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THE DESIGN AND SYNTHESIS OF SELECTIVE KINASE INHIBITORS BASED ON THE INDOLOCARBAZOLE NATURAL PRODUCT K252A

A Dissertation
Presented to the Faculty of the Graduate School
of
Yale University
in Candidacy for the Degree of
Doctor of Philosophy

by
Dejah Thoris Petsch

Dissertation Director: Professor John L. Wood

December 1999

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ABSTRACT

THE DESIGN AND SYNTHESIS OF SELECTIVE KINASE INHIBITORS BASED ON THE INDOLOCARBAZOLE NATURAL PRODUCT K252A

Dejah Thoris Petsch

Yale University

1999

The total syntheses of several K252a analogs are described. Methyl derivative (-)-**52a** showed inhibitory activity against protein tyrosine kinases with IC_{50} values ranging from high nM to low μM and possessed a better selectivity profile than the natural product (+)-K252a (**1**). Benzyl derivatives (-)-**80a** and (+)-**80b** and octenyl derivative (-)-**98** were also produced, but were not found to be inhibitors of protein tyrosine kinases. In addition, a potent but unselective dimer (-)-**100a,b** of K252a was prepared.

Benzyl derivative (-)-**80a** was found to be an excellent inhibitor (IC_{50} = 2 nM) of engineered protein tyrosine kinases. Isopropyl, isobutyl, *sec*-butyl and phenethyl analogs were prepared in order to ascertain the effect of different sized alkyl groups on binding affinity and selectivity for mutated protein tyrosine kinases (I338G v-Src, T339G c-Fyn, T314A c-Abl, F80G CDK2 and F89G CAMKIIa). Isobutyl derivative (-)-**111b** showed extremely potent inhibition of Src and Fyn mutants (IC_{50} = 230 to 550 pM) and also showed selectivity as high as 140,000-fold for the engineered kinase. In addition, modest selectivities were observed for F80G CDK2 and F89G CAMKIIa by inhibitors (-)-**111b** and (-)-**120**, respectively.

For My Family

ACKNOWLEDGEMENT

I would first like to thank my advisor Professor John L. Wood for the great deal of time and effort he invests in each member of his research group and the generosity with which he shares his substantial talent for organic synthesis. One of the most important things about John is that he provides his students with not only the tools of an experienced synthetic chemist, but also those of a well rounded scientist. Over the past five years, it has been a pleasure to share John's enjoyment of not only the exciting results, but also the daily successes and failures produced in a synthesis project. It has certainly been a privilege to have the opportunity to learn from a synthetic chemist as talented as John.

I am also indebted to the other members of my thesis committee, Professors Frederick E. Ziegler and Alanna Schepartz. My semesters spent as a teaching assistant for their sophomore organic chemistry classes were not only enjoyable, but a learning experience as well, which I regard as one of the highlights of my graduate career.

I would also like to thank my undergraduate research advisor Professor Katherine H. Freeman. Kate was a superb mentor and invested her valuable time and energy into my future as a scientist, showing a great deal of trust and respect for a sophomore who had never done research before.

I owe my greatest thanks to the Wood Group as a whole. It has been highly interesting and exciting to watch a group of about five evolve and grow to over twenty scientists who share a common love of synthesis. In the beginning of my tenure in the Wood labs I received a great deal of help and support from senior group members Dr. Brian M. Stoltz and Dr. Derek A. Pflum. I very much appreciated their taking the time to answer many questions with an incredible level of patience. In addition, Brian and Derek's excellent work on the total synthesis of K252a and the rhodium carbenoid-initiated Claisen rearrangement respectively, set the stage and the standard for my own project. I was also fortunate to have the opportunity to work with two very talented undergraduates, Steven N. Goodman, and Elizabeth Hawkins both of whom provided not only starting materials, but also ideas for the project and, along with Brian and Derek, made my first year in lab an enjoyable one.

Although I am thankful to each and every member of the Wood Group, past and present, three people stand out as having made my tenure infinitely better than it would otherwise have been. First, my lab partner Alexandra A. Holubec has

been a wonderful friend and colleague who always had the time to discuss everything from how to fix a reaction that wouldn't work to what the next W6 t-shirt design should be. Alex has a rare gift for making those around her happy which I will certainly miss, and I can't imagine a person with whom I would have rather shared a bench for three years. Second, George A. Moniz is a brilliant chemist and an extremely nice person in spite of what he says to the contrary. George possesses the considerable talent of being able to explain almost anything in an extremely clear and understandable manner. In addition, George and Alex are both very funny people and I certainly won't forget either George's impressions ("Hi-ho Corey the Frog here...") or Alex's practical jokes ("What are you eating under there?"). I am also very grateful to George and Alex for taking the time to proofread this thesis, thereby greatly improving its quality. Lastly, postdoc Dr. Julie A. Dixon joined the group and in a whirlwind year not only held my hand through the entire job interview process, but proved herself an invaluable resource and friend.

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Finally, I would like to thank my family. My parents, who are both PhD organic chemists themselves, for inspiring me to be a chemist in the first place, my sister Jaelieth for being a chemist in spite of liking rock climbing better, and last but not least, my husband Steven who has for many years served not only as my most accurate critic, but also as my best friend. Thank you all.

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LIST OF ABBREVIATIONS

ATP	adenosine triphosphate
aq.	Aqueous
Bn	Benzyl
br	Broad
t-Bu	*tert*-Butyl
calcd	Calculated
CAMKIIa	Ca^{+2}/calmodulin dependent protein kinase IIa
cAMP	cyclic adenosine monophosphate
cat.	Catalytic amount
CDK2	cyclin dependent protein kinase 2
CHOC	Chinese Hamster Ovary cell
CI	Chemical ionization
CSA	Camphorsulfonic acid
d	doublet
dec.	Decomposition
DAG	Diacyl glycerol
DCC	Dicyclohexylcarbodiimide
DMA(C)	*N,N*-Dimethylacetamide
DMAP	4-Dimethylaminopyridine
DMB	3,4-Dimethoxybenzyl
DMF	*N,N*-Dimethylformamide
DMS	Dimethyl sulfide
DMSO	Dimethyl sulfoxide
EDCI	1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride
ee	Enantiomeric excess
EI	Electron impact
equiv	Equivalent
Et	Ethyl
FAB	Fast atom bombardment
FTIR	Fourier Transform Infrared (Spectrometer)
h	hour
HMDS	bis(trimethylsilyl)amide
HOBt	1-hydroxybenzotriazole hydrate
HPLC	High performance liquid chromatography

Hz	Hertz
IC_{50}	50% inhibitory concentration
IP_3	D-*myo*-inositol-1,4,5-triphosphate
IR	Infrared (spectrum)
kD	kilo Dalton
m	Multiplet or medium
m	Mass
µM	micromolar
Me	Methyl
min	minutes
mol	Mole
mp	Melting point
Ms	Mesyl (methanesulfonyl)
MTPA	α-Methoxy-α-trifluoromethylphenylacetic acid
NBS	*N*-Bromosuccinimide
nM	nanomolar
NMR	Nuclear magnetic resonance
p-ABSA	*p*-acetamidobenzenesulfonyl azide
Ph	Phenyl
PhH	Benzene
PIP_2	L-α-Phosphatidyl-D-*myo*-inositol-4,5-biphosphate
PKA	Protein kinase A
PKC	Protein kinase C
pM	picomolar
PP1	4-amino-1*tert*-butyl-3-(*p*-methylphenyl)pyrazolo[3,4-*d*]pyrimidine
ppm	Parts per million
q	quartet
s	Singlet or strong
t	Triplet
TFA	Trifluoroacetic acid
THF	Tetrahydrofuran
TLC	Thin layer chromatography
p-TSA	*p*-Toluenesulfonic acid
w	Weak
z	Charge
Δ	Heat at reflux

CHAPTER ONE

Protein Kinases and Their Inhibition by the Indolocarbazole Class of Natural Products

1.1 Background and Introduction.

1.1.1 Protein Kinase Overview.

Protein kinases are enzymes that phosphorylate proteins. Specifically, amino acid residues that contain an alcohol (serine, threonine, and tyrosine) can be substrates for phosphorylation reactions.[1] The large super-family of protein kinases consists of over 2,000 members and can be divided into two broad categories: protein serine/threonine kinases and protein tyrosine kinases. Members of these two groups may, in turn, be further subdivided by factors such as similarities in catalytic domain, regulation mode, substrate specificity, whether or not the kinase is receptor-bound and overall structure.

One of the keys to ultimately understanding the role of protein kinases is structure. For example, much is known about the structure of protein kinase A (PKA) which is activated in the cell by cyclic adenosine monophosphate (cAMP). In fact, all known effects of cAMP in eukaryotic cells result from the activation of protein kinases.[2]

PKA has two regulatory and two catalytic subunits resulting in a four-membered quaternary structure. In the holoenzyme the two regulatory units are bound to the catalytic sites rendering them inactive. Binding of cAMP to the

regulatory units causes a conformational change to the catalytic sites, thus activating them and allowing for the phosphorylation of appropriate serine and threonine residues in the substrate.[2]

The kinase domain of the catalytic unit is highly conserved among protein kinases and consists of 12 subdomains (see Figure 1.1.1). The kinase domain folds into two lobes, the smaller of which contains the *N*-terminus and consists mainly of anti-parallel β-sheets holding in place the MgATP complex which serves as the source of the phosphate group. The larger lobe contains the C-terminus and is made up primarily of α-helices. This portion of the enzyme binds the substrate and initiates the phosphate transfer.[1]

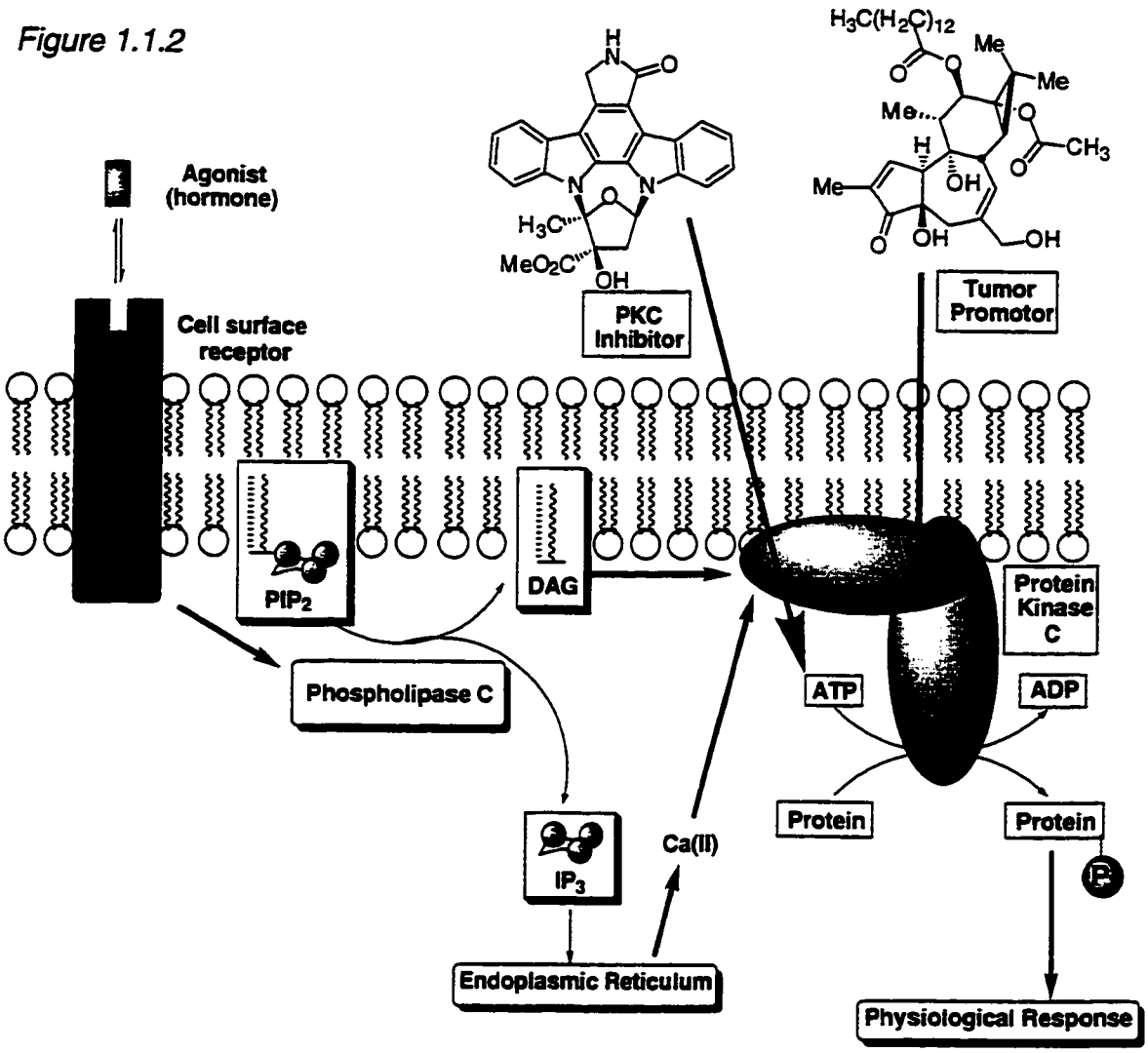
Figure 1.1.1

Kinases play an important role in signal transduction pathways, making them a topic of great interest throughout the scientific community. For example, some carcinogenic viral kinase forms are known to elevate cytoplasmic tyrosine residue phosphorylation, which is the protein switch for proto-oncogenes. In addition, the protein kinase C (PKC) family is activated by phorbol esters, the most effective tumor promoters yet discovered.

1.1.2. Protein Kinase C.

The PKC family is comprised of serine/threonine-specific kinases that are approximately 70-80 kD in size. Members of this kinase family are an important part of the sequence that enhances the release of glucose in the bloodstream in response to stress. PKC activation is initiated by the specific binding of an extracellular agonist to a receptor on the surface of the cell. Epinephrine and norepinephrine are released in response to stress by the adrenal glands activating (1) β-adrenergic receptors that respond by initiating an increase in intracellular cAMP via the enzyme adenylate cyclase and (2) α-adrenergic receptors that stimulate phospholipase C to release inositol triphosphate (IP_3), diacylglycerol (DAG) and calcium ions.[3] PKC is activated by DAG and Ca^{2+}, thereby inactivating glycogen synthase by phosphorylation (see Figure 1.1.2).[4] This response causes glucose to be released into the bloodstream.

Figure 1.1.2


Agonist
(hormone)
Cell surface
receptor
PIP2
DAG
Phospholipase C
IP3
Endoplasmic Reticulum
Ca(II)
PKC
Inhibitor
Tumor
Promotor
Protein
Kinase
C
ATP
ADP
Protein
Protein
P
Physiological Response

1.1.3 Src Family of Protein Kinases.

Another protein kinase that has been the subject of numerous studies is the non-receptor tyrosine kinase Src. Src is the product of the first proto-oncogene to be structurally characterized, and plays the role of a protein switch that can be turned on by a number of receptor-mediated signals to which it responds through changes in kinase activity.[5] When Src is switched on, it phosphorylates proteins which contain a kinase catalytic domain. This activity is controlled by multiple regulatory interactions that are mediated by other parts of its polypeptide chain. There are nine members of the Src family of protein kinases: Fyn, Yes, Fgr, Lyn, Hck, Lck, Blk, Yrk and Src itself. One reason Src is important is because it has a widespread cellular distribution, and every eukaryotic cell type studied to date contains one or more of its homologs.

Members of the Src family share the following N to C terminal domain organization, with each domain individually named as a Src-homology (SH) region.[6] Closest to the N-terminal is the SH4 domain, a myristylation and membrane localization signal containing a 50-70 residue segment that is unique to Src. The next segment, the SH3 domain, is a small β-barrel module that presents a non-polar groove complementary to the short peptides of the target proteins. The SH2 domain polypeptide segments contain a phosphotyrosine residue, and the SH2 and SH3 domains together serve to mediate protein-protein interactions in cellular signaling cascades. The combination of SH2 and SH3 domains can be found in many proteins outside the Src family. Finally, the SH1 domain is the catalytic domain of the protein that possesses the kinase activity. In addition, the enzyme has a linker loop which joins the SH2 and kinase domains and a C-terminal tail that includes a tyrosine residue critical to auto-regulation.[7]

Src is autoregulated by intramolecular interactions that lock the molecule in a closed and inactive conformation. Proline residues in the SH2 kinase linker portion are bound to the SH3 domain, and the SH2 domain binds a phosphotyrosine residue (tyrosine 527) making the kinase catalytic domain inaccessible to other proteins.[8] V-Src, the oncogene product from the Rous Sarcoma Virus, an avian retrovirus, is a constitutively active kinase with a deleted carboxyl terminus. Mutations in either the SH2 or the SH3 domain can also affect the stability of the closed, inactive conformation, leading to a constitutively active Src molecule which has been linked to cancer.[9]

Src changes its conformation to an open and active state by dephosphorylation of residue 527 or by apposition of a high affinity ligand for the SH2 or SH3 domain that disrupts the intramolecular interactions that maintain the closed and inactive state.

Protein kinases play vital roles in cellular signal transduction. Thus, gaining a better understanding of how enzymes such as PKC and Src function is an active area of research in the field of biochemistry.

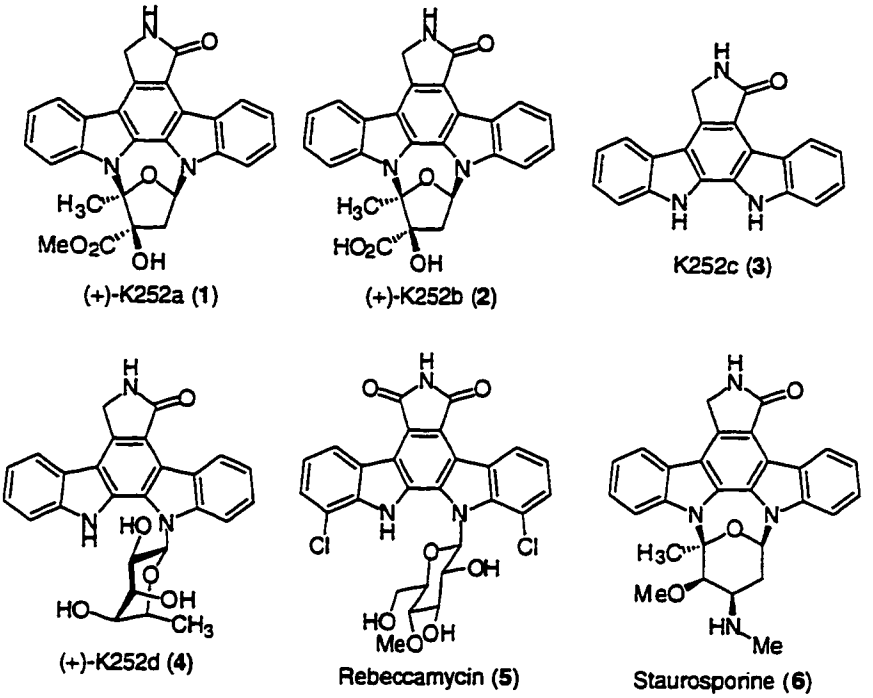
1.2 The Indolocarbazole Family of Natural Products.

1.2.1 Isolation and Biological Activity.

In 1985, the first furanosylated indolocarbazole was discovered independently by two Japanese research groups. The compound was first identified by Sezaki and co-workers from *Actinomadura* sp. SF-2370, and subsequently named SF-2370 (**1**).[10] One year later Kase and co-workers described the isolation of a compound from *Norcardiopsis* sp. K-252, termed

K252a, that was found to be identical to that isolated by Sezaki.[11] In addition, three related compounds K252b-d (**2-4**) were also isolated (see Figure 1.2.1).[12]

Figure 1.2.1



The indolocarbazole family includes not only furanosylated congeners like K252a and K252b, but also compounds that contain only the indolocarbazole core (i.e., K252c (**3**)), monoglycosyl linked indolocarbazoles such as K252d (**4**) and rebeccamycin (**5**), as well as pyranosylated moieties like staurosporine (**6**).

Of particular interest in Kase's studies was the observation that K252a inhibits the action of PKC with an IC_{50} value of 32 nM. Soon afterward, Tamaoki discovered that staurosporine also inhibits PKC but with a slightly higher inhibitory power (IC_{50} = 2.7 nM).[13] The discovery of the exciting biological activity of this family of natural products led to their consideration as lead structures in the development of chemotherapeutics against cancer,[14] Alzheimer's disease,[15] and other neurodegenerative disorders.[16] Following the isolation of K252a and staurosporine,

a number of functionalized indolocarbazoles have been discovered and found to possess varied biological profiles.[17]

The indolocarbazoles K252a and staurosporine, two of the most powerful PKC inhibitors isolated to date, are presumed to act by blocking the ATP binding site and thereby preventing protein phosphorylation.[18] Unfortunately, this mode of kinase inhibition results in low selectivity due to the ubiquitous cellular use of ATP and thereby the concurrent inhibition of several kinases. The preparation of indolocarbazole derivatives possessing selectivity toward specific malfunctioning kinases associated with a disease state has long been a key goal in the kinase inhibitory field.

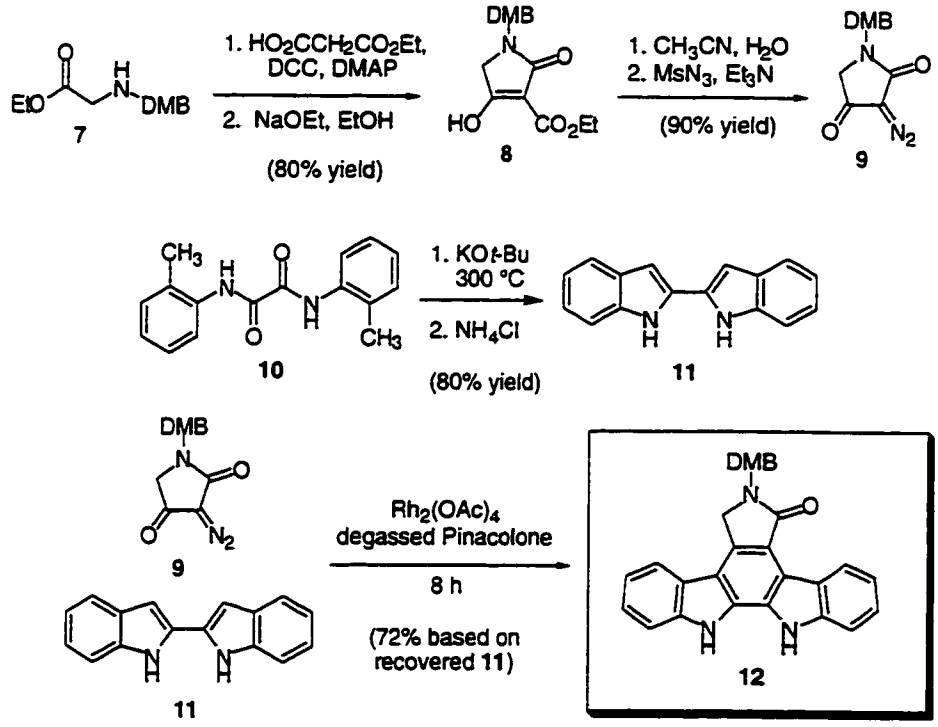
1.2.2 The Total Synthesis of (+)-K252a.

