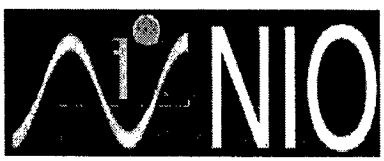
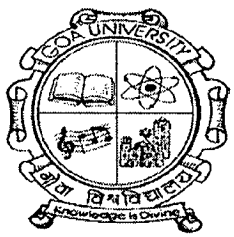
SYNTHETIC STUDIES TOWARDS BIOACTIVE NATURAL PRODUCTS AND THEIR ANALOGUES

By

Mr. PRAKASH T. PARVATKAR

M. Sc.





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All the suggested corrections are incorporated.

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CERTIFICATE

Certified that the work incorporated in the thesis entitled "**Synthetic Studies Towards Bioactive Natural Products and their Analogues**" submitted by **Mr. Prakash T. Parvatkar**, was carried out by the candidate under our supervision and the same has not been submitted elsewhere for the award of a degree.

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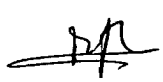
DECLARATION

I hereby declared that the work embodied in the thesis entitled "**Synthetic Studies Towards Bioactive Natural Products and their Analogues**" is the result of investigation carried out by me under the guidance of Dr. P. S. Parameswaran (NIO) and Dr. S. G. Tilve (Goa University) and it has not previously formed basis for any other titles.

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



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All the teaching and non-teaching staff of the Department of Chemistry, Goa University and all the scientists and others officials of National Institute of Oceanography has been extremely helpful and I thank them all. I also acknowledge the Librarian and the staff members of Library, NIO and Goa University for their constant help.

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I am greatly indebted to my parents and family members for morale support, encouragement, valuable suggestions, great affection and selfless sacrifice. Without their constant support and inspiration, it would have been impossible for me to be where I am at present.

I close with thanks giving to almighty for the showers of blessing during the hours of trial.

Mr. Prakash T. Parvatkar

Dedicated To My

Beloved Parents

CONTENTS

GENERAL REMARKS

1) The compound numbers, figure numbers, scheme numbers and reference numbers given in each chapter refer to that particular chapter only.

2) All melting points and boiling points were recorded using Thiele's tube and are uncorrected.

3) Commercial reagents were used without further purification.

4) All solvents were distilled prior to use and then dried using standard procedure.

5) Petroleum ether refers to the hydrocarbon fraction collected in the boiling range 60 – 80 ^{0}C.

6) All reagents were prepared using literature methods.

7) Chromatographic purification was conducted by column chromatography using either silica gel (60 – 120 mesh size) or neutral alumina.

8) Thin layer chromatography (TLC) were carried out on glass plates using silica gel G and were developed in iodine.

9) The IR spectra were recorded on Shimadzu FT-IR spectrophotometer.

10) ^{1}H NMR (300 MHz) and ^{13}C NMR (75 MHz) spectra were recorded on a Brucker AVANCE 300 instrument and the multiplicities of carbon signals were obtained from DEPT experiment.

11) The high resolution mass spectra (HRMS) were recorded on MicroMass ES-QTOF mass spectrometer.

DEFINATION OF ABBREVIATIONS

1) General Abbreviations

g	Gram/s
mg	Milligram/s
mmol	Millimole
mL	Milliliter
Mp / mp	Melting point
b.p.	Boiling point
dil.	Dilute
Eq.	Equation/s
lit.	Literature
d	Day/s
h	Hour/s
min	Minute/s
sec	Second/s
Z	Zussamen (together)
E	Eentegegen (opposite)
R	Rectus
S	Sinister
Fig.	Figure
conc.	Concentrated
glac.	Glacial
sat.	Saturated
aq.	Aqueous
anhyd.	Anhydrous
$h\nu$	Irradiation
^0C	Degree Celcius
%	Percentage
RT / r.t.	Room temperature
Expt.	Experiment
Temp.	Temperature
MW / μW	Microwave

o	Ortho
m	Meta
p	Para
MS	Molecular sieves
psi	Pounds per square inch
cat.	Catalytic
atm.	Atmospheric
et al.	Et alia (and others)
TLC / tlc	Thin layer chromatography

2) Compound Abbreviations

Ac	Acetyl
Ac_2O	Acetic anhydride
TBAF	Tetrabutyl ammonium fluoride
Ar	Aryl
Boc	*tert*-Butyl carbonyl
Bn	Benzyl
Bz	Benzoyl
t-Bu	*tert*-Butyl
TFA	Trifluoro acetic acid
TFAA	Trifluoro acetic anhydride
TEA	Triethyl amine
AcOH	Acetic acid
MeOH	Methanol
EtOH	Ethanol
m-CPBA	*m*-Chloroperbenzoic acid
p-TsOH	*p*-Toluene sulfonic acid
ICZs	Indolocarbazoles
DMSO	Dimethyl sulfoxide
DMF	N,N-Dimethylformamide
THF	Tetrahydrofuran
Et	Ethyl

Me	Methyl
LDA	Lithium diisopropylamide
LAH	Lithium aluminium hydride
NBS	*N*-Bromosuccinimide
EtOAc	Ethyl acetate
n-BuLi	*n*-Butyl lithium
t-BuOK / KTBT	Potassium tertiary butoxide
O_3	Ozone
Pd/C	Palladium on activated charcoal
Ph	Phenyl
PMB	*p*-Methoxybenzyl
PPh_3 / TPP	Triphenylphosphine
TBAF	Tetrabutylammonium fluoride
Ms	Methane sulfonyl
TMS	Trimethylsilyl
TMSCN	Cyanotrimethyl silane
Ts	p-Toluene sulfonyl
Py	Pyridine
TEP	Triethyl phosphite
PPA	Polyphosphoric acid
DCM	Dichloromethane
DCE	1,2-Dichloroethane
DME	1,2-Dimethoxyethane
DMS	Dimethyl sulphate
DDQ	2,3-Dichloro-5,6-dicyanobenzoquinone
Pet ether	Petroleum ether
TsCl	Tosyl chloride
AIBN	Azobisisobutyronitrile
DMAP	4-Dimethyl amino pyridine
HMPA	Hexamethylphosphoramide
DIAD	Diisopropyl azodicaboxylate
DEAD	Diethyl azodicarboxylate

DCC	Dicyclohexyl cabodiimide
CAN	Cerric ammonium nitrate
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DMP	Dess-Martin periodinane
DIBALH	Diisobutyl aluminium hydride
MOM	Methoxymethyl ether
Boc_2O	*tert*-Butyl dicarbonate
i-PrOH	Iso-propanol
TBHP	*tert*-Butyl hydroperoxide
DNA	Deoxyribonucleic acid
DHP	Dihydropyran

3) Spectroscopic Abbreviations

IR	Infrared
υ_{max}	Frequency maximum
cm^{-1}	Frequency in wavenumber
UV	Ultra violet
NMR	Nuclear magnetic resonance
$CDCl_3$	Deuterated chloroform
DMSO-d_6	Deuterated dimethyl sulfoxide
DEPT	Distortionless Enhancement by Polarization Transfer
ppm	Parts per million
δ	Delta (Chemical shift in ppm)
MHz	Megahertz
Hz	Hertz
J	Coupling constant
br s	Broad singlet
s	Singlet
d	Doublet
t	Triplet
q	Quartet
m	Multiplet

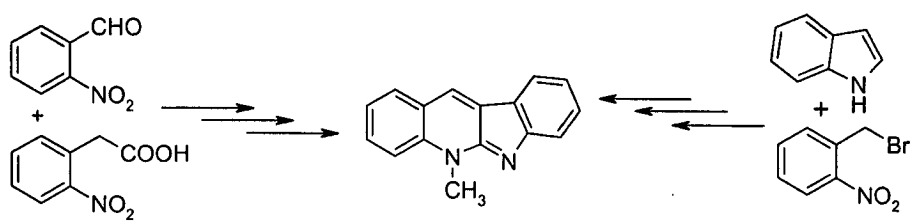
dd	Doublet of doublet
HRMS	High Resolution Mass Spectrum
M^+	Molecular ion
m/z	Mass to charge ratio

ABSTRACT OF THE THESIS

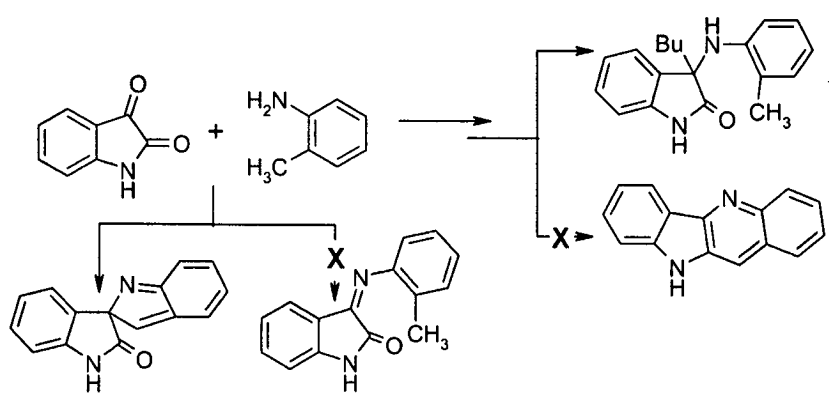
The thesis entitled "Synthetic Studies Towards Bioactive Natural Products and Their Analogues" is divided into three chapters which describes our efforts towards the development of simple methods for the synthesis of various indoloquinoline and bis-indole alkaloids and further to evaluate the biological activities of some of these compounds.

Chapter 1 deals with the synthetic studies of cryptolepine, isocryptolepine and neocryptolepine which are all tetracyclic heteroaromatic compounds containing indoloquinoline framework and isolated from the roots of West African plant *Cryptolepis sanguinolenta*. These alkaloids, due to their wide spectrum of biological activities, have been targets of synthetic chemists in recent years.

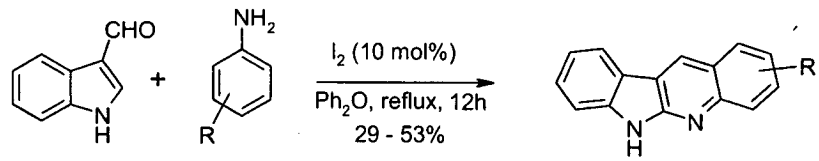
Towards the synthesis of neocryptolepine (cryptotackieine), we have developed two distinct approaches. The first approach involves the Perkin reaction and double reduction – double cyclization as the key steps while the second approach describes its synthesis *via* alkylation reaction and reductive cyclization.



However, we could not succeed in synthesizing cryptolepine and isocryptolepine under different reaction conditions attempted and the unexpected products obtained during the process were fully characterized using spectral data.



Chapter 2 describes the use of molecular iodine for the synthesis of various indoloquinolines and their biological activity evaluation. The first part deals with the synthesis of indoloquinolines using iodine as a catalyst. Recently the use of iodine receives considerable attention as an inexpensive, non-toxic, and easily available catalyst for different organic transformations and we have developed a novel one-pot method for the synthesis of indoloquinolines using 10 mol% iodine as a catalyst. The probable mechanism for its formation is also described.



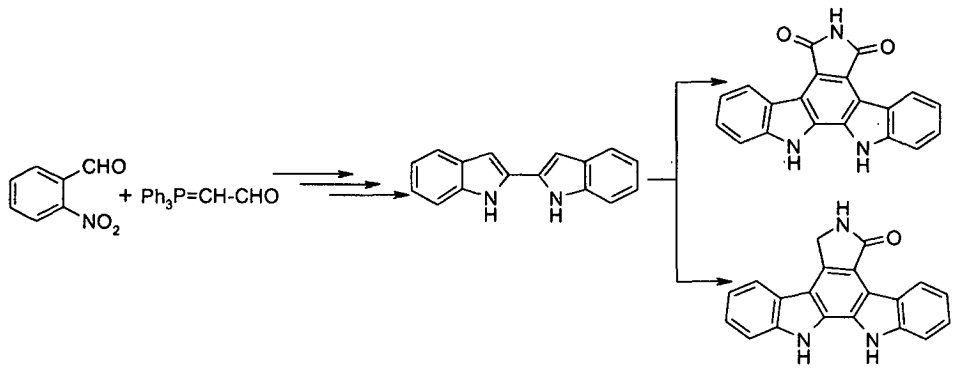
R = H, 2-CH_3, 3-CH_3, 4-CH_3, 3-Br, 2,3-benzo, 3,4-benzo

7 of 8 examples

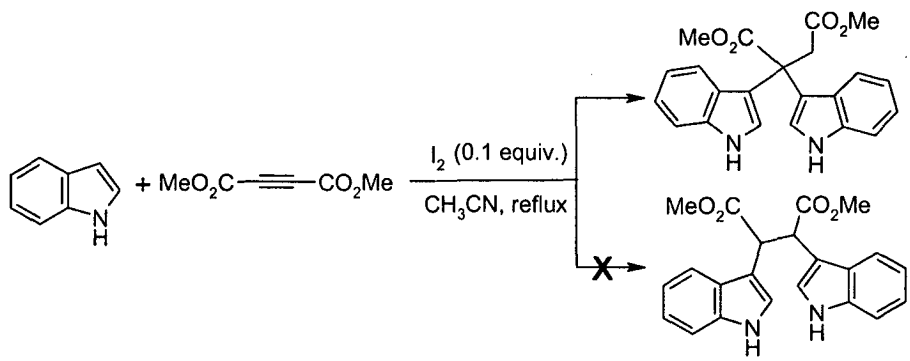
Second part involves the biological activity evaluation of some of these indoloquinolines. The *in vitro* antiproliferative activity i.e. cell growth inhibition activity of these compounds were tested against human hepatocellular carcinoma HepG2 and human breast carcinoma MCF-7 cell lines and the IC_{50} values were calculated by plotting the graph of concentration (mg/mL) against % cell survival.

Compd.	**HepG2 IC_{50} (mg/mL)**	**MCF-7 IC_{50} (mg/mL)**
6*H*-Indolo[2,3-*b*]quinoline	>1	>1
8*H*-Indolo[2,3-*b*]benzo[*h*]quinoline	0.0951	0.0717
4-Methyl-6*H*-Indolo[2,3-*b*]quinoline	0.0098	0.0059
3-Bromo-6*H*-Indolo[2,3-*b*]quinoline	0.0486	0.0369

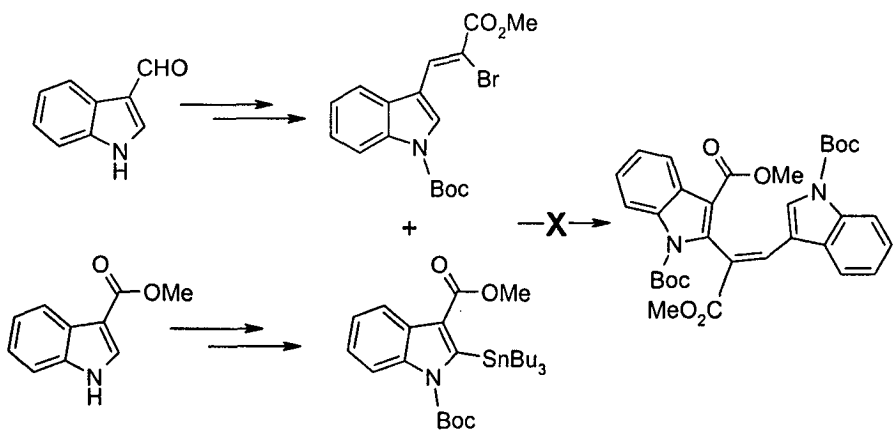
Chapter 3 describes our approach towards the synthesis of marine natural products acryriaflavin A, staurosporinone and caulersin. The first part involves the synthesis of 2,2'-biindole *via* consecutive Wittig reaction and double reductive cyclization which constutes the formal synthesis of naturally occurring indolocarbazole alkaloids – arcyriaflavin A and staurosporinone.



Next, we attempted the total synthesis of arcyriaflavin A but we ended up in getting unexpected bis-indole compounds which is found to be analogues to those of naturally occuring bis-indole alkaloids isolated from North Sea bacterium.



Second part deals with our efforts towards the synthesis of bis-indole marine natural product caulersin utilizing Wittig reaction and Stille coupling as the key steps.



LIST OF PUBLICATIONS

1) **Parvatkar, P. T.**; Parameswaran, P. S.; Tilve, S. G. Double reductive cyclization: a facile synthesis of the indoloquinoline alkaloid cryptotackieine. *Tetrahedron Lett.* **2007**, *48*, 7870-7872.

2) **Parvatkar, P. T.**; Parameswaran, P. S.; Tilve, S. G. An Epeditious I_2-catalyzed Entry into 6*H*-indolo[2,3-*b*]quinoline system of Cryptotackieine. *J. Org. Chem.* **2009**, *74*, 8369-8372.

3) **Parvatkar, P. T.**; Parameswaran, P. S.; Tilve, S. G. Isolation, Biological Activities and Synthesis of Indoloquinoline Alkaloids: Cryptolepine, Isocryptolepine and Neocryptolepine. *Curr. Org. Chem.* **2010** (Review Article Accepted).

4) **Parvatkar, P. T.**; Kadam, H. K.; Parameswaran, P. S.; Tilve, S. G. A facile Synthesis of 2,2'-Biindole: Formal Synthesis of Arcyriaflavin-A and Staurosporinone (K-252c). *Lett. Org. Chem.* **2010** (Communicated).

5) **Parvatkar, P. T.**; Parameswaran, P. S.; Tilve, S. G. Recent Developments in the Synthesis of Five- and Six-Membered Heterocycles Using Molecular Iodine. (Manuscript under preparation).

CONFERENCE PUBLICATIONS

Oral Presentation

1) Paper entitled "Synthesis of Cryptotackieine and 2,2'-Biindole" presented at Royal Society of Chemistry-West India Section **2007**, Goa University, Goa.

2) Paper entitled "Synthetic Studies in Bioactive Natural Products and their Analogues" presented at National Organic Symposium Trust, J-NOST **2009**, IIT, Kanpur.

Poster Presentation

1) Paper entitled "One-Pot Synthesis of 2,2-Biindole and its Analogues" presented at International Conference on Advances in Drug Discovery Research, **2007**, Aurangabad.

2) Paper entitled "Novel One-Pot Synthesis of Indoloquinolines" presented at Chemical Research Society of India, CRSI-11, **2009**, NCL, Pune.

3) Paper entitled "An Efficient Synthesis of Indoloquinoline Alkaloid – Neocryptolepine (Cryptotackieine) presented at Royal Society of Chemistry-West India Section **2010**, Goa University, Goa.

CHAPTER 1

SYNTHETIC STUDIES TOWARDS INDOLOQUINOLINE ALKALOIDS: CRYPTOLEPINE, ISOCRYPTOLEPINE AND NEOCRYPTOLEPINE

SYNTHETIC STUDIES TOWARDS INDOLOQUINOLINE ALKALOIDS: CRYPTOLEPINE, ISOCRYPTOLEPINE AND NEOCRYPTOLEPINE

Section A: Literature Review

1. Introduction

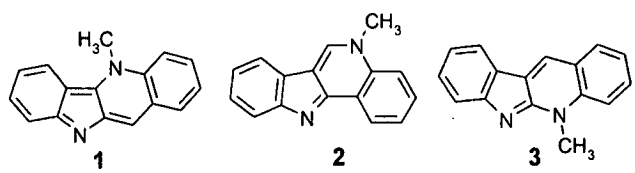
1.1. General

In recent years, indoloquinoline alkaloids have received considerable attention due to their promising DNA intercalating[1] and antimalarial properties.[2-4] According to World Health Organization (WHO), about 3.3 billion people are at risk of malaria. Every year, this leads to about 250 million malaria cases, causing nearly a million deaths, mostly of children under 5 years, justifying its classification as a dreaded infectious disease along with tuberculosis and AIDS.[5]

The roots of the West African plant *Cryptolepis sanguinolentine*[6-19] has long been used in folk medicine for the treatment of infectious diseases, amoebiasis, fever and malaria. Since 1974, a decoction of this plant is being used in the clinical therapy of rheumatism, urinary tract infections, malaria and other diseases.[20-23] Chemical examination indicated this plant to be a rich source of several indoloquinoline alkaloids.[6-19]

1.2. Isolation

So far 13 alkaloids including cryptolepine **1**, isocryptolepine **2** and neocryptolepine **3** have been reported from the roots of the West African plant *C. sanguinolenta* (Figure 1).



1 **2** **3**

Fig.1

Among these, cryptolepine **1** is a rare example of natural product whose synthesis was reported prior to its isolation from nature. It was synthesized in 1906 by Fichter and Boehringer[24] for possible use as a dye while its isolation from *C. triangularis* was

reported only in 1929.[25] Subsequently, in 1951, Gellert *et al.*[6] reported this compound from the roots of *C. sanguinolenta.*

In 1995, two research groups, *i.e.,* Pousset *et al.*[10] and Sharaf *et al.*[26] independently reported a related alkaloid **2** and named it as isocryptolepine and cryptosanguinolentine. Isocryptolepine **2** is an angularly-fused alkaloid with indolo[3,2-*c*]quinoline ring system whereas cryptolepine **1** is a linearly-fused alkaloid with indolo[3,2-*b*]quinoline ring system.

Subsequently in 1996, a new linearly-fused indolo[2,3-*b*]quinoline alkaloid **3** was reported by two independent research groups and named it as neocryptolepine by Pieter's group[9] and cryptotackieine by Schiff's group.[26]

Other alkaloids reported from the plant *C. sanguinolenta* include quindoline[7] **4**, cryptospirolepine[13] **5**, cryptolepicarboline[27] **6**, cryptomisrine[28] **7**, 11-isopropylcryptolepine[17] **8**, cryptolepinone[13-15] **9** and bis-cryptolepine[9] **10** (Figure 2).

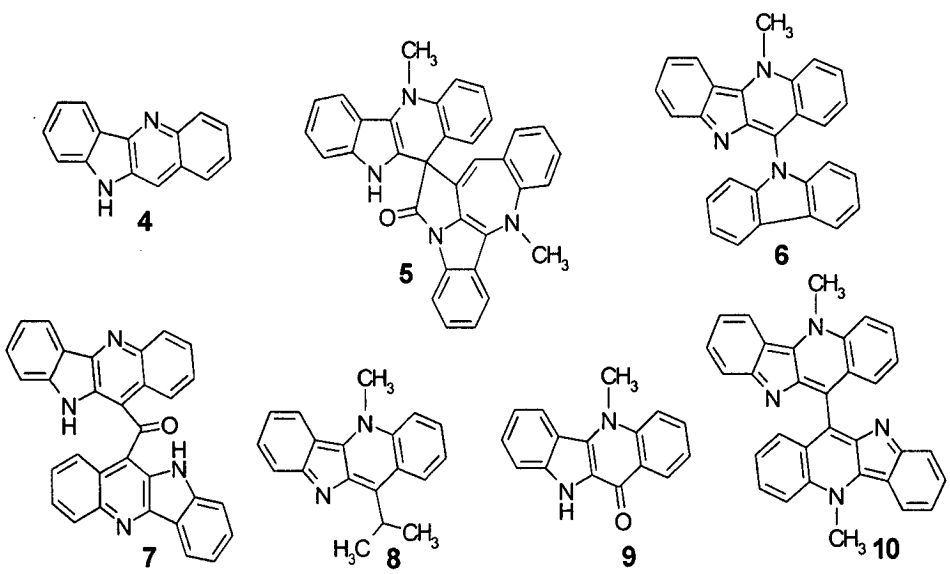


Fig.2

1.3 Brief Biological activities

The tetracyclic heteroaromatic compounds **1** and **3** are linearly fused indoloquinolines, while compound **2** has angularly-fused ring system. All the three compounds exhibit promising antiplasmodial activity[2 - 4, 29] against chloroquine-resistant *P. falciparum* and cryptolepine has been used as a lead compound for synthetic antiplasmodial agents. These alkaloids also intercalate with DNA double

helix, causing dramatic changes in DNA conformation leading to inhibition of DNA replication and transcription.[1] Cryptolepine binds 10-fold more tightly to DNA than other alkaloids and proves to be much more cytotoxic toward B16 melanoma cells. In addition, these compounds as well as some of their methyl derivatives have also shown promising antimuscarinic, antibacterial, antiviral, antimicotic, antihyperglycemic and cytotoxic properties *in vitro* and antitumor activity *in vivo*.[19,23,31-34]

These alkaloids, due to their wide spectrum of biological activities, have been targets of synthetic chemists in recent years.

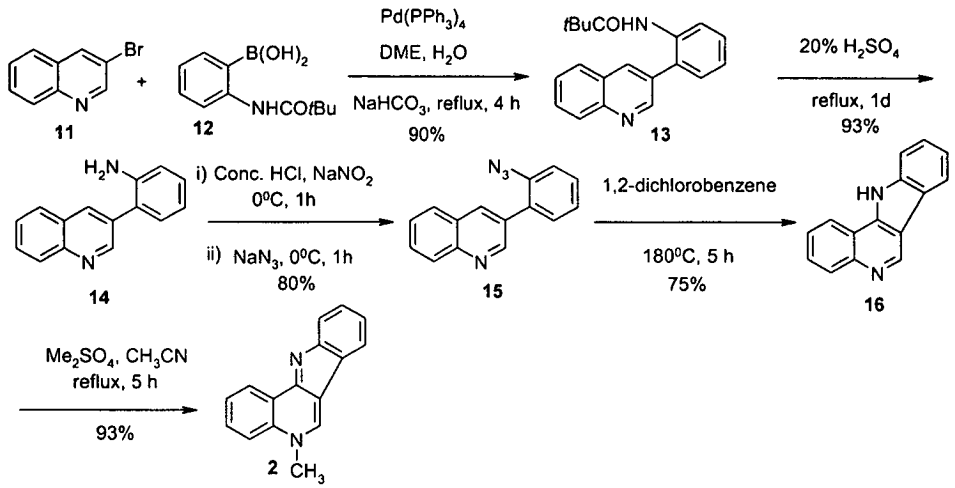
2. Synthesis

The synthetic methods used for the preparation of indoloquinoline alkaloids may be classified under the following six major categories based on the method of formation of the ring system – palladium-catalyzed coupling reaction, aza-Wittig reaction, transition-metal mediated reductive cyclization, photochemical reactions, Graebe-Ullmann reaction and other miscellaneous methods.

2.1. Palladium-catalyzed coupling reaction

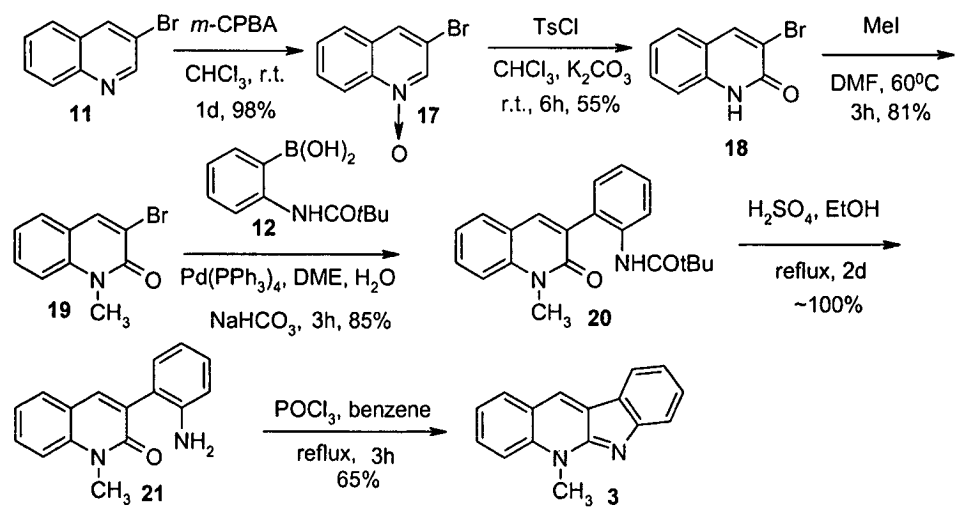
Pd-catalyzed coupling reactions[35] have become a powerful tool for the synthetic chemists particularly for the synthesis of biologically active natural products and for the preparation of versatile organic building blocks. Palladium catalyst possess a higher activity than other metal alternatives (Cu, Ni or Fe) enabling the conversion of less reactive substrates and performance at relatively low temperature.

Timari *et al.*[36] reported the synthesis of isocryptolepine and neocryptolepine using Suzuki procedure (Scheme 1 & 2).



Scheme 1

The reaction of 3-bromoquinoline **11** with *N*-pivaloylaminophenyl boronic acid **12** in presence of Pd(0) catalyst afforded the desired biaryl compound **13** which on hydrolysis with sulfuric acid gave amine **14**. The compound **14** was converted to azide **15** which on thermal cyclization gave exclusively the indolo[3,2-*c*]quinoline **16**. Regioselective methylation on quinoline nitrogen using dimethyl sulfate yielded the target molecule isocryptolepine **2** (Scheme 1).

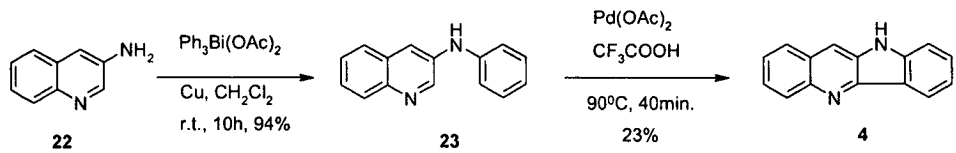


Scheme 2

3-Bromo-1*H*-2-quinoline **18** was prepared from 3-bromo-quinoline **11** *via* its *N*-oxide **17** which on treatment with methyl iodide gave *N*-methyl compound **19**. Coupling reaction of **19** with **12** in presence of Pd(0) catalyst afforded the biaryl compound **20**.

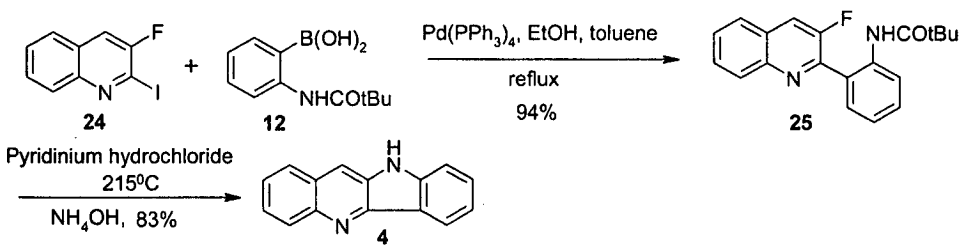
Hydrolysis of **20** with sulfuric acid followed by cyclization using $POCl_3$ furnished the neocryptolepine **3** (Scheme 2).

Fan and Ablordeppy[37] described the synthesis of 10*H*-indolo[3,2-*b*]quinoline **4** *via* *N*-arylation of 3-bromoquinoline **22** with triphenylbismuth diacetate using metallic copper followed by oxidative cyclization of the resultant anilinoquinoline **23** using palladium acetate (Scheme 3).



Scheme 3

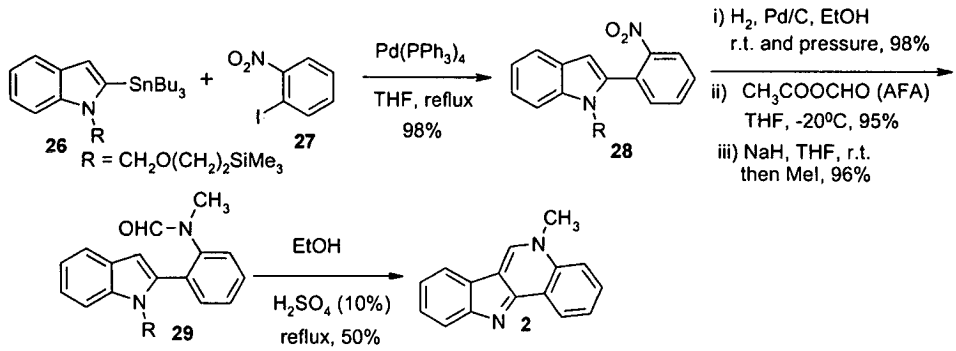
Arzel *et al.*[38] described the first halogen-dance reaction[39] in quinoline series and its application to a synthesis of quindoline (Scheme 4).



Scheme 4

Pd-catalyzed cross-coupling reaction between boronic acid **12** and 3-fluoro-2-iodoquinoline **24** using Suzuki procedure[40-41] afforded the biaryl compound **25** which underwent cyclization on treatment with boiling pyridinium chloride to give quindoline **4**.

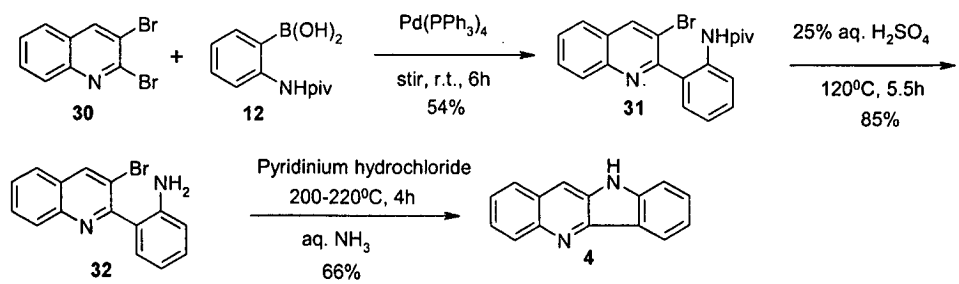
Murray *et al.*[42] achieved the synthesis of isocryptolepine as depicted in scheme 5.



Scheme 5

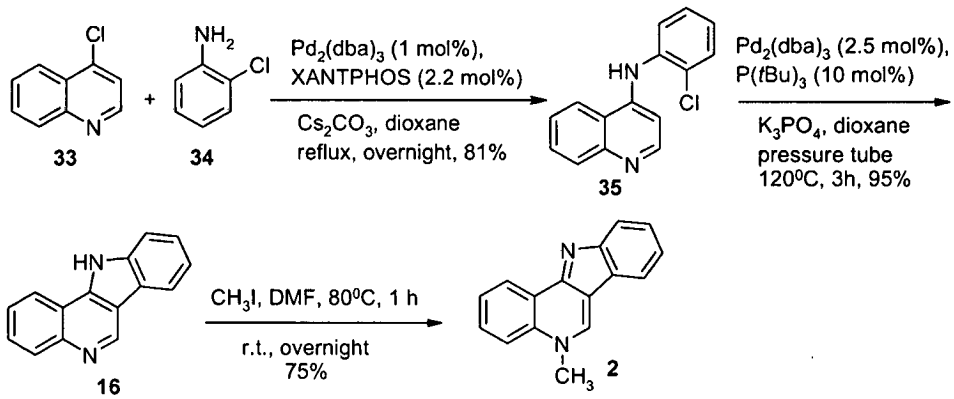
Pd(0)-catalyzed Stille coupling reaction of 2-tributylstannyl-*N*-protected indole **26** with 2-iodonitrobenzene **27** gave 2-(*o*-nitrophenyl)indole **28** which on reduction, *N*-formylation and *N*-methylation afforded the desired formamide **29**. Final ring closure was achieved by refluxing compound **29** in ethanol in presence of sulfuric acid to give isocryptolepine **2**.

Csanyi *et al.*[43] accomplished the synthesis of quindoline **4** by a regioselective coupling reaction of 2,3-dibromoquinoline[44] **30** with **12** taking into consideration the fact that the α-heteroaryl halogen atom is more reactive than the β-halogen atom[45] to give *N*-pivaloyl-2-(2'-anilino)-3-bromoquinoline **31**. Hydrolysis of **31** afforded the free amine **32** which underwent cyclization when heated at 200-220^{0}C in presence of pyridinium hydrochloride to give quindoline **4** (Scheme 6).



Scheme 6

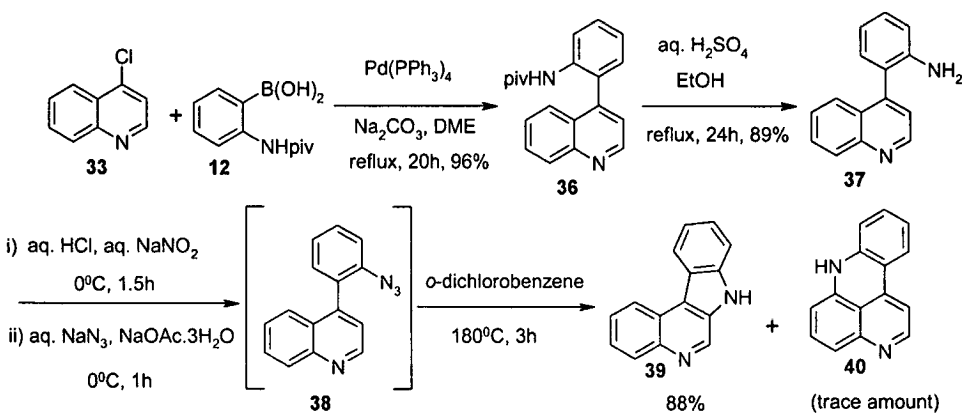
Jonckers *et al*[46] described the Pd-catalyzed 'amination-arylation' approach for the synthesis of isocryptolepine (Scheme 7).



Scheme 7

This approach consists of two consecutive Pd-catalyzed reactions – a selective Buchwald-Hartwig[47-48] reaction of 2-chloroquinoline **34** with 4-chloroquinoline **33** followed by an intramolecular arylation[49-51] of the resulting compound **35** to afford the 11*H*-indolo[3,2-*c*]quinoline **16**.

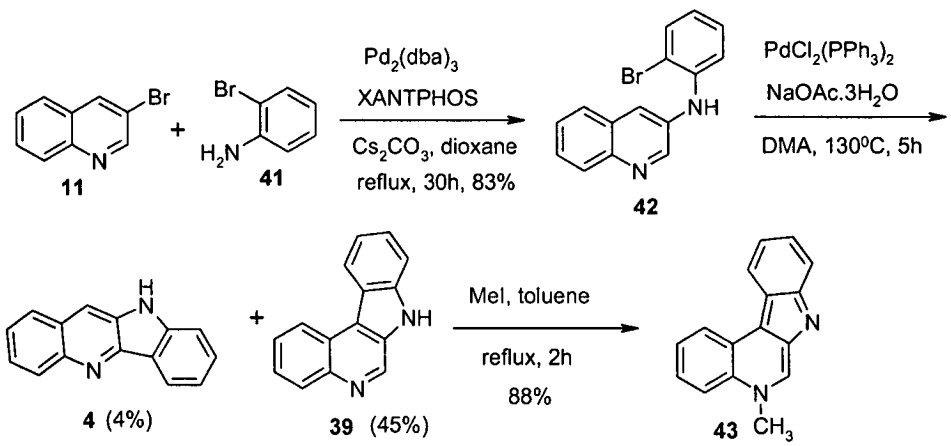
Hostyl *et al.*[52] reported the synthesis of isoneocryptolepine, a missing indoloquinoline isomer in the alkaloid series cryptolepine, neocryptolepine and isocryptolepine *via* two routes – 1) Suzuki arylation with an intramolecular nitrene insertion (Scheme 8) and 2) With a combination of a selective Buchwald-Hartwig-amination with an intramolecular Heck-type reaction (Scheme 9).



Scheme 8

Suzuki reaction of **33** with **12** under Gronowitz conditions[53-54] yielded compound **36** which on hydrolysis provided amine **37**. Diazotization of the resulting amine **37** followed by introduction of azido group and then thermal decomposition of azide **38**

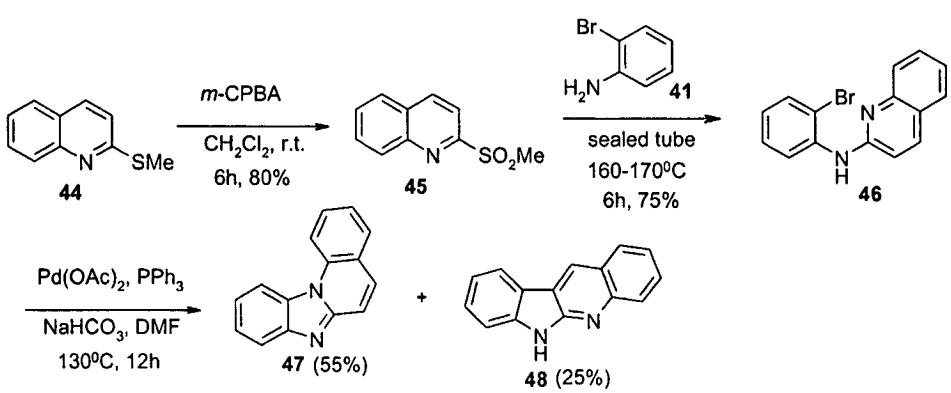
in boiling *o*-dichlorobenzene yielded the target molecule **39** as the major product and **40** in trace amount (Scheme 8).



Scheme 9

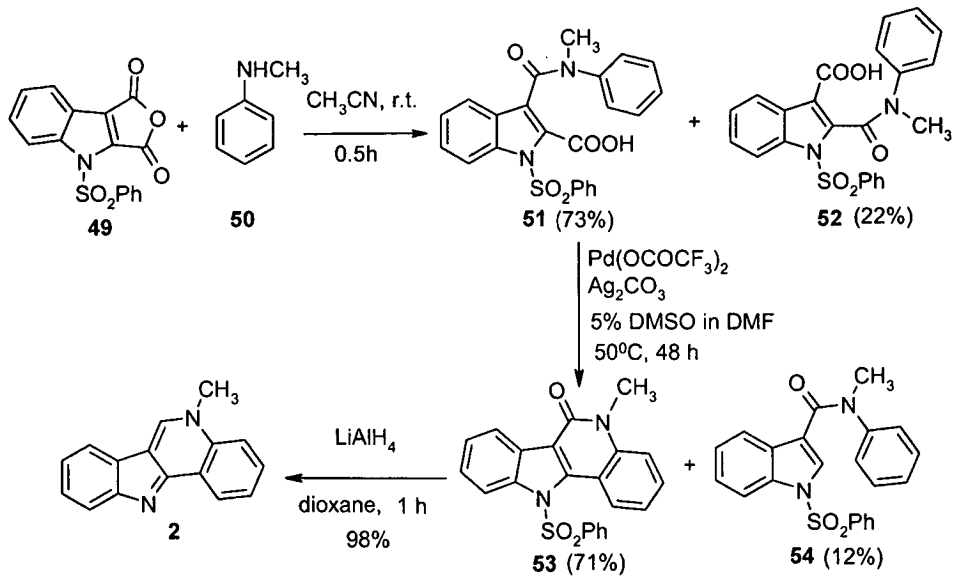
Regioselective amination of **11** with **41** in presence of Pd(0) catalyst gave compound **42** which on Heck-type cyclization yielded predominantly 7*H*-indolo[2,3-*c*]quinoline **39** and small amount of quindoline **4**. Selective *N*-methylation[55] of **39** using methyl iodide in refluxing toluene afforded the isoneocryptolepine **43** (Scheme 9).

Venkatesh *et al.*[56] reported the synthesis of benzimidazo[1,2-*a*]quinoline **47** *via* Pd-catalyzed intramolecular heterocyclization of 2-(2-bromoanilino)quinoline **46** in which 6*H*-indolo[2,3-*b*]quinoline **48** (precursor to neocryptolepine) was formed as a minor product (Scheme 10).



Scheme 10

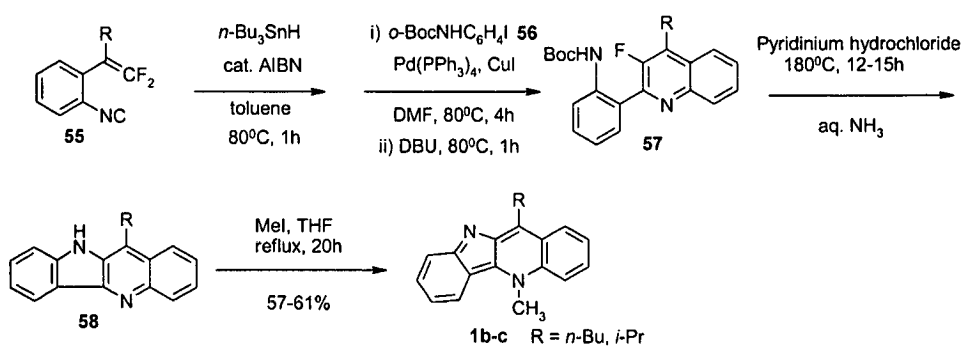
Miki and co-workers[57] have developed a simple approach towards isocryptolepine by applying Mayer's method[58-60] (Scheme 11).



Scheme 11

Reaction of **49** with *N*-methyl aniline **50** in acetonitrile afforded a mixture of acids **51** and **52** respectively. The decarboxylative Heck-type cyclization of **51** was achieved using $Pd(OCOCF_3)_2$ and Ag_2CO_3 to give the required compound **53** in 71% yield and decarboxylation product **54** in 22% yield. The compound **53** was converted to **2** by treatment with $LiAlH_4$ in hot dioxane.

Mori and Ichikawa[61] reported the synthesis of 11-alkylated cryptolepines via radical cyclization and Stille coupling reaction (Scheme 12).



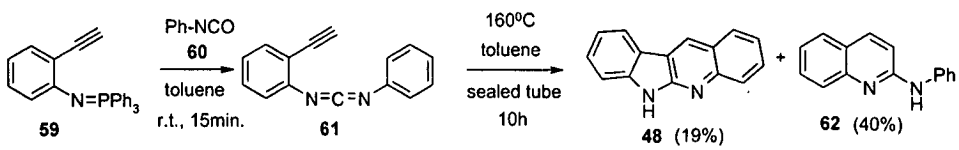
Scheme 12

o-Isocyano-substituted β,β-difluorostyrenes **55** on treatment with tributyltin hydride in presence of catalytic amount of AIBN and subsequent Pd-catalyzed coupling reaction with **56** afforded the 2,4-disustituted-3-fluoroquinolines **57** which on cyclization followed by methylation furnished the 11-butyl and 11-isopropyl cryptolepines **1b-c**.

2.2. Aza-Wittig reaction

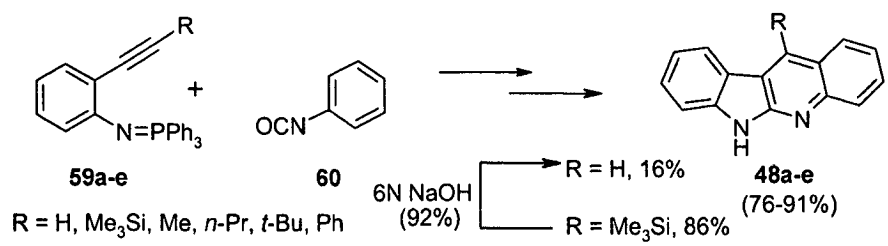
Aza-Wittig reaction[62] has become one of the important reactions in organic synthetic strategies directed towards the construction of acyclic and cyclic compounds as the reaction is mostly carried out in neutral conditions, in the absence of catalyst, generally at mild temperature and usually proceeds in high yield.

Alajarin and co-workers[63] described the synthesis of neocryptolepine using aza-Wittig reaction of the iminophosphorane **59** with phenyl isocyanate **60** to yield carbodiimide **61** and triphenylphosphine oxide which without purification was subjected to thermal treatment to give **46** and 2-anilinoquinoline **62** in 19% and 40% yield respectively (Scheme 13).



Scheme 13

Shi *et al.*[64] prepared various derivatives of 6*H*-indolo[2,3-*b*]quinoline **48** using the above methodology[63] (Scheme 14).



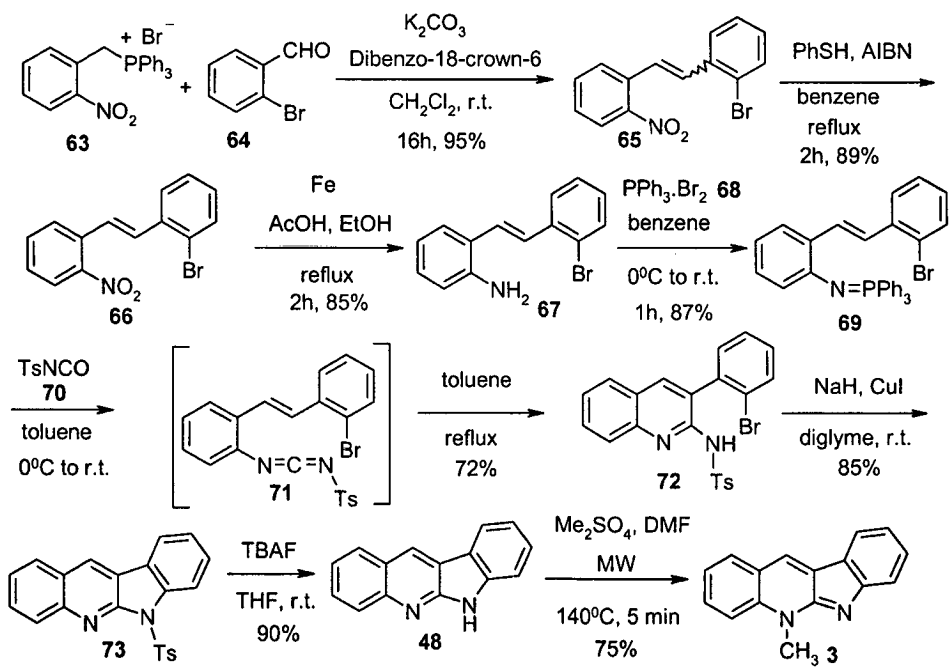
Scheme 14

The introduction of trimethylsilyl group at the acetylenic terminus provided an efficient route to **48** by suppressing the competing pathway toward the 2-anilinoquinoline **62** as the trimethylsilyl group serve as a surrogate for the hydrogen

atom in directing the reaction toward the indoloquinoline. A subsequent protodesilylation using NaOH furnishes **48** in good yield. Simallarly the derivatives of **48** with substitutents at C-11 position are prepared by treating the corresponding iminophosphoranes with phenyl isocynate.

Using the methodology of Alajarin *et al.*[63], Jonckers and co-workers[65] also prepared various cryptolepines with substituents on A-ring or D-ring and were evaluated for their cytotoxicity, antiplasmodial and antitrypanosomal activities.

Molina and co-workers[66] reported the synthesis of neocryptolepine *via* Staudinger, aza-Wittig and electrocyclization reactions (Scheme 15).

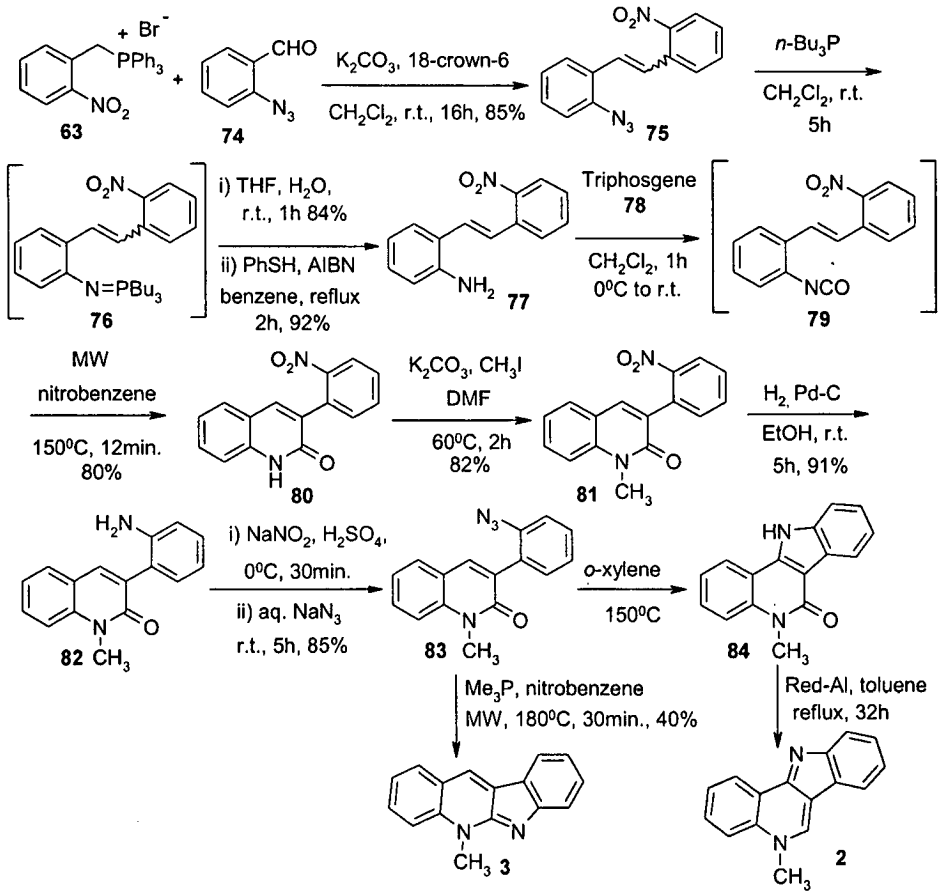


Scheme 15

The iminophosphorane **69** was prepared by condensing 2-nitrobenzyl triphenyl phosphonium bromide **63** with 2-nitrobenzaldehyde **64** in presence of K_2CO_3 followed by reduction of nitro group with iron and then treatment of resultant amino-stilbene derivative **67** with triphenylphosphine dibromide **68**. An aza-Wittig reaction of **69** with tosyl isocyanate **70** afforded the carbodiimide **71** which on heating underwent electrocyclic ring closure to give compound **72**. Treatment of **72** with NaH

in presence of CuI and subsequent detosylation using TBAF yielded **48**. Microwave-promoted methylation with DMS in DMF provided the target molecule **3**.

Fresneda and co-workers[67] devised a divergent synthetic approach to the alkaloids isocryptolepine and neocryptolepine which was based on the formation of key common intermediate 1-methyl-(*o*-azidophenyl)quinoline-2-one **83** (Scheme 16).



Scheme 16

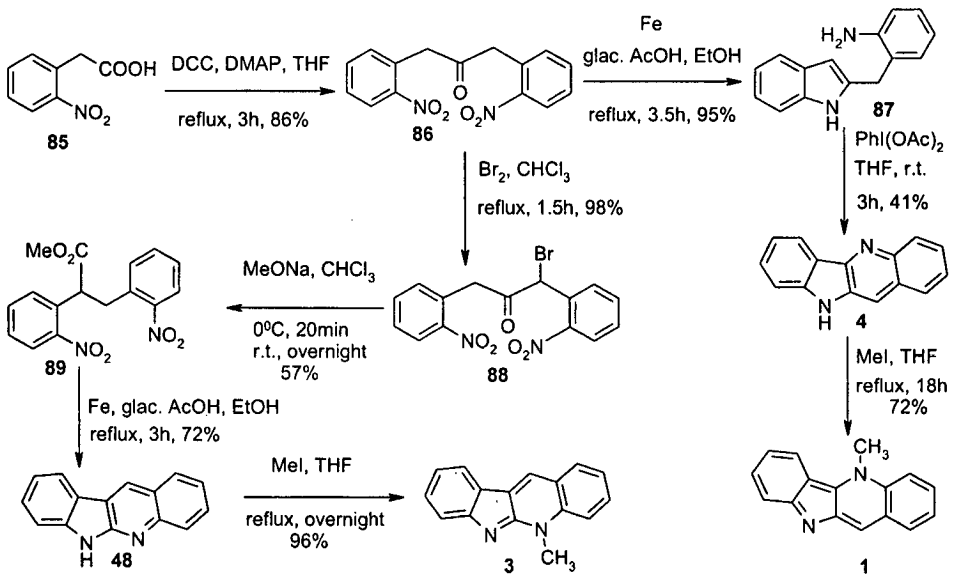
The key intermediate **83** was prepared using **63** and 2-azidobenzaldehyde **74** as the starting materials which underwent Wittig reaction in presence of K_2CO_3 to give compound **75**. Reaction of **75** with *n*-Bu_3P followed by hydrolysis of the resultant iminophosphorane **76** afforded amino-stilbene derivative **77** which on treatment with triphosgene **78** yielded the corresponding *o*-vinylsubstituted isocyanate **79**. Electrocyclic ring closure of **79** was achieved *via* microwave irradiation to give quinoline-2-one derivative **80** which was converted to **83** by four step sequence –

methylation, catalytic hydrogenation and diazotization followed by reaction with sodium azide. Selective indolization was achieved either by intramolecular aza-Wittig reaction of the iminophosphorane derived from **83** and PPh_3 under microwave irradiation to give neocryptolepine **3** or by nitrene-insertion process followed by reduction with Red-Al to give isocryptolepine **2**.

2.3. Trasition metal-mediated reductive cyclization

Reductive cyclization[68] using transition metal is an effective protocol for the synthesis of compounds containing quinoline ring and thus being used by several research groups for the synthesis of indoloquinolines.

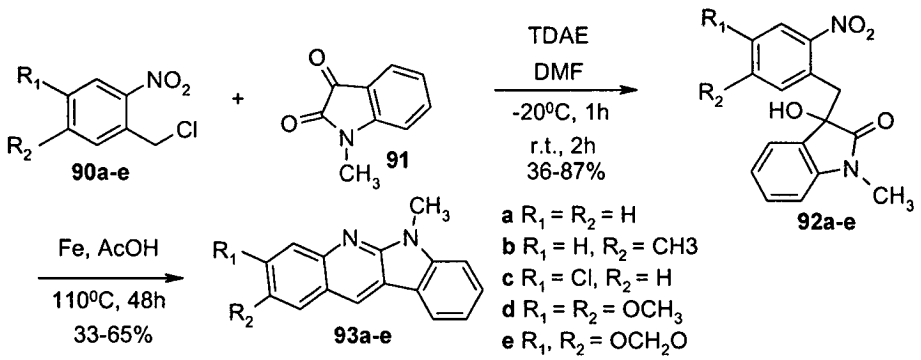
Ho and co-workers[69] reported the synthesis of cryptolepine and neocryptolepine from common intermediate 1,3-bis-(2-nitrophenyl)propan-2-one **86** (Scheme 17).



Scheme 17

The key intermediate **86** was readily obtained from 2-nitrophenyl acetic acid **85** by reaction with DCC in presence of DMAP. The approach to **1** involved the reduction of nitro groups with Fe powder followed by oxidative cyclization and subsequent *N*-methylation[37] while **3** was obtained *via* bromination, Favorskii rearrangement of the resultant bromo compound **88** followed by reduction-cyclization using Fe powder and finally *N*-methylation using the reported method.[37]

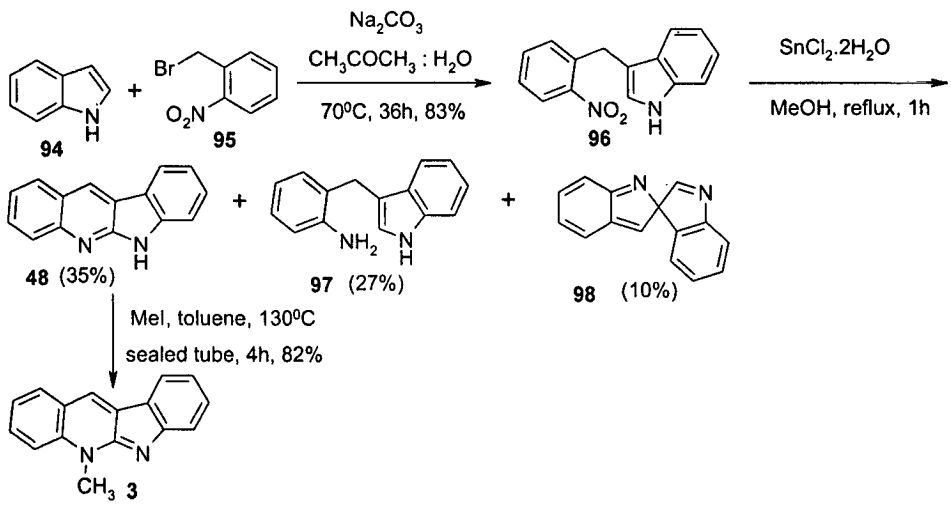
Amiri-Attou *et al.*[70] described the synthesis of analogues of neocryptolepine *via* one-pot reduction-cyclization-dehydration reaction (Scheme 18).



Scheme 18

Reaction of *o*-nitrobenzyl chlorides **90a-e** with 1-methyl isatin **91** in presence of tetrakis(dimethyl-amino)ethylene (TDAE)[71-72] afforded the corresponding α-hydroxy lactams **92a-e** which on treatment with iron underwent reduction-cyclization and dehydration in one-pot to give the respective 6-methyl-6*H*-indolo[2,3-*b*]quinolines **93a-e.**

Sharma and Kundu[73] achieved the synthesis of neocryptolepine using indole **94** and 2- nitrobenzyl bromide **95** as the starting materials (Scheme 19).



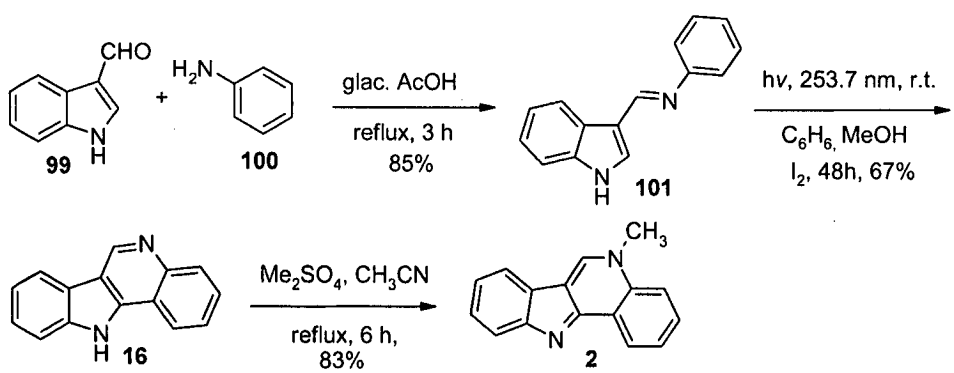
Scheme 19

Alkylation of indole with 2-nitrobenzyl bromide **95** yielded compound **96** which on treatment with $SnCl_2.2H_2O$ afforded **48** in 35% yield along with other two compounds **97** and **98** in 27% and 10% respectively.

2.4. Photochemical reactions

Photochemical reactions[74] are valuable in organic chemistry as they proceed differently than thermal reactions and have the advantage of forming thermodynamically disfavored products by overcoming large activation barriers and allow reactivity otherwise inaccessible by thermal methods. Photochemical substrate activation often occurs without additional reagents which prevents the formation of any by products and thus become important in the context of green chemistry.

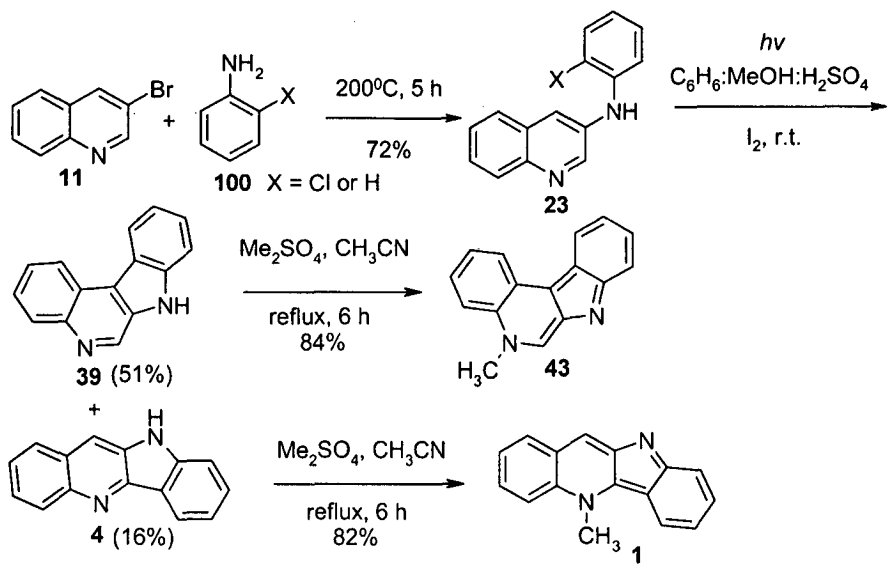
Kumar *et al.*[75] described the synthesis of isocryptolepine using photo-cyclization as the main step (Scheme 20)



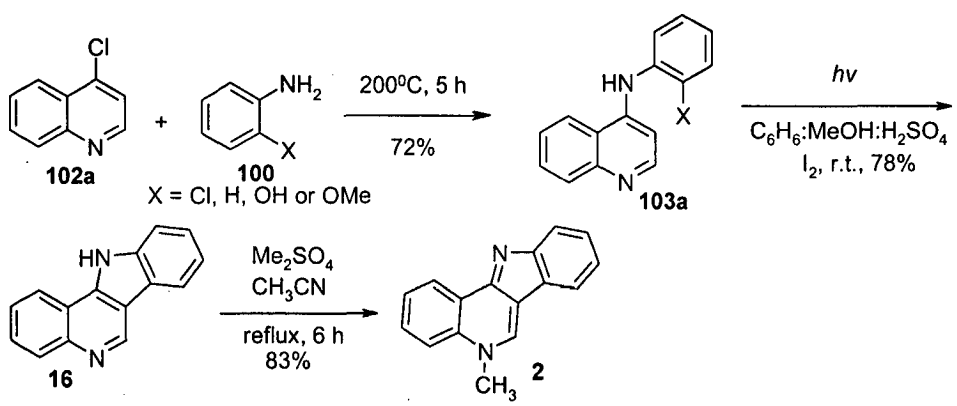
Scheme 20

Schiff's base **101** obtained by heating indole-3-carboxyaldehyde **99** with aniline **100** in acetic acid when irradiated at 253.7nm underwent cyclization to give 11*H*-indolo[3,2-*c*]quinoline **16**.

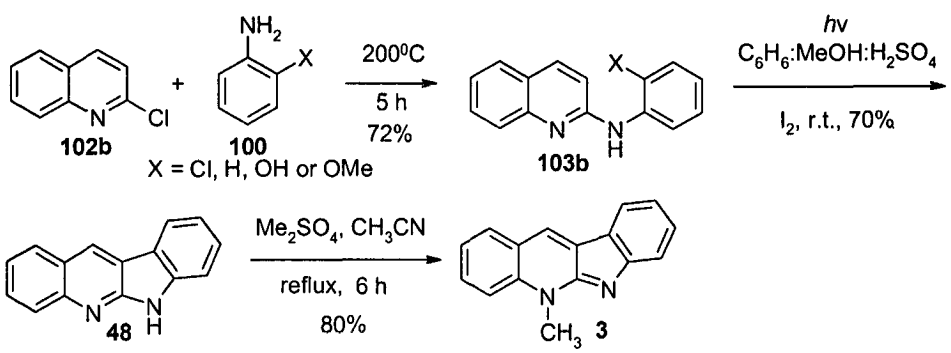
Dhanabal *et al.*[76] reported the synthesis of cryptolepine **1**, isocryptolepine **2** and neocryptolepine **3** *via* heteroatom directed photoannulation technique (Scheme 21-23).



Scheme 21



Scheme 22

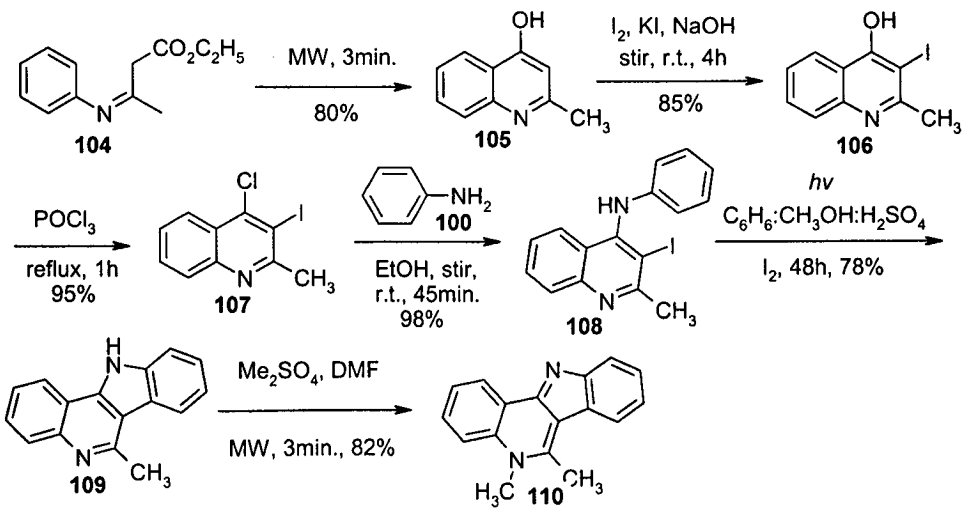


Scheme 23

Nucleophilic substitution of 3-bromoquinoline **11** with aniline **100** was achieved by heating at 200^{0}C and the resultant anilinoquinoline **23** was subjected to photochemical cyclization. Interestingly both linearly-fused and angularly-fused products **4** & **39** were obtained which on methylation gave cryptolepine **1** and isoneocryptolepine **43** respectively (Scheme 21).

Synthesis of isocryptolepine **2** and neocryptolepine **3** were obtained by photocyclization of respective anilinoquinolines **103a** and **103b** and subsequent methylation at quinoline nitrogen. Anilinoquinolines **103a-b** were obtained from corresponding chloroquinolines **102a-b** (Scheme 22 & 23).

Pitchai *et al.*[77] reported a simple photo-induced method for the synthesis of methyl derivative of isocryptolepine (Scheme 24).



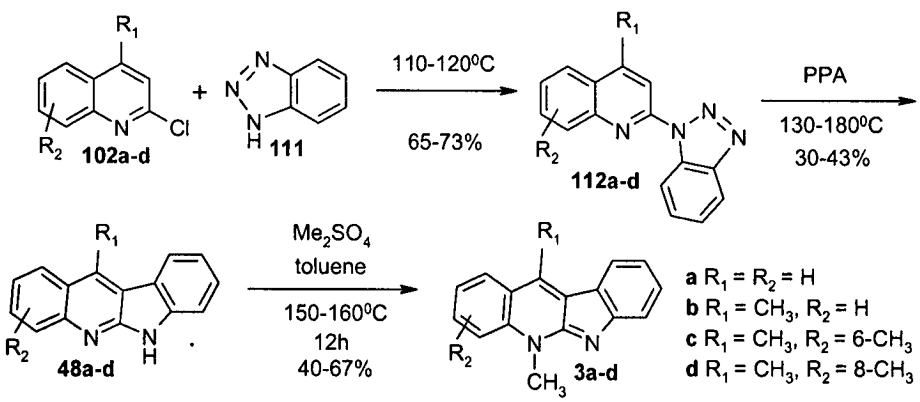
Scheme 24

4-Hydroxy-2-methyl quinoline **105** was prepared by microwave irradiation of β-anilinocrotonate **104** and then converted to 3-iodo-4-hydroxy-2-methylquinoline **106** using known procedure[78] which on treatment with $POCl_3$ afforded the corresponding chloro compound **107**. The amination reaction of **107** with aniline afforded the compound **108** which on photo irradiation and subsequent *N*-methylation yielded the methyl derivative of isocryptolepine.

2.5. Graebe-Ullmann reaction

Graebe-Ullmann reaction[79-80] has been widely used for the synthesis of carbazoles as the phenyl benzotriazoles formed in the reaction is unstable and readily undergo cyclization upon pyrolysis (catalyzed by acid) or on photolysis. Few research groups have exploited this reaction for the synthesis of indoloquinolines using haloquinolines instead of halopyridines as one of the starting material.

