82 (s, 3H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.2, 156.2, 146.8, 143.8, 138.9, 137.5,

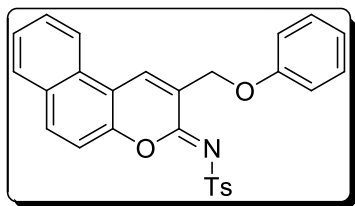
132.7, 129.5, 127.8, 125.9, 120.2, 119.7, 117.7, 116.7, 114.0, 110.0, 64.5, 56.0, 21.8; FT-IR (KBr) 2967, 2923, 1631, 1577, 1532, 1488, 1285, 1248, 1158, 1085, 1034, 832 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{20}\text{BrNO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 514.0319, found: 514.0314.



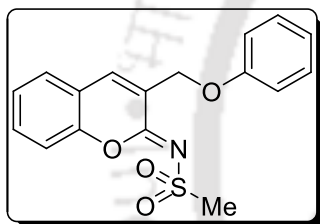
(Z)-4-Methyl-N-(6-methyl-3-(phenoxyethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7v): colourless solid; yield: 77%; mp 234-235 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.01 (d, $J=8.4$ Hz, 2H), 7.84 (s, 1H), 7.35-7.27 (m, 7H), 6.99-6.95 (m, 3H), 4.93 (s, 2H), 2.39 (s, 3H), 2.38 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 156.4, 150.4, 143.5, 138.9, 137.6, 136.0, 133.2, 129.9, 129.5, 128.2, 127.8, 125.9, 121.8, 119.1, 116.3, 115.0, 64.3, 21.7, 21.0; FT-IR (KBr) 3038, 2921, 2851, 1630, 1597, 1545, 1495, 1419, 1312, 1301, 1286, 1247, 1218, 1157, 1087, 1048, 833 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{24}\text{H}_{21}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 420.1264, found: 420.1259.



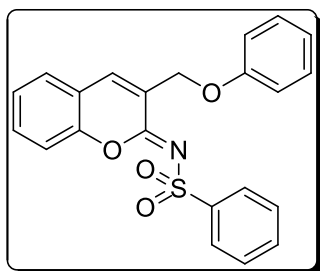
(Z)-N-(6,8-Di-tert-butyl-3-(phenoxyethyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7w): yellow solid; yield 63%; mp 160-161 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.65 (s, 1H), 7.62 (d, $J = 7.6$ Hz, 2H), 7.31-7.27 (m, 3H), 7.19-7.14 (m, 3H), 6.99 (d, $J = 8.4$ Hz, 3H), 4.96 (s, 2H), 2.35 (s, 3H), 1.33 (s, 9H), 1.25 (s, 9H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 163.0, 153.4, 152.1, 144.2, 143.2, 140.8, 138.9, 137.7, 129.8, 129.3, 128.2, 126.9, 125.7, 124.6, 121.5, 120.8, 120.1, 64.2, 34.8, 34.5, 31.5, 29.7, 21.7; FT-IR (KBr) 2959, 1629, 1601, 1573, 1488, 1277, 1233, 1211, 1151, 1083, 1024, 766, 690 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{31}\text{H}_{35}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 518.2360, found: 518.2372.



(Z)-4-Methyl-N-(2-(phenoxyethyl)-3H-benzo[f]chromen-3-ylidene)benzenesulfonamide (7x); yellow solid; yield 67%; mp 216-217 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.62 (s, 1H), 8.23 (d, $J = 8.4$ Hz, 1H), 8.05 (d, $J = 8.0$ Hz, 2H), 7.98 (d, $J = 8.8$ Hz, 1H), 7.89 (d, $J = 7.6$ Hz, 1H), 7.69-7.65 (m, 1H), 7.58-7.50 (m, 2H), 7.33 (t, $J = 7.2$ Hz, 3H), 7.06-6.98 (m, 3H), 5.00 (s, 2H), 2.40 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 156.1, 151.7, 143.6, 139.0, 133.4, 133.3, 130.8, 129.9, 129.5, 129.2, 128.7, 128.6, 127.7, 126.9, 125.3, 121.9, 121.8, 116.0, 115.0, 114.0, 64.4, 21.7; FT-IR (KBr) 3048, 2922, 2851, 1634, 1613, 1597, 1548, 1513, 1315, 1248, 1215, 1156, 1085, 807 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{27}\text{H}_{21}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 456.1264, found: 456.1272.

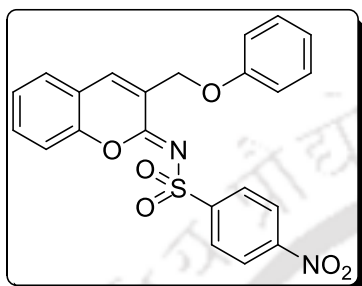


(Z)-N-(3-(Phenoxyethyl)-2H-chromen-2-ylidene)methanesulfonamide (7y); pale yellow solid; yield 70%; mp 166-167 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.19 (s, 1H), 7.53-7.51 (m, 3H), 7.35-7.26 (m, 3H), 7.02-6.95 (m, 3H), 7.34-7.30 (m, 4H), 4.91 (s, 2H), 3.18 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 156.5, 152.0, 137.6, 132.1, 129.8, 128.3, 126.0, 125.6, 121.8, 119.1, 117.0, 114.8, 64.2, 43.0; FT-IR (KBr) 3060, 2932, 2851, 1633, 1562, 1498, 1456, 1406, 1309, 1249, 1215, 1145, 1045, 974, 830 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{17}\text{H}_{15}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 330.0795, found: 330.0803.



(Z)-N-(3-(Phenoxyethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7z); colourless solid; yield 67%; mp 188-189 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.13 (d, $J = 7.2$ Hz, 2H), 7.91 (s, 1H), 7.56-7.50 (m, 5H),

7.40 (d, $J = 8.4$ Hz, 1H), 7.34-7.27 (m, 4H), 6.98 (d, $J = 8.4$ Hz, 2H), 4.93 (s, 2H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 156.3, 152.1, 141.8, 137.7, 132.8, 132.1, 129.8, 129.6, 128.9, 128.4, 127.6, 126.1, 126.0, 121.8, 120.4, 119.2, 116.5, 115.5, 114.9, 64.2; FT-IR (KBr) 3053, 2923, 2857, 2126, 1632, 1588, 1551, 1498, 1459, 1320, 1305, 1252, 1208, 1158, 1084, 1048, 827 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{22}\text{H}_{17}\text{NO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 392.0951, found: 392.0957.



(Z)-4-Chloro-N-(3-(phenoxymethyl)-2H-chromen-2-ylidene)benzenesulfonamide (7aa): yellow solid; yield 39%; mp 212-213 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.36 (d, $J = 7.2$ Hz, 2H), 8.27 (d, $J = 7.6$ Hz, 2H), 8.01 (s, 1H), 7.62-7.56 (m, 2H), 7.58 (d, $J = 8.0$ Hz, 1H), 7.41-7.37 (m, 1H), 7.31-7.28 (m, 2H), 7.01-6.95 (m, 3H), 4.92 (s, 2H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.8, 156.0, 151.8, 143.5, 139.5, 137.4, 131.8, 129.3, 128.3, 127.6, 126.0, 125.8, 123.3, 119.3, 116.1, 112.4, 63.8, 21.5, 21.3; FT-IR (KBr) 3100, 3064, 2919, 2851, 1630, 1600, 1589, 1532, 1497, 1487, 1452, 1350, 1303, 1249, 1215, 1249, 1215, 1160, 1086, 828 cm^{-1} . HRMS (ESI): Calcd for $\text{C}_{22}\text{H}_{16}\text{N}_2\text{O}_6\text{S}$ $[\text{M}+\text{H}]^+$: 437.0802, found: 437.0808.

Crystal Data and Structure Refinement for 7j at 296(2) K.

Identification code	GM-131
Empirical formula	$\text{C}_{24}\text{H}_{21}\text{N}\text{O}_5\text{S}$
Formula weight	435.49
Temperature	296(2) K
Wavelength	0.71073 Å
Crystal system	Monoclinic
Space group	$\text{P}2(1)/c$ loop_ _symmetry_equiv_pos_as_xyz

	x, y, z	
	-x, y+1/2, -z+1/2	
	-x, -y, -z	
	'x, -y-1/2, z-1/2	
Unit cell dimensions	a = 9.2913(18) Å	α(°) = 90.00
	b = 11.1324(19) Å	β(°) = 91.227(11)
	c = 20.607(4) Å	γ(°) = 90.00
Volume	2130.9(7) Å ³	
Z	4	
Density	1.357 Mg/m ³	
Absorption coefficient	0.188 mm ⁻¹	
F(000)	912.0	
Crystal size	0.36 x 0.24 x 0.20 mm	
Theta range for data collection	1.98 to 20.59°	
Index ranges	-8 ≤ h ≤ 9, -11 ≤ k ≤ 11, -20 ≤ l ≤ 17	
Reflections collected	2143	
Independent reflections	1721 [R (int) = 0.0474]	
Completeness to theta = 20.59°	99.1%	
Absorption correction	Multi-scan	
Refinement method	Full-matrix least-squares on F ²	
Data / restraints / parameters	2143/ 0 /283	
Goodness-of-fit on F ²	1.019	
Final R indices [I > 2σ(I)]	R1 = 0.0324, wR2 = 0.0780	
R indices (all data)	R1 = 0.0468, wR2 = 0.0857	

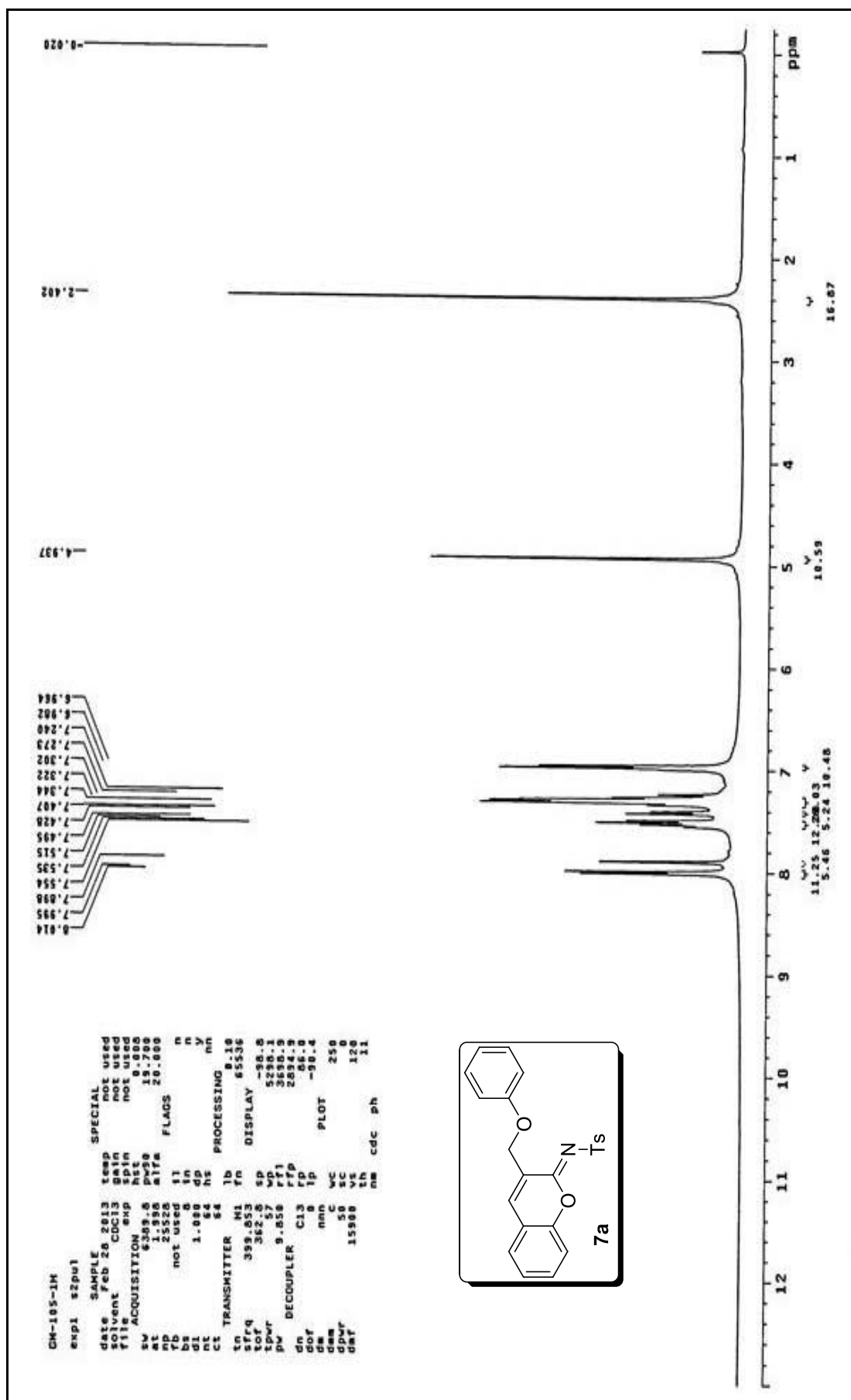
2.4 References

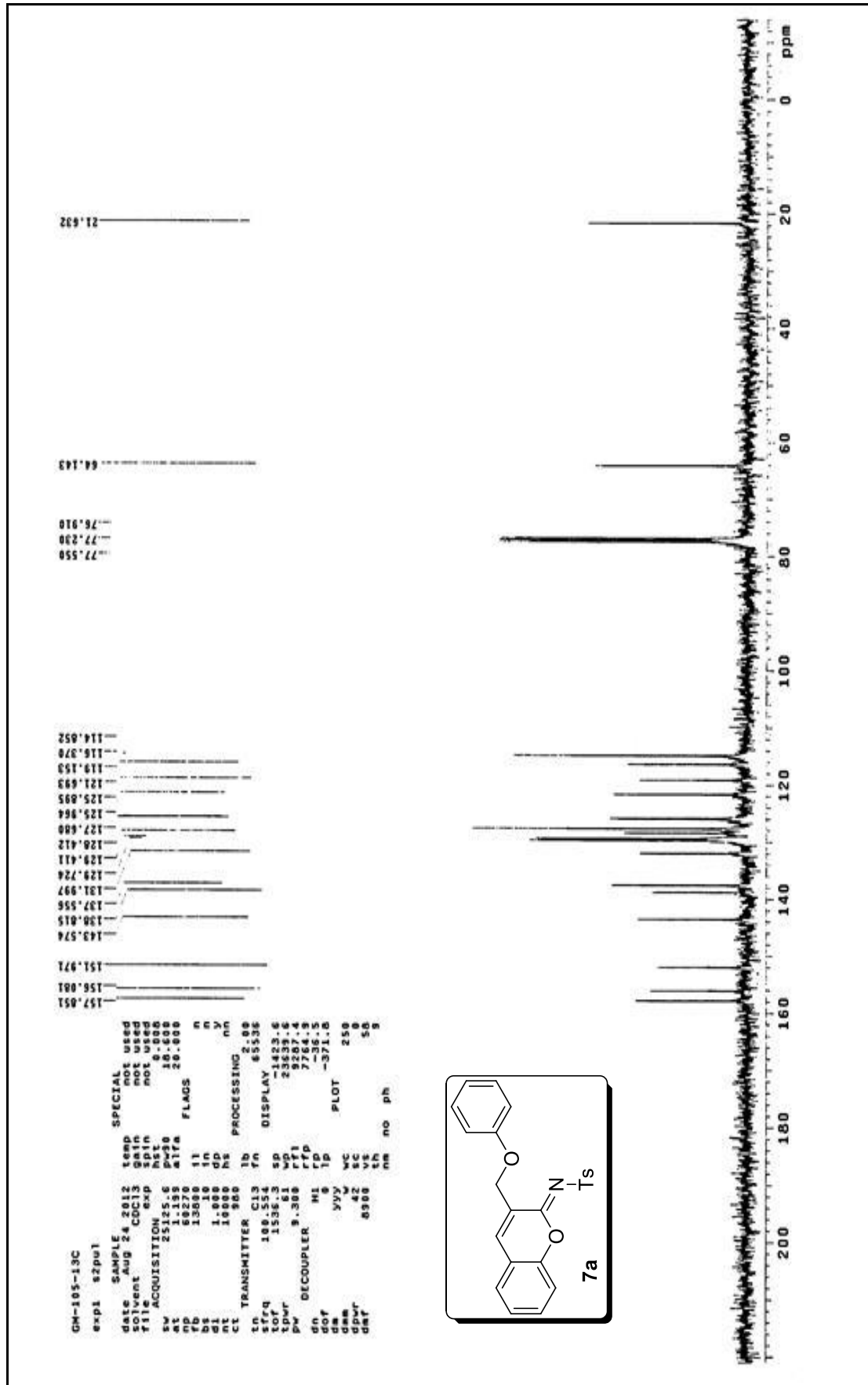
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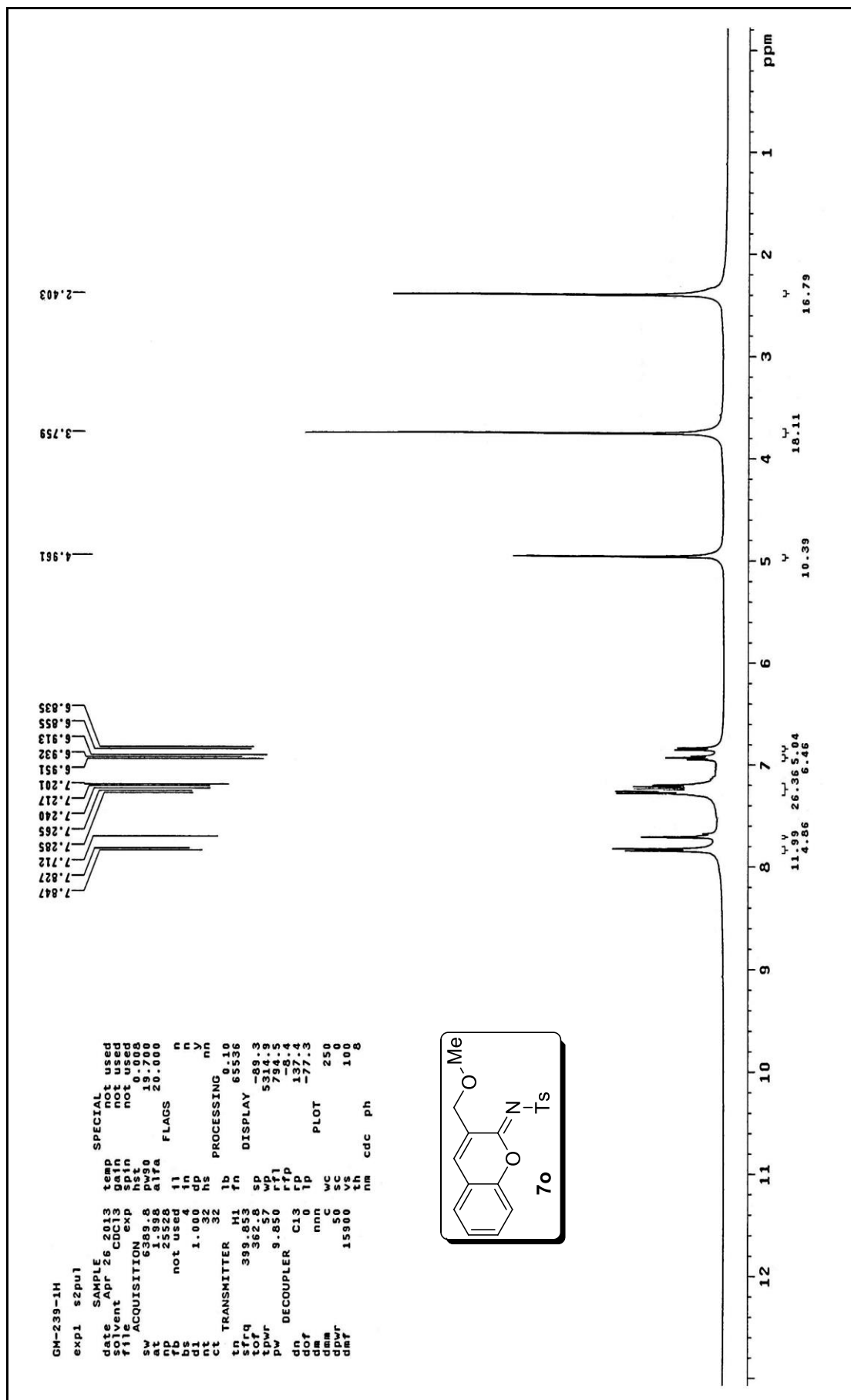
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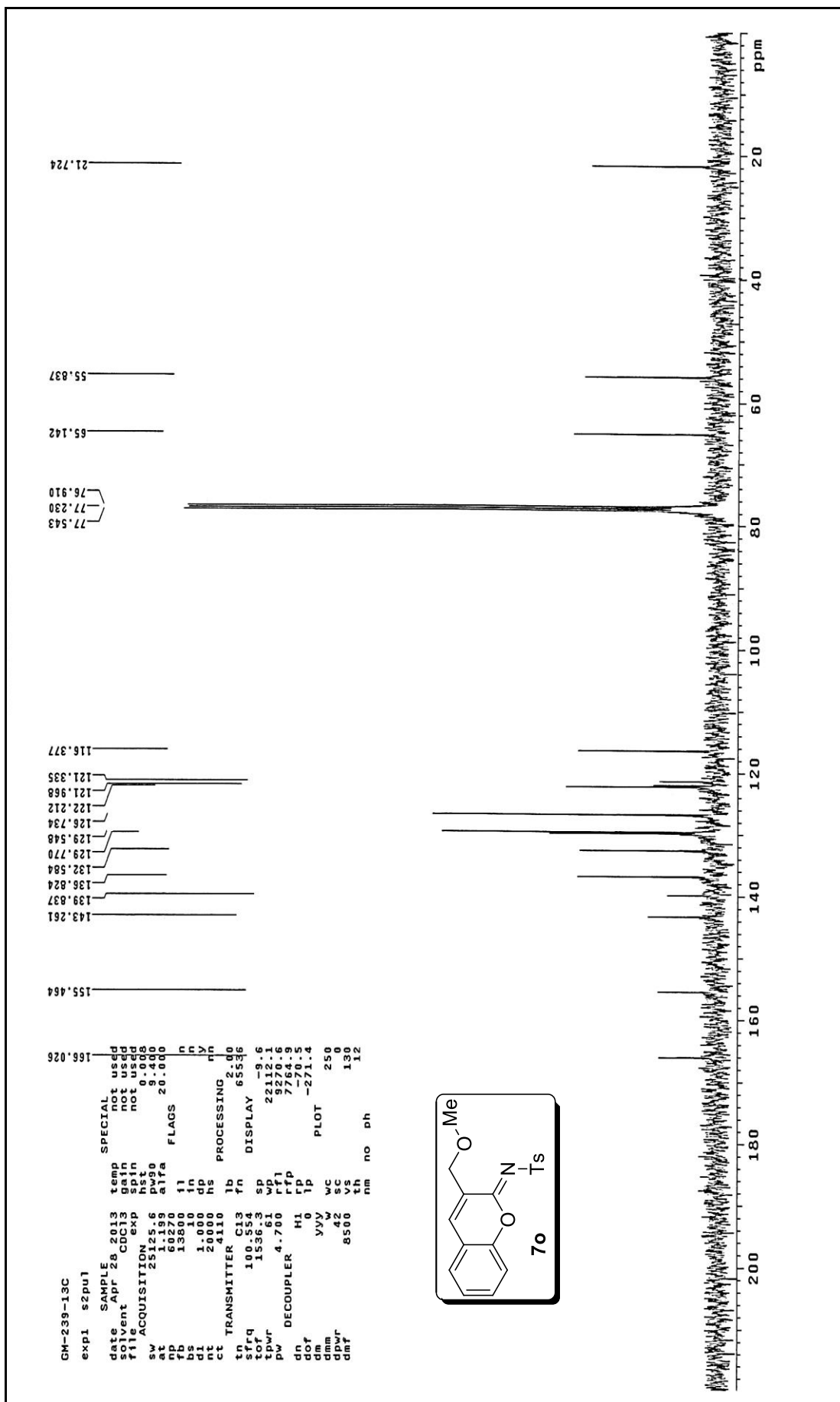
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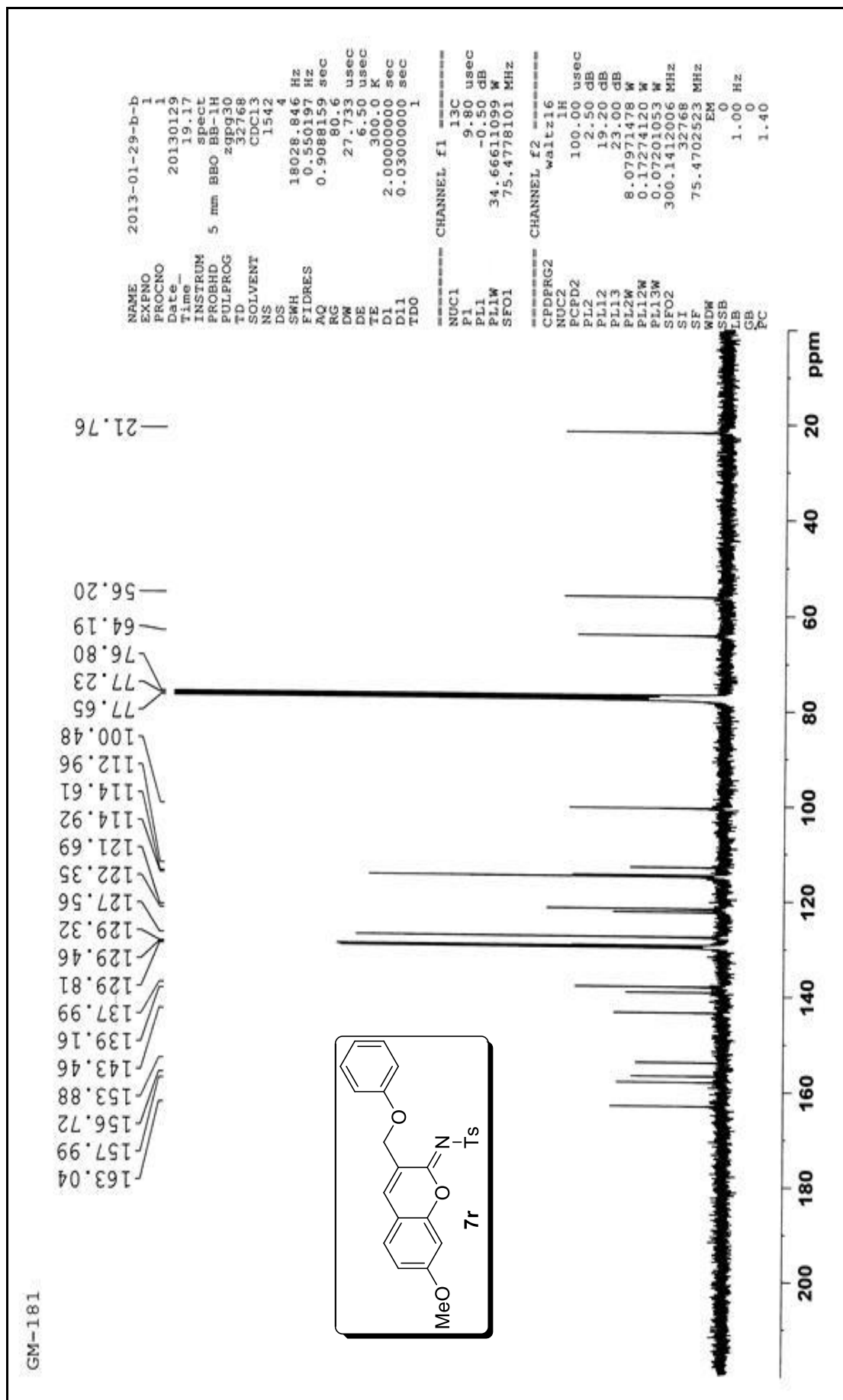
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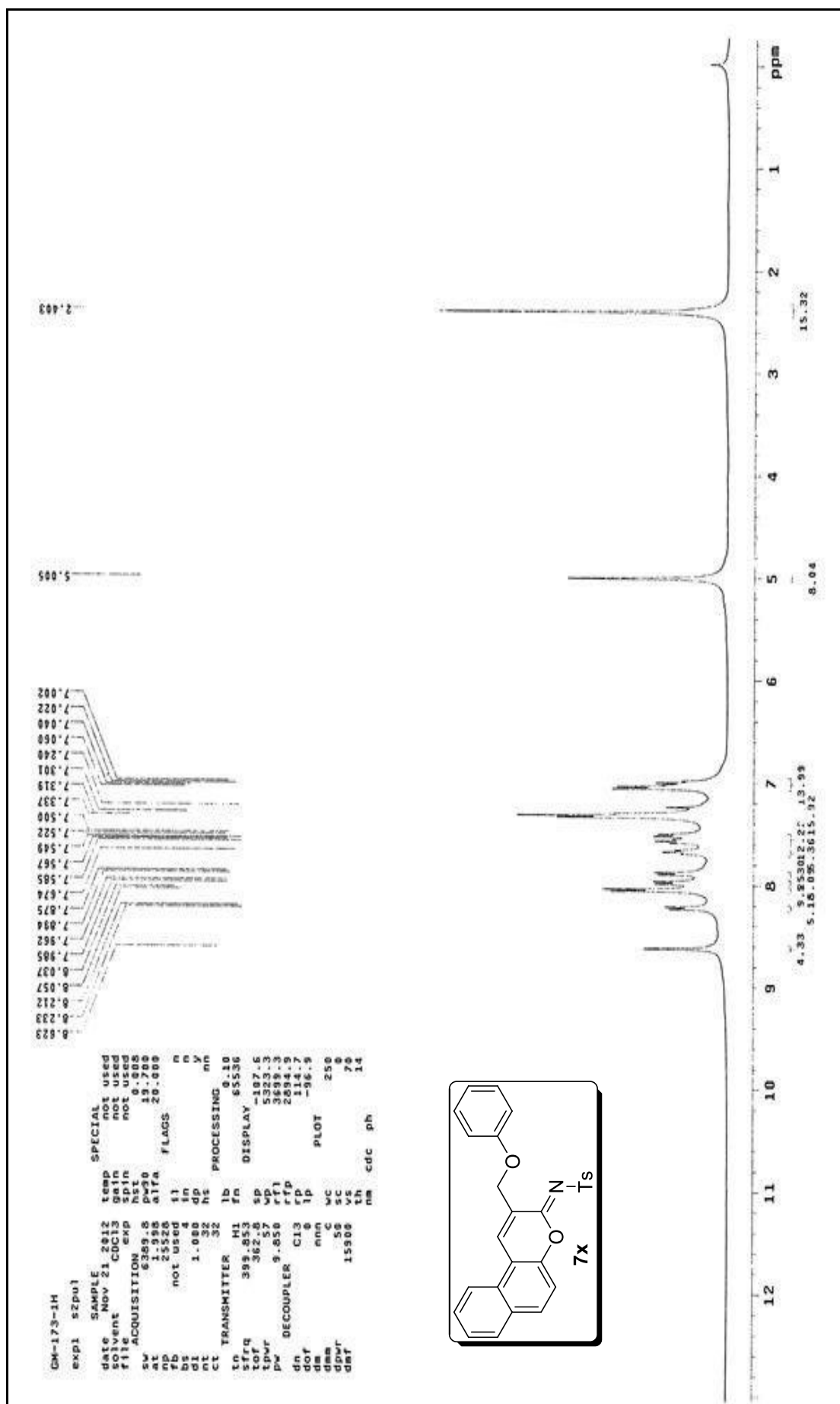


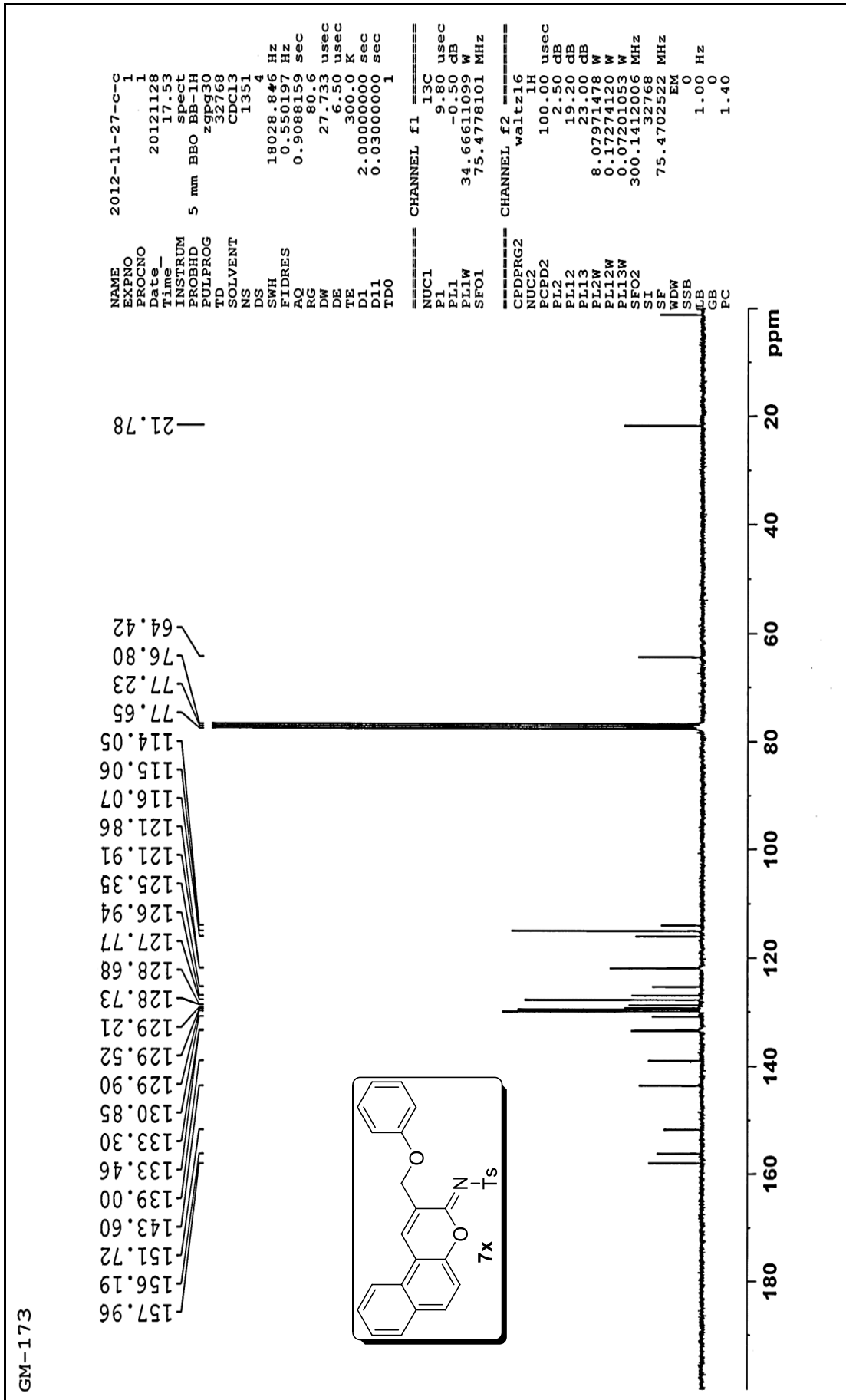


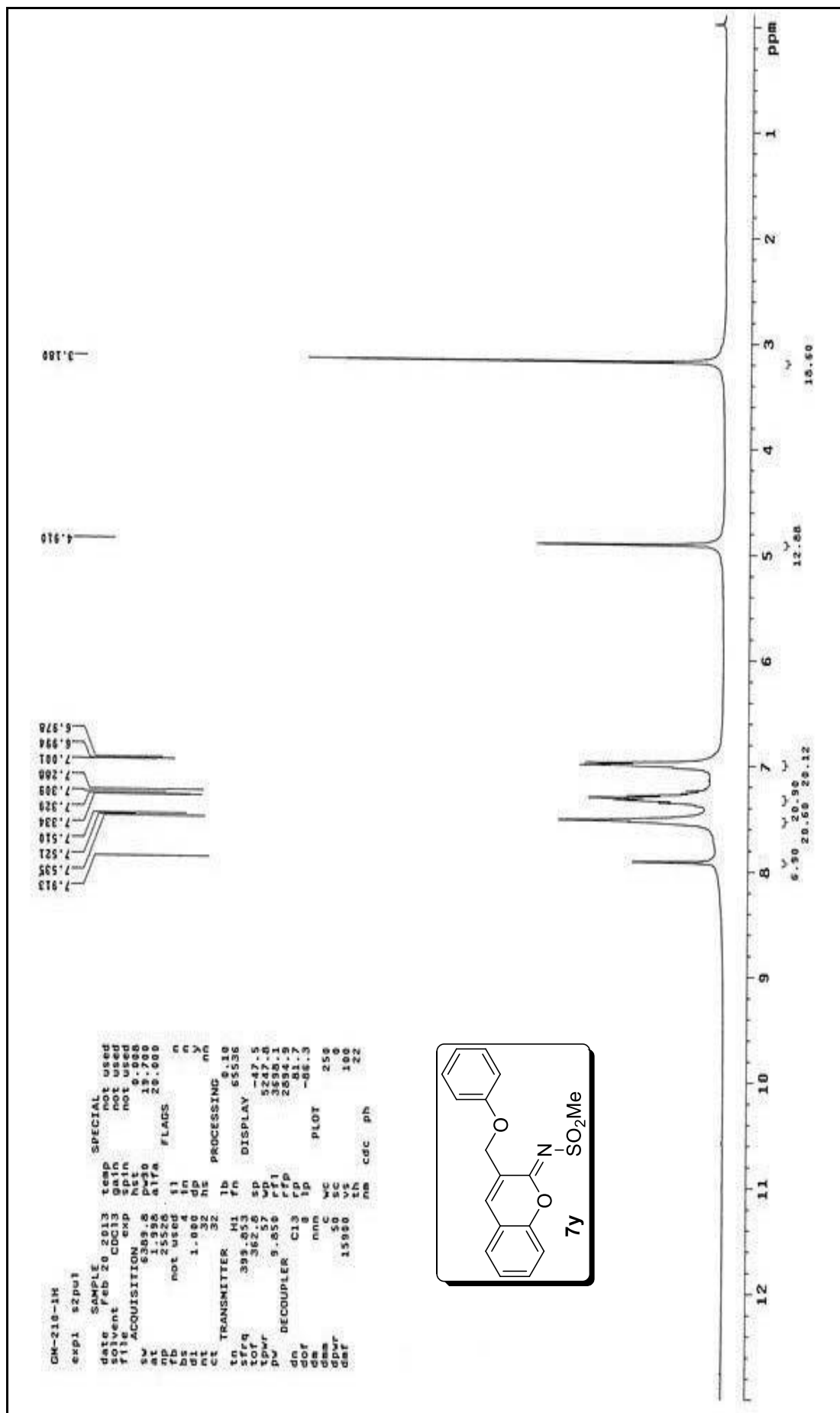


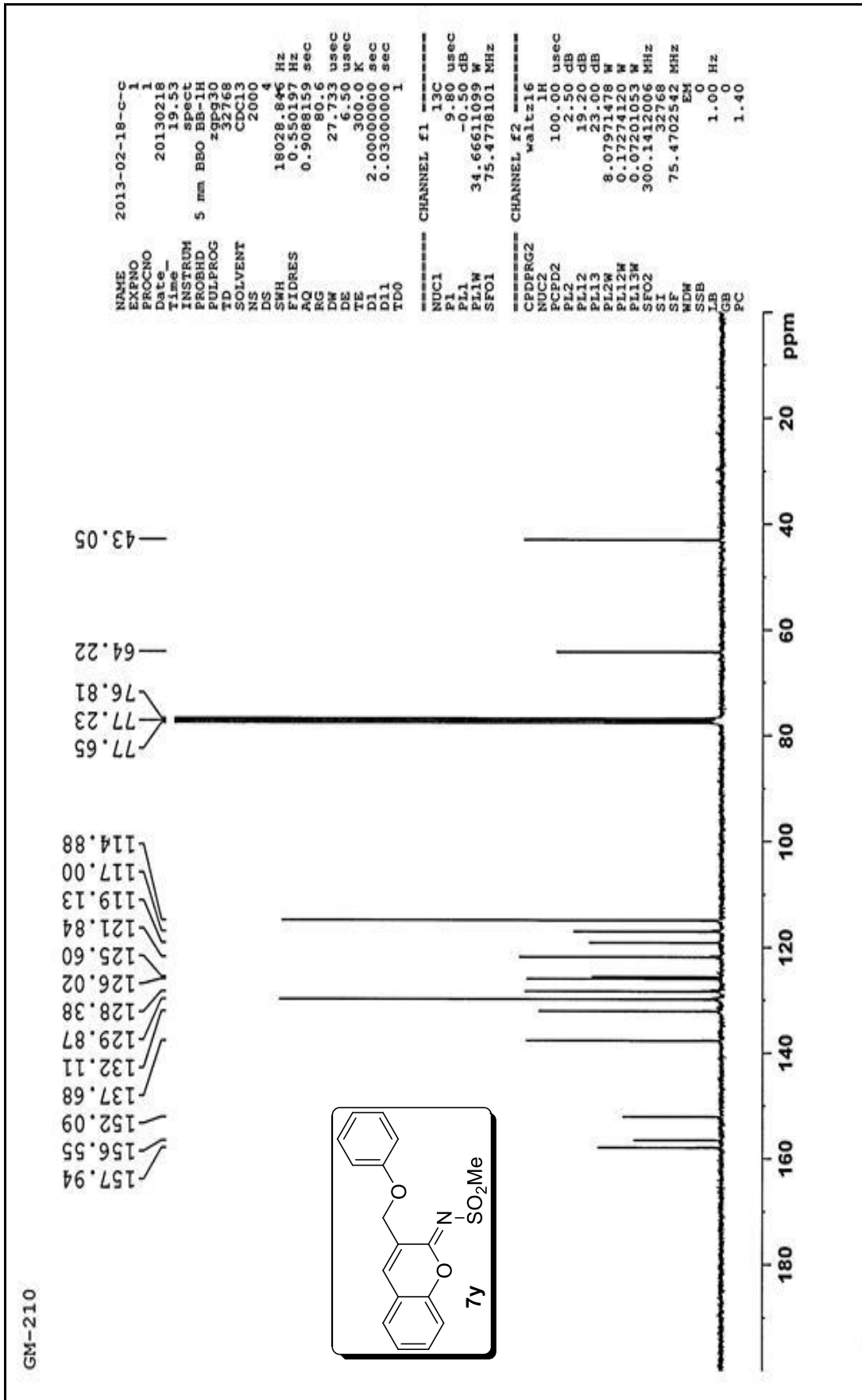












Copper-Catalyzed Four-Component Cascade Synthesis of 3*N*-Sulfonylamidine Coumarins