In 1994, almost 17 years after the initial discovery of the first indolocarbazole by Ōmura, the Wood Group began a synthetic effort focused on the total synthesis of indolocarbazole natural products. K252a was selected as the initial target, owing to the interesting bis-*N*-furanosyl attachment to the aglycon moiety, its potent biological activity, and its relatively unexplored chemistry as compared to staurosporine. The synthesis was completed in 1995, and a brief overview follows.

N-dimethoxybenzyl substituted glycine ester **7**[19] was exposed to coupling with ethyl hydrogen malonate mediated by DCC/DMAP, followed by Dieckmann cyclization (NaOEt/EtOH) to produce lactam **8** (see Scheme 1.2.1). A single-pot decarboethoxylation/diazo-transfer reaction was effected by first heating ester **8** in wet acetonitrile and then treating the cooled reaction mixture (0 °C) with MsN_3 and triethylamine.[20] The overall process involved a single purification step, could be conveniently carried out on a 20 g scale, and resulted in an approximate 50% overall yield of diazo lactam **9** from **7**. The core indolocarbazole portion of the aglycon, 2,2'-

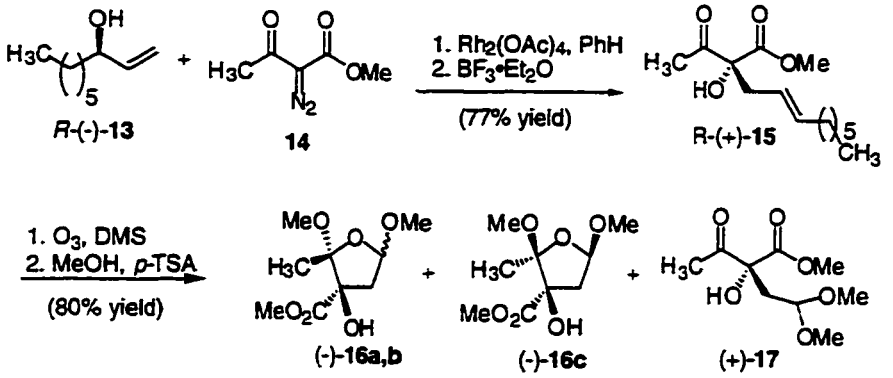
biindole (**11**) was prepared from oxaltoluidide **10** via a double Madelung cyclization according to an excellent procedure recently published by Bergman.[21] In the key aglycon forming reaction, a 1:1 mixture of biindole **11** and diazo lactam **9** was treated with $Rh_2(OAc)_4$ (1.0 mol %) in degassed pinacolone at reflux for 8h producing a 36% yield of protected aglycon **12** and 50% unreacted biindole (72% yield based on recovered **11**).

Scheme 1.2.1



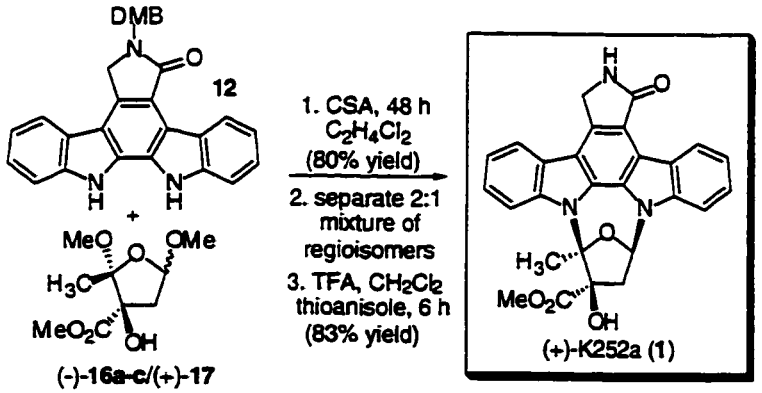
With aglycon **12** in hand, attention turned to the preparation of the furanose, which began with exposure of *R*-(-)-**13** to diazoester **14** and catalytic $Rh_2(OAc)_4$ (PhH, 80 °C, 20 min), followed by introduction of $BF_3 \bullet Et_2O$ to the cooled reaction mixture furnishing alcohol (+)-**15** in 77% yield (see Scheme 1.2.2). Ozonolysis of olefin (+)-**15** followed by acid-mediated cyclization produced a mixture of carbohydrates (i.e., (-)-**16a-c**/(+)-**17**) in 80% yield.

Scheme 1.2.2



Cycloglycosidation of indolocarbazole **12** with the mixture of alcohols (-)-**16a-c**/(+)-**17** then produced a 2:1 mixture of regioisomers that, upon chromatographic separation and deprotection, produced (+)-**1**, a compound identical in all respects to the natural material (see Scheme 1.2.3).[22]

Scheme 1.2.3



Thus, the first total synthesis of furanosylated indolocarbazole K252a (**1**) was completed in the Wood Group by developing new rhodium carbenoid chemistry in the preparation of aglycon **12** and furanose **16**. The total synthesis required only twelve steps from commercially available materials, with a longest linear sequence of seven steps and an overall yield of 21% from ethyl glycinate. The remarkable

stereo- and regioselective cycloglycosidation served as the cornerstone of the approach and its great efficiency prompted the pursuit of staurosporine (**6**), other pyranosylated indolocarbazoles, and analogs of K252a itself.

1.3 Prior Efforts Toward Indolocarbazole Analogs.

1.3.1 Sites of Natural Indolocarbazole Derivatization.

Considerable effort has been focused on the production of analogs of both K252a and staurosporine, however, prior to our work, the lack of efficient synthetic approaches to this class of molecules had limited the effort to simple analogs of structures that are readily derived from the natural material. The preparation of natural product analogs is often generally limited to functionalization of the natural material since the production of a wide variety of analogs through total synthesis is usually not practical. In the case of K252a the synthesis developed in our group is quite efficient and concise, allowing analogs to be readily prepared that are not availiable from derivatization of the natural material.

As summarized in Figure 1.3.1, the published analogs of K252a and staurosporine have been prepared via single and double Friedel-Crafts alkylation (R_3, R_4), oxidation of the lactam methylene (R_1), and acylation of the lactam nitrogen (R_2). In addition, the literature indicates that derivatization of the carbohydrate moiety has been limited to manipulation of the resident functionality. Thus, synthesis, in particular the highly efficient approach developed in our laboratories, provides a unique and viable means of accessing a wide variety of analogs that have yet to be explored for their medicinal potential.

Figure 1.3.1

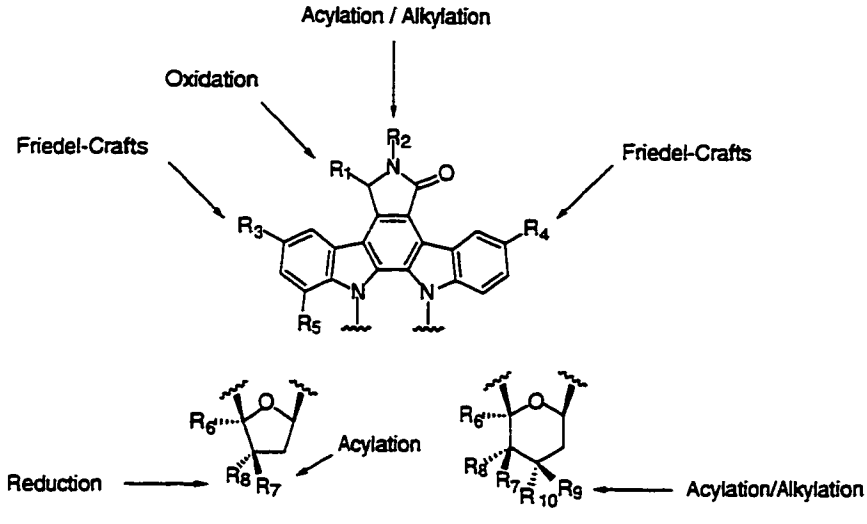


Figure 1.3.1 illustrates a key to the data outlined in Tables 1.1.1 and 1.1.2[23] which represent a compilation of the analogs of staurosporine and K252a that were known when we began this work. In this table structural elements which differ from those found in the natural substances are illustrated, and it is interesting to note that the only known analogs with functionalization at the lactam methylene have been the result of oxidation to the maleimide or aminal, while the carbohydrate methylene has not yet been derivatized.

Table 1.3.1 Known Analogs of K252a (Differences from natural structure are tabulated).

CMP #	R1	R2	R4	R7	R8	Misc.	ref. # 11
Ia		Me					A
Ib		PMB					B
Id	=O	NH_2		R8			C
Ie		3,4-OMB					D
Ig	=O	CH_2CH_2Br			CH_2OH		C
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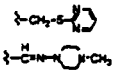
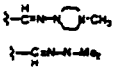
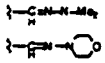
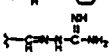
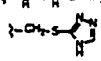
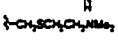
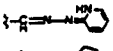
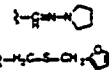
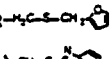
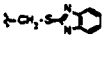
Ii		Ac		OAc			E
Ij		Ac		OAc			E
Ik		Ac		OAc			E
Il		Ac		OAc			E
Im		Ac		OAc			E
In		Ac		OAc			E
Io		Ac		OAc			E
Ip		Ac		OAc			E
Iq		Ac		OAc			E
Ir		Ac		OAc			E
Is		Ac		OAc			E
It		Ac		OAc			E
Iu		Ac		OAc			E
Iv		Ac		OAc			E
Iw		Ac		OAc			C, E
Ix	=O	NH_2			CH_2NH_2		G
Iy		Me		R8			G
Iz							G
Iaa	=O	CH_2CH_2OH			CH_2OH		C
Iab	=O	CH_2CH_2Br		R8			G
Iac	=O	Me			CH_2OH		C
Iad	=O	NH_2		R8			G
Iae		Ac		OAc			H
Iaf	=O						J
Iag		Ac	Ac	OAc			J
Iah	=O	CH_2Ph					J
Iai		Me	O-n-Pr				J
Iaj		Ac		R8			J
Iak		Ac	CH_2SEt	OAc		R3=CH_2OH	C, K
Ial		Ac	CH_2SEt	OAc		R3=R4	C, K
Iam		Ac	CH_2OH	OAc		R3=R4	C, K
Ian		Ac		OAc		R3=R4	C, K
Iao	=O	Me		H	H	R8=H	N
Iap					CH_2NH_2		O
Iaq				R8			O
Iar		CH_2Ph		H	H	R8=H	T
Ias		CH_2Ph		H	H		T
Iat	=O	CH_2Ph		H	H		T

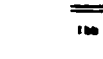
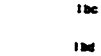
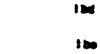
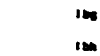
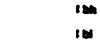
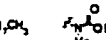
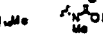
CMP #	R1	R2	R4	R7	R8	Misc.	ref. # 11
Iba	=O						AB
Ibc	=O						AB
Ibd	=O						AB
Ibe	=O						AB
Ibf	=O						AB
Ibg	=O						AB
Ibh	=O						AB
Ibi	=O						AB
Ibj	=O						G, W, AB
Ibk	=O						AB
Ibl	=O	Me			OMe		G, AB
Ibm	=O	Me					AB
Ibn	=O	Bn			OBn		AB
Ibo	=O	Bn					AB
Ibp		Ac			OAc		AC
Ibq		Ac			OAc		AC
Ibr		Ac			OAc		AC
Ibs		Ac	CH_2OEt		OAc		AC
Ibt							AC
Ibu		Ac	CHO		OAc	R3=R4	C, K, AC
Ibv		Ac			OAc		AC
Ibw		Ac			OAc		AC
Ibx							AD
Iby		Ac	Me		OAc		AD
Ibz		Ac		OAc			AD
Ica		Ac		OAc			C, E, AD
Icb		Ac		OAc			E, AD
Icc		Ac	Ac	OAc		R3=R4	AC, AD
Icd		Ac		OAc		R3=NH_2	AD
Ice		Ac		OAc		R3=R4	C, K, AC, AD
Icf		Ac		OAc			C, E, AC, AD
Icg	=O						AD
Ich	=O	NH_2					G, P, Q, W, AB, AD
Ici	=O	NH_2					AD
Icj	=O						AD
Ick		Ac	NO_2	OAc		R3=R4	AD
Icl	=O						AC, AD
Icm		Ac	NO_2	OAc			AC, AD
Icn		Ac	OAc	OAc		R3=R4	AD
Ico		Ac	OAc	OAc			AA, AC, AD
Icp		Ac		OAc			AD
Icq		Ac		OAc			AC, AD
Icr		Ac	Ac	OAc			AA, AC, AD
Ics							AD
Ict		Cl					AD
Icu		Ac		OAc			AA, AC, AE
Icv		CH_2Ph		H	H	R8=H	AF
Icw	=O				CH_2OH		M, P, U
Icx	=O						U
Icz	=O				CH_2NH_2		W
Icy				H	H	R8=H	Y
Ida		Me		OMe			F, I, L, R, S, V, X, Z
Idb	=O				CH_2OAc		AB

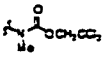
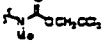
Table 1.3.2 Known Analogs of Staurosporine (Differences from natural structure are tabulated).

CMP #	R1	R2	R4	R9	Misc.	ref. #
II a		Me			R3=R 4	A
II b		Me			R3=R 4	A
II c		Me			R3=R 4	A
II d		Me			R3=R 4	A
II e		Me			R3=R 4	A
II f		Me			R3=R 4	A
II g	=O	Me	CHO		R3=R 4	A
II h		Me			R3=R 4	A
II i		Me			R3=R 4	A
II j		Me			R3=R 4	A
II k		Me			R3=R 4	A
II l		Me			R3=R 4	A
II m		Me			R3=R 4	A
II n		Me			R3=R 4	A
II o		Me			R3=R 4	A
II p		Me			R3=R 4	A
II q		Me			R3=R 4	A
II r		Me	NMe_2		R3=R 4	A
II s		Me	NH_2		R3=R 4	A
II t		Me	NO_2		R3=R 4	A
II u		Me				A
II v		Me				A
II w		Me				A
II x		Me	NMe_2			A
II y		Me	NH_2			A
II z		Me	NO_2			A
II aa		Me	Et		R3=R 4	A
II ab		Me			R3=R 4	A
II ac		Me	CHO		R3=R 4	A
II ad						A
II ae		$-(CH_2)_2$-OH				A

CMP #	R1	R2	R4	R9	Misc.	ref. #
II af						A
II ag		$-CH_2CHO$				A
II ah						A
II ai						A
II aj		Me	$-O-(CH_2)_8Me$		R3=R 4	A
II ak		Me	$-O-(CH_2)_{10}Me$		R3=R 4	A
II al		Me			R3=R 4	A
II am		Me			R3=R 4	A
II an		Me			R3=R 4	A
II ao		Me			R3=R 4	A
II ap	=O	Me	$-CH_2OH$		R3=R 4	A
II aq		Me	$CH_3SCH_2CH_2NMe_2$		R3=CH_2OH	A
II ar		Me	CH_2SEt		R3=R 4	A
II as		Me	$CH_2OCH_2CH_2NMe_2$			A
II at		Me	$CH_2SCH_2CH_2NMe_2$			A
II au		Me				A
II av		Me				A
II aw		Me				A
II ax		Me				A
II ay		Me				A
II az		Me			R3=R 4	A
II ba		Me			R3=R 4	A
II bb		Me			R3=R 4	A
II bc		Me			R3=R 4	A
II bd		Me	OAc		R3=R 4	A
II be		Me			R3=R 4	A
II bf		Me			R3=R 4	A
II bg		Me			R3=R 4	A
II bh		Me	NMe_2		R3=R 4	A
II bi		Me	NH_2		R3=R 4	A
II bj		Me				A
II bk		Me				A
II bl		Me	NMe_2			A
II bm		Me	NH_2			A
II bn		Me				A
II bo		Me	Et			A

Table 1.3.2 cont. Known Analogs of Staurosporine (Differences from natural structure are tabulated).

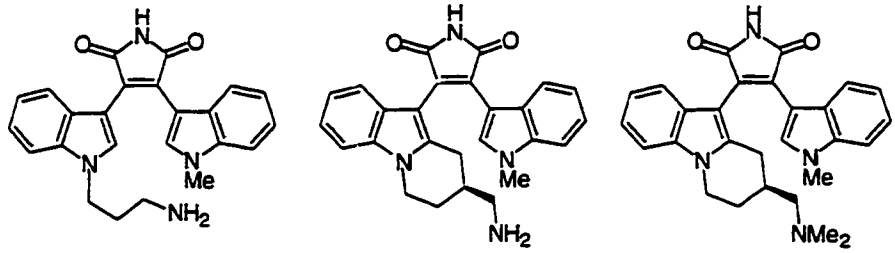
CMP #	R1	R2	R4	R9	Misc.	ref. #
‡ bp		Me	Me		R3=R4	A
‡ bq		Me	Me			A
‡ br		Me	CH_2OH		R3=R4	A
‡ bs		Me	CH_2OH			A
‡ bt						A
‡ bu		$(CH_2)_3OH$				A
‡ bv		CH_2CH_2OH				A
‡ bw						A
‡ bx		CH_2OH				A
‡ by		Me				A, H
‡ bz		Me				A, H, K
‡ ca		CH_2OCH_2Ph				I
‡ cb	=O	CH_2OCH_2Ph				I
‡ cd	=O	CH_2OCH_2Ph			R3=R4	I
‡ ce	=O	CH_2OCH_2Ph			R3=R4	I
‡ cf	=O	CH_2OCH_2Ph			R3=R4, R6=CH_3	I
‡ cg		CH_2Ph				K
‡ ch		Me			R6= 	K
‡ ci	=O	Me				K
‡ cj	=O	NH_2				K
‡ ck	OH	Me				K
‡ cl						K
‡ cm		Me	OH			K
‡ cn		Me	OH		R3=R4	K
‡ co		Me	Ac		R3=R4	A,K
‡ cp		Me	Ac			K
‡ cq		Ac	Ac			K
‡ cr		CH_2Ph				K
‡ cs		$(CH_2)_3Me$				K
‡ ct		Me	Ac		R3=R4	K
‡ cu		Me		NMe_2		K
‡ cv		Me	OH		R3=R4	K
‡ cw		Me	OH			K
‡ cx		Me				K
‡ cy	OH	Me				K
‡ cz		$(CH_2)_3Me$				K

CMP #	R1	R2	R4	R9	Misc.	ref. #
‡ da						K
‡ db						M
‡ dc				=O	R7=H	Q
‡ dd				H	R7=OAc	Q
‡ de				H	R7=OH	Q
‡ df				H	R7= =O	Q
‡ dg	=O	CH_2Ph			R7=R8	Y
‡ dh	=O	CH_2Ph	CH_3I		R7-R8	Y
‡ di	=O	CH_2Ph		H	R7=R8=H	AB
‡ dj		CH_2Ph				AD
‡ dk	=O	CH_2Ph				AD
‡ dl	=O	CH_2Ph				AD
‡ dm		CH_2Ph				AD
‡ dn		Ac	CHO			AG
‡ do		Ac	CHO		R3=R4	S, X, AJ
‡ dp		Ac	NO_2		R3=CH_2OH	AJ
‡ dq		Ac	NO_2		R3=CHO	AJ
‡ dr		Ac	CH_2OH		R3=R4	X, AJ
‡ ds		Ac	NO_2			S, X, AJ
‡ dt		Ac				S, X, AJ
‡ du	=O	Me				AM
‡ dv		CHO				AM
‡ dw		Me				AM
‡ dx	=O	Me				AM
‡ dy	=O	NH_2				AM
‡ dz		CHO				AM
‡ ea		Me				AM
‡ eb		Bn				M, T, V, AA, AP

1.3.2 Bis(indolyl)maleimide Analogs.[24,25]

Nixon and co-workers at Roche Bioscience used the potent, but non-selective PKC inhibitor staurosporine as a structural lead for a series of bis(indolyl)maleimides of which the most potent (**18**) showed a 350-fold selectivity for PKC over PKA.

Figure 1.3.2



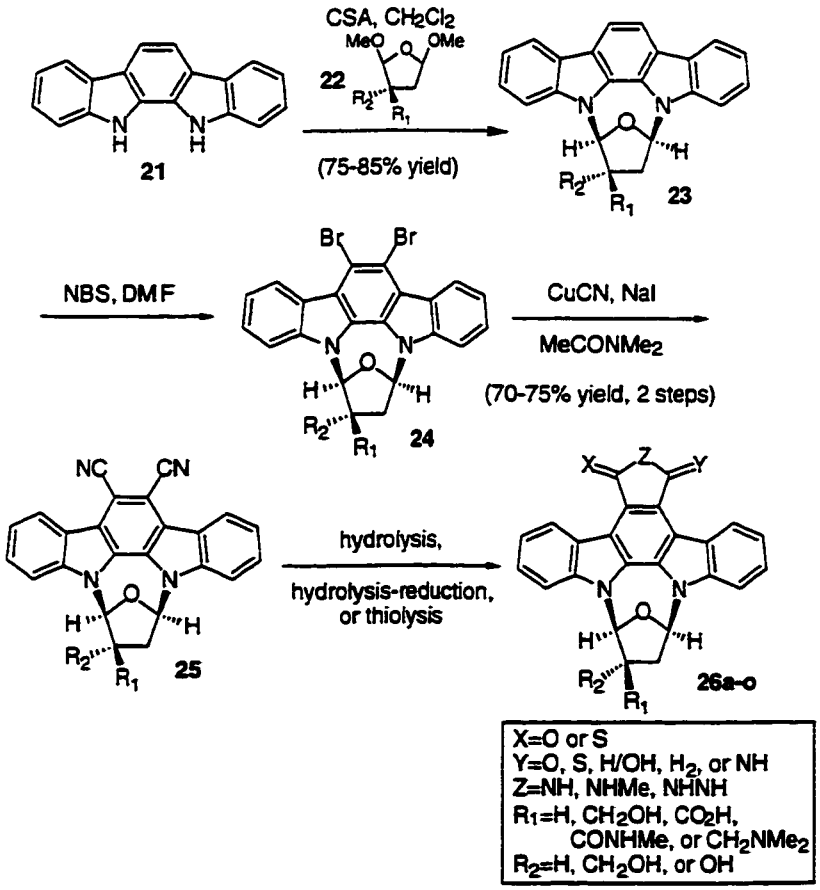
18 Ro 31-7549 **19** Ro 31-8425 **20** Ro 31-8830

The aminopropyl bis(indolyl)maleimide Ro 31-7549 (**18**, Figure 1.3.2) was found to be a potent inhibitor of rat brain PKC (IC_{50} = 75 nM), with improved selectivity for that enzyme over staurosporine (**6**). A further potency improvement was achieved by conformational restriction of the primary amine relative to the bis(indolyl)maleimide. The most potent of these inhibitors, Ro 31-8425 (**19**), showed a 350-fold selectivity for PKC over PKA, a 2400-fold selectivity for PKC over Ca^{2+}/calmodulin-dependent protein kinase, and a 160-fold selectivity for PKC over phosphorylase kinase. Ro 31-8830 (**20**), the tertiary amine of Ro 31-8425, was five-fold less active against PKC and had a selectivity profile similar to the primary amine, but also showed anti-inflammatory activity in a phorbol ester-induced model of inflammation.