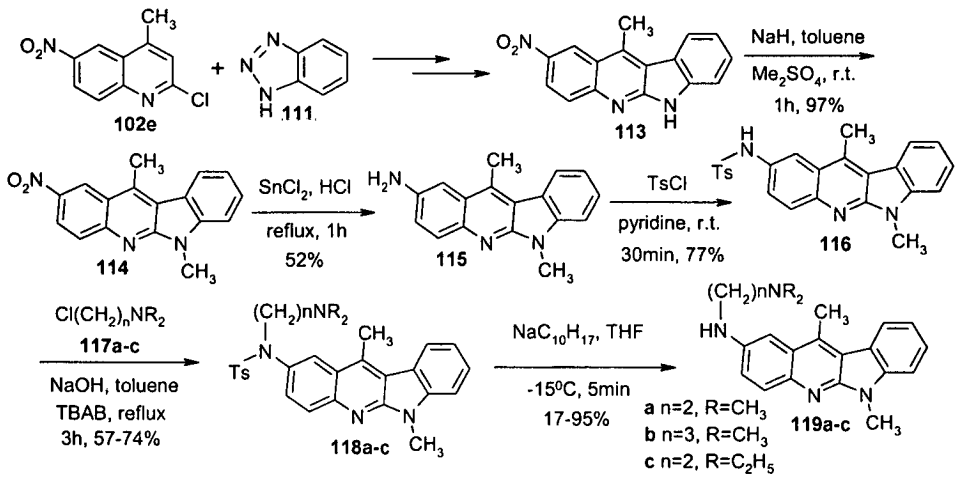
Peczynska-Czoch and co-workers[31] reported the synthesis of various derivatives of neocryptolepines *via* Graebe-Ullmann reaction (Scheme 25) and evaluated for their *in vitro* antimicrobial and cytotoxic activities.



Scheme 25

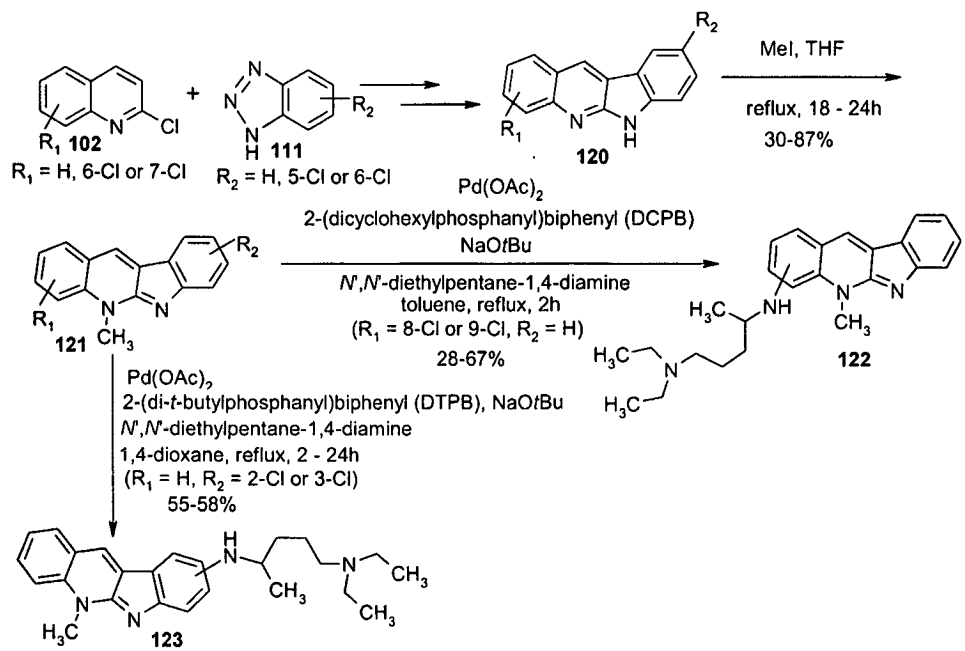
Triazoles **112a-d** were prepared by heating the corresponding chloroquinolines **102a-d** with benzotriazoles **111** at 110-120^{0}C. Decomposition of the triazoles **112a-d** by heating at 130-180^{0}C in presence of PPA yielded the respective indoloquinolines **48a-d** which on methylation using DMS afforded the neocryptolepines **3a-d**.

Godlewska *et al.*[81] reported the synthesis of nitro-substituted 6*H*-indolo[2,3-*b*]quinolines **113a-b** using the above methodology[31] and then indole nitrogen was methylated using NaH and DMS to give the respective analogues of neocryptolepines **114a-b**. The nitro group was reduced to corresponding amines using $SnCl_2$ which on treatment with *p*-toluenesulfonyl chloride afforded sulfonamides **116a-b**. Alkylation with (dialkylamino)alkyl chlorides and subsequent reaction with naphthylsodium yielded the respective amines **119** (Scheme 26).



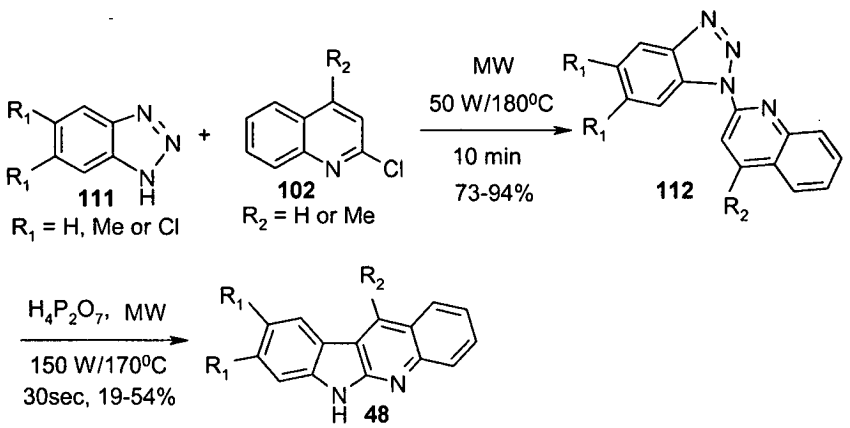
Scheme 26

Sayed *et al.*[82] described the synthesis of neocryptolepines with A or D-ring substitutions using the methodology of Peczynska-Czoch and co-workers[31] and the side chain was introduced on 2-, 3-, 8- & 9-positions using Pd-catalyzed amination reaction (Scheme 27). All these compounds were screened for *in vitro* antiplasmodial activity against a chloroquine-sensitive *P. falciparum* strain and for cytotoxicity on a human cell (MRC5) line.



Scheme 27

Vera-Luque *et al.*[83] achieved the synthesis of 6*H*-indolo[2,3-*b*]quinolines *via* modified Graebe-Ullmann reaction under microwave irradiation (Scheme 28).

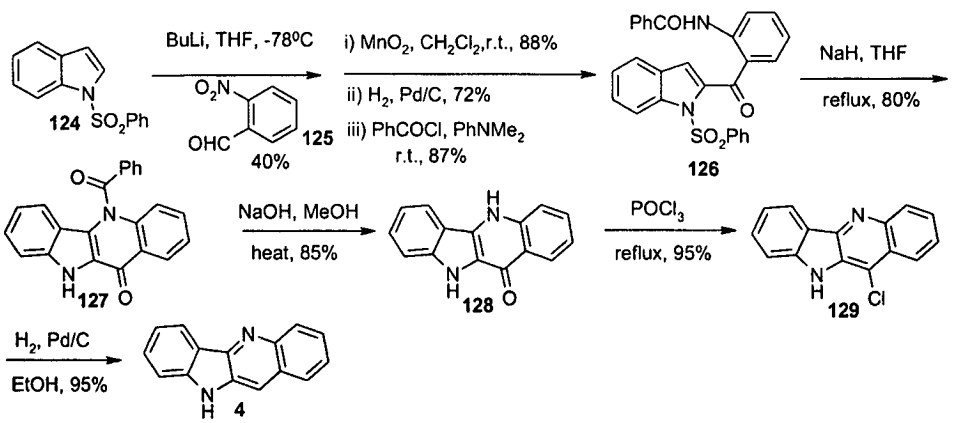


Scheme 28

Microwave irradiation of benzotriazoles **111** and 2-chloroquinoline **102** afforded the respective triazoles **112a-d**. The subsequent microwave irradiation of the resultant triazoles **112a-d** in presence of acid gave the respective 6*H*-indolo[2,3-*b*]quinolines **48a-d**.

2.6. Other miscellaneous methods

Cooper *et al.*[84] described the synthesis of quindoline utilizing the intramolecular β-nucleophilic substitution as the main step (Scheme 29).

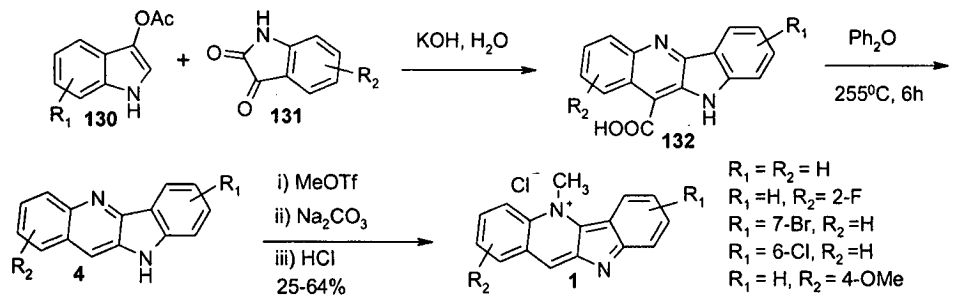


Scheme 29

Amido ketone **126** was prepared by directed lithiation of **124** followed by addition of **125**, subsequent oxidation of the resultant alcohol with MnO_2, reduction of nitro

group using catalytic hydrogenation and *N*-benzoylation using benzoylchloride. Cyclization of **126** was achieved using NaH which on deprotection using NaOH in MeOH and subsequent reaction with $POCl_3$ followed by catalytic hydrogenolysis of the resultant chloro compound **129** gave quindoline **4**.

Bierer and co-workers[23,85] reported the synthesis of cryptolepine and its analogues by utilizing the procedures of Holt and Petrow[86] and Deguitis and Ezyaskaite[87] (Scheme 30).

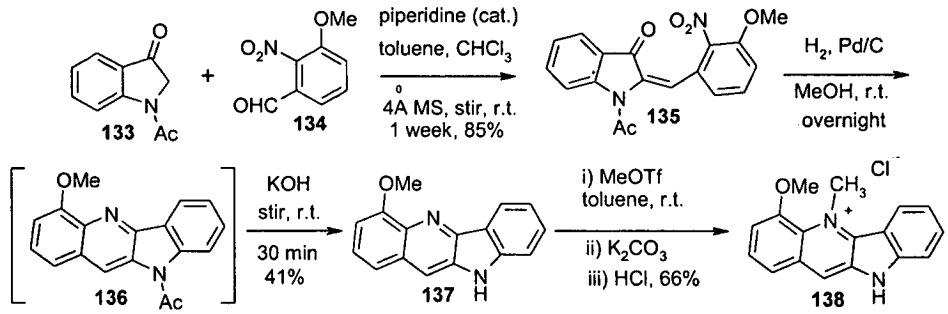


Scheme 30

Reaction of substituted indolyl acetates **130** with isatin derivatives **131** gave the respective quindoline carboxylic acids **132** which were decarboxylated by heating at 255^{0}C in Ph_2O and the subsequent quindolines **4** were alkylated using the method of Fichter and Boehringer[24] to give the respective cryptolepines **1**. All these compounds were evaluated for their antihyperglycemic activities *in vitro* and in an non-insulin-dependent diabetes mellitus (NIDDM) mouse model.

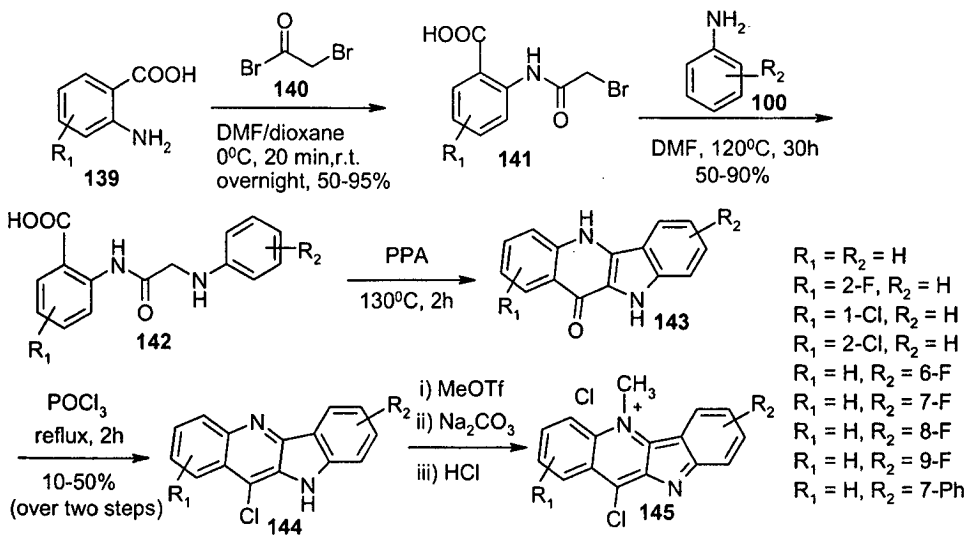
Several other research groups[88-90] have reported the synthesis of cryptolepine analogues using the above methodology[23,83] and were screened for their antimalarial and cytotoxic activities.

Bierer and co-workers[85] have reported the synthesis of 4-methoxy cryptolepine hydrochloride and a series of 11-chlorocryptolepine analogues as shown below (Scheme 31 & 32) and evaluated for their antimalarial and antihyperglycemic activities.



Scheme 31

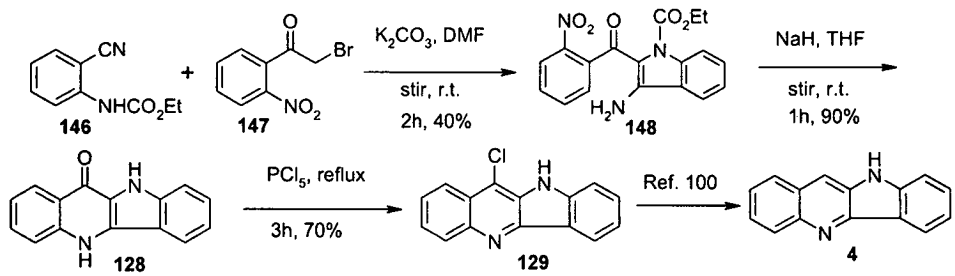
Condensation of **133** with **134** using catalytic amount of piperidine gave compound **135** as a mixture of *E/Z* isomers which on hydrogenation and subsequent deprotection using KOH followed by alkylation afforded the methoxy cryptolepine hydrochloride **138** (Scheme 31).



Scheme 32

Compound **141** formed by stiring anthranilic acids **139** and bromoacetyl bromide on treatment with substituted anilines **100** provided the anthranilic acid derivatives **142**. Acid-promoted cyclization of **142** with PPA gave quindolones **143** which when refluxed in $POCl_3$ afforded the corresponding 11-chloroquindolines **144**. *N*-methylation of **144** was achieved using methyl iodide to give the respective 11-chlorocryptolepines **145** (Scheme 32).

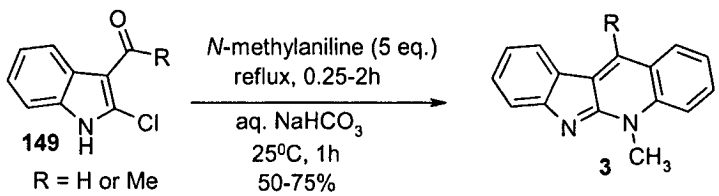
Radl and co-workers[91] reported the synthesis of quindoline **4** *via* intermediate **148** by treating anthranilonitrile **146** with phenacyl bromide **147** in presence of K_2CO_3 (Scheme 33).



Scheme 33

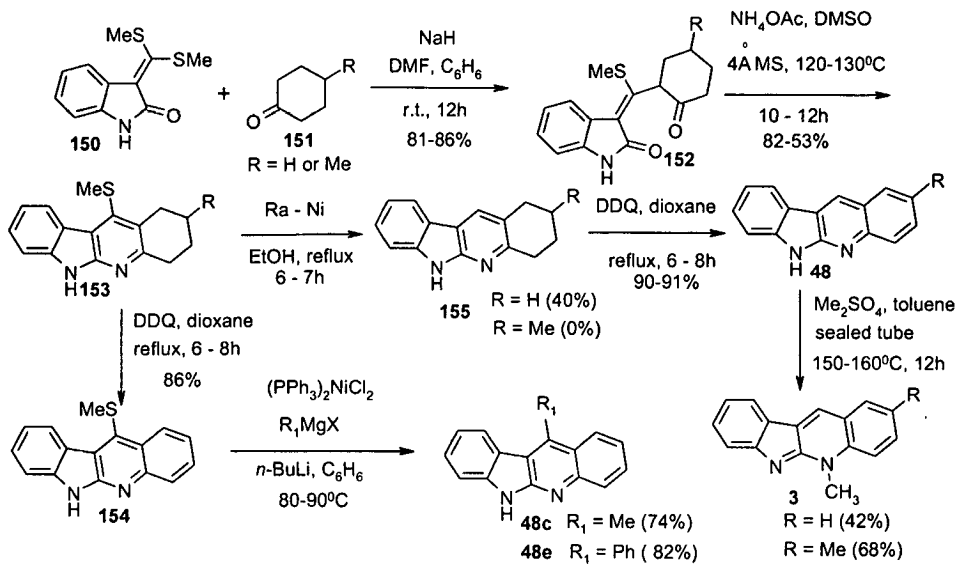
Nucleophilic cyclization of **148** with NaH gave the required tetracyclic compound **128** which on treatment with PCl_5 afforded the corresponding chloro compound **129** in 70% yield.

Engqvist and Bergman[92] achieved the synthesis of neocryptolepine by simply heating the chloroindole derivative **149** with excess *N*-methylaniline at reflux temperature (Scheme 34).



Scheme 34

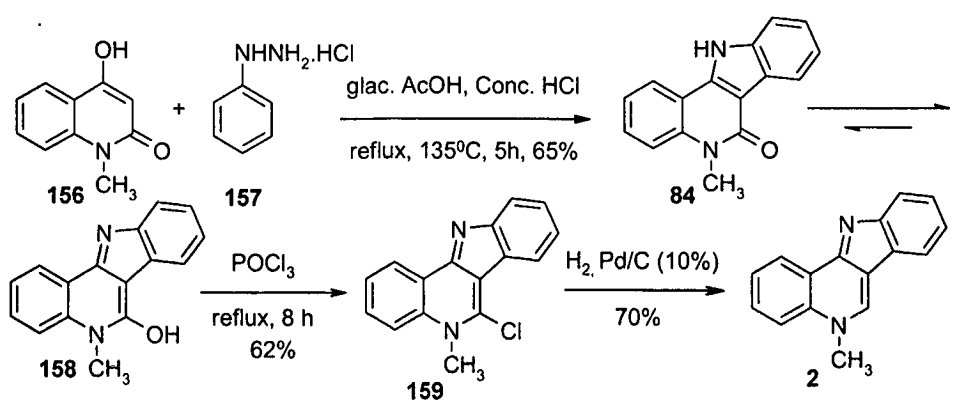
Sundaram *et al.*[93] reported the synthesis of 6*H*-indolo[2,3-*b*]quinoline **48** using conjugate addition and the heterocyclization as the main steps (Scheme 35).



Scheme 35

Reaction of **150** with cyclohexanones **151** in presence of NaH underwent conjugate addition to give the corresponding adduct **152** which on heterocyclization with ammonium acetate yielded compound **153**. Dethiomethylation of **153** with Ra-Ni and subsequent dehydrogenation with DDQ afforded the **48**. The 11-sustituted 6*H*-indolo[2,3-*b*]quinolines were prepared by treating compound **153** with DDQ and subsequent displacement of methylsulfanyl group with Grignard reagents in presence of bis(triphenylphosphino)nickel dichloride complex.

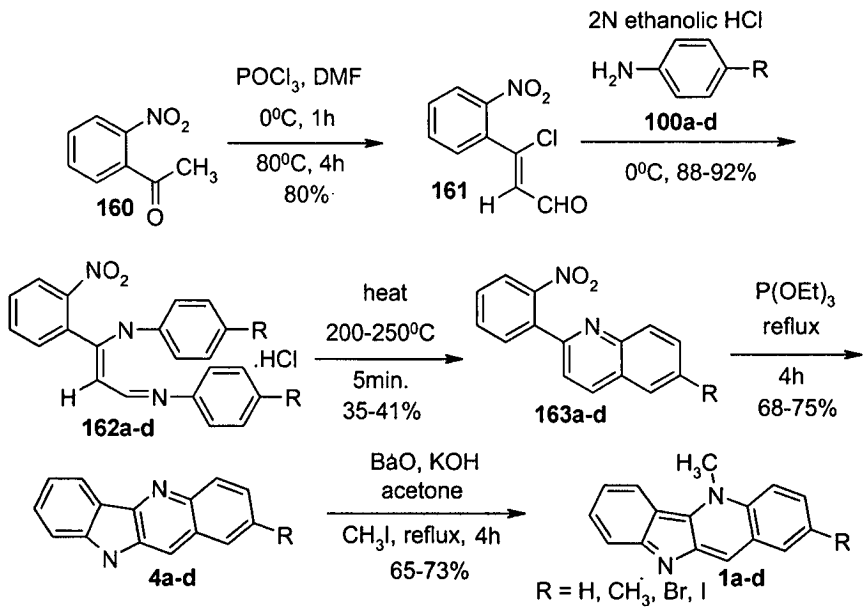
Dhanabal *et al.*[94] described the synthesis of isocryptolepine using a Fischer indole cyclization as the key step (Scheme 36).



Scheme 36

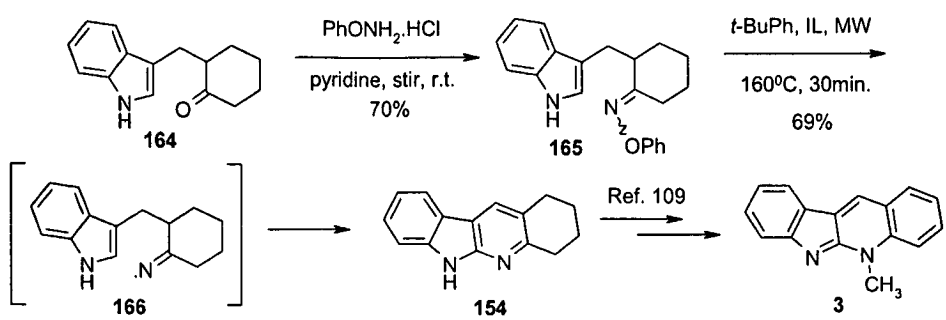
Fischer indole reaction of **156** with **157** gave the indoloquinoline **84** which exist predominantly in the hydroxy form **158** as confirmed by IR. The enol **158** when refluxed in $POCl_3$ afforded the corresponding chloride **159** which on catalytic hydrogenation yielded the isocryptolepine **2**.

Dutta *et al.*[95] developed a general method for the synthesis of various 2-substituted cryptolepines which involves regioselective thermal cyclization and reductive cyclization using triethyl phosphite as the key steps (Scheme 37).



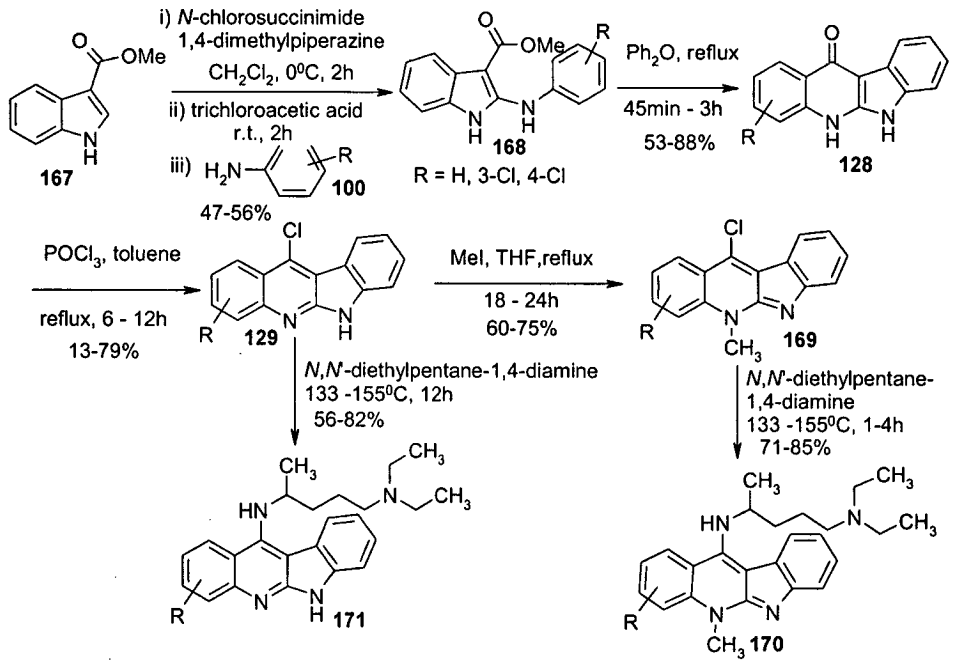
Scheme 37

Portela-Cubillo *et al.*[96] described the microwave-mediated formal synthesis of neocryptolepine *via* radical intermediate (Scheme 38).



Scheme 38

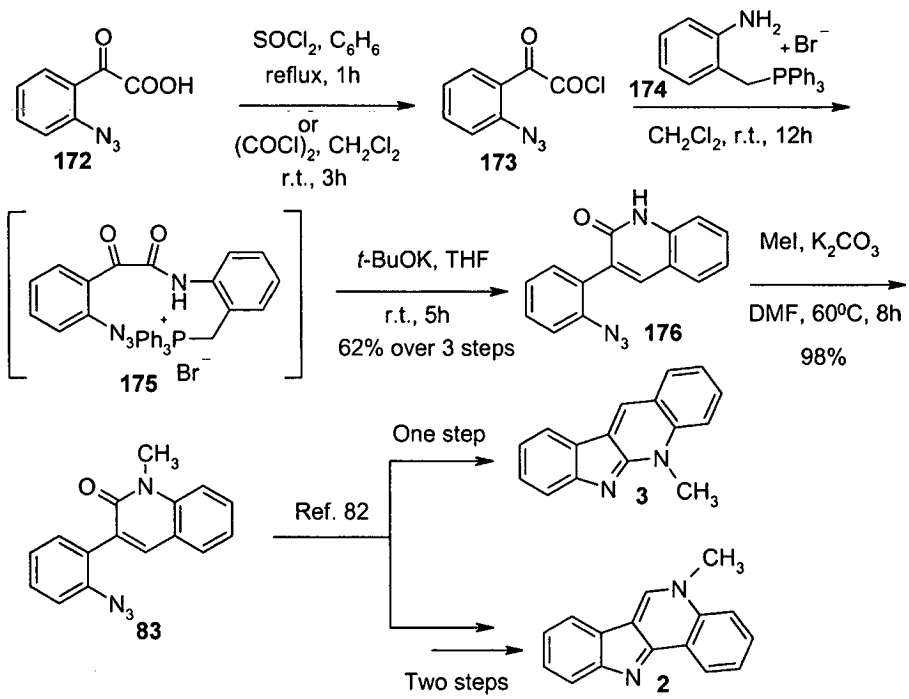
Sayed *et al.*[82] reported the synthesis of aminoalkylamino-substituted neocryptolepines using the procedure of Bergman and co-workers[97] (Scheme 39) and evaluated for their *in vitro* antiplasmodial activity against a chloroquine-sensitive *P. falciparum* strain and for cytotoxicity on a human cell line (MRC5).



Scheme 39

The key intermediate **168** was obtained *via* chlorination of **167** with NCS in presence of 1,4-dimethyl piparazine followed by addition of aniline which underwent cyclization when refluxed in Ph_2O to give compound **128** and then converted to 11-chloro-6*H*-indolo[2,3-*b*]quinolines **129** using $POCl_3$. Methylation using methyl iodide and subsequent amination *via* S_NAr reaction yielded the corresponding aminoalkylamino-substituted neocryptolepine derivatives.

Kraus and Guo[98] achieved a formal synthesis of neocryptolepine **3** and isocryptolepine **2** from a common intermediate **83** using an intramolecular Wittig reaction and regioselective methylation as the key steps (Scheme 40).



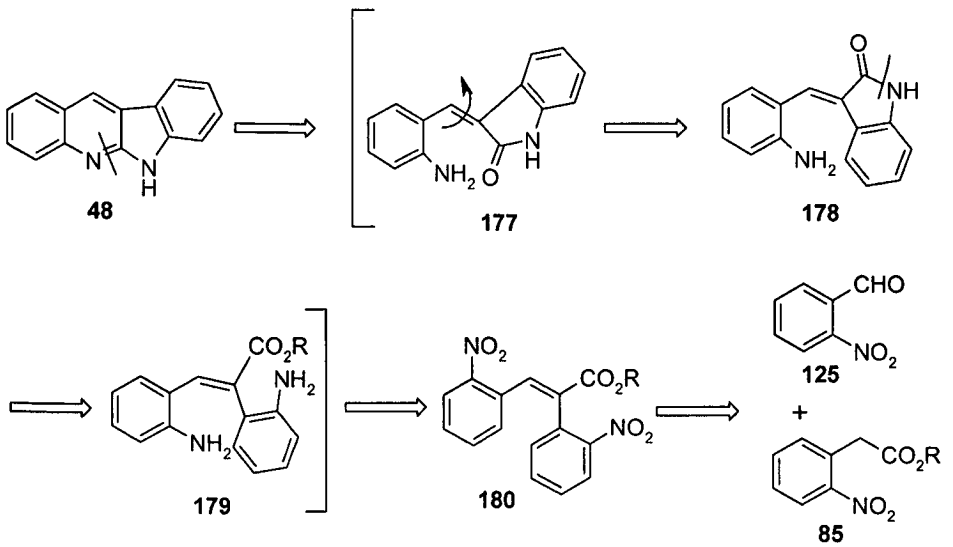
Scheme 40

The acid **173**, prepared from isatin[99] as converted to acid chloride **174** by two different methods, one using thionyl chloride and the other using oxalyl chloride. Condensation of 2-(aminobenzyl)triphenylphosphonium bromide with **174**, followed by intramolecular Wittig reaction in presence of potassium *tert*-butoxide at room temperature afforded lactam **177** in 62% overall yield from compound **173**. Methylation of **177** gave a known intermediate **83** which constitutes the formal synthesis of isocryptolepine **2** and neocryptolepine **3**, respectively.

Section B: Present Synthetic Work (Results and Discussion)

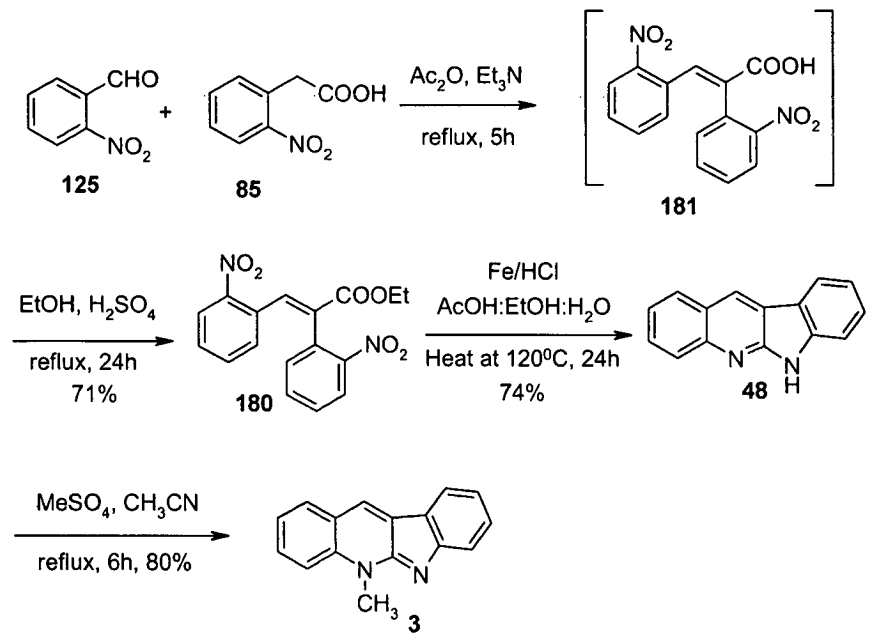
An interesting biological activities of indoloquinolines (as described in Section I) generated an interest in developing new synthetic pathways to these tetracyclic heteroaromatic compounds – neocryptolepine, isocryptolepine and cryptolepine.

Our retrosynthetic analysis of 6*H*-indolo[2,3-*b*]quinoline **48**, the immediate chemical precursor of the alkaloid neocryptolepine indicated that it should be possible for us to prepare **48** from α,β-unsaturated ester **180** *via* double reduction, isomerization and double cyclization reaction (Scheme 41). The key intermediate **180** could in turn be obtained from *o*- nitrobenzaldehyde and *o*-nitrophenylacetic acid using the Perkin reaction.



Scheme 41

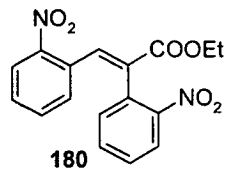
Accordingly, we started the synthesis from commercially available *o*-nitrobenzaldehyde and *o*-nitrophenylacetic acid as depicted below (Scheme 42).



Scheme 42

Condensation of *o*-nitrobenzaldehyde and *o*-nitrophenylacetic acid in refluxing acetic anhydride (Ac_2O) in presence of triethyl amine (NEt_3) yielded the corresponding α,β-unsaturated acid **181** which was extracted with aqueous Na_2CO_3 and then acidified with 6N HCl. The compound **181** without characterization was esterified using ethanol in presence of catalytical amount of sulfuric acid to give the key intermediate i.e. α,β-unsaturated ester **180** as a crystalline white solid in 71% overall yield. In its IR spectrum, the strong peak at 1701 cm^{-1} indicated the presence of conjugated ester carbonyl group while the peaks at 1611, 1524 and 1325 cm^{-1} indicated the presence of C-C double bond and nitro group respectively. The PMR spectrum showed six different signals between δ 1.28 – 8.22 ppm. The triplet at δ 1.28 integrating for three protons was assigned to methyl group while the quartet at δ 4.29 integrating for two protons was attributed to $-OCH_2-$ group. The peaks between 7.02 – 8.16 were attributed to aromatic protons and the singlet at δ 8.22 integrating for one proton was assigned to vinylic proton. The structure was further confirmed by ^{13}C NMR, DEPT experiment and HRMS. The detailed spectroscopic data is described below.

Spectroscopic data:



180

IR (KBr): υ_{max} = 1701, 1611, 1570, 1524, 1325, 751 cm^{-1}.

^{1}H NMR ($CDCl_3$, 300 MHz): (Fig. 3)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
1.28	t	7.2	3	-$OCH_2C\underline{H}_3$
4.29	q	7.2	2	-$OC\underline{H}_2$-
7.02-7.08	m	-	2	Ar-<u>H</u>
7.28-7.41	m	-	3	Ar-<u>H</u>
8.12-8.16	m	-	3	Ar-<u>H</u>
8.22	s	-	1	=C<u>H</u>-

^{13}C NMR ($CDCl_3$, 75 MHz): (Fig. 4)

δ 14.0 (-$OCH_2\underline{C}H_3$), 61.8 (-$O\underline{C}H_2$-), 124.6 (Ar-<u>C</u>H), 124.7 (Ar-<u>C</u>H), 129.3 (Ar-<u>C</u>H), 129.4 (Ar-<u>C</u>H), 131.0 (Ar-<u>C</u>H), 131.6 (Ar-<u>C</u>H), 131.7 (Ar-<u>C</u>H), 133.0 (Ar-<u>C</u>H), 133.1 (Ar-<u>C</u>H), 133.5 (Ar-<u>C</u>H), 133.6 (=<u>C</u>-CO_2Et), 137.2 (=<u>C</u>H-), 147.9 (Ar-<u>C</u>-NO_2), 149.3 (Ar-<u>C</u>-NO_2), 164.7 (-<u>C</u>=O).

HRMS: *m/z* $[M+Na]^+$ 365.0746 (calcd for $C_{17}H_{14}N_2O_6$, 365.0750).

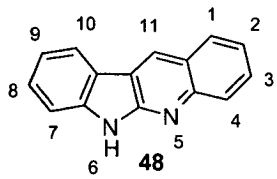
Melting Point: 124^{0}C.

Thus based on the mode of formation and spectral data, the structure **3** was assigned to it.

The next task was to prepare 6*H*-indolo[2,3-*b*]quinoline **48** using reduction and cyclization protocol as suggested in the retro-synthesis (Scheme 41). So, the compound **3** was treated with iron in presence of catalytical amount of HCl in refluxing AcOH and the target compound was obtained in 74% yield as a yellow crystalline solid. In this step, four reactions had taken place in a tandem manner i.e. reduction of both nitro groups, cyclization, isomerization of the intermediate *E*-amide

to the *Z*-amide followed by a second cyclization. IR spectrum showed strong band at 3144 cm^{-1} indicating the presence of –NH functionality. In its ^1H NMR, all the peaks appeared in the aromatic region between δ 7.27 – 9.05 and the –NH proton appeared at δ 11.70. The detailed spectral and physical data i.e. IR, ^1H NMR, ^{13}C NMR, HRMS and melting point are given below.

Spectroscopic data:



48

IR (KBr): υ_{max} = 3144 (-NH), 1614, 1460, 1406, 1231 cm^{-1}.

^1H NMR (DMSO-d_6, 300 MHz): (Fig. 5)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.27	m	-	1	H-9
7.46-7.57	m	-	3	H-2, 7 & 8
7.72	ddd	8.1, 7.2 & 0.9	1	H-3
7.98	d	8.4	1	H-1
8.11	d	8.1	1	H-4
8.27	d	7.8	1	H-10
9.05	s	-	1	H-11
11.70	s	-	1	-N**H**

^{13}C NMR (DMSO-d_6, 75 MHz): (Fig. 6)

δ 111.39 (C-7), 118.37 (C-10b), 120.14 (C-9), 120.76 (C-11a), 122.29 (C-10), 123.20 (C-2), 124. 15 (C-10a), 127.45 (C-1), 128.01 (C-11), 128.67 (C-4), 129.13 (C-3 & C-8), 141.93 (C-6a), 146.79 (C-4a) and 153.36 (C-5a).

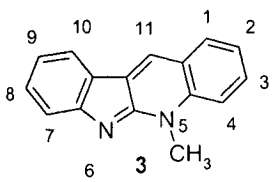
HRMS: *m/z* $[M+H]^+$ 219.0926 (calcd for $C_{15}H_{11}N_2$, 219.0922).

Melting Point: >300 ^0C; Lit.[63, 66] 342-346 ^0C.

On the basis of the above observations with respect to its mode of formation, spectral analysis and melting point, the structure **48** was confirmed for the compound.

Now the final step was the conversion of 6*H*-indolo[2,3-*b*]quinoline **48** into neocryptolepine **3** which was achieved by regioselective methylation on quinoline nitrogen of compound **48** using dimethyl sulfate in refluxing acetonitrile in 80% yield using reported method.[76] Disappearance of –NH peak at 3144 cm^{-1} in its IR spectrum indicates the formation of $-NCH_3$ bond. The PMR spectrum showed a singlet at δ 4.55 integrating for three protons which was assigned to $-NCH_3$ group. The remaining peaks between δ 7.41 – 9.49 were attributed to the aromatic protons. In ^{13}C NMR, the peak at δ 37.0 was assigned to $-NCH_3$ carbon while all the other peaks between δ 113.5 – 148.2 were attributed to aromatic carbons. The complete spectral analysis of the compound is described below.

Spectroscopic data:



IR (KBr): υ_{max} = 2960, 1574, 1496, 1261, 746 cm^{-1}.

^{1}H NMR (DMSO-d_6, 300 MHz): (Fig. 7)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
4.55	s	-	3	-N-C**H**$_3$
7.41	t	7.5	1	H-9
7.60	t	7.5	1	H-2
7.71-7.78	m	-	2	H-3 & H-8
8.03-8.06	m	-	1	H-7
8.28	m	-	2	H-1 & H-4
8.35	d	7.8	1	H-10
9.49	s	-	1	H-11

^{13}C NMR (DMSO-d_6, 75 MHz): (Fig. 8)

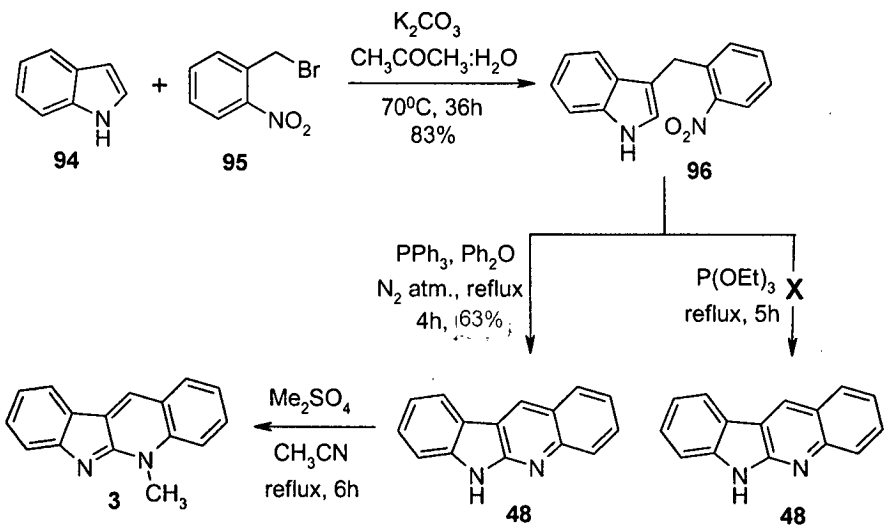
δ 37.0 (-N-**C**H_3), 113.5 (C-4), 116.7 (C-7), 120.4 (C-10b), 122.5 (C-9), 122.6 (C-11a), 123.2 (C-10a), 123.4 (C-10), 126.0 (C-2), 130.3 (C-8), 131.2 (C-11), 133.6 (C-1), 135.7 (C-3), 136.1 (C-4a), 141.3 (C-6a) and 148.2 (C-5a).

HRMS: *m/z* $[M+H]^+$ 233.1067 (calcd for $C_{16}H_{12}N_2$, 233.1078).

Melting Point: 104–106^{0}C; Lit.[31] 108-110^{0}C.

So, based on the above spectral data and closeness of melting point with the literature melting point, the structure **3** was assigned to it. The overall yield of **3** was 42% over the three steps.

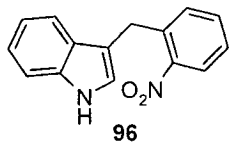
We have also developed another route for the synthesis of cryptotackieine (neocryptolepine) using alkylation and reductive cyclization as the main steps (Scheme 43).



Scheme 43

o-Nitrobenzyl bromide **95** was prepared by reacting *o*-nitro-toluene with *N*-bromo-succinimide in refluxing carbon tetrachloride in presence of benzoyl peroxide. 3-Substituted indole derivative **96**[73] was prepared by reacting indole **94** with freshly prepared *o*-nitrobenzyl bromide **95** in presence of potassium carbonate at 70^{0}C in acetone-water (4:1) solvent. IR spectrum showed the strong bands at 3418, 1520 and 1348 cm^{-1} indicating the presence of –NH and $-NO_2$ functionality. In its ^{1}H NMR, the singlet at δ 4.47 integrating for two protons was assigned to methylene group while the peaks between δ 7.02 – 7.94 were attributed to the aromatic protons and broad singlet at δ 8.06 was assigned to –NH proton of indole moiety.

Spectroscopic data:



96

IR ($CHCl_3$): υ_{max} = 3418 (-NH), 2924, 1520, 1348, 1093, 855, 742 cm^{-1}.

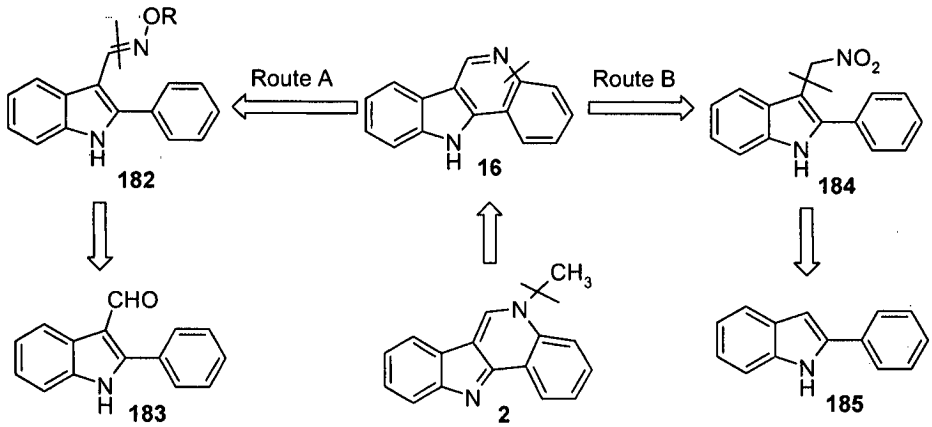
^{1}H NMR ($CDCl_3$, 300 MHz): (Fig. 9)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
4.47	s	-	2	-C**H**$_2$-
7.02	s	-	1	Ar-**H**
7.10	t	7.8	1	Ar-**H**
7.22	t	7.5	1	Ar-**H**
7.33-7.49	m	-	5	Ar-**H**
7.92	d	7.8	1	Ar-**H**
8.06	br s	-	1	-N**H**

Once the sufficient amount of compound **96** is in hand, the next step is the reductive cyclization to get the desired product **48**. Initially, the compound **96** was refluxed in triethyl phosphite for 5 hours, we are expecting compound **48** to form *via* nitrene intermediate but the TLC of the reaction mixture showed many spots whose purification was not attempted. The reductive cyclization was then carried out using triphenyl phosphine in refluxing diphenyl ether to afford the 6*H*-indolo[2,3-*b*]quinoline **48** in 63% yield. Regioselective methylation on quinoline nitrogen was done as described earlier in scheme 2. Spectroscopic data of **48** & **3** are identical with those reported earlier.

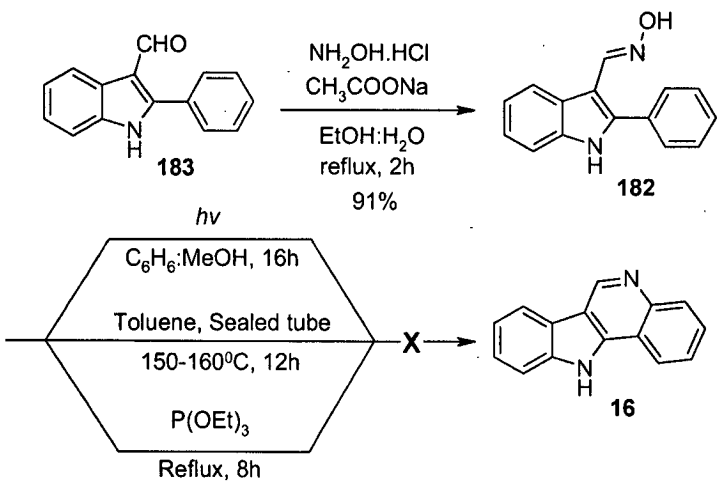
This methodology can be used for the synthesis of analogues of cryptotackieine with subtituents either on indole ring or on quinoline ring using easily available substituted indoles and *o*-nitrobenzyl halides as the starting materials.

Synthesis of isocryptolepine (also known as cryptosanguinolentine) was undertaken based on the retro-synthetic analysis as shown below (Scheme 44).



Scheme 44

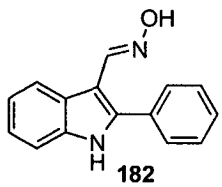
Initially, we attempted the route A for the synthesis of 11*H*-indolo[3,2-*c*]quinoline **16**, the precursor to isocryptolepine from commercially available 2-phenyl-indole-3-carboxyaldehyde as shown below (Scheme 45).



Scheme 45

The aldehyde **183** on treatment with hydroxyl amine hydrochloride in presence of sodium carbonate gave the required oxime **182** in 91% yield. The IR spectrum showed a strong band at 3420 and 3230 cm^{-1} indicating the presence of –NH and –OH functionality respectively. The formation of the compound was further confirmed by melting point and NMR data.

Spectroscopic data:



182

IR (KBr): υ_{max} = 3420 (-NH), 3230 (-OH), 3057, 1631, 1448, 960, 744 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 10)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.14	dd	6.9 & 7.5	1	Ar-**H**
7.22	dd	6.9 & 7.8	1	Ar-**H**
7.44-7.61	m	-	6	Ar-**H**
8.10	d	7.5	1	Ar-**H**
8.29	s	-	1	-N=C**H**-
10.70	s	-	1	-O**H**
11.74	s	-	1	-N**H**

^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 11)

δ 106.3 (Ar-C), 111.9 (Ar-C), 120.9 (Ar-C), 122.3 (Ar-C), 123.2 (Ar-C), 126.0 (Ar-C), 129.0 (Ar-C), 129.4 (4 × Ar-C), 131.8 (Ar-C), 136.8 (Ar-C), 140.1 (Ar-C) and 144.8 (-C=N-).

Melting Point: 182-184 ^{0}C; Lit.[100] 184-185^{0}C.

Based on the above observations with respect to its spectral data and similarity of melting point with that of literature melting point, the structure **182** was confirmed.

Now, the next step was the cyclization of oxime **182** to give the required compound **16** and for this we carried out several reactions as described in scheme 5 above. When the cyclization was tried under UV light, starting material disappears completely as monitored by TLC. But when the product was analysed, it was found that instead of cyclized product **16**, we got the hydrolyzed product i.e. indole-3-carboxyaldehyde **183** in 82% yield whose structure was confirmed by comparing

melting melting point, IR, ^{1}H NMR and ^{13}C NMR with the starting 2-phenyl-indole-3-carboxyaldehyde.

Melting Point: 248-252^{0}C (Commercially available 2-phenyl-indole-3-carboxyaldehyde – 249-253^{0}C).

IR (KBr): υ_{max} = 3200 (-NH), 2980, 1632 (-CO), 1392, 1244, 788 cm^{-1}

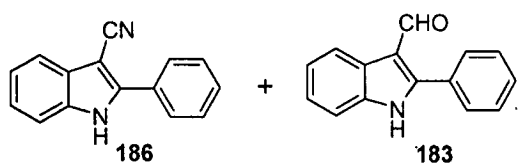
^{1}H NMR (DMSO-d$_6$, 300 MHz): (Fig.12)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.23-7.32	m	-	2	Ar-**H**
7.52	d	7.2	1	Ar-**H**
7.60-7.62	m	-	3	Ar-**H**
7.77-7.93	m	-	2	Ar-**H**
8.22	d	6.9	1	Ar-**H**
9.98	s	-	1	-C**H**O
12.39	br s	-	1	-N**H**

^{13}C NMR (DMSO-d$_6$, 75 MHz): (Fig. 13)

δ 112.4 (Ar-**C**), 113.9 (Ar-**C**), 121.5 (Ar-**C**), 122.9 (Ar-**C**), 124.2 (Ar-**C**), 126.2 (Ar-**C**), 129.4 (3 × Ar-**C**), 130.3 (3 × Ar-**C**), 136.4 (Ar-**C**), 149.5 (Ar-**C**) and 185.9 (-**C**HO).

When the cyclization of oxime was carried out in sealed tube at 150-160^{0}C in toluene, the mixture of 2-phenyl-indole-3-cyanide **186** and 2-phenyl-indole-3-carboxyaldehyde **183** were obtained in the ratio of 1:0.3 instead of expected product **16** in 68% yield (The ratio is based on ^{1}H NMR).



186 **183**

^{1}HNMR (DMSO-d$_6$, 300 MHz): (Fig. 14)

The peak at δ 12.58 was assigned to the –N**H**- of 2-phenyl-indole-3-cyanide while the signal at δ 12.38 was attributed to the –N**H**- of 2-phenyl-indole-3-carboxyaldehyde.

The peak at δ 9.96 was assigned to the –C**H**O of 2-phenyl-indole-3-carboxyaldehyde. The signals at δ 7.25 – 8.22 was attributed to aromatic protons.

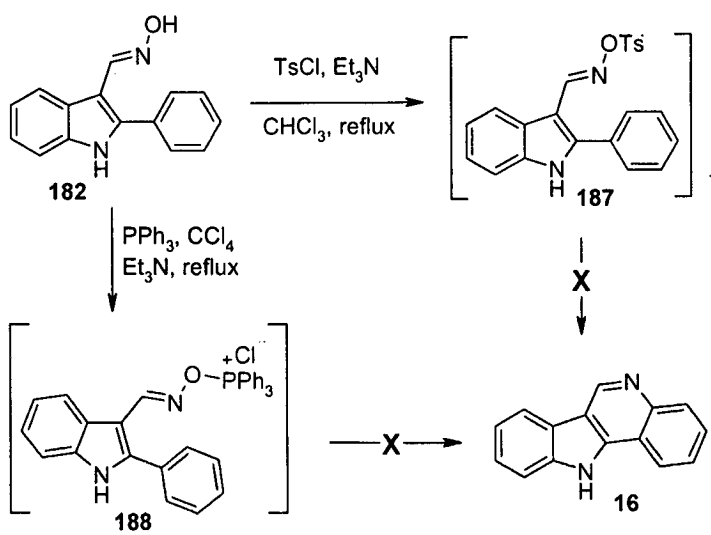
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 15)

The peak at δ 185.9 (d) was assigned to –**C**=O of 2-phenyl-indole-3-carboxyaldehyde while the signal at δ 81.9 (s) was attributed to –**C**N of 2-phenyl-indole-3-cyanide. The remaining signals are due to the aromatic carbons.

IR (KBr): υ_{max} = 3215 (-NH), 3186 (-NH), 2224 (-CN), 1632 (-CO), 1454, 1377, 1246, 741 cm^{-1}

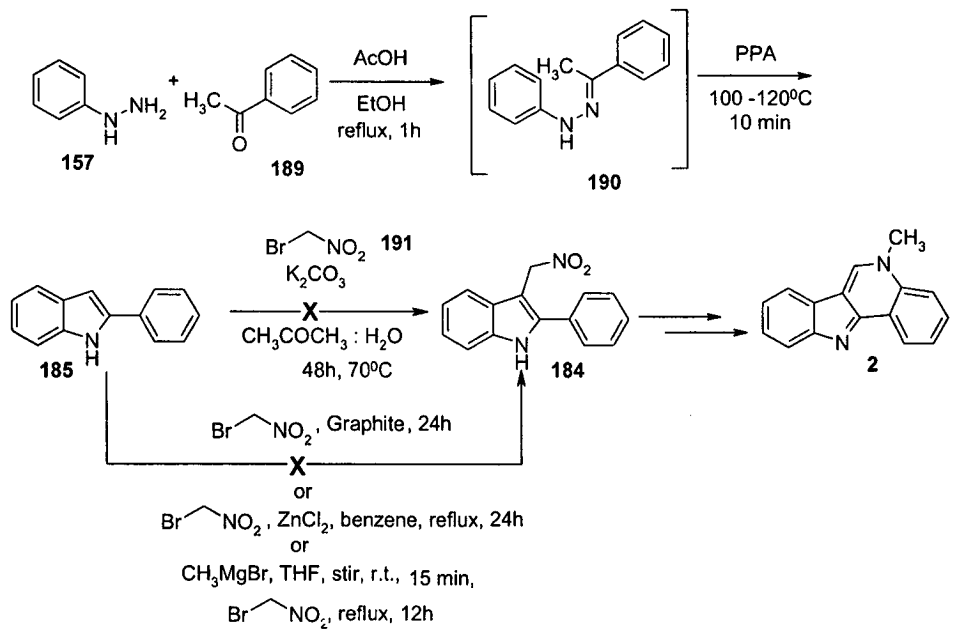
When the cyclization was attempted using triethyl phosphite, again the mixture of 2-phenyl-indole-3-cyanide and 2-phenyl-indole-3-carboxyaldehyde were obtained in the ratio of 1:0.1 in 63% yield as analyzed by ^{1}HNMR.

Since the cyclization of oxime under different reaction conditions did not afforded the expected product i.e. indolo[3,2-*c*]quinoline, we thought that it may be worthwhile if instead we convert oxime into oxime ether which had a greater chance of cyclization.[96,101] Accordingly we carried out the reactions as described in scheme 46 below. But again the reaction failed to deliver the expected product **16** (monitored by TLC).



Scheme 46

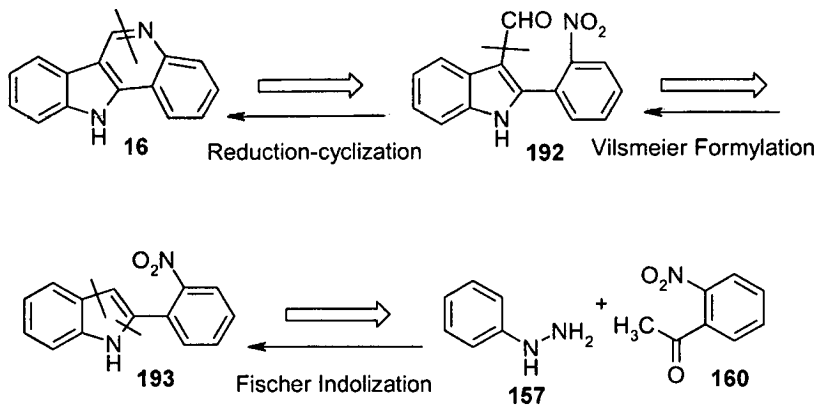
As all our efforts to carry out cyclization of oxime under varying reaction conditions failed to deliver the expected product, we attempted an alternate route i.e route B for the synthesis of isocryptolepine **2** which involves alkylation and reductive cyclization as the key steps (Scheme 47)



Scheme 47

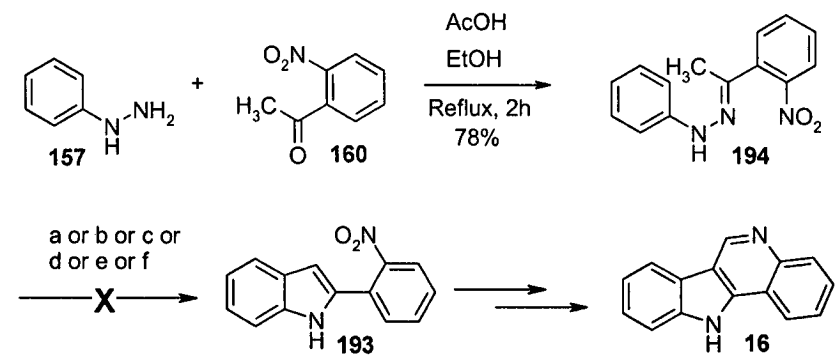
The required compound 2-phenyl indole[101] **185** was prepared starting from phenyl hydrazine **157** and acetophenone **189** *via* intermediate **190** using Fischer indolization. When the alkylation of 2-phenyl indole **185** with nitro-bromomethane **190** was attempted in acetone–water as a solvent in presence of potassium carbonate, the reaction did not yield the product and the starting material remained unchanged. So, alkylation reaction was attempted using graphite or zinc chloride as the catalyst as described in scheme 47 above. But again the reaction failed to give the product and the starting materials remained intact (monitored by TLC). Even the reaction using Grignard's reagent failed to yield the product.

Simultaneously, we also attempted the syntheses of isocryptolepine **2** which was based on retro-synthetic pathway as depicted below involving Fischer indolozation, Vilsmeier formylation and reduction-cyclization protocol (Scheme 48).



Scheme 48

Towards this end, we attempted the following reactions (Scheme 49).

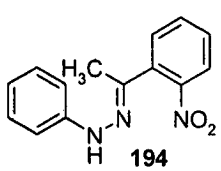


Reaction Conditions: a) PPA, 120-130^{0}C, 3h
b) CF_3SO_3H/P_2O_5, 80^{0}C, 5h
c) AcOH/HCl, 140^{0}C, 10h
d) p-TsOH, Ph_2O, reflux, 8h
e) $POCl_3$, reflux, 16h
f) TFA, reluux, 12h

Scheme 49

Phenyl hydrazine **157** and *o*-nitro-acetophenone **160** when refluxed in ethanol in presence of acetic acid for 3 hours afforded the corresponding hydrazone **194** in 76% yield as a thick brown oil.

Spectroscopic data:



IR ($CHCl_3$): υ_{max} = 3439, 1643, 1526, 1348, 1247 cm^{-1}.

^1H NMR ($CDCl_3$, 300 MHz): (Fig. 16)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.20	s	-	3	-C**H**$_3$
2.58	s	-	1	-N**H**
6.91	t	7.2	1	Ar-**H**
7.11	d	7.8	1	Ar-**H**
7.26-7.31	m	-	2	Ar-**H**
7.44-7.49	m	-	1	Ar-**H**
7.61	d	3.3	2	Ar-**H**
7.77	d	6.0	1	Ar-**H**

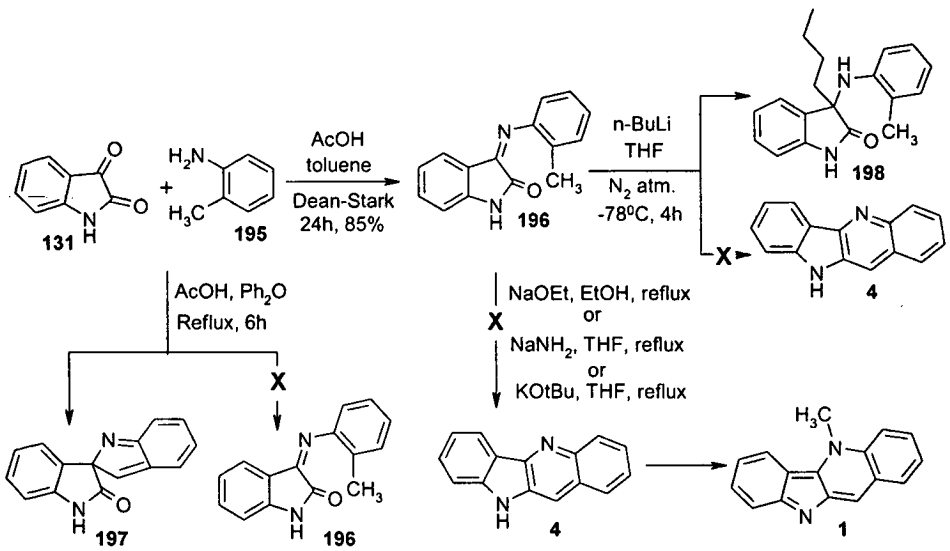
^{13}C NMR ($CDCl_3$, 75 MHz): (Fig. 17)

δ 14.8 (-C**H**$_3$), 113.3 (2 × Ar-**C**), 120.7 (Ar-**C**), 124.3 (Ar-**C**), 128.3 (Ar-**C**), 129.3 (2 × Ar-**C**), 130.2 (Ar-**C**), 132.5 (Ar-**C**), 134.2 (Ar-**C**), 135.3 (Ar-**C**), 138.8 (Ar-**C**), 144.5 (Ar-**C**).

Thus, on the basis of mode of formation and spectral analysis, the structure **194** is suggested for the compound.

Once the sufficient amount of hydrazone **194** was in our hand, the next step was the Fischer indolization to get the cyclized product 2-(*o*-nitrophenyl)indole **193**. For this, we tried various acids (PPA, CF_3SO_3H, AcOH/HCl, *p*-TsOH, TFA or POCl3), but in all cases the reaction failed to give the product and the starting hydrazone was recovered after work up.

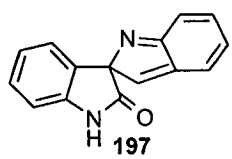
Towards the synthesis of cryptolepine **1**, which is a linearly-fused alkaloid with indolo[3,2-*b*]quinoline ring system, we attempted the following reactions (Scheme 50).



Scheme 50

When isatin **131** and *o*-toluidine **195** was refluxed in diphenyl ether in presence of acetic acid, we are expecting Schiff's base **196** to form but when the product was analyzed it was found that compound **197** was formed instead of compound **196** in 19% yield. The structure was assigned by comparing the melting point, ^{1}H NMR and ^{13}C NMR with that of reported values for this spiro-compound. The structure was further confirmed by HRMS. The detailed spectral data are discussed below.

Spectroscopic data:



197

IR (KBr): υ_{max} = 3209 (-NH), 1703 (-CO), 1597, 1402, 1340, 752 cm^{-1}.

^{1}H NMR (DMSO-d$_6$, 300 MHz): (Fig. 18)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position	Literature Values (δ ppm)
7.22-7.26	m	-	2	Ar-**H**	7.22-7.27
7.27-7.37	m	-	3	Ar-**H**	7.34-7.37
7.41-7.46	dd	7.5 & 7.8	1	Ar-**H**	7.41-7.45
7.74-7.77	m	-	1	Ar-**H**	7.73-7.76

8.10	d	7.8	1	Ar-**H**	8.09-8.11
8.54-8.57	m	-	1	Ar-**H**	8.54-8.56
11.36	br s	-	1	-N**H**	11.37

^{13}C NMR (DMSO-d_6, 75 MHz): (Fig. 19)

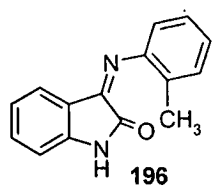
δ 98.7, 114.0, 115.8, 115.9, 120.7, 123.4 (2 × C), 123.9, 124.1, 129.9, 130.1, 133.8, 134.5, 134.7 and 147.5 (-**C**=O).

Melting Point: 248-252^{0}C; Lit.[102] ≥250^{0}C.

Based on the spectral data and similarity of melting point with the literature[102] melting point, the structure **197** was confirmed for the compound.

The Schiff's base **196** was then prepared by reacting isatin **131** and *o*-toluidine **195** in refluxing toluene using Dean-Stark apparatus. IR spectrum showed strong bands at 3240 and 1745 cm^{-1} due to the –NH and carbonyl functionality. In its ^{1}H NMR, the singlet at δ 2.18 was assigned to methyl group while the peaks between δ 6.53 – 7.35 were attributed to the aromatic protons and broad singlet at δ 8.74 was assigned to –NH proton.

Spectroscopic data:



196

IR (KBr): υ_{max} = 3240 (-NH), 1745 (-CO), 1612, 1337, 1205, 758 cm^{-1}.

^{1}H NMR ($CDCl_3$, 300 MHz): (Fig. 20)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.18	s	-	3	-C**H**$_3$
6.53	d	7.5	1	Ar-**H**
6.77	t	7.5	1	Ar-**H**
6.86	d	7.5	1	Ar-**H**
6.93	d	7.5	1	Ar-**H**
7.18-7.35	m	-	4	Ar-**H**

2.30	s	-	3	-C**H**$_3$
4.21	br s	-	1	-N**H**
5.84	d	7.8	1	Ar-**H**
6.59	t	7.5	1	Ar-**H**
6.74	t	7.5	1	Ar-**H**
6.91	d	7.8	1	Ar-**H**
7.02-7.06	m	-	2	Ar-**H**
7.23	t	7.8	2	Ar-**H**
8.86	br s	-	1	-N**H**

^{13}C NMR ($CDCl_3$, 75 MHz): (Fig. 23)

δ 13.8 (**C**H$_3$), 17.7 (**C**H$_2$), 22.6 (**C**H$_2$), 24.7 (**C**H$_2$), 40.8 (**C**H$_3$), 64.7 (-**C**-), 110.5 (Ar-**C**), 111.6 (Ar-**C**), 118.3 (Ar-**C**), 122.9 (Ar-**C**), 123.0 (Ar-**C**), 123.7 (Ar-**C**), 126.9 (Ar-**C**), 128.8 (Ar-**C**), 130.2 (Ar-**C**), 130.6 (Ar-**C**), 140.0 (Ar-**C**), 143.3 (Ar-**C**), 180.8 (-**C**=O).

HRMS: *m/z* $[M+Na]^+$ 317.1641 (calcd for $C_{19}H_{22}N_2O$, 317.1630).

Melting Point: 178-180^{0}C.

Thus on the basis of above observation with respect to its spectral analysis, the compound could have structure **198**.

Conclusion:

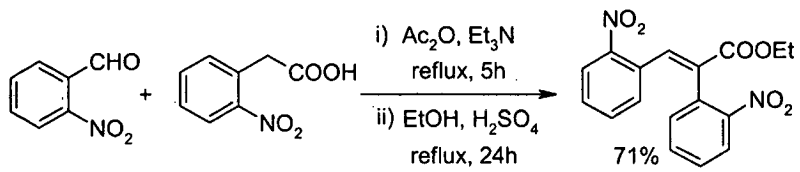
1) We have achieved a new and efficient method for the synthesis of alkaloid cryptotackieine (neocryptolepine) in good overall yield *via* the Perkin reaction followed by 'one pot' double reduction, double cyclization as the key reactions.

2) We have developed another simple method for the preparation of neocryptolepine using alkylation and reductive cyclization as the main steps which can be extended to prepare derivatives of neocryptolepine for their biological evaluation.

3) We attempted the synthesis of isocryptolepine and cryptolepine by unconventional routes without success.

Experimental Section:

1.01 Preparation of α,β-unsaturated ester derivative:

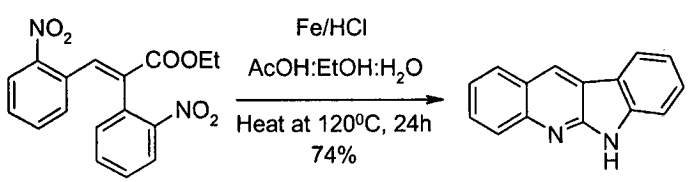


Procedure:

A mixture of o-nitrobenzaldehyde (1.06g, 7.01mmol), o-nitrophenylacetic acid (1.27g, 7.01mmol), Et_3N (1.1mL), and Ac_2O (15mL) were heated at reflux temp. for 5h. The mixture was allowed to cool and poured into water (50mL). This was then extracted with $CHCl_3$ (3 × 15mL) and the combined organic extracts were again extracted with sat. Na_2CO_3 solution (3 × 15mL). The combined Na_2CO_3 extract was acidified with 1:1HCl, filtered and dried. The solid obtained was dissolved in 15mL of EtOH and cat. amt. of H_2SO_4 (5 drops) were added and refluxed for 24h. The reaction mixture was cooled to room temp. and after 6h, the α,β-unsaturated ester derivative **2** (1.71g, 71%) precipitated as a crystalline white solid and was isolated by filtration.

Melting Point: 124-125^{0}C.

1.02 Synthesis of 6*H*-Indolo [2, 3-*b*] quinoline:



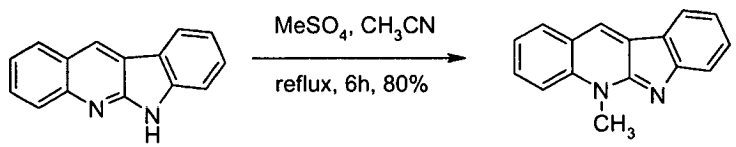
Procedure:

Ester derivative **2** (0.55g, 1.61mmol) and Fe powder (3.60g) were added to a mixture of EtOH (10mL), acetic acid (10mL), and H_2O (5mL). To this mixture 5 drops of conc. HCl were added and the suspension was heated at 120^{0}C while stirring for 24h. The mixture was allowed to cool to room temp. and then filtered through celite. The filtrate was diluted with water (50mL) and then extracted with $CHCl_3$ (3 × 15mL). The combined organic extract was washed with 10% aqueous $NaHCO_3$ (25mL)) and H_2O (3 × 15mL), dried over anhydrous Na_2SO_4 and concentrated to dryness to give a

yellow solid. The solid was washed with Et_2O and air dried to give 6*H*-indolo [2, 3-*b*] quinoline **3** (0.26g, 74%).

Melting Point: > 300^{0}C; Lit.[63, 66] 346^{0}C.