Coumarins are an important organoleptic heterocyclic scaffold and widely exist in nature¹ with numerous applications in the field of material and medicinal sciences (Figure 1).² Coumarin derivatives exhibit various medicinal properties, such as anticoagulant,³ anti-cancer,⁴ anti-microbial,⁵ anti-HIV,⁶ anti-inflammatory,⁷ anti-oxidant,⁸ antibiotic,⁹ anti-diabetic¹⁰ and anti-Alzheimer properties.¹¹ In addition, the compounds containing coumarin motifs showed potent inhibitors of HIV integrase,¹² Heat Shock Protein 90,¹³ Casein Kinase 2 (CK2),¹⁴ Human Monoamine Oxidases¹⁵ and Cholinesterase.¹⁶ Further, coumarin as plant sources are utilized in perfumes.¹⁷ They also find applications as dyes in food flavoring stuffs.¹⁸ In addition, coumarins serve as an excellent fluorescent probes in biology and medicine¹⁹ as well as dyes in laser technology.²⁰

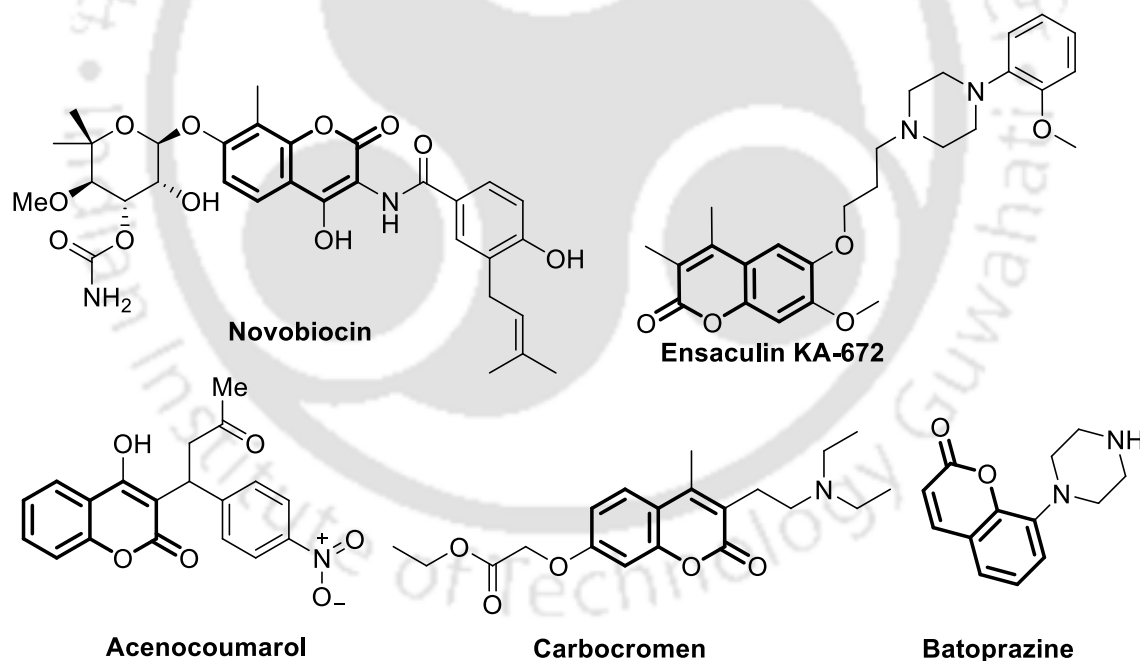
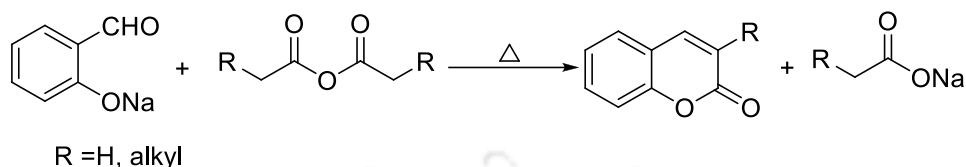


Figure 1. Some examples of biologically important coumarin derivatives

3.1 Strategies for Coumarin Synthesis

3.1.1 Perkin Reaction

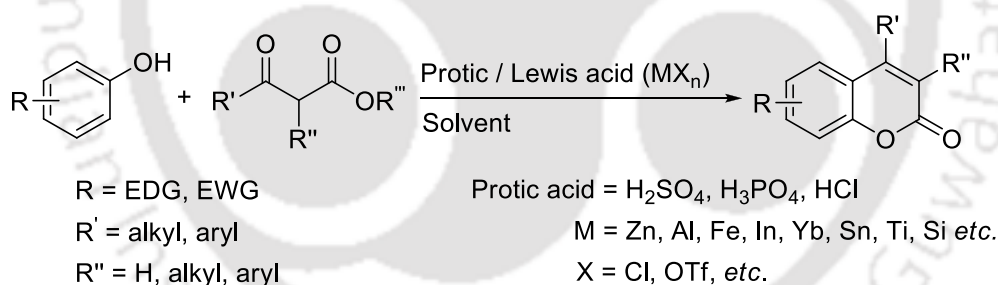
In 1868, Perkin described the formation of coumarins from the reaction of sodium salt of salicylaldehyde with acetic anhydride under heating (Scheme 1).²¹



Scheme 1

3.1.2 Von Pechmann Reaction

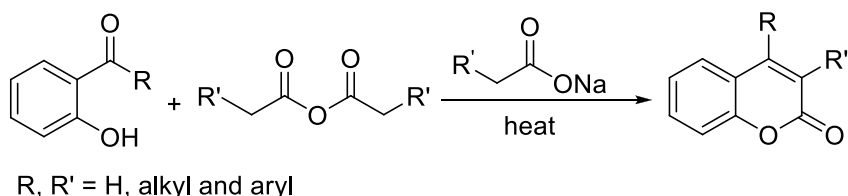
Von Pechmann and co-workers reported the acid catalyzed condensation of nucleophilic phenols with ethyl acetoacetate to produce coumarins (Scheme 2).^{22a} Similar results were also obtained while using maleic acid instead of β -ketoesters.^{22b} The reaction work well with phenols having electron rich groups, whereas phenols bearing electron withdrawing substituents failed to produce the target heterocycles. In addition, the requirement of elevated temperature limit their applications.^{22c}



Scheme 2

3.1.3 Kostanecki Acylation

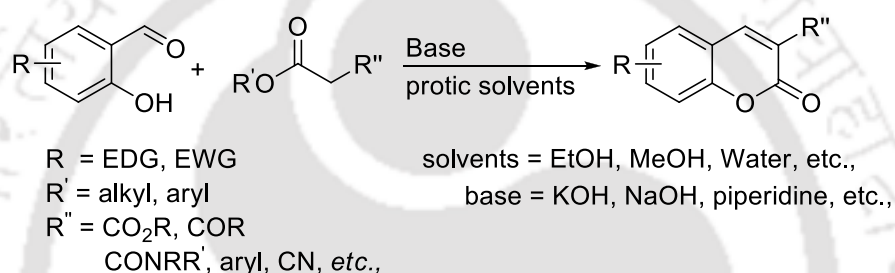
In 1901, Kostanecki synthesized coumarin from 2-hydroxyarylketone and anhydride in the presence of sodium carboxylate *via* *O*-acylation followed by aldol condensation (Scheme 3).²³ In this reaction, R' is aryl, coumarins are formed exclusively. In contrast, R' is alkyl, chromone is formed significantly as byproduct.



Scheme 3

3.1.4 Knoevenagel Condensation

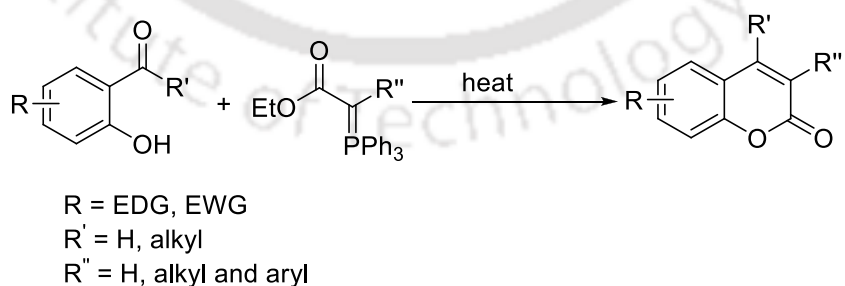
Knoevenagel condensation of *o*-hydroxybenzaldehydes with active methylene compounds such as diethyl malonates, malonic acid, malononitrile, ethyl trifluoroacetate, *N*-acetylglycine, etc., afford 3-substituted coumarin derivatives (Scheme 4).²⁴ The reaction is commonly performed using strong bases in protic solvents at high temperatures.



Scheme 4

3.1.5 Wittig Reaction

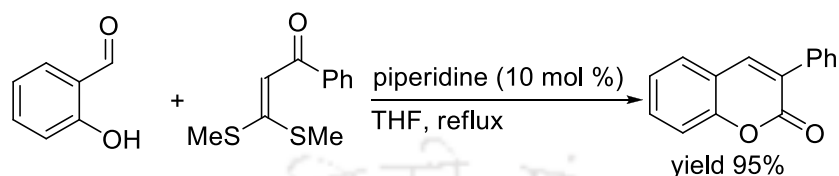
The condensation of *o*-hydroxybenzaldehyde with Wittig ylide gives coumarin analogue (Scheme 5).²⁵ This reaction is moisture sensitive and effective at high temperature with limited substrate scope.



Scheme 5

3.1.6 Synthesis of Coumarin from α -Aroylketene Dithioacetals

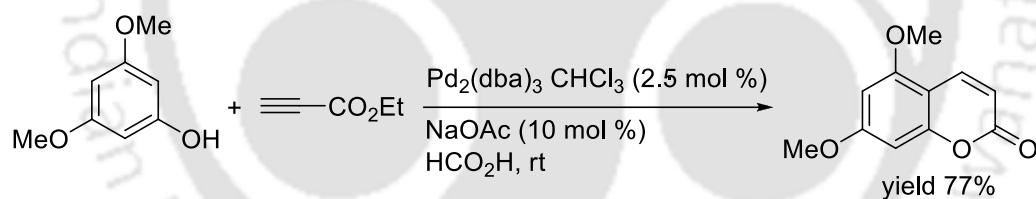
Rao and co-workers reported the synthesis of coumarins from *o*-hydroxybenzaldehydes and α -aroylketene dithioacetals in the presence of catalytic amount of piperidine in THF at reflux temperature (Scheme 6).²⁶



Scheme 6

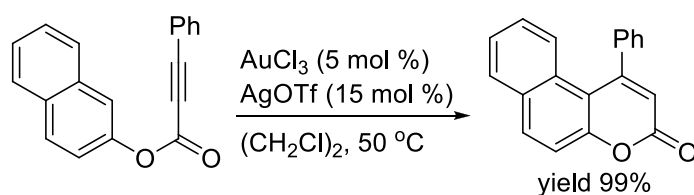
3.1.7 Carbometalation Reaction

Trost and co-workers accomplished the synthesis of coumarins from phenols and alkyl propiolates using 2.5 mol % $\text{Pd}_2(\text{dba})_3 \cdot \text{CHCl}_3$ in the presence of NaOAc in HCOOH at ambient temperature (Scheme 7).^{27a} This reaction proceeds *via* electrophilic palladation of phenols and carbometalation with alkynoates to give cinnamic esters, which further undergoes cycloisomerization to furnish the corresponding coumarin derivatives.^{27b} Later, Kitamura and co-workers demonstrated this strategy using Pt based catalytic system.^{27c}



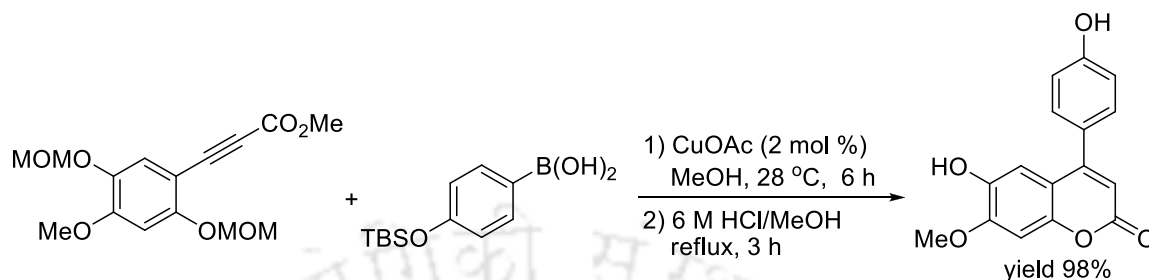
Scheme 7

Shi and He described the synthesis of coumarins using AuCl_3 with AgOTf in CH_2Cl_2 *via* intramolecular C-H functionalization of aryl propiolates (Scheme 8).^{27d} This transformation has been subsequently demonstrated using Hf^{27e} and Pd^{27f} based catalytic systems. In addition, several cascade approaches have been developed for the synthesis of coumarins having 3-iodo,^{27g} 3-organoselenenyl,^{27h} 3-carbonyl²⁷ⁱ and 3-difluoroacetyl^{27j} derivatives.



Scheme 8

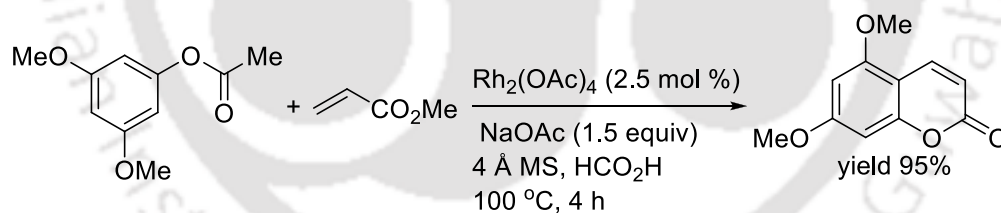
In 2008, Yamamoto and Kirai reported Cu-catalyzed hydroarylation of aryl boronic acids with MOM-protected methyl 3-(2-methoxymethoxy)aryl)propiolates using 2-10 mol % CuOAc in MeOH, followed by refluxing with 6 M HCl to afford coumarins in 75-98% yields (Scheme 9).²⁸



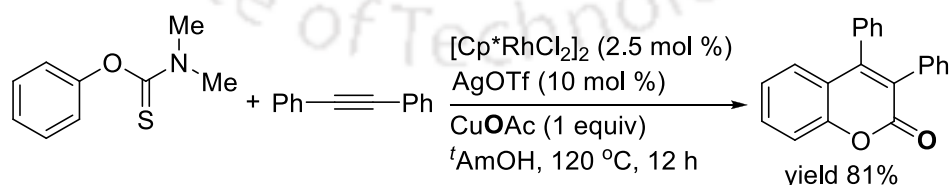
Scheme 9

3.1.8 C-H Activation

In 2013, Maity and co-workers reported Pd-catalyzed synthesis of coumarins from phenols and methyl acrylates using Pd(OAc)₂ with 1,10-phenanthroline in the presence of Cu(OAc)₂ as co-oxidant in 1,2-dichloroethane.^{29a} Later, Sudalai and co-workers reported the synthesis of coumarins from acetyl phenolates and alkyl acrylates using Rh₂(OAc)₄ in the presence of NaOAc in HCOOH *via* C-H bond activation (Scheme 10).^{29b} The reaction tolerates both electron donating and electron withdrawing substitutes with moderate to high yields.



Scheme 10

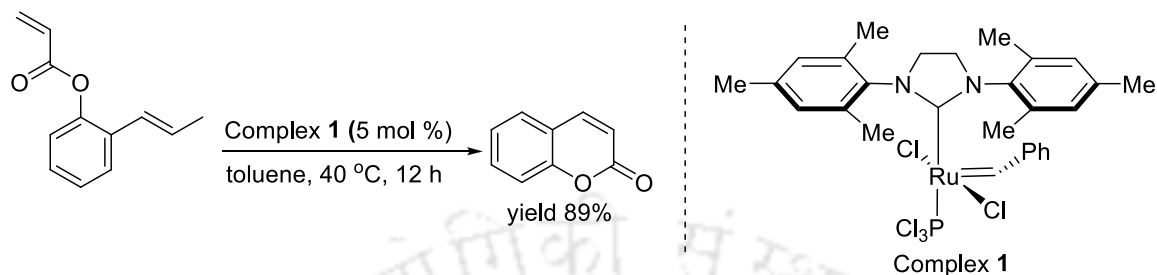


Scheme 11

Xia and co-workers reported the annulation of aryl thiocarbomates with 1,2-disubstituted symmetrical alkynes using [Cp*RhCl₂]/AgOTf in the presence of Cu(OAc)₂ in tert-amyl alcohol to afford functionalized coumarin analogues in moderate to high yields (Scheme 11).^{29c}

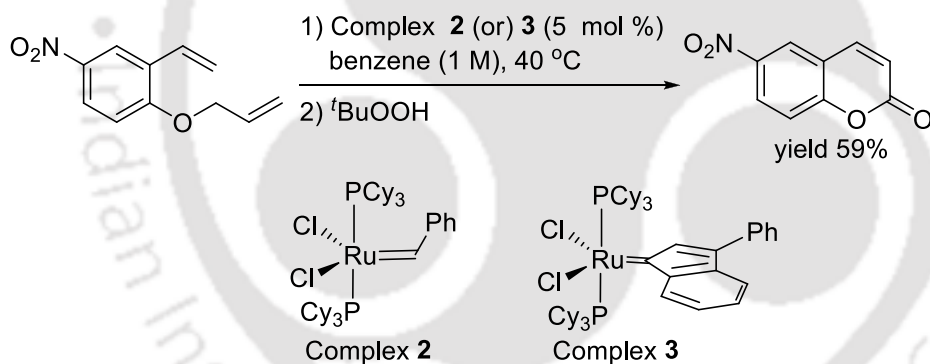
3.1.9 Ring Closing Metathesis

In 2003, Grubbs and co-workers reported the synthesis of coumarins *via* ring closing metathesis using ruthenium carbene complex **1** in toluene at 40 °C with moderate to good yields (Scheme 12).^{30a}



Scheme 12

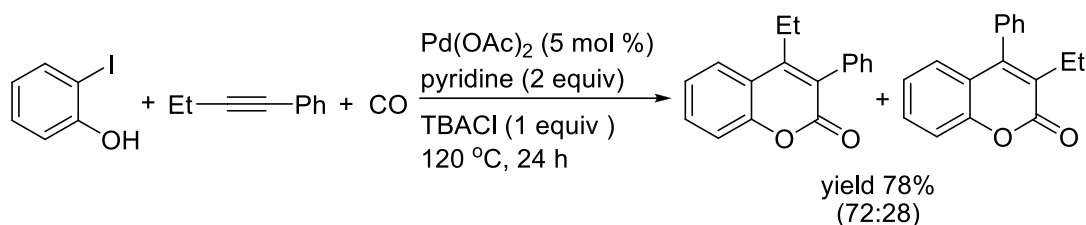
Schmidt and Krehl reported the synthesis of coumarins from 2-allyloxy styrene using ruthenium alkene complexes **2** and **3** *via* ring closing metathesis and subsequent allylic oxidation (Scheme 13).^{30b} The reactions tolerate electron donating and -withdrawing aryl substituents to afford corresponding cyclic products with moderate yields.



Scheme 13

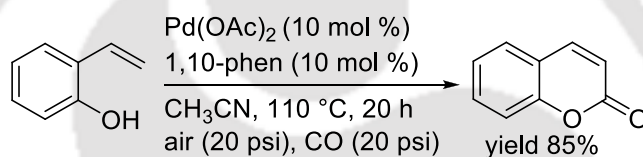
3.1.10 Metal-Catalyzed Carbonylation Reactions

Kadnikov and Larock reported the synthesis of coumarins from alkynes and *o*-iodophenols using Pd(OAc)₂ with pyridine in the presence of TBACl in DMF under CO atmosphere (Scheme 14).^{31a} The reaction involves carbonylative annulation of alkynes with *ortho*-iodophenols. In case of unsymmetrical alkynes, a mixture of regioisomers is formed.



Scheme 14

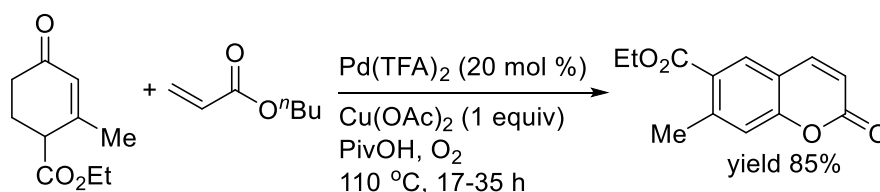
In 2012, Alper and co-workers reported the synthesis of coumarins from 2-hydroxy styrene and carbon monoxide using Pd(OAc)₂ with dppb in the presence of benzoquinone as oxidant in CH₃CN at 110 °C in good to high yields *via* oxidative carbonylation reaction (Scheme 15).^{31b} The authors achieved the same transformation of aryl substituent at alkene using Pd(OAc)₂/1,10-phenanthroline under oxygen atmosphere in CH₃CN at 110 °C. This transformation has also been subsequently demonstrated employing Pd(OAc)₂,^{31c} [Cp*Rh(III)]^{31d} and Cp*Co(CO)I₂^{31e} based catalytic systems.



Scheme 15

3.1.11 Cross-coupling Reactions

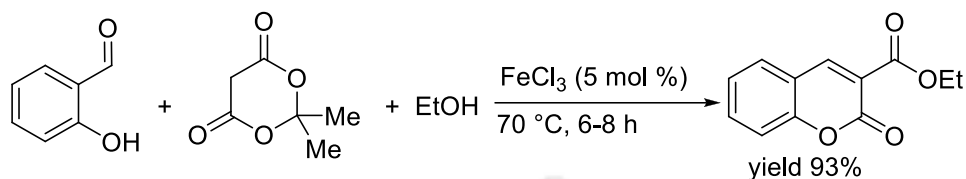
In 2009, Roy and co-workers showed the synthesis of 3-C-linked mannopyranosyl coumarins from 2-iodophenols and alkene functionalized mannopyranose esters using Pd(OAc)₂ in the presence of TBAB and NaHCO₃ in DMF *via* domino Heck coupling and lactonization sequence.^{32a} Costa and co-workers studied the mechanism using electron spray mass spectroscopy.^{32b} In 2013, Hong and co-workers reported the synthesis of coumarin from the substituted cyclohexanone and α,β -unsaturated esters using Pd(TFA)₂ and Cu(OAc)₂ in pivalic acid *via* dehydrogenation of cyclohexanones, oxidative Heck coupling and cyclization reaction sequence (Scheme 16).^{32c}



Scheme 16

3.1.12 Multicomponent Reactions

Shang and co-workers studied MCR of salicylaldehyde, Meldrum's acid and alcohol using FeCl₃ at 70 °C to provide functionalized coumarin-3-carboxylic ester derivatives in high yields (Scheme 17).³³



Scheme 17

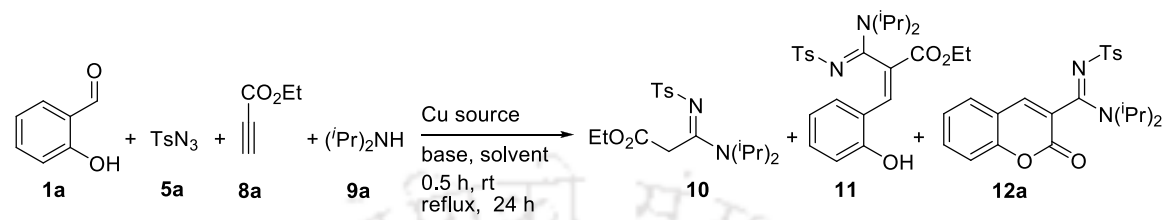
3.2 Present Study

MCR affords a powerful tool for the conversion of simple substrates into complex molecules with structural diversity.³⁴ On the other hand, microwave organic synthesis affords the advantages of greater reactivity.³⁵ Recently, click chemistry has been explored for the formation of ketenimine and subsequent reaction with nucleophiles for the construction of diverse structural scaffolds.³⁶ The present study describes an effective route for synthesis of coumarin-3-*N*-sulfonylimidate derivatives using microwave-assisted copper-catalyzed four-component tandem reaction of readily available synthetic precursors such as alkyl propiolates, sulfonyl azides and 2-hydroxybenzaldehydes.

Firstly, the reaction conditions was optimized using salicylaldehyde **1a**, tosyl azide **5a**, ethyl propiolates **8a**, and diisopropylamine **9a** as model substrates using a series of copper salts in the presence of different bases in various solvents (Table 1). The reaction using 10 mol % CuI and 1.2 equiv K₃PO₄ for 0.5 h in 1,4-dioxane at ambient temperature produced amidine³⁷ **10** as the sole product whereas the aldehyde **1a** failed to react (entry 1). However, increasing the reaction temperature to reflux for 24 h led to the coupling of all the substrates to afford a mixture of the ester **11** and coumarin **12a** in 19% and 30%, respectively, along with amidine **10** in 48% (entry 2). Subsequent screening of the base led to increase the yield of desired heterocycle **12a** to 50% in the presence of K₂CO₃, whereas Cs₂CO₃ showed inferior results (entries 3-5). In contrast, Na₂CO₃, and organic bases such as DBU, Et₃N and 2,6-lutidine are not effective (entries 5-8). In a set of copper sources screened, CuI, CuCl, CuBr and Cu(acac)₂, the former afforded the best result (entries 3, 9-11). In case of solvents, dioxane exhibited superior result compared to that of

DMSO, toluene and DMF (entries 3, 12-14). Recrystallization of **11** in MeOH yielded single crystal whose structure was confirmed by X-ray analysis (Figure 2).

Table 1. Optimization of the Reaction Conditions^a



Entry	[Cu]	Base	Solvent	Yield (%) ^b		
				10	11	12a
1	CuI	K ₃ PO ₄	1,4-dioxane	<99	n.d.	n.d. ^c
2	CuI	K ₃ PO ₄	1,4-dioxane	48	19	30
3	CuI	K₂CO₃	1,4-dioxane	34	14	50
4	CuI	Cs ₂ CO ₃	1,4-dioxane	38	39	16
5	CuI	Na ₂ CO ₃	1,4-dioxane	83	17	n.d.
6	CuI	Et ₃ N	1,4-dioxane	<99	n.d.	n.d.
7	CuI	Lutidine	1,4-dioxane	<99	n.d.	n.d.
8	CuI	DBU	1,4-dioxane	75	25	n.d.
9	CuBr	K ₂ CO ₃	1,4-dioxane	44	12	37
10	CuCl	K ₂ CO ₃	1,4-dioxane	30	25	39
11	Cu(acac) ₂	K ₂ CO ₃	1,4-dioxane	60	25	9
12	CuI	K ₂ CO ₃	DMSO	71	n.d.	23
13	CuI	K ₂ CO ₃	Toluene	50	8	36
14	CuI	K ₂ CO	DMF	64	19	n.d.

^a Aldehyde **1a** (0.5 mmol), tosyl azide **5a** (0.6 mmol), ethyl propiolate **8a** (0.5 mmol), amine **9a** (0.6 mmol), Cu source (10 mol %), base (0.6 mmol), solvent (1 mL), 0.5 h, rt; reflux, 24 h. ^b Determined by 400 MHz ¹H NMR. ^c 0.5 h, rt. n.d.= not detected.

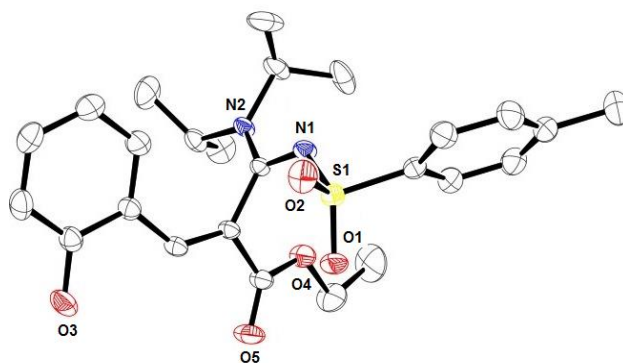


Figure 2. ORTEP diagram of (*Z*)-3-((*Z*)-*N,N*-diisopropyl-*N'*-tosylcarbamimidoyl)-4-(2-hydroxyphenyl)-2-oxobut-3-enoate **11** (CCDC 1040055). Thermal ellipsoids are drawn at the 50% probability level (Hydrogen atoms have been omitted for clarity).

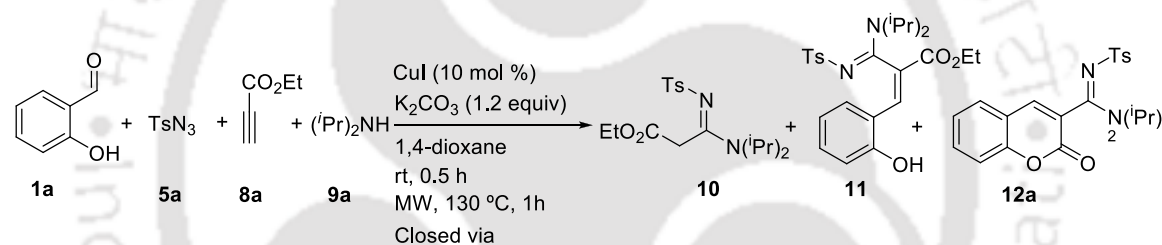
Interestingly, the use of microwave-heating led to the formation **12a** in high yields and table 2 summarizes the results. The reactivity and selectivity are greater under microwave heating compared to the conventional heating. The best results have been obtained using 10 mol % of CuI in the presence of 1.2 equiv of K₂CO₃ in dioxane at 130 °C with microwave power 150 W in closed vial (entry 1). Further increase of the temperature to 140 °C led to drop the yield to 81% due to decomposition of **12a** (entry 1). Similarly, decrease of the catalyst loading (5 mol %) (entry 2) or reaction temperature (120 °C) (entry 4) or increase of base (2 equiv) (entry 6) led to inferior results. Control experiment confirmed that the target product was not formed in the absence of the copper catalyst. The reaction in sealed tube without microwave irradiation produced **12a** in 34% yield (entry 7).

With optimized conditions, the reaction of a series of substituted salicylaldehydes was explored (Table 3). Aldehydes **1b-d** with 3-chloro, 3-methoxy and 3-*tert*-butyl groups underwent reaction to afford coumarins **12b-d** in 60-83% yields. The reaction of the aldehydes **1e-g** bearing 4-alkoxy group afforded the desired **12e-g** in 25-67% yields. Aldehydes **1h-j** and **1l-n** having functionality at 5-position with bromo, chloro, fluoro, methoxy and methyl substituents readily reacted to provide coumarin derivatives **12h-j** and **12l-n** in 65-78% yields. The substrates **1k** and **1o** having strong electron withdrawing groups such as 5-CHO and 5-NO₂ showed no reaction. However, the aldehydes **1p-q** having sterically hindered substituents such as 5-iodo-3-*tert*-butyl and 3,5-di-*tert*-butyl groups underwent reaction to offer **12p-q** in 76-80% yields. Moreover, 2-naphthaldehyde reacted smoothly to furnish fused benzene ring substituted coumarin **12r** in good yield,

which was crystallized in MeOH-CH₂Cl₂ (1:1) and its structure was determined using single crystal X-ray analysis (Figure 2). These results suggest that the aldehydes having electron donating groups afford the best results compared to that having withdrawing substituents.

Next the reaction of different sulfonyl azides was studied using salicylaldehyde **1a**, ethyl propiolate **8a** and diisopropylamine **9a** as representative examples (Table 5). The reaction of methanesulfonyl azide **5b** produced coumarin **12s** in 72% yield, while phenyl sulfonyl azide **3c** underwent reaction to give **12t** in 80% yield. Similarly, aryl sulfonyl azides **5d-f** bearing 4-chloro, 4-methoxy and 2,4,6-trimethyl substituents underwent reaction to afford the corresponding coumarin analogues **12u-w** in 68-82% yields. These results clearly suggests that the reaction can be compatible to both alkyl and aryl sulfonyl azides in good yields.

