Synthetic Studies on Fused Pyridine/Dihydropyridine Based Heterocyclic Systems

THESIS

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by

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CERTIFICATE

This is to certify that the thesis entitled "**Synthetic Studies on Fused Pyridine/Dihydropyridine Based Heterocyclic Systems**" submitted by **T YADAGIRI** (ID. No. **2012PHXF0511H)** for award of Ph.D. of this institute embodies original research work done by him under my supervision.

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Name in capital letters : **Prof. ANUPAM BHATTACHARYA**

Designation : Associate Professor

Date :

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Date: **T YADAGIRI**

Abstract

The main aim of the research discussed in this thesis was to develop alternative synthetic routes to fused pyridine/dihydropyridine based heterocyclic compounds. The thesis is divided into five chapters. First chapter gives a general introduction to the research topic, whereas the final chapter contains summary, conclusions and future perspectives.

Chapter 1 (Introduction, objective & materials and methods): This chapter gives a general introduction about various fused pyridine based systems already known in literature and also emphasizes on their biological and physical properties. Main objective of this research work along with materials and methods is also mentioned in this chapter.

Chapter 2 (One pot two step Nazarov-Schmidt rearrangement for the synthesis of fused δ-lactam systems): Main emphasis here is the synthetic route developed for fused thieno/phenyl-dihydropyridine compounds using simple organic transformations.

Chapter 3 (Synthesis and anticancer activity of 1, 4-disubstituted imidazo[4,5-*c*]quinolines): This chapter describes three step synthetic protocol developed for the said compounds using modified Pistet-Spengler approach. This chapter also discusses the anticancer activity results obtained for 1, 4-disubstituted imidazo[4,5-*c*]quinolines.

Chapter 4 (Synthesis and anticancer activity of fused chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds): Work described in this chapter elaborates a new route developed for synthesis of the a forementioned compounds and also their anticancer activity.

Chapter 5 (Summary and conclusions): Discusses the main conclusions of the research work and gives the future perspective.

Table of contents

Table of contents

Table of contents

List of tables

List of figures

List of figures

List of abbreviations

ATP	Adenosine Triphosphate
BVDV	Bovine Viral Diarrhea Virus
°C	Degree symbol
CSA	Camphor sulphonic acid
$CDCl_3$	Chloroform-d
CCDC	Cambridge Crystallographic Data Centre
^{13}C-NMR	Carbon Nuclear Magnetic Resonance
COX	Cyclooxygenase
CK2	Casein Kinase 2
d	doublet
DCM	Dichloromethane
DCE	1,2-Dichloroethane
dd	doublet of doublet
DDPP	Diphenylphosphino-2-(diphenylphosphinomethyl)pyrrolindine
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
DFT	Density FunctionalTheory
DMF	Dimethylformamide
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid
DPE	Diphenyl ether
ESI	Electrospray Ionization
ESIPT	Excited-State Intramolecular Proton Transfer

HIV	Human Immunodeficiency Virus
DMEM	Dulbecco's Modified Eagle's Medium
HCC	Hepatocellular Carcinoma
HCV	Hepatitis C Virus
^{1}H-NMR	Proton Nuclear Magnetic Resonance
HRMS	High Resolution Mass Spectroscopy
5-HT_4R	5-Hydoxytryptamine Receptor 4
Hz	Hertz
IC_{50}	Half Maximal Inhibitory Concentration
IR	Infrared Spectroscopy
J	Coupling Constant
LRMS	Low Resolution Mass Spectroscopy
LCMS	Liquid Chromatography-Mass Spectrometry
m	multiplet
MALDI	Matrix-Assisted Laser Desorption Ionization
MHz	Mega Hertz
mg	milligram
MIC	Minimum Inhibitory Concentration
mmol	millimolar
M.p	Melting point
MTT assay	[3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide] assay
mL	microliter
MS	Mass Spectrum

NADP	Nicotinamide Adenine Dinucleotide Phosphate
NBS	*N*-Bromosuccinimide
NCS	*N*-Cholorosuccinimide
nm	nanometer
nM	nanomolar
NMR	Nuclear Magnetic Resonance
NSAIDs	NonsteroidalAnti-Inflammatory Drugs
ODCB	*o*-dichlorobenzene
PPA	Polyphosphoric acid
PTSA	p-Toluenesulfonic acid
q	quartet
t	triplet
TBAF	Tetrabutylammoniumfluoride
TEA	Triethylamine
TFA	Trifluoroaceticacid
THF	Tetrahydrofuran
THTP	Tetrahydrothieno pyridine
TLC	Thin Layer Chromatography
TLR	Toll-Like Receptor
TMS	Tetramethylsilane
TosMIC	Toluenesulfonylmethylisocyanide
μM	micromolar
UV	Ultraviolet

CHAPTER 1

Importance of fused pyridine/dihydropyridine systems

1. Introduction

Pyridine possesses one of the most elementary heteroaromatic structure and replacement of CH of benzene with N in case of pyridine leads to far reaching changes in its reactivity pattern. Pyridine which is far less susceptible towards reaction with electrophiles displays preference towards nucleophiles as well as undergoes electrophilic addition. Some useful products like 'pyridinium' salts or *N*-oxides have no existence in benzene chemistry.

In living systems, pyridine is found in various naturally occurring compounds such as NADP/NADPH (the redox system), vitamins pyridoxine and niacin, and alkaloids such as anabasine and niacin. Owing to this, pyridine is used in numerous pharmaceutical actives and agrochemical products.

1.1. Pyridine/dihydropyridine as commercially available drugs (shown in figure 1.1)

Some of the most important pyridine containing pharmaceuticals are proton pump inhibitors; lansoprazole, rabeprazole and omeprazole, which also comprise of benzaimidazole moiety attached to a sulfoxide unit at the 2^{nd} position. Other pyridine containing pharmaceutical agents include thiazolidinone based class 2-diabetes drugs such as rosiglitazone and pioglitazone. These molecules bind to the peroxisome proliferator-activated receptors, which migrate to the DNA on activation and help in regulating the transcription of genes controlling the carbohydrates and fatty acid metabolism. Desloratadine, a pyridine containing dual antagonist of platelet aggregating factor and of histamine, is used for the treatment of allergies. Recently a new class of NSAIDs, etoricoxib has been introduced commercially bearing a simple pyridine core. The molecule shows a 160-fold selectivity for COX-2 over COX-1.[1]

Dihydropyridines are found in many drug substances owing to high stability and their structural spacing especially in 1,4-form. One of the first drug molecules bearing this scaffold

was nifedipine, used as an antihypertensive and antianginal compound. Several analogues with similar structure have been commercialized like amlodipine and clevidipine.

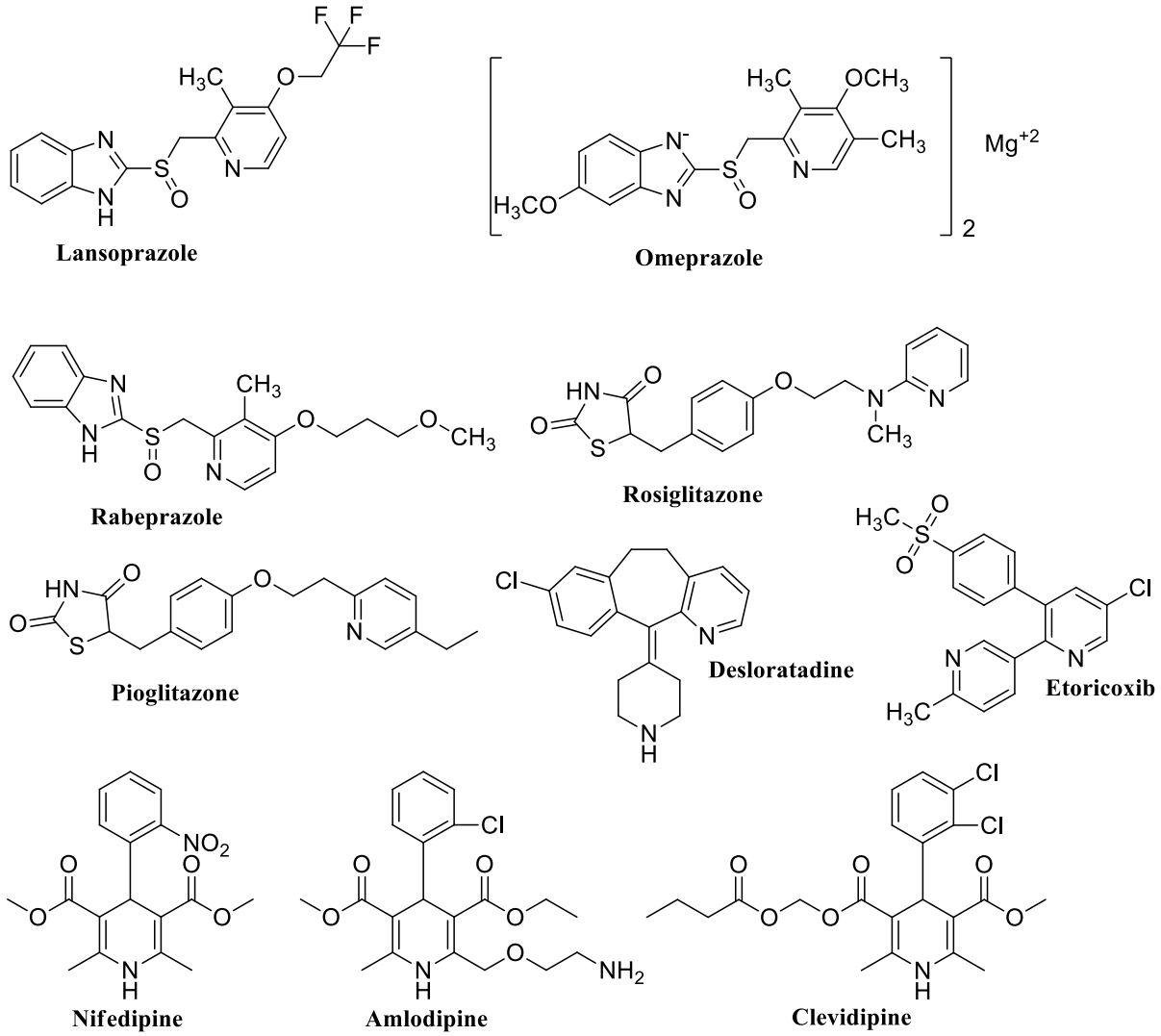
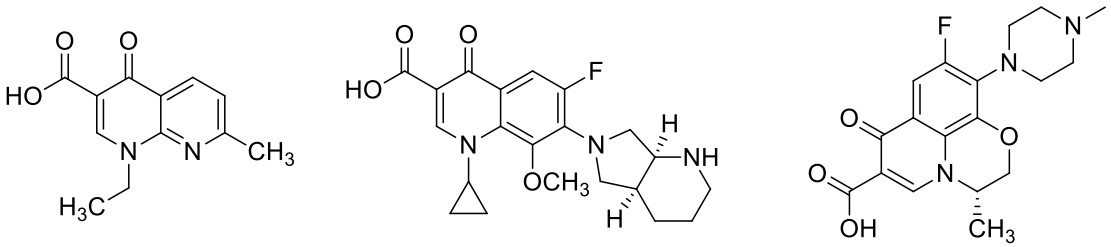


Figure 1.1: Commercial drugs based on pyridine and di-hydropyridine scaffolds

1.2. Quinolines as commercially available drugs

Although pyridines/dihydropyridines especially in an isolated arrangement are a vital component in several drugs, the quinoline/quinolone scaffolds bearing fused pyridine/pyridinone-benzene structures are also becoming increasingly common. For example nalidixic acid with a naphthyridone core has been effectively used against both gram-positive and gram-negative bacteria. Usually, quinolone based antibiotics act as DNA-

gyrase and/or topoisomerase inhibitors. Two third-generation fluoroquinolone antibiotics, moxifloxacin and levofloxacin are among the top selling drugs.

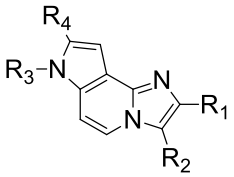


Nalidixic acid **Moxifloxacin** **Levofloxacin**

Figure 1.2: Commercial drugs based on quinoline skeleton

1.3. Diverse biological activity displayed by fused pyridine(quinoline)/dihydropyridine systems

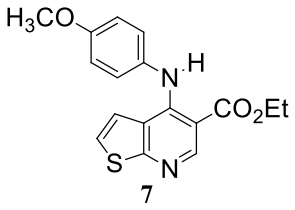
In a work reported by Chezal *et al.,* diverse imidazo[1,2-*a*]pyrrolo[2,3-*c*]pyridine compounds were synthesized and screened against bovine viral diarrhea virus (BVDV). Among the molecules screened compounds **1-6 (Figure 1.3)** displayed significant anti-BVDV activity.[2]



R_1 = Me; R_2 = H; R_3 = H; R_4 = CO_2Et **(1)**
R_1 = Ph; R_2 = H; R_3 = H; R_4 = CO_2Et **(2)**
R_1 = $CH(OEt)_2$; R_2 = H; R_3 = H; R_4 = CO_2Et **(3)**
R_1 = Me; R_2 = H; R_3 = Me; R_4 = CO_2Me **(4)**
R_1 = Me; R_2 = H; R_3 = H; R_4 = CO_2Me **(5)**
R_1 = Me; R_2 = H; R_3 = H; R_4 = CO_2Bu **(6)**

Figure 1.3: Imidazo[1,2-*a*]pyrrolo[2,3-*c*]pyridine compounds screened against BVDV

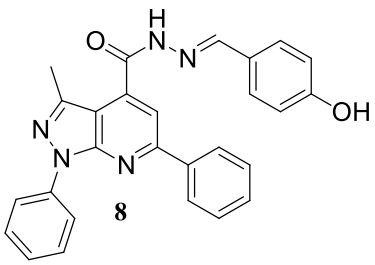
Bernadino *et al.,* described synthesis and antiparasitic activity of thieno[2,3-*b*]pyridine compounds. The study showed significant activity by *p*-methoxy substituted derivative **(Figure 1.4)**.[3]



7

Figure 1.4: Ethyl 4-((4-methoxyphenyl)amino)thieno[2,3-*b*]pyridine-5-carboxylate displaying significant anti-parasitic activity

Chagas disease caused by *Trypanosomacruzi* was targeted by 1*H*-pyrazolo[3,4-*b*]pyridine derivatives, in a study reported by Dias and co-workers. The study revealed compound **8** as the most potent molecule, with activity higher than nufurtimox, a medicine currently used for treating the Chagas disease **(Figure 1.5)**.[4]



8

Figure 1.5: *N*'-(4-Hydroxybenzylidene)-3-methyl-1,6-diphenyl-1*H*-pyrazolo[3,4-*b*]pyridine-4-carbohydrazide displays higher activity against Chagas disease than nufurtimox.

Hayakawa and coworkers carried out synthesis and biological evaluation of imidazo[1,2-*a*]pyridine compounds as novel PI3 kinase P110α inhibitors. Among the compounds screened, 3-{1-[(4-fluorophenyl)sulfonyl]-1*H*-pyrazol-3-yl}-2-methylimidazo[1,2-*a*]pyridine **(9)** and 6-chloro-3-{2-[(2-methyl-5-nitrophenyl)sulfonyl]-1,3- thiazol-4-yl}imidazo[1,2-a]pyridine hydrochloride**(10)** gave IC_{50} value of 0.0018 μM and 0.0028 μM, respectively **(Figure 1.6)**.[5]

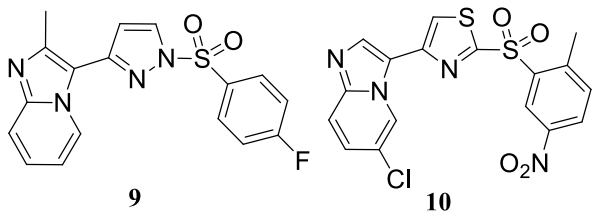


Figure 1.6: Imidazo[1,2-*a*]pyridine compounds displaying highest activity againstPI3 kinase.

In a study to target drug resistant *Staphylococcus epidermidis*, Leal *et al*., used 1*H*-pyrazolo[3,4-*b*]pyridine and thieno[2,3-*b*]pyridine compounds. Most active compounds were **11**, **12**, **13** and **14** where 1*H*-pyrazolo[3,4-*b*]pyridine skeleton was present **(Figure 1.7)**.[6]

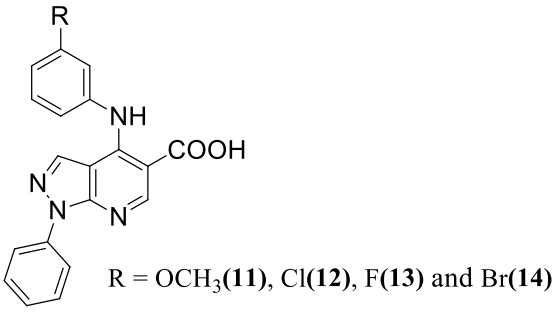
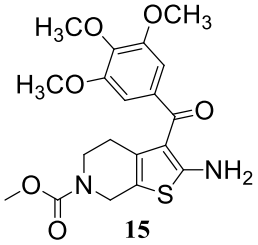


Figure 1.7: 1*H*-Pyrazolo[3,4-*b*]pyridine derivatives showing highest activity against *Staphylococcus epidermidis*.

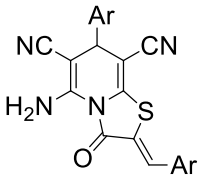
Tubulin polymerization inhibitor based on the 2-amino-3-(3,4,5-trimethoxybenzoyl)-4,5,6,7-tetrahydrothieno[*b*]pyridine molecular skeleton were synthesized and screened for antiproliferative activity, inhibition of tubulin polymerization, and cell cycle effects by Romangoli *et al*., The most active compound in this series, 2-amino-3-(3,4,5-trimethoxybenzoyl)-6-methoxycarbonyl-4,5,6,7-tetrahydrothieno[*b*]pyridine **(15)**, inhibited cancer cell growth in a panel of four cancer cell lines. IC_{50}-values ranged from 25 to 90 nM and the molecule was found to interact strongly with tubulin by binding to the colchicine site **(Figure 1.8)**.[7]



15

Figure 1.8: 2-Amino-3-(3,4,5-trimethoxybenzoyl)-6-methoxycarbonyl-4,5,6,7-tetrahydro-thieno[*b*]pyridine acting as tubulin polymerization inhibitor.

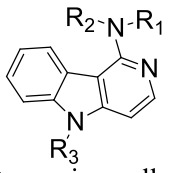
Shi *et al*., reported a green synthesis of thiazolo[3,2-*a*]pyridine derivatives in water using malononitrile, aromatic aldehydes and 2-mercaptoacetic acid. Compounds obtained were subjected to the experiments of antioxidant activity. Most of the compounds displayed capacities for scavenging free radicals **(Figure 1.9)**.[8]



Ar = various substituted phenyl rings

Figure 1.9: Diversethiazolo[3,2-*a*]pyridine derivatives as antioxidants.

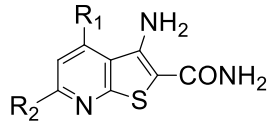
Wang and co-workers have shown synthesis of *N*-alkyl-5*H*-pyrido[4,3-*b*]indol-1-amines and their application as urotensin-II receptor antagonists **(Figure 1.10)**.[9]



$R_1/R_2/R_3$: various alkyl and aryl groups

Figure 1.10: *N*-Alkyl-5*H*-pyrido[4,3-*b*]indol-1-amine derivatives as urotensin-II receptor antagonists.

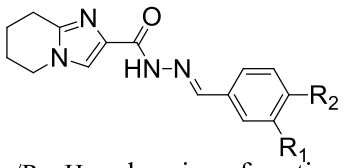
In a SAR study done by Wu and co-workers, a series of thienopyridine molecules were screened for IκB Kinase β (IKKβ) inhibition activity. Emphasis was given on the structural optimization at C4 and C6 of structure **(Figure 1.11).**The study disclosed preference for C4 position (R_1) by small alkyl and certain aromatic groups and polar groups with proper orientation at C6 position (R_2) for efficient enhancement of compound potency. IC_{50}s as low as 40 nM were displayed by the most potent analogues **(Figure 1.11)**.[10]



R_1: various alkyl and aryl groups
R_2: H or CH_3

Figure 1.11: Thienopyridine molecules as IκB Kinase β (IKKβ) inhibitors.

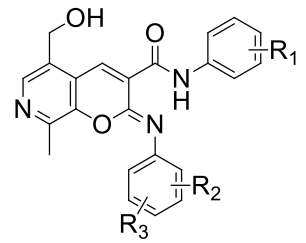
5,6,7,8-Tetrahydroimidazo[1,2-*a*]pyridine derivatives were studied by Ozdemir and coworkers for potential antifungal activity. Among the compounds screened, 5,6,7,8-tetrahydroimidazo[1,2-*a*]pyridine-2-carboxylic acid-(4-cyanobenzylidene) showed very strong inhibitory activity against the screened *Candida* species **(Figure 1.12)**.[11]



R_1/R_2: H and various functional groups

Figure 1.12: 5,6,7,8-Tetrahydroimidazo[1,2-*a*]pyridine derivatives as antifungal compounds.

In a study by Zhuravel *et al*., diverse 2-imino-5-hydroxymethyl-8-methyl-2*H*-pyrano[2,3-*c*]pyridine-3-carboxamides were screened for antibacterial and antifungal activities. Most of the compounds displayed comparable or even better potency than the standard drugs ciprofloxacin, fluconazole **(Figure 1.13)**.[12]

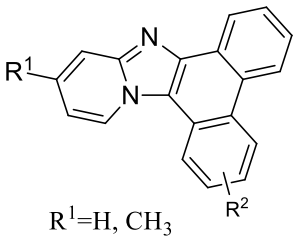


$R_1/R_2/R_3$: H and various alkyl and aryl groups

Figure 1.13: 2-Imino-5-hydroxymethyl-8-methyl-2*H*-pyrano[2,3-*c*]pyridine-3-carboxamides screened for antibacterial and antifungal activities.

1.4. Diverse photophysical properties showed by fused pyridine(quinoline) systems

Banerjee *et al*., recently reported synthesis and photophysical activity evaluation of fused phenthro-imidazo[1,2-*a*]pyridine derivatives. The molecules synthesized displayed good fluorescent property in solution as well as solid phase, which were further utilized in the imaging of live cells **(Figure 1.14)**.[13]



R^1=H, CH_3

R^2=H, CH_3, OCH_3, Et, Ph, ^{t}-Bu

Figure 1.14: Fused phenthro-imidazo[1,2-*a*]pyridine derivatives used for live cells imaging.

In a study by Wang and co-workers, synthesis and properties of two series of tetracyclic pyrazolo[3,4-*b*]pyridine-based coumarin chromophores were reported. The compounds exhibited high fluorescence quantum yields and good photochemical, thermal and electrochemical stabilities. Additionally, living cell imaging was also explored by laser scanning confocal microscopy **(Figure 1.15)**.[14]

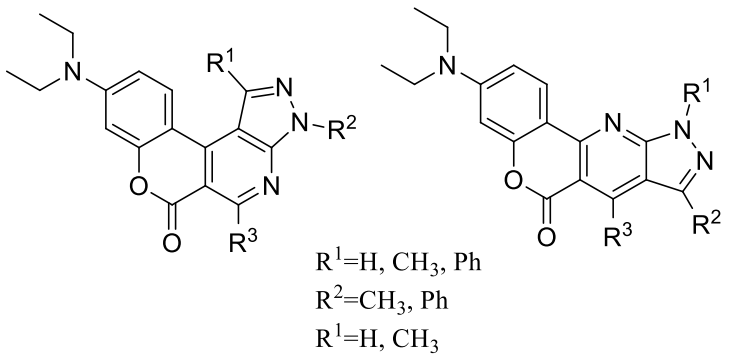


Figure 1.15: Tetracyclic pyrazolo[3,4-*b*]pyridine-based coumarin chromophores

Linear and non-linear optical properties of diverse imidazo [1,2-*a*]pyridines were studied by Daniel Gryko and co-workers. Extensive photophysical investigation was carried out on molecules bearing electron donating and electron withdrawing group at the 2nd position. The compounds showed fluorescence quantum yield ranging from 0.2 to 0.7, with a dependence on the substitution of the phenyl ring. A new quadrupolar system was also reported with two imidazo[1,2-*a*]pyridineunits on the outside and a 1,4-dicyanobenzene moiety at its center. This system showed a large Stokes-shifted luminescence and an acceptable two photon absorption response in the NIR region **(Figure 1.16)**.[15]

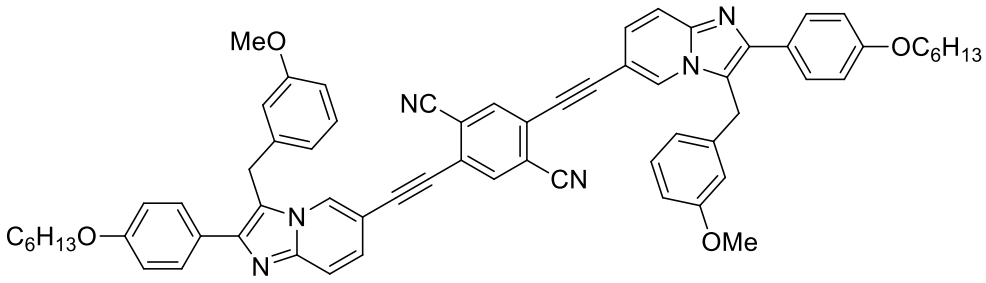


Figure 1.16: A quadrupolar system containing imidazo[1,2-*a*]pyridine unit with capability to show two photon absorption.

Kim *et al*., reported synthesis of a fluorescent core containing pyrazolo[1,5-*a*]pyridine-fused pyrimidine, *via* a one-pot silver catalyzed cascade cyclization. The molecules showed good

solvatochromism with a turn-on fluorescence in the lipophilic environment. The molecule with highest fluorescence was used to study lipid droplets in living cells **(Figure 1.17)**.[16]

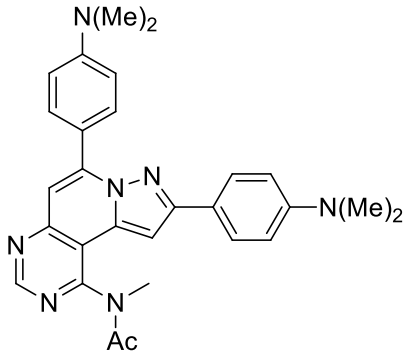


Figure 1.17: Pyrazolo[1,5-*a*]pyridinefused pyrimidine molecule used for studying lipid droplets in living cells.

Pyrazolopyridine annulated heterocycles were synthesized and the effect of substituents on their photophysical properties was studied by Toche and co-workers. The work revealed interesting substituents dependent fluorescence properties of pyrazolopyridine which can be used as NLO materials **(Figure 1.18)**.[17]

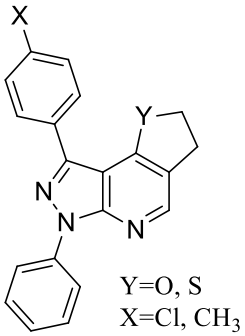


Figure 1.18: Pyrazolopyridine annulated heterocycles studied for photophysical properties.

Fused pyridine systems have also been investigated for selective ion sensing. Cheng and co-workers reported one-pot reaction for the synthesis of 2-carbonyl-3-(pyridylamino)imidazo-[1,2-*a*]pyridines. Among the synthesized molecules, 1-(6-chloro-3-(5-chloropyridin-2-ylamino)imidazo[1,2-*a*]pyridin-2-yl)-2-ethylbutan-1-one was found to be selective

fluorescent sensor for mercury ion both in aqueous as well as organic (acetonitrile) media **(Figure 1.19)**.[18]

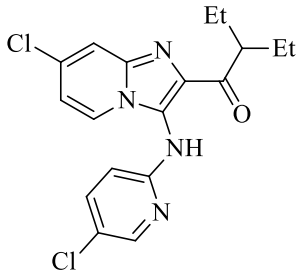
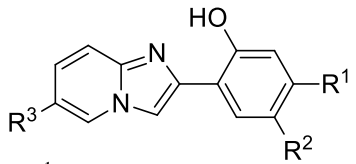


Figure 1.19: Selective Hg^{2+} sensing by1-(6-chloro-3-(5-chloropyridin-2-ylamino)imidazo-[1,2-*a*]pyridin-2-yl)-2-ethylbutan-1-one.

Synthesis and fluorescence properties of diverse 2-(2′-hydroxyphenyl)imidazo[1,2-*a*]pyridines were studies by Mutai *et al*. Faint dual emission ($\Phi \approx 0.01$) due to the normal and excited-state intramolecular proton transfer (ESIPT) fluorescence was shown by all the compounds in solution phase **(Figure 1.20)**. However in poly(methylmetharylate) matrix efficient ESIPT fluorescence (Φ up to 0.6) was displayed by the same compounds. It was noticed that the presence of electron-donating and electron-withdrawing groups in the phenyl ring caused blue and red shifts of the ESIPT fluorescence emission band, respectively. When same substituents were introduced in theimidazopyridinehalf, results in fluorescence shifted in the opposite directions.[19]



R^1=H, OCH_3
R^2=H, OCH_3, CH_3
R^3=H, Cl, Br, F, CH_3, OCH_3, CF_3, CN

Figure 1.20: ESIPT fluorescence displayed by various 2-(2′-hydroxyphenyl)imidazo[1,2-*a*]pyridines in poly(methylmetharylate) matrix.

1.5. Objectives

The main objective of this thesis is to develop newer methods for the synthesis of diverse fused pyridine/dihydropyridine compounds.

Chapter II: Development of a simple route for the synthesis of biologically important fused δ-lactam systems by application of one pot two step Nazarov-Schmidt rearrangement.

Chapter III: Introduction of potent groups at 4^{th} position of imidazo[4,5-*c*]quinolines *via* aldimine formation followed by 6-*endo*-*trig* cyclization and *in*-*situ* oxidation. anticancer activity evaluation of the resulting compounds.

Chapter IV: $FeCl_3$ catalyzed synthesis of chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one by using 7-amino indole, 4-hydroxy coumarin and various aldehydes. Screening of anticancer activity of the synthesized compounds.

1.6. Material and Methods

All starting materials were purchased from Aldrich, Alfa Aesar, Acros, Spectrochem, SRL, AVRA and Sd Fine (India) and used directly without further purification. Solvents were dried using standard methods and distilled before use. Visualization on TLC was achieved using of UV light (254 nm) or iodine. Melting points were recorded on a Stuart SMP 30 melting point apparatus. ^{1}H NMR (300 MHz and 400 MHz) and ^{13}C (75 MHz and 101 MHz) spectra were recorded in $CDCl_3$ and DMSO solution with TMS as internal standard. IR spectra were recorded as KBr plates on Jasco FT/IR-4200 spectrometer. Mass spectra were recorded on Shimadzu LCMS-2020.High resolution mass spectra were recorded on Bruker microTOF and Agilent 1100/LC MSD Trap SL version QII instrument. Column chromatography was performed on silica gel (100–200 mesh, SRL. India).

Cell line culture and drug treatment (done in collaboration with Dr. Balaram Ghosh, Department of Pharmacy, BITS-Pilani Hyderabad campus)

B16F10 (murine melanoma) cells were cultured in Dulbecco's Modified Eagle Medium (DMEM) (Himedia Laboratories Pvt. Ltd., Mumbai, India). Culture medium was supplemented with 10% heat inactivated fetal bovine serum (Himedia Laboratories Pvt. Ltd., Mumbai, India) and 1 % of antibiotic solution (10 U Penicillin and 10 mg Streptomycin per ml, Himedia Laboratories Pvt. Ltd., Mumbai, India). Cells were cultured at 37°C in humidified atmosphere with 5% CO_2. Stock solutions of all the synthesized compounds were prepared in DMSO at a concentration of 10 mM and stored.

1.7. References

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CHAPTER 2

One pot two step Nazarov-Schmidt rearrangement for the synthesis of fused δ-lactam systems

2.1. Introduction

Inherent stability of amide linkages and their propensity to exist in diverse molecules, gives lactam rings a privileged position in the domain of medicinal chemistry. Some of the most sought after drug molecules containing this stable linkage includes β-lactam antibiotics: penicillin, cephalosporin, monobactam and carbapenem. While β-lactam compounds gets the maximum coverage owing to their diverse biological role, increasing attention is garnered by molecules bearing δ-lactam rings.[1-4] Imming *et al*., in their work on comparative analysis of various lactams, concluded δ-lactam to be as effective as β-lactam in serine proteinase inhibition and thus a promising candidate for development of new antibiotics.[5] It has also been noticed that lactams are most effective as drug candidates when they are fused with other heterocyclic units. For example, in penicillin and cephalosporin class of antibiotics, β-lactam is fused with thiazolidine and 1,3-thiazinane ring, respectively. Some of the well-known fused δ-lactam compounds are dihydroquinolin-2-one alkaloids **(Figure 2.1)** such as Excavatine B (**1**), Megistoguinone II (**2**), Meloscine (**3**), Yaequinone (**4**), etc., known for their myriad biological activities.[6-8]

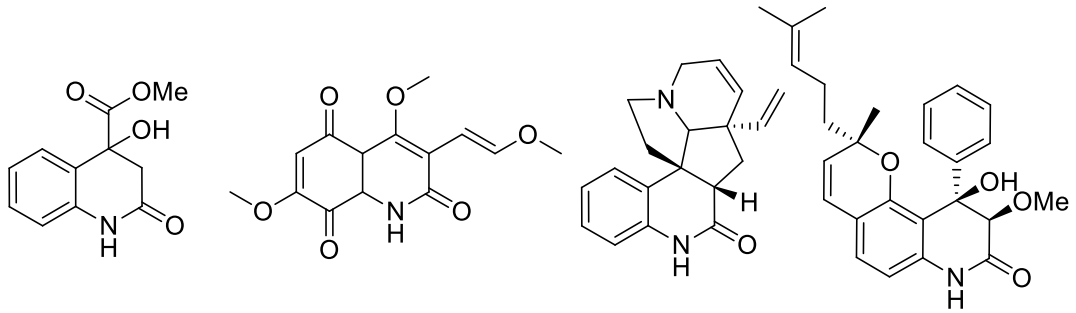
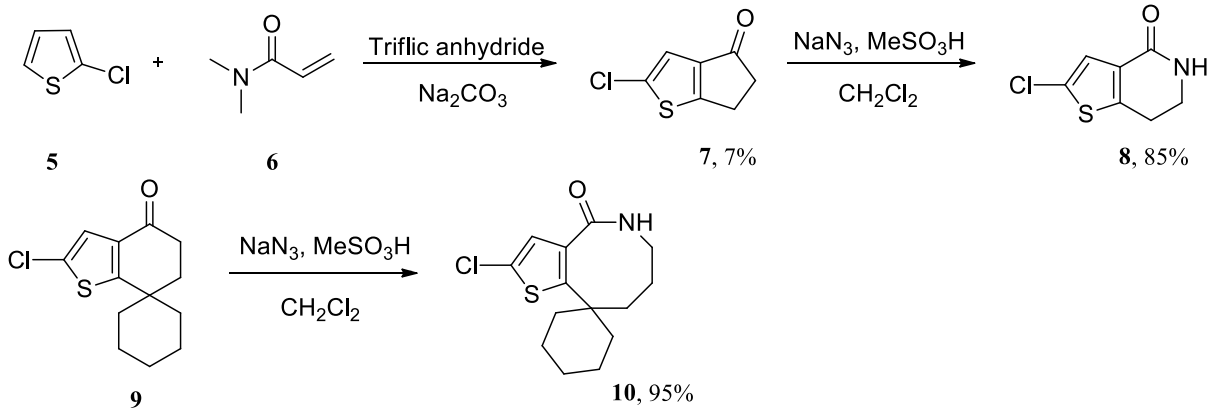


Figure 2.1: Well-known fused δ-lactam compounds are dihydroquinolin-2-one alkaloids.

In continuation with the on-going research in our laboratory in the field of fused aromatic systems, we wanted to explore synthesis of fused thiophene-δ-lactam compounds. It was articulated that bringing together biologically important scaffolds thiophene and δ-lactam

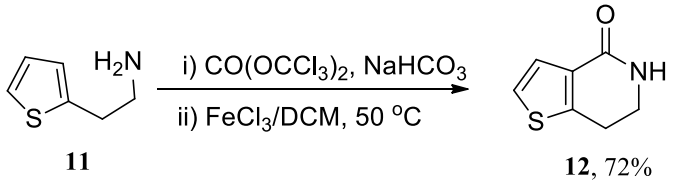
will bestow the resulting structure with both molecular rigidity as well as flexibility. Additionally, prospect of diverse biological properties was also assumed to be a reasonable outcome. While several approaches are known for the synthesis of these molecules, the need to fuse aromatic and non-aromatic cyclic framework makes them long and tedious.[9-17]

Lindvall *et al*., reported two step synthesis of the dihydrothieno[3,2-*c*]pyridin-4(5*H*)-ones using chlorothiophene **(5)** and *N,N'*-dimethylacrylamide **(6)** as the starting compounds. Initial reaction between **5** and **6** in the presence of triflic anhydride yielded 2-chloro-5,6-dihydro-4*H*-cyclopenta[*b*]thiophen-4-one **(7)**, 2-chloro-5,6-dihydro-4*H*-spiro[benzo[b]thiophene-7,1'-cyclohexan]-4-one **(9)**, which was converted to the final molecule **(8, 10)** by treatment with sodium azide (**Scheme 2.1**).[10]



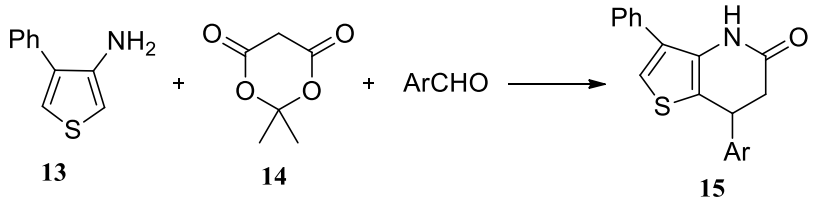
Scheme 2.1: Synthesis of dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one system by Lindvall *et al*.

George *et al*., reported synthesis of the 6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one **(12)** by using carboxylation of 2-(thiophen-2-yl)ethanamine (**11**) as the first step followed by cyclisation in the presence of $FeCl_3$ (**Scheme 2.2**).[11]



Scheme 2.2: Synthesis of dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one system by George *et al*.

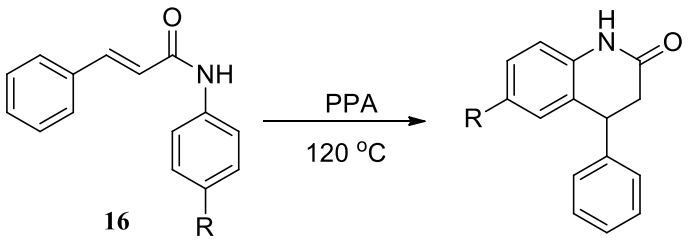
During their studies on fused thieno-dihydropyridone systems, Lichitsky *et al*., reported synthesis of substituted 6,7-dihydro-4*H*-thieno[3,2-*b*]pyridine-5-onemolecules. The synthesis involved Michael addition of 3-amino-thiophene (**13**) to arylmethylene derivative of Meldrum's acid (**14**) followed by the intramolecular cyclization. Final molecules were obtained by elimination of CO_2 and acetone **(Scheme 2.3)**.[12]



15(Ar=2-OEt-C_6H_4, 4-OMe-C_6H_4, 3-OMe-C_6H_4, 4-Cl-C_6H_4, Thiophen-2-yl, Thiophen-3-yl)
71%, 68%, 40%, 65% 51%, 62%

Scheme 2.3: Synthesis of 7-phenyl-6,7-dihydrothieno[3,2-*b*]pyridin-5(4*H*)-one system by Lichitsky *et al*.

Literature reports also show synthesis of fused phenyl δ-lactam systems. Conley *et al*., showed single step conversion of *N*-phenylcinnamide to give 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one. The reaction was carried out using polyphosphoric acid at 120 °C **(Scheme 2.4)**.[13]

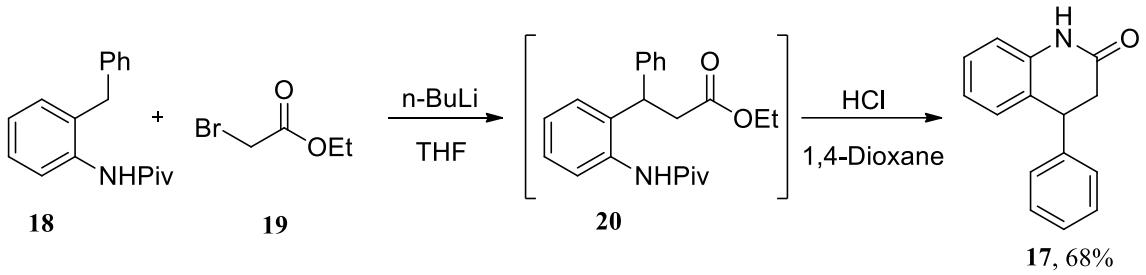


17 (R=H, CH_3, OCH_3, Br)
PPA = Polyphosphoric Acid 94%, 83%, 90%, 83%

Scheme 2.4: Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one system by Conley *et al*.

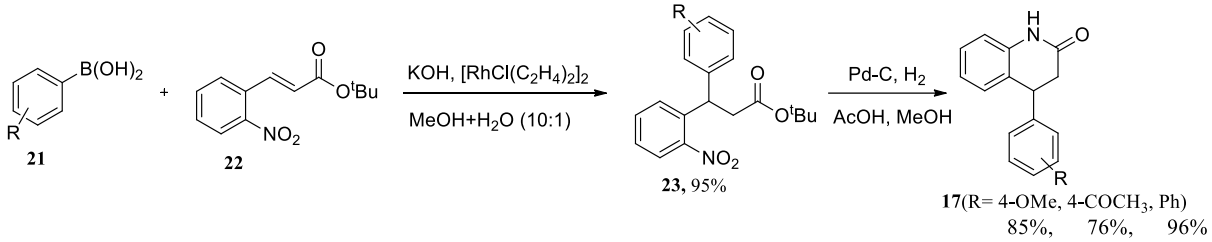
Kim *et al*., reported a synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one **(17)** using *N*-pivaloyl aniline **(18)** and α-bromoethyl acetate **(19)**. Starting materials **18** and **19** were

converted to ethyl 3-phenyl-3-(2-pivalamidophenyl)propanoate **(20)** in the presence of *n*-BuLi. Subsequently, intermediate **20** was treated with HCl to generate the final molecule **17 (Scheme 2.5)**.[14]



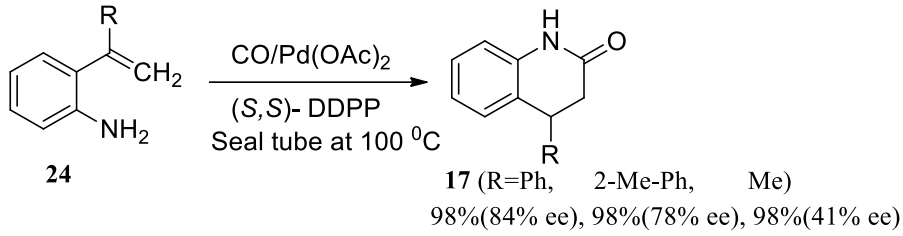
Scheme 2.5: Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one system by Kim *et al*.

Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one **(17)** was reported by Paquin *et al*., in two steps **(Scheme 2.6)**.[15] The first step involved 1,4-addition of aryl boric acid **(22)** to α,β-unsaturated ester **(21)** by using a rhodium catalyst, $[RhCl(C_2H_4)]_2$ under basic condition. The compound obtained was subjected to reduction of its existing nitro functional group, followed by intramolecular amide bond formation to give the target molecule.



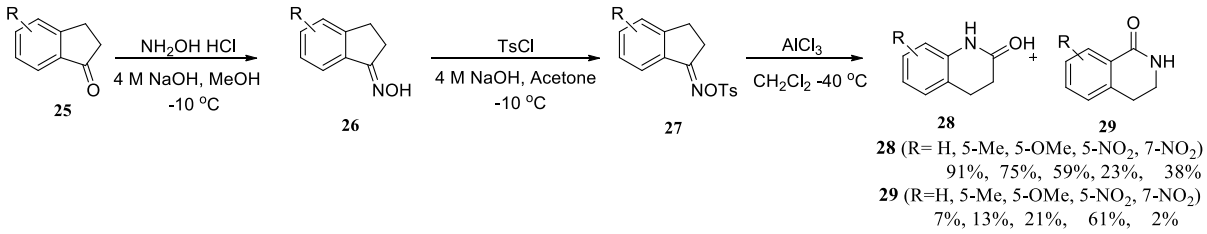
Scheme 2.6: Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one system by Paquin *et al*.

Dong and Alper reported the enantioselective cyclocarbonylation of various 2-vinylanilines **(24)** for the synthesis of diverse 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (**Scheme 2.7**).[16] The reaction was catalysed by $Pd(OAc)_2$/(2*S*, 4*S*)-(-)-4-diphenylphosphino-2-(diphenylphosphinomethyl)pyrrolindine was carried out under an atmosphere of CO (500 psi) and H_2 (100 psi) at 100 °C in sealed tube for 48 hours.



Scheme 2.7: Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one system by Dong *et al*.

In a report by Lee *et al*., Beckmann rearrangement was used as the key step for the final synthesis of various 3,4-dihydroquinolin-2(1*H*)-one molecules **(Scheme 2.8)**.[17] The synthesis commenced from substituted 1-indanones **(25)** which were converted to corresponding oximes **(26)** by treatment with hydroxyl amine. Further, oximes were converted to respective tosyloximes **(27)** under basic condition using NaOH. In the final step, $AlCl_3$ was used to carry out Beckmann rearrangement to yield the target *N*-aryl amides.

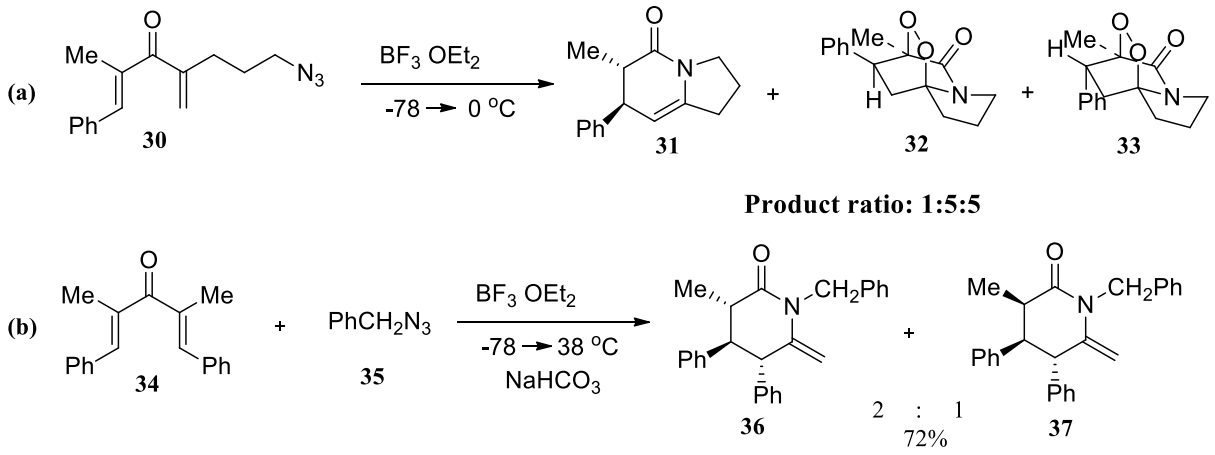


Scheme 2.8: Synthesis of 4-phenyl-3,4-dihydroquinolin-2(1*H*)-one system by Lee *et al*.

Use of one-pot reaction sequences have emerged as an important area of research in organic chemistry. This approach allows use of simple chemical compounds to generate relatively complex products and also proves environmentally beneficial by moderating the use of chemicals. Nazarov and Schmidt reactions are well explored name reactions used for the synthesis of cyclopentanone and amide derivatives, respectively.[18-26] While lot of papers on their one-pot/tandem/domino reactions are known,[27-36] very few literature reports have shown their mutual one-pot execution for organic synthesis.[37-39] During their first attempt towards domino Nazarov-Schmidt rearrangement, West and co-workers reported formation

of target molecule **31** only in trace amounts **(Scheme 2.9a)**.[37] Major products obtained were diastereomeric endoperoxides **32** and **34**. However their subsequent attempt with simple 1,4-dien-3-ones **(34)** and various azides **(35)** resulted in formation of 3,4-dihydropyridin-2-ones **(36, 37)** in moderate to good yields **(Scheme 2.9b). (**Only one representative example is shown in the Scheme**)**.[38]

Though the devised method was successful, it was never extended to the synthesis of fused aromatic/non-aromatic frameworks.

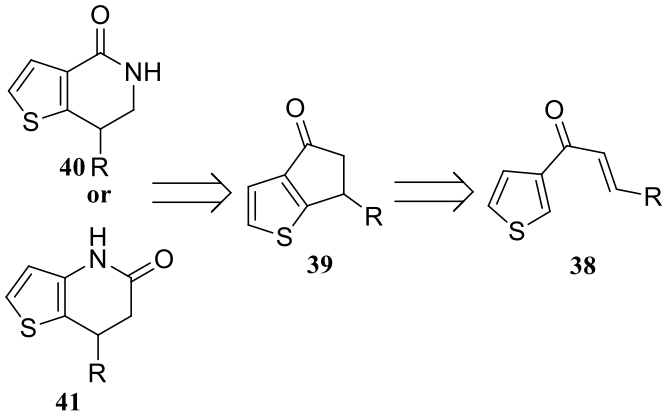


Scheme 2.9 (a and **b):** Domino Nazarov-Schmidt rearrangement by West and co-workers, *en-route* to dihydropyridones.

2.2. Results and Discussion

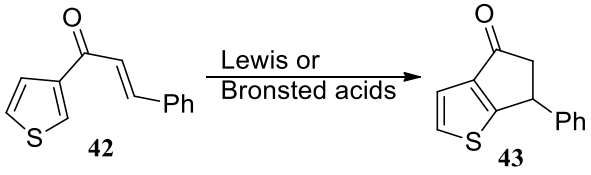
The synthesis of the target molecules was planned based on retrosynthetic strategy shown in **Scheme 2.10**. Here compound **39** was envisaged as the intermediate, *en-route* to the final compounds **40** and/or **41**. Synthesis of **39** was planned by execution of Nazarov reaction. It was articulated that introduction of azide ion, after formation of compound **39** will allow the resulting intermediate to undergo Schmidt rearrangement and would yield the target molecules. Given the common requirement of acidic conditions in both the reactions,

introduction of azide nucleophile prior to isolation of the fused cyclopentanone adduct, would allow development of a one-pot strategy.



Scheme 2.10: Retrosynthetic strategy for synthesis of fused thieno *δ*-lactam systems.

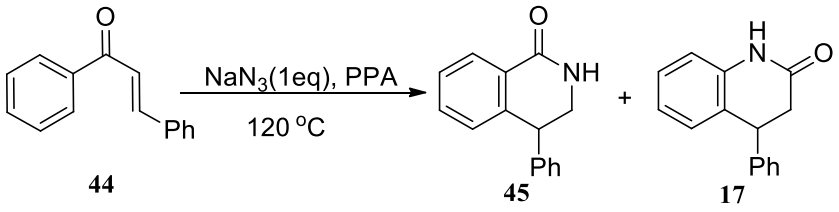
The devised synthetic strategy was initiated with the preparation of thiophene substituted α,β-unsaturated ketones **(38)**.[40-41] The chalcones thus obtained were purified and thoroughly characterized, before attempting any further reactions. In order to develop the appropriate conditions for the synthesis of target molecules, 3-phenyl-1-(thiophen-3-yl)prop-2-en-1-one **(42)** was chosen as the model substrate. Initially, Nazarov reaction was attempted using already established literature protocols **(Scheme 2.11)**.[42-44] Investigations were carried out with acetic acid (neat)/formic acid (neat)/triflic acid (in $Cl(CH_2)_2Cl$, 1,4-dioxane and toluene) /Eaton's reagent (in toluene)/*p*-toluenesulphonic acid (in toluene)/polyphosphoric acid (neat)/$FeCl_3$ (in 1,4-dioxane)/$AlCl_3$ [in 1,4-dioxane] at 120 ^oC, in sealed tubes. Formation of compound **43** was noticed, only in case of $FeCl_3$, triflic acid and PPA. It was isolated and subjected to spectroscopic measurements, for characterization of structure.



Scheme 2.11: Synthesis of compound **43**

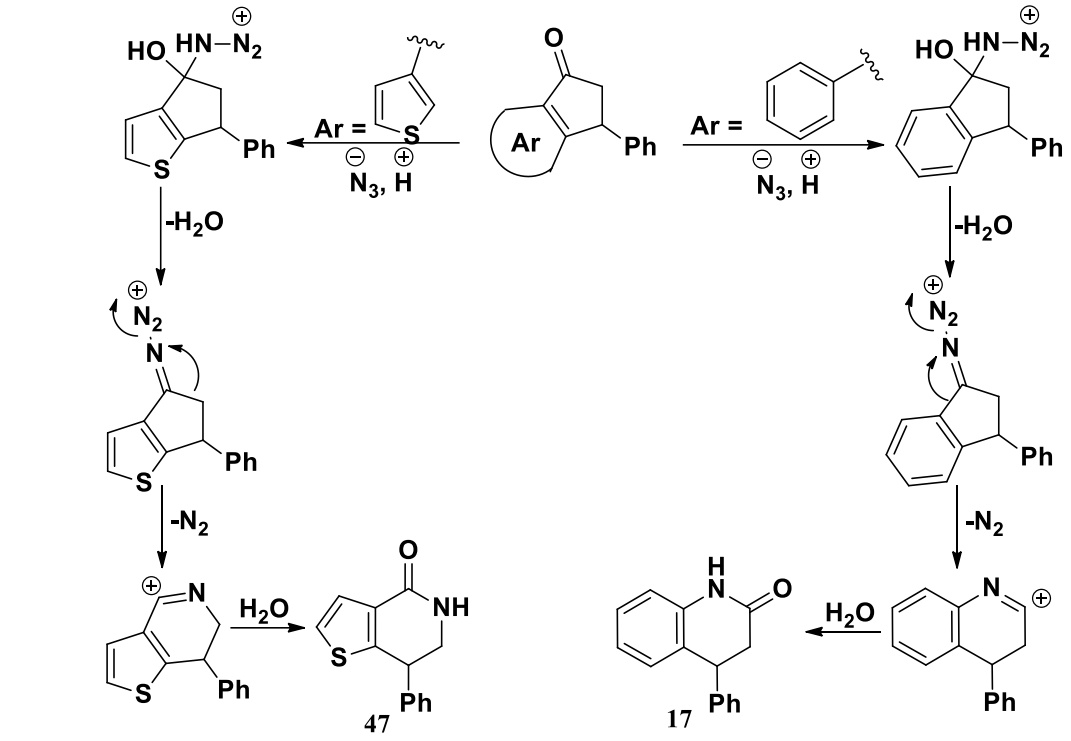
With the conditions for Nazarov reaction in hand, we embarked on development of an appropriate method for its one pot execution along with Schmidt rearrangement **(Table 2.1)**. Our attempt involved synthesis of fused thiophenecyclopentanone intermediate and then addition of sodium azide. The entire process was designed to be carried out without separation of **43**. Reaction temperature kept at 120 ^{o}C, based on the conditions established for synthesis of intermediate **43**. Final product was not obtained when $FeCl_3$ and triflic acid (in 1,4-dioxane and 120 °C) were used, even after allowing the reaction mixture to stir for 24 hours. When PPA was used both as a catalyst and solvent, reaction was complete after 8 hours (extra 6 hours required for the completion of Nazarov reaction) and only *N*-alkyl amide **47** was obtained in 40% yield. The most important evidence for the formation of **47** was provided by ^{1}H NMR spectroscopy, NH peak for *N*-alkyl amide is generally seen at δ ~6-7 ppm,[45] whereas for *N*-aryl amide product, the same peak is seen at δ ~ 8-10 ppm.[46] In case of **46**, NH peak was observed at δ 6.07 ppm, indicating formation of *N*-alkyl compound. Additionally, ^{13}C NMR spectrum showed amide carbonyl at δ 163.5 ppm. According to IR data 1679 cm^{-1} indicated that amide bond. Result obtained was surprising, as very few literature reports indicate alkyl migration compared to aryl migration, in case of Schmidt rearrangement.[47-50, 10]

This unusual result prompted us to use the same reaction condition on benzylideneacetophenone **(44) (Scheme 2.12)**, which yielded the usual *N*-aryl amide product **(17)**.[51-53] Structure of compound **17** was established by ^{1}H and ^{13}C NMR spectroscopy. In ^{1}H NMR spectrum NH peak of **17** was observed at δ 8.64 ppm, indicating the formation of *N*-aryl amide system [12, 17, 54] and the amide carbonyl was shown by ^{13}C NMR at δ 170.7 ppm. IR data tells that amide bond stretching value 1675 cm^{-1}.



Scheme 2.12: Synthesis of compound **17**

Based on these preliminary studies, a plausible mechanism was proposed for the formation of both *N*-alkyl/*N*-aryl amide systems **(Scheme 2.13)**. The unusual regioselectivity observed in case of thiophene system can be explained in terms stability of putative transition state **A** in comparison to the more strained transition state **B (Figure 2.2)**. To support the proposed mechanism **(Scheme 2.13)** and the transition states **(A-C)**, we have performed density functional theory (DFT) calculations with major focus on the ring expansion pathway. This step leads to the formation of the *N*-alkyl or *N*-aryl amides cation and concomitant removal of a N_2 molecule. The vibration along the N···N_2 bond were critically considered to determine the true transition states for the ring expansion step. Interestingly, we could only optimize the transition states **A** and **C** through DFT calculations; but the transition state **B** failed to get optimized in the proposed structure even after several attempts using higher basis sets and different hybrid density functionals. We assume that the structural instability of the proposed transition state **B** arises due to strained formation of three fused rings in the thiophene system which leads to the observed divergence in the DFT calculations. Additionally, inefficient 2p-3p overlap which is responsible for π-electron cloud delocalization in thiophene may also affect the migration tendency. Such a scenario most likely does not emerge in case of phenyl system, where phenonium cation in the transition state **C (Figure 2.2)** is more effective in delocalizing the positive charge.[55]



Scheme-2.13: Plausible mechanism for one pot two step Nazarov-Schmidt reaction.

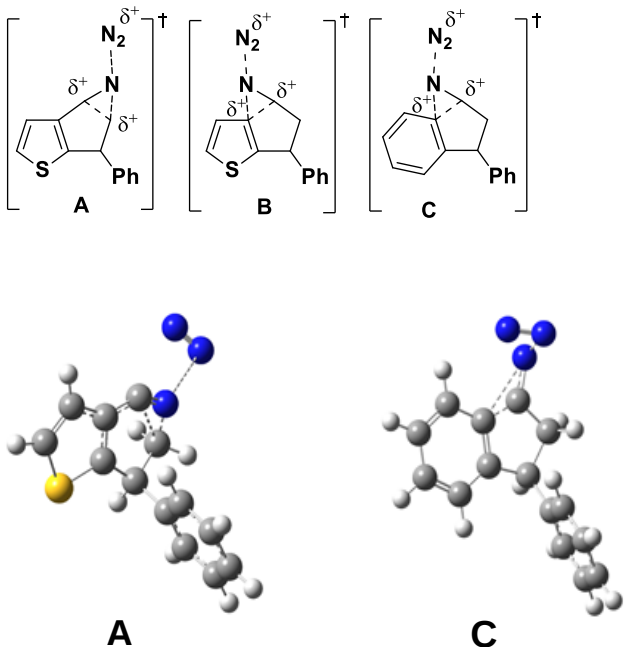
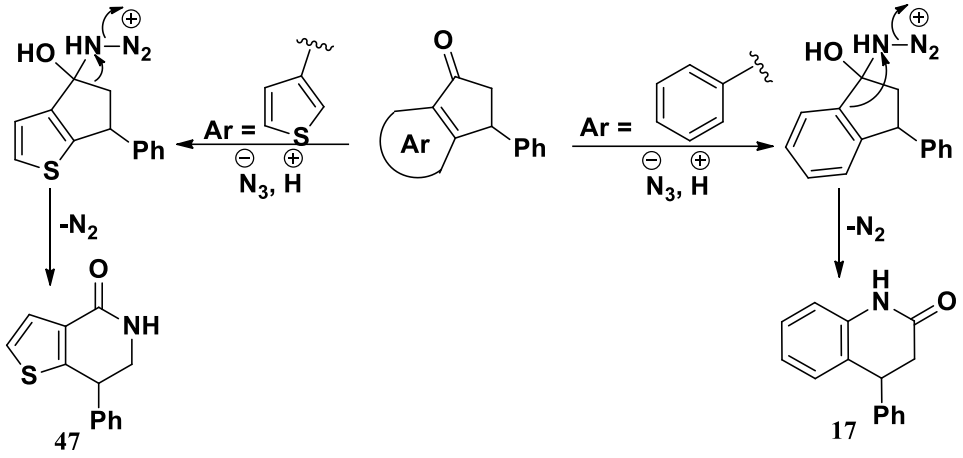


Figure 2.2: Structures of plausible transition states for ring expansion and N_2 removal step. (color coding: N: blue, S: yellow, C: grey, H: white); M06/6-31+G (d,p) basis set level of theory and QST3 method.

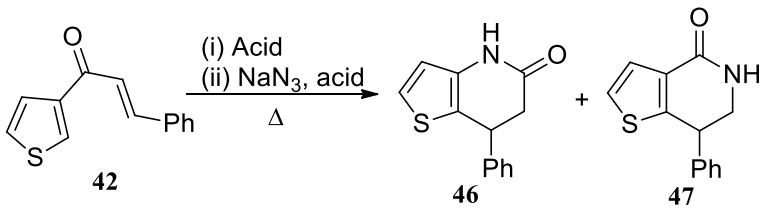
An alternative mechanism based on Baeyer-Villiger reaction is also possible, where azidohydrin directly converts to the final molecule **(Scheme 2.14)**.[56] Nonetheless, it is difficult to give a satisfactory explanation for the opposite migration tendencies of thiophene and phenyl systems.



Scheme 2.14: Alternative mechanism based on Baeyer-Villiger reaction.

Further, studies were carried out to optimize the reaction conditions. Reactions carried out by varying the amount of sodium azide **(entry 4 and 5)**, resulted in the formation of **47** in 65% and 63% yields, respectively. Increasing or decreasing the reaction temperature by 10 ^{o}C **(entry 6 and 7)**, did not impact the overall reaction, as the final yields obtained were 62% and 65%, respectively. On attempting the reaction in open tube **(entry 8)**, final yield was decreased to 20%. Based on the screening results, reaction in PPA at 120 °C with two equivalents of NaN_3 in sealed tube was chosen as the preferred condition for implementation of the designed protocol.

Table 2.1: Optimization of the reaction conditions



Entry	NaN_3	Acid	Temp	Yield[a](%)	
				47	**46**
1	1 eq	Triflicacid[b]	120^o C	0	0
2	1 eq	PPA[c]	120^o C	40	0
3	1 eq	$FeCl_3$(30%)[b]	120^o C	0	0
4	**2 eq**	**PPA**	**120^o C**	**65**	**0**
5	3 eq	PPA	120^o C	63	0
6	2 eq	PPA	110^o C	62	0
7	2 eq	PPA	130^o C	65	0
8	2 eq	PPA[d]	120^o C	20	0

[a] Yield of isolated product; [b] Only compound **43** was obtained;

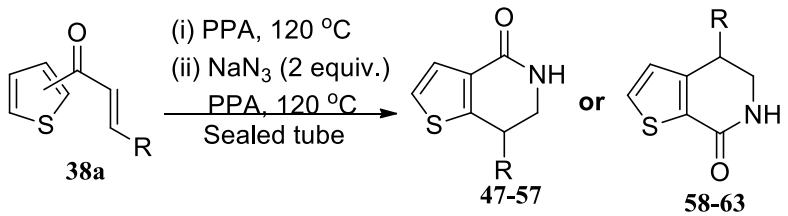
[c] polyphosphoric acid; [d] reaction was carried out in an open tube

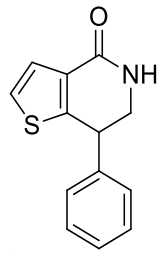
Using the optimized conditions **(Table 2.1; entry 4)**, reactions were carried out on various 3-aryl substituted-1-(thiophen-3-yl)prop-2-en-1-one and 2-aryl substituted-1-(thiophen-3-yl)prop-2-en-1-one derivatives **(Table 2.2)**. Our initial attempt was to look at the effect of benzene rings bearing electron releasing and electron withdrawing groups, on the overall yield of the reaction **(47-63)**. As expected presence of electron releasing group **(48)** decreased the yield of the reaction whereas no significant difference was noticed from the completely unsubstituted counterpart **(47)**, when nitro (*o*-/*p*-) group was present **(49-50)**. Further, reactions were carried out with molecules bearing halogens as substituents on the benzene ring **(51-54)**. Surprisingly, yields were comparatively less than the unsubstituted counterpart for molecules bearing fluorine atom **(51-52)**, whereas yields were moderately impacted by the presence of chlorine or bromine as substituents **(53-54)**. Attempts were also made to study the impact of di-substituted benzene ring on the overall yield **(55-57)**. While yields did not change substantially for molecules bearing *o*-, *p*- or *o*-, *o′*- di-substituted systems **(55 & 57)**, the presence of substituents on *ortho* and *meta* position of the benzene ring reduced the

overall yield of the reaction to 52% **(56)**. Reaction with 3-alkyl substituted-1-(thiophen-3-yl)prop-2-en-1-one compounds were not successful, despite numerous attempts and in all the cases starting molecules were degraded under the optimized conditions.

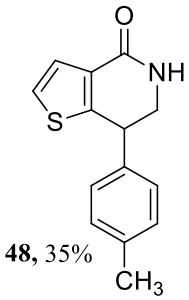
Our initial success with 3-acetylthiophene system prompted us to apply the established method for thiophene substituted α,β-unsaturated ketones, prepared from aromatic aldehydes and 2-acetylthiophene. Initial attempt was made to synthesize 4-phenyl-4*H*-cyclopenta[b]thiophen-6(5*H*)-one, to check the applicability of Nazarov reaction on 2-substituted thiophene system. After the desired compound was isolated and characterized, further attempts were made to apply the established one-pot two step protocol on diverse 3-aryl-1-(thiophen-2-yl)prop-2-en-1-one compounds. Gratifyingly, all the compounds screened generated the final *N*-alkyl amide products in 44-61% yields **(Table 2.2; entry 58-63)**. Comparatively low yield here can be justified on the basis of less reactivity of C-3 position of thiophene ring towards electrophilic substitution reaction. Attempts to react 2-aryl substituted-1-(thiophen-3-yl)prop-2-en-1-one compounds bearing electron releasing groups on phenyl ring, were not successful as the initial Nazarov reaction failed to generate fused thiophene-cyclopentanone compound.

Table 2.2: Synthesis of 7-aryl substituted 6,7-dihydrothieno[3,2-*c*]pyridine-4(5*H*)-one and 4-aryl substituted 5, 6-dihydrothieno[2,3-*c*]pyridine-7(4*H*)-one [a]

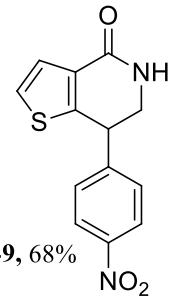




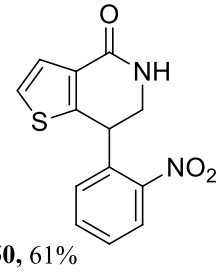
47, 65%



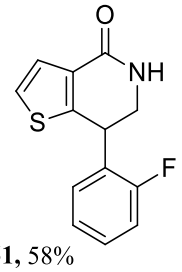
48, 35%



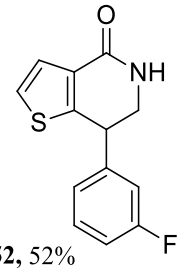
49, 68%



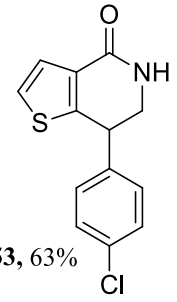
50, 61%



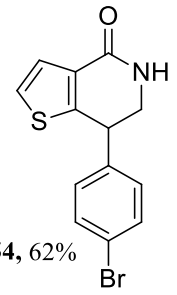
51, 58%



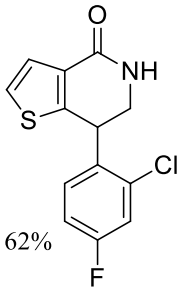
52, 52%



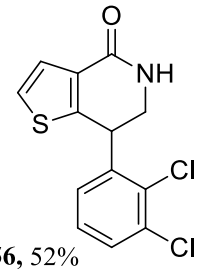
53, 63%



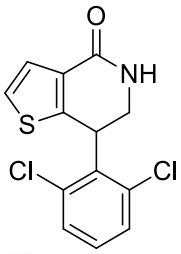
54, 62%



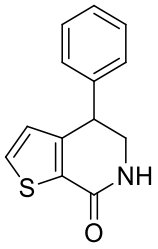
55, 62%



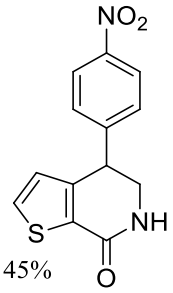
56, 52%



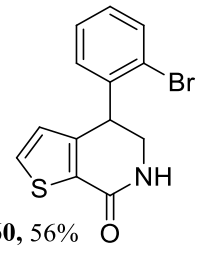
57, 62%



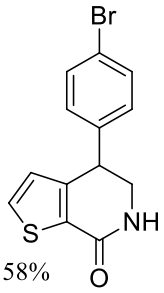
58, 44%



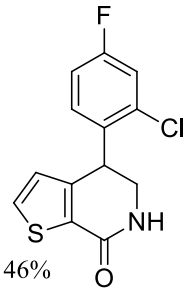
59, 45%



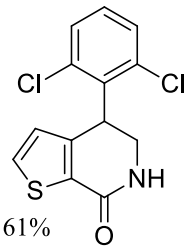
60, 56%



61, 58%



62, 46%



63, 61%

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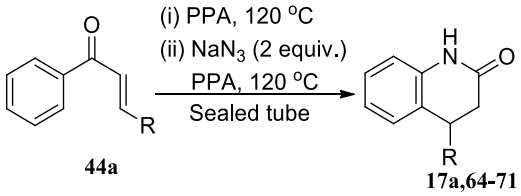
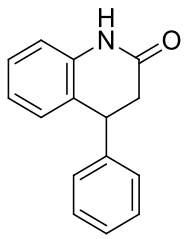
[a] Reaction condition: 0.46 μmol of **38a**, 0.9 μmol of NaN_3 and 1 mL PPA.

Our success with various 2/3-substituted thiophene α,β-unsaturated ketones and the outcome of **Scheme 2.13** inspired us to further explore the reactions with phenyl substituted substrates **(Table 2.3)**. As preliminary results clearly indicated preference for aryl migration over alkyl migration by phenyl rings, it was felt that this reaction can be used for synthesis of various dihydroquinolinones. Accordingly, several chalcones were synthesized using well established literature procedure and subjected to reaction under the optimized conditions. While diverse chalcones, bearing electron releasing and donating groups were screened under the reaction condition, only halogen substituted substrates resulted in the formation of expected *N*-aryl amide products **(17, 64-71)**. Among the compounds screened, best results were obtained with substrates bearing 2,4 dichloro, and 2-chloro,4-fluro substituted benzene ring **(67-68)**.

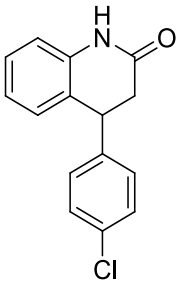
Table 2.3: Synthesis of 4-aryl substituted 3,4-dihydroquinolin-2(1*H*)-one [a]



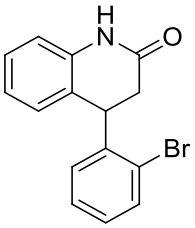
44a **17a,64-71**



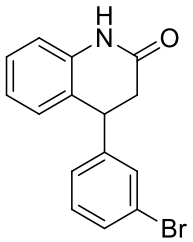
17, 56%



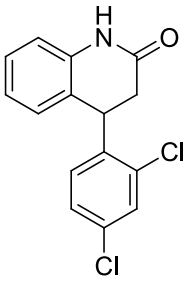
64, 48%



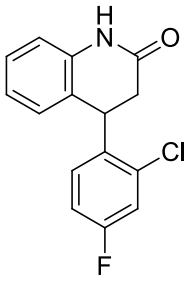
65, 43%



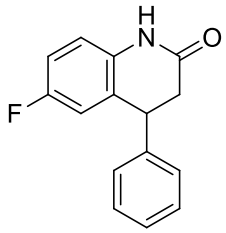
66, 49%



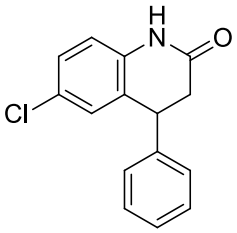
67, 64%



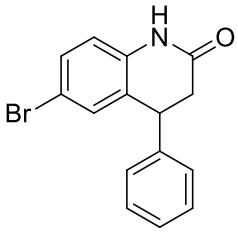
68, 62%



69, 60%



70, 58%



71, 59%

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[a] Reaction condition: 0.46 μmol of **44a**, 0.9 μmol of NaN_3 and 1 mL PPA.