1.3.3 Simplified Carbohydrate Derivatives of K252a.[26,27]

McCombie and his group of investigators at Schering-Plough prepared a number of derivatives of K252a that they tested *in vitro* for their ability to inhibit a mixture of isozymes of partially purified rat brain PKC. The condensation of indolo[2,3-a]-carbazole (**21**) with 2,5-dimethoxytetrahydrofuran derivatives gave cyclofuranosylated compounds (e.g., **23**) that were converted *via* dibromo compounds to the dinitriles (e.g., **25**). Hydrolysis, hydrolysis-reduction and thiolysis afforded imides, lactams and their thio analogs (see Scheme 1.3.1).

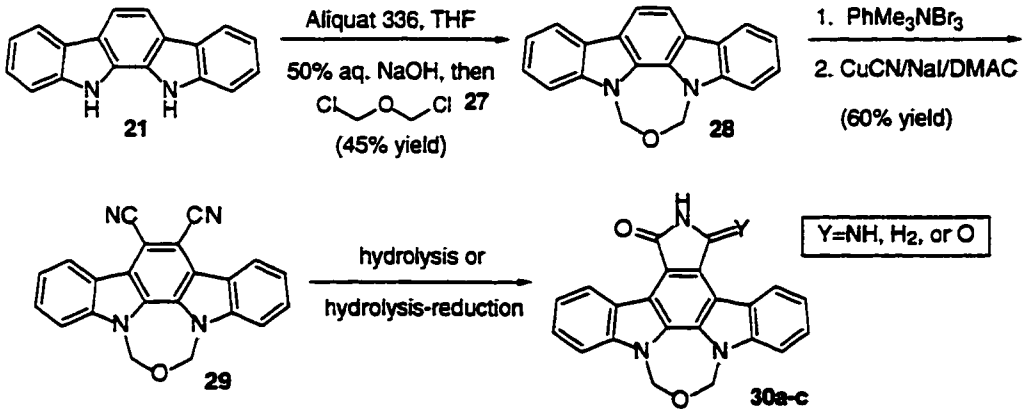
Scheme 1.3.1



In general, the McCombie group found that the high potency characteristic of the natural products (K252a (**1**): IC_{50} = 32 nM, staurosporine (**6**): IC_{50} = 3 nM) could be achieved with structurally simpler systems. In the aglycon portion of the molecule, the imides, lactams, and aminoimides were comparably potent (IC_{50} = 13-19 nM), while the thio analogs and the hydroxy lactam were somewhat less active (IC_{50} = 42-58 nM). In marked contrast, *N*-alkylation of the lactam or ring expansion to the cyclic hydrazide resulted in complete loss of activity. The carbohydrate portion of the molecule was also found to be quite tolerant of substitutions (IC_{50} = 0.5-20 nM). This suggested potential for tailoring characteristics such as cellular penetration, absorption and tissue distribution which are important in secondary assays and for *in vivo* activity.

Additional work in the McCombie labs showed that the carbohydrate was not neccessary to retain high potency, but could be replaced by a simple three-atom bridge.

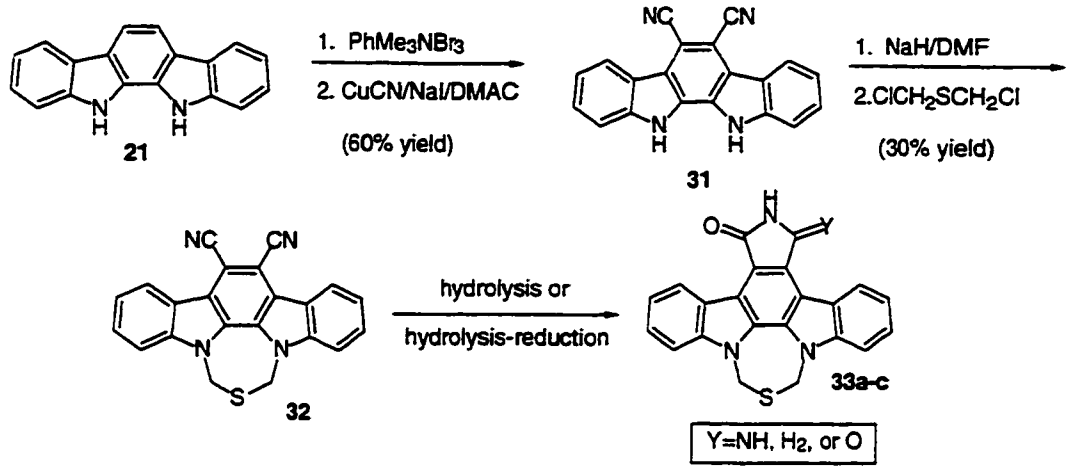
Scheme 1.3.2



Indolocarbazole **21** was parlayed into the desired cyclic ether by direct alkylation/cyclization, followed by bromination/cyanation, and hydrolysis or hydrolysis/reduction (see Scheme 1.3.2). In the case of the sulfur-linked bridge similar chemistry was applied with a different order of events. Nitrile **31** was bis-

alkylated and subjected to hydrolysis or hydrolysis/reduction to provide analogs **33a-c** (see Scheme 1.3.3). PKC activity data showed no loss in inhibitory power. Compounds such as **30a-c** gave IC_{50} values between 10-24 nM, while sulfur analogs (i.e., **33a-c**) gave similar values (IC_{50} = 7-24 nM).

Scheme 1.3.3

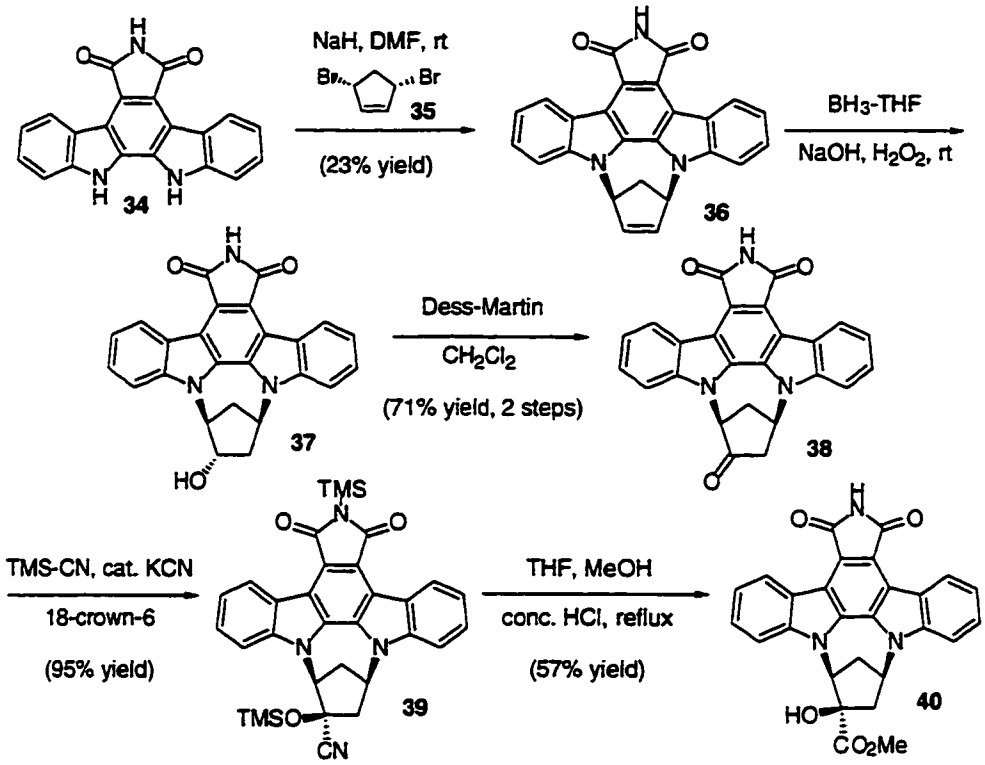


Disappointingly, in spite of the highly potent analogs prepared by the McCombie group, there was a lack of specificity for individual PKC isozymes or for PKC over other serine/threonine protein kinases.

1.3.4 Carbocyclic Carbohydrate Analogs.[28]

The synthesis of some cyclopentane-bridged indolocarbazoles, representing carbocyclic analogs of the natural product K252a, was recently achieved by Simpkins and co-workers (see Scheme 1.3.4). These analogs were shown to be potent inhibitors of PKC, and ring expansion to a staurosporine type derivative was demonstrated.

Scheme 1.3.4



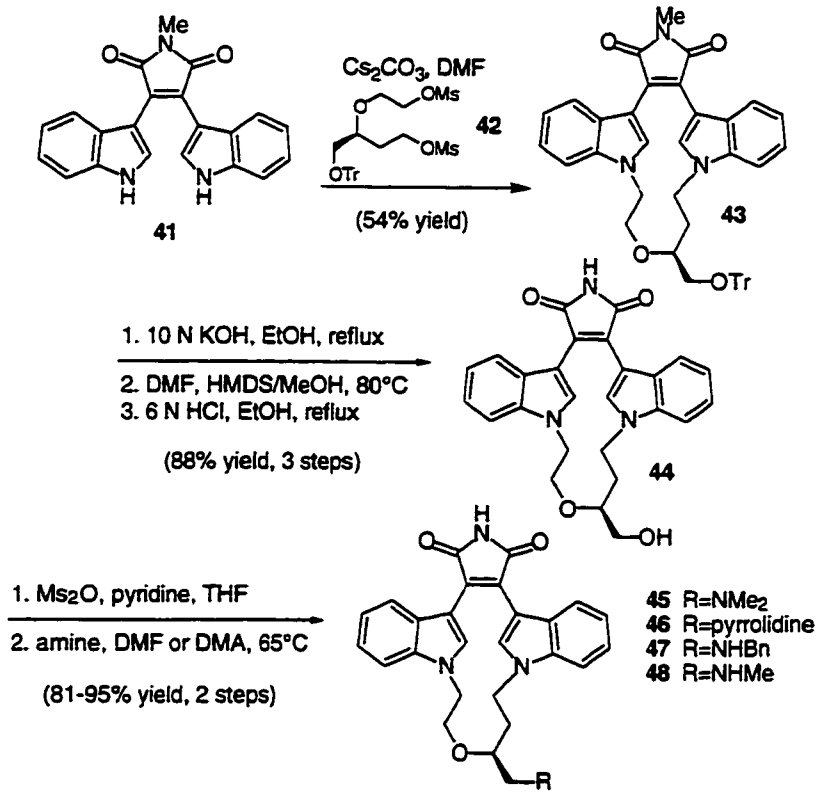
The condensation of indolocarbazole **34** with dibromide **35** was followed by hydroboration and Dess-Martin oxidation to provide ketone **38**. Preparation of the cyanohydrin followed by hydrolysis provided a carbocyclic, maleimide derivative of K252a. Compound **40** was found to be a potent inhibitor of PKC (IC_{50} = 53 nM) in *in vitro* assays, although the issues of isozyme and kinase selectivity have not been addressed.

1.3.5 Macrocyclic Bisindolylmaleimides.[29,30]

Macrocyclic bisindolylmaleimides have recently been identified by a group at Eli Lilly as competitive reversible inhibitors of PKC β_1 and β_2. Through a highly convergent and stereoselective synthesis of only 11 steps and 26% overall yield

(>98% ee), compounds are being advanced to the clinic for evaluation as a treatment of retinopathy associated with diabetic complications (see Scheme 1.3.5).

Scheme 1.3.5

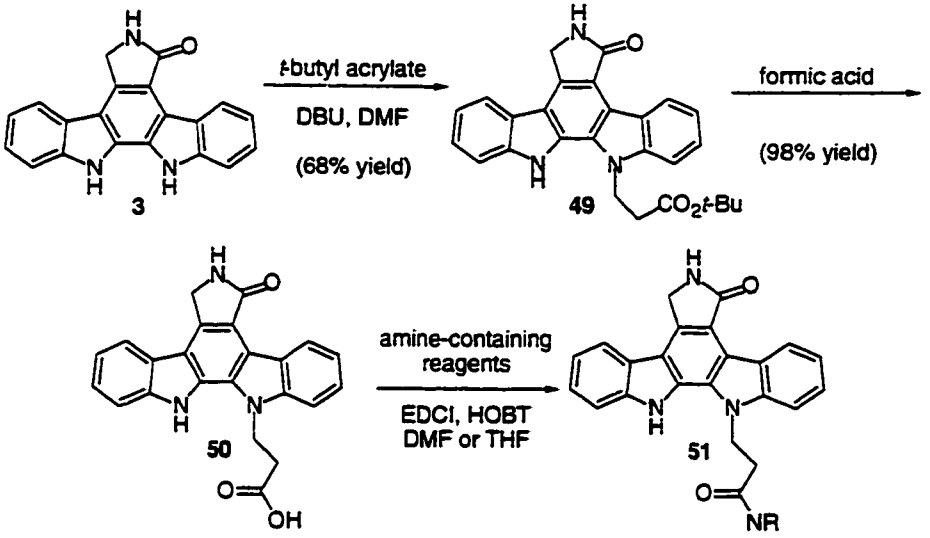


In the forward synthetic sense, bisindolylmaleimide **41** and bisalkylating agent **42** were coupled in the presence of 6 equivalents of Cs_2CO_3, followed by hydrolysis and ammonolysis. Finally, detritylation provided alcohol **44** which upon mesylation and displacement with the appropriate amines afforded compounds **45-48** which are potent and selective inhibitors of protein kinase C isoforms β_1 and β_2. The PKA/PKC IC_{50} ratio as a measure of kinase selectivity was >10,000, while PKCα/PKCβ_2 IC_{50} ratio as a measure of isozyme selectivity was >20.

1.3.6 Substituted Aglycon Variants.[31]

Lown and co-workers found that their previously prepared K252c could be used as a nucleophile in a regioselective Michael-type reaction, furnishing acid **50** in two steps (see Scheme 1.3.6). Coupling to a variety of amine-containing reagents then provided the desired analogs (**51**), which were found to inhibit PKC at high nM to low μM values and showed some selectivity for PKC over PKA. In addition, a group from Gödecke used a very similar approach to compounds with a wider variety of side chains and obtained similar results in terms of biological activity.

Scheme 1.3.6



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CHAPTER TWO

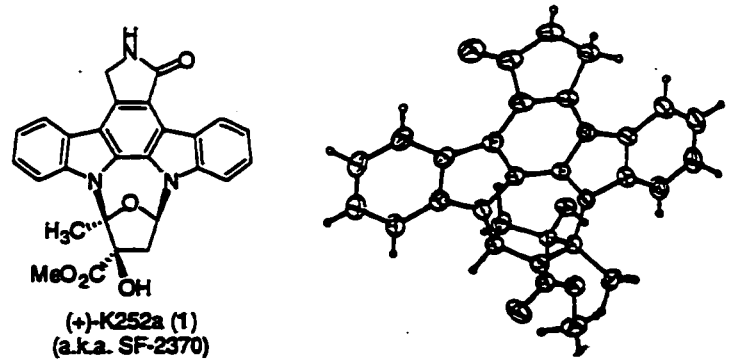
The Design and Synthesis of C(7) Alkyl Analogs of (+)-K252a and Their Application to Selective Protein Kinase Inhibition

2.1 Background.

2.1.1 Introduction.

In 1995, following our completion of the total synthesis of the furanosylated indolocarbazole (+)-K252a (**1**), we decided to explore the possibility of extending that methodology to include analogs. The synthesis developed in our group was short and efficient, two key factors which we believed would allow us to readily prepare a significant number of derivatives not available *via* manipulation of the natural material.

Figure 2.1.1



(+)-K252a (1)
(a.k.a. SF-2370)

As described in Chapter 1, K252a (**1**) was isolated in 1985 by Sezaki and co-workers and was given the name SF-2370.[1] One year later Kase independently isolated the same compound, describing not only the structure elucidation by single crystal X-ray analysis (Figure 2.1.1), but also the ability of **1** to inhibit protein kinase C (PKC) at nanomolar concentrations (IC_{50} = 32 nM).[2]

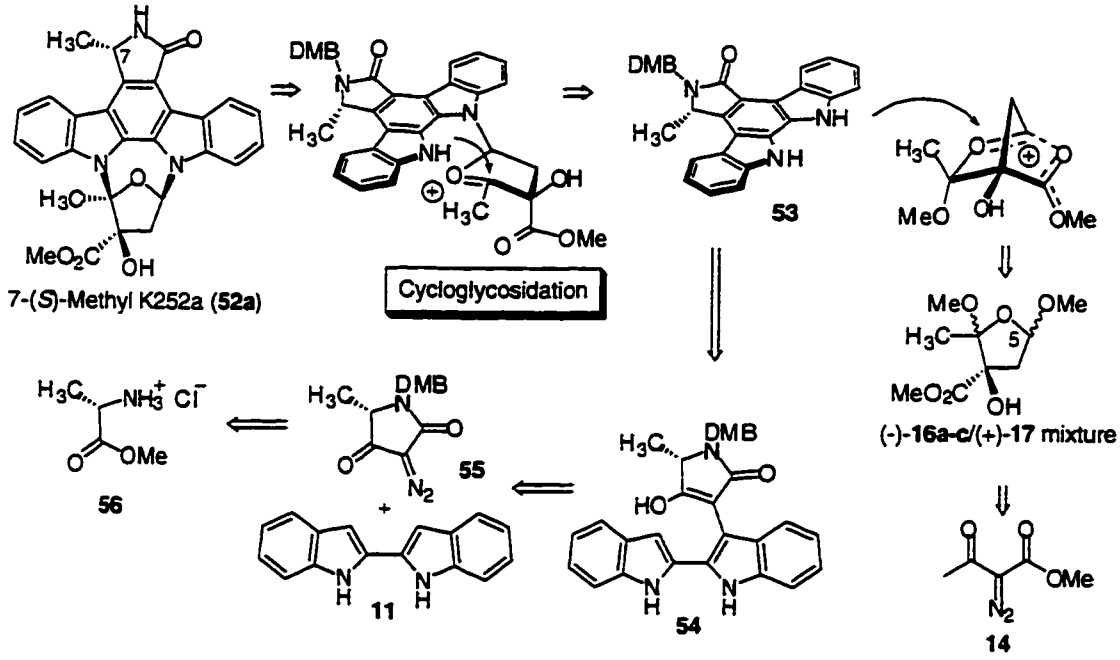
2.1.2 Evaluating Potential Routes to K252a Analogs.

The convergent synthesis of K252a, as discussed in Chapter 1, relied on a late-stage coupling of aglycon and furanose moieties to provide the fully elaborated indolocarbazole.[3] Therefore, in planning a synthesis of K252a analogs, we believed that derivatization of either the top or bottom portion of the molecule would be possible. The synthetic design of our analogs, like the synthesis of K252a itself, was based on this most simplifying disconnection.

In evaluating potential analogs, it was important to recall the work of others in the field. It had already been shown that substitution of the lactam nitrogen led to complete loss of biological activity.[4] Similarly, substituting the aromatic rings of the indolocarbazole core was also reported to be unsuccessful in providing both potent and selective inhibitors.[5] When looking at our synthesis we believed that the C(7) methylene of the lactam was a position at which derivatization would be possible *via* our synthetic pathway (see **52**, Scheme 2.1.1). In the K252a synthesis, the initial starting material for the preparation of the aglycon was a methyl ester derivative of glycine. We planned to execute the same synthetic strategy starting with a variety of different amino acids that would be readily availiable in enantiopure form. Thus, we decided first to attempt to extend our synthesis to include a simple methyl group at the C(7) postition, starting from a commercially available derivative of L-alanine.

As outlined retrosynthetically in Scheme 2.1.1, we envisioned methyl analog **52** as arising via cycloglycosidative coupling of aglycon **53** with our previously prepared mixture of carbohydrates (-)-**16a-c**/(+)-**17**. Aglycon **53** was expected to derive from cyclopropanation of 2,2'-biindole (**11**) with diazo lactam **55**, which in turn is availiable from the amino ester **56**, followed by ring opening and electrocyclization of an intermediate such as enol **54**. As in our K252a synthesis, the carbohydrate mixture (-)-**16a-c**/(+)-**17** derives from methyl acetoacetate **14**.

Scheme 2.1.1



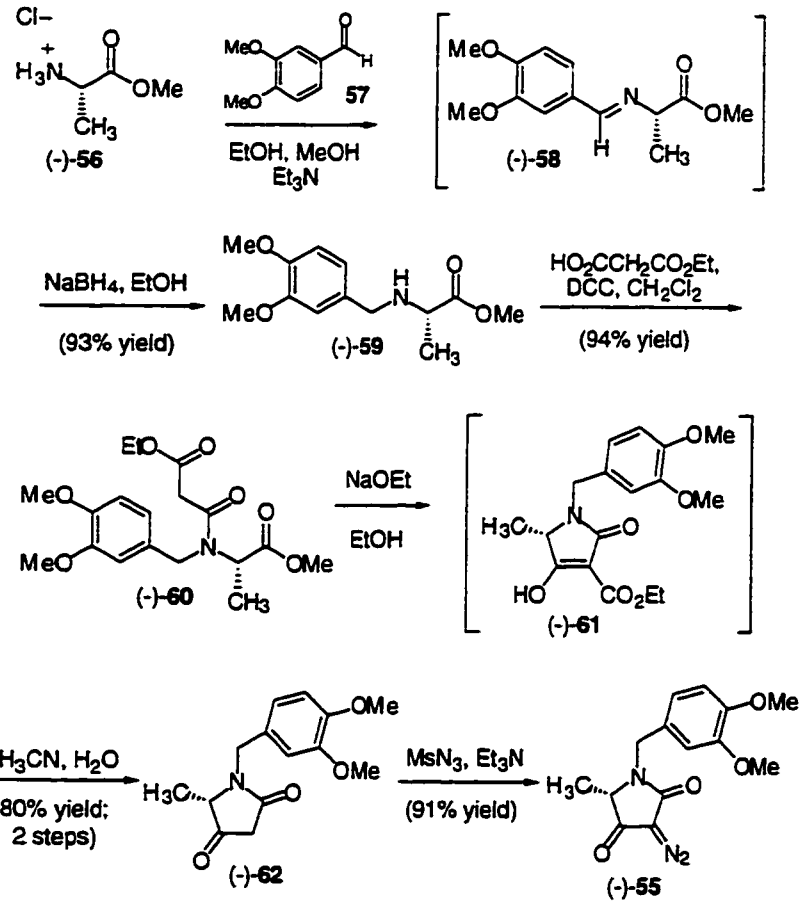
2.2 Synthesis of Methyl K252a Analogs.