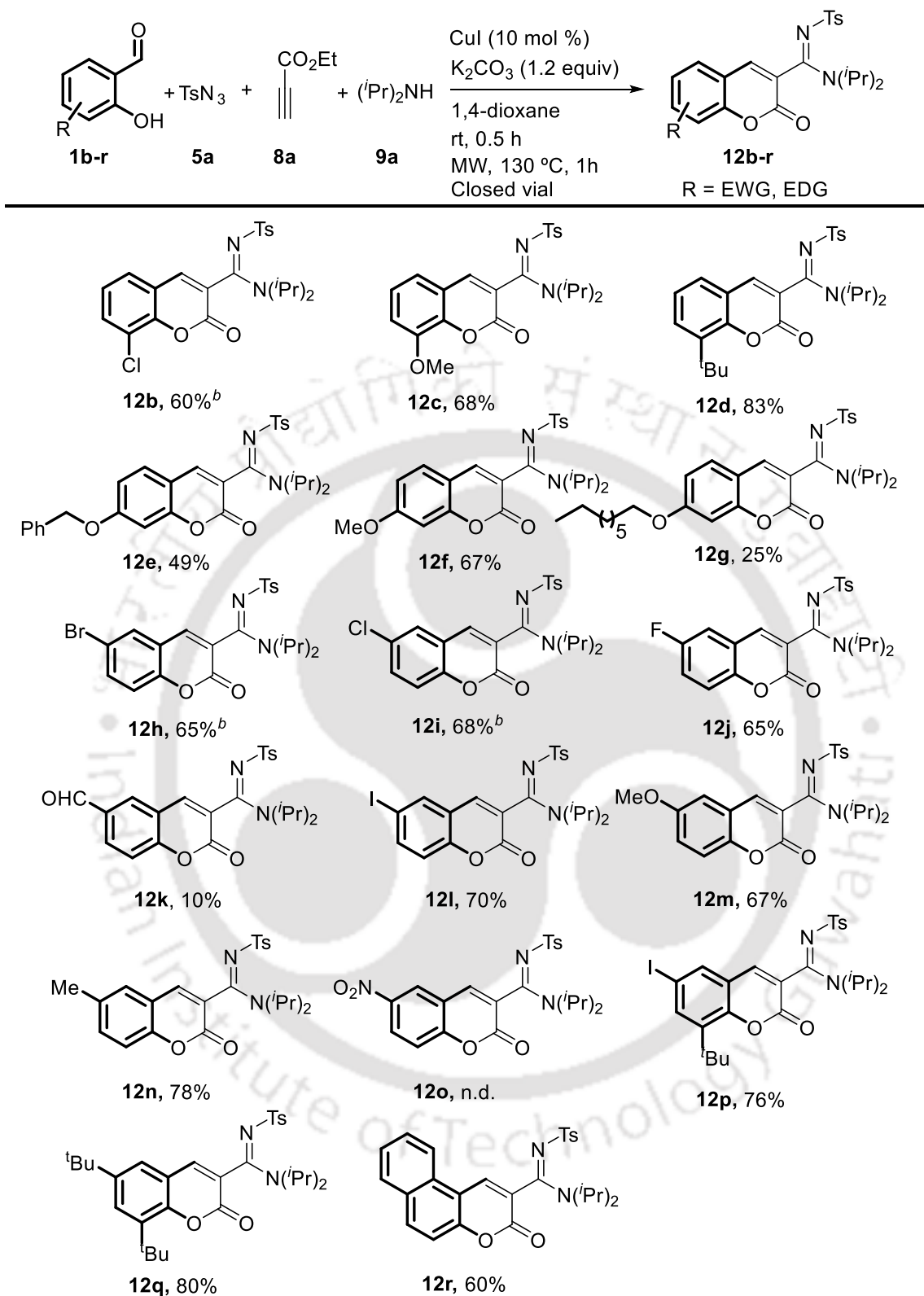
Table 2. Optimization of Reaction Conditions Using Microwave Heating^a



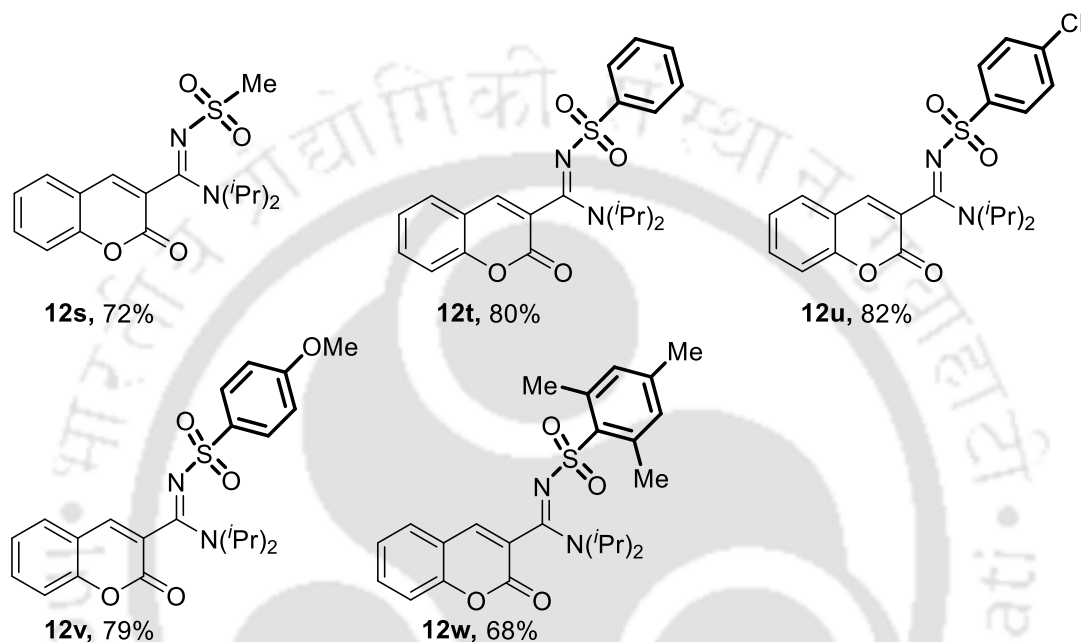
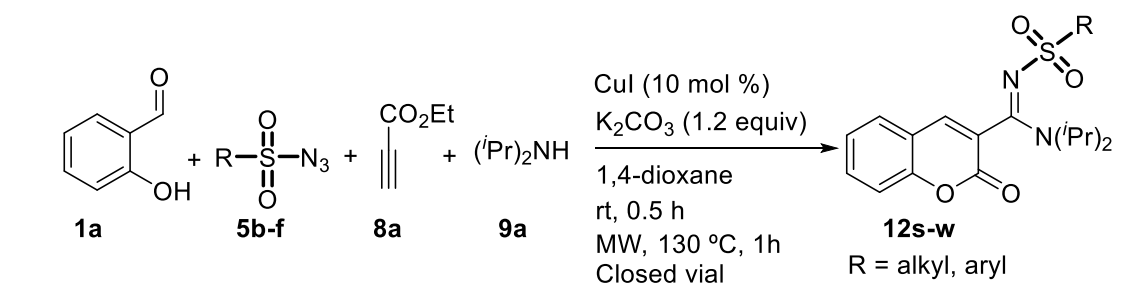
Entry	<i>T</i> (°C)	Yield (%) ^b		
		10	11	12a
1	130	n.d.	7	88
2	130	n.d.	3	70 ^c
3	130	n.d.	25	72 ^d
4	120	14	54	30
5	140	n.d.	n.d.	81
6	130	16	18	63 ^e
7	130	27	38	34 ^f

^aAldehyde **1a** (0.5 mmol), tosyl azide **5a** (0.6 mmol), ethyl propiolate **8a** (0.5 mmol), amine **9a** (0.6 mmol), CuI (10 mol %), K₂CO₃ (0.6 mmol), 1,4-dioxane (1 mL), 0.5 h, rt; MW, 130 °C, 1 h. ^b Determined by 400 MHz ¹H NMR. ^c 5 mol % CuI. ^d 0.5 h ^e 2 equiv K₂CO₃. ^f Sealed tube without MW, 1 h. n.d. = not detected.

Table 3. Reaction of Substituted Salicylaldehydes^a



^aAldehyde **1** (0.5 mmol), **5a** (0.6 mmol), **8a** (0.5 mmol), **9a** (0.6 mmol), CuI (10 mol %), K₂CO₃ (0.6 mmol), 1,4-dioxane (1 mL), rt, 0.5 h, air; MW, 130 °C, 1 h. ^b Ester (~5%) was obtained as byproduct. n.d. = not detected.

Table 4. Reaction of Sulfonyl Azides^a

^aAldehyde **1a** (0.5 mmol), **5b-f** (0.6 mmol), **8a** (0.5 mmol), **9a** (0.6 mmol), CuI (10 mol %), K₂CO₃ (1.2 equiv), 1,4-dioxane (1 mL), rt, 0.5 h, air; MW, 130 °C, 1 h.

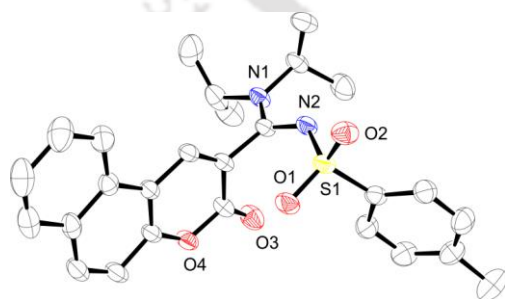
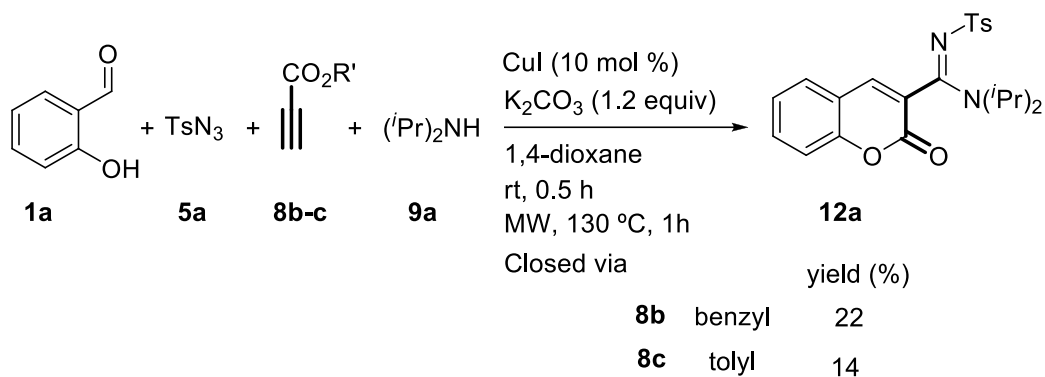
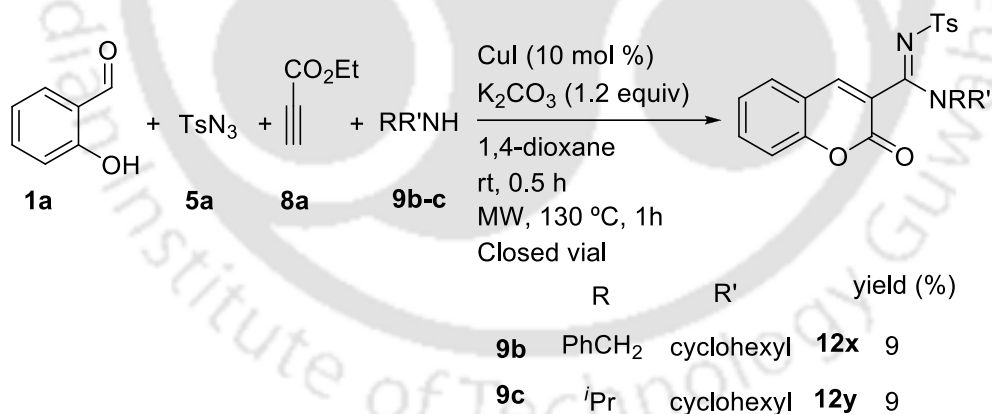


Figure 3. ORTEP diagram of the compound (*Z*)-*N,N*-Diisopropyl-3-oxo-*N'*-tosyl-3*H*-benzo[*f*]chromene-2-carboximidamide **12r** (CCDC 1040054). Thermal ellipsoids are drawn at the 50% probability level (Hydrogen atoms have been omitted for clarity).



Scheme 18

Next, the reaction of different propiolates was investigated (Scheme 18). Benzyl propiolate **8b** underwent reaction to give target heterocycle in 22% yield, while *p*-tolyl propiolate **8c** produced the target heterocycle **12a** in 14% yield. Finally, the reaction with different amines was performed (Scheme 19). Cyclohexylamine and morpholine failed to produce the coupled products due to decomposition. However, benzyl cyclohexylamine **9b** underwent reaction to give **12x** as a 3:1 mixture of isomers in 9% yield. Likewise isopropyl cyclohexylamine **9c** gave **12y** as a 1:1 mixture of isomers in 9% yield. These results suggest that the nature of propiolate and secondary amine plays a crucial role in this coupling reaction.



Scheme 19

To gain insight into the catalytic pathway, the reaction of **1a**, **5a**, **8a** and **9a** was stopped at 10 min and the resulting mixture was analyzed using ESI mass analysis, and found three major species **10**, **11** and **12a** (Figure 4). Furthermore, the substrates **5a**, **8a** and **9a** readily underwent coupling to provide **10** that could be readily reacted with **1a** to furnish **12a** in 78% yield (Scheme 20a). In addition, the ester **11** readily underwent

cyclization to furnish **12a** in 85% yield (Scheme 20b). These results suggest that reaction may take place *via* the intermediates **10** and **11** to yield the target heterocycle **12a**.

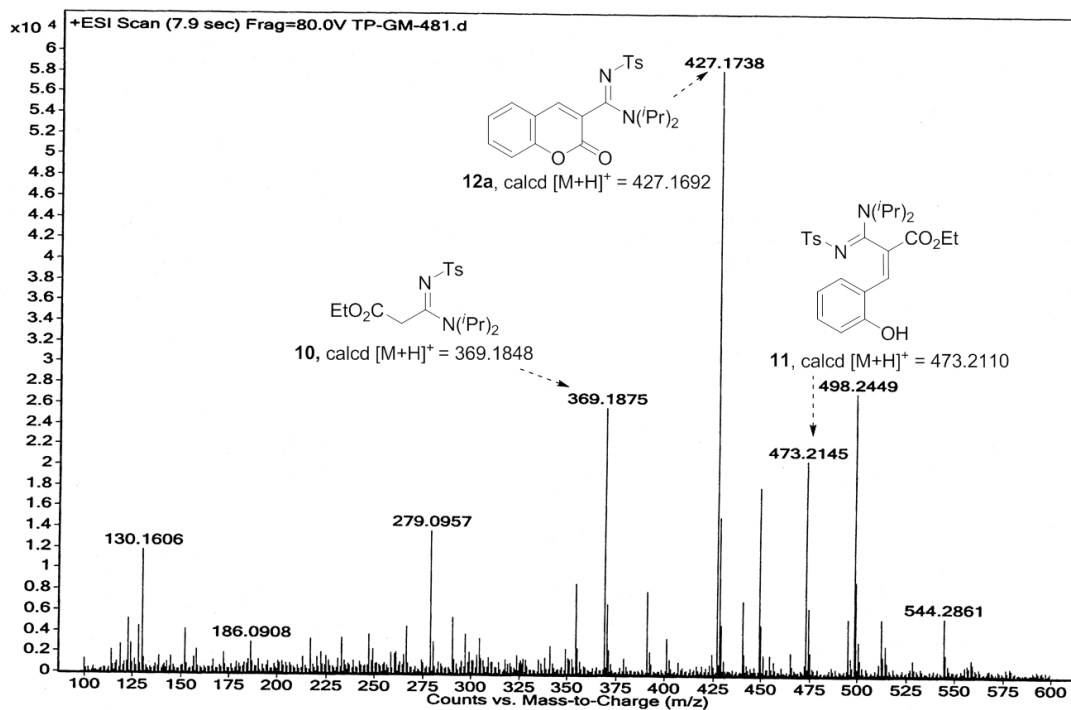
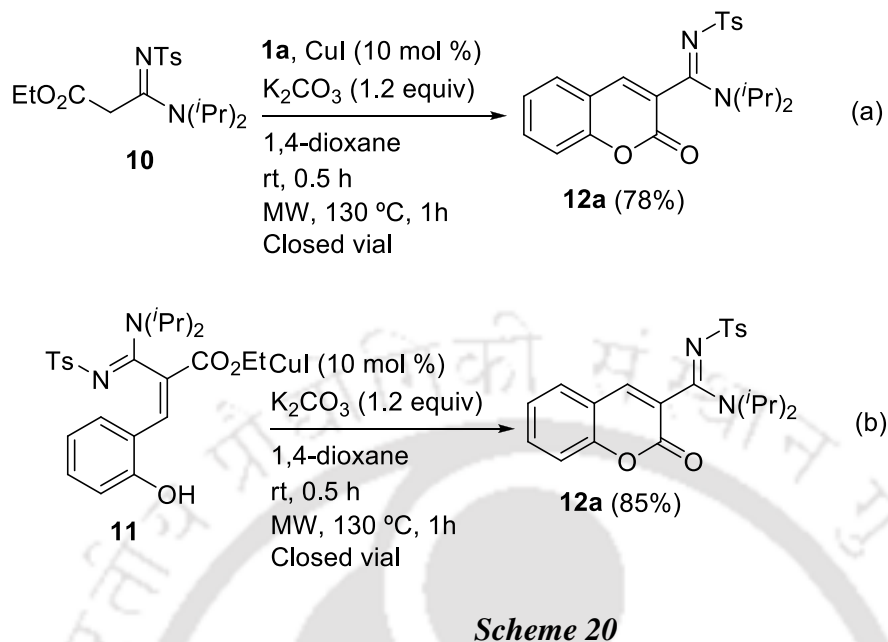
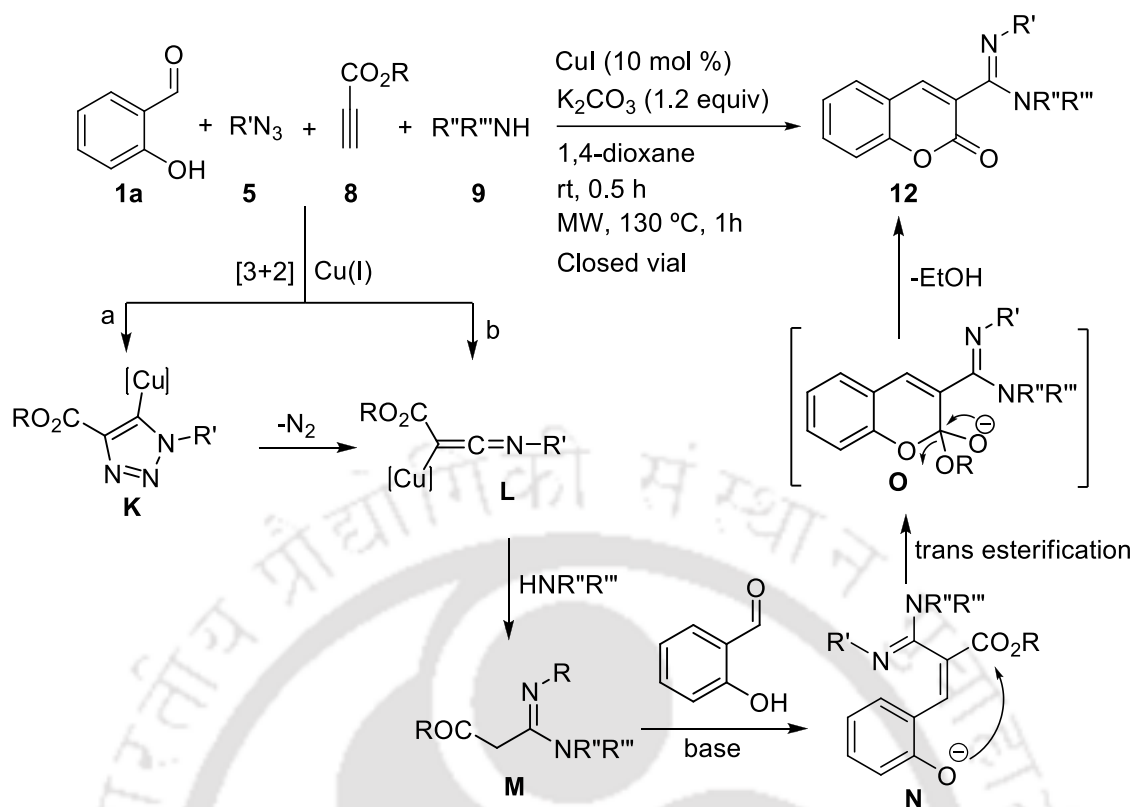


Figure 4. ESI Mass Analysis of the Reaction Mixture of **1a**, **5a**, **8a**, and **9a** after 0.1 h.



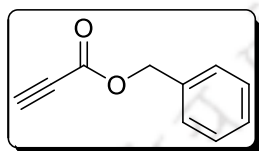
Scheme 21

The proposed catalytic cycle is shown in Scheme 21. The reaction may take place *via* the intermediates **M** and **N** to provide the target coumarin motifs. Thus, the cycloaddition of propiolates **8** with sulfonyl azides **5** may produce ketenimine **L** *via* intermediate **K**. Nucleophilic addition of the amine **9** to the intermediate **J** may furnish **G** that can react with the salicylaldehyde **1a** to give **N**, which may cyclize to produce coumarin derivatives by trans-esterification.

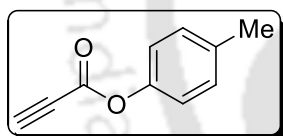
In conclusion, copper-catalyzed four-component synthesis of 3-*N*-sulfonylamidine coumarin is developed *via* the coupling of salicylaldehydes, propiolates, sulfonyl azides and secondary amines. The reaction using microwave irradiation is found to be superior to the conventional heating process. The greater reactivity, mild reaction condition and high selectivity constitute the significant practical advantages. This study may open new avenue for the further development of multicomponent studies for the synthesis of highly functionalized coumarin derivatives

4.2 Experimental Section

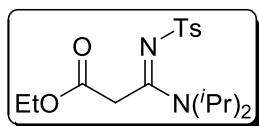
General Information. The reaction was performed in closed vial using *CEM Discover LabMate* microwave reactor equipped with surface sensor for temperature measurement. CuI (98%) of Aldrich, CuBr (97%) of Alfa Aesar and CuCl (98%) of Rankem were used as received. 2,6-Lutidine (98%), ethyl propiolate (99%), Cs₂CO₃ (99%), K₃PO₄ (98%), Na₂CO₃ (99%) and DBU (98%) were purchased from commercial suppliers and used as received. Solvents were purchased from commercial source and purified prior to use.³⁸ Substituted salicylaldehydes^{39,40} and sulfonyl azides⁴¹ were prepared.



Benzyl propiolate 8b:⁴² analytical TLC on silica gel, 1:19 ethyl acetate-hexane $R_f = 0.50$; colorless liquid; yield 70% (561 mg); ¹H NMR (400 MHz, CDCl₃) δ 7.40-7.37 (m, 5H), 5.23 (s, 2H), 2.90 (s, 1H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 152.8, 134.7, 128.93, 128.9, 128.8, 75.3, 74.7, 68.1; FT-IR (neat) 2961, 2924, 2855, 2120, 1716, 1605, 1383, 1223, 1020, 749, 696, 668, 562 cm⁻¹.

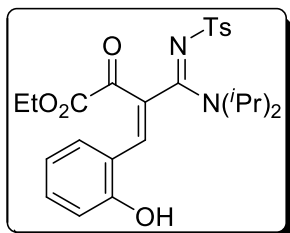


***p*-Tolyl propiolate 8c:**⁴³ analytical TLC on silica gel, 1:19 ethyl acetate-hexane $R_f = 0.30$; colorless liquid; yield 53% (424 mg); ¹H NMR (400 MHz, CDCl₃) δ 7.20 (d, $J = 8.4$ Hz, 2H), 7.03 (d, $J = 8.4$ Hz, 2H), 3.06 (s, 1H), 2.35 (s, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 151.4, 147.8, 136.6, 130.3, 121.1, 76.8, 74.5, 21.1; FT-IR (neat) 2962, 2924, 2855, 2125, 1731, 1504, 1384, 1217, 1197, 1018, 909, 807, 744, 607, 502 cm⁻¹.



Ethyl (Z)-3-(diisopropylamino)-3-(tosylimino)propanoate (10):³⁷ analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.40$; colorless solid; mp 122-123 °C. ¹H NMR (400 MHz, CDCl₃) δ 7.80 (d, $J = 8.0$ Hz, 2H), 7.24 (d, $J = 7.6$ Hz, 2H), 4.16-4.11 (m, 4H), 3.95-3.88 (m, 1H), 3.61 (br. s, 1H), 2.38 (s, 3H), 1.38 (d, $J = 6.8$ Hz, 6H), 1.27-1.21 (m, 9H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 166.9, 157.9, 141.7, 141.1, 129.0, 126.2, 61.7, 48.4, 37.6, 21.4, 20.2, 19.9, 14.0; FT-IR (KBr) 3015, 2983,

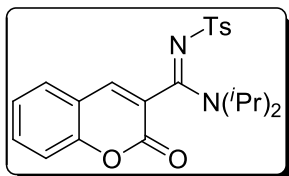
2936, 2907, 1729, 1547, 1484, 1441, 1369, 1325, 1276, 1201, 1135, 1083, 1055, 960, 885, 810, 763, 715, 663, 551, 542 cm⁻¹; HRMS (ESI) *m/z* calcd for C₁₈H₂₉N₂O₄S [M+H]⁺: 369.1848, found: 369.1875.



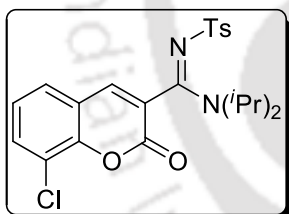
(Z)-3-((Z)-*N,N*-diisopropyl-*N'*-tosylcarbamimidoyl)-4-(2-hydroxyphenyl)-2-oxobut-3-enoate (11): analytical TLC on silica gel, 2:3 ethyl acetate-hexane *R_f* = 0.32; colorless solid; mp 206-207 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.22 (s, 1H), 7.87 (s, 1H), 7.72 (d, *J* = 8.4 Hz, 2H), 7.43 (dd, *J* = 8.4, 1.6 Hz, 1H), 7.11-7.06 (m, 3H), 6.86 (d, *J* = 7.6 Hz, 1H), 6.62 (t, *J* = 8.0 Hz, 1H), 4.30-4.22 (m, 2H), 4.09-4.01 (m, 1H), 3.63-3.55 (m, 1H), 2.28 (s, 3H), 1.57 (d, *J* = 6.8 Hz, 3H), 1.49 (d, *J* = 6.8 Hz, 3H), 1.30 (t, *J* = 7.2 Hz, 3H), 1.11 (d, *J* = 6.4 Hz, 3H), 0.74 (d, *J* = 6.8 Hz, 3H); ¹³C{¹H} NMR (100 MHz, CDCl₃) δ 165.0, 160.9, 156.8, 141.8, 40.5, 136.7, 132.5, 129.0, 129.0, 126.6, 122.9, 119.8, 119.6, 116.7, 61.7, 52.4, 48.4, 21.5, 20.2, 19.7, 19.0, 14.2; FT-IR (KBr) 3390, 2973, 2924, 2853, 1717, 1700, 1618, 1606, 1536, 1463, 1443, 1367, 1279, 1253, 1219, 1142, 1084, 1036, 1017, 906, 813, 776, 760, 675, 596, 554 cm⁻¹; HRMS (ESI) *m/z* calcd for C₂₅H₃₃N₂O₅S [M+H]⁺: 473.2110, found: 473.2117.

General Procedure for the Synthesis of Coumarins

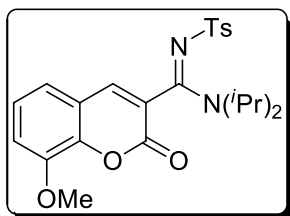
To a stirred solution of sulfonyl azide **5** (0.6 mmol), propiolate **8** (0.5 mmol) and CuI (0.05 mmol, 9.5 mg) in 1,4-dioxane (1 mL) was added secondary amine **9** (0.5 mmol), salicylaldehyde **1** (0.5 mmol) and K₂CO₃ (0.6 mmol, 82.9 mg) at room temperature under air. After 0.5 h (arrested N₂ bubbles), the reaction vial was sealed with a cap and stirred at 130 °C [150 W] for 1 h using microwave-irradiation. The solvent was then evaporated under *vacuo* and the residue was diluted with CH₂Cl₂ (30 mL), and washed with saturated NH₄Cl (10 mL) and water (10 mL). Drying (Na₂SO₄) and evaporation of the solvent gave a residue that was purified on silica gel column chromatography using n-hexane/ethyl acetate as eluent.



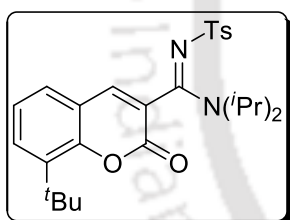
(Z)-*N,N*-Diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12a): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.40$; colorless solid; yield 75% (160 mg); mp 257-258 °C; ^1H NMR (600 MHz, CDCl_3) δ 7.72 (s, 1H, H-4), 7.65 (d, $J = 7.8$ Hz, 2H, Ar-H (tosyl)), 7.58 (t, $J = 7.8$ Hz, 1H, H-7), 7.54 (d, $J = 7.8$ Hz, 1H, H-5), 7.34-7.30 (m, 2H, H-6 and H-8), 7.16 (d, $J = 7.8$ Hz, 2H, Ar-H (tosyl)), 3.89-3.85 (m, 1H, -N-CH-), 3.70-3.66 (m, 1H, -N-CH-), 2.36 (s, 3H, -CH₃ (tosyl)), 1.58 (d, $J = 6.6$ Hz, 3H, -CH₃ (isopropyl)), 1.42 (d, $J = 7.2$ Hz, 3H, -CH₃ (isopropyl)), 1.26 (d, $J = 6.6$ Hz, 3H, -CH₃ (isopropyl)), 1.14 (d, $J = 6.6$ Hz, 3H, -CH₃ (isopropyl)); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 157.5 (C), 157.4 (C), 154.2 (C), 142.1 (C), 141.6 (CH), 140.8 (C), 132.9 (CH), 129.2 (CH), 129.0 (CH), 126.5 (CH), 125.1 (CH), 123.3 (C), 118.2 (C), 117.1 (CH), 52.8 (CH), 48.6 (CH), 21.6 (CH₃), 20.7 (CH₃), 20.2 (CH₃), 20.1 (CH₃), 19.7 (CH₃); FT-IR (KBr) 3035, 2969, 2932, 1717, 1628, 1608, 1547, 1445, 1371, 1279, 1251, 1210, 1142, 1087, 1013, 901, 812, 780, 759, 676, 595, 553 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{27}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 427.1692, found: 427.1692.



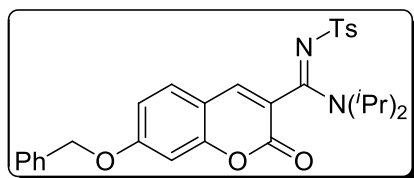
(Z)-8-Chloro-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12b): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.37$; colorless solid; 60% (138 mg); mp 285-286 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.75 (s, 1H), 7.66-7.62 (m, 3H), 7.48 (d, $J = 7.6$ Hz, 1H), 7.30 (d, $J = 8.0$ Hz, 1H), 7.17 (d, $J = 7.6$ Hz, 2H), 3.86-3.81 (m, 1H), 3.72-3.66 (m, 1H), 2.38 (s, 3H), 1.60 (d, $J = 6.8$ Hz, 3H), 1.43 (d, $J = 7.2$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.16 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.8, 156.3, 149.9, 142.3, 141.2, 140.6, 133.2, 129.2, 127.5, 126.6, 125.2, 124.0, 122.1, 119.5, 52.9, 48.7, 21.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3079, 2998, 2973, 2927, 1727, 1623, 1600, 1547, 1476, 1442, 1367, 1270, 1208, 1138, 1087, 1055, 1014, 903, 838, 787, 756, 682, 593, 551 cm^{-1} ; HRMS (APCI) m/z calcd for $\text{C}_{23}\text{H}_{26}\text{ClN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 461.1302, found: 461.1301.



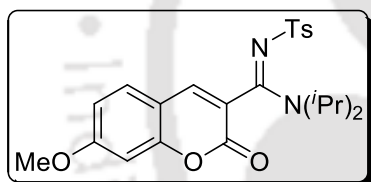
(Z)-*N,N*-Diisopropyl-8-methoxy-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12c): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.50$; colorless solid; yield 68% (155 mg); mp 294-295 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.71 (s, 1H), 7.66 (d, $J = 8.0$ Hz, 2H), 7.26 (d, $J = 10.0$ Hz, 1H), 7.17-7.11 (m, 4H), 3.97 (s, 3H), 3.88-3.84 (m, 1H), 3.69-3.65 (m, 1H), 2.37 (s, 3H), 1.59 (d, $J = 6.8$ Hz, 3H), 1.41 (d, $J = 6.8$ Hz, 3H), 1.25 (d, $J = 6.8$ Hz, 3H), 1.14 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.3, 156.9, 147.4, 143.9, 142.0, 141.7, 140.8, 129.1, 126.6, 124.9, 123.5, 120.3, 118.8, 114.8, 56.5, 52.7, 48.5, 21.6, 20.6, 20.2, 20.1, 19.7; FT-IR (KBr) 3080, 3032, 2995, 2967, 2936, 2844, 1723, 1623, 1610, 1578, 1543, 1489, 1453, 1369, 1267, 1213, 1175, 1135, 1104, 1085, 1059, 1016, 975, 950, 904, 839, 809, 787, 752, 677, 595, 551 cm^{-1} ; HRMS (APCI) m/z calcd for $\text{C}_{24}\text{H}_{29}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]^+$: 457.1797, found: 457.1797.



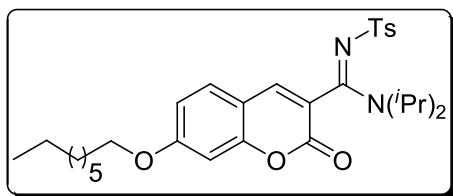
(Z)-8-(*tert*-Butyl)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12d): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.53$; colorless solid; yield 83% (200 mg); mp 249-250 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.77 (s, 1H), 7.59 (d, $J = 8.0$ Hz, 3H), 7.42 (d, $J = 7.6$ Hz, 1H), 7.26 (t, $J = 8.0$ Hz, 1H), 7.13 (d, $J = 8.0$ Hz, 2H), 3.87-3.84 (m, 1H), 3.71-3.68 (m, 1H), 2.33 (s, 3H), 1.61 (d, $J = 7.2$ Hz, 3H), 1.48 (s, 12H), 1.27 (d, $J = 6.0$ Hz, 3H), 1.15 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.7, 156.6, 152.8, 143.2, 141.9, 140.7, 138.1, 130.6, 129.1, 127.5, 126.5, 124.6, 121.4, 118.4, 52.7, 48.5, 35.1, 30.0, 21.5, 20.6, 20.2, 20.0, 19.6; FT-IR (KBr) 2996, 2973, 2936, 2881, 1727, 1626, 1593, 1542, 1449, 1432, 1370, 1278, 1213, 1172, 1142, 1122, 1085, 1059, 1016, 941, 909, 819, 801, 754, 678, 592, 549 cm^{-1} ; HRMS (APCI) m/z calcd for $\text{C}_{27}\text{H}_{35}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 483.2318, found: 483.2318.

**(Z)-7-(Benzyloxy)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-**

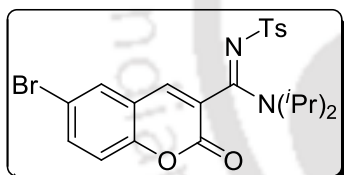
2*H*-chromene-3-carboximidamide (12e): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.30$; yellow solid; yield 49% (131 mg); mp 222-223 °C. ^1H NMR (600 MHz, CDCl_3) δ 7.66 (s, 1H), 7.64 (d, $J = 7.8$ Hz, 2H), 7.42-7.41 (m, 5H), 7.36 (d, $J = 6.0$ Hz, 1H), 7.14 (d, $J = 7.2$ Hz, 2H), 6.96 (d, $J = 7.8$ Hz, 1H), 6.86 (s, 1H), 5.15 (s, 2H), 3.89-3.87 (m, 1H), 3.66 (s, 1H), 2.35 (s, 3H), 1.58 (d, $J = 6.6$ Hz, 3H), 1.43 (d, $J = 6.0$ Hz, 3H), 1.25 (d, $J = 6.0$ Hz, 3H), 1.13 (d, $J = 6.0$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 162.8, 157.9, 157.7, 156.1, 142.0, 141.9, 140.9, 137.8, 130.1, 129.1, 129.0, 128.6, 127.6, 126.5, 119.4, 114.0, 112.0, 102.1, 70.8, 52.7, 48.5, 21.7, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 2967, 2923, 2856, 1729, 1616, 1538, 1451, 1373, 1267, 1243, 1122, 1082, 808, 754, 679, 619, 550 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{30}\text{H}_{33}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]$: 533.2105, found: 533.2112.

**(Z)-*N,N*-Diisopropyl-7-methoxy-2-oxo-*N'*-tosyl-2*H*-**

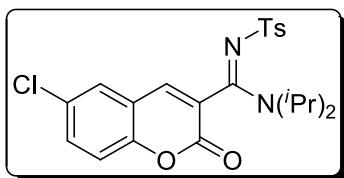
chromene-3-carboximidamide (12f): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.58$; colorless solid; yield 67% (153 mg); mp 265-266 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.67 (s, 1H), 7.65 (d, $J = 8.0$ Hz, 2H), 7.45 (d, $J = 8.4$ Hz, 1H), 7.17 (d, $J = 7.6$ Hz, 2H), 6.90 (dd, $J = 8.8, 1.2$ Hz, 1H), 6.81 (s, 1H), 3.91-3.86 (m, 4H), 3.70-3.64 (m, 1H), 2.37 (s, 3H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.43 (d, $J = 6.8$ Hz, 3H), 1.26 (d, $J = 6.8$ Hz, 3H), 1.14 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 163.8, 157.9, 157.7, 156.2, 141.95, 141.91, 140.9, 130.0, 129.1, 126.5, 119.4, 113.3, 111.8, 101.0, 56.1, 52.7, 48.5, 21.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3093, 3052, 2972, 2934, 2849, 1730, 1614, 1540, 1512, 1473, 1459, 1444, 1371, 1271, 1244, 1202, 1125, 1080, 1057, 1024, 972, 901, 854, 807, 763, 726, 675, 593, 550 cm^{-1} ; HRMS (APCI) m/z calcd for $\text{C}_{24}\text{H}_{29}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]^+$: 457.1797, found; 457.1798.