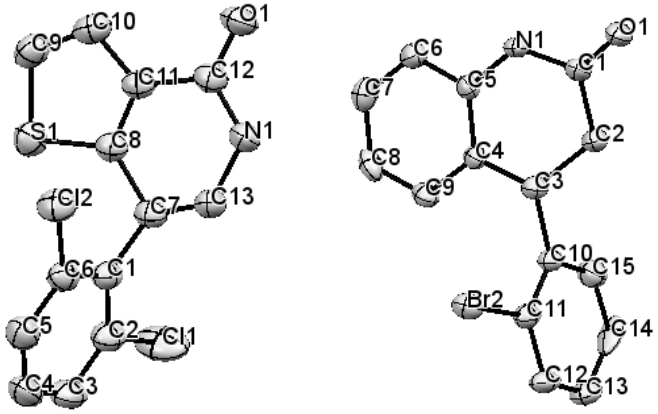
Single crystals of $C_{13}H_9Cl_2NOS$ **(57)**, $C_{15}H_{12}BrNO$ **(65)**, were detected by X-Ray crystallography. Suitable crystals were selected on Xta LAB Pro: Kappa dual offset/far diffractometer. The crystals for compound **57** and 6**5** were kept at 298.15 K and 293(2) K, during data collection. Using Olex2,[57] the structure was solved with the ShelXS structure solution program using direct methods and refined with the ShelXL refinement package using Least Squares minimization.[58-60] Selected crystallographic data and structure refinement are shown in **Table 2.4** (full details appears as tables in the supplementary information) and molecular structures of compounds **57** and 6**5** are shown in **Figure 2.3** as ORTEP diagrams. It further confirmed the structure of both *N*-alkyl/*N*-aryl amides determined previously by ^{1}H-NMR spectroscopy.

Table 2.4: Crystallographic information and structure refinement for **57** and 6**5**

	57	**65**
Empirical formula	$C_{13}H_9Cl_2NOS$	$C_{15}H_{12}BrNO$

Formula weight	298.17	302.17
Temperature	298.15K	293(2) K
Wavelength	0.71073 Å	0.71073 Å
Crystal system	Triclinic	Orthorhombic
Space group	P-1	C2/c
Unit cell dimensions	a=6.7476(6)Å, α=93.6960(10)° b=8.1541(7)Å, β=102.4670(10)° c=13.1499(12)Å, γ=114.0080(10)°	a=23.962(2)Å, α = 90°.00 b=5.0912(6) Å, β= 107.100(11)° c=21.337(2)Å, γ = 90°.00
Volume	635.86(10) Å^3	2488.0(5) Å^3
Z	2	8
Density (calculated)	1.557 Mg/m^3	1.613 Mg/m^3
Absorption coefficient	0.659 mm^{-1}	3.290 mm^{-1}
F(000)	304.0	1216.0
Theta range for data collection	3.22 to 51.974 °	10.146 to 49.97 °
Index ranges	-8 ≤ h ≤ 8, -10 ≤ k ≤ 10, -16 ≤ l ≤ 16	-26 ≤ h ≤ 28, -5 ≤ k ≤ 6, -25 ≤ l ≤ 24
Reflections collected	6415	7456
Independent reflections	2463 [Rint = 0.0217, Rsigma = 0.0223]	2155 [Rint = 0.0214, Rsigma = 0.0192]
Data/restraints/parameters	2155/0/163	2155/0/163
Goodness-of-fit on F^2	1.145	1.041

Final R indices I>2sigma(I)	R1 = 0.0411, wR2 = 0.1343	R1 = 0.0330, wR2 = 0.0839
R indices (all data)	R1 = 0.0462, wR2 = 0.1403	R1 = 0.0379, wR2 = 0.0865
Largest diff. peak and hole	0.41/-0.44e.Å^{-3}	1.01/-0.35 e. Å^{-3}



57 **65**

Figure 2.3: Representation of the molecular structure of compounds **57** and **65**. Displacement ellipsoids are drawn with 50% probability and hydrogen atoms are omitted for clarity.

While the overall method looks limited in terms of the substrate scope, as halogen substituted compounds are more preferred. Given the fact that aryl halides are favoured substrates in the ubiquitous cross coupling reactions; the synthesized halogenated systems will enable easy access to diverse fused δ-lactam systems.

2.3. Conclusions

In conclusion, we have developed a simple one pot two step synthetic route to fused δ-lactam compounds by consecutive application of Nazarov and Schmidt rearrangement reactions. Initial studies with various 3-aryl substituted-1-(thiophen-3-yl)prop-2-en-1-

one and 3-aryl substituted-1-(thiophen-2-yl)prop-2-en-1-one as substrates, resulted in the formation of unusual thiophene fused *N*-alkyl amide compounds in modest to good yields. With 3-aryl substituted phenylprop-2-en-1-one as substrates under the optimized conditions, *N*-aryl amide compounds were formed as the final products instead of corresponding *N*-alkyl analogues. These results also indicate the importance of strained versus relaxed transition states and *p*-orbital overlaps in controlling the aryl migratory tendencies in Schmidt rearrangement reactions. Further work is currently under progress in our laboratory for the development of pyridine and heteroaryl versions of the fused δ-lactam molecules, using the developed methodology.

2.4. Experimental

General information

All starting materials were purchased from various chemical manufacturers and were used directly. Solvents were dried and distilled using standard methods, before use. Column chromatography was performed on silica gel (100–200 mesh, SRL. India). Visualization on TLC was achieved by use of UV light (254 nm) or iodine. ^{1}H NMR (300MHz and 400 MHz) and ^{13}C (75 MHz and 101 MHz) spectra were recorded in $CDCl_3$ and DMSO solution with TMS as internal standard. IR spectra were recorded as KBr plates on Jasco FT/IR-4200 instrument. Melting points were recorded on a Stuart SMP 30 melting point apparatus and are uncorrected. Mass spectra were recorded on Shimadzu LCMS-2020.

2.5. Procedure for the preparation of the compounds

General method for synthesis of 3-aryl-1-(thiophen-3-yl)prop-2-en-1-one (38a)

To a mixture of 1-(thiophen-3-yl)ethanone (0.7 mmol) and aldehyde (0.77 mmol) in 1.5 mL of ethanol, was added 2 mL NaOH (20 mol%). The solution was stirred at 30 ^{o}C for 30 minutes under nitrogen atmosphere. On the completion of the reaction as indicated by the TLC, the reaction mixture was poured into ice cold water. The

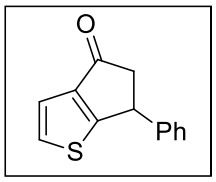
precipitate thus formed, was filtered and used directly for the next step without any purification.

Method for synthesis of 6-phenyl-5,6-dihydro-4*H*-cyclopenta[*b*]thiophen-4-one and 4-phenyl-4,5-dihydro-4*H*-cyclopenta[*b*]thiophen-6-one (43)

A mixture of 3-phenyl-1-(thiophen-3-yl)prop-2-en-1-one [or 3-phenyl-1-(thiophen-2-yl)prop-2-en-1-one] (200mg, 0.93 mM) and polyphosphoric acid (2 mL) were taken in a sealed tube and allowed to stir for 6 hours at 120 °C. The reaction mixture was then cooled to room temperature and stirred with water (10 mL) for 30 minutes. The aqueous solution thus obtained was extracted with ethyl acetate (3 x 10 mL) and the ensuing organic layer was successively washed with brine and dried over anhydrous Na_2SO_4 (1 g). Finally, the dried organic layer was concentrated under reduced pressure and the residue obtained was purified by column chromatography, using 8% EtOAc and hexane as the eluent to give the corresponding product.

The ^{1}H and ^{13}C NMR data for 6-phenyl-5,6-dihydro-4*H*-cyclopenta[*b*]thiophen-4-one was found to be consistent with the data already reported for same compound.[2]

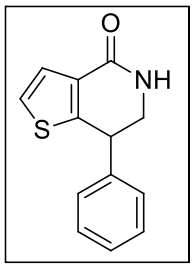
4-Phenyl-4,5-dihydro-4*H*-cyclopenta[*b*]thiophen-6-one (43): Yield: 67%; colourless semi-solid; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate = 9:1]; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.93(dd, *J* = 4.0 Hz and 16.0 Hz, 1H), 3.51(dd, *J* = 8.0 Hz and 16.0 Hz, 1H), 4.54 (dd, *J* = 4.0 Hz and 8.0 Hz, 1H), 6.91 (d, *J* = 4.0 Hz, 1H), 7.16 (d, *J* = 8.0 Hz, 2H), 7.29 (d, *J* = 8.0 Hz, 1H), 7.33 (t, *J* = 8.0 Hz, 2H), 7.91 (d, *J* = 4.0 Hz, 1H);[3] **^{13}C NMR** (101 MHz, $CDCl_3$): δ 42.5, 51.5, 124.0, 127.1, 127.2, 128.9, 140.9, 141.2, 142.3, 171.1, 196.2.



General method for synthesis of 7-aryl-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one(47)/4-aryl-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (58) and 4-aryl-3,4-dihydroquinolin-2(1*H*)-one (17, 64-71):

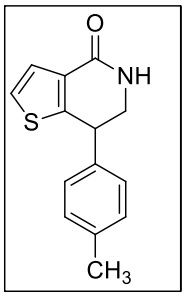
In a sealed tube, a solution of 3-aryl-1-(thiophen-3-yl)prop-2-en-1-one (0.46 mmol) in 1mL of polyphosphoric acid was stirred for 6 hours under nitrogen atmosphere. The temperature of the reaction mixture was maintained at 120 °C. On the formation of the fused cyclopentanone compound, as indicated by the TLC, NaN_3 (0.9 mmol) was added and the reaction mixture was left to stir for additional 8 hours. Progress of the reaction was monitored by the TLC. On completion of the reaction, the contents were cooled to room temperature and diluted with 10 mL of (3:1) water-ethyl acetate mixture. It was then passed through a celite pad and the resulting aqueous-organic layers were separated. The aqueous layer was further extracted with (2 x 10 mL) ethyl acetate. All the organic layers were combined and washed with brine and dried over Na_2SO_4 (1 g). It was finally concentrated under reduced pressure and the residue thus obtained was purified by silica gel column chromatography.

7-Phenyl-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (47). Yield: 65%; light brown solid;



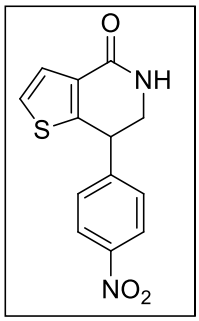
M.p. 150-154 °C; $\mathbf{R_f}$ = 0.3 [hexane / ethyl acetate = 7:3]; $\mathbf{\nu_{max}}$ **(KBr)/cm^{-1}**: 3206, 3061, 2878, 1679, 1482, 1454, 1322, 1290, 1193; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.68 – 3.75 (m, 1H), 3.83 (ddd, *J* = 12.2, 5.6, 3.2 Hz, 1H), 4.47 (dd, *J* = 9.0, 5.7 Hz, 1H), 6.07 (bs, 1H), 7.20 (d, *J* = 5.2 Hz, 1H), 7.29 [d (merged with residual $CHCl_3$ peak), 2H], 7.35-7.40 (m, 3H), 7.51 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 42.0, 48.9, 124.4, 126.0, 128.0, 128.1, 128.9, 132.4, 140.0, 150.4, 163.5; **LRMS-ESI** (m/z): 230.00 $[M + H]^+$.

7-(*p*-Tolyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (48). Yield: 35%; light brown semi



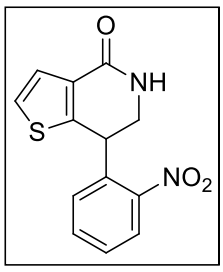
solid; $\mathbf{R_f}$ = 0.4 [hexane / ethyl acetate = 7:3]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3198, 3056, 2926, 2854, 1664, 1482, 1430, 1322, 1256, 1195; **^{1}H NMR** (400 MHz, $CDCl_3$): δ ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.38 (s, 3H), 3.67 – 3.75 (m, 1H), 3.81 (ddd, *J* = 12.2, 5.7, 3.3 Hz, 1H), 4.43 (dd, *J* = 9.2, 5.7 Hz, 1H), 6.11 (bs, 1H), 7.18-7.19 [m, 5H(1H - thiophene merged with 4H - phenyl ring)], 7.51 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 29.7, 41.7, 49.0, 124.3, 125.9, 127.9, 129.5, 132.3, 136.9, 137.8, 150.9, 163.6; **LRMS-ESI** (m/z): 244.06 $[M + H]^+$.

7-(4-Nitrophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (49). Yield: 68%; light



brown solid; **M.p**. 162-165 °C; $\mathbf{R_f}$ = 0.2 [hexane / ethyl acetate = 6:4]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3305, 3206, 3061, 2860, 1667, 1598, 1427, 1296, 1262, 1192; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.70 – 3.76 (m, 1H), 3.95 – 4.01 (m, 1H), 4.56 (t, *J* = 6.4 Hz, 1H), 7.08 (bs, 1H), 7.25 (d, *J* = 5.2 Hz, 1H), 7.44 (d, *J* = 8.6 Hz, 2H), 7.51 (d, *J* = 5.2 Hz, 1H), 8.20 (d, *J* = 8.6 Hz, 2H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.3, 48.4, 124.1, 125.1, 126.1, 128.9, 132.9, 147.3, 147.5, 147.9, 163.6; **LRMS-ESI** (m/z): 275.00 $[M + H]^+$. [Compound isolated contained some aromatic impurities which persisted in spite of repeated chromatography and crystallization attempts.]

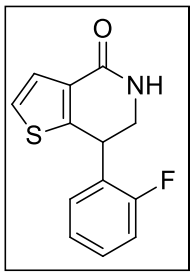
7-(2-Nitrophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (50). Yield: 61%; brown



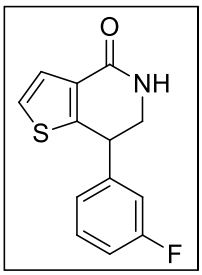
semi solid; $\mathbf{R_f}$ = 0.1 [hexane / ethyl acetate = 5:5]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3246, 3045, 2845, 1676, 1588, 1456, 1276, 1272, 1165; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.74 – 3.83 (m, 1H), 4.18 (ddd, *J* = 12.8, 5.7, 1.8 Hz, 1H), 5.10 (t, *J* = 5.1 Hz, 1H), 6.10 (bs, 1H), 7.19 (d, *J* = 7.7 Hz, 1H), 7.28 (app doublet, 1H, merged with residual $CHCl_3$ peak), 7.49 (t, 1H), 7.46-7.56 (merged multiplet and doublet, 3H), 8.02 (d, *J* = 8.0 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 35.9,

48.0, 125.1, 125.4, 126.0, 128.8, 130.5, 133.4, 133.7, 134.6, 147.4, 148.7, 163.4; **LRMS-ESI** (m/z): 274.97 $[M + H]^+$.

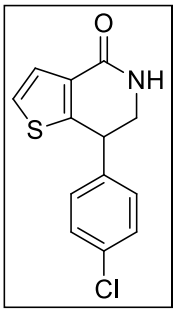
7-(2-Fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (51). Yield: 58%; brown semi solid; $\mathbf{R_f}$ = 0.3 [hexane / ethyl acetate = 8:2]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3202, 3085, 2924, 2853, 1668, 1582, 1485, 1454, 1310, 1217, 1193; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.72 (dd, *J* = 12.3, 6.7 Hz, 1H). 3.88 – 3.95 (m, 1H), 4.80 (t, *J* = 6.1 Hz, 1H), 6.29 (bs, 1H), 7.03 – 7.15 (m, 3H), 7.23 (d, *J* = 8.0 Hz, 1H), 7.28-7.34 (m, 1H),7.53 (d, *J* = 8.0 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 34.2, 47.3, 115.6, 124.4, 124.5, 126.0, 126.9, 129.4, 129.5, 133.0, 148.3, 160.3, 163.5; **LRMS-ESI** (m/z): 248.20 $[M + H]^+$.



7-(3-Fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (52). Yield: 52%; light brown solid; **M.p**.110-114 °C; $\mathbf{R_f}$ = 0.3 [hexane / ethyl acetate = 8:2]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3210, 2935, 2908, 1675, 1486, 1266; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.66 – 3.74 (m, 1H), 3.84-3.89 (m, 1H), 4.42 – 4.47(m, 1H), 6.17 (bs, 1H), 6.95 – 7.06 (m, 3H), 7.20 (d, *J* = 5.2 Hz, 1H), 7.33 (td, *J* = 8.0, 6.1 Hz, 1H), 7.50 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.6, 48.7, 114.8, 115.1, 123.6, 124.7, 126.0, 142.4, 149.3, 162.9, 163.4; **LRMS-ESI** (m/z): 248.24 $[M + H]^+$.

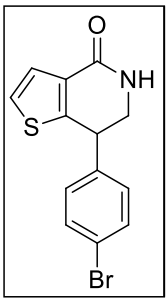


7-(4-Chlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (53). Yield: 63%; light yellow semi solid; $\mathbf{R_f}$ = 0.2 [hexane / ethyl acetate = 7:3]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3208, 3065, 2913, 2360, 1675, 1492, 1326, 1192; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.63 – 3.72 (m, 1H), 3.83-3.88 (m, 1H), 4.45 (dd, *J* = 8.5, 5.6 Hz, 1H), 6.06 (s, 1H), 7.21-7.23 [merged doublets, 3H(1H - thiophene merged with 2H - phenyl ring)], 7.23 (d, *J* = 4.6 Hz, 2H), 7.35 (d, *J* = 8.5 Hz, 2H),



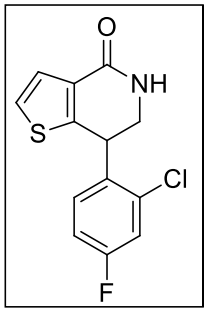
7.52 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.4, 48.9, 124.6, 126.1, 129.1, 129.3, 132.5, 133.9, 138.4, 149.6, 163.4; **LRMS-ESI** (m/z): 264.00 $[M + H]^+$.

7-(4-Bromophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (54). Yield: 62%; brown



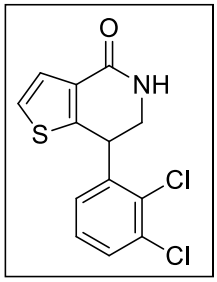
solid; **M.p**. 112-117 °C; **R_f** = 0.2 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3356, 3136, 1663, 1622, 1421, 1058; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.62 – 3.70 (m, 1H), 3.81-3.86 (m, 1H), 4.41 (dd, *J* = 8.5, 5.6 Hz, 1H), 5.95 (s, 1H), 7.15 (d, *J* = 8.0 Hz, 2H), 7.20 (d, *J* = 5.2 Hz, 1H), 7.47 – 7.50 [merged doublets, 3H(1H - thiophene merged with 2H - phenyl ring)]; **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.4, 48.8, 122.0, 124.7, 126.1, 129.7, 132.0, 132.5, 139.0, 149.5, 163.5; **HRMS-ESI** (m/z): Calcd for $C_{13}H_{11}BrNOS$ $[M + H]^+$ 307.9745, found 307.9805.

7-(2-Chloro-4-fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (55). Yield:



62%; light brown semi solid; **R_f** = 0.3 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3210, 3183, 3070, 2973, 1678, 1594, 1489, 1382, 1235; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.64-3.69 (m, 1H), 3.98 (ddd, *J* = 12.5, 5.5, 2.0 Hz,1H), 4.93 (t, *J* = 5.4 Hz, 1H), 5.98 (bs, 1H), 6.89 – 6.95 (m, 1H), 6.96 – 7.01(m, 1H), 7.21 (d, *J* = 8.0 Hz, 1H), 7.26 (d, *J* = 5.2 Hz, 1H), 7.55 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 37.0, 47.1, 114.4, 117.2, 124.9, 126.0, 130.5, 133.2, 133.4, 133.7, 147.8, 161.8 163.3; **LRMS-ESI** (m/z): 282.05 $[M + H]^+$.

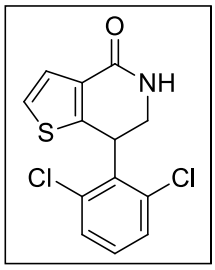
7-(2,3-Dichlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (56). Yield: 52%; light



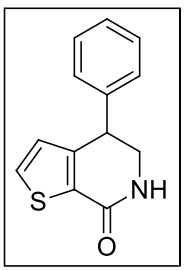
yellow semi solid; **R_f** = 0.2 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3198, 3080, 2912, 2360, 1684, 1531, 1482, 1446, 1421, 1308, 1180; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.66 – 3.72 (m, 1H), 4.04 (ddd, *J* = 12.5, 5.4, 1.7 Hz, 1H), 5.01 (t, *J* = 5.1 Hz, 1H), 5.58 (bs, 1H), 6.88 (d, *J* = 8.0 Hz, 1H), 7.14 (t, *J* = 7.9 Hz, 1H), 7.28 [d (merged with residual $CHCl_3$ peak), 1H], 7.44 (d, *J* =

8.0 Hz, 1H), 7.56 (d, *J* = 5.3 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.2, 46.8, 125.1, 126.0, 127.5, 127.6, 129.8, 131.3, 133.4, 133.6, 139.5, 147.4, 163.1; LRMS-ESI (m/z): 298.01$[M + H]^+$.

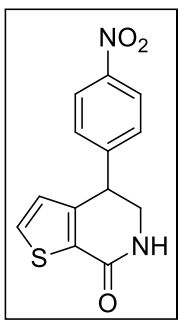
7-(2,6-Dichlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (57). Yield: 62%; light yellow semi solid; **R_f** = 0.2 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3317, 3236, 2952, 1656, 1523, 1431, 1315, 1287; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.60 (ddd, *J* = 11.6, 6.9, 4.6 Hz, 1H), 4.33 (t, *J* = 12.0 Hz, 1H), 5.63 (dd, *J* = 13.2, 6.9 Hz, 1H), 6.36 (bs, 1H), 7.14 (d, *J* = 5.2 Hz, 1H), 7.27 (d, *J* = 7.9 Hz, 1H), 7.34 (d, *J* = 7.0 Hz, 1H), 7.45 (d, *J* = 8.0 Hz, 1H), 7.48 (d, *J* = 5.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 39.0, 43.8, 123.5, 126.1, 128.7, 129.8, 130.7, 130.9, 133.8, 135.8, 136.6, 149.7, 163.4; **HRMS-ESI** (m/z): Calcd for $C_{13}H_{10}Cl_2NOS$ $[M + H]^+$ 297.9860, found 297.9900.



4-Phenyl-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (58). Yield: 44%; light brown semi solid; **R_f** = 0.4 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3367, 3178, 1654, 1609, 1524, 1432, 1394, 1243, 1124; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.04 (dd, *J* = 16.2, 11.6 Hz, 1H), 3.15 (dd, *J* = 16.5, 5.5 Hz, 1H), 4.94 (dd, *J* = 11.6, 5.3 Hz, 1H), 5.73 (bs, 1H), 6.92 (d, *J* = 4.9 Hz, 1H), 7.33 – 7.44 (m, 5H), 7.54 (d, *J* = 4.9 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 33.9, 57.9, 126.5, 126.7, 128.5, 129.0, 130.7, 131.7, 140.7, 143.8, 163.2; **LRMS-ESI** (m/z): 230.01 $[M + H]^+$.

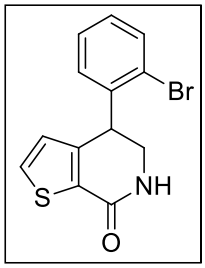


4-(4-Nitrophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (59). Yield: 45%; brown solid; **M.p**.180-183 °C; **R_f** = 0.3 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3375, 3185, 1588, 1558, 1482, 1401, 1313, 1249; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.65 – 3.74 (m, 1H), 3.89 – 3.97 (m, 1H), 4.38 – 4.46 (m, 1H), 6.74 (d, *J* = 5.0 Hz, 1H), 7.24 (bs, 1H), 7.39 (d, *J* = 8.6 Hz, 2H),

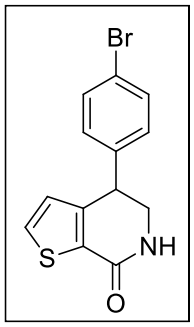


7.56 (d, *J* = 5.0 Hz, 1H), 8.18 (d, *J* = 8.6 Hz, 2H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.6, 48.6, 124.0, 126.7, 129.0, 132.0, 145.6, 147.4, 147.6, 163.3; **LRMS-ESI** (m/z): 275.18 $[M + H]^+$.

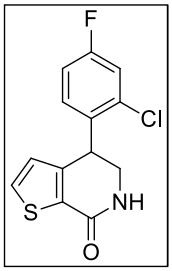
4-(2-Bromophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (60). Yield: 56%; light brown semi solid; **R_f** = 0.3 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3375, 3045, 1667, 1568, 1568, 1489, 1417, 1313, 1249, 1168; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.65 (ddd, *J* = 12.5, 6.6, 3.1 Hz, 1H), 3.92 (ddd, *J* = 12.5, 5.8, 2.4 Hz, 1H), 4.82 (t, *J* = 6.2 Hz, 1H), 6.43 (bs, 1H), 6.80 (d, *J* = 5.0 Hz, 1H),6.95 (d, *J* = 7.6 Hz, 1H),7.16 (t, *J* = 7.6 Hz, 1H), 7.23 (t, *J* = 7.5 Hz, 1H),7.56 (d, *J* = 5.0 Hz, 1H), 7.64 (d, *J* = 7.9Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 40.6, 47.4, 124.2, 127.0, 127.8, 129.1, 129.7, 131.8, 132.3, 133.2, 138.9, 146.2, 163.3; **LRMS-ESI** (m/z): 307.15 $[M + H]^+$.



4-(4-Bromophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (61). Yield: 58%; brown solid; **M.p**. 102-106 °C; **R_f** = 0.4 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3360, 3176, 1659, 1622, 1408, 1068; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.62 – 3.69 (m, 1H), 3.80 – 3.87 (m, 1H), 4.29 (dd, *J* = 8.6, 5.8 Hz, 1H), 6.48 (bs, 1H), 6.72 (d, *J* = 5.0 Hz, 1H), 7.11 (d, *J* = 8.4 Hz, 2H), 7.49 (d, *J* = 8.4 Hz, 2H), 7.54 (d, *J* = 5.0 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 41.6, 49.0, 121.5, 126.9, 129.8, 130.1, 131.6, 132, 139.1, 146.9, 163.2; **LRMS-ESI** (m/z): 307.95 $[M + H]^+$. [Compound isolated contained some aromatic impurities which persisted in spite of repeated chromatography and crystallization attempts.]

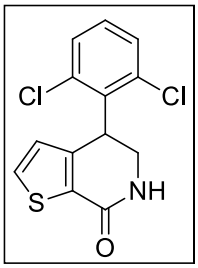


4-(2-Chloro-4-fluorophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (62). Yield: 46%; light brown semi solid; **R_f** = 0.3 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3354, 3082, 2926, 1657, 1492, 1400, 1354, 1313, 1265, 1216, 1122; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.61-3.67 (m, 1H), 3.94 (m, 1H), 4.80

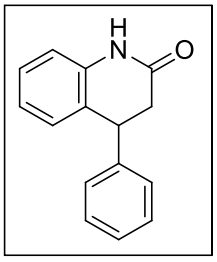


(t, *J* = 5.9 Hz, 1H), 6.24 (bs, 1H), 6.81 (d, *J* = 5.0 Hz, 1H), 6.90 – 6.93 (m, 2H),7.21 (d, *J* = 8.0 Hz, 1H), 7.59 (d, *J* = 5.0 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 37.4, 47.3, 114.3, 117.2, 126.9, 130.6, 132.0, 132.4, 133.3, 134.0, 145.9, 161.5, 163.0; **LRMS-ESI** (m/z): 282.08 $[M + H]^+$.

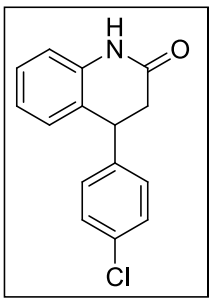
4-(2,6-Dichlorophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (63). Yield: 61%; brown semi solid; **R_f** = 0.3 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3323, 3016, 1667, 1534, 1464, 1323, 1267; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.52 – 3.59 (m, 1H), 4.25 (t, *J* = 12 Hz, 1H), 5.42 (dd, *J* = 13.5, 6.8 Hz, 1H), 6.47 (bs, 1H), 6.56 (d, *J* = 5.0 Hz, 1H), 7.23-7.34 (m, 2H), 7.39-7.48 [m, 2H(contains 1H from thiophene)]; **^{13}C NMR** (101 MHz, $CDCl_3$): δ 39.3, 44.0, 126.1, 128.7, 129.5, 129.7, 130.5, 131.2, 133.9, 146.2, 163.2; **LRMS-ESI** (m/z): 298.00 $[M + H]^+$.



4-Phenyl-3,4-dihydroquinolin-2(1*H*)-one (17). Yield: 56%; light yellow solid; **M.p**. 180-182 °C; **R_f** = 0.4 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3447,3217, 2967, 3009, 1675, 1646, 1481, 1371, 1101, 856; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.89 – 2.99 (m, 2H), 4.30 (t, *J* = 7.5 Hz, 1H), 6.86 (d, *J* = 7.8 Hz, 1H),6.91-6.99 (m, 2H), 7.21 (t, *J* = 6.9 Hz, 3H), 7.26 (d, *J* = 4.0 Hz, 1H), 7.34 (t, *J* = 8.0 Hz, 2H), 8.66 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.4, 42.0, 115.6, 123.4, 126.7, 127.2, 127.8, 128.0, 128.4, 128.9, 137.0, 141.4, 170.7; **LRMS-ESI** (m/z): 224.08 $[M + H]^+$.

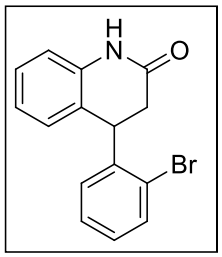


4-(4-Chlorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (64). Yield: 48%; light yellow solid; **M.p**. 184-186 °C; **R_f** = 0.5 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3224, 3128,1680, 1557, 1548, 1239, 1105, 872; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.90 (qd, *J* = 16.2, 7.3 Hz, 2H), 4.27 (t, *J* = 8.0 Hz, 1H), 6.90 (t, *J* = 8.0 Hz, 2H), 6.97 (t, *J* = 8.0 Hz, 1H), 7.11 (d, *J* = 8.0 Hz, 2H), 7.21 (t,



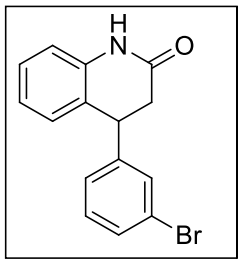
J = 8.0 Hz, 1H), 7.29 (d, *J* = 8.0 Hz, 2H), 9.07 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.3, 41.4, 115.9, 123.5, 126.0, 127.8, 128.3, 129.1, 129.2, 133.0, 137.0, 140.0, 170.6; **LRMS-ESI** (m/z): 258.73 $[M + H]^+$.

4-(2-Bromophenyl)-3,4-dihydroquinolin-2(1*H*)-one (65). Yield: 47%; light brown solid;



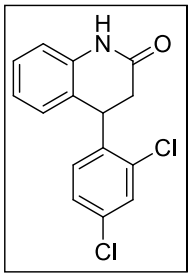
M.p. 200-204 °C; **R_f** = 0.4 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3424, 3008, 2898, 1682, 1548, 1178, 1143, 1045, 798; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.89-3.00 (m, 2H), 4.86 (t, *J* = 8.0 Hz, 1H), 6.94 – 7.02 (m, 4H) 7.14 (t, *J* = 8.0 Hz, 1H), 7.25 (q, *J* = 8.0 Hz, 2H), 7.65 (d, *J* = 8.0 Hz, 1H), 9.47 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 37.1, 41.0, 115.8, 123.6, 124.4, 125.2, 128.0, 128.3, 128.5, 128.8, 129.1, 133.3, 137.4, 140.5, 170.5; **HRMS-ESI** (m/z): Calcd for $C_{15}H_{13}BrNO$ $[M + H]^+$ 302.0181, found 302.0175.

4-(3-Bromophenyl)-3,4-dihydroquinolin-2(1*H*)-one (66). Yield: 49%; light brown oil; **R_f** =



0.6 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3445, 3194, 3056, 2922, 1672, 1588, 1485, 1381, 1249, 1170, 1103, 1072, 1009, 899, 757; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.84-2.98 (m, 2H), 4.27 (t, *J* = 8.0 Hz, 1H), 6.91(t, *J* = 8.0 Hz, 2H), 6.99 (t, *J* = 8.0 Hz, 1H), 7.11 (d, *J* = 7.7 Hz, 1H), 7.18-7.25 (m, 2H), 7.33 (s, 1H), 7.40 (d, *J* = 8.0 Hz, 1H), 8.98 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.3, 41.7, 115.9, 123.0, 123.6, 125.7, 126.5, 128.3, 128.4, 130.4, 130.5, 130.9, 137.0, 143.9, 170.4; **LRMS-ESI** (m/z): 302.01, 304.08 $[M + H]^+$.

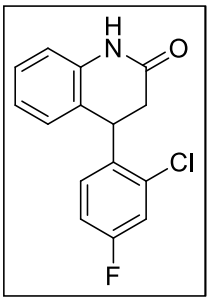
4-(2,4-Dichlorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (67). Yield: 53%; light yellow oil;



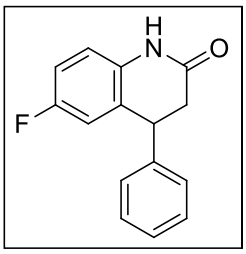
R_f = 0.5 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3246, 3035, 2872, 1657, 1534, 1467, 1324, 1056, 883, 776, 687; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.92 (qd, *J* = 16.3, 6.8 Hz, 2H), 4.78 (t, *J* = 6.7 Hz, 1H), 6.85 (d, *J* = 8.4 Hz, 1H), 6.93 (d, *J* = 7.8 Hz, 2H), 7.00 (t, *J* = 7.5 Hz, 1H), 7.14 (d, *J* = 8.4

Hz, 1H), 7.22 –7.28 (m, 1H), 7.45 (s, 1H), 9.23 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 36.8, 38.1, 116.0, 123.7, 124.6, 127.7, 128.4, 128.6, 129.8, 129.9, 133.6, 134.3, 137.4, 137.5, 170.3; **LRMS-ESI** (m/z): 292.03 $[M + H]^+$.

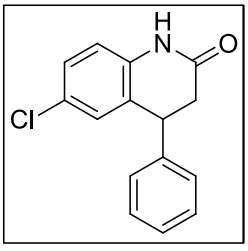
4-(2-Chloro-4-fluorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (68). Yield: 52%; light yellow solid; **M.p**. 184-187 °C; **R_f** = 0.6 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3446, 3065, 2922, 1677, 1598, 1546, 1487, 1383, 1237, 1039, 903, 862, 818, 754; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.86 – 2.99 (m, 2H), 4.79 (t, *J* = 6.8 Hz, 1H), 6.89 (d, *J* = 8.0 Hz, 3H), 6.96 (d, *J* = 8.0 Hz, 1H), 7.03 (t, *J* = 8.0 Hz, 1H), 7.21 (d, *J*= 8.0 Hz, 1H), 7.28 (t, *J*= 8.0 Hz, 1H; also includes $CDCl_3$ residual solvent peak), 8.54 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 36.9, 38.0, 114.6,115.7,117.3, 123.7, 124.9, 128.4, 129.9,130.7, 134.2, 134.7, 137.3, 161.4, 169.9; **LRMS-ESI** (m/z): 276.04 $[M + H]^+$.



6-Fluoro-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (69). Yield: 55%; colourless oil; **R_f** = 0.6 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3275, 3018, 2879, 1675, 1491, 1389, 1135, 1078, 989, 879,755, 635; **^{1}H NMR** (400 MHz, $CDCl_3$): δ ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.90-2.92 (merged qd, 2H), 4.27 (t, *J* = 7.8 Hz, 1H), 6.61 (d, *J* = 8.0 Hz, 1H), 6.84– 6.93 (m, 2H), 7.20 – 7.22 (d, *J* = 8.0 Hz, 2H), 7.30 – 7.38 (m, 3H), 9.45 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.0, 42.1, 114.6, 115.2, 116.8, 127.5, 127.8, 128.6, 129.1, 133.2, 140.6,158.9, 170.9; **LRMS-ESI** (m/z): 242.34 $[M + H]^+$.

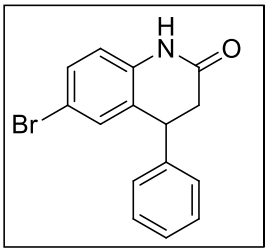


6-Chloro-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (70). Yield: 47%; colourless oil; **R_f** = 0.6 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3334, 3156, 2913, 1670, 1545, 1229, 1145, 872, 734; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.90-2.92 (d, *J* = 8.0 Hz, 2H), 4.26 (t, *J* = 8.0 Hz, 1H), 6.79 (d, *J* = 8.0



Hz, 1H), 6.89 (s, 1H), 7.16-7.19 (m, 3H), 7.31 (d, *J* = 8.0 Hz, 1H), 7.35 – 7.38 (m, 2H), 8.58 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$):δ 37.9, 41.5, 116.6, 127.6, 127.7, 128.0, 128.3, 128.4, 128.5, 129.1, 135.6, 140.3, 170.2; **LRMS-ESI** (m/z): 258.81 $[M + H]^+$.

6-Bromo-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (71). Yield: 45%; light yellow oil; **R_f** = 0.6 [hexane / ethyl acetate = 7:3]; **ν_{max} (KBr)/cm^{-1}**: 3412, 2965, 2867, 1662, 1543,1463,1176, 889, 767; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 2.88–2.99 (merged qd, 2H), 4.29 (t, *J* = 7.6 Hz, 1H), 6.76 (d, *J* = 8.4 Hz, 1H), 7.06 (d, *J* = 2.0 Hz, 1H), 7.21 (d, *J* = 8.0 Hz, 2H), 7.33 – 7.39 (m, 4H), 8.52 (bs, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 38.1, 41.9, 115.9, 117.1, 127.6, 127.7, 128.9, 129.1, 130.9, 131.2, 136.1, 140.5, 170.2; **LRMS-ESI** (m/z): 302.03, 304.10 $[M + H]^+$.



2.6. Computational Details

The geometries of the reactants and products were optimized in the gas phase using M06/6-31+G (d,p) level of theory and QST3 method was used to optimize the predicted transition state structures as implemented in the Gaussian 09 software.[61,62] Calculated structures were identified as true transition states for this reaction from the single imaginary frequency of vibration along the N···N_2 bond.

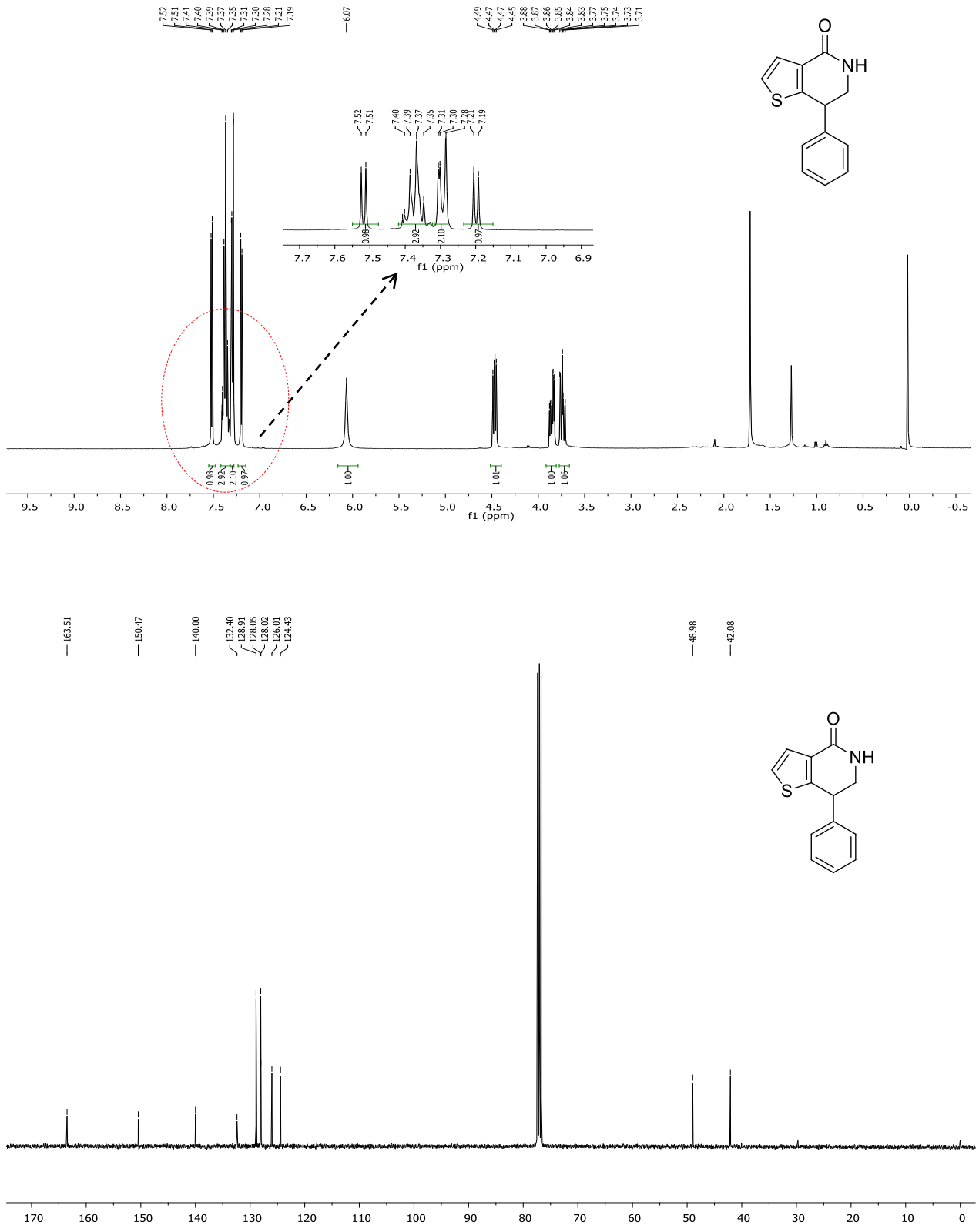
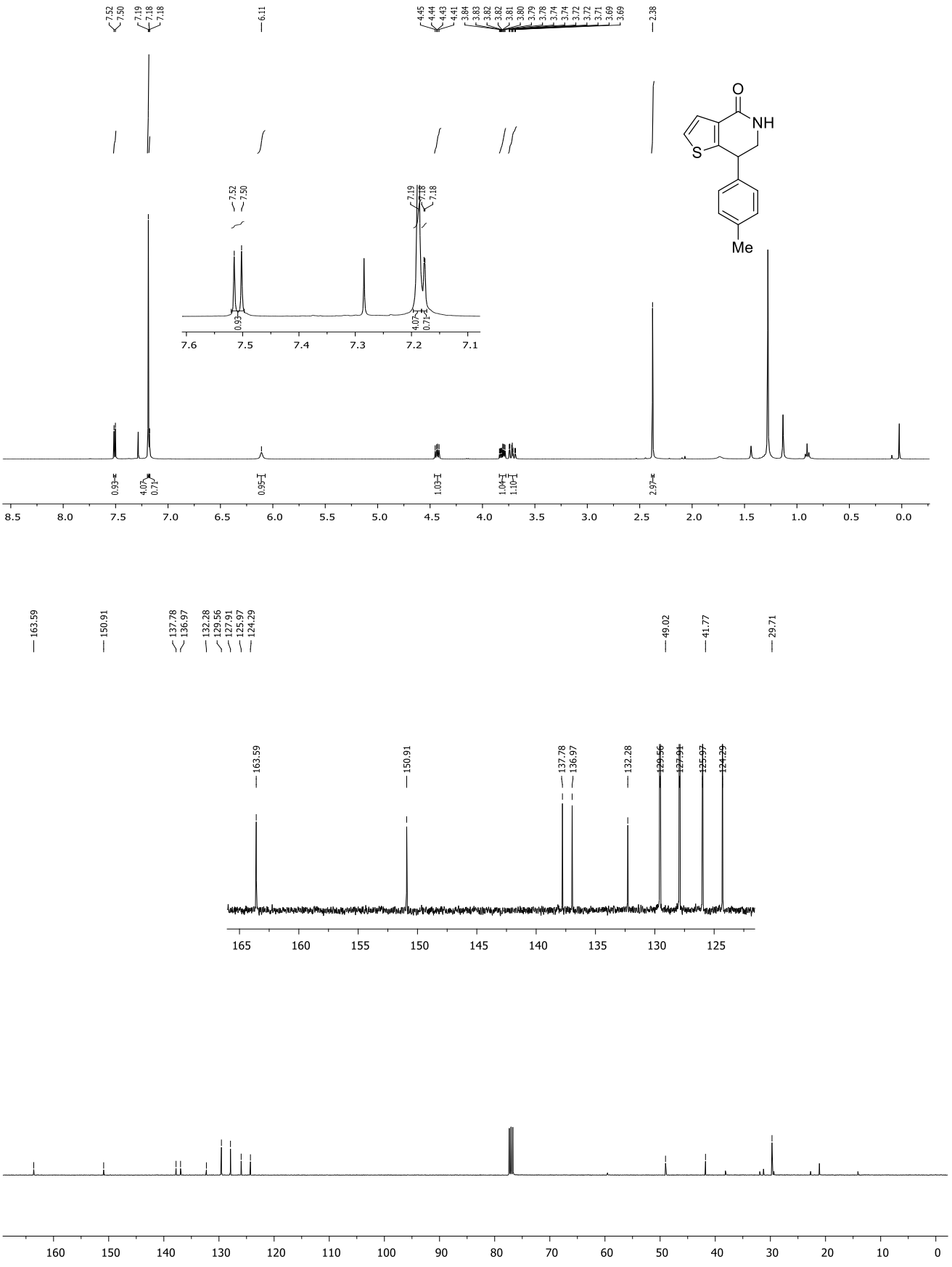
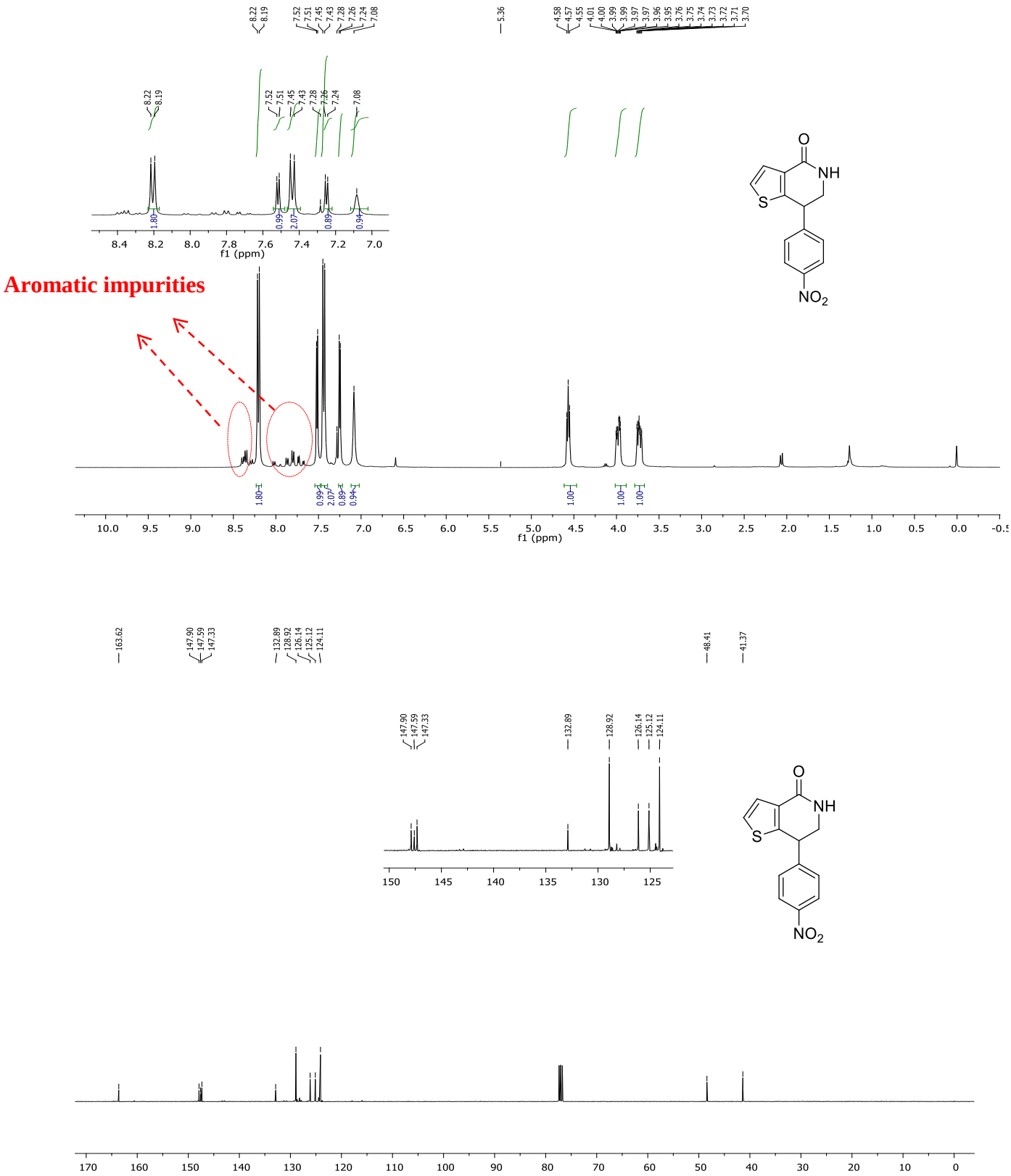


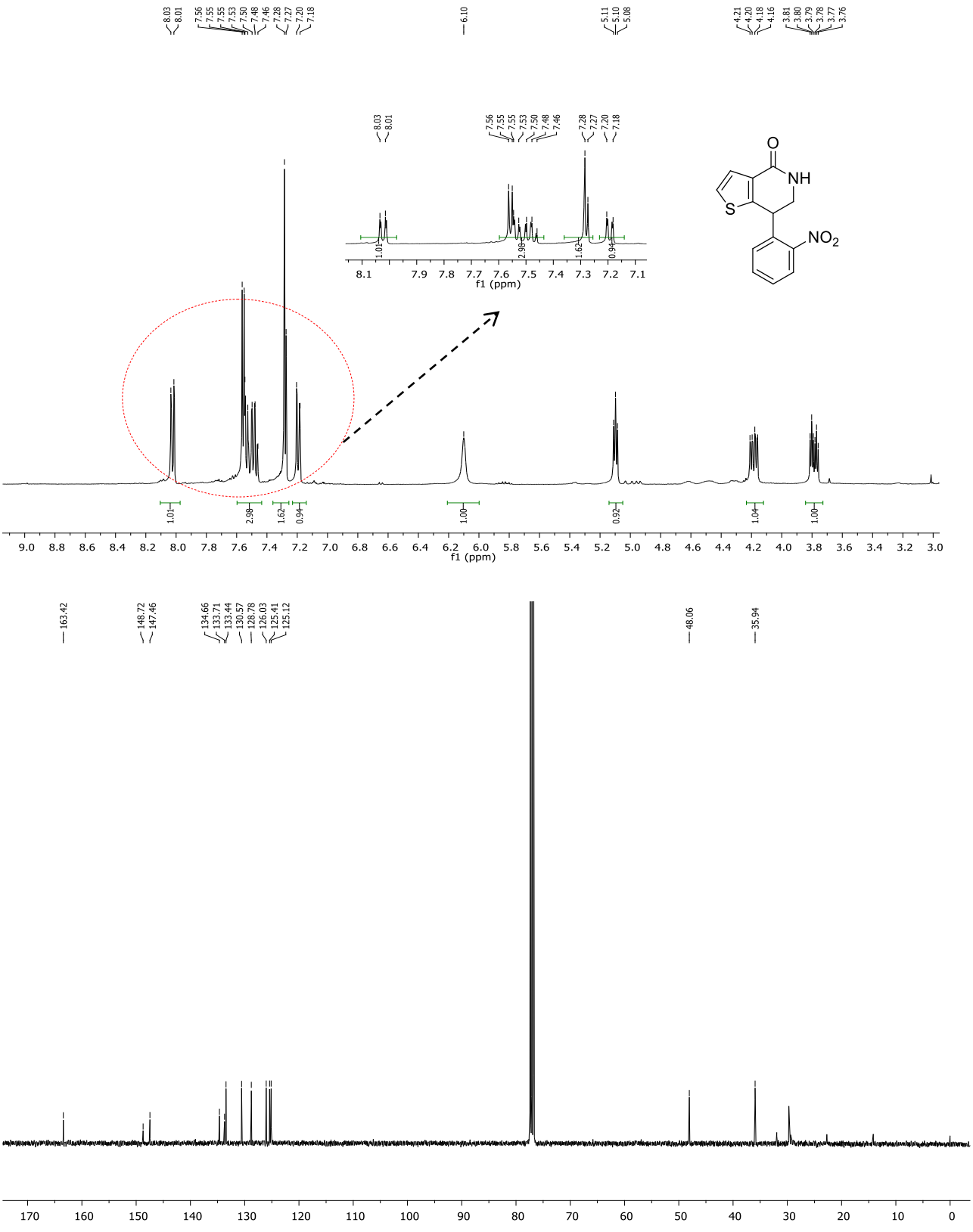
Figure 2.4: ^{1}H and ^{13}C NMR of compound **7-Phenyl-6,7-dihydrothieno[3,2-c]pyridin-4(5*H*)-one (47)** in $CDCl_3$



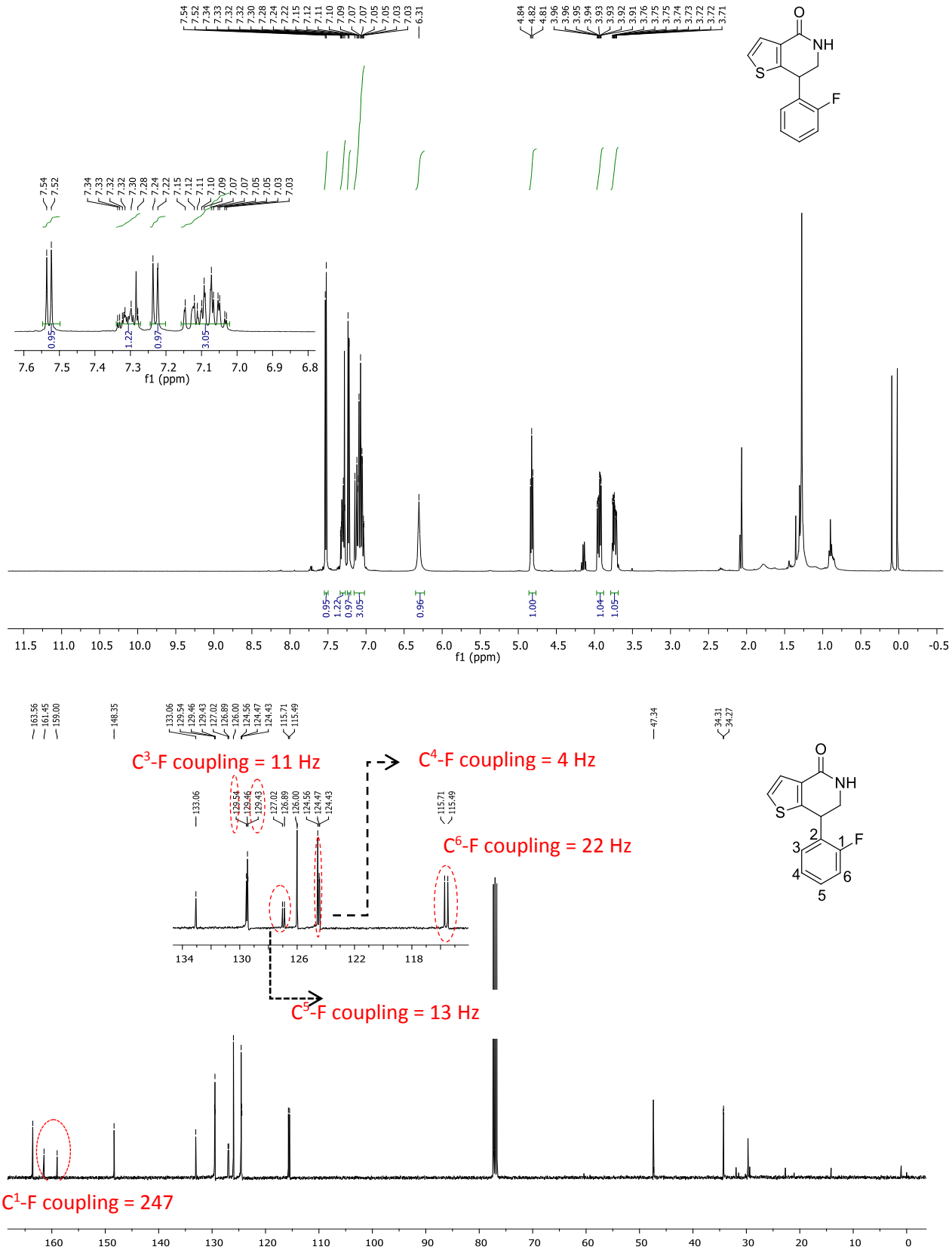
^{1}H and ^{13}C NMR of compound **7-(p-Tolyl)-6,7-dihydrothieno[3,2-c]pyridin-4(5H)-one (48)** in $CDCl_3$



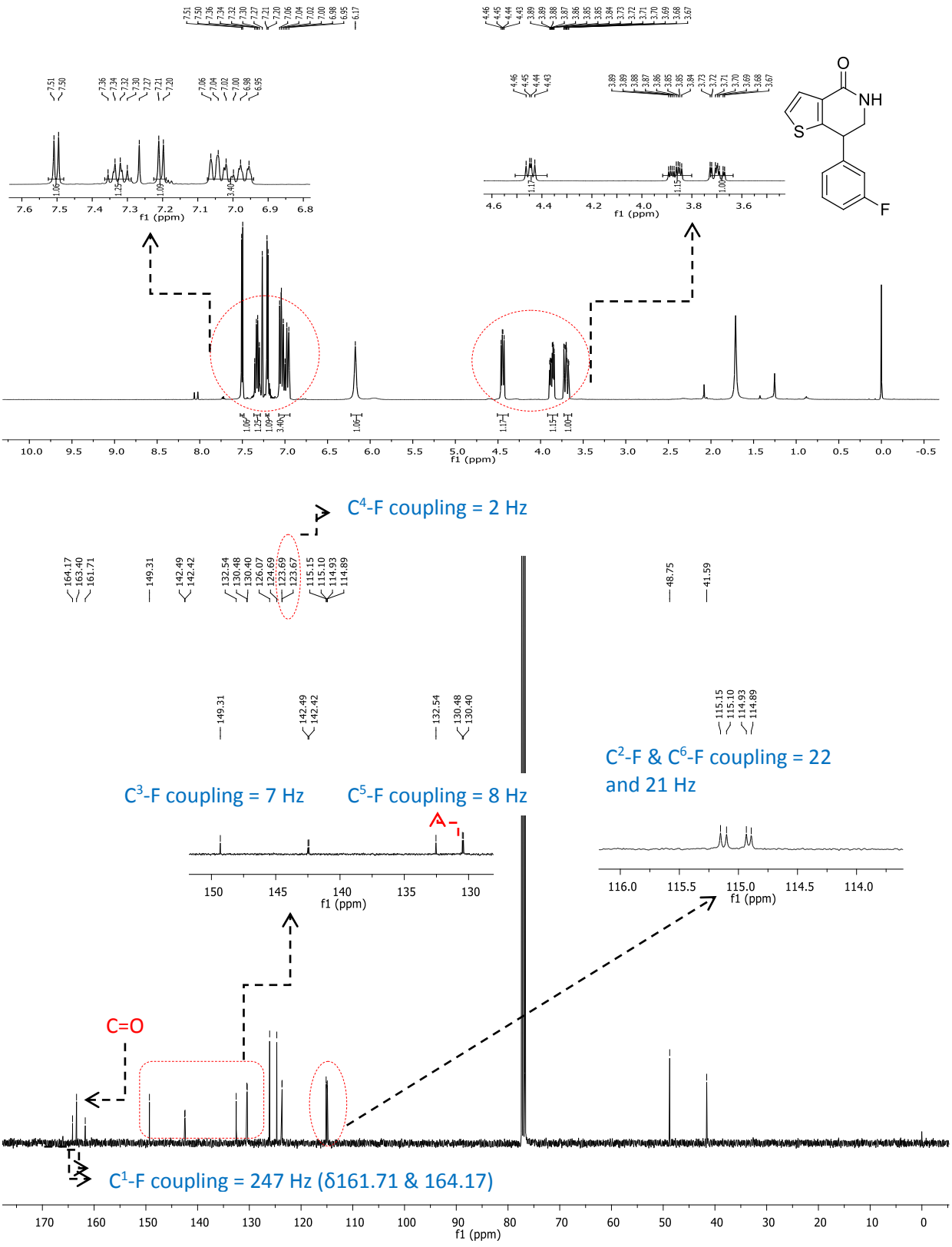
^{1}H and ^{13}C NMR of compound **7-(4-Nitrophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (49)** in $CDCl_3$



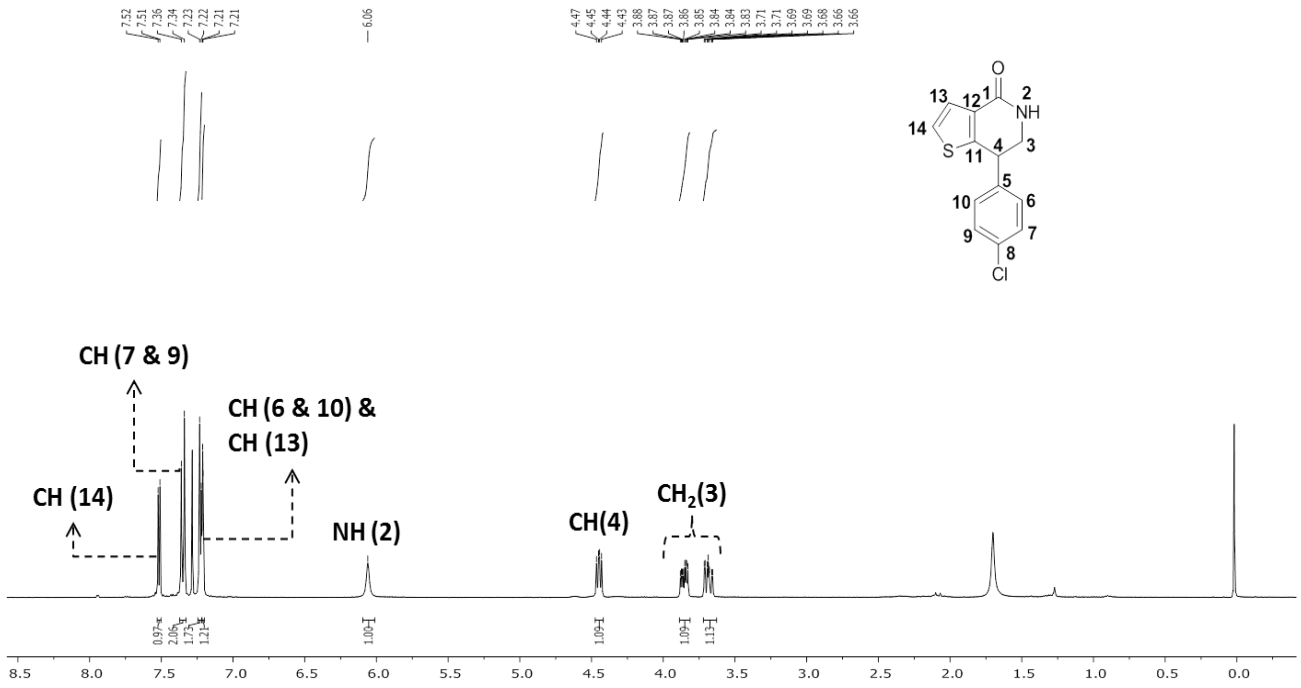
^{1}H and ^{13}C NMR of compound **7-(2-Nitrophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (50)** in $CDCl_3$

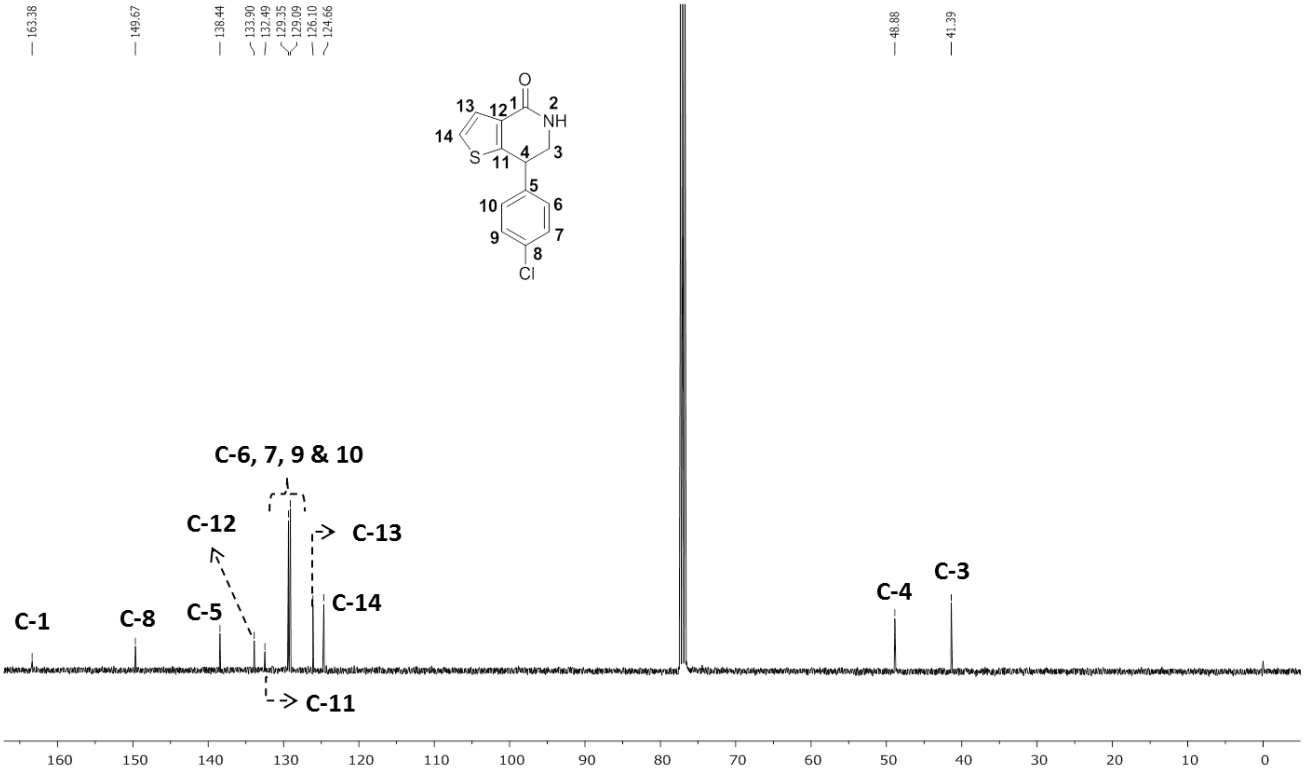


^{1}H and ^{13}C NMR of compound **7-(2-Fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (51)** in $CDCl_3$

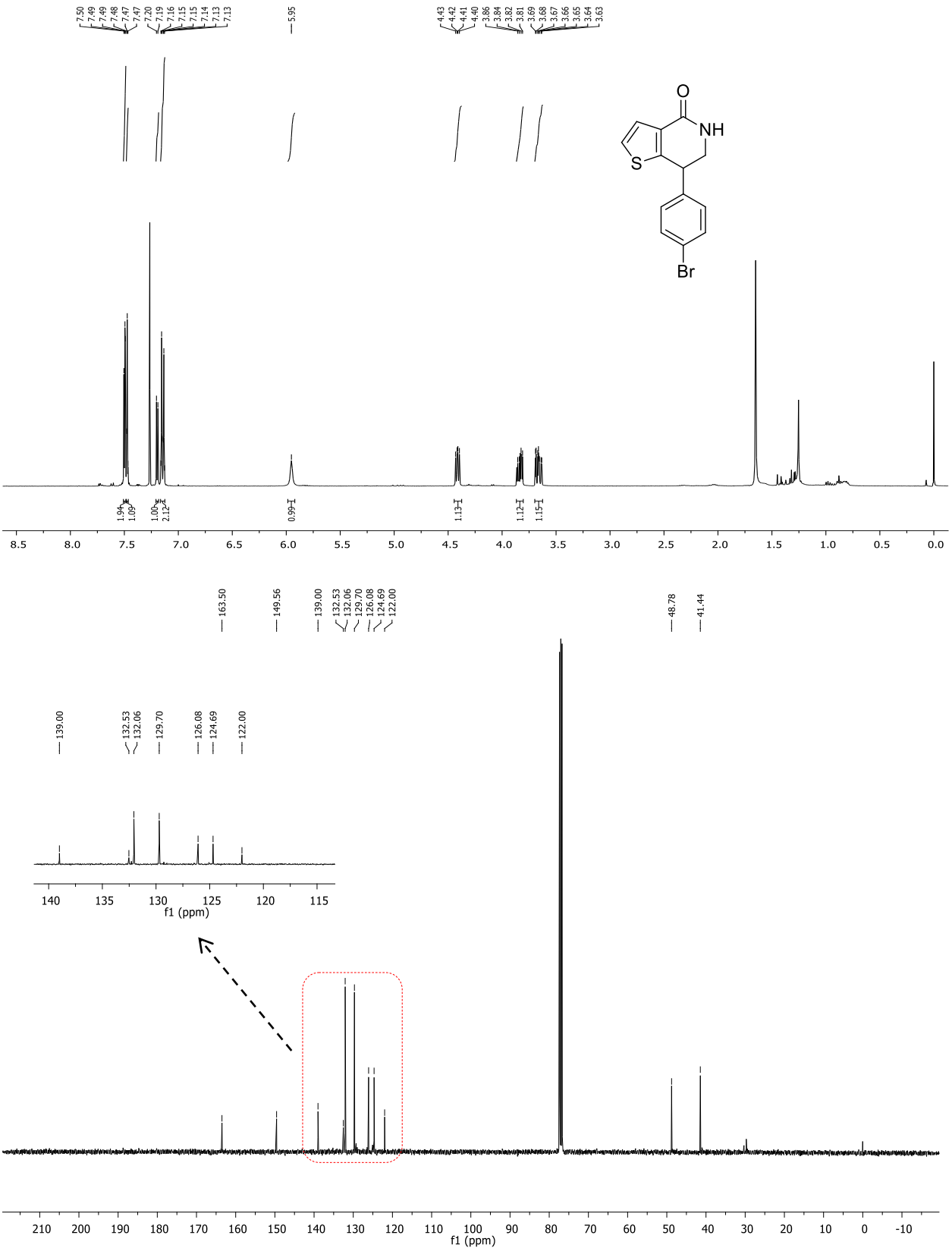


^{1}H and ^{13}C NMR of compound **7-(3-Fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (52)** in $CDCl_3$