1.03 Synthesis of Neocryptolepine (Cryptotackieine):

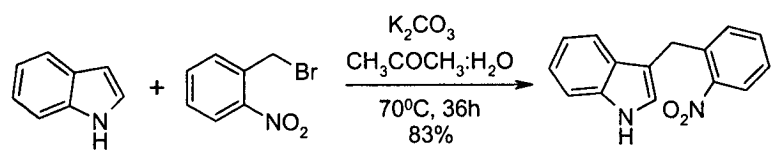


Procedure:

6*H*-Indolo[2,3-*b*]quinoline (0.15g, 0.70 mmol) and dimethyl sulfate (0.5 mL) in CH_3CN (5mL) were heated at reflux temperature for 6 hours. H_2O (15 mL) was added and then alkalized with aq. K_2CO_3. This was extracted with $CHCl_3$ (3 × 15mL), dried over anhydrous Na_2SO_4 and concentrated in vacuo. The crude product was purified by column chromatography using 15% ethyl acetate in hexanes as an eluent to give the product (0.13g) as an orange solid in 80% yield.

Melting Point: 104–106^{0}C; Lit.[31] 108-110^{0}C.

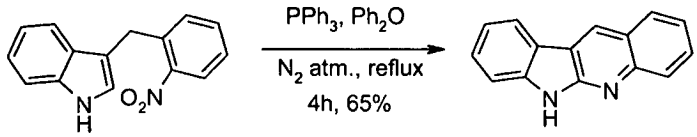
1.04 Preparation of 3-substituted Indole:



Procedure:

The mixture of indole (1.39g, 11.87 mmol), 2-nitrobenzyl bromide (0.51g, 2.37 mmol) and K_2CO_3 (0.66g, 4.75 mmol) were heated at 70^{0}C in 4:1 acetone-water (10mL) for 36 hours. H_2O (20mL) was added, extracted with Et_2O (3 × 20mL), dried over anhyd. Na_2SO_4 and concentrated under reduced pressure. The crude reaction mixture was chromatographed on silica gel (60-120 mesh) and the excess indole was removed using 5% ethyl acetate in hexanes as an eluent. Further elution with 10% ethyl acetate in hexanes afforded the product[73] (0.49g) as a brown thick gel in 83% yield.

1.05 Synthesis of 6*H*-Indolo [2, 3-*b*] quinoline:

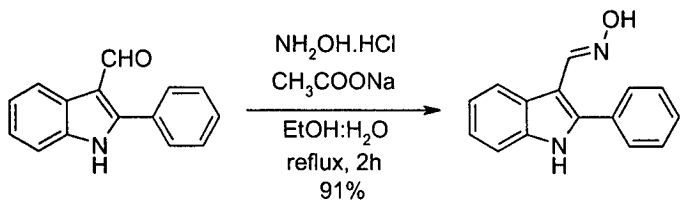


Procedure:

3-Substituted indole (0.40g, 1.62 mmol) and PPh_3 (0.85g, 3.24 mmol) were refluxed in Ph_2O under N_2 atm. for 4 hours. After cooling, reaction mixture was chromatographed on alumina and diphenyl ether was removed using hexanes as an eluent. Further elution with 20% ethyl acetate in hexanes afforded the 6*H*-indolo[2,3-*b*]indoloquinoline (0.22g) as a yellow solid in 63% yield.

Melting Point: > 300^{0}C; Lit.[63, 66] 346^{0}C.

1.06 Preparation of Oxime:

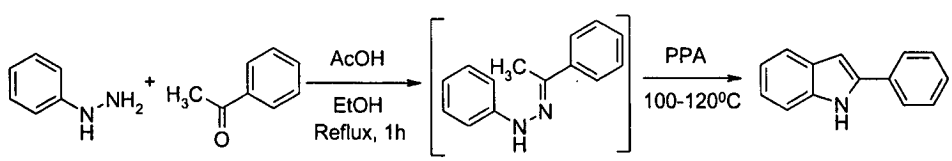


Procedure:

2-Phenylindole-3-carboxyaldehyde (1.02g, 4.63 mmol), hydroxylamine hydrochloride (0.64g, 9.27 mmol) and sodium acetate (1.52g, 18.53 mmol) were refluxed in aq. ethanol (20mL) for 3 hours. After cooling, H_2O (20mL) was added and extracted with EtOAc (3 × 15mL), dried over anhyd. Na_2SO_4 and solvent was removed under reduced pressure to give the yellow crystalline solid (0.99g) in 91% yield.

Melting Point: 182-184 ^{0}C; Lit.[100] 184-185^{0}C.

1.07 Preparation of 2-Phenyl Indole:

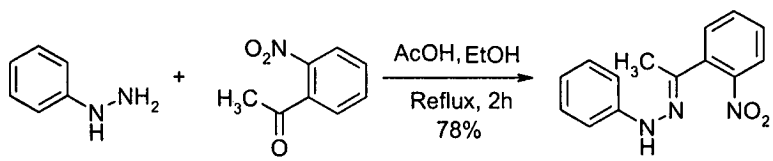


Procedure:

A mixture of acetophenone (2.38g, 19.85 mmol) and phenyl hydrazine (2.14g, 19.85 mmol) were refluxed in ethanol in presence of 5 drops of glac. AcOH for 1 hour. The reaction mixture was cooled to 0^0C and the solid which comes out was filtered, washed first with dilute HCl and then with cold ethanol to give hydrazone as a white solid. To this was added polyphosphoric acid (20g) and heated at 100-120^{0}C for 10 min. After cooling, H_2O (50mL) was added and stirred to complete the solution of PPA. The solid was filtered and washed with water. The crude product was refluxed in ethanol in presence of decolorizing charcoal (1.01g) for 10 min and then filtered. On cooling, the product crystallizes out as a white solid which was filtered and washed with cold ethanol. Yield – 79% (3.03g).

Melting Point: 176-178 ^{0}C; Lit.[101] 178-180^{0}C.

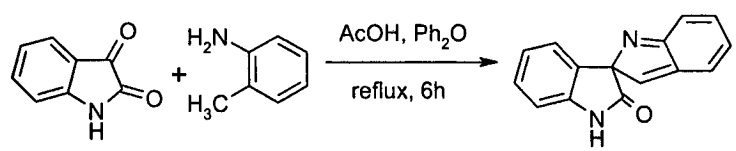
1.08 Preparation of 2-Nitro-phenyl hydrazone:



Procedure:

A mixture of 2-nitroacetophenone (0.56g, 3.45 mmol) and phenyl hydrazine (0.37g, 3.45 mmol) were refluxed in ethanol in presence of 5 drops of glac. AcOH for 2 hours. After cooling, H_2O (30mL) was added and extracted with $CHCl_3$ (3 × 15mL), dried over anhyd. Na_2SO_4 and concentrated in vacuo. The product was purified by column chromatography using 10% ethyl acetate as the eluent to afford the product (0.65g) as sticky solid in 78% yield.

1.09 Synthesis of Spiro compound:



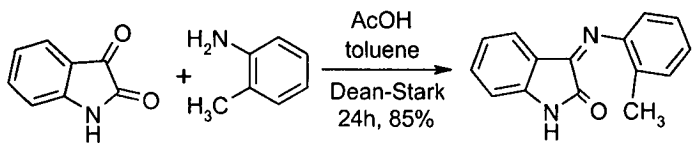
Procedure:

The mixture of isatin (0.51g, 3.49 mmol) and *o*-toluidine (0.37g, 3.49 mmol) were refluxed in Ph_2O (10mL) in presence of 5 drops of AcOH for 6 hours. The crude

reaction mixture was chromatographed on silica gel (60-120 mesh) and diphenyl ether was removed using hexanes as an eluent. Further elution with 20% ethyl acetate in hexanes afforded the spiro compound (0.15g) as a white solid in 19% yield.

Melting Point: 248-252^{0}C; Lit.[102] ≥250^{0}C.

1.10 Preparation of Schiff's base of Isatin:

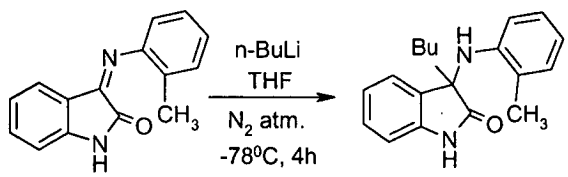


Procedure:

The mixture of isatin (1.53g, 10.43 mmol) and *o*-toluidine (1.11g, 10.43 mmol) were refluxed in toluene in presence of few drops of AcOH using Dean-Stark apparatus for 24 hours. The reaction mixture was concentrated to about 1/3rd of its volume and the solid which comes out was filtered through Buckner funnel, washed with Et_2O to give the product (2.09g) as an orange crystalline solid in 85% yield.

Melting Point: 168-170^{0}C

1.11 Alkylation of Schiff's base of Isatin:



Procedure:

Schiff's base (0.47g, 1.10 mmol) in THF (15mL) was cooled to -78^{0}C under N_2 atmosphere and 1.6M *n*-BuLi (2.5mL) were added with the help of syringe and stirred at this temp. for 4 hours and then allowed to come to room temp. To this was added aq. NH_4Cl (20mL) and extracted with EtOAc (3 × 15mL), dried over anhyd. Na_2SO_4 and concentrated in vacuo. The crude mixture was purified by column chromatography using 30% ethyl acetate in hexanes as the eluent to give the light yellow crystalline solid (0.43g) in 82% yield.

Melting Point: 178-180^{0}C.

Spectra:

Fig. 3: ^{1}H NMR spectrum of **180**

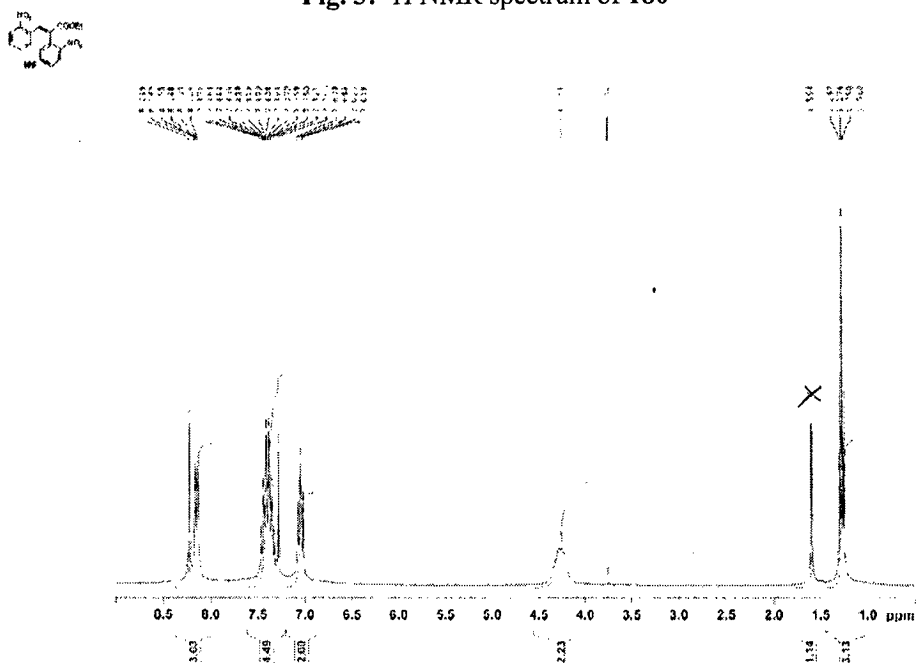


Fig. 4: ^{13}C NMR spectrum of **180**

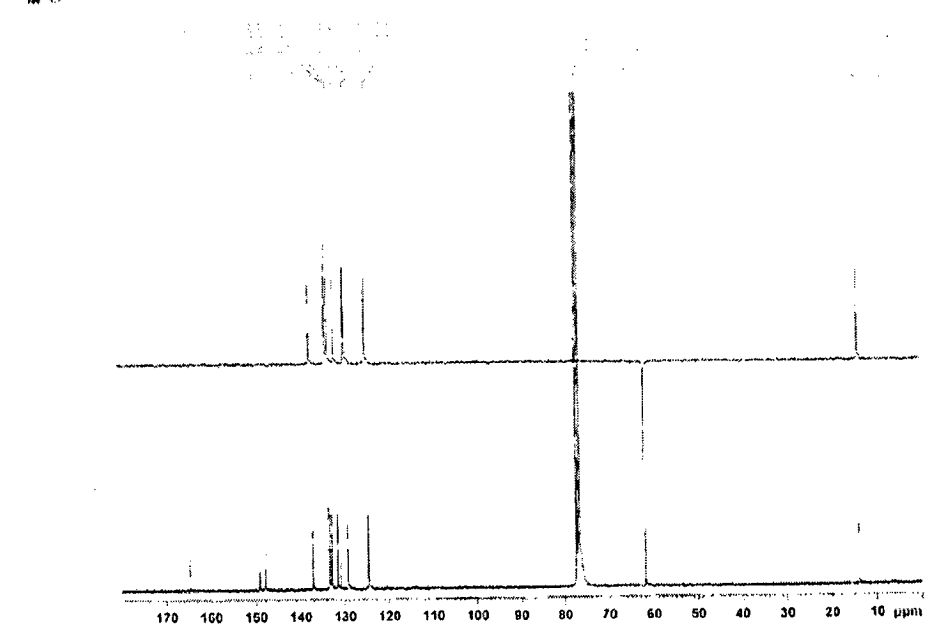


Fig. 5: ^{1}H NMR spectrum of **48**

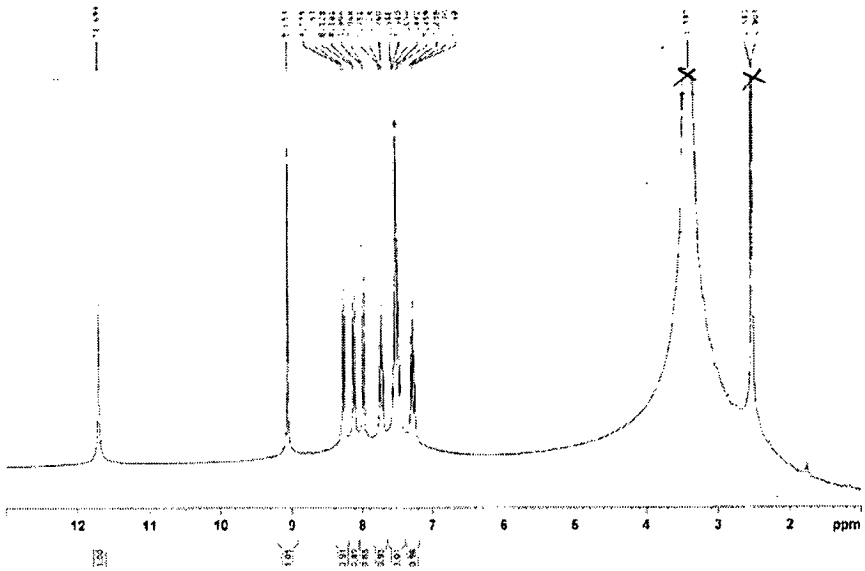


Fig.6: ^{13}C NMR spectrum of **48**

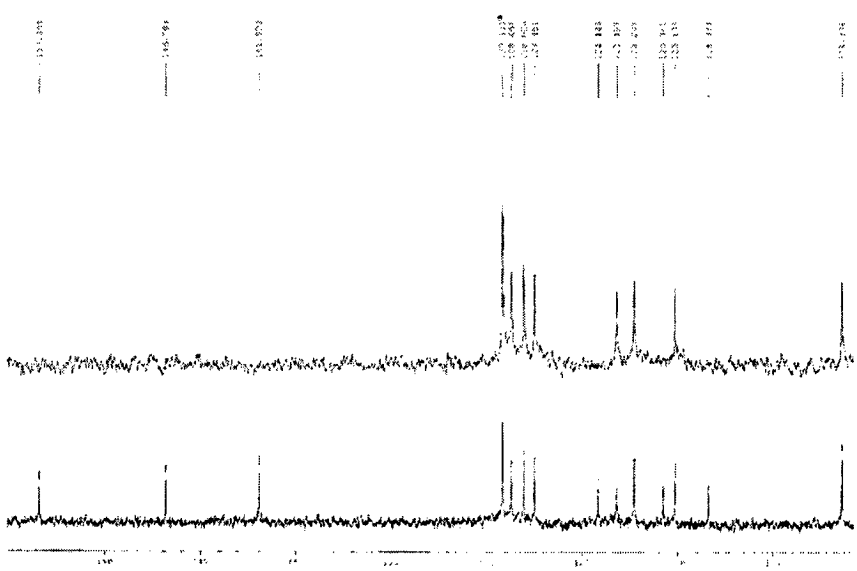


Fig. 7: ^{1}H NMR spectrum of **3**

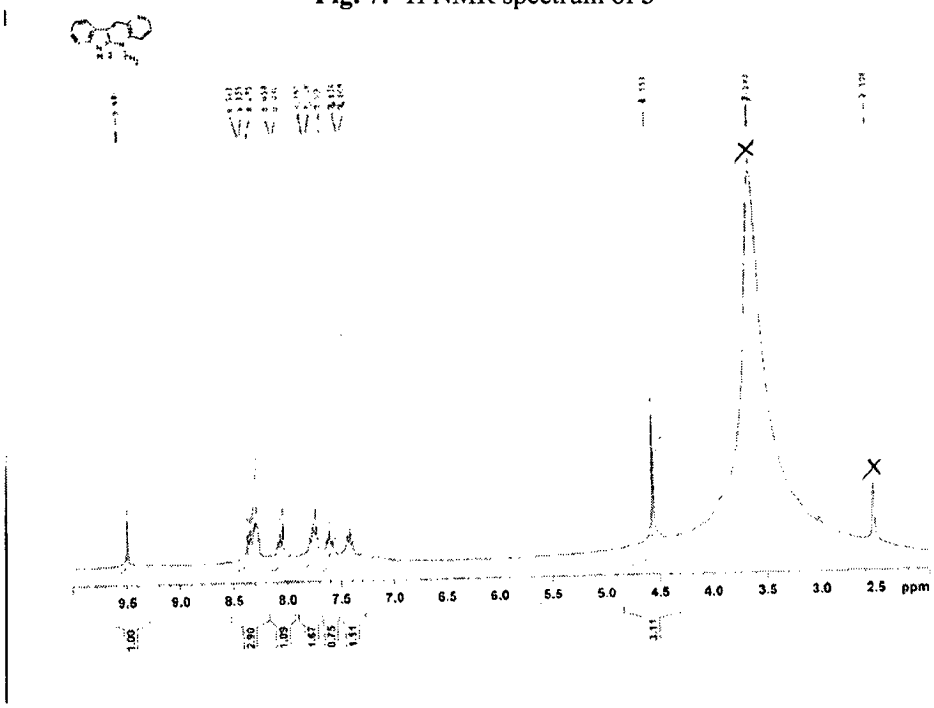


Fig. 8: ^{13}C NMR spectrum of **3**

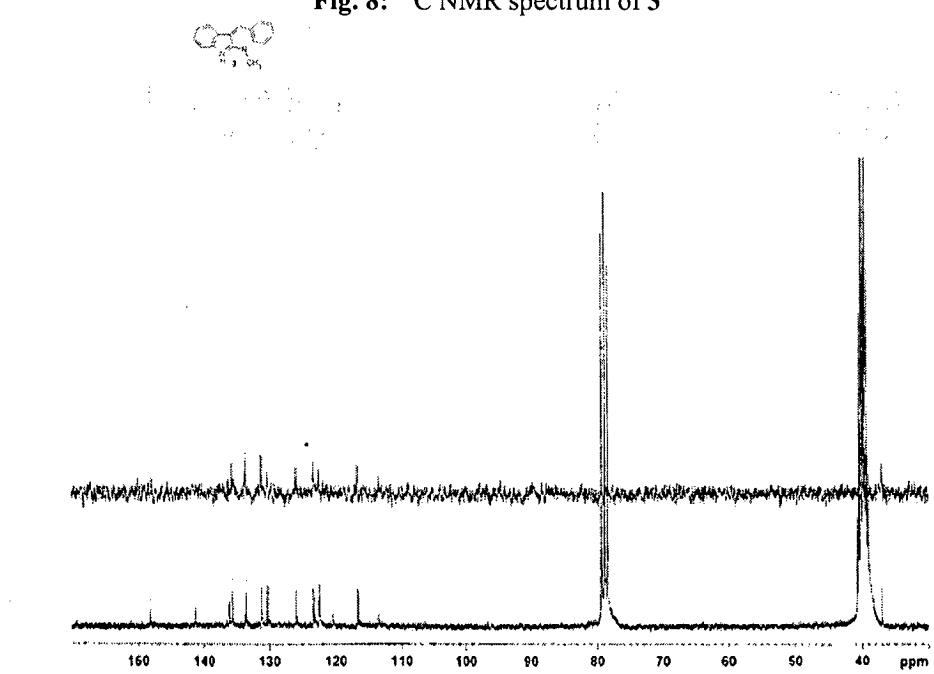


Fig. 11: ^{13}C NMR spectrum of **182**

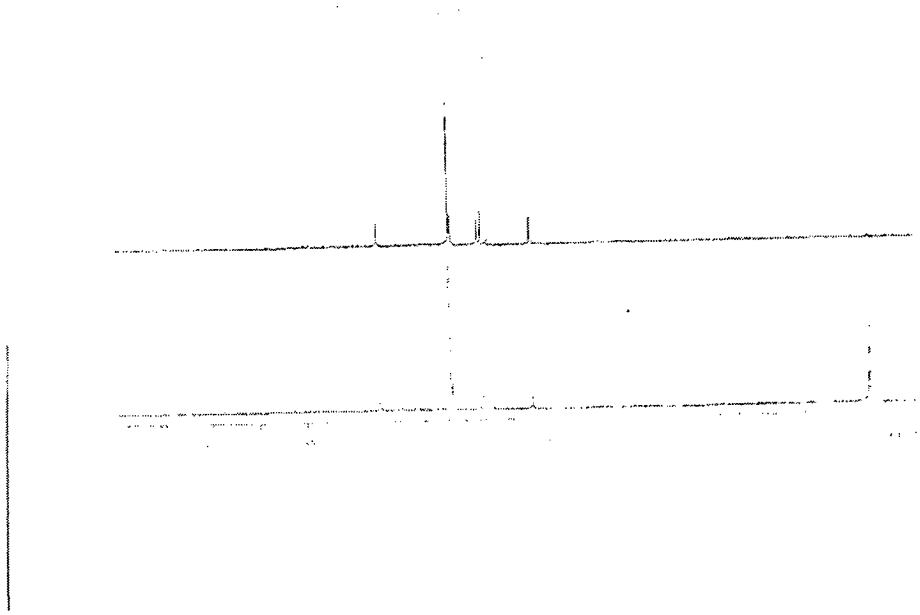


Fig. 12: ^{1}H NMR spectrum of **183**

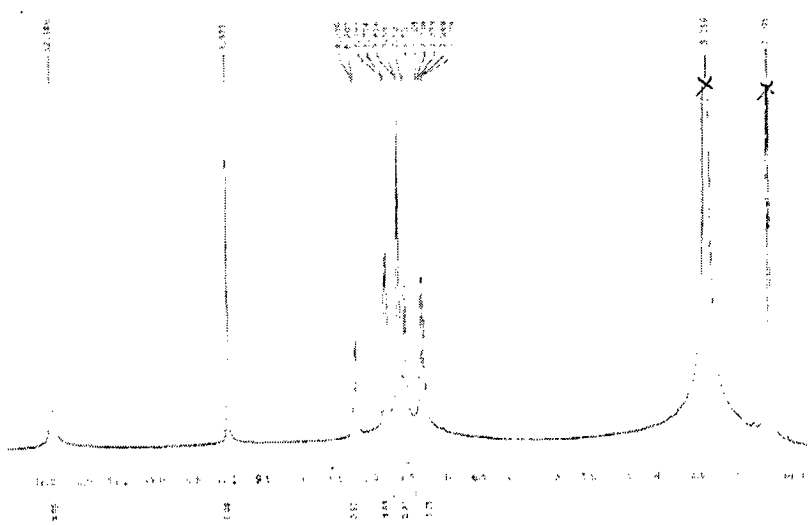


Fig. 13: ^{13}C NMR spectrum of **183**

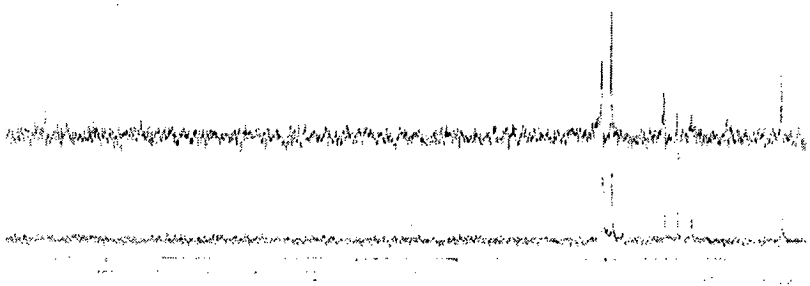
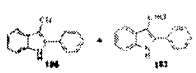


Fig. 14: ^{1}H NMR spectrum of **183 + 186**



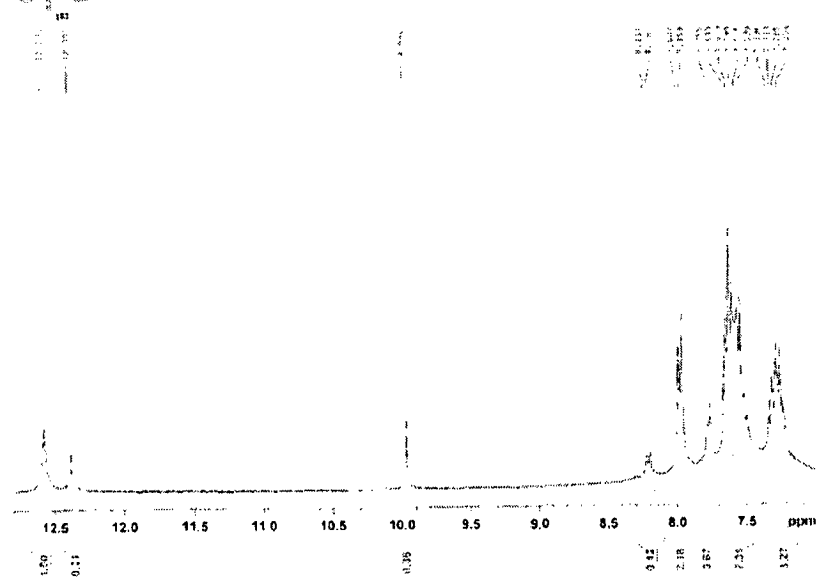


Fig. 15: ^{13}C NMR spectrum of **183 + 186**

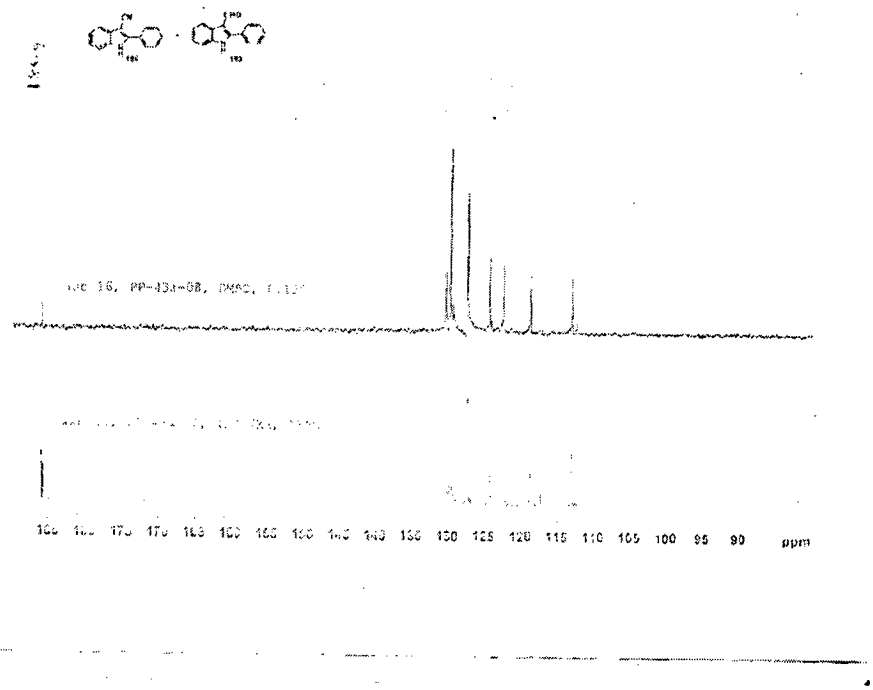


Fig. 16: ^{1}H NMR spectrum of **194**

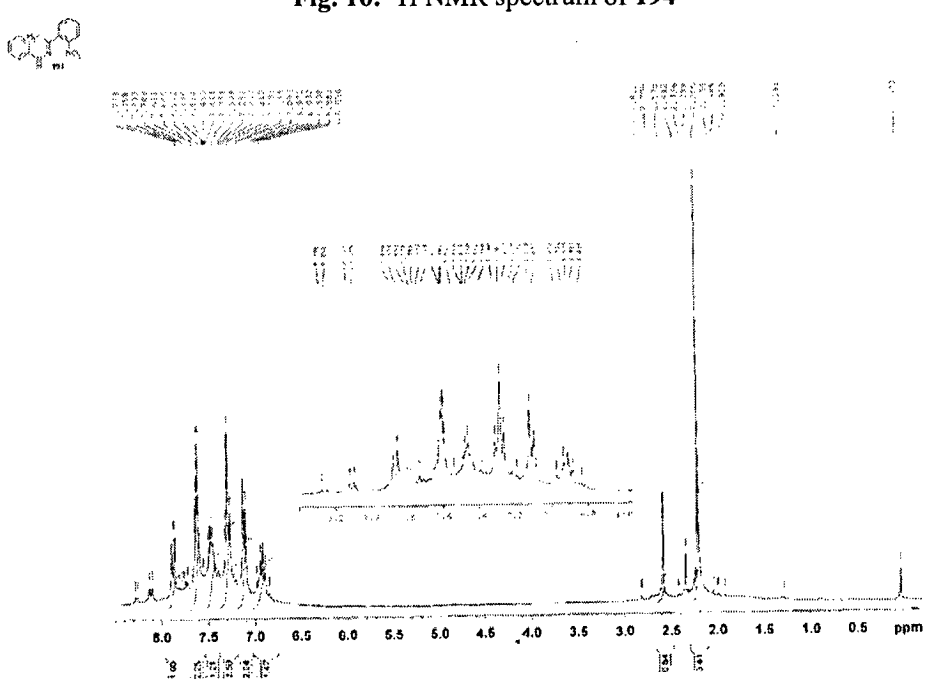


Fig. 17: ^{13}C NMR spectrum of **194**

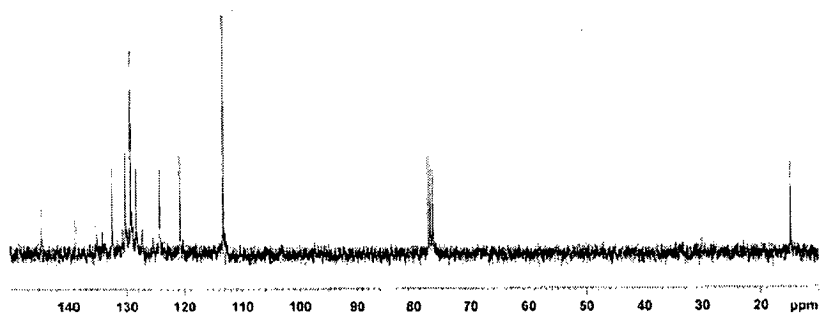


Fig. 18: ^{1}H NMR spectrum of **197**

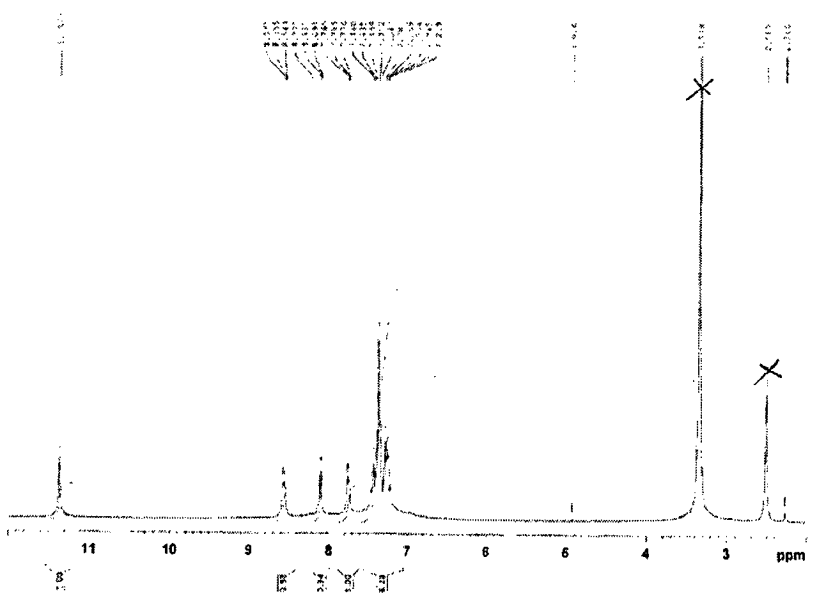


Fig. 19: ^{13}C NMR spectrum of **197**

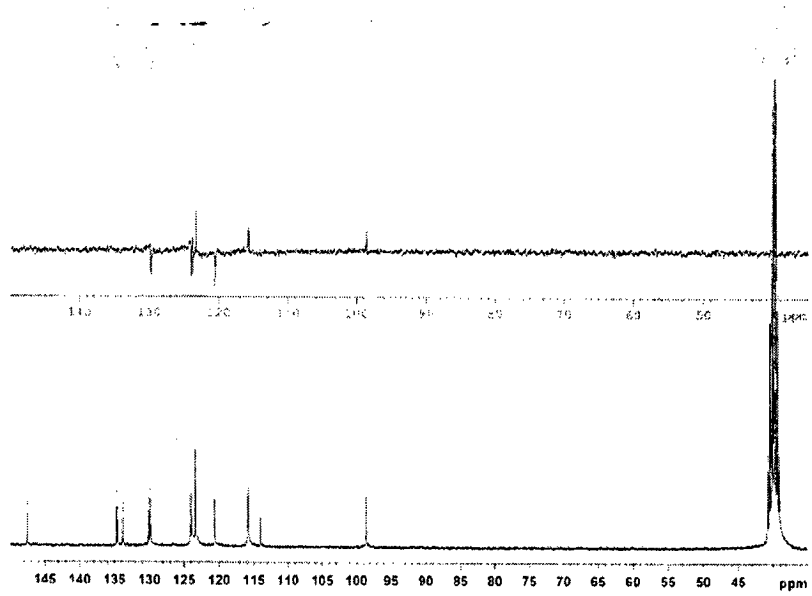


Fig. 20: ^{1}H NMR spectrum of **196**

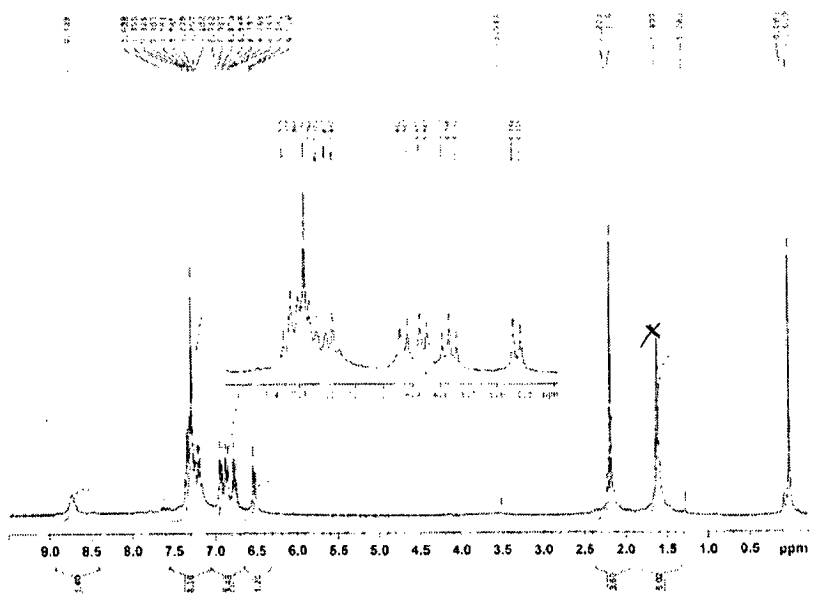


Fig. 21: ^{13}C NMR spectrum of **196**

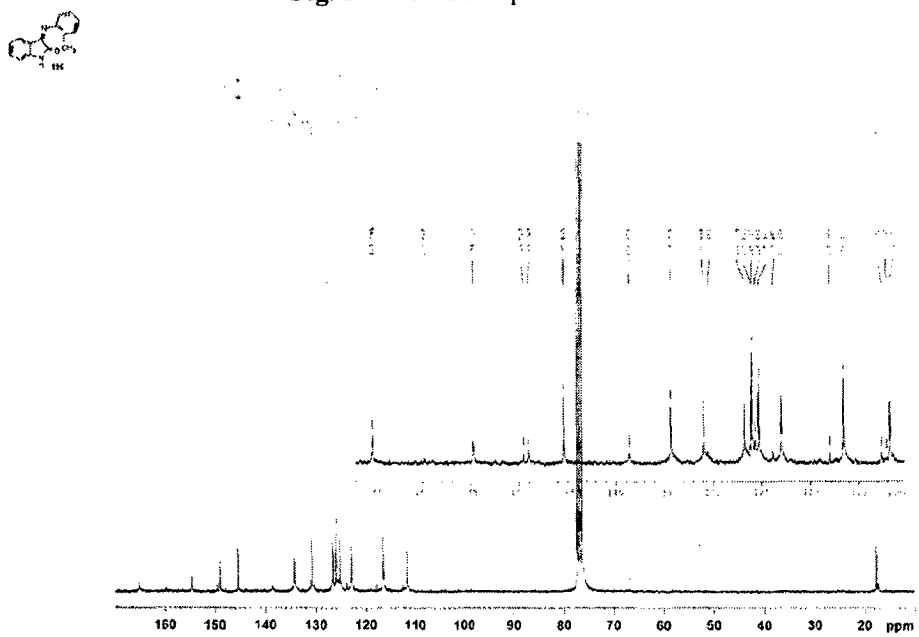


Fig. 22: ^{1}H NMR spectrum of **198**

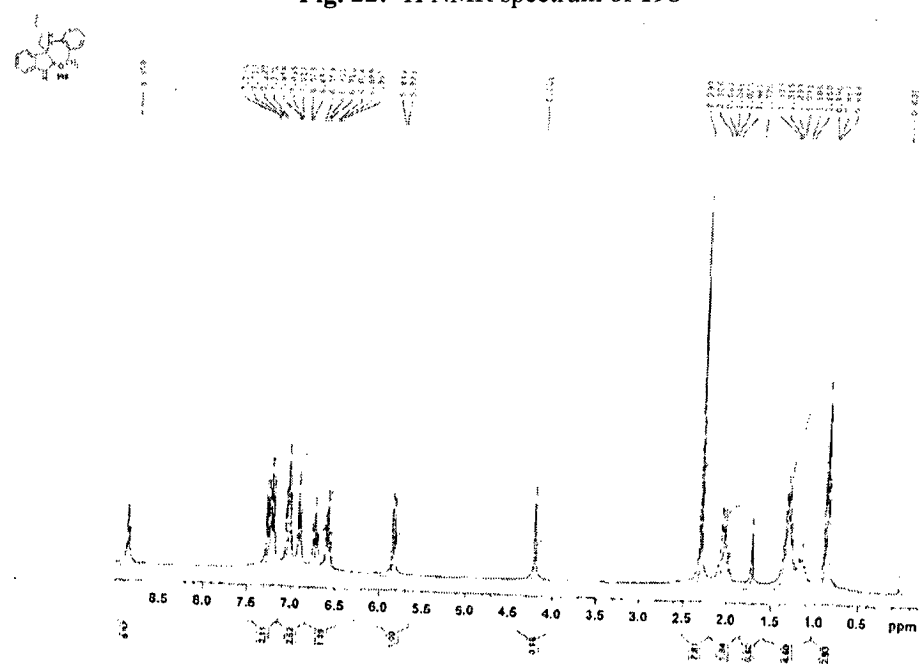
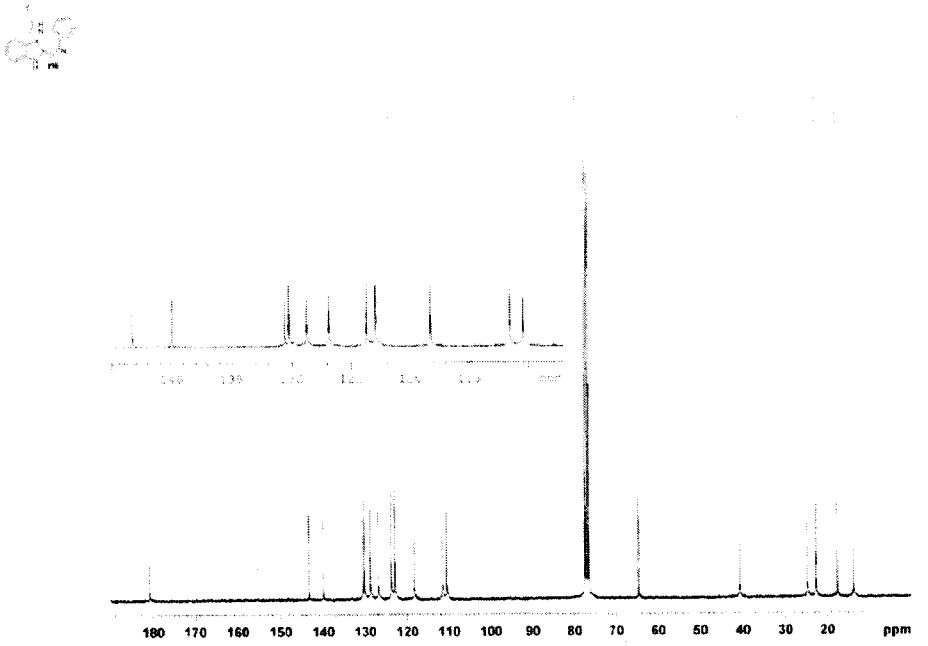


Fig. 23: ^{13}C NMR spectrum of **198**



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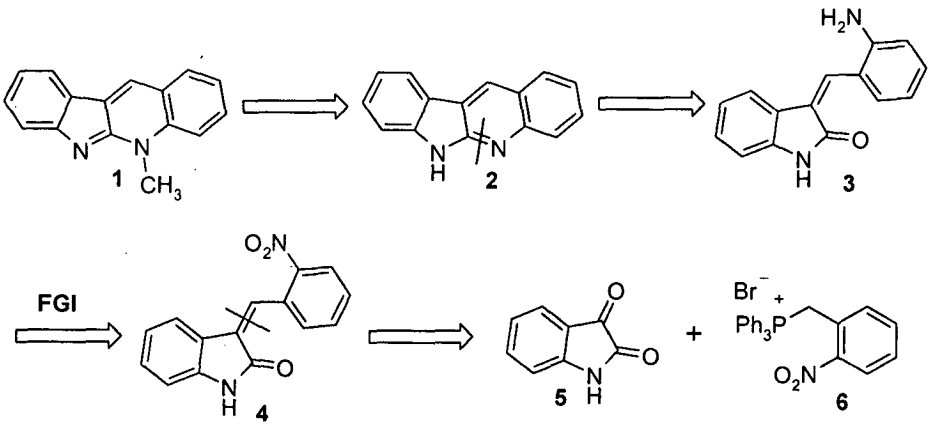
CORRIGENDA

We developed another new method for the synthesis of neocryptolepine in high yield using Wittig reaction and reduction-cyclization-dehydration approach as the key steps. Our retro-synthetic analysis of 6*H*-indolo[2,3-*b*]quinoline **2** (precursor to neocryptolepine) showed that, it could be prepared in one-pot from intermediate **4** *via* reduction-cyclization-dehydration approach. The intermediate **4** in turn be obtained by Wittig reaction from easily available starting materials (Scheme 1).



Scheme 1

Thus, condensation of (2-nitrobenzyl)triphenylphosphonium bromide **6** with isatin **5** in presence of triethyl amine yielded the corresponding Wittig product **4** in 92% yield. The PMR spectrum shows the formation of **4** as a mixture of *cis* / *trans* in the ratio of 1:0.45. The singlet at δ10.69 and 10.52 is assigned to –NH while a singlet at δ 7.83 and 8.16 were attributed to vinylic protons. All the aromatic protons appeared between δ 6.72 – 8.31. The structure was further confirmed by ^{13}C NMR and LC-MS. The detailed spectroscopic data are described below.

^{1}H NMR (DMSO-d_6, 400 MHz): (Fig. i) –

Trans-isomer – δ 10.69 (s, -NH), 8.31 (d, J = 1.2*Hz*, 1H), 7.83 (s, 1H), 7.64 - 7.89 (m, 4H), 6.72 – 7.25 (m, 4H).

Cis-isomer – δ 10.52 (s, -NH), 8.29 (d, J = 0.8*Hz*, 1H), 8.16 (s, 1H), 7.64 - 7.89 (m, 4H), 6.72 – 7.25 (m, 4H).

^{13}C NMR (DMSO-d_6, 75 MHz): (Fig. ii)

Trans-isomer – δ 111.2, 121.4, 122.2, 123.3, 125.2, 128.8, 129.6, 130.7, 130.9, 131.6, 132.4, 135.5, 143.9, 148.4, 168.9 (-C=O).

Cis-isomer – δ 110.6, 121.7, 122.3, 124.1, 126.1, 129.4, 129.8, 130.9, 131.4, 131.9, 132.3, 134.2, 142.5, 147.9, 167.6 (-C=O).

IR (KBr): υ_{max} = 3175 (-NH), 1705 (-C=O), 1616, 1522, 1340, 1232, 866, 735 cm^{-1}

LC-MS: *m/z* $[M+H]^+$ 267

Melting Point: 228 – 232^{0}C

On the basis of the above observations with respect to its mode of formation, and spectral analysis, the structure **4** was confirmed for the compound.

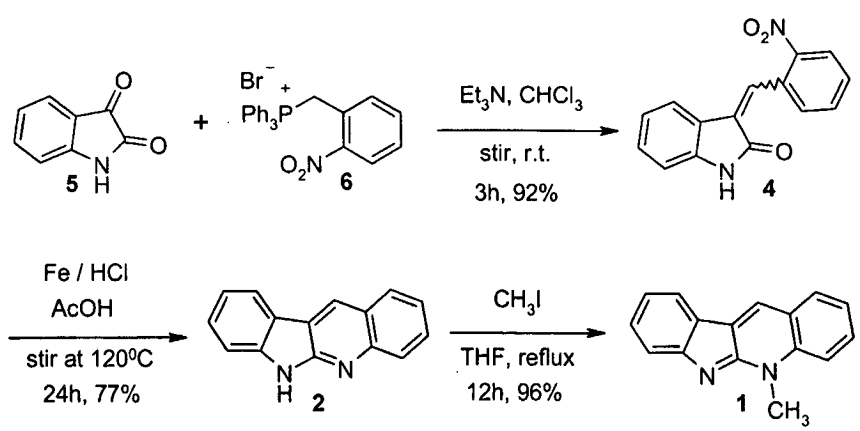
Now, the next step is the conversion of **4** to 6*H*-indolo[2,3-*b*]quinoline **2** and this was achieved using Fe/AcOH in presence of catalytic amount of HCl in 77% yield. In this step, reduction of nitro group, cyclization and dehydration took place in one-pot to give the aromatized product **2**. Spectral data of the 6*H*-indolo[2,3-*b*]quinoline **2** matches with that of our earlier reported data.

Melting Point: > 300^{0}C; Lit.[63, 66] 346^{0}C.

Finally, the compound **2** is converted to neocryptolepine **1** *via* regioselective methylation using methyl iodide.[69]

Melting Point: 106–108^{0}C; Lit.[31] 108-110^{0}C.

The overall yield of **1** in this three step sequence is 68% (Scheme 1) which is found to be highest so far among all the reported methods.



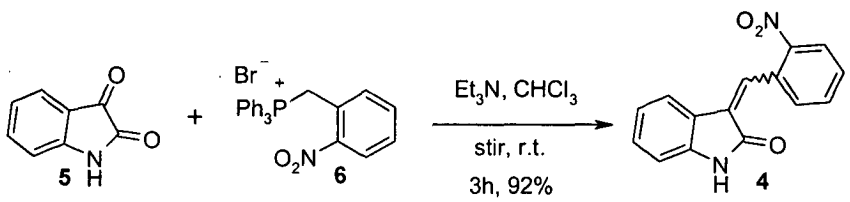
Scheme 2

Conclusion:

We have developed a short, simple and high yielding method which can be elaborated to prepare the derivatives of neocryptolepine for their biological evaluation.

Experimental Section:

Preparation of Intermediate 4:

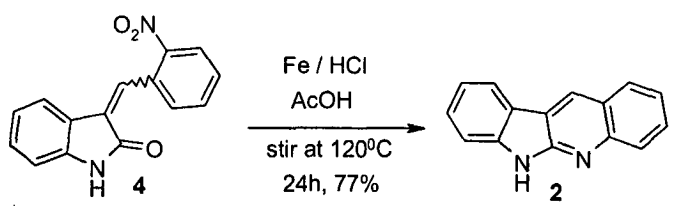


Procedure:

To a mixture of isatin (0.30g, 2.04mmol) and o-nitrobenzyl triphenylphosphonium bromide (1.17g, 2.45mmol) in $CHCl_3$ (10mL) was added Et_3N (0.5mL) and stirred at room temp. for 3h. The solid which comes out was filtered and dried to give the product **4** (0.50g) as a bright red solid in 92% yield.

Melting Point: ^{0}C.

Synthesis of *6H*-Indolo [2, 3-*b*]quinoline:



Procedure:

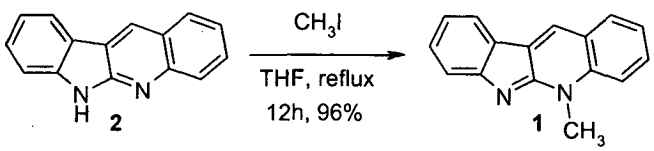
Fe powder (3.01g) was added over a period of 30 minutes to a stirred solution of compound **4** (0.3574g, 1.34mmol) in AcOH (30mL). To this mixture 5 drops of conc. HCl were added and the suspension was stirred at 120^{0}C for 24h. The mixture was allowed to cool to room temp. and then filtered through celite. The filtrate was diluted with water (50mL) and then extracted with $CHCl_3$ (4 × 25mL). The combined organic

extract was washed with 10% aqueous $NaHCO_3$ (25mL)) and H_2O (3 × 15mL), dried over anhydrous Na_2SO_4 and concentrated to dryness to give a yellow solid. The solid

was washed with Et_2O and air dried to give 6*H*-indolo [2, 3-*b*] quinoline **2** (0.23g) as a yellow solid in 77% yield.

Melting Point: > 300^{0}C; Lit.[63, 66] 346^{0}C.

Synthesis of Neocryptolepine (Cryptotackieine):



Procedure:

MeI (0.5mL) was added to a solution of 6*H*-Indolo[2,3-*b*]quinoline **2** (0.20g, 0.92 mmol) in THF (5mL) and refluxed for 14 hours. The solvent was evaporated and the crude product was purified by column chromatography using 20% ethyl acetate in hexanes as an eluent to give the **1** (0.2g) as an orange solid in 96% yield.

Melting Point: 104–106^{0}C; Lit.[31] 108-110^{0}C.

Fig. i: ^{1}H NMR spectrum of **4**

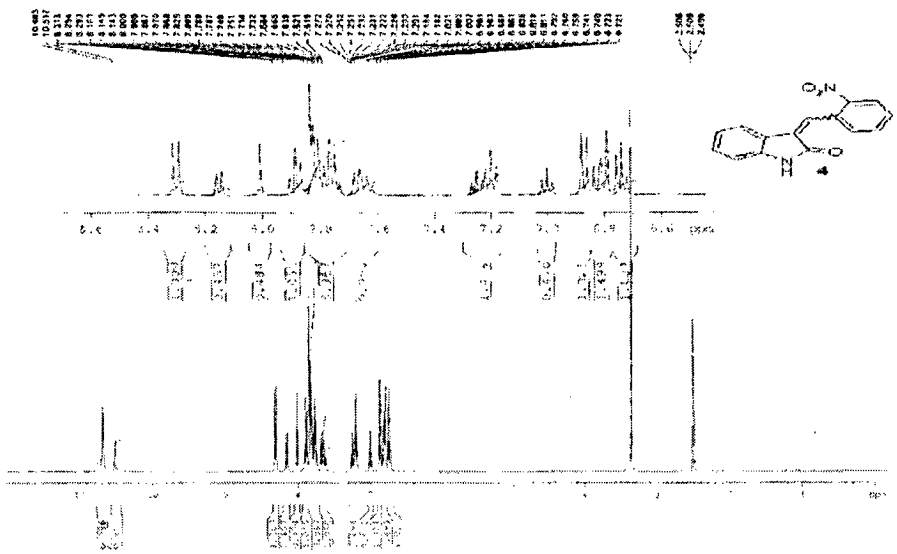
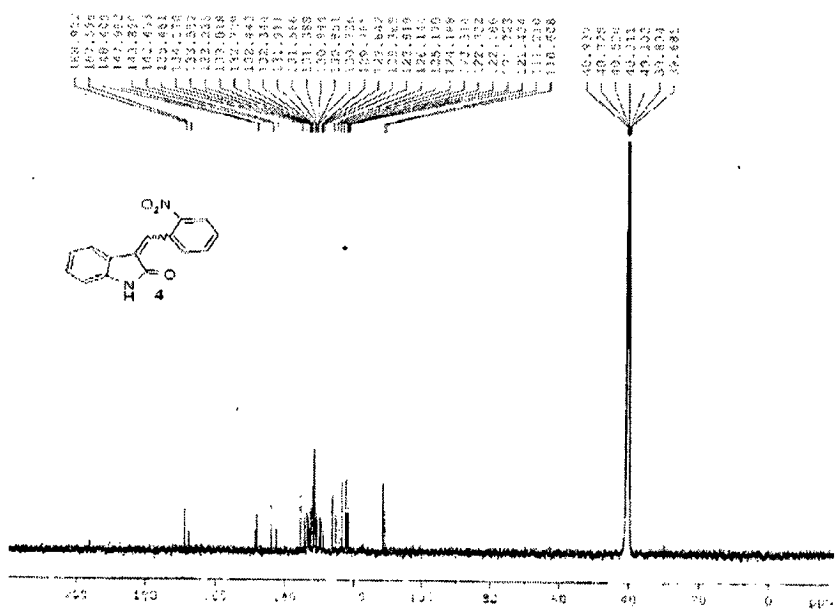


Fig. ii: ^{13}C NMR spectrum of **4**



CHAPTER 2

USE OF MOLECULAR IODINE FOR THE SYNTHESIS OF INDOLOQUINOLINES AND *IN VITRO* ANTIPROLIFERATIVE ACTIVITY STUDY OF SELECTED INDOLOQUINOLINES

USE OF MOLECULAR IODINE FOR THE SYNTHESIS OF INDOLOQUINOLINES AND *IN VITRO* ANTIPROLIFERATIVE ACTIVITY STUDY OF SELECTED INDOLOQUINOLINES

Section A: Use of Molecular Iodine for the Synthesis of 6*H*-Indolo[2,3-*b*]quinoline and their Derivatives.

Introduction:

Iodine is non-metallic and has the following physical properties –
Mp – 112.5^{0}C; Bp – 184.4^{0}C; Specific gravity – 4.933; Electron affinity – 3.06 eV
Vapour pressure – 0.3 mmHg at 20^{0}C.

Iodine is a bluish black solid with an irritating pungent odour and it readily sublimes to a deep-violet-coloured vapor. Its name originates from the Greek word *iodes*, which means violet. Commercially, it is available as a brown solid.

Iodine is a relatively rare element in the earth's crust, but the solubility of its compounds has caused it to concentrate in the oceans. It is the heaviest essential element known to be needed by all living organisms, though in trace amounts, and its inadequacy has led to many deficiency problems in several land animals and humans. The primary function of iodine in the body is to provide a substrate for the synthesis of the thyroid hormones, thyroxine and triiodothyronine, which are crucial for normal growth and development. Iodine deficiency causes goiter and is also the leading cause of mental retardation.

Iodine is non-combustible, but it is a strong oxidizer, especially for –SH groups to disulfides, iodination of aromatic rings in tyrosine and histidine in proteins. Based on this oxidizing ability of iodine, tincture of iodine and povidone-iodine has long been used as disinfectants.

From several places in which iodine occurs in nature only two are used as source for iodine –

1) The caliche (nitrate-bearing earth), found in Chile
2) The iodine containing brines of gas and oil fields, especially in Japan and the United States.

The production of iodine from sea-water *via* electrolysis is not used due to the sufficient abundance of iodine-rich brine. Another source of iodine was kelp (seaweeds) used in the 18th and 19th centuries, but it is no longer economically viable.

In organic chemistry, the alkaline solution of iodine has been used for the detection of acetyl group and is known as the Lieben iodoform reaction. Iodine is a common general stain used in thin-layer chromatography and it is also sometimes used to activate magnesium when preparing Grignard reagents. The major application of iodine is as a co-catalyst for the production of acetic acid by the Mansanto and Cativa processes.

For last few years, iodine has received considerable attention as an inexpensive, non-toxic and a readily available catalyst for various organic transformations which have been well reviewed[1,2] recently in the year 2006. Iodine has high tolerance to air as well as moisture and can be easily removed from the reaction mixture by washing with reducing agents. The development of safe, atom efficient acid-catalyzed organic process is one of the most important challenges for green chemistry. While acid catalysis remains the most widely used type of catalysis, the commonly used acid catalysts continue to present serious problems through health and safety hazards and through separation based on destructive aqueous quenches. Moreover, the mild Lewis acidity associated with iodine enhanced its usage in organic synthesis to perform several organic transformations using catalytic to stoichiometric amounts.

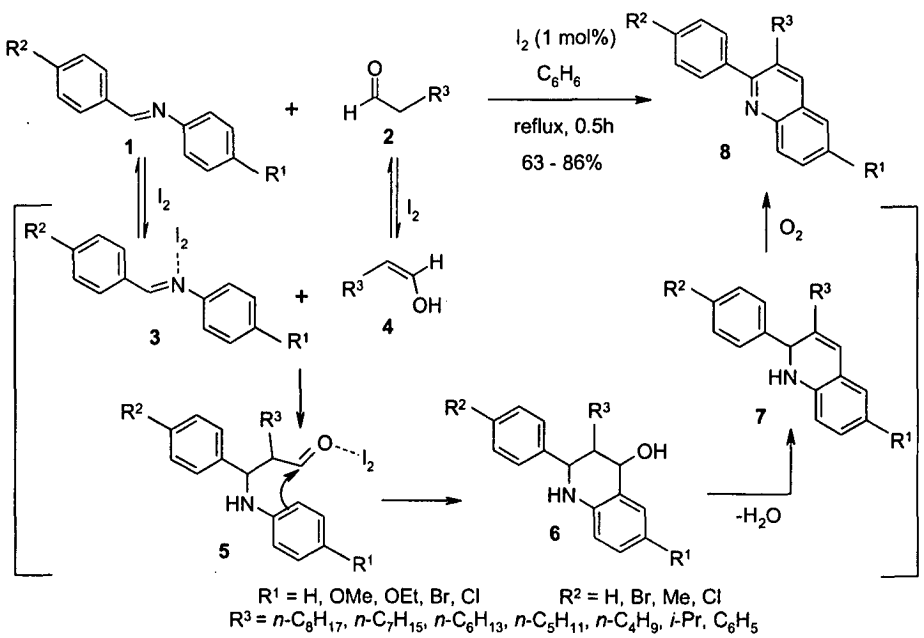
Many of the reactions using iodine are associated with mild reaction conditions, greater stereo- and regioselectivities, short reaction times and mostly carried out *via* multicomponent and domino reaction sequence. Multicomponent[3-7] and domino[8-12] reactions allow the creation of several bonds in a single operation and are one of the important synthetic tools for the creation of molecular diversity and complexicity.[13,14] They also have advantages in terms of user and environmental friendliness because of the step reduction and a high atom-economy associated to their use.

In this review, we focus on the use of catalytical amount of molecular iodine for the construction of various heterocyclic compounds either as a single ring system or condensed to other ring with a mechanistic approach (from 2006 onwards). Heterocyclic rings occur as the key structural subunits in numerous biologically active natural products and has also been used as chiral auxiliaries, chiral catalysts and ligands for asymmetric catalysis. Many important heterocycles, such as benzofurans,

furans, benzothiophenes, thiophenes, benzopyrans, benzoselenophenes, selenophenes, indoles, quinolines, isoquinolines, α-pyrones, isocoumarins, isoxazoles, chromones, β-lactams, 2,3-dihydropyrroles, pyrroles, furopyridines, spiro[4.5]trienones, furanones, isochromenes etc. have been prepared *via* iodine-mediated domino or one-pot multicomponent reactions. To best of our knowledge, the use of molecular iodine for the synthesis of heterocycles has not been reviewed recently, except for iodocyclization by Mphahlele.[15] However, its application in the protection – deprotection[16] of functional groups and electrophilic iodination[17] of organic compounds has been reviewed in detail.

Literature Review:

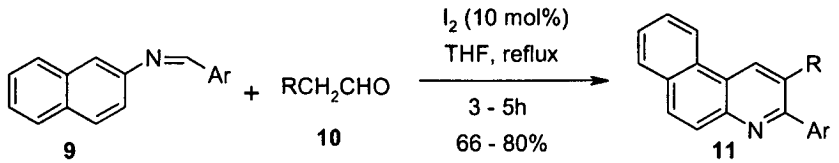
Lin *et al.*[18] prepared a series of substituted quinolines *via* iodine-mediated one-pot domino reaction of imines **1** with enolizable aldehydes **2** (Scheme 1).



Scheme 1

The in situ generated enol **4** reacts with the iodine-activated imine **3** to form intermediate **5** which, underwent intramolecular Friedel-Crafts cyclization and subsequent dehydration and then oxidation to yield the aromatized products **8**.

Wang and co-workers[19] developed a general route to prepare the benzo[*f*]quinolines *via* iodine-catalyzed reaction of Schiff's base with alkyl aldehydes (Scheme 2).

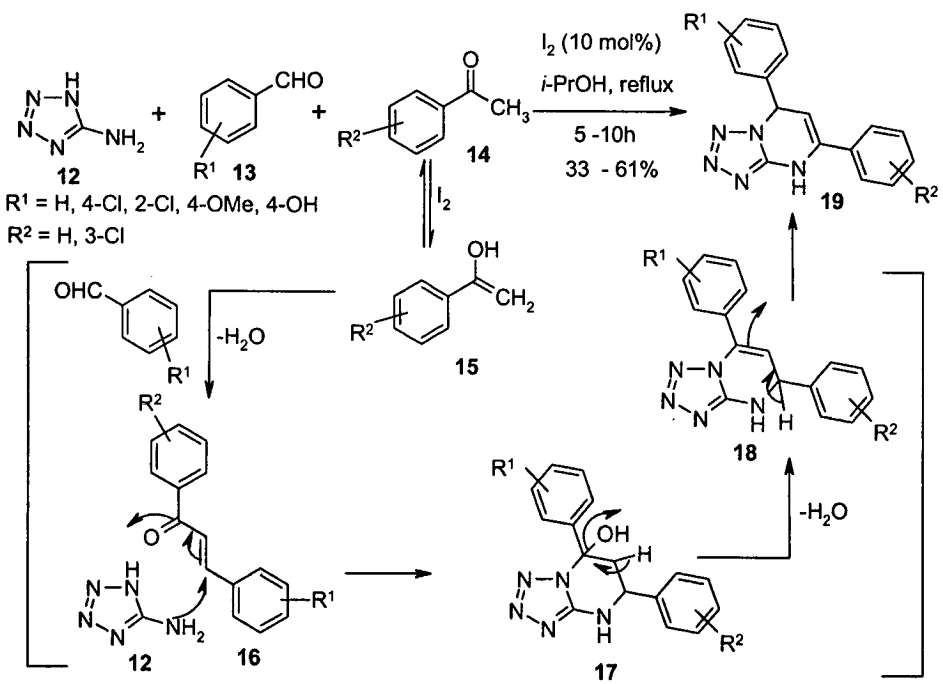


Ar = 4-ClC_6H_4, $4FC_6H_4$, 4-BrC_6H_4, 2,4-$Cl_2C_6H_3$, 2-thiophenyl, 4-$NO_2C_6H_4$, 3-BrC_6H_4

R = Me, Et, C_6H_5, *n*-C_5H_{11}

Scheme 2

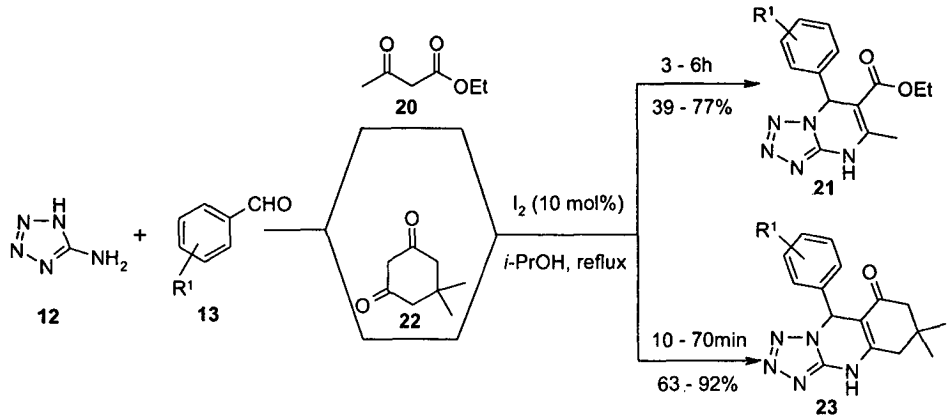
Zeng and Cai[20] accomplished a convenient approach for the construction of diverse tetrazolopyrimidines and tetrazoloquinazolines (Scheme 3 and 4).



Scheme 3

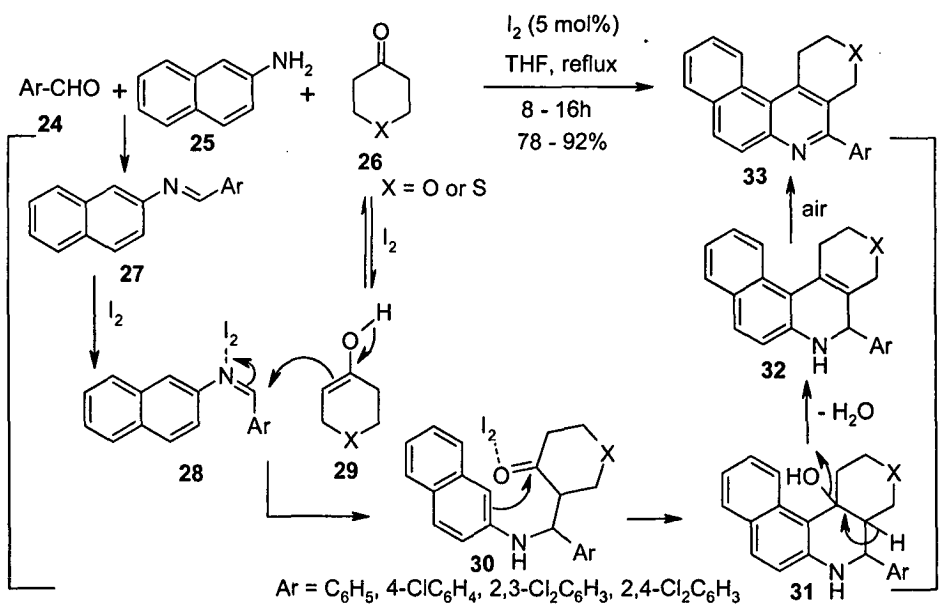
In situ formed enols **15** reacts with aldehydes **13** to form chalcones **16**. 1,4-Addition of the free amino group of 5-aminotetrazole **12** with **16** followed by intramolecular cyclization gave intermediate **17**. Elimination of water and subsequent isomerization of double bond furnishes the corresponding tetrazolopyrimidines **19** (Scheme 3).

Similarly, tetrazolopyrimidines **21** and tetrazoloquinazolines **23** were prepared starting from **20** or **22**, 5-aminotetrazole **12** and various aldehydes **13** (Scheme 4).



Scheme 4

Wang and co-workers[21] described the preparation of pyranoquinoline, thiopyranoquinoline derivatives *via* iodine catalyzed one-pot three component reaction (Scheme 5).