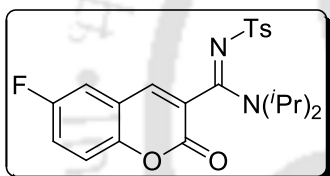
(Z)-*N,N*-Diisopropyl-7-(octyloxy)-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12g): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.32$; pale yellow solid; yield 25% (69 mg); mp 198-199 °C. ^1H NMR (600 MHz, CDCl_3) δ 7.66 (s, 1H), 7.65 (d, $J = 7.8$ Hz, 2H), 7.43 (d, $J = 8.4$ Hz, 1H), 7.17 (d, $J = 7.8$ Hz, 2H), 6.88 (dd, $J = 9.0, 1.8$ Hz, 1H), 6.79 (s, 1H), 4.03 (t, $J = 6.6$ Hz, 2H), 3.91-3.89 (m, 1H), 3.68-3.66 (m, 1H), 2.37 (s, 3H), 1.84-1.80 (m, 2H), 1.58 (d, $J = 6.6$ Hz, 3H), 1.49-1.44 (m, 2H), 1.43 (d, $J = 7.2$ Hz, 3H), 1.37-1.29 (m, 8H), 1.26 (d, $J = 6.6$ Hz, 3H), 1.14 (d, $J = 6.6$ Hz, 3H), 0.90 (t, $J = 6.6$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 163.4, 158.0, 157.8, 156.2, 142.0, 141.9, 141.0, 130.0, 129.1, 126.6, 119.1, 113.7, 111.6, 101.5, 69.0, 52.7, 48.5, 32.0, 29.5, 29.4, 29.1, 26.1, 22.8, 21.6, 20.7, 20.2, 20.1, 19.7, 14.3; FT-IR (KBr) 2967, 2922, 2855, 1728, 1619, 1539, 1373, 1294, 1272, 1251, 1138, 1081, 806, 789, 682, 552 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{30}\text{H}_{33}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]^+$: 555.2887, found: 555.2895.



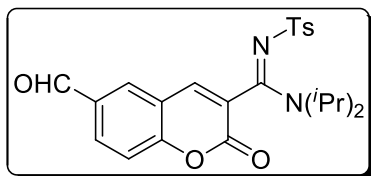
(Z)-6-Bromo-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12h): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.70$; yellow solid; yield 65% (164 mg); mp 308-309 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.67 (d, $J = 6.4$ Hz, 5H), 7.25 (d, $J = 8.8$ Hz, 1H), 7.19 (d, $J = 8.0$ Hz, 2H), 3.85-3.82 (m, 1H), 3.70-3.66 (m, 1H), 2.38 (s, 3H), 1.57 (d, $J = 6.4$ Hz, 3H), 1.41 (d, $J = 6.4$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.15 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.9, 156.8, 153.0, 142.1, 140.7, 140.0, 135.5, 131.2, 129.2, 126.5, 124.6, 119.7, 118.8, 117.6, 52.9, 48.6, 21.6, 20.6, 20.1, 19.7; FT-IR (KBr) 3110, 3045, 2980, 2966, 2925, 1726, 1623, 1602, 1542, 1471, 1448, 1372, 1271, 1241, 1207, 1136, 1083, 1016, 941, 904, 828, 809, 761, 683, 621, 549 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{26}\text{BrN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 505.0797, found: 505.0800.



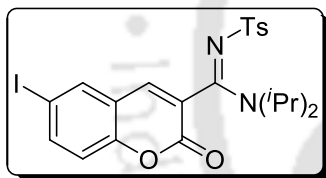
(Z)-6-Chloro-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12i): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.58$; yellow solid; yield 68% (156 mg); mp 314-315 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.67 (d, $J = 6.0$ Hz, 3H), 7.52 (s, 2H), 7.31 (d, $J = 9.6$ Hz, 1H), 7.19 (d, $J = 7.6$ Hz, 2H), 3.85-3.80 (m, 1H), 3.69-3.64 (m, 1H), 2.37 (s, 3H), 1.57 (d, $J = 7.2$ Hz, 3H), 1.40 (d, $J = 6.8$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.15 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.0, 156.8, 152.6, 142.1, 140.7, 140.1, 132.7, 130.4, 129.2, 128.2, 126.5, 124.6, 119.2, 118.5, 52.9, 48.6, 21.6, 20.7, 20.1, 19.7; FT-IR (KBr) 3044, 2981, 2925, 1727, 1625, 1544, 1473, 1447, 1413, 1373, 1271, 1241, 1208, 1134, 1084, 1058, 1017, 920, 837, 828, 809, 762, 684, 549 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{26}\text{ClN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 461.1302, found: 461.1296.



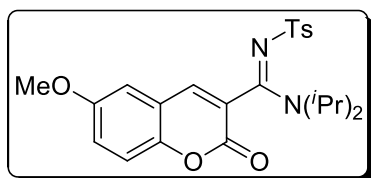
(Z)-6-Fluoro-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12j): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.47$; yellow solid; yield 65% (144 mg); mp 303-304 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.68 (d, $J = 8.4$ Hz, 3H), 7.33-7.28 (m, 2H), 7.24 (d, $J = 6.4$ Hz, 1H), 7.20 (d, $J = 8.0$ Hz, 2H), 3.86-3.81 (m, 1H), 3.70-3.65 (m, 1H), 2.38 (s, 3H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.41 (d, $J = 6.4$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.16 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 159.9 (d, $J_{\text{C-F}} = 244.5$ Hz), 157.2, 156.9, 150.4, 142.1, 140.7, 140.3, 129.2, 126.5, 124.7, 120.4 (d, $J_{\text{C-F}} = 24$ Hz), 118.9 (d, $J_{\text{C-F}} = 13.5$ Hz), 118.7 (d, $J_{\text{C-F}} = 9$ Hz), 114.3 (d, $J_{\text{C-F}} = 24$ Hz), 52.8, 48.6, 21.6, 20.7, 20.1, 19.7; ^{19}F NMR (376 MHz, CDCl_3 , CF_3COOH as internal reference = -76.55) -116.6; FT-IR (KBr) 3045, 2984, 2925, 1725, 1577, 1545, 1490, 1432, 1376, 1267, 1211, 1136, 1085, 1018, 883, 775, 755, 686, 549 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{25}\text{FN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 445.1597, found: 445.1589.



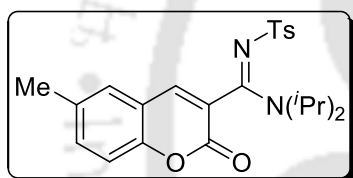
(Z)-6-Formyl-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12k): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.33$; colorless solid; yield 10% (23 mg); mp 261-262 °C; ^1H NMR (600 MHz, CDCl_3) δ 10.04 (s, 1H), 8.11 (dd, $J = 8.4, 1.8$ Hz, 1H), 8.08 (d, $J = 1.2$ Hz, 1H), 7.82 (s, 1H), 7.69 (d, $J = 7.8$ Hz, 2H), 7.50 (d, $J = 8.4$ Hz, 1H), 7.20 (d, $J = 8.4$ Hz, 2H), 3.89-3.85 (m, 1H), 3.72-3.67 (m, 1H), 2.38 (s, 3H), 1.58 (d, $J = 7.2$ Hz, 3H), 1.41 (d, $J = 7.2$ Hz, 3H), 1.28 (d, $J = 6.6$ Hz, 3H), 1.17 (d, $J = 6.6$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 190.0, 157.8, 156.7, 156.6, 142.3, 140.6, 140.5, 133.3, 133.0, 131.4, 129.3, 126.5, 124.9, 118.6, 118.3, 53.0, 48.7, 21.7, 20.8, 20.2, 20.1, 19.7; FT-IR (KBr) 2981, 2965, 2924, 2852, 1737, 1700, 1623, 1547, 1447, 1373, 1269, 1168, 1136, 1114, 1084, 1056, 103, 947, 907, 644, 810, 762, 684, 549 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{24}\text{H}_{27}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]^+$: 455.1641, found: 455.1640.



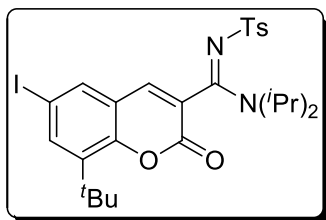
(Z)-6-Iodo-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12l): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.66$; colorless solid; yield 70% (193 mg); mp 277-278 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.86-7.83 (m, 2H), 7.67 (d, $J = 7.6$ Hz, 2H), 7.63 (s, 1H), 7.19 (d, $J = 8.4$ Hz, 2H), 7.12 (d, $J = 8.4$ Hz, 1H), 3.85-3.82 (m, 1H), 3.70-3.66 (m, 1H), 2.38 (s, 3H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.41 (d, $J = 6.8$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.15 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 156.8, 153.8, 142.2, 141.3, 140.7, 139.9, 137.3, 129.2, 126.5, 124.4, 120.2, 119.0, 87.8, 52.9, 48.6, 21.6, 20.7, 20.1, 19.7; FT-IR (KBr) 2981, 2924, 2850, 1727, 1543, 1451, 1371, 1275, 1140, 1083, 1057, 1014, 943, 903, 828, 810, 761, 683, 550 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{26}\text{IN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 553.0658, found: 553.0654.



(Z)-*N,N*-Diisopropyl-6-methoxy-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12m): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.34$; colorless solid; yield 67% (153 mg); mp 264-265 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.71 (s, 1H), 7.66 (d, $J = 7.2$ Hz, 2H), 7.28 (s, 1H), 7.17 (d, $J = 6.8$ Hz, 3H), 6.97 (s, 1H), 3.86 (s, 4H), 3.70-3.67 (m, 1H), 2.37 (s, 3H), 1.59 (d, $J = 6.8$ Hz, 3H), 1.43 (d, $J = 6.4$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.15 (d, $J = 6.0$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.6, 157.5, 156.5, 148.7, 142.0, 141.5, 140.8, 129.2, 126.6, 123.5, 121.0, 118.5, 118.1, 110.7, 56.0, 52.7, 48.5, 21.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3035, 2966, 2932, 1713, 1578, 1550, 1497, 1448, 1367, 1280, 1212, 1142, 1089, 1037, 1017, 965, 904, 879, 814, 806, 768, 755, 680, 595, 553 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{24}\text{H}_{29}\text{N}_2\text{O}_5\text{S}$ $[\text{M}+\text{H}]^+$: 457.1797, found: 457.1805

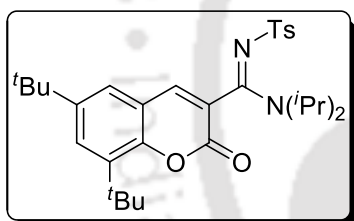


(Z)-*N,N*-Diisopropyl-6-methyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide (12n): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.41$; yellow solid; yield 78% (172 mg); mp 295-296 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.70 (s, 1H), 7.66 (d, $J = 8.4$ Hz, 2H), 7.40 (d, $J = 8.4$ Hz, 1H), 7.33 (s, 1H), 7.24 (d, $J = 8.4$ Hz, 1H), 7.17 (d, $J = 7.6$ Hz, 2H), 3.88-3.83 (m, 1H), 3.70-3.64 (m, 1H), 2.42 (s, 3H), 2.37 (s, 3H), 1.59 (d, $J = 7.2$ Hz, 3H), 1.43 (d, $J = 6.8$ Hz, 3H), 1.26 (d, $J = 6.8$ Hz, 3H), 1.14 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.65, 157.61, 152.3, 141.9, 141.7, 140.8, 134.8, 133.9, 129.1, 128.8, 126.5, 122.9, 117.9, 116.7, 52.7, 48.5, 21.6, 20.8, 20.6, 20.2, 20.0, 19.6; FT-IR (KBr) 3080, 3025, 2982, 2965, 2924, 1718, 1627, 1580, 1542, 1492, 1447, 1373, 1269, 1211, 1175, 1135, 1084, 1061, 1018, 948, 906, 843, 810, 772, 685, 592, 570, 548 cm^{-1} ; HRMS (ESI): m/z calcd for $\text{C}_{24}\text{H}_{29}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 441.1848, found: 441.1864.



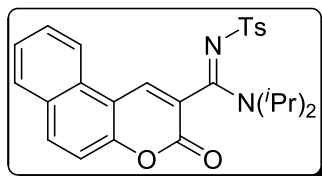
(Z)-8-(*tert*-Butyl)-6-iodo-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-

2*H*-chromene-3-carboximidamide (12p): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.61$; yellow solid; yield 76% (231 mg); mp 244-245 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.80 (s, 1H), 7.73 (s, 1H), 7.65 (s, 1H), 7.62 (d, $J = 8.0$ Hz, 2H), 7.16 (d, $J = 8.4$ Hz, 2H), 3.84-3.80 (m, 1H), 3.71-3.68 (m, 1H), 2.35 (s, 3H), 1.60 (d, $J = 6.8$ Hz, 3H), 1.49-1.46 (m, 12H), 1.28 (d, $J = 6.0$ Hz, 3H), 1.15 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) 157.1, 156.1, 152.6, 142.0, 141.5, 140.7, 140.6, 139.2, 135.8, 129.2, 126.5, 122.6, 120.3, 88.2, 52.9, 48.6, 35.3, 29.9, 21.6, 20.7, 20.2, 20.1, 19.6; FT-IR (KBr) 2966, 2924, 1737, 1630, 1541, 1479, 1369, 1282, 1211, 1147, 1086, 1017, 970, 904, 828, 765, 748, 674, 664, 592, 551 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{27}\text{H}_{34}\text{IN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 609.1284, found: 609.1288.



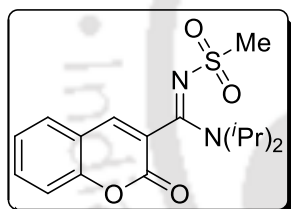
(Z)-6,8-*tert*-Butyl-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-

chromene-3-carboximidamide (12q): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.40$; colorless solid; yield 80% (215 mg); mp 242-243 °C; ^1H NMR (400 MHz, CDCl_3) δ 7.76 (s, 1H), 7.62 (d, $J = 8.0$ Hz, 3H), 7.36 (s, 1H), 7.15 (d, $J = 8.0$ Hz, 2H), 3.88-3.85 (m, 1H), 3.71-3.67 (m, 1H), 2.34 (s, 3H), 1.61 (d, $J = 6.4$ Hz, 3H), 1.49-1.47 (m, 12H), 1.36 (s, 9H), 1.26 (d, $J = 6.4$ Hz, 3H), 1.14 (d, $J = 6.0$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) 158.0, 157.0, 151.0, 147.3, 143.7, 141.9, 140.9, 137.6, 129.2, 128.4, 126.6, 123.7, 121.3, 117.9, 52.7, 48.6, 35.3, 34.9, 31.5, 30.1, 21.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 2961, 2871, 1731, 1629, 1586, 1543, 1479, 1451, 1371, 1271, 1018, 953, 903, 842, 810, 767, 689, 594, 552 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{31}\text{H}_{43}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 539.2944, found: 539.2958.



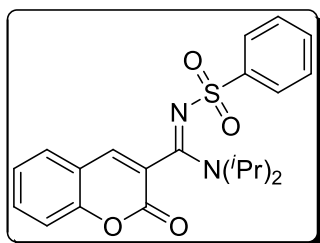
(Z)-*N,N*-Diisopropyl-3-oxo-*N'*-tosyl-3*H*-benzo[*f*]chromene-

2-carboximidamide (12r): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.46$; colorless solid; yield 60% (143 mg); mp 290-291 °C. ^1H NMR (400 MHz, CDCl_3) δ 8.44 (s, 1H), 8.20 (d, $J = 8.4$ Hz, 1H), 8.05 (d, $J = 9.2$ Hz, 2H), 7.93 (d, $J = 8.0$ Hz, 1H), 7.68 (t, $J = 8.0$ Hz, 1H), 7.64 (d, $J = 8.0$ Hz, 2H), 7.58 (t, $J = 8.0$ Hz, 1H), 7.46 (d, $J = 8.8$ Hz, 1H), 7.12 (d, $J = 8.0$ Hz, 2H), 3.94-3.91 (m, 1H), 3.74-3.71 (m, 1H), 2.35 (s, 3H), 1.66 (d, $J = 6.8$ Hz, 3H), 1.50 (d, $J = 6.8$ Hz, 3H), 1.30 (d, $J = 6.8$ Hz, 3H), 1.16 (d, $J = 6.8$ Hz, 3H). $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.9, 157.5, 154.3, 142.0, 140.9, 137.6, 134.4, 130.5, 129.4, 129.2, 128.7, 126.5, 122.0, 121.9, 116.9, 112.6, 52.8, 48.6, 21.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3058, 2991, 2934, 1716, 1573, 1539, 1477, 1440, 1367, 1279, 1213, 1139, 1087, 1015, 955, 901, 849, 817, 669, 552 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{27}\text{H}_{29}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 477.1848, found: 477.1860

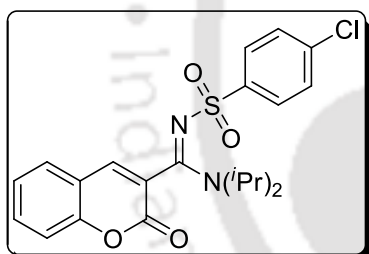


(Z)-*N,N*-Diisopropyl-*N'*-(methylsulfonyl)-2-oxo-2*H*-

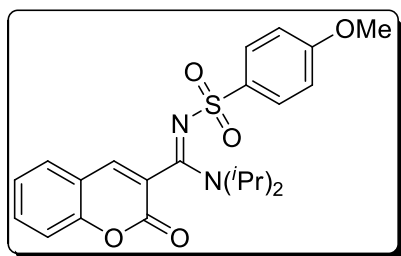
chromene-3-carboximidamide (12s): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.25$; colorless solid; yield 72% (126 mg); mp 249-250 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.75 (s, 1H), 7.58-7.53 (m, 2H), 7.36 (d, $J = 8.4$ Hz, 1H), 7.30 (t, $J = 8.0$ Hz, 1H), 3.92-3.89 (m, 1H), 3.75-3.72 (m, 1H), 2.96 (s, 3H), 1.61 (d, $J = 6.4$ Hz, 6H), 1.30 (d, $J = 6.4$ Hz, 3H), 1.17 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.8, 157.7, 154.2, 141.4, 132.8, 129.0, 125.0, 123.4, 118.1, 117.1, 52.6, 48.3, 43.1, 20.6, 20.2, 20.1, 19.8; FT-IR (KBr) 2924, 2853, 1719, 1608, 1541, 1449, 1376, 1275, 1214, 1112, 1061, 1018, 961, 908, 816, 756, 726, 518 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{17}\text{H}_{23}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 351.1379, found: 351.1376.



(Z)-N,N-Diisopropyl-2-oxo-N'-(phenylsulfonyl)-2H-chromene-3-carboximidamide (12t): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.36$; colorless solid; yield 80% (165 mg); mp 259-260 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.77 (d, $J = 6.8$ Hz, 2H), 7.73 (s, 1H), 7.59-7.53 (m, 2H), 7.44-7.42 (m, 1H), 7.38-7.30 (m, 4H), 3.88-3.85 (m, 1H), 3.69-3.66 (m, 1H), 1.58 (d, $J = 6.8$ Hz, 3H), 1.41 (d, $J = 6.8$ Hz, 3H), 1.26 (d, $J = 6.0$ Hz, 3H), 1.14 (d, $J = 6.0$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.5, 157.4, 154.2, 143.6, 141.6, 132.9, 131.6, 129.0, 128.6, 126.5, 125.1, 123.3, 118.2, 117.1, 52.9, 48.6, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3036, 2969, 2934, 1720, 1628, 1608, 1545, 1482, 1444, 1366, 1278, 1262, 1143, 1088, 1013, 901, 802, 755, 690, 612, 547 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{22}\text{H}_{25}\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 413.1535, found: 413.1535.

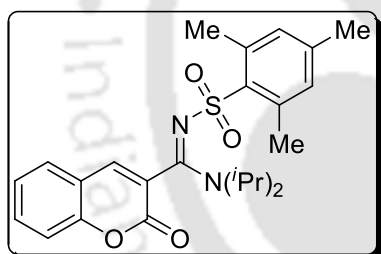


(Z)-N'-(4-Chlorophenyl)sulfonyl-N,N-diisopropyl-2-oxo-2H-chromene-3-carboximidamide (12u): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.40$; colorless solid; yield 82% (183 mg); mp 293-294 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.76 (s, 1H), 7.75 (d, $J = 8.8$ Hz, 2H), 7.62-7.55 (m, 2H), 7.37-7.32 (m, 4H), 3.92-3.89 (m, 1H), 3.71-3.68 (m, 1H), 1.57 (d, $J = 6.4$ Hz, 3H), 1.39 (d, $J = 7.2$ Hz, 3H), 1.28 (d, $J = 6.4$ Hz, 3H), 1.17 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.6, 157.5, 154.2, 142.2, 141.5, 137.8, 133.1, 129.0, 128.8, 128.0, 125.1, 123.4, 118.1, 117.2, 53.0, 48.7, 20.6, 20.1, 19.8; FT-IR (KBr) 3033, 2980, 2928, 1714, 1627, 1609, 1550, 1483, 1444, 1368, 1295, 1271, 1213, 1144, 1088, 1012, 903, 819, 804, 765, 747, 655, 623, 550 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{22}\text{H}_{24}\text{ClN}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 447.1145, found: 447.1149.



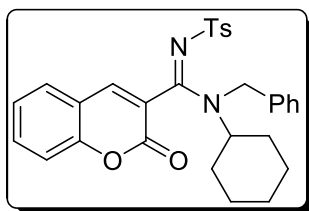
(*Z*)-*N,N*-Diisopropyl-*N'*-((4-methoxyphenyl)sulfonyl)-

2-oxo-2*H*-chromene-3-carboximidamide (12v): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.70$; colorless solid; yield 79% (175 mg); mp 259-260 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.73 (s, 1H), 7.71 (d, $J = 8.4$ Hz, 2H), 7.61-7.54 (m, 2H), 7.35-7.31 (m, 2H), 6.86 (d, $J = 8.8$ Hz, 2H), 3.91-3.86 (m, 1H), 3.82 (s, 1H) 3.72-3.67 (m, 1H), 1.59 (d, $J = 6.8$ Hz, 3H), 1.44 (d, $J = 6.8$ Hz, 3H), 1.27 (d, $J = 6.8$ Hz, 3H), 1.15 (d, $J = 6.4$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 162.0, 157.5, 157.3, 154.2, 141.5, 135.8, 132.9, 129.0, 128.5, 125.0, 123.3, 118.2, 117.1, 113.7, 55.6, 52.7, 48.5, 20.7, 20.2, 20.1, 19.7; FT-IR (KBr) 3034, 2975, 2931, 2839, 1718, 1627, 1609, 1597, 1548, 1496, 1445, 1371, 1353, 1314, 1142, 1128, 1087, 1060, 1014, 901, 809, 790, 758, 677, 596, 556 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{27}\text{N}_2\text{O}_4\text{S}$ [$\text{M}+\text{H}$]: 443.1641, found: 443.1641.



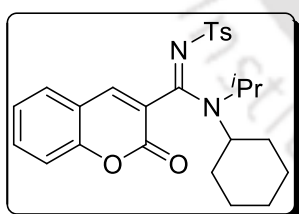
(*Z*)-*N,N*-Diisopropyl-*N'*-(mesitylsulfonyl)-2-oxo-2*H*-

chromene-3-carboximidamide (12w): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.43$; yellow solid; yield 68% (154 mg); mp 289-290 °C. ^1H NMR (400 MHz, CDCl_3) δ 7.73 (s, 1H), 7.57 (t, $J = 7.6$ Hz, 1H), 7.51 (d, $J = 7.6$ Hz, 1H), 7.32-7.25 (m, 2H), 6.74 (s, 2H), 3.82-3.79 (m, 1H), 3.70-3.66 (m, 1H), 2.48 (s, 6H), 2.24 (s, 3H), 1.60 (d, $J = 6.8$ Hz, 3H), 1.47 (d, $J = 7.2$ Hz, 3H), 1.27 (d, $J = 6.4$ Hz, 3H), 1.14 (d, $J = 6.8$ Hz, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (100 MHz, CDCl_3) δ 157.2, 157.1, 154.1, 140.9, 140.8, 138.3, 137.7, 132.8, 131.3, 129.0, 124.9, 123.3, 118.2, 116.9, 52.7, 48.4, 22.8, 21.0, 20.7, 20.4, 20.1, 19.9; FT-IR (KBr) 3037, 2970, 2933, 1721, 1626, 1607, 1540, 1540, 1481, 1445, 1364, 1350, 1281, 1212, 1129, 1061, 1008, 971, 902, 854, 797, 758, 673, 596, 522 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{25}\text{H}_{31}\text{N}_2\text{O}_4\text{S}$ [$\text{M}+\text{H}$] $^+$: 455.2005, found: 455.2005.



(Z)-*N*-Benzyl-*N*-cyclohexyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-

carboximidamide (12x): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.35$; colorless solid; yield 9% (22 mg, 3:1 isomers); mp 205-206 °C. ^1H NMR (600 MHz, CDCl_3) δ 7.88 (s, 1H), 7.64 (d, $J = 8.4$ Hz, 0.5H), 7.64-7.60 (m, 2H), 7.52-7.50 (m, 0.6H), 7.42 (d, $J = 8.4$ Hz, 1H), 7.36 (t, $J = 7.2$ Hz, 1H), 7.33-7.31 (m, 4H), 7.29-7.26 (m, 3H), 7.25-7.19 (m, 2H), 7.17 (d, $J = 7.2$ Hz, 1H), 7.10 (d, $J = 7.2$ Hz, 0.6H), 7.00 (d, $J = 7.8$ Hz, 2H), 5.02 (d, $J = 15.6$ Hz, 1H), 4.67 (s, 0.3H), 4.61 (d, $J = 15.6$ Hz, 1H), 4.56 (d, $J = 17.4$ Hz, 0.3H), 4.40 (d, $J = 16.8$ Hz, 0.3H), 3.58-3.54 (m, 1H), 2.38 (s, 0.9H), 2.32 (s, 3H), 2.02 (d, $J = 12.6$ Hz, 1H), 1.76-1.71 (m, 0.6H), 1.63-1.59 (m, 3.6H), 1.55-1.53 (m, 2H), 1.39-1.33 (m, 3H), 1.14-1.07 (m, 2.4H), 1.00 (t, $J = 12.6$ Hz, 1.3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 160.5, 159.4, 157.6, 157.4, 154.4, 154.1, 143.7, 142.6, 142.4, 142.0, 140.5, 140.3, 137.7, 136.9, 133.2, 133.0, 129.2, 129.1, 129.0, 128.9, 128.6, 127.8, 126.8, 126.7, 126.6, 126.5, 125.2, 125.0, 122.3, 121.9, 118.1, 117.8, 117.3, 116.9, 60.7, 58.4, 49.2, 47.5, 32.1, 31.8, 31.0, 30.32, 30.3, 25.84, 25.8, 25.62, 25.6, 25.1, 21.7, 21.6; FT-IR (KBr) 3033, 3015, 2958, 2936, 2925, 2852, 1714, 1625, 1609, 1574, 1543, 1452, 1420, 1372, 1282, 1185, 1243, 1088, 1065, 997, 980, 907, 686, 555 cm^{-1} ; HRMS (ESI) m/z calcd for $\text{C}_{30}\text{H}_{31}\text{N}_2\text{O}_4\text{S}$ [M+H]: 515.1999, found: 515.2005.



(Z)-*N*-Cyclohexyl-*N*-isopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-

3-carboximidamide (12y): analytical TLC on silica gel, 2:3 ethyl acetate-hexane $R_f = 0.35$; colorless solid; yield 9 % (20 mg, 1:1 isomers); mp 271-272 °C. ^1H NMR (600 MHz, CDCl_3) δ 7.74 (s, 1H), 7.72 (s, 1H), 7.66 (t, $J = 8.4$ Hz, 4H), 7.61-7.56 (m, 4H), 7.37-7.31 (m, 4H), 7.17-7.16 (m, 4H), 3.90-3.87 (m, 1H), 3.74-3.72 (m, 1H), 3.37-3.33 (m, 1H), 3.18 (s, 1H), 2.88-2.87 (m, 1H), 2.47-2.39 (m, 2H), 2.38 (s, 3H), 2.37 (s, 3H), 2.02 (d, $J = 12.0$ Hz, 1H), 1.85 (d, $J = 12.0$ Hz, 1H), 1.77-1.72 (m, 2H), 1.67 (d, $J = 6.6$ Hz, 3H), 1.41 (d, $J = 7.2$ Hz, 3H), 1.25-1.17 (m, 7H), 1.13 (d, $J = 6.6$ Hz, 3H), 1.10-1.00 (m, 9H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 157.6, 157.52, 157.5, 154.3, 142.1, 141.6,

141.5, 140.83, 140.8, 132.9, 129.21, 129.2, 129.1, 129.07, 126.6, 125.1, 123.4, 123.3, 118.24, 118.2, 117.2, 117.1, 61.7, 57.9, 52.9, 49.8, 30.8, 30.3, 29.9, 29.3, 28.6, 26.7, 26.5, 25.62, 25.6, 25.4, 25.1, 21.7, 20.8, 20.2, 19.7; FT-IR (KBr) 2964, 2931, 2849, 1722, 1607, 1540, 1482, 1445, 1383, 1371, 1272, 1143, 1087, 906, 811, 754, 729, 690, 575, 547 cm⁻¹; HRMS (ESI) *m/z* calcd for C₂₆H₃₁N₂O₄S [M+H]: 467.1999, found: 467.2005.

Crystal Data and Structure Refinement for 11 at 296(2) K

Identification code	GM-349c	
Empirical formula	C ₂₅ H ₃₂ N ₂ O ₅ S	
Formula weight	472.59	
Temperature	296(2) K	
Wavelength	0.71073 Å	
Crystal system	'Monoclinic'	
Space group	'P 21/c'	
	loop_	
	_symmetry_equiv_pos_as_xyz	
	'x, y, z'	
	'-x, y+1/2, -z+1/2'	
	'-x, -y, -z'	
	'x, -y-1/2, z-1/2'	
Unit cell dimensions	<i>a</i> = 8.2829 (12) Å	$\alpha(^{\circ})$ = 90.00
	<i>b</i> = 10.0890 (15) Å	$\beta(^{\circ})$ = 91.266 (8)
	<i>c</i> = 30.145 (4) Å	$\gamma(^{\circ})$ = 90.00
Volume	2518.5 (6) Å ³	
Z	4	
Density	1.246 Mg/m ³	
Absorption coefficient	0.165 mm ⁻¹	
<i>F</i> (000)	1008.0	
Crystal size	0.40 x 0.38 x 0.20 mm	
Theta range for data collection	2.13 to 28.50°	
Index ranges	-10 ≤ <i>h</i> ≤ 10, -12 ≤ <i>k</i> ≤ 13, -39 ≤ <i>l</i> ≤ 31	
Reflections collected	5919	

Independent reflections	4360 [R (int) = 0.0459]
Completeness to theta = 28.50°	99.5%
Absorption correction	Multi-scan
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	5919/ 0 /356
Goodness-of-fit on F^2	1.043
Final R indices [$I > 2\sigma(I)$]	$R1 = 0.0459$, $wR2 = 0.1239$
R indices (all data)	$R1 = 0.0672$, $wR2 = 0.1365$

Crystal Data and Structure Refinement for 12r at 296(2) K.

Identification code	GM-411A
Empirical formula	C ₂₇ H ₂₈ N ₂ O ₄ S
Formula weight	476.58
Temperature	296(2) K
Wavelength	0.71073 Å
Crystal system	Monoclinic
Space group	P 21/n
	loop_
	_symmetry_equiv_pos_as_xyz
	'x, y, z'
	'-x+1/2, y+1/2, -z+1/2'
	'-x, -y, -z'
	'x-1/2, -y-1/2, z-1/2'
Unit cell dimensions	$a = 11.9970(5)$ Å α (°) = 90.00
	$b = 18.4263(7)$ Å β (°) = 108.212(2)
	$c = 12.1195(4)$ Å γ (°) = 90.00
Volume	2544.93 (17) Å ³
Z	4
Density	1.244 Mg/m ³
Absorption coefficient	0.162 mm ⁻¹
$F(000)$	1008.0
Crystal size	0.48 x 0.38 x 0.24 mm

Theta range for data collection	1.08 to 28.33°
Index ranges	-15<=h<=15, -24<=k<=24, -16<=l<=16
Reflections collected	6311
Independent reflections	3603 [R (int) = 0.0506]
Completeness to theta = 28.330°	99.6%
Absorption correction	Multi-scan
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	6311/ 0 /312
Goodness-of-fit on F^2	0.955
Final R indices [$I > 2\sigma(I)$]	$R1 = 0.0506$, $wR2 = 0.1445$
R indices (all data)	$R1 = 0.0972$, $wR2 = 0.1713$