2.2.1 Preparation of the Aglycon.

In the synthetic sense, L-(-)-alanine methyl ester hydrochloride (**56**), (see Scheme 2.2.1) was monoprotected as the dimethoxybenzyl derivative via

reductive amination with 3,4-dimethoxybenzaldehyde (**57**) to provide the imine as a white powder which was used directly in a reduction with $NaBH_4$ to provide the protected amine in 93% yield.[6] The protected amine (-)-**59** was then coupled to ethyl hydrogen malonate to furnish the cyclization precursor (-)-**60** in 94% yield.

Scheme 2.2.1

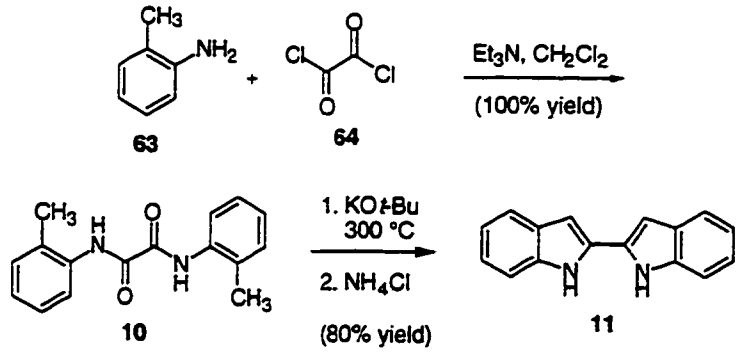


At this stage, we became concerned that in order to effect the planned Dieckmann cyclization of (-)-**60** we would need to use strongly basic conditions which might epimerize the stereogenic center. Following the literature precedent of Klutchko, the ester ((-)-**60**) was briefly warmed to reflux (5 min) in a solution of NaOEt in EtOH (< 1.0 equivalent).[7] The resultant lactam ((-)-**61**), without further purification, was subjected to decarboethoxylation in a refluxing mixture of CH_3CN

and H_2O to provide ketolactam (-)-**62** in 80% yield over two steps. Diazo transfer from mesyl azide with Et_3N in CH_3CN led to the desired diazo lactam (-)-**55** in 91% yield.[8] This sequence allowed us to prepare 25 gram quantities of diazo lactam in good yield with only one chromatographic purification.

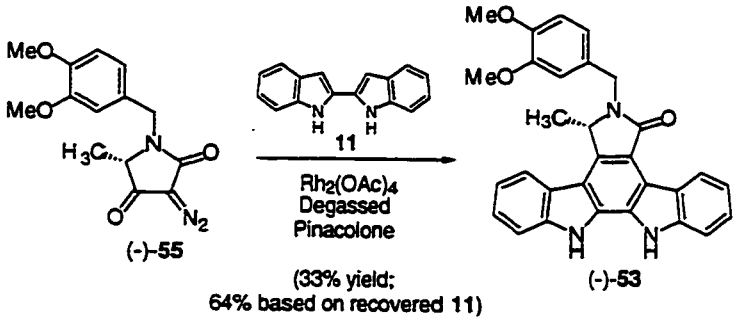
Having assembled (-)-**55** we turned to its coupling partner 2,2'-biindole (**11**), which is readily accessible via the method of Bergman from commercially available *o*-toluidine (**63**) and oxalyl chloride (**64**). Thus, coupling of **63** and **64** furnished oxaltoluidide **10**, which was transformed to the desired biindole via a double Madelung cyclization upon treatment with KO*t*Bu and heating to 300°C in a Wood's Metal bath (see Scheme 2.2.2).[9]

Scheme 2.2.2



The desired aglycon (-)-**53** was then prepared by coupling equimolar amounts of diazo lactam (-)-**55** and 2,2'-biindole (**11**) in the presence of 1 mol % $Rh_2(OAc)_4$. Key to the success of this reaction was degassing the pinacolone solvent for 2 hours prior to warming the mixture to reflux. This reaction was performed on a multigram scale and unreacted **11** was recovered and recycled, providing the aglycon in 64% yield based on recovered biindole (see Scheme 2.2.3).

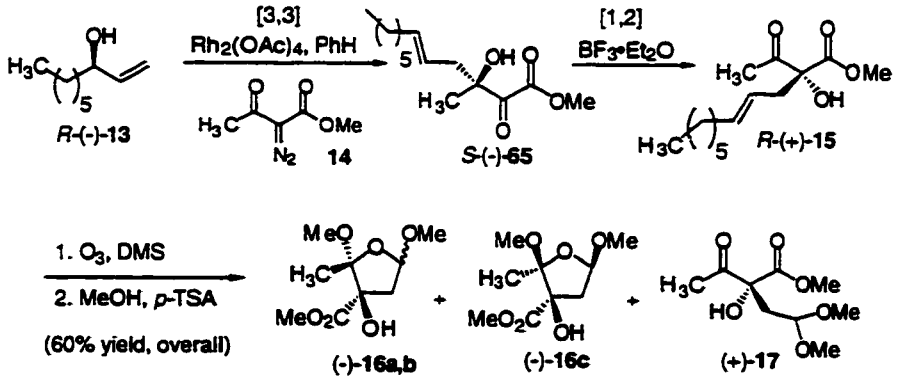
Scheme 2.2.3



2.2.2 Preparation of the Carbohydrate.

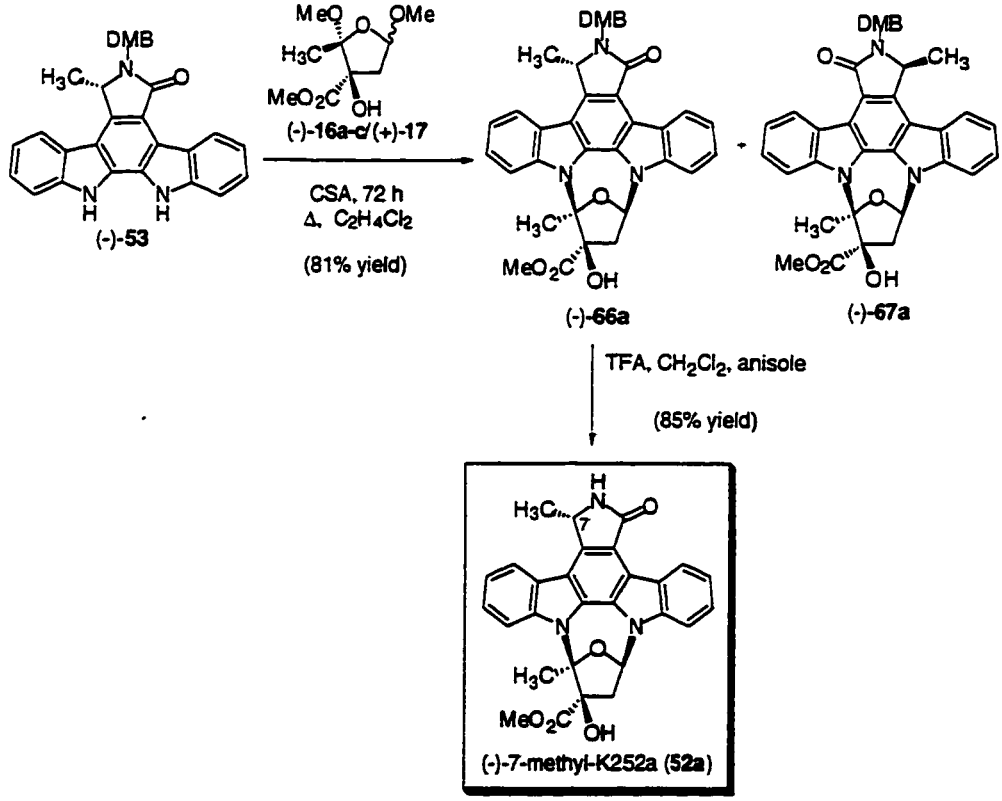
The furanose unit (-)-**16a-c**/(+)-**17** was prepared using the sequence developed in our synthesis of **1**, wherein *R*-(-)-1-nonen-3-ol (**13**)[10] served as the source of asymmetry in a tandem [3,3]/[1,2] rearrangement (i.e., **13**→**65**→**15**; Scheme 2.2.4). Reductive ozonolysis and cyclization of the derived β-keto ester (+)-**15** furnished the illustrated cycloglycosidation substrate (-)-**16a-c**/(+)-**17** as a mixture of alcohols in 60% overall yield (92% ee) for the four steps.

Scheme 2.2.4



In accord with our previous approach to furanosylated indolocarbazoles, aglycon (-)-**53** was treated with the carbohydrate mixture (-)-**16a-c**/(+)-**17** over a 24 hour period in the presence of a catalytic amount of CSA. Continued reflux over 48 additional hours in 1,2-dichloroethane gave the expected 2:1 mixture of regioisomers (-)-**66a** and (-)-**67a** in 81% yield.[3,11] The mixture of regioisomers was separated by HPLC, and the synthesis was completed by deprotection of amide (-)-**66**. Removal of the dimethoxybenzyl group was achieved with TFA/CH_2Cl_2 in the presence of a cation scavenger, affording the desired analog (-)-**52a** in 85% yield.[12,13] The epimeric analog (+)-7-*(R)*-methyl K252a (**52b**) was also prepared using the above method starting from D-(+)-alanine methyl ester hydrochloride (**56**).

Scheme 2.2.5



2.2.3 Proof of Stereochemistry.

With an approach to analogs firmly established, the one synthetic question remaining to be answered was whether the stereocenter at C(7) had been epimerized. Although we had taken steps to minimize the potential for epimerization and were able to detect only trace quantities of other diastereomers in the crude NMR of the glycosidation mixture, we set out to quantify the extent of epimerization, if indeed any had occurred during the sequence. Since we believed the step most likely to result in epimerization was the Dieckmann cyclization, we first focused our attention on decarboxylated product (-)-**62**. Reduction of ketolactam (-)-**62** to a 1.5:1 mixture of diastereomeric alcohols was achieved with $NaBH_4$ as outlined in Scheme 2.2.6, followed by comparison of the derived mixture of Mosher esters (-)-**69a,b** to a sample prepared from the reduction and esterification of material prepared from (+)-**62**.[14] ^{19}F NMR as shown in Figure 2.2.1 established an ee of 90% for this material.

Scheme 2.2.6

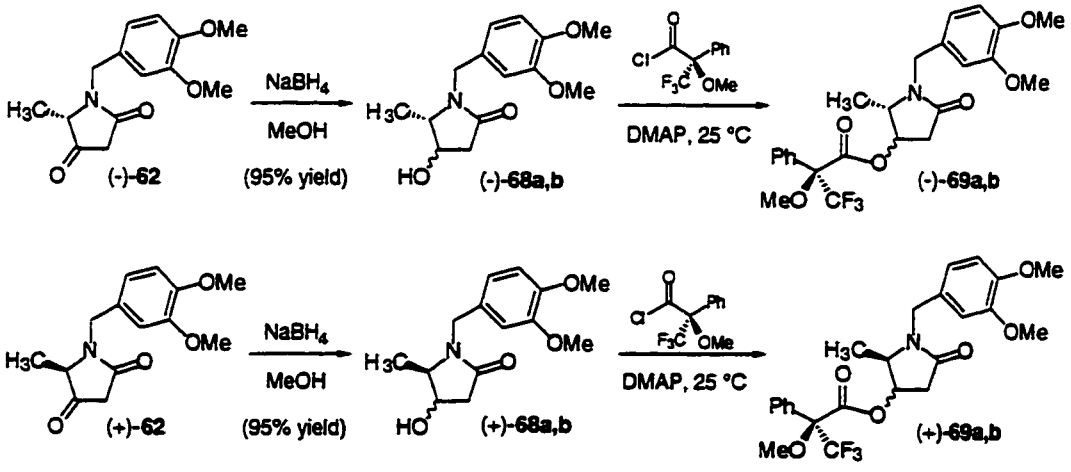
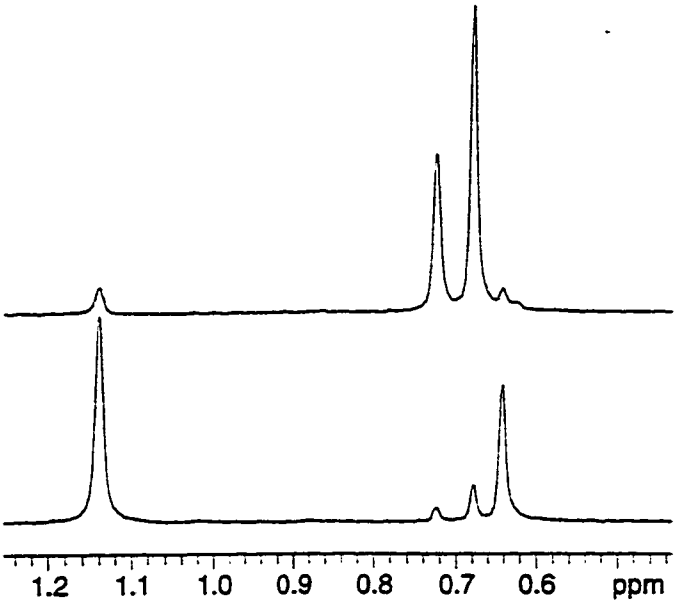


Figure 2.2.1



^{19}F NMR (490 MHz, benzene-d_6) comparison

of Mosher esters **69a,b** derived from:

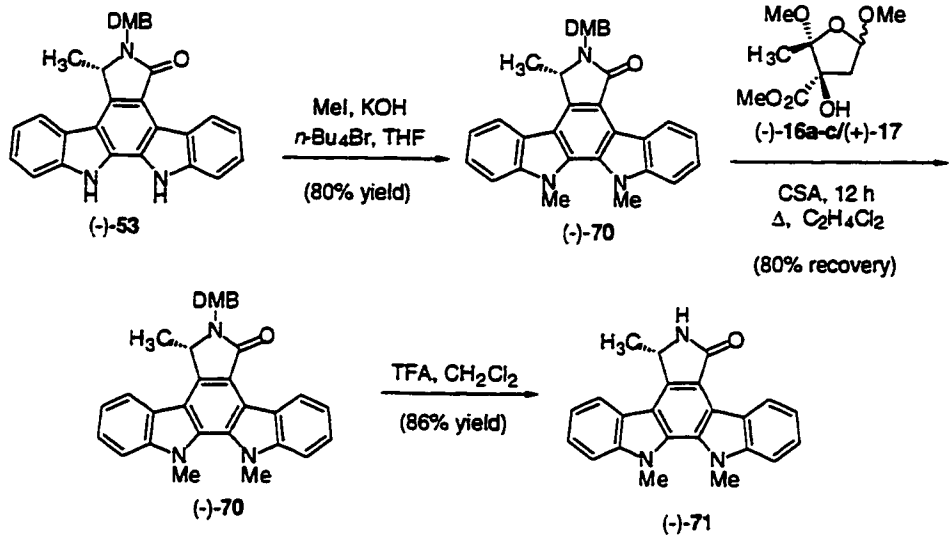
top; (-)-**68a,b**

bottom; (+)-**68a,b**

Observation of only trace quantities of other diastereomers in the crude NMR of the glycosidation mixture also supported the theory that the C(7) position of aglycons (-)- and (+)-**53** and analogs (-)- and (+)-**52** had not been further epimerized following the cyclization. Attempts to further prove the diastereomeric purity of our analogs led to derivatization of the aglycon (-)-**53** by the following route. Bis-methylation of the indole nitrogens under phase-transfer alkylation conditions provided (-)-**70** in good yield. Aglycon (-)-**70** was now effectively protected against reaction with the carbohydrate mixture, but was nonetheless subjected to the conditions of the glycosidation followed by deprotection to afford amide (-)-**71**. Chiral HPLC (Chiracel OD column) of (-), (+), and (±)-**71** established ee values

ranging from 88-90%, thus confirming that only minor epimerization had occured at the C(7) position through the entire synthetic pathway.

Scheme 2.2.7



2.2.4 Evaluation of Biological Activity.

With two methyl derivatized K252a analogs in hand we investigated their biological activity at the Yale University Boyer School for Molecular Medicine with the aid of Dr. Vincent Marchesi, whose group had developed a simple, rapid, and inexpensive way to analyze the effects of individual compounds on the different stages of mammalian cell division. In this assay, Chinese Hamster ovary cells (CHOC) trapped in prophase by nocodazole, can be harvested in high yield and cultured under conditions in which essentially 100% of the cells pass from prophase through metaphase, anaphase, and telophase within a 75-minute period. By adding compounds to the media at different points, the effects on the mechanisms that regulate each stage of mitotic progression can be assessed.

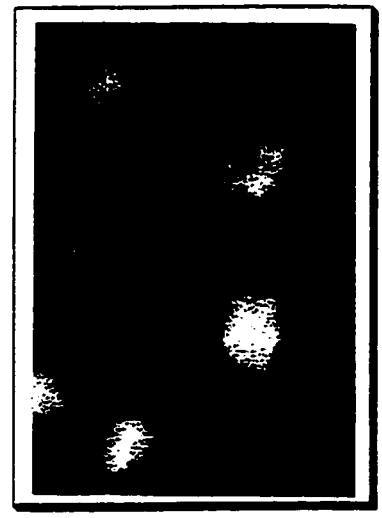
Recent experiments in the Marchesi lab indicate that inhibitors of protein kinases (e.g., K252a) and phosphotases have striking and remarkably specific

effects on different stages of mitosis.[15] The ability of K252a (**1**) to cause mitosing Chinese Hamster Ovary cells to skip anaphase and undergo mitotic arrest provides an opportunity to investigate the kinases that are active during this phase of the cell cycle. When the analogs described above, (+)-7-(*R*)- and (-)-7-(*S*)-methyl K252a (**52a,b**) were subjected to the CHOC assay, they were found to block mitosis at concentrations between 0.5 and 1.0 μM. This can be compared to a value of 0.25 μM for both synthetic and natural K252a.

Photomicrographs of the cells show distinct differences.[16] The untreated cells appear in sets of distinct pairs, and the nuclei are clearly dividing in preparation for splitting of the cell into two daughter cells. In the cells which have been treated with K252a, however, the nuclei of the cells look diffuse and it is clear that no cell division is imminent (see Figure 2.2.2).

Figure 2.2.2

CHO cells synchronized at prophase and incubated at 37°C for 40 min in growth medium



CHO cells synchronized at prophase and incubated at 37°C for 40 min with 1.0 μM (-)-**52a** or (+)-**52b**

With the information in hand that C(7) derivatives of K252a still affect mitosis in cells, and therefore still retained some of their biological activity, we decided to determine whether or not any of these compounds might be a specific kinase inhibitor.

Given that the Marchesi group's assay was not suited to evaluating selectivity for individual kinases, we turned to a group of biochemists at Kinetix Pharmaceuticals who were also interested in selective inhibition of protein kinases, specifically of members of the Src family of protein kinases.[17] After testing K252a, (-)-7-(*S*)-methyl K252a (**52a**) and (+)-7-(*R*)-methyl K252a (**52b**) against a panel of five protein tyrosine kinases (Lck, Fyn, ZAP-70, Syk, and Itk), it was found that all three compounds were kinase inhibitors (see Table 2.2.1).[18]

Table 2.2.1

Kinase	IC_{50} (μM)		
	(+)-**1**	(-)-**52a**	(+)-**52b**
Lck	1.25	2.70	5.22
Fyn	4.59	6.22	10.71
ZAP-70	0.32	4.29	10.00
Syk	0.04	0.19	3.55
Itk	0.35	5.07	2.22

Of the three compounds tested, K252a (**1**) was found to be the most potent inhibitor of protein tyrosine kinases. As expected, K252a was also quite unselective in its inhibition of this family of kinases. Analog (+)-**52b** was not a very potent inhibitor with IC_{50} values in the low μM range. Analog (-)-**52a**, however, was more potent and also showed a better selectivity profile than either (+)-**1** or (+)-**52b**. With these results in hand, we speculated about whether varying the alkyl group at the C(7) position would affect the observed selectivity. Based on the more promising

profile of (-)-**52a**, we decided to focus our efforts on the *S* or L-series of amino acids.

To this end, we prepared a C(7)-*S*-benzyl analog with the goal of establishing that our chemistry could be extended to include larger and more varied alkyl groups, as well as to determine if an aromatic moiety would have any effect on binding. For example, if the ATP binding site of a particular kinase contained phenylalanine, tyrosine, or tryptophan residues, a favorable interaction with the benzyl group of our analog might be possible.

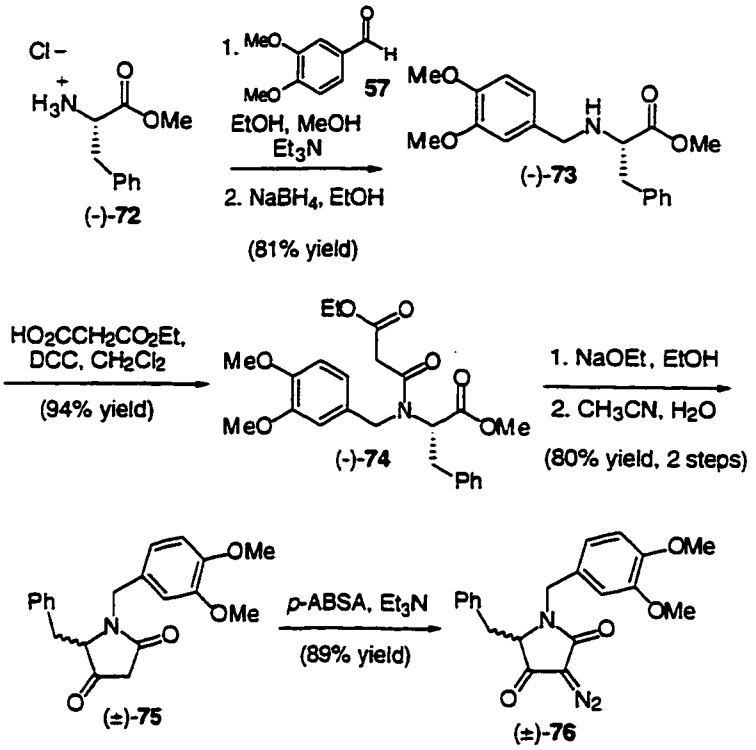
2.3 Synthesis of Benzyl K252a Analogs.