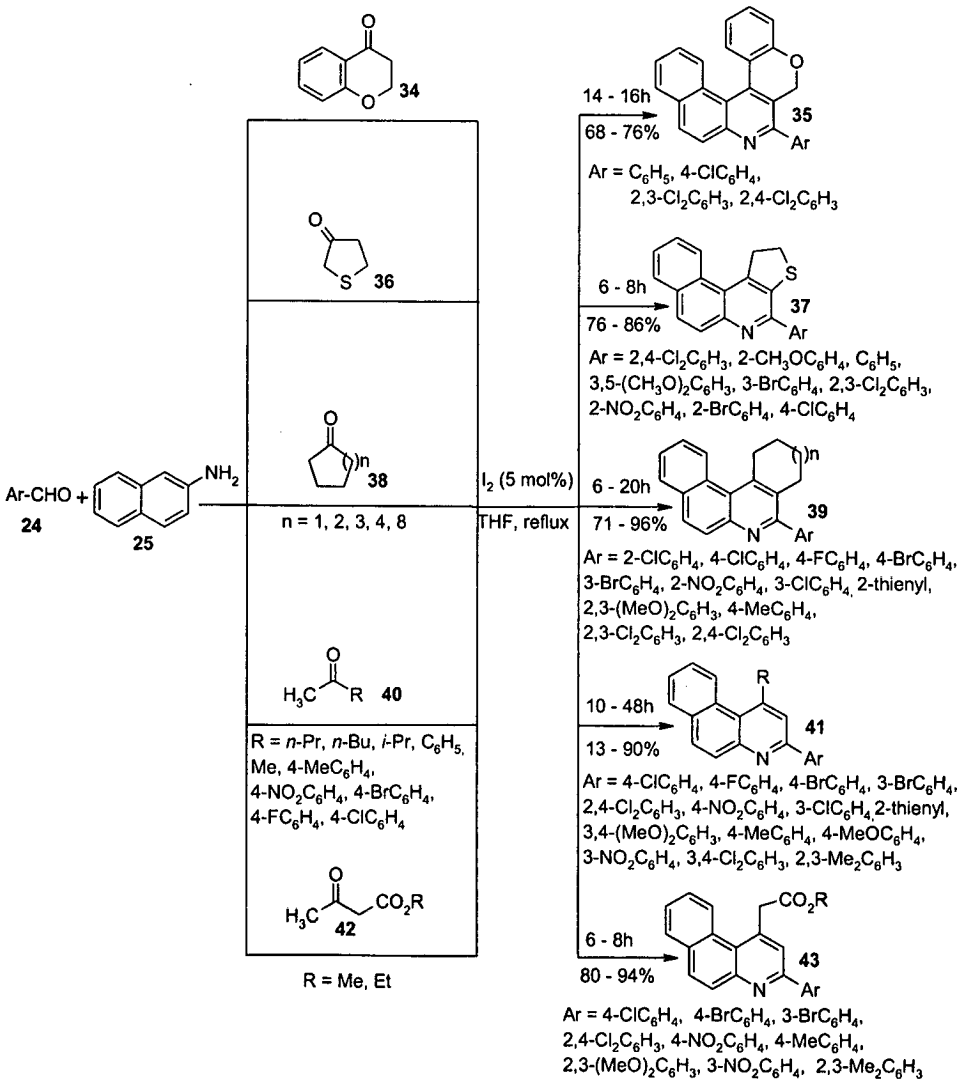


Scheme 5

The reaction of aromatic aldehydes **24** and naphthalene-2-amine **25** and tetrahydropyran-4-one or tetrahydrothiopyran-4-one **26** in presence of iodine afforded the respective 1*H*-5-aryl-benzo[*f*]pyrano[3,4-*c*]quinoline or 1*H*-5-aryl-

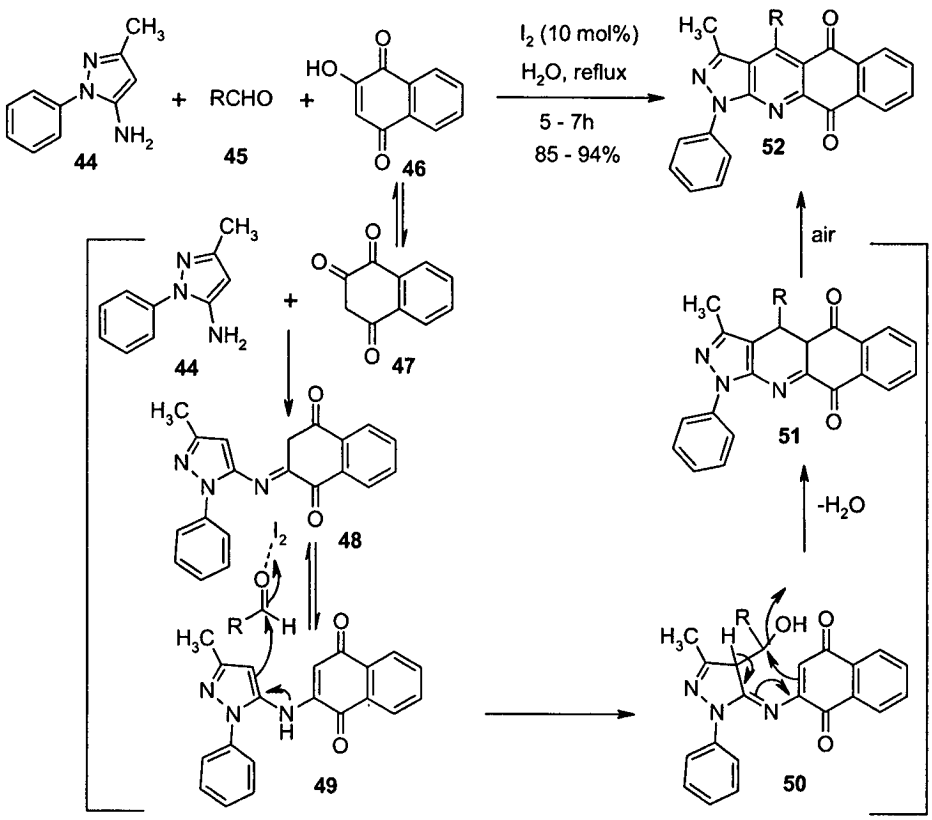
benzo[*f*]thiopyrano[3,4-*c*]quinoline derivatives **33**. In presence of iodine, the ketone is in equilibrium with the enol form **29** which, immediately attacks the iodine activated Schiff's base to form intermediate **30**. Intramolecular Friedel-Crafts cyclization followed by dehydration and oxidation provided the aromatized compounds **33** (Scheme 5).

Analogously, the naphtho[2,7]naphthyridines[21] **35**, thienoquinolines[21] **37**, bezo[*f*]quinolines[22] and benzo[*a*]phenanthridines[22] **39** and 3-aryl-1-substituted benzo[*f*]quinolines[23,24] **41** and **43** were prepared as shown below (Scheme 6).



Scheme 6

Wu and co-workers[25] developed an efficient one-pot method for the preparation of 4-aryl-3-methyl-1*H*-benzo[*h*]pyrazolo[3,4-*b*]quinoline-5,10-diones *via* three component condensation reaction of 3-methyl-1-phenyl-1*H*-pyrazol-5-amine, aldehydes and 2-hydroxynaphthalene-1,4-dione using 10 mol% of iodine in water (Scheme 7).

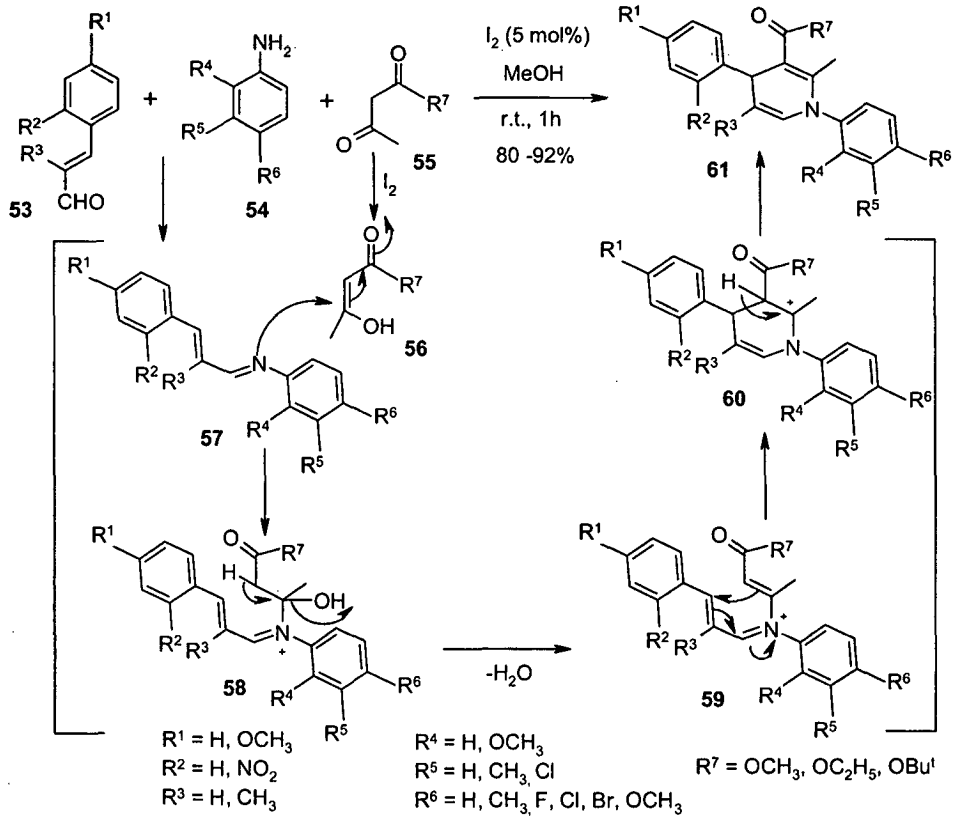


Scheme 7

Dione **46** is in equilibrium with its keto form **47** which, reacts with **44** to form intermediate **48** and then tautomerizes to give intermediate **49**. Nucleophilic attack of **49** on iodine-activated carbonyl carbon of aldehydes **45** followed by intramolecular cyclization and subsequent oxidation yielded the desired products **52**.

Kumar and co-workers[26] described the formation of *N*-aryl-1,4-dihydropyridines (1,4-DHPs) **61** *via* iodine-catalyzed three component reaction of substituted cinnamaldehydes **53**, anilines **54** and 2-keto esters **55** in methanol (Scheme 8). All the

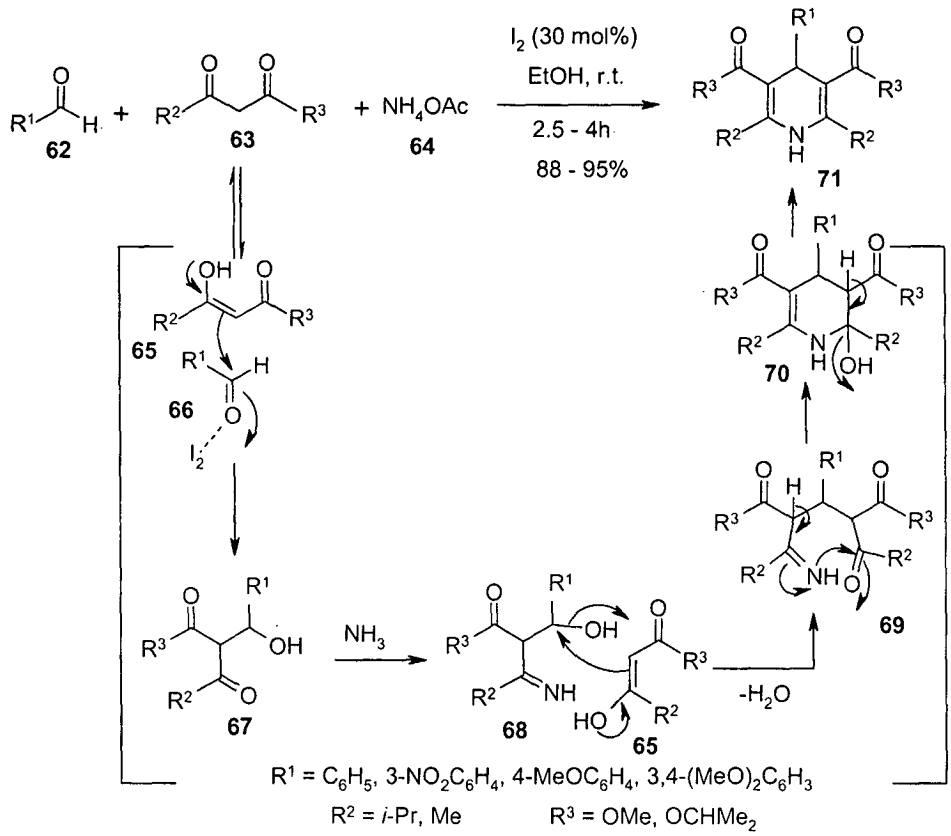
synthesized 1,4-DHPs were screened for their antidyslipidemic and antioxidant activity *in vivo* and *in vitro*.



Scheme 8

Schiff's base **57** formed by treating **53** and **54** underwent 1,4-addition with the enol **56** to generate the intermediate **58**. Elimination of water followed by intramolecular cyclization and subsequent loss of proton afforded the corresponding compounds **61**.

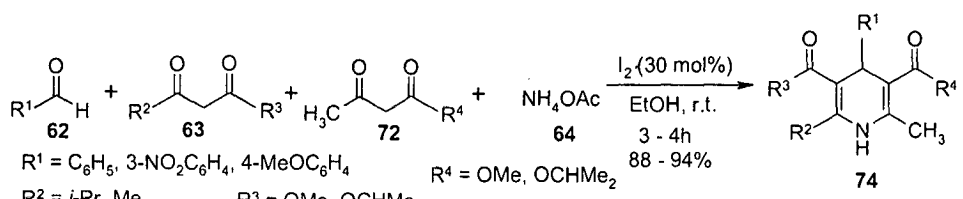
Akbari *et al.*[27] reported the synthesis of symmetrically substituted 1,4-dihydropyridines *via* multicomponent reactions of aldehydes, 1,3-dicarbonyl compounds and ammonium acetate using 30 mol% of iodine (Scheme 9).



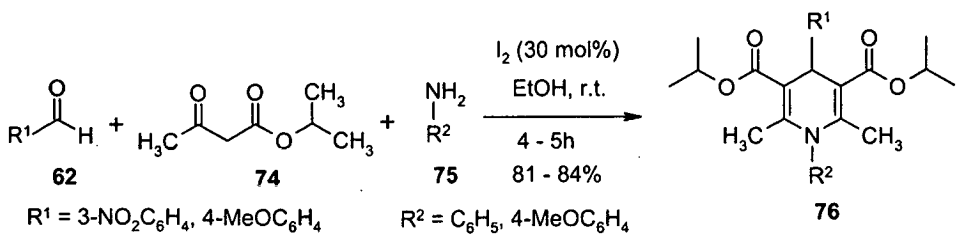
Scheme 9

1,3-Dicarbonyl compounds **63** are in equilibrium with its enol form **65** which, attacks the iodine-activated carbonyl carbon of **66** to generate intermediate **67**. Intermediate **68** formed by condensation of NH_3 with **67** was then attacked by another molecule of **65** to give **69**. Intramolecular cyclization followed by elimination of water afforded the cyclized products **71**.

In a similar fashion, various unsymmetrically substituted 1,4-DHPs **74** (Scheme 10) and *N*-substituted 1,4-DHPs **76** (Scheme 11) have been prepared using iodine as a catalyst.

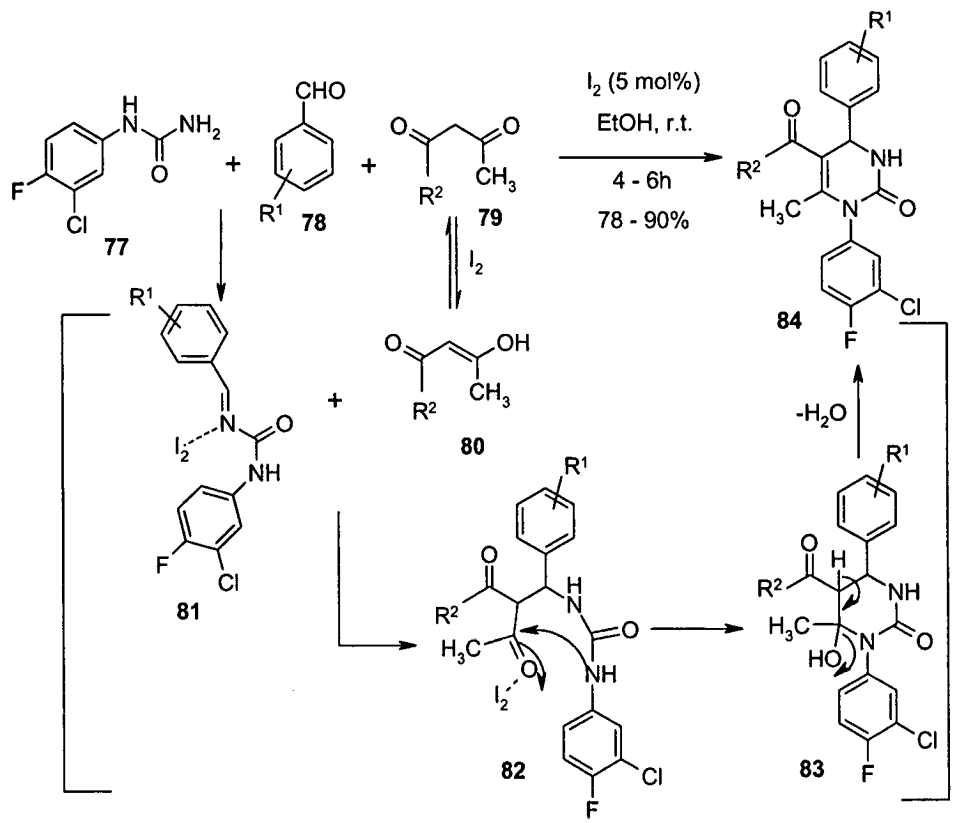


Scheme 10



Scheme 11

Zalavadiya *et al.*[28] demonstrated the three component domino approach for the synthesis of dihydropyrimidines (DHPMs) from aromatic aldehydes, 1,3-dicarbonyl compounds and *N*-(3-chloro-4-fluorophenyl)urea using 5 mol% of iodine (Scheme 12) and these were screened for their *in vitro* antimycobacterial activity.

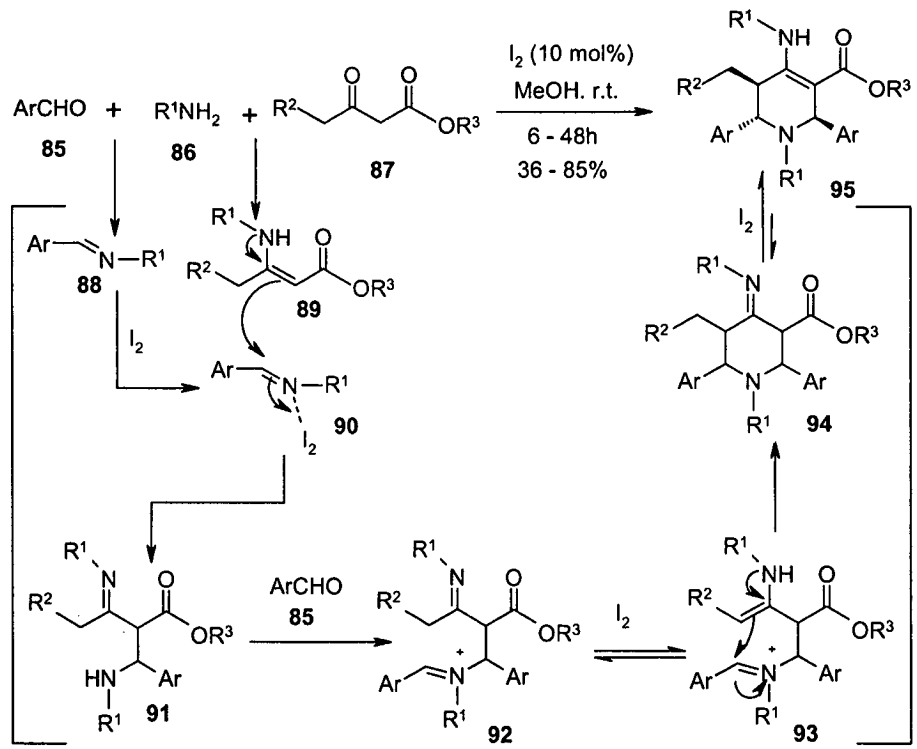


Scheme 12

In presence of iodine, 1,3-diketone **79** is in equilibrium with its enol form **80** which, attacks the iodine activated *N*-acylinium ion intermediate **81** formed in situ by the

condensation of **77** with **78** to give intermediate **82**. Intramolecular cyclization of **82** followed by dehydration resulted in the formation of DHPMs **84**.

Khan and co-workers[29] described an iodine-catalyzed one-pot five component approach for the preparation of highly functionalized piperidine derivatives (Scheme 13).



Ar = C_6H_5, 4-MeC_6H_4, 3,4,5-$(MeO)_3C_6H_2$, 4-ClC_6H_4, 4-BrC_6H_4, 4-FC_6H_4, 3-$NO_2C_6H_4$, 2-$NO_2C_6H_4$

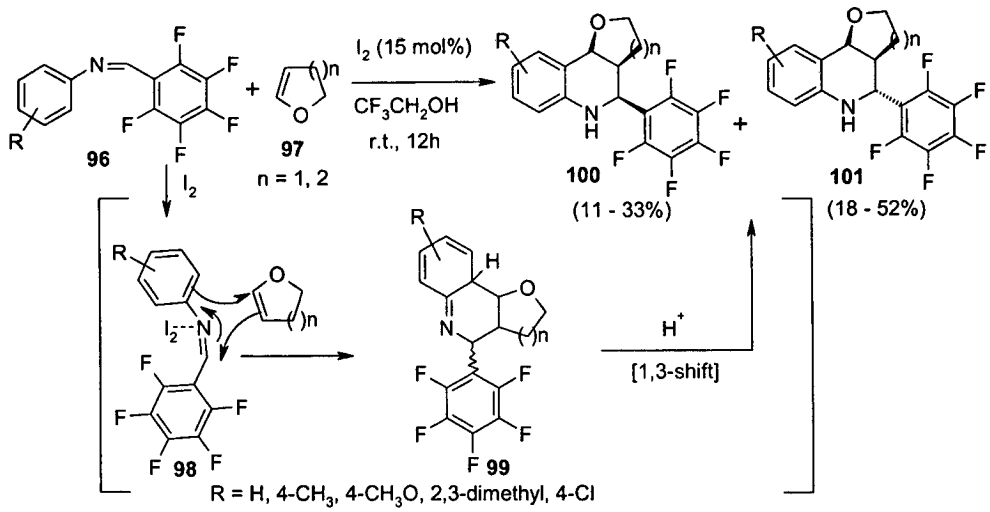
R^1 = C_6H_5, -MeC_6H_4, 4-EtC_6H_4, 4-$MeOC_6H_4$, 4-BrC_6H_4, 4-$NO_2C_6H_4$, *n*-C_4H_9, $C_6H_5CH_2$

R^2 = H, Me, Et, C_6H_5 R^3 = Me, Et, *t*-Bu, allyl

Scheme 13

Enamine **89** generated in situ by the reaction of β-keto esters **87** with amines **86** underwent Mannich-type reaction with iodine-activated Schiff's base **90** to give the intermediate **91**. The intermediate **91** then reacts with aldehydes **85** to form **92** which, in presence of iodine tautomerizes to give the **93**. Intramolecular Mannich-type reaction of **93** resulted in the formation of **94** which, tautomerizes to furnish the corresponding products **95**.

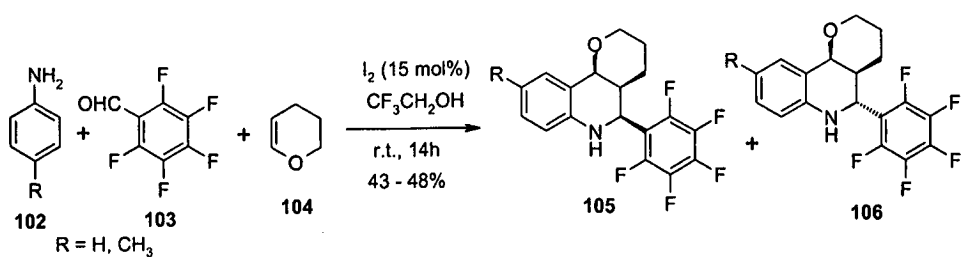
Jin *et al.*[30] reported the iodine-catalyzed imino-Diels-Alder reaction of pentafluorobenzylidineaniline **96** with 3,4-dihydro-2*H*-pyran (DHP) or 2,3-dihydrofuran (DHF) to afford the tetrahydroquinolines as stereoisomers (Scheme 14).



Scheme 14

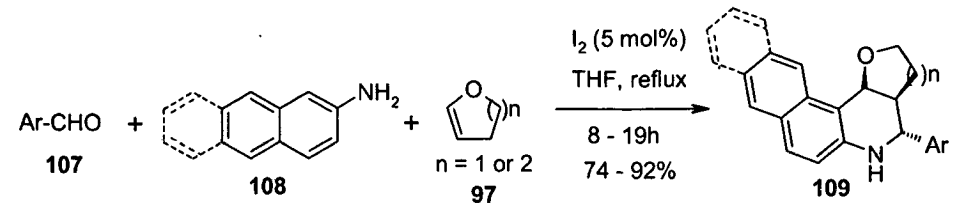
Iodine which acts as a Lewis acid coordinates to the nitrogen atom of the polar C=N bond of Schiff's base **98** which, then underwent hetero-Diels-Alder reaction with DHP or DHF to furnish a series of 2-pentafluorophenyl pyrano[3,2-*c*] and furo[3,2-*c*]tetrahydroquinolines in moderate yields.

One-pot, three component reaction of pentafluorobenzaldehyde, aniline and DHP which avoids the separate step of preparing the imine is also reported (Scheme 15).



Scheme 15

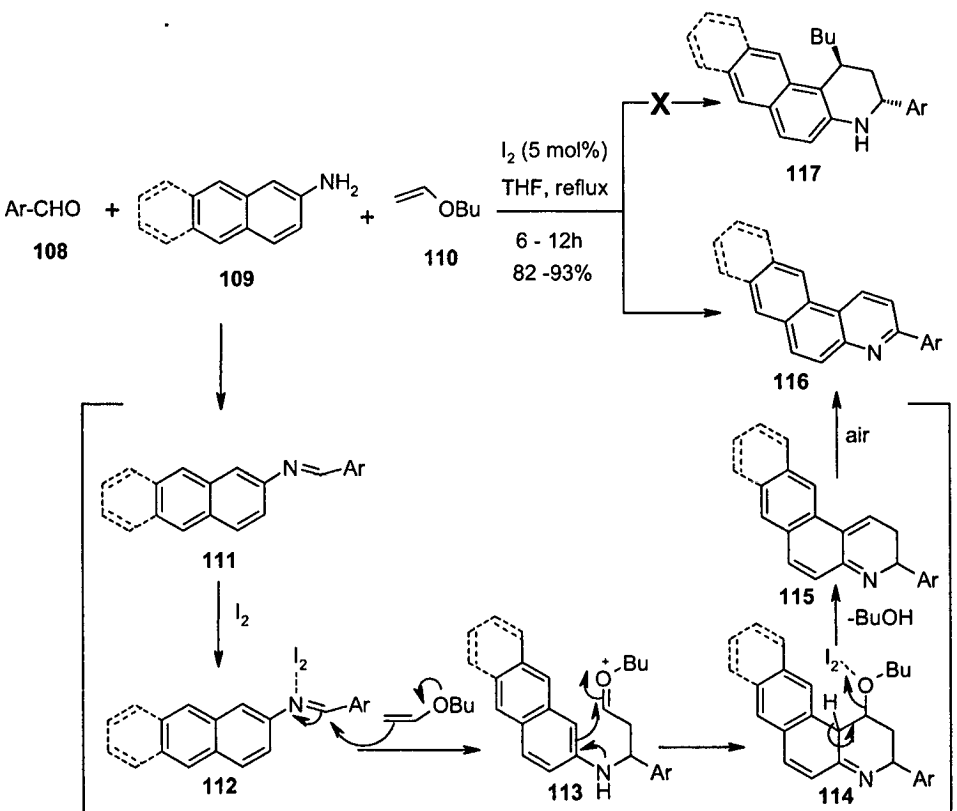
Wang and co-workers[31] demonstrated the formation of pyranoquinoline and furoquinoline derivatives *via* three component reactions of aromatic aldehydes, naphthalen-2-amine or anthracen-2-amine and 2,3-dihydrofuran or 3,4-dihydro-2*H*-pyran (Scheme 16).



Ar = C_6H_5, 4-FC_6H_4, 4-$MeOC_6H_4$, 4-$NO_2C_6H_4$, 4-BrC_6H_4, 3-BrC_6H_4, 3-FC_6H_4, 3-ClC_6H_4, 3,4-$Cl_2C_6H_3$, 2-$MeOC_6H_4$, 3,4-$(MeO)_2C_6H_3$, 4-ClC_6H_4, 3-$NO_2C_6H_4$, 2-thienyl

Scheme 16

When the same reaction was carried out with vinyl ether i.e. *n*-butylvinyl ether **110** instead of cyclic ether **97**, the aromatized compounds **116** were obtained in high yields (Scheme 17).



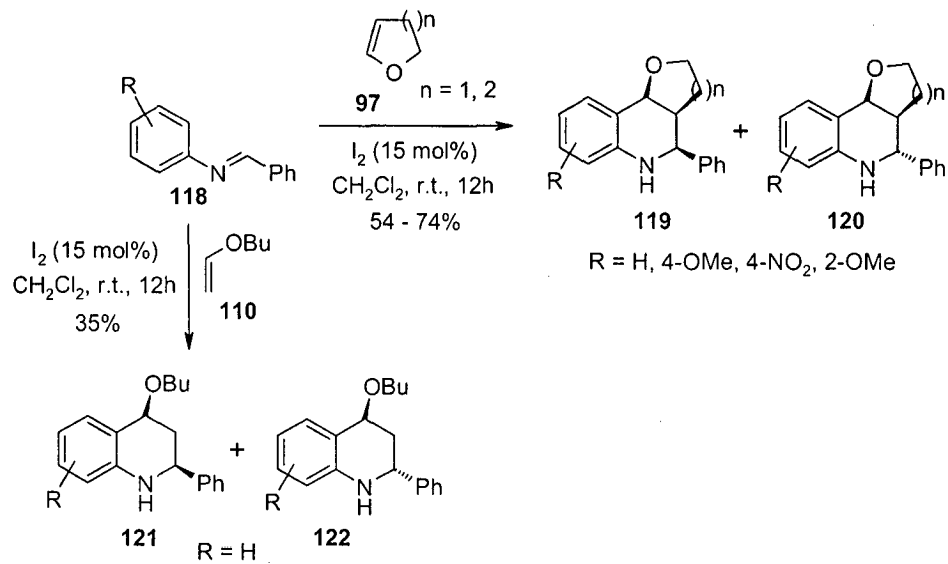
Ar = C_6H_5, 4-FC_6H_4, 4-$MeOC_6H_4$, 4-$NO_2C_6H_4$, 4-BrC_6H_4, 3-BrC_6H_4, 2,3,$Cl_2C_6H_3$, 3,4-$Cl_2C_6H_3$, 3,4-$OCH_2OC_6H_3$, 3,4-$(MeO)_2C_6H_3$, 4-MeC_6H_4, 4-ClC_6H_4

Scheme 17

The vinyl ether **110** attacks the iodine-activated Schiff's base **112** to generate intermediate **113**. Intramolecular Friedel-Crafts cyclization followed by expulsion of

BuOH induced by iodine and subsequent air oxidation resulted in the formation of aromatized 3-arylbenzo[*f*]quinolines **116**.

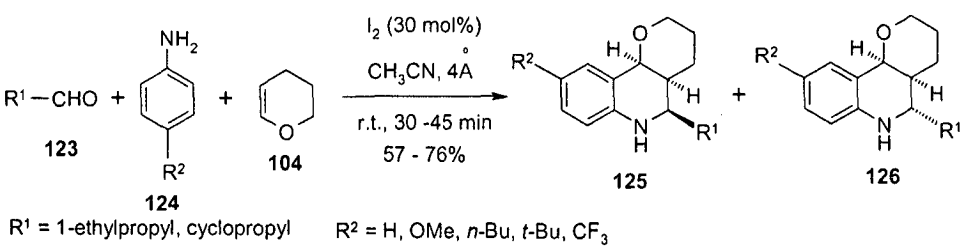
Yan and co-workers[32] reported the preparation of tetrahydroquinolines *via* iodine-mediated imino-Diels-Alder reaction (Scheme 18).



Scheme 18

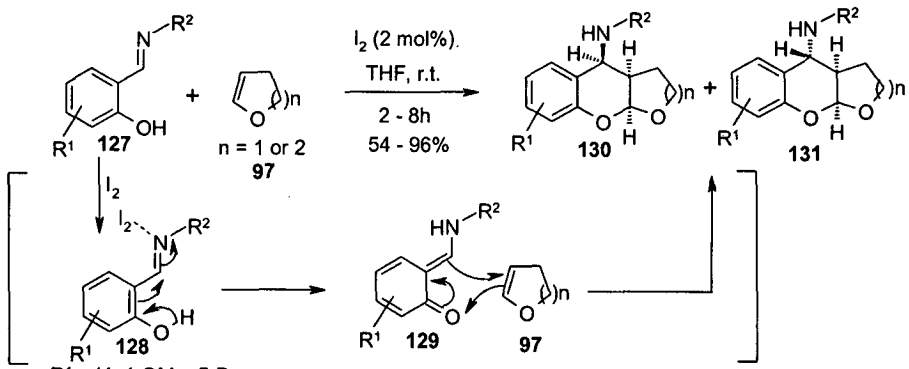
The reaction of imine **118** with cyclic ethers **97** or acyclic ether **110** afforded the respective tetrahydroquinoline derivatives as a mixture of cis- and trans-isomers.

Rai *et al.*[33] described the iodine-catalyzed one-pot, three component synthesis of pyranoquinolines from 3,4-dihydro-2*H*-pyran and in situ generated imines *via* aza-Diels-Alder reaction (Scheme 19).



Scheme 19

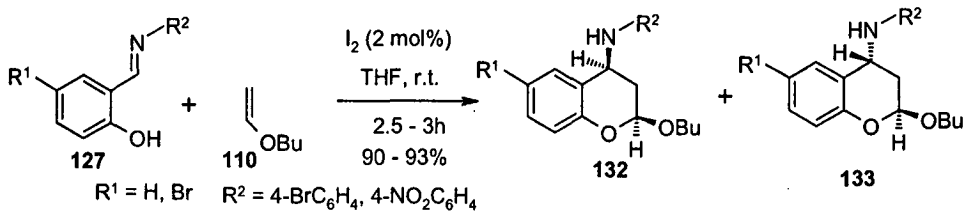
Wang and co-workers[34] prepared several cis-fused pyranobenzopyrans and furanobenzopyrans by reacting *o*-hydroxybenzaldimines **127** with 3,4-dihydro-2*H*-pyran (DHP) or 2,3-dihydrofuran (DHF) **97** (Scheme 20).



Scheme 20

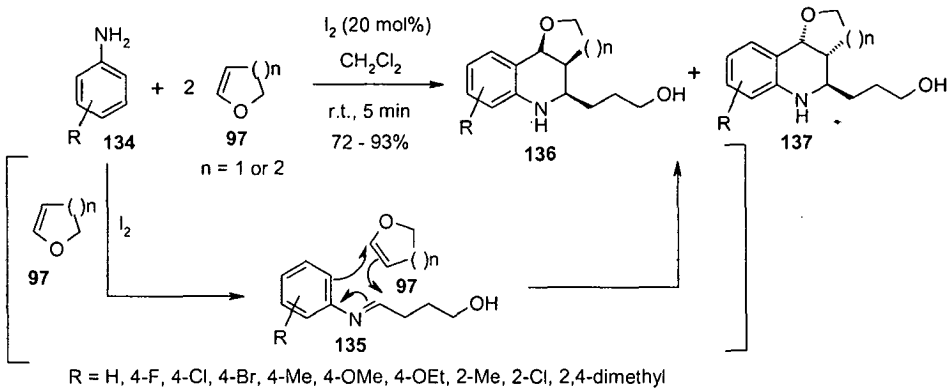
Iodine promotes the transformation of *o*-hydroxybenzaldimines **127** to oxadiene **129** which underwent [4+2] cycloaddition with **97** to furnish the corresponding pyranobenzopyrans or furanobenzopyrans as a mixture of diastereomers in 54 – 96 % yields.

Analogously, the reaction of **127** with acyclic vinyl ether **110** gave the respective 2-butoxy-4-*N*-arylaminobenzopyrans as a mixture of diastereoisomers **132** and **133** in excellent yields (Scheme 21).



Scheme 21

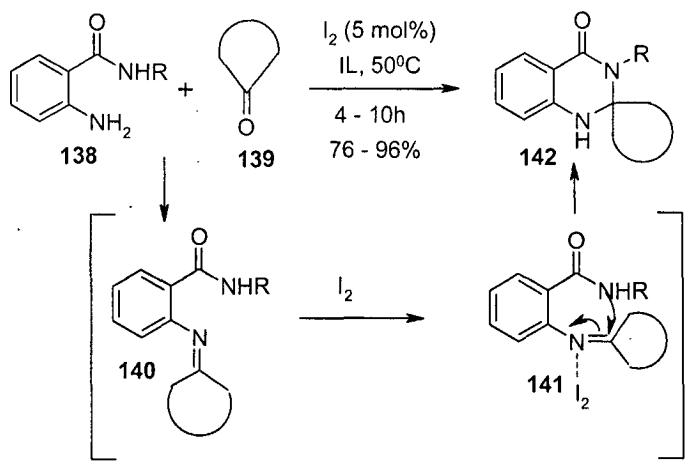
Lin *et al.*[35] demonstrated an efficient method for the preparation of 1,2,3,4-tetrahydroquinolines *via* an iodine-mediated domino reaction of anilines with cyclic enol ethers under mild reaction conditions (Scheme 22).



Scheme 22

2-Azadienes **135** formed in situ by reacting cyclic enol ethers **97** with anilines **134** underwent aza-Diels-Alder reaction with another molecule of cyclic enol ethers **97** in presence of iodine which acts as a mild Lewis acid to form the corresponding tetrahydroquinolines as a mixture of endo/exo isomers.

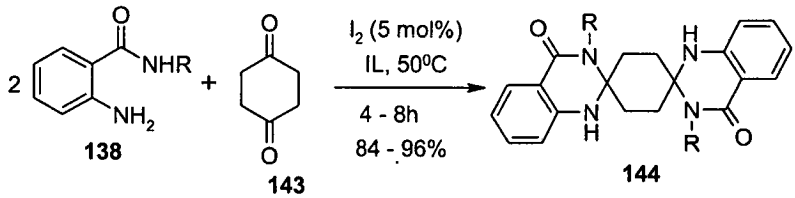
Wang and co-workers[36] reported the synthesis of quinazoline-4-(1*H*)-one derivatives in high yields catalyzed by iodine in ionic liquids (Scheme 23).



Scheme 23

Intramolecular nucleophilic attack of amino group on iodine-activated Schiff's base **141** formed by reacting **138** with **139** gave the desired cyclized product **142**.

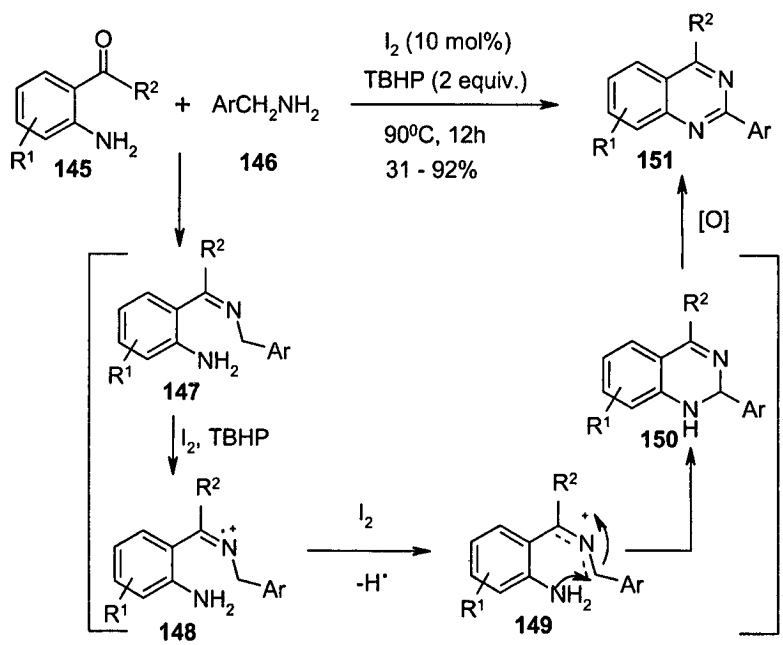
Similarly, dispirocyclic compounds containing quinazolin-4-(1*H*)-one derivatives **144** were prepared by reacting cyclohexane-1,-dione **143** with two molecules of **138** (Scheme 24).



R = H, 4-MeOC$_6$H$_4$(CH$_2$)$_2$, 4-FC$_6$H$_4$, C$_6$H$_5$, 4-MeC$_6$H$_4$, C$_6$H$_5$CH$_2$, 4-MeOC$_6$H$_4$CH$_2$, C$_6$H$_5$(CH$_2$)$_2$

Scheme 24

Zhang *et al.*[37] developed a novel tandem approach for the synthesis of various 2-phenylquinazolines (Scheme 25).



R^1 = H, 5-Cl, 5-Br, 4,5-(OMe)$_2$

R^2 = H, C$_6$H$_5$, 4-FC$_6$H$_4$, 4-BrC$_6$H$_4$, 4-MeC$_6$H$_4$, 2,5-Me$_2$C$_6$H$_3$, *n*-Bu, hexadecyl, *i*-Pr, *t*-Bu, cyclopropyl, cyclopentyl, 4-chlorostyryl

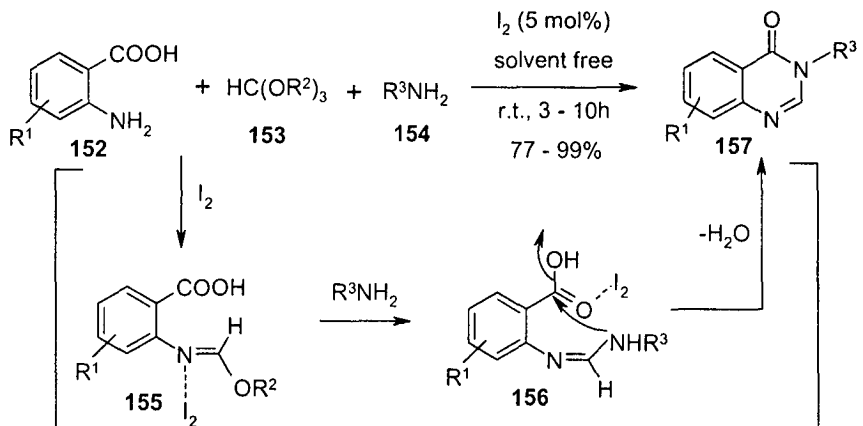
Ar = 4-MeC$_6$H$_4$, 3-MeC$_6$H$_4$, 2-MeC$_6$H$_4$, 4-MeOC$_6$H$_4$, 4-ClC$_6$H$_4$, 4-FC$_6$H$_4$, 4-CF$_3$C$_6$H$_4$, naphthalen-1-yl

Scheme 25

Oxidation of in situ generated Schiff's base **147** resulted in the formation of intermediate **149** *via* SP^3 C-H functionalization.[38,39] Intramolecular cyclization of **149**

and subsequent oxidation of the resultant intermediate **150** provided the respective 2-phenylquinazoline derivatives **151**.

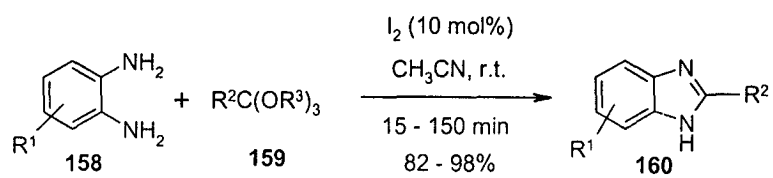
Wang and co-workers[40] accomplished the one-pot synthesis of 3,4-dihydroquinazolin-4-ones *via* three component reaction of anthranilic acids, ortho esters and amines using 5 mol% of iodine under solvent-free conditions (Scheme 26).



Scheme 26

Iodine facilitates the formation of imidic ester intermediate **155** by reacting anthranilic acids **152** with ortho esters **153** which, react rapidly with amines **154** to generate amidine intermediate **156**. Intramolecular attack of the amino group at iodine activated carbonyl carbon afforded the respective cyclized products **157**.

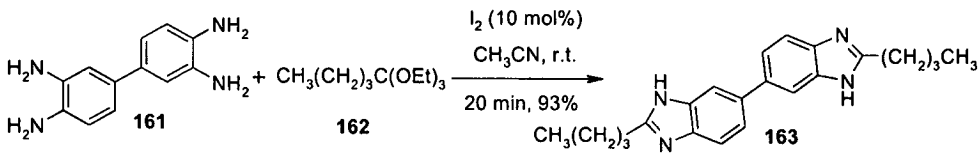
Zhang and co-workers[41] developed an efficient route for the synthesis of 2-substituted benzimidazoles (Scheme 27).



Scheme 27

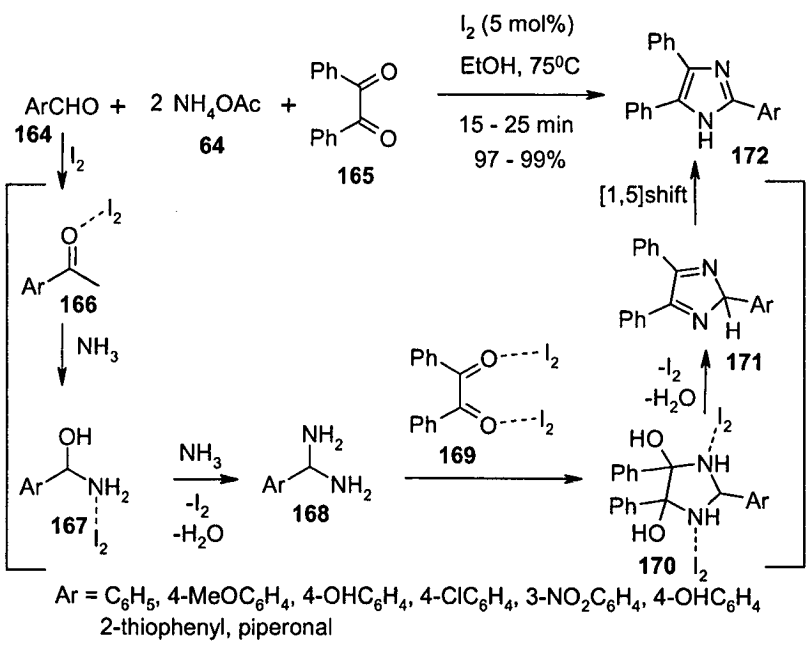
Condensation of 1,2-phenylene diamines **158** with orthoesters **159** in presence of 10 mol% of iodine afforded the respective 2-substituted benzimidazoles **160** in high to excellent yield.

Under similar reaction conditions, bis-benzimidazole **163** was prepared by reacting 3,3'-diaminobenzidine **161** with two equivalents of triethyl orthovalerate **162** in excellent yield (Scheme 28).



Scheme 28

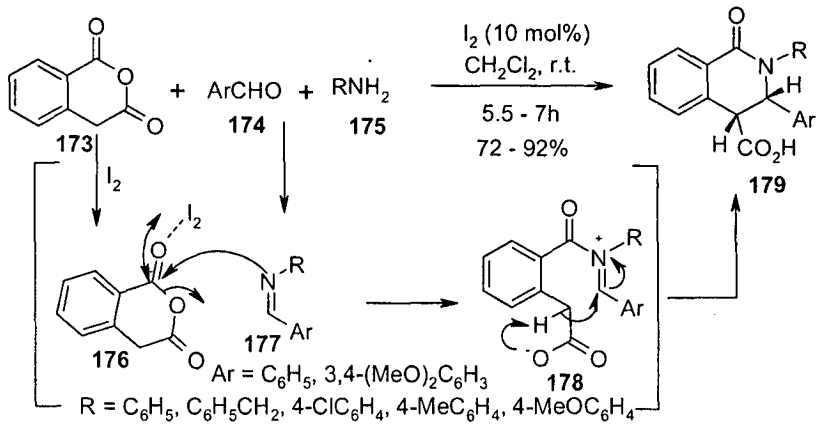
Kidwai and co-workers[42] accomplished the one-pot synthesis of 2,4,5-trisubstituted & 1,2,4,5-tetrasubstituted imidazoles using 5 mol% iodine (Scheme 29).



Scheme 29

Iodine catalyzes the reaction by bonding with the carbonyl oxygen which, facilitate the formation of a diamine intermediate **168** and then condenses with the iodine-activated carbonyl carbon of 1,2-diketone **169** to form the cyclized intermediate **170**. Dehydration of **170** followed by [1,5] sigmatropic shift of the intermediate **171** afforded the respective imidazoles **172**.

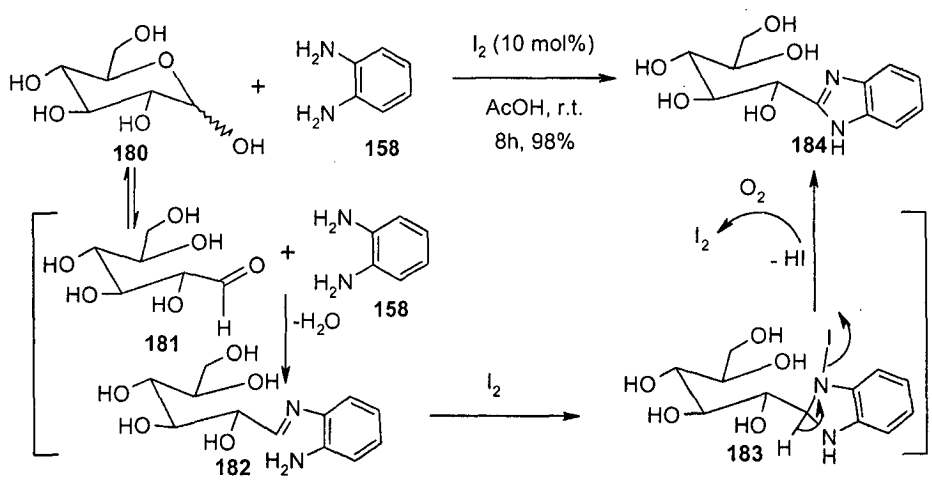
Yadav and co-workers[43] described the iodine-promoted one-pot approach for the synthesis of cis-1-oxo-1,2,3,4-tetrahydroisoquinoline-4-carboxylic acid by the three component reaction of aldehydes, amines and homophthalic anhydride under mild reaction conditions (Scheme 30).



Scheme 30

In situ generated Schiff's base **177** attacks the iodine-activated carbonyl carbon of **176** to form intermediate **178** which, gets converted to the desired products **179**.

Lin *et al.*[44] achieved a convenient method for the synthesis of aldo-benzimidazoles and aldo-naphthimidazoles using iodine as an oxidant or promoter (Scheme 31 & 32).

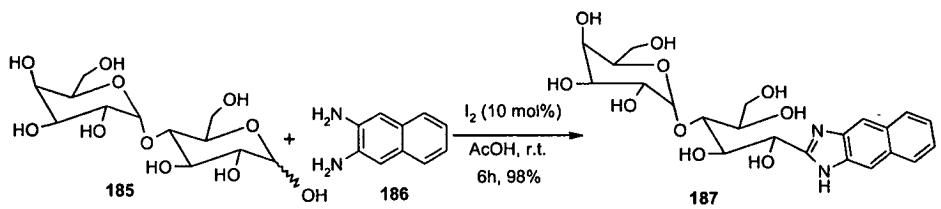


Scheme 31

The condensation of **181** with **158** forms the Schiff's base **182** which, subsequently underwent intramolecular nucleophilic addition by the other amino group and *N*-

iodination of the imine moiety of Schiff's base to give **183.** Finally, the lose of HI provided the aldo-imidazole **184** in excellent yield (Scheme 31).

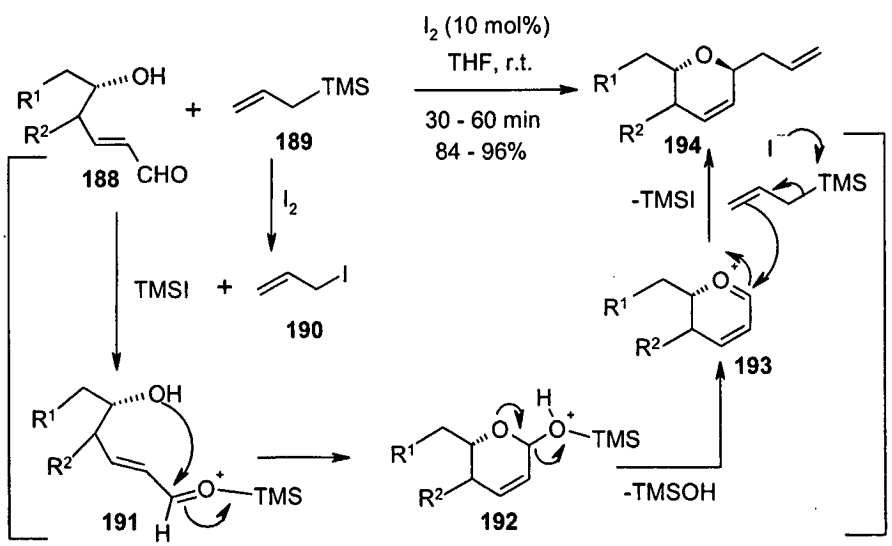
The oxidative condensation of **185** with 2,3-naphthalenediamine **186** using 10 mol% iodine as the promoter gave the aldo-naphthimidazole **187** in 98% yield (Scheme 32).



Scheme 32

Similarly, the various other aldo-benzimidazoles and aldo-naphthimidazoles have been prepared by condensing different aldoses i.e. mono-, di- and tri-saccharides with *o*-phenylenediamines and 2,3-naphthalenediamine using iodine as a catalyst.

Mohapatra and co-workers[45] reported the synthesis of trans-2,6-disubstituted-3,-dihydropyrans by treating δ-hydroxy-α,β-unsaturated aldehydes with allyltrimethyl silane in presence of 10 mol% of iodine (Scheme 33).



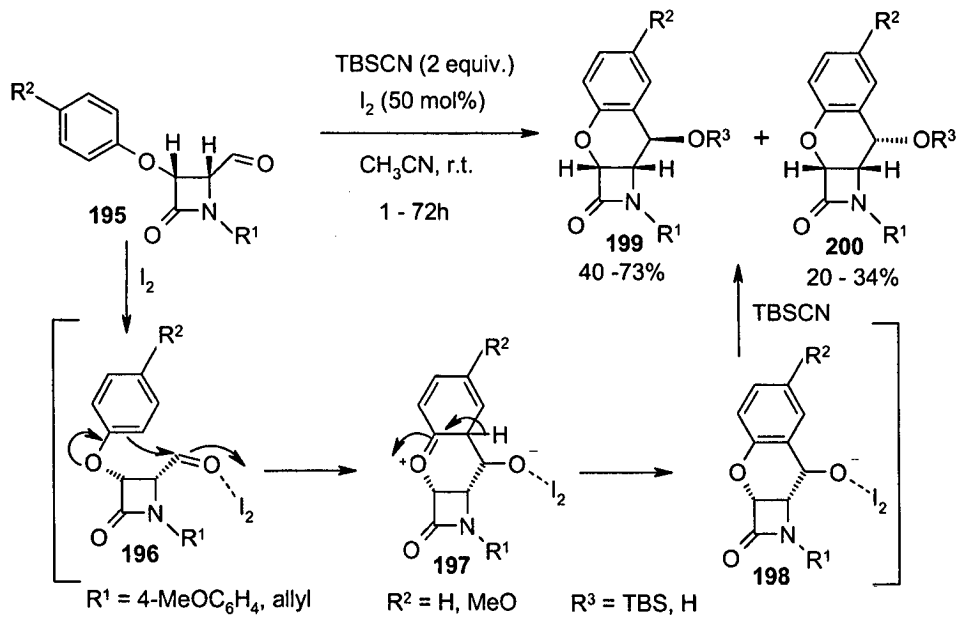
R^1 = 4-$MeOC_6H_4CH_2OCH_2$, 4-$MeOC_6H_4CH_2O(CH_2)_2CHOMe$, $C_6H_5CH_2OCH_2$, OAc, $OCH_2C_6H_5$, OTBDPS

R^2 = H, Me, OAc, $C_6H_5CH_2O$, OMe, OEt, 4-$MeOC_6H_4CH_2O$

Scheme 33

Activation of the carbonyl group of **188** by in situ formed TMSI generates intermediate **191**. Nucleophilic attack of hydroxyl group on activated carbonyl carbon followed by expulsion of trimethyl silyl hydroxide forms the oxonium intermediate **192**. The final products **194** were formed by the initial attack of I^- on the trimethyl silyl group to generate nucleophile which, attacks the oxonium intermediate **193**.

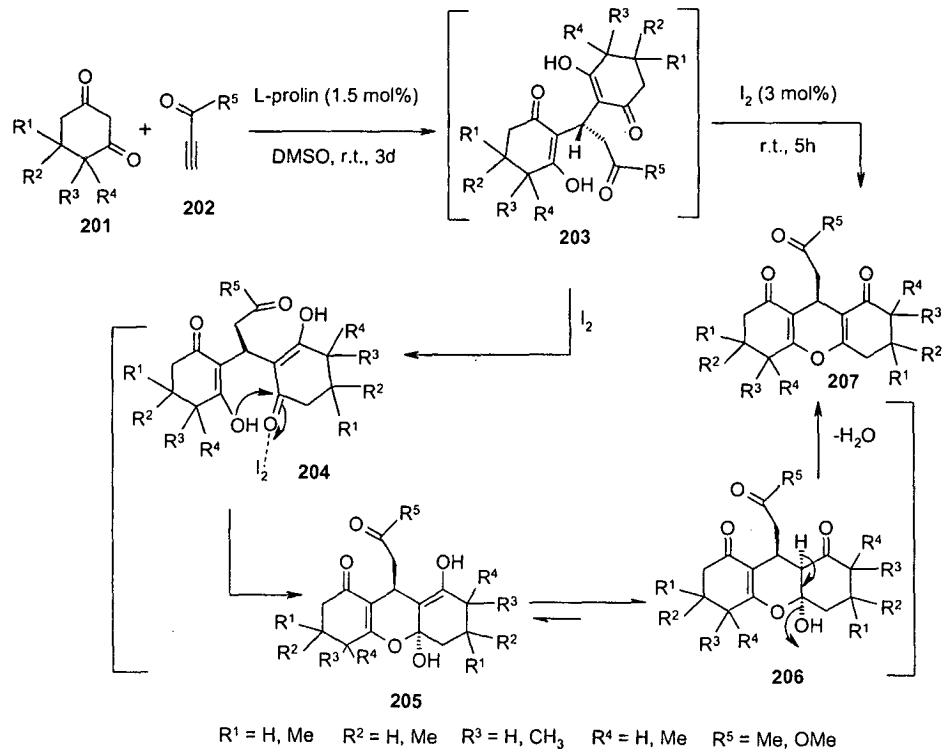
Alcaide and co-workers[46] demonstrated the iodine-mediated reaction of 3-aryloxy-β-lactams to give the cyclized product β-lactam-fused chromenes as a mixture of cis- and trans-isomers (Scheme 34).



Scheme 34

Iodine promotes the reaction by coordinating with the oxygen of aldehydic carbonyl group which, underwent intramolecular Friedel-Crafts cyclization to form six-membered Wheland-type intermediate **197**. Deprotonation followed by iodine-silicon exchange generates the desired products.

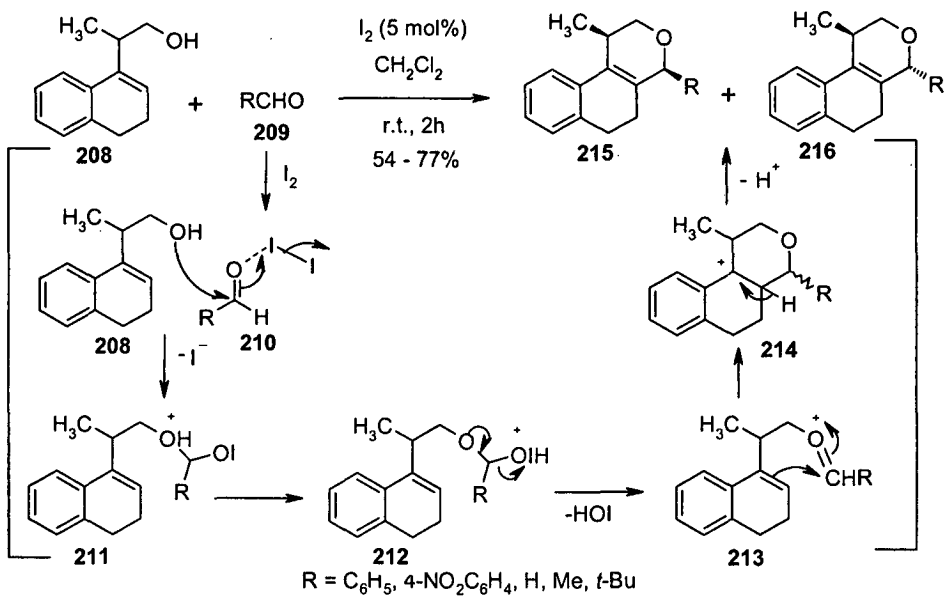
Luna *et al.*[47] described the synthesis of 9-substituted-1,8-dioxooctahydroxanthenes *via* sequential, tandem Michael-iodine-catalyzed cyclization (Scheme 35).



Scheme 35

Activation of carbonyl group of **203** by I_2 followed by intramolecular nucleophilic attack of the other enol moiety resulted in the formation of hemiketal intermediate **205** which exist in its keto form **206**. Elimination of a water molecule afforded the corresponding octahydroxanthene **207**.

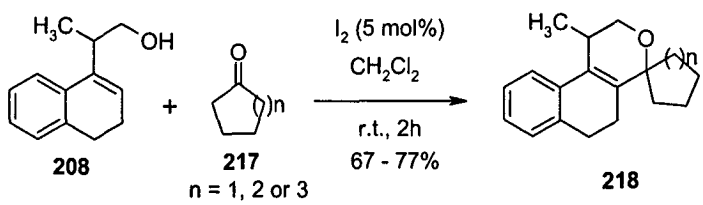
Silva Jr. and Quintiliano[48] described the iodine-induced Prins cyclization for the preparation of hexahydrobenzo[*f*]isochromenes (Scheme 36).



Scheme 36

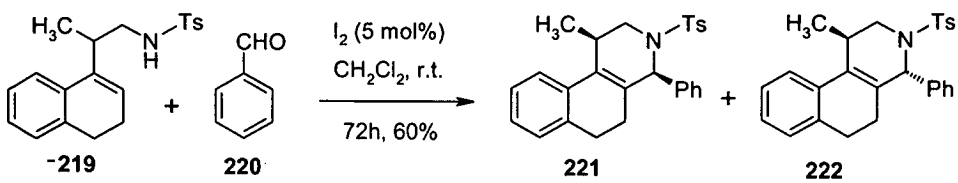
Homobenzylic alcohol **208** attacks the iodine-activated aldehydes **210** to form intermediate **211**. Elimination of HOI from **212** and subsequent intramolecular cyclization afforded the six-membered cyclic intermediate **214**. Loss of proton gave the compounds **215** or **216**. When R = Ph, mixture of diastereomers (cis/trans) were obtained in 77% yield while a single diastereomers were formed when R = $NO_2C_6H_4$ or Me in 68% and 78% yield, respectively.

Similarly, the Prins cyclization of **208** with the cyclic ketones **217** provided the spiro compounds **218** in good yields (Scheme 37).



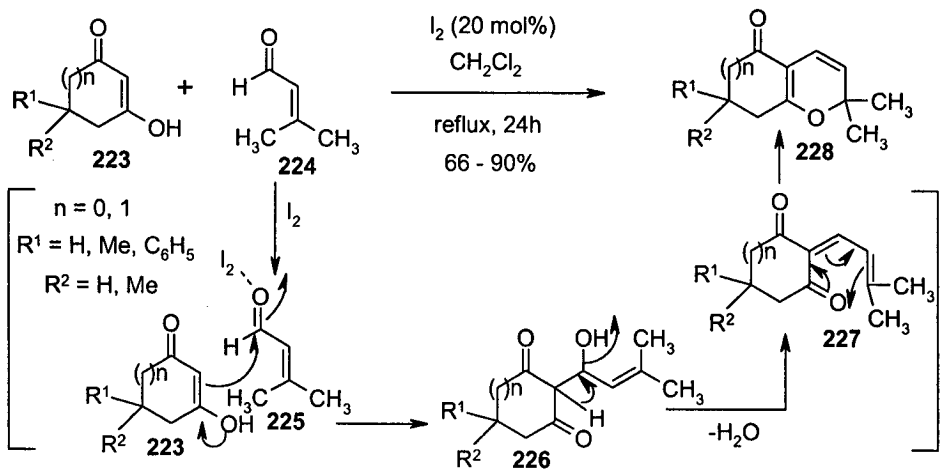
Scheme 37

The aza-Prins cyclization of **219** with **220** using 20 mol% of iodine afforded the mixture of product **221** and **222** in 60% yield and has a structure similar to Populene D[49] (Scheme 38).



Scheme 38

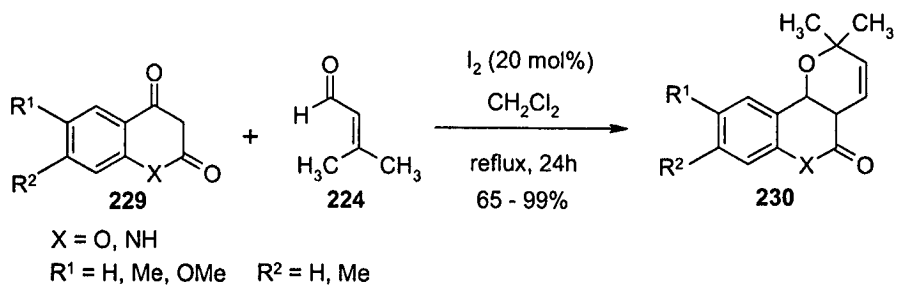
Jung *et al.*[50] developed a one-pot method for the synthesis of 2*H*-pyrans *via* iodine-mediated domino Knoevenagel - 6π-electrocyclization reactions (Scheme 39 & 40).



Scheme 39

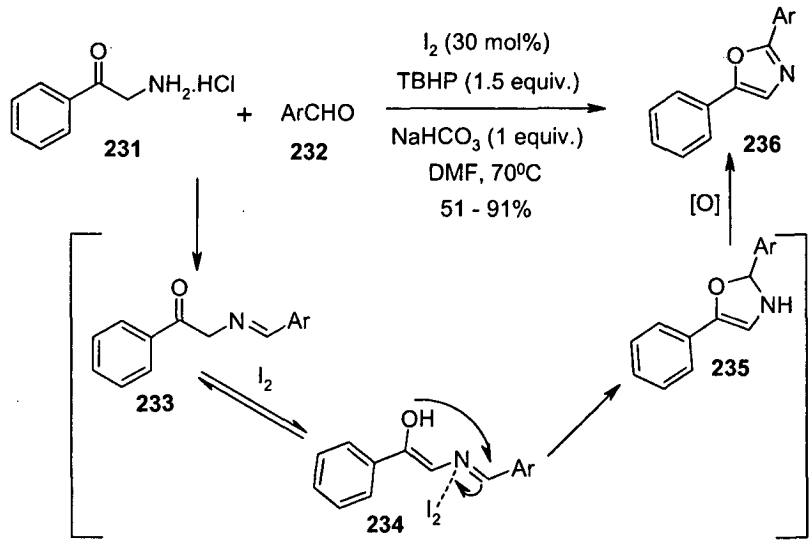
The dimedone **223** attacks the iodine activated aldehydes **225** to generate intermediate **226** which readily eliminates water on heating to give **227**. Electrocyclization of **227** gave the cycloadduct **228** (Scheme 39).

When the reactions were carried out by treating compounds **229** with **224**, the angular products **230** were obtained in good to excellent yield (Scheme 40).



Scheme 40

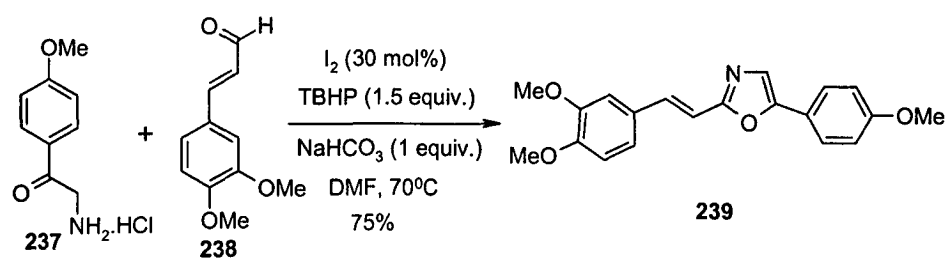
Wan *et al.*[51] demonstrated the synthesis of 2,5-disubstituted oxazoles *via* tandem oxidation cyclization protocol catalyzed by iodine (Scheme 41).



Scheme 41

The in situ generated Schiff's base **233** is in equilibrium with its enol form **234**. Intramolecular nucleophilic attack of oxygen atom on iodine activated Schiff's base **234** followed by oxidation of the resultant cyclized intermediate **235** provided the corresponding products **236**.

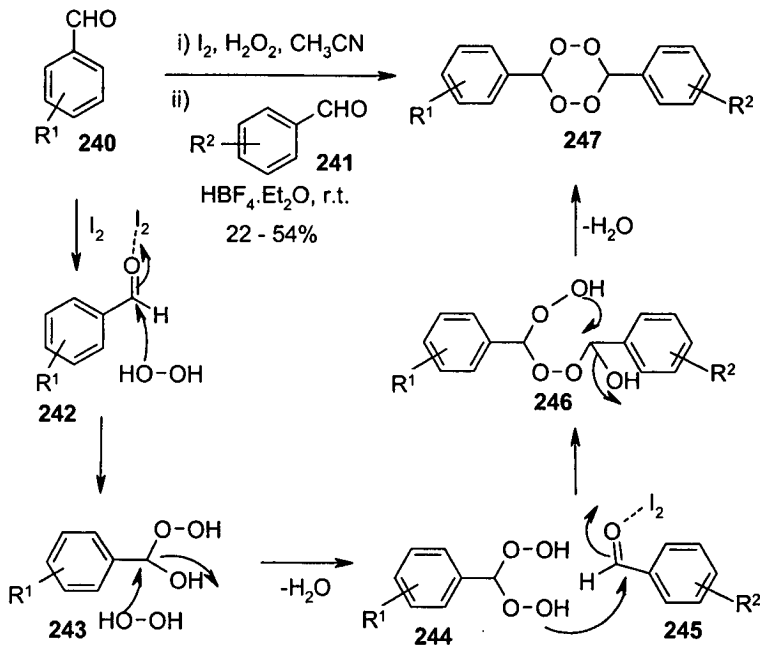
Using the above methodology, annuloline **239** – the first isolated natural product containing an oxazole sub-structure was prepared by reacting **237** with **238** in 75% yield (Scheme 42).



Scheme 42

Kumar *et al.*[52] accomplished the one-pot synthesis of symmetrically and asymmetrically substituted 3,6-diphenyl[1,2,4,5]tetraoxanes using 10 mol% iodine

(Scheme 43) and these were evaluated for *in vitro* antimalarial activity and cytotoxicity.

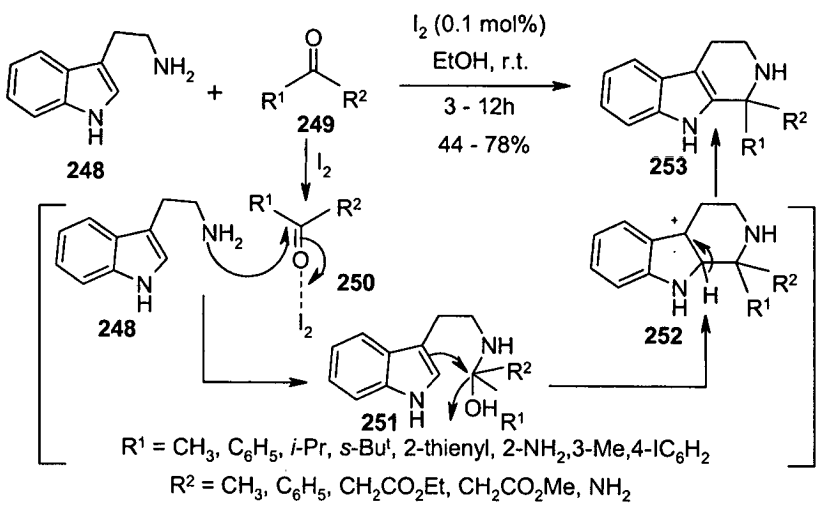


$R^1 = R^2$ = *o*-Me, *m*-Me, *p*-Me, *p*-Cl, *p*-Br, *p*-F, *p*-OMe, *p*-Et, *p*-*n*-Pr, *p*-*i*-Pr *p*-*n*-Bu, *p*-*t*-Bu, *p*-CO_2Me

Scheme 43

Hydrogen peroxide attacks the iodine-activated carbonyl carbon of aldehydes **242** to form intermediate **243** which was again attacked by H_2O_2 to generate gem-bis-hydroperoxides intermediate **244**. Condensation of **244** with other aldehydes **245** followed by intramolecular cyclization of the resultant intermediate **246** afforded the respective tetraoxanes **247**.