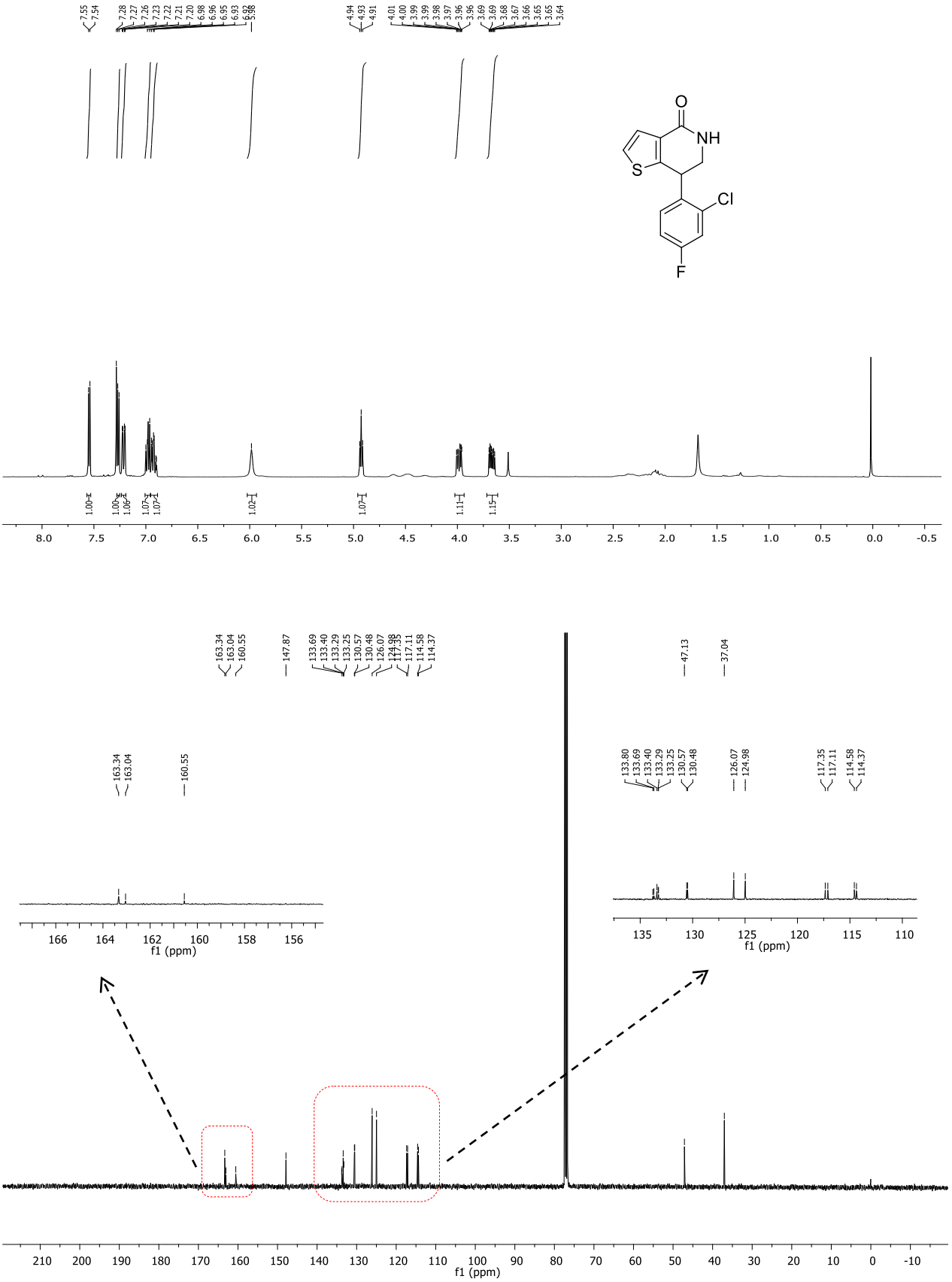




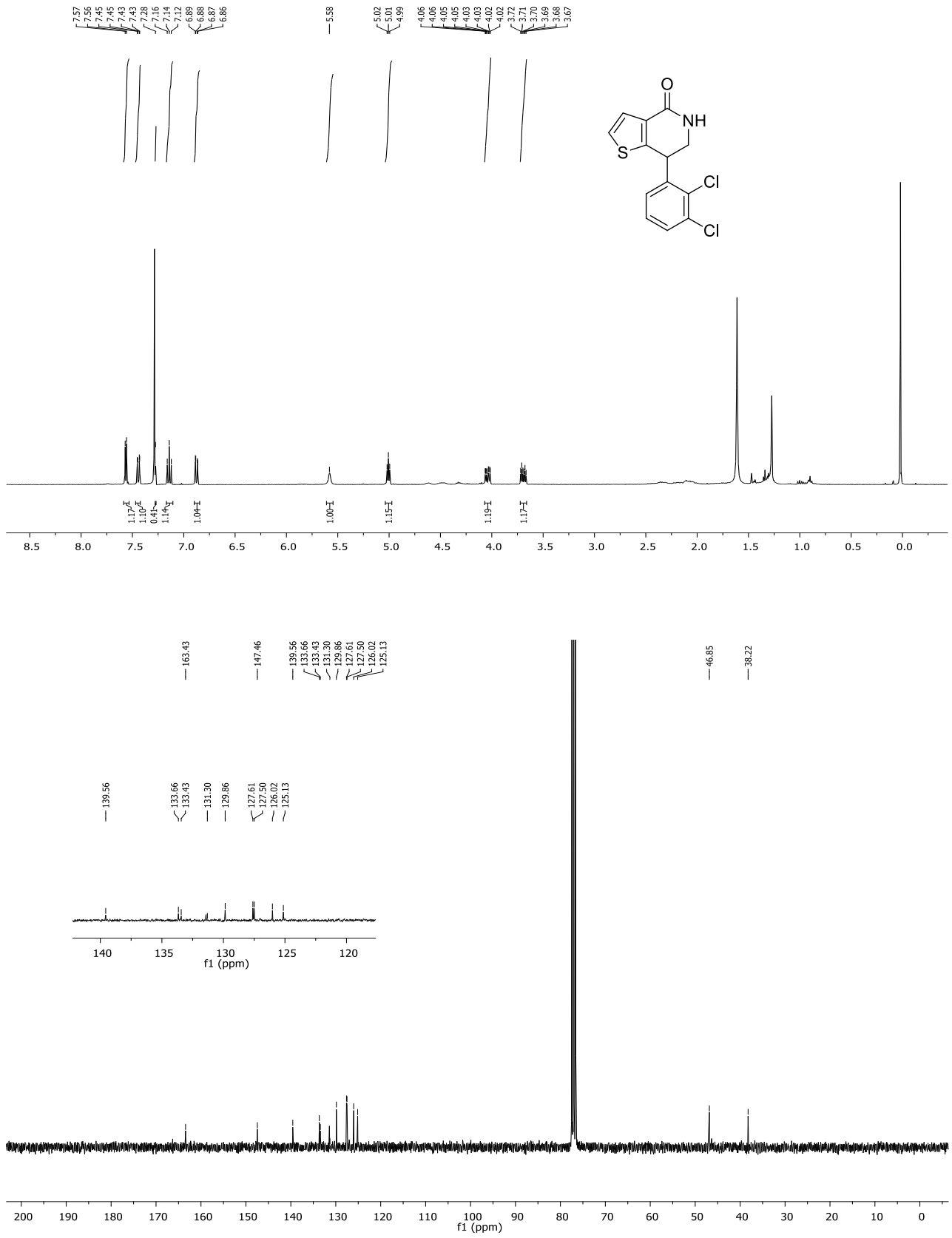
^{1}H and ^{13}C NMR of compound **7-(4-Chlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (53)** in $CDCl_3$



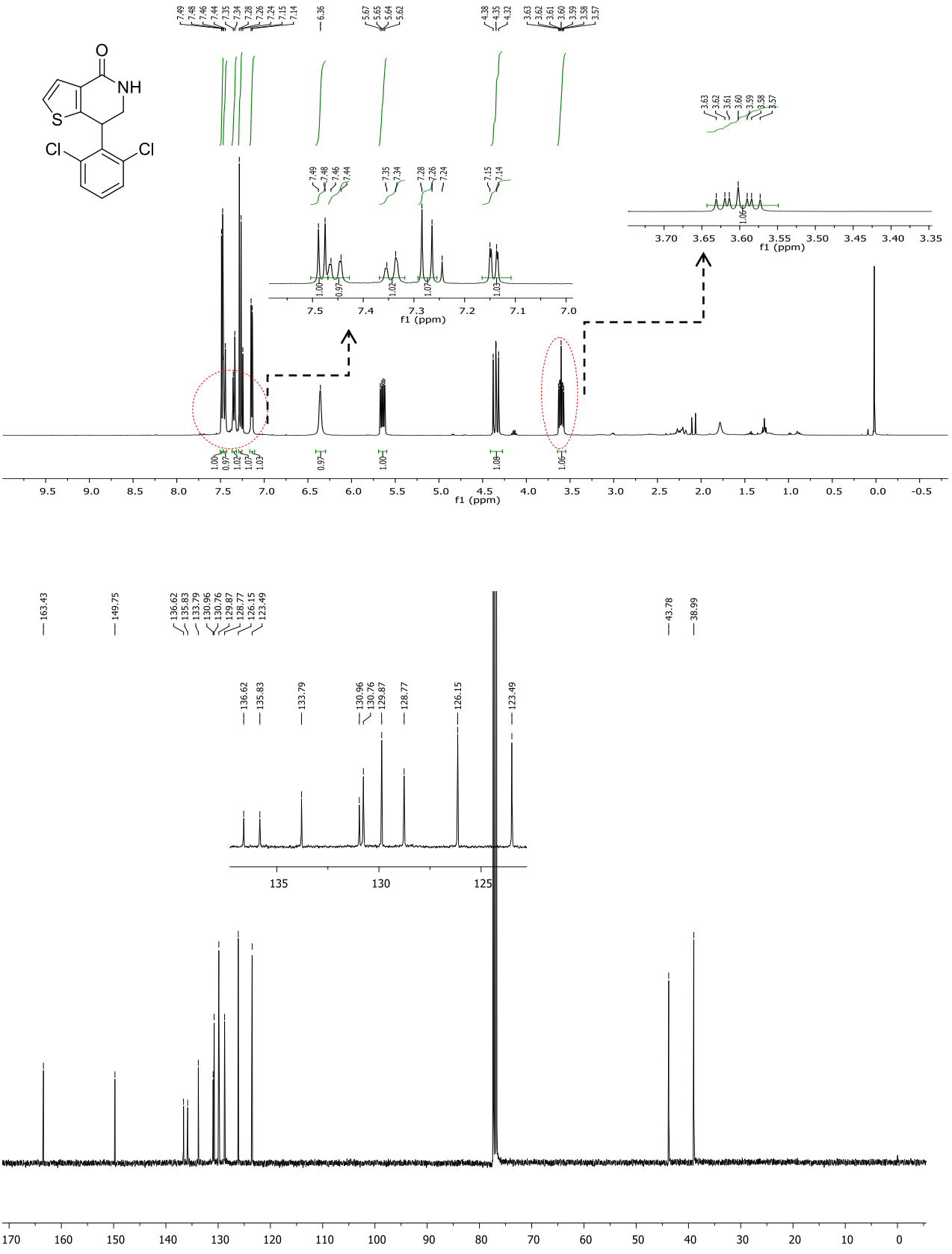
^{1}H and ^{13}C NMR of compound **7-(4-Bromophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (54)** in $CDCl_3$



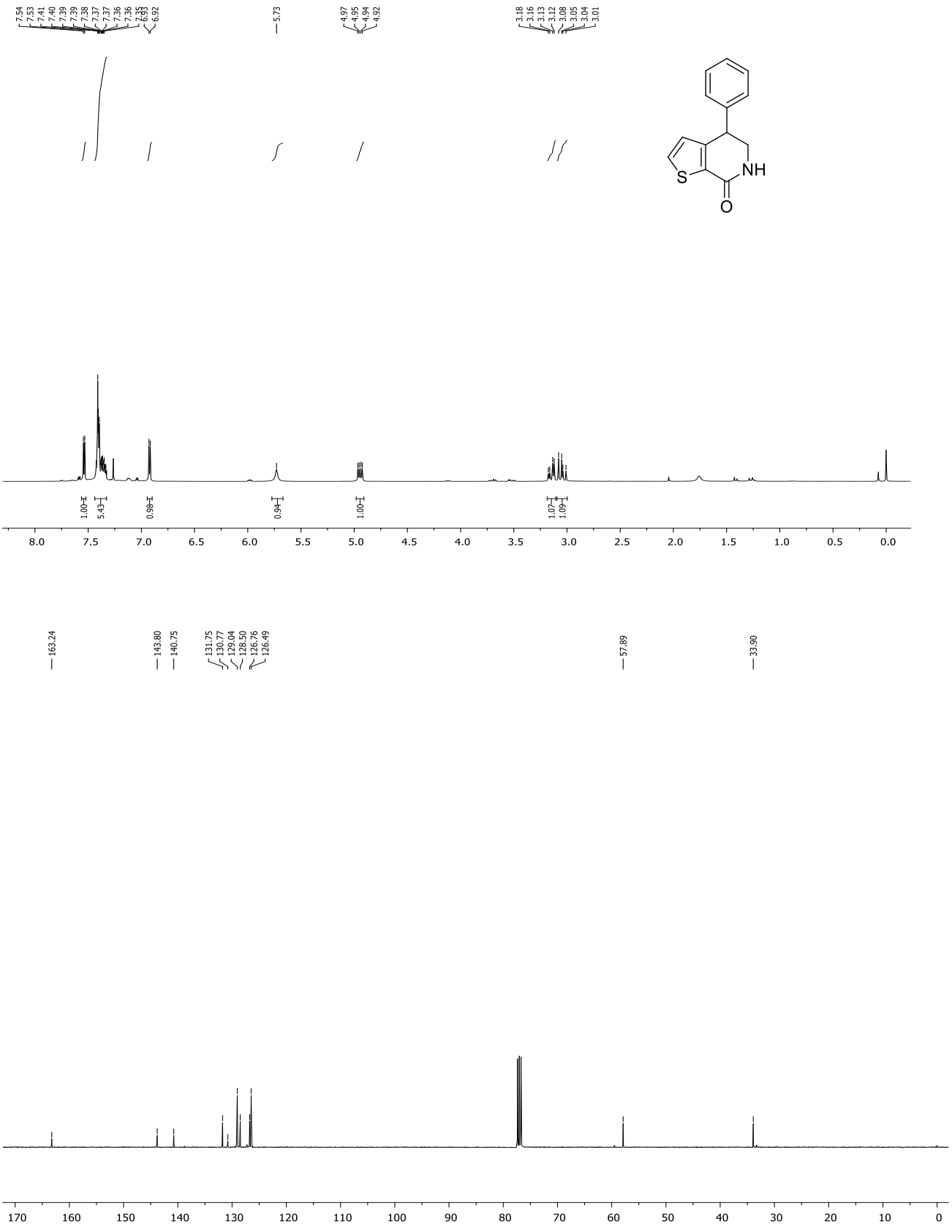
^{1}H and ^{13}C NMR of compound **7-(2-Chloro-4-fluorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (55)** in $CDCl_3$



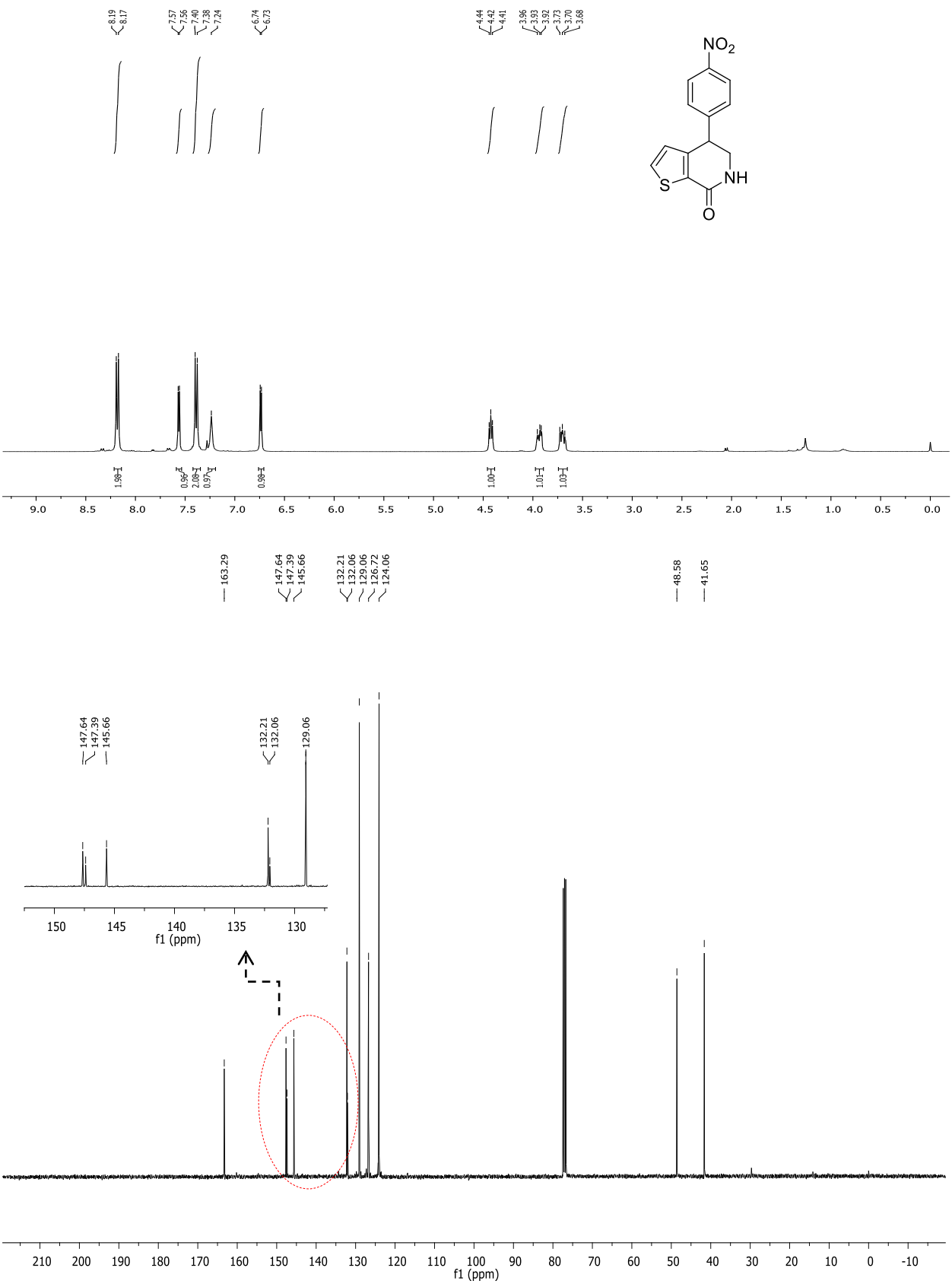
^{1}H and ^{13}C NMR of compound **7-(2,3-Dichlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (56)** in $CDCl_3$



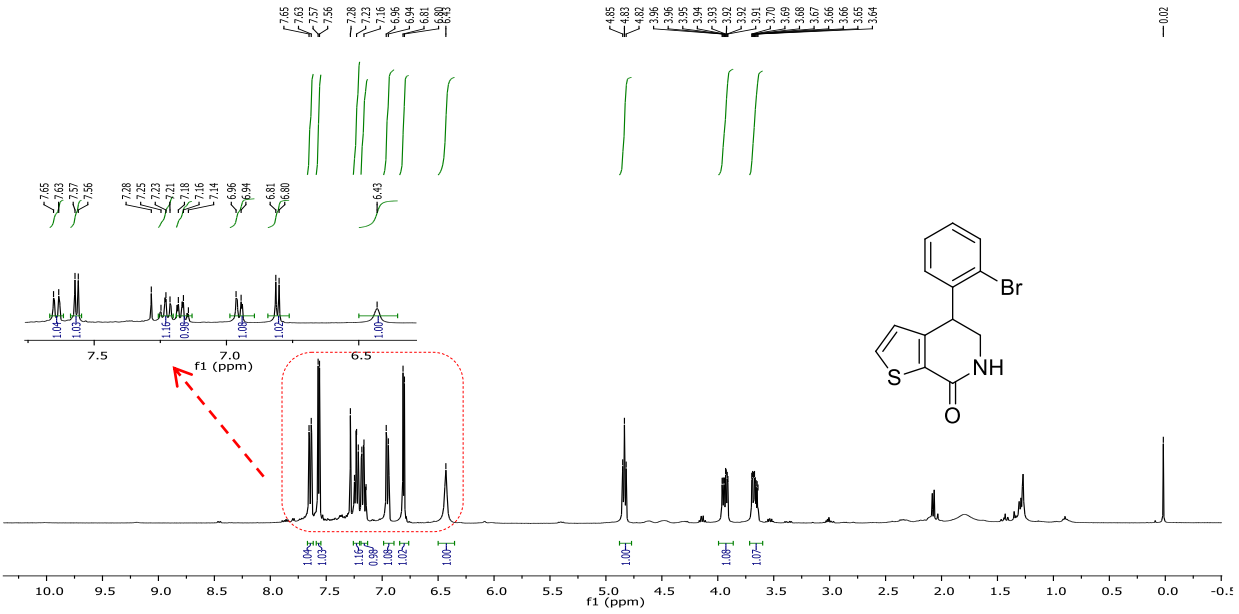
^{1}H and ^{13}C NMR of compound **7-(2,6-Dichlorophenyl)-6,7-dihydrothieno[3,2-*c*]pyridin-4(5*H*)-one (57)** in $CDCl_3$

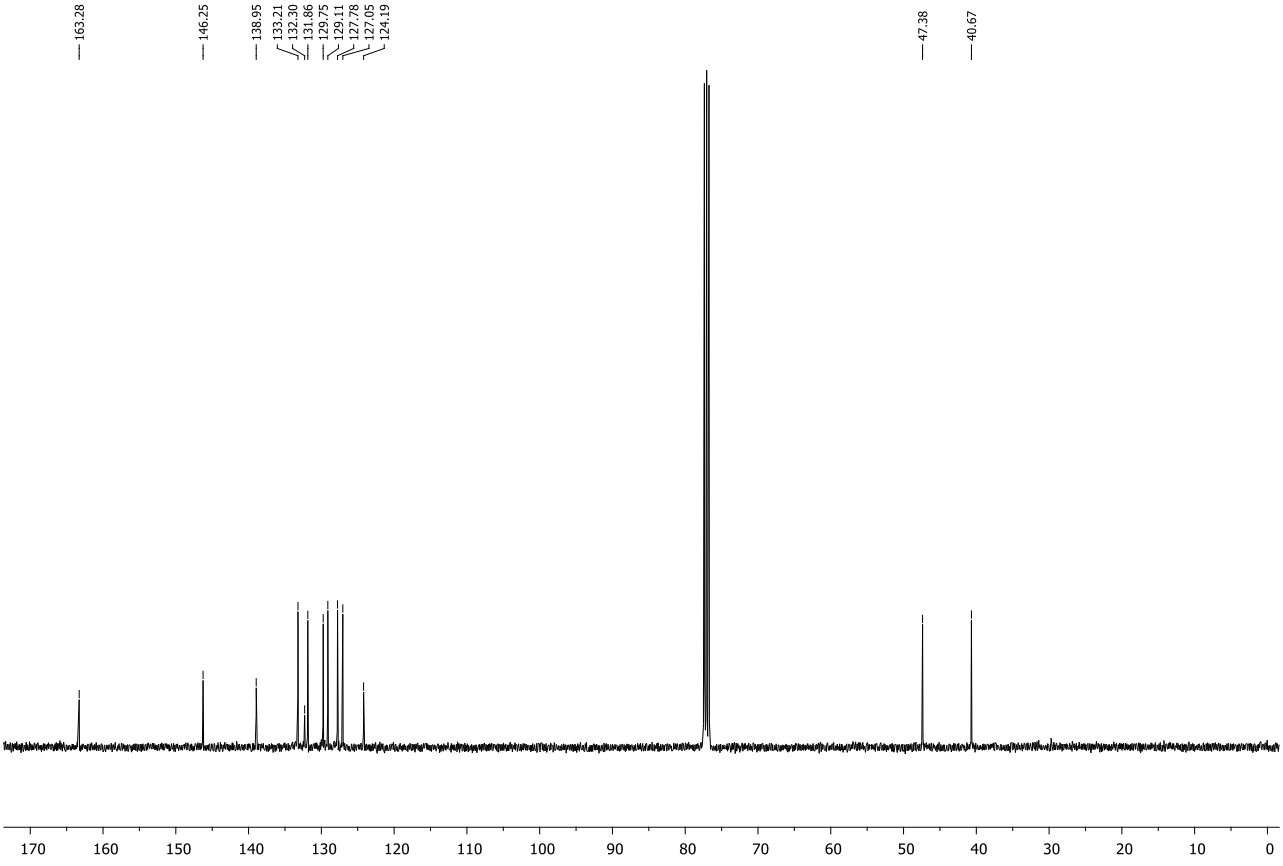


^{1}H and ^{13}C NMR of compound **4-Phenyl-5,6-dihydrothieno[2,3-c]pyridin-7(4H)-one (58)** in $CDCl_3$

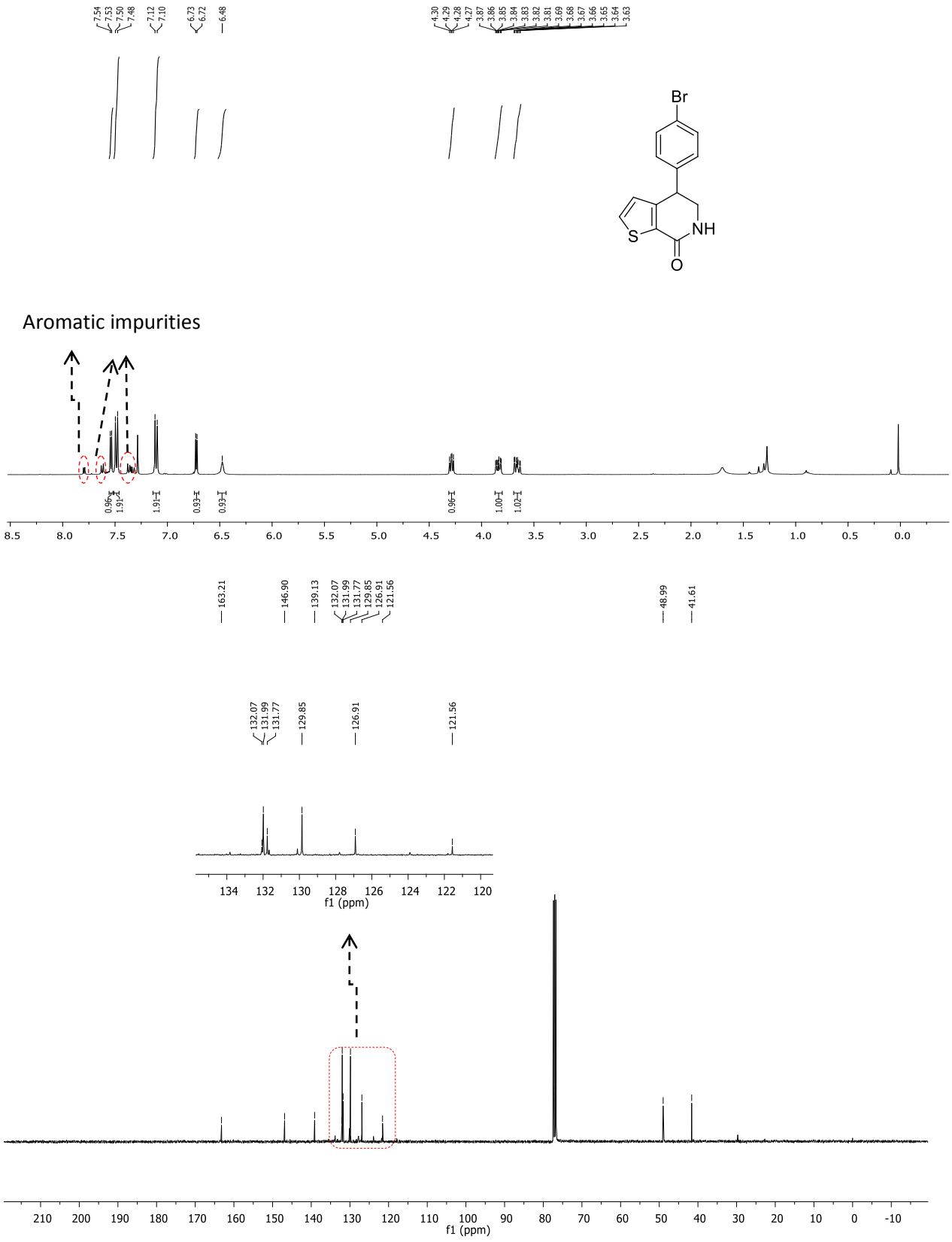


^{1}H and ^{13}C NMR of compound **4-(4-Nitrophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (59)** in $CDCl_3$

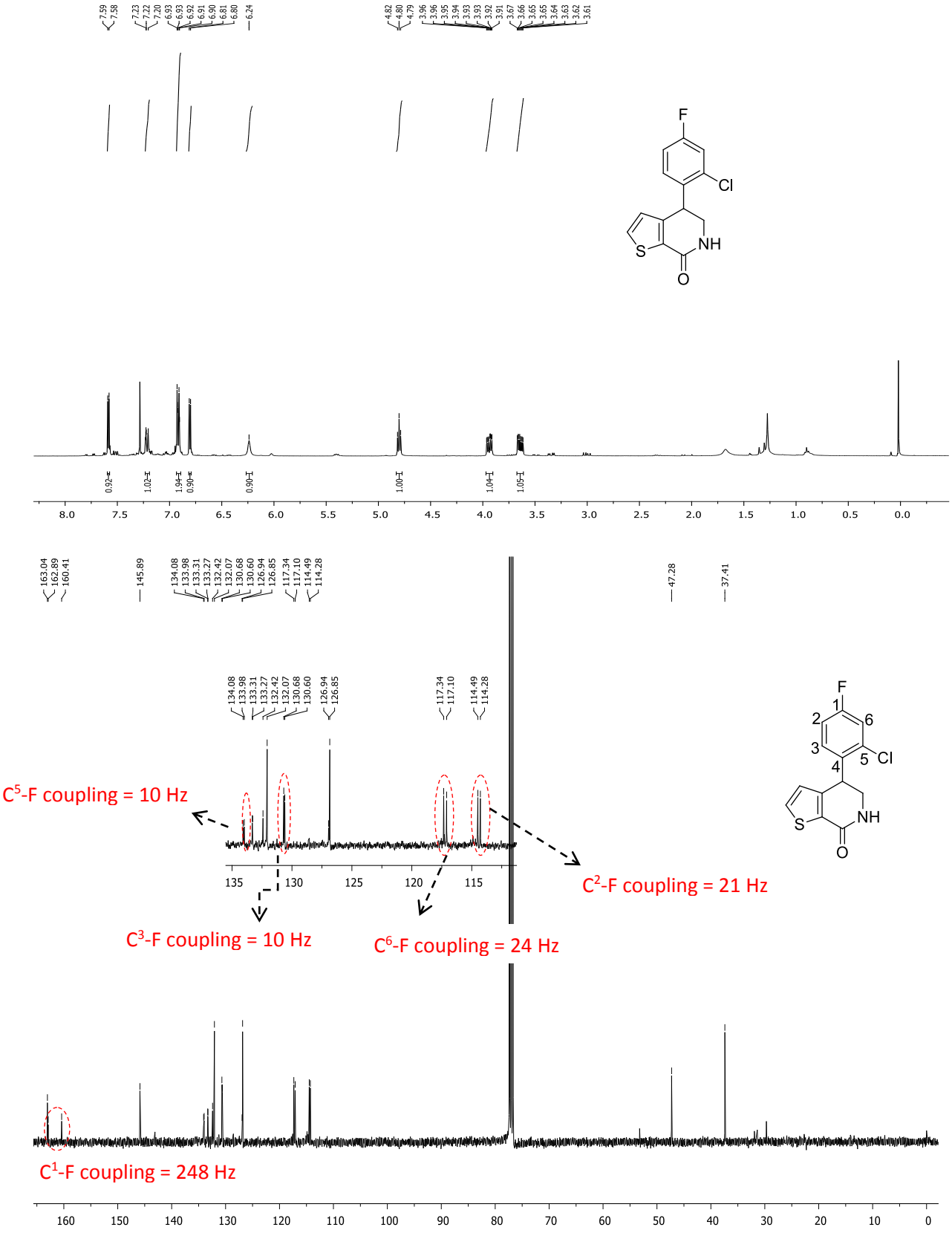




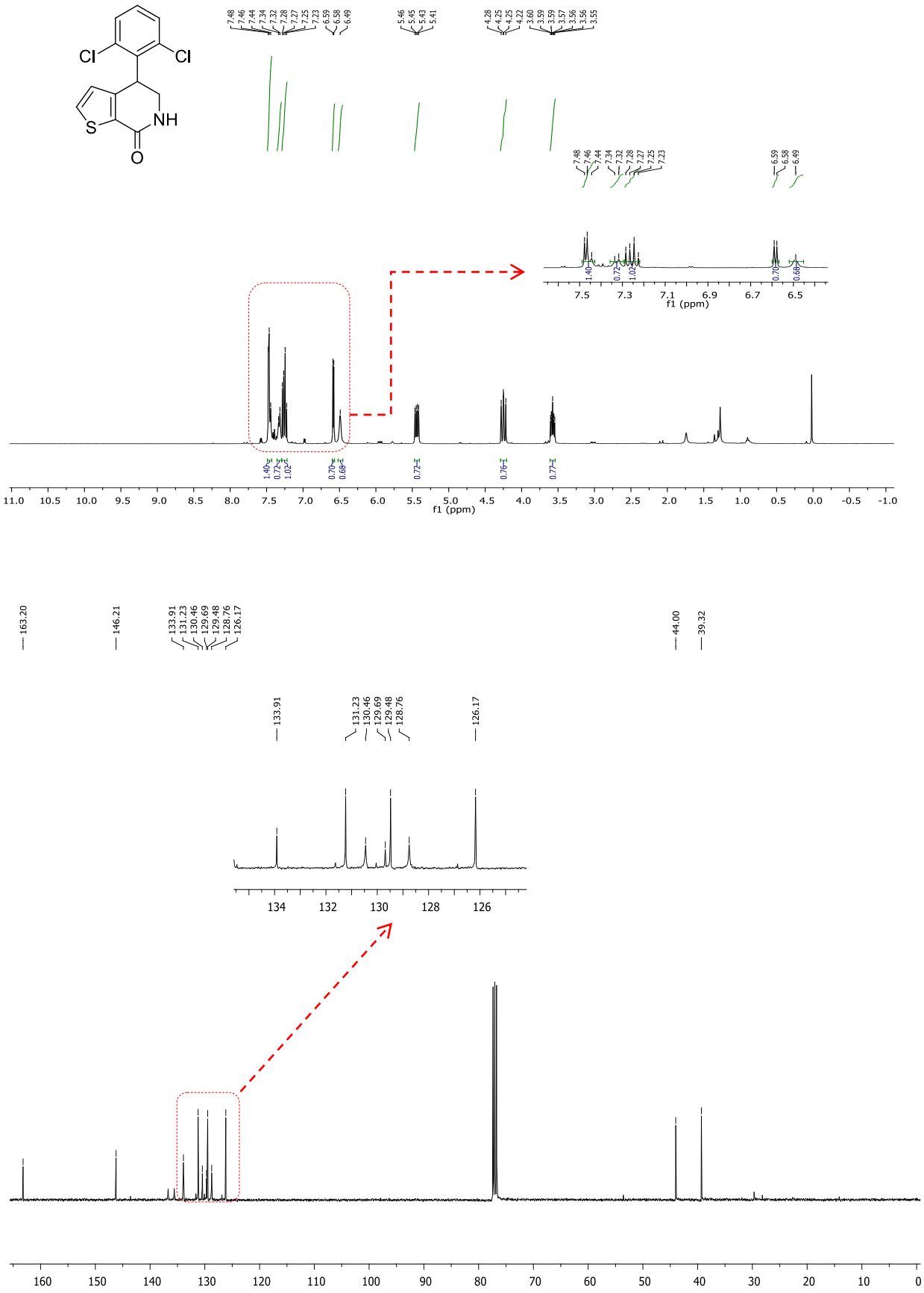
^{1}H and ^{13}C NMR of compound **4-(2-Bromophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (60)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **4-(4-Bromophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (61)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **4-(2-Chloro-4-fluorophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (62)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **4-(2,6-Dichlorophenyl)-5,6-dihydrothieno[2,3-*c*]pyridin-7(4*H*)-one (63)** in $CDCl_3$

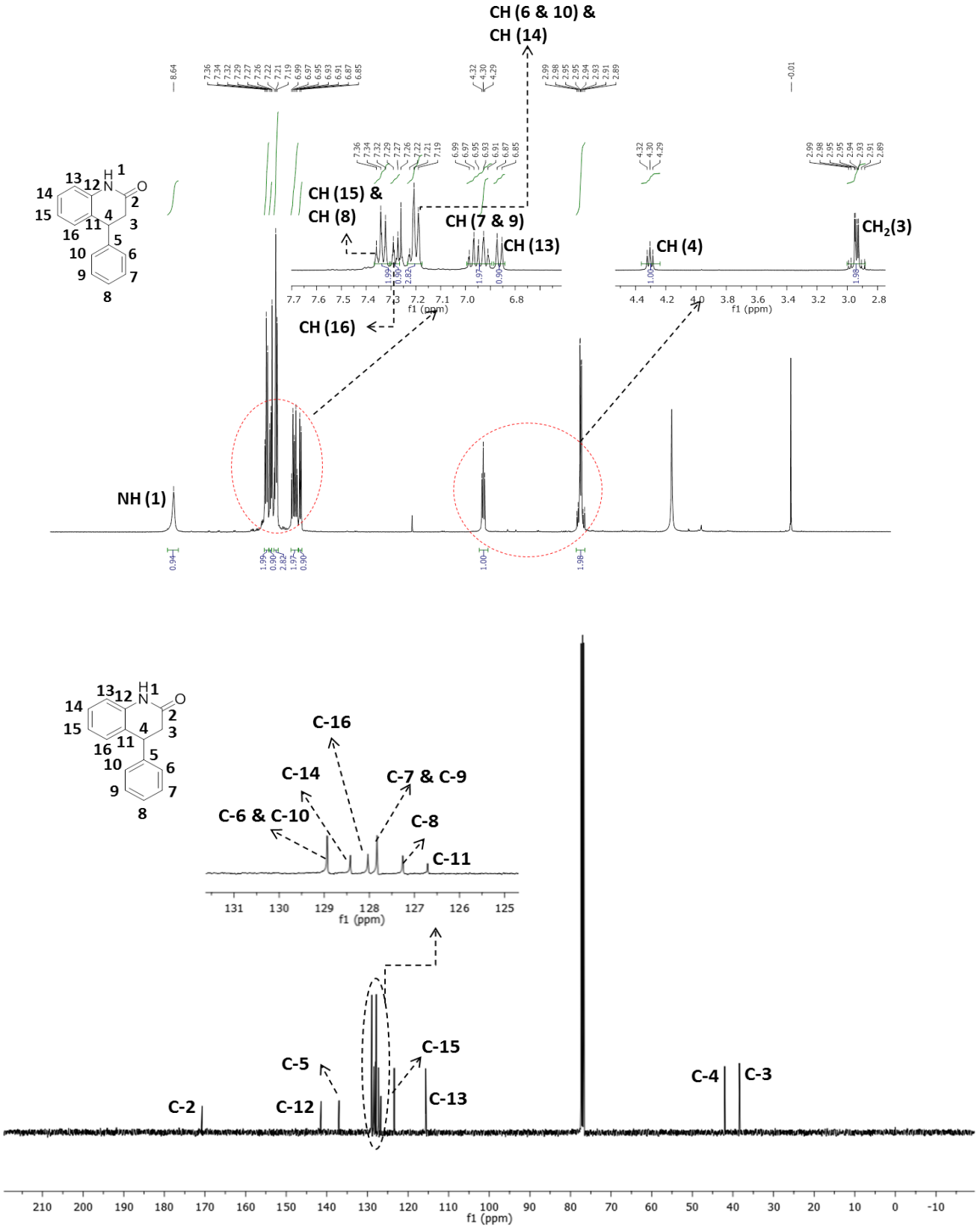
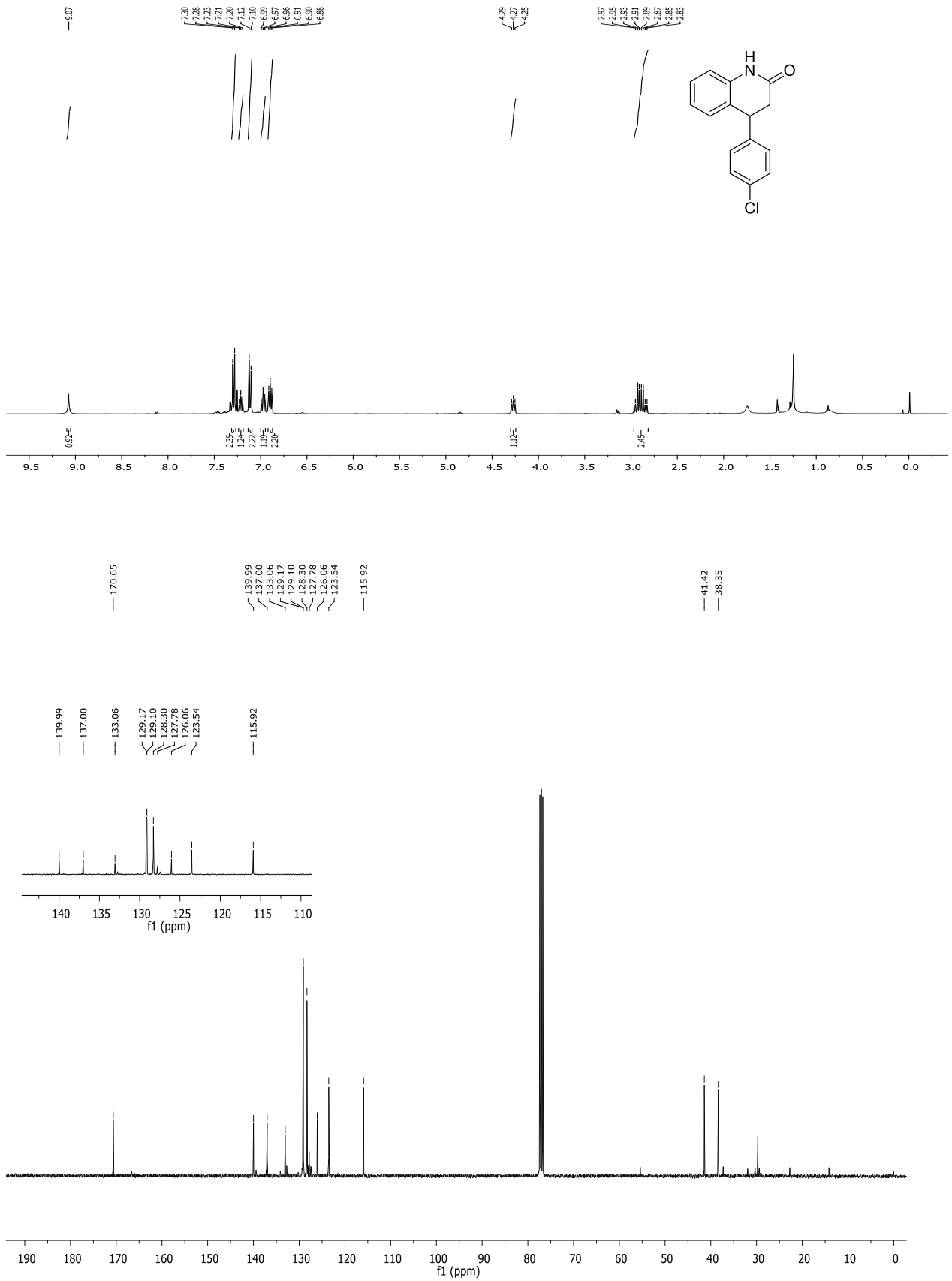
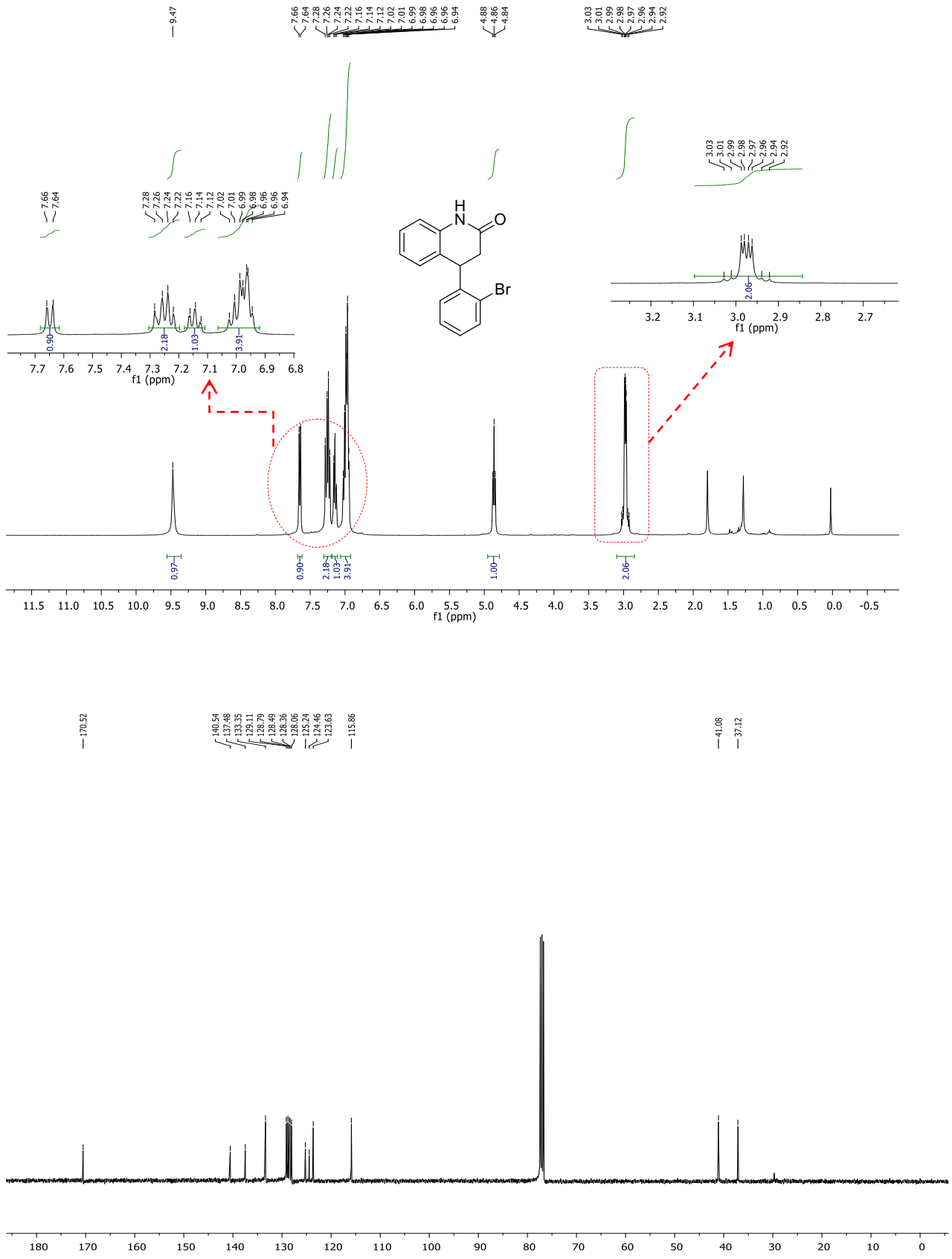


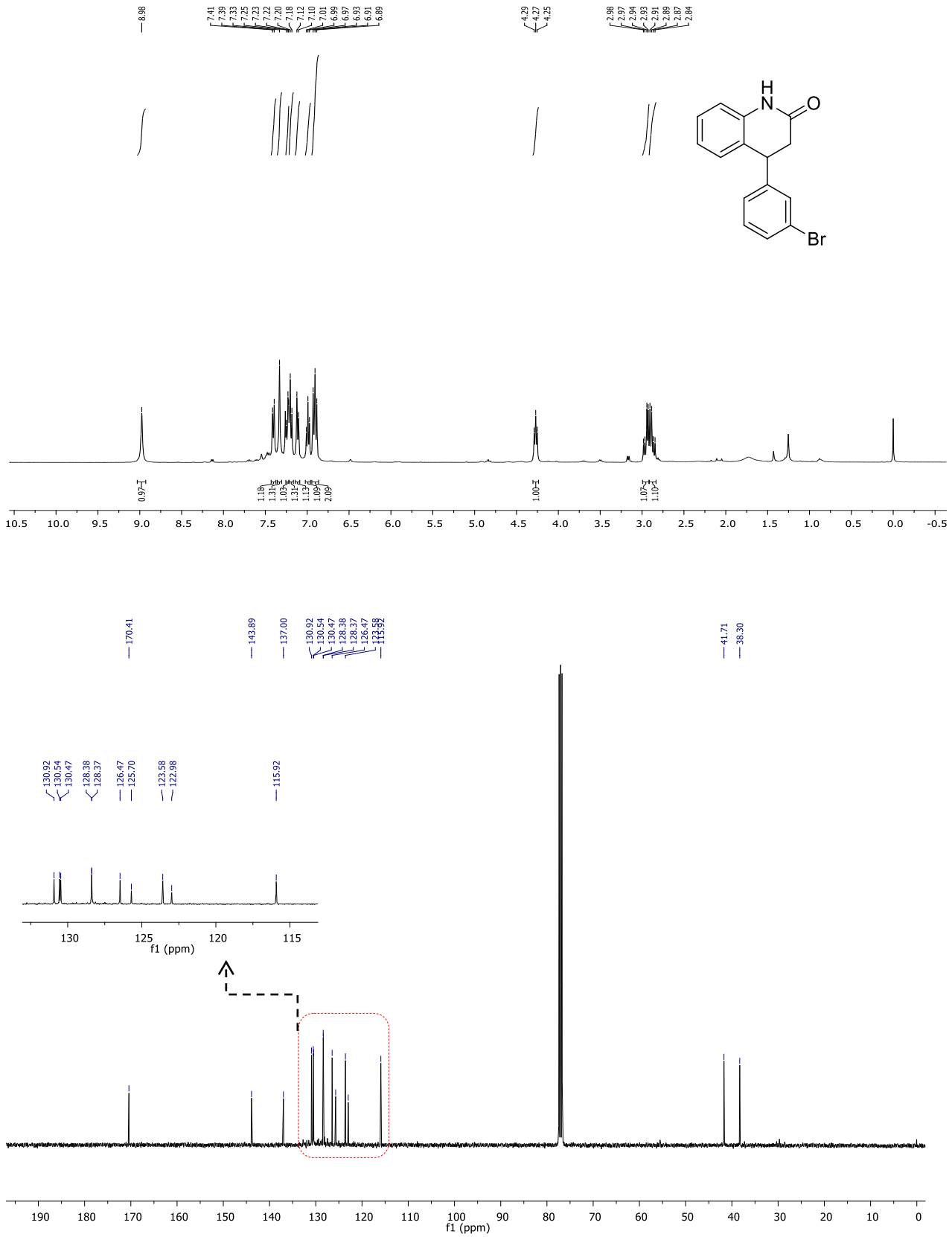
Figure 2.5: ^{1}H and ^{13}C NMR of compound **4-Phenyl-3, 4-dihydroquinolin-2(1*H*)-one (17)** in $CDCl_3$



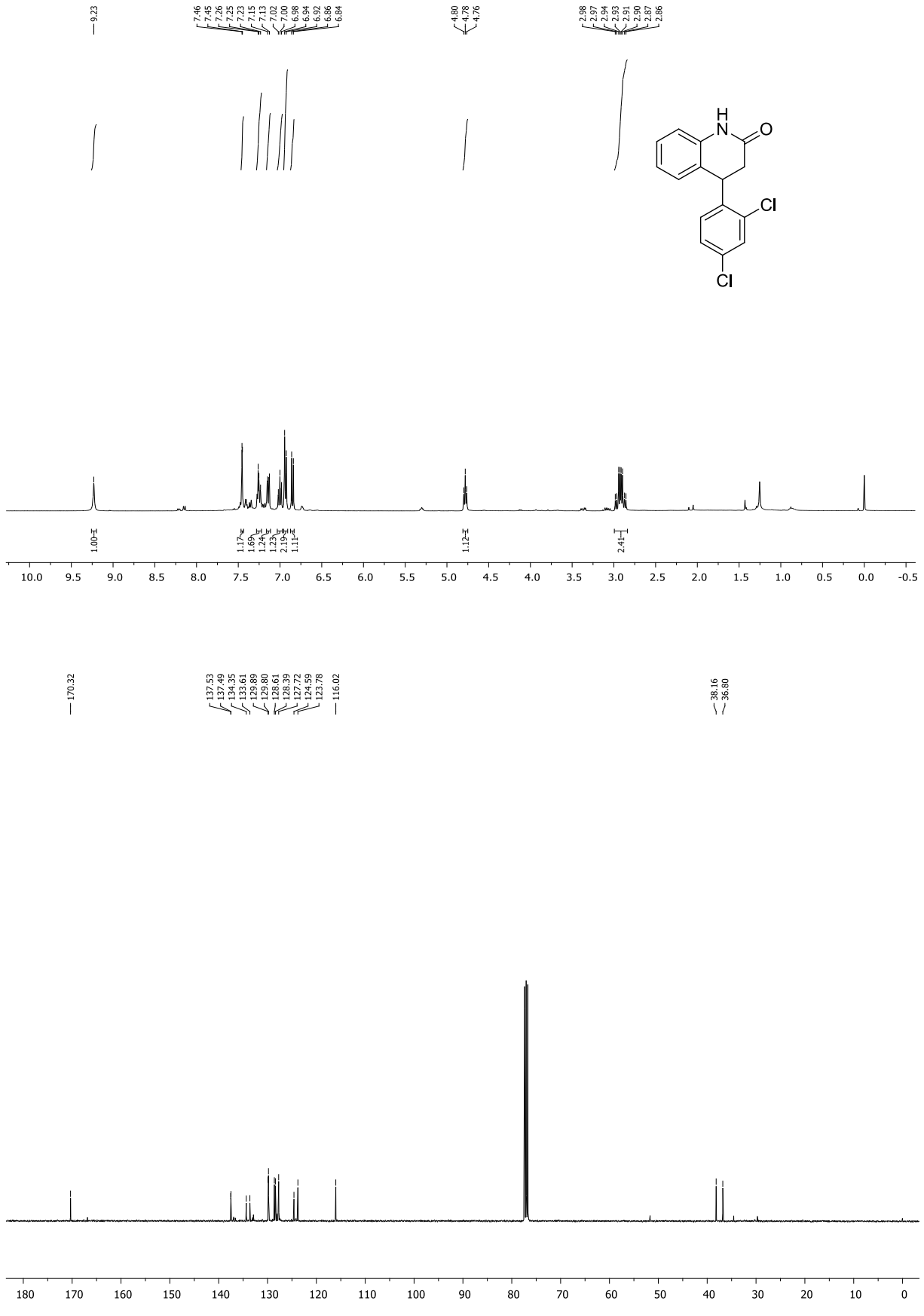
^{1}H and ^{13}C NMR of compound **4-(4-Chlorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (64)** in $CDCl_3$



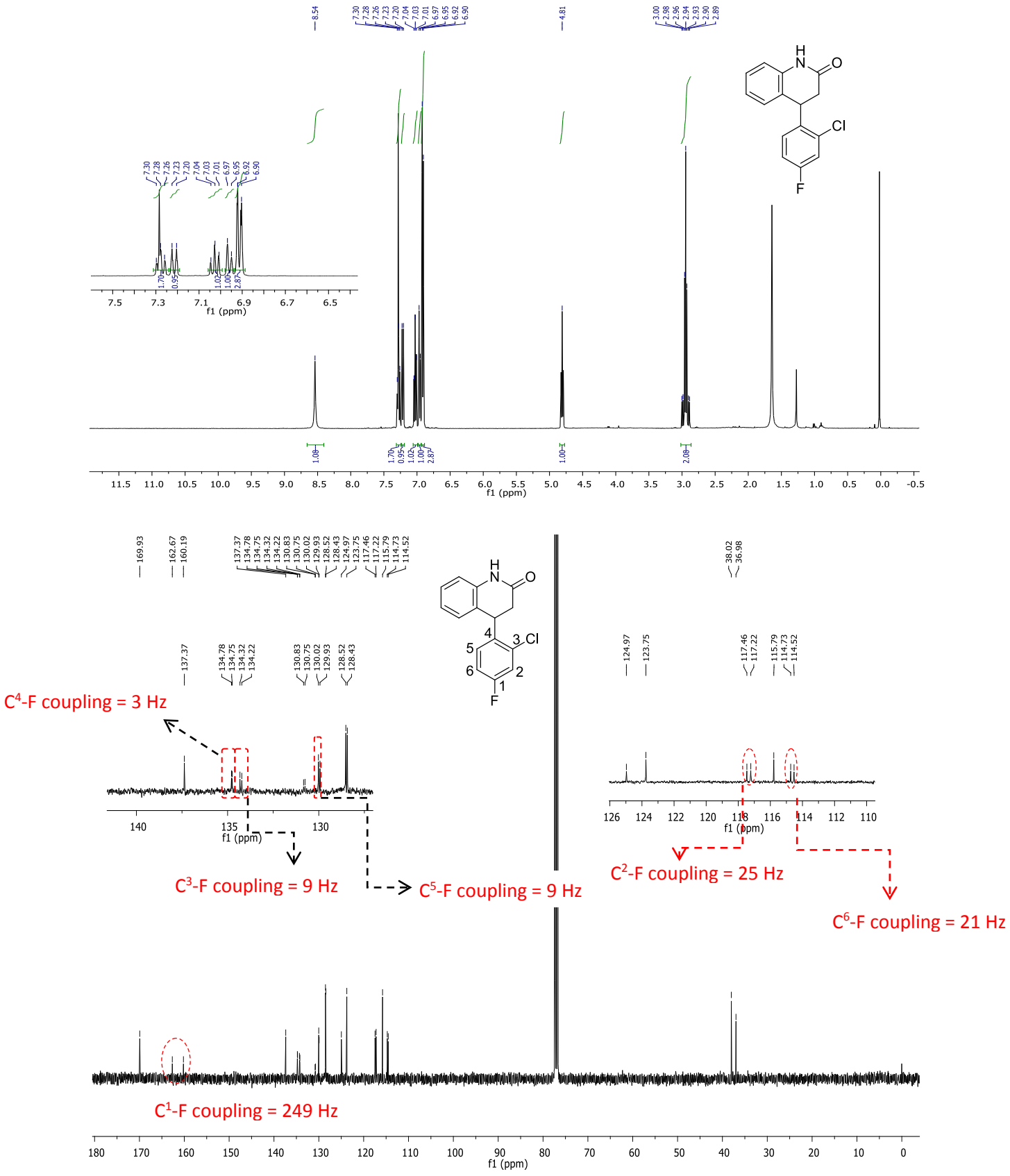
^{1}H and ^{13}C NMR of compound **4-(2-Bromophenyl)-3,4-dihydroquinolin-2(1*H*)-one (65)** in $CDCl_3$



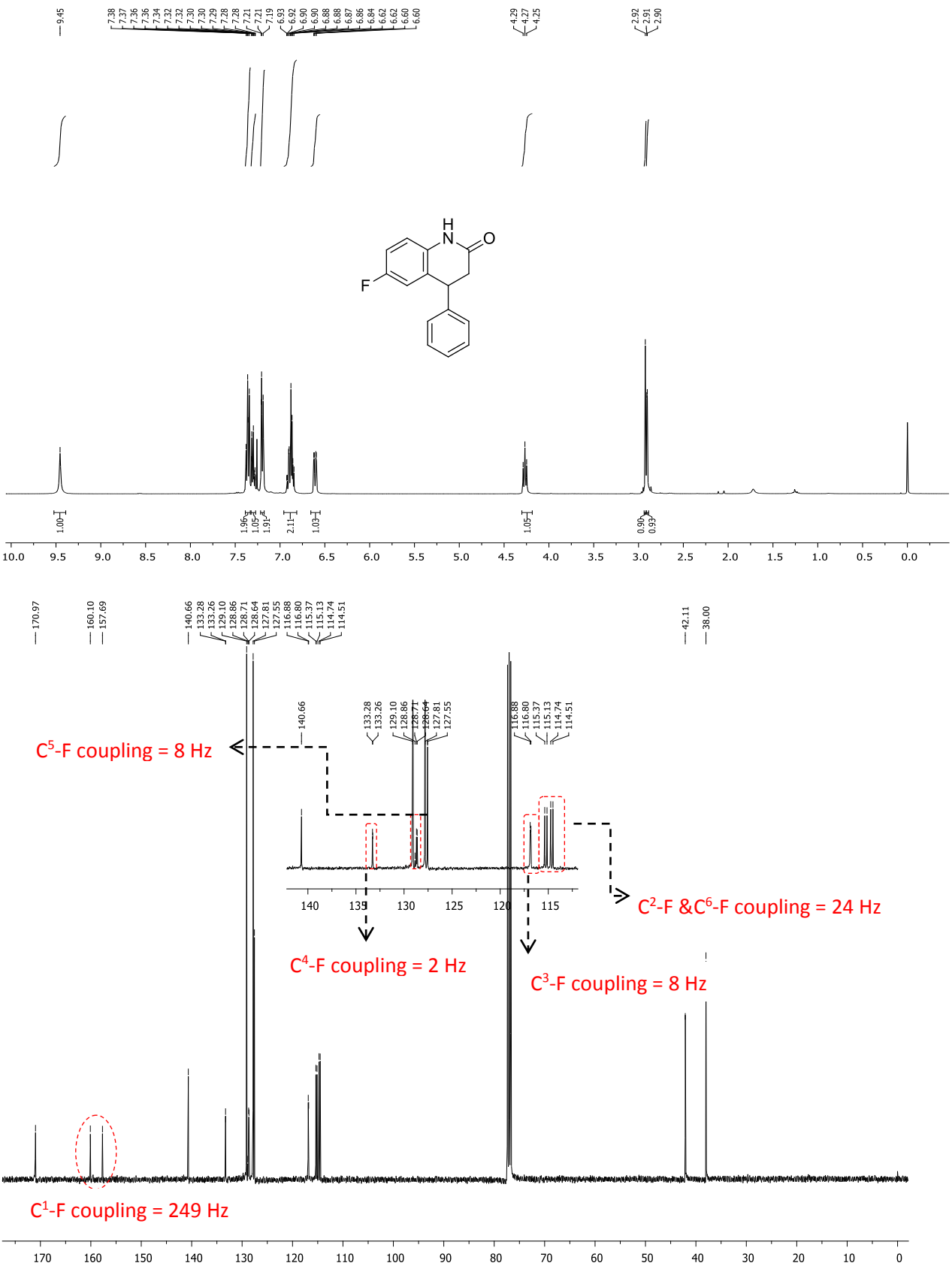
^{1}H and ^{13}C NMR of compound **4-(3-Bromophenyl)-3,4-dihydroquinolin-2(1*H*)-one (66)** in $CDCl_3$



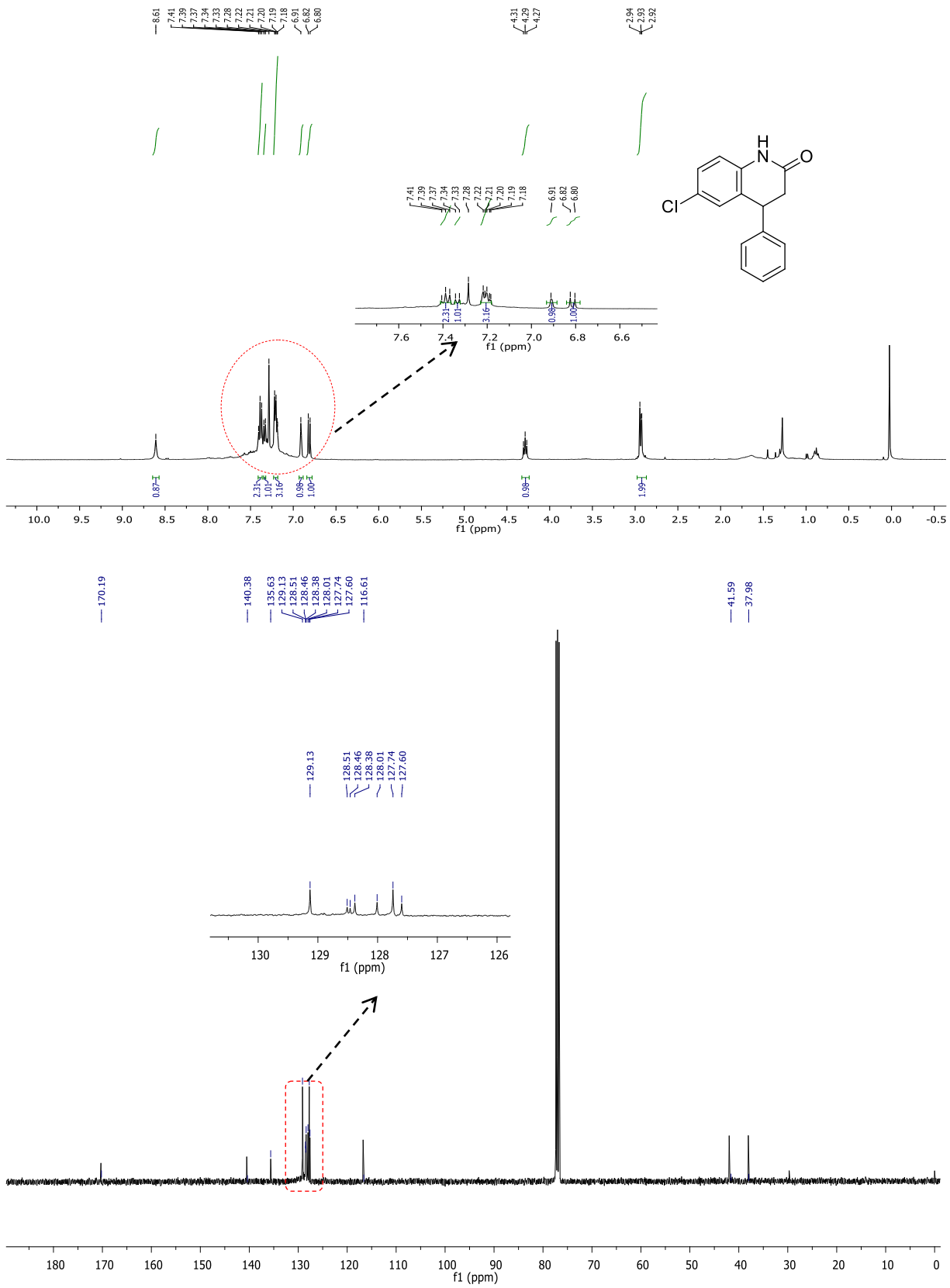
^{1}H and ^{13}C NMR of compound **4-(2,4-Dichlorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (64)** in $CDCl_3$



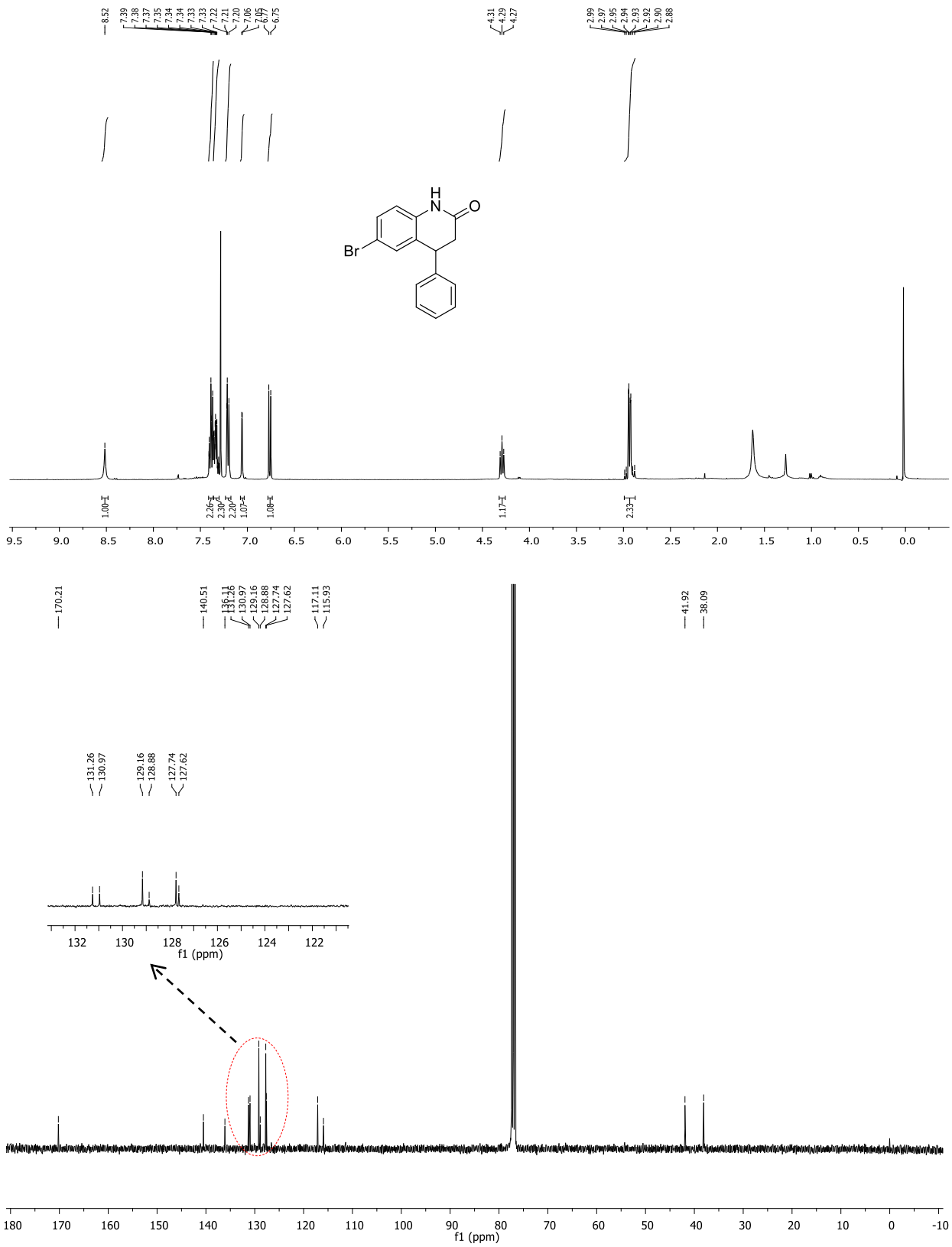
^{1}H and ^{13}C NMR of compound **4-(2-Chloro-4-fluorophenyl)-3,4-dihydroquinolin-2(1*H*)-one (68)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **6-Fluoro-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (69)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **6-Chloro-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (70)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **6-Bromo-4-phenyl-3,4-dihydroquinolin-2(1*H*)-one (71)** in $CDCl_3$

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CHAPTER 3

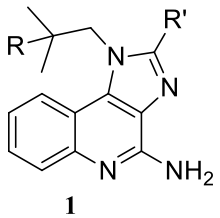
Synthesis and anticancer activity of 1, 4-disubstituted imidazo[4,5-*c*]quinolines

3.1. Introduction

Synthesis of fused heterocycles bearing structurally different units remains an important challenge in the domain of organic chemistry. Fused systems are not only bestowed with properties due to their respective structural units, but also acquire additional attributes owing to changed electronic environment. Most frequently encountered examples in the area of fused heterocycles are indole, quinoline and isoquinoline. Compounds containing these fused heterocyclic units are known to possess diverse biological and physical properties.[1-5]

Since the last few years our group has been actively involved in exploring the area of fused quinoline based heterocyclic systems. Towards this endeavour we have reported the syntheses and applications of structural motifs containing fused quinoline-pyrrole, quinoline-thiophene, quinoline-furan and quinoline-oxazole based systems.[6-10] Fused quinoline-pyrrole compounds exhibited anti-tubercular activity,[6] additionally selective Zn^{2+} and F^- sensing was also displayed by two of the analogues.[8,9] In continuation of our efforts, herein, we wish to describe our findings in the synthesis of 1,4-disubstituted imidazo[4,5-*c*]quinolines using modified Pictet-Spengler approach.

Fused quinoline and imidazole based systems are well known for their multitude of activities.[10-16] Prominent examples include imiquimod and its structural analogue resiquimod, a prescription medicine used as immune response modifier **(Figure 3.1)**.[17-18]



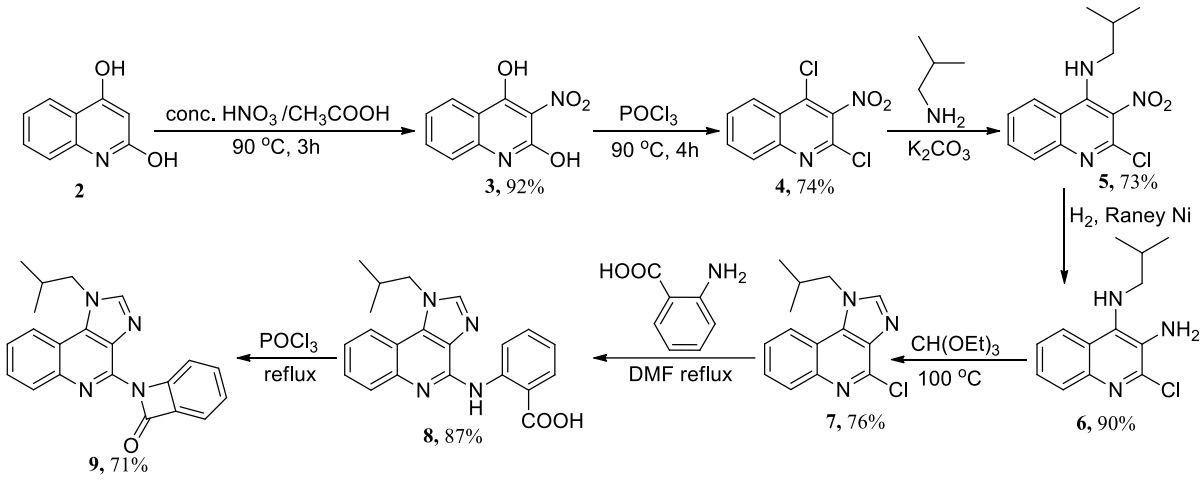
1

R = R' = H (Imiquimod) 1a
R = OH; R' = $CH_2OCH_2CH_3$ (Resiquimod) 1b

Figure 3.1: Chemical structures of imiquimod and resiquimod

Most common strategy observed in literature pertaining to the synthesis of imidazo[4,5-*c*]quinolines involve reaction on 3,4-disubstituted quinoline to form the imidazole ring.[19-21] Importance of this strategy stems from the fact that formation of imidazole prior to quinoline ring will require reaction at less reactive C-4 position, thus methods used generally avoid this difficult step.

In a work reported by Kayamar *et al.*, a series of 7-(1-isobutyl-1*H*-imidazo[4,5-*c*]quinolin-4-yl)-7-azabicyclo[4.2.0]octa-1,3,5-trien-8-ones were synthesized starting from2,4-dihydroxyquinoline **(2)** by following a multistep protocol.[20] Key step involved was synthesis of 4-chloro-1-isobutyl-1*H*-imidazo[4,5-*c*]quinoline **(7)** by reaction between 2-chloro-N^4-isobutylquinoline-3,4-diamine **(6)** and triethoxy methane **(Scheme-3.1)**.