2.3.1 Preparation of C(7) Benzyl Analogs.

For the preparation of benzyl analogs, L-(-)-phenylalanine methyl ester hydrochloride (**72**, Scheme 2.3.1) was protected as the dimethoxybenzyl derivative giving rise to amine (-)-**73** in 81% yield. Coupling to ethyl hydrogen malonate then afforded ester (-)-**74** (94% yield). Dieckmann cyclization was effected under basic conditions (NaOEt/EtOH) and followed, without further purification, by decarboethoxylation in a refluxing mixture of CH_3CN and H_2O furnishing ketolactam (±)-**75** in 80% yield over two steps. Disappointingly, in the benzyl case the optical rotation of (±)-**75** was observed to be zero, and it was not possible to effect the Dieckmann cyclization without concommitant epimerization of the C(7) stereocenter. In spite of our inability to maintain stereochemical integrity, we decided to continue to pursue the C(7) benzyl derivatives with the hope of separating a mixture of diastereomers later in the synthesis. To this end, diazo transfer from *p*-ABSA, a

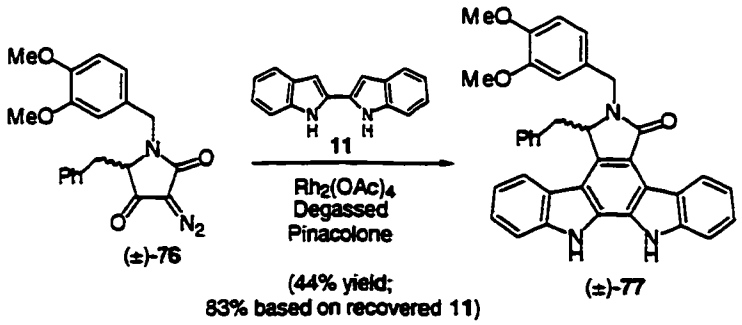
diazo transfer reagent which is easily prepared[19] and much less shock sensitive than MsN_3, afforded the desired diazo lactam (±)-**76** in 89% yield.

Scheme 2.3.1



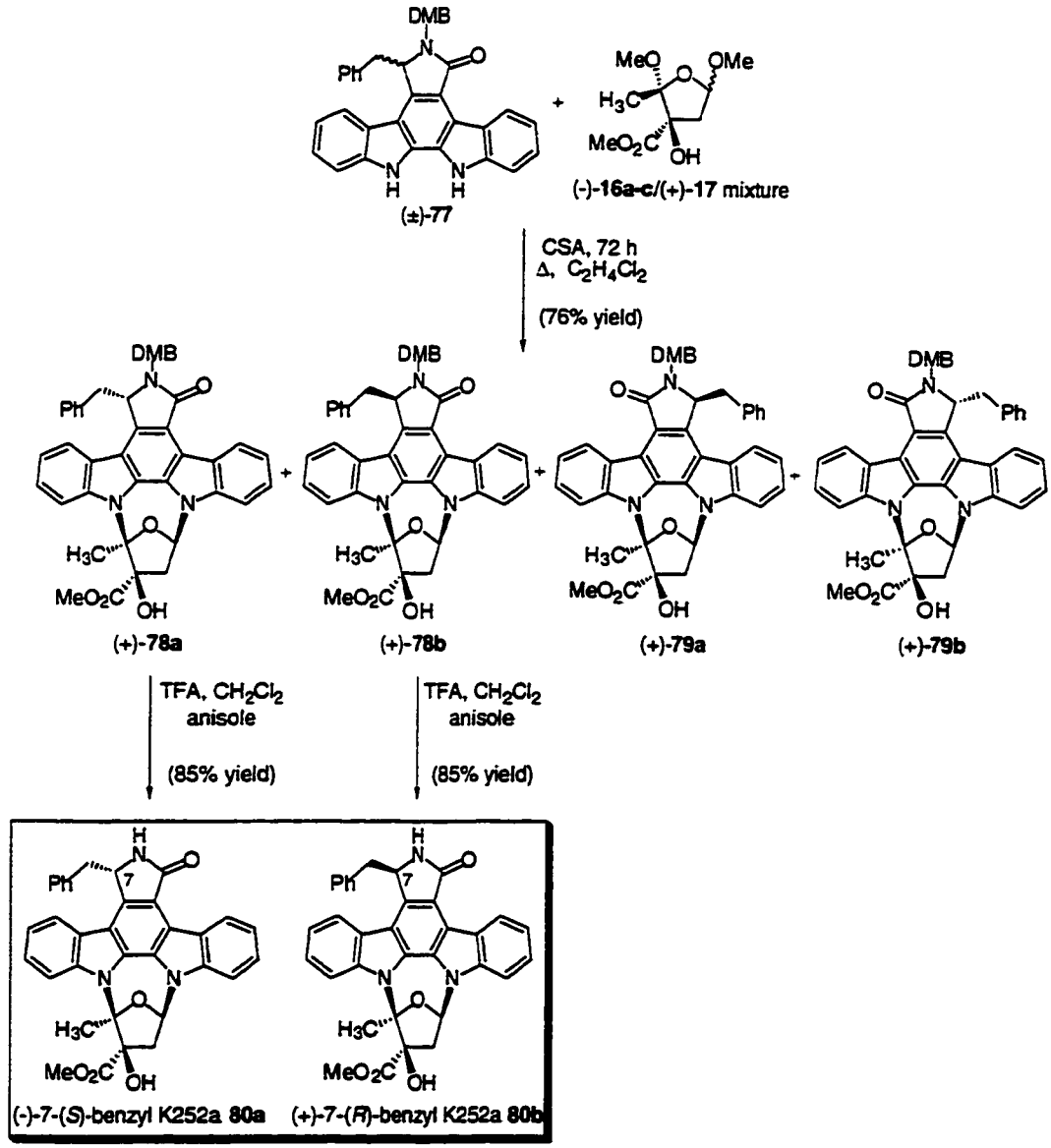
Aglycon (±)-**77** was produced (83% yield based on recovered **11**) by treatment of a 1:1 mixture of diazo lactam (±)-**76** and 2,2'-biindole (**11**) with 1 mol % $Rh_2(OAc)_4$ (see Scheme 2.3.2).

Scheme 2.3.2



As before, aglycon (±)-**77** was subjected to cycloglycosidation with the carbohydrate mixture (-)-**16a-c**/(+)-**17** in the presence of a catalytic amount of CSA in refluxing 1,2-dichloroethane, to give an expected 2:2:1:1 mixture of regioisomers and diastereomers (+)-**78a,b**, and (+)-**79a,b** in 76% yield (see Scheme 2.3.3).

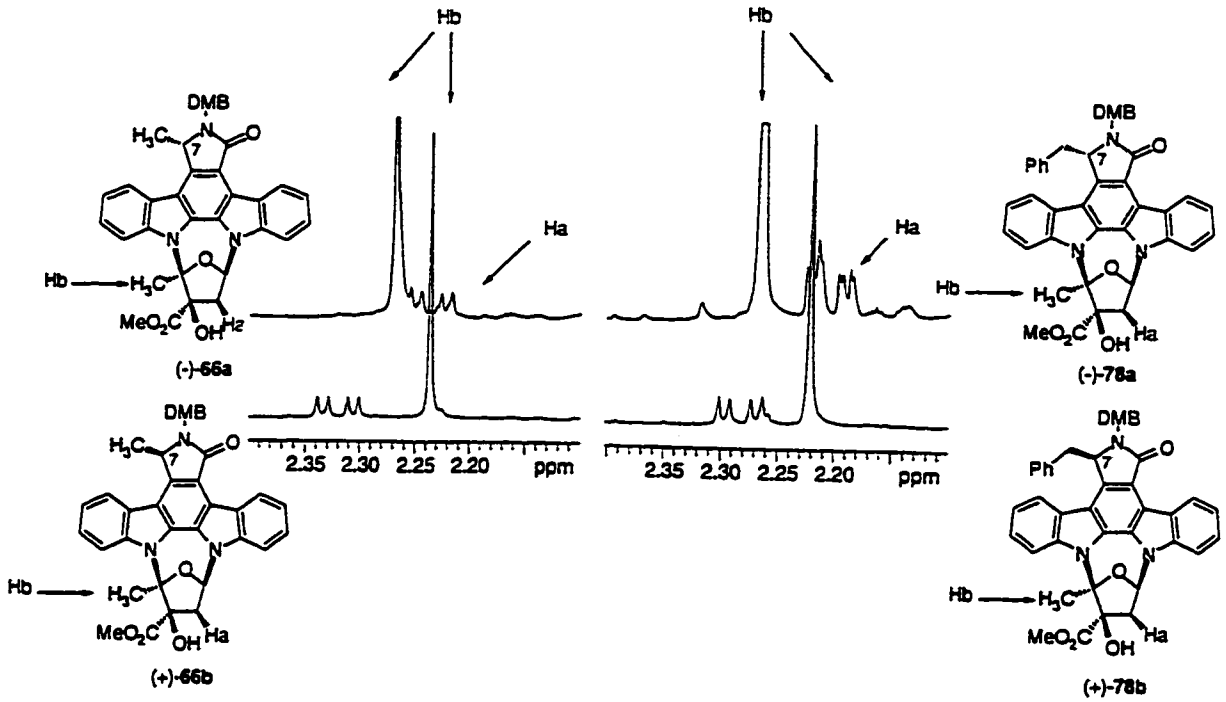
Scheme 2.3.3



The relative stereochemistry of these four compounds was determined by comparison to ^{1}H NMR spectra of (-)-**66a** and (+)-**66b**. The chemical shift of proton

H_a attached to the sugar methylene is significantly shifted based on the stereochemistry at the C(7) position (see Figure 2.3.1). Chromatographic separation of the mixture, followed by deprotection of amides (+)-**78a** and (+)-**78b** with TFA/CH_2Cl_2 in the presence of anisole afforded the desired analogs (-)-**80a,b** in 85% yield.

Figure 2.3.1



^{1}H NMR (500 MHz, acetone-d_6) comparison:

left top; (-)-**66a** right top; (+)-**78a**

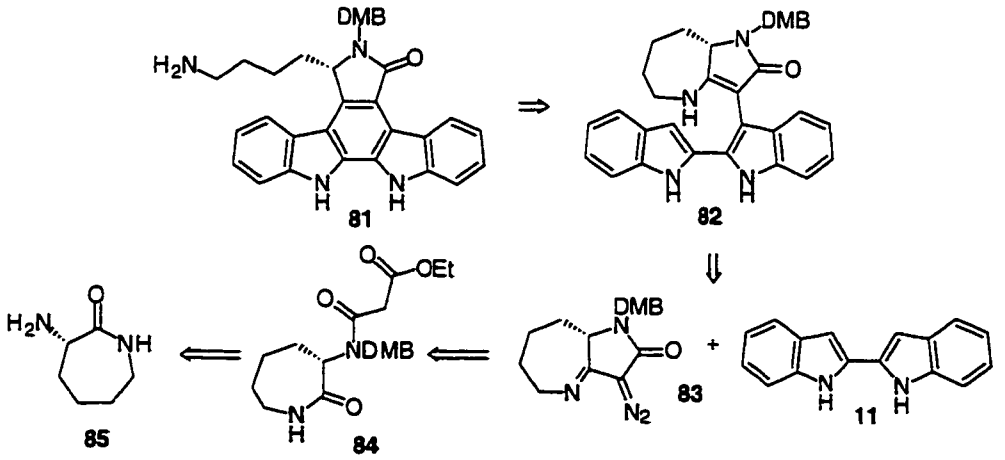
left bottom; (+)-**66b** right bottom; (+)-**78b**

2.4 Synthesis of a K252a Derivative Containing a Linker.

2.4.1 Synthesis of a K252a Analog With a Linker: First-Generation Approach.

Our success in preparing both methyl and benzyl K252a analogs led us to an interest in the use of K252a as a biological probe. It was thought that if a K252a derivative was prepared with a long alkyl linker it could be coupled to a resin, fluorescent label, or biotin and be used by a collaborating group of biochemists to discover more about the manner in which K252a binds its kinase targets. In our first approach we believed that a terminal amine or alcohol would provide a suitable site for derivatization. In an attempt to avoid complicated protecting group chemistry we decided to employ an approach where the terminal amine was masked as the amide of a lactam ring.

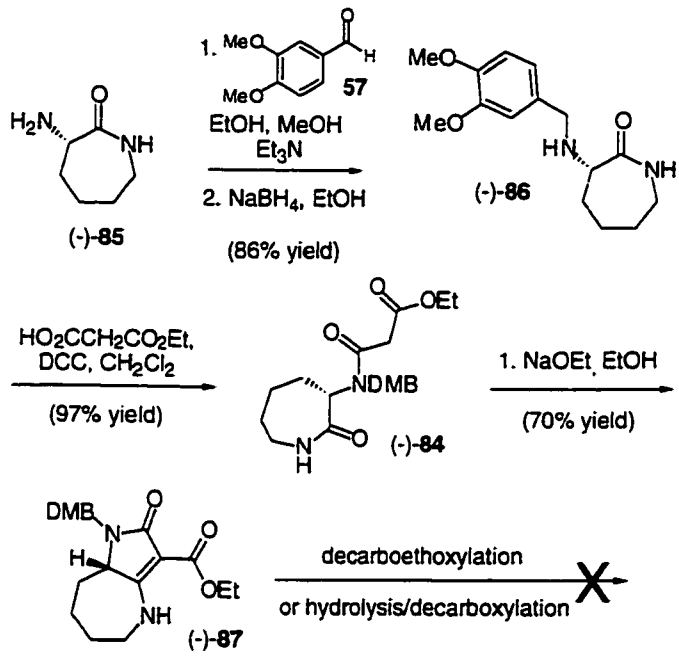
Scheme 2.4.1



Retrosynthetically, when looking at the aglycon, we believed the amine could be unveiled during the coupling of diazo lactam and biindole, where instead of a loss of water, the amine side chain could act as the leaving group (see Scheme 2.4.1).

Aglycon **81** was envisioned to arise through electrocyclization and loss of amine from an intermediate such as **82**. Enamine **82** would come from the coupling of diazo imine **83** and 2,2'-biindole (**11**), while diazo compound **83** itself could derive from cyclization of ester **84**. The ester, in turn, was expected to arise from commercially available caprolactam derivative **85**.

Scheme 2.4.2

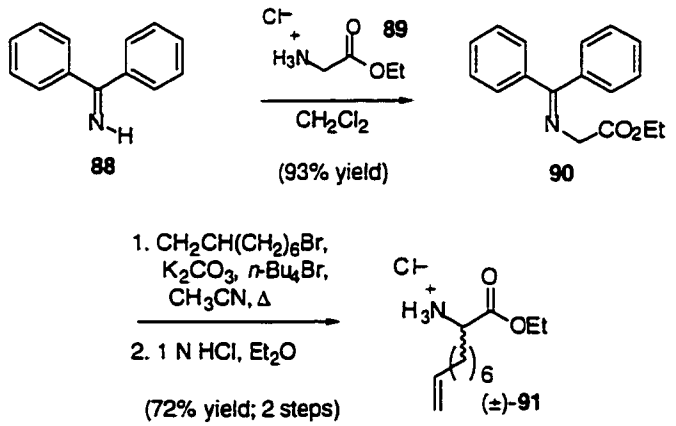


In the forward sense, L-(-)-α-amino-ε-caprolactam (**85**, Scheme 2.4.2) was protected as the dimethoxybenzyl derivative in 86% yield. The protected amine (-)-**86** was then activated by DCC and coupled to ethyl hydrogen malonate furnishing ester (-)-**84** in 97% yield. Ester (-)-**84** was warmed to reflux in a solution of NaOEt in EtOH for 5 minutes producing the desired lactam (-)-**87** in 70% yield. Unfortunately, all attempts to decarboethoxylate (-)-**87** or hydrolyze and decarboxylate the corresponding acid failed in our hands and we were forced to explore a new approach to a linked analog.

2.4.2 Synthesis of a K252a Analog With a Linker: Second-Generation Approach.

In our second approach we modified our plan such that the terminal amine or alcohol would be unveiled at a later stage in the synthesis. Again, attempting to avoid protecting group chemistry, we employed an approach where the terminal amine was masked as an olefin. An appropriate amino acid derivative ((±)-**91**) was prepared using the method of O'Donnell,[20] where commercially available benzophenone imine (**88**) undergoes an imine exchange reaction with glycine ethyl ester hydrochloride (**89**) to produce glycine enolate equivalent **90**.[21] Ester **90** was alkylated under phase-transfer conditions, followed by selective hydrolysis of the imine to afford the desired amino ester (±)-**91** (see Scheme 2.4.3).

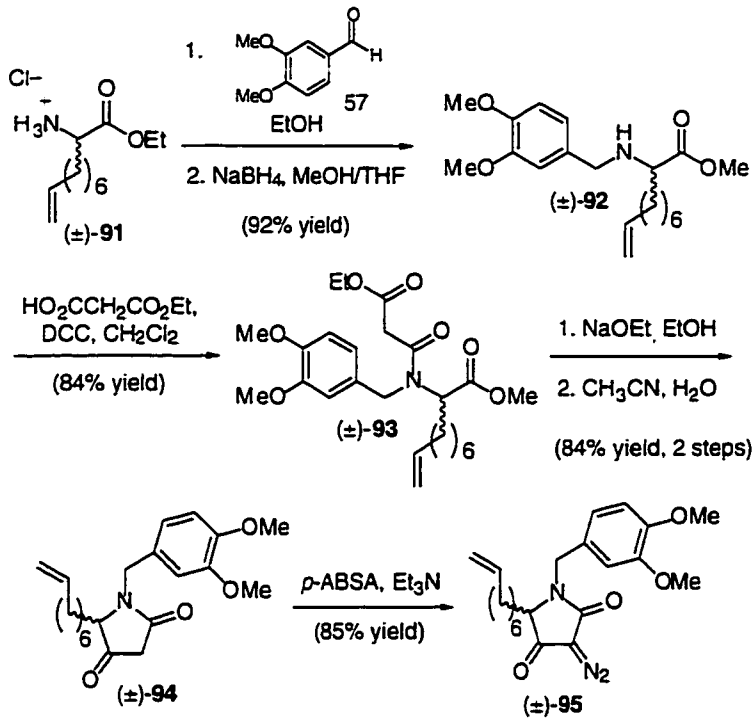
Scheme 2.4.3



Rather than try to perform the alkylation reaction in an enantioselective manner or use classical resolution chemistry to provide enantiopure **91**, we chose to prepare racemic aglycon from (±)-**91** and use the enantioenriched carbohydrate mixture to resolve the aglycon, as with the benzyl derivatives. To this end, protection of (±)-**91** as the dimethoxybenzyl derivative (±)-**92** followed by coupling to ethyl hydrogen malonate furnished the Dieckmann cyclization precursor (±)-**93** in 77%

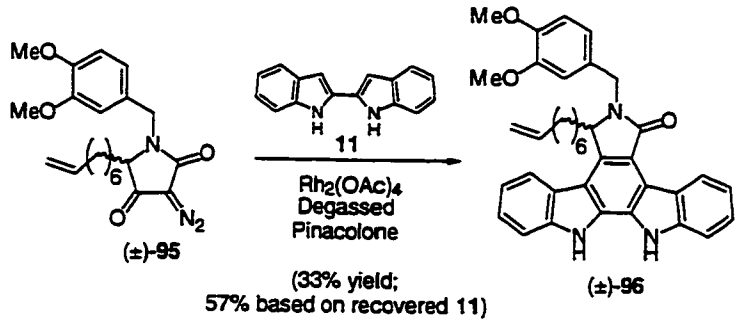
yield for the two steps. Ester (±)-**93** was warmed to reflux in a solution of NaOEt in EtOH followed by decarboethoxylation (CH_3CN/H_2O, Δ) to provide ketolactam (±)-**94** in 84% yield over two steps. Finally, diazo transfer from *p*-ABSA produced the desired diazo lactam (±)-**95** in 85% yield.

Scheme 2.4.4



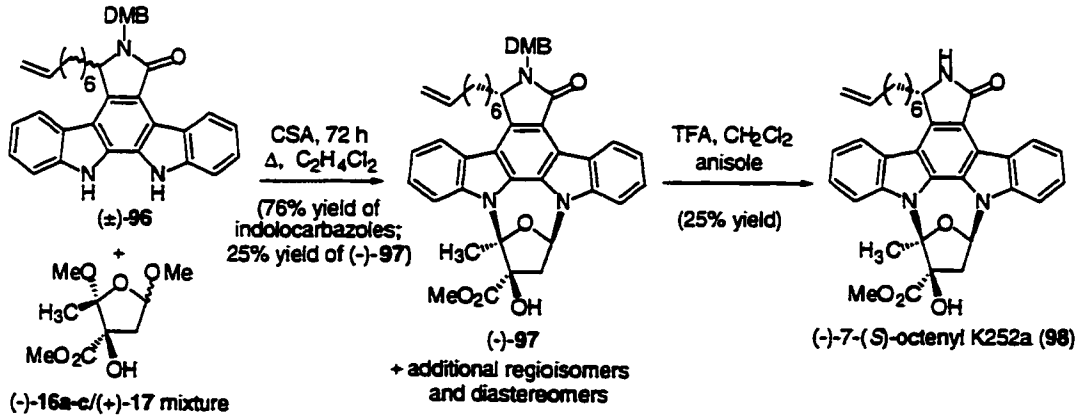
As before, racemic aglycon (±)-**96** was prepared by coupling equimolar amounts of diazo lactam (±)-**95** with 2,2'-biindole (**11**) in the presence of 1 mol % $Rh_2(OAc)_4$ providing octenyl aglycon (±)-**96** (57% yield based on recovered **11**, see Scheme 2.4.5).

Scheme 2.4.5



In accord with our previous approach to benzyl K252a analogs, aglycon (±)-**96** was subjected to cycloglycosidation with the carbohydrate mixture (-)-**16a-c**/(+)-**17** under our standard conditions, providing the expected 2:2:1:1 mixture of regioisomers and diastereomers, the stereochemistry of which could again be assigned by comparison of NMR spectra. The mixture was separated by HPLC, followed by deprotection of amide (-)-**97** (TFA/CH_2Cl_2, anisole) affording C(7) octenyl analog (-)-**98** in 25% yield.