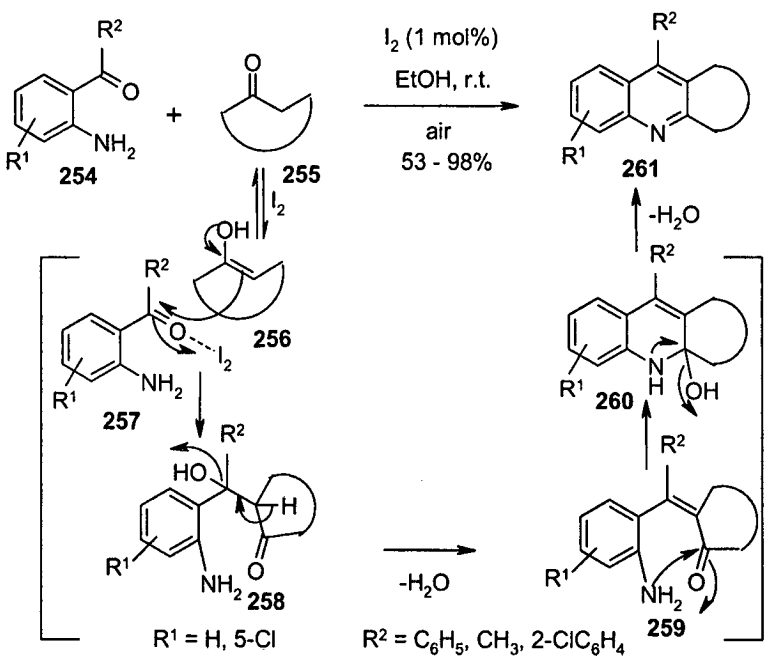
Lingam and co-workers[53] prepared various 1,1-disubstituted tetrahydro-β-carbolines *via* an iodine-induced Pictet-Spengler reaction[54] (Scheme 44).



Scheme 44

Iodine catalyzes the reaction by coordinating with the carbonyl oxygen of ketones **249** which are then attacked by the amino group of tryptamine **248** to generate the intermediate **251**. Intramolecular Friedel-Crafts cyclization of **251** followed by lose of proton yielded the desired products **253**.

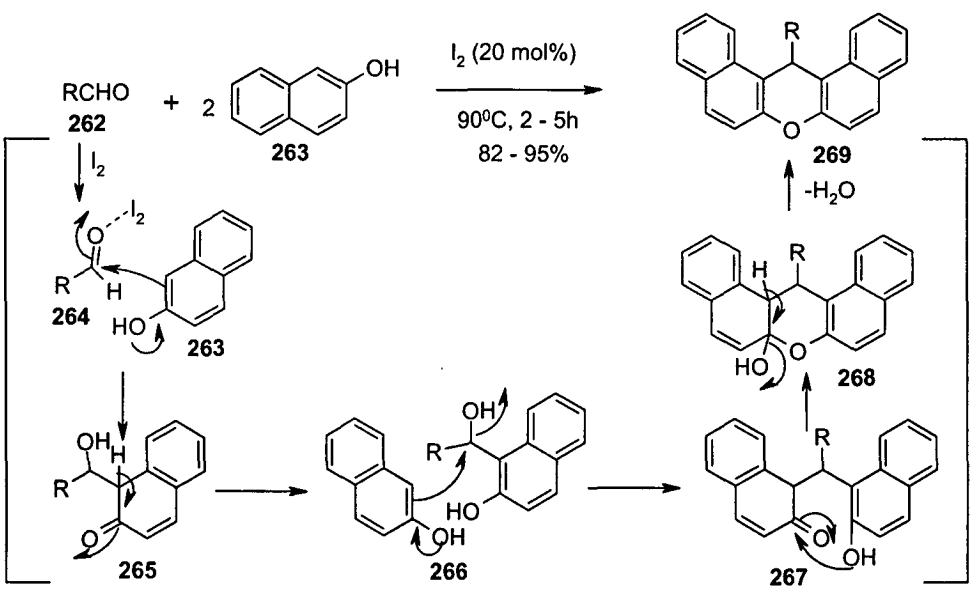
Wu and co-workers[55] reported the synthesis of quinolines *via* Friedlander annulation[56,57] (Scheme 45).



Scheme 45

In situ formed enols **256** underwent Aldol condensation with iodine-activated 2-aminobenzophenones **257** to give intermediate **258** which readily loses water to form α,β-unsaturated ketone intermediate **259**. Final ring closure by the attack of amino group on the carbonyl carbon generates the required compounds **261**.

Das and coworkers[58] described a convenient method for the synthesis of 14-aryl or alkyl-14*H*-dibenzo[*a,j*]xanthenes **269** by one-pot condensation of 2-naphthol with aromatic or aliphatic aldehydes in presence of 20 mol% iodine under solvent-free condition (Scheme 46).



R = C_6H_5, 4-MeC_6H_4, 4-EtC_6H_4, 4-OHC_6H_4, 4-$MeOC_6H_4$, 4-$CF_3C_6H_4$, 3-MeO,4-OH-C_6H_3 4-ClC_6H_4, 4-FC_6H_4, 3-$NO_2C_6H_4$, 4-$NO_2C_6H_4$, Et, *i*-Pr

Scheme 46

Friedel-Crafts alkylation of β-naphthol **263** with iodine activated aldehydes **264** followed by another Friedel-Crafts alkylation of other molecule of β-naphthol with intermediate **266** generates **267**. Intramolecular cyclization and subsequent loss of water gave the required aromatized products **269**.

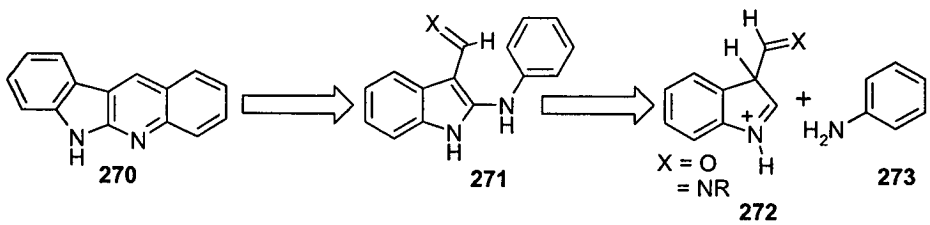
Pash and co-workers[59] reported the synthesis of 14-aryl or alkyl-14*H*-dibenzo[*a,j*]xanthenes using the above methodology but instead of 20 mol% of catalyst, 2.5 mol% of iodine has been used.

Objective:

The objective of the present study is to explore the use of iodine as a catalyst for the preparation of 6*H*-indolo[2,3-*b*]quinolines and further to evaluate the biological activities of these indoloquinolines.

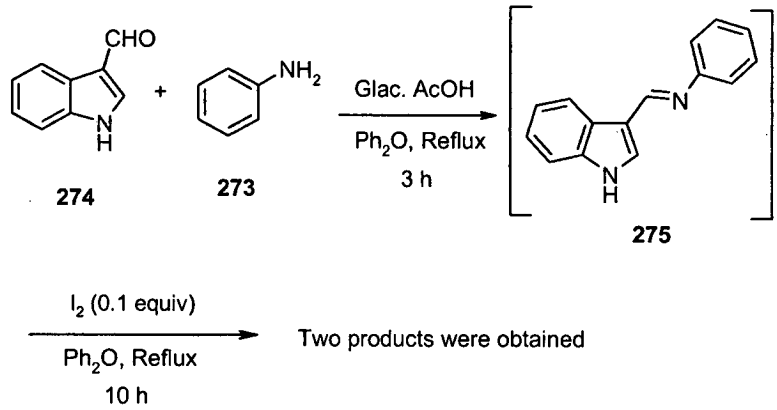
Present Work:

Our retro-synthetic analysis (Scheme 47) for the formation of 6*H*-indolo[2,3-*b*]quinoline indicated that if 3*H*-indolinium cation **272** is generated, it should be possible for nitrogen of aniline to make a nucleophilic attack followed by annulation and subsequent oxidation should lead to 6*H*-indolo[2,3-*b*]quinoline.



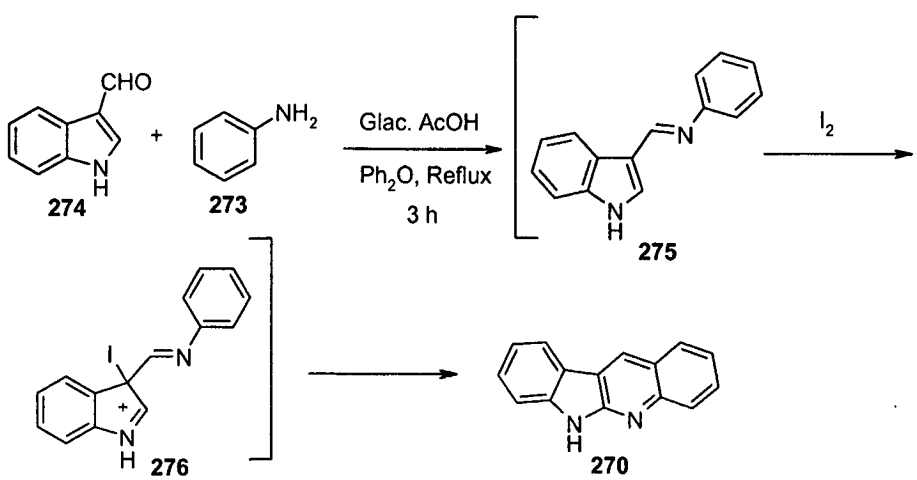
Scheme 47

To test the above hypothesis, we initially heated indole-3-carboxyaldehyde **274** with excess aniline **273** in refluxing diphenyl ether in presence of acetic acid. It was observed that only Schiff's base **275** was formed (monitored by TLC by comparison with the authentic sample prepared as per the reported[60] method). As mentioned earlier, the use of iodine has received considerable attention as an inexpensive, environmentally tolerable and readily available mild Lewis acid for different organic transformations. So, a few crystals of iodine were added to the above reaction mixture containing Schiff's base and the heating was continued for further 10 hours (monitored by TLC). This resulted in the formation of two products which were separated by column chromatography (Scheme 48).



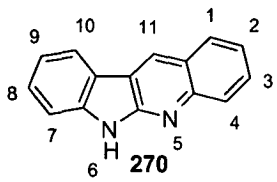
Scheme 48

The less polar product did not melt up to 300^{0}C. In its IR spectrum, it showed a strong band at 3142 cm^{-1} which could be due to N-H stretching. Its PMR (Fig. 1) indicated the presence of 9 protons in the ratio of 1:3:1:1:1:1:1 in the aromatic region from δ 7.20 to 11.70 ppm. The downfield signal for single proton at δ 11.70 was assigned to the NH proton of the indole ring. The downfield nature of the signal suggested that indeed quinoline ring was formed. It was envisaged that once the iodine was added to the Schiff's base, there might be the formation of 3-iodo-indolinium cation as suggested in our retro synthetic pathway (Scheme 47). Further the nucleophilic attack of aniline on 3-iodo-indolinium cation followed by annulation and subsequent oxidation may lead to the formation of 6*H*-indolo[2,3-*b*]quinoline (Scheme 49).



Scheme 49

Assuming that expected indoloquinoline **270** was formed, we assigned the remaining signal in the PMR spectrum. The downfield singlet observed at δ 9.05 was assigned to proton on C-11. The doublet at δ 8.27 (J = 7.8 Hz) was assigned to proton on C-10. The doublets seen at δ 7.98 & 8.11 were assigned to the proton on C-1 and C-4 respectively. The ddd observed at δ 7.72 was attributed to proton at C-3. The multiplet seen at δ 7.46 – 7.57 for 3 protons could be attributed to H-2, H-7 & H-8 of indole ring. The proton seen at δ 7.27 as multiplet was assigned to H-9. Thus the PMR data fitted well for the proposed structure. So the spectral data of compound **270** was compared with the literature data[61] of 6*H*-indolo[2,3-*b*]quinoline and it matches exactly as shown in the table below.



270

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 1)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (J Hz)	No. of Protons (H)	Position	Literature Values (δ ppm)
7.27	m	-	1	H-9	7.27
7.46-7.57	m	-	3	H-2, H-7 & H-8	7.48-7.53
7.72	ddd	8.1, 7.2 & 0.9	1	H-3	7.72
7.98	d	8.4	1	H-1	7.98
8.11	d	8.1	1	H-4	8.11
8.27	d	7.8	1	H-10	8.26
9.05	s	-	1	H-11	9.05
11.70	s	-	1	-NH	11.70

The structure was further confirmed by ^{13}C NMR, DEPT studies and HRMS as mentioned below.

^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 2)

δ 111.39 (C-7), 118.37 (C-10b), 120.14 (C-9), 120.76 (C-11a), 122.29 (C-10), 123.20 (C-2), 124.15 (C-10a), 127.45 (C-1), 128.01 (C-11), 128.67 (C-4), 129.13 (C-3 & C-8), 141.93 (C-6a), 146.79 (C-4a) and 153.36 (C-5a).

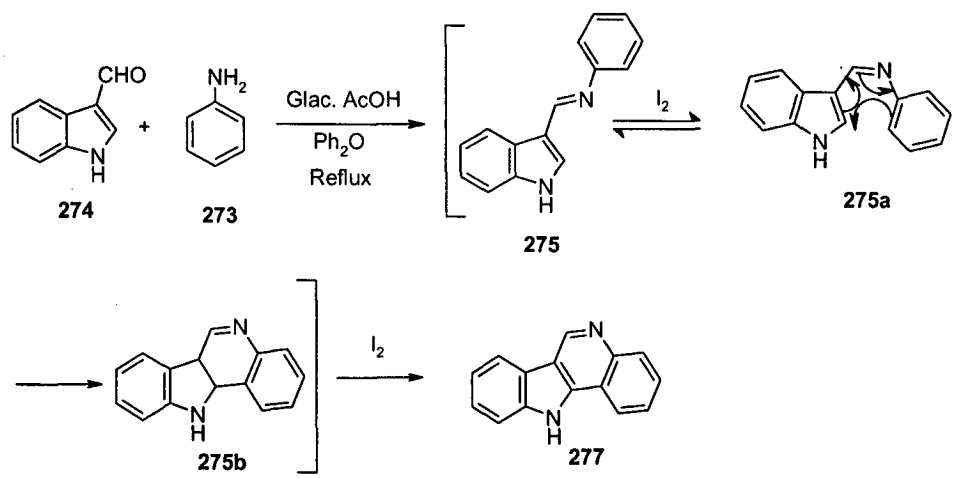
IR (KBr): υ_{max} = 3142, 3090, 1614, 1580, 1460, 1408, 1329, 1230, 1126, 908, 820, 787, 737, 696 cm^{-1}

HRMS: *m/z* $[M+H]^+$ 219.0926 (calcd for $C_{15}H_{11}N_2$, 219.0922).

Melting Point: >300 ^{0}C; Lit.[61] 342-346 ^{0}C.

Based on the mode of formation and physical & spectroscopic data which were in full agreement with the literature data,[61] the structure **270** was assigned to the less polar solid. The yield was found to be 23% (based on 3-formyl indole).

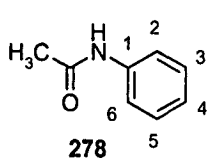
The other possibility was that, there might be the isomerization of Schiff's base from *E*- to *Z*-isomer on addition of iodine which then could undergo electrocyclization and subsequent oxidation to provide 11*H*-indolo[3,2-*c*]quinoline (Scheme 50).



Scheme 50

However, this was not to be the case as the NMR data did not match with that of 11*H*-indolo[3,2-*c*]quinoline[60] **277**. Surprisingly, the melting point and spectral data matched very well with that of acetanilide **278**.

Acetanilide 278



IR (KBr): υ_{max} = 3294, 1665, 1599, 1435, 1369, 1323, 756 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 3)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.04	s	-	3	-NHCOC<u>H</u>$_3$
7.01	m	-	1	H-4
7.28	t	7.8	2	H-3 & H-5
7.58	d	7.8	2	H-2 & H-6
9.91	s	-	1	-NH

^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 4)

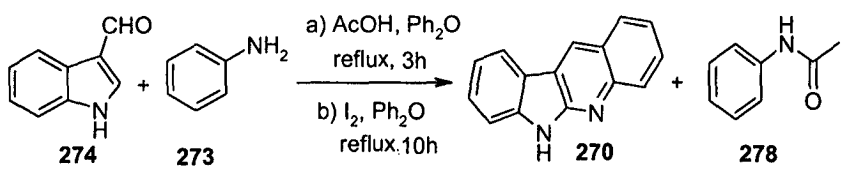
δ 24.4 (-NHCO<u>C</u>H$_3$), 119.3 (Ar-<u>C</u>H), 119.4 (Ar-<u>C</u>H), 123.4 (Ar-<u>C</u>H), 129.1 (Ar-<u>C</u>H, 2 × C), 139.8 (Ar-<u>C</u>H), 168.7 (-<u>C</u>=O).

Melting Point: 114^{0}C (Commercially available acetanilide – 113 – 115^{0}C).

Based on the spectroscopic data and similarity of melting point with that of commercially available acetanilide, the structure **278** was assigned to the more polar solid.

In order to optimize the reaction conditions, the reaction was studied with different concentrations of these reagents i.e. iodine and aniline as shown in Table 1.

Table 1: Reaction of indole-3-carboxyaldehyde with aniline



274 273 270 278

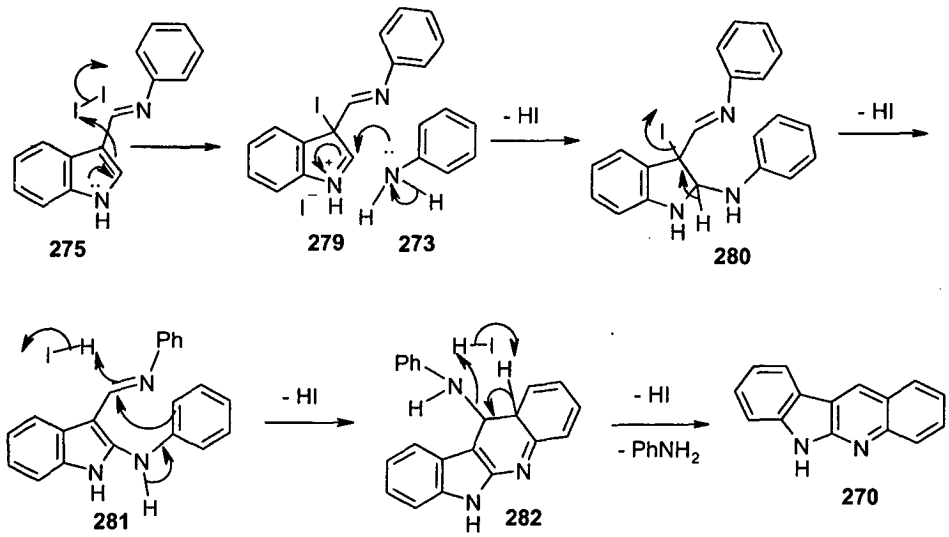
Entry	I_2 (Equiv.)	105 (Equiv.)	Yield (%) of 102
1	0.0	2	0
2	0.05	2	20
3	0.1	2	23
4	0.2	2	21
5	0.5	2	18
6	1.0	2	12
7	0.1	1	0

8	0.1	1.5	19
9	0.1	3	23
10	0.1	4	23

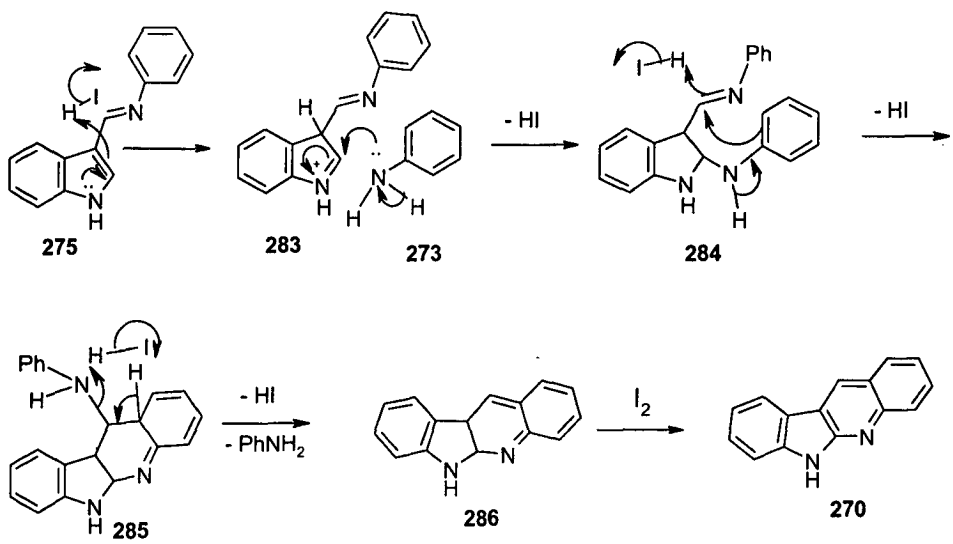
Initially the reactions were carried out by keeping the concentration of aniline constant i.e. two equivalents and varying the concentration of iodine from 0 to 1 equivalent (Entries 1 – 6). It was observed that in the absence of iodine (entry 1), product **270** was not formed and only acetanilide **278** was formed (monitored by TLC). When the reaction was carried out with 0.05 equivalent of iodine (entry 2), 20% of the product **270** was obtained while the yield of the product was maximum (23%) when the concentration of iodine was 0.1 equivalent (entry 3). As the concentration of the iodine is increased from 0.2 to 1.0 equivalent (entries 4 – 6) there was gradual decrease in the yield of the product **270**.

Secondly the reactions were carried out by varying the amount of aniline **273** from 1 to 4 equivalents and keeping the concentration of iodine as 0.1 equivalent (Entries 7 – 10). It was observed that with one equivalent of aniline **273**, no product **270** was forming (entry 7) and when the concentration of aniline was increased from 1 to 1.5 equivalent, product **270** was obtained in 19% yield (entry 8). The maximum yield of the product **270** was obtained when the concentration of aniline was two or more than two equivalents (entries 3, 9 & 10).

Based on the above observations, the optimum concentrations of iodine and aniline were found to be 0.1 and 2 equivalents respectively. The probable mechanisms for the formation of the **270** are given in scheme 51 & 52 respectively.



Scheme 51



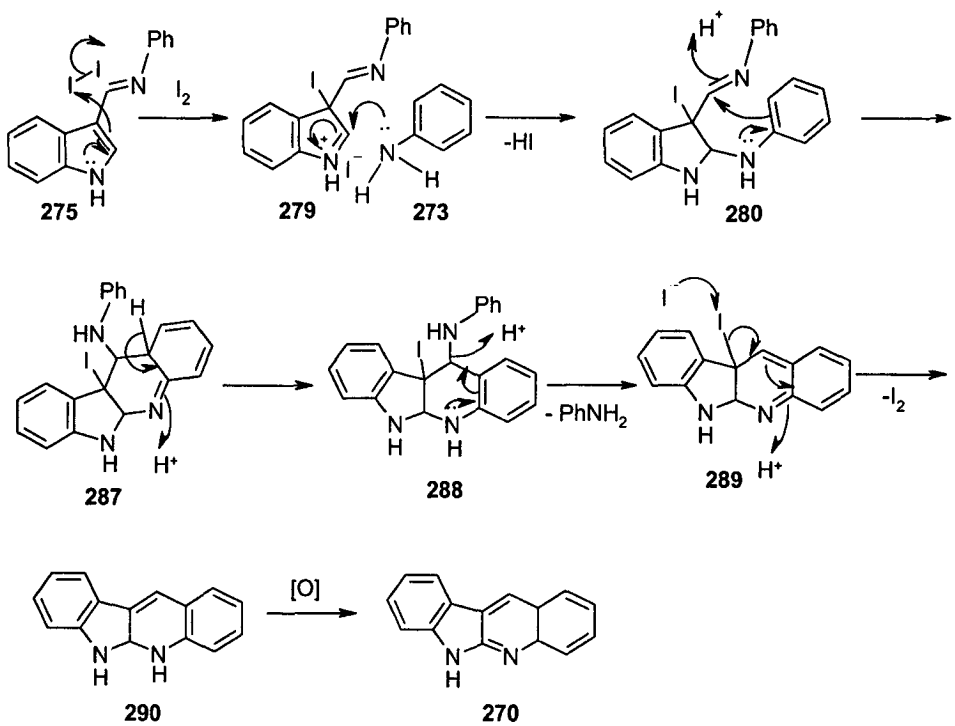
Scheme 52

As per the above two probable mechanisms, we came to the conclusion that iodine initializes the reaction by electrophilic attack on Schiff's base **275** to generate 3-iodo-indolinium cation **279.** Nucleophilic attack by nitrogen of the aniline **273** on compound **279** may give 2-*N*-phenyl substituted indole **280**. Annulation of **280** followed by expulsion of aniline provides 6*H*-indolo[2,3-*b*]quinoline **270** (Scheme 51). Further the HI generated from iodine may lead to the formation of 3-hydro-

indolinium cation **283** which is then attacked by nitrogen of aniline **273** to give the compound **284**. Annulation of **284** followed by expulsion of aniline provides dihydro-6*H*-indolo[2,3-*b*]quinoline **286** which subsequently gets oxidized by I_2 to give 6*H*-indolo[2,3-*b*]quinoline **270** (Scheme 52).

As seen in the probable mechanisms described in schemes 51 & 52, I_2 initializes the reaction and HI generated from I_2 in the reaction media may have catalyzed the reaction further to give the desired product **270**. So, we carried out the reaction using HI instead of I_2 as a catalyst, but the formation of product was not observed (monitored by TLC). As the reaction did not yield the desired product using HI, the probable mechanism described in scheme 52 was discarded.

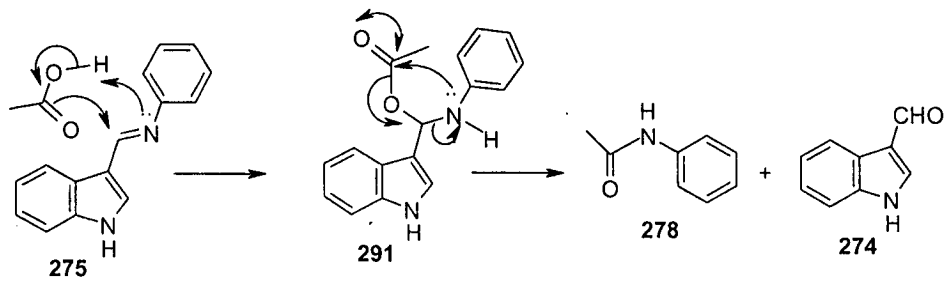
If we considered the mechanism described in scheme 51, it was observed that one equivalent of iodine is required for the formation of product **270**. But the results described in table 1 showed that the catalytical amount of iodine i.e. 0.1 equivalent gives the better yield. So the mechanism described in scheme 51 was also discarded and new plausible mechanism for the formation of **270** was postulated (Scheme 53).



Scheme 53

Thus, initial electrophilic attack of iodine on Schiff's base **275** generates 3-iodo-indolinium cation **279**. Subsequent nucleophilic attack by aniline on **279** will lead to 2-*N*-phenyl substituted indole **280**. Intramolecular electrophilic substitution leading to annulated structure **288** *via* **287** followed by expulsion of aniline may form **289**. Further, departure of iodine followed by oxidation could lead to aromatized heterocycle **270** (Scheme 53).

The probable mechanism for the formation of the by product acetanilide **278** is shown in scheme 54.

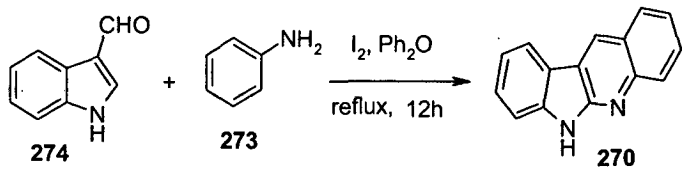


Scheme 54

As the acetanilide **278** was forming due to the presence of acetic acid in the reaction medium, the reaction was carried out in absence of it. It was observed that the yield of the product was increased from 23 to 45% but the time required for the formation of the Schiff's base was 18 hours instead of 3 hours in presence of acetic acid and the total time required for the formation of product **270** was 26 hours instead of 13 hours.

As acetic acid was used for the formation of Schiff's base **275**, we thought of using iodine itself as an agent for its formation and further transformations. Thus the mixture of indole-3-carboxyaldehyde **274**, aniline **273** and iodine were refluxed in diphenyl ether instead of adding the iodine later after the formation of Schiff's base **275**. To our delight, the reaction was complete in 12 hours with almost the same yield. As the yield of the product had increased in the absence of acetic acid, we studied the influence of the amount of iodine and aniline on the yield as shown in table 2.

Table 2: Reaction of indole-3-carboxyaldehyde with aniline in absence of AcOH



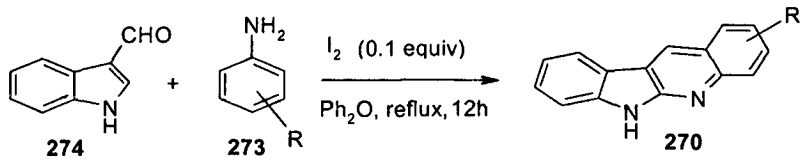
274 273 270

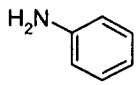
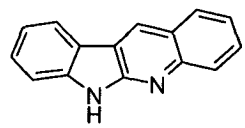
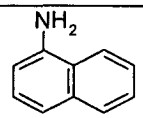
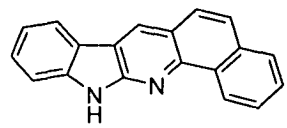
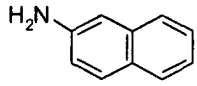
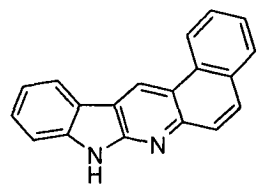
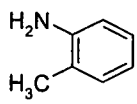
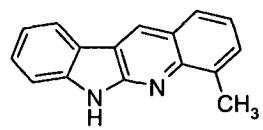
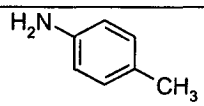
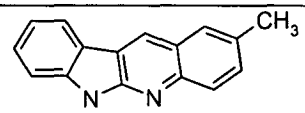
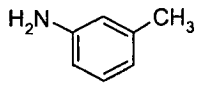
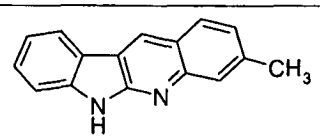
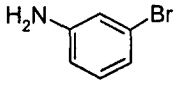
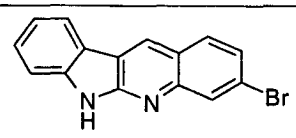
Entry	I_2 (Equiv.)	273 (Equiv.)	Yield (%) of 270
1	0.1	1	0
2	0.1	1.5	30
3	0.1	2	45
4	0.1	3	45
5	0.05	2	34
6	0.3	2	37

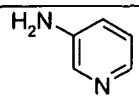
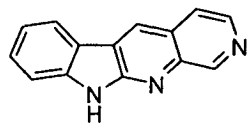
However, no further improvements in the yields were observed. The modest yield may be due to the decomposition of the Schiff's base intermediate under the reaction condition and also some of the product might be lost during purification by column chromatography due to its low solubility.

The potential biological activities[64-68] of these indoloquinolines prompted us to check the feasibility of making the library of such compounds (Table 3). As methyl substituted indolo[2,3-*b*]quinolines[64] have shown promising anticancer activity, we prepared 2-, 3- and 4-methyl substituted indolo[2,3-*b*]quinolines from corresponding toluidines in 38 – 41% yield (entries d – f). Next, the reactions were carried out with naphthylamines (α & β) to get the corresponding annulated pentacyclic benzo-indolo[2,3-*b*]quinolines in 48 and 53% yield (entries b & c). The reaction with *m*-bromo-aniline gave 3-bromo-indolo[2,3-*b*]quinoline in 44% yield (entry g). The reaction was also studied with amino heterocycle i.e. 3-amino pyridine to obtain the corresponding product i.e. 6*H*-indolo[2,3-*b*][1,7]naphthyridine in 29% yield (entry h).

Table 3: Synthesis of different indoloquinolines using I_2 as a catalyst



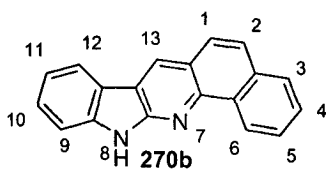
Entry	Substrate 273	Product 270	Yield (%)
a			45
b			48
c			53
d			41
e			38
f			40
g			44

h			29

The common route amongst the reported methods for the synthesis of 6*H*-indolo[2,3-*b*]quinoline involves building of indole ring on a quinoline precursor, while the present method describes the construction of quinoline ring on an indole precursor using iodine as a catalyst in one-pot. This methodology can be used for the synthesis of different indolo[2,3-*b*]quinolines with substituents either on the indole nucleus or on the quinoline nucleus.

Spectroscopic data:

8H-Indolo[2,3-*b*]benzo[*h*]quinoline 270b



IR (KBr): υ_{max} = 3348, 3053, 1609, 1491, 1462, 1385, 1364, 1258, 1234, 1147, 1105, 1018, 899, 812, 797, 746, 685 cm^{-1}

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 5)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.31	m	-	1	H-11
7.57	m	-	2	H-4 & H-10
7.74	m	-	2	H-2 & H-9
7.81	d	9.0	1	H-5
8.02	m	-	2	H-1 & H-3
8.31	d	7.5	1	H-12
9.11	s	-	1	H-13
9.26	d	9.0	1	H-6
12.01	s	-	1	-NH

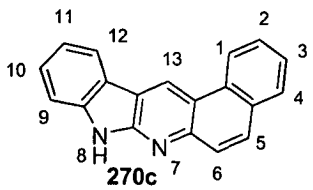
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 6)

δ 111.6 (C-9), 117.4 (C-12b), 120.2 (C-11), 120.8 (C-13a), 121.2 (C-12a), 122.1 (C-6), 123.9 (C-4), 124.4 (C-12), 126.7 (C-2), 127.2 (C-1), 128.2 (C-3, C-10 & C-5), 128.4 (C-13), 131.0 (C-2a), 133.7 (C-6a), 141.2 (C-8a), 144.4 (C-6b), 152.4 (C-7a).

HRMS: *m/z* $[M+H]^+$ 269.1070 (calcd for $C_{19}H_{13}N_2$, 269.1079).

Melting Point: 264-268 ^{0}C.

8H-Indolo[2,3-*b*]benzo[*f*]quinoline 270c



270c

IR (KBr): υ_{max} = 3400, 3152, 1614, 1519, 1445, 1398, 815, 740 cm^{-1}

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 7)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.32	m	-	1	H-11
7.57	m	-	2	H-5 & H-9
7.64	m	-	1	H-6
7.78	dd	6.9 & 7.5	1	H-10
7.94	d	9.0	1	H-4
8.06	dd	8.4 & 9.0	2	H-2 & H-3
8.43	d	7.2	1	H-12
9.03	d	8.4	1	H-1
10.03	s	-	1	H-13
11.87	s	-	1	-NH

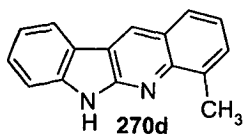
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 8)

δ 111.6 (C-9), 117.2 (C-12b), 119.9 (C-11), 120.2 (C-13a), 121.1 (C-12a), 122.3 (C-2), 123.0 (C-11), 123.8 (C-12), 126.4 (C-4), 127.5 (C-5), 127.8 (C-13), 128.2 (C-1), 129.0 (C-10), 130.0 (C-6), 130.4 (C-4a), 131.0 (C-13b), 141.3 (C-8a), 146.4 (C-6a) and 152.8 (C-7a).

HRMS: *m/z* $[M+H]^+$ 269.1070 (calcd for $C_{19}H_{13}N_2$, 269.1079).

Melting Point: >300 ^{0}C.

4-Methyl-6H-Indolo[2,3-*b*]quinoline 270d



270d

IR (KBr): υ_{max} = 3142, 3090, 1614, 1580, 1460, 1408, 1329, 1230, 1126, 908, 820, 787, 737, 696 cm^{-1}.

^{1}HNMR (DMSO-d$_6$, 300 MHz): (Fig. 9)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.77	s	-	3	-C<u>H</u>$_3$
7.26	dd	6.9 & 7.2	1	H-8
7.37	dd	7.2 & 7.5	1	H-2
7.46-7.52	m	-	2	H-3 & H-9
7.59	d	6.9	1	H-7
7.95	d	7.8	1	H-10
8.25	d	7.5	1	H-1
9.01	s	-	1	H-11
11.80	s	-	1	-NH

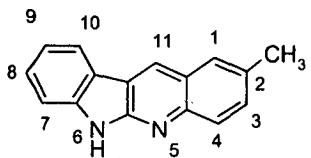
^{13}CNMR (DMSO-d$_6$, 75 MHz): (Fig. 10)

δ 18.9 (-<u>C</u>H$_3$), 111.3 (C-7), 118.0 (C-10b), 120.0 (C-9), 120.7 (C-11a), 122.2 (C-10), 122.8 (C-1), 123.9 (C-2), 127.1 (C-10a), 128.4 (C-11), 128.5 (C-8), 129.2 (C-3), 134.6 (C-4), 141.9 (C-6a), 145.8 (C-4a) and 152.8 (C-5a).

HRMS: *m/z* $[M+H]^+$ 233.1076 (calcd for $C_{16}H_{13}N_2$, 233.1078).

Melting Point: 230-232 ^{0}C.

2-Methyl-6H-Indolo[2,3-*b*]quinoline 270e



270e

IR (KBr): υ_{max} = 3400, 3121, 1614, 1519, 1471, 1404, 1232, 848, 740 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 11)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.50	s	-	3	-C<u>H</u>$_3$
7.25	m	-	1	H-8
7.49	m	-	2	H-9 & H-10
7.56	d	8.7	1	H-3
7.86	s	-	1	H-1
7.88	d	8.7	1	H-4
8.24	d	7.8	1	H-10
8.93	s	-	1	H-11
11.59	s	-	1	-NH

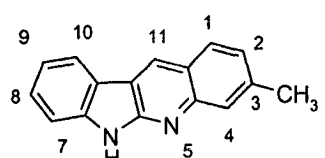
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 12)

δ 21.4 (-<u>C</u>H$_3$), 111.3 (C-7), 118.3 (C-10b), 120.0 (C-9), 120.7 (C-11a), 122.2 (C-10), 124.1 (C-4), 127.2 (C-10a & C-11), 127.2 (C-1), 128.5 (C-3), 131.4 (C-8), 132.2 (C-2), 141.8 (C-6a), 145.2 (C-4a) and 152.9 (C-5a).

HRMS: *m/z* $[M+H]^+$ 233.1087 (calcd for $C_{16}H_{13}N_2$, 233.1078).

Melting Point: >300 ^{0}C.

3-Methyl-6H-Indolo[2,3-*b*]quinoline 270f



270f

IR (KBr): υ_{max} = 3402, 3138, 1614, 1497, 1462, 1232, 908, 798, 740 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 13)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
2.55	s	-	3	-C<u>H</u>$_3$
7.26	m	-	1	H-8
7.32	d	7.8	1	H-2
7.48	m	-	2	H-3 & H-9
7.76	s	-	1	H-4
7.99	d	8.1	1	H-10
8.22	d	7.5	1	H-1
8.97	s	-	1	H-11
11.63	s	-	1	-NH

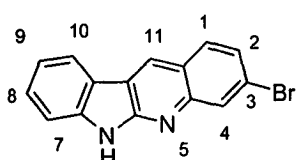
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 14)

δ 22.0 (-<u>C</u>H$_3$), 111.3 (C-7), 117.6 (C-10b), 120.0 (C-9), 120.9 (C-11a), 122.0 (C-10), 122.2 (C-1), 125.4 (C-2), 126.4 (C-10a), 127.7 (C-4), 128.3 (C-11), 128.8 (C-8), 138.9 (C-3), 141.7 (C-6a), 147.0 (C-4a) and 153.4 (C-5a).

HRMS: *m/z* $[M+H]^+$ 233.1078 (calcd for $C_{16}H_{13}N_2$, 233.1078).

Melting Point: 228-230 ^{0}C.

3-Bromo-6H-Indolo[2,3-*b*]quinoline 270g



270g

IR (KBr): υ_{max} = 3375, 3132, 1614, 1508, 1472, 1358, 1242, 939, 798, 740 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 15)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.30	dd	6.9 & 7.2	1	H-8
7.52-7.62	m	-	2	H-3 & H-9
7.65	d	8.1	1	H-7

7.84	d	7.5	1	H-2
8.02	d	8.7	1	H-10
8.44	d	7.5	1	H-1
9.22	s	-	1	H-11
11.90	s	-	1	-NH

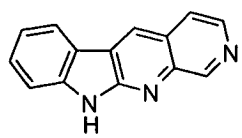
^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 16)

δ 111.6 (C-7), 119.6 (C-10b), 120.4 (C-9), 120.5 (C-11a), 122.3 (C-1 & C-10), 122.9 (C-3), 126.7 (C-10a), 127.2 (C-2), 127.8 (C-11), 129.3 (C-4 & C-8), 129.5 (C-6a), 142.2 (C-4a), and 153.4 (C-5a).

HRMS: *m/z* $[M+H]^+$ 297.0037 (calcd for $C_{15}H_{10}N_2Br$, 296.0027).

Melting Point: 260-264 ^{0}C.

6*H*-Indolo[2,3-*b*][1,7]naphthyridine 270h



270h

IR (KBr): υ_{max} = 3302, 3130, 1508, 1357, 1226, 935, 815, 738 cm^{-1}.

^{1}HNMR (DMSO-d_6, 300 MHz): (Fig. 17)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
7.37	m	-	1	H-8
7.53	t	7.2	1	H-9
7.74	d	8.1	1	H-3
7.80	m	-	1	H-1
8.38	d	7.8	1	H-10
8.54	d	8.4	1	H-2
9.01	s	-	1	H-11
9.68	s	-	1	H-4
13.0	s	-	1	-NH

^{13}CNMR (DMSO-d_6, 75 MHz): (Fig. 18)

δ 112.5 (C-7), 117.4 (C-10b), 120.5 (C-9), 120.9 (C-10), 121.7 (C-1), 123.5 (C-10a), 126.2 (C-11), 134.1 (C-8), 137.1 (C-11a), 139.3 (C-2), 139.6 (C-4), 140.5 (C-6a), 145.7 (C-4a), and 148.9 (C-5a).

HRMS: *m/z* $[M+H]^+$ 220.0864 (calcd for $C_{14}H_{10}N_3$, 220.0874).

Melting Point: >300 ^{0}C.

Conclusion:

To conclude, we have developed a new one-pot method for the assembly of substituted indoloquinolines by sequential imination, nucleophilic addition and annulation catalyzed by iodine. Though the yields are moderate, this method is easy and short which makes it attractive.

Section B: *In Vitro* Antiproliferative Activity Study of Selected Indoloquinolines

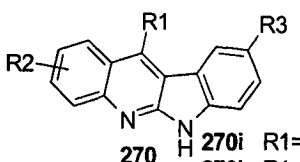
Introduction:

Indolo[2,3-*b*]quinolines share many biological properties with the natural alkaloid neocryptolepine including the ability to interact with DNA and to inhibit topoisomerase II activity. The indolo[2,3-*b*]quinoline derivatives also display promising antimicrobial and cytotoxic activities.[62,63] In the following section, these biological activities of indolo[2,3-*b*]quinolines are being listed and discussed.

Literature Report:

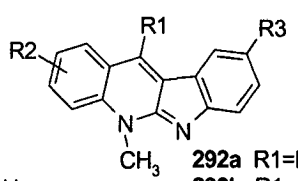
Peczynska-Czoch and co-workers[64] evaluated the antimicrobial activity against a variety of micro-organisms and cytotoxicity against the KB cell line of a series of 6*H*-indolo[2,3-*b*]quinoline and 5*H*-indolo[2,3-b]quinoline derivatives as described in table 4 below.

Table 4: Antimicrobial Activity and Cytotoxicity of 6*H*- and 5*H*-Indolo[2,3-*b*]quinolines



270

270i R1=R2=R3=H
270j R1=CH_3, R2=R3=H
270k R1=CH_3, R2=2-CH_3, R3=H
270l R1=CH_3, R2=4-CH_3, R3=H
270m R1=CH_3, R2=H, R3=CH_3
270n R1=CH_3, R2=2-CH_3, R3=CH_3



292

292a R1=R2=R3=H
292b R1=CH_3, R2=R3=H
292c R1=CH_3, R2=2-CH_3, R3=H
292d R1=CH_3, R2=4-CH_3, R3=H
292e R1=CH_3, R2=H, R3=CH_3
292f R1=CH_3, R2=2-CH_3, R3=CH_3

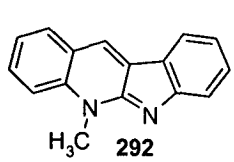
Compd.	MIC[a] (µmol/mL)						ID_{50}[b] (µmol/mL)
	1	2	3	4	5	6	
270i	-	-	-	-	-	-	0.6
292a	-	-	0.25	0.25	0.12	0.06	0.006
270j	-	-	-	-	-	-	0.2
292b	-	-	0.12	0.06	0.03	0.06	0.004
270k	-	-	-	-	-	-	0.1
292c	-	-	0.06	0.03	0.015	0.03	0.003
270l	-	-	-	-	-	-	0.1

292d	-	-	0.25	0.12	0.12	0.06	0.009
270m	-	-	-	-	-	-	0.1
292e	-	-	0.12	0.06	0.06	0.12	0.003
272n	-	-	-	-	-	-	0.1
292f	-	-	0.03	0.015	0.015	0.03	0.002

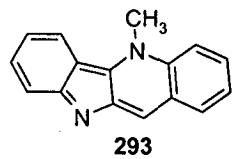
[a] (1) *Escherichia coli* PCM 271, (2) *Pseudomonas aerudomonas* PCM 499, (3) *Staphylococcus aureus* PCM 458, (4) *Micrococcus luteus* PCM 525, (5) *Candida albicans* (clinical isolate), (6) *Trichophyton mentagrophytes* (clinical isolate); MICs were determined at the concentration range of 0.01-5 μmol/mL.
[b]Cytotoxicity expressed as ID_{50} values was determined *in vitro* against KB cell line.

Cimanga and co-workers[65] tested the antiplasmodial activity of cryptolepine and neocryptolepine against three *Plasmodium falciparum* strains (Table 5).

Table 5: Antiplasmodial Activity (IC_{50} ng/mL) of cryptolepine and neocryptolepine against three Plasmodium falciparum Strains[a]



292



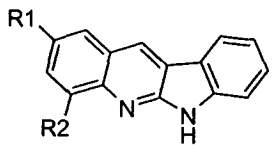
293

Compd.	***P. falciparum* strain**		
	D-6	**K-1**	**W-2**
292	35 ± 0.7	51 ± 0.1	65 ± 1.3
293	27 ± 0.3	33 ± 0.1	41 ± 0.5

[a]Values are expressed as mean ± standard deviation (S.D.)

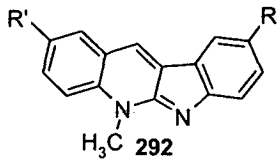
Jonckers *et al.*[66] evaluated in vitro antiplasmodial activity against a chloroquine-sensitive and a chloroquine-resistant *P. falciparum* strain and cytotoxicity on a human cell lines (MRC-5 cells) of neocryptolepine derivatives (Table 6).

Table 6: *In vitro* Cytotoxicity and Antiplasmodial Activity



270

270o R1=OCH_3, R2=H
270p R1=Cl, R2=H
270q R1=F, R2=H
270r R1=Cl, R2=Cl



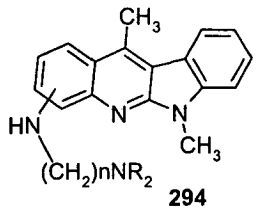
292

292a R'=R=H
292g R'=H, R=OCH_3
292h R'=H, R=Br
292i R'=H, R=Cl
292j R'=H, R=F
292k R'=H, R=I
292l R'=H, R=CH_3
292m R'=H, R=NO_2
292n R'=H, R=SCH_3
292o R'=H, R=CN
292p R'=CN, R=H
292q R'=CN, R=Cl
292r R'=CN, R=OCH_3
292s R'=CN, R=CF_3
292t R'=CN, R=F

Compd.	Cytoxicity (MRC-5 cells) IC_{50} (μM)	*P. falciparum* (chloroquine sensitive) IC_{50} (μM)	*P. falciparum* (chloroquine sensitive) IC_{50} (μM)
292[a]	11.0 ± 1.4	27.3 ± 5.7	14.0 ± 1.7
292g	4.0 ± 0.1	4.3 ± 0.6	4.7 ± 0.6
270[o]	>32	>32	>32
292h	>32	6.0 ± 6.1	4.0 ± 0.1
292i	16.5 ± 0.7	21.0 ± 8.9	5.0 ± 0.1
270p	>32	>32	>32
292j	15.0 ± 0.1	19.3 ± 3.8	4.7 ± 0.6
270q	>32	>32	>32
292k	16.0 ± 0.1	17.7 ± 5.1	6.3 ± 0.6
292l	0.95 ± 0.07	2.7 ± 2.1	2.3 ± 0.6
292m	>32	29.0 ± 1.7	>32
292n	5.0 ± 0.1	4.0 ± 1.0	3.7 ± 0.6
2920	16.0 ± 0.1	17.0 ± 1.0	15.3 ± 0.6
292p	>32	>32	>32
292q	>32	>32	>32
292r	>32	28.3 ± 3.5	17.0 ± 6.2
292s	>32	14.0 ± 2.6	6.7 ± 1.1
292t	11.0 ± 5.7	16.3 ± 1.2	14.7 ± 4.9
270r	>32	>32	>32

Godlewska *et al.*[67] screened the antimicrobial and cytotoxic activities of indolo[2,3-*b*]quinoline derivatives as shown below (Table 7).

Table 7: Antimicrobial and Cytotoxic Activities of 6*H*-Indolo[2,3-*b*]quinoline derivatives



294

294a n=2, R=2-CH_3
294b n=2, R=9-CH_3
294c n=3, R=2-CH_3
294d n=3, R=9-CH_3
294e n=2, R=2-C_2H_5
294f n=2, R=9-C_2H_5

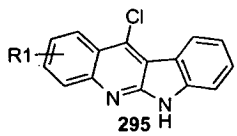
Compd.	MIC* (mM)		ID_{50}** (μM) against KB cells
	Micrococcus luteus	*Saccharomyces cerevisiae*	
294a	0.5	0.5	6.8 ± 1.9
294b	1.0	1.0	2.1 ± 1.5
294c	0.25	1.0	6.2 ± 1.4
294d	1.0	1.0	8.4 ± 2.4
294e	0.06	0.5	7.4 ± 1.4
294f	0.25	1.0	5.3 ± 1.6

MIC* - Minimal Inhibitory Concentration

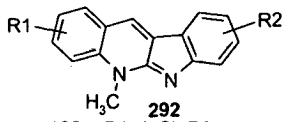
ID_{50}** - the dose of compound that inhibits proliferation rate of the tumor cells by 50% as compared to control untreated cells.

Sayed *et al.*[68] reported the antiplasmodial activity against a chloroquine-sensitive *P. falciparum* strain and cytotoxicity on a human cell (MRC-5) line of a series of substituted neocryptolepine derivatives (Table 8)

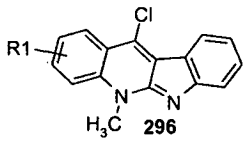
Table 8: Cytotoxicity and Antiplasmodial Activity of substituted neocryptolepines



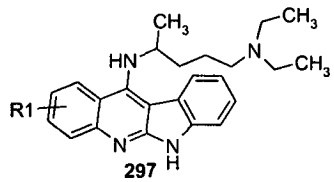
295 R1=H, 1-Cl, 3-Cl, 2-Cl



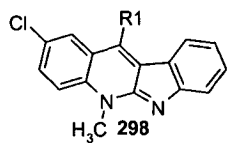
292 R1=2-Cl, 3-Cl, 8-Cl, 9-Cl; R2=H



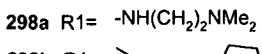
296 R1=H, 1-Cl, 3-Cl, 2-Cl



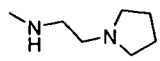
297 R1=1-Cl, 3-Cl, 2-Cl



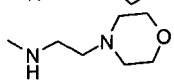
298 R1=



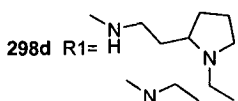
298a R1= -NH(CH$_2$)$_2$NMe$_2$



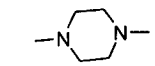
298b R1=



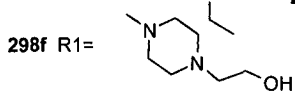
298c R1=



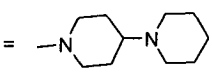
298d R1=



298e R1=



298f R1=



298g R1=

Compd.	Cytotoxicity (MRC-5) IC_{50} µM	Antiplasmodial Activity (*P. falciparum* strain) IC_{50} µM
292u	>64	52
292v	21	41.11
292w	>64	>64
292x	>64	29
295b	>64	>64
295c	>64	>64
295d	>64	>64
296a	5	>64
296b	34	39
296c	27	>64
296d	>64	>64
297a	20	1.2
297b	15	0.14
297c	>64	0.12
298a	>64	0.4

298b	>64	3
298c	>64	33
298d	15	0.38
298e	3	2.46
298f	2	2.34
298g	2	<0.25

Results and Discussion:

In vitro antiproliferative activity (cell growth inhibition activity) of some of the synthesized indoloquinoline compounds i.e. 6H-Indolo[2,3-*b*]quinoline **270a**, 8H-Indolo[2,3-*b*]benzo[*h*]quinoline **270b**, 4-Methyl-6H-Indolo[2,3-*b*]quinoline **270d** and 3-Bromo-6H-Indolo[2,3-*b*]quinoline **270g** were evaluated against human hepatocellular carcinoma HepG2 and human breast carcinoma MCF-7 cell lines (obtained from American Type Culture Collection – Manassas, VA, USA).

HepG2 and MCF-7 cells were plated at a density of 10000 cells per well in 96 well cell culture plate and allowed to adhere for 24 hours at 37^{0}C. This was then treated with various concentrations of compounds diluted in culture medium for further 48 hours. In the control cells, culture medium consisting of corresponding concentration of DMSO was added. Thereafter, cell survival was assessed. The optical density was taken on a microplate reader at 570 nm using 630 nm as a reference filter. Absorbance given by untreated cells was taken as 100% cell growth. All assays were performed in triplicates. The detailed results are given in tabular form below.

For HepG2 Cells

DMSO

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	0	100	1.6776
0.0001	3.081	94.560	1.5856
0.01	3.081	91.987	1.5423
0.1	3.106	85.902	1.4404
1	2.487	82.722	1.3871

Compound 270a

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	1.6776	100	0
0.0001	1.5897	94.819	3.789
0.01	1.0375	71.700	6.899
0.1	1.0267	61.211	0.855
1	1.0219	61.020	6.571

Compound 270b

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	1.6776	100	0
0.0001	1.4605	87.199	9.826
0.01	1.3501	80.520	3.810
0.1	0.8209	49.017	5.447
1	0.4308	25.646	2.256

Compound 270d

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	1.6776	100	0
0.0001	1.5649	93.314	5.715
0.01	0.6185	36.883	1.574
0.1	0.0817	4.868	0.126
1	0.0612	3.652	0.135

Compound 270g

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	1.6776	100	0
0.0001	1.5334	91.436	2.584

Compound 270d

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	2.7485	100	0
0.0001	2.3563	85.803	5.427
0.01	1.1079	40.338	2.331
0.1	0.1438	5.237	1.200
1	0.1291	4.724	0.278

Compound 270g

Conc. (mg/mL)	Average Reading	% Cell Survival	Standard Deviation
0	2.7485	100	0
0.0001	2.2643	82.427	1.762
0.01	1.7615	64.130	1.925
0.1	0.6149	22.320	2.422
1	0.1542	5.606	0.267

The concentration of the compounds that inhibits the cell growth of the human cancer cells by 50% as compared to control untreated cells i.e. IC_{50} values were calculated by plotting the graph of concentration (mg/mL) against cell survival (%) as shown below (Figures 1 & 2).

Fig. 19: Antiproliferative activity of compounds in HepG2 cells

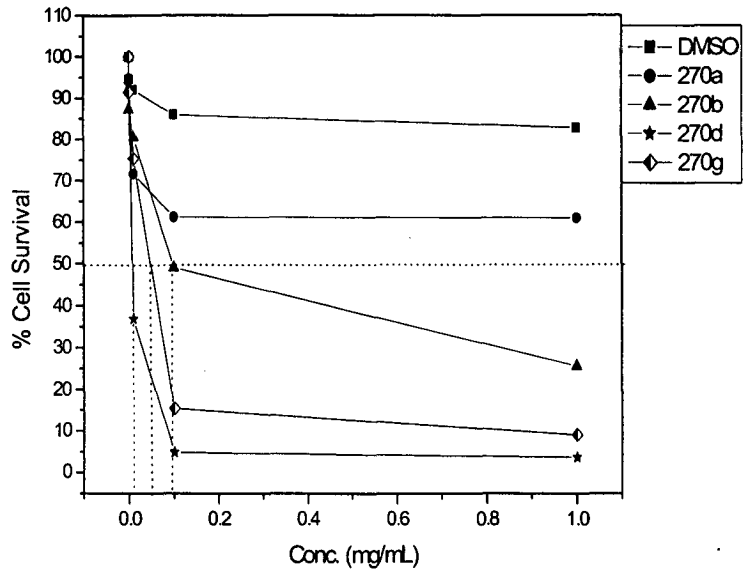
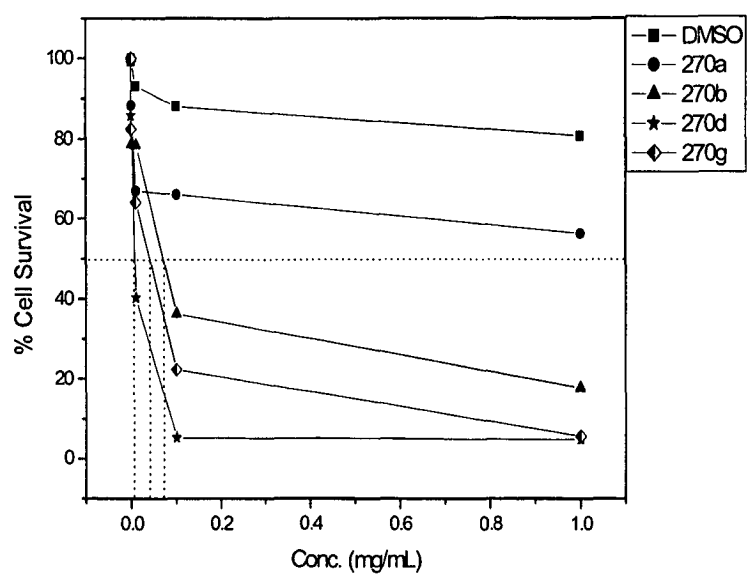
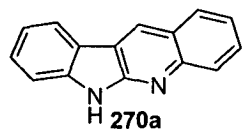
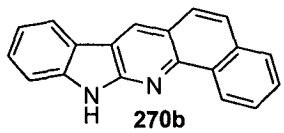
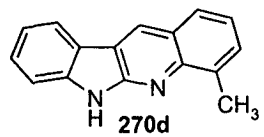
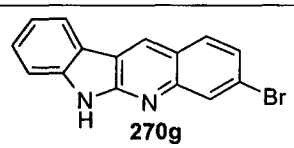


Fig. 20: Antiproliferative activity of compounds in MCF-7 cells



The detail results of *in vitro* antiproliferative activity of these indoloquinolines are given in table 9 below.

Table 9: Antiproliferative Activity of Indoloquinoline Derivatives against HepG2 and MCF-7 cell lines

Compd.	HepG2 IC_{50} (mg/mL)	MCF-7 IC_{50} (mg/mL)
 **270a**	>1	>1
 **270b**	0.0951	0.0717
 **270d**	0.0098	0.0059
 **270g**	0.0486	0.0369

The results suggest that the cell growth inhibitory activity of compound **270d** is very high (< 0.01 mg/mL) in both the cell lines (HepG2 and MCF-7). The compound **270a** showed less activity (> 1 mg/mL) to both cell lines (HepG2 and MCF-7). There was cell type difference in the toxicity of compound **270b**, as it was more toxic to MCF-7 than to HepG2 cells at higher concentrations. Compound **270g** shows moderate activity against both cell lines (< 0.1 mg/mL). These results indicate the limited activity of compounds at lower concentration and differential activity at higher concentration.

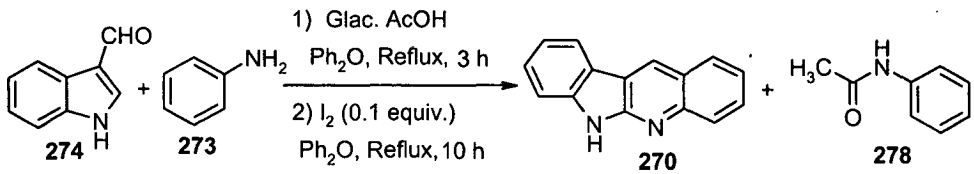
Conclusion:

It can be concluded that the methyl substituted 6*H*-indolo[2,3-*b*]quinoline shows high activity against HepG2 and MCF-7 cell lines than the other substituted 6*H*-indolo[2,3-*b*]quinoline derivatives and this higher activity is probably due to the presence of alkyl group. From the present work, alkyl substituted 6*H*-indolo[2,3-*b*]quinoline is therefore considered as the most promising lead for potentially new anticancer agents.

Experimental Section:

Section A: Use of Molecular Iodine for the Synthesis of 6*H*-Indolo[2,3-*b*]quinoline and their Derivatives.

1.01 Reaction of indole-3-carboxyaldehyde with aniline in presence of AcOH:



Procedure: To a mixture of indole-3-carboxyaldehyde **274** (0.50 g, 3.46 mmol) and aniline **273** (0.64 g, 6.91 mmol) in diphenyl ether (20 mL) was added glacial acetic acid (5 mL) and heated under reflux temperature for 3 hours. The reaction mixture was cooled to room temperature, iodine (0.08 g, 0.35 mmol) was added and heating was continued at reflux temperature for 10 hours. After cooling, reaction mixture was chromatographed on silica gel column and diphenyl ether was removed using hexanes as the eluent. Further elution with 20% ethyl acetate in hexanes afforded the 6*H*-indolo[2,3-*b*]quinoline **270**.

Yield: 23% (0.17 g).

Nature: Yellow solid

Melting Point: >300 ^{0}C; Lit.[60] 342-346 ^{0}C.

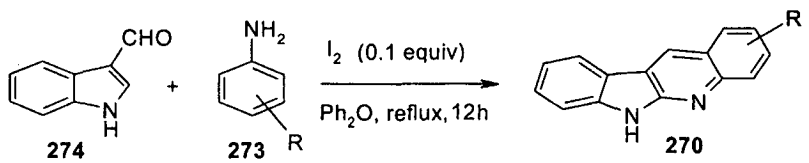
Finally, the elution with 30% ethyl acetate in hexanes afforded **278**.

Yield: 33% (0.154 g).

Nature: White solid.

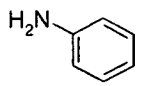
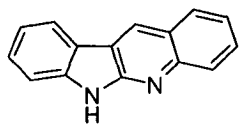
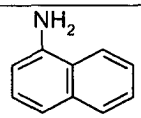
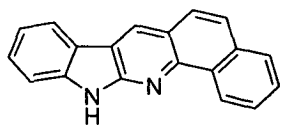
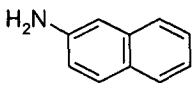
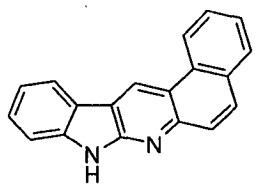
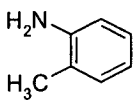
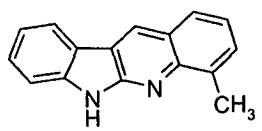
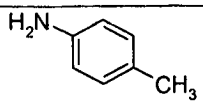
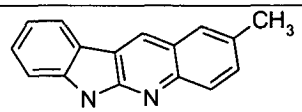
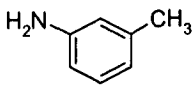
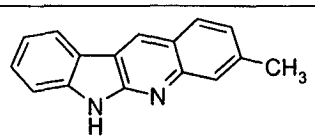
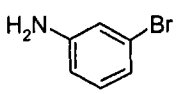
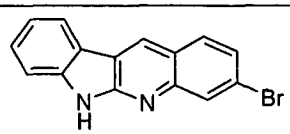
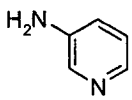
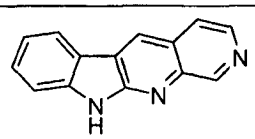
Melting Point: 114-115^{0}C (Commercially available acetanilide – 114 -116^{0}C).

1.02 Reaction of indole-3-carboxyaldehyde with aryl amines in absence of AcOH



General Procedure: Indole-3-carboxyaldehyde **274** (3.46 mmol), aryl amines **273a-h** (6.92 mmol) and iodine (0.35 mmol) was refluxed in diphenyl ether (20 mL) for 12 hours. After cooling, reaction mixture was chromatographed on alumina and diphenyl

ether was removed using hexanes as an eluent. Excess aryl amines (except 3-amino-pyridine which remains at the bottom of the product) were eluted using 5% ethyl acetate in hexanes. Further elution with 20% ethyl acetate in hexanes afforded the indoloquinolines **270a-h**.

Entry	Substrate 273	Product 270	Yield (%)	Nature	M.P. (^{0}C)
a			45	Yellow Solid	>300
b			48	Gray Solid	264 - 268
c			53	Gray Solid	>300
d			41	Yellow Solid	230 - 234
e			38	Yellow Solid	>300
f			40	Yellow Solid	228 - 230
g			44	Brown Solid	260 – 264
h			29	Brown Solid	>300

Section B: *In Vitro* Antiproliferative Activity Study of Selected Indoloquinolines

This study was done at NCCS, Pune.

2.01 Materials and methods:

Human hepatocellular carcinoma HepG2 and human breast carcinoma MCF-7 cell lines were obtained from American Type Culture Collection (Manassas, VA, USA), and maintained at NCCS in-house National Cell repository. Cells were maintained as a monolayer in culture medium consisting of nutrient media DMEM supplemented with heat inactivated fetal bovine serum (10 %), penicillin (100 U/mL) and streptomycin (100 µg/mL) (Invitrogen Life Technologies, MD, USA) at 37 °C in 5 % CO_2 and humidified air atmosphere. Stock solutions of the compounds were prepared in DMSO at a concentration of 100 mg/mL. Afterwards the samples were diluted to the required concentration in cell culture media. The 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) was dissolved (1 mg/mL) in DMEM (without phenol red).

2.02 Antiproliferative Activity:

MTT Cell Proliferation Assay: HepG2 and MCF-7 cells were plated at a density of 10,000 cells per well in 96 well tissue culture plates. Cells were allowed to adhere for 24 h at 37 °C and then treated with various concentrations (0.00001, 0.01, 0.1, 1.0 mg/mL) of compounds diluted in culture medium, for additional 48 h. In the control cells, culture medium consisting of corresponding concentration of DMSO was added. Thereafter, cell proliferation was assessed by replacing culture medium with 50 µl DMEM media containing 1 mg/mL MTT and subsequently incubated for additional 4 h at 37 °C. Medium was then aspirated off and formazan crystals were solubilized in 100 µl of iso-propanol. The optical density was taken on a microplate reader at 570 nm using 630 nm as a reference filter. Absorbance given by untreated cells was taken as 100 % cell growth. All assays were performed in triplicates.