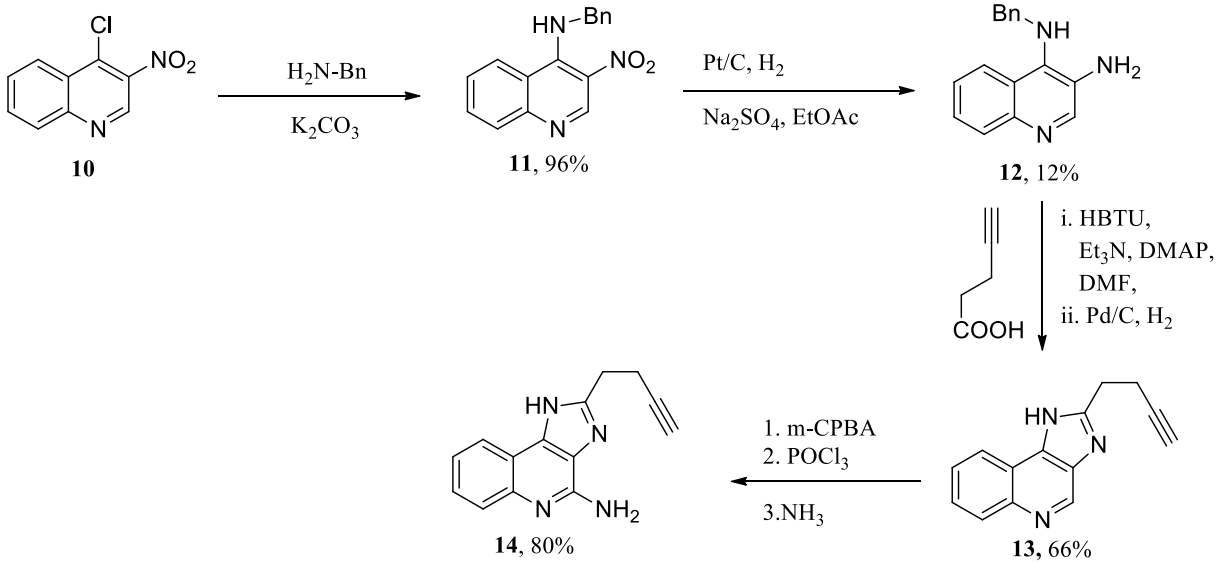


Scheme 3.1: Synthesis of imidazo[4,5-*c*]quinoline system by Kayamar *et al.*

David and co-workers in a separate work explored the synthesis of 1-benzyl-2-(but-3-ynyl)-1*H*-imidazo[4,5-*c*]quinolin-4-amine and related molecules.[21] The synthesis started from 4-chloro-3-nitroquinoline **(10)**, which was converted to *N*-benzyl-3-nitroquinolin-4-amine **(11)** by treatment with benzylamine. Further, nitro group present in molecule **11** was reduced using catalytic amount of Pt/C and Na_2SO_4 to generate N^4-benzylquinoline-3,4-diamine **(12)**. Compound **12** was subsequently treated with pentynoic acid in presence of HBTU,

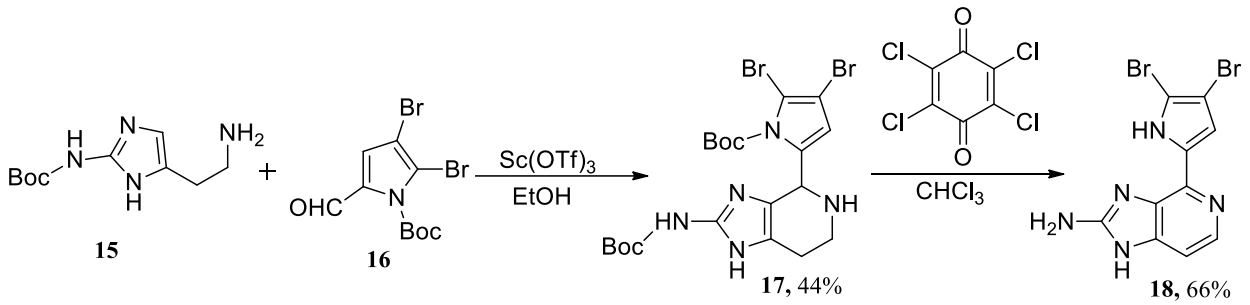
triethylamine and catalytic amount of DMAP to form 1-benzyl-2-(but-3-ynyl)-1*H*-imidazo-[4,5-*c*]quinolin-4-amine **(13)**.In the final step compound **13** was converted to 1-benzyl-2-(but-3-ynyl)-1*H*-imidazo[4,5-*c*]quinolin-4-amine **(14)** by treatment with 3-chloroperbenzoic acid for making quinoline N-oxide on **13**, then it was treated with $POCl_3$ for making a 4-chloro substituted imidazo[4,5-*c*]quinoline derivative and it was reacted with ammonia to get a target molecule **14**.

Both the above mentioned strategies relied mainly on formation of imidazole ring as the last step while attempting synthesis of imidazo[4,5-*c*]quinolines.



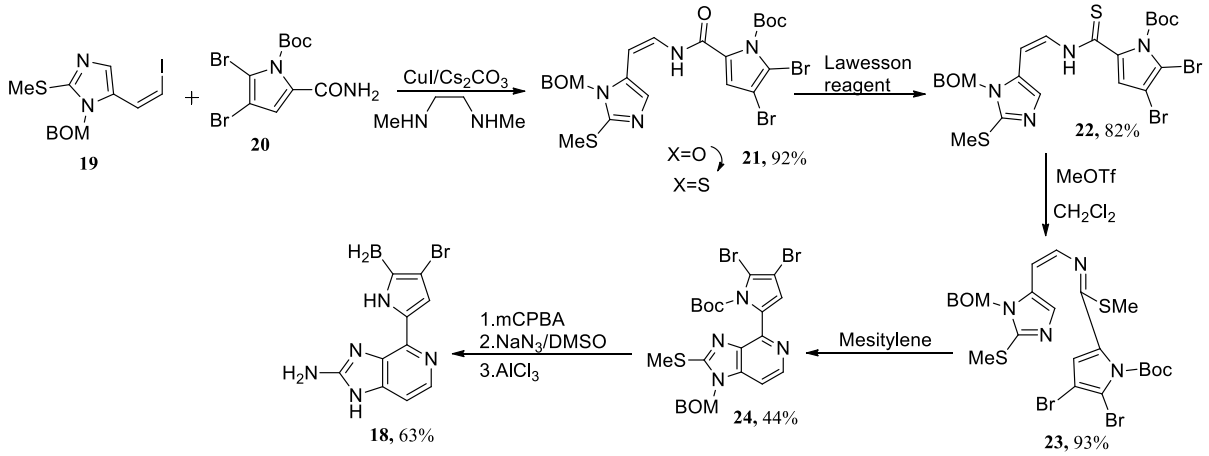
Scheme 3.2: Synthesis of imidazo[4,5-*c*]quinoline system by Shukla *et al.*

However, examples are known in literature involving reaction at C-4 position of imidazole ring during formation of fused imidazo-pyridine system.[22,23] While attempting synthesis of natural product ageladine A (isolated from marine sponge *Agelasnakamurai*), Shengule *et al*., successfully carried out a transformation at C-4 position of imidazole **(17)** using $Sc(OTf)_3$ in moderate yields **(Scheme 3.3)**.[22]



Scheme 3: Shengule *et al.* synthesis of Ageladine A.

In another attempt, Meketa *et al*., used 6π-2-azatriene electrocyclization to functionalize imidazole's C-4 position to accomplish the synthesis of the same natural product.[23] Interestingly though, both the reactions were carried out on imidazoles where the more reactive C-2 position was already blocked **(Scheme 3.4)**.



Scheme 3.4: Meketa *et al.* synthesis of Ageladine A.

Thus, in order to devise a more versatile approach at access imidazoquinolinesour endeavour was focused towards the functionalization of C-4 position of imidazole, *en-route* to 1,4-disubstitutedimidazo[4,5-*c*]quinolines, while leaving its C-2 position completely unsubstituted.

Literature reports also indicate protein kinase inhibition by imidazo[4,5-*c*]quinolines.[24] Protein kinases are usually found to be involved in various pathological

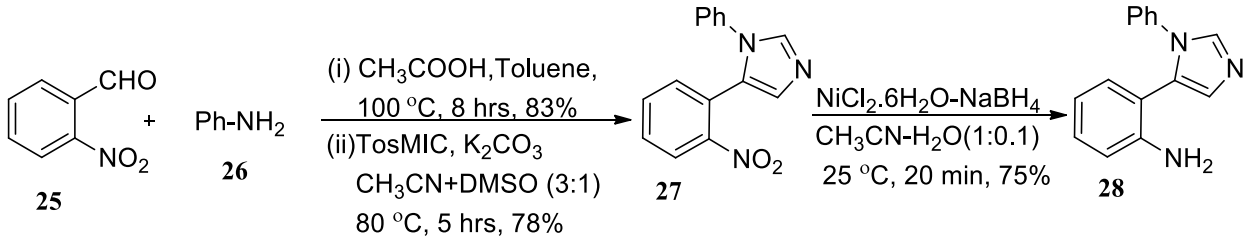
processes specifically malignancies. Different protein kinases have been found to be deregulated in chronic myelogenous leukaemia, gastrointestinal stromal tumours, various other sarcomas and cancers as well as non-malignant disorders. [25] Hence, the exploration of anticancer activity of the target molecules in the current study was amply warranted.

3.2. Current work

In this chapter, a detailed discussion of a four step synthetic strategy leading to synthesis of 1,4-disubstituted imidazo[4,5-*c*]quinolines *via* modified Pictet-Spengler approach starting from 2-nitrobenzaldehyde (**25**) is presented. These newly created fused quinolines were subsequently evaluated for anticancer activity in collaboration with the research group of Dr. Balaram Ghosh (Department of Pharmacy, BITS-Pilani-Hyderabad campus).

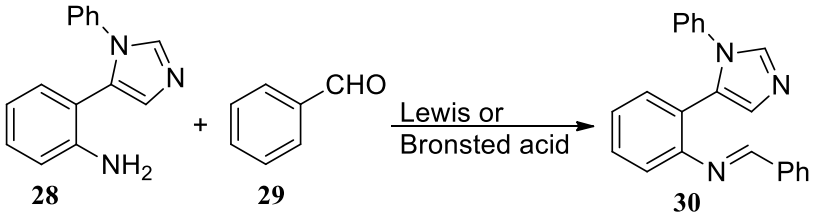
3.3. Results and discussion

Synthesis of the target molecules was designed starting from 2-nitrobenzaldehyde (**25)**. It involved usage of van Leusen's imidazole synthesis[26-27] and subsequent application of modified Pictet-Spengler synthesis. We began our quest for the synthesis of imidazo[4,5-*c*]quinolines by first preparing the intermediate **28** using 2-nitrobenzaldehyde and aniline (**25)** as reactants (**Scheme-3.5**). The condensation of compound **25** with aniline **(26)** resulted in an imine intermediate,[28-29] which upon treatment with TosMIC(toluenesulfonylmethylisocyanide) in presence of potassium carbonate gave rise to N-1 substituted imidazole **(27)**.[26-27] Next, the reduction of the nitro functionality present in **27** was effected using mixed $NiCl_2$-$NaBH_4$ system to obtain the crucial intermediate **28** of our designed synthesis **(Scheme 3.5)**.[30]



Scheme 3.5: Synthesis of 2-(1-phenyl-1*H*-imidazol-5-yl)aniline (**28)**

In subsequent studies, the compound **28** and benzaldehyde **(29)** were used as model reactants to optimize the conditions for the desired condensation-cyclization sequence leading to the framework of compound **31**. Based on our prior experience,[7] initial reactions were attempted with various Lewis and Bronsted acids and using methanol as solvent **(Scheme 3.6)**. However, to our disappointment, the reaction resulted in the formation of only the corresponding imine **30** in 60-74% yields, under these conditions. Further increment in the reaction temperature 140 °C (in xylene) was also found to be futile. These results corroborate our assumption about the low reactivity of the C-4 position of imidazole towards functionalization.



Scheme 3.6: Proposed synthesis of 1,4-disubstituted imidazo[4,5-*c*]quinolines using various Lewis and Bronsted acids

In order to address the challenge posed by the low reactivity of the C-4 position we expanded our investigation to screen a range of Lewis acids at elevated temperature (150 °C) to produce imidazo-quinoline ring in one step **(Table 3.1)**. Furthermore, the solvent was also changed to nitrobenzene because of its oxidizing nature,[7] which might facilitate the desired

oxidative cyclization step. To our delight, the reaction attempted using $Cu(TFA)_2$ resulted in required product formation in 46% yield **(entry 1; Table 3.1),** the authenticity of which was established using spectroscopic data. In ^1H NMR spectrum of compound **31**, disappearance of C-4 proton (at δ=7.26 ppm) from imidazole ring clearly indicated functionalization of the stated site **(Figure 3.2)**. It was further evident by a downfield shift of C-2 and C-6 protons owing to de-shielding effect of the newly formed quinoline ring. The doublet at δ 8.71 ppm for C-2' and C-6' protons, established association of benzene ring at C-4 position of the fused imidazole-quinoline ring. Disappearance of N-H stretching frequencies in IR spectrum between 3200-3400 cm^{-1}, was also found to be in agreement with the proposed cyclization and aromatization leading to compound **31**. Finally, the high resolution mass spectrum (HRMS) data of **31** was found to be 322.1328 $[M + H],^+$ which unambiguously helped us to assign the structure as 1,4-diphenyl-1*H*-imidazo[4,5-*c*]quinoline.

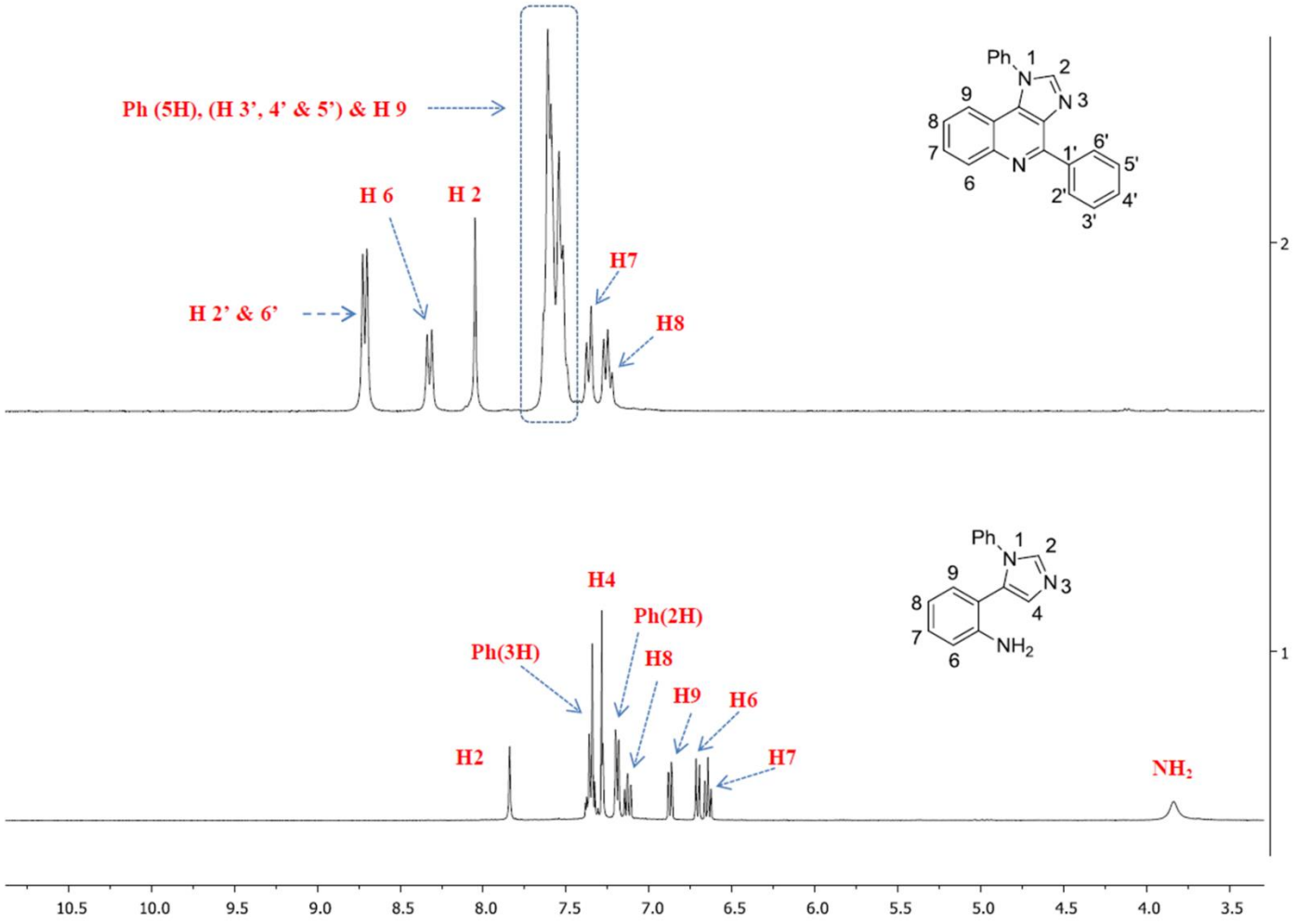
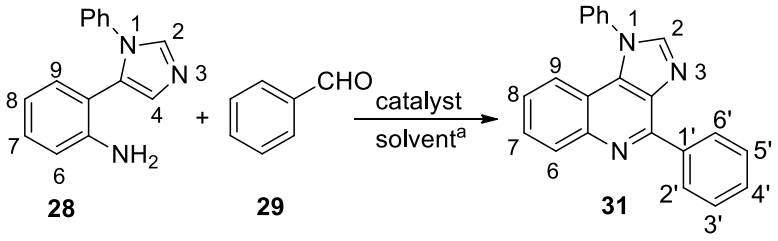


Figure 3.2: Overlay of ^{1}H NMR spectra of 1,4-diphenyl-1*H*-imidazo[4,5-*c*]quinoline **(31)** and 2-(1-phenyl-1*H*-imidazol-5-yl)aniline **(28)**

Once the isolated product was fully characterized we proceeded with optimizing other reaction parameters. Interestingly, when the same reaction was attempted by using DPE (diphenyl ether) as solvent, instead of nitrobenzene, no products were formed **(entry 2; Table 3.1)** under similar conditions. Further screening with other potential catalysts using nitrobenzene as solvent led to $Yb(OTf)_3$ as being the most efficient catalyst for the desired transformation,yielding **31** in 73% **(entry 12; Table 3.1)**. When $Yb(OTf)_3$ was used in 10 mol% and 30 mol% **(entry 15-16; Table 3.1)**, yields obtained were 47% and 72%, respectively. Based on the above results, we established the use of $Yb(OTf)_3$ at 20 mol% in

nitrobenzene at 150 °C as the best conditions for the formation of fused quinolines **31** from amine **28**.

Table 3.1: Screening of catalysts for synthesis of quinoline ring



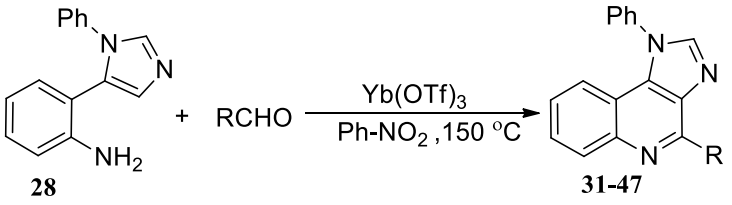
S.No.	Catalyst (0.2 mol%)	Yield(%)[b]
1	$Cu(TFA)_2$	46
2	$Cu(TFA)_2$/DPE[c]	0
3	$Cu(OTf)_2$	41
4	$Cu(OAc)_2$	0
5	CuI	0
6	$Pd(OAc)_2$	0
7	$PdCl_2(TPP)_2$	42
8	$Pd(TFA)_2$	51
9	$AlCl_3$	50
10	$ZnCl_2$	63
11	$FeCl_3$	60
12	$Yb(OTf)_3$	73
13	PTSA	50
14	$Fe(acac)_3$	0
15[d]	$Yb(OTf)_3$	47
16[e]	$Yb(OTf)_3$	72

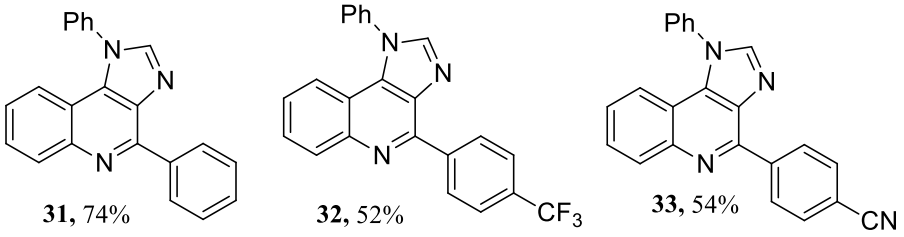
[a] Nitrobenzene is used as solvent except in entry 2; [b] Isolated yields;

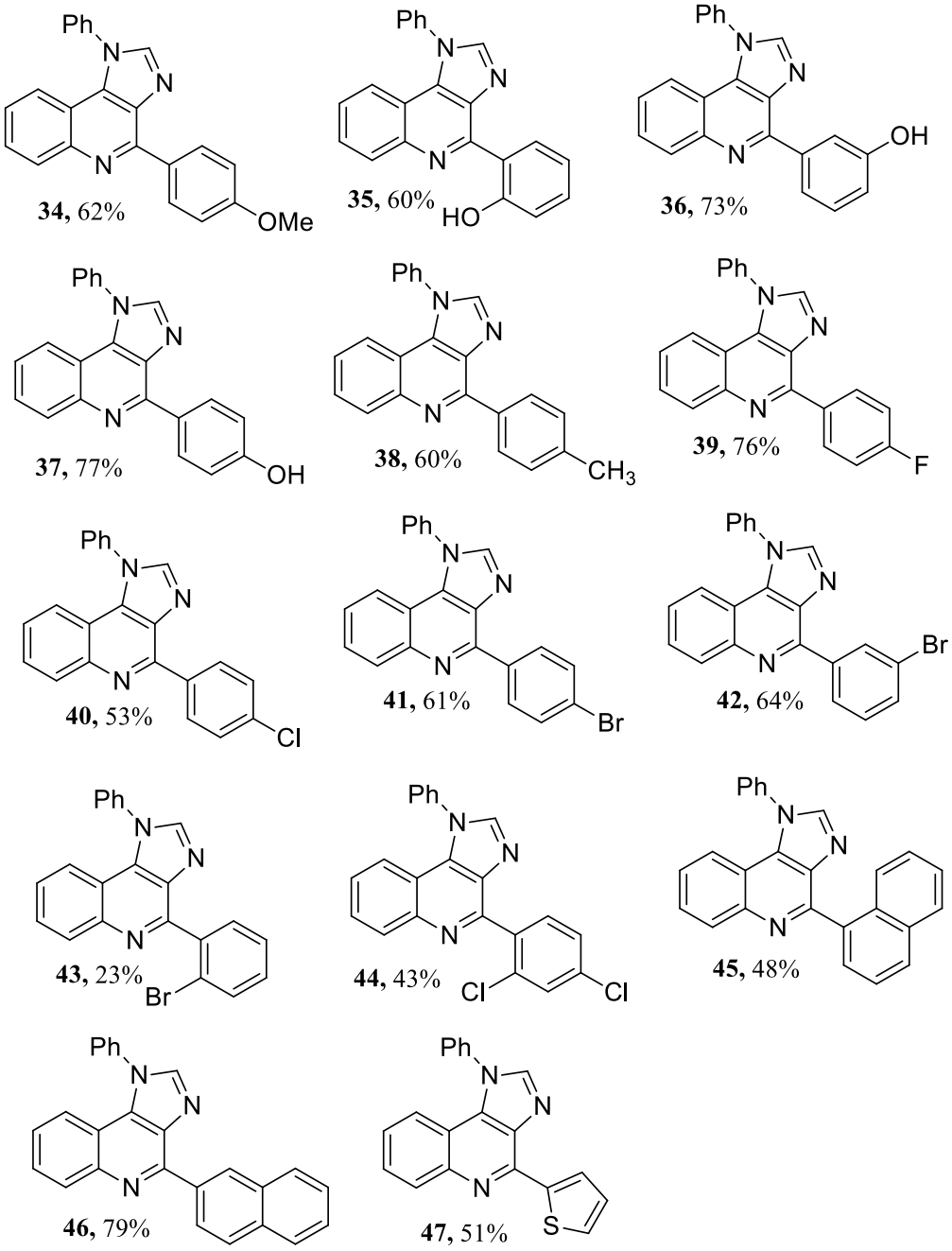
[c] DPE(diphenylether) as solvent; [d] catalyst used 0.1 eqv; [e] catalyst used 0.3 eqv.

The optimized conditions were subsequently used with various aldehydes in order to demonstrate the substrate scope for the quinoline ring formation **(Table 3.2)**. Wide array of aromatic aldehydes were reacted with 2-(1-phenyl-1*H*-imidazol-5-yl)aniline in the presence of $Yb(OTf)_3$ using nitrobenzene as solvent. Use of aromatic aldehydes possessing electron donating groups (*e.g.* OH, OCH_3 and CH_3) resulted in 1,4-disubstituted imidazo[4,5-*c*]quinolines in high yields **(Table 3.2)**. However, the yields were slightly inferior in the case of aldehydes with electron withdrawing groups (*e.g.* CF_3, CN). Interestingly, *p*-fluoro benzaldehyde gave higher yield compared to corresponding chloro or bromo analogues. Aldehydes with bromo substitution (*e.g. o*-, *m*-, *p*-) resulted in 23%, 64% and 61% yields, respectively, which partially reflect relative steric influence of bromine atom. Thiophene carboxaldehyde, 1-naphthaldehyde and 2-naphthaldehyde also produced corresponding final products in moderate to good yields. However, despite numerous attempts, the use of aliphatic aldehydes does not lead to the desired cyclized products, as most of the reactions attempted were terminated at imine stage without proceeding to the final product under these conditions.

Table 3.2: Synthesis of various 1-phenyl-4-substituted imidazo[4,5-*c*]quinolines[a]







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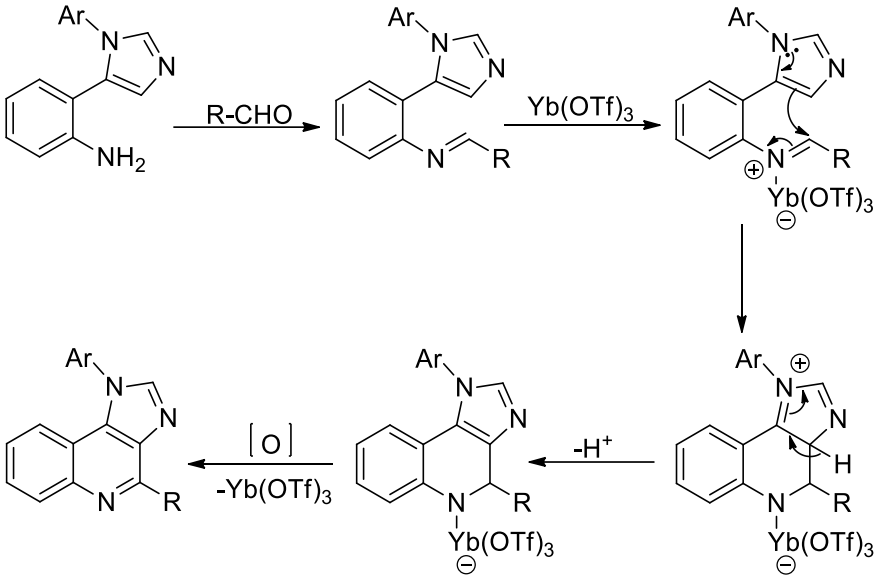
[a]Reaction condition: 0.5 mmol of **28**, 0.6 mmol of aldehyde, $Yb(OTf)_3$ (20 mol%) and nitrobenzene (1.5 mL).

The mechanism of formation of imidazo[4,5-*c*]quinolines appears to proceed through cationic intermediates in accordance with previous report **(Scheme 3.7)**.[7] The electrophilic attack of imidazole ring via position 4 followed by oxidative aromatization are envisioned as key steps in the creation of compound **31**. Given the requirement of high temperature in the

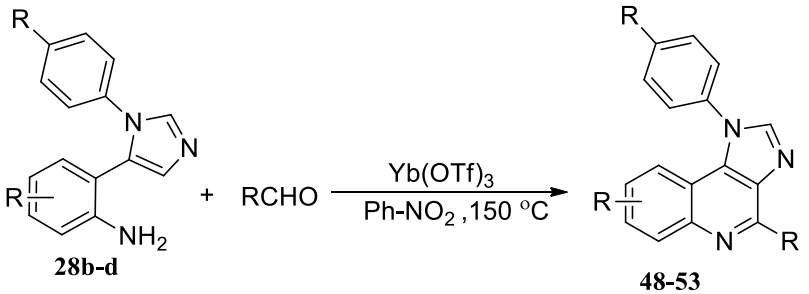
reaction, an alternative C-H activation step could also be envisaged, which requires detailed investigation in the future.

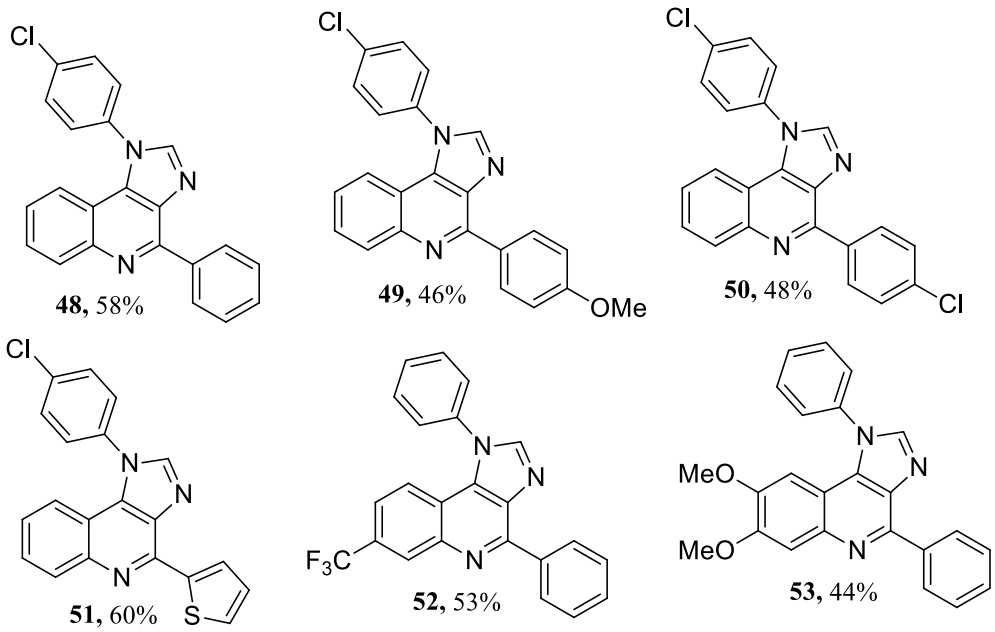


Scheme 3.7: Plausible mechanism for formation of the final compound.

In order to demonstrate the flexibility of the synthetic strategy, reactions were also carried out between 2-(1-(4-chlorophenyl)-1*H*-imidazol-5-yl)aniline **(28b) /** 4,5 or 6-substituted 2-(1-phenyl-1*H*-imidazol-5-yl)anilines **(28c-d)** and various aromatic aldehydes **(Table 3.3)**. All the reactions proceeded successfully, resulting in the synthesis of corresponding imidazo[4,5-*c*]quinoline compounds in moderate yields.

Table 3.3: Synthesis of various 1-(4-chlorophenyl)-4-substituted imidazo[4,5-*c*]quinolines[a]



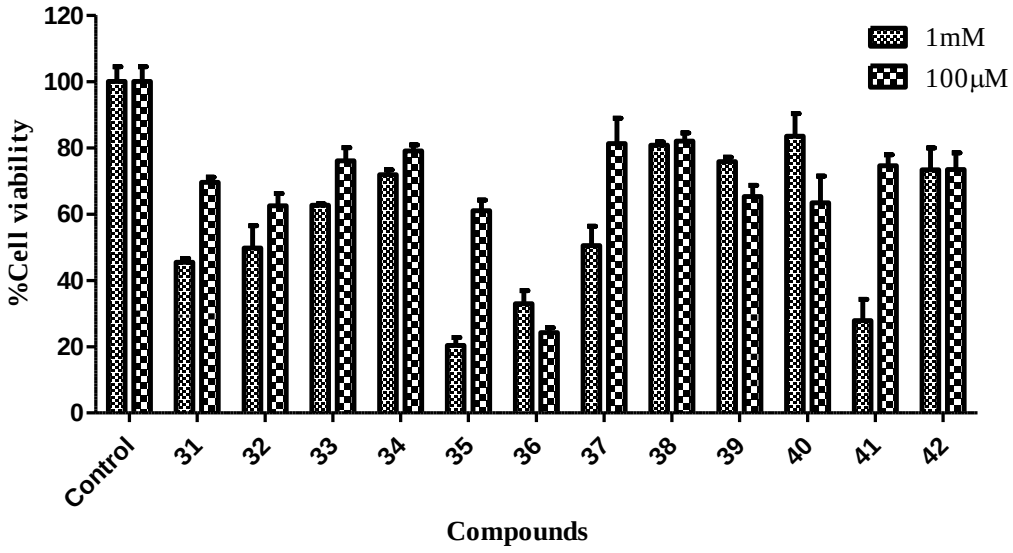


[a] Reaction condition: 0.5 mmol of **28b**, 0.6 mmol of aldehyde, $Yb(OTf)_3$ (20 mol%) and nitrobenzene (1.5 mL).

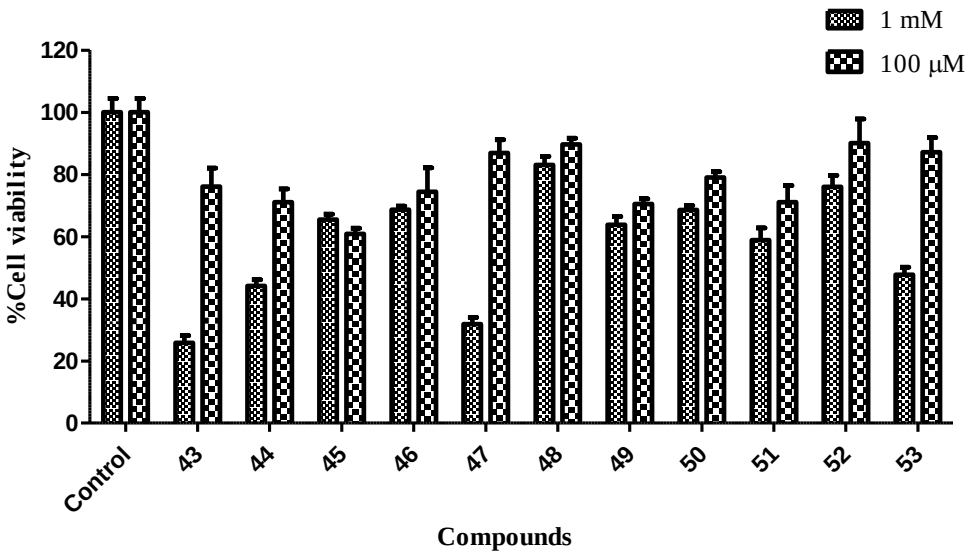
Final compounds after thorough characterization were screened for their anticancer activity using MTT (microculturetetrazolium) assay **(Figure 3.3)**.[31] Standard anticancer drug doxorubicin was used as a positive control to validate the assay. Among twenty three compounds, six most active compounds (**35, 36, 41, 42, 43** and **47**) were explored further with longer range of concentration to find out their IC_{50} value **(Figure 3.4)**. The experimental results, led us to the identification of 1-phenyl-1*H*-imidazo[4,5-*c*]quinoline nucleus as an important structural feature to possess anticancer activity. Compound **35, 36, 41** and **43** shows better activity than **47** suggesting that substituted phenyl rings are more effective than heterocyclic ring system as substituent R on 1-phenyl-1*H*-imidazo[4,5-*c*]quinoline moiety. However, difference in substitution on phenyl ring at position R shows higher impact on biological activity of molecules. Introduction of bromine at ortho position instead of para position, results in a drop of IC_{50} value from 443.3 µM to 103.3 µM, making **43** the most active compound in this series. However, bromine substitution at meta position gave IC_{50} value of

1253 μM. Overall, synthesized molecules while displaying anticancer activity showed weaker potency compared to Doxorubicin.

The compound **43** shows optimum structural requirement for anticancer activity. This compound can be further modified to discover more potent molecules. It can be further explored to check apoptotic activity as well as to find out downstream regulation of various cancer cellular biomarkers.



A



B

Figure 3.3 (A and B): Cytotoxicity of twenty three novel compounds was evaluated in murine melanoma cell line (B16F10). Cell viability was measured by *in vitro* MTT assay. Cells were treated with drug molecules for 24 hours at two concentrations 1 mM and 100 µM (n = 3). Data represent mean values of measurements ± s.d.

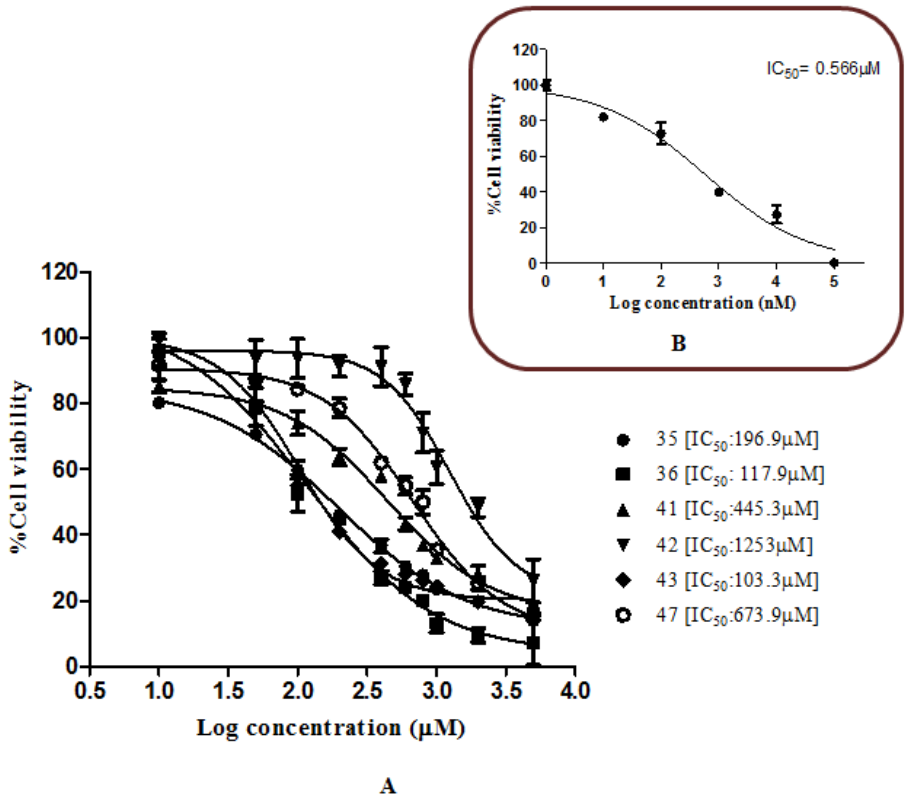


Figure 3.4: (**A**) Promising novel compounds (**35, 36, 41, 42, 43, 47**) were explored in a range of ten different concentrations in B16F10 cells (n = 2). After 24 hours of drug treatment, cells were treated with MTT reagent and cell viability was measured. Data were analyzed and plotted in dose-response format representing mean values of measurements ± s.d. IC_{50} was calculated using nonlinear regression analysis. (**B)** Cytotoxicity of Doxorubicin was measured to validate the assay procedure. B16F10 cells were treated with doxorubicin within 0.001 µM to 100 µM concentration. Cell viability was measured in the same way as for other novel chemical entities. IC_{50} value for Doxorubicin was found to be 0.566 µM which corresponds to reported literature.[32]

3.4. Conclusion

In conclusion we have developed a simple and straight forward method for the preparation of 1,4-disubstituted imidazo[4,5-*c*]quinolines. The method developed is flexible enough to allow substituents at imidazole as well as quinoline rings present in the molecule. The final step utilized can be carried out without introduction of any additive or co-catalyst. Given the diverse application of substituted imidazo[4,5-*c*]quinolines, we feel that the developed method will provide opportunities for easy synthesis of various fused imidazole-quinoline compounds for myriad purposes. Some of the synthesized compounds also display significant anticancer activity. This indicates ample scope for screening hitherto relatively unexplored imidazo[4,5-*c*]quinoline moiety as potential anticancer compounds.

3.5. Experimental

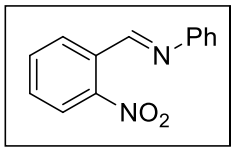
All the compounds and reagents required were purchased from commercial sources and were used without further purification. Solvents were dried and distilled using standard procedures, prior to use. ^{1}H NMR (300 & 400 MHz) and ^{13}C (75 & 101 MHz) spectra were recorded in $CDCl_3$ and DMSO using $(CH_3)_4Si$ as internal standard. IR spectra were recorded as KBr plates on Jasco FT/IR-4200 instrument. Melting points were recorded on a Stuart SMP 30 melting point apparatus and are uncorrected. The mass spectroscopy data were obtained at the Department of Chemistry, Dalhousie University, Canada, by Mr. Xiao Feng.

3.6. Procedures for preparation of compounds 27 and 28

Synthesis of (*E*)-N-(2-Nitrobenzylidene)aniline: To a mixture of 2-nitrobenzaldehyde (2 g, 0.0132 mol) and aniline (1.478 g, 0.0158 mol) in 30 mL of toluene, catalytic amount of acetic acid was added. The solution was stirred at 100 ^{o}C for 8 hours. The reaction mixture was cooled to room temperature on completion and solvent was removed under reduced pressure. The residue obtained, was diluted with water (2 x 5 mL) and extracted with ethyl acetate (2 x 10

mL). The separated organic layer was then washed with brine solution and dried over anhydrous Na_2SO_4. Subsequently, organic layer was concentrated in vacuo and resulting residue purified by column chromatography using 15% ethylacetate/hexane as eluent.

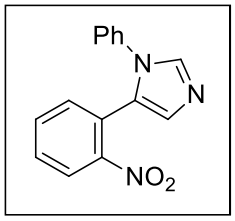
Yield: 83%(2.4 g); yellow solid; **M.p**. 60-62 ^oC; $\mathbf{R_f}$ = 0.7 [hexane / ethyl acetate =8:2];



ν_{max}(KBr)/cm^{-1}: 3056, 2332, 1569, 1522, 1346, 1189, 858, 765, 699; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.26 – 7.35 (m, 3H), 7.39 – 7.49 (m, 2H), 7.62 (t, *J* = 7.7 Hz, 1H), 7.74 (t, *J* = 7.5 Hz, 1H), 8.07 (d, *J* = 8.1 Hz, 1H), 8.32 (d, *J* = 7.8 Hz, 1H), 8.95 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$) *δ* 121.24, 124.57, 126.96, 129.32, 129.76, 131.11, 131.24, 133.64, 149.31, 151.08, 155.88; **LRMS-ESI** (m/z): 227.05 [M + H]$^+$.

Synthesis of 5-(2-Nitrophenyl)-1-phenyl-1*H*-imidazole (27): To a mixture of (*E*)-N-(2-nitrobenzylidene)aniline (2 g, 0.088 mol) and toluenesulfonylmethylisocyanide (2.07 g, 0.0106 mol) in 30 mL of CH_3CN:DMSO (3:1), K_2CO_3 (3.669 g, 0.0265 mol) was added. The solution was then stirred at 80 ^oC for 6 hours. On completion of the reaction as indicated by TLC, the reaction mixture was poured into water (2 x 5 mL) and extracted with ethyl acetate (2 x 10 mL). The separated organic layer was then washed with brine and dried over anhydrous Na_2SO_4 (1 g). Organic layer was subsequently concentrated in vacuo and residue was purified by column chromatography using 40% ethyl acetate/ hexane as eluent.

Yield: 78%(1.84 g); Light brown solid; **M.p**. 107-110 ^oC; $\mathbf{R_f}$ = 0.3 [hexane / ethyl acetate = 5:5]; **ν_{max}(KBr)/cm^{-1}**: 3118, 1520, 1349,1159, 773, 696; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.09

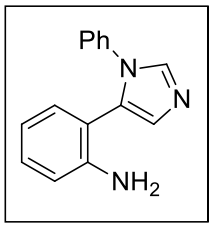


(dd, *J* = 6.3, 2.9 Hz, 2H), 7.25 (s, 1H), 7.32 - 7.34 (m, 3H), 7.48 (dd, *J* = 15.0, 7.6 Hz, 2H), 7.61 (t, *J* = 7.5 Hz, 1H), 7.81 (s, 1H), 7.84 (d, *J* = 8.2 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 124.42, 124.58, 125.04, 128.30,

128.43, 128.46, 129.56, 129.61, 132.99, 133.17, 135.62, 138.73, 148.66; **LRMS-ESI** (m/z): 266 $[M + H]^+$.

Synthesis of 2-(1-Phenyl-1*H*-imidazol-5-yl)aniline (28): To a solution of 5-(2-nitrophenyl)-1-phenyl-1*H*-imidazole (1.8 g, 0.0067 mol) in Acetonitrile/water (1:0.1) at 0 ^oC, $NiCl_2$ $6H_2O$ (0.318 g, 0.00134 mol) and $NaBH_4$ (1.0138 g, 0.027 mol) were added. The reaction mixture was then stirred for 20 minutes, initially at 0 ^oC for 10 minutes and then at room temperature for the remaining duration. On completion of the reaction, resulting mixture was diluted with cold water and passed through celite pad. The solution obtained was then diluted with water and extracted with ethyl acetate. The organic layer was subsequently washed with brine and dried over Na_2SO_4. Dried organic layer was concentrated in vacuo and residue was purified by column chromatography with 50% ethylacetate/ hexane as eluent.

Yield:75% (1.19 g); brown liquid; $\mathbf{R_f}$ = 0.2 [hexane / ethyl acetate = 3:7]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 3475, 3370, 3206, 2359, 1613, 1495, 1460, 1251, 744, 697; **^{1}H NMR** (400 MHz, $CDCl_3$) δ

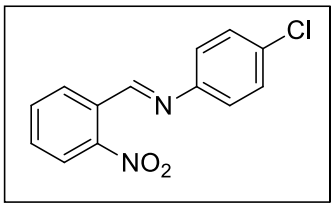


3.81 (s, 2H), 6.62 (td, *J* = 7.5, 1.1 Hz, 1H), 6.69 (dd, *J* = 8.1, 0.9 Hz, 1H), 6.85 (dd, *J* = 7.6, 1.5 Hz, 1H), 7.11 (ddd, *J* = 8.1, 7.4, 1.6 Hz, 1H), 7.15 – 7.19 (m, 2H), 7.26 (s, 1H), 7.30 – 7.37 (m, 3H), 7.82 (s, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 114.39, 115.35, 118.06, 124.61, 127.86, 129.36, 129.40, 129.59, 129.77, 131.78, 136.45, 138.29, 145.46; **LRMS-ESI** (m/z): 236.05 $[M + H]^+$.

(*E*)-4-Chloro-N-(2-nitrobenzylidene)aniline: To a mixture of 2-nitrobenzaldehyde (1.1 g, 0.0085 mol) and *p*-chloro aniline (1.5 g, 0.010 mol) in 15 mL of toluene, catalytic amount of acetic acid was added. The solution was stirred at 100 ^oC for 8 hours. The reaction mixture was cooled to room temperature on completion and solvent was removed under reduced pressure. The residue obtained, was diluted with water (2 x 5mL) and extracted with ethyl acetate (2 x 10 mL). The separated organic layer was then washed with brine solution and dried over

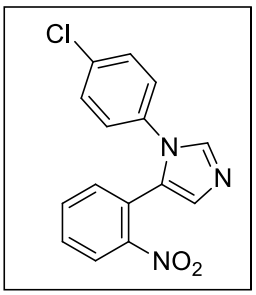
anhydrous Na_2SO_4. Subsequently, organic layer was concentrated in vacuo and resulting residue purified by column chromatography using 15% ethylacetate / hexane as eluent.

Yield:87%; yellow solid; **M.p**. 68-70 ^oC; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1];



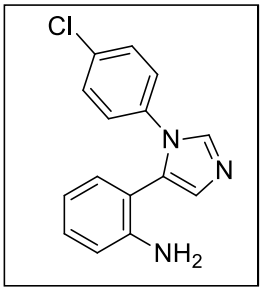
$\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 2998, 2359, 1569, 1523, 1485, 1339, 1186, 1090, 858, 829, 787, 743, 702, 662; **^{1}H NMR** (300 MHz, $CDCl_3$) δ 7.22 (d, *J* = 8.2 Hz, 2H), 7.38 (d, *J* = 8.0 Hz, 2H), 7.63 (t, *J* = 7.3 Hz, 1H), 7.74 (t, *J* = 7.3 Hz, 1H), 8.07 (d, *J* = 7.9 Hz, 1H), 8.28 (d, *J* = 7.4 Hz, 1H), 8.91 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): *δ*=122.57, 124.63, 129.40, 129.72, 130.84, 131.43, 132.54, 133.68, 149.27, 149.47, 156.26; **LRMS-ESI** (m/z): 261.05 $[M + H]^+$.

1-(4-Chlorophenyl)-5-(2-nitrophenyl)-1*H*-imidazole: Yield:79%; block semisolid; $\mathbf{R_f}$ = 0.3 [hexane / ethylacetate = 5:5]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 3123, 3064, 1687, 1523, 1495, 1353, 1091, 1018, 845, 818, 782, 754, 658; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.03 –7.08 (m, 2H), 7.24 (d, *J*



= 1.0 Hz, 1H), 7.29 –7.34 (m, 2H), 7.45 (dd, *J* = 7.6, 1.4 Hz, 1H), 7.51–7.56 (m, 1H), 7.64 (td, *J* = 7.6, 1.3 Hz, 1H), 7.77 (d, *J* = 1.0 Hz, 1H), 7.90 (dd, *J* = 8.1, 1.1 Hz, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): *δ* 124.24, 124.71, 126.31, 128.34, 129.76, 129.83, 130.00, 133.06, 133.21, 134.28, 134.31, 138.71, 148.75; **LRMS-ESI** (m/z): 300.05 $[M + H]^+$.

2-(1-(4-Chlorophenyl)-1*H*-imidazol-5-yl)aniline (28b): Yield: 84%; yellow solid; **M.p**. 112



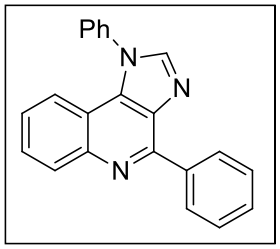
-114 ^oC; $\mathbf{R_f}$ = 0.1 [hexane / ethyl acetate = 7: 3]; $\mathbf{\nu_{max}}$ **(KBr)/cm^{-1}**: 3460, 3373 , 3216, 2360, 1624, 1495, 1311, 1092, 916, 828, 752, 659; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 3.80 (s, 2H), 6.65 (td, *J* = 7.5, 1.1 Hz, 1H), 6.69 (dd, *J* = 8.1, 0.8 Hz, 1H), 6.85 (dd, *J* = 7.6, 1.3 Hz, 1H), 7.09 -7.11 (m, 1H), 7.13 (dt, *J* = 4.6, 1.9 Hz, 2H), 7.25 (s, 1H), 7.28 -7.30 (m, 1H), 7.30 - 7.32 (m, 1H),

7.78 (s, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 114.05, 115.41, 118.19, 125.79, 129.38, 129.56, 129.87, 130.01, 131.75, 133.71, 135.01, 138.13, 145.52; **LRMS-ESI** (m/z): 270.05 $[M + H]^+$.

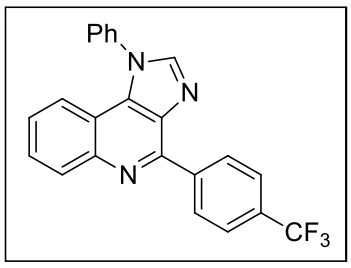
3.7. General method for synthesis of 4-substituted imidazo[4,5-*c*]quinolines

To a mixture of 2-(1-phenyl-1*H*-imidazol-5-yl)aniline (0.5 mmol) and aldehyde (0.6 mmol) in 1.5 mL of nitrobenzene was added $Yb(OTf)_3$ (20 mol%). The solution was stirred at 150 °C for 6 hours under nitrogen atmosphere. On completion of the reaction as indicated by TLC, the reaction mixture was poured into water and extracted with ethyl acetate. The organic layer was then washed with brine (2 mL) and dried over Na_2SO_4 (1 g). Further, dried organic layer was concentrated in vacuo and resulting residue was purified by column chromatography using hexane/ethyl acetate as the eluent to give the corresponding product.

1,4-Diphenyl-1*H*-imidazo[4,5-*c*]quinoline (31). Yield: 74%; light yellow solid; **M.p**. 153-157 °C; **R_f** = 0.9 [hexane / ethyl acetate =9:1]; **ν_{max} (KBr)/cm^{-1}**: 3079, 2360, 1556, 1502, 1450, 1365, 1294, 1204, 1074; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.20 – 7.29 (m, 1H), 7.36 (d, *J* = 8.2 Hz, 1H), 7.49 – 7.63 (m, 9H), 8.05 (s, 1H), 8.32 (d, *J* = 8.4 Hz, 1H), 8.71 (d, *J* = 7.5 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.0, 120.1, 125.7, 127.1, 127.7, 128.5, 129.6, 130.1, 130.1, 130.5, 134.7, 135.7, 136.6, 137.7, 143, 144.5, 151.7; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{16}N_3$ $[M + H]^+$, 322.1344 found 322.1328.

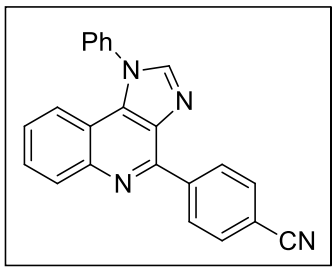


1-Phenyl-4-(4-(trifluoromethyl)phenyl)-1*H*-imidazo[4,5-*c*]quinoline (32). Yield: 52%; yellow solid; **M.p**. 182-185 °C; **R_f** = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3100, 2360, 1511, 1322, 1171, 1127, 1065; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.35 (t, *J* = 7.6 Hz, 1H), 7.44 (d, *J* = 8.3 Hz, 1H), 7.57 – 7.65 (m, 2H), 7.66 - 7.74 (m, 4H), 7.87 (d, *J* = 7.9 Hz, 2H), 8.11 (s, 1H), 8.35 (d, *J* = 8.3 Hz, 1H), 8.86 (d, *J* = 7.9 Hz, 2H);



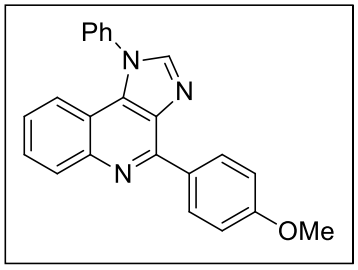
^{13}C NMR (75 MHz, $CDCl_3$): δ 117.3, 120.2, 125.3, 125.3, 125.4, 125.4, 126.3, 127.2, 127.9, 130.2, 130.2, 130.8, 134.9, 135.7, 136.6, 143.1, 144.6, 150.1; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{15}F_3N_3$ $[M + H]^+$, 390.1218 found 390.1230.

4-(1-Phenyl-1*H*-imidazo[4,5-*c*]quinolin-4-yl)benzonitrile (33). Yield: 54%; light yellow solid; **M.p**. 242-245 °C; $\mathbf{R_f}$ = 0.3 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3078, 2360,



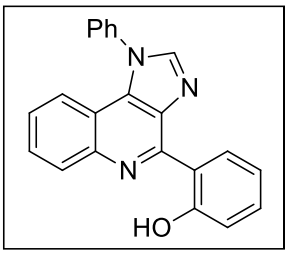
2225, 1565, 1501,1314,1210; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.36 (d, *J* = 8.1 Hz, 1H), 7.42 (t, *J* = 8.1 Hz, 1H), 7.60 - 7.72 (m, 6H), 7.90 (d, *J* = 8.1 Hz, 2H), 8.13 (s, 1H), 8.39 (d, *J* = 8.1 Hz, 1H), 8.93 (d, *J* = 8.1 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 112.8, 117.3, 119.1, 120.2, 126.6, 127.1, 128.2, 130.3, 130.4, 130.5, 132.2, 135.2, 135.6, 136.5, 141.6, 143.3, 144.3, 149.1; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{15}N_4$ 347.1297 $[M + H]^+$, found 347.1303.

4-(4-Methoxyphenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (34). Yield: 62%; white solid; **M.p**. 175-178 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3107, 3046, 2837,



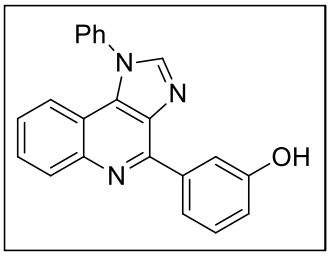
2360, 1571, 1370, 1308, 1173, 1030; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 3.93 (s, 3H), 7.15 (d, *J* = 8.3 Hz, 2H), 7.27 (t, *J* = 7.5 Hz, 1H), 7.38 (d, *J* = 8.2 Hz, 1H), 7.54 – 7.62 (m, 3H), 7.67 (d, *J* = .5 Hz, 3H), 8.08 (s, 1H), 8.34 (d, *J* = 8.2 Hz, 1H), 8.76 (d, *J* = 8.3 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 55.4, 113.9, 116.9, 120.1, 125.4, 127.2, 127.7, 130.1, 130.2, 130.3, 131.5, 134.7, 135.6, 136.8, 140.1, 142.8, 144.6, 151.3, 160.9; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{18}N_3O$ 352.1450 $[M + H]^+$, found 352.1445.

2-(1-Phenyl-1*H*-imidazo[4,5-*c*]quinolin-4-yl)phenol (35). Yield: 60%; light yellow solid;



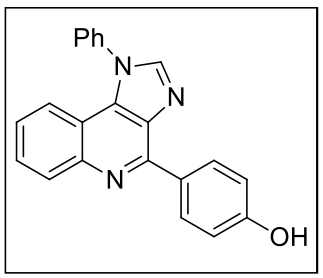
M.p. 206-210 °C; $\mathbf{R_f}$ = 0.8 [hexane / ethyl acetate = 9:1]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3390, 3100, 3060, 2360, 1591, 1505, 1300, 1245, 1157; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 6.97 -7.04 (m, 2H), 7.32 (s, 2H),7.49 (s, 3H), 7.59 (s, 4H), 8.01 (s, 2H), 9.62 (d, *J* = 6.7 Hz, 1H), 15.79 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 116.5, 118.0, 118.6, 118.9, 120.2, 126.1, 127.2, 128.1, 128.3, 130.3, 130.4, 132.1, 132.1, 134.7, 135.1, 136.4, 141.0, 142.9, 152.1, 161.5; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{16}N_3O$ 338.1293 $[M + H]^+$, found 338.1293.

3-(1-Phenyl-1*H*-imidazo[4,5-*c*]quinolin-4-yl)phenol (36). Yield: 73%; white solid; **M.p**.



195-198 °C; $\mathbf{R_f}$ = 0.5 [hexane / ethyl acetate = 6:4**]; $\boldsymbol{\nu_{max}}$ (KBr)/cm^{-1}**: 3481, 3050, 1577, 1557, 1518, 1502, 1448, 1373,1304, 1261, 1219, 1183, 1086; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 6.97 - 7.02 (m, 1H), 7.27 – 7.32 (m, 1H), 7.40 (d, *J* = 8.3 Hz, 1H), 7.47 (t, *J* = 7.9 Hz, 1H), 7.54 – 7.60 (m, 2H), 7.60 – 7.63 (m, 1H), 7.67 (dd, *J* = 8.0, 2.5 Hz, 3H), 8.01 (dd, *J* = 7.6, 0.7 Hz, 1H), 8.13 (d, *J* = 0.8 Hz, 1H), 8.33 (d, *J* = 9.3 Hz, 2H), 9.14 (s, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 116.7, 117.1, 117.2, 120.2, 121.7, 125.9, 127.1, 128.0, 130.0, 130.2, 130.3, 130.4, 134.7, 135.4, 136.6, 138.7, 143.2, 144.7, 152.5, 156.8; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{16}N_3O$ 338.1293 $[M + H]^+$, found 338.1311.

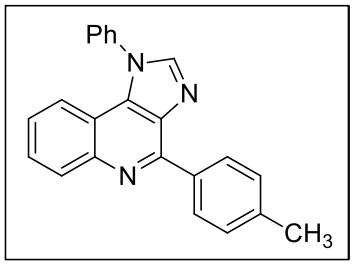
4-(1-Phenyl-1*H*-imidazo[4,5-*c*]quinolin-4-yl)phenol (37). Yield: 77%; Light yellow solid,



M.p. 190-193 °C; $\mathbf{R_f}$ = 0.5 [hexane / ethyl acetate = 6:4]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3449, 3108, 1607, 1573, 1509, 1440, 1371, 1323, 1300, 1283, 1241, 1170; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.04 (d, *J* = 8.7 Hz, 2H), 7.31 (d, *J* = 8.1 Hz, 1H), 7.44 (dd, *J* = 22.4, 7.6 Hz, 2H), 7.60 – 7.67 (m, 3H), 7.69 – 7.73 (m, 3H), 8.12 (s, 1H), 8.34 (d, *J* = 8.1 Hz, 1H), 8.59 (d, *J* = 8.7 Hz, 2H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 115.5, 117.0, 120.2, 125.7, 127.9, 128, 129,

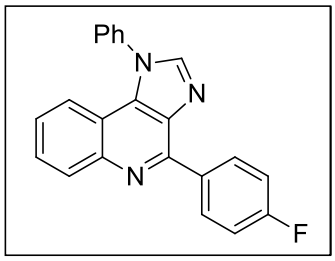
130.4, 130.6, 131.7, 134.6, 135.3, 137, 144.4, 144.5, 150.2, 159.6; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{16}N_3O$ 338.1293 $[M + H]^+$, found 338.1310.

1-Phenyl-4-p-tolyl-1*H*-imidazo[4,5-*c*]quinoline (38). Yield: 60%; light yellow solid; **M.p**.



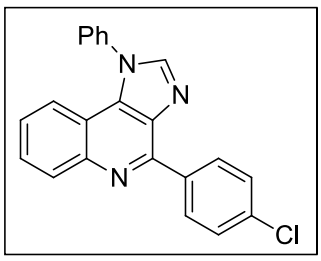
210-212 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3099, 2360, 1567, 1505, 1366, 1294, 1205; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 2.41 (s, 3H), 7.20 (s, 1H), 7.25 (d, *J*= 7.2 Hz, 1H), 7.34 (t, *J* = 8.2 Hz, 3H), 7.50 - 7.64 (m, 6H), 8.03 (s, 1H), 8.54 (d, *J* = 8.0 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 21.6, 116.9, 120.1, 122.5, 123.7, 125.8, 127.2, 128.0, 129.3, 130.0, 130.2, 135.0, 135.1, 135.6, 136.7, 140.1, 143.2, 151.8; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{18}N_3$ 336.1501 $[M + H]^+$, found 336.1501.

4-(4-Fluorophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (39). Yield: 76%; white solid;



M.p. 182-186 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3060, 2360, 1595, 1502, 1292, 1209, 1154; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.30 (d, *J* = 8.2 Hz, 3H), 7.39 (d, *J* = 8.3 Hz, 1H), 7.55 - 7.60 (m, 2H), 7.63 (d, *J* = 7.7 Hz, 1H), 7.67 (d, *J* = 4.6 Hz, 3H), 8.07 (s, 1H), 8.31 (d, *J* = 8.4 Hz, 1H), 8.78 (dd, *J* = 8.2, 5.8 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 115.2, 115.5, 117.0, 120.1, 125.8, 127.1, 127.7, 130.2, 130.5, 131.9, 132.0, 133.9, 133.9, 134.8, 135.5, 136.7, 142.9, 144.6, 150.4.165.4, 165.5; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}FN_3$ 340.1250 $[M + H]^+$, found 340.1252.

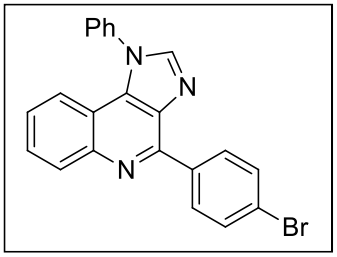
4-(4-Chlorophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (40). Yield: 53%; light yellow



solid; **M.p**. 187-192 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3043, 2360, 1566, 1489, 1317, 1209, 1083; **^{1}H NMR**(300 MHz, CDCl3): δ 7.16 – 7.21 (m, 1H), 7.29 (s, 1H), 7.45 – 7.57 (m, 8H), 7.97 (s, 1H), 8.21 (d, *J* = 8.2 Hz, 1H), 8.62 (dd, *J*

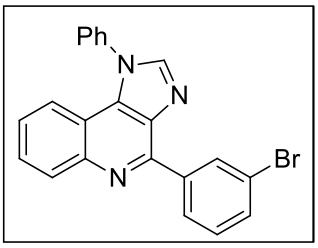
= 8.6, 2.4 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.1, 120.2, 126.0, 127.1, 127.2, 127.8, 128.6, 130.2, 130.5, 131.3, 134.7, 135.5, 135.7, 136.1, 136.6, 143.0, 144.4, 150.2; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}ClN_3$ 356.0955 $[M + H]^+$, found 356.0953.

4-(4-Bromophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (41). Yield: 61%; yellow solid;



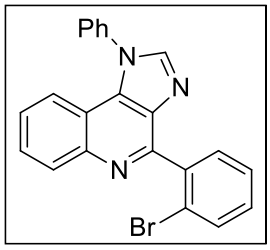
M.p. 94-96 ˚C; **R_f** = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3125, 2872, 2360, 1623, 1558, 1483, 1283, 1112; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.32 (t, *J* = 7.5 Hz, 1H), 7.40 (d, *J* = 8.3 Hz, 1H), 7.62 (dd, *J* = 13.3, 3.0 Hz, 3H), 7.65 – 7.79 (m, 5H), 8.09 (s, 1H), 8.35 (d, *J* = 8.3 Hz, 1H), 8.66 (d, *J* = 8.3 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.1, 120.2, 124.4, 126.0, 126.2, 127.1, 127.2, 127.9, 130.2, 130.4, 131.5, 131.6, 134.9, 135.5, 136.6, 143.0, 144.3, 150.2; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}BrN_3$ 400.0449 $[M + H]^+$, found 400.0427.

4-(3-Bromophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (42). Yield: 64%; white solid;



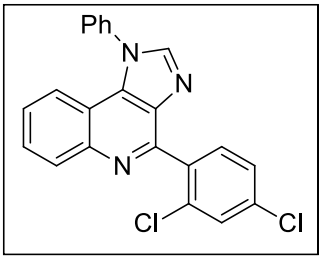
M.p. 195-200 ˚C; **R_f** = 0.9 [hexane / ethyl acetate = 9:1**]; ν_{max} (KBr)/cm^{-1}**: 3050, 2360, 1697, 1554, 1510, 1367, 1311, 1247, 1217; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.28 - 7.34 (m, 1H), 7.40 (d, *J* = 8.2 Hz, 1H), 7.48 (t, *J* = 7.9 Hz, 1H), 7.56 – 7.61 (m, 2H), 7.62 – 7.71 (m, 5H), 8.09 (s, 1H), 8.34 (d, *J* = 8.4 Hz, 1H), 8.75 (d, *J* = 7.8 Hz, 1H), 8.90 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.2, 120.2, 122.2, 126.1, 127.1, 127.9, 128.7, 129.9, 130.2, 130.6, 132.5, 132.6, 134.9, 135.6, 136.6, 139.6, 143.1, 144.4, 149.8; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}BrN_3$ 400.0449 $[M + H]^+$, found 400.0446.

4-(2-Bromophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (43). Yield: 23%; light black solid; **M.p**. 146-150 ˚C; **R_f** = 0.9 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3078, 2922, 2360, 1593, 1501, 1441, 1294, 1203, 1022; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.21 – 7.26 (m,



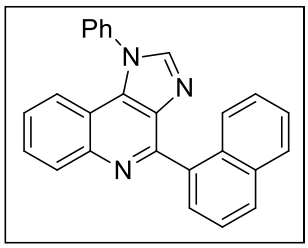
2H), 7.36 – 7.39 (m, 2H), 7.47 – 7.58 (m, 7H), 7.65 (d, *J* = 8.0 Hz, 1H), 7.93 (s, 1H), 8.23 (d, *J* = 8.4 Hz, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.5, 120.3, 122.7, 126.3, 127.1, 127.4, 127.8, 130.2, 130.2, 130.3, 130.8, 131.5, 133.2, 134.0, 136.2, 136.6, 139.0, 143.4, 144.5, 153.8; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}BrN_3$ 400.0449 $[M + H]^+$, found 400.0436.

4-(2,4-Dichlorophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (44). Yield: 43%; light yellow solid; **M.p**. 175-179˚C; **R_f** = 0.8 [hexane / ethyl acetate = 8.5:1.5]; **ν_{max} (KBr)/cm^{-1}**:



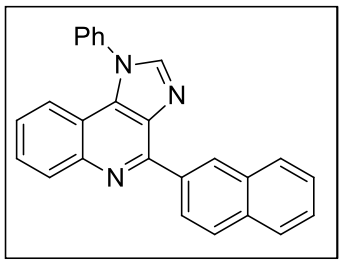
3091, 2923, 2389, 1588, 1564, 1498, 1450, 1371, 1323, 1296, 1257, 1209, 1140, 1102, 1066, 1035; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.38 (t, *J* = 7.6 Hz, 1H), 7.46 (t, *J* = 8.4 Hz, 2H), 7.60 – 7.69 (m, 8H), 8.05 (s, 1H), 8.31 (d, *J* = 8.4 Hz, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.6, 120.4, 120.6, 127.2, 127.3, 128.0, 130.1, 130.3, 130.5, 131.0, 132.6, 134.2, 134.4, 135.6, 135.7, 136.4, 136.7, 143.5, 144.7, 151.6; **LRMS-ESI** (m/z): 390 $[M + H]^+$ [all attempts to get the HRMS of this compound resulted in removal of a chlorine atom and the mass shown corresponds to monochlorophenyl substituted compound 356.0994 ($M + H^+$)].

4-(Naphthalen-1-yl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (45). Yield: 47%; light yellow solid; **M.p**. 176-180 ˚C; **R_f** = 0.8 [hexane / ethyl acetate = 9:1]; **ν_{max} (KBr)/cm^{-1}**: 3053, 2360,



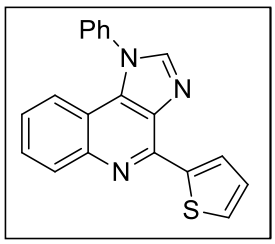
1559, 1507, 1369, 1307, 1247, 1212, 1069; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.37 – 7.46 (m, 2H), 7.49 (d, *J* = 7.6 Hz, 1H), 7.54 (d, *J* = 7.8 Hz, 2H), 7.64 - 7.66 (m, 2H), 7.71 (t, *J* = 6.5 Hz, 5H), 7.95 – 8.00 (m, 2H), 8.05 (d, *J* = 10.8 Hz, 3H), 8.46 (s, 1H); **^{13}C NMR** (300 MHz, $CDCl_3$): δ 117.2, 120.3, 125.3, 125.9, 126.1, 126.2, 126.3, 127.1, 128.0, 128.4, 128.8, 128.9, 129.6, 129.7, 129.8, 130.2, 131.8, 133.8, 134.2, 136.4, 136.6, 137.0, 137.2, 143.5; **HRMS-ESI** (m/z): Calcd for $C_{26}H_{18}N_3$ 372.1501 $[M + H]^+$, found 372.1503.

4-(Naphthalen-2-yl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (46). Yield: 79%; yellow solid;



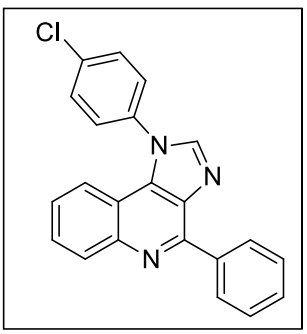
M.p. 178-181 °C; $\mathbf{R_f}$ = 0.8 [hexane / ethyl acetate = 9:1]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3042, 2360, 1558, 1508, 1308, 1205; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.19 (dd, *J* = 13.7, 5.8 Hz, 1H), 7.30 (d, *J* = 8.1 Hz, 1H), 7.37 – 7.62 (m, 8H), 7.76 – 7.88 (m, 1H), 7.98 (dd, *J* = 12.5, 8.2 Hz, 3H), 8.29 (d, *J* = 8.3 Hz, 1H), 8.75 (d, *J* = 8.5 Hz, 1H), 9.26 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 117.1, 120.2, 125.8, 126.0, 126.9, 127.2, 127.6, 127.7, 128.0, 129.4, 130.1, 130.2, 130.4, 130.6, 133.4, 134.1, 134.9, 135.1, 136.0, 136.8, 142.8-, 143.0, 144.7, 151.5; **HRMS-ESI** (m/z): Calcd for $C_{26}H_{18}N_3$ 372.1501 $[M + H]^+$, found 372.1508.