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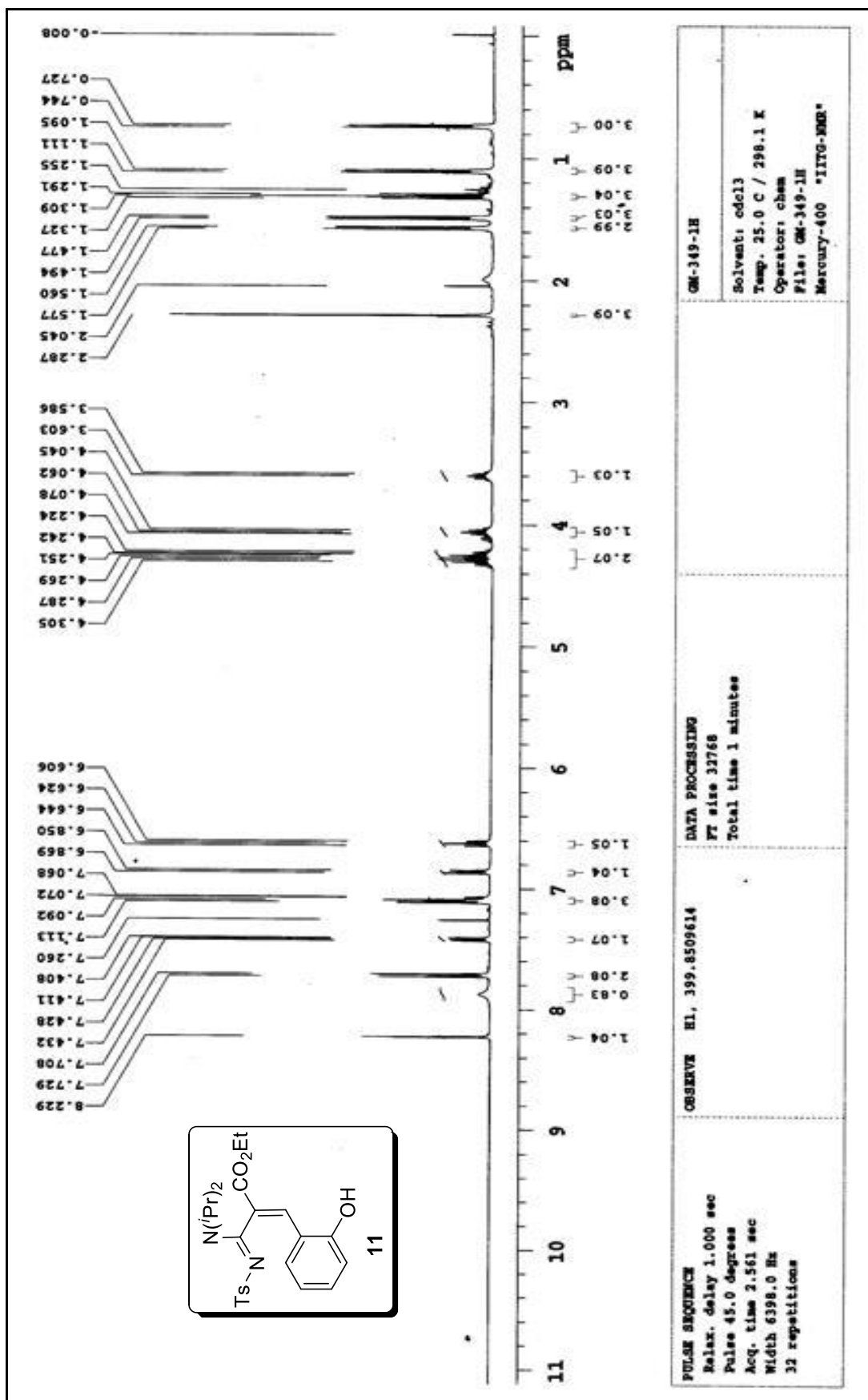
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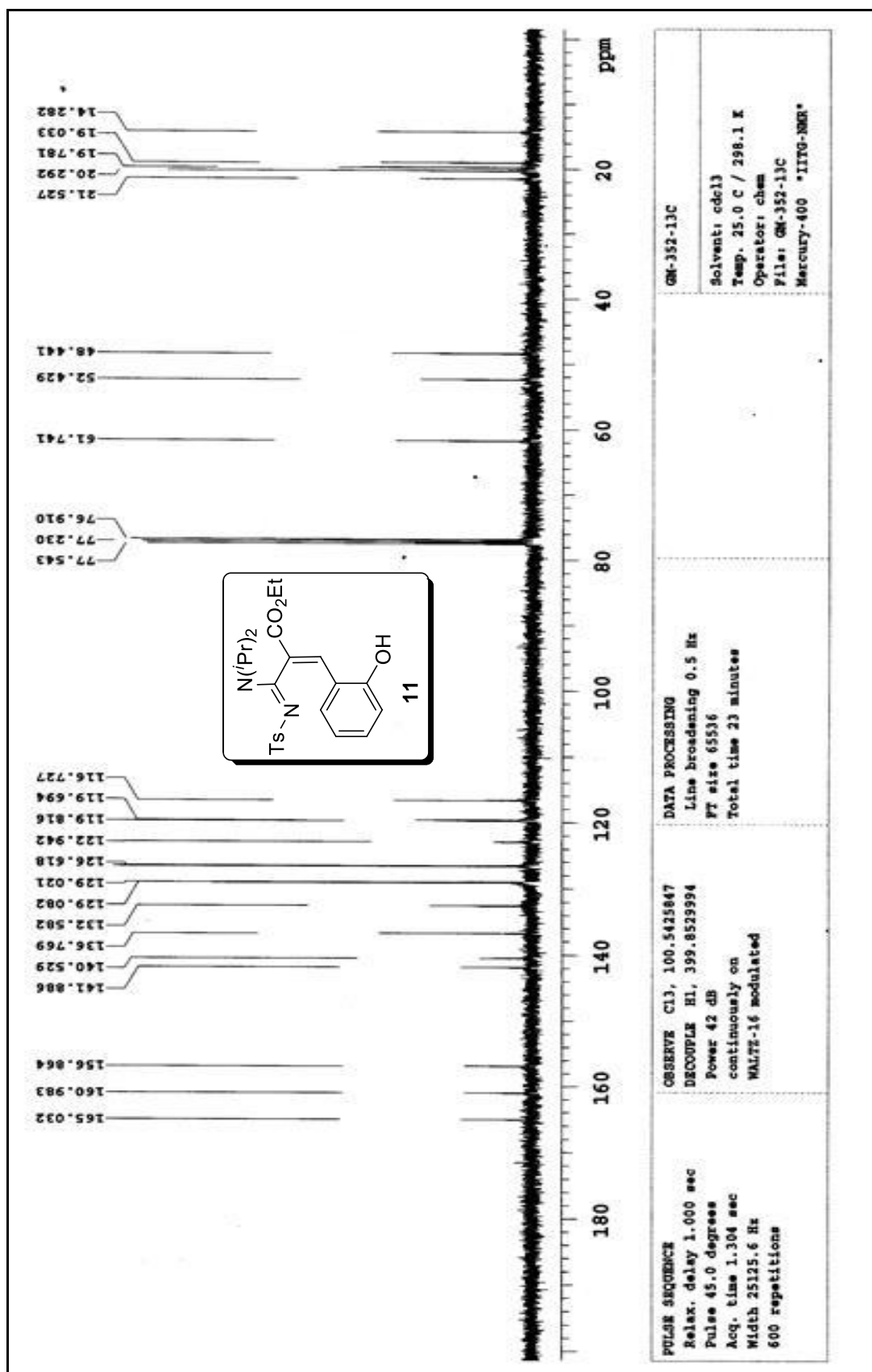
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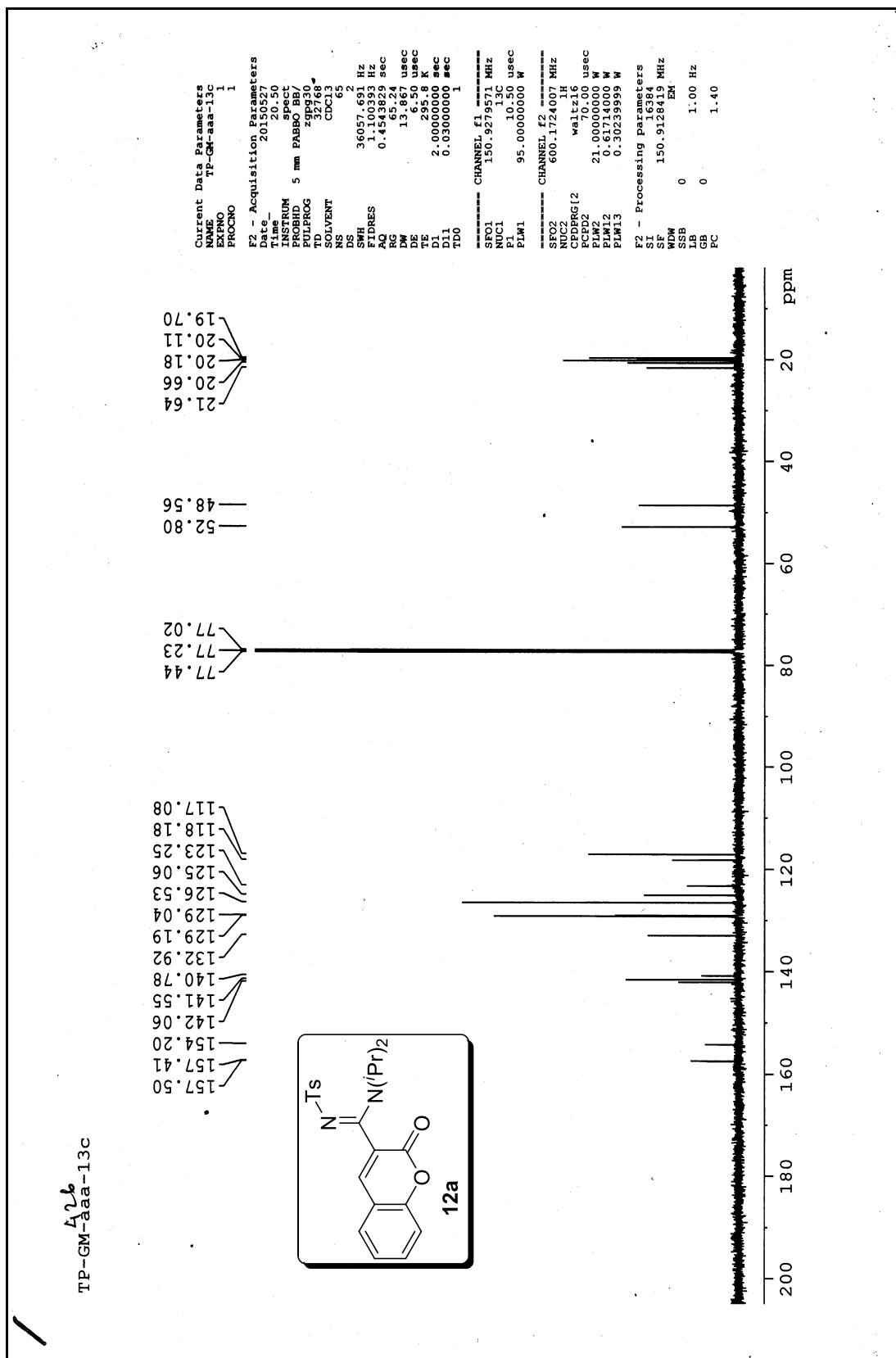
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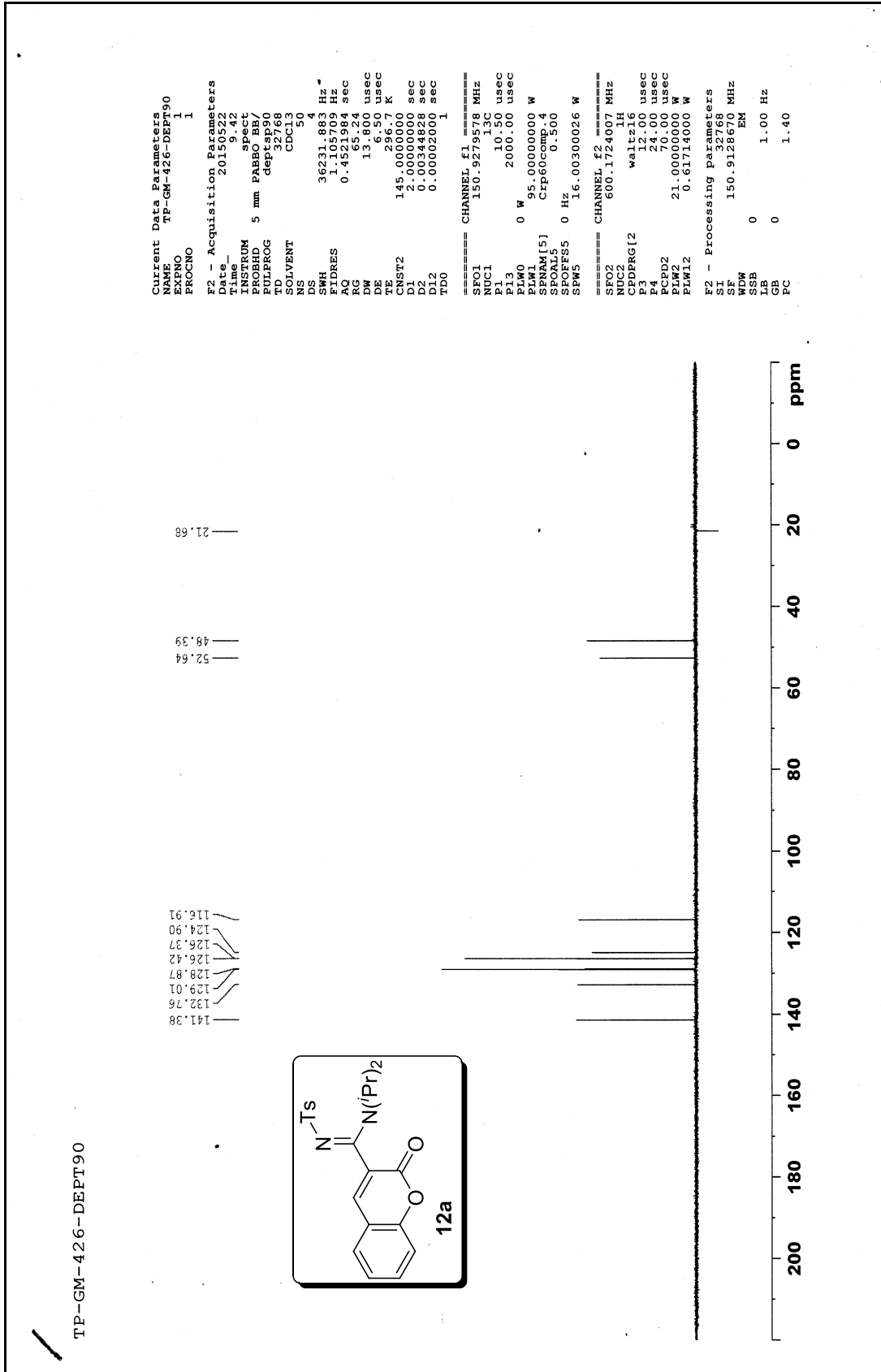
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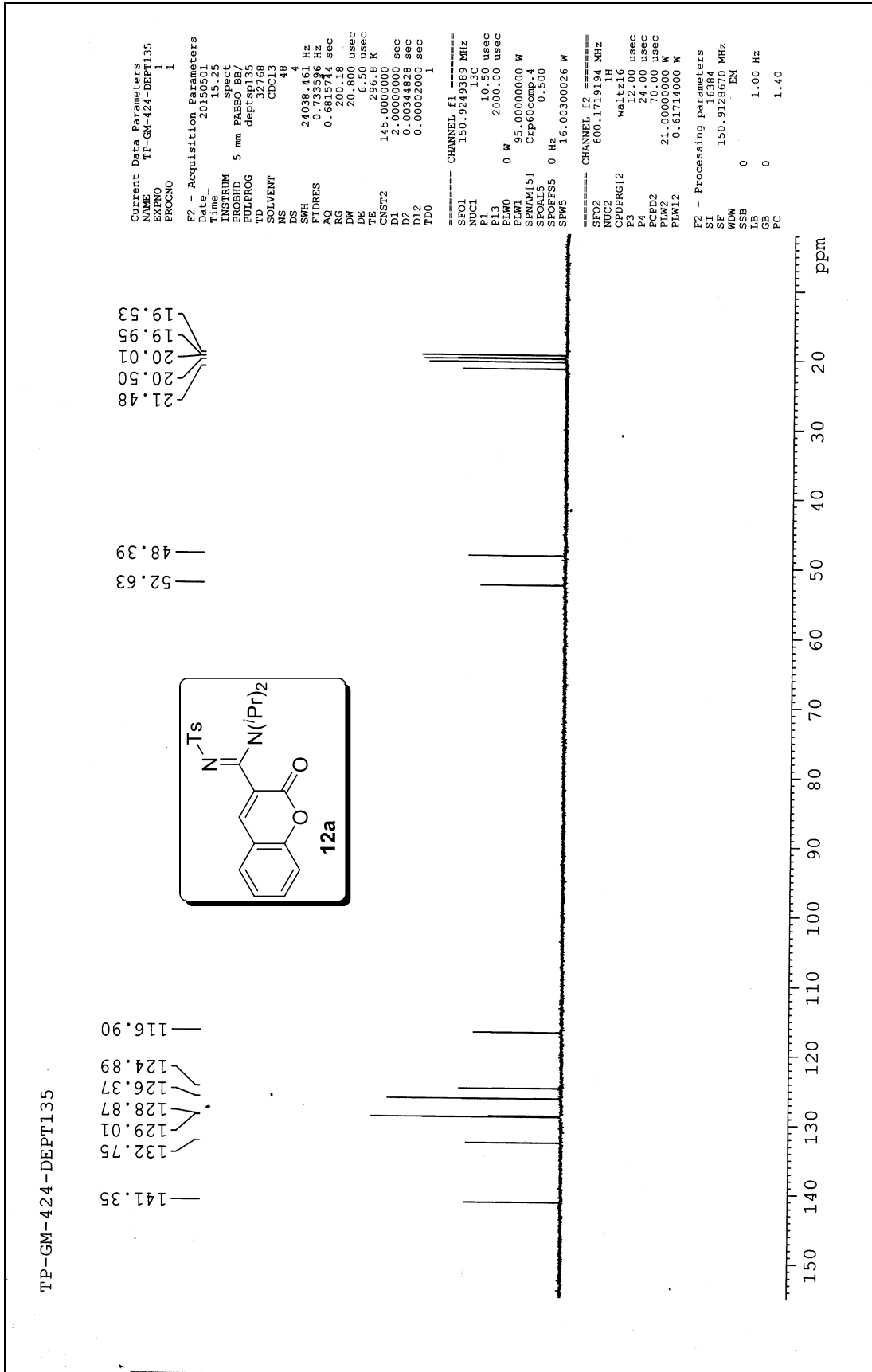
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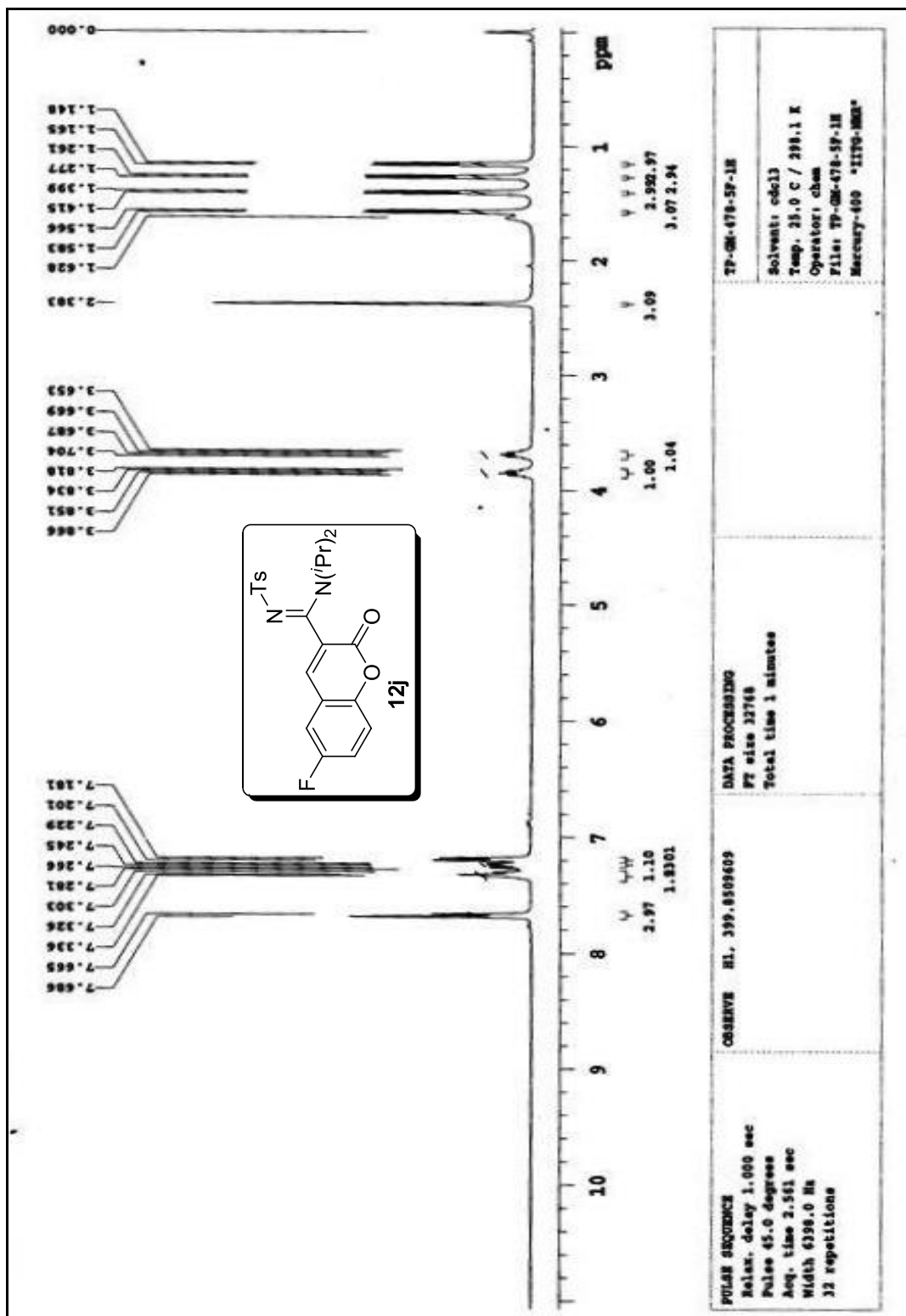