Scheme 2.4.6



2.4.3 Evaluation of Biological Activity.

At this stage we again turned to the biochemists at Kinetix to test (-)-7-(*S*)-benzyl K252a (**80a**) and (-)-7-(*S*)-octenyl K252a (**98**) against the same panel of protein tyrosine kinases (Lck, Fyn, ZAP-70, Syk, and Itk). Disappointingly, it was found that neither compound was an inhibitor of this group of kinases (see Table 2.4.1).

Table 2.4.1

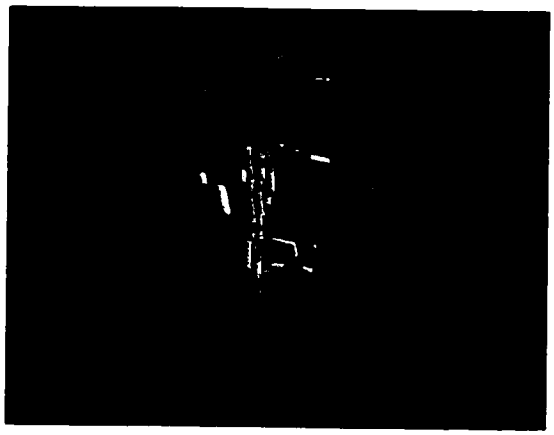
Kinase	IC_{50} (μM)		
	(+)-**1**	(-)-**80a**	(-)-**98**
Lck	1.25	>9	>9
Fyn	4.59	>9	>9
ZAP-70	0.32	>9	>9
Syk	0.04	6.77	>9
Itk	0.35	>9	>9

The lack of kinase activity observed for (-)-**80a** and (-)-**98** was attributed to the size of the alkyl group. The small methyl group at this position was tolerable, but a much larger benzyl or octenyl substituent resulted in a complete loss of activity.

At this time, X-ray crystal structure data of another indolocarbazole, staurosporine (**6**), bound to protein kinases PKA and cyclin-dependent protein kinase 2 (CDK2) became available in the literature.[22] Careful study of the X-ray data led us to believe that there was little free space around the aglycon portion of staurosporine and in particular, the C(7) position appeared to be blocked from both sides (see Figure 2.4.1). It is clear that the side chains of the valine and phenylalanine residues in the ATP binding site of CDK2 are blocking both sides of

the C(7) carbon. Shown here in white, bulky isopropyl and benzyl groups are close in space to C(7) of staurosporine. If we assume a similar mode of binding for K252a, steric hindrance between the side chains and the C(7) alkyl group would be detrimental to binding. Based on these structural reports and our loss of kinase activity, we were forced to abandon our plan to use K252a as a biological probe linked at the C(7) position.

Figure 2.4.1



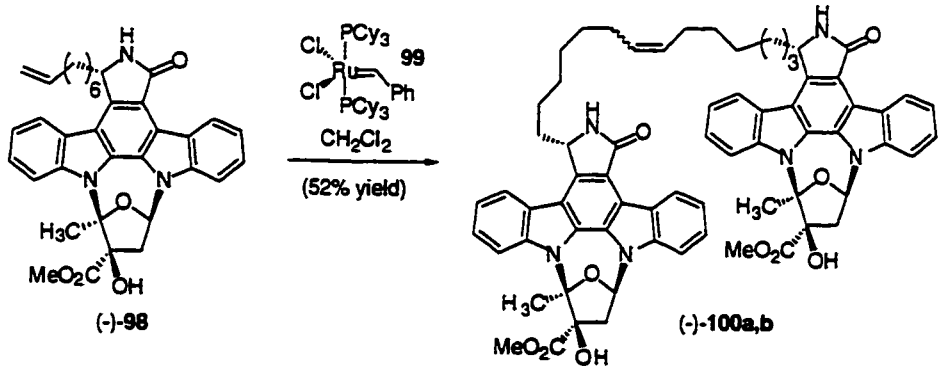
2.5 Synthesis of a Novel Dimer of (+)-K252a.

2.5.1 Synthesis of a K252a Dimer.

Although it did not seem possible to prepare a potent and selective C(7) substituted derivative of K252a, we were intrigued by a report describing the activation of signal transduction pathways and gene transcription by an FK-506 dimer produced *via* olefin metathesis.[23] With olefin analog (-)-**98** in hand we decided to pursue the synthesis of a K252a dimer.

Olefin (-)-**98** was treated with 0.20 mol % Grubbs catalyst **99** in CH_2Cl_2 for 24 hours at room temperature.[24] The dimeric product (-)-**100a,b** was isolated in 52% yield as a 1:1 mixture of olefin isomers.

Scheme 2.5.1



To our surprise, the mixture of olefins (-)-**100a,b** was a much more potent inhibitor than the monomeric octenyl derivative (-)-**98** (see Table 2.5.1). In fact, in the case of the kinase Lck the dimer had very similar inhibitory power to the natural product (IC_{50} = 1.25 μM as compared to 1.32 μM). Intrigued by this result, we speculated that (-)-**100a,b** might be involved in the inhibition of a process which involves kinase dimerization and can therefore be facilitated by a dimeric inhibitor.

Unfortunately, (-)-**100a,b** also had an uninteresting selectivity profile similar to the natural material.

Table 2.5.1

kinase	IC_{50} (μM)		
	(+)-**1**	(-)-**98**	(-)-**100a,b**
Lck	1.25	>9	1.32
Fyn	4.59	>9	5.87
ZAP-70	0.32	>9	0.95
Syk	0.04	>9	0.76
Itk	0.35	>9	4.01

2.6 Conclusion.

The total synthesis of a number of K252a analogs was completed by extension of the indolocarbazole synthesis developed in these laboratories. Methyl, benzyl, and octenyl analogs were prepared, as well as a novel dimer of K252a. Methyl derivative (-)-**52a** showed inhibition in the high nM to low μM range (IC_{50} = 190 nM to 6.22 μM) and also showed a better selectivity profile than either (+)-K252a (**1**), or its C(7) epimer (+)-**52b**. Benzyl and octenyl derivatives were not inhibitors of protein tyrosine kinases. In addition, a potent but unselective dimer of K252a was prepared.

2.7 Experimental Section.

2.7.1 Material and Methods.

Unless stated otherwise, reactions were performed in flame-dried glassware under a nitrogen atmosphere, using freshly distilled solvents. Diethyl ether (Et_2O) and tetrahydrofuran (THF) were distilled from sodium/benzophenone ketyl. Methylene chloride (CH_2Cl_2), benzene, and triethylamine (Et_3N) were distilled from calcium hydride. Boron trifluoride etherate ($BF_3 \bullet OEt_2$), 1,2-dichloroethane and pinacolone were purchased from the Aldrich Chemical Co. in Sure/Seal™ containers and used without further purification. All other commercially obtained reagents were used as received.

Unless stated otherwise, all reactions were magnetically stirred and monitored by thin-layer chromatography (TLC) using E. Merck silica gel 60 F254 pre-coated plates (0.25 mm). Preparative TLC was also performed using E. Merck silica gel 60 F254 pre-coated plates (0.25 mm). Column or flash chromatography (silica) was performed with the indicated solvents using silica gel (particle size 0.032-0.063 mm) purchased from Fisher Scientific. In general, the chromatography guidelines reported by Still were followed.[25]

All melting points were obtained on a Haacke-Buchler variable temperature melting point apparatus (model: MFB 595 8020) and are uncorrected. Infrared spectra were recorded on a Midac M-1200 FTIR. ^1H and ^{13}C NMR spectra were recorded on a Bruker AM-500 spectrometer. Chemical shifts are reported relative to solvent residuals: chloroform (^1H, δ 7.27 ppm, ^{13}C, δ 77.0 ppm), acetone (^1H, δ 2.04 ppm, ^{13}C, δ 206.0 ppm) or methyl sulfoxide (^1H, δ 2.49 ppm, ^{13}C, δ 39.5 ppm). High resolution mass spectral analyses were performed at The University of

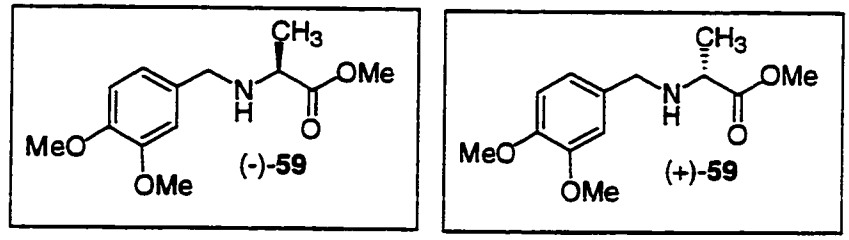
Illinois Mass Spectrometry Center. High performance liquid chromatography (HPLC) was performed on a Waters model 510 system using a Rainin Microsorb 80-199-C5 column or a Rainin Dynamax SD-200 system with a Rainin Microsorb 80-120-C5 column. Optical rotations were measured on a Perkin-Elmer 241 polarimeter.

The determination of enantiomeric excess by Mosher ester derivatization involved esterification of the corresponding alcohols with *R*-(+)-MTPA (DCC, CH_2Cl_2 or $(COCl)_2$, DMF, CH_2Cl_2) followed by purification and 500 MHz ^1H NMR analysis in benzene-d_6. Where possible an identical analysis was performed employing a racemic mixture of alcohols.

In some compounds containing a tertiary amine, spectra indicate a mixture of rotamers. Although these rotamers could be observed to coalesce at high temperatures, characterization spectra were obtained at room temperature.

2.7.2 Preparative Procedures:

Preparation of Amines (+) and (-)-59.

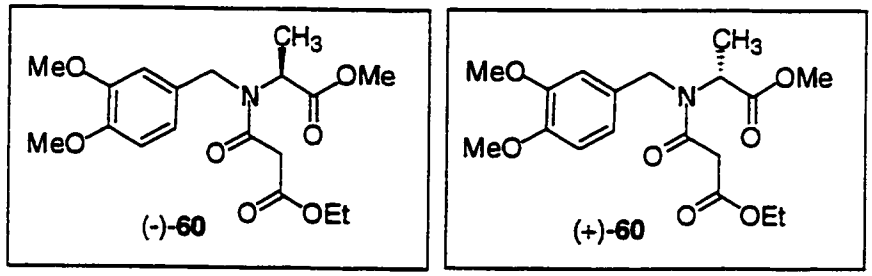


Amine (-)-59. To a solution of L-(-)-alanine methyl ester hydrochloride (**56**) (18.1 g, 130 mmol, 1.0 equiv), in MeOH (45 mL) was added Et_3N (18.1 mL, 130 mmol, 1.0 equiv) followed by addition of 3,4-dimethoxybenzaldehyde (15.1 g, 90.8 mmol, 0.7 equiv) as a solution in EtOH (230 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in EtOH (310 mL) and $NaBH_4$ (4.9 g, 130 mmol, 1.0 equiv) was added portionwise over a 15-minute period. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, the EtOH was removed under reduced pressure and the residue was dissolved in 1 N HCl (200 mL). The acid solution was washed with EtOAc (100 mL) and then brought to pH 10 using 10 N aqueous NaOH (10 mL). The basic solution was extracted with CH_2Cl_2 (3 x 100 mL) and the combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield the protected amine (-)-**59** (21.3 g, 93% yield) as a clear, colorless oil: $[\alpha]_D^{20}$ -36.00° (*c* 1.00, MeOH); IR (thin film/NaCl) 3329 (br w), 2945 (m), 2836 (w), 1735 (s), 1598 (w), 1513 (s), 1457 (m), 1262 (s), 1199 (s), 1031 (s), 980 (w), 855 (w), 808 (w), 762 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.88-6.78 (comp m, 3H), 3.87 (s, 3H), 3.84 (s, 3H), 3.72 (m, 4H), 3.60 (d, J = 12.6 Hz, 1H), 3.37 (q, J = 7.0 Hz, 1H), 1.81 (br s, 1H), 1.30 (d, J = 7.0 Hz, 3H); ^{13}C NMR

(125 MHz, $CDCl_3$) δ 175.5, 148.4, 147.5, 131.8, 119.7, 110.9, 110.5, 55.2, 55.1, 55.1, 51.1, 51.0, 18.5; high resolution mass spectrum (EI) *m/z* 253.1315 [calc'd for $C_{13}H_{19}NO_4$ (M+) 253.1314].

Amine (+)-**59** proved identical in all respects to (-)-**59** with the exception of the sign of the optical rotation.

Preparation of Amides (-)-60 and (+)-60.

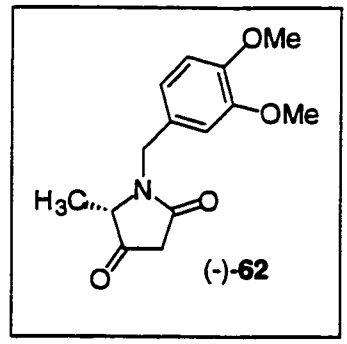


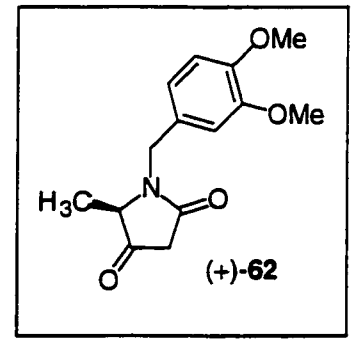
Amide (-)-60: A three-necked flask equipped with an addition funnel was charged with the amine (-)-**59** (25 g, 99 mmol, 1.0 equiv), ethyl hydrogen malonate (13.1 g, 99 mmol, 1.0 equiv), and CH_2Cl_2 (400 mL). The flask was cooled to 0°C and a solution of 1,3-dicyclohexylcarbodiimide (20.4 g, 99 mmol, 1.0 equiv) in CH_2Cl_2 (160 mL) was added dropwise through the addition funnel over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h at rt. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (500 mL). The organic layer was separated and the aqueous layer was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to yield an oily residue which was taken up in a minimum of acetone (10 mL) and filtered to remove the remaining urea. The filtrate was concentrated at reduced pressure to furnish amide (-)-**60** (34 g, 94% yield) as a clear, light yellow oil: $[\alpha]_D^{20}$ -39.69° (*c* 0.65, MeOH);

IR (thin film/NaCl) 2948 (m), 2839 (w), 1741 (s), 1654 (s), 1516 (s), 1366 (w), 1317 (w), 1261 (s), 1149 (s), 1029 (s), 920 (w), 853 (w), 807 (w), 767 (w), 734 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.87-6.76 (comp m, 3H), 4.38-4.74 (comp m, 3H), 4.17 (q, J = 7.1 Hz, 2H), 3.88 (s, 2.4H), 3.87 (s, 2.4H), 3.85 (s, 0.6H), 3.83 (s, 0.6H), 3.68 (s, 3H), 3.57-3.37 (comp m, 3H), 1.40 (d, J = 7.2 Hz, 3H), 1.29 (t, J = 7.3 Hz, 0.6H), 1.25 (t, J = 7.1 Hz, 2.4H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.6, 167.3, 167.0, 149.3, 148.5, 128.6, 119.6, 118.6, 111.2, 110.9, 110.7, 109.5, 61.6, 61.4, 56.0, 55.9, 55.9, 55.8, 54.4, 52.4, 52.2, 50.7, 50.6, 46.3, 41.8, 41.4, 15.9, 14.5, 14.0; high resolution mass spectrum (EI) *m/z* 367.1632 [calc'd for $C_{18}H_{25}NO_7$ (M+) 367.1631].

Amide (+)-**60** proved identical in all respects to (-)-**60** with the exception of the sign of the optical rotation.

Preparation of Lactams (-)-62 and (+)-62.



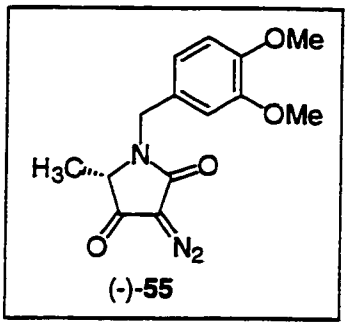


Lactam (-)-62. A three-necked flask was charged with absolute EtOH (18 mL). Sodium metal (294 mg, 12.8 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester (-)-**60** (5 g, 13.6 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in the remaining absolute EtOH (17 mL). The mixture was brought to reflux for 5 min in a preheated

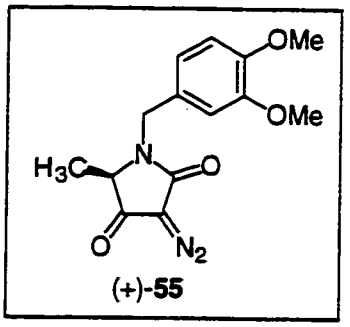
oil bath and then allowed to slowly cool to rt following the removal of the bath. The EtOH was removed under reduced pressure and the residue was dissolved in H_2O (100 mL). The aqueous layer was washed with EtOAc (50 mL) and acidified to a pH of 2 with 2 N HCl (10 mL). The acidic solution was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield a yellow oil. A suspension of the oil in CH_3CN (1130 mL) and H_2O (1 mL) was warmed to reflux open to the air for three hours. The mixture was cooled to rt and the CH_3CN/H_2O mixture was removed at reduced pressure to yield lactam (-)-**62** as a dark yellow oil (2.8 g, 80% yield) which solidified upon standing to a dark yellow glassy solid: $[\alpha]_D^{20}$ -29.61° (*c* 0.25, MeOH); IR (thin film/NaCl) 2940 (br m), 1766 (m), 1690 (s), 1603 (w), 1512 (m), 1424 (m), 1252 (s), 1147 (m), 1026 (m), 916 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.80 (m, 3H), 5.17 (d, *J* = 14.7 Hz, 1H), 3.97 (d, *J* = 14.7 Hz, 1H), 3.86 (s, 3H), 3.85 (s, 3H), 3.78 (q, *J* = 6.9 Hz, 1H), 3.09 (s, 2H), 1.31 (d, *J* = 6.9 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.7, 168.3, 149.4, 148.9, 127.8, 120.7, 111.4, 111.1, 61.7, 56.0, 55.9, 43.6, 40.5, 15.1; high resolution mass spectrum (EI) *m/z* 263.1160 [calc'd for $C_{14}H_{17}NO_4$ (M+) 263.1158].

Lactam (+)-**62** proved identical in all respects to (-)-**62** with the exception of the sign of the optical rotation.

Preparation of Diazo lactams (-)-55 and (+)-55.



(-)-55

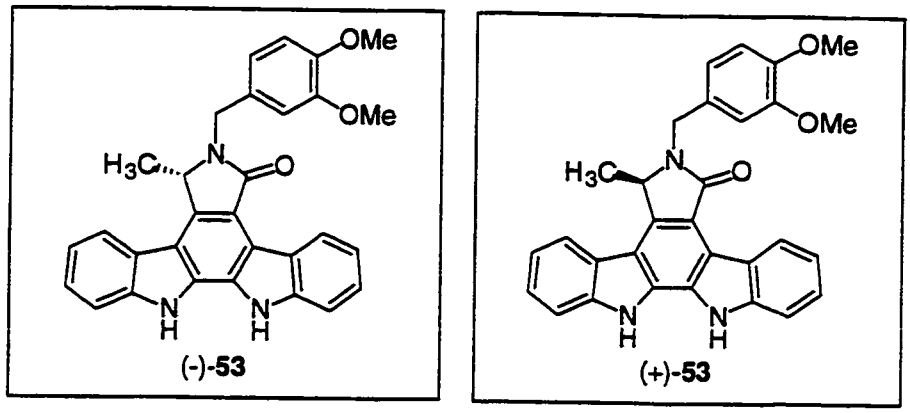


(+)-55

Diazo lactam (-)-55. A stirred solution of lactam (-)-**62** (1.5 g, 5.7 mmol, 1.0 equiv), MsN_3 (715 mg, 6.3 mmol, 1.1 equiv), and CH_3CN (40 mL) was cooled to 0°C and Et_3N (0.95 mL, 6.8 mmol, 1.2 equiv) was added dropwise to the mixture. After gradually warming to rt, the mixture was stirred for an additional 3 h and the volatiles were removed under reduced pressure. The residue was dissolved in CH_2Cl_2 (20 mL), and washed with 1 N aqueous NaOH (20 mL). The organic layer was separated, concentrated, dissolved in EtOAc (50 mL), and filtered through a plug of silica gel to provide diazo lactam (-)-**55** (1.5 g, 91% yield) as a bright yellow foam: $[\alpha]_D^{20}$ -94.13° (*c* 0.8, MeOH); IR (thin film/NaCl) 2929 (m), 2857 (w), 2126 (s), 1684 (s), 1515 (m), 1401 (m), 1356 (m), 1260 (m), 1232 (w), 1139 (w), 1026 (w), 736 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.79 (comp m, 3H), 5.09 (d, *J* = 14.9 Hz, 1H), 4.04 (d, *J* = 14.9 Hz, 1H), 3.88 (s, 6H), 3.76 (q, *J* = 6.9 Hz, 1H), 1.37 (d, *J* = 6.9 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 189.5, 161.3, 149.4, 148.9, 128.0, 120.6, 111.2, 111.1, 59.6, 55.9, 55.9, 44.2, 15.3; high resolution mass spectrum (EI) *m/z* 289.1065 [calc'd for $C_{14}H_{15}N_3O_4$ (M+) 289.1063].

Diazo lactam (+)-**55** proved identical in all respects to (-)-**55** with the exception of the sign of the optical rotation.

Preparation of Aglycons (-)-53 and (+)-53.