1-Phenyl-4-(thiophen-2-yl)-1*H*-imidazo[4,5-*c*]quinoline (47). Yield: 51%; light yellow solid; **M.p**. 138-140 °C; $\mathbf{R_f}$ = 0.8 [hexane / ethyl acetate = 9:1]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3095, 2350,



1563, 1494, 1450, 1367, 1315, 1209, 1088, 1046; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.22 (t, *J* = 7.2 Hz, 1H), 7.30 (dd, *J* = 10.4, 6.2 Hz, 2H), 7.56 - 7.68 (m, 7H), 8.06 (s, 1H), 8.23 (d, *J* = 6.0 Hz, 1H), 8.86 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ116.9, 120.3, 125.4, 127.1, 127.7, 128.3, 129.0, 130.1, 130.9, 134.3, 134.4, 136.6, 142.5, 142.9, 144.5, 144.5, 146.3; **HRMS-ESI** (m/z): Calcd for $C_{20}H_{14}N_3S$ 328.0908 [M + H]+, found 328.0914.

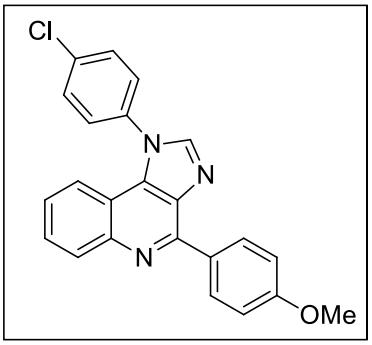
1-(4-chlorophenyl)-4-phenyl-1*H*-imidazo[4,5-*c*]quinoline (48). Yield: 58%; yellow solid;



M.p. 168-171°C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; $\boldsymbol{\nu_{max}}$ **(KBr)/cm^{-1}**: 3060, 1562, 1498, 1367, 1297, 1208, 1091; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.29 - 7.35 (m, 1H), 7.39 (d, *J* = 8.2 Hz, 1H), 7.52 (d, *J* = 7.0 Hz, 3H), 7.61 (t, *J* = 7.5 Hz, 5H), 8.02 (s, 1H), 8.33 (d, *J* = 8.2 Hz, 1H), 8.66 (d, *J* = 8.1 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 116.8, 119.9, 125.9, 127.8, 128.4, 128.5, 129.7, 129.9, 130.4, 130.6, 134.6, 135.2,

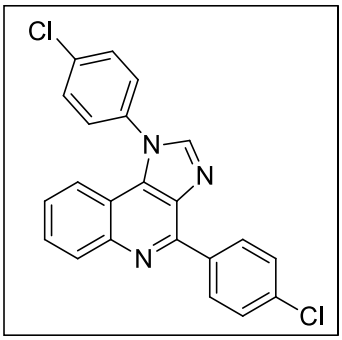
135.7, 136.2, 137.5, 142.7, 144.6, 151.8; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{15}ClN_3$ 356.0955 $[M + H]^+$, found 356.0966.

1-(4-Chlorophenyl)-4-(4-methoxyphenyl)-1*H*-imidazo[4,5-*c*]quinoline (49). Yield: 46%; yellow solid; **M.p**. 165-168 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9: 1]; $\mathbf{\nu_{max}}$ **(KBr)/cm^{-1}**:



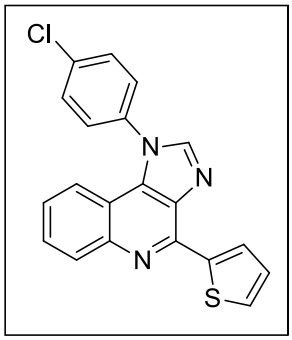
3010, 2360, 1685, 1602, 1513, 1426, 1299, 1260, 1167, 1024; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 3.90 (s, 3H), 6.63 (d, *J* = 8.8 Hz, 1H), 6.73 (s, 1H), 7.33 (d, *J* = 3.7 Hz, 2H), 7.56 (d, *J* = 8.5 Hz, 2H), 7.59 - 7.66 (m, 2H), 7.69 (d, *J* = 8.4 Hz, 2H), 8.09 (d, *J* = 9.5 Hz, 2H), 9.65 (d, *J* = 9.1 Hz, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 55.4, 101.7, 102.0, 107.0, 114.6, 114.6, 115.8, 120.1, 126.0, 128.5, 129.0, 130.6, 133.6, 134.2, 134.7, 134.8, 136.7, 141.1, 143.1; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{17}ClN_3O$386.1060 $[M + H]^+$, found 386.1067.

1,4-Bis(4-chlorophenyl)-1*H*-imidazo[4,5-*c*]quinoline (50). Yield: 48%; yellow solid; **M.p**. 224-226 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; $\mathbf{\nu_{max}}$ **(KBr)/cm^{-1}**: 3010, 2960, 2360, 1495,



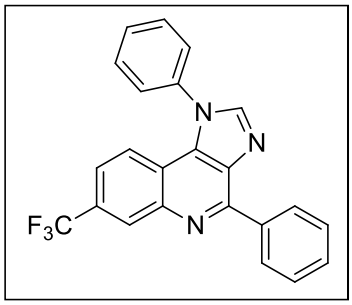
1315, 1208, 1092, 1014; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 7.18 (s, 1H), 7.32 (d, *J* = 7.9 Hz, 1H), 7.47 (t, *J* = 8.4 Hz, 4H), 7.57 (s, 2H), 7.59 (s, 1H), 7.97 (s, 1H), 8.30 (d, *J* = 7.7 Hz, 1H), 8.63 (d, *J* = 7.9 Hz, 2H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 116.9, 120.0, 126.2, 128.4, 128.7, 130.5, 131.3, 135.1, 135.6, 136.4, 142.8, 147.9, 148.7, 148.7, 150.2; **HRMS-ESI** (m/z): Calcd for $C_{22}H_{14}Cl_2N_3$ 390.0565 $[M + H]^+$, found 390.0559.

1-(4-Chlorophenyl)-4-(thiophen-2-yl)-1*H*-imidazo[4,5-*c*]quinoline (51). Yield: 60%; yellow solid; **M.p**. 218-220 °C; $\mathbf{R_f}$ = 0.9 [hexane / ethyl acetate = 9:1]; $\mathbf{\nu_{max}}$ **(KBr)/cm^{-1}**: 3095, 2360, 1563, 1494, 1450, 1367, 1315, 1209, 1088, 1046; **^{1}H NMR** (300 MHz, $CDCl_3$): δ 6.47



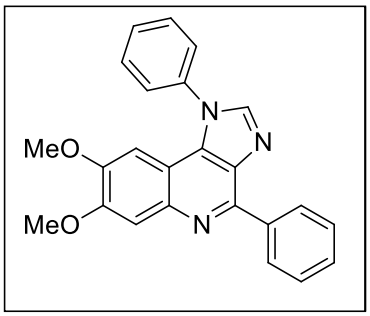
(s, 1H), 7.13 (s, 1H), 7.23 (d, *J* = 7.5 Hz, 1H), 7.32 (d, *J* = 8.2 Hz, 1H), 7.55 (d, *J* = 7.6 Hz, 2H), 7.60 (d, *J* = 9.2 Hz, 1H), 7.66 (d, *J* = 7.7 Hz, 2H), 8.01 (s, 1H), 8.15 (s, 1H), 11.10 (s, 1H); **^{13}C NMR** (75 MHz, $CDCl_3$): δ 110.4, 112.0, 116.3, 120.0, 121.2, 124.7, 127.8, 128.4, 129.0, 129.9, 130.4, 130.9, 132.8, 133.9, 135.1, 136.2, 142.2, 145.2; **HRMS-ESI** (m/z): Calcd for $C_{20}H_{13}ClN_3S$ 362.0519 [M + H]+, found 362.0527.

1,4-Diphenyl-7-(trifluoromethyl)-1*H*-imidazo[4,5-*c*]quinoline (52). Yield: 53%; White solid; **M.p**. 198-201 °C; **R_f** = 0.4 [hexane / ethyl acetate = 8:2]; **ν_{max} (KBr)/cm^{-1}**: 2931, 1596, 1513, 1370, 1316, 1279, 1132; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.50 (s, 2H), 7.58-7.63 (m, 5H),



7.69 – 7.75 (m, 3H), 8.18 (s, 1H), 8.64 (s, 1H), 8.73 (d, *J* = 7.3 Hz, 2H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 119.0, 121.1, 121.4, 121.4, 122.7, 125.5, 127.1, 128.3, 128.4, 128.6, 129.2, 129.6, 129.9, 130.0, 130.4, 130.5, 134.3, 136.4, 136.9, 137.4, 143.6, 143.9, 153.2; **HRMS-ESI** (m/z): Calcd for $C_{23}H_{15}F_3N_3$ 390.1218 $[M + H]^+$, found 390.1213.

7, 8-dimethoxy-1,4-diphenyl-1*H*-imidazo[4,5-*c*]quinoline (53). Yield: 44%. white solid; **M.p**. 191-193°C; **R_f** = 0.4 [hexane / ethyl acetate = 8:2]; **ν_{max} (KBr)/cm^{-1}**: 3022, 2929, 1578,



1565, 1487, 1455, 1442, 1326, 1280, 1245, 1144, 1107, 1044, 1012; **^{1}H NMR** (400 MHz, $CDCl_3$) δ 3.55 (s, 3H), 4.06 (s, 3H), 6.66 (s, 1H), 7.51 (t, *J* = 7.4 Hz, 1H), 7.64 (ddd, *J* = 15.5, 10.2, 5.9 Hz, 4H), 7.67 – 7.74 (m, 4H), 8.09 (s, 1H), 8.63 (d, *J* = 7.1 Hz, 2H); **^{13}C NMR** (101 MHz, $CDCl_3$) δ 55.4, 56.1, 99.3, 110.1, 111.3, 127.7, 128.5, 129.1, 129.5, 129.9, 130.0, 134.5, 135.0, 136.8, 138.1, 141.3, 142.3, 148.7, 149.7, 150.3; **HRMS-ESI** (m/z): Calcd for $C_{24}H_{20}N_3O_2$ 382.1556 $[M + H]^+$, found 382.1549.

3.8. Cell line culture and drug treatment

B16F10 (murine melanoma) cells were cultured in Dulbecco's Modified Eagle Medium (DMEM) (Himedia Laboratories Pvt. Ltd., Mumbai, India). Culture medium was supplemented with 10% heat inactivated fetal bovine serum (Himedia Laboratories Pvt. Ltd., Mumbai, India) and 1% of antibiotic solution (10 U Penicillin and 10 mg Streptomycin per ml, Himedia Laboratories Pvt. Ltd., Mumbai, India). Cells were cultured at 37°C in humidified atmosphere with 5% CO_2. Stock solutions of all 23 synthesized compounds were prepared in DMSO at a concentration of 10 mM and stored. Stock solution of doxorubicin (ZhejingHisun Pharmaceutical Co. Ltd.) was prepared at a concentration of 100 mM using DMSO and stored.

3.9. MTT Assay

Anticancer activity was determined using MTT assay.[31,33] Briefly, cells were seeded at 10,000 cells/well in 100 µl of medium in 96-well plates and incubated for 24 hours. Cells were treated with drug compounds at two concentrations (1 mM and 100 µM) in triplicates and incubated for 24 hours. Stock solutions were diluted in such a way that final DMSO concentration was 1%. 50 µl of 5 mg/mL 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT; Himedia Laboratories Pvt. Ltd., Mumbai, India) was added and incubated for 4 hours. Formazan crystals were dissolved in 150 µl of DMSO and evaluated spectrophotometrically at 570 nm and 650 nm using Spectramax M4 (Molecular Devices, USA).

For determination of IC_{50} values of some selected compounds same procedure was followed as described above where five drug compounds were tested at 10 concentrations as 5 mM, 2 mM, 1 mM, 800 µM, 600 µM, 400 µM, 200 µM, 100 µM, 50 µM and 10 µM. The experiment was repeated same as previous with another batch of cells.

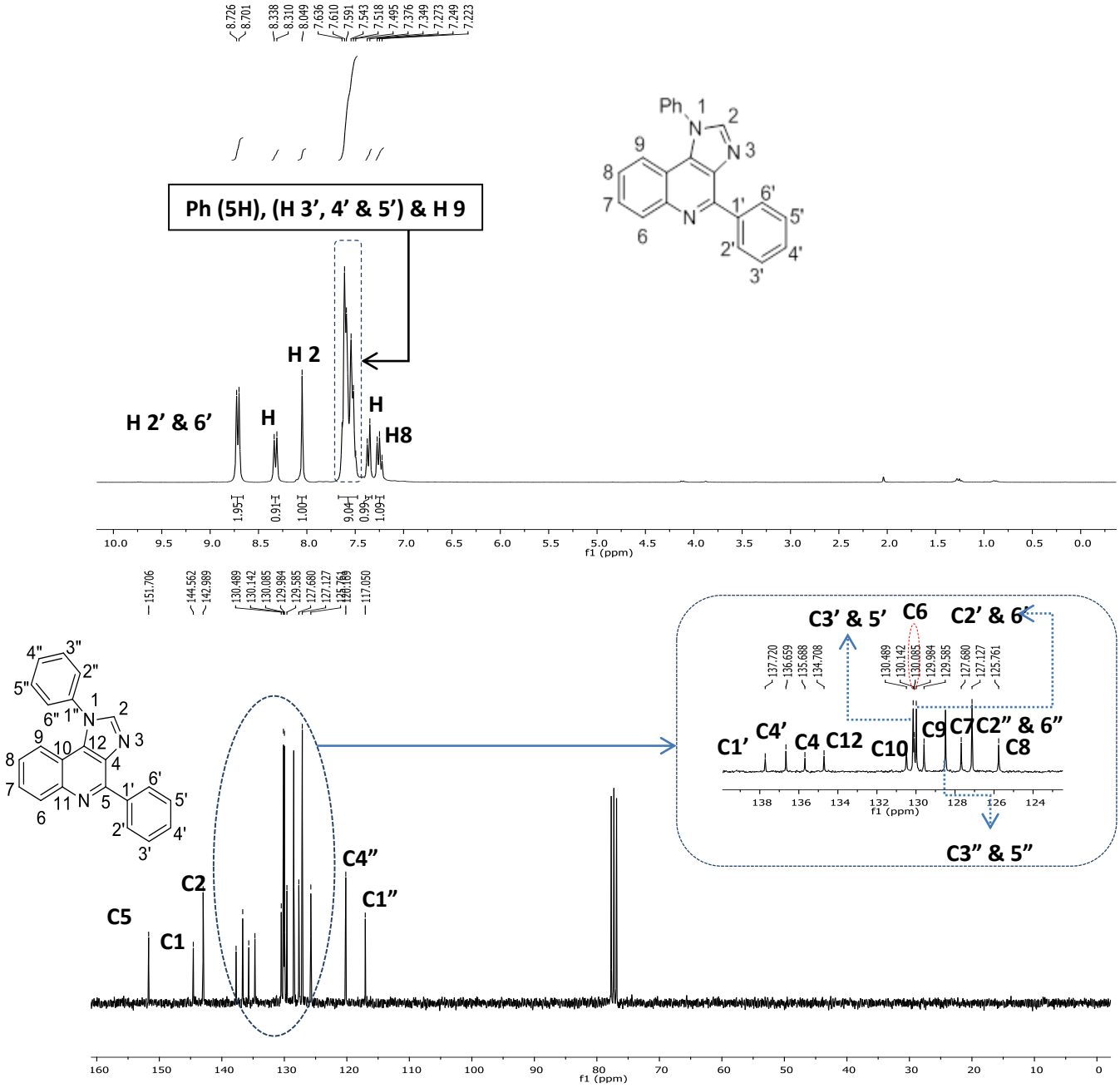
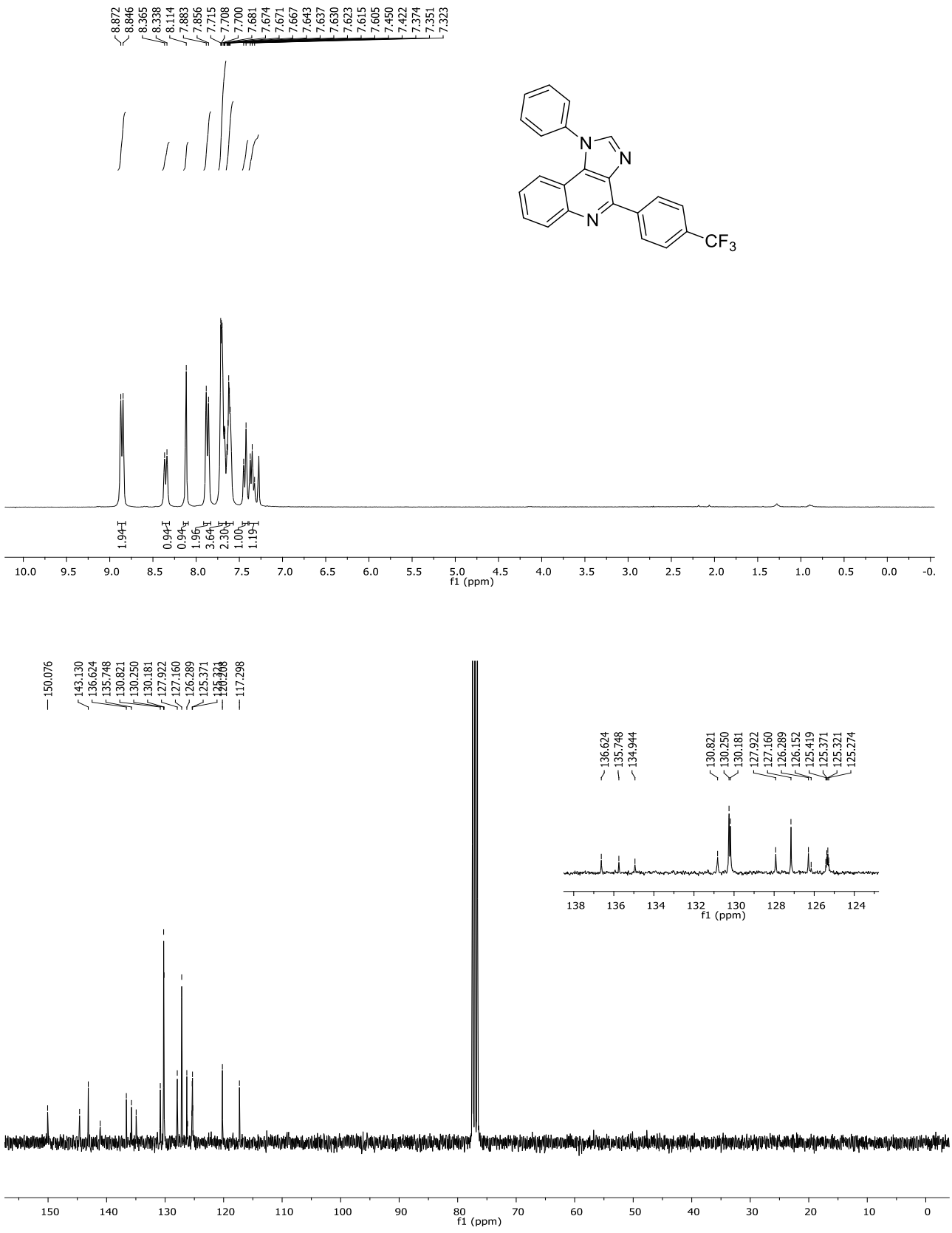
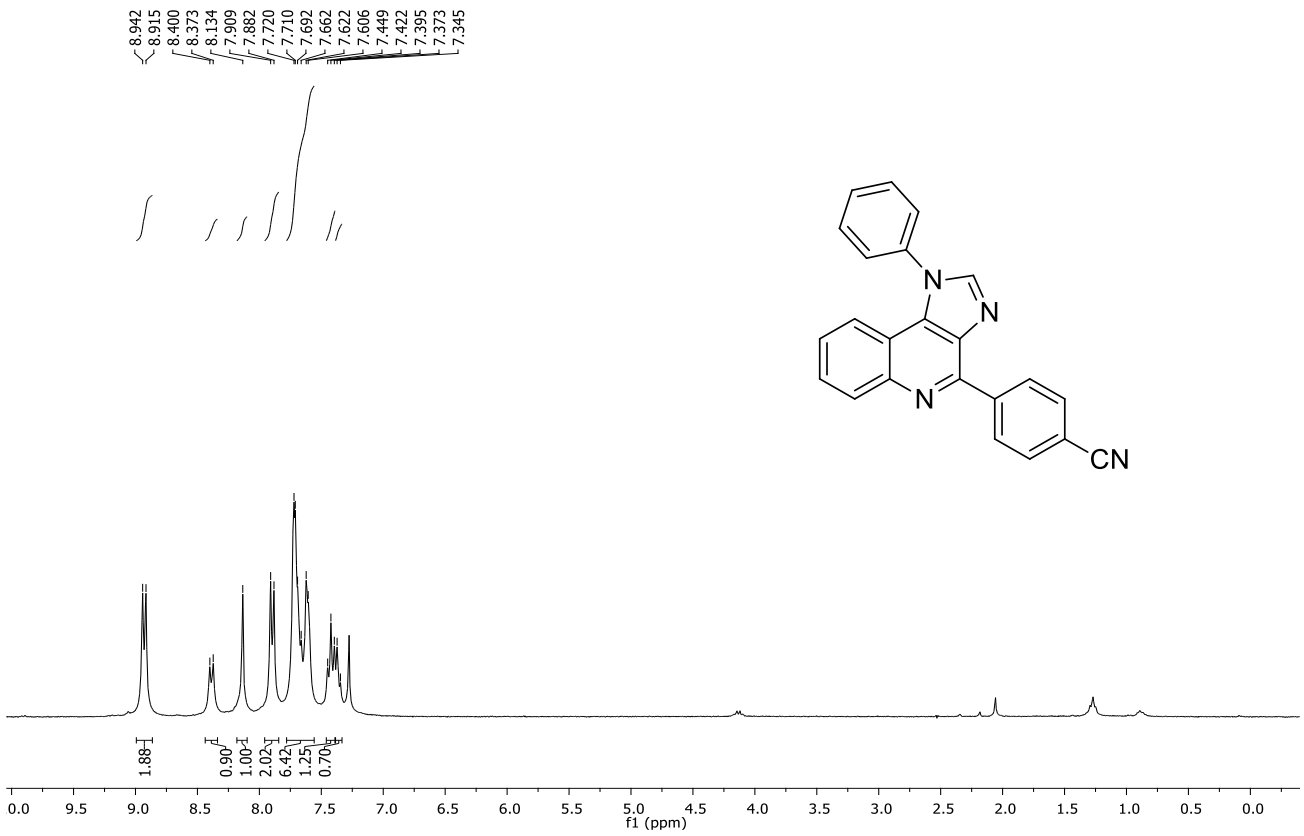
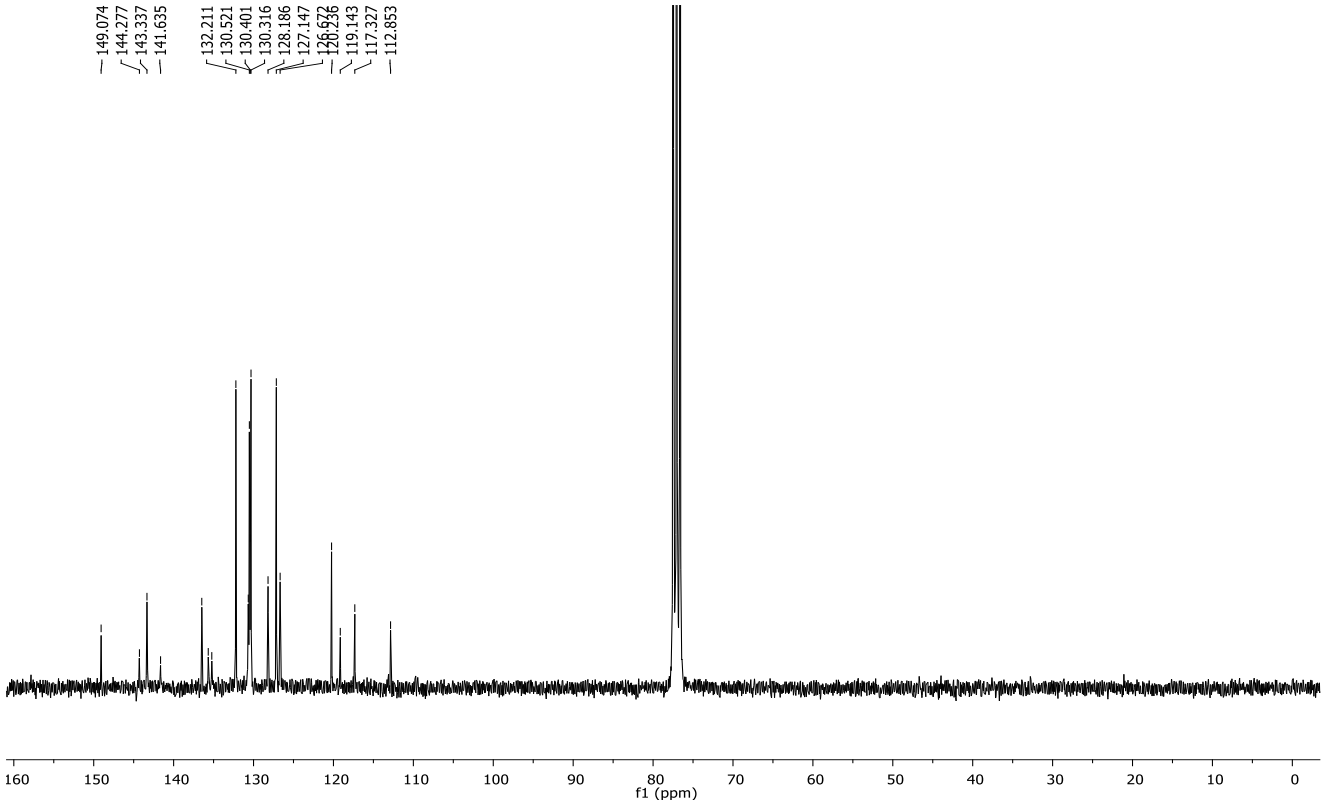


Figure 3.5. ^{1}H and ^{13}C NMR of compound **1,4-Diphenyl-1*H*-imidazo[4,5-*c*]quinoline (31)** in $CDCl_3$

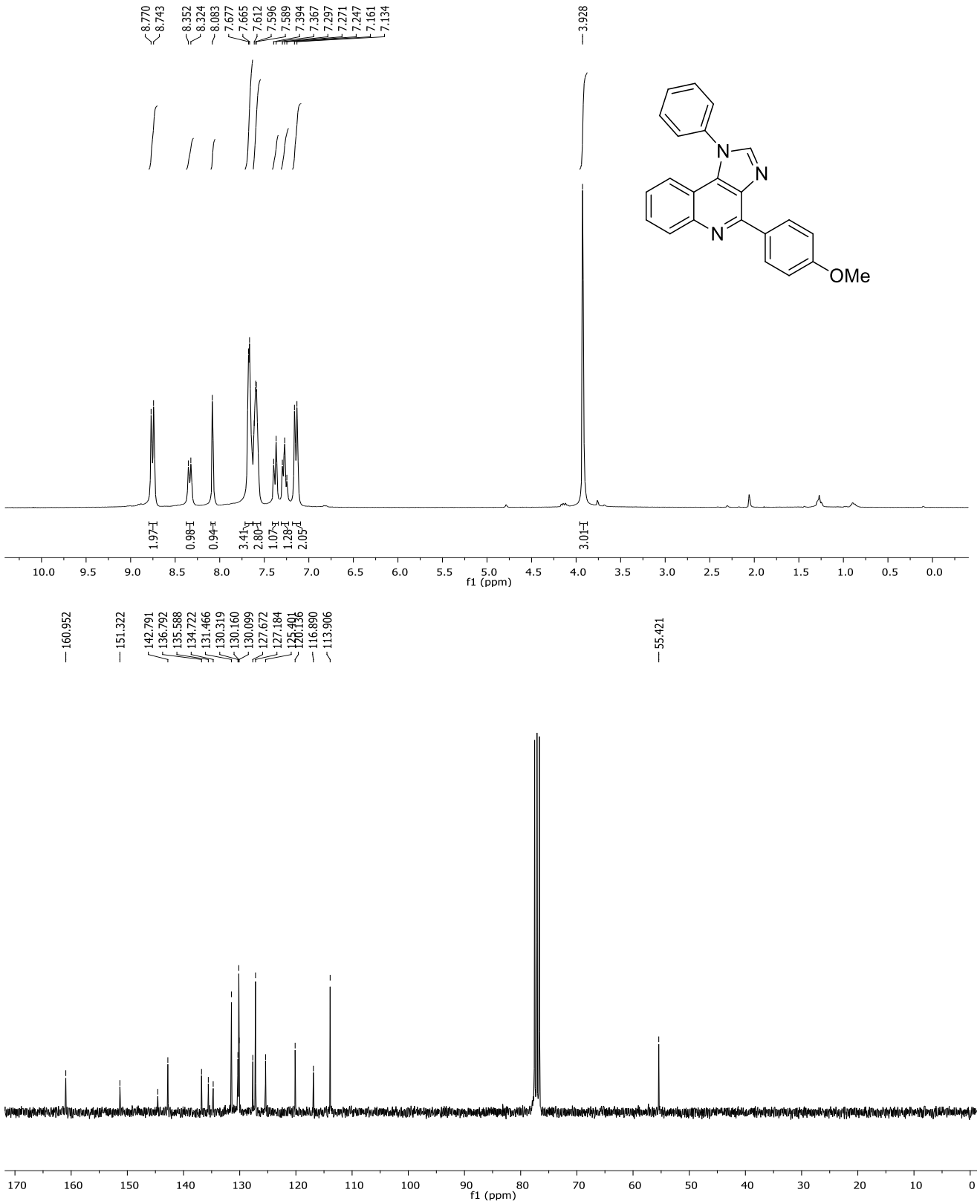


^{1}H and ^{13}C NMR of compound **1-Phenyl-4-(4-(trifluoromethyl)phenyl)-1H-imidazo[4,5-c]quinoline (32)** in $CDCl_3$

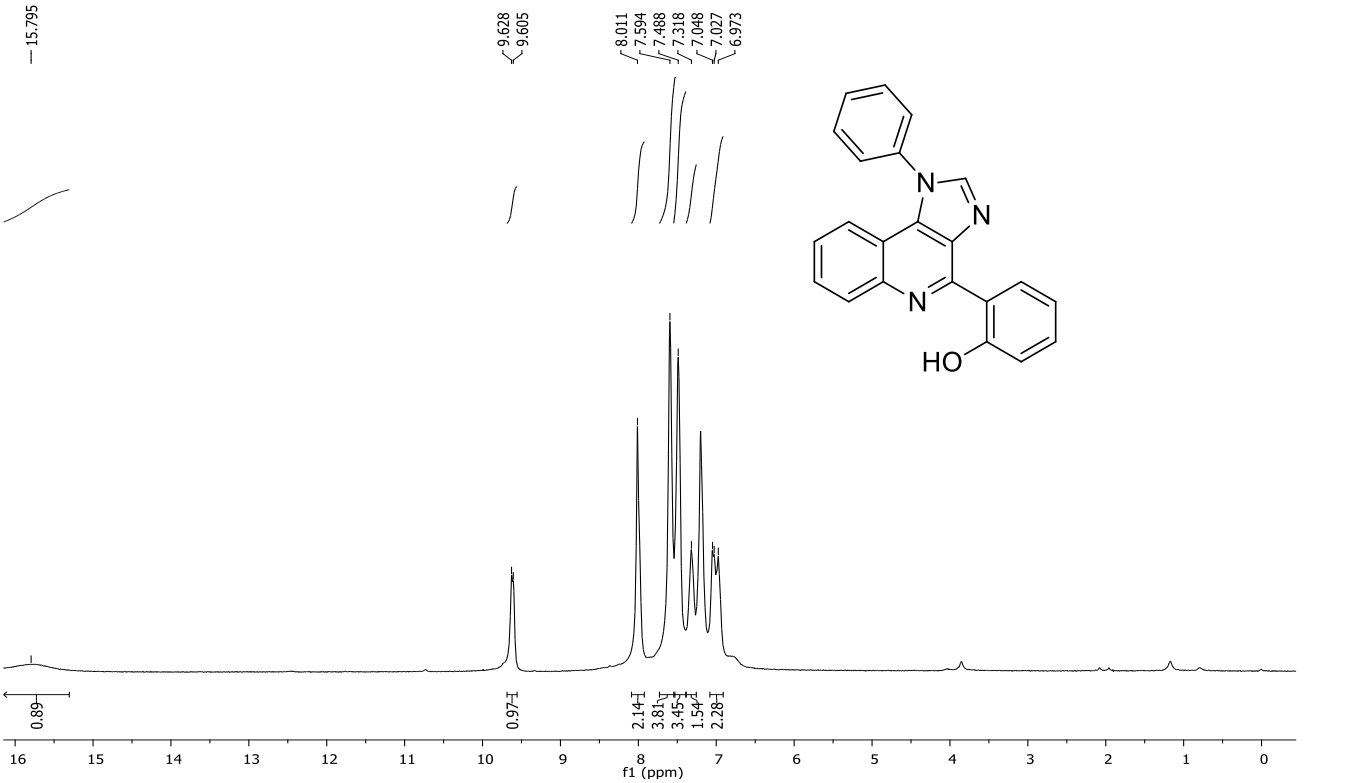


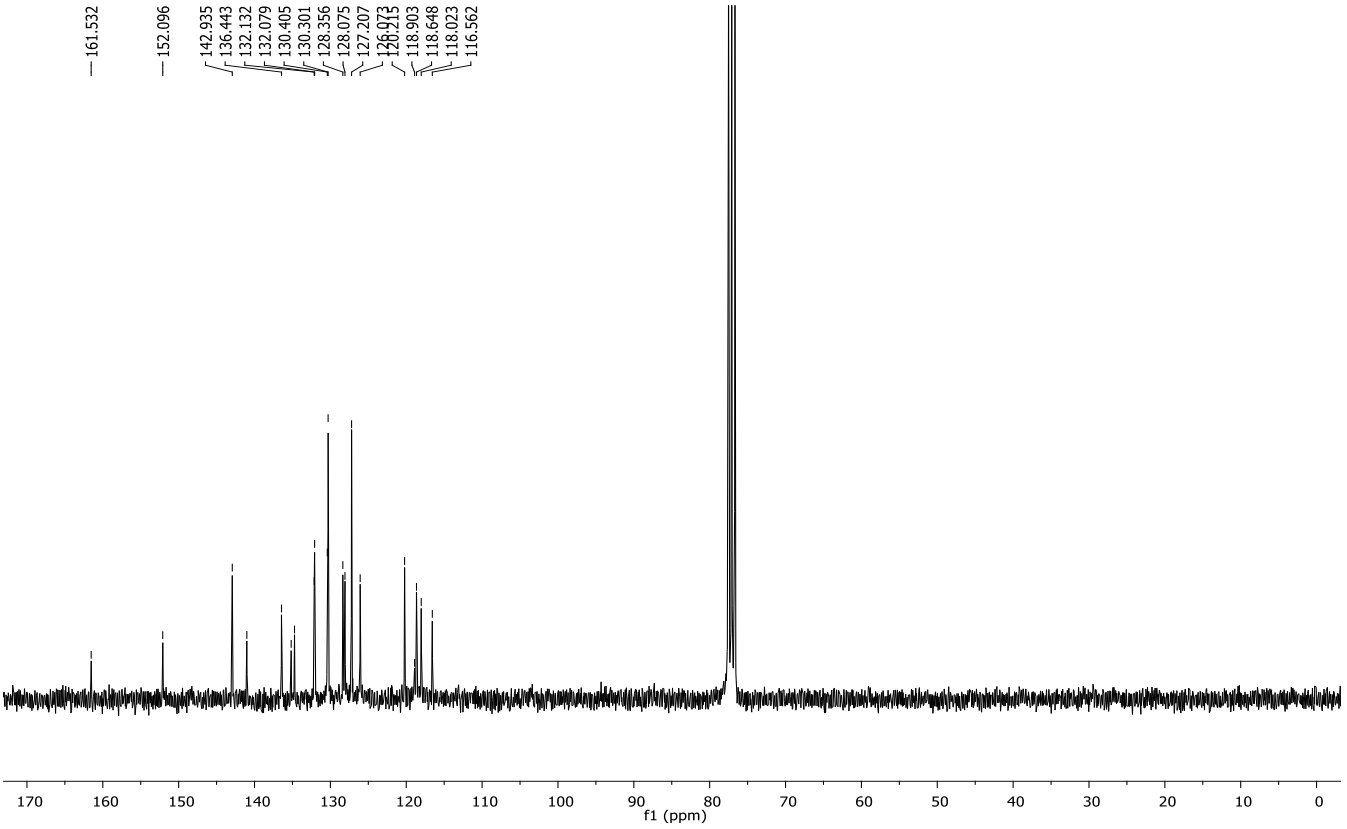


^{1}H and ^{13}C NMR of compound **4-(1-Phenyl-1H-imidazo[4,5-c]quinolin-4-yl)benzonitrile (33)** in $CDCl_3$

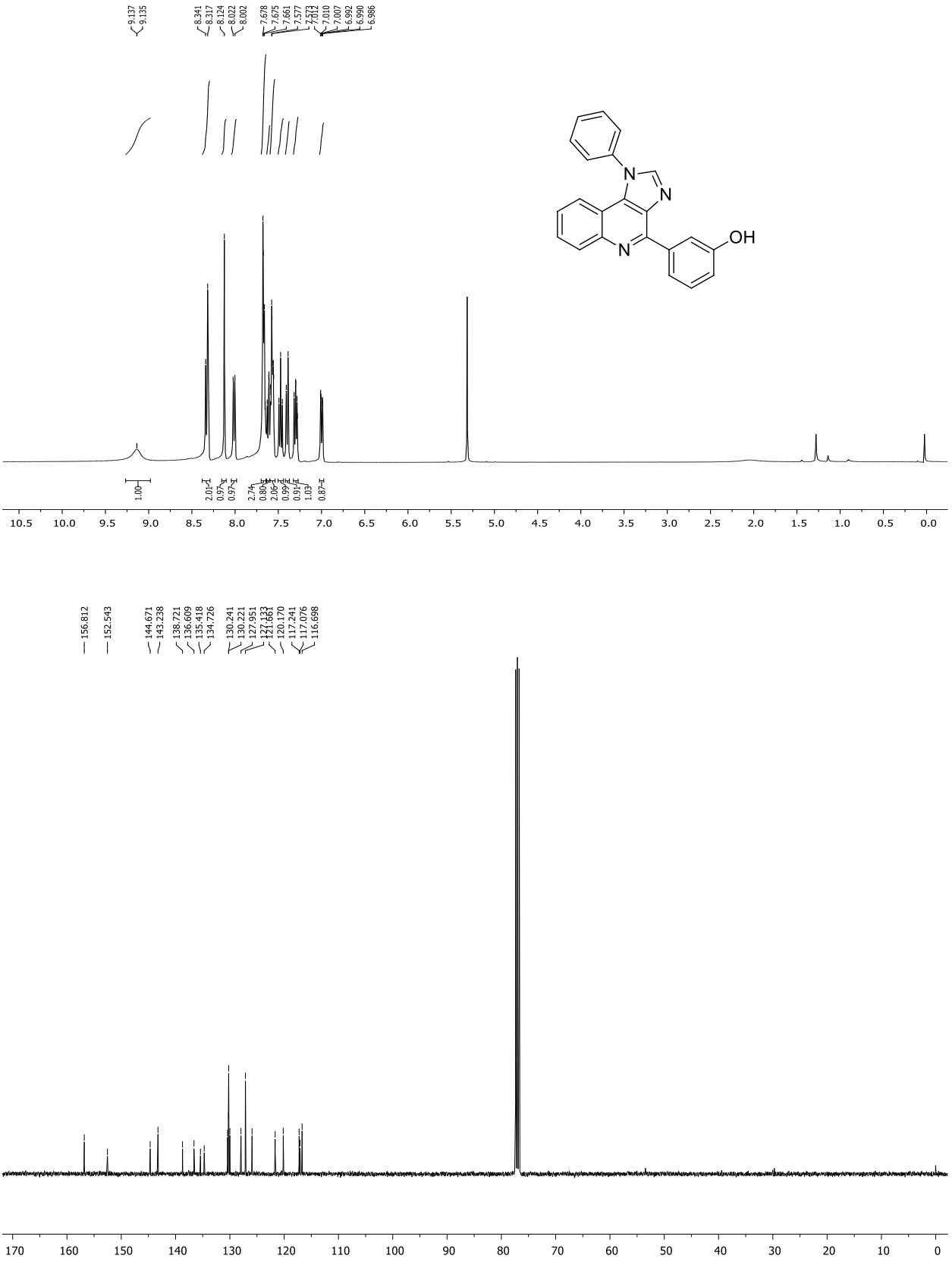


^{1}H and ^{13}C NMR of compound **4-(4-Methoxyphenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (34)** in $CDCl_3$

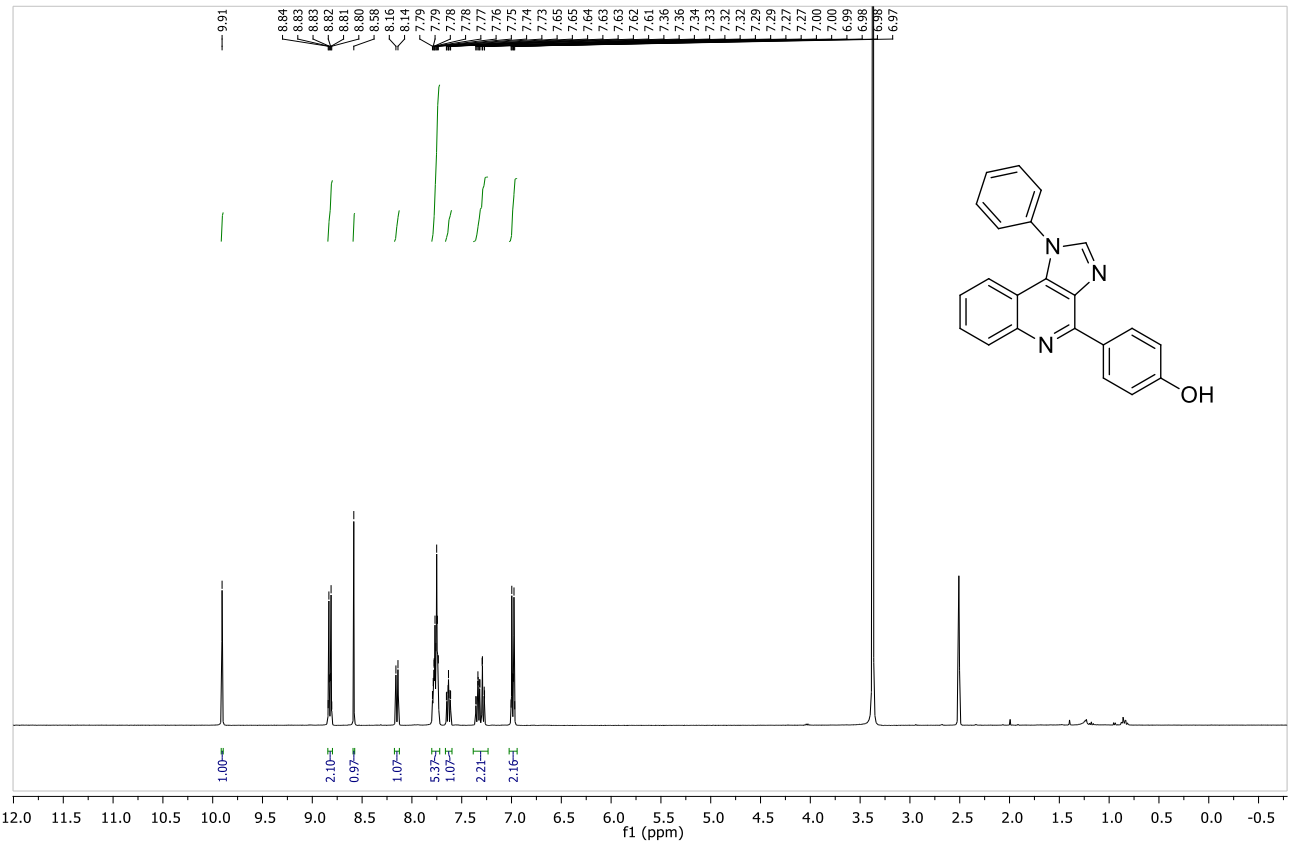


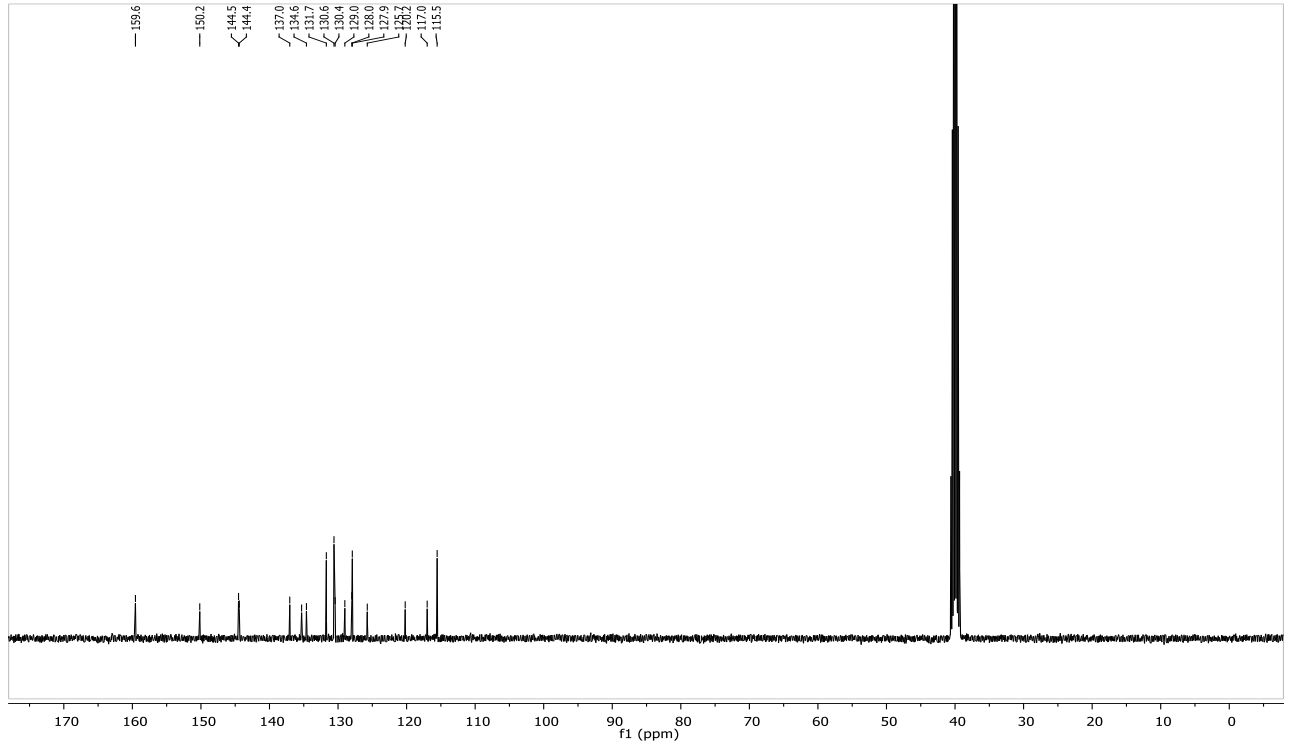


^{1}H and ^{13}C NMR of compound **2-(1-Phenyl-1H-imidazo[4,5-c]quinolin-4-yl)phenol (35)** in $CDCl_3$

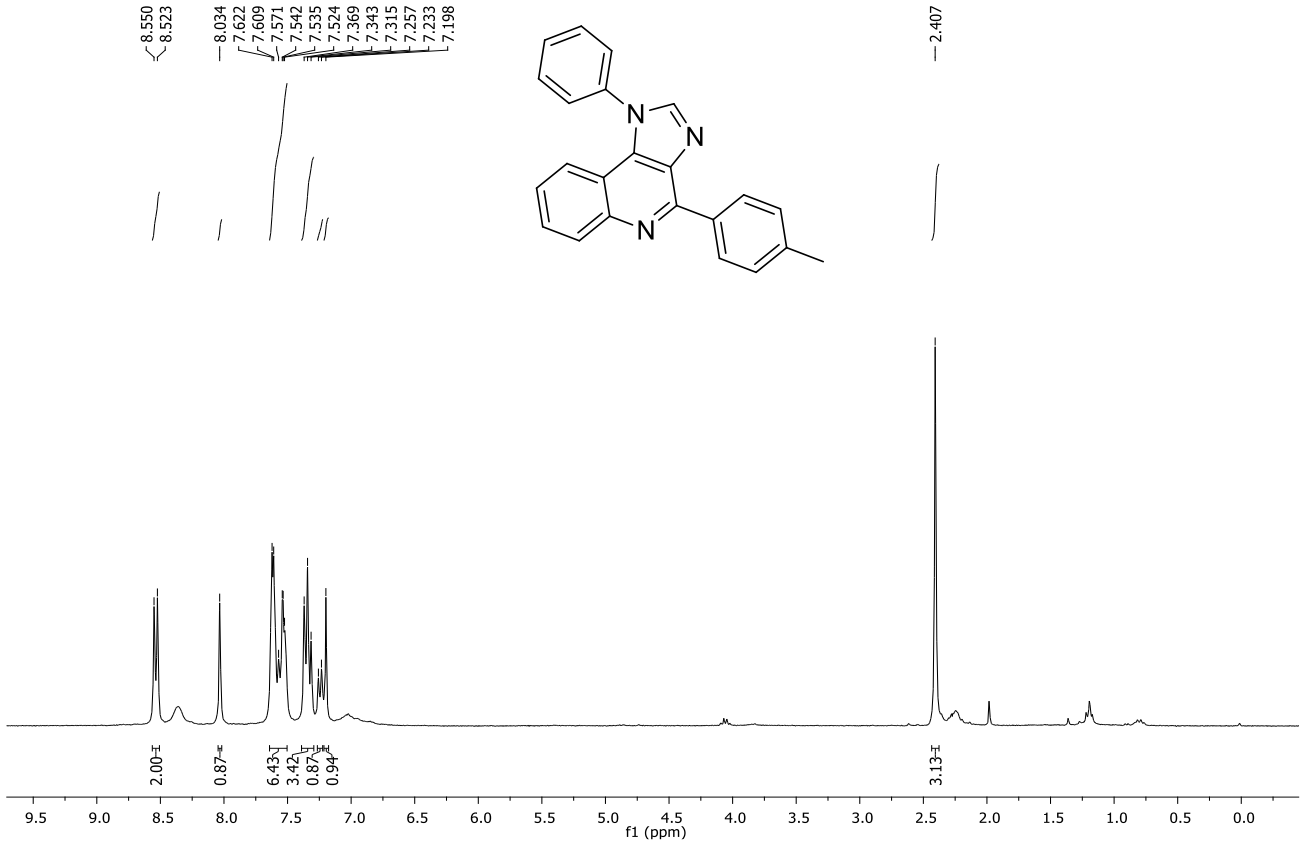


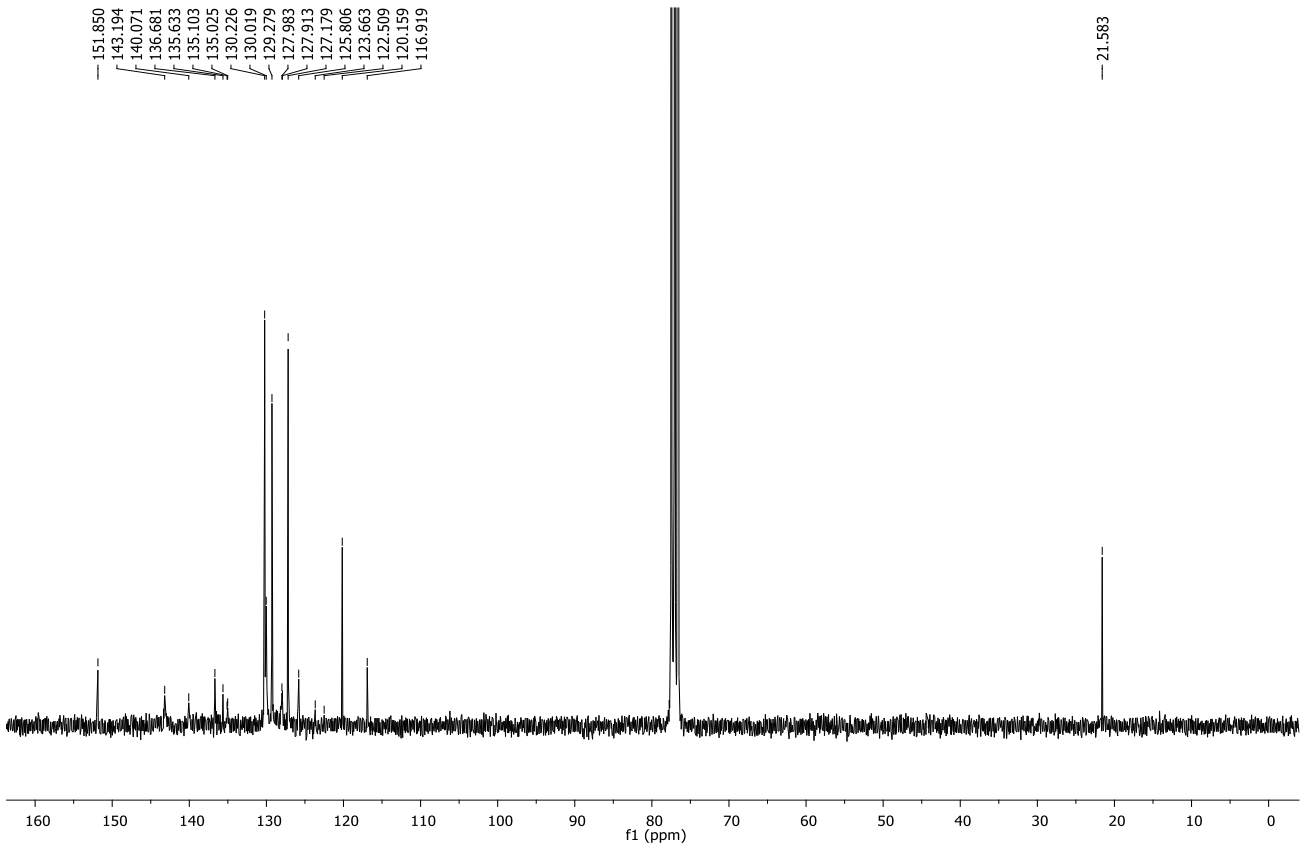
^{1}H and ^{13}C NMR of compound **3-(1-Phenyl-1H-imidazo[4,5-c]quinolin-4-yl)phenol (36)** in $CDCl_3$



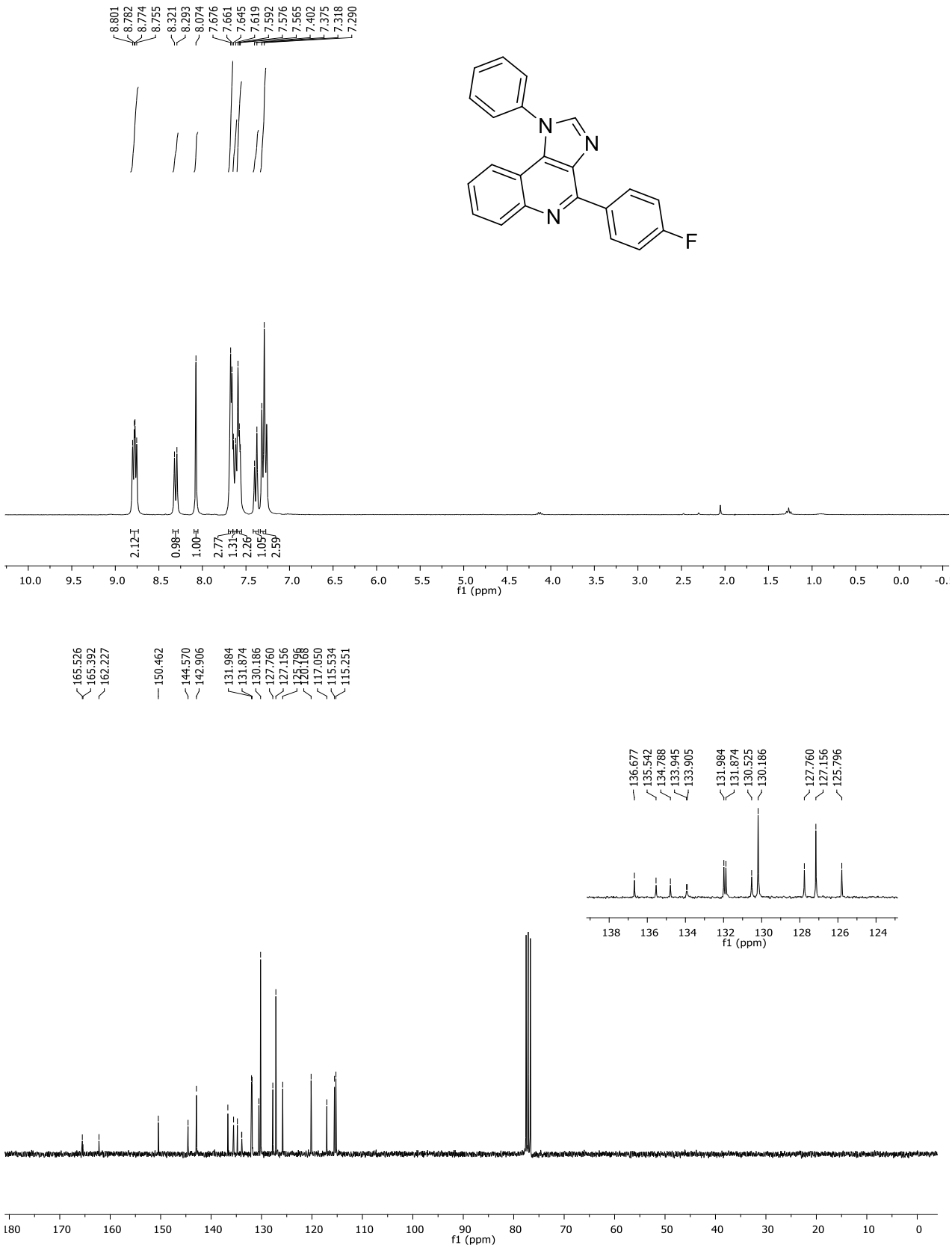


^{1}H and ^{13}C NMR of compound **4-(1-Phenyl-1H-imidazo[4,5-c]quinolin-4-yl)phenol (37)** in $CDCl_3$

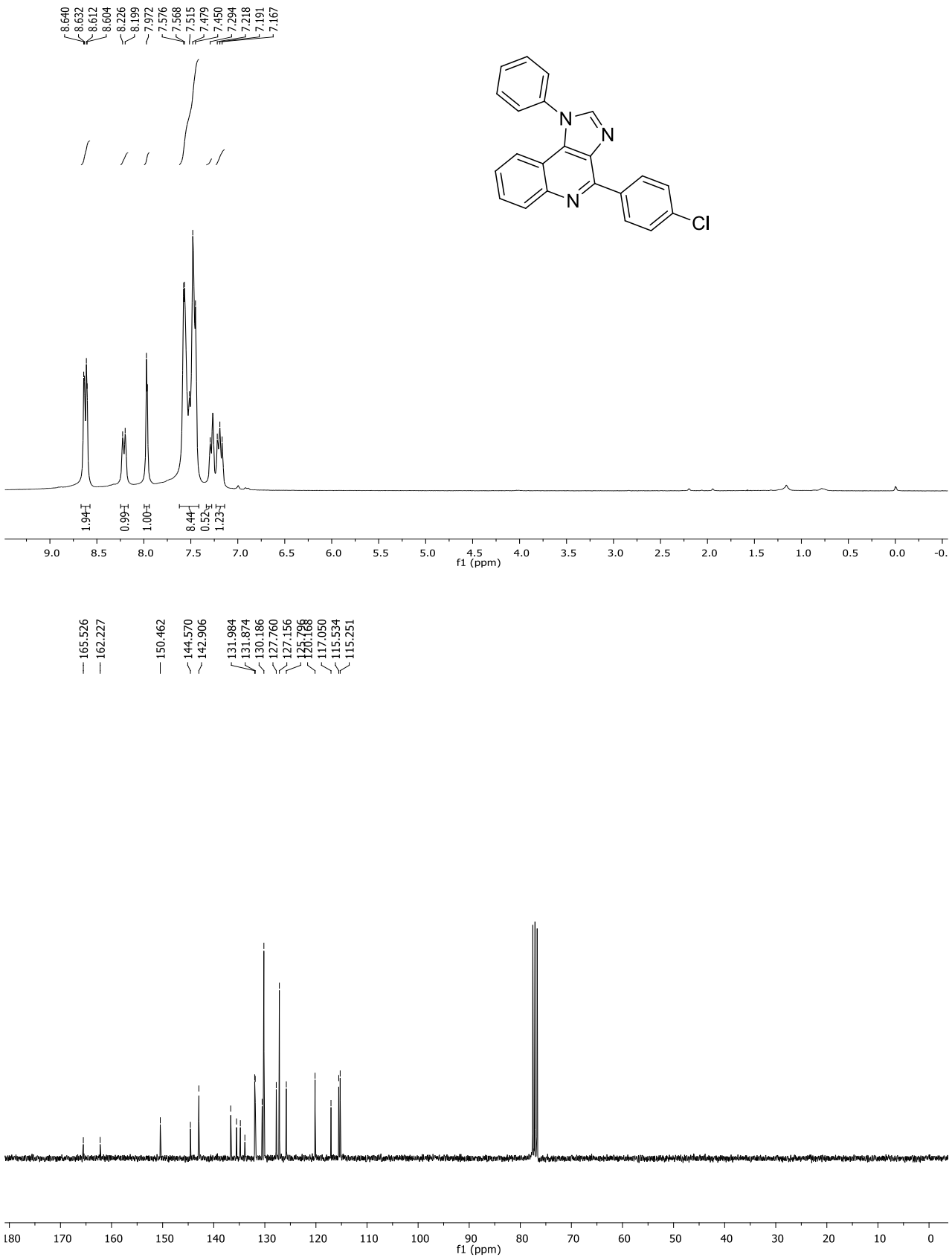




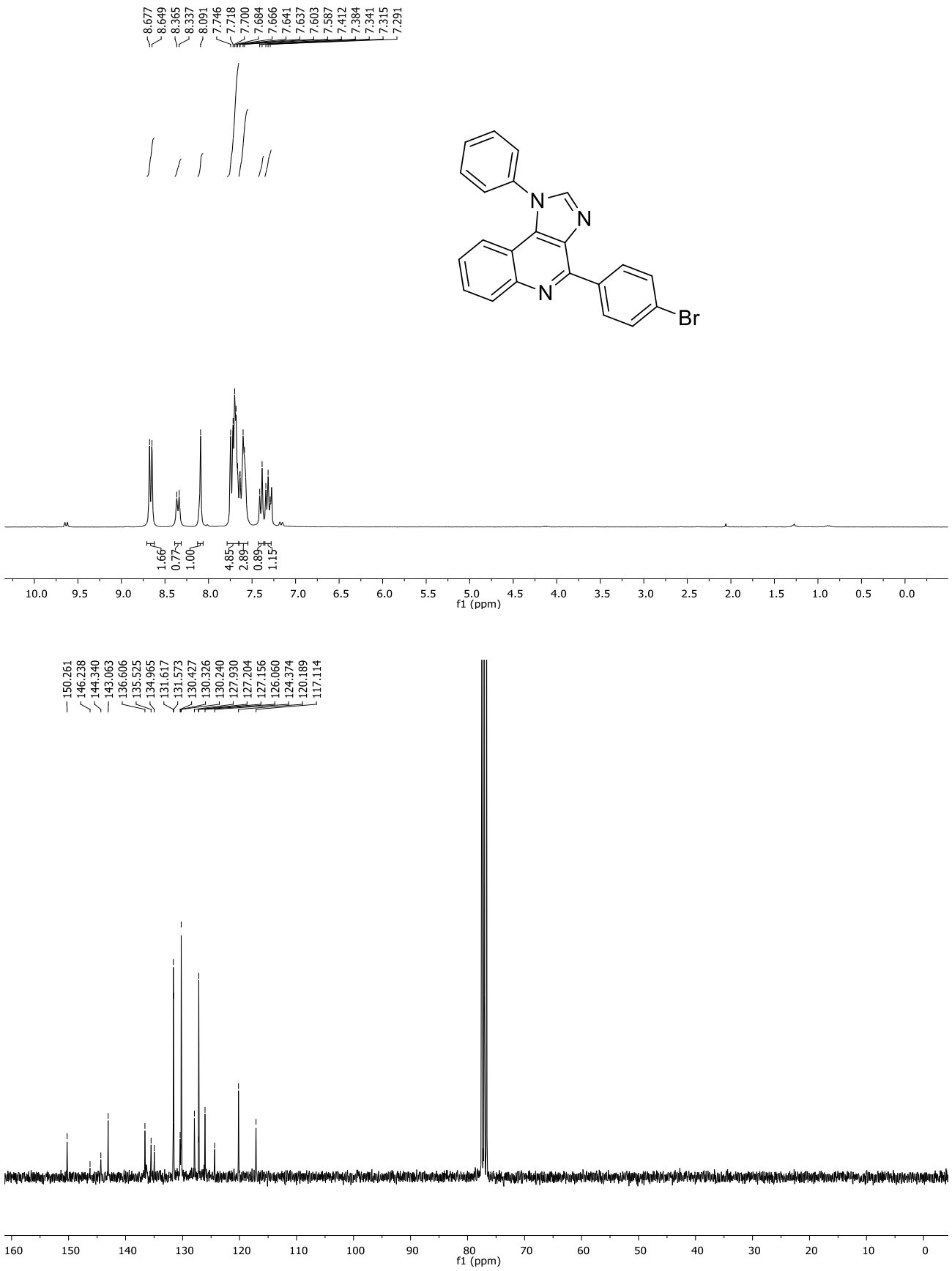
^{1}H and ^{13}C NMR of compound **1-Phenyl-4-p-tolyl-1H-imidazo[4,5-c]quinoline (38)** in $CDCl_3$



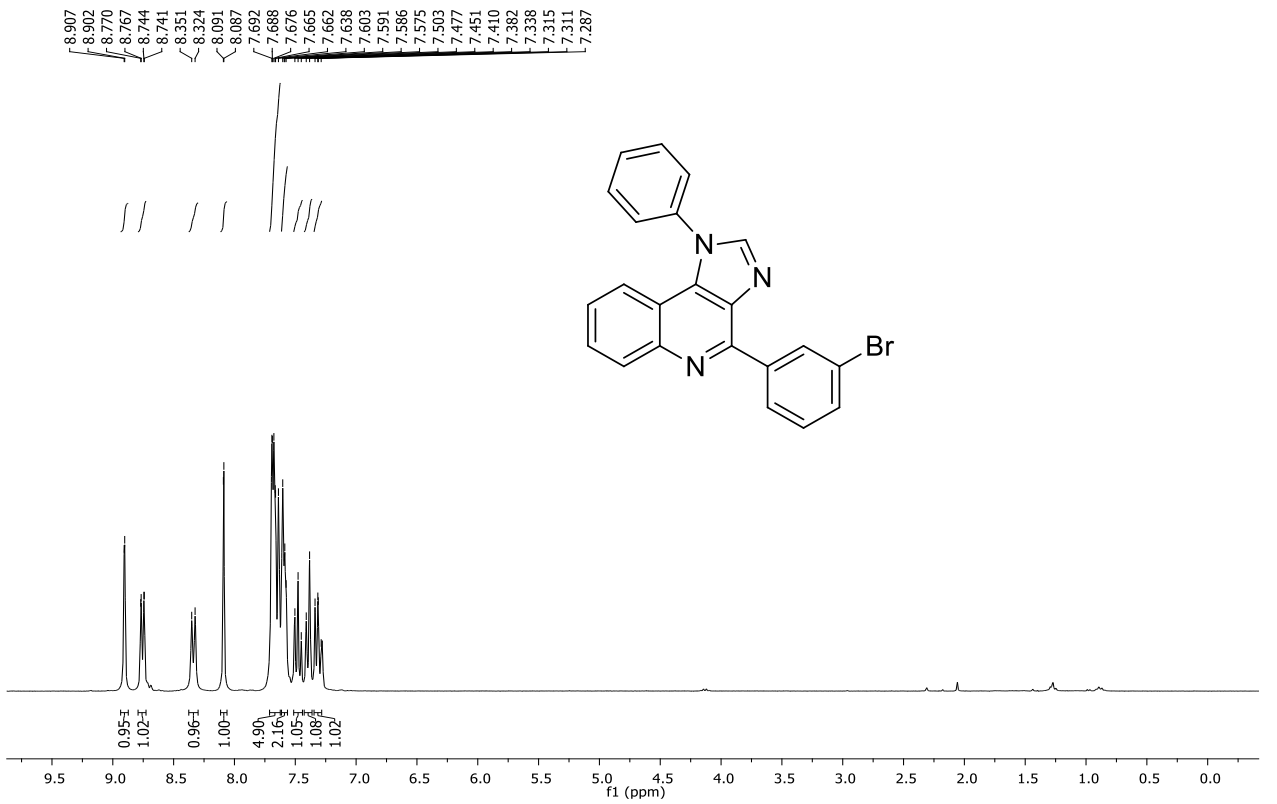
^{1}H and ^{13}C NMR of compound **4-(4-Fluorophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (39)** in $CDCl_3$

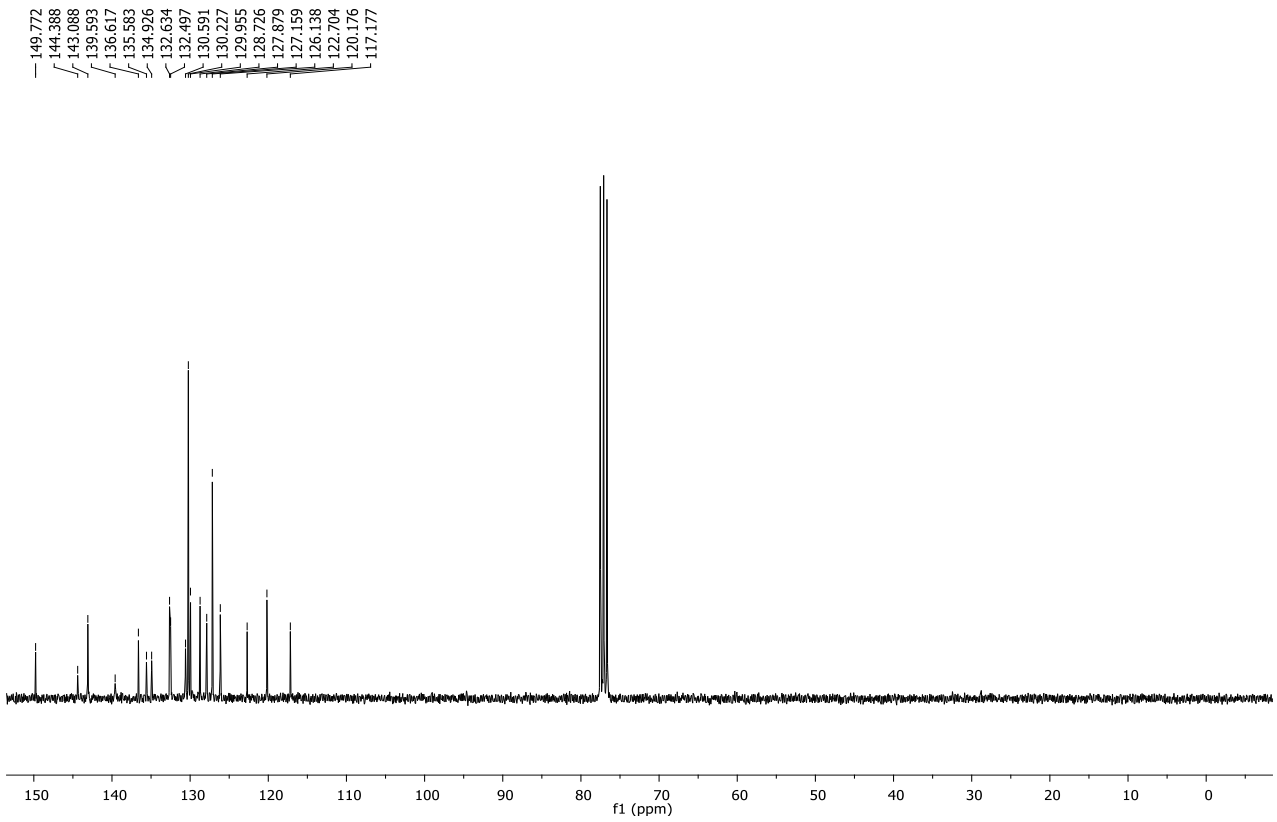


^{1}H and ^{13}C NMR of compound **4-(4-Chlorophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (40)** in $CDCl_3$