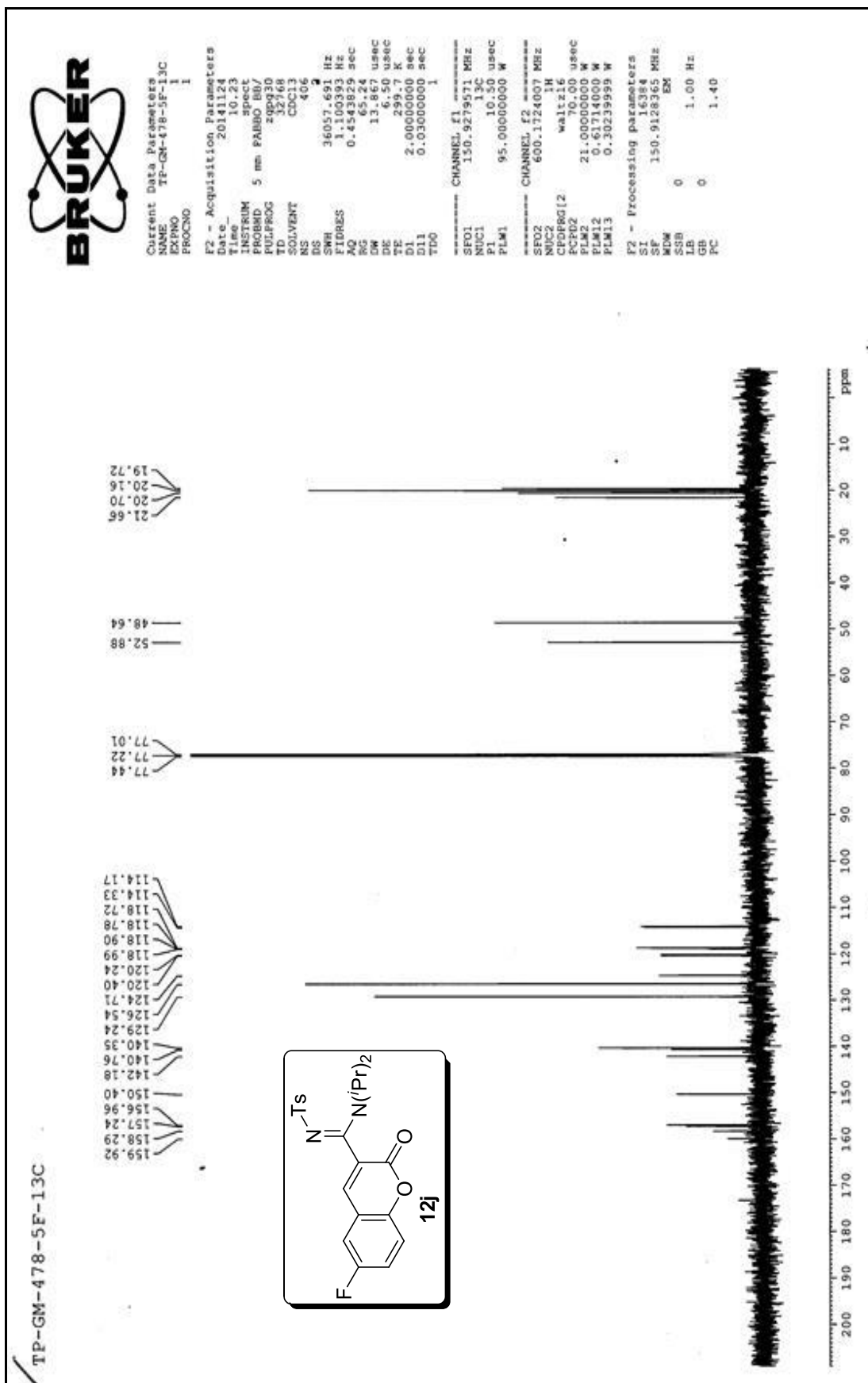


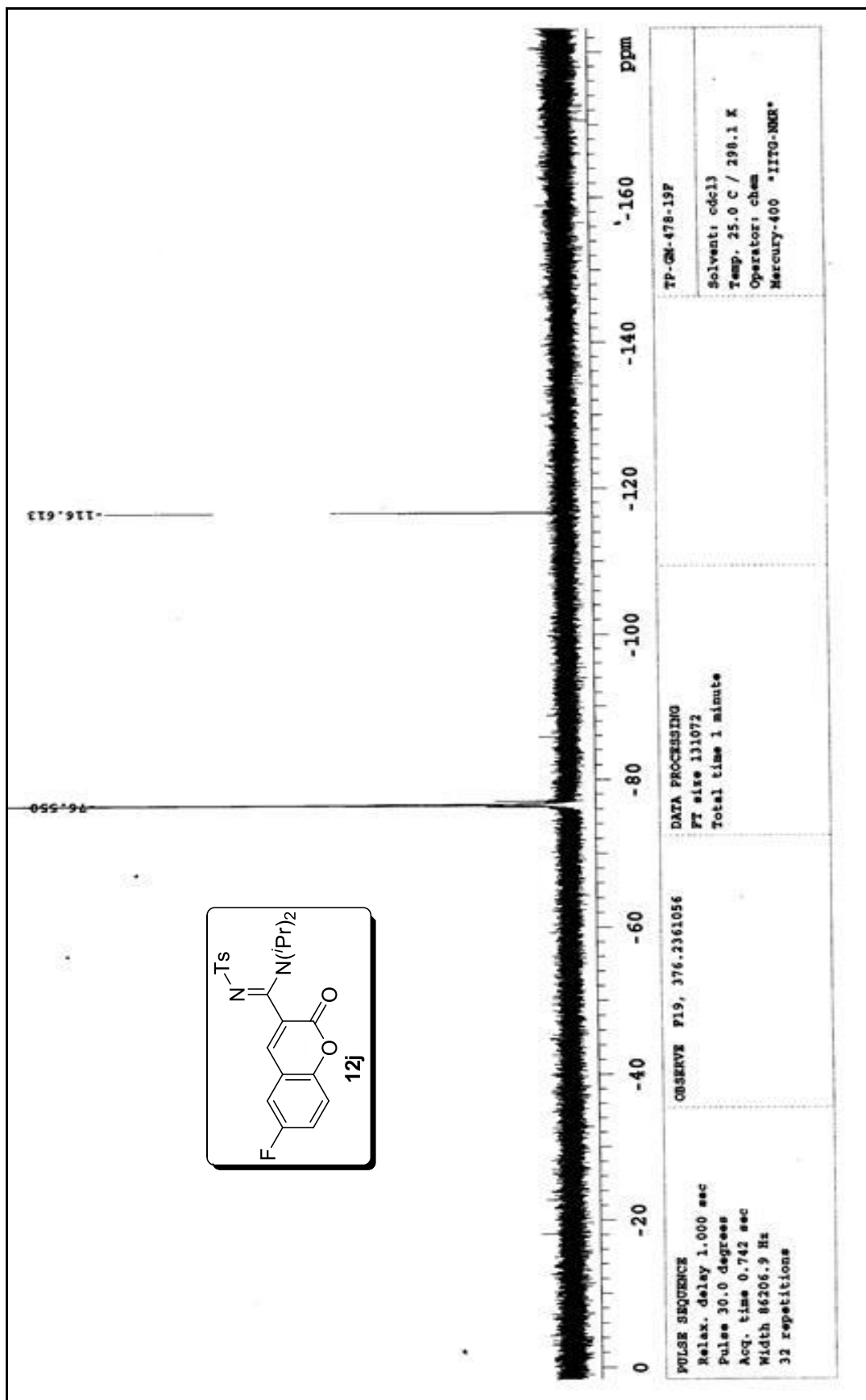


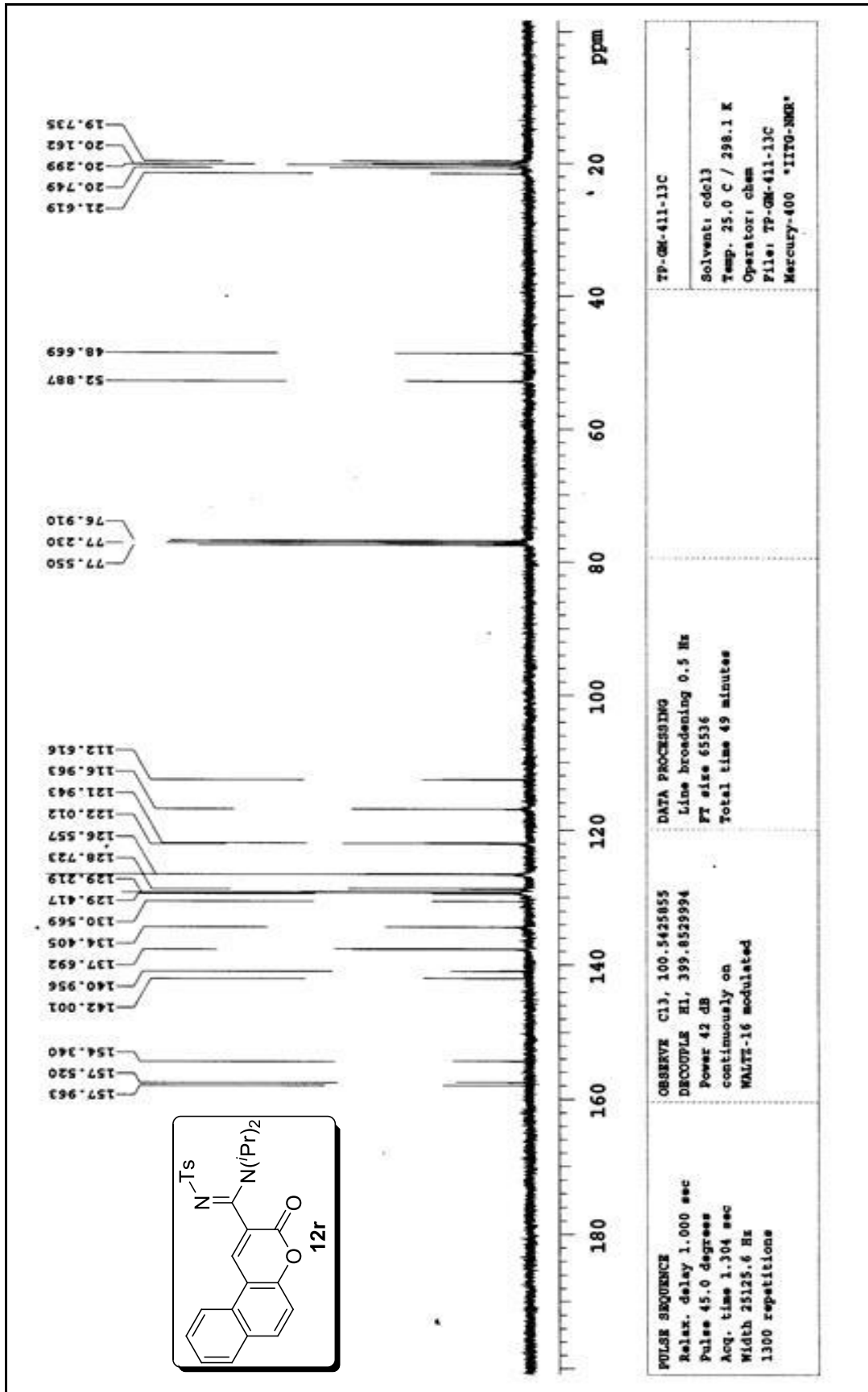


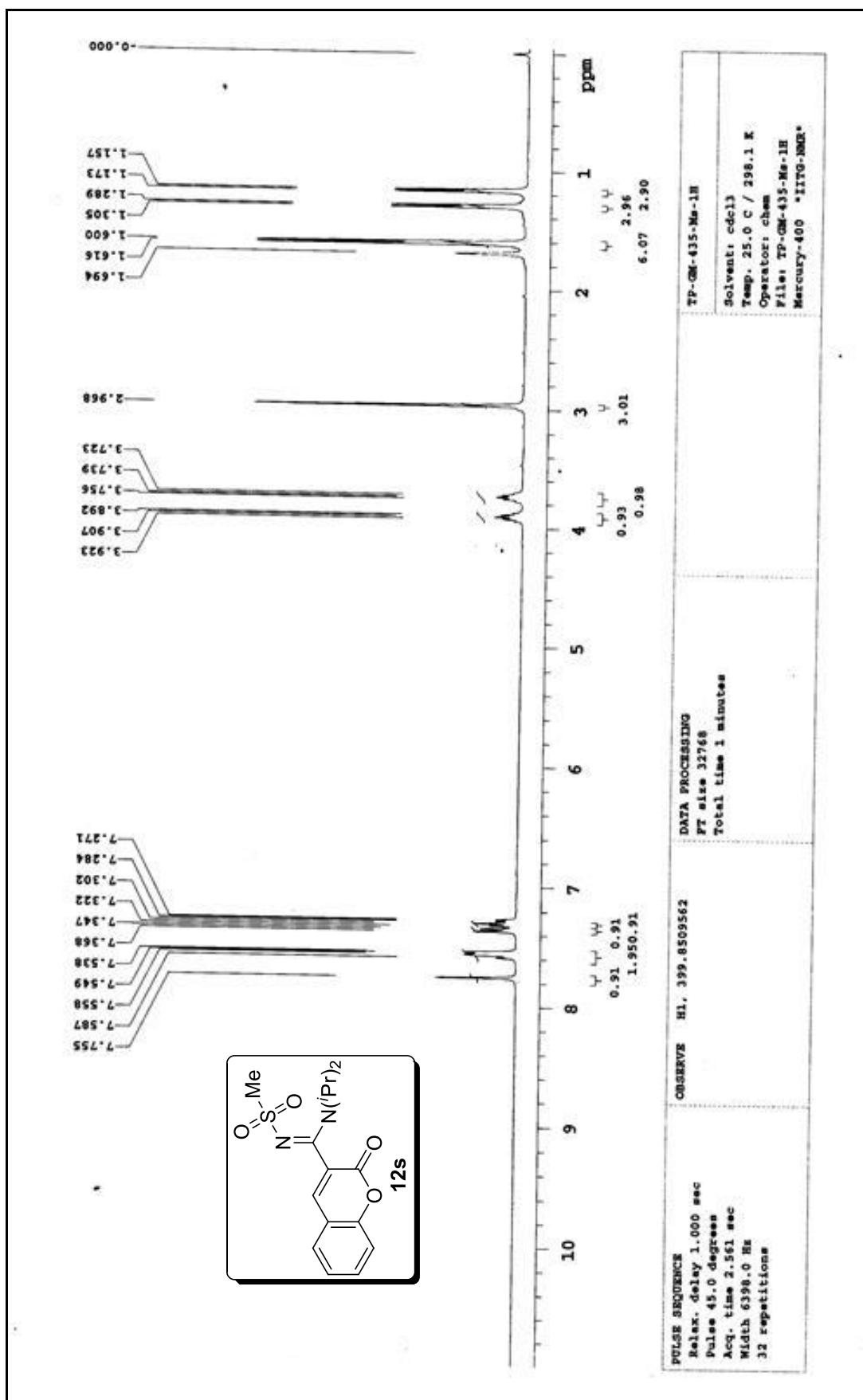


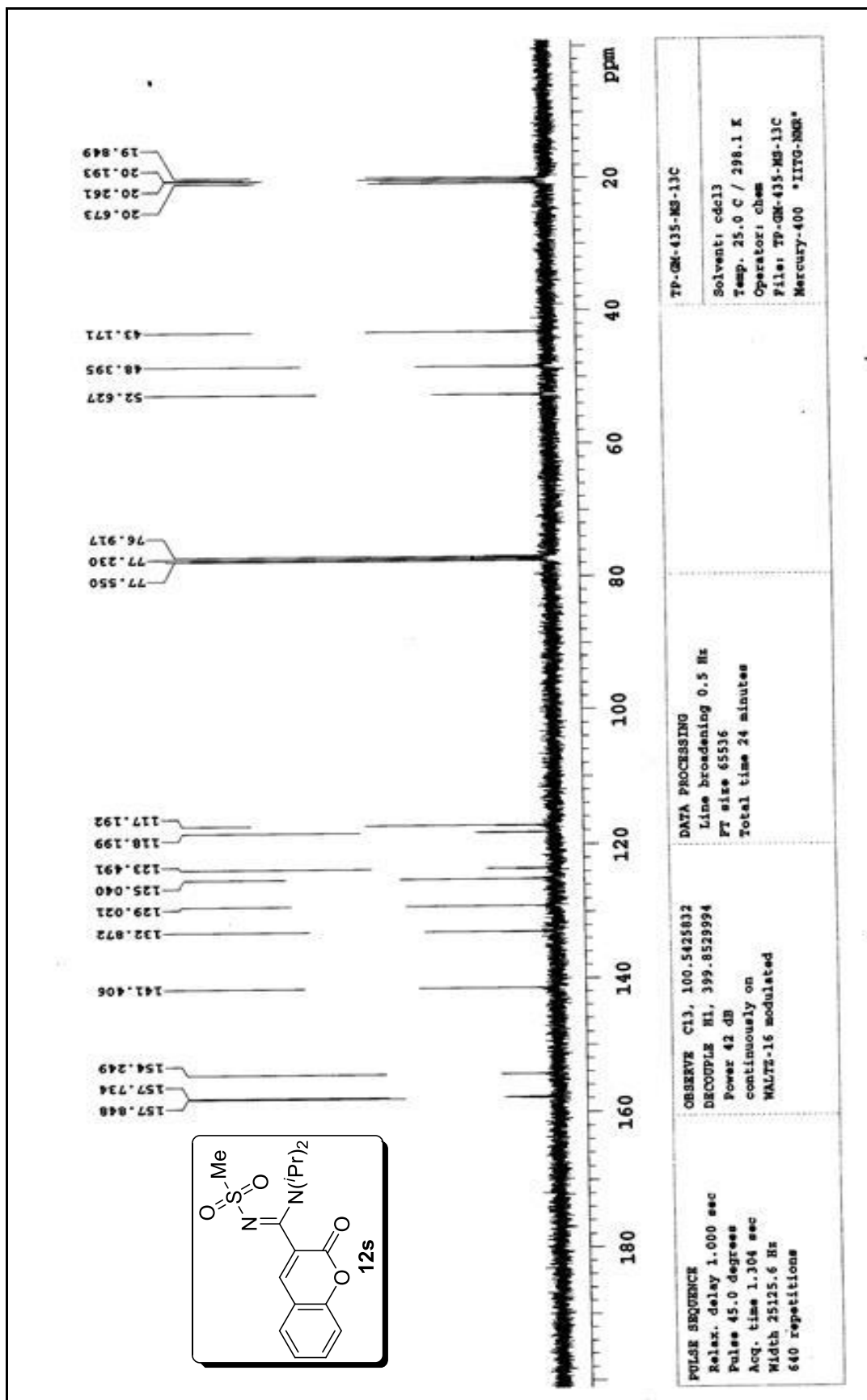




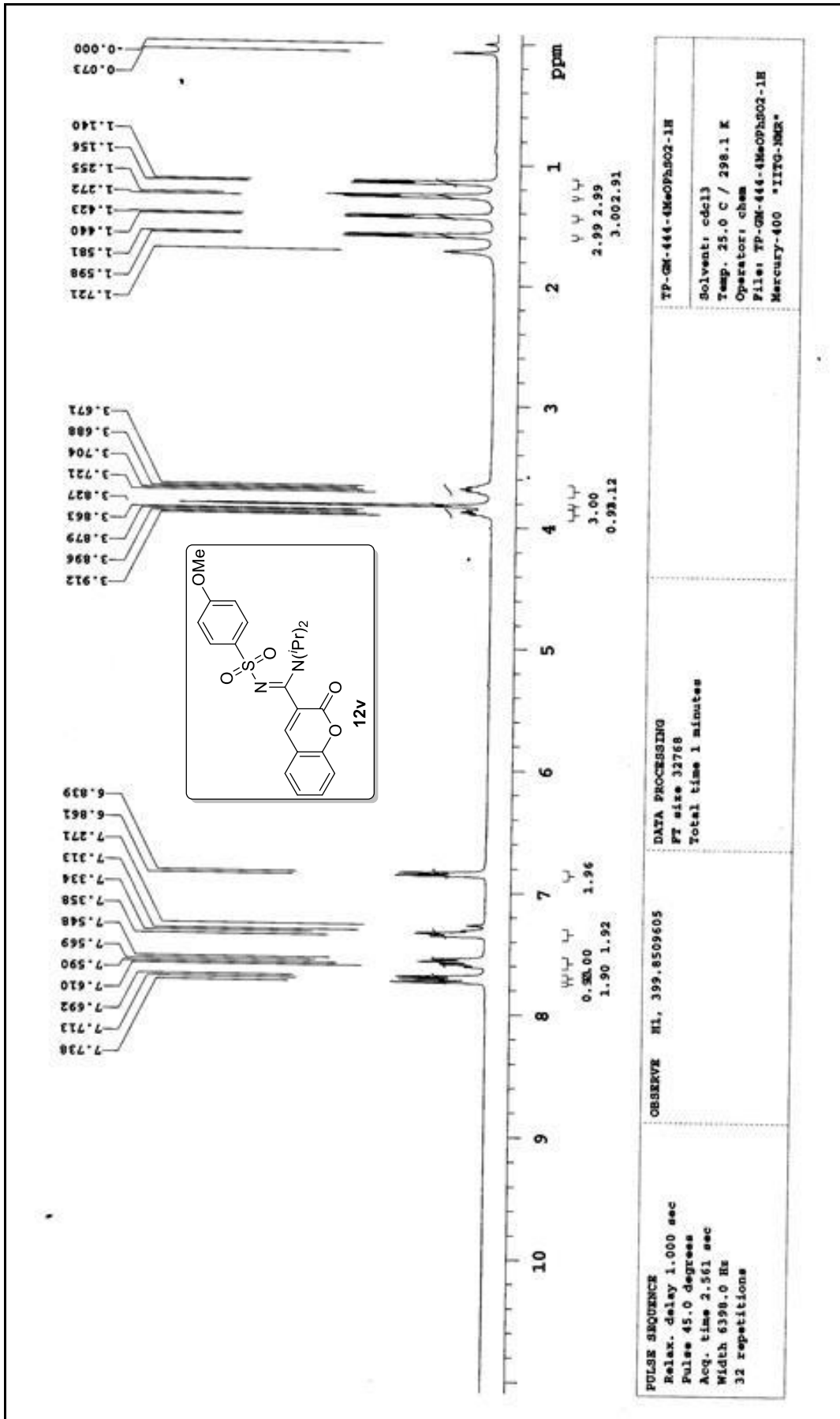


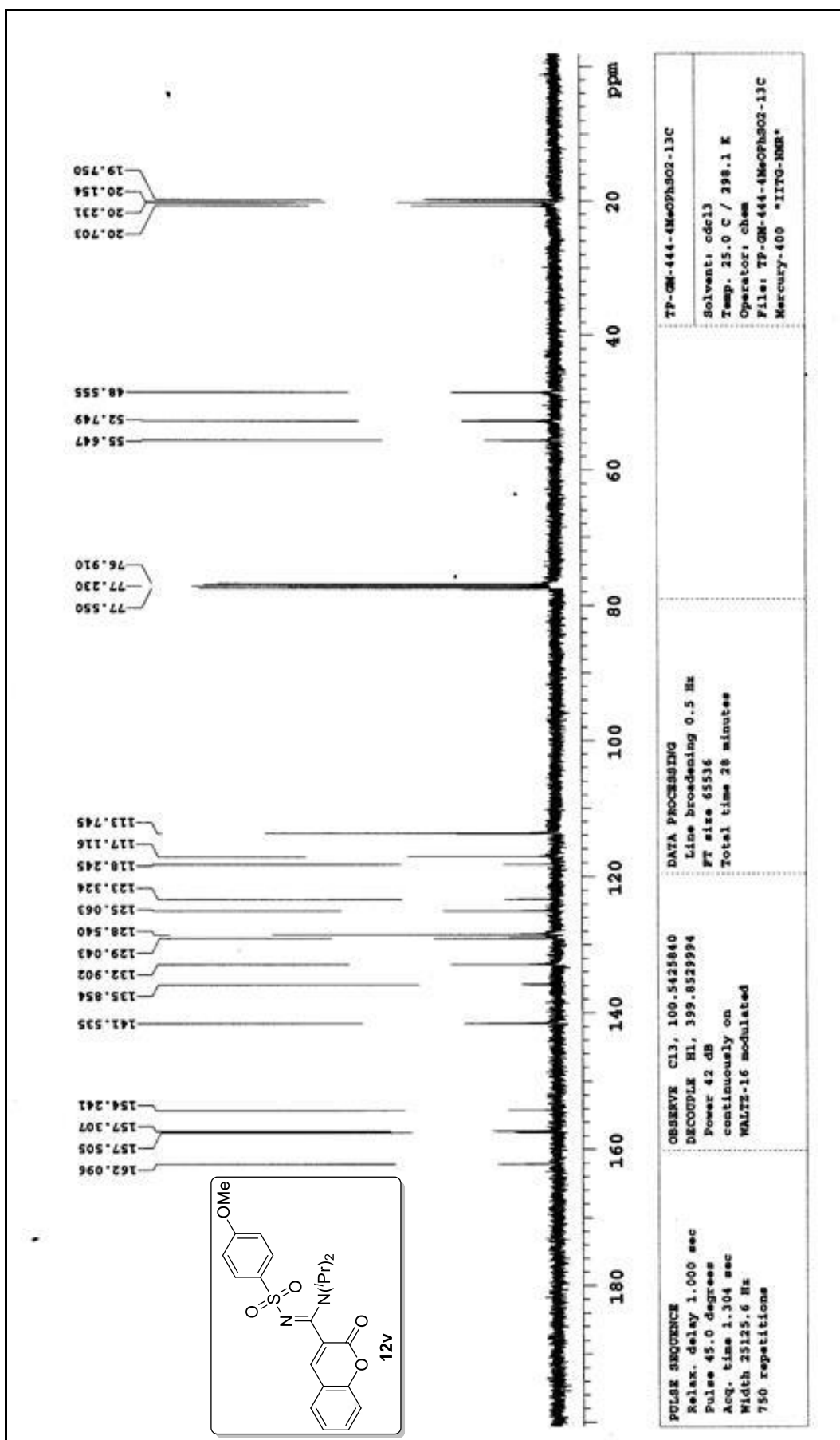


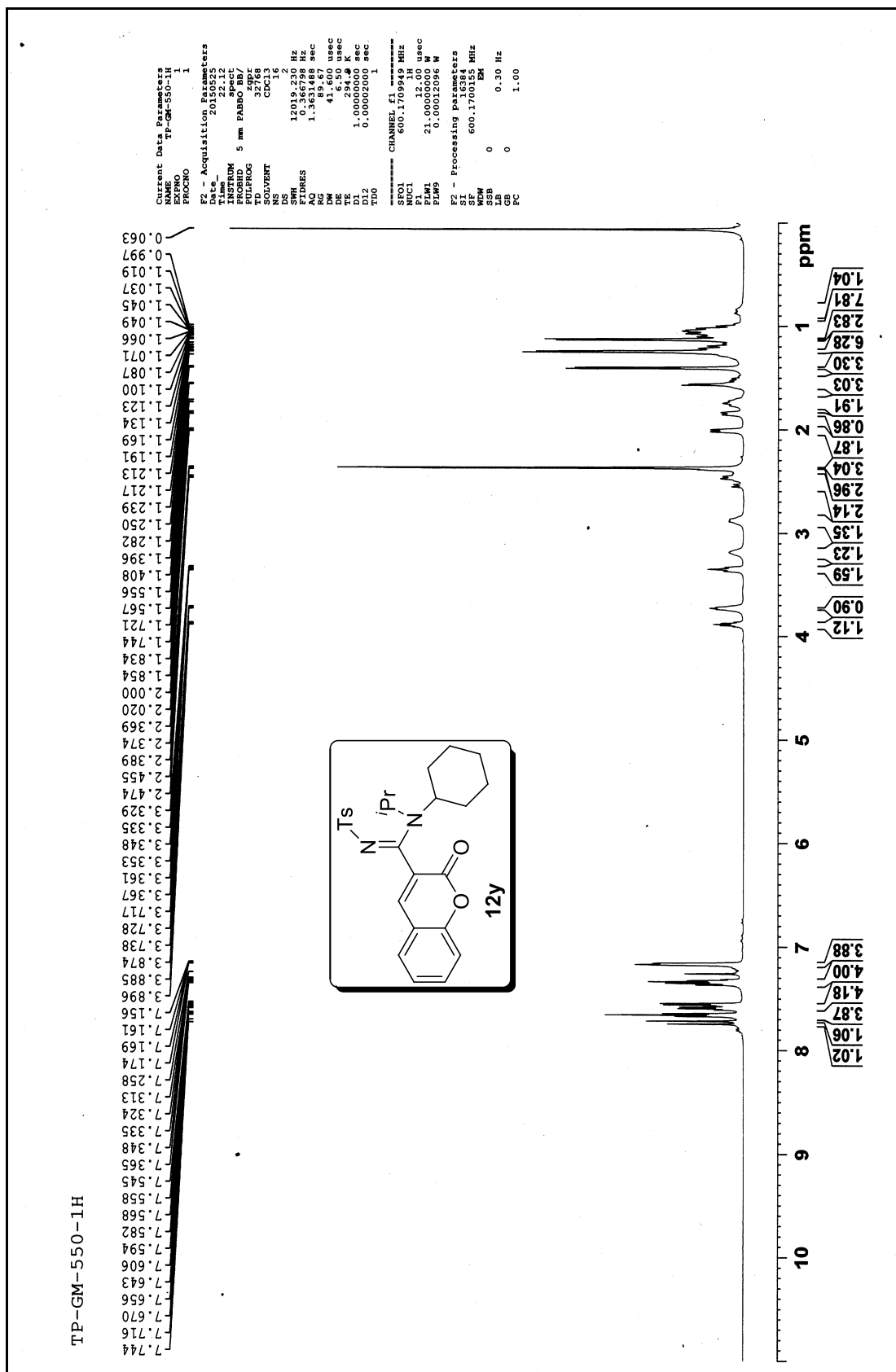


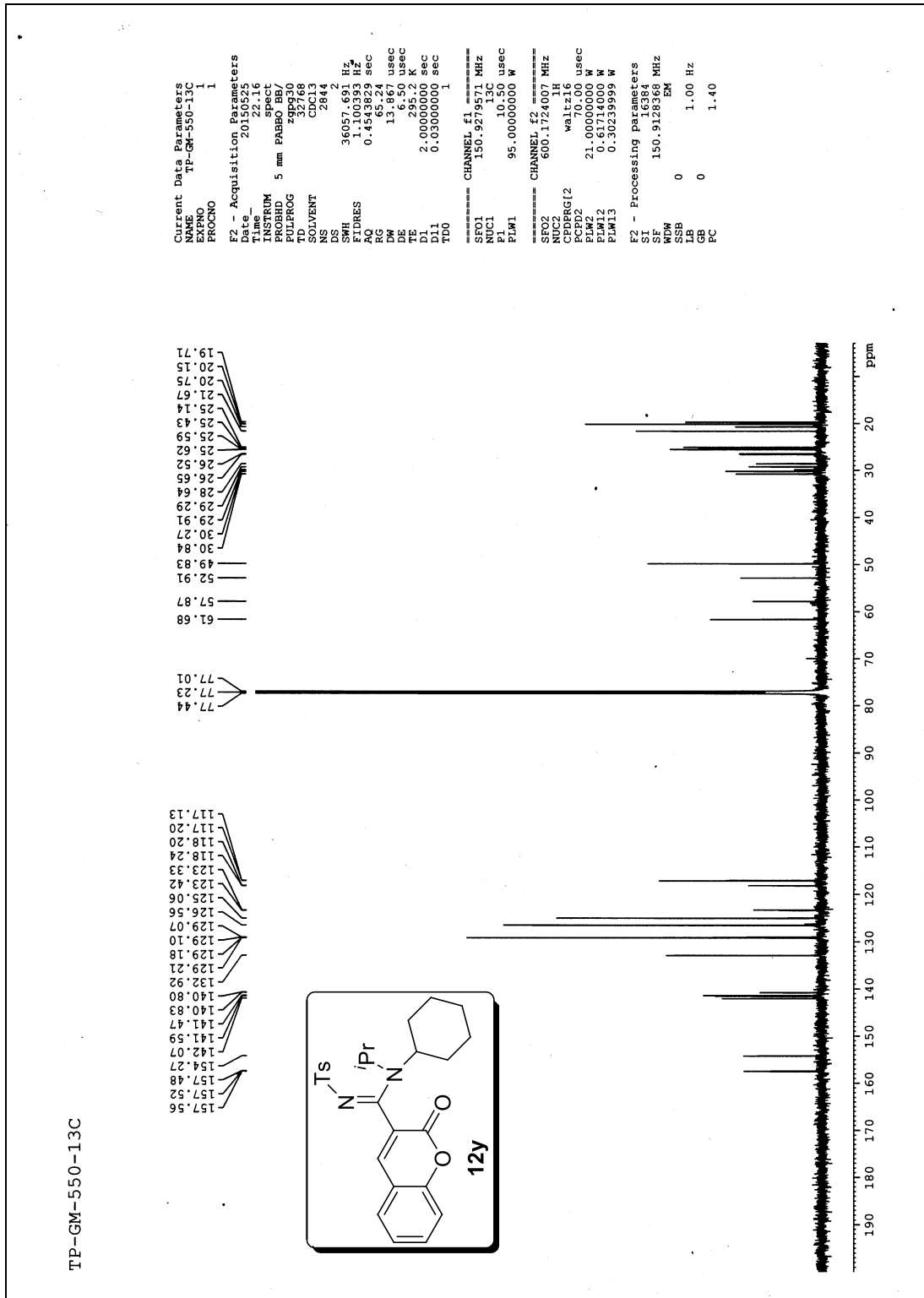


Synthesis of 3N-Sulfonylamidine Coumarins









Study on Interaction of Solvatochromic Fluorescent Probe (Z)-7-(Diethylamino)-N,N-diisopropyl-2-oxo-N'-tosyl-2H-chromene-3-carboximidamide with Bovine Serum Albumin

Serum albumins are the multifunctional abundant proteins in blood plasma of circulatory system.¹ They transport and depot many endogenous and exogenous compounds in the body by hydrophobic, hydrophilic and ionic interactions.^{1,2} Human serum albumin (HSA), bovine serum albumin (BSA), equine serum albumin and rat serum albumin are commonly studied proteins, which are isolated from blood serum of human, calf, horse and rat, respectively. BSA is a readily available and easily extractable large globular heart-shaped water soluble protein (66.4 kDa), which consists of 583 amino acid residues.³ Due to its stability and excellent structural homology with HSA, it is highly preferable for the biological assays.⁴ Tertiary structure of BSA protein comprised of three domains (I, II and III), where each domains is further sub-divided into two sub-domains (IA and IB; IIA and IIB; IIIA and IIIB) stabilized by 17 pairs of disulfide bridges (Figure 1).^{5,6} The primary binding sites (I and II) are positioned in the hydrophobic cavities of subdomains IIA and IIIA, which influence the specific physiological activity of the bioactive molecules on BSA. The binding site I influences strong hydrophobic interactions, is also referred as Sudlow's site I or the warfarin

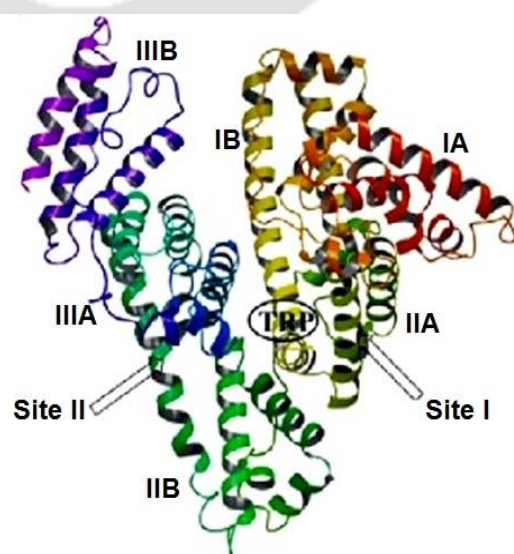


Figure 1. Structure of bovine serum albumin (BSA)

binding site, whereas binding site II influences hydrophobic, hydrogen bonding and electrostatic interactions, is also referred as Sudlow's site II or the binding site of ibuprofen.^{7,8} BSA has two tryptophan moieties namely Trp-134 and Trp-213 residues, which have intrinsic fluorescence. Trp-134 residue located in hydrophilic subdomain IB near the surface of albumin molecule, which is exposed to solvent environment, whereas Trp-213 residue located deeply in the hydrophobic cavity of subdomain IIA.^{2,9}

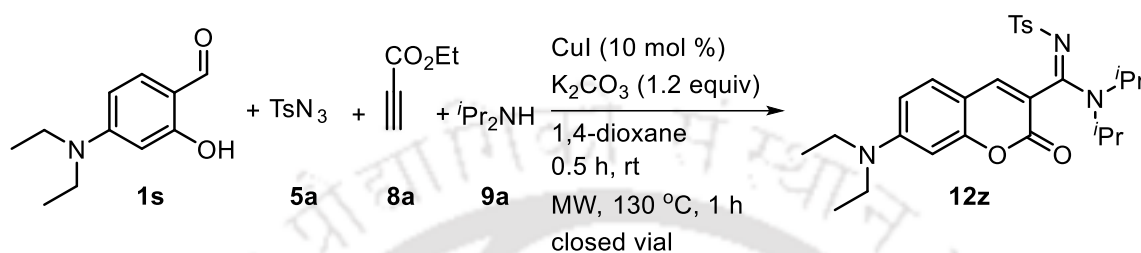
Generally, studies on interaction of small bioactive molecules with BSA provided useful information about the forces involved and the topology of the binding sites.⁷⁻¹¹ Recently, these studies have drawn greater attention and also afforded the deep insights in the understanding of different biological systems as well as protein immobilization. Often, molecular interactions are investigated using versatile fluorescence spectroscopic techniques, which are highly sensitive and relatively easy to use.^{11,12} These methods have received considerable attention among modern chemists in utilizing fluorescent dyes that exhibits unique and varied photophysical properties.¹³ In addition, these techniques can be applied for drug discovery by a wide array of format ranging from high-throughput screening assays.¹⁴ Moreover, the emission properties of the fluorescent dyes are highly sensitive to physical changes in the local environment including solvent polarity, pH, viscosity and biological analytes.¹¹ Specifically, solvatochromic fluorophore possesses an emission property, which is being highly sensitive to the nature of local micro-environments.^{11,15} These fluorophores are widely used in the applications to study the protein structural dynamics that detect protein-binding interactions, which can lead to design and development of drug discovery.¹⁶

4.1 Present Study

We present here the interaction of a new solvatochromic fluorescent probe (*Z*)-7-(diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide with BSA using UV-visible, fluorescent and picosecond time-resolved spectroscopy. In addition, the docking study of probe with BSA has been performed for understanding the nature of protein-binding interactions.

4.1.1 Synthesis of Fluorophore 12z

Initially, the target fluorophore **12z** has been synthesized using microwave-assisted copper(I)-catalyzed reaction from 4-(diethylamino)-2-hydroxybenzaldehyde (**1s**), ethyl propiolate (**8a**), tosyl azide (**5a**) and diisoprylamine (**9a**) (Scheme 1).¹⁷ The probe was further characterized using ¹H & ¹³C NMR, IR and HRMS analyses.



Scheme 1

4.1.2 Photophysical Studies

4.1.2.1 Absorption and Fluorescence Studies

Since the sensitivity of solvent microenvironment with fluorophore **12z** can be recognized by examining absorption and emission spectra in different solvents, the absorption and emission spectra of **12z**, (5×10^{-6} M) were recorded in some of the representative both non-polar and polar solvents of different polarity at room temperature, which are strongly dependent on the solvent polarities. Figure 2 shows the normalized absorption and fluorescence spectra of **12z** in some of the representative solvents having low (e.g., toluene, $\epsilon = 2.38$)¹⁸ to high polarities (e.g., DMSO, $\epsilon = 46.7$).¹⁸ Typical spectroscopic data of probe **12z** such as wavelength of both absorption and fluorescence maxima ($\lambda_{\text{max}}^{\text{ab}}$ and $\lambda_{\text{max}}^{\text{fl}}$ respectively), solvent polarity function (Δf), Stokes shift ($\Delta \bar{\nu}$) and lifetimes (τ) in different solvents are summarized in Table 1. We observed that the solvent polarity impacts a minor influence on the absorptive properties (Figure 2a). The fluorescence emission of fluorophore **12z** in different solvents of varying polarities occurs in the region 439-467 nm, which exhibits gradual shift (28 nm) from blue ($\lambda_{\text{max}}^{\text{fl}} = 439$ nm) in toluene to red in MeOH ($\lambda_{\text{max}}^{\text{fl}} = 467$ nm) (Figure 2b). The sensitivity of the solvent polarity on the fluorophore is due to the charge shift away from the diethylamino group (electron donating group) in excited state towards imidate and carboxylate groups (electron acceptor group).¹⁹ Figure 3 plots the wavenumbers for the absorption fluorescence

maxima ($\bar{\nu}_{\text{abs}}$ and $\bar{\nu}_{\text{fl}}$ respectively) of fluorophores **12z** against the polarity functions, Δf (calculated from eq (1)) of the solvents. It is observed that $\bar{\nu}_{\text{abs}}$ follows linear relationship with Δf for toluene, DMSO, CH₃CN and MeOH, while $\bar{\nu}_{\text{fl}}$ follows linear relationships with Δf for all solvents.

Table 1. Photophysical Parameters of Fluorophore **12z** in Different Solvents

Solvents	Δf	$\lambda_{\text{abs}}^{\text{max}}$ /nm	$\lambda_{\text{em}}^{\text{max}}$ /nm	$\Delta\bar{\nu}$ /cm ⁻¹	Life time (fl.)	
					τ /ns	χ^2
Toluene	0.013	396	439	2473	$\tau_1 = 0.62$ (6.83%) $\tau_2 = 2.12$ (93.17)	0.99
THF	0.210	388	448	3452	2.85	0.99
EtOAc	0.200	387	445	3368	2.83	1.03
CHCl ₃	0.148	402	447	2504	2.87	1.00
DMSO	0.263	398	465	2620	1.42	1.04
CH ₃ CN	0.305	396	458	3418	2.65	1.05
MeOH	0.309	401	467	3524	1.17	1.01

The Lippert-Mataga polarity parameter (Δf) has been considered as the measure of the polarity of different solvents and solvents mixtures used and was calculated using the following equation (2),^{11,20}

$$\Delta f = \frac{\epsilon - 1}{2\epsilon + 1} - \frac{n^2 - 1}{2n^2 + 1} \quad \text{eq (1)}$$

In different solvents, absorption and fluorescence spectra involve the electronic transition between the ground and excited electronic states, the Stokes shift ($\Delta\bar{\nu}$) is expected to follow a linear relation with (Δf), as suggested by the Lippert-Mataga equation as²⁰

$$\Delta\bar{\nu} = \Delta\bar{\nu}_0 + \frac{2(\mu_e - \mu_g)^2}{hca^3} \Delta f \quad \text{eq (2)}$$

where $\Delta\mu = \mu_e - \mu_g$ is the difference between the excited state and ground-state dipole moments, μ_e and μ_g , respectively, h is Planck's constant, c is the velocity of light, and a is

the Onsager radius for the interaction sphere of the dipole in a solvent. Figure 4 shows the Lippert-Mataga plot of $\Delta\bar{\nu}$ against the Δf values of the solvents. It is seen from this figure that though for almost all the solvents and solvent mixtures, the $\Delta\nu$ versus Δf plot is linear within experimental error, the $\Delta\nu$ values for the nonpolar solvents are unusually small and largely deviated from the above linear correlation.

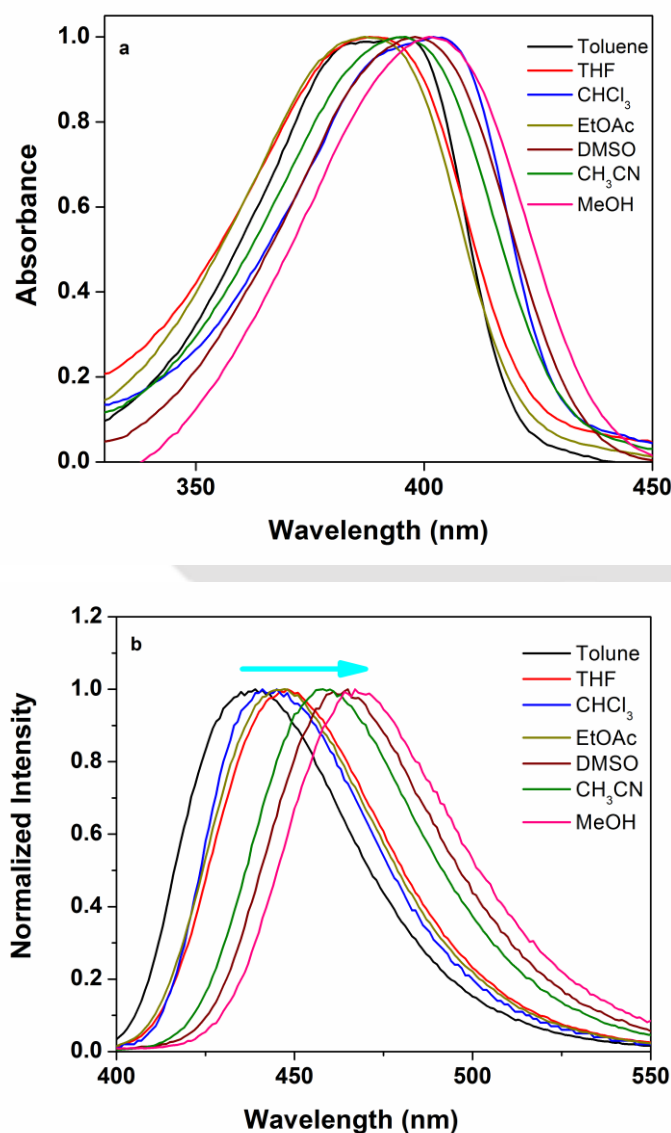


Figure 2. Normalized absorption (a) and fluorescence spectra (b) of **12z** in some of the representative solvents of varying polarities. The solvents are toluene, THF (tetrahydrofuran), CHCl₃ (chloroform), ethyl acetate, DMSO (dimethylsulfoxide), CH₃CN and MeOH (methanol). (Concentration = 5×10^{-6} M, rt; $\lambda_{\text{ex}} = 390$ nm).

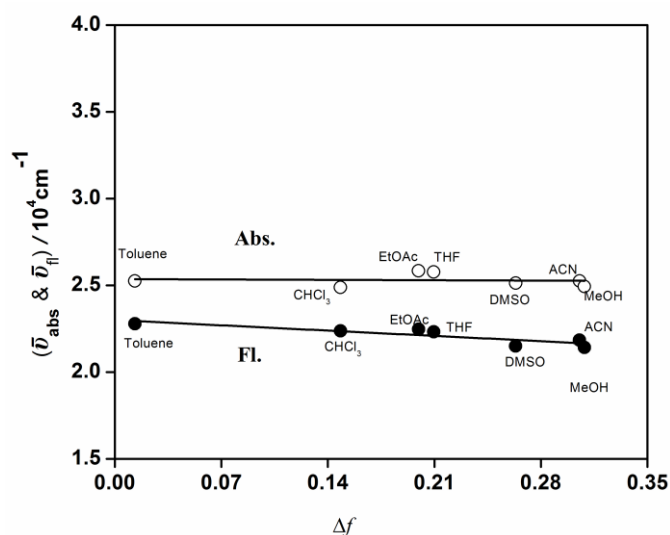


Figure 3. Plot of absorption (Abs.) and fluorescence (Fl.) maxima of fluorophores **12z** ($\bar{\nu}_{\text{abs}}$ and $\bar{\nu}_{\text{fl}}$ respectively) against solvent polarity functions, Δf

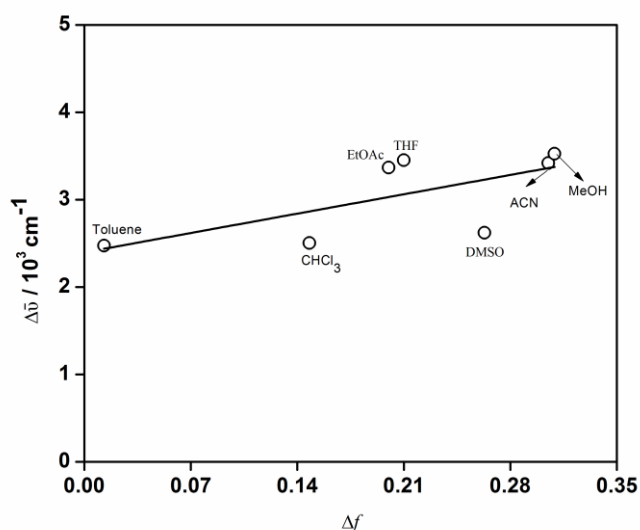


Figure 4. Lippert-Mataga plot of Stokes shift ($\Delta\bar{\nu}$) versus the Lippert solvent parameter $\Delta f = f(\epsilon) - f(n^2)$. The straight line represented the best linear fit.

4.1.2.2 Fluorescence Lifetimes

The time resolved fluorescence decay profiles of fluorophore **12z** in different solvents of varying polarities were measured at room-temperature by keeping the wavelength of measurement at $\lambda_{\text{max}}^{\text{fl}}$ of the probe in respective solvents (Figure 5). We observed that excluding toluene as solvent, the fluorescence decay of fluorophore **12z** in all other solvents are single exponential. The fluorescence lifetimes (τ) of the fluorophore **12z** thus

measured in different solvents are listed in Table 1. The fluorescence lifetimes (τ) of the fluorophore **12z** in toluene exhibits a biexponential decays with a lifetimes of 0.62 ns (6.83%) and 2.12 ns (93.17%).²¹ The lifetimes (τ) of fluorophore **12z** in solvents such as THF, ethyl acetate, CHCl_3 and CH_3CN are in the range of 2.64-2.87 ns, whereas fluorophore of **12z** in DMSO and MeOH exhibit monoexponential decay with lifetimes of 1.42 ns and 1.17 ns, respectively.

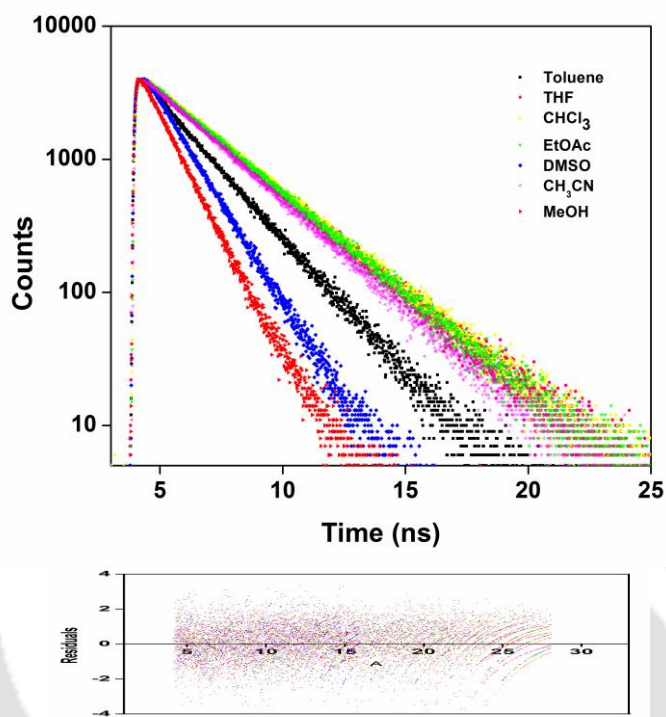


Figure 5. Time resolved fluorescence decay profiles of **12z** different solvent

4.1.3 Interaction Studies of Fluorophore **12z** with BSA

4.1.3.1 Absorption Spectra

We studied the interaction of fluorophore and BSA using UV-visible spectra. Absorption spectra of fluorophore **12z** has been recorded in PBS buffer (pH 7.0) at different concentrations of BSA ranges from 0-20 μM (Figure 6). The absorption spectra of free **12z** showed absorption band at 419 nm. Thus, upon addition of BSA concentration (0-20 μM) to the solution of fluorophore **12z**, we observed that the hyperchromicity in the absorption spectral maxima of fluorophore with hypsochromic shift from 419 nm to 415 nm. This result clearly indicates the presence of interaction between **12z** and BSA.

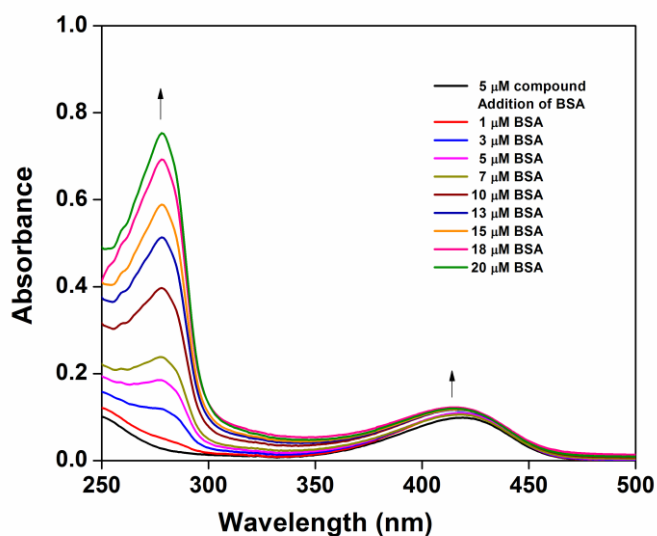


Figure 6. Changes in the absorption spectra of fluorophore **12z** (5 μM) in the presence of increasing BSA concentration (0–20 μM) in 10 mM PBS buffer (pH 7.0).

4.1.3.2 Fluorescence Spectra

We investigated the interaction of fluorophore **12z** with BSA using the fluorescence technique, which is more sensitive than absorption spectra. The effect of BSA on the fluorophore **12z** is shown in Figure 7. In the absence of BSA, fluorophore **12z** exhibits fluorescence band at 487 nm in 10 mM PBS buffer (pH = 7.0) when excited n at 414 nm

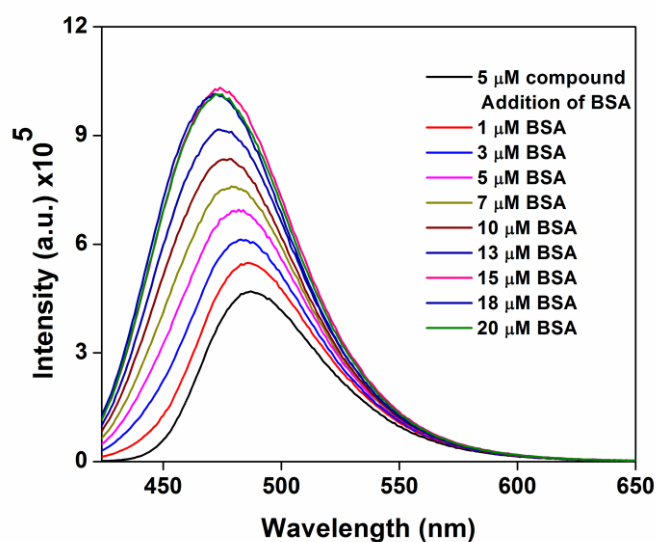


Figure 7. Emission spectra of fluorophore **12z** (5 μM) in the presence of increasing BSA concentration (0–20 μM) in 10 mM PBS buffer (pH 7.0) . [λ_{ex} = 414 nm].

(Table 2). Thus upon addition of BSA concentrations (0-20 μM) to the fluorophore **12z** solutions, fluorescence intensity of probe **12z** increased up to 2.2 folds with substantial hypsochromic shift 15 nm. The observed hypsochromic shift in the emission spectra of the fluorophore **12z** in the presence of BSA indicates that the probe **12z** is moving from hydrophilic environment to deep inside the hydrophobic pocket of BSA protein.

Table 2. Fluorescence Lifetime (τ , ns) of fluorophore **12z** with the increase in addition of BSA Concentration.

[12z] / μM	[BSA] / μM	$\lambda_{\text{abs}}^{\text{max}}$ /nm	$\lambda_{\text{em}}^{\text{max}}$ /nm	Life time	
				τ/ns	χ^2
5	0	419	487	$\tau_1 = 0.56$ (96.01)	1.07
				$\tau_2 = 3.53$ (3.91)	
5	1	418	486		
5	3	278, 418	483	$\tau_1 = 0.59$ (68.19)	1.04
				$\tau_2 = 3.84$ (31.81)	
5	5	278, 417	482		
5	7	278, 417	479		
5	10	278, 415	478		
5	13	278, 415	474		
5	15	278, 415	474	$\tau_1 = 0.71$ (31.91)	1.03
				$\tau_2 = 3.98$ (68.09)	
5	18	278, 415	472		
5	20	278, 415	472		

4.1.3.3 Binding Constant

To investigate the binding of fluorophore **12z** with BSA, association constant or binding constant was calculated from the Benesi-Hilderbrand plot,²² which was constructed using the following equation (3).

$$1/(I - I_0) = 1/\{(I_\infty - I_0) K_{\text{B-H}} [\text{BSA}]\} + 1/(I_\infty - I_0) \quad (3)$$

where, I_0 and I are the fluorescence intensities of the fluorophore in the absence and presence of BSA, respectively. I_∞ represents the fluorescence intensity on complete complex formation of fluorophore with BSA. K_{B-H} is the binding constant or association constant. The binding constant (K_{B-H}) of **12z** with BSA is $4.20 \times 10^4 \text{ M}^{-1}$, which was calculated from slope of the Benesi-Hilderbrand plot (Figure 8).

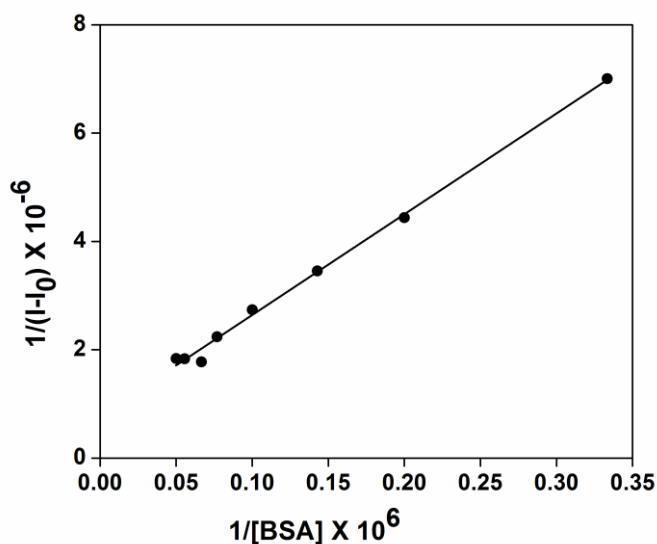


Figure 8. Benesi-Hilderbrand plot for fluorophore **12z** (5 μM) in the presence of increasing BSA concentration (0–20 μM)

4.1.3.4 Fluorescence Lifetime

The time resolved fluorescence decay profiles of fluorophore **12z** with BSA at different concentrations (0, 3 and 15 μM) is shown in figure 9. Time-resolved fluorescence analysis indicates that fluorophore **12z** alone exhibits a biexponential decay with the lifetimes of 0.56 ns (96.01%) and 3.53 ns (3.91%). Addition of BSA (3 μM) to the solution of fluorophore **12z** led to increase the biexponential decay significantly with the lifetimes of 0.59 ns (68.19%) and 3.84 ns (31.81%). Similarly, biexponential decay of **12z** further increased with the lifetimes of 0.71 ns (31.91%) and 3.98 ns (68.09%) when the concentration of BSA is enhanced to 15 μM . These observed changes in fluorescence decay profiles clearly support the complex formation of fluorophore **12z** with BSA (Table 2).