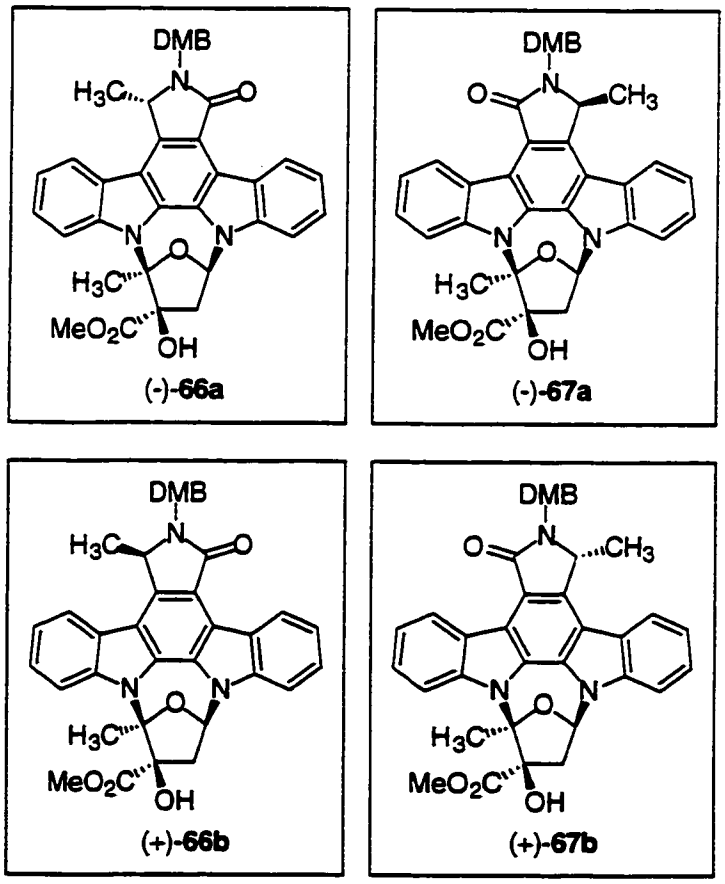


Aglycon (-)-53. To a three-necked flask equipped with a condenser were added the diazo lactam (-)-**55** (800 mg, 2.8 mmol, 1.0 equiv), 2,2'-biindole (**11**) (650 mg, 2.8 mmol, 1.0 equiv), $Rh_2(OAc)_4$ (12 mg, 0.028 mmol, 0.01 equiv) and pinacolone (28 mL). The whole was degassed by bubbling a stream of N_2 through the solution for 2 h. The mixture was then warmed to reflux for an additional 8 h. The reaction mixture was concentrated to a dark brown foamy solid, which was adsorbed onto silica gel and subjected to flash chromatography (50% EtOAc/hexanes eluent), affording unreacted **11** (335 mg, 52% yield) as a white powder, and aglycon (-)-**53** (430 mg, 33% yield; 64% yield based on recovered **11**), isolated as a pale yellow solid: mp 245-248 °C (dec.); $[\alpha]_D^{20}$ -31.11° (*c* 0.45, MeOH); IR (thin film/NaCl) 3323 (br w), 2931 (w), 1649 (s), 1514 (s), 1320 (s), 1236 (s), 1231 (s), 1140 (m), 1024 (m), 812 (w), 746 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 10.92 (s, 1H), 10.70 (s, 1H), 9.53 (d, J = 8.0 Hz, 1H), 8.06 (d, J = 7.9 Hz, 1H), 7.64 (d, J = 8.3 Hz, 1H), 7.60 (d, J = 8.0 Hz, 1H), 7.41 (m, 2H), 7.25 (t, J = 7.5 Hz, 2H), 7.07 (m, 1H), 6.99 (m, 1H), 6.88 (m, 1H), 5.36 (d, J = 15.2 Hz, 1H), 5.15 (m, 1H), 4.46 (d, J = 15.2 Hz, 1H), 3.74 (s, 3H), 3.73 (s, 3H), 1.75 (d, J = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 170.0, 150.6, 149.8, 140.9, 140.9, 137.9, 132.2, 129.3,

127.0, 126.9, 126.8, 126.3, 126.0, 124.5, 123.4, 122.8, 121.1, 121.0, 120.2, 119.5, 117.6, 115.2, 112.9, 112.9, 112.6, 111.9, 56.2, 56.1, 44.0, 18.9; high resolution mass spectrum (EI) *m/z* 475.1896 [calc'd for $C_{30}H_{25}N_3O_3$ (M+) 475.1896].

Aglycon (+)-**53** proved identical in all respects to (-)-**53** with the exception of the sign of the optical rotation.

Preparation of Indolocarbazoles (-)-66a, (+)-66b, (-)-67a and (+)-67b.



Indolocarbazoles (-)-66a and (-)-67a. To a refluxing solution of aglycon (-)-**53** (700 mg, 1.47 mmol, 1.0 equiv) and camphorsulfonic acid (34 mg, 0.147

mmol, 0.1 equiv) in 1,2-dichloroethane (50 mL) was added, via addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)-**17** (638 mg, 2.9 mmol, 2.0 equiv) in 1,2-dichloroethane (32 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% aqueous $NaHCO_3$ (20 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 20 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:1 mixture of regioisomers (-)-**66a** and (-)-**67a** (753 mg, 81% yield). Separation of the regioisomers was achieved using HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent).

(-)-**66a:** mp 280-282 °C (dec.); $[\alpha]_D^{20}$ -14.35° (*c* 0.23, MeOH); IR (thin film/NaCl) 3343 (br w), 3003 (w), 2950 (w), 1733 (s), 1670 (s), 1586 (m), 1514 (m), 1455 (s), 1393 (s), 1351 (m), 1314 (m), 1259 (s), 1201 (m), 1138 (m), 1026 (m), 800 (w), 747 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.50 (d, *J* = 7.8 Hz, 1H), 8.06 (d, *J* = 7.9 Hz, 1H), 8.00 (d, *J* = 8.5 Hz, 1H), 7.81 (d, *J* = 8.3 Hz, 1H), 7.51 (ddd, *J* = 1.1, 7.2, 8.2 Hz, 1H), 7.43 (ddd, *J* = 1.1, 7.2, 8.3 Hz, 1H), 7.33-7.28 (comp m, 2H), 7.16 (dd, *J* = 4.9, 7.4 Hz, 1H), 7.08 (m, 1H), 6.99 (m, 1H), 6.92 (m, 1H), 5.37 (d, *J* = 15.2 Hz, 1H), 5.19 (q, *J* = 6.5 Hz, 1H), 4.45 (d, *J* = 15.2 Hz, 1H), 4.01 (s, 3H), 3.77 (s, 3H), 3.76 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.23 (s, 3H), 2.22 (dd, *J* = 5.0, 14.1 Hz, 1H), 1.78 (d, *J* = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.7, 169.1, 150.4, 149.5, 141.2, 138.2, 137.4, 131.8, 129.8, 127.2, 127.1, 126.3, 125.6, 125.3, 124.9, 124.0, 122.5, 121.2, 120.8, 120.3, 119.7, 117.3, 115.8, 115.2, 112.6, 109.1, 100.0, 86.2, 86.0, 85.7, 55.9, 53.2, 43.6, 43.2, 43.1, 23.2, 18.6; high resolution mass spectrum (EI) *m/z* 631.2314 [calc'd for $C_{37}H_{33}N_3O_7$ (M+) 631.2319].

53.2, 43.6, 43.2, 43.1, 23.2, 18.6; high resolution mass spectrum (EI) *m/z* 631.2314 [calc'd for $C_{37}H_{33}N_3O_7$ (M+) 631.2319].

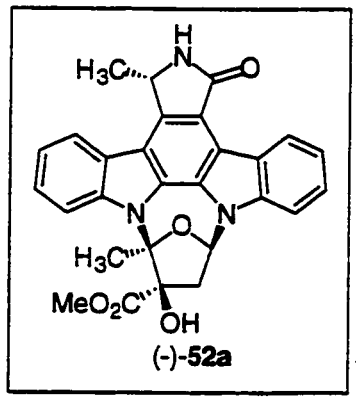
(+)-**66b**: mp >310 °C (dec.); $[\alpha]_D^{20}$ +58.75° (*c* 0.8, MeOH); IR (thin film/NaCl) 3479 (br w), 3361 (br w), 3062 (w), 2946 (w), 2835 (w), 1728 (s), 1660 (s), 1583 (m), 1516 (m), 1454 (m), 1257 (m), 1140 (m), 1034 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.50 (d, *J* = 8.0 Hz, 1H), 8.10 (d, *J* = 7.8 Hz, 1H), 8.00 (d, *J* = 8.5 Hz, 1H), 7.81 (d, *J* = 8.3 Hz, 1H), 7.51 (ddd, *J* = 1.2, 7.1, 8.3 Hz, 1H), 7.44 (ddd, *J* = 1.2, 7.2, 8.4 Hz, 1H), 7.31 (q, *J* = 7.2 Hz, 2H), 7.16 (dd, *J* = 5.0, 7.4 Hz, 1H), 7.09 (m, 1H), 7.02 (m, 1H), 6.93 (m, 1H), 5.38 (s, 1H), 5.37 (d, *J* = 15.1 Hz, 1H), 5.22 (q, *J* = 6.4 Hz, 1H), 4.45 (d, *J* = 15.2 Hz, 1H), 4.01 (s, 3H), 3.78 (s, 3H), 3.77 (s, 3H), 3.53 (dd, *J* = 7.5, 14.1 Hz, 1H), 2.35 (dd, *J* = 5.0, 14.2 Hz, 1H), 2.19 (s, 3H), 1.74 (d, *J* = 6.4 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 169.2, 150.7, 149.8, 141.5, 138.5, 137.7, 132.0, 129.8, 127.4, 127.3, 126.5, 125.7, 125.5, 124.8, 124.2, 122.9, 121.2, 121.1, 120.5, 120.1, 117.6, 115.8, 115.5, 113.0, 109.2, 100.5, 86.4, 85.9, 56.1, 56.1, 56.1, 53.5, 43.9, 43.3, 23.5, 18.9; high resolution mass spectrum (EI) *m/z* 631.2314 [calc'd for $C_{37}H_{33}N_3O_7$ (M+) 631.2319].

(-)-**67a**: mp >300 °C (dec.); $[\alpha]_D^{20}$ -21.76° (*c* 0.40, MeOH); IR (thin film/NaCl) 3055 (w), 2959 (w), 2838 (w), 1735 (s), 1678 (s), 1590 (m), 1517 (m), 1451 (m), 1403 (s), 1218 (m), 1234 (s), 1138 (m), 1023 (m), 751 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.74 (d, *J* = 7.7 Hz, 1H), 8.13 (d, *J* = 7.9 Hz, 1H), 7.94 (d, *J* = 8.5 Hz, 1H), 7.89 (d, *J* = 8.3 Hz, 1H), 7.52 (ddd, *J* = 1.0, 7.3, 8.2 Hz, 1H), 7.44 (ddd, *J* = 1.4, 7.0, 8.5 Hz, 1H), 7.31 (m, 2H), 7.15 (dd, *J* = 4.9, 7.4 Hz, 1H), 7.09 (m, 1H), 7.01 (m, 1H), 6.93 (m, 1H), 5.36 (d, *J* = 15.2 Hz, 1H), 5.30 (s, 1H), 5.19 (q, *J* = 6.5 Hz, 1H), 4.46 (d, *J* = 15.2 Hz, 1H), 4.01 (s, 3H), 3.78 (s, 3H),

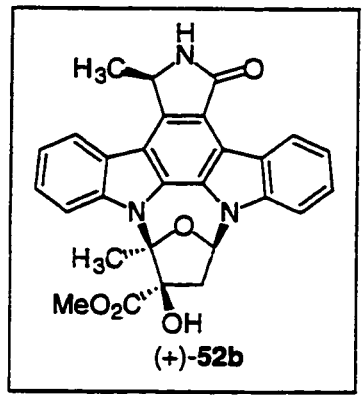
3.76 (s, 3H), 3.55 (dd, J = 7.5, 14.1 Hz, 1H), 2.40 (dd, J = 4.9, 14.1 Hz, 1H), 2.20 (s, 3H), 1.75 (d, J = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.7, 169.4, 150.5, 149.6, 141.2, 138.5, 138.0, 132.0, 127.7, 127.2, 127.1, 126.2, 126.0, 123.3, 123.0, 121.2, 120.9, 120.4, 119.6, 118.2, 116.0, 114.9, 114.8, 112.7, 110.0, 100.2, 86.4, 85.7, 56.0, 56.0, 55.6, 53.3, 43.8, 43.4, 29.1, 23.4, 18.7; high resolution mass spectrum (EI) *m/z* 631.2320 [calc'd for $C_{37}H_{33}N_3O_7$ (M+) 631.2319].

(+)-**67b**: mp 267-271 °C (dec.); $[\alpha]_D^{20}$ +62.69° (*c* 0.26, MeOH); IR (thin film/NaCl) 3479 (w), 3365 (br w), 3057 (w), 2933 (w), 2835 (w), 1738 (s), 1676 (s), 1588 (m), 1511 (m), 1454 (m), 1243 (m), 1140 (m), 1022 (m), 743 (m) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.75 (d, J = 8.1 Hz, 1H), 8.11 (d, J = 7.9 Hz, 1H), 7.94 (d, J = 8.5 Hz, 1H), 7.87 (d, J = 8.3 Hz, 1H), 7.51 (ddd, J = 1.0, 7.2, 8.2 Hz, 1H), 7.44 (ddd, J = 1.4, 7.1, 8.5 Hz, 1H), 7.31 (q, J = 7.7 Hz, 1H), 7.15 (dd, J = 4.9, 7.4 Hz, 1H), 7.08 (m, 1H), 6.99 (m, 1H), 6.91 (m, 1H), 5.38 (d, J = 15.2 Hz, 1H), 5.23 (s, 1H), 5.15 (q, J = 6.5 Hz, 1H), 4.45 (d, J = 15.2 Hz, 1H), 4.01 (s, 3H), 3.77 (s, 3H), 3.75 (s, 3H), 3.50 (dd, J = 7.5, 14.0 Hz, 1H), 2.27 (ddd, J = 1.9, 5.0, 14.0 Hz, 1H), 2.22 (s, 3H), 1.78 (d, J = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.7, 169.4, 150.5, 149.6, 141.1, 138.4, 138.0, 131.9, 127.7, 127.5, 127.2, 127.1, 126.1, 126.0, 125.9, 123.2, 123.1, 121.2, 120.9, 120.4, 119.6, 118.1, 114.8, 112.7, 110.0, 100.1, 86.4, 86.0, 56.0, 56.0, 55.4, 53.3, 43.7, 43.1, 29.1, 23.3, 18.7; high resolution mass spectrum (EI) *m/z* 631.2313 [calc'd for $C_{37}H_{33}N_3O_7$ (M+) 631.2319].

Preparation of (-)-7-(*S*)-Methyl K252a (52a) and (+)-7-(*R*)-Methyl K252a (52b).



(-)-**52a**



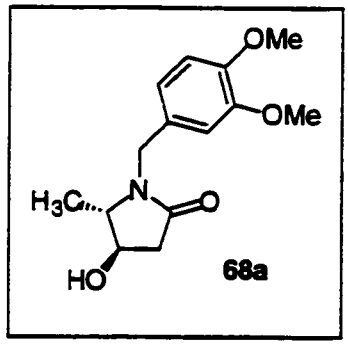
(+)-**52b**

Representative Procedure: (-)-7-(*S*)-Methyl K252a (52a). A solution of protected amide (-)-**66a** (60 mg, 0.095 mmol, 1.0 equiv) and anisole (1.03 g, 9.5 mmol, 100 equiv) in CH_2Cl_2 (2 mL) was treated dropwise with TFA (2 mL). After stirring at rt for 12 h, the reaction was quenched with 20% aqueous $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**52a** (39 mg, 85% yield) as a pale yellow solid: mp 276-281 °C (dec.); $[\alpha]_D^{20}$ -4.00° (*c* 0.1, MeOH); IR (thin film/NaCl) 3407 (br w), 3266 (br w), 3053 (w), 3002 (w), 2950 (w), 1733 (m), 1669 (s), 1679 (m), 1450 (s), 1392 (s), 1308 (m), 1257 (m), 1199 (m), 1141 (m), 1083 (m), 748 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.41 (d, *J* = 7.9 Hz, 1H), 8.19 (d, *J* = 7.7 Hz, 1H), 8.02 (d, *J* = 8.5 Hz, 1H), 7.79 (d, *J* = 8.3 Hz, 1H), 7.63 (s, 1H), 7.50-7.44 (comp m, 2H), 7.35 (t, *J* = 7.5 Hz, 1H), 7.28 (t, *J* = 7.5 Hz, 1H), 7.15 (dd, *J* = 4.9, 7.4 Hz, 1H), 5.43 (q, *J* = 6.5 Hz, 1H), 5.28 (s, 1H), 4.01 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.24 (m, 4H), 1.79 (d, *J* = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 171.6, 141.4, 139.7, 138.4, 130.1, 127.5, 127.4, 126.4, 125.7, 125.5, 125.3, 124.4, 122.7, 121.4, 117.6, 116.0, 109.2,

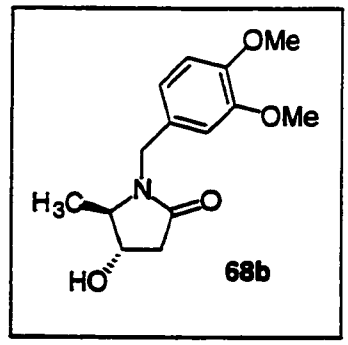
100.3, 86.5, 86.2, 53.4, 53.3, 53.2, 43.4, 23.4, 21.3; high resolution mass spectrum (EI) *m/z* 481.1637 [calc'd for $C_{28}H_{23}N_3O_5$ (M+) 481.1638].

(+)-**52b**: mp 295-298 °C (dec.); $[\alpha]_D^{20}$ +41.02° (*c* 0.1, MeOH); IR (thin film/NaCl) 3407 (br w), 2964 (w), 2928 (m), 2861 (w), 1738 (m), 1676 (s), 1588 (m), 1449 (s), 1393 (m), 1315 (m), 1202 (m), 1084 (w), 877 (w), 743 (m) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.41 (d, *J* = 7.8 Hz, 1H), 8.21 (d, *J* = 7.9 Hz, 1H), 8.02 (d, *J* = 8.5 Hz, 1H), 7.79 (d, *J* = 8.3 Hz, 1H), 7.65 (s, 1H), 7.51-7.44 (comp m, 2H), 7.36 (t, *J* = 7.5 Hz, 1H), 7.28 (t, *J* = 7.5 Hz, 1H), 7.15 (dd, *J* = 5.0, 7.4 Hz, 1H), 5.45 (q, *J* = 6.4 Hz, 1H), 5.39 (s, 1H), 4.02 (s, 3H), 3.52 (dd, *J* = 7.5, 14.1 Hz, 1H), 2.34 (dd, *J* = 5.0, 14.1 Hz, 1H), 2.21 (s, 3H), 1.76 (d, *J* = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 171.6, 141.5, 139.7, 138.4, 129.9, 127.5, 127.4, 126.4, 125.6, 125.4, 125.1, 124.2, 122.9, 121.2, 120.4, 117.7, 115.8, 109.1, 100.5, 86.4, 85.9, 53.5, 53.3, 53.2, 43.3, 23.5, 21.4; high resolution mass spectrum (EI) *m/z* 481.1633 [calc'd for $C_{28}H_{23}N_3O_5$(M+) 481.1638].

Preparation of Alcohols (-)-68a-b.



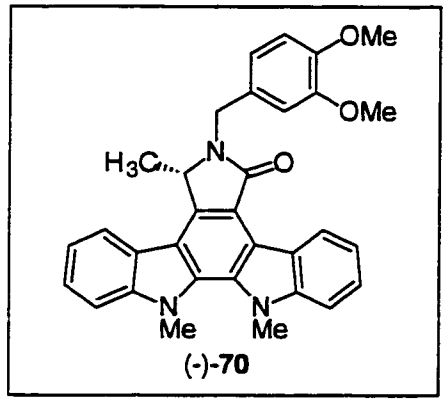
68a



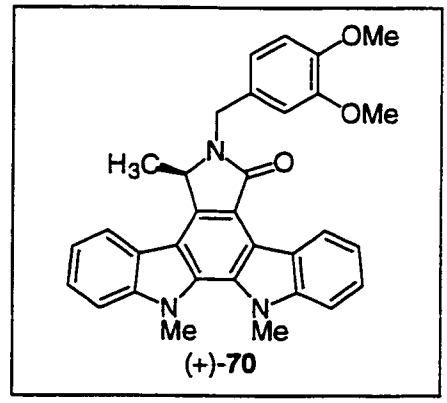
68b

Alcohols (-)-68a-b. To a solution of lactam (-)-**62** (1 g, 3.8 mmol, 1.0 equiv) in MeOH (20 mL), $NaBH_4$ (144 mg, 3.8 mmol, 1.0 equiv) was carefully added in one portion. The mixture was stirred at rt for 30 min and MeOH was then removed under reduced pressure to afford an oily yellow residue. The oil was taken up in 1 N HCl (20 mL) and the acidic solution was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to provide (-)-**68a-b** (954 mg, 95% yield) as an inseparable 1.5:1 mixture of diastereomers of a clear, colorless oil: $[\alpha]_D^{20}$ -40.90° (*c* 0.85, MeOH); IR (thin film/NaCl) 3395 (br w), 3055 (w), 2923 (m), 2851 (w), 1681 (s), 1515 (m), 1418 (m), 1265 (s), 1143 (w), 1029 (m), 739 (s) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.80-6.75 (comp m, 3H), 4.91 (d, *J* = 14.9 Hz, 1H), 4.31 (m, 0.6H), 4.04 (m, 0.4H), 3.89 (d, *J* = 14.5 Hz, 1H), 3.85 (s, 3H), 3.55 (m, 0.6H), 3.39 (m, 0.4H), 2.77 (dd, *J* = 6.2, 17.3 Hz, 0.4H), 2.65 (dd, *J* = 6.5, 17.1 Hz, 0.6H), 2.47 (d, *J* = 3.2 Hz, 0.6H), 2.43 (d, *J* = 3.2 Hz, 0.4H), 1.20 (d, *J* = 6.6 Hz, 1.8H), 1.12 (d, *J* = 6.6 Hz, 1.2H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 173.0, 172.4, 149.2, 149.1, 148.4, 148.4, 129.1, 128.8, 120.2, 120.1, 111.1, 111.0, 111.0, 111.0, 71.4, 67.1, 61.7, 57.2, 55.9, 55.8, 43.6, 43.5, 40.2, 39.7, 16.6, 12.4; high resolution mass spectrum (EI) *m/z* 265.1314 [calc'd for $C_{14}H_{19}NO_4$(M+) 265.1314].

Preparation of Indolocarbazoles (-) and (+)-70.