Spectra:

Fig. 1: ^{1}H NMR spectrum of **270a**

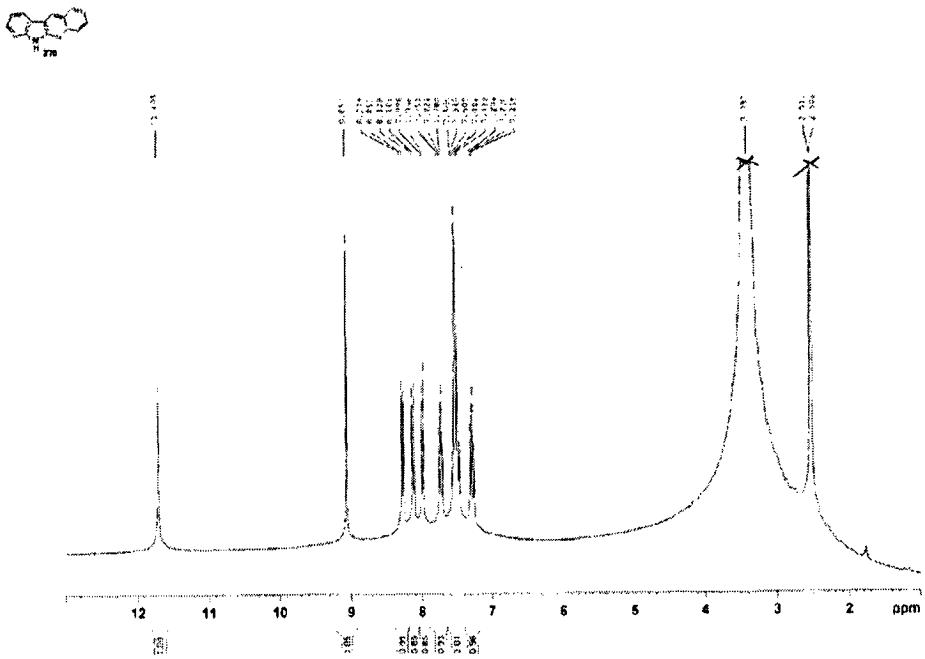


Fig. 2: ^{13}C NMR spectrum of **270a**

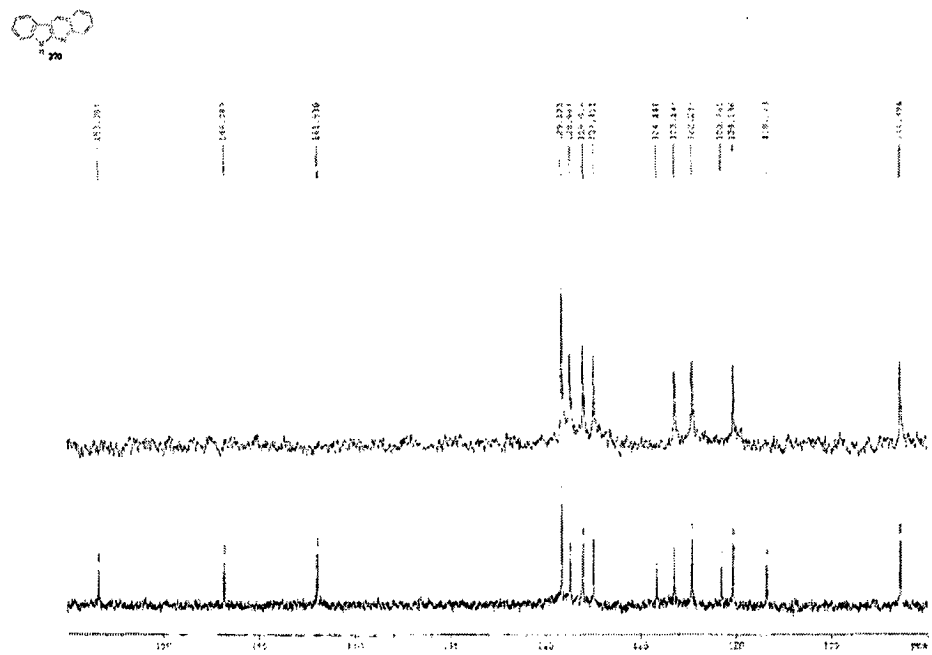


Fig. 3: ^{1}H NMR spectrum of **278**

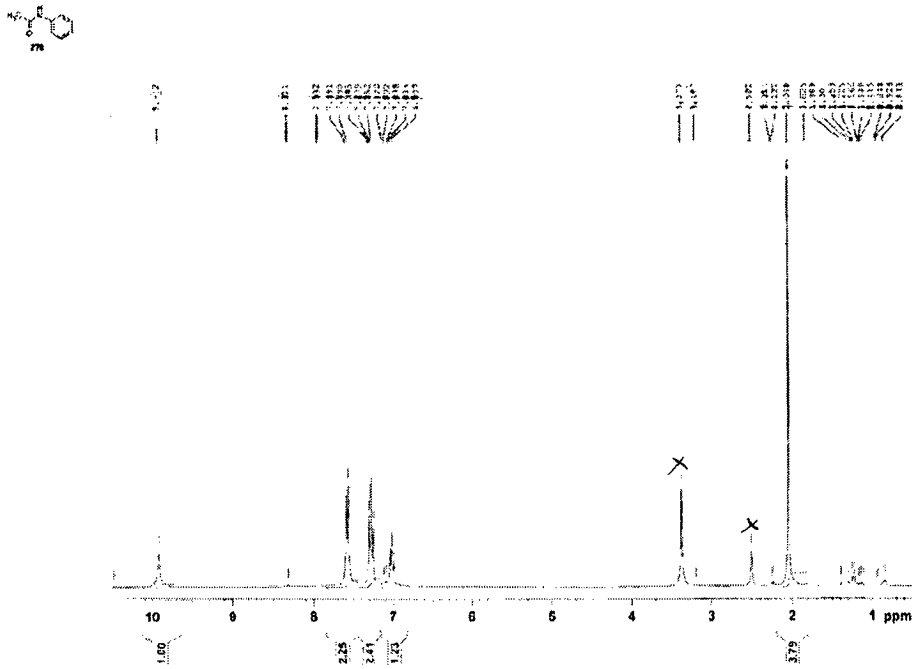


Fig. 4: ^{13}C NMR spectrum of **278**

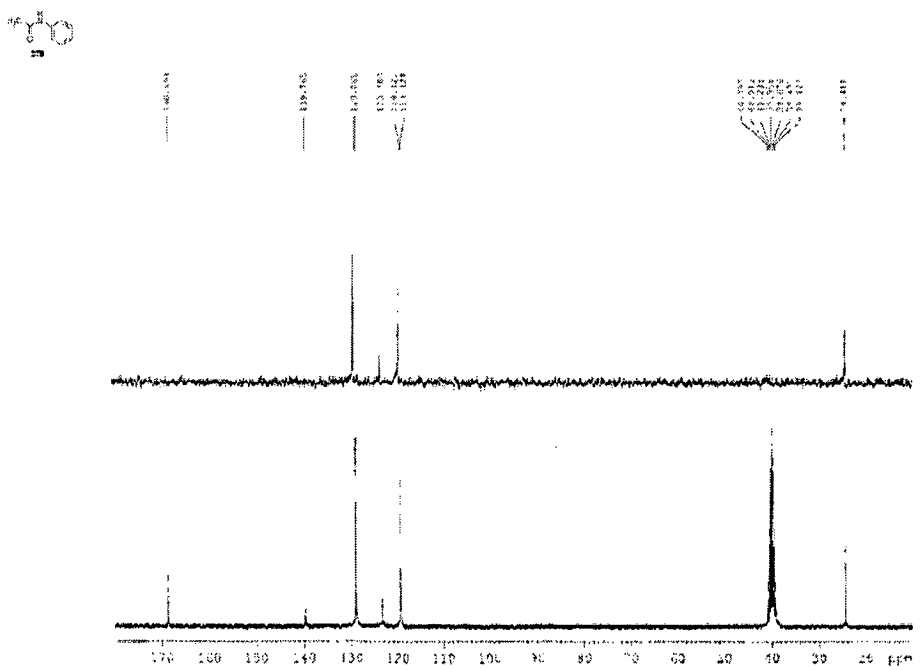
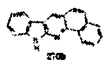


Fig. 5: ^{1}H NMR spectrum of **270b**



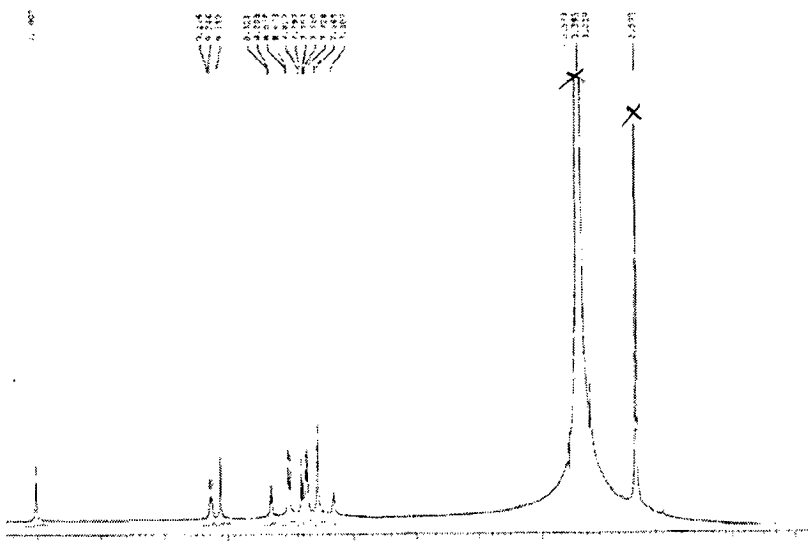
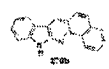


Fig. 6: ^{13}C NMR spectrum of **270b**



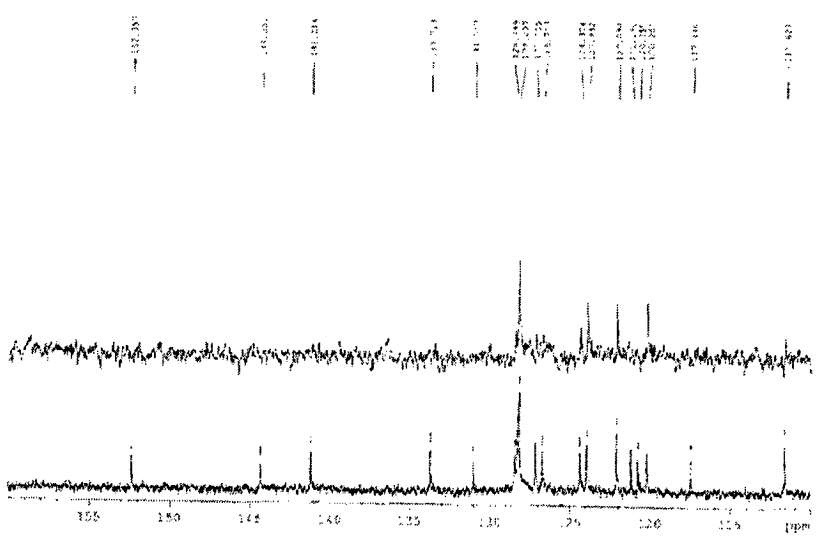


Fig. 7: ^{1}H NMR spectrum of **270c**

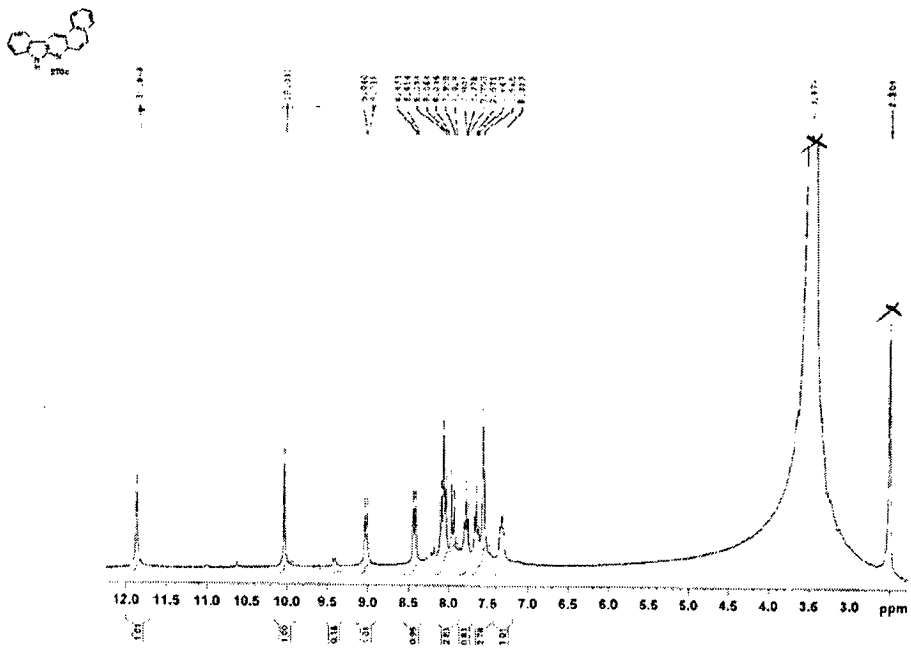


Fig. 8: ^{13}C NMR spectrum of **270c**

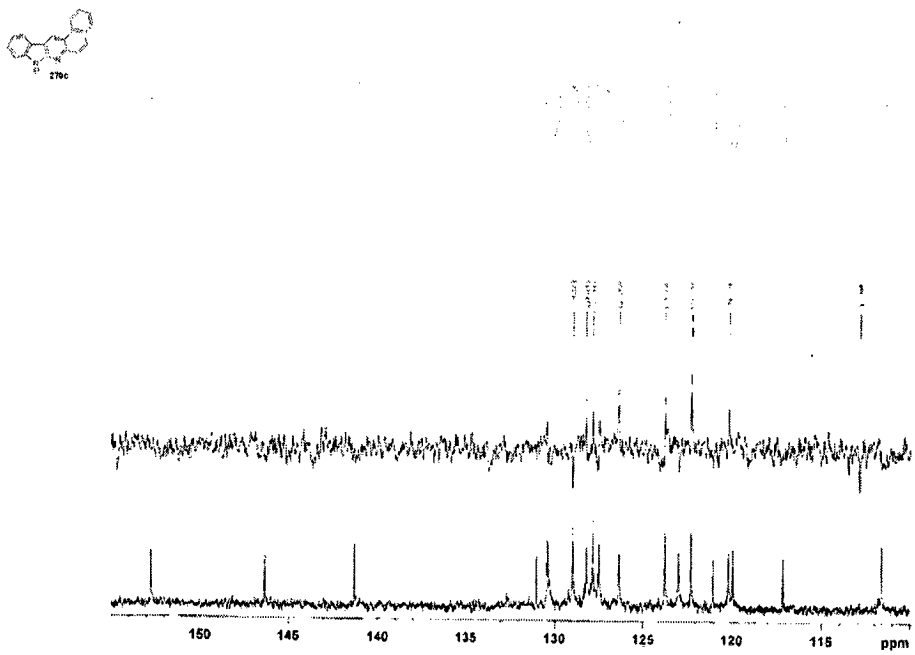


Fig. 9: ^{1}H NMR spectrum of **270d**

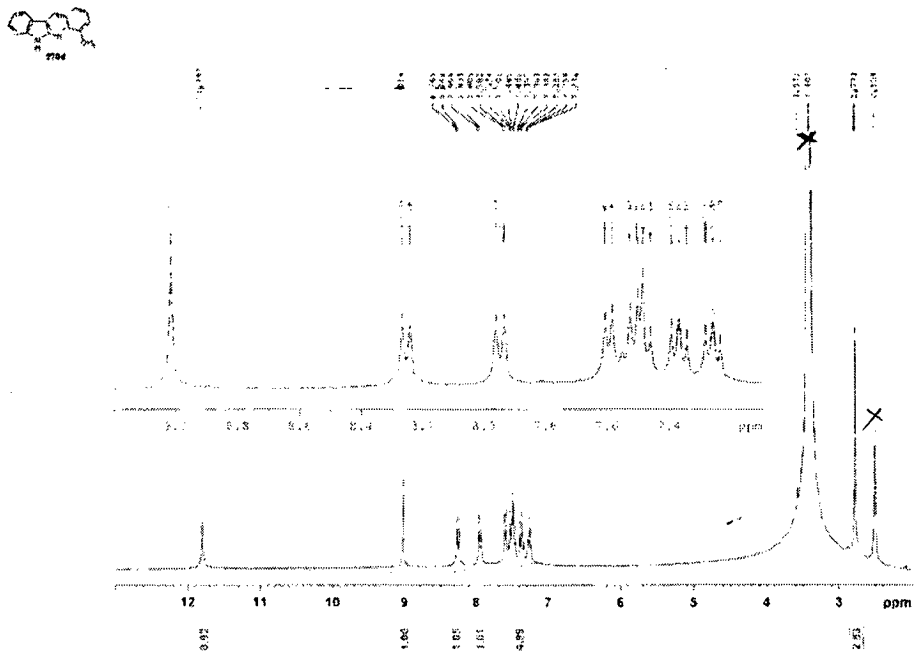


Fig. 10: ^{13}C NMR spectrum of **270d**

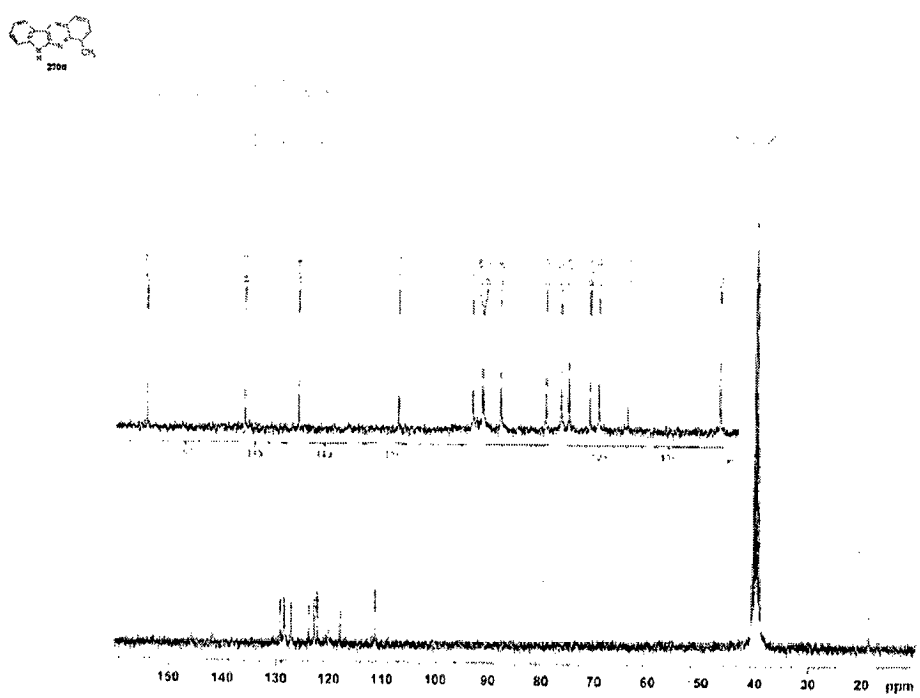


Fig. 11: ^{1}H NMR spectrum of **270e**

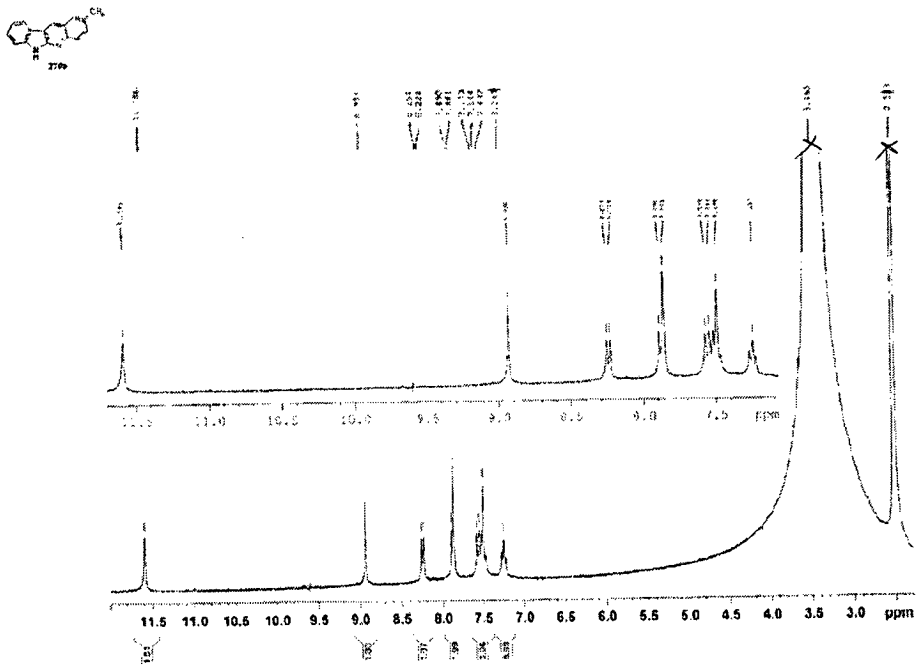


Fig. 12: ^{13}C NMR spectrum of **270e**

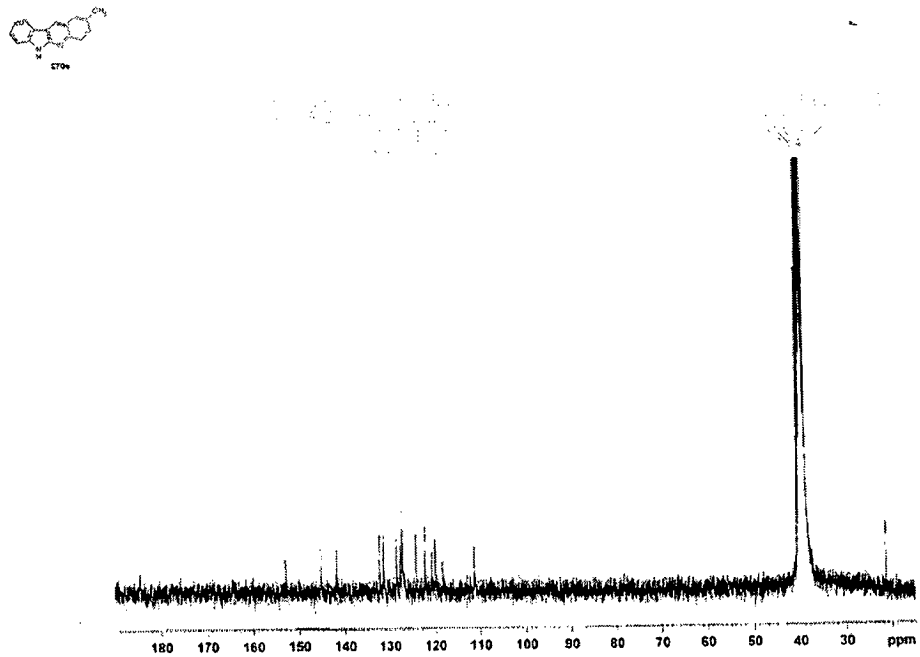


Fig. 13: ^{1}H NMR spectrum of **270f**

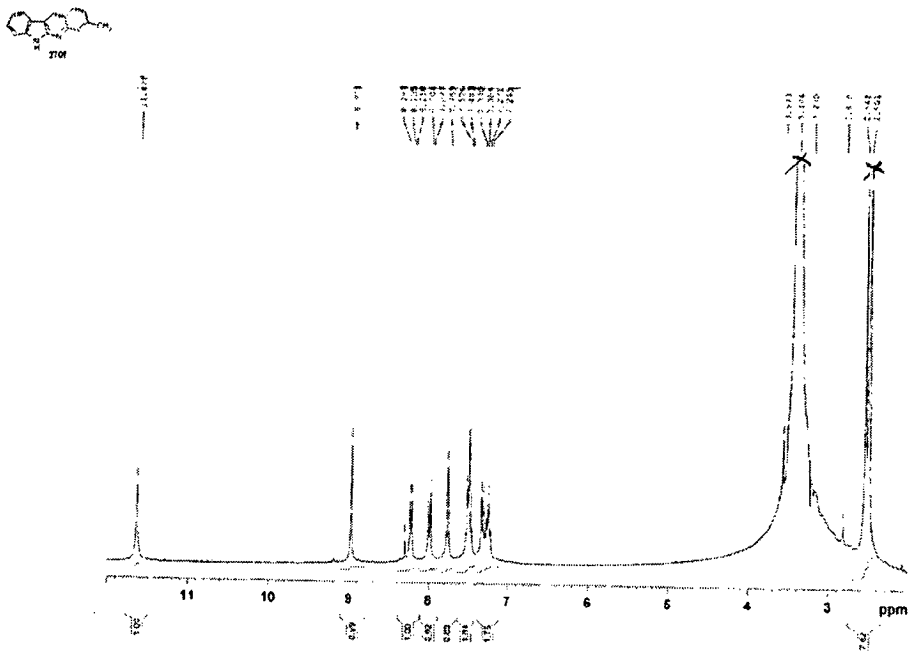


Fig. 14: ^{13}C NMR spectrum of **270f**

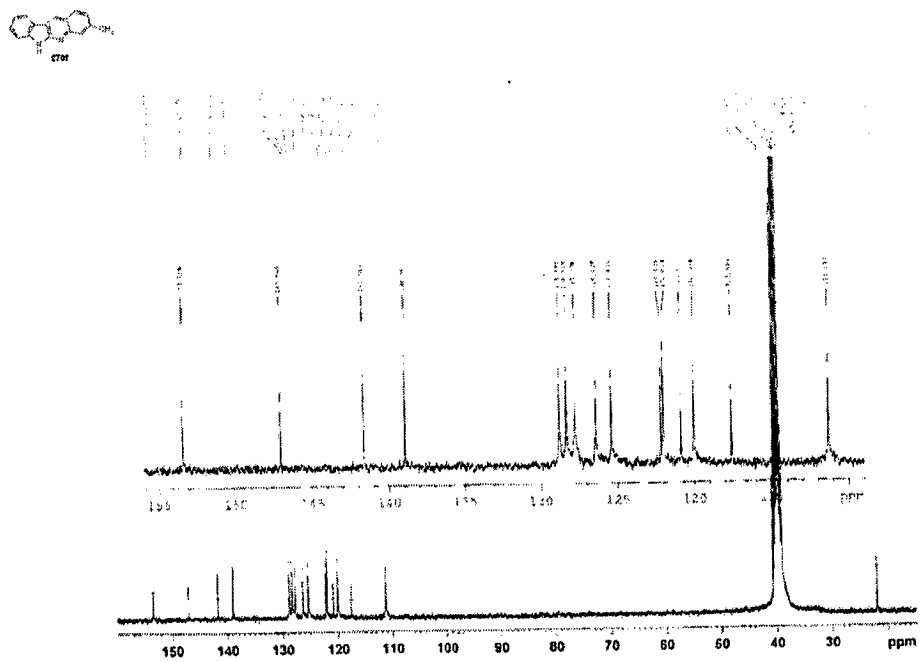


Fig. 15: ^{1}H NMR spectrum of **270g**

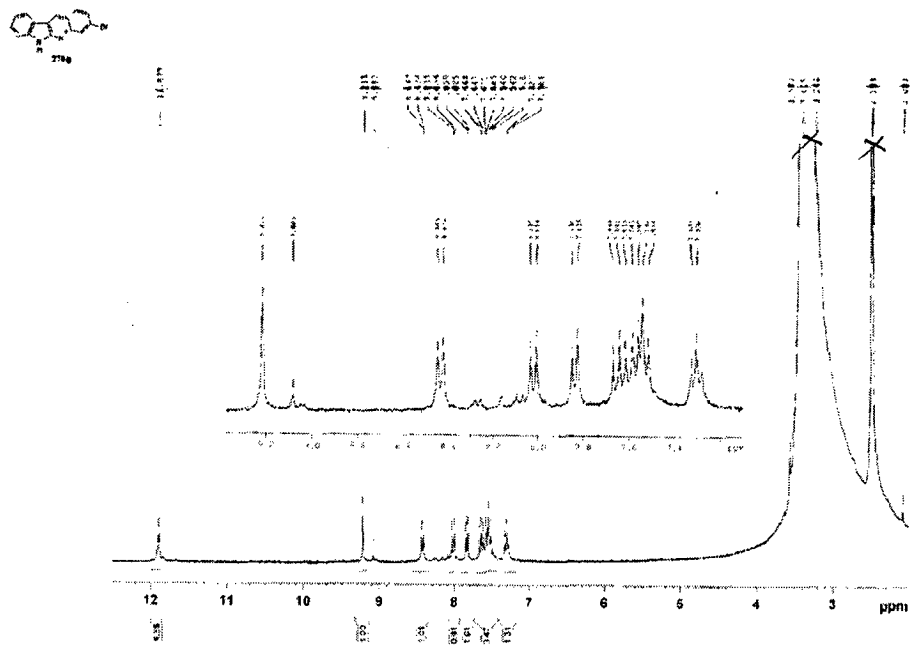


Fig. 16: ^{13}C NMR spectrum of **270g**

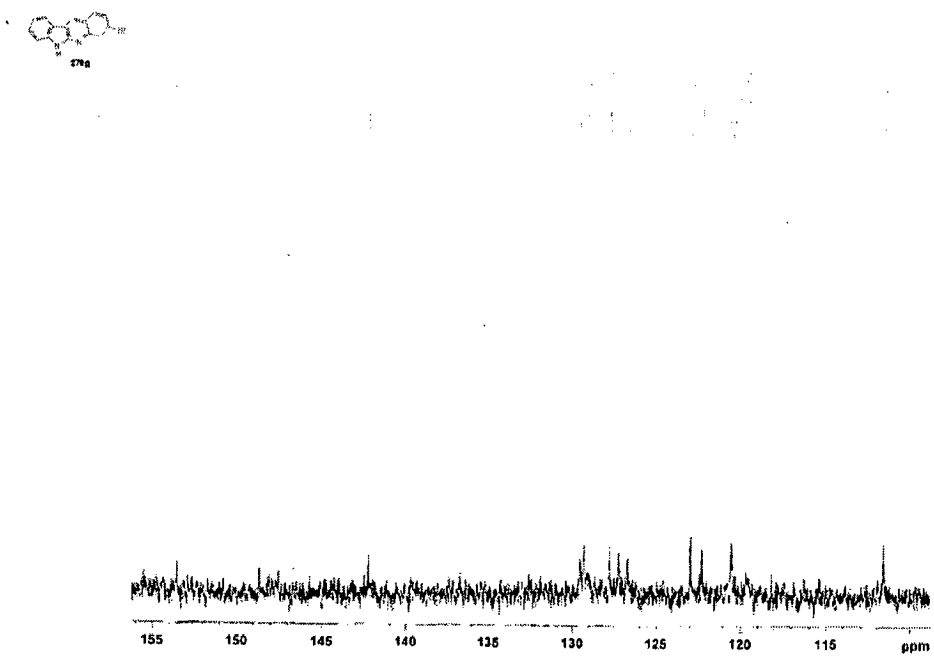


Fig. 17: ^{1}H NMR spectrum of **270h**

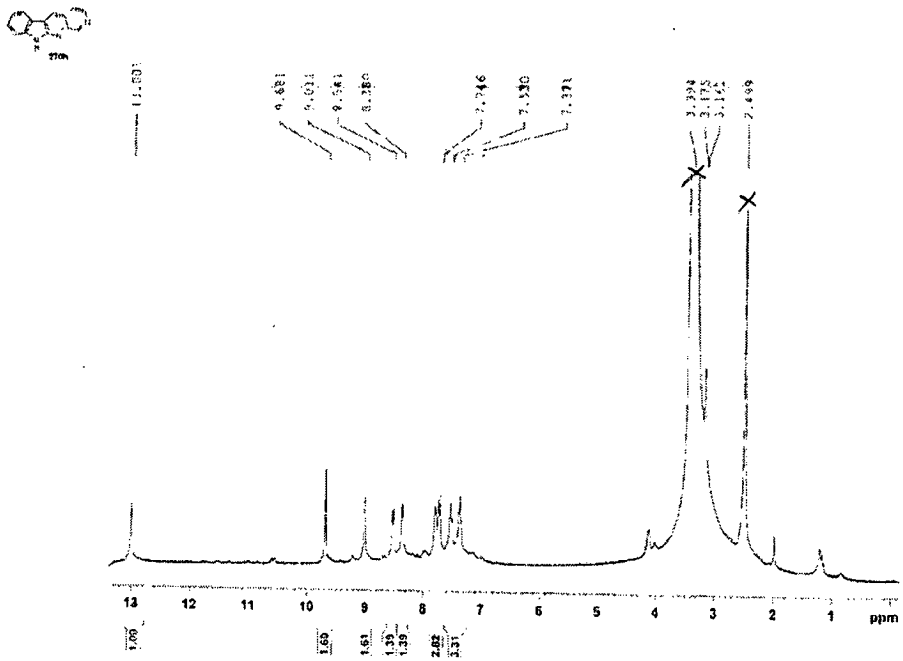
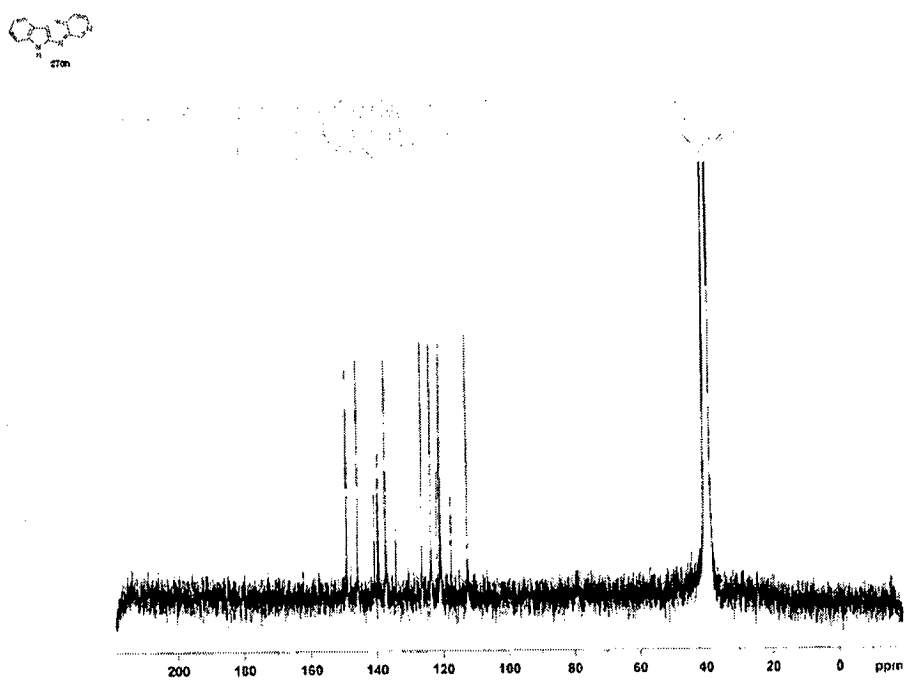


Fig. 18: ^{13}C NMR spectrum of **270h**



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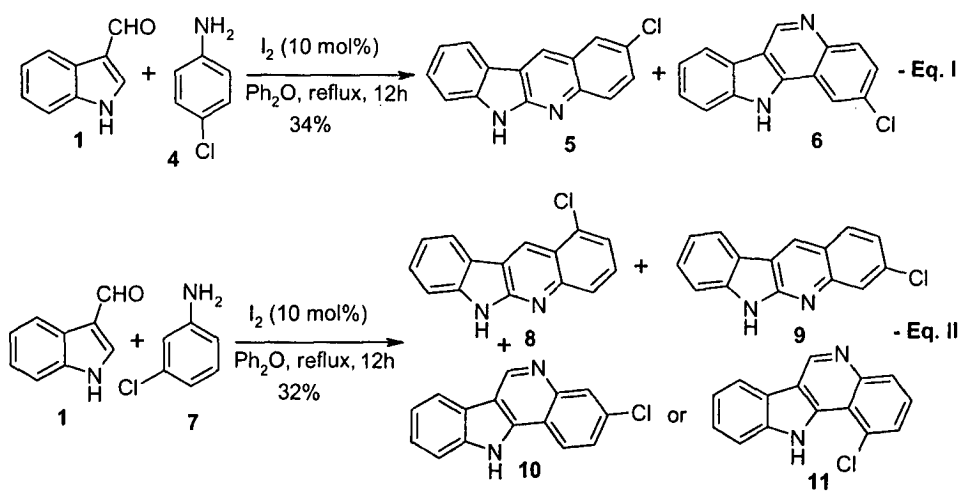
CORRIGENDA

We were interested to make D-ring chloro-substituted 6*H*-indolo[2,3-*b*]quinolines using iodine as a catalyst to check their bioactivity. Accordingly, indole-3-carboxyaldehyde **1** and 4-chloroanilines **4** was refluxed in diphenyl ether using 10 mol% iodine as a catalyst. We were expecting only linear indoloquinolines to form, however the careful NMR analysis of the product obtained from 4-chloroaniline revealed that, it is a mixture of 2-chloro-6*H*-indolo[2,3-*b*]quinoline **5** and 3-chloro-5*H*-indolo[3,2-*c*]quinoline **6** in a 1:1 ratio (Scheme 2 – Eq. I). When the reaction was carried with 3-chloroaniline, a mixture of three compounds *viz.* 3-chloro-6*H*-indolo[2,3-*b*]quinoline **8**, 1-chloro-6*H*-indolo[2,3-*b*]quinoline **9** and 2-chloro-5*H*-indolo[3,2-*c*]quinoline **10** was formed in a 1:1:1 ratio (Scheme 2 – Eq. II). The structure of the compounds were determined by ^{1}H NMR by comparison with the literature data of the related compounds and by LC-MS. LC-MS of compounds shown in eq. I displays two peaks at retention time 3.91 and 5.72 with same *m/z* 253 (^{35}Cl, M+H)$^+$ and 255 (^{37}Cl, M+H)$^+$ and for compounds in eq. II shows three peaks, one at retention time 3.82 and two overlapping peaks at retention time 5.70 and 5.73 with same *m/z* 253 (^{35}Cl, M+H)$^+$ and 255 (^{37}Cl, M+H)$^+$.

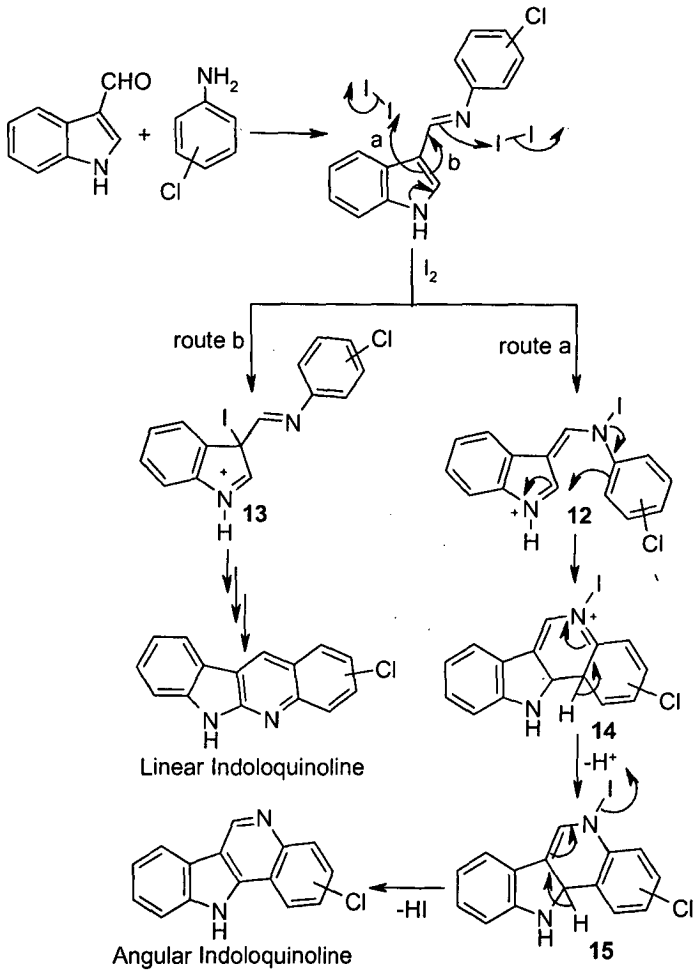


Scheme 2

The reaction shown in eq. II yielded three products out of four possible products, as evident from three singlets at δ 12.8, 11.9 and 11.8 due to –NH protons and three singlets at δ 9.1, 9.3 and 9.6 due to C-ring protons. In this, the signals at δ 9.1 and 9.3 may be attributed to the linear indoloquinolines **8** & **9** while a singlet at δ

9.6 may be due to angular indoloquinoline **10** or **11**. Of the angular indoloquinolines **10** and **11**, the compound **10** is expected to show a singlet in the region δ 8.0 - 8.2 due to the presence of *peri* proton next to chlorine while in compound **11**, it is expected to show a double doublet. The ^{1}H NMR of the product mixture had two singlets in the region δ 8.0 - 8.2. One singlet was attributed to linear indoloquinoline **9** while other was attributed to angular indoloquinoline **10**. The formation of **11** was also disfavored due to steric crowding. Thus, the products formed from 3-chloro-aniline are **8**, **9** and **10**.

In our earlier section, we have detailed a probable mechanism for the exclusive formation of linear indolquinoline. However, the formation of both linear and angular indoloquinolines in case of chloro-substituted anilines indicated that a different mechanism may be operating for the formation of angular indoloquinoline. A probable mechanism to account for the formation of both the products is shown below (Scheme 3).



Scheme 3

Initial electrophilic addition of iodine may lead to the formation of *N*-iodo-indolonium intermediate **12** (route a) and 3-iodo-indolinium cation **13** (route b). The formation of intermediate **12** may be facilitated due to electron withdrawing chloro-substituent. Intramolecular cyclization of intermediate **12** will lead to angular indoloquinoline while intermediate **13** would furnish the linear indoloquinoline *via* intermolecular attack as reported earlier.[13] The products obtained were tested for biological activity without further purification as described below.

Biological Activities:

In vitro antiproliferative activity (cell growth inhibition activity) of these chloro-substituted indoloquinolines (**3e** & **3f**) were evaluated against human

hepatocellular carcinoma HepG2 and human breast carcinoma MCF-7 cell lines (obtained from American Type Culture Collection – Manassas, VA, USA).

The results of *in vitro* antiproliferative activity of indoloquinolines against two different cells (HepG-2 and MCF-7) are presented in table 1 below.

Table 1: Antiproliferative Activity of Indoloquinoline Derivatives against HepG2 and MCF-7 cells

Compds.	HepG2 IC_{50} (mg/mL)	MCF-7 IC_{50} (mg/mL)
Compds. (**5**+**6**) – **3e**	0.0519	0.0428
Compds. (**8**+**9**+**10**) – **3f**	0.8283	0.0714

The results suggest that the cell growth inhibitory activity of compound **3e** and **3f** against HepG2 and MCF-7 cells are moderately active.

Conclusion:

We have synthesized chloro-substituted linear and angular indoloquinolines in a one-pot experiment and evaluated their cytotoxicity against HepG2 and MCF-7 cells.

Experimental:

General procedure:

Indole-3-carboxyaldehyde **1** (3.46 mmol), chloroanilines **4** / **7** (6.92 mmol) and iodine (0.35 mmol) was refluxed in diphenyl ether (20 mL) for 12 hours. After cooling, reaction mixture was chromatographed on alumina and diphenyl ether was removed using hexanes as an eluent. Excess chloroanilines were eluted using 5% ethyl acetate in hexanes as an eluent. Further elution with 20% ethyl acetate in hexanes afforded the mixture of linear and angular indoloquinolines.

2-Chloro-6*H*-indolo[2,3-*b*]quinoline (5) & 3-chloro-5*H*-indolo[3,2-*c*]quinoline (6) as a inseparable mixture after column chromatography: Yield 34% (0.2964g); white solid; LC-MS shows two peaks at retention time 3.91 and 5.72 with same *m/z* 253 (^{35}Cl, $M+H)^+$ and 255 (^{37}Cl, $M+H)^+$ respectively; HRMS *m/z* $[M+H]^+$ 253.0533 (calcd for $C_{15}H_{10}ClN_2$, 253.0532); IR (KBr) 3142, 3090, 1614, 1580, 1460, 1408, 1329, 1230, 1126, 908, 820, 787, 737, 696 cm^{-1}. ^1H NMR (300 MHz, DMSO-d_6) of compound **(5) – (Fig. i)** - δ 7.29 (bt, 1H), 7.53 (m, 3H), 7.97 (d, 1H, *J* = 9.2 Hz), 8.12 (d, 1H, *J* = 9.2 Hz), 8.24 (d, 1H, *J* = 8.0 Hz), 9.02 (s, 1H), 11.79 (s, 1H, -NH). ^{13}C NMR (75 MHz, DMSO-d_6) – **(Fig. ii)** - δ 112.5, 118.3, 119.1, 120.8, 122.5, 124.7, 126.5, 127.2, 127.5, 128.8, 129.1, 129.2, 144.1, 145.7, 153.4. ^1H NMR (300 MHz, DMSO-d_6) of compound **(6) – (Fig. i)** - δ 7.36 (bt, 1H), 7.72 (m, 3H), 8.20 (d, 1H, *J* = 2.3 Hz), 8.32 (d, 1H, *J* = 7.8 Hz), 8.61 (s, 1H),), 9.61 (s, 1H), 12.81 (s, 1H, -NH). ^{13}C NMR (75 MHz, DMSO-d_6) – **(Fig. ii)** - δ 111.6, 115.3, 120.3, 120.4, 121.4, 121.6, 122.0, 127.2, 129.3, 129.4, 130.5, 132.0, 139.3, 142.1, 145.0.

1-Chloro-6*H*-indolo[2,3-*b*]quinoline (8), 3-chloro-6*H*-indolo[2,3-*b*]quinoline (9) & 2-chloro-5*H*-indolo[3,2-*c*]quinoline (10) as a inseparable mixture after column chromatography: Yield 32% (0.2790g); white solid; LC-MS shows three peaks, one at retention time 3.82 and two overlapping peaks at retention time 5.70 and 5.73 with same m/z 253 (^{35}Cl, $M+H)^+$ and 255 (^{37}Cl, $M+H)^+$ respectively; HRMS *m/z* $[M+H]^+$ 253.0532 (calcd for $C_{15}H_{10}ClN_2$, 253.0532); IR (KBr) 3142, 3090, 1614, 1580, 1460, 1408, 1329, 1230, 1126, 908, 820, 787, 737, 696 cm^{-1}. ^1H NMR (300 MHz, DMSO-d_6) of compounds **(8 & 9) – (Fig. iii)** - δ 7.30 (m, 2H), 7.52 (m, 7H), 7.98 (d, 1H, *J* = 8.7 Hz), 8.0 (s, 1H), 8.15 (d, 1H, *J* = 10.8 Hz), 8.16 (s, 1H), 7.73 (m, 1H), 9.08 (s, 1H), 9.29 (s, 1H), 11.83 (s, 1H, -NH), 11.89 (s, 1H, -NH). ^{13}C NMR (75 MHz, DMSO-d_6) – **(Fig. iv)** - δ 112.4, 119.4, 120.8, 122.5, 123.0, 123.6, 124.3, 126.4, 126.7, 126.8, 129.1, 140.3, 144.9, 146.9, 153.1.

^1H NMR (300 MHz, DMSO-d_6) of compound **(10) – (Fig. iii)** - δ 7.36 (bt, 1H), 7.73 (m, 2H), 8.26 (s, 1H), 8.33 (d, 1H, *J* = 7.8 Hz), 8.43 (d, 1H, *J* = 7.8 Hz), 8.55 (d, 1H, *J* = 8.7 Hz), 9.63 (s, 1H), 12.85 (s, 1H, -NH). ^{13}C NMR (75 MHz, DMSO-d_6) – **(Fig. iv)** - δ 111.6, 115.0, 115.7, 120.3, 120.5, 121.6, 121.7, 121.9, 127.3, 129.3, 131.3, 133.6, 139.3, 141.9, 145.6.

Fig. i: ^{1}H NMR spectrum of **(5+6)**

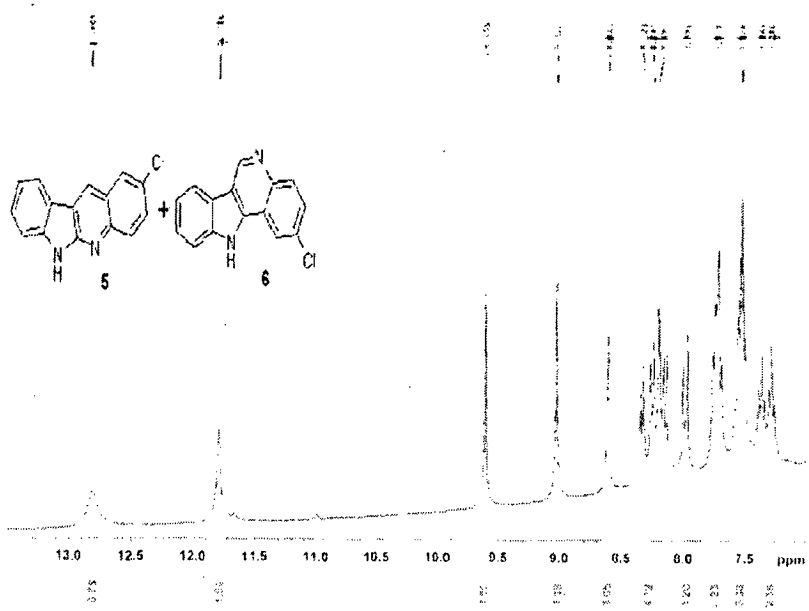


Fig. ii: ^{13}C NMR spectrum of **(5+6)**

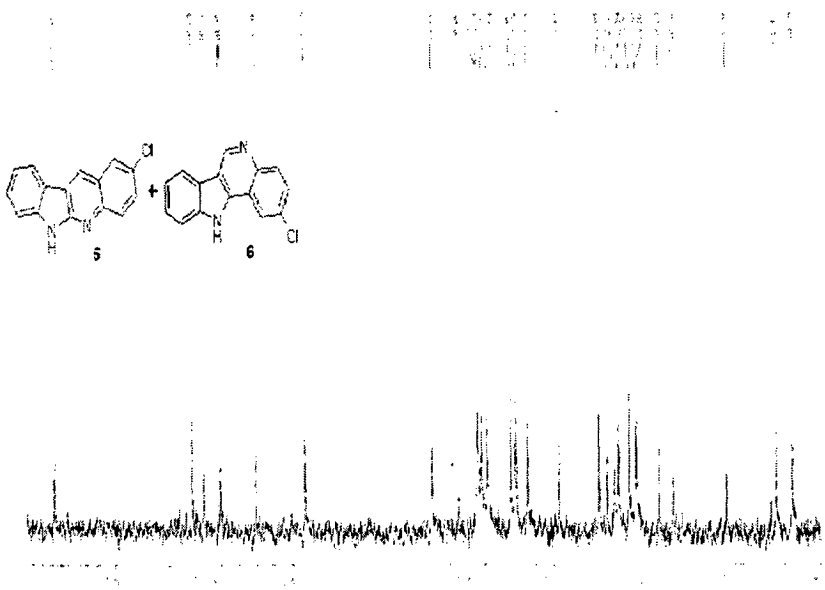


Fig. iii: ^{1}H NMR spectrum of **(8+9+10)**

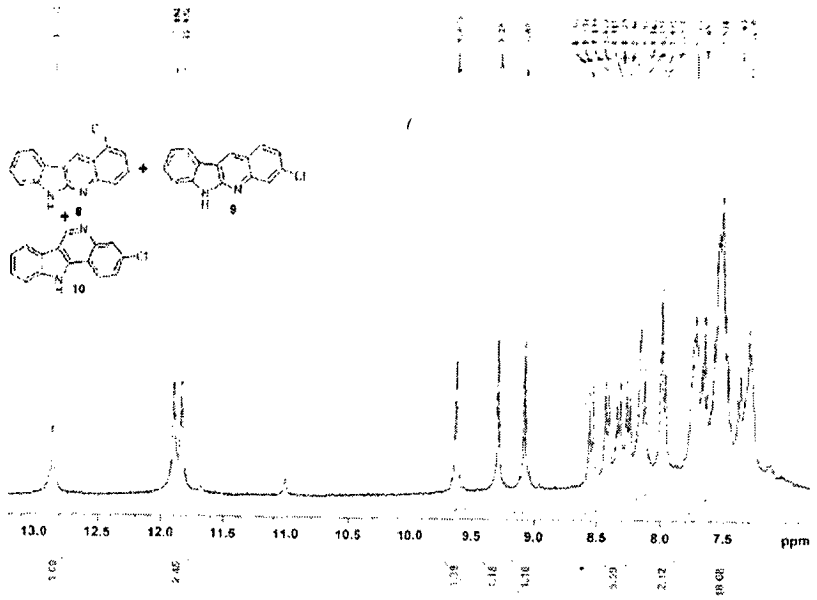
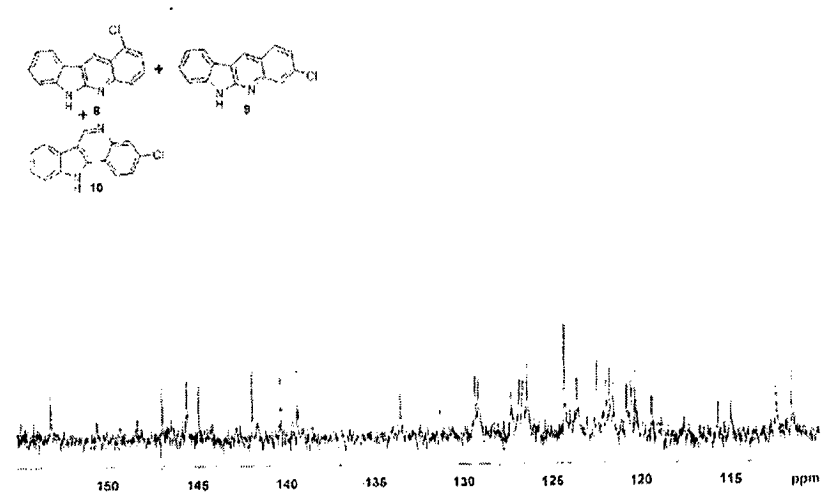


Fig. iv: ^{13}C NMR spectrum of **(8+9+10)**



CHAPTER 3

SYNTHETIC STUDIES TOWARDS MARINE NATURAL PRODUCTS ARCYRIAFLAVIN A, STAUROSPORINONE AND CAULERSIN

SYNTHETIC STUDIES TOWARDS MARINE NATURAL PRODUCTS ARCYRIAFLAVIN A, STAUROSPORINONE AND CAULERSIN

Section A: Synthetic Studies Towards Indolocarbazole Alkaloids – Arcyriaflavin- A and Staurosporinone

Introduction:

Indolocarbazoles (ICZs) are a class of compounds that are under current study due to their potential as anti-cancer drugs. Different arrangements are possible for indole and carbazole to yield the five isomeric ring systems for ICZs – indolo[2,3-*a*]carbazole **1**, indolo[2,3-*b*]carbazole **2**, indolo[2,3-*c*]carbazole **3**, indolo[3,2-*a*]carbazole **4** and indolo[3,2-*b*]carbazole **5** (Fig. 1). However, almost all of the ICZs isolated from nature are indolo[2,3-a]carbazoles which were obtained either from soil organisms, slime molds or marine sources.[1-3] The indolo[2,3-*a*]carbazole alkaloids display wide range of biological activities such as antifungal, antimicrobial, antitumor, cytotoxic and antihypertensive activities as well as inhibition of various serine-threonine and tyrosine specific protein kinases.[4]

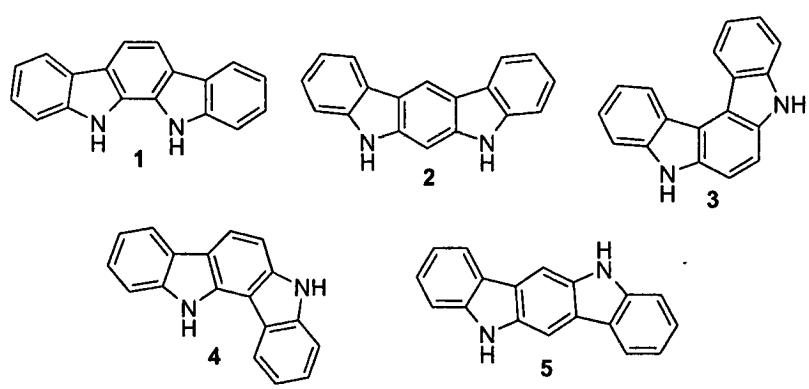


Fig. 1

The isolation of an indolocarbazole alkaloid from a natural source was first reported in 1977 by Omura *et al.*[5] The alkaloid, initially named AM-228, was extracted from the cultures of *Streptomyces staurosporeus* AM-2282, which was collected from soil sample and soon after it was renamed as staurosporine (STA) **6**. Thereafter, a variety of ICZs have been isolated from different organisms, including

bacteria, fungi and invertebrates. The indolocarbazole alkaloid **7** was first isolated from the fungi, *Nocardiopsis sp.*[6] and was named as staurosporinone or K-252c. The structure elucidation showed that K-252c represents the aglycon of staurosporine, therefore it is also referred as staurosporine aglycon. Later in 1980, another indolocarbazole alkaloid similar to staurosporinone was isolated from the fungi, *Arcyrai denudata*[7] by Steglich and co-workers and named it as arcyriaflavin A **8**. In 1994, the alkaloids **7** and **8** were again isolated from a common source i.e. specimen of the marine ascidian, *Eudistoma sp.*, collected off the coast of West Africa.[8]

Staurosporine **6**, staurosporinone **7** and arcyriaflavin A **8** (Fig. 2) are in fact the derivatives of indolo[2,3-*a*]pyrrolo[3,4-*c*]carbazole ring system, but for simplicity, they will be designated as indolo[2,3-*a*]carbazoles.

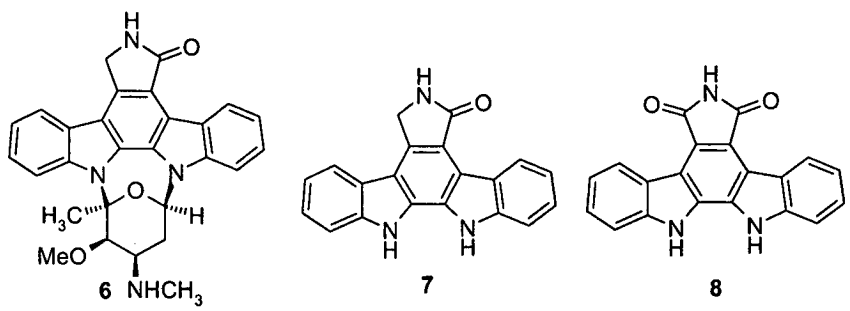
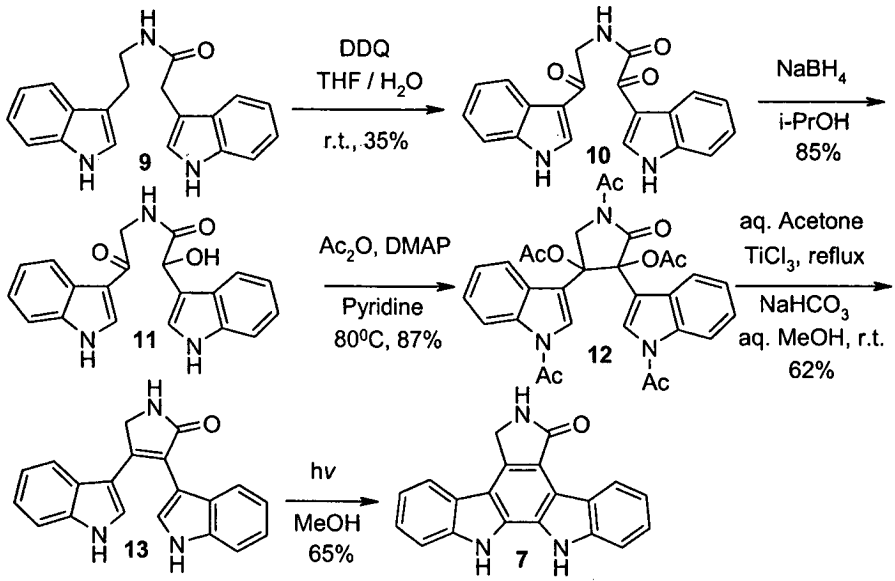


Fig. 2

Natural products incorporating the indolo[2,3-*a*]carbazole ring system have been, since their isolation, the target of synthesis owing to their diverse and in some instances extraordinary biological properties, such as inhibition of protein kinase C (PKC), platelet aggression and cytotoxic activity. A number of synthetic approaches have been developed to prepare staurosporinone and arcyriaflavin A which have been briefly discussed below.

Literature Synthetic Methods:

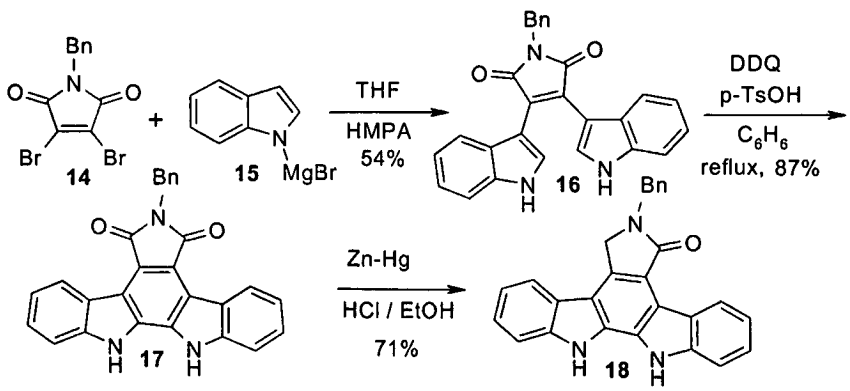
Winterfeldt *et al.*[14] described the first synthesis of staurosporinone **7** starting from tryptamine and 3-indolyl-acetate chloride by sequential base- and photo-induced cyclization (Scheme 1).



Scheme 1

Acylation of tryptamine by 3-indolylacetyl chloride provided the amide **9** in 77% yield which was oxidized with DDQ to give the corresponding diketone **10**. Reduction of **10** with $NaBH_4$ afforded the hydroxyl ketone **11** which was then heated at 80^0C in acetic anhydride in presence of DMAP and pyridine to afford the cyclized product i.e. pentaacetyl derivative **12**. Reduction of **12** with $TiCl_3$ followed by deacylation and photoinduced cyclization furnished the target molecule **7**.

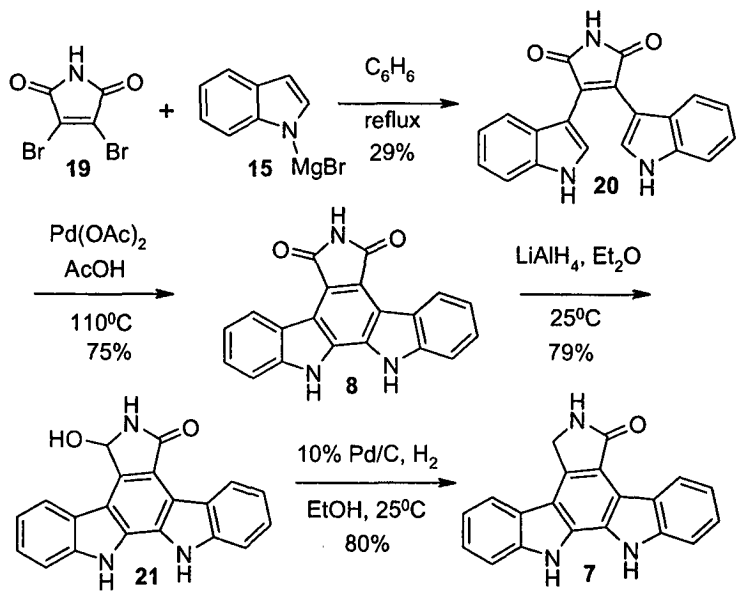
Weinreb and co-workers[15] reported the synthesis of *N*-benzylarcyriaflavin A **17** using double alkylation and oxidative cyclization and then converted it to *N*-benzylstaurosporinone **18** using Clemensen reduction (Scheme 2).



Scheme 2

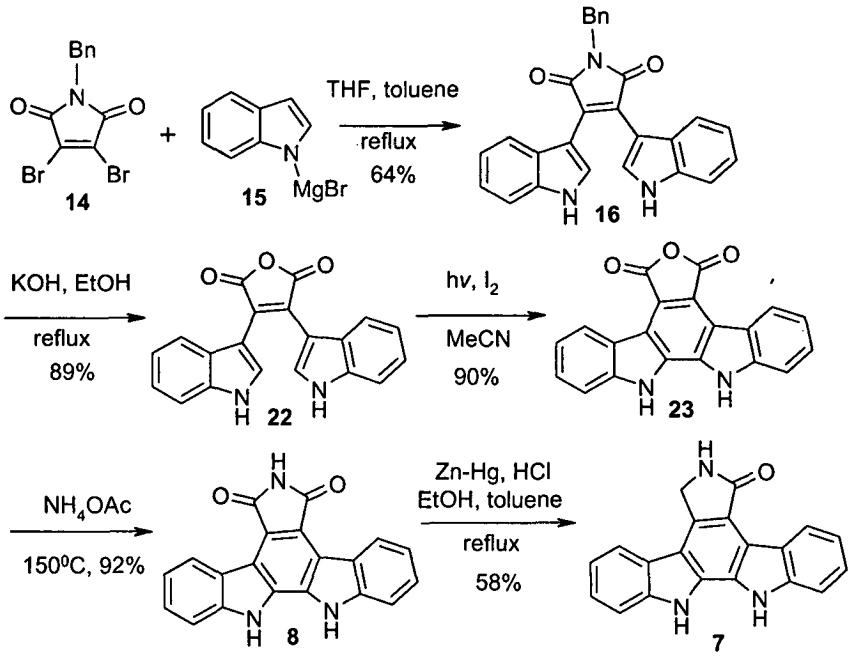
Reaction of *N*-benzyldibromomaleimide **14** with indolylmagnassium bromide **15** led to the bisindolylmaleimide **16** which on oxidative cyclization using a mixture of DDQ and *p*-TsOH provided the *N*-benzyl arcyriaflavin A **17**. Reduction with Zn-Hg (Clemmensen reduction) afforded the *N*-benzylstaurosporinone **18** in 71% yield.

Hill and co-workers[16] achieved the synthesis of arcyriaflavin A **8** using a palladium-mediated oxidative cyclization as the key step and then transformed it into staurosporinone **7** *via* reduction with $LiAlH_4$ followed by hydrogenolytic deoxygenation of the hydroxyl lactam (Scheme 3).



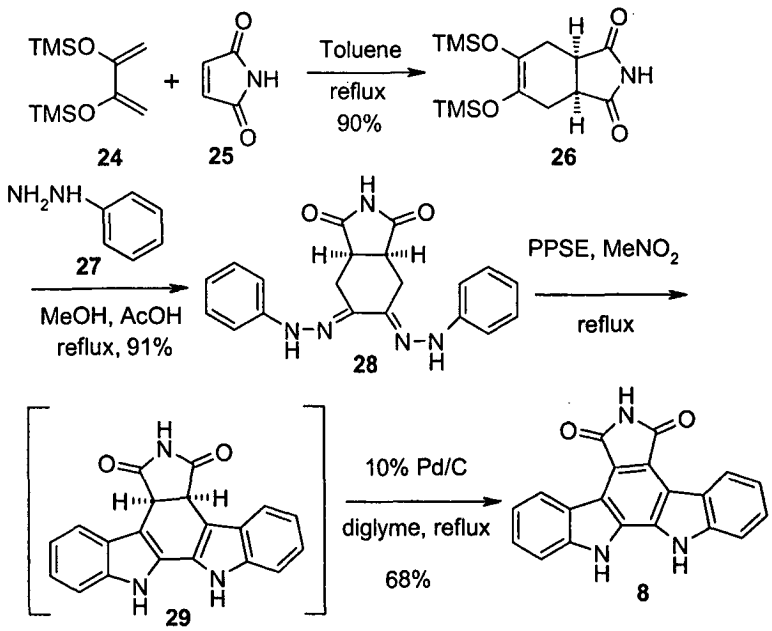
Scheme 3

Xie and Lown[17] reported a facile synthesis of arcyriaflavin A **8** involving photochemically induced oxidative cyclization as the key step which was then converted to staurosporinone **7** by reduction with zinc amalgam (Scheme 4).



Scheme 4

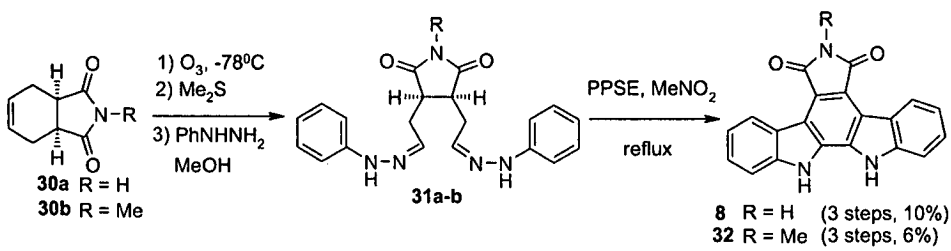
Bergman and Pelcman[18] described the synthesis of arcyriaflavin A using Diels-Alder and double Fischer indolization as main steps (Scheme 5).



Scheme 5

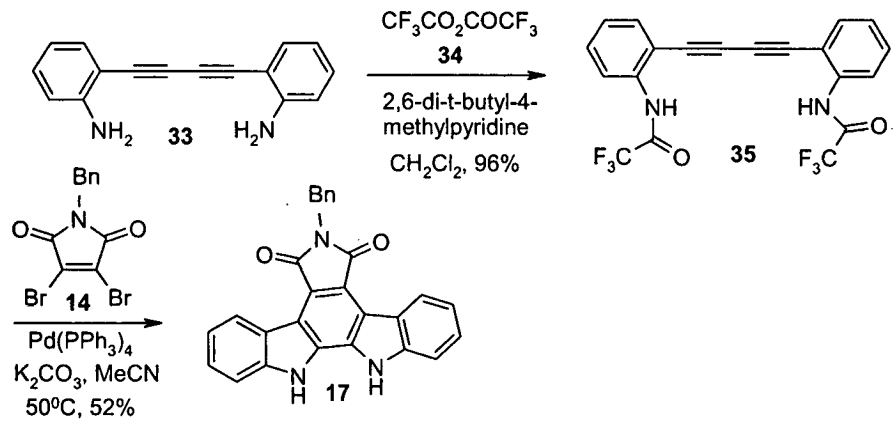
A Diels-Alder cycloaddition of 2,3-bis(trimethylsilyloxy)butadiene **24** with maleimide **25** gave compound **26**. Subsequent condensation with phenylhydrazine **27** in presence of acetic acid afforded the bis-(phenylhydrazone) **28** which underwent double Fischer indolization when treated with polyphosphoric acid trimethylsilyl ester (PPSE) to give compound **29** which without isolation was dehydrogenated to furnish arcyriaflavin A **8**.

Gribble and Berthel[19] achieved the synthesis of Arcyriaflavin A **8** and 6-methyl Arcyriaflavin A **32** from commercially available cyclohexene imides **30a-b** which were oxidized to labile dialdehydes and then treated with two equivalents of phenylhydrazine to give the corresponding bis-(phenylhydrazones) **31a-b**. Subsequent double Fischer indolization of the resultant bis-(phenylhydrazones) **31a-b** with polyphosphoric acid trimethylsilyl ether (PPSE) in nitromethane under reflux[18] afforded the arcyriaflavin A **8** and 6-methyl arcyriaflavin A **32** respectively (Scheme 6).



Scheme 6

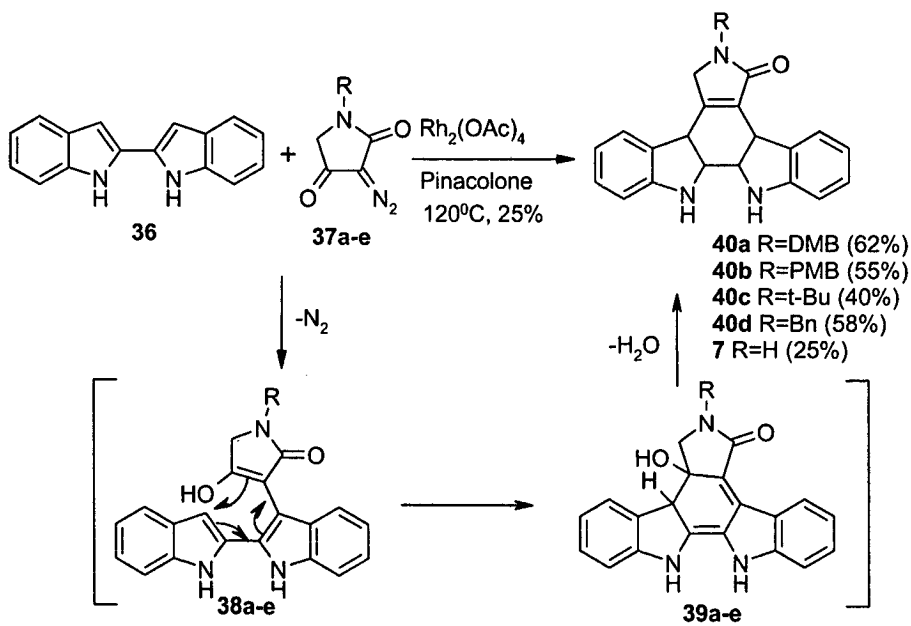
Saulnier *et al.*[20] reported the synthesis of *N*-benzylarcyriaflavin A starting from diacetylene derivative **33** (Scheme 7).



Scheme 7

Diacetylene derivative **33** on treatment with trifluoroacetic anhydride **34** afforded the bistrifluoroacetanilide **35**. The reaction of **35** with *N*-benzyldibromomaleimide **14** in presence of Pd(0) catalyst furnishes the *N*-benzylarcyriaflavin A **17** in 52% yield.

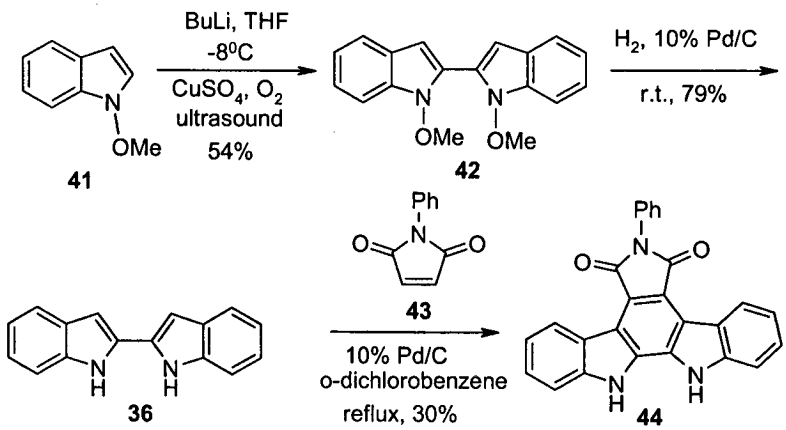
Wood *et al.*[21] accomplished the synthesis of staurosporinone **7** and the other *N*-substituted analogues **40a-d** using 2,2'-bisindole **36** and diazolactams **37a-e** as the starting materials (Scheme 8).



Scheme 8

The 2,2'-bisindole **36** was prepared by a double Madelung cyclization as reported by Bergman[22] while diazolactams **37a-e**[23] were prepared from *N*-substituted glycene esters by DCC/DMAP promoted coupling with ethyl hydrogen malonate, followed by Dieckmann cyclization and the subsequent treatment with mesyl azide (MsN_3). Coupling of diazolactams **37a-e** with 2,2'-bisindole **36** in presence of catalytic amount of $Rh_2(OAc)_2$ provided staurosporinone **7** and the other analogues **40a-d**.