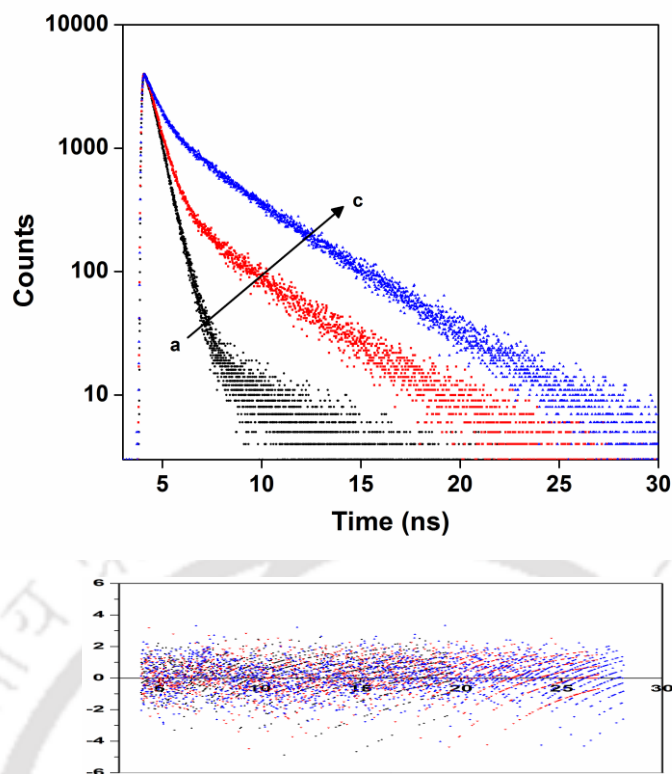


Figure 9. Time-resolved fluorescence decay profiles and residual plot for the titration of fluorophore **12z** (5 μM) with BSA. [BSA] a) 0 μM , b) 3 μM and c) 15 μM .

4.1.3.5 Absorption Job's Plot

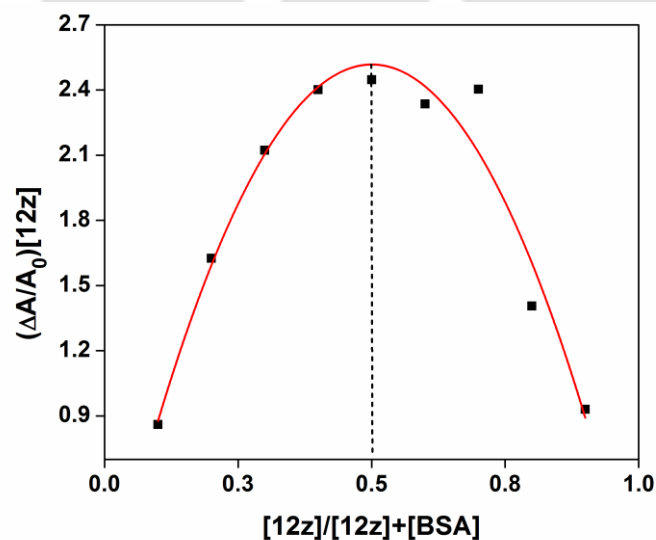


Figure 10. Absorption Job's plot of fluorophore **12z** in the presence of BSA protein indicates a 1:1 stoichiometry of complex formation of probe with BSA.

To quantify the stoichiometry of complex formation of fluorophore **12z** with BSA protein, absorption measurement was carried out using UV-visible spectroscopy for Job's plot by the plotting of [Probe-BSA protein] vs X_{probe} . Where [Probe-BSA protein] = $\Delta A/A_0 \times [probe]$, $\Delta A = A - A_0$ and X_{probe} is the mole fraction of probe, $X_{probe} = [probe]/([probe] + [BSA])$. The Job's plot of fluorophore **12z** (Figure 10) showed the point of maximum at the mole fraction of ~ 0.5 , which clearly suggested a 1:1 binding stoichiometry for complex formation between **12z** and BSA protein.

4.1.3.6 Study of Fluorescence Anisotropy

Next, the fluorescence anisotropy and polarization changes of fluorophore **12z** ($5 \mu\text{M}$) in the presence of increasing BSA concentration ($0\text{--}20 \mu\text{M}$) were measured to confirm the binding between probe and BSA. Thus, an enhancement of fluorescence anisotropy for the fluorophore **12z** from 0.10 in 10 mM PBS buffer ($\text{pH} = 7.0$) to 0.29 in the presence of BSA indicated that the probe **12z** bound strongly inside the hydrophobic pocket of BSA protein and experienced a highly restricted rotational motion (Figure 11).^{11,23}

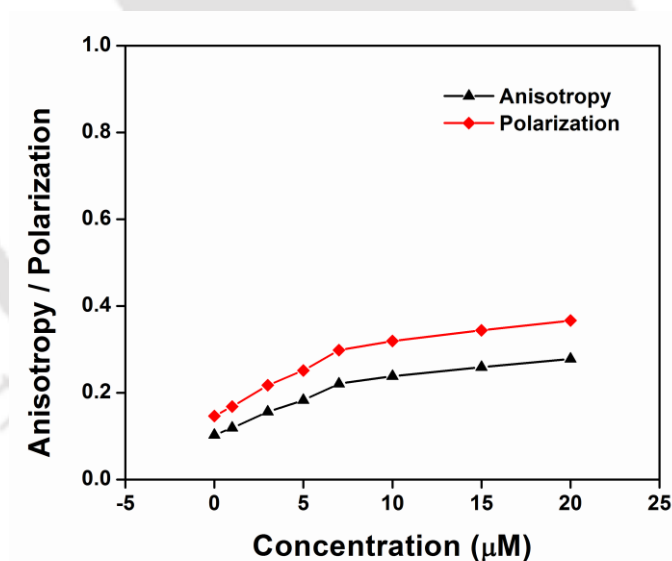


Figure 11. Fluorescence anisotropy and polarization changes of fluorophore **12z** ($5 \mu\text{M}$) in the presence of increasing concentration of BSA ($0\text{--}20 \mu\text{M}$).

4.1.3.7 Molecular Docking Study

To understand more details on molecular binding, docking study of fluorophore **12z** with BSA was carried out. When the molecules bind at the hydrophilic pockets of proteins, no

considerable shift in fluorescence spectrum was observed.²⁴ On the other hand, if the molecules enter into the hydrophobic pocket of the proteins, blue shift in the fluorescence spectrum was observed.²⁴ It is clearly observed from the fluorescence studies that fluorophore **12z** is in the hydrophobic environment (Section 4.1.2.2). This was proved by the docking study of fluorophore **12z** with BSA (Figure 12).

The fluorophore **12z** interact with hydrophobic and polar amino acid residues such as His337, Leu304, Thr305, Glu339, Tyr340, Phe373, Lys377, Val380, Asp381, Gln384 and Ser442 which are present in the hydrophobic pocket in the BSA protein *via* hydrogen bonding and van der Waal's interaction (Figure 12). The fluorophore **12z** forms one hydrogen bond with polar amino acid residue His337 through the sulfonyl oxygen (2.84 Å), which clearly indicates that BSA acts as hydrogen bond donor and fluorophore **12z** acts as hydrogen bond acceptor. In addition, **12z** involves the hydrophobic interaction with amino acid residues Leu304, Thr305, Glu339, Tyr340, Phe373, Lys377, Val380, Asp381, Gln384 and Ser442. This result clearly agrees with the experimental result of higher spectral shift in fluorescence of spectrum of fluorophore **12z** with BSA in PBS buffer (Table 2).²⁵

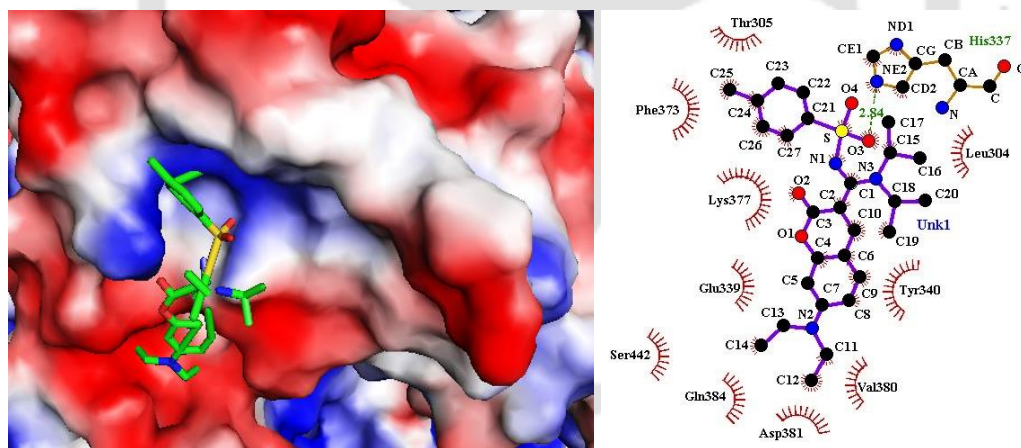


Figure 12. Docking pose and Molecular docking analysis results of fluorophore **12z** with BSA: amino acid surrounding ligands

The change in free energy, intermolecular energy, internal energy and torsional energy obtained from the docking study of BSA-fluorophore **12z** interaction are listed in Table 3. The change in free energy ($\Delta G = -6.40$ kcal/mol) obtained from the docking experiment is good agreement with free energy ($\Delta G = -6.31$ kcal/mol) calculated from experiment.

Table 3. Change in Free Energy ΔG (kcal/mol) and other Energies (kcal/mol) Obtained from Docking Study of Fluorophore **12z** with BSA.

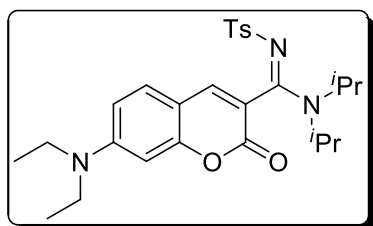
Molecule	ΔG^a	$E_{\text{intermolecular}}$	E_{internal}	$E_{\text{torsional}}$
12z	-6.40 (-6.31)	-9.09	-2.40	2.68

^a Change in free energy obtained from experiment is included in the parenthesis.

In summary, a new solvatochromic fluorophore **12z** was synthesized from the 4-(diethylamino)-2-hydroxybenzaldehyde (**1s**), ethyl propiolate (**8a**), tosyl azide (**5a**) and diisoprylamine (**9a**) using microwave assisted copper catalyzed four component reaction. Sensitivity of fluorophore **12z** in different solvents with varying polarities was studied using absorption, fluorescence and time resolved fluorescence decay spectroscopy. The interaction of BSA with fluorophore **12z** was studied using the spectra of absorption, fluorescence, time resolved fluorescence decay and fluorescent anisotropy. The docking study of **12z** with BSA was performed. The fluorescence intensity of **12z** was increased with increasing concentration of BSA. The hypsochromic shift in fluorescence spectrum of **12z** suggests that ligand binds at hydrophobic pocket of BSA. These results are unanimously agreement with docking experiment. From the docking study, it is revealed that BSA acts as hydrogen bond donor and fluorophore act as hydrogen bond acceptor which plays a crucial role in molecular binding.

4.2 Experimental Section

General Information. CuI (98%) and 4-(diethylamino)-2-hydroxybenzaldehyde (98%) of Aldrich used as received. ethyl propiolate (99%) and K_2CO_3 (98%), were purchased from commercial suppliers and used as received. Solvents were purchased from commercial source and purified prior to use.¹¹ Tosyl azides¹⁴ was prepared according to the literature procedures. BSA, $NaHPO_4$ and $NaH_2PO_4 \cdot H_2O$ from Merck, India were used as received. Milli-Q (18.2 Ω) were obtained from purification system. All the photophysical experiments were performed using freshly prepared solutions. Recrystallization of fluorophore **12z** was done by slow evaporation method using a 1:1 mixture of dichloromethane and methanol at ambient temperature.



Synthesis of (Z)-7-(diethylamino)-N,N-diisopropyl-2-oxo-N'-tosyl-2H-chromene-3-carboximidamide **12z**.

To a stirred solution of ethyl propiolate **8a** (0.5 mmol, 49 mg), tosyl azide **5a** (0.6 mmol, 118.2 mg) and CuI (0.05 mmol, 9.5 mg) in 1,4-dioxane (1 mL) was added diisopropylamine **9a** (0.5 mmol, 61 mg), 4-(diethylamino)-2-hydroxybenzaldehyde **1s** (0.5 mmol, 96.5 mg) and K₂CO₃ (0.6 mmol, 82.9 mg) at room temperature under air. After 0.5 h (arrested N₂ bubbles), the reaction vial was sealed with a cap and stirred at 130 °C, [150 W] for 1 h using microwave-irradiation. The solvent was then evaporated under *vacuo* and the residue was diluted with CH₂Cl₂ (30 mL), and washed with saturated NH₄Cl (10 mL) and water (10 mL). Drying (Na₂SO₄) and evaporation of the solvent gave a residue that was purified on silica gel column chromatography using n-hexane/ethyl acetate as eluent; Yellow solid; yield 25%; mp 207-208°C. ¹H NMR (600 MHz, CDCl₃) δ 7.63 (d, *J* = 6.0 Hz, 2H), 7.56 (s, 1H), 7.31(s, 1H), 7.14 (d, *J* = 12.0 Hz, 2H), 6.61 (s, *J* = 12.0 Hz, 1H), 6.44 (s, 1H), 3.96-3.91 (m, 1H), 3.67-3.61 (m, 1H), 3.43 (q, *J* = 6.0 Hz, 4H), 2.36 (s, 3H), 1.58 (d, *J* = 6.0 Hz, 3H), 1.45 (d, *J* = 6.0 Hz, 3H), 1.24-1.20 (m, 9 H), 1.10 (d, *J* = 6.0 Hz, 3H); ¹³C{¹H} NMR (150 MHz, CDCl₃) δ 159.0, 158.4, 157.1, 151.6, 142.7, 141.7, 141.1, 130.2, 129.0, 126.7, 114.6, 109.4, 107.4, 97.4, 52.5, 48.4, 45.2, 21.7, 20.8, 20.4, 20.1, 19.7, 12.6; FT-IR (KBr) 3434, 2925, 1715, 1602, 1525, 1414, 1355, 1253, 1133, 1085, 757, 668, 550 cm⁻¹. HRMS (ESI) *m/z* calcd for C₂₇H₃₆N₃O₄S [M+H]⁺: 498.2427; found: 498.2435.

Photophysical Study of Fluorophore **12z**

UV-visible and Fluorescence Measurements

Absorption spectra of fluorophore **12z** (5 x 10⁻⁶ M) was recorded in different solvents of varying dielectric constants on Perkin-Elmer Lambda-750 UV-visible spectrophotometer using 1 cm path length cell at 298 K. Fluorescence spectra of **12z** (5 x 10⁻⁶ M) were recorded in different solvents of varying dielectric constants on Horiba Fluoromax-4 spectrofluorometer using 1 cm path length cell at 298 K. Time resolved fluorescence decays profiles of **12z** (5 x 10⁻⁶ M) were recorded on Picosecond Time-resolved cum Steady State Luminescence Spectrometer at 298 K.

Study on Interaction of Fluorophore 12z with BSA

Preparation of BSA Solution

BSA standard solution was prepared using Milli-Q water. A BSA stock solution (1000 μM) was prepared by dissolving 0.198 g of BSA in 3 mL Milli-Q water.

General procedure for studying the interaction with BSA

The interaction of fluorophore **12z** with BSA was studied by titration of **12z** (5×10^{-6} M, 1% DMF in 10 mM PBS, pH = 7.0) with different concentrations of BSA (ranging from 0, 1, 3, 5, 7, 10, 13, 15, 18, 20 μM). The total volume of the final solution for each sample was 3 mL. 2-3% of DMF does not induce structural changes to biomolecules. Each and every sample solution was thoroughly mixed prior to spectral experiments.

UV-Visible Study

The UV-Visible absorbance measurements were recorded using Perkin-Elmer Lambda-750 UV-visible spectrophotometer using a cell of 1 cm path length at 298 K. All the UV-visible studies were carried out in 10 mM phosphate buffer (1% DMF) of pH 7.0 at 298 K. All the experiments were carried out under ambient condition with freshly prepared sample solutions at wavelength regime of 200-600 nm.

Fluorescence Study

Fluorescence experiments were recorded using Horiba Fluoromax-4 spectrofluorometer using a cell of 1 cm path length at 298 K. All the fluorescence studies were carried out in 10 mM phosphate buffer (1% DMF) of pH 7.0 at 298 K. Steady state anisotropy of the solutions was recorded using Fluoromax-4 spectrophotometer at 298 K. Time resolved fluorescence decays profiles of **12z** (5×10^{-6} M, 1% DMF in 10 mM PBS, pH = 7.0) with different concentrations of BSA (ranging from 0, 3, 15 μM) were recorded on Picosecond Time-resolved cum Steady State Luminescence Spectrometer at 298 K.

Docking Study

The crystal structure of BSA (PDB ID:3VO3) was downloaded from the RSCB Protein Data Bank.²⁶ Five cycle of energy minimization was done using Swiss pdb viewer²⁷

before using it for molecular modeling and docking screening. The final energy minimized structure was saved in pdb format, which was to be used for the docking experiments. This structure was converted to pdbqt format in autodock 4.1 and polar hydrogens, kollman charges, gastegier charges and atomic solvation charges were accordingly assigned.²⁸ The optimized ligand (fluorophore) was next converted to pdbqt. For docking a grid of 0.375Å was centered around the potential ligand binding pocket of BSA. Genetic algorithm was chosen for the docking simulations and number of generation was fixed at 27000. Energy evaluations and GA runs were set at 2500000 and 20, respectively. These data were fed to the racoon VS software of autodock to perform uninterrupted docking with probe **12z**.²⁹ After docking simulation, the BSA-probe **12z** molecular models sorted, on the basis of lowest binding energy. The BSA-probe **12z** molecular model further analyzed in molecular visualization software pyMOL v0.99.^{30a} The molecular interaction between bound probe **12z** and BSA residues was done by LigPlot+.^{30b}

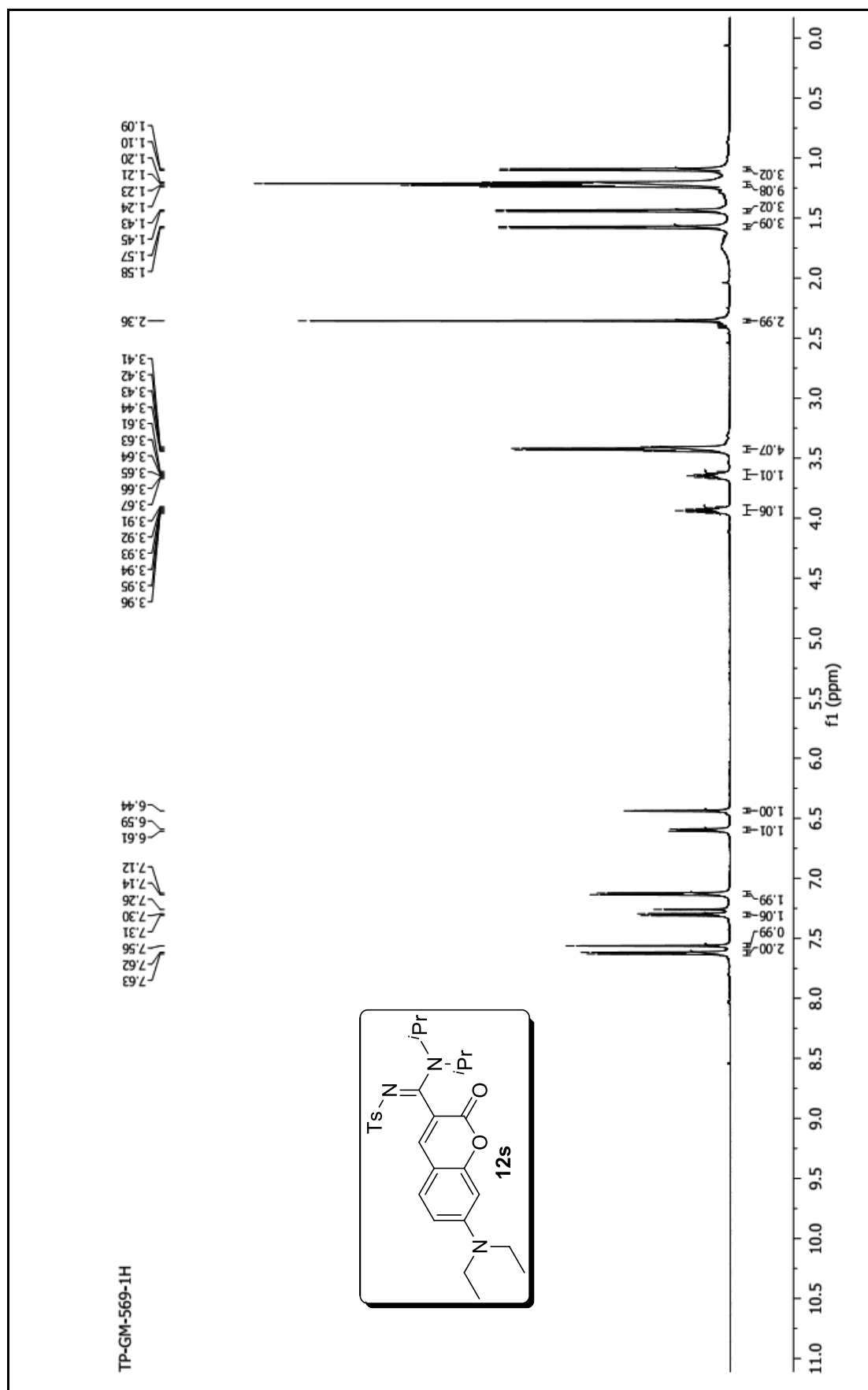
4.5 References

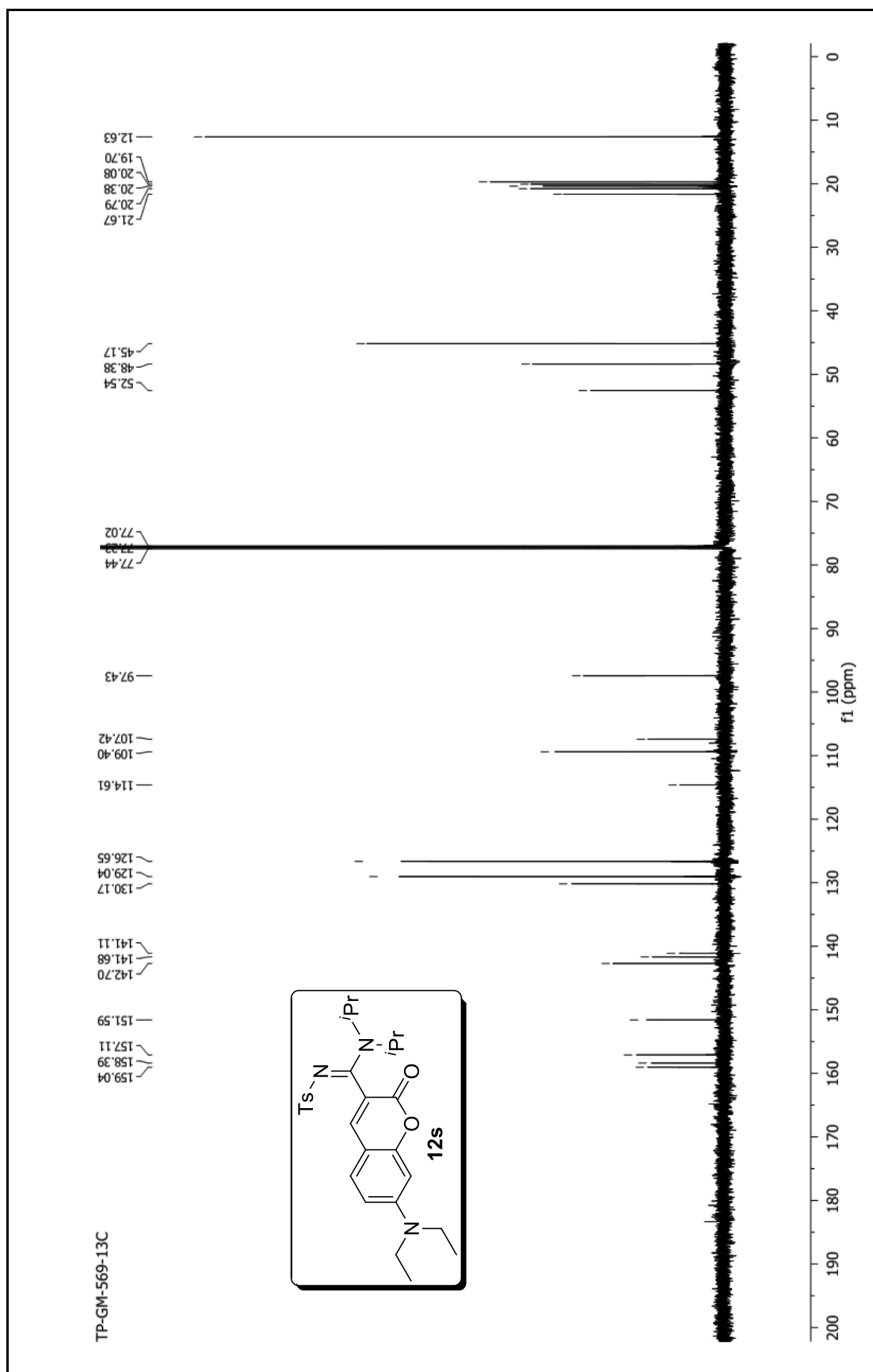
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Anticancer Activity of Iminocoumarin Aryl Methyl Ethers Against Triple Negative Breast Cancer Cell (TNBC) Line MDA-MB-231

Worldwide, the largest group of malignant disease is known as cancer or carcinoma, which is the most frequent cause of death in the direction of developing community. Usually, cancers are a large family of diseases dynamically involves in the uncontrolled cell division and subsequently, results in rapid tumor formation of metastasis to any part of the body, thereby disabling the vital functions of organs. In 2007, heterogeneous entity disease breast cancer was speculated in 1.3 million women and also increases the rate of mortality. The death rate often intensifies due to malignancy of an aggressive subtype, heterogeneous triple negative breast cancer (TNBC).^{1,2} The breast cancer cells (TNBC) lack receptors such as estrogen, progesterone and human epidermal growth receptor 2 (HER2), was characterized by immunohistochemical confirmation.^{3,4} Further, important parameters such as race, genetics, age, waist or hip ratios, breast feeding patterns, and disparity can progress towards various fashion of development of TNBC.⁵ Especially, breast cancer is commonly observed in African-American women and black ethnicities.^{6,7} In general, lack of breast feeding after parturition develops the risk of TNBC in females. Besides, the disparities in living lifestyle may promotes the increase of co-morbidity thereby playing a part in association with the biology of TNBC.⁸⁻¹⁰ In mice, involution of breast tissues after parturition or lactation followed by the activation of cyclooxygenase 2 (COX2), also referred as prostaglandin G/H synthase 2 and tissue inflammation envisages poor survival with invasive breast cancer. During the wound healing programmes, the deposition of high-risk cross-linked, aligned fibrillar collagen also predicts the risk of survivability.¹¹ Further, diminution of mortality rate due to breast cancer was observed globally in high income countries. Thus survivability of women with TNBC is always deprived or critical due to biological differences.¹²

Scientifically, protein kinase C (PKC) is a family of serine/ threonine lipid dependent protein kinases which involves a various important physiological processes such as cell proliferation, differentiation, signal transduction, modulation of gene expression and apoptosis.¹³⁻¹⁵ All these physiological functions in the cells are due to the phosphorylation

of target proteins. However, limited experiments demonstrated the activation of PKC after exogenous treatment of phorbol ester increased the cell motility resulted in the fibrosarcoma cells. Conversely, regulatory effect and distribution by the activation of PKC abrogates the mobility of cells in Walker carcinosarcoma cells, bladder carcinoma cells followed by ER⁻ breast cancer cell lines.¹⁶ Till date, TNBC a sub-type possessing aggressive biology has poor prognosis and treatment failures owing to the increased resistance against expression of targeted receptors. Even though chemotherapy is considered to be the most effective option,¹⁷ there is an urgent need for the development of new lead drugs for the cure of an infectious disease and also for the prevention and incidences of death.

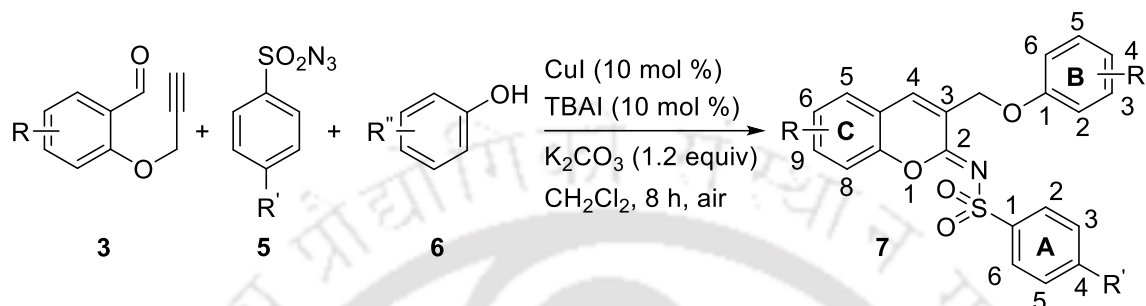
The majority of new drugs are small functionalized heterocyclic molecules used in standard therapy which can play a vital role for the lead drug discoveries and also to serve as a potential drug candidates.¹⁸⁻²⁵ The development of effective new drugs rely greatly on high efficiency, low cost and short synthetic sequences. In accordance, multicomponent reactions (MCRs) are considered to be one of the promising tools for the development of new drugs by tackling structural complexities as well as introduction of new functionalities through one-pot process. Iminocoumarins are one of the important structural scaffolds 2-imino derivatives of coumarin exhibiting widespread biological and medicinal properties such as antitumour activity against the Ehrlich ascites tumour, inhibitors of dynamins I and II GTPase, protein-tyrosine kinase p56lck, and HIV-1 integrase.²⁶⁻³¹ Hence, recent research suggested that derivatization of coumarin through MCRs has a significant evidences that it can act as an anti-tumour or chemotherapeutic agent against breast carcinoma.

5.1 Present Study

Development of effective drug candidates³² for triple breast cancer rely on high efficiency, low cost and short synthetic sequences are greatly driving the attention among the synthetic organic chemists and biologists. In this present study, we presented a series of iminocoumarin analogues which has been accomplished using copper-catalyzed multicomponent reactions followed by screening of anticancer activity of the compounds against MDA-MB-231 Triple Negative Breast Cancer cell line.

The iminocoumarin aryl methyl ethers analogues **7a-h**, **7j**, **7l-n**, **7r-x**, **7z** and **7ab-ad** using Cu(I)-catalyzed three component reaction of corresponding ynals **3**, sulfonyl azides

5 and phenols **6** via cascade [3+2] cycloaddition, ketenimie rearrangement, 1,4-addition and aldol condensation reaction (Scheme 1).³³ All the multicomponent variations R of **3**, R' of **5** and R'' of **6** afforded the corresponding iminocoumarin analogous using 10 mol % CuI with 10 mol % tetrabutylammoniumiodide (TBAI) in the presence of 1.2 equiv K₂CO₃ in CH₂Cl₂ under air at ambient temperature.

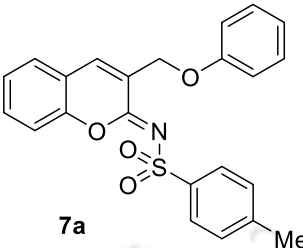
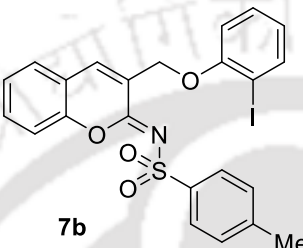
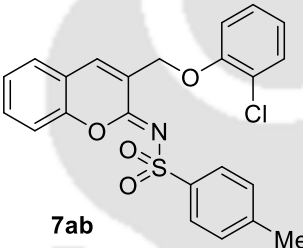
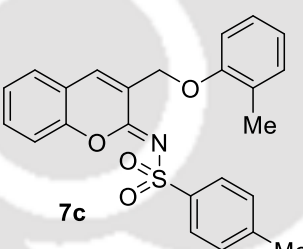
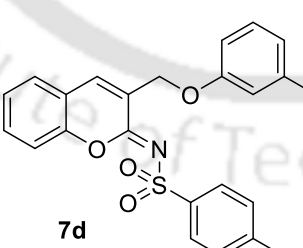
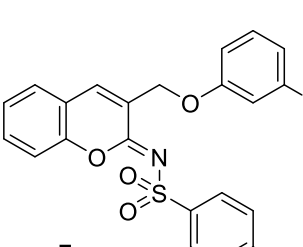


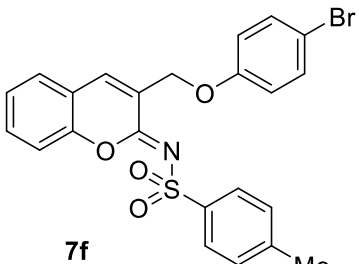
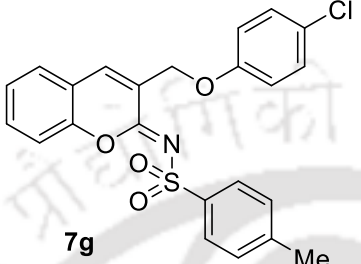
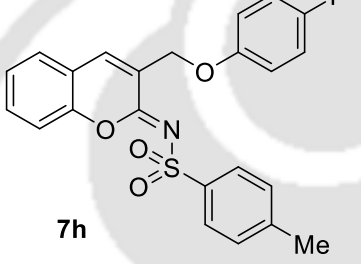
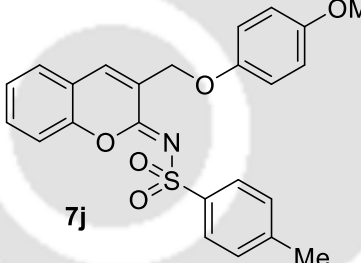
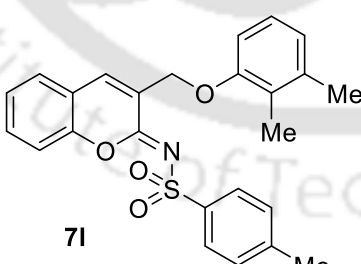
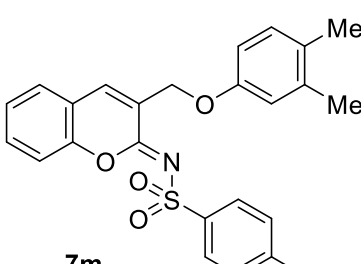
Scheme 1

5.1.1 Anticancer activity

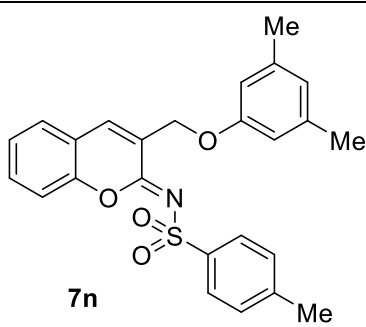
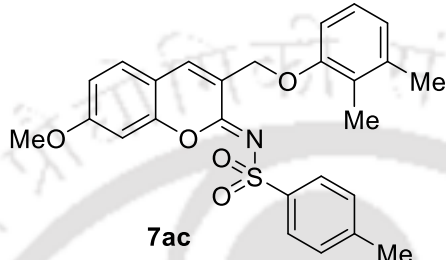
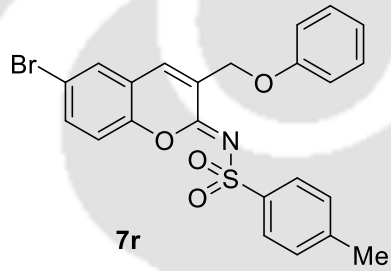
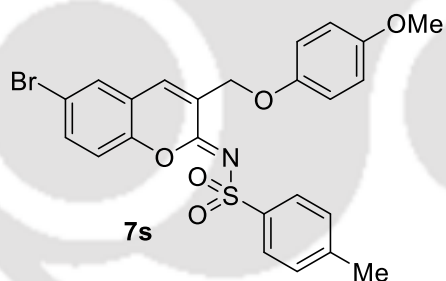
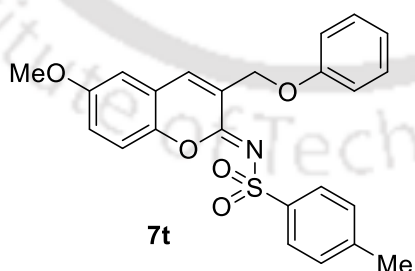
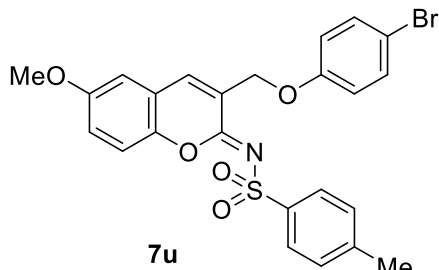
The anticancer activity of the synthetic analogues **7a-h**, **7j**, **7l-n**, **7r-x**, **7z** and **7ab-ad** was evaluated against human metastatic triple negative breast cancer cell line MDA-MB-231 for 48 h in a dose dependent manner using MTT assay. Based on structural variations such as R, R' and R'', the tested compounds has been categorized as *N*-sulfonyl derivatives **7a**, **7z**, phenolic substitutions **7b-h**, **7j**, **7l-n** and **7ab**, and derivatives of ynals **7r-x**, **7ac-ad** with their IC₅₀ (μM) values are shown in Table 1. The obtained IC₅₀ values against MDA-MB-231 cell lines revealed that iminocoumarin analogues having 2-I, 3-Br and 3,5-Me₂ substituents on ring **B** of **7b**, **7d** and **7n** have superior anticancer activity with its IC₅₀ value 0.62 μM, 0.68 μM and 1.61 μM respectively. Consequently, the analogues **7e** bearing 3-Me on ring B, **7r** with 4-Br on ring B and 6-OMe on ring C, and **7x** having 5,6-fused benzene ring on C showed higher activities with IC₅₀s, 4.59–7.75 μM against metastatic TNBC cell line. Further, the analogues **7ab**, **7s** and **7w** showed mild anticancer activity with IC₅₀s, 12.48-13.54 μM. In addition to iminocoumarin analogues **7a**, **7c**, **7f-m**, **7ac**, **7v** and **7ad** are found to be less effective (IC₅₀ > 30 μM). The viability assay depicted that iminocoumarin analogues **7b**, **7d**, **7e**, **7n** and **7u** are inhibiting the proliferation and causing death of highly robust breast cancer cells and leads us to explore the cause of the cell death.