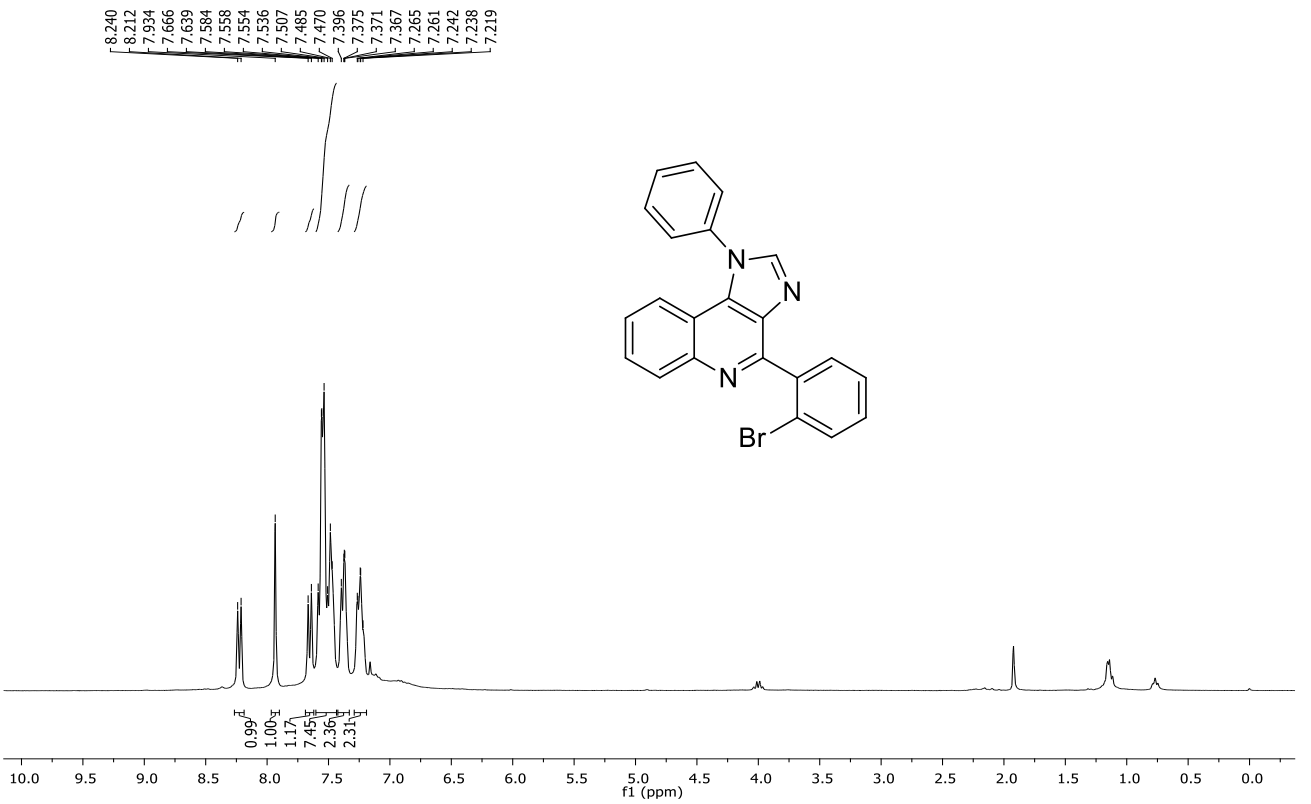


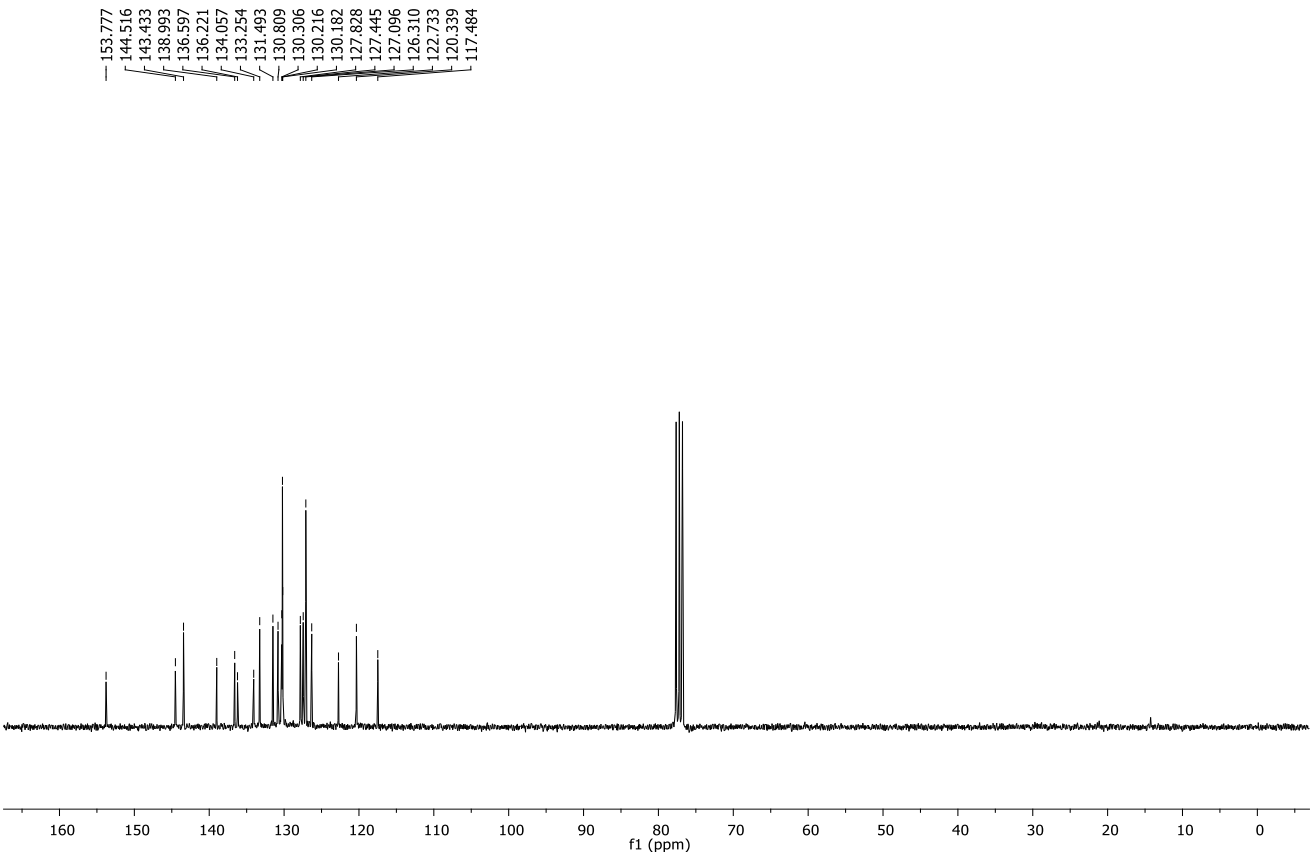
^{1}H and ^{13}C NMR of compound **4-(4-Bromophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (41)** in $CDCl_3$



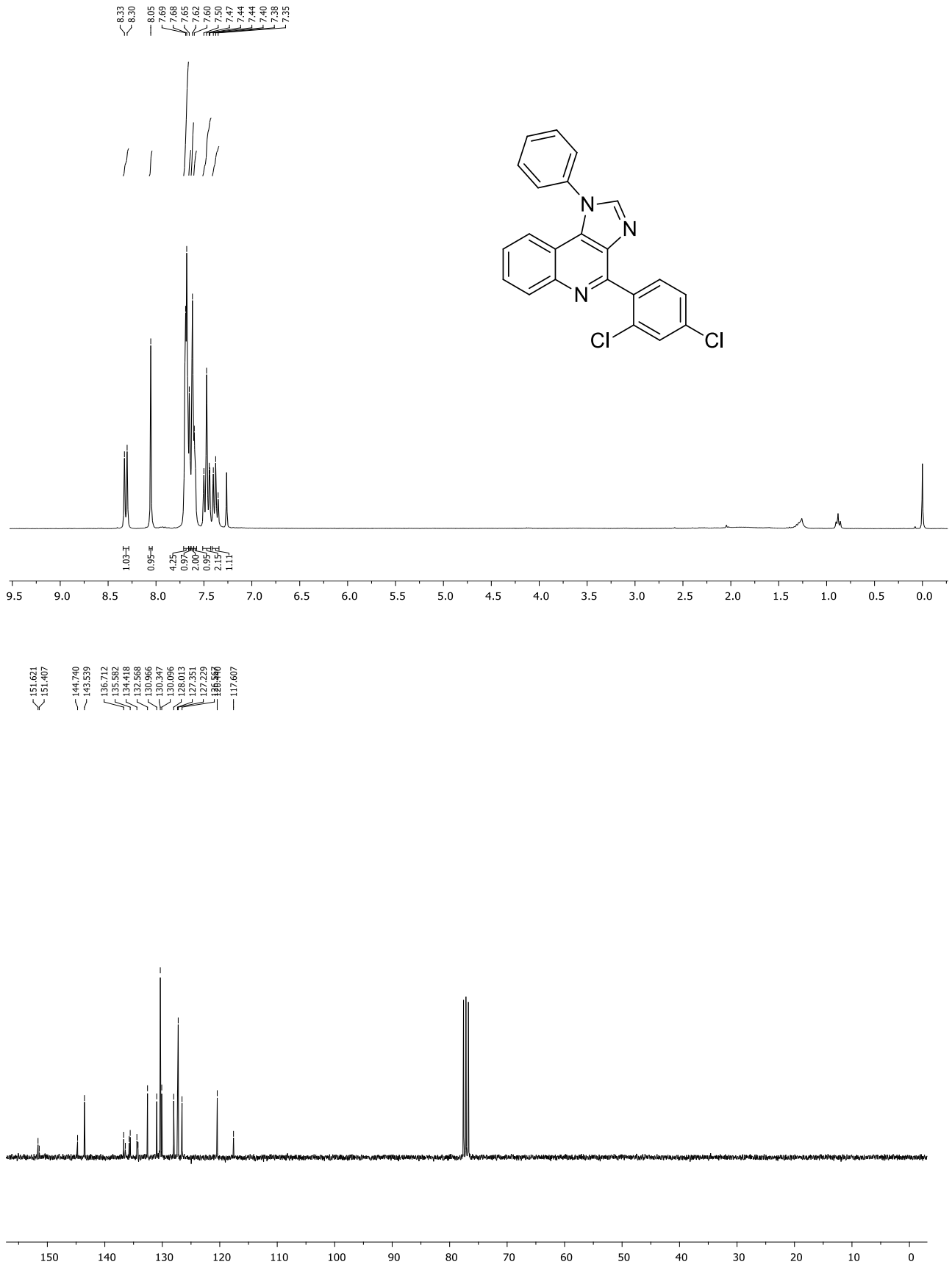


^{1}H and ^{13}C NMR of compound **4-(3-Bromophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (42)** in $CDCl_3$

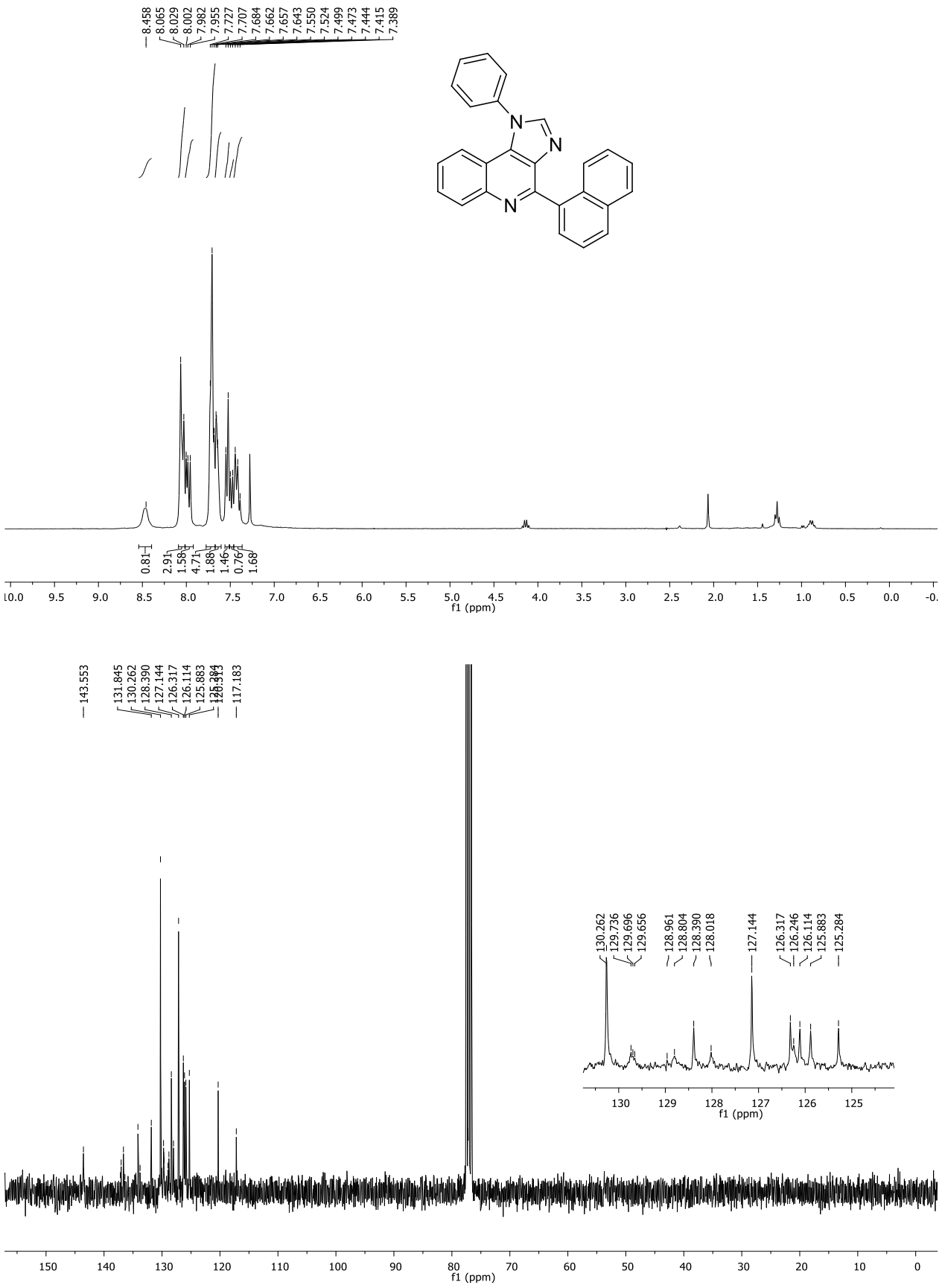




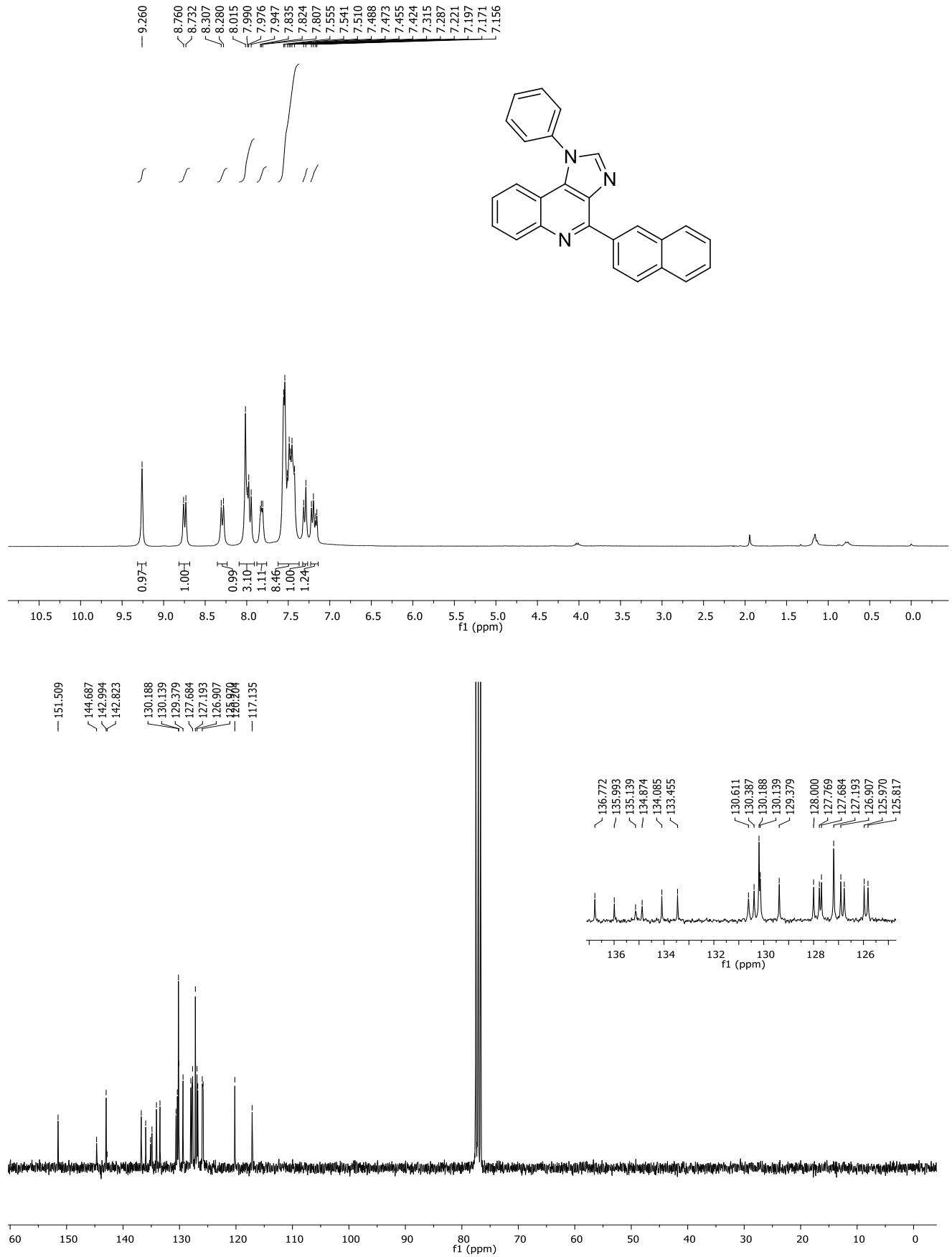
^{1}H and ^{13}C NMR of compound **4-(2-Bromophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (43)** in $CDCl_3$



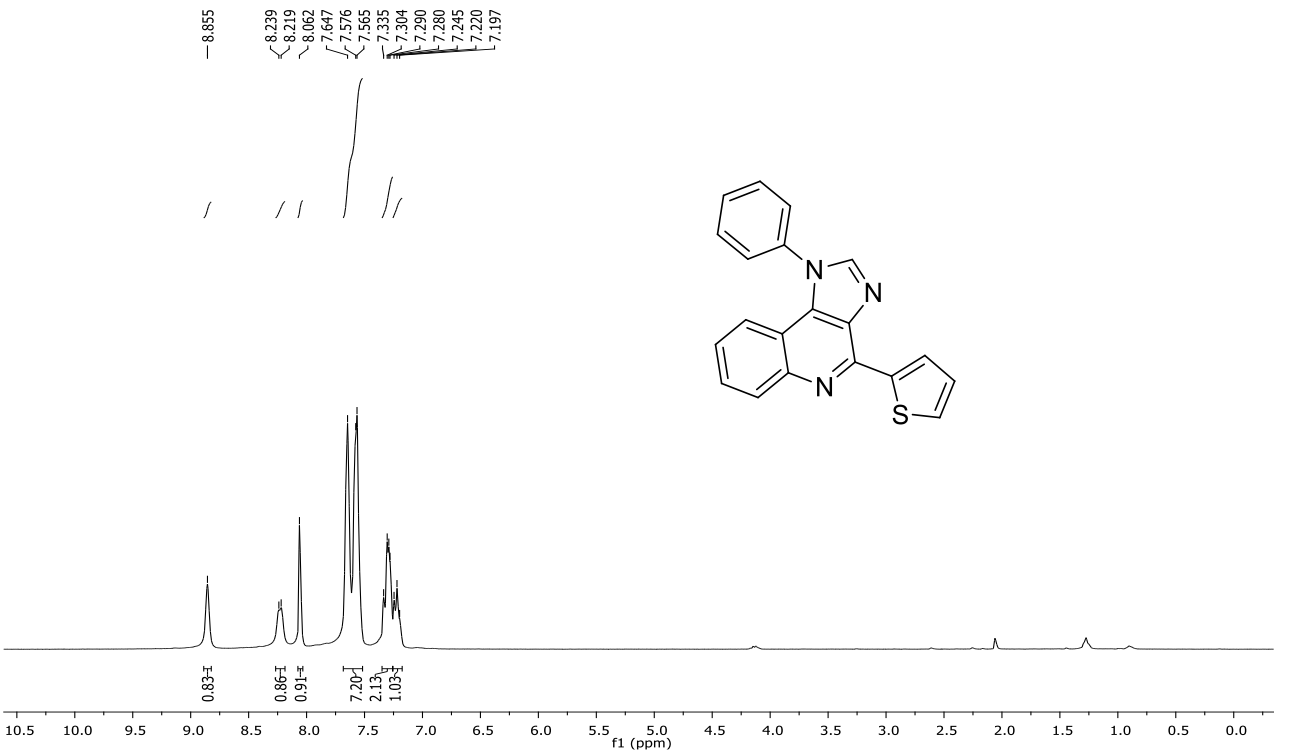
^{1}H and ^{13}C NMR of compound **4-(2,4-Dichlorophenyl)-1-phenyl-1H-imidazo[4,5-c]quinoline (44)** in $CDCl_3$

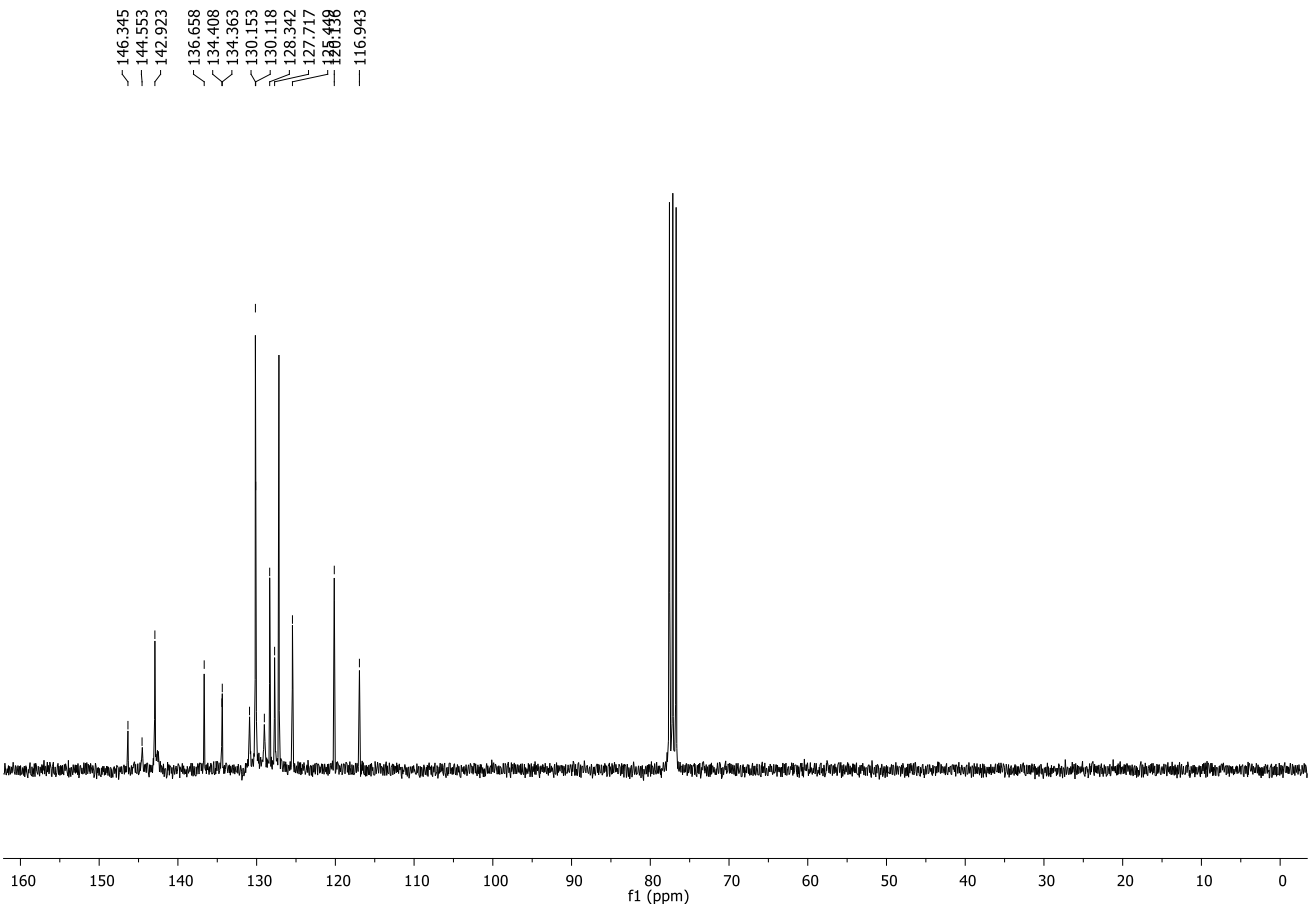


^{1}H and ^{13}C NMR of compound **4-(Naphthalen-1-yl)-1-phenyl-1H-imidazo[4,5-c]quinoline (45)** in $CDCl_3$

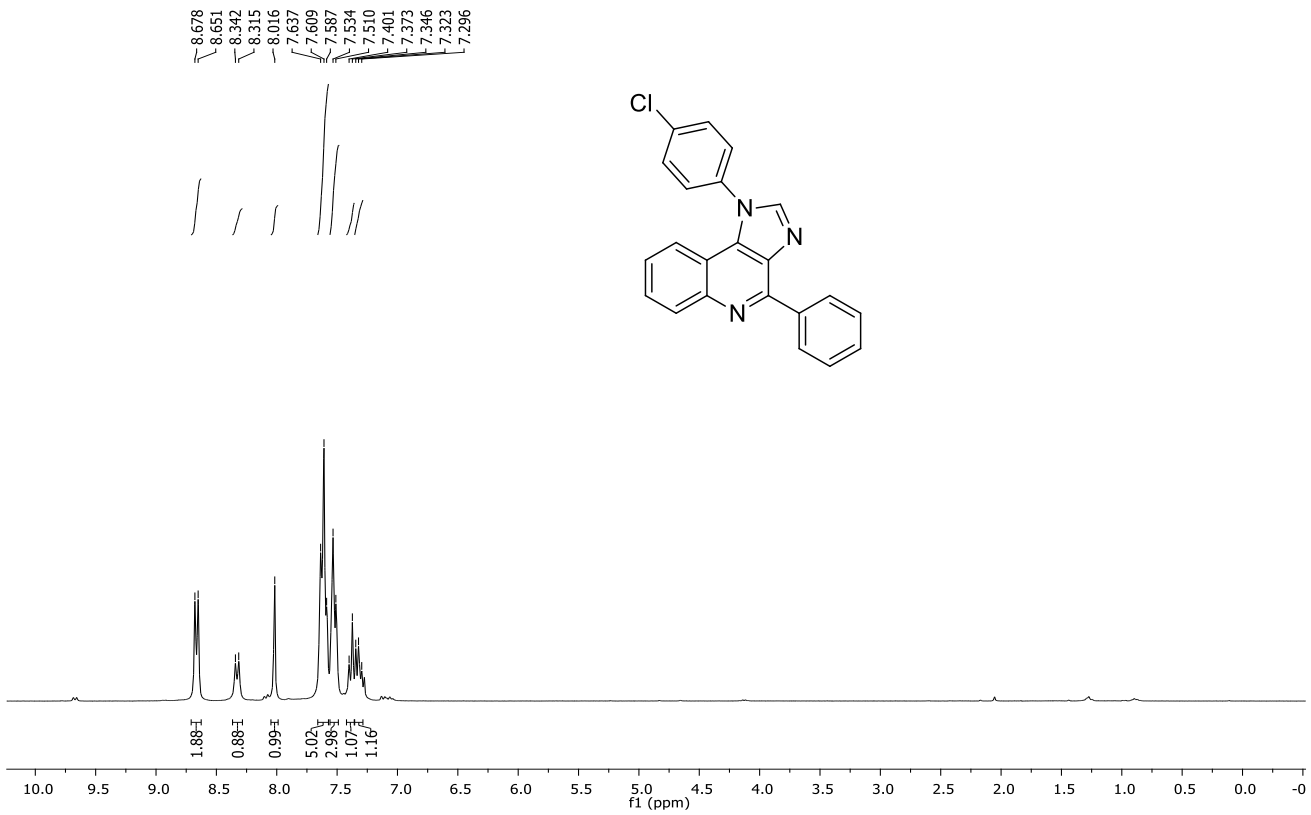


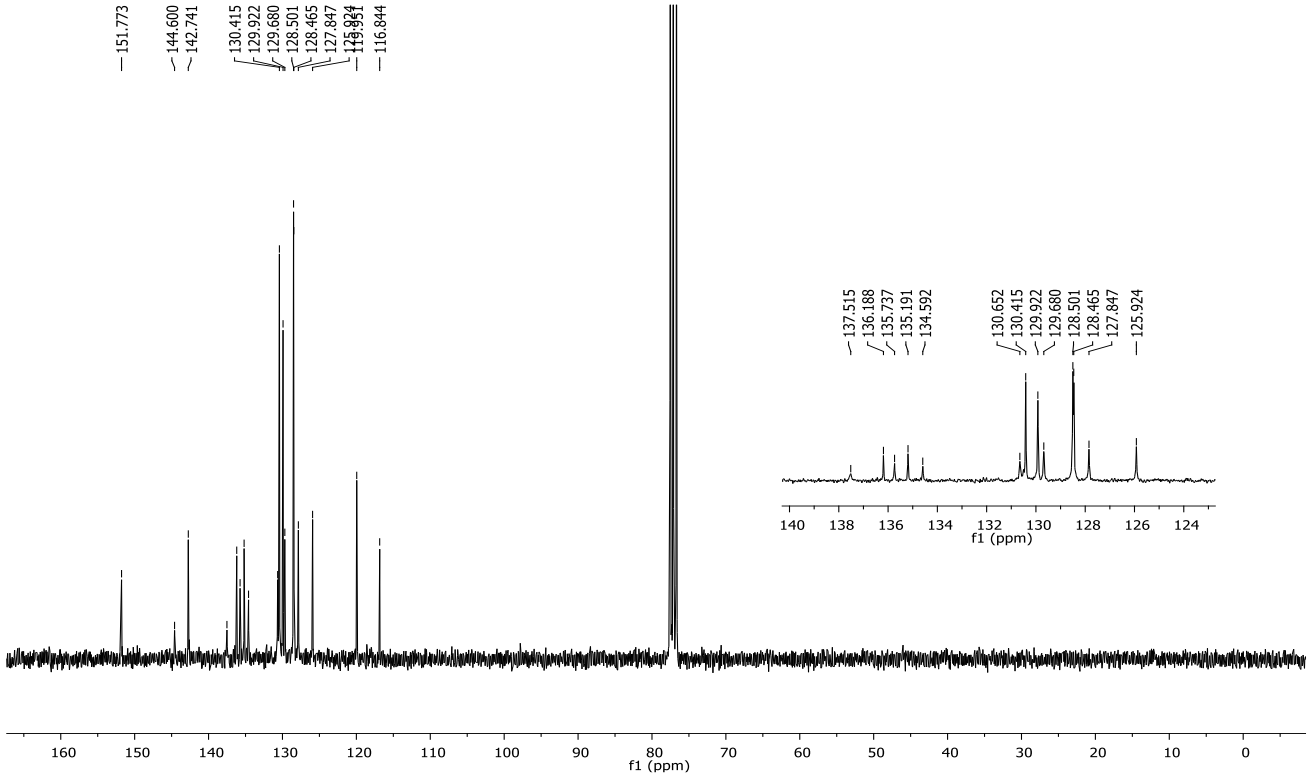
^{1}H and ^{13}C NMR of compound **4-(Naphthalen-2-yl)-1-phenyl-1H-imidazo[4,5-c]quinoline (46)** in $CDCl_3$



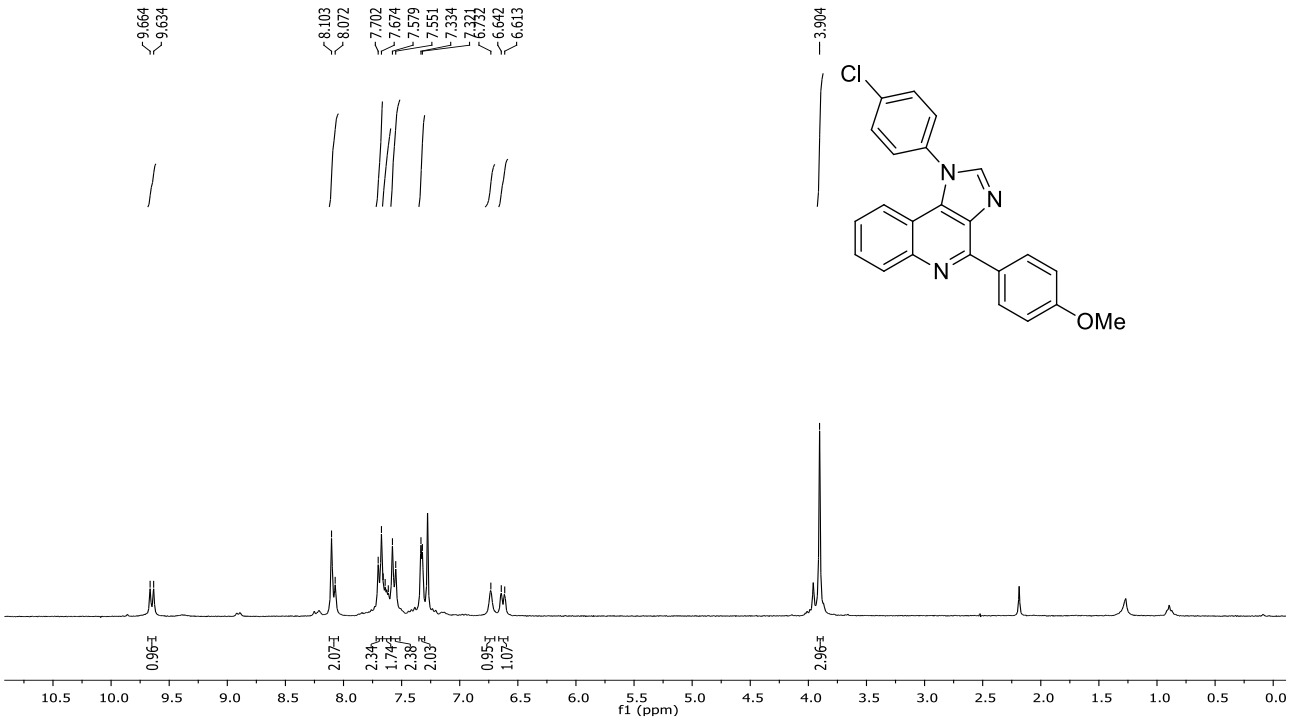


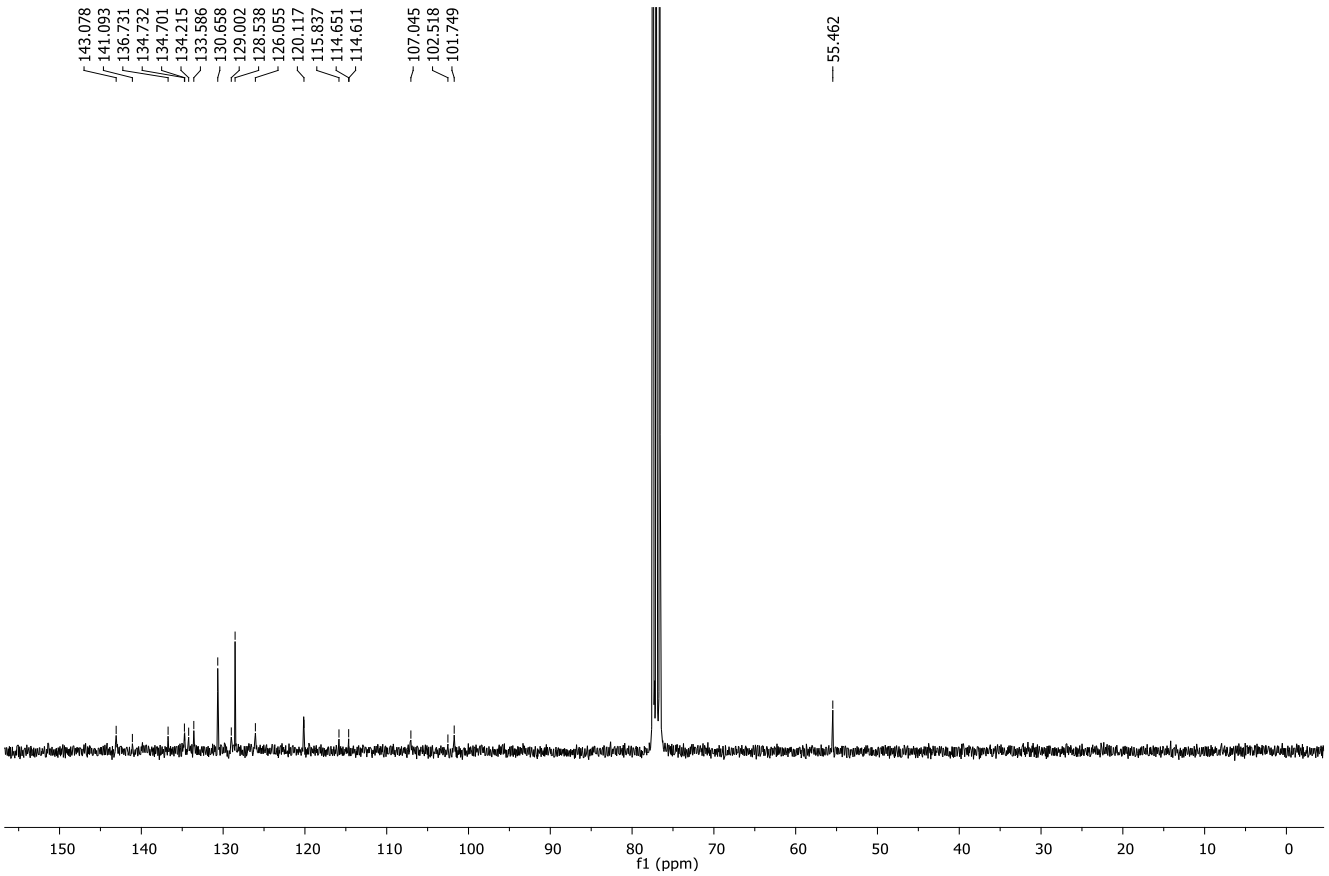
^{1}H and ^{13}C NMR of compound **1-Phenyl-4-(thiophen-2-yl)-1H-imidazo[4,5-c]quinoline (47)** in $CDCl_3$



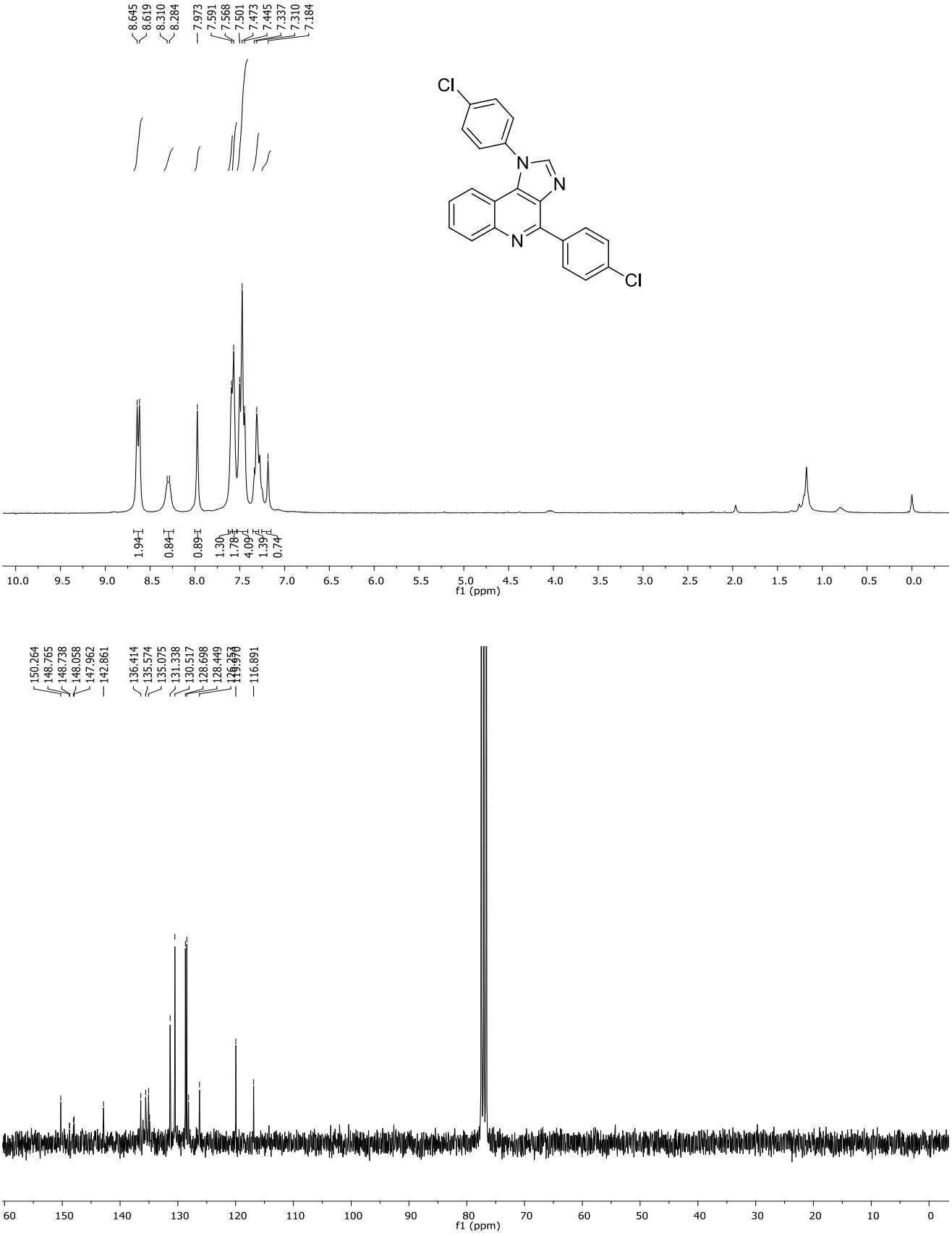


^{1}H and ^{13}C NMR of compound **1-(4-Chlorophenyl)-4-phenyl-1*H*-imidazo[4,5-*c*]quinoline (48)** in $CDCl_3$

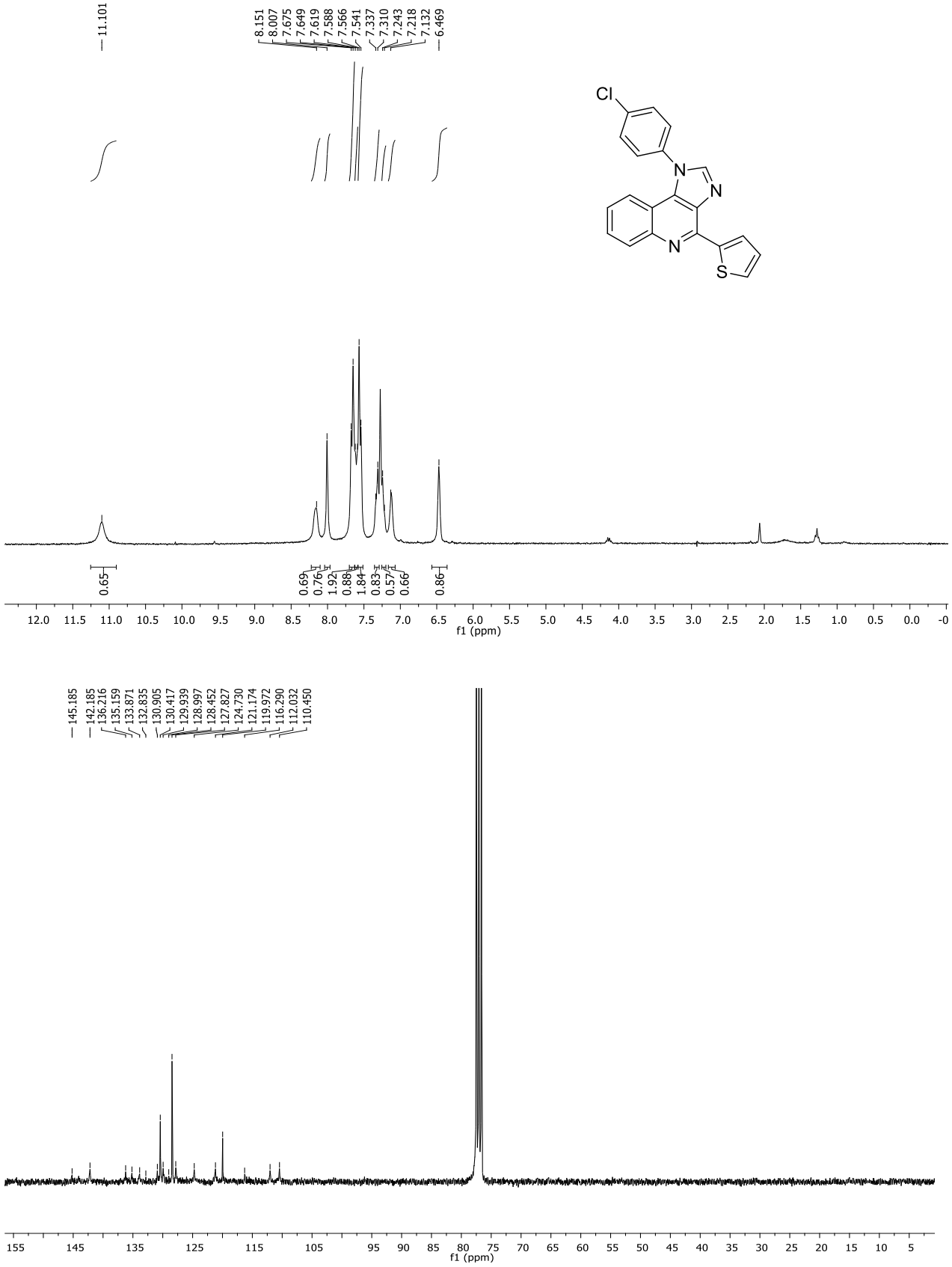




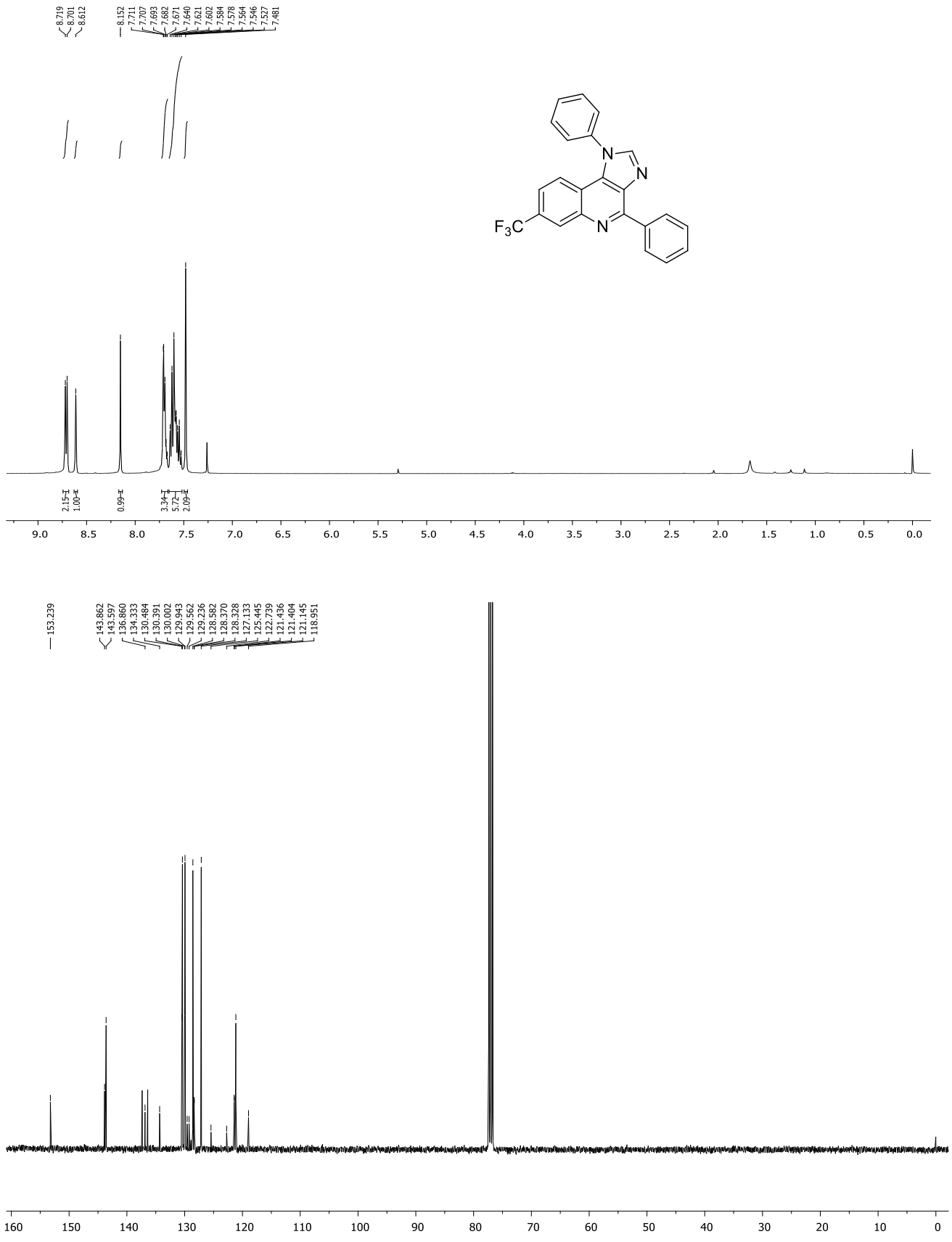
^{1}H and ^{13}C NMR of compound **1-(4-Chlorophenyl)-4-(4-methoxyphenyl)-1*H*-imidazo[4,5-*c*] quinoline (49)** in $CDCl_3$



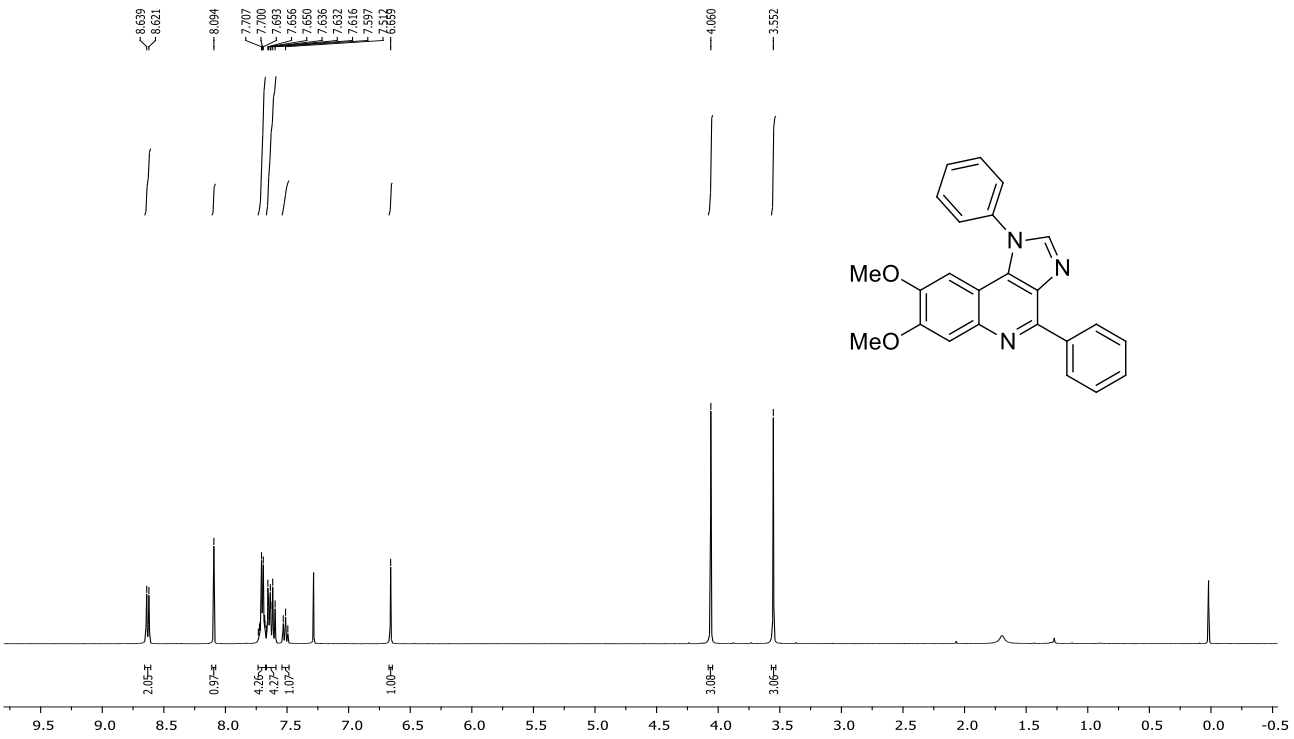
^{1}H and ^{13}C NMR of compound **1,4-Bis(4-chlorophenyl)-1*H*-imidazo[4,5-*c*]quinoline (50)** in $CDCl_3$

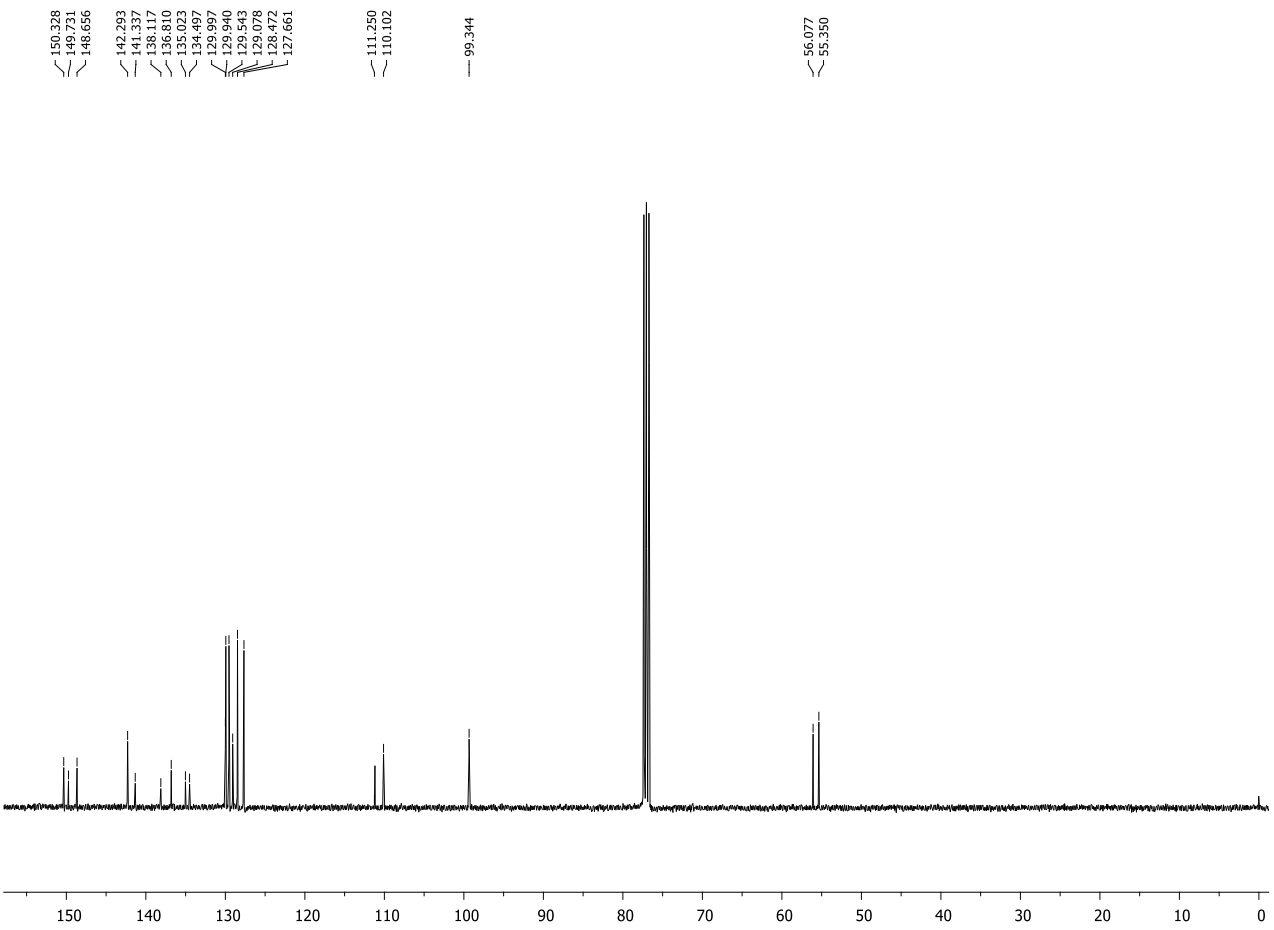


^{1}H and ^{13}C NMR of compound **1-(4-Chlorophenyl)-4-(thiophen-2-yl)-1*H*-imidazo[4,5-*c*]quinoline (51)** in $CDCl_3$



^{1}H and ^{13}C NMR of compound **1,4-Diphenyl-7-(trifluoromethyl)-1*H*-imidazo[4,5-*c*]quinoline (52)** in $CDCl_3$





^{1}H and ^{13}C NMR of compound **7,8-Dimethoxy-1,4-diphenyl-1*H*-imidazo[4,5-*c*]quinoline (53)** in $CDCl_3$

3.10. References

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CHAPTER 4

Synthesis and anticancer activity of fused chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds

4.1. Introduction

Coumarins are one of the most recognizable scaffolds in the domain of organic/medicinal chemistry. They are found in numerous natural products both as a multi-substituted single unit as well as in fused or combined formats with other compounds. Coumarin based compounds represent an unusual group of structurally diverse secondary metabolites **(Figure 4.1)**. These well-known aromatic lactones, isolated from a variety of plant sources, are known to possess diverse bioactivities, such as anticancer,[1] antiviral,[2] topoisomerase I inhibitor,[3] HIV integrase inhibitor,[4]etc.

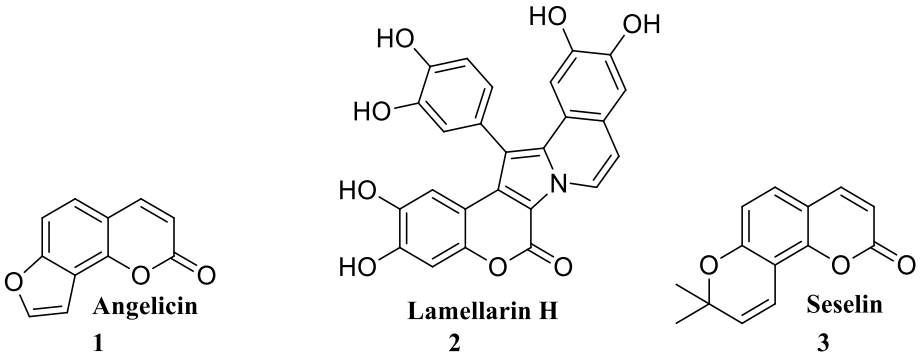
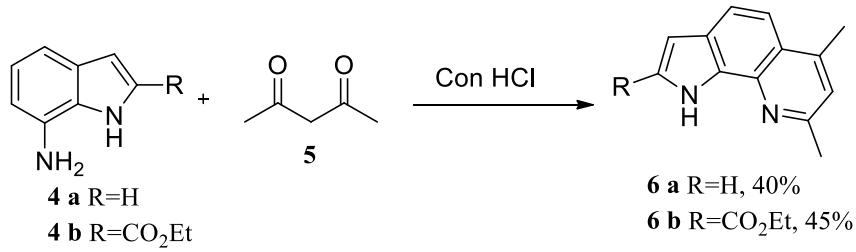


Figure 4.1: Coumarin based molecules from natural sources

In continuation with our research program in the synthesis and applications of fused heterocyclic systems,[5-8] we initiated an exploration in the area of coumarin fused heterocyclic systems. The target envisaged and synthesized was chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one, possessing unique assembly of coumarin, pyrrole and quinoline units. It was felt that bringing together biologically important scaffolds like coumarin, pyrrole and quinoline in a putative planar framework will give rise to unique features in the proposed molecule. Additionally, it was also articulated that the proximity of nitrogen atoms belonging to pyrrole and quinoline, may also serve as a metal binding motif within the molecule.

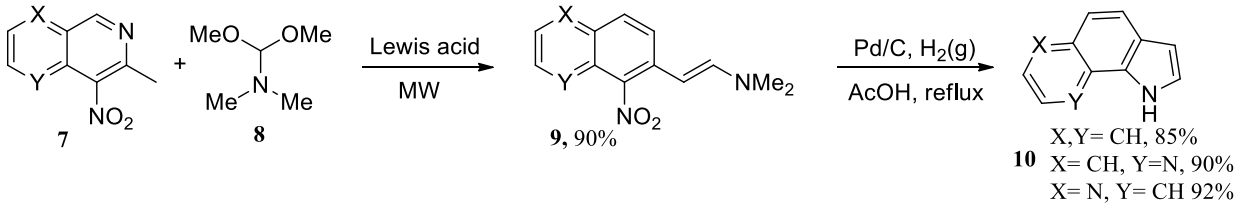
Since the proposed compound represented a hitherto unknown scaffold, the literature examples refer to consisted of synthetic strategies towards pyrrolo[3,2-*h*]quinoline compounds and fused coumarin-dihydroquinoline systems.

Only few papers have been reported in the literature pertaining to synthesis of pyrrolo[3,2-*h*]quinoline compounds. El Ouar *et al*., have reported the synthesis of these molecules in modest yields by reacting ethyl-7-aminoindole-2-carboxylate with various 1,3-diketones at 220 ^{o}C, in the presence of conc. HCl (**Scheme 4.1**).[9]



Scheme 4.1: Synthesis of 6,8-dimethyl-1H-pyrrolo[3,2-*h*]quinoline system by El Ouar *et al*.

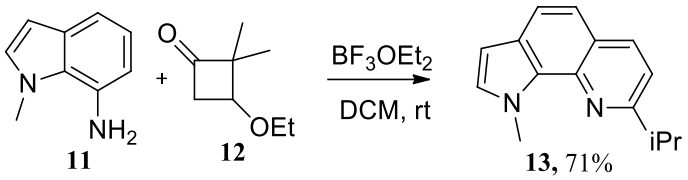
Steve Ley and co-workers, reported synthesis of pyrrolo[3,2-*h*]quinoline **(10)** in their effort towards microwave assisted Leimgruber-Batchoindole synthesis. The molecule was synthesized using 7-methyl-8-nitro-1,6-naphthyridine **(7)** and 1,1'-dimethoxy-*N*,*N*-demthylmethanamine **(8)** as the starting compounds, followed by reduction and cyclization of the intermediate *N*,*N*'-dimethyl-2-(8-nitro-1,6-naphthyridin-7-yl)ethenamine **(9)** to the final product **10** (**Scheme 4.2**).[10]



Scheme 4.2: Synthesis of pyrrolo[3,2-*h*]quinoline derivative systems byJ. Siu *et al*.

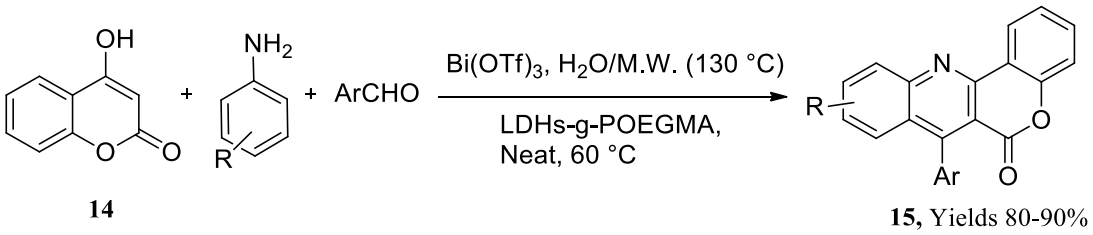
$BF_3.OEt_2$ catalyzed synthesis of several 1,8-di-substituted pyrrolo[3,2-*h*]quinoline compounds were reported by Lin *et al*., in their attempt to generate diverse pyridine derivatives (**Scheme 4.3**).[11] Herein we have shown a representative example which uses 1-

methyl-1*H*-indol-7-amine **(11)** and 3-ethoxy cyclobutanone **(12)** as the starting materials to yield 8-isopropyl-1-methyl-1*H*-pyrrolo[3,2-*h*]quinoline **(13)** as the final product.



Scheme 4.3: Synthesis of pyrrolo[3,2-*h*]quinoline system by Lin *et al*.

Few examples pertaining to synthesis of fused coumarin-dihydroquinoline systems **(15)** are known in the literature.[12, 13] These examples report application of multicomponent 6π-electrocyclization reactions *en-route* to formation of fused coumarin-dihydroquinoline systems **(Scheme 4.4**). In a green synthetic route devised by Reddy *et al*. various chromene incorporated dihydroquinoline molecules were prepared by using reusable poly(oligoethylene glycol methacrylate)-*g*-supported layered double hydroxides (LDHs-g-POEGMA) as catalyst.[12] 4-hydroxy coumarin **(14)**, diverse anilines and aromatic aldehydes were used as the starting molecules. Using a similar approach, Khan *et al*., reported synthesis of **15** with $Bi(OTf)_3$ as the catalyst under aqueous conditions and microwave irradiation.[13]



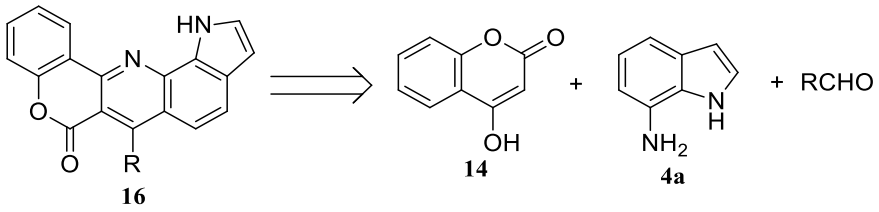
Scheme 4.4: Synthesis of 6π-electrocyclization *en-route* to formation of fused coumarin-dihydroquinoline systems.

Given the structural complexity of the target molecule, most of the above mentioned synthetic approaches looked inappropriate. However, taking cue from the strategies adopted Khan *et al*.,[12] we decided to use a multicomponent approach involving use of 7-aminoindole (**4a**), aromatic aldehydes and 4-hydroxy coumarin (**14**) to generate the target molecule. On

completion of the synthesis, anticancer activity evaluation of the compounds was planned, as nitrogen bearing planar aromatic systems display DNA intercalation behavior an important feature of anticancer drug molecules.[14-18]

4.2. Results and discussion

Prior to execution of the synthetic strategy, retrosynthetic analysis of the target molecule was carried out **(Scheme 4.5)**, revealing as precursors 4-hydroxy coumarin **(14)**, 7-amino indole **(4a)** and various aldehydes to form the desired chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one scaffold (**16**).

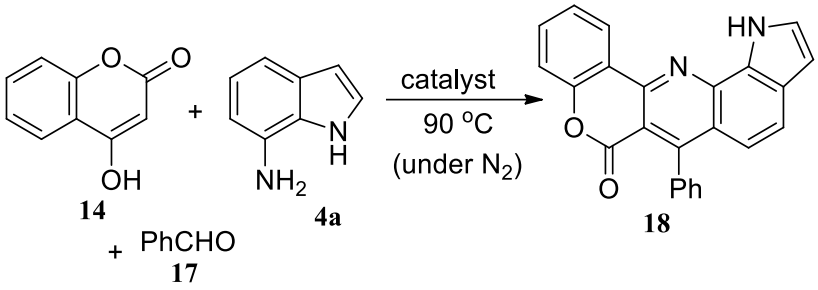


Scheme 4.5: Retrosynthetic analysis of 6-substituted chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one.

4-Hydroxy coumarin **(14)**, 7-amino indole **(4a)** and benzaldehyde **(17)** were chosen as the starting materials for initiating the synthetic studies. In our attempts to synthesize the target molecule, $CuCl_2$ was used as catalyst and 1,4-dioxane as the solvent at 90 °C (**entry 1; Table 4.1**). This choice of reaction conditions was based on a literature example, where the aforementioned conditions were used for the quinoline synthesis, a crucial component of the target molecule.[19] Our initial attempt (**entry 1**) was fruitful in generating the target molecule, although in low yield. Thus, it was necessary to screen other reagents (**entry 2-11**). Some of the reagents (**entry 5, 7, 9 and 10**) didn't generate the desired compound and the reactions resulted in complex mixtures, which were difficult to separate. Such outcome is understandable, as highly reactive N-1 hydrogen of indole can undergo several side reactions easily under the given conditions. The best result was obtained when the reaction was carried out using $FeCl_3$ (30 mol%) in 1,4-dioxane at 90 °C. Reactions attempted (**entry 12-17**) with

different amounts of catalyst or by changing the temperature of the reaction or by changing the solvents, did not improve the yield. Based on the screening results, reaction with $FeCl_3$ (30 mol%) in 1,4-dioxane at 90 °C was chosen as the optimal reaction condition and was used for further synthetic studies.

Table 4.1: Optimization of the reaction conditions*

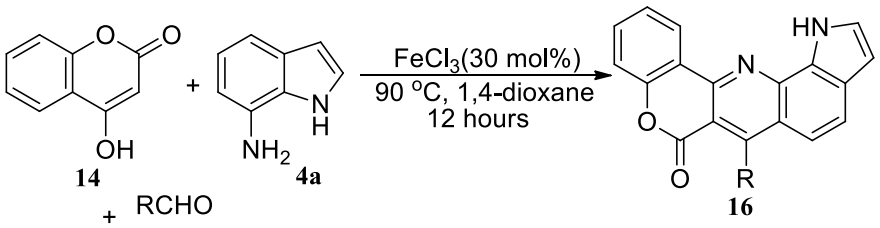


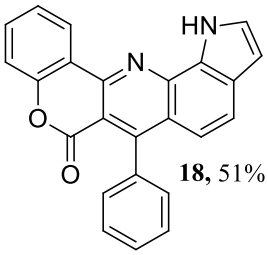
S.NO	Catalyst	Solvent	Yield(%) **(18)**
1	$CuCl_2.2H_2O$	1,4-dioxane	5
2	CuI	1,4-dioxane	10
3	CuBr	1,4-dioxane	8
4	$Cu(OTFA)_2$	1,4-dioxane	5
5	$AlCl_3$	1,4-dioxane	0
6	$FeCl_3$ (30%)	1,4-dioxane	51
7	$Fe(NO_3)_2.9H_2O$	1,4-dioxane	0
8	$ZnCl_2$	1,4-dioxane	5
9	PTSA	1,4-dioxane	0
10	$BF_3.OEt_2$	1,4-dioxane	0
11	$Yb(OTf)_3$	1,4-dioxane	6
12	$FeCl_3$ (10%)	1,4-dioxane	20
13	$FeCl_3$ (20%)	1,4-dioxane	38
14	$FeCl_3$ (40%)	1,4-dioxane	45
15	$FeCl_3$(30%)/110°C	1,4-dioxane	51
16	$FeCl_3$ (30%)	Nitrobenzene	0
17	$FeCl_3$ (30%)	DMF	18

*All the reaction were carried out at 90 °C for 12 hours

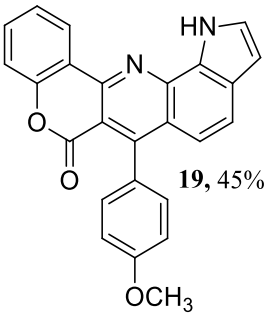
After optimization, reactions were carried out between 4-hydroxycoumarin, 7-amino indole and diverse aldehydes. Most of the reactions involving aromatic aldehydes were successful and gave moderate yields 25-45% **(Table 4.2)**. However, reactions involving aliphatic aldehydes did not result in the formation of the expected products because of aliphatic aldehyde unstable and low boiling properties at higher temperature. All the final products bearing various substituents on the benzene ring gave lower yields compared to their completely unsubstituted counterparts. Aromatic aldehydes with electron releasing substituents **(19-21)** gave better yields compared to aldehydes with electron withdrawing substituents **(22-23)**. On screening mono-halogenated benzaldehydes **(24-31)**, comparatively low yields were obtained for *p*-fluoro and *o*- bromosubstituted analogues. Reactions were also carried out with di-substituted benzaldehydes **(29-31)**, which resulted in yields between 26-41%. Interestingly, sterically hindered 2,6-dichlorobenzaldehyde **(31)**, gave the highest yield (41%) among the three, which clearly shows that steric effects do not play a significant role in determining the course of the reaction. Further attempts to increase the scope of the reaction were carried out with naphthalene-1-carboxaldehyde, naphthalene-2-carboxaldehyde, pyridine-2-carboxaldehyde, 2-chloro-pyridine-3-carboxaldehyde and thiophene-2-carboxaldehyde. All the reactions resulted in the formation of the final products **(32-36)** in 27-37% yields. While the overall yield of the products formed was modest at best, the complexity of the starting compounds, along with that of the final products and use of a minimum number of steps to carry out the transformations, makes the overall process useful.