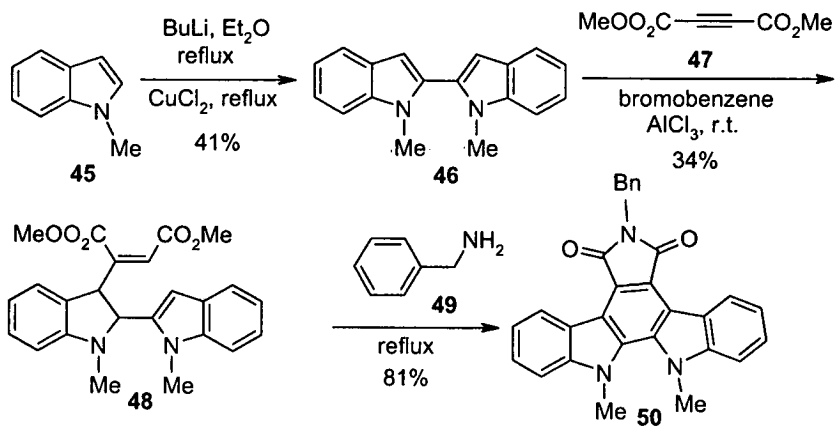
Somei and Kodama[24] described a synthesis of 6-phenylarcyriaflavin A **44** *via* oxidative coupling and Diels-Alder cycloaddition as the main steps (Scheme 9).



Scheme 9

Deprotonation of 1-methoxyindole **41** with BuLi followed by a novel oxidative coupling using ultrasound technique under anhydrous $CuSO_4$ and oxygen atmosphere gave 2,2'-bis-(1-methoxyindole) **42** in 54% yield. Catalytic hydrogenolysis of **42** provided 2,2'-bisindole **36** and subsequent Diels-Alder cycloaddition with *N*-phenylmaleimide **43** in presence of catalytic amount of 10% Pd/C afforded 6-phenylarcyriaflavin A.

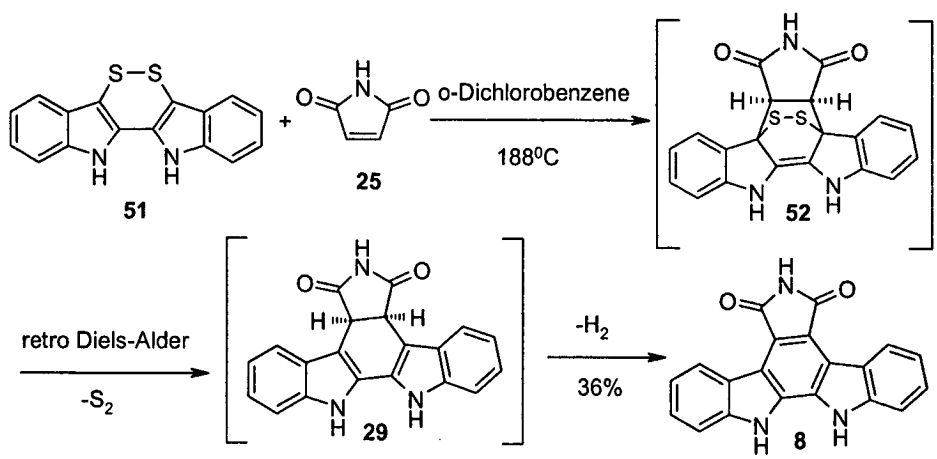
Pindur *et al.*[25] achieved a synthesis of 6-benzyl-12,13-dimethylarcyriaflavin A **50** in which the key steps are oxidative coupling, Michael addition and cyclization (Scheme 10).



Scheme 10

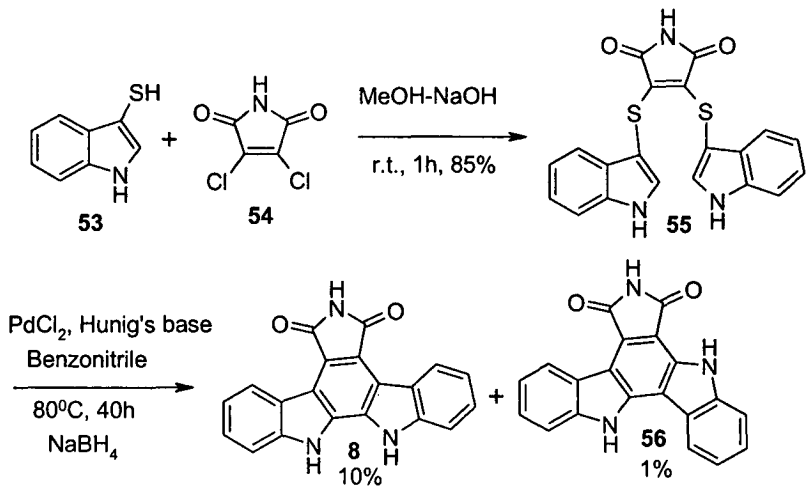
The reaction of 2,2'-bis(1-methylindole) **46** with dimethyl acetylenedicarboxylate (DMAD) **47** in presence of aluminium trichloride afforded the 2,2'-bisindol-3-yl-substituted dimethyl maleate **48** which on cyclization with benzylamine led directly to 6-benzyl-12,13-dimethylarcyriaflavin A **50**.

Lobo and co-workers[26] reported a synthesis of arcyriaflavin A **8** starting from 2,2'-bisindolyl-3,3'-dithiete[27] **51** and maleimide **25** *via* Diels-Alder reaction followed by a retro-Diels-Alder reaction with extrusion of S_2 and dehydrogenation by dissolved oxygen or extruded sulfur (Scheme 11).

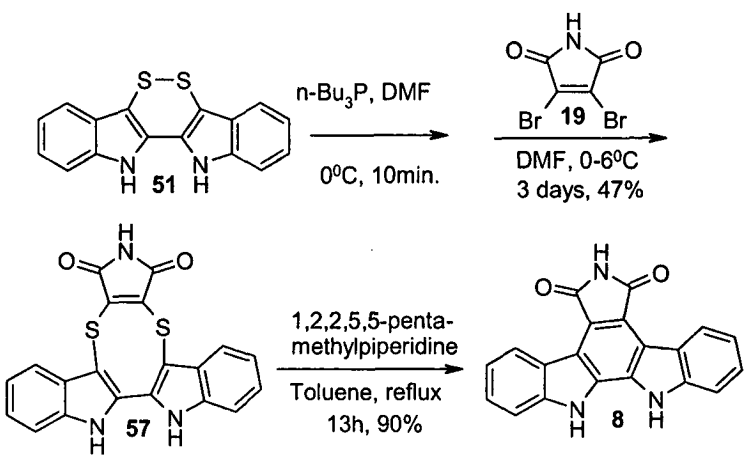


Scheme 11

The same group[28,29] developed the synthesis of arcyriaflavin A **8** by a 2-fold sulfur extrusion reaction (Scheme 12 & 13).

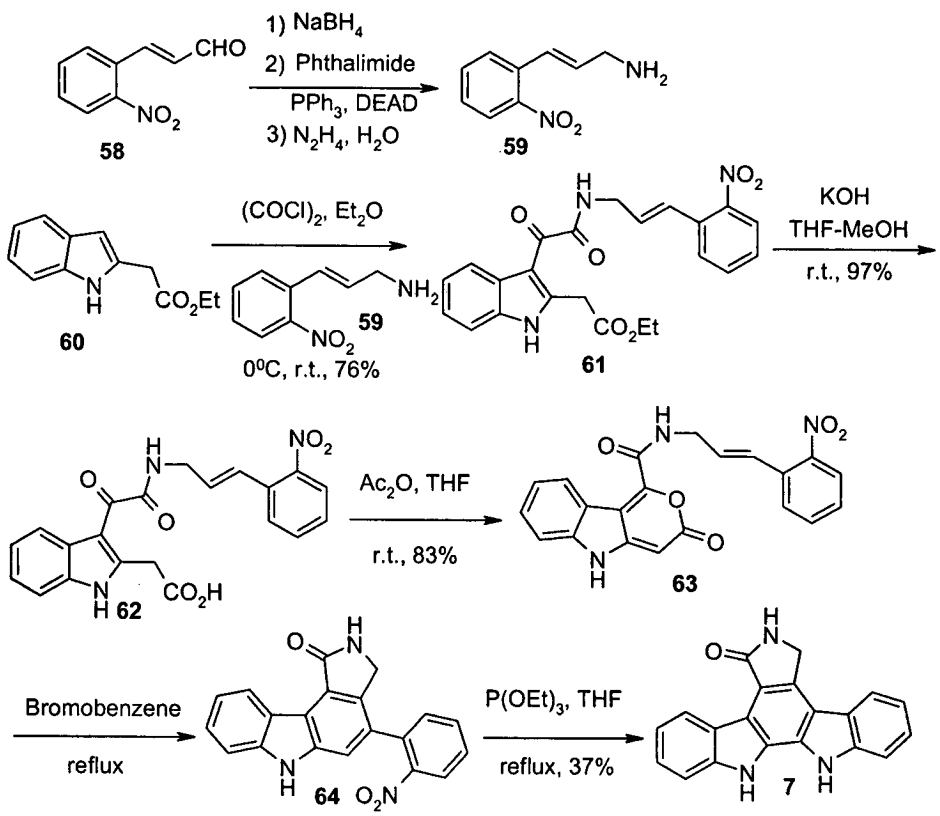


Scheme 12



Scheme 13

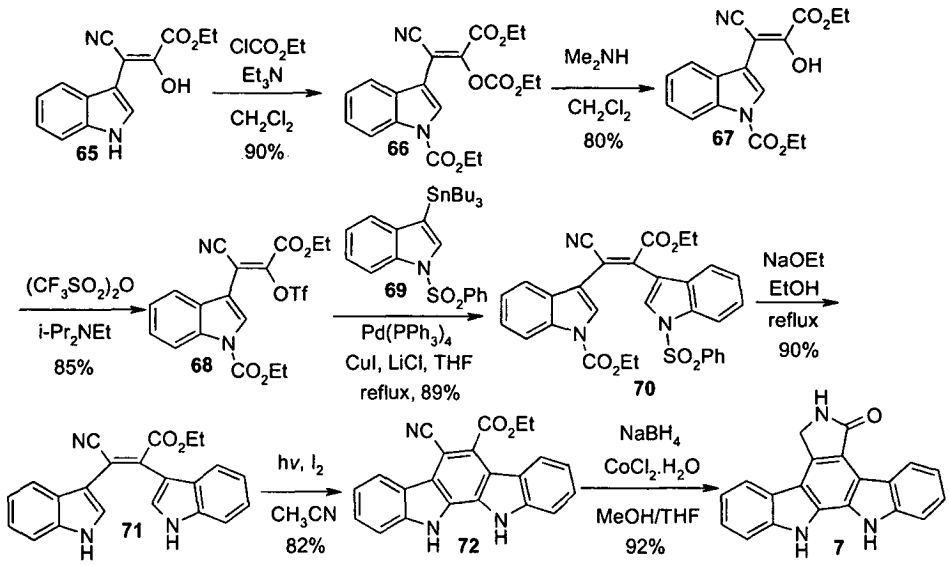
Moody and Rahimtoola[30] reported a synthesis of staurosporinone **7** using intramolecular Diels-Alder reaction and reductive cyclization as the key steps (Scheme 14).



Scheme 14

Reaction of ethyl indol-2-yl acetate **60** with oxalyl chloride followed by quenching with *o*-nitrocinnamylamine **59** yielded the 2,3-disubstituted indole **61**. Hydrolysis of the ester **61** followed by cyclodehydration of the keto acid **62** with acetic anhydride led to the pyrano[4,3-*b*]indol-3-one **63**. Intramolecular Diels-Alder reaction of **63** followed by reductive cyclization of the resultant carbazole **64** with triethyl phosphite provided staurosporinone **7**.

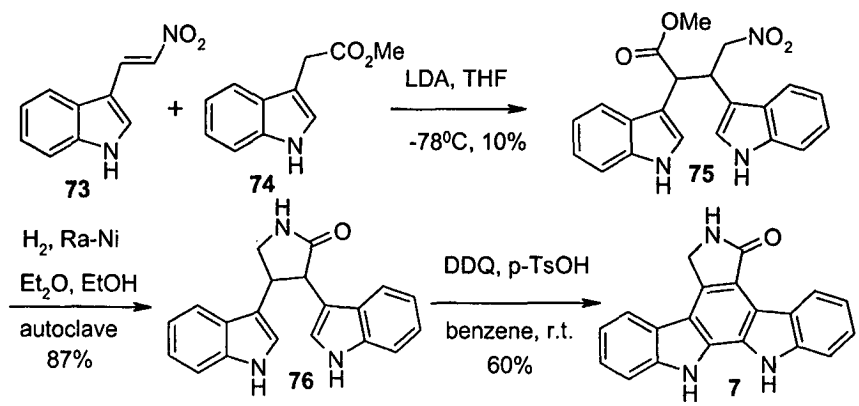
Beccalli *et al.*[31] described a synthesis of staurosporinone **7** involving palladium-catalyzed cross coupling and oxidative photocyclization as the major reactions (Scheme 15).



Scheme 15

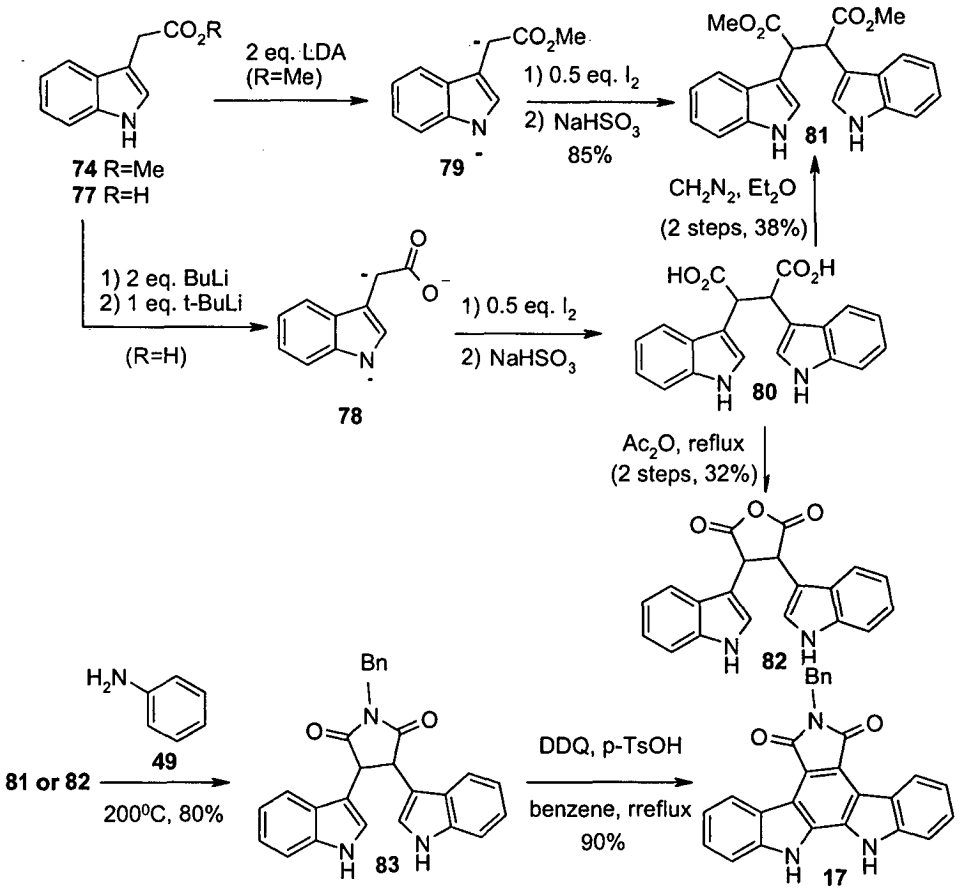
Reaction of **65** with ethyl chloroformate gave compound **66** which was selectively deethoxycarbonylated with dimethylamine and then treated with trifluromethanesulfonic anhydride to give the corresponding triflate **68**. The Pd-catalyzed coupling with 1-phenylsulfonyl-3-tributylstannylindole **69** provided 1,2-bis-(indol-3-yl)-cis-alkene **70** which on deprotection and subsequent oxidative photocyclization afforded 5,6-disustituted indolo[2,3-*a*]carbazole **72**. Reductive cyclization of **72** with $NaBH_4$ and cobalt(II)chloride provided staurosporinone **7**.

Mahboobi *et al.*[32] reported a synthesis of staurosporinone **7** using Michael addition and oxidative cyclization as the main steps (Scheme 16).



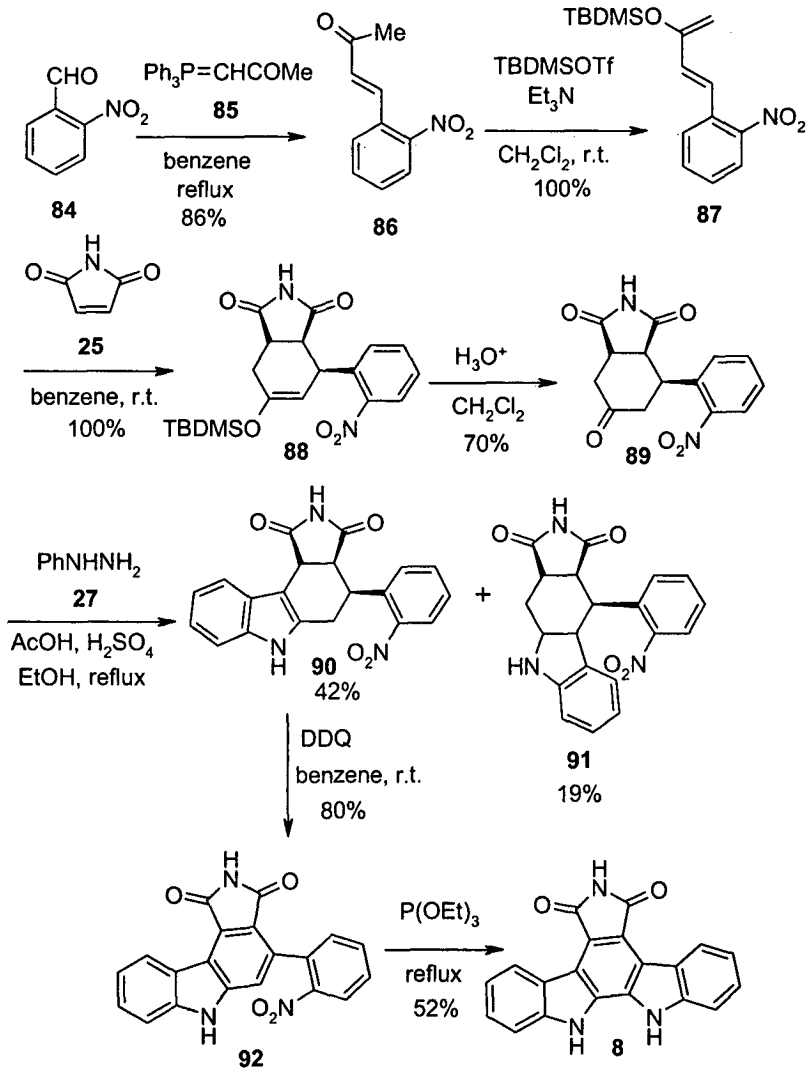
Scheme 16

Bergman and coworkers[33] described a synthesis of 6-benzylarcyriaflavin A **17** which involves iodine-promoted oxidative coupling as the key step (Scheme 17).



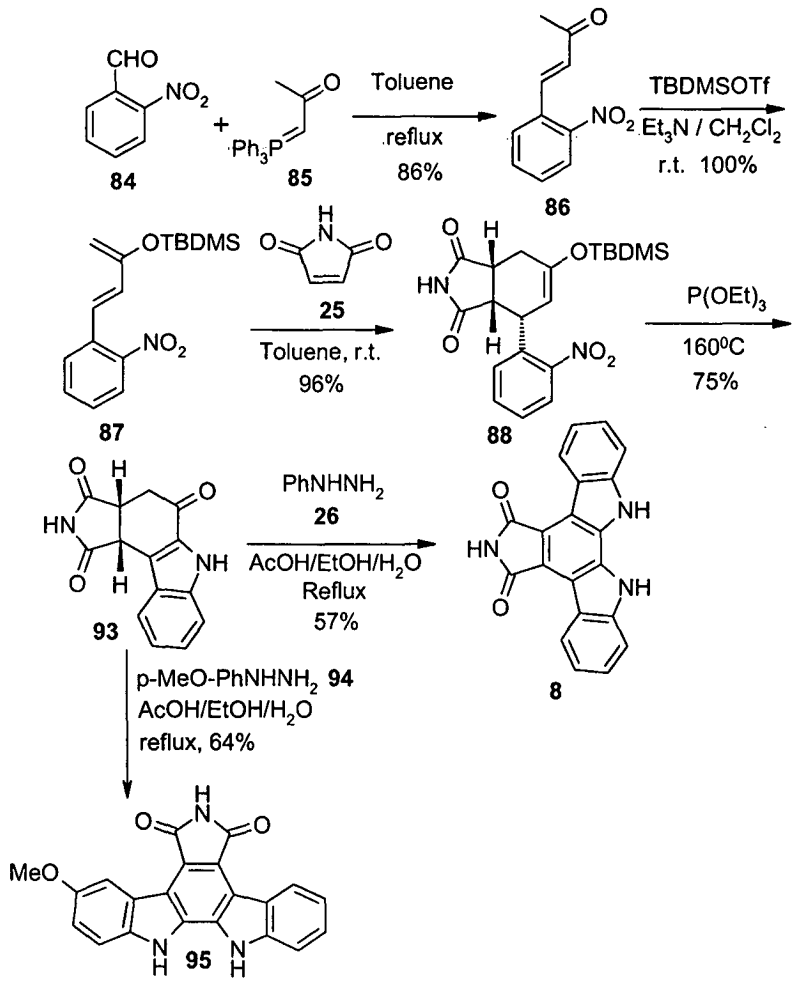
Scheme 17

Tome and co-workers[34] achieved a synthesis of arcyriaflavin A *via* successive Wittig reaction, Diels-Alder cycloaddition, Fischer indolization and nitrene insertion (Scheme 18).



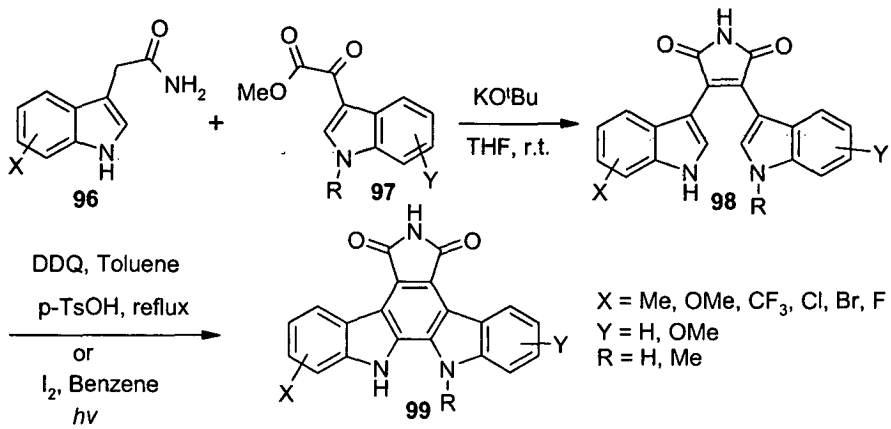
Scheme 18

The same group[35] developed a new high-yielding method for the synthesis of the alkaloid arcyriaflavin A and its 3-substituted unsymmetrical analogue *via* sequential aryl-siloxy-diene Diels-Alder reaction, nitro-reductive silyl enol ether-mediated indolization and Fischer indolization (Scheme 19).



Scheme 19

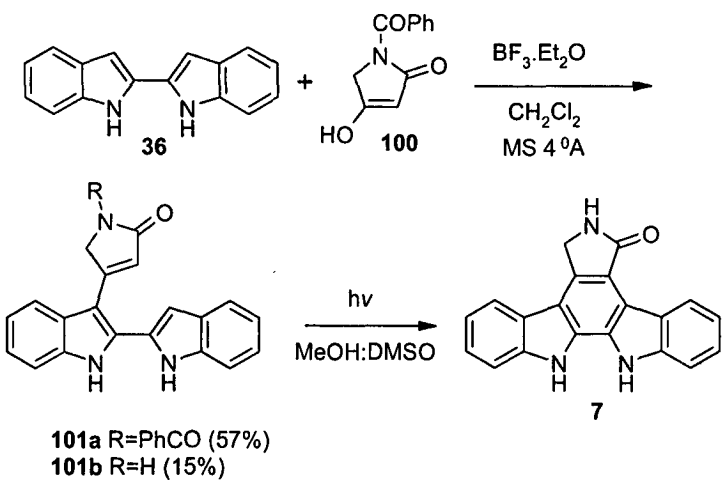
Zhu *et al.*[36] reported the synthesis of arcyriaflavin A and its analogues using oxidative cyclization as the main step (Scheme 20).



96 **97** **98** **99**

Scheme 20

Prabhakar and co-workers[37] described a short synthesis of staurosporinone *via* condensation of biindole with 3-keto-γ-lactam and photocyclization (Scheme 21).

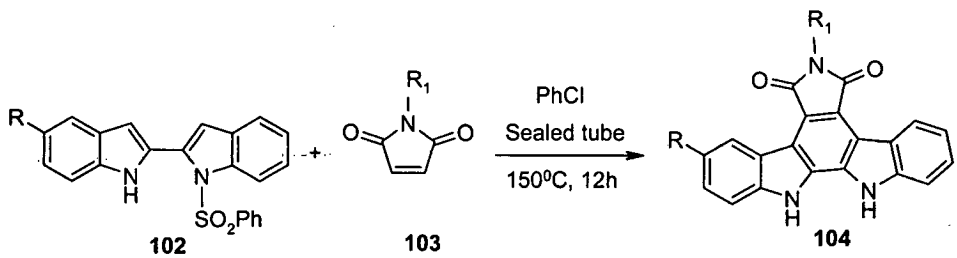


36 **100** **7**

101a R=PhCO (57%)
101b R=H (15%)

Scheme 21

Kuethe and Davies[38] reported an improved synthesis of indolo[2,3-*a*]carbazoles *via* [4+2] cycloaddition using *N*-sulfonyl-2,2'-biindoles and *N*-protected maleimides as the starting materials (Scheme 22). Earlier Wallace and co-workers[39] had reported the [4+2] cycloaddition approach to indolo[2,3-*a*]carbazoles in low yields using 2,2'-biindole and maleimides or dimethyl acetylene dicarboxylate (DMAD) as the starting materials.



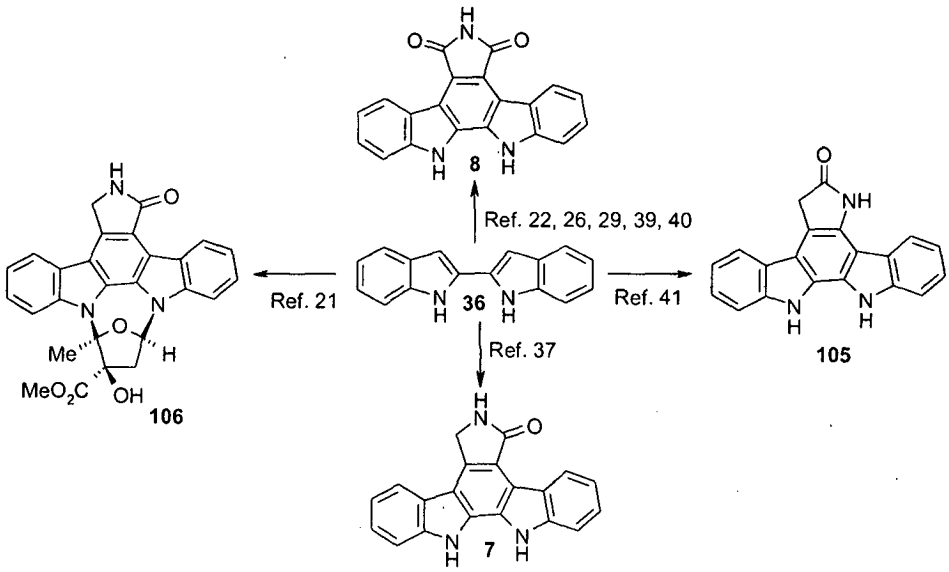
R = Cl, R_1 = Bn (50%)
R = Cl, R_1 = Me (47%)
R = Me, R_1 = Ph (42%)

Scheme 22

Results and Discussion (Present Synthetic Work):

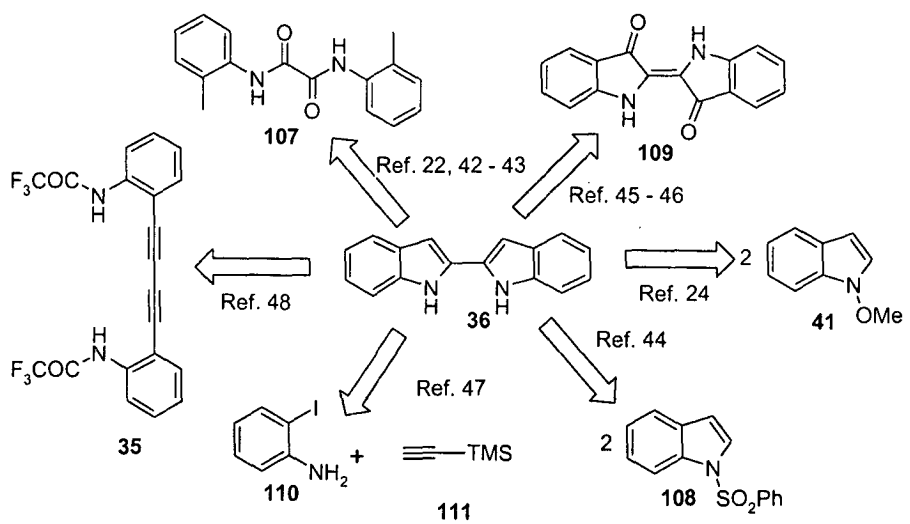
Careful examination of the literature methods revealed that there are large numbers of methods available for the synthesis of these important indolocarbazole alkaloids. However, we felt that there is enough scope for us to develop a convenient method.

In recent years, domino reactions have attracted considerable attention of synthetic organic chemists as they provide easy entry to complex molecules by including two or more transformations in one-pot. So, we decided to synthesize 2,2'-biindole in one-pot as it is a core structural motif of several biologically active indolocarbazole alkaloids like staurosporine, staurosporinone, rebeccamycin, tjipanazole and arcyriaflavins. Some of these alkaloids have been prepared from 2,2'-biindole as depicted below (Scheme 23).



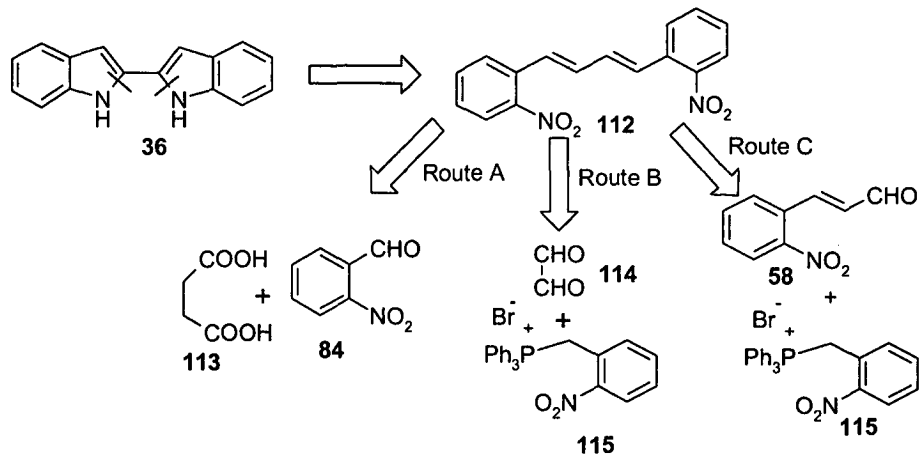
Scheme 23

2,2'-Biindole **36** was first synthesized by Madelung[42] from *o*-toluidine and thereafter a number of diverse synthetic approaches[22, 24, 43-48] have been developed (Scheme 24).



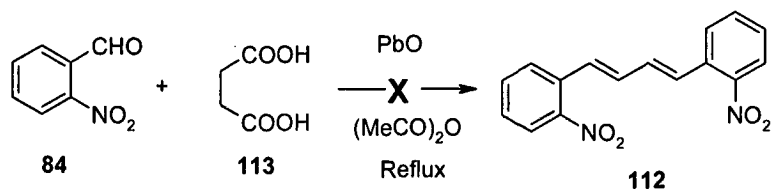
Scheme 24

Our approach to 2,2'-biindole **36** is based on the retro-synthetic analysis depicted below (Scheme 25), through the preparation of 1,4-bis-(*o*-nitro-phenyl)-1,3-butadiene **112**, a precursor for double reductive cyclization to be obtained either by Wittig reaction or Perkin reaction.



Scheme 25

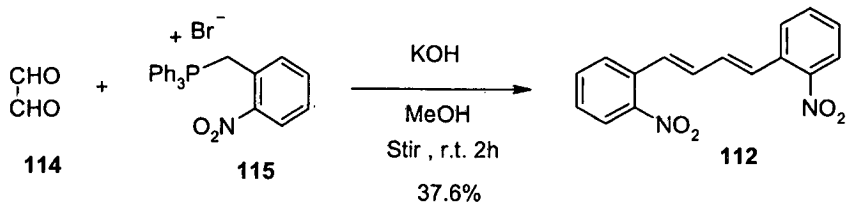
Initially, we tried to synthesize 1,4-bis-(*o*-nitro-phenyl)-1,3-butadiene using 2-nitro-benzaldehyde and succinic acid (route A) as the starting material (Scheme 26).



Scheme 26

We are expecting nitro-dimer **112** to form *via* double Perkin reaction followed by decarboxylation, but the reaction did not furnish the product under the reaction condition employed.

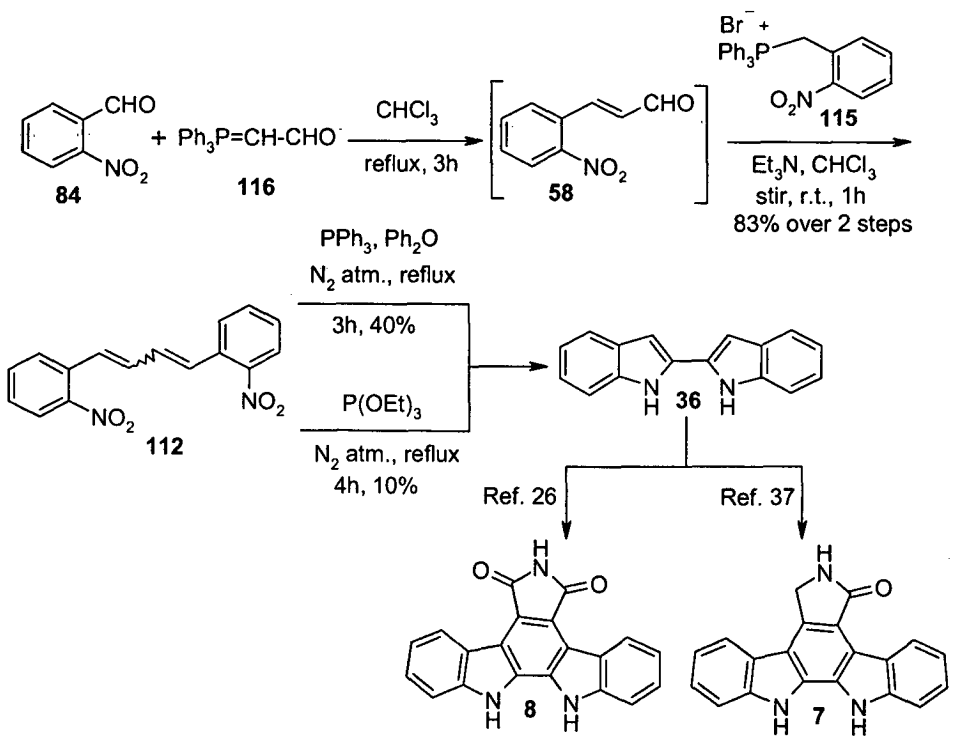
As above strategy did not afford the required product, we attempted an alternate route i.e. route B for the synthesis of nitro-dimer **112** which involves double Wittig reaction (Scheme 27).



Scheme 27

When the reaction was carried out by stirring *o*-nitrobenzyl-triphenylphosphonium bromide **115** (which was prepared from *o*-nitro-benzylbromide and triphenyl phosphine as the starting materials) and aqueous glyoxal **114** in presence of KOH, the 1,4-bis-(*o*-nitro-phenyl)-1,3-butadiene **112** is obtained in only 37.6% yield. The IR spectrum showed strong bands at 1518 and 1348 cm^{-1} indicating the presence of nitro group. Due to the presence of *s*-cis and *s*-trans isomers of the compound, the ^1H NMR spectrum (Fig. 3) showed complex multiplets between δ 6.60 – 7.82 ppm. Finally, the formation of compound was confirmed by HRMS (Fig. 4) showing $[M+Na]^+$ peak at *m/z* 319.0692 for $C_{16}H_{12}N_2O_4Na$ (calcd for $C_{16}H_{12}N_2O_4Na$, 319.0695).

The low yield of the Wittig product may be due to the aqueous nature of glyoxal, so we thought of another route i.e. route C for the synthesis of nitro-dimer **112** which was then converted to 2,2'-biindole which constitutes the formal synthesis of naturally occurring indolocarbazole alkaloids – arcyriaflavin A and staurosporinone (Scheme 28).



Scheme 28

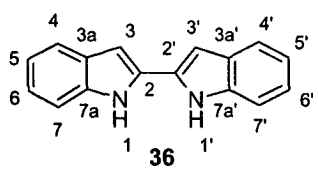
To begin with, *o*-nitro-benzaldehyde **84** was condensed with (triphenylphosphoranylidene)-acetaldehyde **116** yielding the Wittig product i.e. *o*-nitro-cinnamaldehyde **58** (monitored by TLC) which without isolation was treated with *o*-nitro-benzyl-triphenylphosphonium bromide **115** in presence of triethyl amine to give 1,4-bis-(*o*-nitro-phenyl)-1,3-butadiene **112** in 83% overall yield over two steps.

Once the sufficient amount of 1,4-bis-(*o*-nitro-phenyl)-1,3-butadiene **112** is in our hand, the next step is the double reductive cyclization to give the target molecule i.e 2,2'-biindole **36**. Initially the reductive cyclization was carried out using triethyl phosphite (TEP), but the yield of the product was only 10%, so the same reaction was attempted using triphenylphosphine in refluxing diphenyl ether. The yield of the product was increased from 10% to 40% using triphenyl phosphine. Further, to improve the yield of the product, the reductive cyclization was carried out by triphenylphosphine under mild reaction conditions i.e. in refluxing toluene using

dichlorodioxomolybdenum(VI) complex[49] as a catalyst. Disappointingly, the reaction failed to give the product. Even the microwave condition using triphenylphosphine failed to give the expected product, and we got some tarry material.

In IR spectrum, it showed a strong band at 3398 cm^{-1} indicating the presence of NH functionality. PMR spectrum showed six signals, all in the aromatic region between δ 6.93 – 11.57. The peaks at δ 11.57 integrating for two protons were attributed to –NH of two indole rings while the peaks between δ 6.93 – 7.56 were assigned to the remaining protons. Further, the assigned structure was confirmed by ^{13}C NMR spectrum and DEPT experiment. Its HRMS spectrum showed $[M+K]^+$ peak at *m/z* 271.0839 for $C_{16}H_{12}N_2K$ (Calcd for $C_{16}H_{12}N_2K$, 271.0838). The detailed spectroscopic data is described below.

Spectroscopic data:



36

IR (KBr): υ_{max} = 3398, 1442, 1396, 1340, 1261, 1070, 931, 800, 625 cm^{-1}

^{1}HNMR (300 MHz, DMSO-d_6): (Fig. 5)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
6.93	s	-	2	H-3 & H-3'
7.01	t	7.5	2	H-5 & H-5'
7.11	t	7.5	2	H-6 & H-6'
7.41	d	7.5	2	H-7 & H-7'
7.56	d	7.5	2	H-4 & H-4'
11.57	s	-	2	-NH

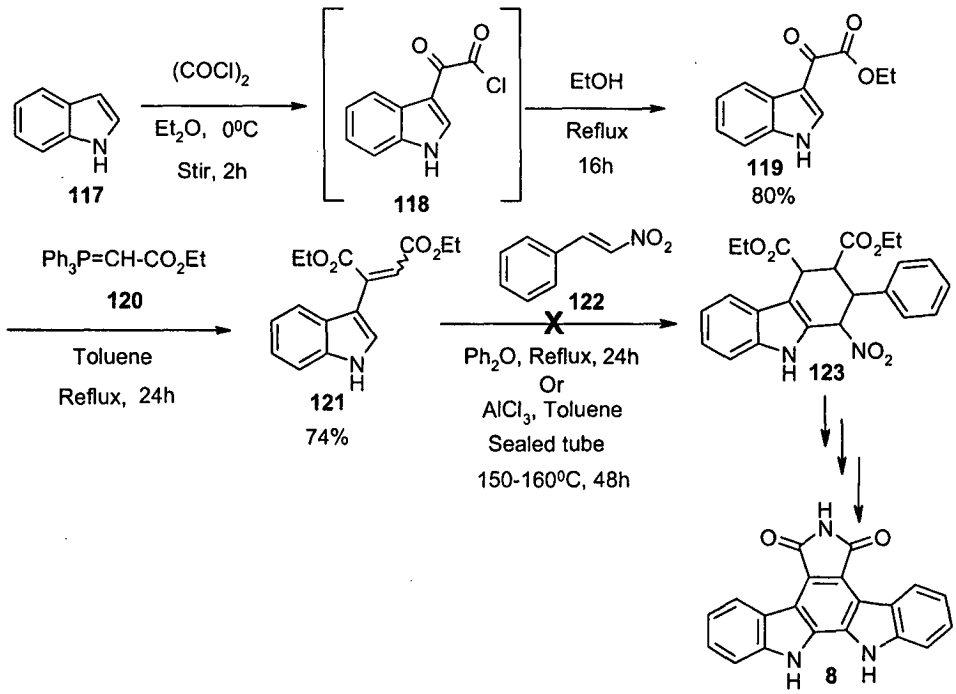
^{13}CNMR (75 MHz, DMSO-d_6): (Fig. 6)

δ 98.9 (C-3 & C-3'), 115.2 (C-5 & C-5'), 119.8 (C-7 & C-7'), 120.5 (C-6 & C-6'), 122.1 (C-4 & C-4'), 128.9 (C-3a & C-3a'), 131.9 (C-2 & C-2') and 137.4 (C-7a & C-7a').

HRMS: *m/z* $[M+K]^+$ 271.0839 (calcd for $C_{16}H_{12}N_2K$, 271.0838).

Melting Point: >300 ^{0}C (Lit.[50] Mp = 311-314^{0}C).

As the yield of the 2,2'-biindole **36** is low, we thought a new method for the synthesis of marine natural product arcyriaflavin A which involves Wittig reaction and Diels-Alder reaction as the key steps (Scheme 29). The arcyriaflavin A then can be converted to staurosporinone by selective reduction.

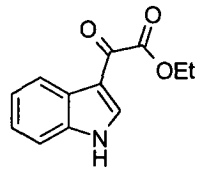


Scheme 29

Thus, indole was treated with oxalyl chloride at 0^{0}C in ether to give the corresponding chloro compound which without isolation was refluxed in ethanol to afford the required keto-ester **119** in 80% yield. In its IR spectrum, the strong peaks at 3329 and 1728 cm^{-1} indicates the presence of –NH and carbonyl functionality. In ^{1}H NMR spectrum (Fig. 7), a triplet at δ 1.34 integrating for three protons was attributed to $-CH_3$ while a quartet at δ 4.36 integrating for two protons was assigned to $-OCH_2-$ group. The signals between δ 7.25 – 8.42 were attributed to the aromatic protons and a broad singlet at δ 12.39 was assigned to the –NH proton of the indole ring. In ^{13}C NMR spectrum (Fig. 8), the peaks at δ 14.4 and 62.1 were assigned to $-CH_3$ and $-OCH_2-$ carbons respectively. The peaks of aromatic carbons appeared between δ

112.8 – 138.6 while the peaks at δ 164.0 and 179.6 were assigned to two carbonyl carbons.

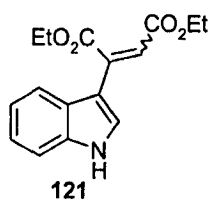
Based on the mode of formation and spectral data described above, the structure **119** is assigned to the compound.



119

Once the required keto-ester **119** was in our hand, the next step was to carry the Wittig reaction to prepare the diene required for the Diels-Alder reaction. So, ester was reacted with (ethoxycarbonyl methylene)triphenyl phosphorane **120** in refluxing toluene for 14 hours to give the corresponding Witttig product in 74% yield. IR spectrum showed strong bands at 3344 and 1703 cm^{-1} indicating the presence of –NH and carbonyl functionality. In PMR spectrum (Fig. 9), the two triplets at δ 1.08 and 1.34 integrating for three protons each were attributed to methyl groups while the two quartets at δ 4.08 and 4.33 were assigned to $-OCH_2-$ groups. The singlet at δ 6.95 integrating for one proton is attributed to vinylic proton. The aromatic protons appeared between δ 7.11 – 7.58 and a –NH proton of indole ring appeared at δ 8.45 as a broad singlet. The structure was further confirmed by ^{13}C NMR and DEPT experiment. In CMR spectrum (Fig. 10), the peaks at δ 13.8, 14.1, 60.7 and 61.8 were attributed to carbons of two $-CH_3$ and two $-OCH_2-$ groups respectively. The peaks between δ 109.3 – 137.5 were assigned to the aromatic and vinylic carbons whereas the two carbonyl carbons appeared at δ 166.1 and 167.4 respectively.

Thus on the basis of mode of formation and spectral data, the compound was assigned the following structure **121**.

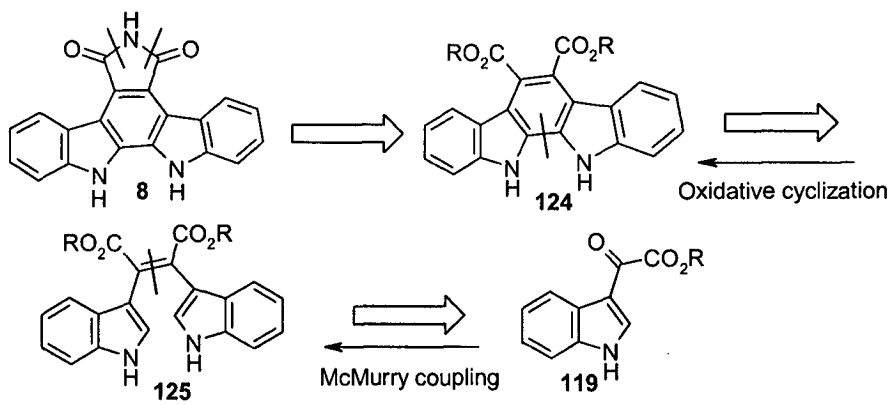


121

With compound **121** in hand, our next task was to prepare compound **123** *via* Diels-Alder reaction. So, the compound **121** and nitro-styrene **122** was refluxed in diphenyl ether for 8 hours, but the reaction did not yield the product. Next, we

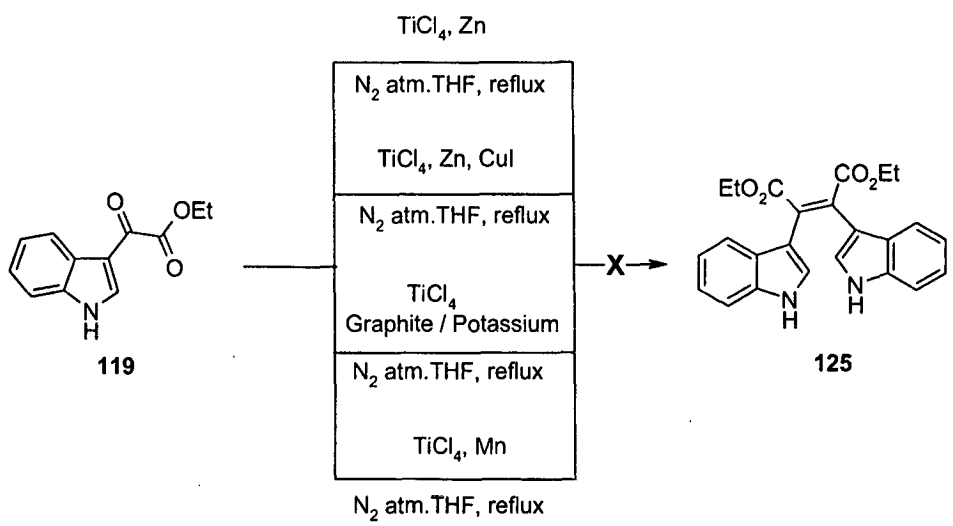
attempted the Diels-Alder reaction in sealed tube in presence of Lewis acid ($AlCl_3$) at 150-160^{0}C in toluene. But unfortunately, the Diels-Alder reaction did not take place under the reaction conditions employed.

Our failure to synthesize arcyriaflavin A by utilizing Diels-Alder reaction prompted us to try a new strategy. Our approach is based on the retro-synthetic pathway (Scheme 34) which involves McMurry coupling and oxidative cyclization as the main steps.



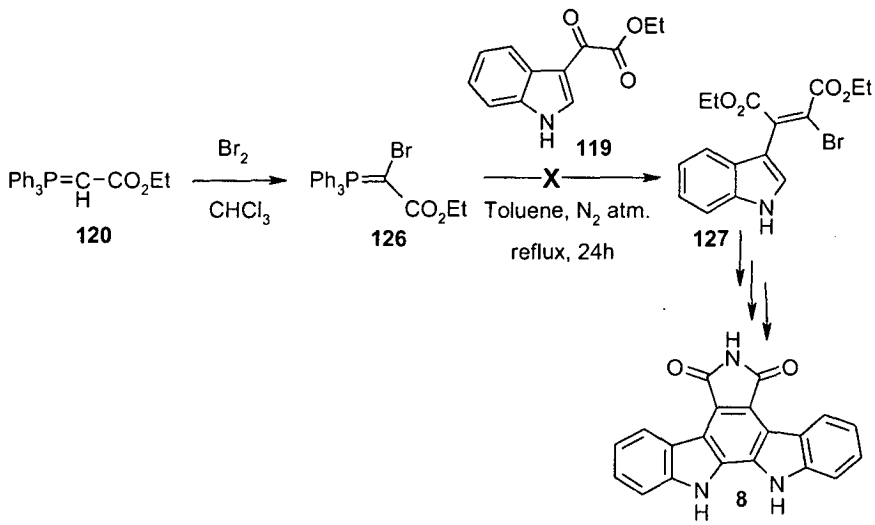
Scheme 30

Towards this end, we attempted a self McMurry coupling of compound **119** under different reaction condition as shown below (Scheme 31)



Scheme 31

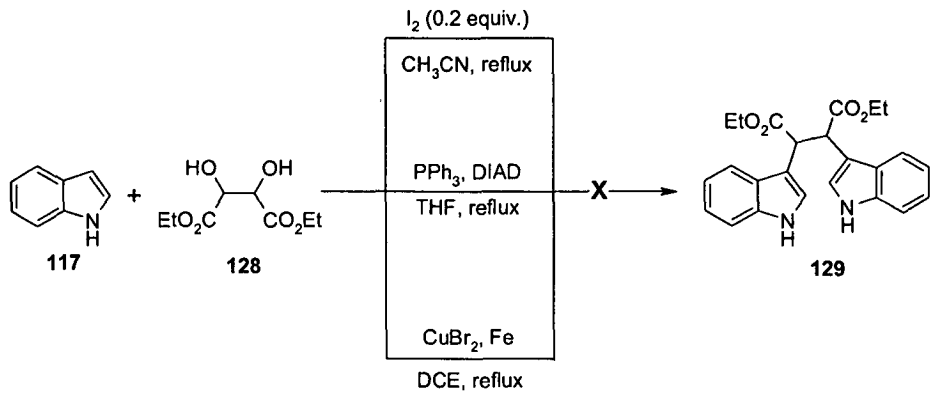
All our efforts to synthesize the compound **125** *via* McMurry coupling under varying reaction conditions failed to give the expected result. So, we attempted an alternate route as depicted below (Scheme 32).



Scheme 32

Carboethoxymethylidene(bromo)triphenyl phosphorane **126** was prepared by adding bromine to (ethoxycarbonyl methylene)triphenyl phosphorane **120** in chloroform at 0^0C using the reported procedure.[51] Next step is to carry the Wittig reaction of this bromo phosphorane **126** with compound **119** to furnish compound **127** which then could be converted to the required compound **8** using Pd-catalyzed coupling reaction. But when the Wittig reaction was attempted in refluxing toluene, the reaction did not take place and the starting material remains unchanged. So, we changed the solvent and the reaction was carried out in refluxing xylene, but again the reaction failed to give the product.

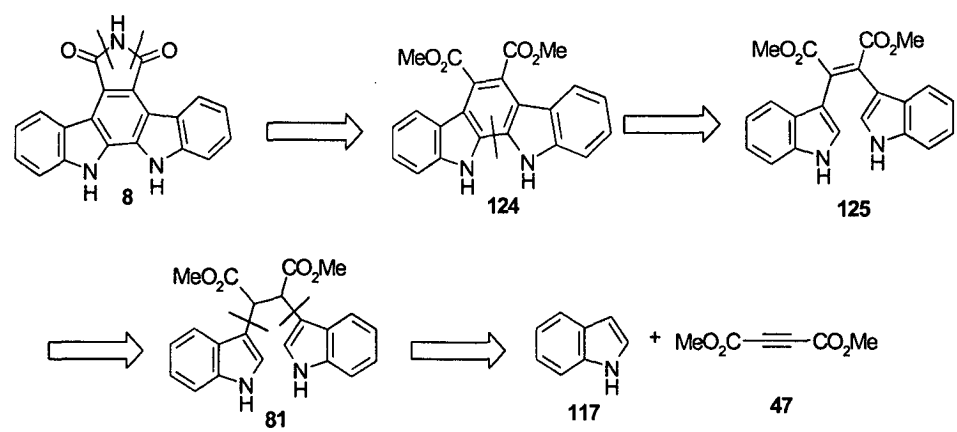
Simultaneously, we also attempted the synthesis of arcyriaflavin A using indole **117** and diethyl tartarate **128** as the starting materials (Scheme 33).



Scheme 33

Unfortunately, the double alkylation reaction of indole did not yield the product under all the reaction conditions tried.

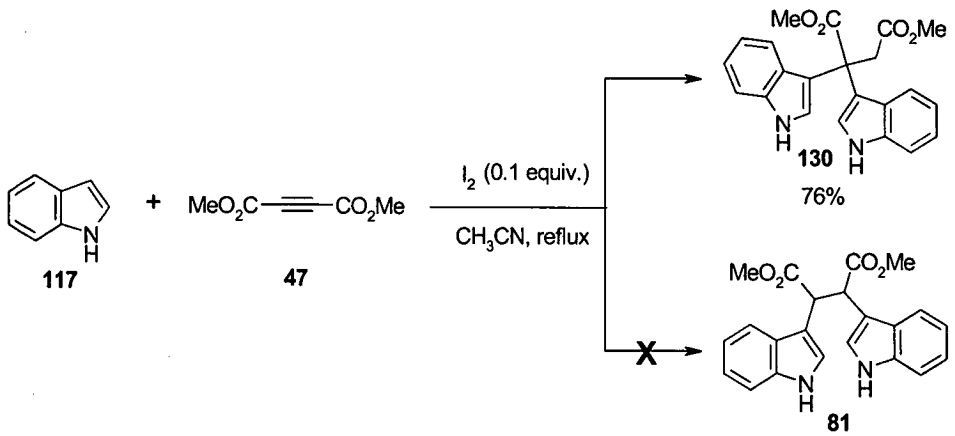
As all our efforts to synthesize arcyriaflavin A either by Diels-Alder reaction (Scheme 29), McMurry coupling (Scheme 31), Wittig reaction (Scheme 32) or alkylation (Scheme 33) were unsuccessful, we tried an alternate route which is based on retro-synthetic pathway as depicted below (Scheme 34). Retro-synthetic analysis suggested that it should be possible to prepare arcyriaflavin A, if we can prepare the intermediate **81** which inturn could be obtained by the reaction of indole **117** with dimethyl acetylene dicarboxylate (DMAD) **47** *via* double Michael reaction.



Scheme 34

When the Michael reaction was carried out with two equivalents of indole **117** and one equivalent of DMAD **47** in presence of 0.1 equivalent of iodine (Scheme 35), we

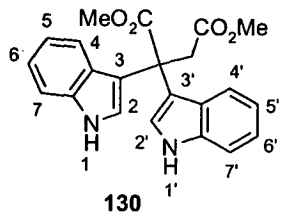
were expecting the compound **81** to form *via* double Michael reactions. But when the product was analyzed using ^{1}H NMR, ^{13}C NMR and DEPT experiments, it was found that Michael adduct 3,3'-bisindolyl succinate **130** was formed instead of expected compound **81** in 76% yield.



Scheme 35

The product obtained showed strong bands at 3400 and 1720 cm^{-1} in its IR spectrum, indicating the presence of NH and carbonyl functionalities. In its PMR spectrum (Fig. 11), the peaks at δ 3.51 and 3.72 integrating for three protons each were assigned to two $-OCH_3$ groups while peak at δ 3.76 integrating for two protons was attributed to methylene protons. The signals between δ 6.87 – 7.41 were assigned to aromatic protons. The broad singlet at δ 8.15 integrating for two protons was attributed to –NH of two indole rings. In ^{13}C NMR (Fig. 12), the peak at δ 42.5 was attributed to methylene carbon while the peak at δ 48.3 was assigned to quaternary aliphatic carbon. The peaks at δ 51.3 and 52.0 were attributed to two methoxy carbons. The peaks due to aromatic carbons were observed between δ 111.4 – 136.7. The two ester carbonyl carbons appeared at δ 171.4 and 173.5 respectively. The multiplicities of carbons were determined by DEPT experiment. Its HRMS spectrum showed $[M+Na]^+$ peak at *m/z* 399.1321 for $C_{22}H_{20}N_2O_4Na$ (Calcd for $C_{22}H_{20}N_2O_4Na$, 399.1321). The detailed spectral data is described below.

Spectroscopic data:



130

IR (KBr): υ_{max} = 3400, 2959, 1720, 1616, 1456, 1334, 1240, 1099, 810 cm^{-1}.

^{1}HNMR (300 MHz, $CDCl_3$): (Fig. 11)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
3.51	s	-	3	-OC**H**$_3$
3.72	s	-	3	-OC**H**$_3$
3.76	s	-	2	-C**H**$_2$-
6.87	t	7.2	2	H-5 & H-5'
7.08	t	7.2	2	H-6 & H-6'
7.25-7.35	m	-	4	H-4, H-4', H-7 & H-7'
7.41	d	2.4	2	H-2 & H-2'
8.15	br s	-	2	-NH

^{13}CNMR (75 MHz, $CDCl_3$): (Fig. 12)

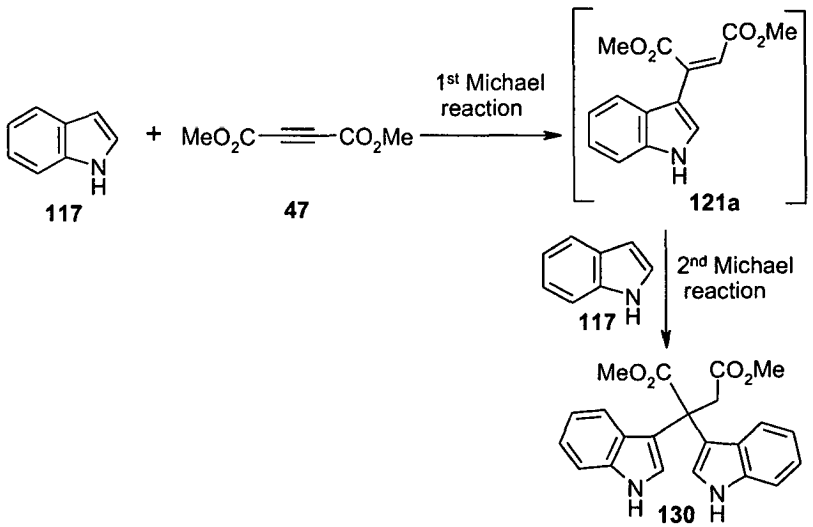
δ 42.5 (-**C**H$_2$-), 48.3 (-C-), 51.3 (-O**C**H$_3$), 52.0 (-O**C**H$_3$), 111.4 (C-3 & C-3'), 115.0 (C-3a, C-3a'), 118.5 (C-2 & C-2'), 120.5 (C-5 & C-5'), 120.9 (C-7 & C-7'), 124.1 (dC-6 & C-6'), 126.0 (C-4 & C-4'), 136.7 (C-7a & C-7a'), 171.4 (-**C**=O) and 173.5 (-**C**=O).

HRMS: *m/z* $[M+Na]^+$ 399.1321 (calcd for $C_{22}H_{20}N_2O_4Na$, 399.1321).

Melting Point: 156-158 ^{0}C; Lit.[52] 158-160^{0}C.

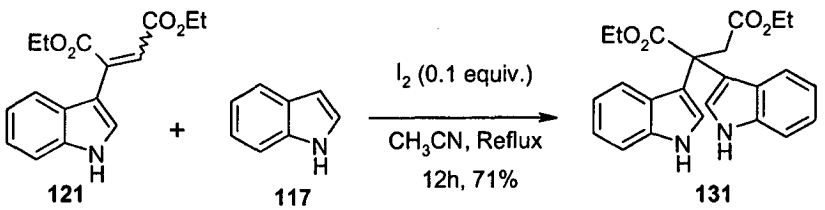
On the basis of above observations with respect to it's spectral analysis and similarity of melting point with the literature[52] melting point, the structure **130** was confirmed for the compound.

The formation of **130** could be accounted by the following sequence of reaction through the intermediate **121a** (Scheme 36).



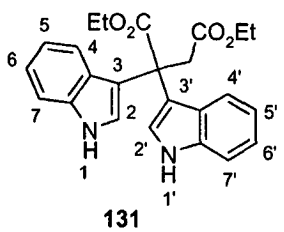
Scheme 36

As we had in our hand corresponding ethyl ester **121** we attempted 2nd Michael reaction on it. Thus compound **121** was treated with indole **117** in presence of catalytic amount of iodine and we got the expected corresponding ethyl ester of **130** i.e. compound **131** in 71% yield (Scheme 37).



Scheme 37

Spectroscopic data:



IR (KBr): υ_{max} = 3398, 2980, 1712, 1458, 1340, 1182, 1103, 1024 cm^{-1}.

^{1}HNMR (300 MHz, $CDCl_3$): (Fig. 13)

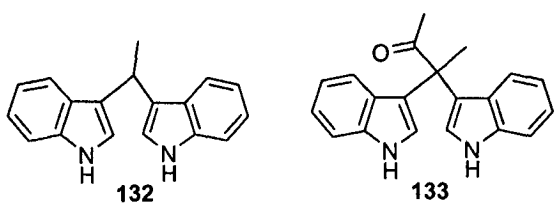
Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
1.06	t	7.2	3	-$OCH_2C\underline{H}_3$
1.19	t	7.2	3	-$OCH_2C\underline{H}_3$
3.75	s	-	2	-$C\underline{H}_2$-
3.95	q	7.2	2	-$OC\underline{H}_2CH_3$
4.20	q	7.2	2	-$OC\underline{H}_2CH_3$
6.87	t	7.2	2	H-5 & H-5'
7.08	t	7.2	2	H-6 & H-6'
7.31-7.34	m	-	4	H-4, H-4', H-7 & H-7'
7.41	d	2.4	2	H-2 & H-2'
8.14	br s	-	2	-NH

^{13}C NMR (75 MHz, $CDCl_3$): (Fig. 14)

δ 13.9 (-$OCH_2\underline{C}H_3$), 14.0 (-$OCH_2\underline{C}H_3$), 42.2 (-C-), 60.2 (-$O\underline{C}H_2CH_3$), 61.2 (-$O\underline{C}H_2CH_3$), 111.0 (C-2 & C-2'), 115.8 (C-3 & C-3'), 119.1 (C-5 & C-5'), 121.1 (C-7 & C-7'), 121.5 (C-6 & C-6'), 124.1 (C4 & C-4'), 126.1 (C-3a & C-3a'), 136.4 (C-7a & C-7a'), 171.1 (-$\underline{C}$=O) and 173.0 (-$\underline{C}$=O).

HRMS: *m/z* $[M+Na]^+$ 427.1636 (calcd for $C_{22}H_{24}N_2O_4Na$, 427.1634).

3,3'-Bis-indole compounds **130** and **131** are found to be similar to those of naturally occurring indole derivatives vibrindole **132** and compound **133** respectively (Fig. 15).



132 **133**

Fig. 15

Conclusion:

1) We have successfully developed a new one-pot approach for the synthesis of 2,2'-biindole using double reductive cyclization as the key step which constitutes the formal synthesis of naturally occurring indolocarbazole alkaloids – arcyriaflavin A and staurosporinone.

2) Secondly, we have achieved a simple and efficient one-pot method for the synthesis of 3,3'-bis(indolyl)succinate *via* double Michael reaction using iodine as a catalyst. This methodology can be extended for the synthesis of derivatives of 3,3'-bis(indolyl)alkanes for their biological evaluation.

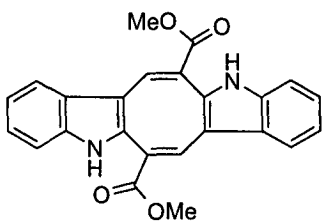
3) Unfortunately, the total synthesis of arcyriaflavin A and staurosporinone using different strategies were unsuccessful.

Section B: Synthetic Studies Towards Bis-indole Marine Natural Product – Caulersin

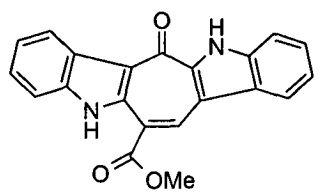
Introduction:

Naturally occurring bis-indole products have emerged as an important structural class due to their high degree of biological activity.[53] Compounds possessing a six-membered carbocyclic ring between the two "parallel" indoles, like staurosporinone **7** or arcyriaflavin A **8** have been discussed earlier in section A of this chapter.

Bis-indole alkaloids with a seven or eight-membered central ring and two "antiparallel" indole cores have been isolated from the algae *Caulerpa* (Fig. 16). Caulerpine[54] **134** with eight-membered central ring was isolated from the green alga *Caulerpa racemose* and was shown to be a plant growth regulator and also exhibits slight in vitro antitumor activity.[55] Caulersin[56] **135** with seven-membered central ring was isolated by Su in 1997 from the alga *Caulerpa serrulata*, but its biological activity was not reported inspite of a total synthesis of caulersin published in 1999.



134



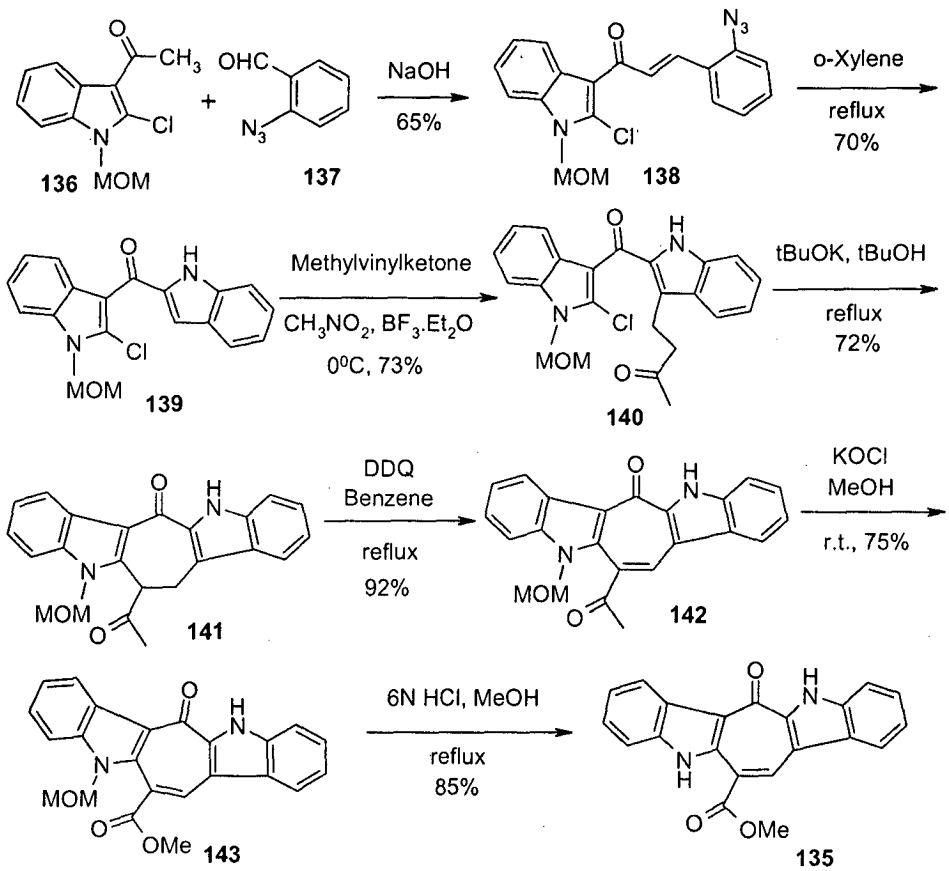
135

Fig. 16

Reported Synthesis:

So far only three methods have been reported for the synthesis of bis-indole marine natural product caulersin.

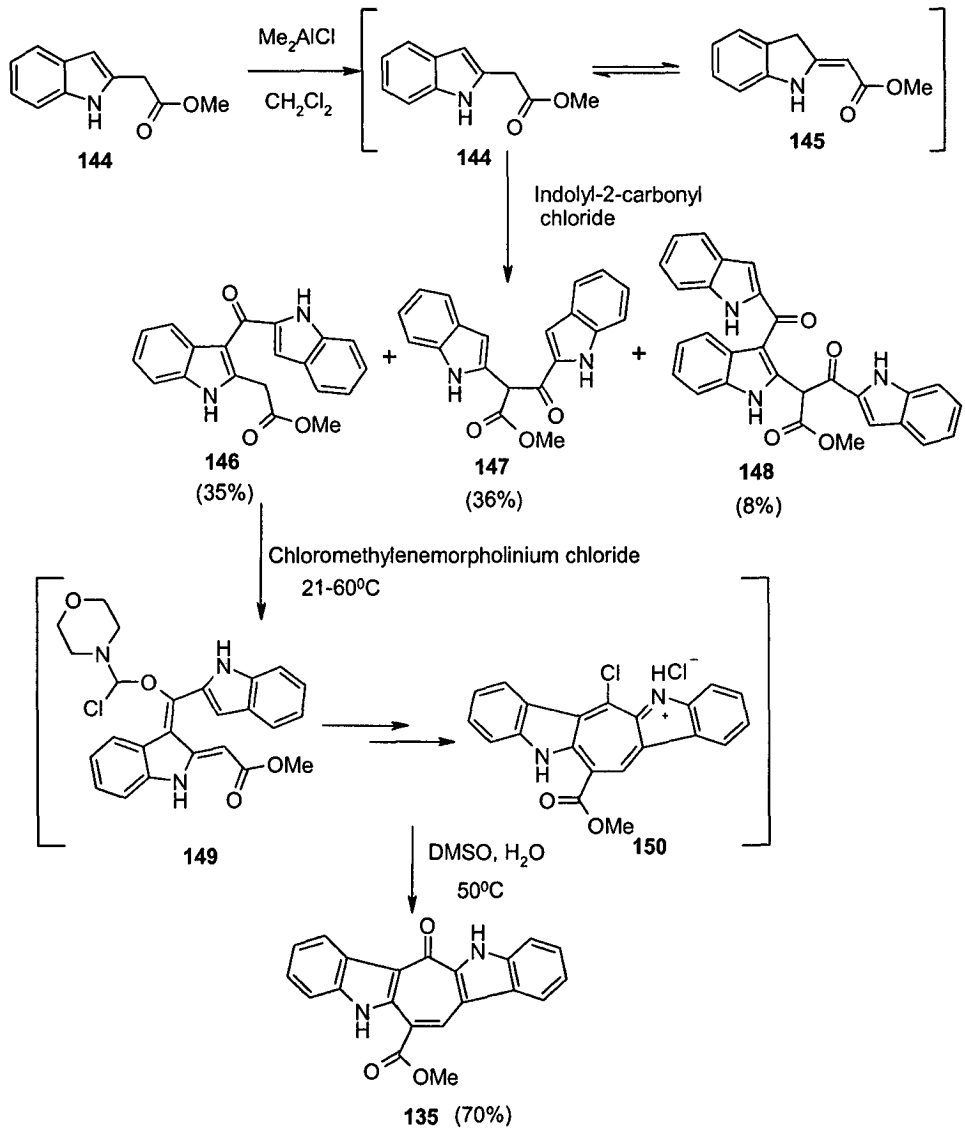
Fresneda and co-workers[57] reported the first synthesis of caulersin in 1999 by seven-step sequence as described below (Scheme 38).