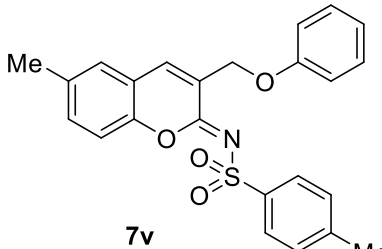
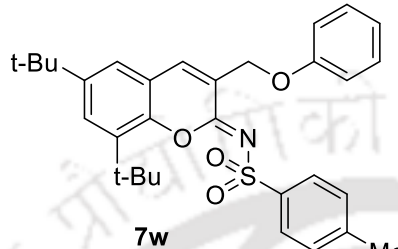
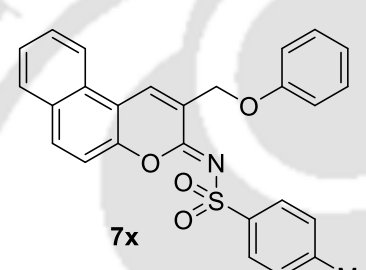
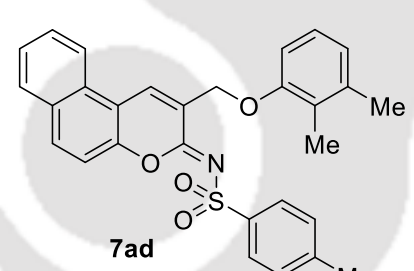
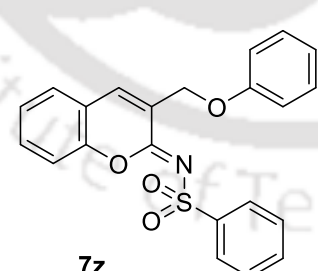
Table 1. Anticancer activity of the Iminocoumarin Analogues against MDA-MB-231 Cell Line

Entry	Compound	Cell viability, IC ₅₀ (μM ± SD)
1	 <p>7a</p>	39.36 ± 2.42
2	 <p>7b</p>	0.62 ± 0.15
3	 <p>7ab</p>	12.62 ± 0.68
4	 <p>7c</p>	46.69 ± 1.77
5	 <p>7d</p>	0.68 ± 0.08
6	 <p>7e</p>	6.13 ± 0.68

Entry	Compound	Cell viability, IC ₅₀ (μM ± SD)
7	 <p>7f</p>	> 50
8	 <p>7g</p>	> 50
9	 <p>7h</p>	> 50
10	 <p>7j</p>	> 50
11	 <p>7i</p>	40.53 ± 1.01
12	 <p>7m</p>	> 50

Anticancer Activity of Iminocoumarin aryl methyl ethers

Entry	Compound	Cell viability, IC ₅₀ (μM ± SD)
13	 <p>7n</p>	1.61 ± 0.30
14	 <p>7ac</p>	> 50
15	 <p>7r</p>	5.72 ± 0.74
16	 <p>7s</p>	12.48 ± 1.24
17	 <p>7t</p>	6.71 ± 0.90
18	 <p>7u</p>	4.69 ± 0.25

Entry	Compound	Cell viability, IC ₅₀ (μM ± SD)
19	 <p>7v</p>	> 50
20	 <p>7w</p>	13.54 ± 0.91
21	 <p>7x</p>	7.75 ± 0.66
22	 <p>7ad</p>	> 50
23	 <p>7z</p>	30.90 ± 2.04

5.1.2 Morphological Analysis

The five potent compounds **7b**, **7d**, **7e**, **7n** and **7u** which showed very good activity against MDA-MB-231 were selected for more detailed examinations. In view of that, the compounds **7b**, **7d**, **7e**, **7n** and **7u** were investigated for identification of apoptosis induced by MDA-MB-231 cells. Morphological analysis of **7b**, **7d**, **7e**, **7n** and **7u**-treated cells for 24 h was studied using light microscopy after staining with acridine orange/propidium iodide. The morphological changes observed in the stained cells revealed that iminocoumarin analogues **7b**, **7d**, **7e**, **7n** and **7u** reduced cell viability (Figure 1). The significant increment of apoptosis and ROS level induced by iminocoumarin analogues in MDA-MB-231 cell line was observed (data not shown).

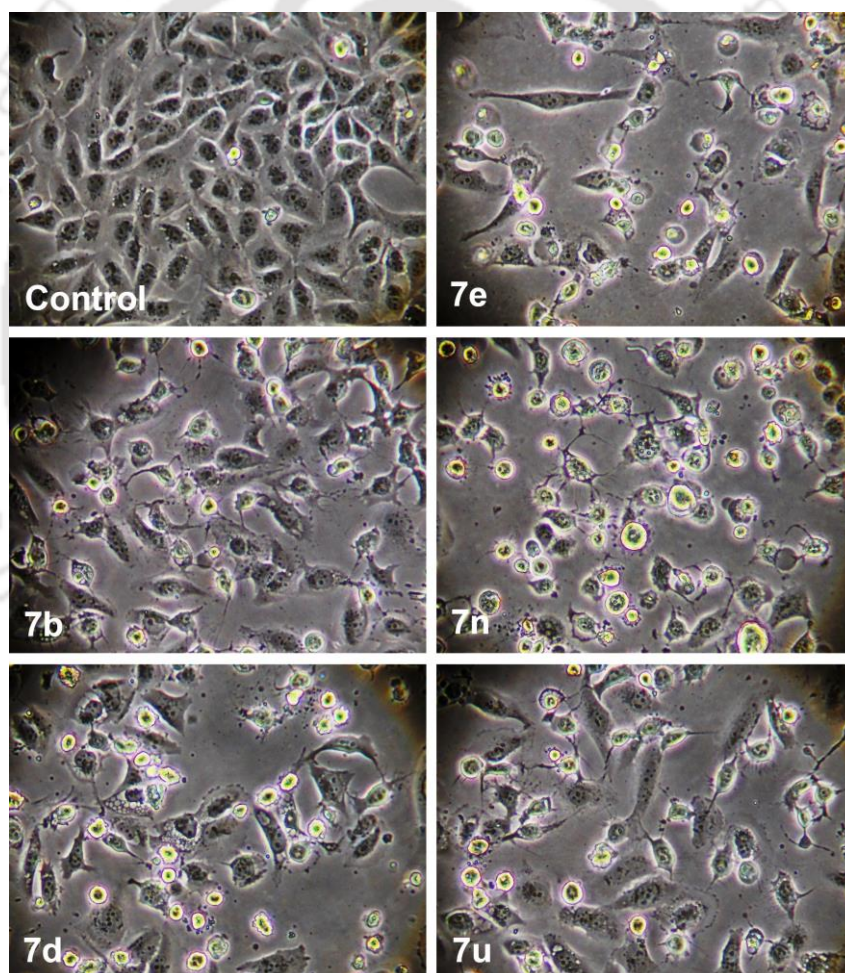


Figure 1. Cellular imaging of compounds **7b**, **7d**, **7e**, **7n** and **7u** against MDA-MB-231 cell line illustrating the morphological analysis.

5.1.3 Molecular Modelling Analysis

A molecular modeling and virtual screening technique was used to screen iminocoumarin analogues as potential agonists of PKC α . Protein Kinase C (C1b domain) PKC³⁴ was chosen as the most appropriate target for our synthesized iminocoumarin analogues. The

Table 2. Docking Results of Iminocoumarin analogues with PKC α (C1b domain)

Entry	Iminocoumarin analogues	Binding Energy (kcal/mol)	Inhibition constant Ki (μ M)
1	7b	-6.90	8.70
2	7c	-6.85	9.52
3	7d	-6.94	8.12
4	7e	-6.88	9.06
5	7f	-6.83	9.81
6	7g	-7.53	3.02
7	7h	-7.03	7.08
8	7j	-7.32	4.33
9	7l	-6.93	8.36
10	7m	-7.09	6.36
11	7n	-7.04	6.96
12	7r	-7.11	6.13
13	7s	-6.66	13.5
14	7t	-6.65	13.30
15	7u	-6.56	15.58
16	7v	-6.90	8.79
17	7w	-5.99	40.97
18	7x	-6.64	13.49
19	7x	-6.76	11.01

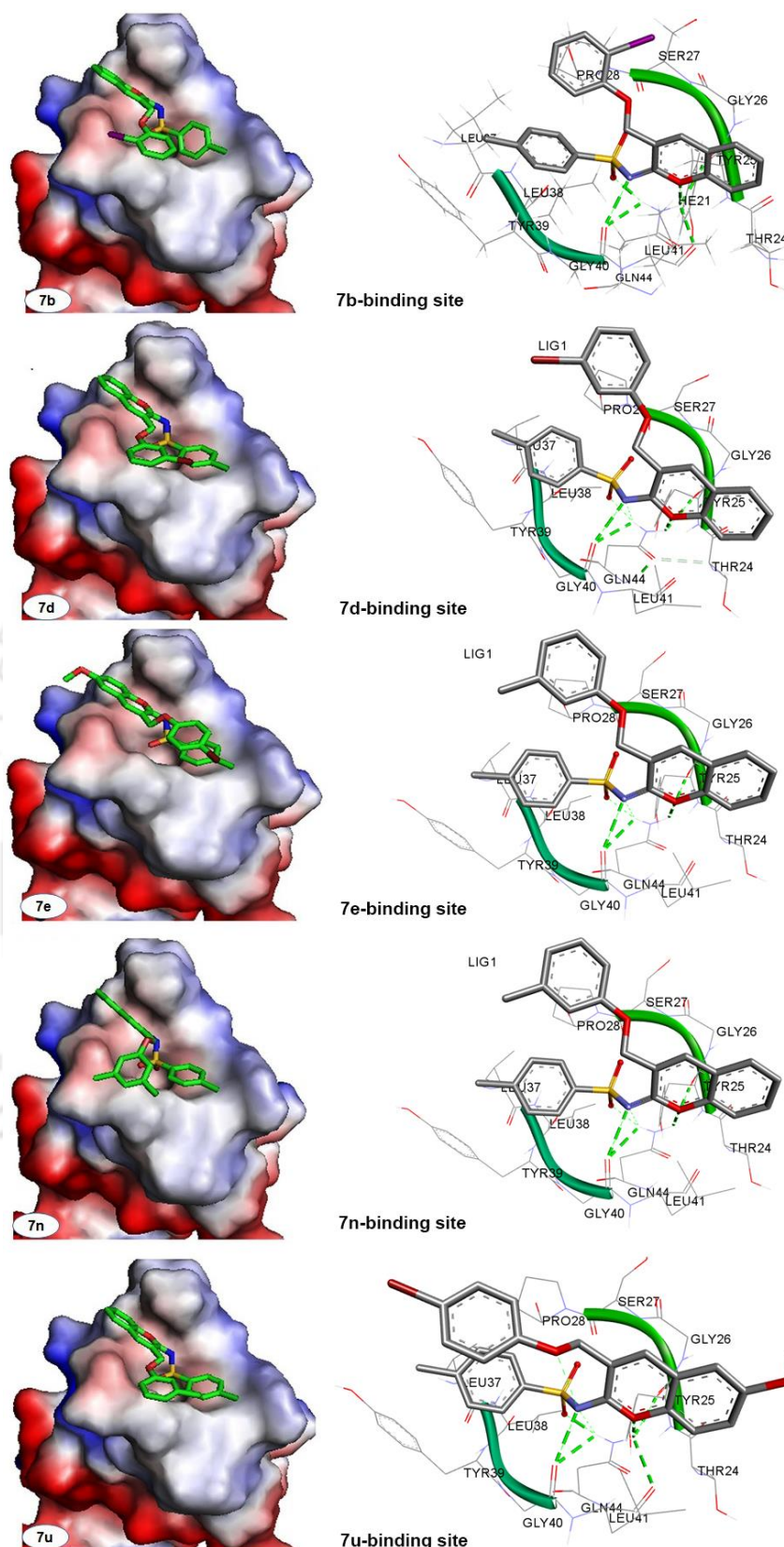
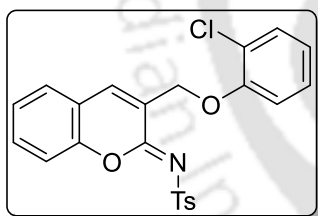


Figure 2. Docking poses and molecular docking analysis results: amino acid surrounding ligands of iminocoumarin analogues **7b**, **7d**, **7e**, **7n** and **7u** with PKC α (C1b domain)

docking results of some of the iminocoumarin analogues PKC α (C1b domain) are given in Table 2. Molecular screening studies of the iminocoumarins against PKC α showed that some of these iminocoumarins **7b**, **7d**, **7e**, **7n** and **7u** bind very strongly to the polar binding pocket of the C1b domain of the PKC α . The molecules **7b**, **7d**, **7e**, **7n** and **7u** exhibit hydrogen bonding interactions with Gly40, Gln44 and hydrophobic interaction with protein residues Leu41, Leu37, Pro28 and Tyr39 (Figuer 2).

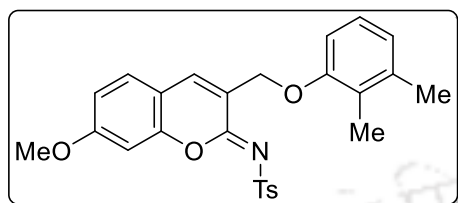
In conclusion, a series of iminocoumarin aryl methyl ether analogues were prepared and tested for their anticancer activity against triple negative breast cancer cell line. Of the various compounds, compound **7b** and **7d** potently inhibited the growth of metastatic TNBC cell line MDA-MB-231 with its IC_{50s} 0.62 \pm 0.05 μ M and 0.68 \pm 0.08 μ M respectively. Docking studies of the compound **7d** with PKC α , a protein kinase C isoforms, exhibits strong hydrogen bonding interactions with Gly40, Gln44 and strong hydrophobic interaction with protein residues Leu41, Leu37 and Tyr39. Iminocoumarin-based therapy acts as promising lead drug candidate has therapeutic potential and pharmacological intervention by elucidating the biological roles in the treatment of massive proliferation of breast cancer.

5.2 Experimental Section

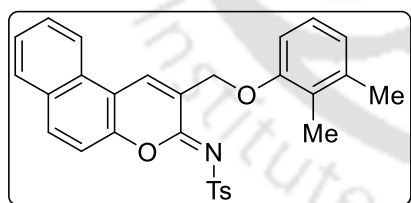


(Z)-N-(3-((2-chlorophenoxy)methyl)-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide (7ab). To a stirred solution of 2-(prop-2-yn-1-yloxy)benzaldehyde (0.5 mmol), tosyl azide (0.6 mmol), 2-chlorophenol (0.6 mmol) and CuI (9.5 mg, 10 mol %, 0.05 mmol) in CH₂Cl₂ (3 mL) using CaCl₂ guard tube under air, K₂CO₃ (152 mg, 1.1 mmol) and TBAI (18.5 mg, 10 mol %, 0.05 mmol) were added. The reaction was stirred at room temperature for 8 h and then the reaction mixture was then diluted using CH₂Cl₂ (30 mL) and washed with brine (1 x 10 mL) and water (2x10 mL). Drying (Na₂SO₄) and evaporation of the solvent gave a residue that was purified by silica gel column chromatography using hexane and ethyl acetate as eluent. Black solid; yield 10%; mp 192-193 °C. ¹H NMR (400 MHz, CDCl₃) δ 8.09 (s, 1H), 8.04 (d, *J* = 8.0 Hz, 2H), 7.60-7.56 (m, 2H), 7.46-7.37 (m, 3H), 7.35 (d, *J* = 8.4 Hz, 2H), 7.24 (d, *J* = 8.4 Hz, 1H), 7.04 (d, *J* = 7.6 Hz, 1H), 6.97 (d, *J* = 8.0 Hz, 1H), 5.00 (s, 2H), 2.43 (s, 3H);

$^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 156.1, 153.6, 152.3, 143.7, 138.9, 137.9, 132.2, 130.7, 129.6, 128.7, 128.2, 127.9, 126.1, 125.8, 123.2, 122.6, 119.4, 116.7, 114.3, 65.3, 21.8; FT-IR (KBr) 3064, 2962, 2923, 2853, 1632, 1597, 1553, 1485, 14550, 1319, 1287, 1261, 1206, 1160, 1082, 1017, 952, 904, 816, 760, 661, 617, 589, 556, 545, 537 cm^{-1} . HRMS (ESI) m/z calcd for $\text{C}_{23}\text{H}_{19}\text{ClNO}_4\text{S}$ $[\text{M}+\text{H}]^+$: 440.0718, found: 440.0727.



(Z)-N-(3-((2,3-dimethylphenoxy)methyl)-7-methoxy-2H-chromen-2-ylidene)-4-methylbenzenesulfonamide 7ac. According to procedure of **7ab**, the compound **7ac** was prepared from 4-methoxy-2-(prop-2-yn-1-yloxy) benzaldehyde, tosyl azide and 2,3-dimethylphenol. Pale yellow solid; yield 25%; mp 193-194 $^{\circ}\text{C}$. ^1H NMR (600 MHz, CDCl_3) δ 8.00 (s, 1H), 7.85 (s, 1H), 7.43-7.26 (m, 3H), 7.03-6.73 (m, 5H), 4.89 (s, 2H), 3.90 (s, 3H), 2.42 (s, 3H), 2.29 (s, 3H), 2.24 (s, 2H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 163.1, 156.7, 156.1, 154.0, 143.4, 139.4, 138.4, 137.7, 129.5, 129.3, 127.6, 126.2, 125.5, 123.3, 123.1, 114.6, 113.1, 109.9, 100.6, 64.9, 56.2, 21.8, 20.3, 12.1; FT-IR (KBr) 2922, 2861, 1638, 1617, 1547, 1505, 1468, 1407, 1356, 1312, 1263, 1241, 1205, 1156, 1133, 1088, 1025, 920, 863, 827, 818, 785, 767, 741, 705, 664, 619, 588, 554, 540 cm^{-1} . HRMS (ESI) m/z calcd for $\text{C}_{26}\text{H}_{26}\text{NO}_5\text{S}$ $[\text{M}+\text{H}]^+$: 464.1526, found: 464.1536.



(Z)-N-(2-((2,3-dimethylphenoxy)methyl)-3H-benzo[f]chromen-3-ylidene)-4-methylbenzenesulfonamide (7ad). According to procedure of **7ab**, the compound **7ad** was prepared from 2-(prop-2-yn-1-yloxy)-1-naphthaldehyde, tosyl azide and 2,3-dimethylphenol. Pale yellow solid; yield 15%; mp 272-273 $^{\circ}\text{C}$. ^1H NMR (600 MHz, CDCl_3) δ 8.77 (s, 1H), 8.27 (d, $J = 12.0$ Hz, 1H), 8.07-8.02 (m, 3H), 7.95 (d, $J = 12.0$ Hz, 1H), 7.23 (t, $J = 12.0$ Hz, 1H), 7.64-7.57 (m, 2H), 7.35 (d, $J = 12.0$ Hz, 2H), 7.08 (t, $J = 12.0$ Hz, 1H), 6.87-6.83 (m, 1H), 5.03 (s, 2H), 2.42 (s, 3H), 2.35 (s, 3H), 2.33 (s, 3H); $^{13}\text{C}\{^1\text{H}\}$ NMR (150 MHz, CDCl_3) δ 156.3, 156.1, 151.9, 143.9, 143.5, 139.2, 138.5, 133.5, 133.2, 131.04, 129.5, 129.3, 128.9, 128.8, 127.8, 127.0,

126.4, 126.2, 125.5, 123.6, 121.8, 116.3, 114.3, 110.2, 65.2, 21.8, 20.3, 12.3; FT-IR (KBr) 3074, 3052, 3017, 2908, 2853, 1633, 1543, 1510, 1468, 1439, 1385, 1313, 1301, 1286, 1259, 1219, 1157, 1106, 1087, 906, 888, 874, 823, 769, 733, 678, 628, 590, 553, 542 cm^{-1} . HRMS (ESI) m/z calcd for $\text{C}_{29}\text{H}_{26}\text{NO}_4\text{S}$ [M+H]: 484.1577, found: 484.1581.

Biology Experiments

Cell culture. The human MDA-MB-231 breast cancer cell line were maintained in RPMI 1640 medium supplemented with 10% heat inactivated fetal calf serum, streptomycin (100 $\mu\text{g}/\text{mL}$) and penicillin (100 $\mu\text{g}/\text{mL}$) at 37 °C in a humidified atmosphere with 5% CO_2 .

MTT Assay

The anticancer activity was determined by cell viability assay. The dye MTT (3-(4,5-Dimethylthiazol-2-yl)-2,5-Diphenyltetrazolium Bromide) was used to measure cellular viability. Ten thousand cells of MDA-MB-231 breast cancer cell line were seeded in each well of 96 well plates containing 200 μL DMEM F12 complete medium and kept for adhesion. Then the cells were washed twice with cell culture grade phosphate buffer saline (PBS) and were subsequently incubated with various iminocoumarin analogues **4a-w** at increasing concentrations from 0-50 $\mu\text{g}/\text{mL}$ in 200 μL serum free medium for 24 h and 48 h. The cells treated with incomplete medium alone were considered as control. After treatment, the cells were washed twice with PBS and incubated with 100 μL of MTT (0.5 mg/mL) for 4 h at 37 °C with 5% CO_2 . Then, MTT solution was removed and formazan crystals were dissolved in 100 μL cell culture grade DMSO. The optical density was determined using a spectrophotometer (SpectraMax M2) at 570 nm. The graphs of percentage inhibition at different concentration were plotted in reference to control.

Apoptosis Assay

Apoptosis was performed to identify apoptotic and dead cells using acridine orange and propidium iodide dye staining. Fifty thousand cells of MDA-MB-231 breast cancer cell line were seeded in 24 well plate containing 1 mL DMEM F12 complete medium and kept for adhesion. Then the cells were washed twice with cell culture grade PBS and were subsequently incubated with various iminocoumarin analogues **7b**, **7d**, **7e**, **7n** and **7u** with the consistent IC_{50} in 1 mL serum free medium for 24 h. Cells treated with incomplete

medium alone were considered as control. After treatment, the cells were washed twice with PBS detached by 0.6% EDTA. Cells were centrifuged and re-suspended in PBS. Acridine orange and propidium iodide is added in the final concentration 1 $\mu\text{g}/\text{mL}$ and 10 $\mu\text{g}/\text{mL}$, respectively. Flow cytometry was performed by using BD FACS Caliber with the excitation wavelength 488 nm. The emission spectra was collected by two filters, FL-1 (525/20 nm) and FL-3 (635/20 nm). Quadrant analysis was done to identify live, early apoptotic, late apoptotic and dead cells in the total population of 10,000 cells by Cell Quest pro software (BD Biosciences, USA).

Oxidative Stress Assay

Fifty thousand cells of MDA-MB-231 breast cancer cell line were seeded in 24 well plate containing 1 mL DMEM F12 complete medium and kept for adhesion, then it was washed twice with cell culture grade PBS. Subsequently, the cultures were incubated with iminocoumarin analogues **7b**, **7d**, **7e**, **7n** and **7u** with the corresponding IC_{50} in 1 mL serum free medium for 24 h. Cells treated with incomplete medium alone were considered as control. After treatment, the cells were washed twice with PBS and incubated with 10 μM DCFDF dye for 30 min. Cells were detached with 0.6% EDTA. Flow cytometry was performed with the help of BD FACS Caliber with the excitation wavelength 488 nm and the emission spectra was collected by 525/20 nm filter (FL-1). Analysis was done with the help of Cell Quest pro software (BD Biosciences, USA).

Molecular Docking Analysis

The 3-D structure of the of $\text{PKC}\alpha$ (PDB ID: 2ELI) was downloaded as NMR structure from Protein data bank in PDB format (<http://www.rcsb.org/pdb/explore.do?structureId=3iw4>). The downloaded structure of $\text{PKC}\alpha$ encompasses the C1b domain, displaying the ligand binding pocket. Five cycle of energy minimization was done using Swiss pdb viewer before using it for molecular modeling and docking screening.³⁵ The final energy minimized structure was saved in pdb format which was to be used for the docking experiments. This structure was converted to pdbqt format in autodock 4.1 and polar hydrogens, kollman charges, gastegier charges and atomic solvation charges were accordingly assigned. The ligands (iminocoumarin analogues) were next converted to pdbqt.³⁶ For docking a grid of 0.375 \AA was centered around the potential ligand binding pocket of the C1b domain. Genetic algorithm was chosen for the docking simulations and

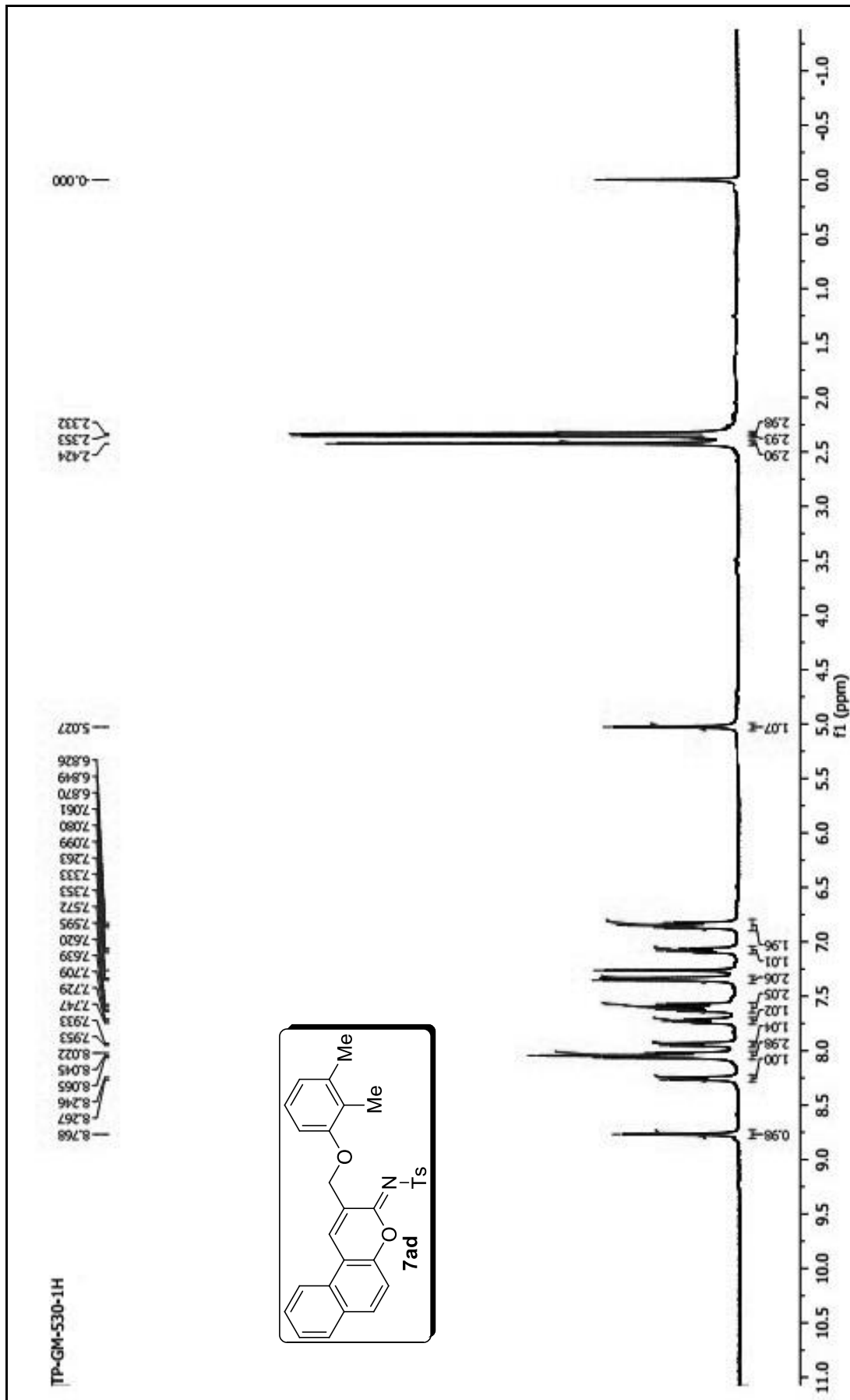
number of generation was fixed at 27000. Energy evaluations and GA runs were set at 2500000 and 20 respectively. These data were fed to the racoon VS software of autodock to perform uninterrupted docking with all iminocoumarin analogues.^{37a} After docking simulation, the PKC-agonist molecular models sorted, on the basis of lowest binding energy. The PKC-agonist molecular model further analyzed in molecular visualization software pyMol v0.99.^{37b} The molecular interaction between bound agonist molecules and PKC residues was done by Discovery Studio 4.5 Visualizer.

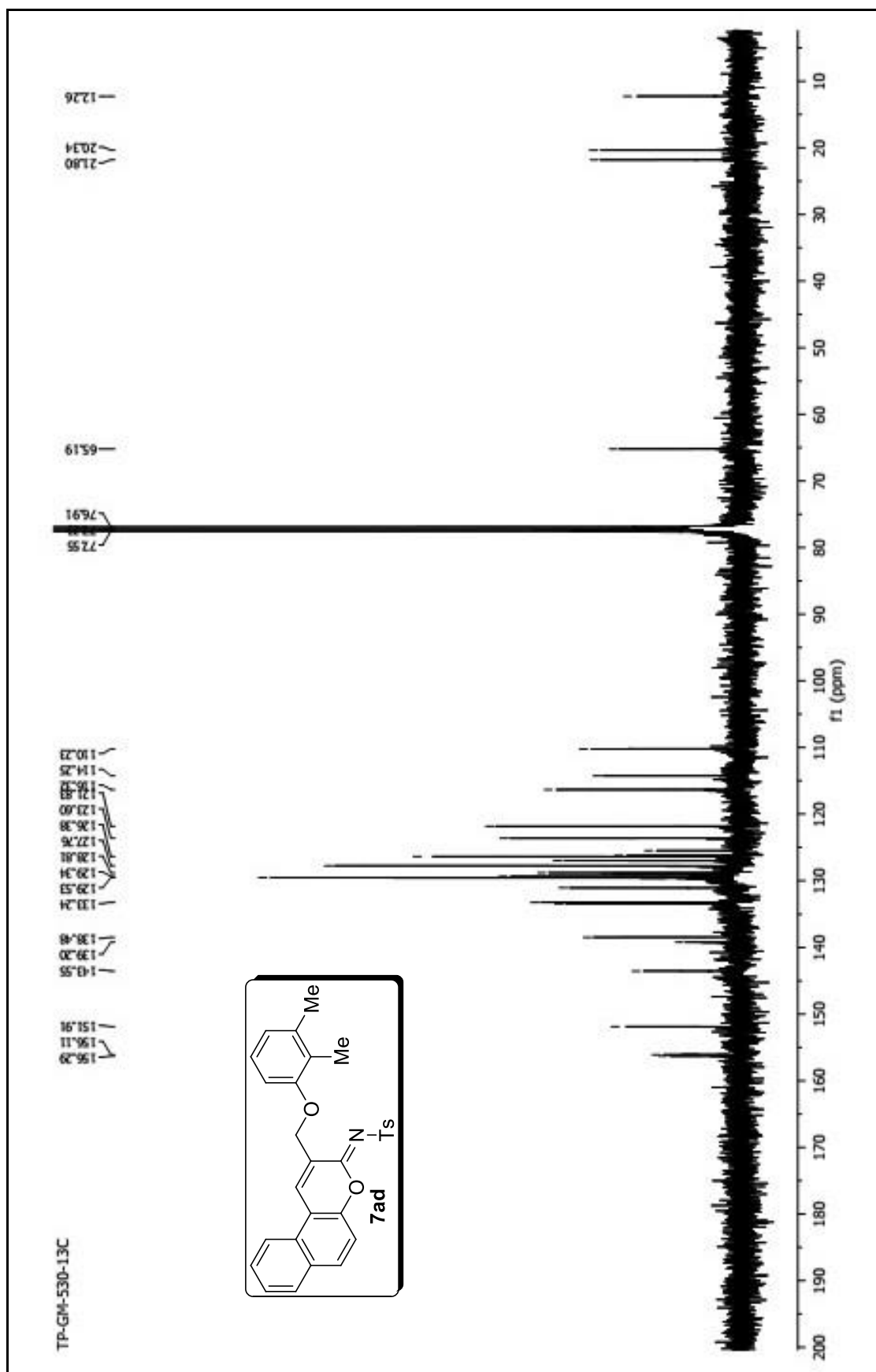
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Conclusions

In chapter I, literature survey of facile copper-catalyzed MCRs for heterocycle syntheses is covered. This reaction provides elegant contributions in heterocycle chemistry in recent years.

In chapter II, copper-catalyzed three-component synthesis of functionalized iminocoumarin aryl methyl ethers is described from ynal, phenols and sulfonyl azide *via* a cascade [3+2]-cycloaddition, 1,3-pseudopericyclic ketenimine rearrangement, 1,4-conjugate addition followed by aldol-type condensation. This protocol is simple and provides an efficient route for the construction of iminocoumarin aryl methyl ethers in good yields.

In chapter III, microwave-assisted copper(I)-catalyzed four-component cascade synthesis of 3-*N*-sulfonylamidine coumarins is presented from readily available salicylaldehydes, propiolates, sulfonyl azides and secondary amines. This reaction provides an effective route for the synthesis of functionalized 3-*N*-sulfonylamidine coumarins in a single operation under mild reaction conditions.

In chapter IV, the study on interaction of solvatochromic probe (*Z*)-7-(diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide with BSA protein is described. The fluorescence intensity of probe is increased with significant hypsochromic shift with increasing concentration of BSA. The docking study of (*Z*)-7-(diethylamino)-*N,N*-diisopropyl-2-oxo-*N'*-tosyl-2*H*-chromene-3-carboximidamide with BSA reveals that it acts as hydrogen bond donor and fluorophore acts as hydrogen bond acceptor, which plays a crucial role in molecular binding.

In chapter V, anticancer activity of iminocoumarin aryl methyl ethers is examined against metastatic triple negative breast cancer cell line MDA-MB-231. (*Z*)-*N*-(3-((2-Iodophenoxy)methyl)-2*H*-chromen-2-ylidene)-4-methylbenzenesulfonamide and (*Z*)-*N*-(3-((3-bromophenoxy)methyl)-2*H*-chromen-2-ylidene)-4-methylbenzenesulfonamide potently inhibit the growth of metastatic TNBC cell line MDA-MB-231 with its IC₅₀s 0.62 ± 0.05 μM and 0.68 ± 0.08 μM, respectively. Docking studies of (*Z*)-*N*-(3-((3-bromophenoxy)methyl)-2*H*-chromen-2-ylidene)-4-methylbenzenesulfonamide with PKCα, a protein kinase C isoforms, exhibited strong hydrogen bonding interactions with Gly40, Gln44 and strong hydrophobic interaction with protein residues Leu41, Leu37 and Tyr39. These results suggest that iminocoumarin-based drug therapy acts as promising lead drug candidate-

List of Publications

1. Novel Copper-Catalyzed Multicomponent Cascade Synthesis of Iminocoumarin Aryl Methyl Ethers.
Murugavel, G.; Punniyamurthy, T. *Org. Lett.* **2013**, *15*, 3828.
2. Microwave-Assisted Copper-Catalyzed Four Component Tandem Synthesis of 3-N-Sulfonylamidine Coumarins.
Murugavel, G.; Punniyamurthy, T. *J. Org. Chem.* **2015**, *80*, 6291.
3. Copper-Catalyzed Nitroaldol (Henry) Reaction: Recent Developments.
Murugavel, G.; Sadhu, P.; Punniyamurthy, T. *Chem. Rec.* **2016**, doi: 10.1002/tcr.201500268

Conferences

1. Novel Copper-Catalyzed Multicomponent Cascade Synthesis of Iminocoumarin Aryl Methyl Ethers.
Murugavel, G.; Punniyamurthy, T. 9th *J-NOST Conference 2013*, December 04-06, IISER Bhopal, India.
2. Multicomponent Cascade Synthesis of Iminocoumarin Aryl Methyl Ethers using Copper-Catalyst.
Murugavel, G.; Punniyamurthy, T. *International Symposium on Nature Inspired Initiatives in Chemical Trends 2014*, March 02-05, IICT Hyderabad, India.
3. Microwave-Assisted Copper Catalyzed Multicomponent Tandem Synthesis of Highly Functionalized Coumarin-3-N-sulfonylimidates.
Murugavel, G.; Punniyamurthy, T. *Nascent Developments in Chemical Sciences 2015*, October 16-18, BITS Pilani, India.