Table 4.2: Structure and yield of the compounds formed





18, 51%



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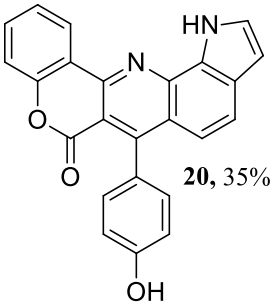

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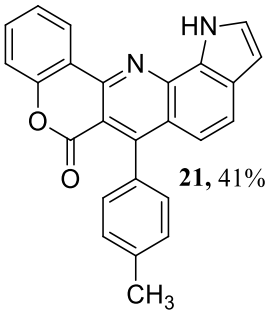

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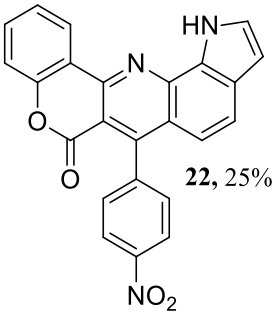

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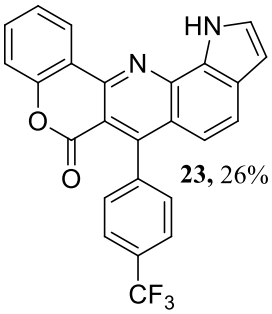

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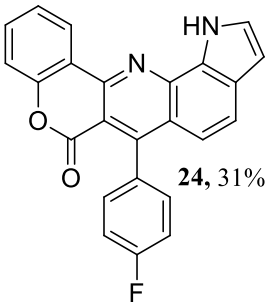

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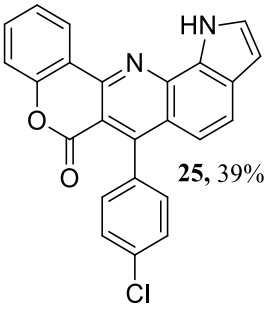

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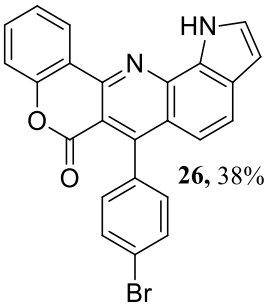

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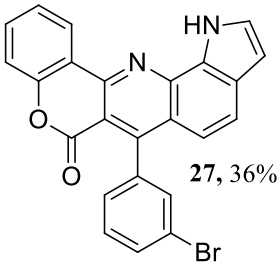

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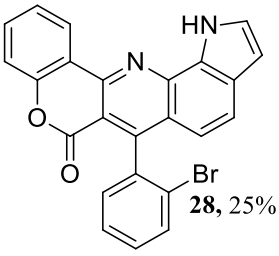

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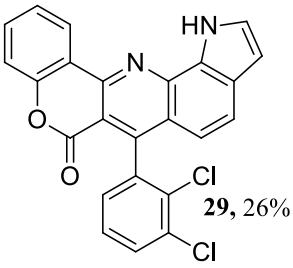

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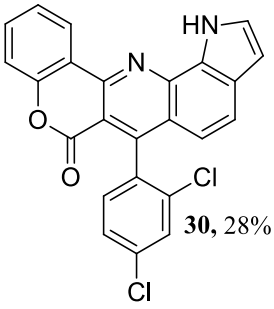

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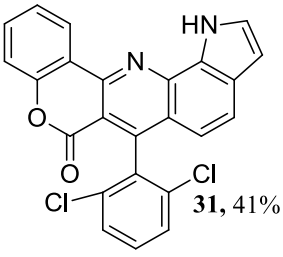

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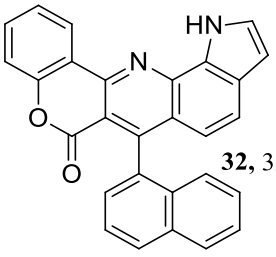
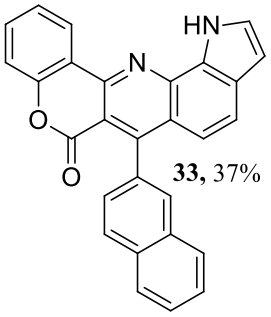
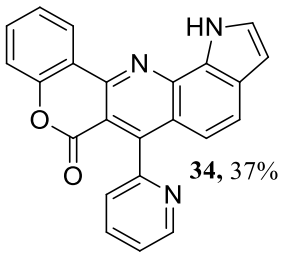
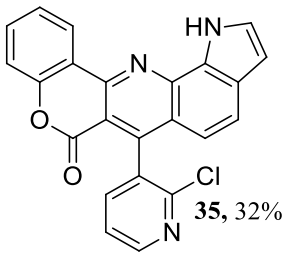
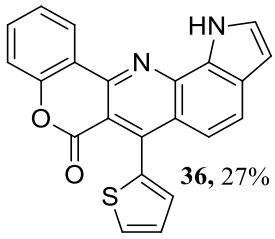


32, 32%

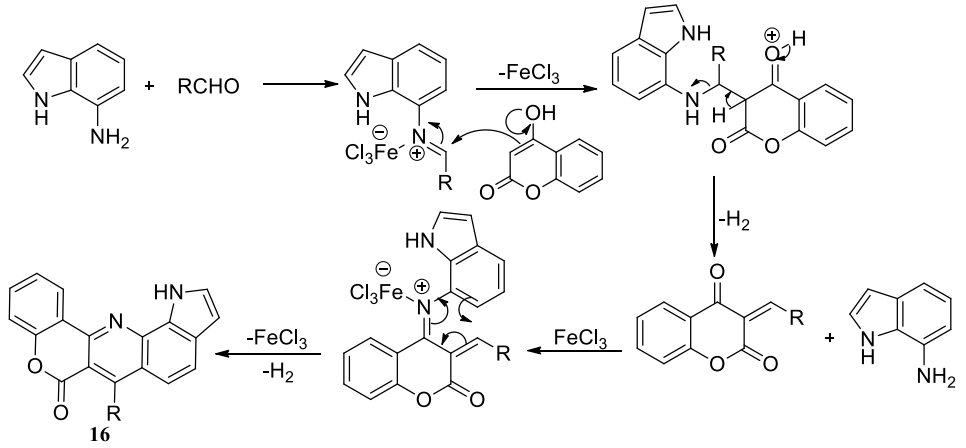






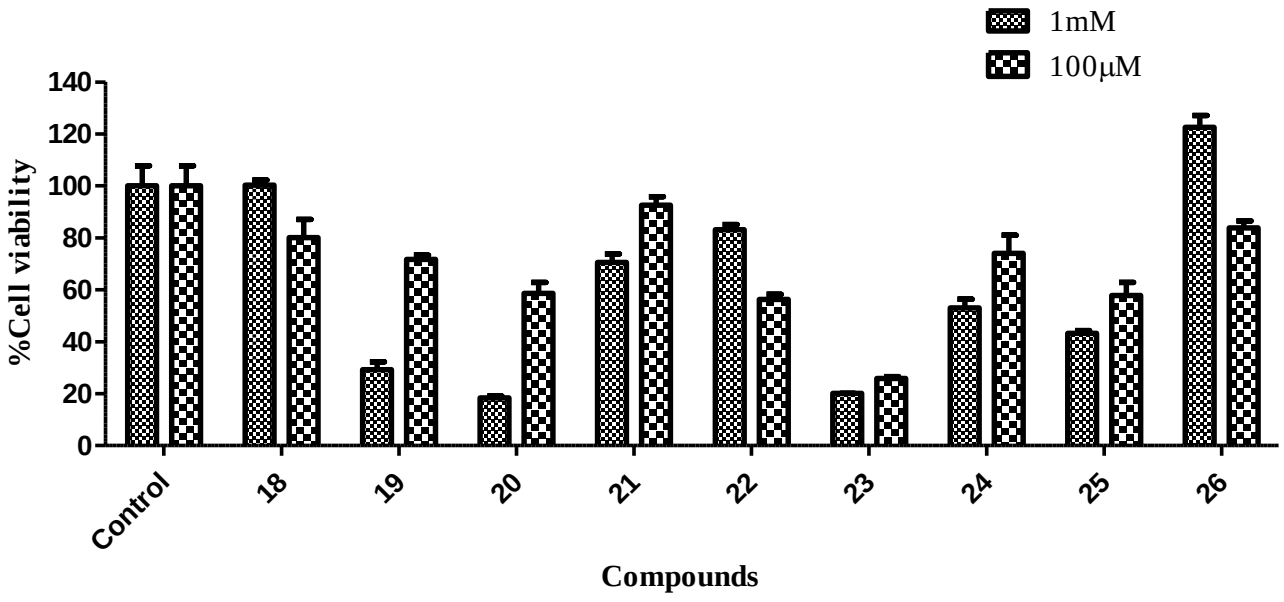


A plausible mechanism has been proposed based on the outcome of the reaction. The reaction is triggered by the imine formation between 7-amino indole and aldehyde, followed by attachment of $FeCl_3$ to the imine nitrogen **(Scheme 4.6)**. Subsequently, iminium ion undergoes reaction with 4-hydroxy coumarin, which eventually generates substituted 3-ethylidenechroman-2,4-dione by expulsion of 7-aminoindole. In the final step, imine formation between chroman-2,4-dione and 7- amino indole followed by attachment of $FeCl_3$ ushers 6π-electrocyclization, which eventually results in the formation of the target molecule.

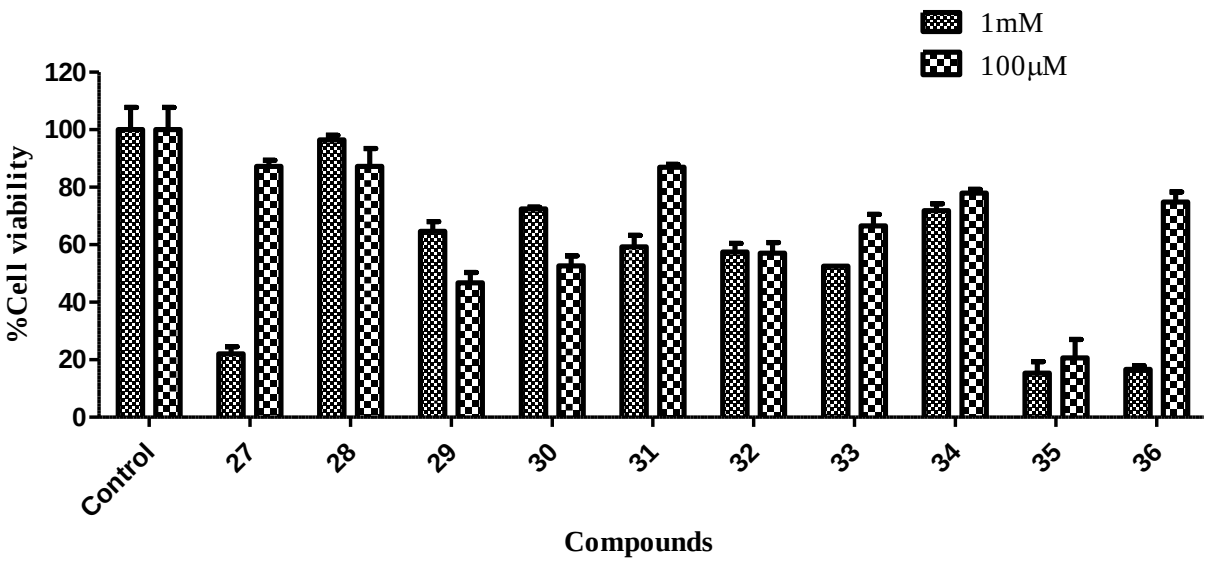


Scheme 4.6: Plausible mechanism for the formation of fused chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds

The molecules were then tested for anticancer activity using the MTT assay against murine melanoma cell line (B16F10) **(Figure 4.2)**. Among the nineteen compounds, the six most active compounds (**19, 20, 23, 27, 35** and **36**) were explored further with longer range of concentration to find out their IC_{50} value (**Figure 4.3**). MTT assay for doxorubicin was performed to validate the assay. While chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds showed poor anticancer activity, compound **35**, its 2-chloropyridine analogue, showed modest activity with IC_{50} of 70.74 μM. This molecule can be further modified with various substituents to enhance its potency.



[A]



[B]

Figure 4.2 [A and B]: Anticancer activity of chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7-one analogues against B16F10 cells. Cell viability was measured by *in vitro* MTT assay.[1,2] Cells were exposed to compounds for 24 hours at two concentrations 1mM and 100 µM (n = 3). Data represent mean values of measurements ± s.d. of two experiments.

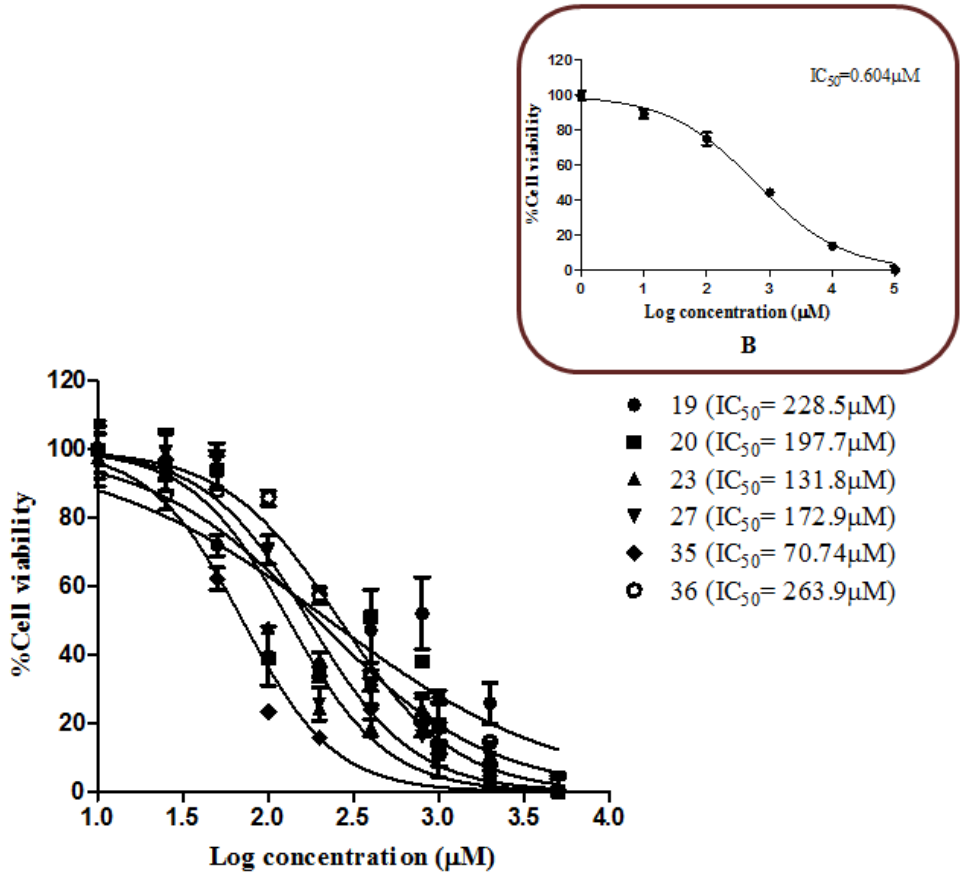


Figure 4.3:(A) Dose response curve of most active six compounds (**19, 20, 23, 27, 35** and **36**) and their IC_{50} values. All six compounds were tested in ten different concentrations and cell viability was measured by MTT assay. Data were analyzed and plotted in dose-response format representing mean values of measurements ± s.d. (n = 2). IC_{50} was calculated using non-linear regression analysis method. **(B)** MTT assay for Doxorubicin was performed to validate the assay procedure. B16F10 cells were treated with Doxorubicin within 0.001 µM to 100 µM concentration range. After 24 hour of treatment cells were treated with MTT reagent and cell viability was measured. IC_{50} value for Doxorubicin was found to be 0.604 µM which corresponds to reported literature.[20]

4.3. Conclusion

In conclusion, we have developed a simple route for the synthesis of 6-substituted chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds from easily available starting molecules. The final compounds brought together the biologically important skeletons coumarin and indole in a fused format. The transformation requires use of an inexpensive and readily available catalyst, $FeCl_3$, at 90 °C in 1,4-dioxane as a solvent. The synthesized compounds, when screened for anticancer activity, showed IC_{50} value of 70.74 µM for the molecule bearing the 2-chloro-3-pyridyl substituent. Given the global impact of cancer, search for newer entities displaying anticancer activity is essential. It is this need that we have attempted to address in this work.

4.4. Experimental

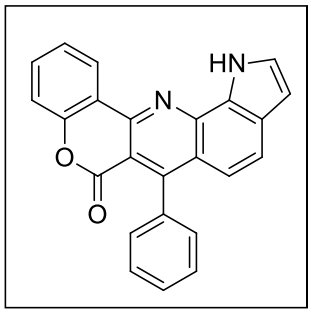
Compounds and reagents required for this work were purchased from commercial sources and used without further purification. All the solvents used were dried and distilled using standard procedures, prior to use. NMR spectra [^1H (400 MHz) and ^{13}C (101 MHz)] were recorded in $CDCl_3$ and DMSO using $(CH_3)_4Si$ as internal standard. KBr plates were used to

record IR spectra on Jasco FT/IR-4200 instrument. Melting points were recorded on a Stuart SMP 30 melting point apparatus and are uncorrected. Mass spectra were recorded on Shimadzu LCMS-2020.

4.5. General procedure for the preparation of fused chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one:

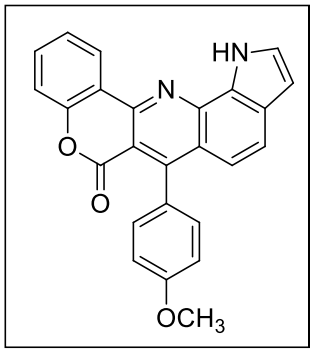
A mixture of the requisite aldehyde (0.8 mmol), 7-aminoindole (1.0 mmol) and 4-hydroxycoumarin (0.8 mmol) and $FeCl_3$ (30% mol) in 1,4-dioxane (dry) were taken in a round bottomed flask fitted with condenser, under N_2 atmosphere. The reaction mixture was heated at 90 ^oC for 12 hours and the progress of the reaction was monitored by TLC. On completion of the reaction, the mixture was cooled to room temperature. It was subsequently poured into water and extracted with ethyl acetate. The organic layer was separated, washed with brine (2 mL) and dried over Na_2SO_4 (1 g). The dried organic layer was concentrated in vacuo and the resulting residue was purified by column chromatography using hexane/ethyl acetate as eluent, to give the corresponding product.

6-Phenylchromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (18). Yield: 51%; yellow solid; **M.p**. 278-282 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate = 8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 3406,



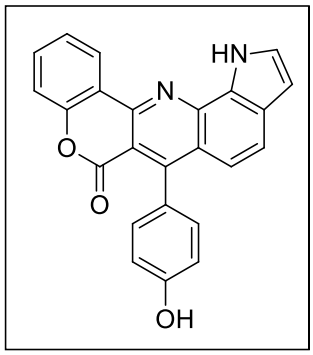
3071, 2923, 1686, 1600, 1583, 1534, 1454, 1424, 1325, 1292, 1185, 1126; **^{1}H NMR** (400 MHz, DMSO): δ 6.78 – 6.74 (m, 1H), 6.91 (d, *J* = 9.0 Hz, 1H), 7.33 (dd, *J* = 7.6, 1.6 Hz, 2H), 7.44 (d, *J* = 7.6 Hz, 1H), 7.57 – 7.49 (m, 4H), 7.72 – 7.66 (m, 1H), 7.76 (dd, *J* = 7.3, 4.9 Hz, 2H), 9.10 (dd, *J* = 7.9, 1.6 Hz, 1H), 12.78 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.5, 111.1, 117.0, 118.4, 120.3, 123.8, 123.3, 124.8, 126.0, 128.0, 128.2, 128.4, 128.5, 129.1, 130.3, 132.7, 138.4, 140.4, 148.7, 152.7, 154.2, 159.4; **LRMS-ESI** (m/z): 363.10 $[M + H]^+$.

6-(4-Methoxyphenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (19). Yield: 45%; yellow solid; **M.p**. 190-193 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



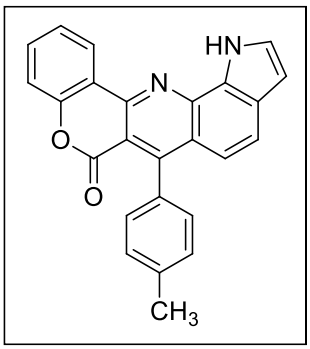
3445, 2922, 2853, 1734, 1508, 1242,1104; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 3.96 (s, 3H), 6.79 (s, 1H), 7.12 (d, *J* = 8.3 Hz, 2H), 7.23 (d, *J* = 9.0 Hz, 1H), 7.28 (d, *J* = 1.4 Hz, 2H), 7.36 (d, *J* = 8.1 Hz, 1H), 7.48 – 7.41 (m, 1H), 7.57 (dd, *J* = 13.5, 5.1 Hz, 2H), 7.72 (d, *J* = 8.9 Hz, 1H), 8.92 (d, *J* = 7.3 Hz, 1H), 9.94 (s, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 55.3, 104.9, 113.7, 116.9, 119.6, 120.1, 122.3, 124.2, 124.8, 125.2, 125.7, 128.8, 129.4, 130.00, 130.3, 131.8, 140.3, 149.0, 152.7, 154.9, 155.3, 159.3, 159.9; **LRMS-ESI** (m/z): 393.15 $[M + H]^+$.

6-(4-Hydroxyphenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (20). Yield: 35%; yellow solid; **M.p**. 235-240 ^{o}C; $\mathbf{R_f}$ = 0.4 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



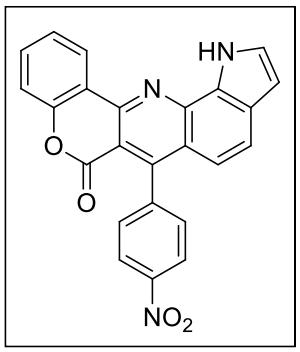
3587,3416, 2919, 2852, 1722, 1597, 1532, 1476, 1400, 1379, 1294, 1223, 1117; **^{1}H NMR** (400 MHz, DMSO): δ 6.76 (d, *J* = 1.7 Hz, 1H), 6.93 (dd, *J* = 6.5, 4.1 Hz, 2H), 7.07 (dd, *J* = 9.0, 4.5 Hz, 1H), 7.13 (dd, *J* = 8.3, 4.3 Hz, 2H), 7.44 (d, *J* = 8.2 Hz, 1H), 7.53 (t, *J* = 9.2 Hz, 1H), 7.68 (t, *J* = 9.5 Hz, 1H), 7.80 – 7.72 (m, 2H), 9.09 (d, *J* = 5.6 Hz, 1H), 9.66 (s, 1H), 12.73 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.4, 107.7, 115.2, 116.9, 120.4, 123.0, 124.7, 126.1, 126.2, 128.0, 128.1, 128.5, 129.9, 131.2, 131.2, 132.6, 132.9, 140.4, 144.9, 148.7, 152.7; **LRMS-ESI** (m/z): 423.05 $[M + H]^+$.

6-(p-Tolyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (21). Yield: 41%; yellow solid; **M.p**. 118-120 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 3445, 2922, 2853, 1734, 1508, 1242; **^{1}H NMR** (400 MHz, DMSO): δ 2.46 (s, 3H), 6.77 – 6.72 (m, 1H), 6.95 (d, *J* = 9.0 Hz, 1H), 7.22 (d, *J* = 8.0 Hz, 2H), 7.35 (d, *J* = 7.8 Hz, 2H), 7.44 (d, *J* = 8.1 Hz, 1H), 7.53 (t, *J* = 7.6 Hz, 1H), 7.71 – 7.65 (m, 1H), 7.75 (dd, *J* = 6.0, 3.2 Hz, 2H), 9.10 (d,



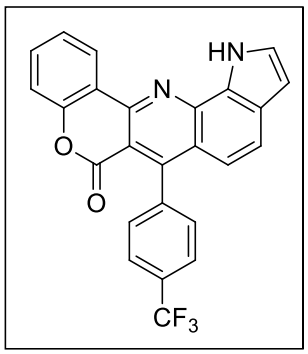
J = 6.3 Hz, 1H), 12.76 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 21.5, 104.5, 111.2, 116.9, 118.5, 120.4, 123.2, 124.0, 124.8, 126.1, 128.2, 128.5, 128.9, 129.1, 130.4, 132.6, 135.4, 137.1, 140.4, 148.7, 152.7, 154.5, 159.3; **LRMS-ESI** (m/z): 477.10 [M + H]$^+$.

6-(4-Nitrophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (22). Yield: 25%; yellow solid; **M.p**. 178-183 ^{o}C; **R_f** = 0.5 [hexane / ethyl acetate = 7:3]; **ν_{max}(KBr)/cm^{-1}**:



3302, 2923, 2852, 1715, 1593, 1537, 1513, 1386, 1339, 1248, 1204, 1118; **^{1}H NMR** (400 MHz, DMSO): δ 6.80 – 6.77 (m, 1H), 6.82 (d, *J* = 8.9 Hz, 1H), 7.47 (d, *J* = 8.2 Hz, 1H), 7.57 (t, *J* = 8.1 Hz, 1H), 7.67 (d, *J* = 8.8 Hz, 2H), 7.72 (d, *J* = 8.2 Hz, 1H), 7.79 (dd, *J* = 7.4, 4.9 Hz, 2H), 8.42 (s, 1H), 8.44 (s, 1H), 9.11 (d, *J* = 7.9 Hz, 1H), 12.84 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.7, 111.0, 117.1, 117.8, 120.2, 123.0, 123.7, 123.8, 125.0, 126.0, 128.5, 129.2, 130.1, 130.3, 132.8, 140.5, 145.9, 147.5, 148.6, 151.5, 152.6, 159.8; **LRMS-ESI** (m/z): 408.05 [M + H]$^+$.

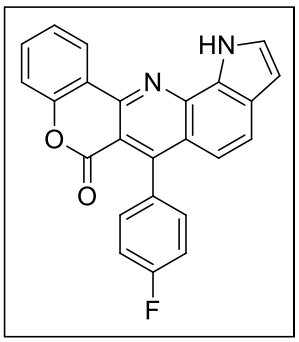
6-(4-(Trifluoromethyl)phenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (23). Yield: 26%; yellow solid; **M.p**. 228-232 ^{o}C; **R_f** = 0.6 [hexane / ethyl acetate =8:2];



ν_{max}(KBr)/cm^{-1}: 3460, 3430, 2924, 2853, 1723, 1612, 1560, 1534, 1460, 1403, 1380, 1322, 1241, 1163, 1112; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 6.83 – 6.80 (m, 1H), 7.02 (d, *J* = 9.0 Hz, 1H), 7.39 (dd, *J* = 8.2, 0.8 Hz, 1H), 7.50 – 7.45 (m, 3H), 7.59 (t, *J* = 2.7 Hz, 1H), 7.62 (ddd, *J* = 8.4, 7.4, 1.7 Hz, 1H), 7.74 (d, *J* = 9.0 Hz, 1H), 7.85 (d, *J* = 8.0 Hz, 2H), 8.94 (dd, *J* = 7.9, 1.5 Hz, 1H), 9.95 (s, 1H); **^{13}C NMR** (101 MHz, CDCl3): δ 29.7, 105.2, 111.1, 117.1, 118.9, 119.9, 122.9, 123.9, 124.5, 125.2, 126.0, 128.4,

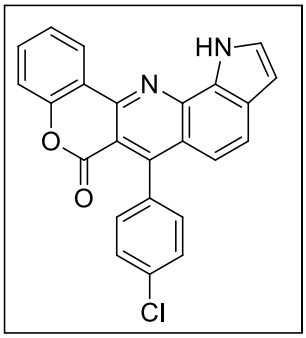
129.0, 130.2, 130.2, 132.1, 140.4, 149.0, 152.6, 153.0, 159.9; **LRMS-ESI** (m/z): 430.20 [M + H]$^+$.

6-(4-Fluorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (24). Yield: 31%; yellow solid; **M.p**. 260-264 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



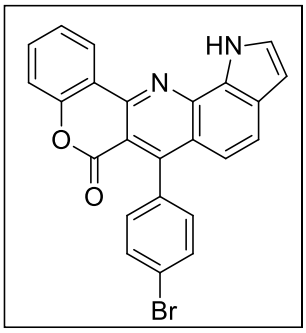
3416, 2919, 2852, 1722, 1597, 1532, 1476, 1400, 1379, 1294, 1223, 1117; **^{1}H NMR** (400 MHz, DMSO): δ 6.77 (dd, *J* = 2.7, 2.0 Hz, 1H), 6.93 (d, *J* = 8.9 Hz, 1H), 7.39 (d, *J* = 7.7 Hz, 4H), 7.44 (d, *J* = 8.2 Hz, 1H), 7.54 (t, *J* = 7.6 Hz, 1H), 7.72 – 7.67 (m, 1H), 7.81 – 7.74 (m, 2H), 9.10 (d, *J* = 7.9 Hz, 1H), 12.79 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.5, 111.4, 115.3, 115.5, 117.0, 118.2, 120.3, 123.4, 123.9, 124.9, 126.0, 128.3, 129.1, 130.3, 130.6, 130.7, 132.7, 134.5, 134.6, 140.4, 148.7, 152.7, 153.2, 159.5, 161.0; **LRMS-ESI** (m/z): 481.10 [M + H]$^+$.

6-(4-Chlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (25). Yield: 39%; yellow solid; **M.p**. 257-260 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



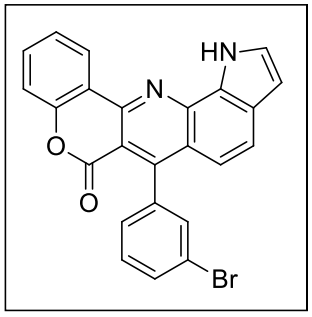
3416, 2919, 2852, 1722, 1597, 1532, 1476, 1400, 1379, 1294, 1223, 1119; **^{1}H NMR** (400 MHz, DMSO): δ 6.77 (d, *J* = 2.5 Hz, 1H), 6.90 (d, *J* = 8.9 Hz, 1H), 7.38 (d, *J* = 8.4 Hz, 2H), 7.44 (d, *J* = 8.2 Hz, 1H), 7.54 (t, *J* = 8.0 Hz, 1H), 7.61 (d, *J* = 8.4 Hz, 2H), 7.68 (t, *J* = 8.5 Hz, 1H), 7.77 (t, *J* = 6.5 Hz, 2H), 9.09 (d, *J* = 9.4 Hz, 1H), 12.79 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 117.0, 118.1, 120.3, 123.5, 123.6, 124.9, 126.0, 128.3, 128.5, 129.2, 130.3, 130.5, 132.7, 132.9, 137.3, 140.4, 148.6, 152.6, 152.7, 159.5; **LRMS-ESI** (m/z): 397.10 [M + H]$^+$.

6-(4-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (26). Yield: 38%; yellow solid; **M.p**. 265-270 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



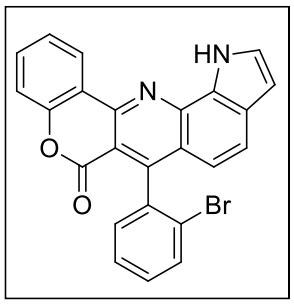
3417, 2922, 2853, 1721, 1596, 1532, 1475, 1400, 1114; **^{1}H NMR** (400 MHz, $CDCl_3$): δ 6.79 (dd, *J* = 2.8, 2.3 Hz, 1H), 7.09 (d, *J* = 9.0 Hz, 1H), 7.22 – 7.18 (m, 2H), 7.37 (dd, *J* = 8.2, 0.8 Hz, 1H), 7.46 – 7.41 (m, 1H), 7.62 – 7.54 (m, 2H), 7.69 – 7.67 (m, 1H), 7.71 (t, *J* = 5.3 Hz, 2H), 8.91 (dd, *J* = 7.9, 1.6 Hz, 1H), 9.90 (s, 1H); **^{13}C NMR** (101 MHz, $CDCl_3$): δ 105.1, 117.0, 119.1, 119.9, 122.1, 122.8, 124.1, 124.4, 125.2, 125.9, 128.9, 129.71, 130.2, 131.4, 132.0, 136.9, 140.3, 149.0, 152.6; **LRMS-ESI** (m/z): 441.05 $[M + H]^+$.

6-(3-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (27). Yield: 36%; yellow solid; **M.p**. 297-303 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; **ν_{max}(KBr)/cm^{-1}**:



3459, 2922, 1725, 1614, 1595, 1560, 1533, 1518, 1470, 1397, 1380, 1364, 1292, 1242, 1219, 1198, 1169, 1116; **^{1}H NMR** (400 MHz, DMSO): δ 6.78 – 6.76 (m, 1H), 6.90 (d, *J* = 8.9 Hz, 1H), 7.37 (d, *J* = 7.7 Hz, 1H), 7.45 (d, *J* = 7.6 Hz, 1H), 7.51 (d, *J* = 7.9 Hz, 1H), 7.54 (t, *J* = 7.6 Hz, 1H), 7.58 (s, 1H), 7.68 (d, *J* = 8.8 Hz, 1H), 7.72 (d, *J* = 9.2 Hz, 1H), 7.76 (t, *J* = 2.7 Hz, 1H), 7.80 (d, *J* = 9.0 Hz, 1H), 9.09 (d, *J* = 7.9 Hz, 1H), 12.80 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.6, 111.1, 117.0, 118.1, 120.3, 121.7, 123.5, 123.6, 124.9, 126.0, 127.7, 128.3, 129.2, 130.3, 130.6, 130.9, 131.0, 132.7, 140.5, 140.8, 148.6, 152.0, 152.6, 159.5;**LRMS-ESI** (m/z): 441.05 $[M + H]^+$.

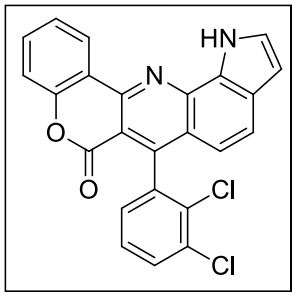
6-(2-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (28). Yield: 25%;



yellow solid; **M.p**. 140-145 ^{o}C; $\mathbf{R_f}$ = 0.5 [Hexane / Ethyl acetate =8:2]; **ν_{max}(KBr)/cm^{-1}**: 3447, 2923, 2848, 1716, 1609, 1456, 1387, 1217; **^{1}H NMR** (400 MHz, DMSO): δ 6.79 – 6.77 (m, 1H), 6.80 (d, *J* = 8.9 Hz, 1H), 7.37 (d, *J* = 7.5 Hz, 1H), 7.48 (t, *J* = 7.7 Hz, 2H),

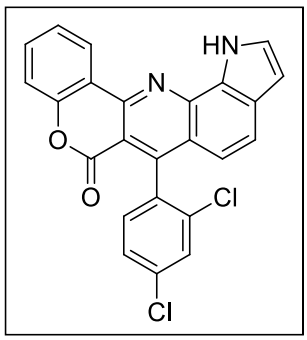
7.57 – 7.54 (m, 1H), 7.60 – 7.57 (m, 1H), 7.74 – 7.68 (m, 1H), 7.80 (dt, *J* = 5.5, 4.9 Hz, 3H), 9.11 (d, *J* = 7.9 Hz, 1H), 12.85 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.7, 111.0, 117.1, 117.7, 120.1, 121.9, 123.1, 123.9, 125.1, 125.9, 128.0, 128.4, 129.4, 130.1, 130.2, 130.3, 132.5, 132.9, 139.2, 140.8, 148.6, 152.3, 152.5, 159.2; **LRMS-ESI** (m/z): 441.05 $[M + H]^+$.

6-(2,3-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (29). Yield:



26%; yellow solid; **M.p**. 267-270 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\boldsymbol{\nu_{max}}$**(KBr)/cm^{-1}**: 3473, 2912, 2852, 1720, 1597, 1564, 1538, 1470, 1405, 1387, 1249, 1212, 1190, 1134; **^{1}H NMR** (400 MHz, DMSO): δ 6.81 – 6.78 (m, 1H), 6.85 (d, *J* = 8.9 Hz, 1H), 7.43 (d, *J* = 8.3 Hz, 1H), 7.48 (d, *J* = 8.2 Hz, 1H), 7.58 (t, *J* = 7.6 Hz, 1H), 7.64 (dd, *J* = 8.2, 2.1 Hz, 1H), 7.75 – 7.70 (m, 1H), 7.79 (t, *J* = 2.7 Hz, 1H), 7.84 (d, *J* = 8.9 Hz, 1H), 7.88 (d, *J* = 2.1 Hz, 1H), 9.11 (d, *J* = 6.3 Hz, 1H), 12.87 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.8, 111.2, 117.2, 117.4, 120.1, 123.0, 124.2, 125.1, 125.9, 127.9, 128.6, 129.1, 129.5, 130.3, 131.6, 132.8, 132.9, 133.9, 136.3, 140.8, 148.6, 149.5, 152.5, 159.4; **LRMS-ESI** (m/z): 431.05 $[M + H]^+$.

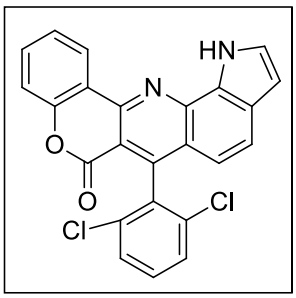
6-(2,4-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (30). Yield:



28%; yellow solid; **M.p**. 273-275 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\boldsymbol{\nu_{max}}$**(KBr)/cm^{-1}**: 3475, 2922, 2852, 1721, 1592, 1562, 1536, 1470, 1402, 1381, 1244, 1216, 1198, 1124; **^{1}H NMR** (400 MHz, DMSO): δ 6.79 (s, 1H), 6.84 (d, *J* = 9.0 Hz, 1H), 7.42 (d, *J* = 8.2 Hz, 1H), 7.47 (d, *J* = 8.2 Hz, 1H), 7.56 (t, *J* = 7.5 Hz, 1H),), 7.63 (d, *J* = 8.2 Hz, 1H), 7.71 (t, *J* = 7.7 Hz, 1H), 7.79 (s, 1H), 7.82 (d, *J* = 8.9 Hz, 1H), 7.87 (s, 1H), 9.10 (d, *J* = 7.8 Hz, 1H), 12.87 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.8, 111.2, 117.1, 117.4, 120.1, 123.0, 124.2, 125.1, 125.9, 127.9, 128.5, 129.1, 129.4,

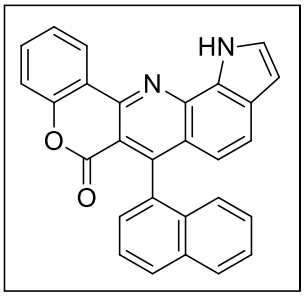
130.3, 131.6, 132.8, 132.9, 133.9, 136.3, 140.7, 148.6, 149.5, 152.5, 159.4; **LRMS-ESI** (m/z): 431.05 $[M + H]^+$.

6-(2,6-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (31). Yield: 41%; light yellow solid; **M.p**. 265-268 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2];



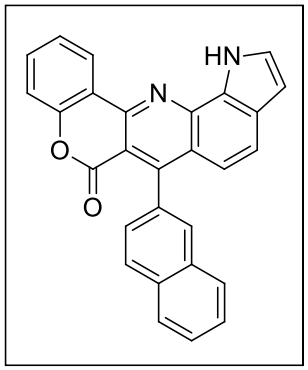
ν_{max}(KBr)/cm^{-1}: 3470, 2925, 2850, 1723, 1586, 1556, 1530, 1476, 1411, 1371, 1267, 1206, 1188, 1126; **^{1}H NMR** (400 MHz, DMSO): δ 6.82 (dd, *J* = 9.1, 5.6 Hz, 2H), 7.50 (d, *J* = 8.1 Hz, 1H), 7.61 (dt, *J* = 14.9, 7.5 Hz, 2H), 7.73 (dd, *J* = 10.9, 7.7 Hz, 3H), 7.82 (t, *J* = 2.6 Hz, 1H), 7.87 (d, *J* = 8.9 Hz, 1H), 9.12 (d, *J* = 7.9 Hz, 1H), 12.93 (s, 1H) ; **^{13}C NMR** (101 MHz, DMSO): δ 104.9, 110.8, 116.5, 117.3, 119.9, 122.3, 124.8, 125.3, 125.9, 128.7, 129.7, 130.3, 131.2, 132.9, 133.1, 135.9, 141.1, 148.1, 152.4, 148.8, 159.2; **LRMS-ESI** (m/z): 431.05 $[M + H]^+$.

6-(Naphthalen-1-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (32). Yield: 32%; yellow solid; **M.p**. 325-330 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; **ν_{max}(KBr)/cm^{-1}**:



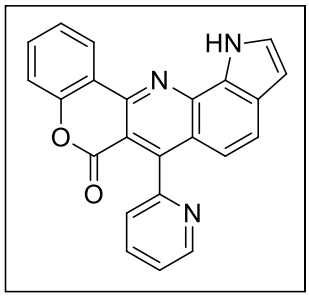
3463, 2921, 2853, 1726, 1594, 1560, 1533, 1458, 1391, 1359, 1240, 1198, 1114; **^{1}H NMR** (400 MHz, DMSO): δ 6.65 (d, *J* = 8.9 Hz, 1H), 6.75 – 6.73 (m, 1H), 7.17 (d, *J* = 8.2 Hz, 1H), 7.27 (t, *J* = 7.6 Hz, 1H), 7.42 (dd, *J* = 10.2, 6.8 Hz, 2H), 7.52 (t, *J* = 7.0 Hz, 1H), 7.57 (t, *J* = 7.6 Hz, 1H), 7.67 (dd, *J* = 17.2, 8.2 Hz, 3H), 7.77 (t, *J* = 2.7 Hz, 1H), 8.08 (t, *J* = 8.1 Hz, 2H), 9.17 (d, *J* = 7.9 Hz, 1H), 12.85 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.6, 112.3, 117.0, 118.4, 120.4, 123.4, 124.4, 124.9, 125.8, 125.9, 126.1, 126.4, 126.8, 128.3, 128.7, 129.2, 130.4, 131.7, 132.7, 133.3, 136.3, 140.6, 148.9, 152.6, 152.7, 159.1; **LRMS-ESI** (m/z): 413.10 $[M + H]^+$.

6-(Naphthalen-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (33). Yield: 37%; yellow solid; **M.p**. 240-245 ^{o}C; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



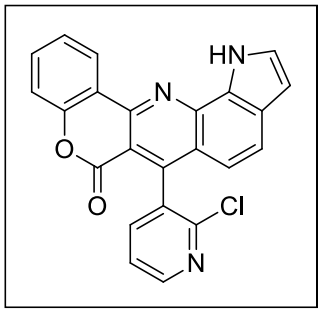
3433, 2923, 2852, 1734, 1536, 1457, 1379, 1260, 1109; **^{1}H NMR** (400 MHz, DMSO): δ 6.70 (dd, *J* = 2.8, 2.0 Hz, 1H), 7.53 (dd, *J* = 8.4, 5.7 Hz, 2H), 7.57-7.63 (m, 2H), 7.79 (d, *J* = 8.5 Hz, 1H), 8.01 (dd, *J* = 6.6, 2.8 Hz, 1H), 8.10 (dd, *J* = 9.2, 6.0 Hz, 2H), 8.27 (d, *J* = 8.6 Hz, 1H), 8.48 (d, *J* = 8.6 Hz, 1H), 8.76 (dd, *J* = 8.6, 1.8 Hz, 1H), 8.97 (s, 1H), 12.34 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 103.7, 116.6, 118.9, 122.0, 123.6, 125.3, 125.4, 126.8, 126.9, 127.2, 128.1, 128.6, 129.1, 131.1, 133.6, 133.9, 136.8, 137.5, 138.3, 153.9; **LRMS-ESI** (m/z): 413.10 $[M + H]^{+}$.

6-(Pyridin-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (34):Yield: 37%; yellow solid; **M.p**. 297-300 ^{o}C; $\mathbf{R_f}$ = 0.5 [hexane / ethyl acetate =7:3]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$:



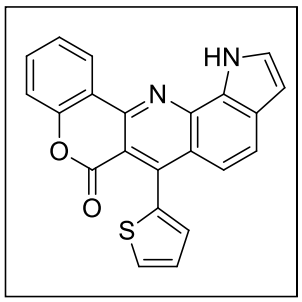
3445, 2922, 2853, 1736, 1561, 1523, 1461, 1397, 1113; **^{1}H NMR** (400 MHz, DMSO): δ 6.79 (t, *J* = 6.1 Hz, 2H), 7.46 (d, *J* = 8.2 Hz, 1H), 7.55 (dt, *J* = 12.2, 7.8 Hz, 3H), 7.73 – 7.68 (m, 1H), 7.78 (dd, *J* = 8.1, 5.7 Hz, 2H), 7.98 (td, *J* = 7.7, 1.7 Hz, 1H), 8.76 – 8.73 (m, 1H), 9.10 (dd, *J* = 7.9, 1.5 Hz, 1H), 12.83 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.7, 110.9, 117.1, 117.9, 120.2, 123.2, 123.3, 123.6, 124.3, 124.9, 125.9, 128.3, 129.2, 130.2, 132.8, 136.6, 140.8, 148.6, 149.3, 151.9, 152.6, 157.1, 159.5; **LRMS-ESI** (m/z): 364.10 $[M + H]^{+}$.

6-(2-Chloropyridin-3-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (35). Yield: 32%; light yellow solid; **M.p**. 290-295 ^{o}C; $\mathbf{R_f}$ = 0.5 [hexane / ethyl acetate =7:3]; $\mathbf{\nu_{max}(KBr)/cm^{-1}}$: 3445, 2922, 2853, 1736, 1561, 1523, 1461, 1397, 1113; **^{1}H NMR** (400 MHz, DMSO): δ 6.88 – 6.79 (m, 2H), 7.49 (d, *J* = 8.1 Hz, 1H), 7.58 (t, *J* = 7.3 Hz, 1H), 7.66 (dd, *J* = 7.4, 4.9 Hz, 1H), 7.73 (t, *J* = 7.7 Hz, 1H), 7.80 (s, 1H), 7.86 (d, *J* = 8.9 Hz, 1H), 7.90



(d, *J* = 7.5 Hz, 1H), 8.61 (d, *J* = 6.5 Hz, 1H), 9.11 (d, *J* = 6.8 Hz, 1H), 12.89 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.8, 111.0, 117.2, 120.0, 122.8, 123.6, 124.4, 125.2, 125.9, 128.6, 129.5, 130.3, 132.9, 133.8, 139.4, 140.8, 148.2, 148.4, 148.7, 149.7, 152.5, 159.6; **LRMS-ESI** (m/z): 398.15 $[M + H]^+$.

6-(Thiophen-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (36). Yield: 27%; yellow solid; **M.p**.145-147 ^oC; $\mathbf{R_f}$ = 0.6 [hexane / ethyl acetate =8:2]; **ν_{max}(KBr)/cm^{-1}**:



3403, 2923, 2853, 1725, 1532, 1458, 1379, 1114; **^{1}H NMR** (400 MHz, DMSO): δ 6.79 (t, *J* = 6.1 Hz, 1H), 7.19 – 7.11 (m, 2H), 7.28 (dd, *J* = 5.0, 3.5 Hz, 1H), 7.45 (d, *J* = 8.2 Hz, 1H), 7.54 (t, *J* = 7.1 Hz, 1H), 7.69 (dd, *J* = 11.0, 4.4 Hz, 1H), 7.79 – 7.75 (m, 1H), 7.86 – 7.80 (m, 2H), 9.11 – 9.03 (m, 1H), 12.81 (s, 1H); **^{13}C NMR** (101 MHz, DMSO): δ 104.6, 111.3, 112.4, 113.8, 117.0, 118.0, 120.2, 123.8, 124.9, 126.1, 127.5, 127.7, 128.1, 128.4, 129.2, 130.2, 132.8, 137.5, 140.3, 146.9, 152.6, 158.7; **LRMS-ESI** (m/z): 369.05 $[M + H]^+$.

4.6. Cell line culture and drug treatment

The murine melanoma cell line (B16F10) was cultured in Dulbecco's Modified Eagle Medium (DMEM) (Himedia Laboratories Pvt. Ltd., Mumbai, India), supplemented with 10% heat inactivated fetal bovine serum (Himedia Laboratories Pvt. Ltd., Mumbai, India) and 1% of antibiotic solution (10000 U Penicillin and 10 mg Streptomycin per ml, Himedia Laboratories Pvt. Ltd., Mumbai, India). Cells were cultured at 37°C in humidified atmosphere with 5% CO_2. Stock solutions of all 19 novel compounds were prepared in DMSO at a concentration of 100 mM and stored. 100 mM stock solution of doxorubicin (ZhejingHisun Pharmaceutical Co. Ltd.) was prepared in DMSO and stored at -20°C.

4.7. MTT Assay

The anticancer activity of the synthesized compounds was determined using the MTT assay. 1×10^4 cells were seeded in 96 well plates and incubated overnight. Cells were treated with synthesized compounds at two concentrations (1 mM and 100 µM) in triplicates and incubated for 24 hours. 50 µl of 5 mg/mL of 23-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT; Himedia Laboratories Pvt. Ltd., Mumbai, India) was added and incubated for 4 hours. 150 µl of DMSO was added to dissolve formazan crystals and evaluated spectrophotometrically at 570 nm and 650 nm using Spectramax M4 (Molecular Devices, USA). The same assay was repeated with another batch of cells.

For the determination of IC_{50} values of six selected compounds, the same procedure was followed as described above, where six compounds (**19, 20, 23, 27, 35, 36**) were tested at 10 concentrations as 5 mM, 2 mM, 1 mM, 800 µM, 400 µM, 200 µM, 100 µM, 50 µM, 25 µM and 10 µM. The experiment was repeated with another batch of cells.

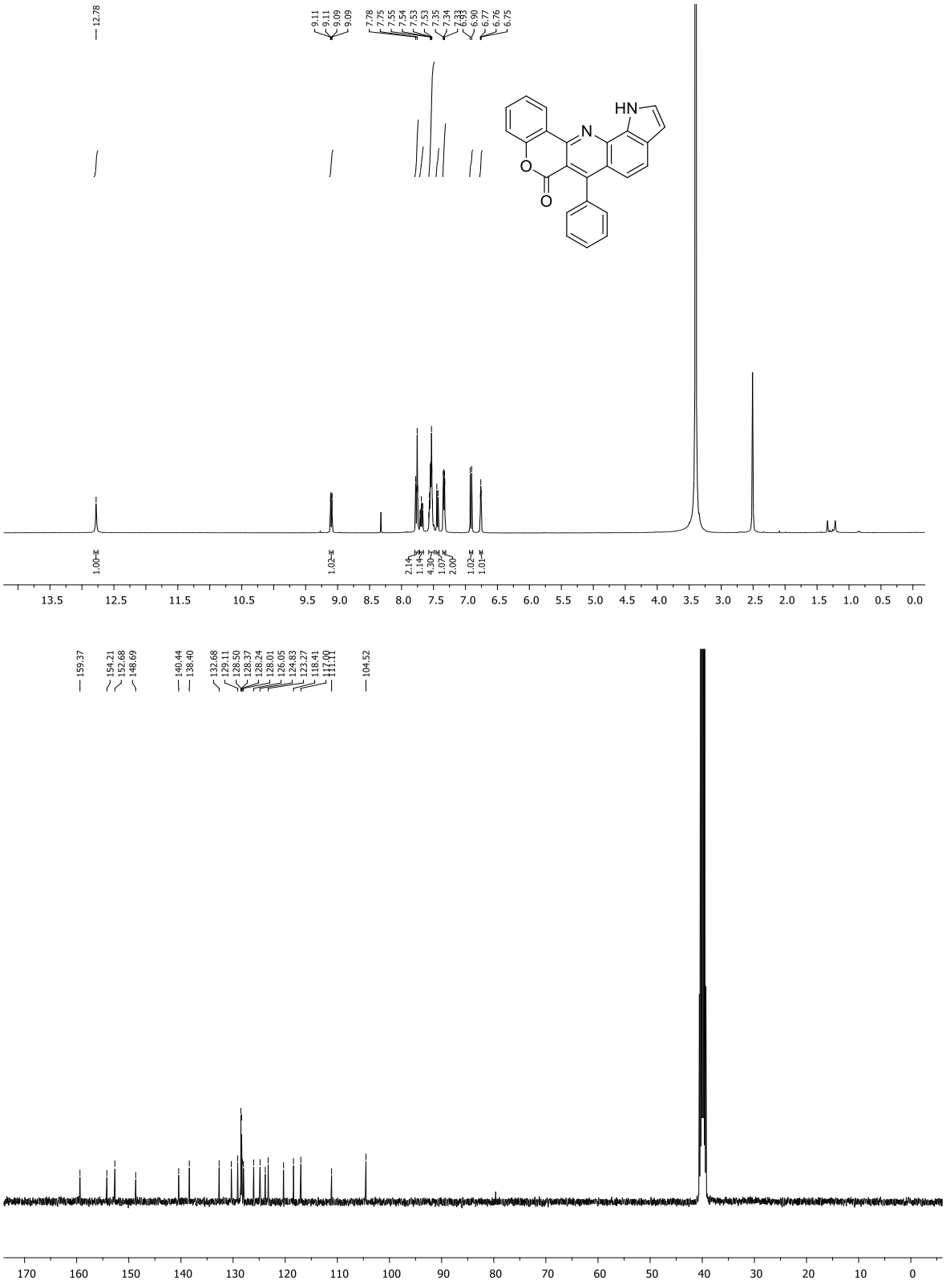
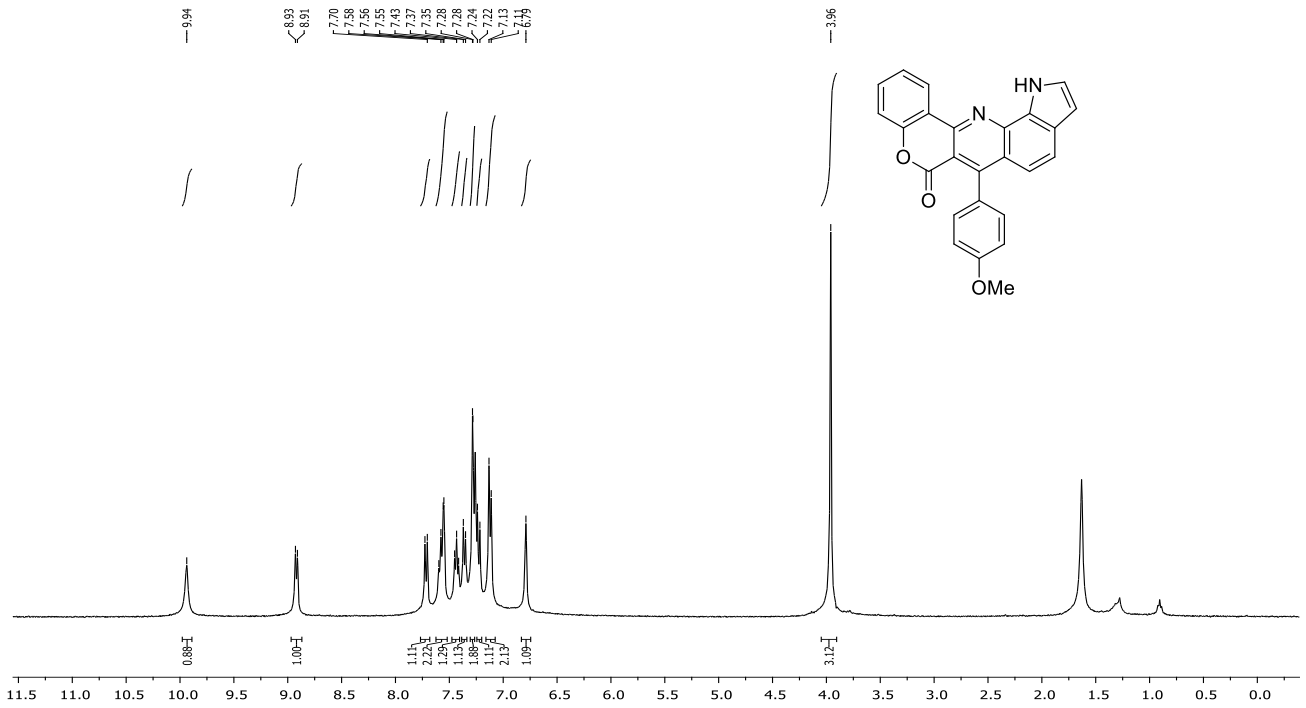
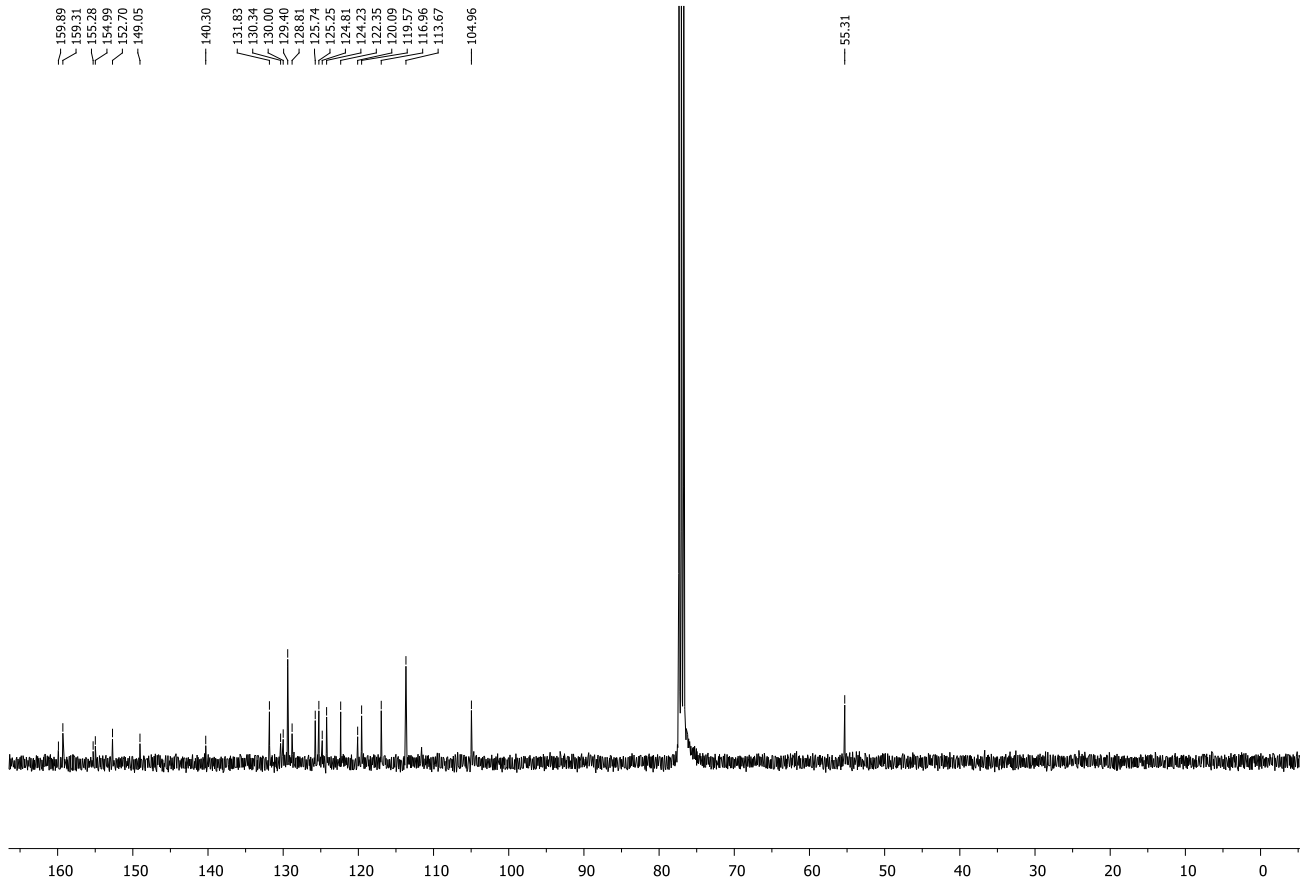
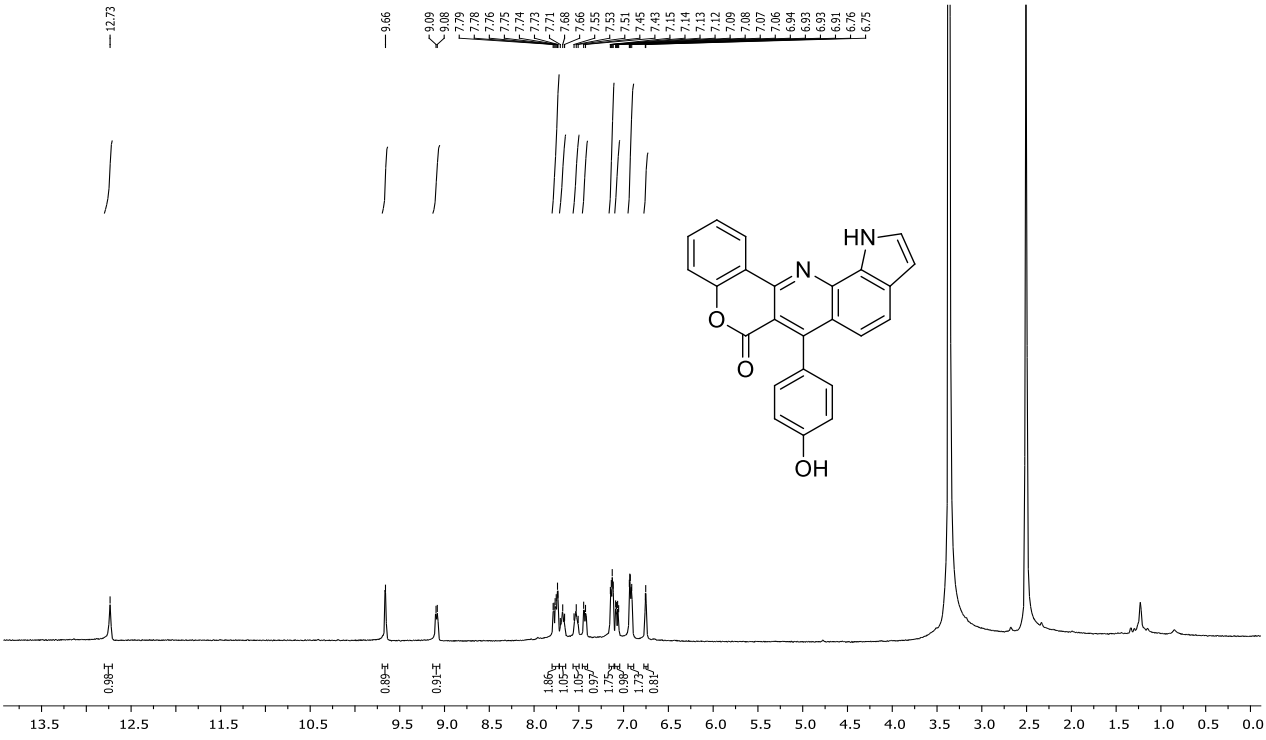


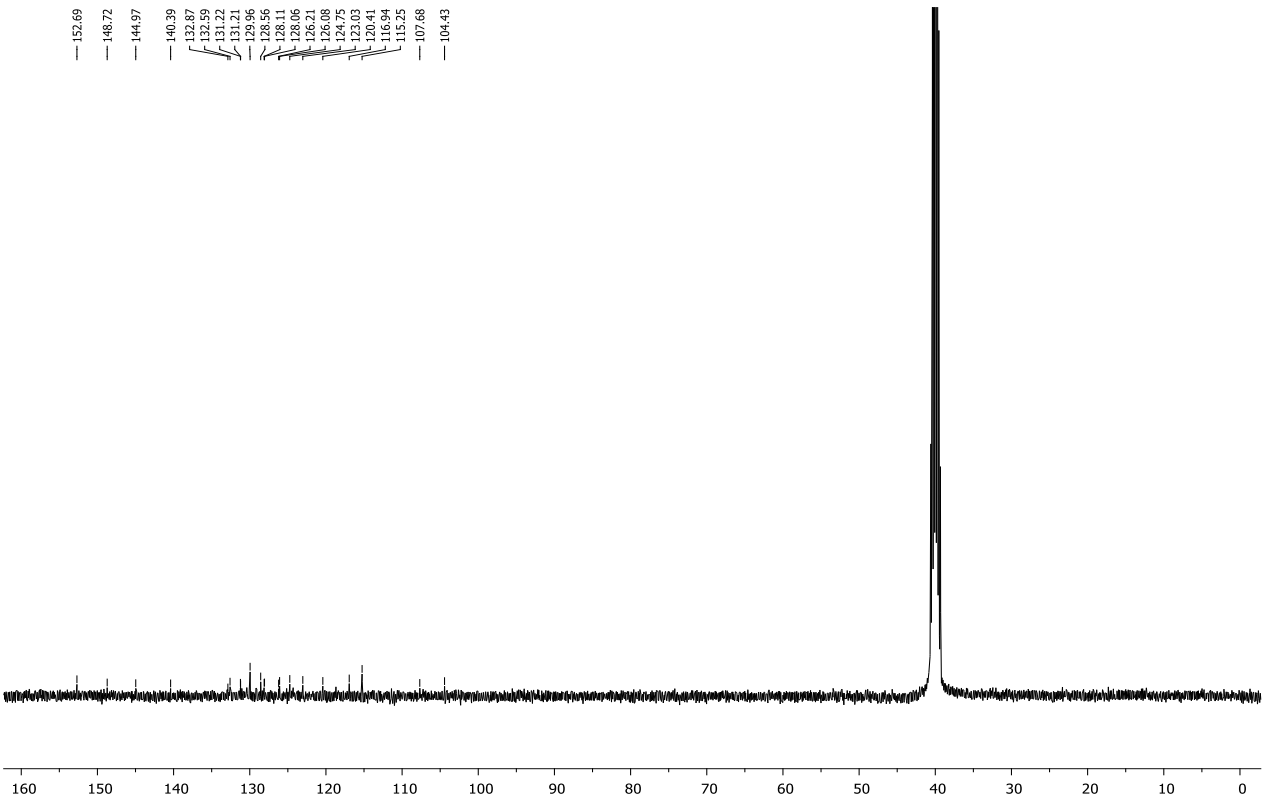
Figure 4.4: ^{1}H and ^{13}C NMR of compound **6-Phenylchromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (18)** in DMSO-d_6



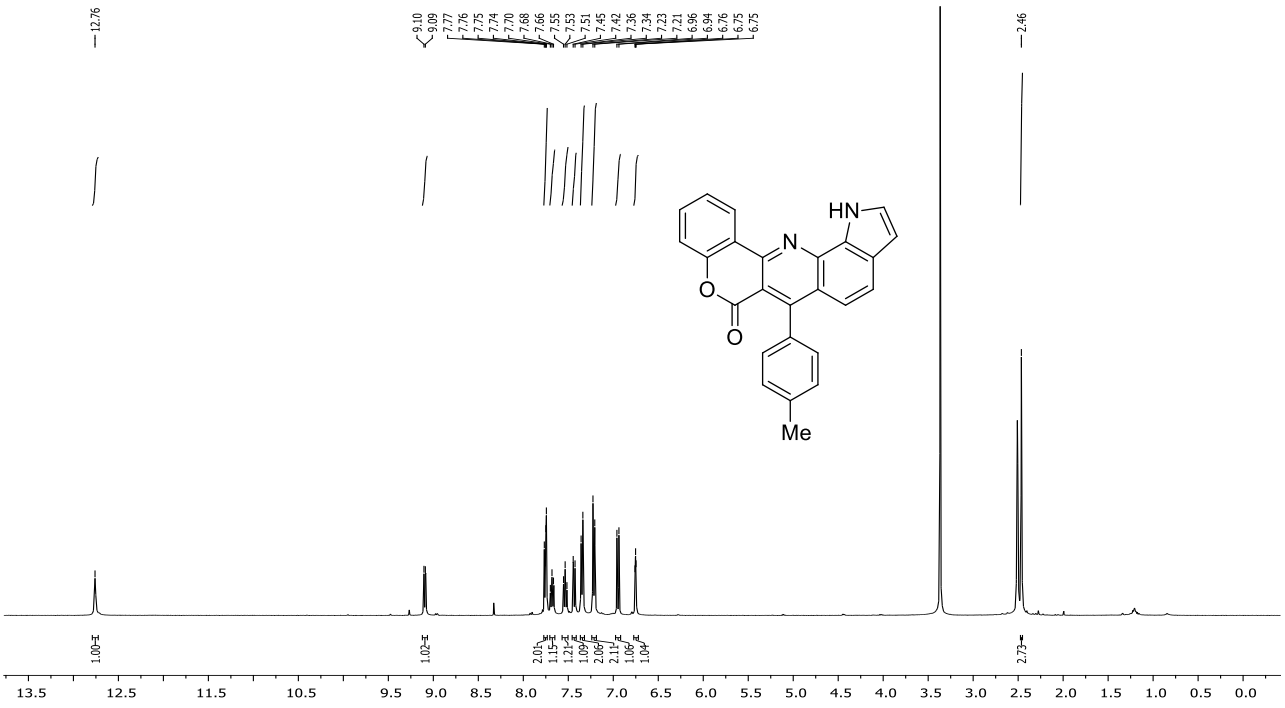


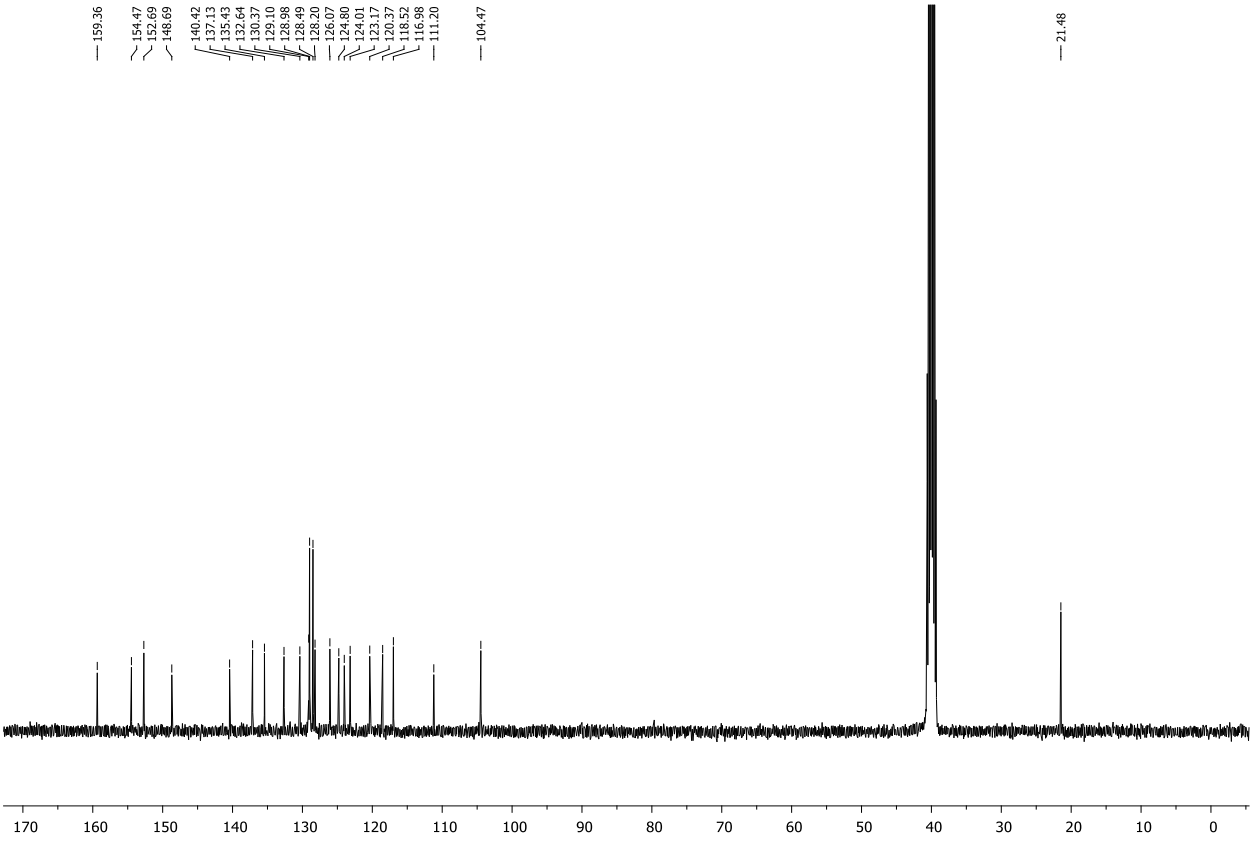
^{1}H and ^{13}C NMR of compound **6-(4-Methoxyphenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (19)** in DMSO-d_6