Scheme 38

Aldol condensation of the indole derivative **136** with *o*-azidobenzaldehyde **137** provided the chalcone **138** in 65% yield which was converted into key intermediate bis(indolyl) ketone **139** by thermal treatment in *o*-xylene at reflux temperature. Michael-type addition of bis(indolyl)ketone **139** to methyl vinyl ketone was performed using boron trifluoride in nitromethane to furnish 3-oxoalkylated product **140** in 73% yield. Central seven-membered carbocyclic ring was formed by intramolecular nucleophilic displacement of the chlorine atom using t-BuOK/t-BuOH in refluxing benzene to afford compound **141** in 72% yield. Dehydrogenation of **141** with DDQ provided **142** in 92% yield which was converted to methyl ester **143** using aqueous KOCl in methanol at 0^0C. The synthesis of caulersin **135** was completed by *N*-deprotection of **143** using 6N HCl in MeOH at reflux temperature.

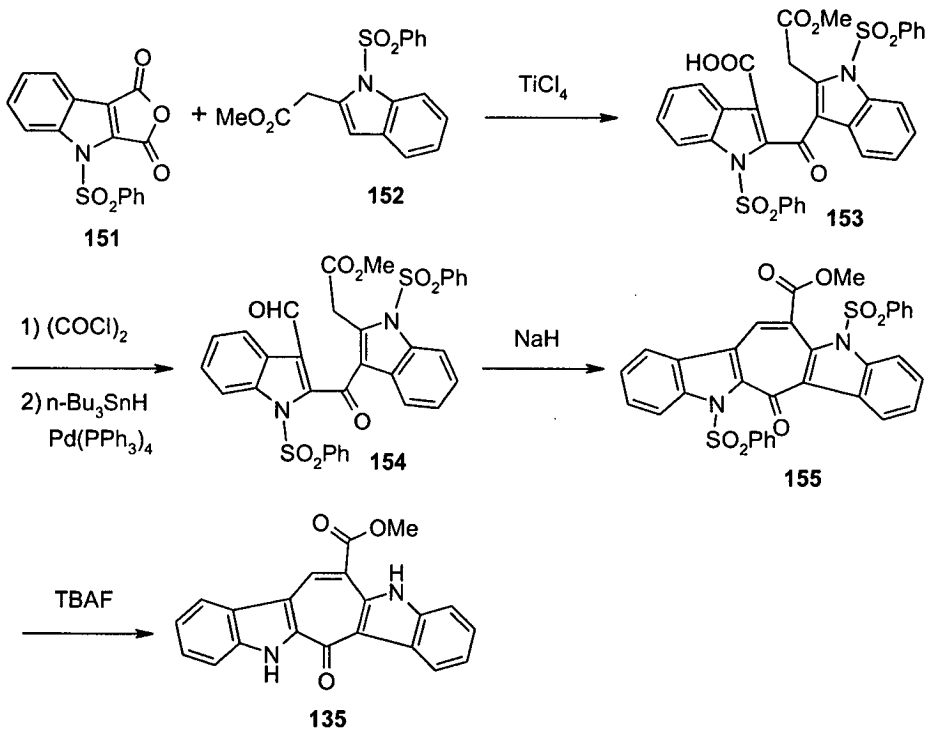
Bergman and co-workers[58] achieved a three-step synthesis by cyclization of the simple keto ester with the Vilsmeier reagent (Scheme 39).



Scheme 39

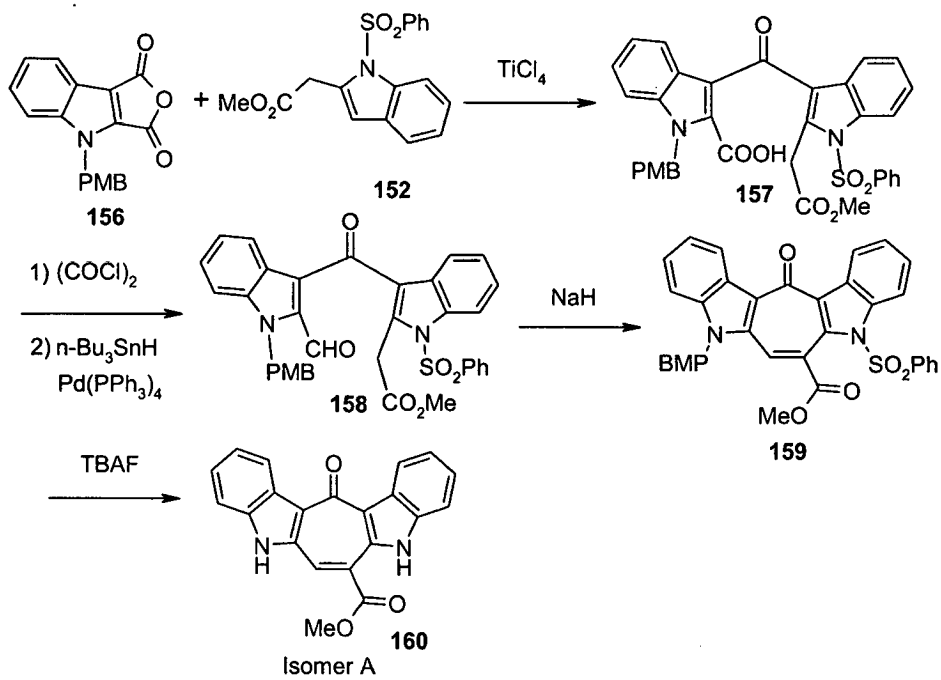
Treatment of indole-2-acetic acid methyl ester **144** with dimethylaluminium chloride, followed by addition of indole-2-carbonyl chloride provided the required 3-acylated bisindole **146** in 35% yield together with two other products arising from the tautomer **145**. Cyclization of the keto ester **146** with the Vilsmeier reagent chloromethylenemorpholinium chloride yielded the target compound **135**. The total yield of caulersin in this facile three-step procedure was 25%.

Miki and co-workers[59] accomplished the synthesis of caulersin (Scheme 44) and its isomer (Schemes 40 - 43) using indole-2,3-dicarboxylic anhydrides and methyl indoleacetate as the starting materials.

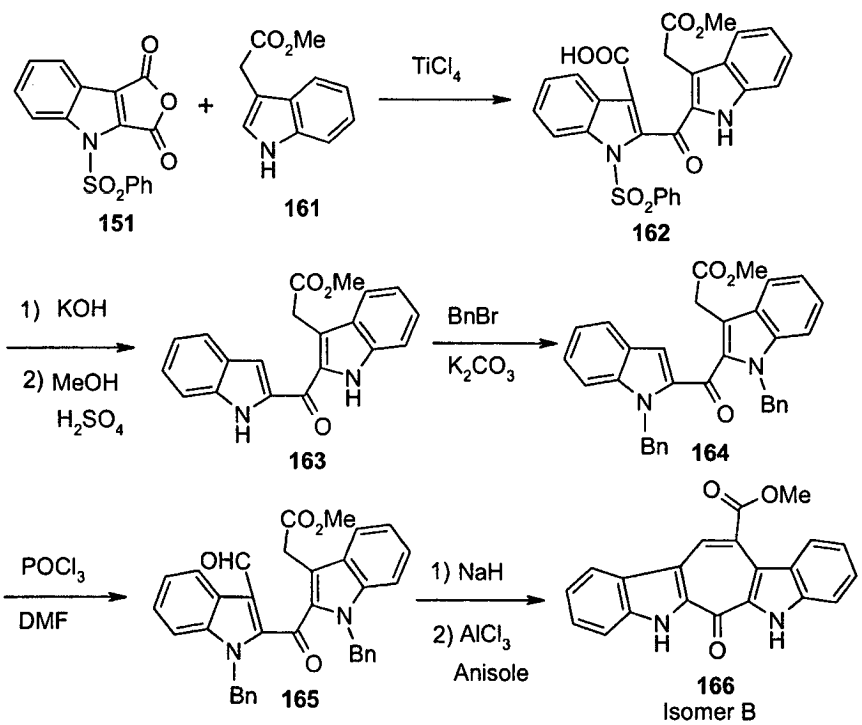


Scheme 40

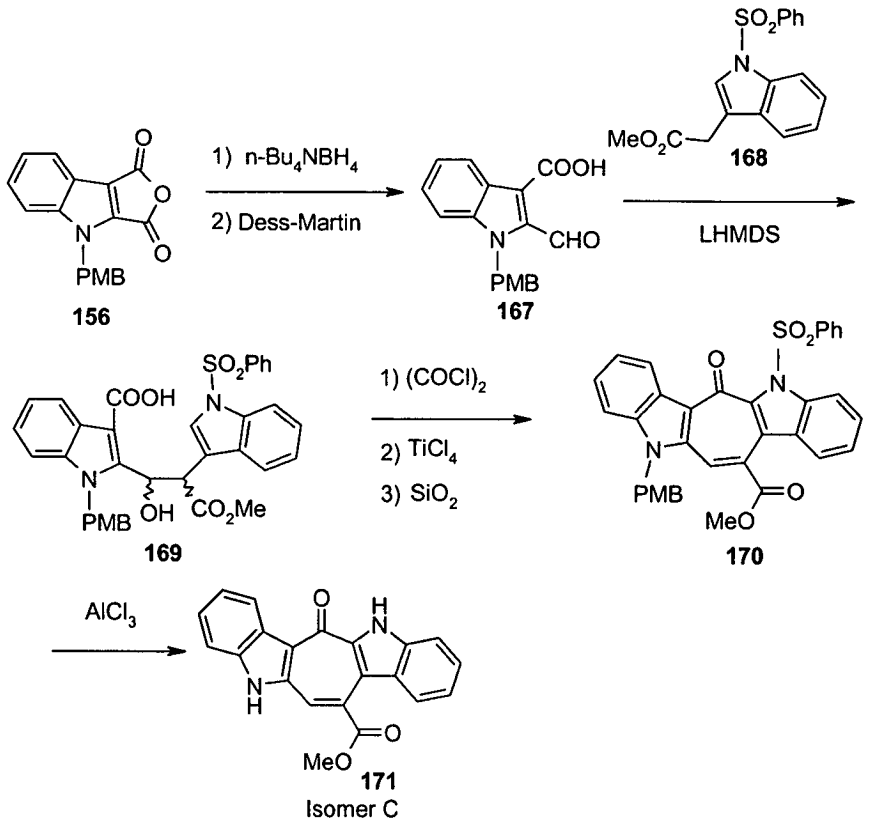
Reaction of 1-benzenesulfonylindole-2,3-dicaroxylic anhydride **151** with methyl 1-benzenesulfonylindole-2-acetate **152** in dichloromethane in presence of titanium (IV) chloride afforded 2-acylindole-3-carboxylic acid **153** in quantitative yield. Treatment of **153** with oxalyl chloride followed by tetrabutyltin hydride in presence of $Pd(PPh_3)_4$ at room temperature in toluene gave aldehyde **154** in 77% yield. Cyclization of **154** to obtain *N,N'*-dibenzenesulfonyl caulersin **155** was achieved using NaH in THF at room temperature. Debenzenesulfonylation of **155** with tetrabutylammonium fluoride in THF gave caulersin **135** in 91% yield. Under similar reaction condition, the three isomers A, B and C were synthesized by reacting indole-2,3-dicarboxylic anhydrides with methyl indoleacetates as described below (Scheme 41 - 43).



Scheme 41



Scheme 42



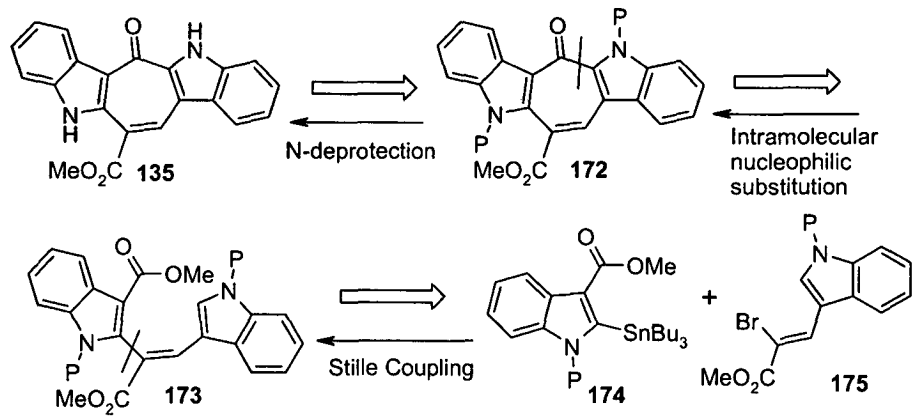
Scheme 43

Objective:

Caulersin represents the only natural product isolated so far containing a bisindole structure bridged by a central troponoid framework. Objective of the present study is to develop a short, simple and high yielding method for the synthesis of caulersin employing Stille coupling as the key step.

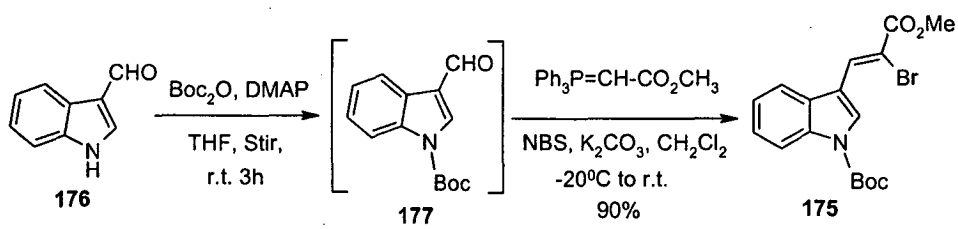
Results and Discussion:

Our approach towards the synthesis of caulersin is based on the retro-synthetic pathway as depicted below (Scheme 44). The main task is the construction of seven-membered carbocyclic ring between two "anti-parallel" indole moieties. By dissecting the molecule as shown in scheme 44, we thought that it should be possible for us to construct seven-membered ring between two indole cores *via* Stille coupling and intramolecular nucleophilic substitution.



Scheme 44

Accordingly, we started the synthesis and the compound **175** and **174** required for the Stille coupling thus were prepared from commercially available starting materials (Schemes 45 & 46).

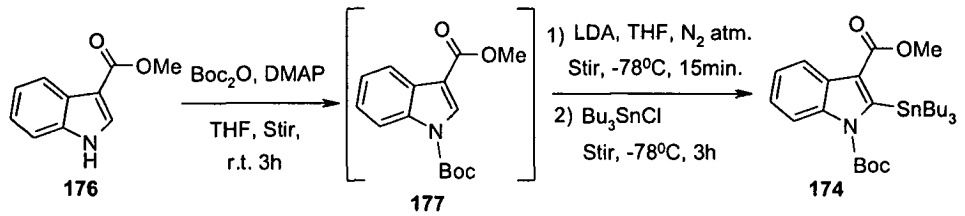


Scheme 45

N-protection of indole-3-carboxyaldehyde **176** was achieved using Boc_2O at room temperature. Wittig reaction of *N*-Boc-indole-3-carboxyaldehyde **177** with in situ prepared bromo phosphoylide[60] afforded the required compound **175** in 90% yield. In its IR spectrum, the strong bands at 1735 and 1670 cm^{-1} could be attributed to carbonyl functionality of ester and carbamate respectively. In ^1H NMR spectrum (Fig. 17), a singlet at δ 1.73 is assigned to three methyls of –Boc group and a peak at δ 3.95 integrating for three protons is attributed to methoxy group. The aromatic protons appeared between δ 7.38 – 8.52 while a vinylic proton appeared as a singlet at δ 8.83. In ^{13}C NMR spectrum (Fig. 18), a peak at δ 28.1 is attributed to methyls of –Boc group whereas the peaks at δ 53.5 and 84.9 is assigned to methoxy carbon and quaternary carbon of –Boc group respectively. All aromatic and vinylic carbons appeared between δ 111.7 – 134.9 and the two carbonyl carbons at δ 149.3 and 163.8 respectively.

Melting point: 114–116^{0}C (Lit.[61] mp = 116^{0}C).

Based on the mode of formation, spectral data and closeness of melting point with the literature[61] melting point, the structure **175** is assigned to the compound.

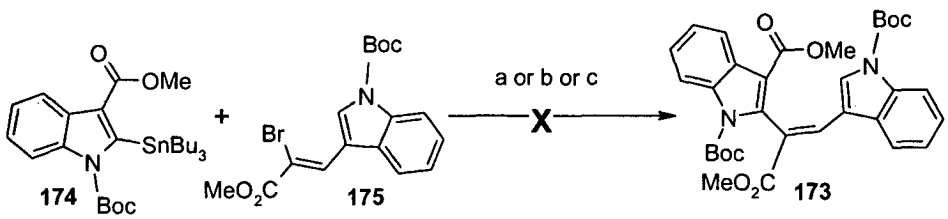


Scheme 46

N-protection of **176** was done using Boc_2O which was lithiated with LDA in THF at -78^{0}C and to this was added tributyl tin chloride and stirred at this temperature for 3 hours to give compound **174** as a colourless thick liquid in 87% yield. The IR spectrum showed a strong bands at 486, 1414 and 1732 due to C-Sn and carbonyl functionality. In its PMR spectrum (Fig. 19), the peaks between δ 0.92 – 1.53 were attributed to the protons of tributyl group whereas the singlet at δ 1.76 integrating for nine protons was assigned to methyl of –Boc group. A singlet at δ 3.97 integrating for three protons was assigned to methoxy group while the peaks between δ 7.30 – 8.12 were assigned to aromatic protons. In CMR spectrum (Fig. 20), the peaks at δ 13.3, 13.7 and 27.4 were attributed to methylene carbons of tributyl group while the peaks at δ 28.2 and 29.1 were assigned to methyl carbons of –Boc and tributyl groups respectively. The peak at δ 51.5 was attributed to the methoxy carbon whereas the quaternary carbon of –Boc group appeared at δ 84.4. The aromatic carbons appeared between δ 114.8 – 151.4 and the carbonyl carbons at δ 154.1 and 166.2 respectively.

Based on the mode of formation and spectral data, the compound could have structure **174**.

With compounds **174** and **175** in hand, the stage was set for the key coupling reaction. To this end, a variety of reaction conditions were attempted (Scheme 51). Unfortunately, all failed to deliver any results.



Reaction condition: a) $Pd_2(dba)_3$, PBu_3, Et_3N, Dioxane, N_2 atm., reflux, 24h

b) $Pd(PPh_3)_4$, Toluene, N_2 atm., reflux, 12h

c) $Pd(PPh_3)_4$, CuI, THF, N_2 atm., reflux, 20h

Scheme 47

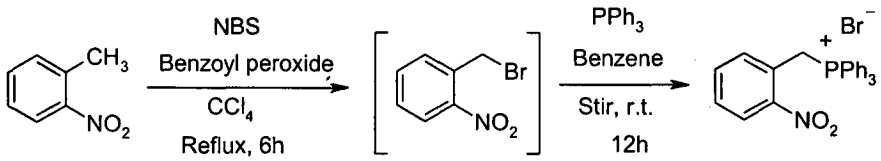
Conclusion:

We were able to synthesize the precursors required for caulersin, however we could not succeeded in synthesizing caulersin as the coupling reaction did not take place under the reaction conditions employed.

Experimental Section:

Section A: Synthetic Studies Towards Indolocarbazole Alkaloids – Arcyriaflavin A and Staurosporinone

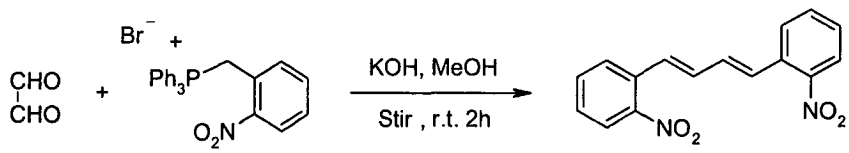
3.01 Preparation of *o*-nitro-benzyl-triphenyl phosphonium bromide:



Procedure:

A solution of *o*-nitrotoluene (3.05g, 22.30 mmol) in CCl_4 (20 mL) containing NBS (3.96, 22.30 mmol) and benzoyl peroxide (0.07g) is refluxed for 6 hours, cooled, filtered and solvent removed by distillation to give crude brown thick liquid. To this, a solution of triphenyl phosphine (5.84g, 22.30 mmol) in benzene (20 mL) was added and stirred at room temperature for 12 hours. The solid obtained was filtered, washed with benzene and dried to give *o*-nitro-benzyl-triphenyl phosphonium bromide (7.46g) in 70% yield. **Melting Point:** 158-160^{0}C (Lit.[62] mp 161-162^{0}C).

3.02 Synthesis of nitro-dimer using double Wittig reaction with glyoxal:

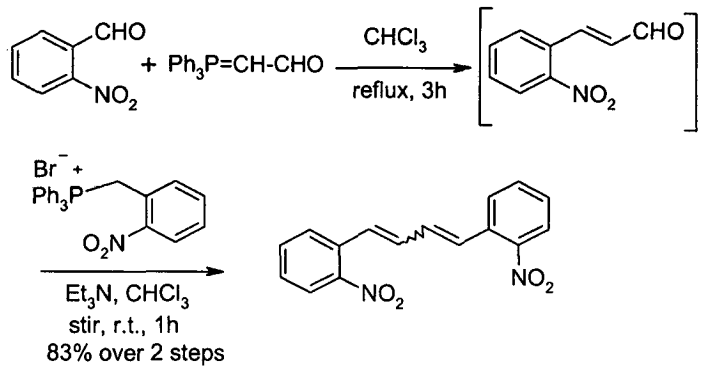


Procedure:

To a mixture of *o*-nitro-benzyl-triphenyl phosphonium bromide (3.05g, 22.30 mmol) and 40% aqueous gyloxal (0.46g, 7.94 mmol) in methanol is added KOH (0.35g, 6.35 mmol) and stirred at room temp. for 2 hours. The yellow solid is filtered through Buchner funnel to obtain nitro-dimer (0.35g) in 37.6% yield.

HRMS: $[M+Na]^+$ peak at *m/z* 319.0692 for $C_{16}H_{12}N_2O_4Na$ (calcd for $C_{16}H_{12}N_2O_4Na$, 319.0695).

3.03 Synthesis of nitro-dimer:



Procedure:

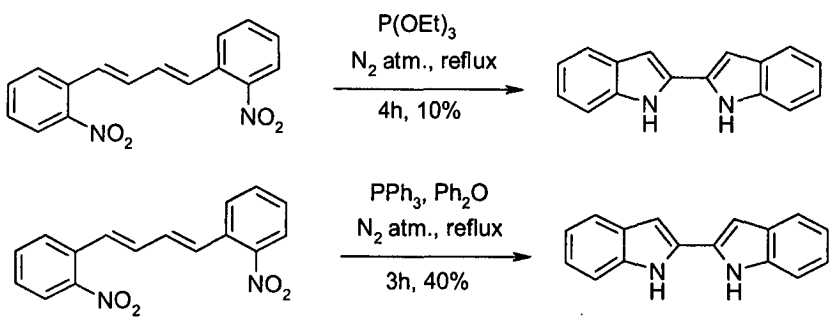
A mixture of *o*-nitro-benzaldehyde (0.52g, 3.46 mmol) and (triphenylphosphoranylidene)-acetaldehyde (1.05g, 3.46 mmol) was refluxed in chloroform (10 mL) for 3 hours. The reaction mixture was allowed to cool to room temperature and to this was added *o*-nitro-benzyl-triphenyl phosphonium bromide (1.65g, 3.46 mmol), Et_3N (1 mL) and stirred at room temperature for 1 hour. The chloroform was removed under vaccum and to this was added methanol. The yellow solid i.e. product which comes out was filtered through Buchner funnel (0.85g, 83% overall yield).

Melting Point: 102 – 104 ^{0}C.

IR (KBr): υ_{max} 1518, 1341, 1142, 952, 858, 748 cm^{-1}.

HRMS: $[M+Na]^+$ peak at *m/z* 319.0692 for $C_{16}H_{12}N_2O_4Na$ (calcd for $C_{16}H_{12}N_2O_4Na$, 319.0695).

3.04 Synthesis of 2,2'-Biindole



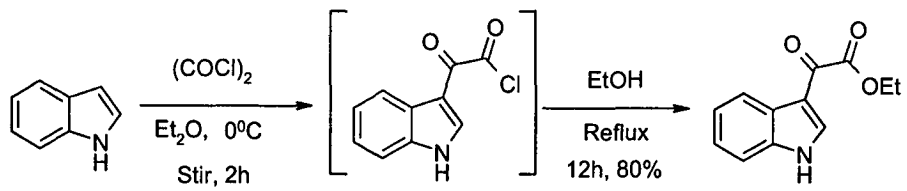
Procedure:

a) Nitro-dimer (0.78g, 2.65 mmol) is refluxed in triethyl phosphite (5 mL) under nitrogen atmosphere for 4 hours. Excess triethyl phosphite was distilled out under reduced pressure and the crude product was chromatographed on silica gel column (60 – 120 mesh). Elution with 20% ethyl acetate in pet ether afforded 2,2'-biindole (0.06g) as a gray solid in 10% yield.

b) A mixture of nitro-dimer (1.50g, 5.10 mmol) and triphenyl phosphine (2.67g, 10.2 mmol) is refluxed in Ph_2O (15 mL) for 3 hours under N_2 atmosphere. After cooling, reaction mixture was chromatographed on silica gel column (60 -120 mesh) and diphenyl ether was removed using pet ether as an eluent. Further elution with 20% ethyl acetate in pet ether afforded 2,2'-biindole (0.47g) as a gray solid in 40% yield.

Melting Point: >300^{0}C (Lit.[50] mp 311-314^{0}C).

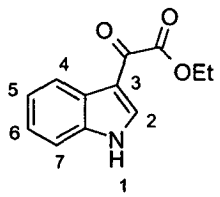
3.05 Synthesis of Keto-ester



Procedure:

Oxalyl chloride (1.14g, 9.01mmol) in diethylether was added dropwise to a solution of indole (1.05g, 9.01 mmol) in diethylether at 0^{0}C for a period of 15 minutes. The reaction was stirred at room temperature for 1 hour. The solid which comes out was filtered and washed with diethyl ether to give a yellow compound. The yellow solid compound was dissolved in excess of ethanol (30 mL) and it was refluxed for 12 hours. The reaction mixture was cooled to room temp. and concentrated in vacuo. The residue was purified by column chromatography on silica gel (60-120 mesh) using hexanes-ethyl acetate (1:1) as eluent to afford indole keto-ester (1.56g, 7.21 mmol, 80%) as a buff colour solid.

Melting Point: 182 – 186^{0}C (Lit.[63] mp 186 - 187^{0}C).



IR (KBr): υ_{max} 3229, 3167, 1728, 1614, 1510, 1431, 1263, 1130, 1020, 762, 659 cm^{-1}

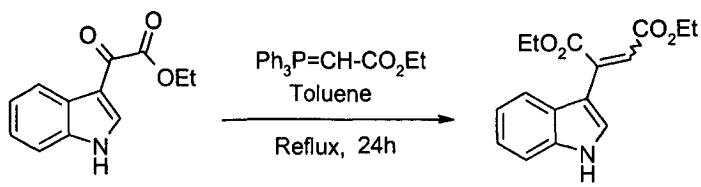
^{1}H NMR (300 MHz, DMSO-d_6): (Fig. 7)

Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
1.34	t	7.2	3	-$OCH_2C\underline{H}_3$
4.36	q	7.2	2	-$OC\underline{H}_2CH3$
7.25-7.33	m	-	2	H-4 & H-7
7.56	t	-	1	H-5
8.17	m	-	1	H-6
8.42	d	3.3	1	H-2
12.39	br s	-	1	-NH

^{13}CNMR (75 MHz, DMSO-d_6): (Fig. 8)

δ 14.4 (-$OCH_2\underline{C}H_3$), 62.1 (-$O\underline{C}H_2CH_3$), 112.8 (C-2), 113.2 (C-5), 121.6 (C-7), 123.3 (C-6), 124.3 (C-4), 125.9 (C-3a), 137.1 (C-7a), 138.6 (C-3), 164.0 (-**C**=O) and 179.6 (-**C**=O).

3.06 Synthesis of Diester:

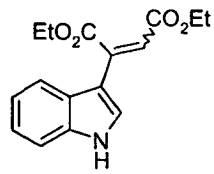


Procedure:

A mixture of keto-ester (1.12g, 5.18 mmol), and phosphorane (1.80g, 5.18 mmol) was refluxed in toluene for 24 hours. The reaction mixture was cooled to room temp. and concentrated in vacuo. The residue was chromatographed on silica gel (60-120 mesh) and the product was separated from the triphenyl phosphine oxide using hexanes-

ethyl acetate (4:1) as an eluent to afford **14** (1.10g, 3.84 mmol, 74%) as a yellow solid.

Melting Point: 74 – 76^{0}C.



IR (KBr): υ_{max} 3344, 2984, 1703, 1605, 1535, 1388, 1234, 1034, 742 cm^{-1}.

^{1}HNMR (300 MHz, $CDCl_3$): (Fig. 9)

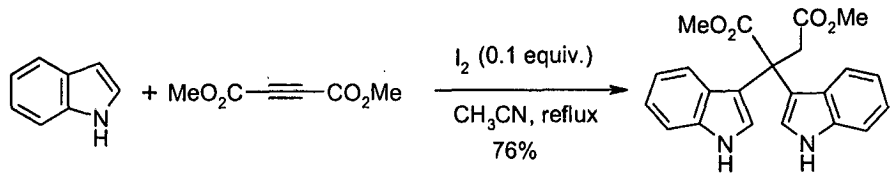
Chemical Shift (δ ppm)	Multiplicity	Coupling Constant (*J* Hz)	No. of Protons (H)	Position
1.08	t	7.2	3	-OCH$_2$C**H**$_3$
1.34	t	7.2	3	-OCH$_2$C**H**$_3$
4.08	q	7.2	2	-OC**H**$_2$CH$_3$
4.33	q	7.2	2	-OC**H**$_2$CH$_3$
6.95	s	-	1	=C**H**-
7.11 – 7.23	m	-	3	Ar-**H**
7.37	d	8.1	1	H-2
7.58	d	2.7		Ar-**H**
8.45	br s	-	1	-NH

^{13}CNMR (75 MHz, $CDCl_3$): (Fig. 10)

δ 13.8 (-OCH$_2$**C**H$_3$), 14.1 (-OCH$_2$**C**H$_3$), 60.7 (-O**C**H$_2$CH$_3$), 61.8 (-O**C**H$_2$CH$_3$), 109.3 (C-3), 111.4 (C-5), 119.7 (C-7), 122.3 (C-6), 125.4 (C-4), 126.5 (C-3a), 127.2 (=**C**H-), 135.5 (=C-), 137.5 (C-7a), 166.1 (-**C**=O) and 167.4 (-**C**=O).

HRMS: *m/z* $[M+Na]^+$ 310.1055 (calcd for $C_{16}H_{17}NO_4$, 310.1055).

3.07 Synthesis of dimethyl-2,2-bis(indol-3'-yl)succinate

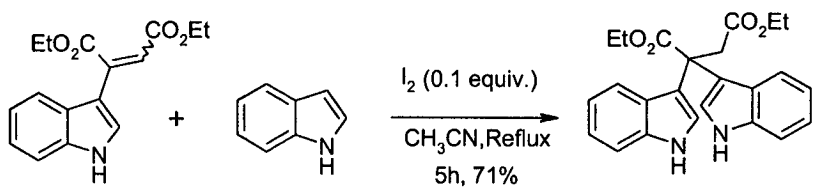


Procedure:

A mixture of indole (0.51g, 4.38 mmol), DMAD (0.31g, 2.19 mmol) and iodine (0.11g, 0.44 mmol) was refluxed in acetonitrile (15mL) for 8 hours. The reaction mixture was cooled to room temp. and concentrated in vacuo. The residue was purified by column chromatography on silica gel (60-120 mesh) using hexanes-ethyl acetate (7:3) as an eluent to afford the product (1.25g, 3.33 mmol, 76%) as a white solid.

Melting Point: 156-158 ^{0}C (Lit.[52] mp 158-160^{0}C).

3.08 Synthesis of bis(indolyl)succinate:

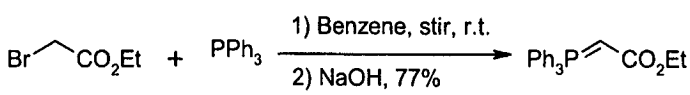


Procedure:

A mixture of (0.82g, 2.87 mmol), indole (0.33g, 2.87 mmol) and iodine (0.07g, 0.29 mmol) was refluxed in acetonitrile (20mL) for 5 hours. The reaction mixture was cooled to room temp. and concentrated in vacuo. The residue was purified by column chromatography on silica gel (60-120 mesh) using hexanes-ethyl acetate (7:3) as an eluent to afford the product (0.82g, 2.04 mmol, 71%) as a white solid.

Melting Point: 136-140^{0}C.

3.09 Preparation of (Ethoxycarbonyl methylene)triphenyl phosphorane:



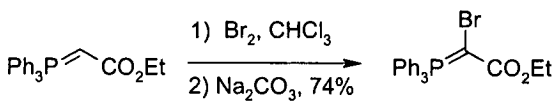
Procedure:

Addition of solution of triphenyl phosphine (10.0g, 38.1 mmol) in dry benzene (30 mL) to a solution of ethyl bromoacetate (6.36g, 38.1 mmol) in benzene (10 mL) at room temp. resulted in an elevation in temp. and precipitation of salt. After allowing the mixture to cool to room temp., it was vigorously shaken and left overnight. The solid obtained was filtered and washed with benzene and dried. Water (150 mL) was added to salt followed by addition of benzene (100 mL) and then neutralized by

aqueous NaOH with constant shaking to a phenolphtheleine end point. The benzene layer was evaporated, dried over anhyd. Na_2SO_4 and concentrated to about 1/3rd volume. Addition of *n*-hexanes resulted in separation of white crystalline product which was filtered and dried to afford phosphorane (10.22g, 77%).

Melting Point: 124-126^{0}C, (Lit.[64] mp 125-127^{0}C).

3.10 Preparation of carboethoxymethylidene(bromo)triphenyl phosphorane:



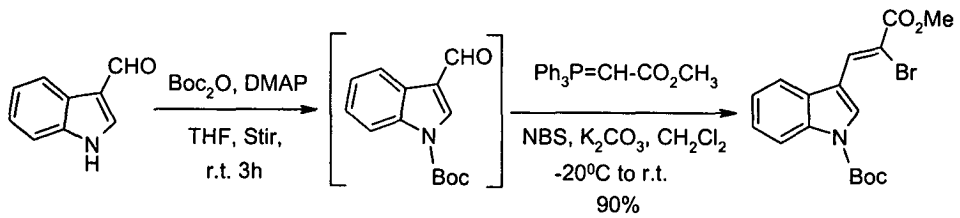
Procedure:

Bromine (1.10g, 6.9 mmol) in $CHCl_3$ (5 mL) was added to (carboethoxymethylene)triphenyl phosphorane (2.0g, 5.7 mmol) in $CHCl_3$ (20 mL) at 0^{0}C, whereupon immediate decolorization occurred. After allowing it to attain room temperature, the solution was concentrated. The residual oil was dissolved in CH_2Cl_2 (30 mL) and solution was extracted three times with an equivalent of Na_2CO_3 in water (10 mL). The organic layer was dried over anhyd. Na_2SO_4 and then concentrated. Addition of *n*-hexane gave bromo phosphorane (1.81g, 74%).

Melting Point: 152-154^{0}C, (Lit.[65] mp 155-156^{0}C).

Section B: Synthetic Studies Towards Bis-indole Marine Natural Product – Caulersin

3.11 Preparation of 3-(2-Bromo-2-methoxycarbonyl ethenyl)-indole-1-carboxylic acid *tert*-butyl ester



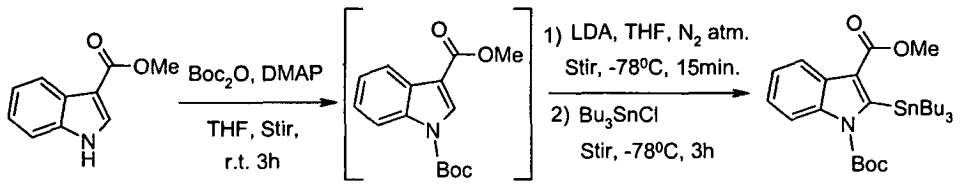
Procedure:

Di-tert-butyl dicarbonate (2.81g, 12.9 mmol) and catalytic amount of DMAP (0.13g, 1.1 mmol) were added to a solution of indole-3-carboxyaldehyde (1.55g, 10.7 mmol) in THF (15 mL) and the mixture were stirred at room temperature for 1 hour. The solvent was removed under vaccum to give *N*-Boc-protected indole-3-carboxyaldehyde which without purification was used for further reaction.

At -20^{0}C, a solution of methyl(triphenyl phosphoranylidene)acetate (7.17g, 21.5 mmol) in dry CH_2Cl_2 (60 mL) was treated with NBS (4.20g, 23.6 mmol). After stirring at -20^{0}C for 30 minutes, *N*-Boc-protected indole-3-carboxyaldehyde (2.63g, 10.7 mmol) and K_2CO_3 (4.94g, 35.7 mmol) were successively added. The reaction mixture was then stirred for 30 hours while allowing the temperature to reach the ambient condition. After filtration through a pad of Celite, the filtrate was concentrated in vacuo. The crude mixture was purified by column chromatography using 5% ethyl acetate in pet ether as the eluent to give 3-(2-Bromo-2-methoxycarbonyl ethenyl)-indole-1-carboxylic acid *tert*-butyl ester (3.67g) as a yellow solid in 90% yield.

Melting Point: 114 – 116^{0}C (Lit.[61] mp 116^{0}C).

3.12 Preparation of organo-tin-compound



Procedure:

Di-tert-butyl dicarbonate (1.51g, 6.9 mmol) and catalytic amount of DMAP (0.0705g, 1.1 mmol) were added to a solution of methyl-indole-3-carboxylate (1.01g, 5.8 mmol) in THF (15 mL) and the mixture were stirred at room temperature for 1 hour. The solvent was removed under vaccum to give *N*-Boc-protected methyl-indole-3-carboxylate which without purification was used for further reaction. To this was added dry THF and cooled to -78^{0}C. LDA was added to the above solution under N_2 atmosphere at -78^{0}C and stirred at this temperature for 15 minutes. Bu_3SnCl (1.87g, 5.8 mmol) was added at -78^{0}C and stirred for 3 hours while allowing the temperature to reach the ambient condition. Water (50 mL) was added and extracted with $CHCl_3$ (3 × 20 mL), dried over anhydrous Na_2SO_4, concentrated in vacuo. The crude product was purified by column chromatography using 2% ethyl acetate in pet ether to afford the product (2.76g) as a colourless thick liquid in 85% yield.

Spectra:

Fig. 3: ^{1}H NMR spectrum of **112**

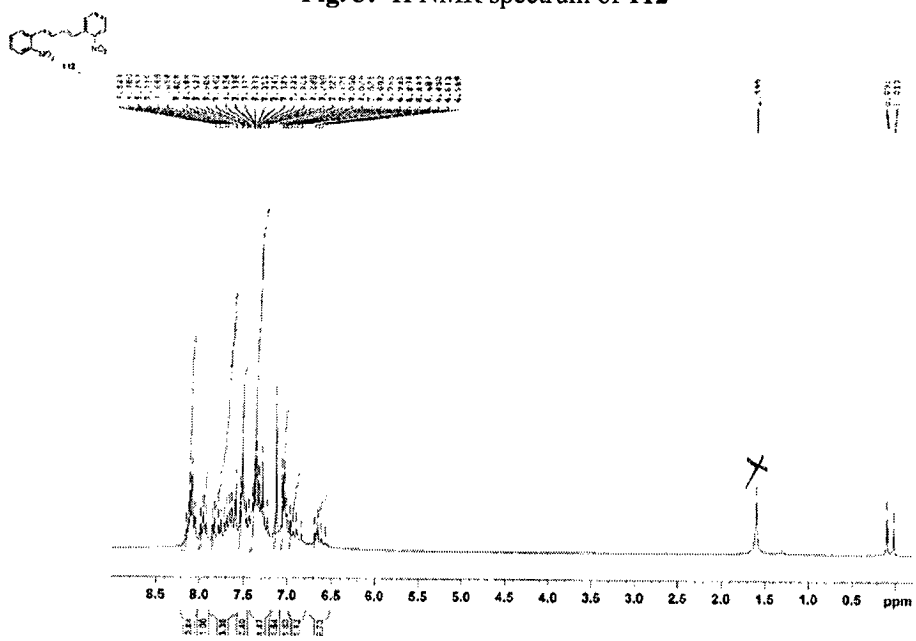


Fig. 4: HRMS spectrum of **112**

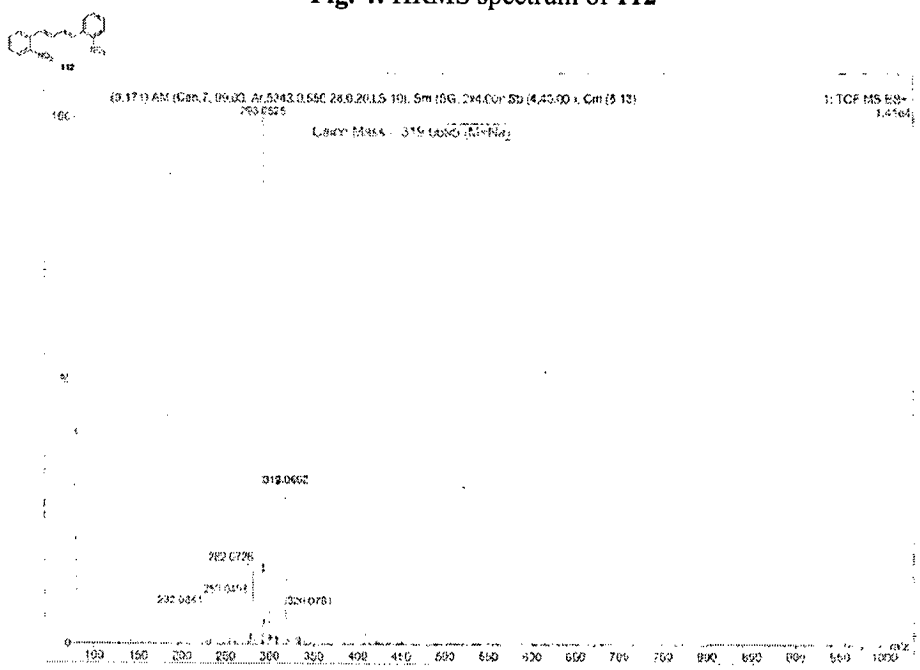


Fig. 5: ^{1}H NMR spectrum of **36**

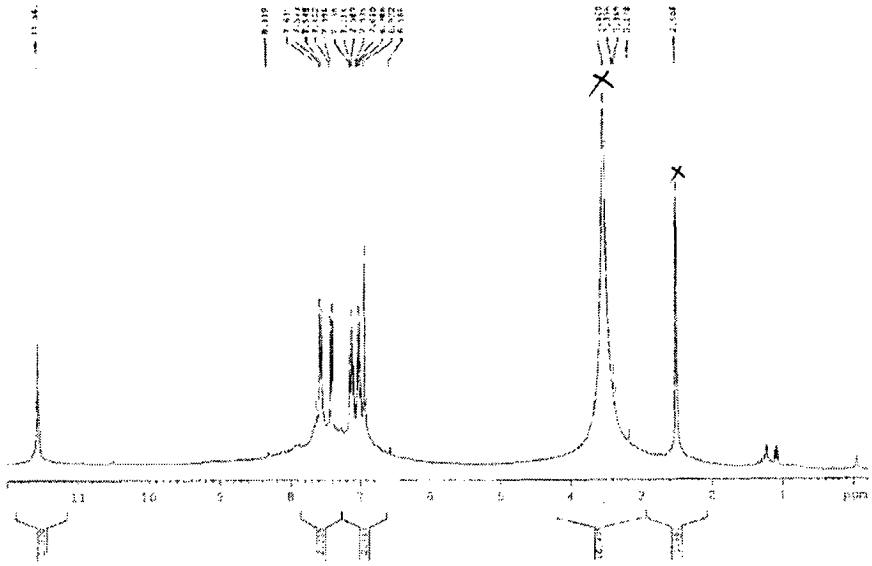
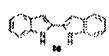


Fig. 6: ^{13}C NMR spectrum of **36**



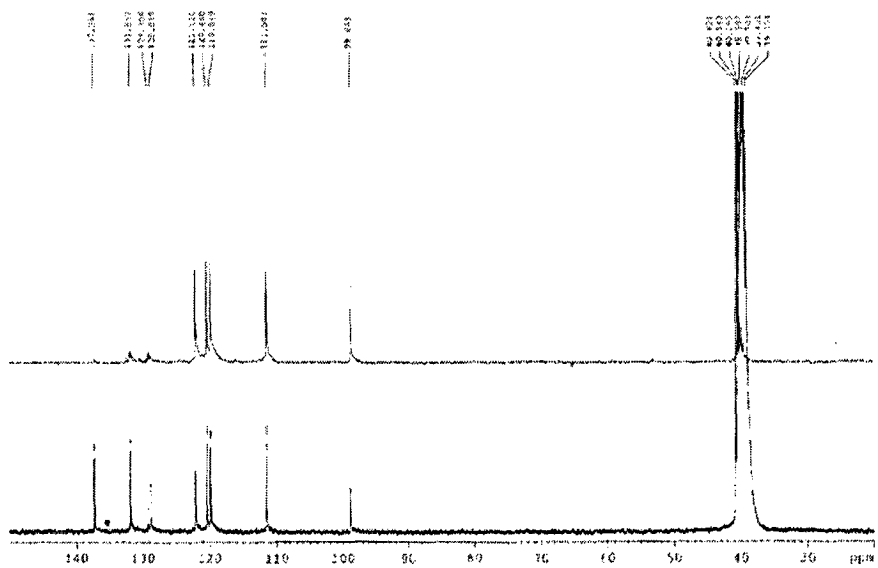


Fig. 7: ^{1}H NMR spectrum of **119**

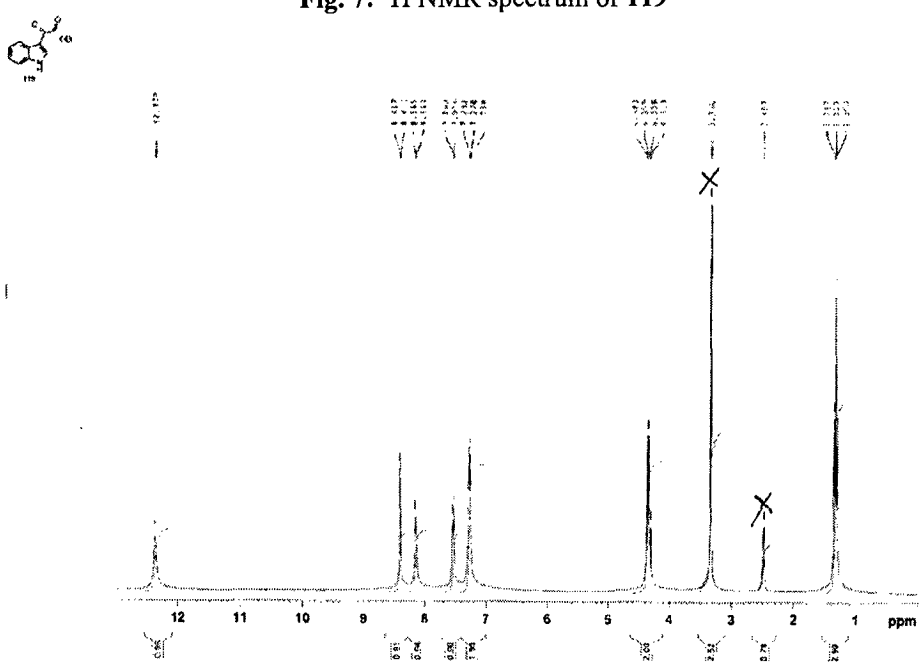


Fig. 8: ^{13}C NMR spectrum of **119**

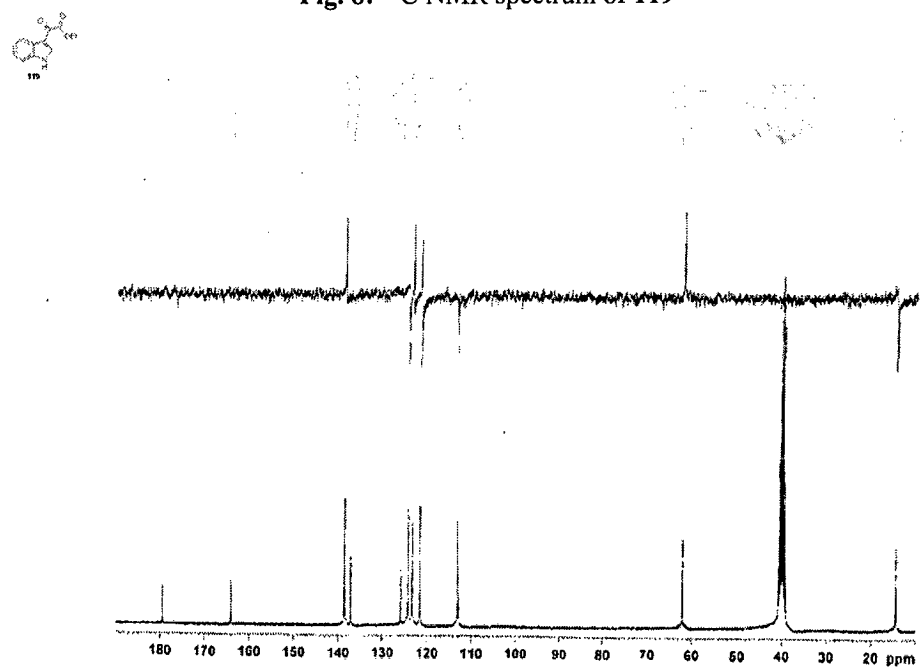


Fig. 9: ^{1}H NMR spectrum of **121**

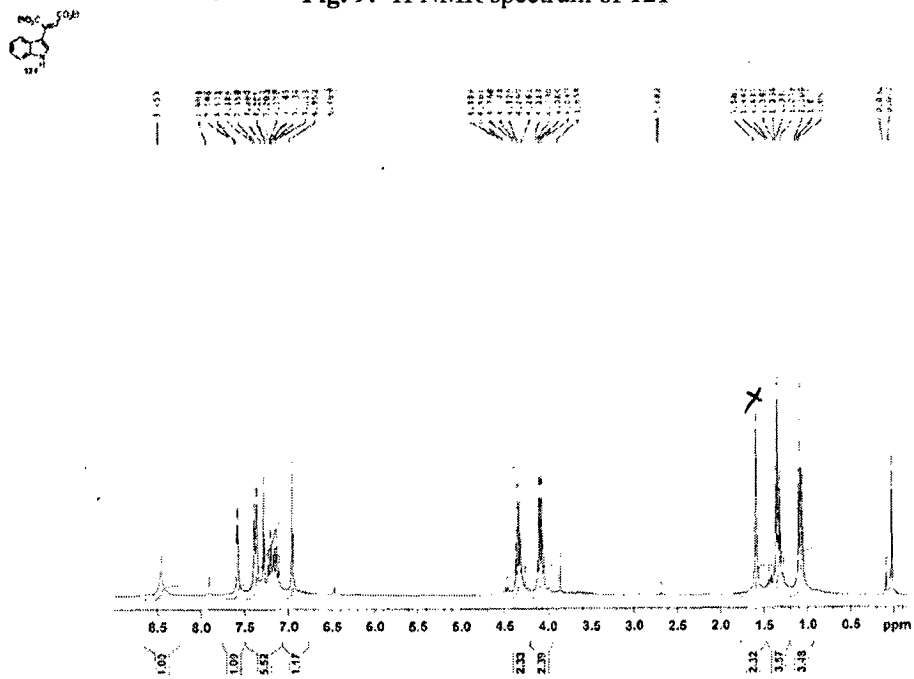


Fig. 10: ^{13}C NMR spectrum of **121**

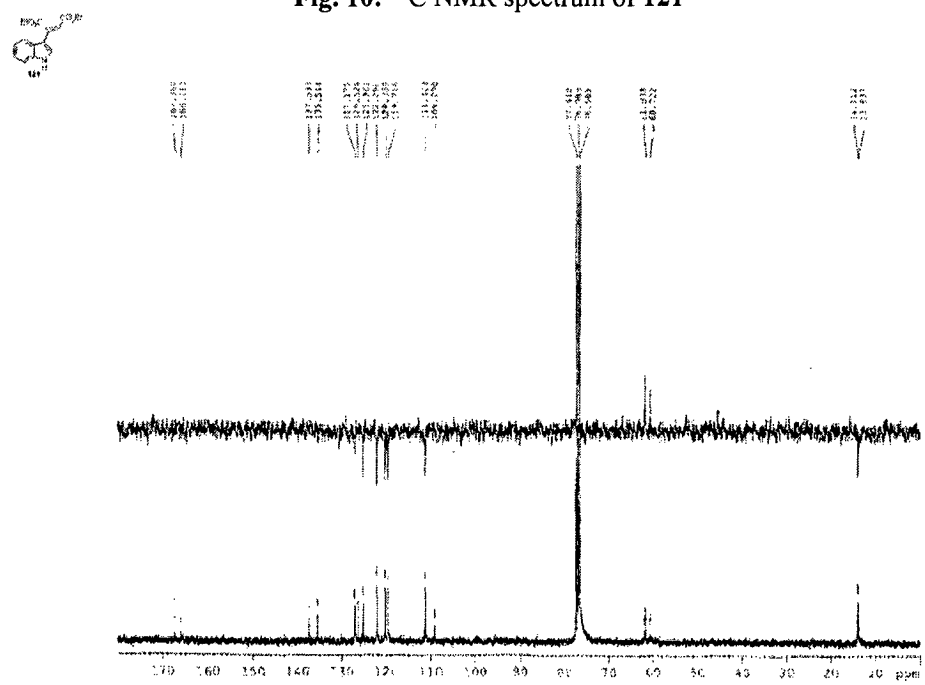


Fig. 11: ^{1}H NMR spectrum of **130**

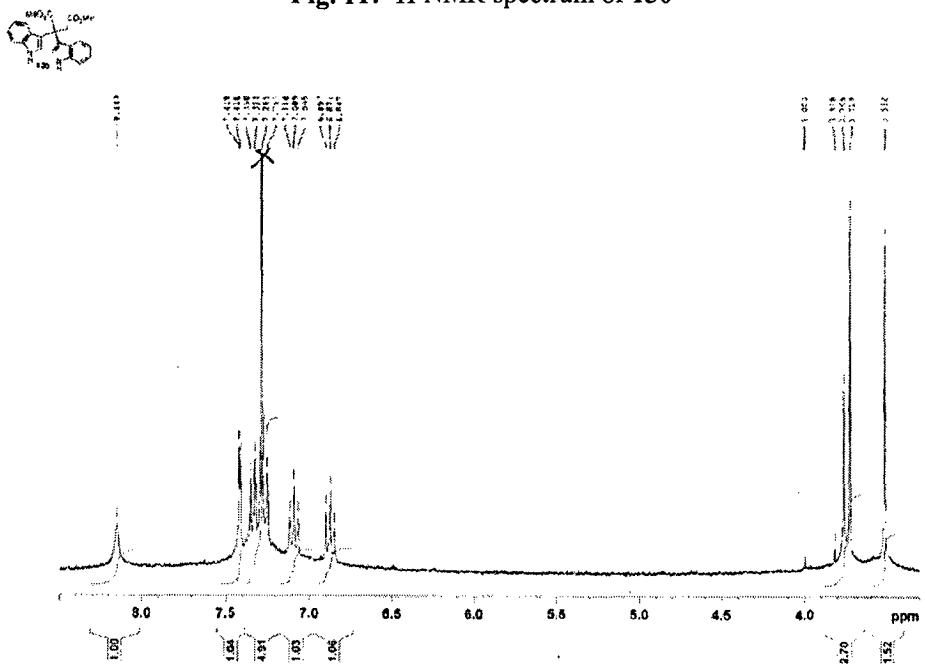


Fig. 12: ^{13}C NMR spectrum of **130**

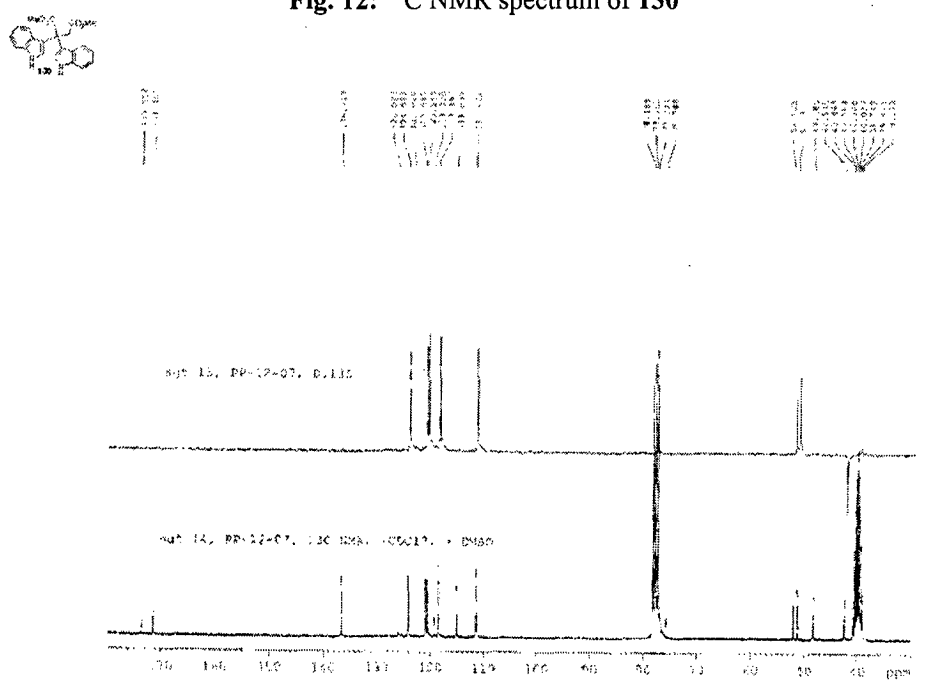


Fig. 13: ^{1}H NMR spectrum of **131**

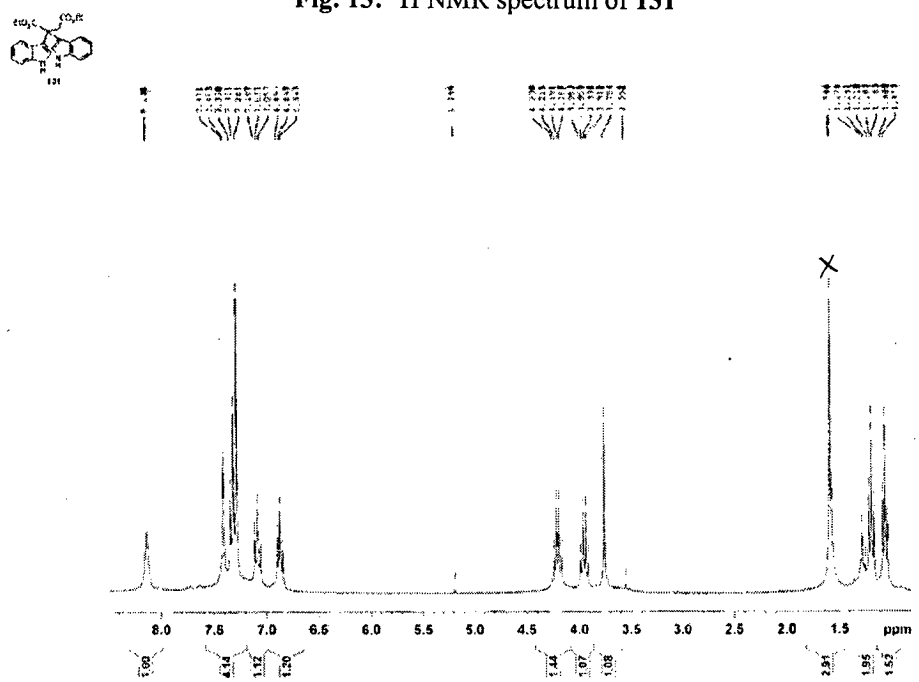


Fig. 14: ^{13}C NMR spectrum of **131**

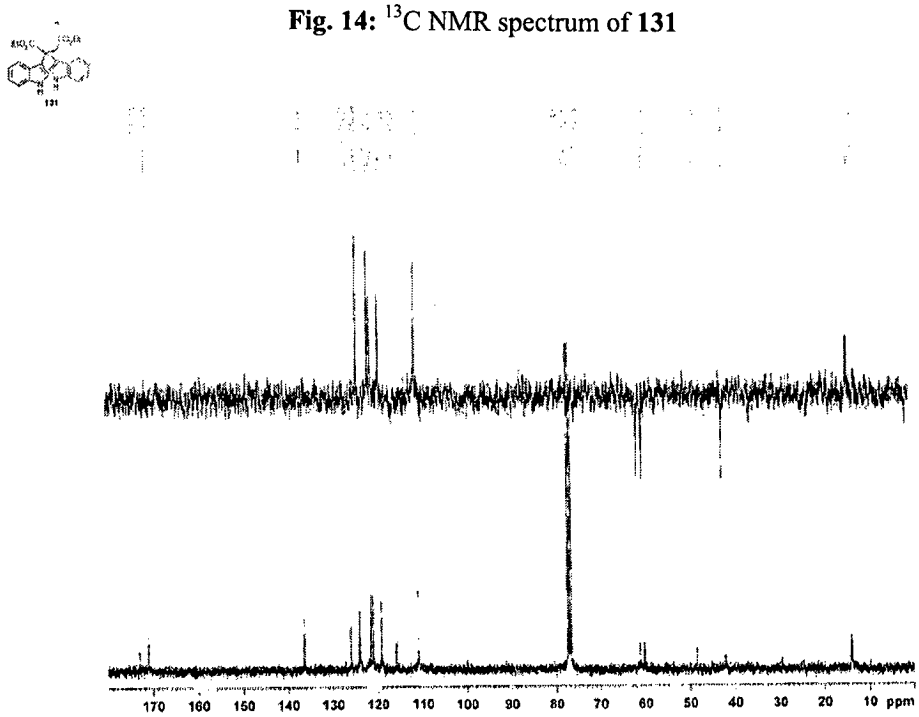


Fig. 17: ^{1}H NMR spectrum of **175**

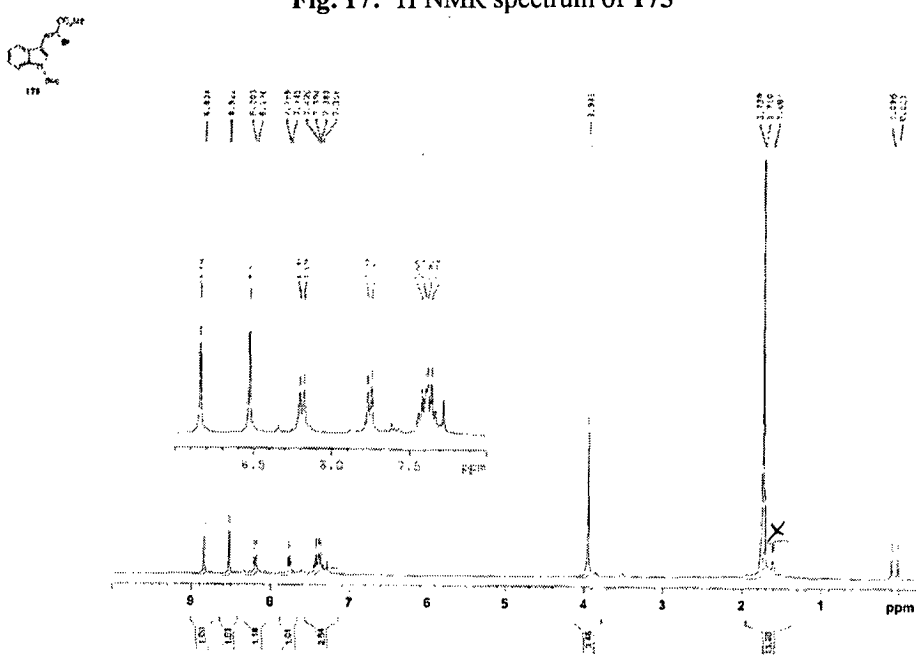


Fig. 18: ^{13}C NMR spectrum of **175**

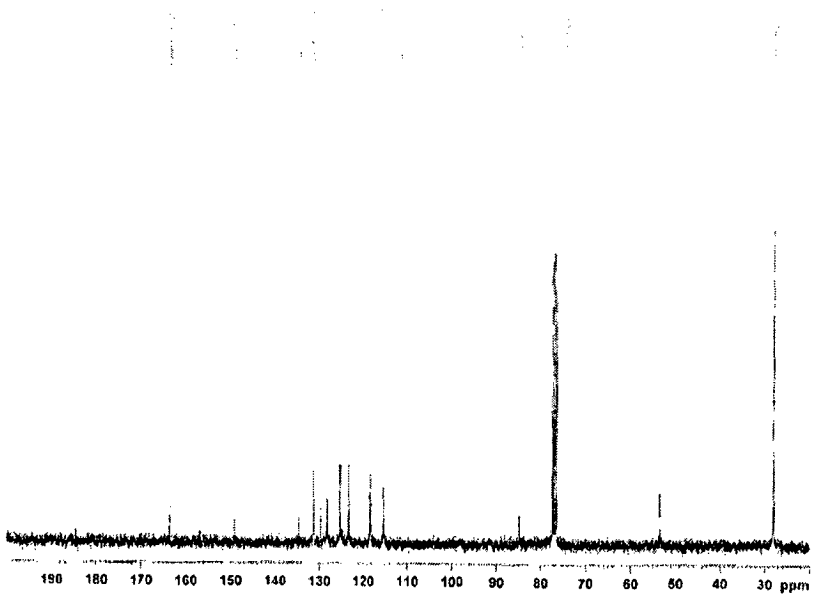


Fig. 19: ^{1}H NMR spectrum of **174**

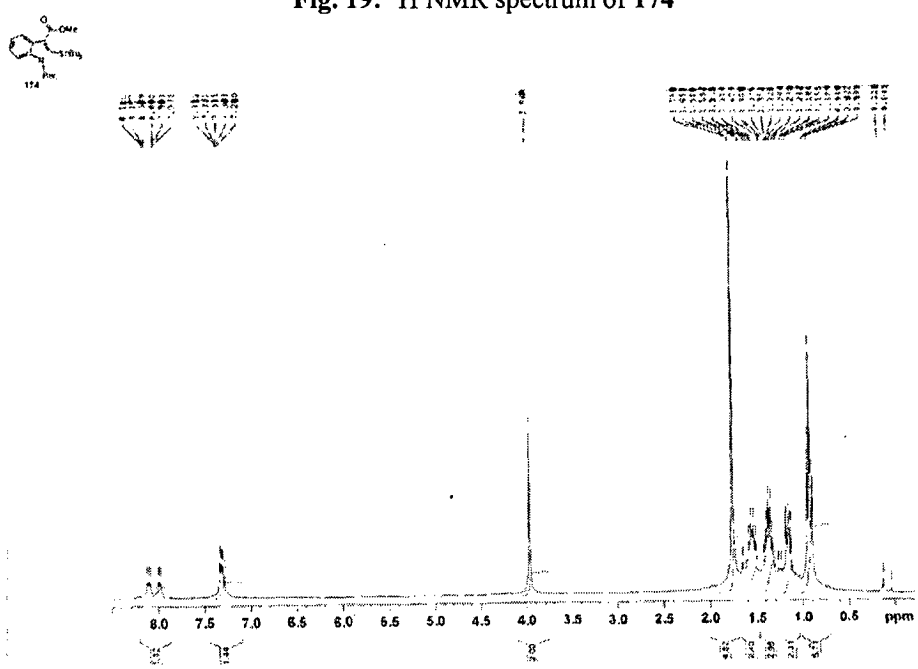
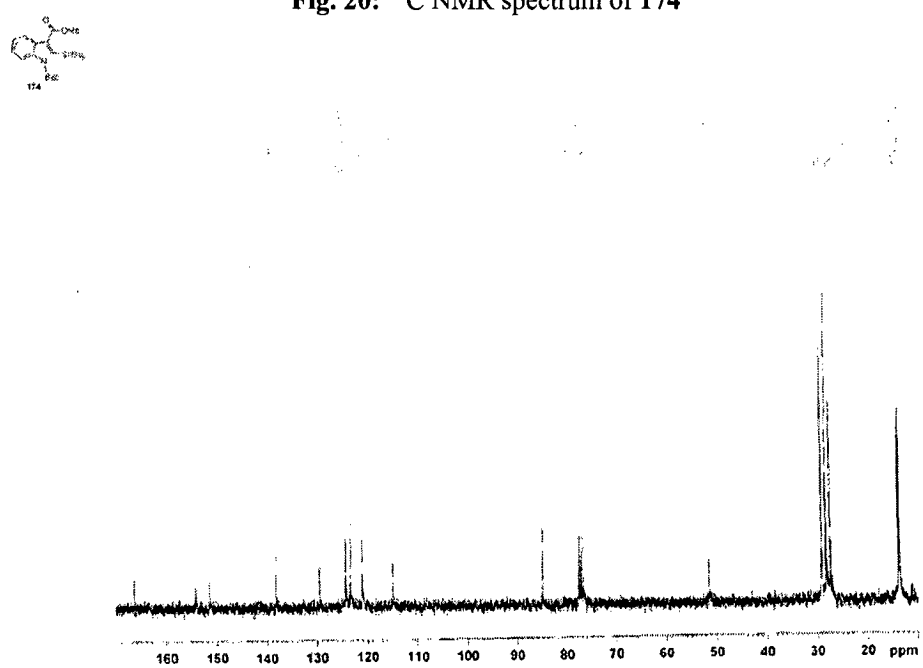


Fig. 20: ^{13}C NMR spectrum of **174**



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