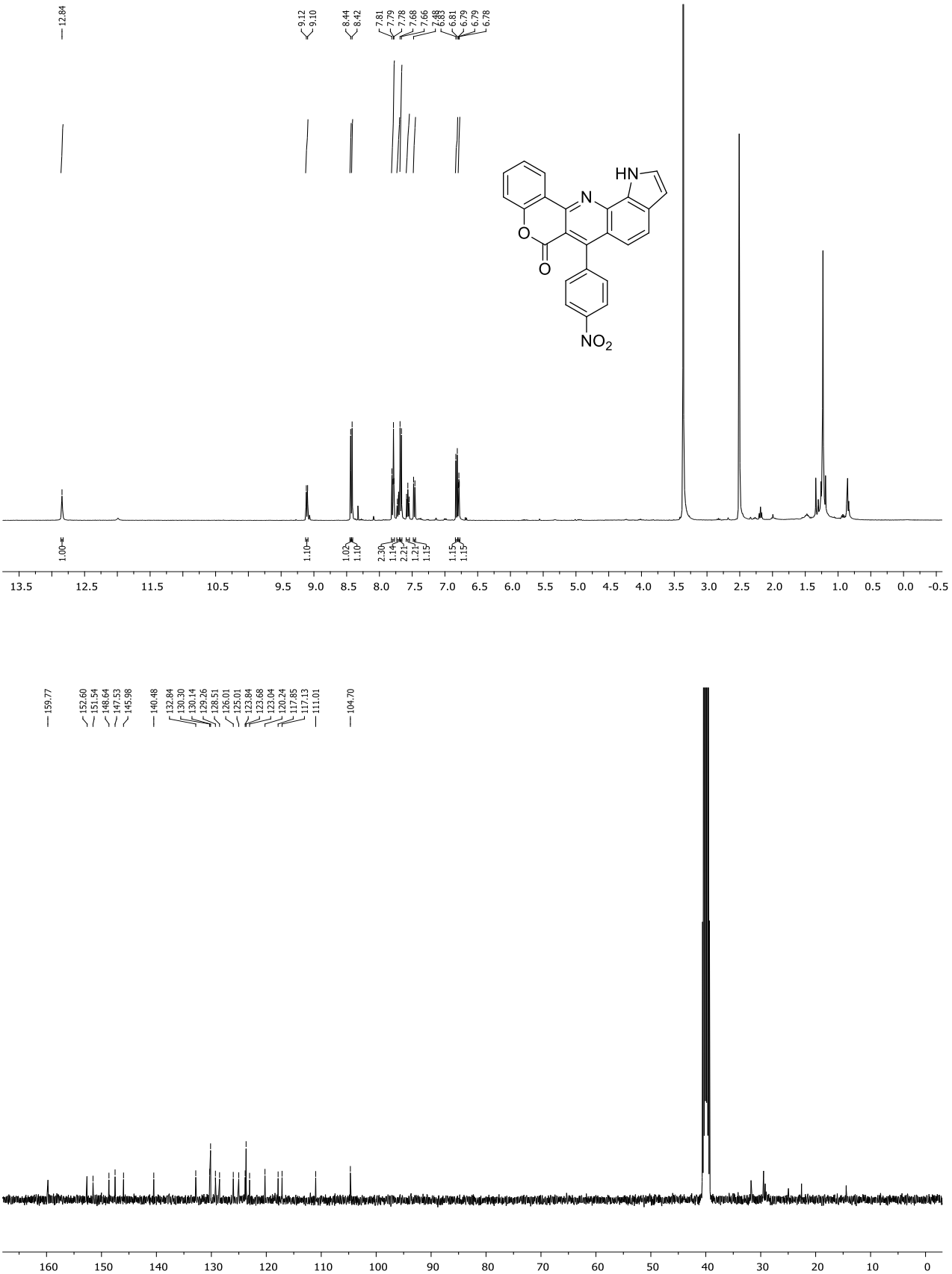


^{1}H and ^{13}C NMR of compound **6-(4-Hydroxyphenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (20)** in DMSO-d_6

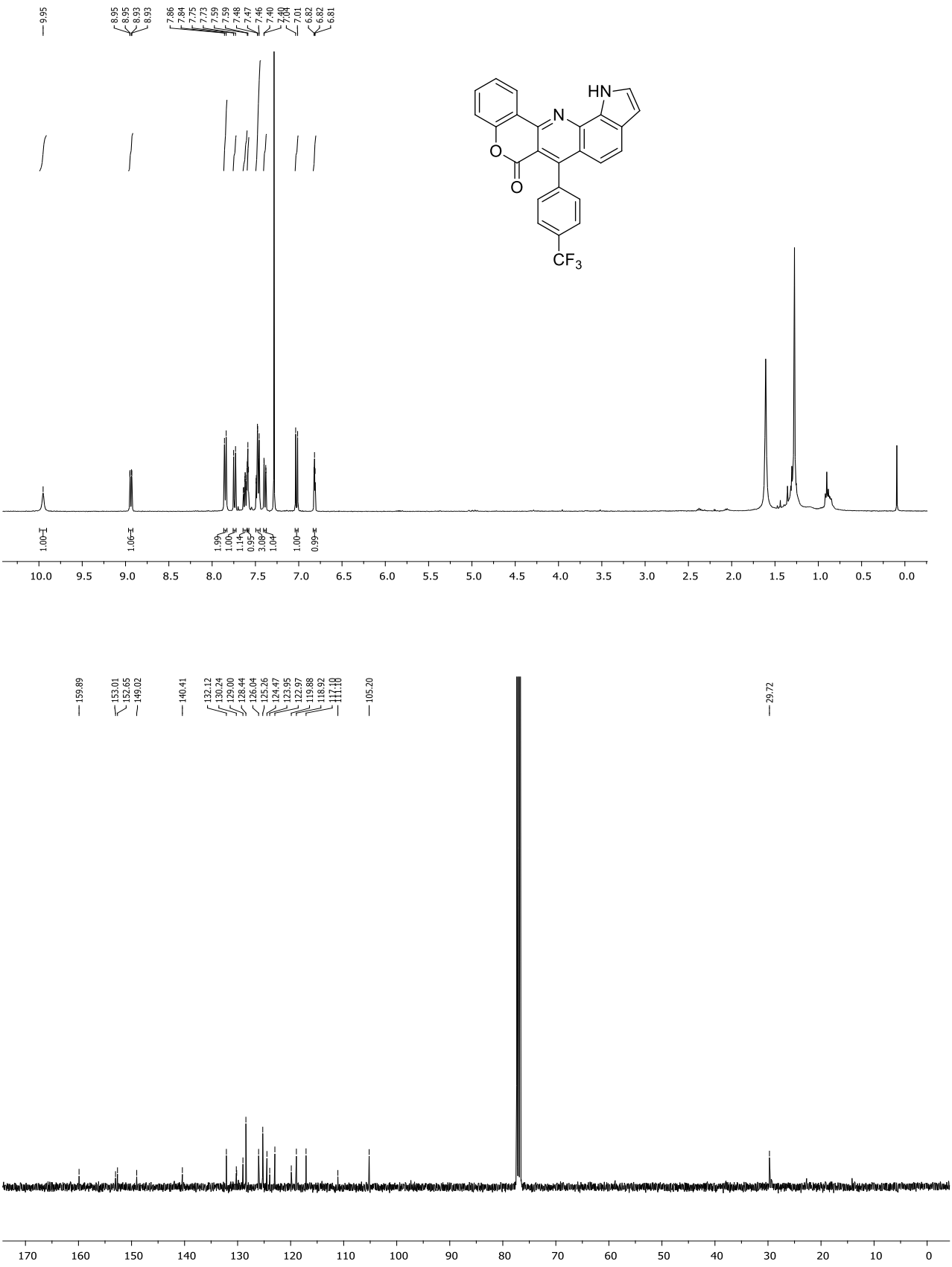




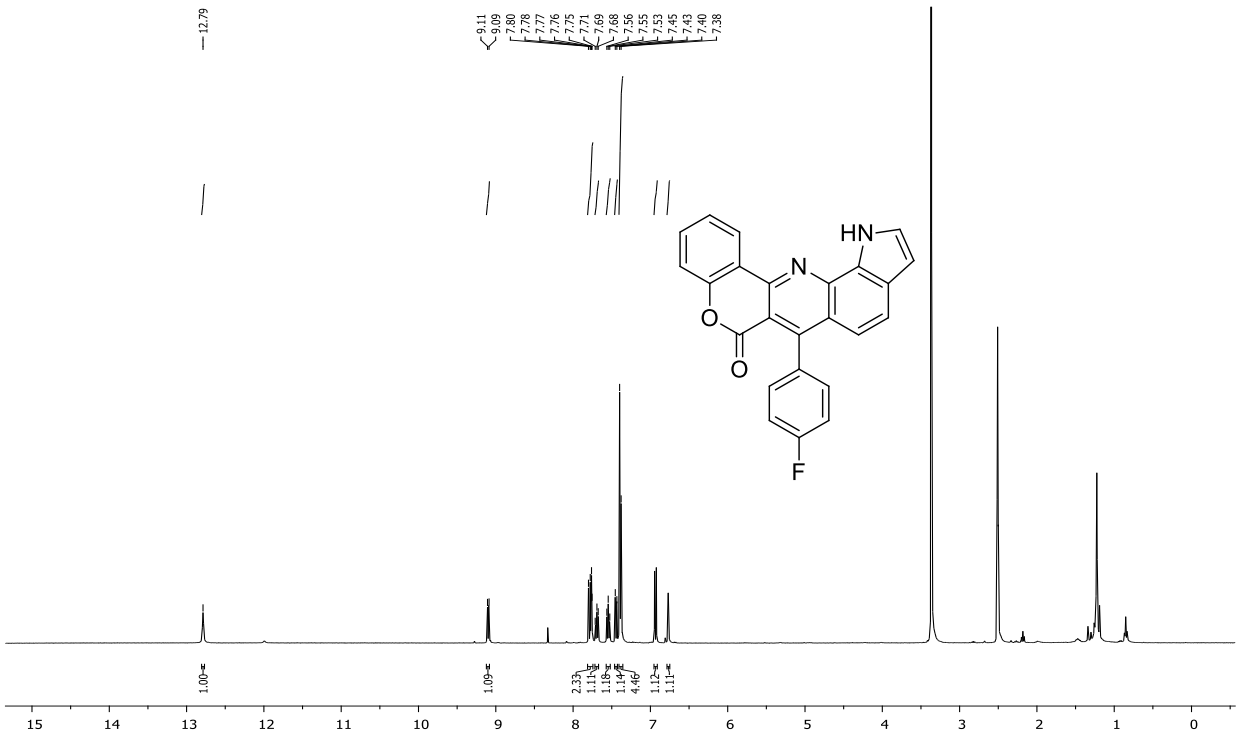
^{1}H and ^{13}C NMR of compound **6-(p-Tolyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (21)** in DMSO-d_6

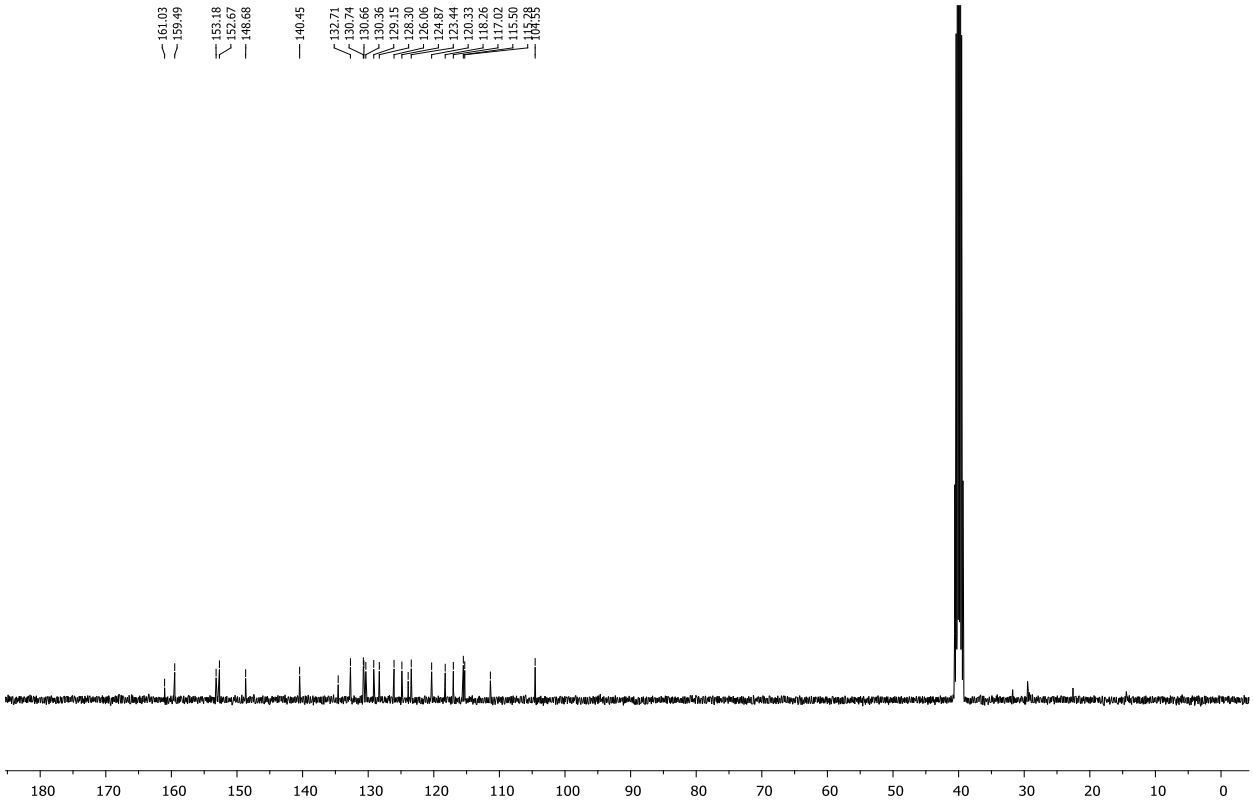


^{1}H and ^{13}C NMR of compound **6-(4-Nitrophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (22)** in DMSO-d_6

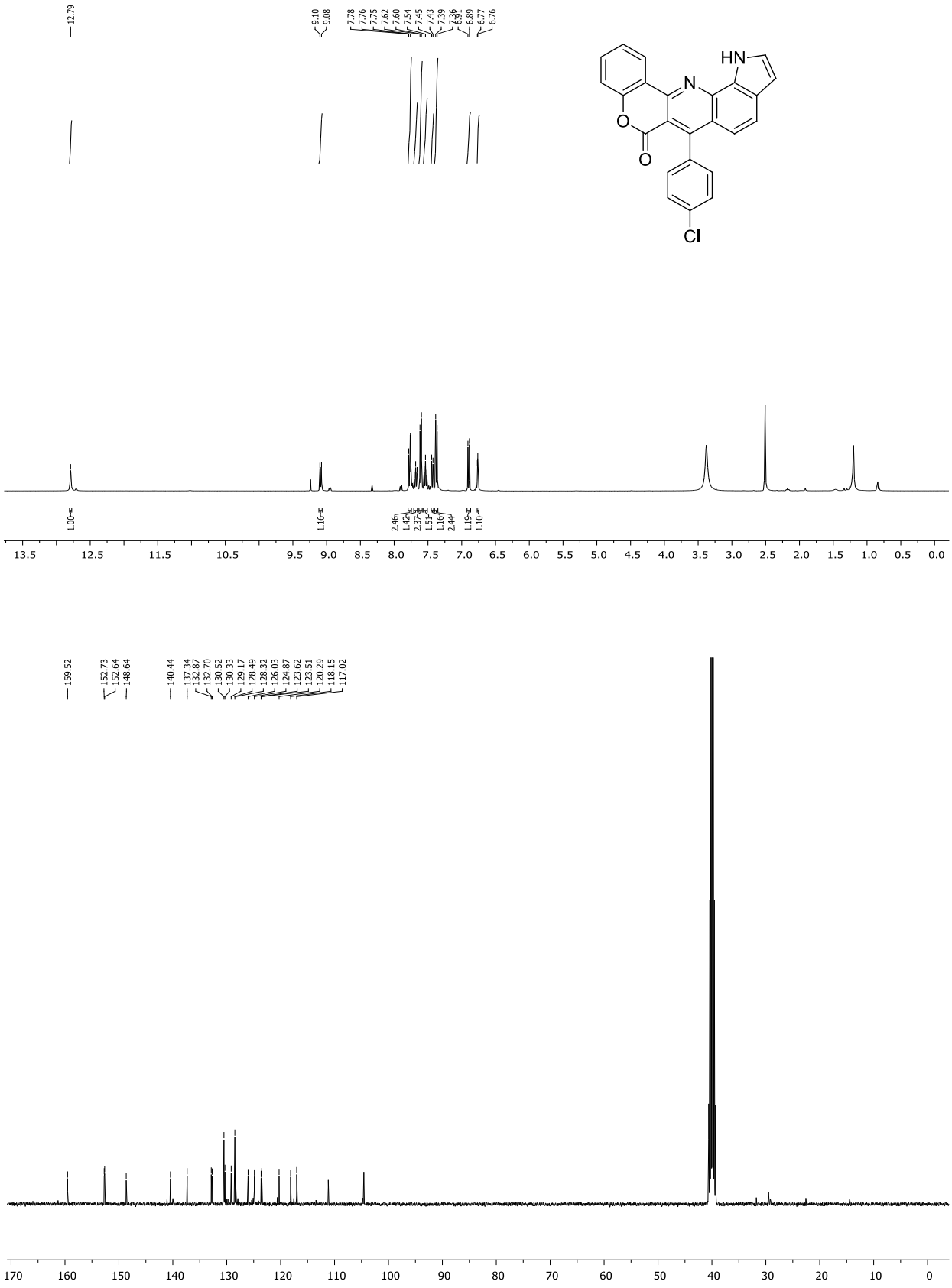


^{1}H and ^{13}C NMR of compound **6-(4-(Trifluoromethyl)phenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (23)** in DMSO-d$_6$

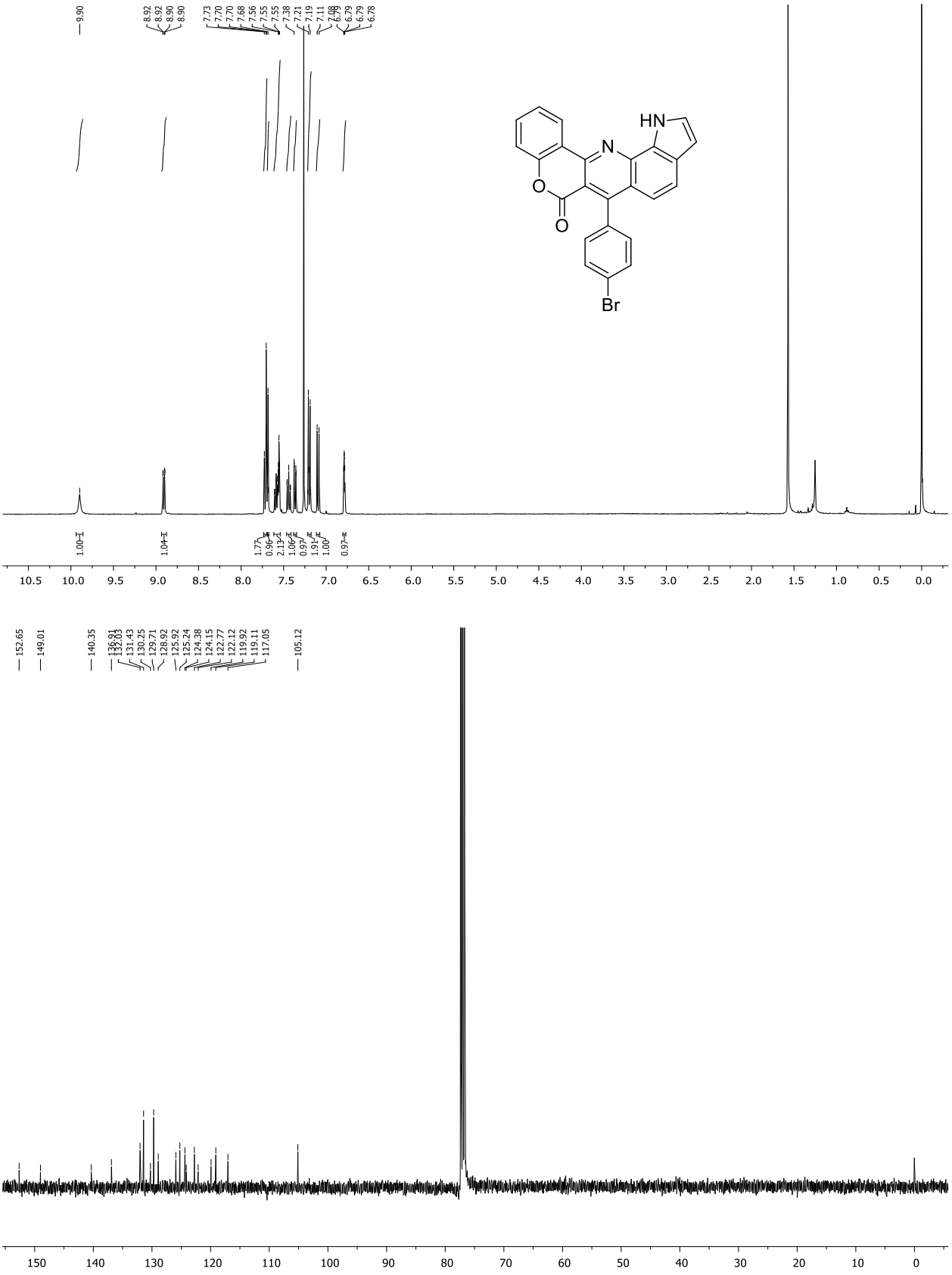




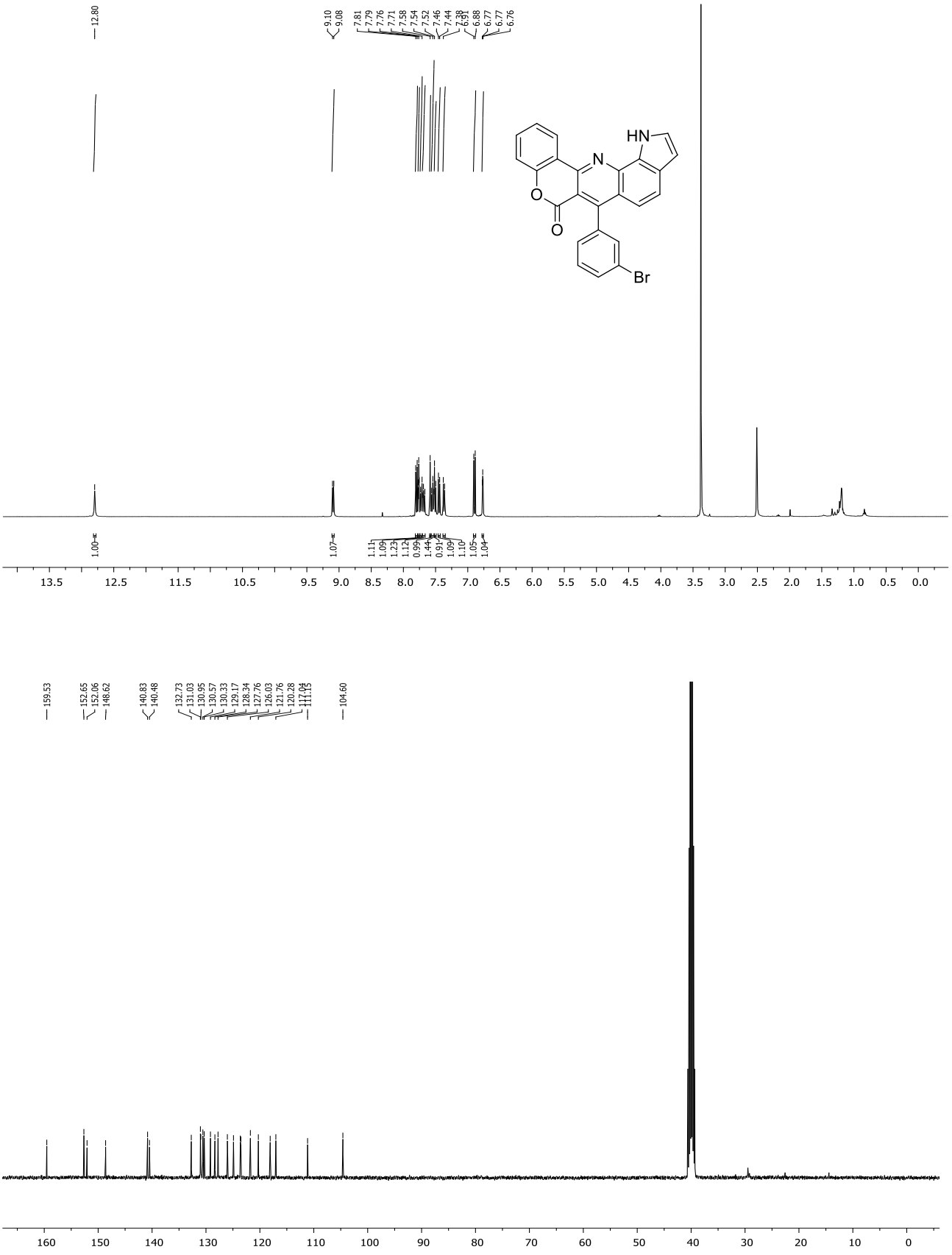
^{1}H and ^{13}C NMR of compound **6-(4-Fluorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (24)** in DMSO-d$_6$



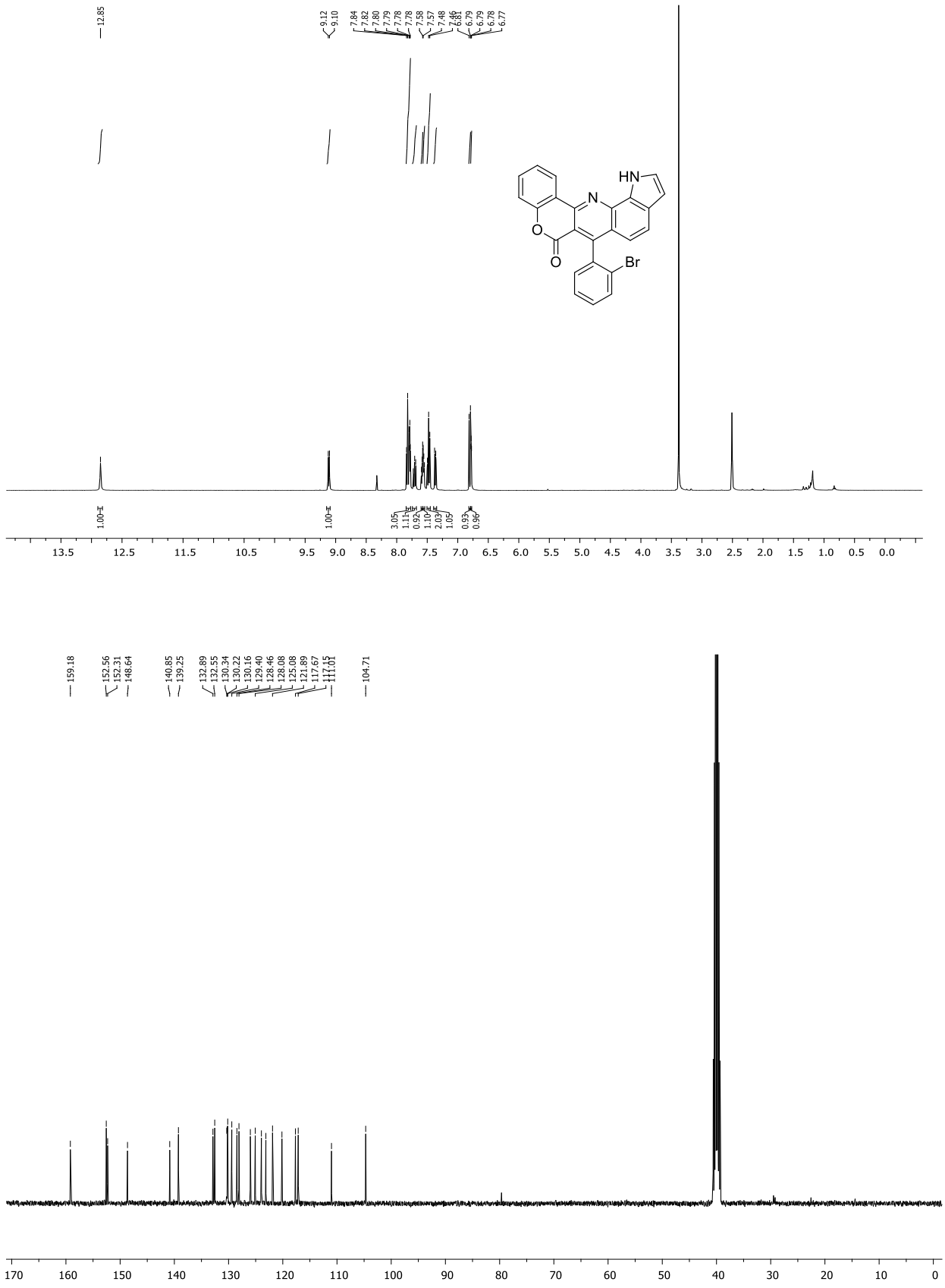
^{1}H and ^{13}C NMR of compound **6-(4-Chlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (25)** in DMSO-d_6



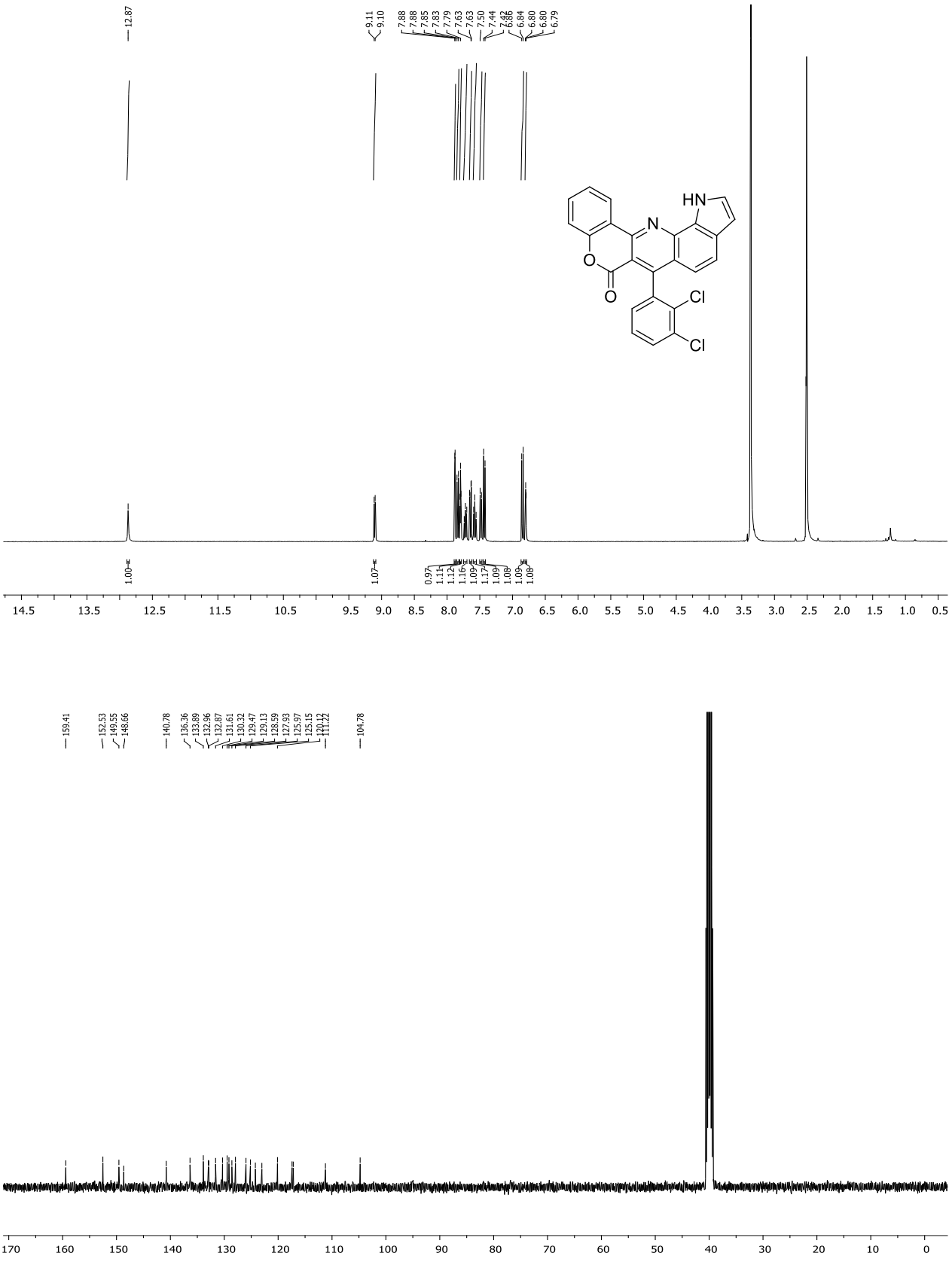
^{1}H and ^{13}C NMR of compound **6-(4-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (26)** in DMSO-d$_6$



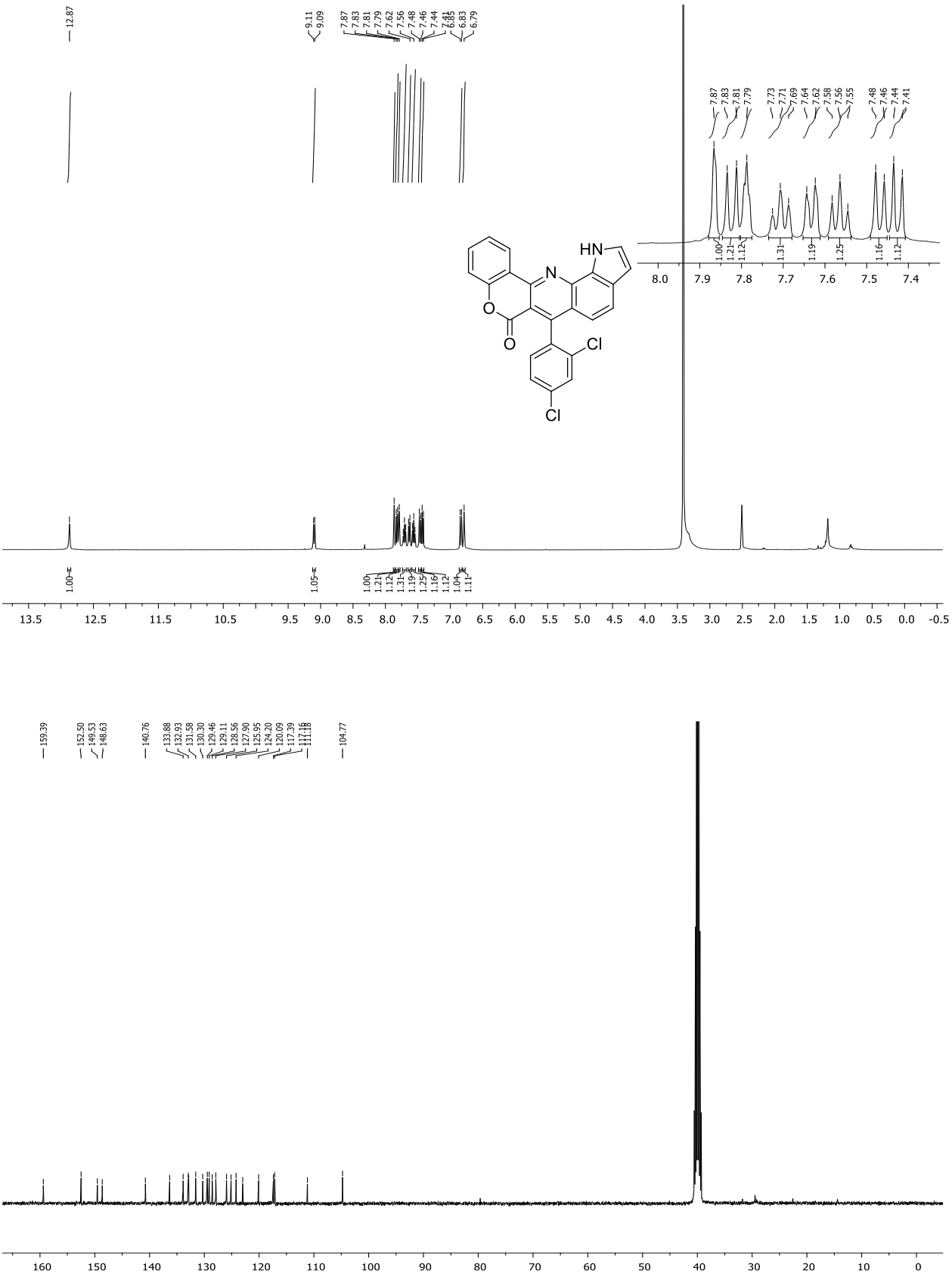
^{1}H and ^{13}C NMR of compound **6-(3-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (27)** in DMSO-d_6



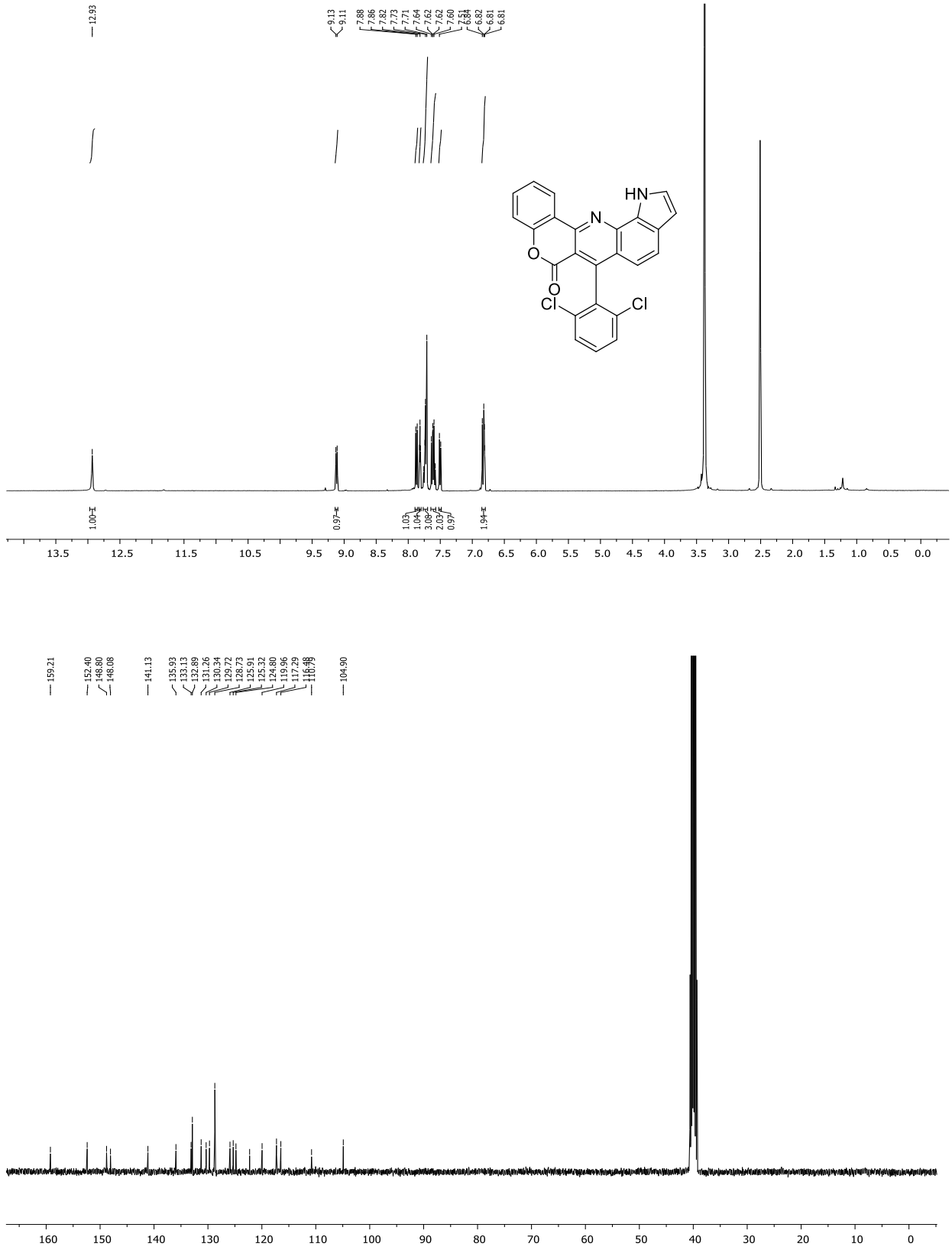
^{1}H and ^{13}C NMR of compound **6-(2-Bromophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (28)** in DMSO-d_6



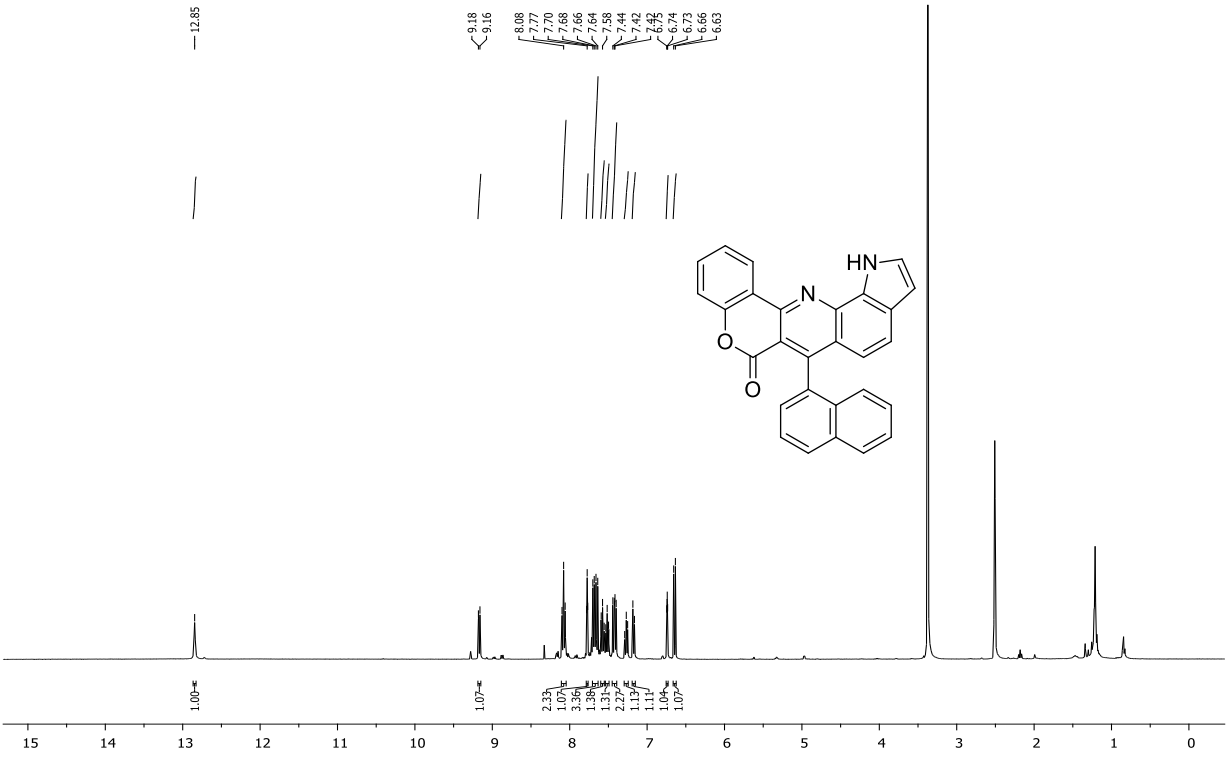
^{1}H and ^{13}C NMR of compound **6-(2,3-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (29)** in DMSO-d$_6$

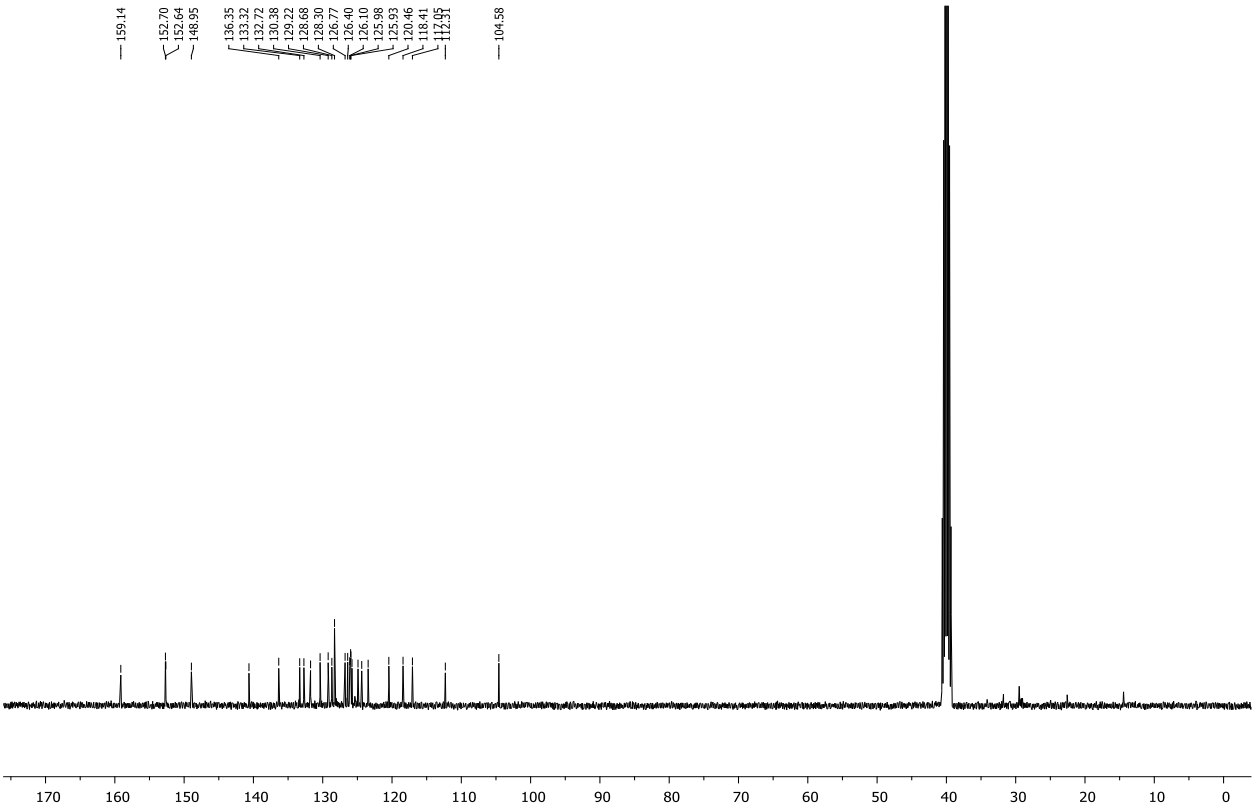


^{1}H and ^{13}C NMR of compound **6-(2,4-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (30)** in DMSO-d$_6$

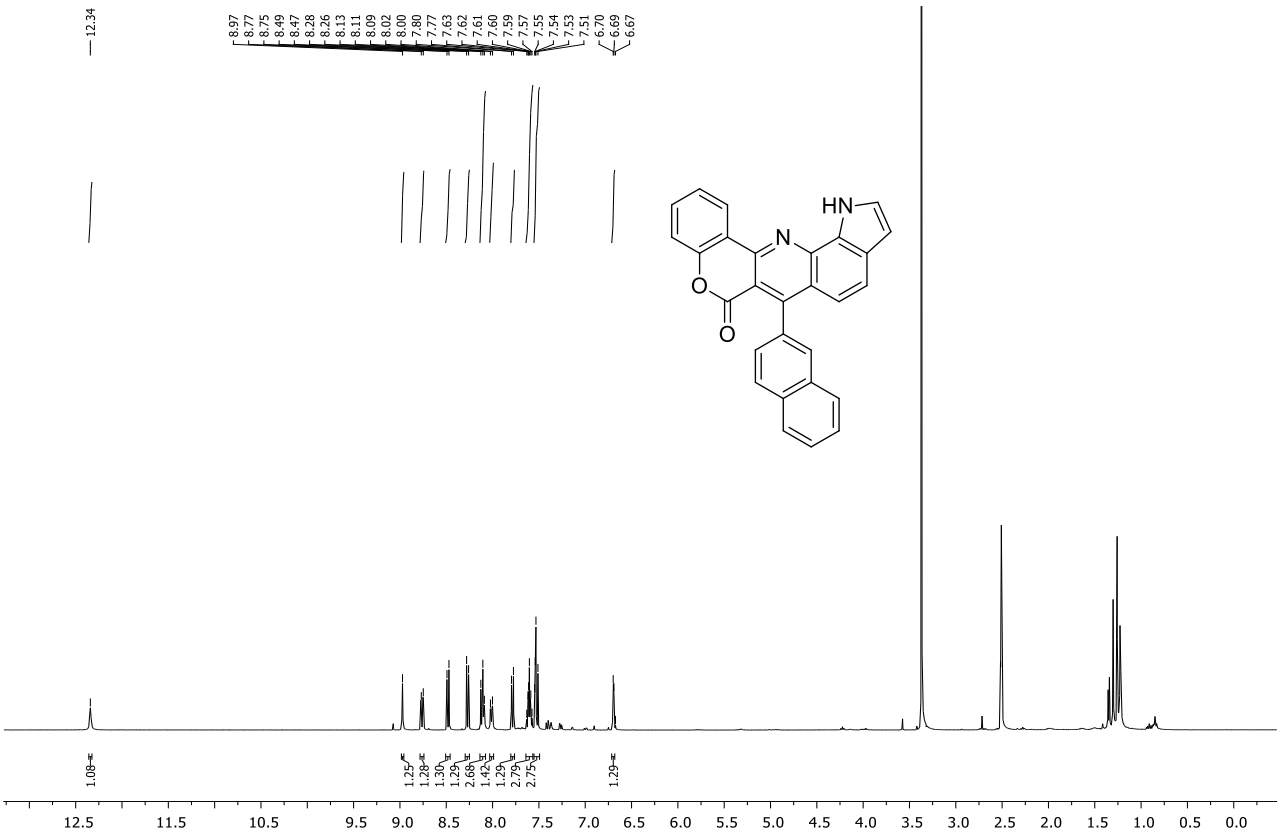


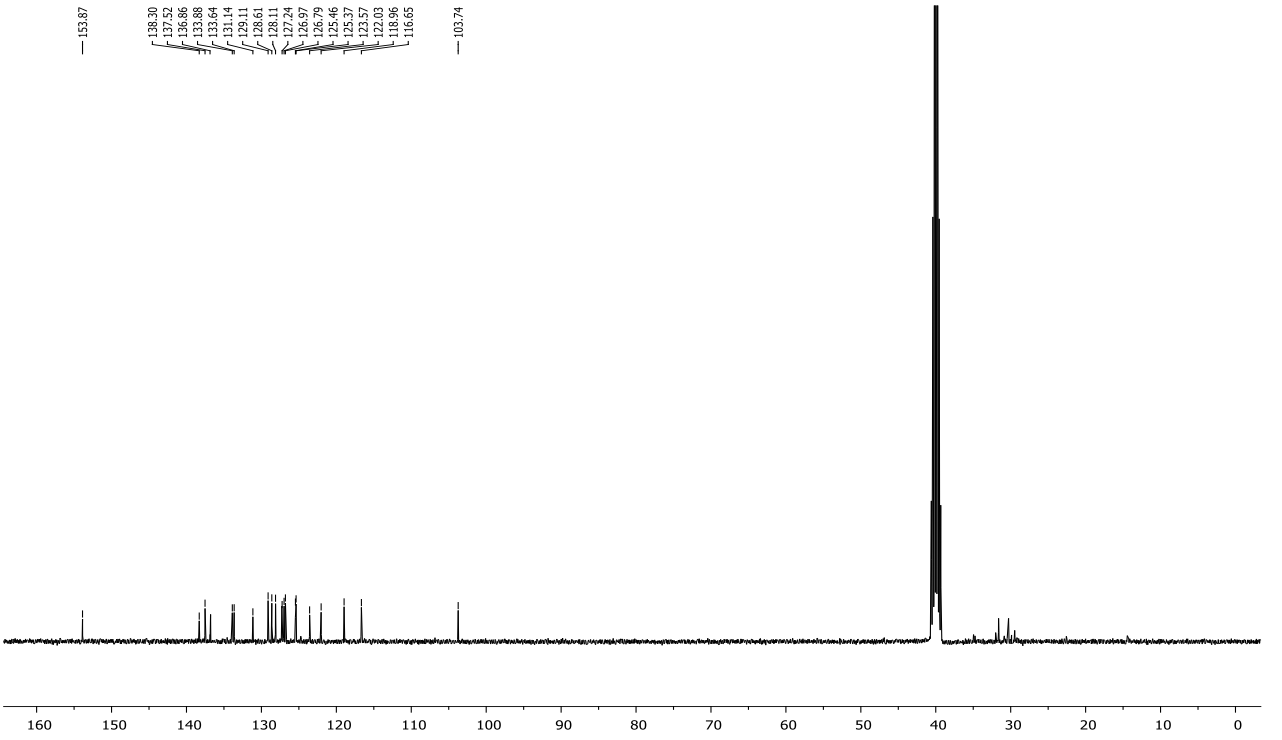
^{1}H and ^{13}C NMR of compound **6-(2,6-Dichlorophenyl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (31)** in DMSO-d$_6$



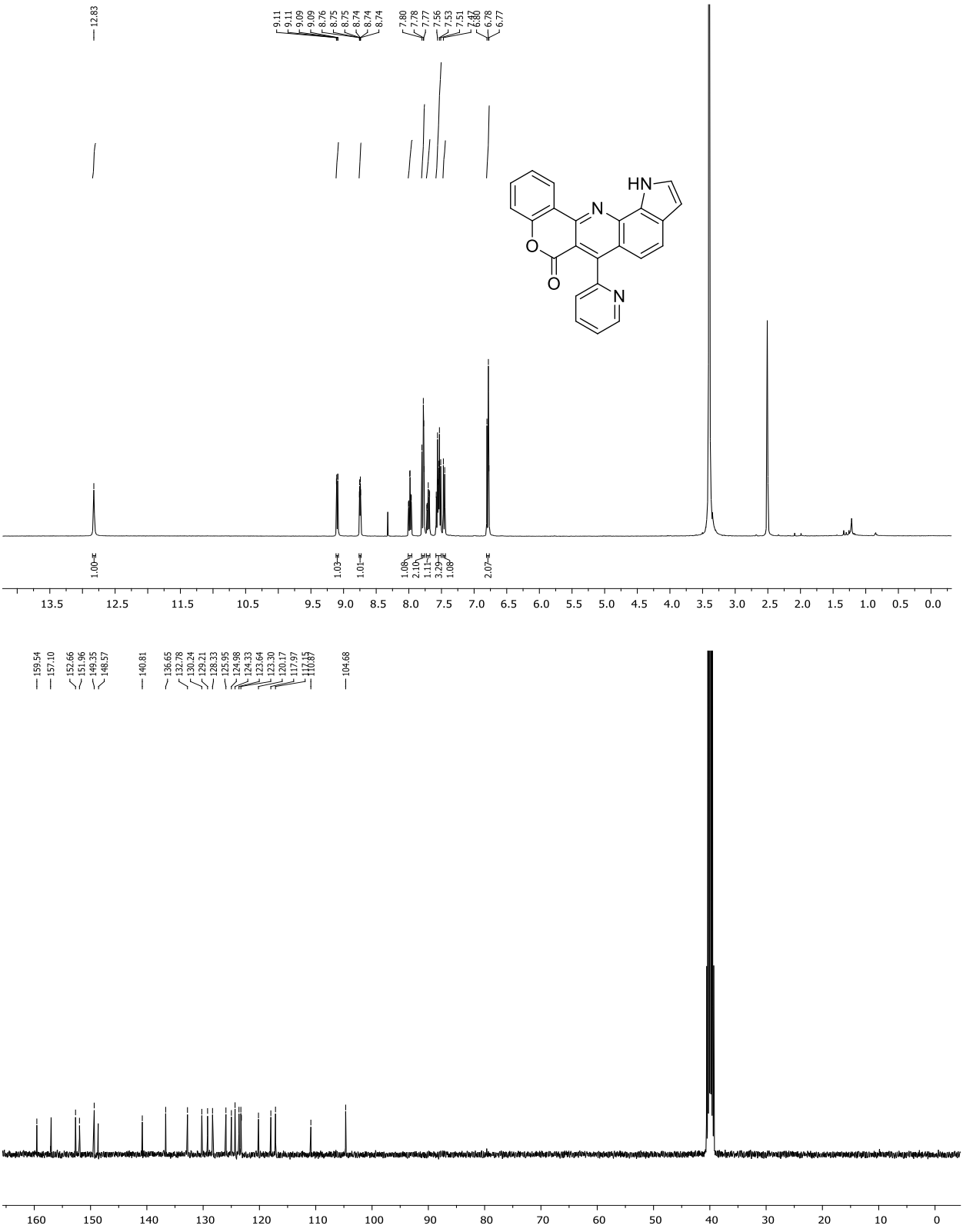


^{1}H and ^{13}C NMR of compound **6-(Naphthalen-1-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (32)** in DMSO-d_6

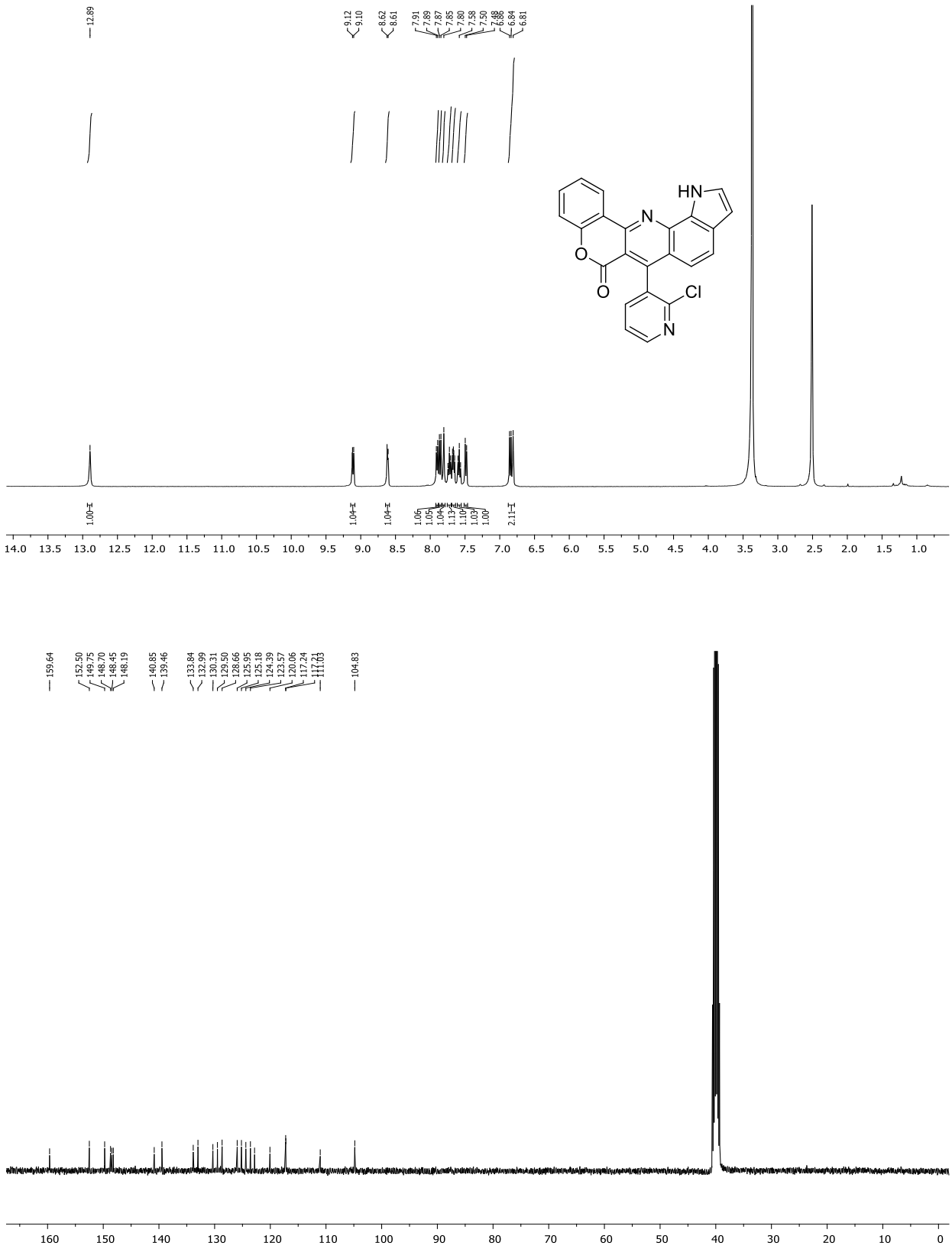




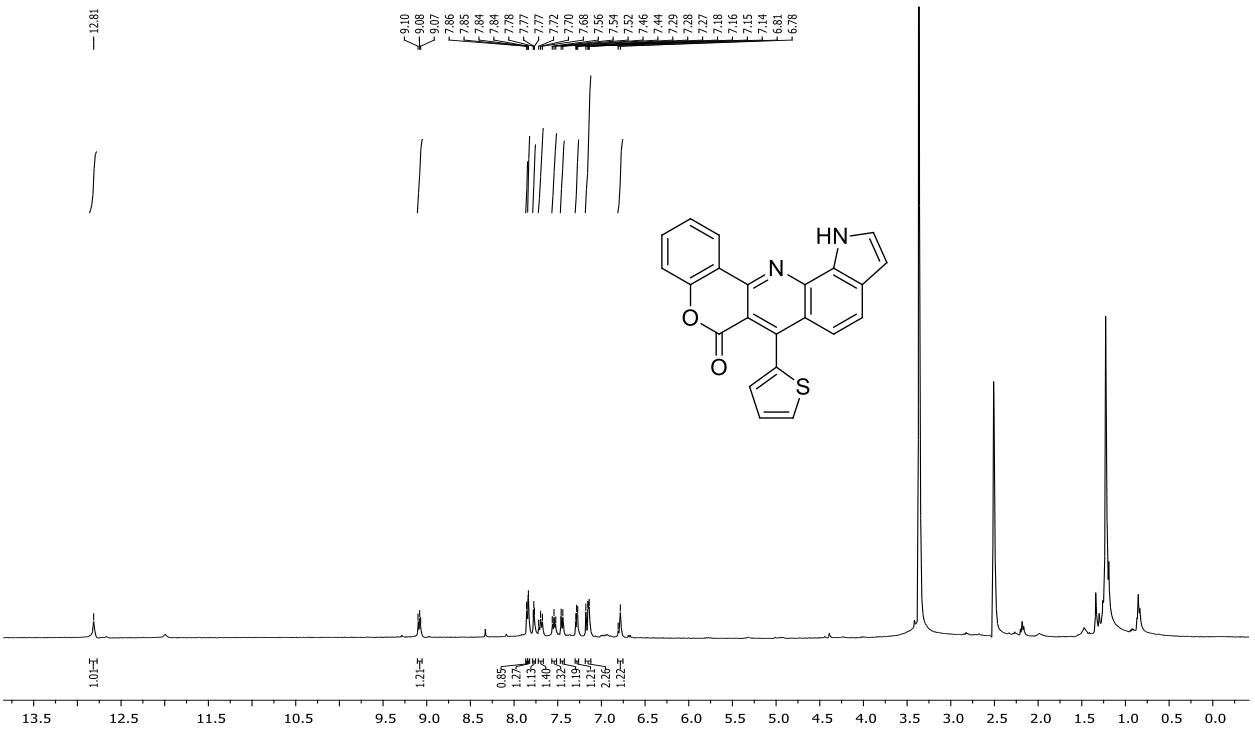
^{1}H and ^{13}C NMR of compound **6-(Naphthalen-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (33)** in DMSO-d_6

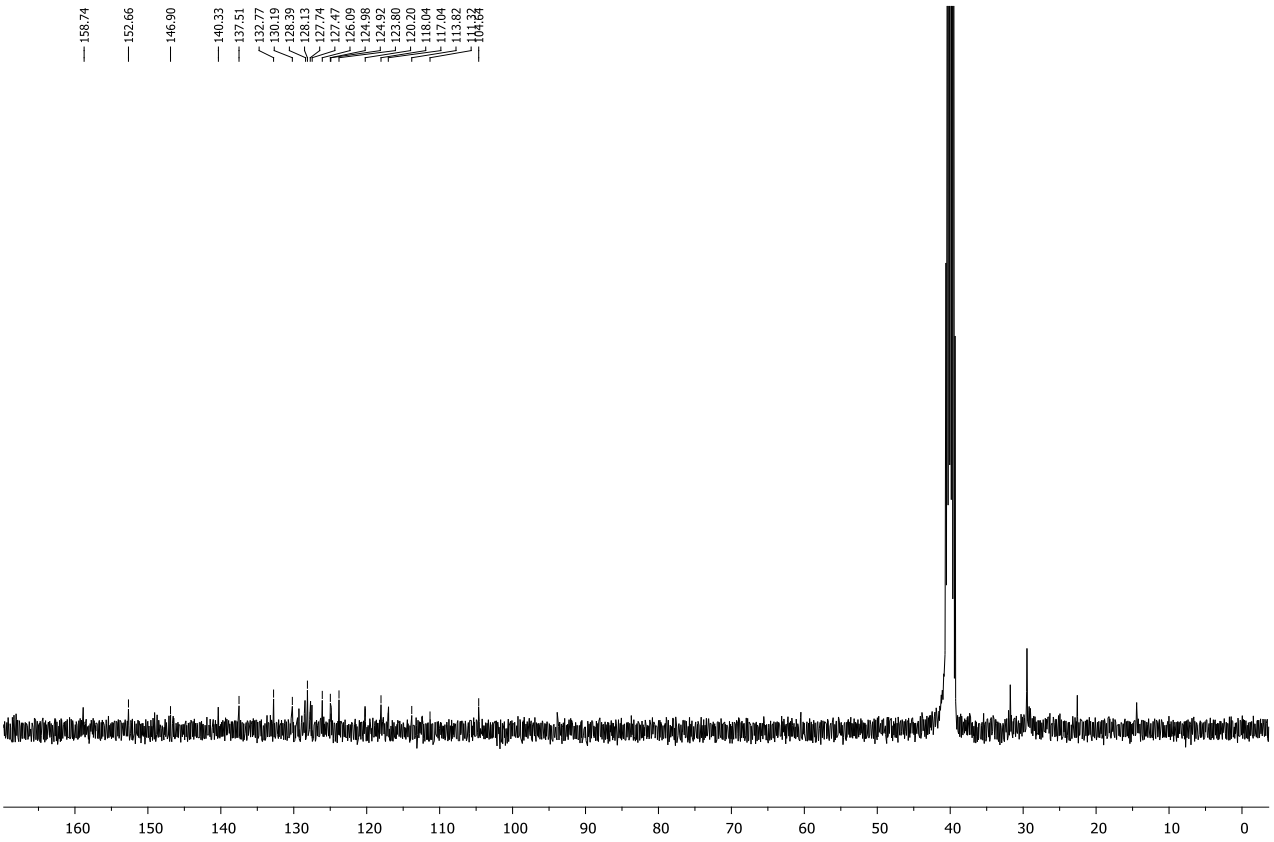


^{1}H and ^{13}C NMR of compound **6-(Pyridin-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (34)** in DMSO-d_6



^{1}H and ^{13}C NMR of compound **6-(2-Chloropyridin-3-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (35)** in DMSO-d$_6$





^{1}H and ^{13}C NMR of compound **6-(Thiophen-2-yl)chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one (4s)** in DMSO-d$_6$

4.8. References:

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CHAPTER 5

Summary and conclusions

5.1. Summary and conclusions

The main aim of this research work was to carry out synthesis of diverse fused heterocyclic systems based on pyridine/dihydropyridine and quinoline scaffolds.

First chapter looks at importance of various fused pyridine and dihydropyridine systems. A general survey of pyridine/dihydropyridine drugs has been given, along with discussion on the literature reports available on biological and photophysical properties of fused pyridines/dihydropyridines.

In chapter two we have described a method to synthesize fused δ-lactam compounds from chalcones, *via* a one pot two step route, involving consecutive application of Nazarov and Schmidt rearrangement reactions. Initial studies were done on 3-phenyl-1-(thiophen-3-yl)prop-2-en-1-one, using polyphosphoric acid (PPA) and sodium azide at 120 $^{\circ}$C, whence the unexpected *N*-alkyl amide product was obtained, instead of usual *N*-aryl analogue. With diverse 3-aryl substituted-1-(thiophen-3-yl)prop-2-en-1-one and 3-aryl substituted-1-(thiophen-2-yl)prop-2-en-1-one as substrates, corresponding *N*-alkyl amide compounds were obtained in modest to good yields. Interestingly, with 3-aryl substituted phenylprop-2-en-1-one as substrates under the optimized conditions, *N*-aryl amide compounds were formed as the final products instead of corresponding *N*-alkyl analogues.

Chapter three describes synthesis and anticancer activity evaluation of fused imidazoquinoline compounds. $Yb(OTf)_3$ has been utilized as a catalyst for the synthesis of 1,4-diaryl substituted imidazo[4,5-*c*]quinolines *via* modified Pictet-Spengler approach. The desired imidazole ring was synthesized from imines using TosMIC (toluenesulfonylmethylisocyanide) and subsequently functionalized at C-4 position yielding imidazoquinoline skeleton. Importantly, the final step was carried out without aid of any prefunctionalization to obtain the resultant compounds in good yields. Synthesized

compounds when screened for anticancer activity revealed highest activity with4-(2-bromophenyl)-1-phenyl-1*H*-imidazo[4,5-*c*]quinoline (IC_{50}: 103.3 μM).

$FeCl_3$ catalyzed synthesis of chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-one compounds is reported in chapter four. The target molecules were synthesized by combining 4-hydroxy coumarin, 7-amino indole and diverse aromatic aldehydes in 1,4-dioxane as a solvent, at 90 °C. The final compounds obtained were subjected to anticancer activity evaluation using murine melanoma cell lines (B16F10). Best result was obtained for compound bearing 2-chloro-3-pyridyl substituent, which showed IC_{50} value of 70.74 μM.

5.2. Future perspectives

- Extending the scope of one pot two step Nazarov-Schmidt rearrangement to other heterocyclic systems and experimental verification of the proposed mechanism.
- Mechanistic studies on $Yb(OTf)_3$ catalyzed synthesis of fused imidazo-quinolines.
- Based on preliminary anticancer screening results some promising structures were noticed, focus can be given on rational drug design approach to get more potent anticancer compounds.

Appendix

PUBLICATIONS

Yadagiri Thigulla, Santosh Ranga, Subhas Ghosal, Jayanty Subbalakshmi, Anupam Bhattacharya "One-Pot Two Step Nazarov-Schmidt Rearrangement for the Synthesis of Fused δ-Lactam Systems" *Chemistry Select*, **2017**, *2*, 9744-9750.

Yadagiri Thigulla, T. Uday Kumar, Prakruti Trivedi, Balaram Ghosh, Anupam Bhattacharya "One-Step Synthesis of Fused Chromeno[4,3-*b*]pyrrolo[3,2-*h*]quinolin-7(1*H*)-One Compounds and their Anticancer Activity Evaluation" *Chemistry Select*, **2017**, *2*, 2718-2721.

Yadagiri Thigulla, Mahesh Akula, Prakruti Trivedi, Balaram Ghosh, Mukund Jha, Anupam Bhattacharya "Synthesis and anti-cancer activity of 1,4-disubstituted imidazo[4,5-*c*]quinolines" *Organic & Biomolecular Chemistry*, **2016**, *14*, 876-883.

Mahesh Akula, **Yadagiri Thigulla**, Connor Davis, Mukund Jha, Anupam Bhattacharya "Synthesis of 4-substituted oxazolo[4,5-*c*]quinolines by direct reaction at C-4 position of oxazole" *Organic & Biomolecular Chemistry*, **2015**, *13*, 2600-2605.

Mahesh Akula, **Yadagiri Thigulla**, Amit Nag, Anupam Bhattacharya "Selective detection of fluoride using fused quinoline systems: effect of pyrrole" *RSC Advances*, **2015**, *5*, 57231-57234.

CONFERENCES

Yadagiri Thigulla, Anupam Bhattacharya; poster: "One pot two step synthesis of fused δ-lactam systems by systematic application of Nazarov-Schmidt rearrangement" CRSI-NSC-21, held at CSIR-IICT Hyderabad, Telangana, India in **2017,** July 13-16.

Yadagiri Thigulla, Mahesh Akula Anupam Bhattacharya; poster: "Synthesis and anti-cancer activity of 1,4-disubstituted imidazo[4,5-*c*]quinolines" NDCS-2015, held at BITS-Pilani, Rajastan, India in **2015,** October 17-19.

Yadagiri Thigulla, Anupam Bhattacharya; poster: "Chalcone mediated synthesis of 4-aryl substitutedpyrrolo-[3,4-*c*]quinoline compounds" CRSI-NSC15, CSIR-NIIST, held at Banaras Hindu University, Varanasi, India in **2013,** February 1-3.

BIOGRAPHY

Mr. T Yadagiri completed his B.Sc and M.Sc degrees in chemistry from Osmania University, Hyderabad. He has worked as a Drug Discovery Chemist in Hetero Drugs Research Foundation, Hyderabad for two years two months (Feb 2010-Apr 2012), prior to joining the Ph.D programme of Department of Chemistry, Birla Institute of Technology and Science-Pilani, Hyderabad Campus in April, 2012. He has published five research papers in international peer reviewed journals and presented his research work in three conferences.

Prof. Anupam Bhattacharya has completed his bachelor's and master's degree in chemistry from University of Delhi. He carried out his doctoral study at the same university under the supervision of Prof. V. S. Parmar. During Ph.D he was a DST-DAAD exchange fellow at Max Plank Institute for Molecular Physiology, Dortmund, Germany, where he has worked under Prof. Oliver Seitz. After completing his postdoctoral studies with Prof. David Zechel at Queens University, Kingston, Canada, he joined BITS-Pilani Hyderabad Campus in the year 2008 where he is currently an associate professor in department of chemistry. His research interest lies in the area of synthetic organic chemistry, bio-organic chemistry and organic molecule based ion sensors.