(-)-70



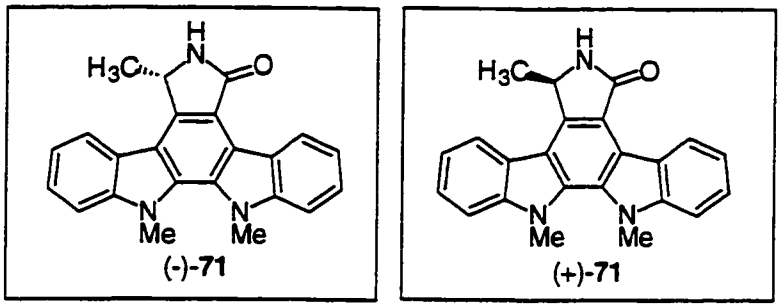
(+)-70

Indolocarbazole (-)-70. To a solution of aglycon (-)-**53** (100 mg, 0.21 mmol, 1.0 equiv) in THF (2 mL) was added powdered KOH (142 mg, 2.53 mmol, 12.0 equiv), MeI (298 mg, 2.1 mmol, 10 equiv), and *n*-Bu_4NBr (7 mg, 0.021 mmol, 0.1 equiv). The mixture was allowed to stir at rt for 10 min, at which time the reaction mixture was filtered through a plug of alumina (EtOAc eluent) providing indolocarbazole (-)-**70** (85 mg, 80% yield) as a pale yellow solid: mp >320 °C (dec.); $[\alpha]_D^{20}$ -43.61° (*c* 0.28, MeOH); IR (thin film/NaCl) 3063 (br w), 3015 (br w), 2939 (w), 2843 (w), 1671 (s), 1585 (m), 1521 (m), 1451 (m), 1413 (m), 1322 (s), 1241 (s), 1134 (m), 1026 (m), 752 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.65 (d, *J* = 7.8 Hz, 1H), 8.05 (d, *J* = 7.8 Hz, 1H), 7.68 (d, *J* = 8.2 Hz, 1H), 7.63 (d, *J* = 8.3 Hz, 1H), 7.55-7.51 (comp m, 2H), 7.34-7.30 (comp m, 2H), 7.09 (m, 1H), 6.99 (m, 1H), 6.91 (m, 1H), 5.34 (d, *J* = 15.2 Hz, 1H), 5.13 (m, 1H), 4.44 (d, *J* = 15.2 Hz, 1H), 4.30 (s, 3H), 4.25 (s, 3H), 3.76 (s, 6H), 1.72 (d, *J* = 6.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.1, 149.5, 148.6, 144.8, 144.3, 137.3, 132.4, 130.6, 130.2, 126.6, 126.2, 125.7, 124.3, 123.2, 121.8, 120.8, 120.8, 120.4, 120.1, 119.2, 116.6, 114.5, 113.3, 110.6, 109.8, 56.0, 56.0, 54.8, 43.5, 37.0,

36.6, 18.1; high resolution mass spectrum (EI) *m/z* 503.2209 [calc'd for $C_{32}H_{29}N_3O_3$(M+) 503.2209].

Indolocarbazole (+)-**70** proved identical in all respects to (-)-**70** with the exception of the sign of the optical rotation.

Preparation of Amides (-) and (+)-71.

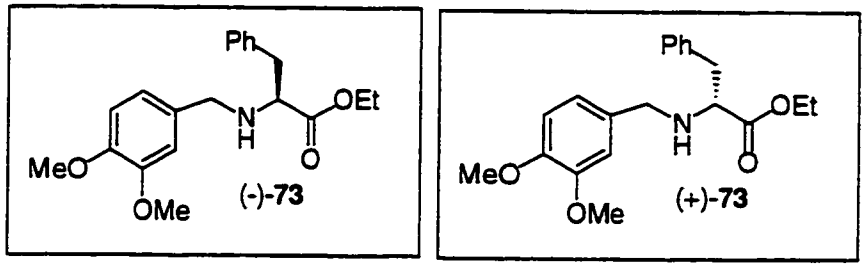


Amide (-)-71. A solution of protected amide (-)-**70** (30 mg, 0.06 mmol, 1.0 equiv) in anisole (1.30 g, 12 mmol, 200 equiv) was treated with TFA (6 mL). After stirring at rt for 12 h, the reaction was quenched with 20% $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**71** (18 mg, 86% yield) as a white solid: mp 310-313 °C (dec.); $[\alpha]_D^{20}$ -43.19° (*c* 0.19, MeOH); IR (thin film/NaCl) 3194 (br w), 3066 (br w), 2926 (br w), 2851 (br w), 1674 (s), 1581 (w), 1470 (m), 1435 (m), 1412 (w), 1319 (m), 1237 (w), 1150 (w), 1086 (w), 1016 (w), 987 (w), 940 (w), 736 (m) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.56 (d, *J* = 7.9 Hz, 1H), 8.19 (d, *J* = 7.8 Hz, 1H), 7.73 (d, *J* = 8.3 Hz, 1H), 7.64-7.51 (comp m, 4H), 7.39 (t, *J* = 7.2 Hz, 1H), 7.35 (t, *J* = 7.1 Hz, 1H), 5.37 (q, *J* = 6.4 Hz, 1H), 4.34 (s, 3H), 4.27 (s, 3H), 1.74 (d, *J* = 6.4 Hz, 3H); ^{13}C NMR (125 MHz, DMSO-d_6) δ

170.7, 143.6, 143.2, 139.0, 131.2, 129.0, 125.8, 125.6, 125.5, 123.0, 122.0, 121.7, 120.6, 119.7, 118.8, 118.3, 115.9, 111.0, 110.4, 51.7, 36.8, 36.6, 20.6; high resolution mass spectrum (EI) *m/z* 353.1531 [calc'd for $C_{23}H_{19}N_3O$ (M+) 353.1528].

Amide (+)-**71** proved identical in all respects to (-)-**71** with the exception of the sign of the optical rotation.

Preparation of Amines (+) and (-)-73.

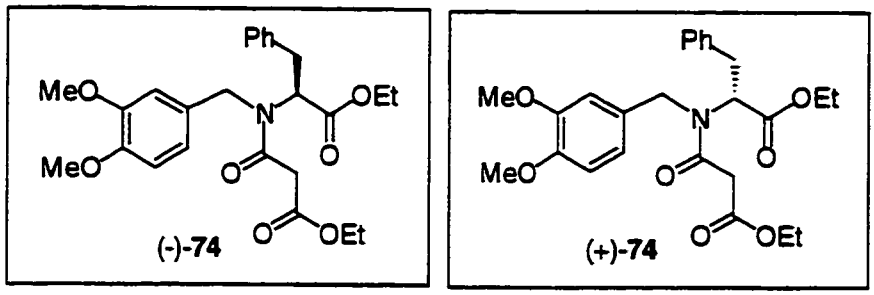


Amine (-)-73. To a solution of L-(-)-phenylalanine methyl ester hydrochloride (**72**) (50.0 g, 233 mmol, 1.0 equiv), in MeOH (75 mL) was added Et_3N (30.4 mL, 233 mmol, 1.0 equiv) followed by addition of 3,4-dimethoxybenzaldehyde (25.3 g, 152 mmol, 0.7 equiv) as a solution in EtOH (360 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in EtOH (550 mL) and $NaBH_4$ (8.2 g, 218 mmol, 1.0 equiv) was added portionwise over a 15 minute period. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, the EtOH was removed under reduced pressure and the residue was dissolved in 1 N HCl (400 mL). The acid solution was washed with EtOAc (400 mL) and then brought to pH 10 using 10 N aqueous NaOH (20 mL). The basic solution was extracted with CH_2Cl_2 (3 x 200 mL) and the combined organic layers were dried over anhydrous

$MgSO_4$, filtered, and concentrated to yield the protected amine (-)-**73** (42.5 g, 81% yield) as a clear, colorless oil: $[\alpha]_D^{20}$ -11.00° (*c* 0.30, MeOH); IR (thin film/NaCl) 3337 (br w), 2943 (m), 2836 (m), 1730 (s), 1516 (s), 1463 (m), 1265 (s), 1236 (s), 1184 (m), 1029 (s), 763 (m), 701 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.30-7.17 (comp m, 5H), 6.81-6.75 (comp m, 3H), 4.13 (q, *J* = 7.2 Hz, 2H), 3.87 (s, 3H), 3.82 (s, 3H), 3.79 (d, *J* = 13.0 Hz, 1H), 3.60 (d, *J* = 13.0 Hz, 1H), 3.53 (t, *J* = 7.0 Hz, 1H), 2.98 (m, 2H), 1.19 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 174.5, 148.8, 147.9, 137.4, 132.1, 129.2, 128.2, 126.5, 120.1, 111.1, 110.8, 61.7, 60.5, 55.8, 55.6, 51.6, 39.7, 14.1; high resolution mass spectrum (EI) *m/z* 343.1784 [calc'd for $C_{20}H_{25}NO_4$ (M+) 343.1784].

Amine (+)-**73** proved identical in all respects to (-)-**73** with the exception of the sign of the optical rotation.

Preparation of Amides (-)-74 and (+)-74.

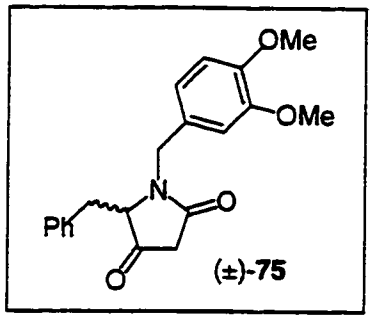


Amide (-)-74: A three-necked flask equipped with an addition funnel was charged with the amine (-)-**73** (4.0 g, 11.6 mmol, 1.0 equiv), ethyl hydrogen malonate (1.53 g, 11.6 mmol, 1.0 equiv), and CH_2Cl_2 (60 mL). The flask was cooled to 0°C and a 1 M solution of 1,3-dicyclohexylcarbodiimide (11.6 mL, 11.6 mmol, 1.0 equiv) in CH_2Cl_2 (40 mL) was added dropwise *via* an addition funnel

over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h at room temperature. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (100 mL). The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to yield an oily residue which was taken up in a minimum of acetone (10 mL) and filtered to remove the remaining urea. The filtrate was concentrated at reduced pressure to furnish amide (-)-**74** (5.0 g, 94% yield) as a clear, light yellow oil: $[\alpha]_D^{20}$ -92.35° (*c* 0.85, MeOH); IR (thin film/NaCl) 2988 (m), 2938 (m), 2838 (w), 1741 (s), 1660 (s), 1515 (s), 1455 (m), 1260 (s), 1230 (s), 1155 (m), 1024 (m), 749 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.30-7.07 (comp m, 5H), 6.90-6.62 (comp m, 3H), 4.39 (d, *J* = 16.6 Hz, 1H), 4.26 (dd, *J* = 5.5, 9.4 Hz, 1H), 4.19 (q, *J* = 7.2 Hz, 1.06H), 4.12 (q, *J* = 7.1 Hz, 0.94H), 3.84 (s, 2.9H), 3.80 (s, 2.2H), 3.73 (s, 0.45H), 3.70 (s, 0.45H), 3.45-3.25 (comp m, 4H), 1.27 (t, *J* = 7.1 Hz, 1.6H), 1.22 (t, *J* = 7.2 Hz, 1.4H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.7, 169.5, 167.3, 167.1, 167.0, 166.6, 149.1, 148.8, 148.5, 148.0, 138.0, 136.5, 129.9, 129.2, 129.1, 128.6, 128.4, 127.8, 127.0, 126.5, 120.1, 119.5, 111.3, 110.9, 110.6, 110.4, 62.3, 61.6, 61.4, 61.3, 61.3, 61.1, 55.8, 55.7, 52.7, 46.5, 41.5, 41.4, 36.0, 35.0, 14.0, 14.0, 13.7; high resolution mass spectrum (EI) *m/z* 457.2110 [calc'd for $C_{25}H_{31}NO_7$ (M+) 457.2101].

Amide (+)-**74** proved identical in all respects to (-)-**74** with the exception of the sign of the optical rotation.

Preparation of Lactam (±)-75.

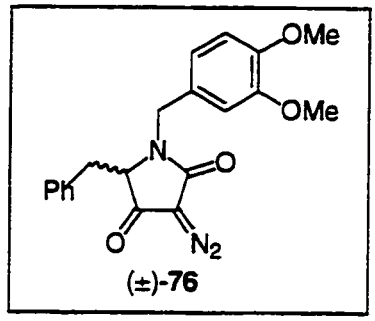


(±)-75

Lactam (±)-75. A three-necked flask was charged with EtOH (80 mL). Sodium metal (1.4 g, 60.7 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester (-)-**74** (29.5 g, 64.6 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in EtOH (80 mL). The mixture was brought to reflux for 5 min and then allowed to cool to rt. The EtOH was removed under reduced pressure and the residue was dissolved in H_2O (500 mL). The aqueous layer was washed with EtOAc (500 mL) and acidified to a pH of 2 with 2 N HCl (50 mL). The acidic solution was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield a yellow oil. A suspension of the oil in CH_3CN (522 mL) and H_2O (1 mL) was warmed to reflux open to the air for three hours. The mixture was cooled to rt and the CH_3CN/H_2O mixture was removed under reduced pressure to yield lactam (±)-**75** as a dark yellow oil (17.4 g, 80% yield) which solidified upon standing to a dark yellow glassy solid: IR (thin film/NaCl) 2933 (br w), 2833 (w), 1766 (m), 1686 (s), 1596 (w), 1511 (s), 1456 (m), 1411 (m), 1265 (s), 1235 (s), 1145 (m), 1025 (m), 760 (w), 699 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.32-7.25 (comp m, 3H), 7.06 (m, 2H), 6.83-6.74 (comp m, 3H), 5.36 (d, J = 14.6 Hz, 1H), 4.02 (t, J = 4.3 Hz, 1H), 3.94 (d, J = 14.8 Hz, 1H), 3.88 (s, 3H), 3.85 (s, 3H), 3.15 (dd, J = 4.4, 14.4 Hz, 1H), 3.07 (dd, J = 4.5, 14.4 Hz, 1H), 2.83 (d, J = 22.2

Hz, 1H), 2.40 (d, J = 22.2 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.3, 168.9, 149.4, 149.0, 134.5, 129.5, 128.8, 127.5, 127.4, 120.9, 111.6, 111.1, 66.6, 55.9, 55.9, 43.9, 41.3, 35.3; high resolution mass spectrum (EI) *m/z* 339.1481 [calc'd for $C_{20}H_{21}NO_4$ (M+) 339.1471].

Preparation of Diazo lactam (±)-76.

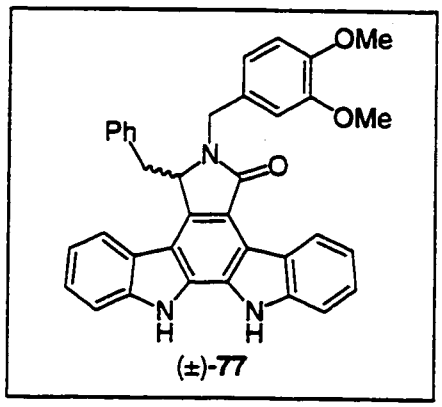


(±)-76

Diazo lactam (±)-76. A stirred solution of lactam (±)-**75** (2.5 g, 7.36 mmol, 1.0 equiv), *p*-ABSA (1.95 g, 8.10 mmol, 1.1 equiv), and CH_3CN (40 mL) was cooled to 0°C and Et_3N (3.1 mL, 22.1 mmol, 3.0 equiv) was added dropwise to the mixture. After gradually warming to rt, the mixture was stirred for an additional 3 h and the volatiles were removed under reduced pressure. The residue was subjected to flash chromatography (50% EtOAc/hexanes eluent) to provide diazo lactam (±)-**76** (2.4 g, 89% yield) as a bright yellow foam: IR (thin film/NaCl) 2938 (w), 2119 (s), 1679 (s), 1521 (m), 1391 (m), 1230 (m), 1145 (w), 1020 (w), 745 (w), 694 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.33-7.26 (comp m, 3H), 7.13 (d, J = 6.6 Hz, 2H), 6.80 (d, J = 8.0 Hz, 1H), 6.64 (m, 2H), 5.22 (d, J = 14.8 Hz, 1H), 3.99 (t, J = 4.8 Hz, 1H), 3.87 (m, 4H), 3.84 (s, 3H), 3.21 (dd, J = 4.4, 14.5 Hz, 1H), 3.10 (dd, J = 5.4, 14.5 Hz, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 188.3, 161.8, 149.3, 148.8, 134.7, 129.4, 128.6, 127.7, 127.2, 120.6, 111.3, 111.0,

65.5, 64.0, 55.8, 55.8, 44.6, 35.6; high resolution mass spectrum (FAB) *m/z* 366.1453 [calc'd for $C_{20}H_{20}N_3O_4$ (M+H) 366.1454].

Preparation of Aglycon (±)-77.

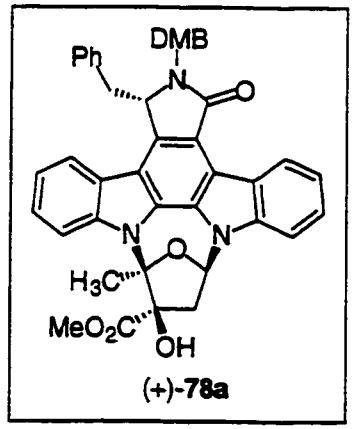


(±)-77

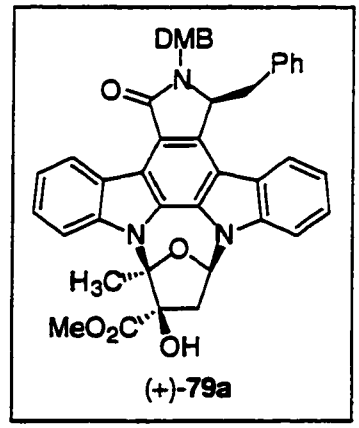
Aglycon (±)-77. To a three-necked flask equipped with a condenser were added the diazo lactam(±)-**76** (2.4 g, 6.6 mmol, 1.0 equiv), 2,2'-biindole (**11**) (1.5 g, 6.5 mmol, 1.0 equiv), $Rh_2(OAc)_4$ (30 mg, 0.07 mmol, 0.01 equiv) and pinacolone (55 mL). The whole was degassed by bubbling a stream of N_2 through the solution for 2 h. The mixture was then warmed to reflux for an additional 8 h. The reaction mixture was concentrated to a dark brown foamy solid, which was adsorbed onto silica gel and subjected to flash chromatography (50% EtOAc/hexanes eluent), affording unreacted **11** (700 mg, 47% yield) as a white powder, and aglycon (±)-**77** (1.58 g, 44% yield; 83% yield based on recovered **11**) was isolated as a pale yellow solid: mp >310 °C (dec.); IR (thin film/NaCl) 3323 (br m), 3060 (w), 2931 (w), 1642 (s), 1578 (w), 1513 (m), 1455 (m), 1411 (s), 1328 (m), 1260 (s), 1235 (m), 1143 (w), 1025 (w), 747 (s) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 11.58 (s, 1H), 11.29 (s, 1H), 9.11 (d, J = 7.9 Hz, 1H), 8.29 (d, J = 7.9 Hz, 1H), 7.82 (d, J

= 8.2 Hz, 1H), 7.69 (d, J = 8.2 Hz, 1H), 7.49 (t, J = 7.6 Hz, 1H), 7.40 (t, J = 7.6 Hz, 1H), 7.32 (t, J = 7.3 Hz, 1H), 7.17 (t, J = 7.7 Hz, 1H), 6.94-6.72 (comp m, 6H), 6.68 (m, 2H), 5.42 (t, J = 4.1 Hz, 1H), 5.26 (d, J = 15.2 Hz, 1H), 4.35 (d, J = 15.2 Hz, 1H), 3.80 (dd, J = 3.9, 14.4 Hz, 1H), 3.71 (s, 3H), 3.69 (s, 3H), 3.57 (dd, J = 4.3, 14.5 Hz, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 169.2, 148.8, 148.0, 139.4, 139.1, 135.6, 133.8, 130.7, 129.0, 127.9, 127.5, 126.1, 125.5, 125.1, 125.0, 125.0, 122.5, 121.9, 121.7, 120.0, 119.9, 118.8, 118.7, 115.2, 113.7, 112.1, 112.0, 111.2, 58.8, 55.5, 55.5, 43.1, 35.4; high resolution mass spectrum (FAB) *m/z* 552.2286 [calc'd for $C_{36}H_{30}N_3O_3$ (M+H) 552.2287].

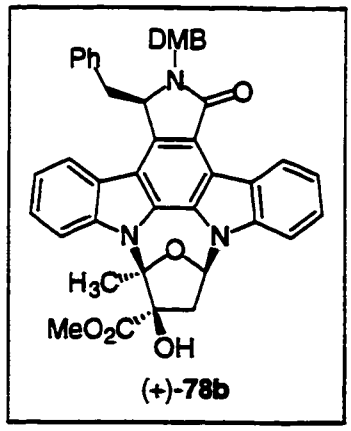
Preparation of Indolocarbazoles (+)-78a, (+)-78b, (+)-79a and (+)-79b.



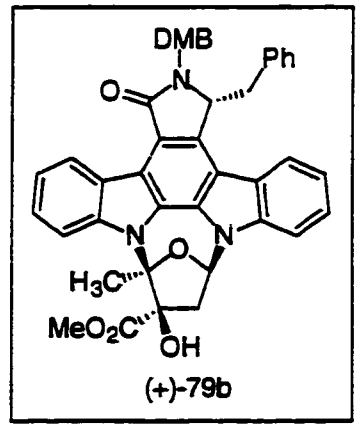
(+)-78a



(+)-79a



(+)-78b



(+)-79b

Indolocarbazoles (+)-78a, (+)-78b, (+)-79a and (+)-79b. To a refluxing solution of aglycon (±)-**78** (570 mg, 1.03 mmol, 1.0 equiv) and camphorsulfonic acid (24 mg, 0.10 mmol, 0.1.0 equiv) in 1,2-dichloroethane (34 mL) was added, via addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)-**17** (455 mg, 2.02 mmol, 2.0 equiv) in 1,2-dichloroethane (12 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% $NaHCO_3$ (20 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 20 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:2:1:1 mixture of indolocarbazoles (+)-**78a**, (+)-**78b**, (+)-**79a** and (+)-**79b** (556 mg, 76% yield). Separation of the mixture was achieved *via* HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent).

(+)-**78a:** mp 250-258 °C (dec.); $[\alpha]_D^{20}$ +87.38° (*c* 0.42, MeOH); IR (thin film/NaCl) 3002 (w), 2951 (w), 2835 (w), 1731 (m), 1676 (s), 1585 (m), 1514 (m), 1452 (s), 1391 (m), 1353 (m), 1314 (m), 1259 (s), 1237 (s), 1182 (m), 1138 (m), 1028 (m), 875 (w), 744 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.37 (d, *J* = 8.1 Hz, 1H), 8.28 (d, *J* = 7.9 Hz, 1H), 8.04 (d, *J* = 8.4 Hz, 1H), 7.78 (d, *J* = 8.3 Hz, 1H), 7.49-7.44 (comp m, 2H), 7.35 (t, *J* = 7.5 Hz, 1H), 7.26 (t, *J* = 7.5 Hz, 1H), 7.15 (dd, *J* = 4.9, 7.3 Hz, 1H), 7.03-7.01 (m, 3H), 6.93-6.80 (comp m, 5H), 5.51 (t, *J* = 4.5 Hz, 1H), 5.44 (d, *J* = 15.1 Hz, 1H), 5.24 (s, 1H), 4.26 (d, *J* = 15.1 Hz, 1H), 4.01 (s, 3H), 3.92 (dd, *J* = 3.8, 14.5 Hz, 1H), 3.74 (s, 3H), 3.73 (s, 3H), 3.58 (dd, *J* = 5.2, 14.5 Hz, 1H), 3.49 (dd, *J* = 7.4, 14.2 Hz, 1H), 2.27 (s, 3H), 2.20 (m, 1H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.5, 169.7, 150.0, 149.2, 141.1, 137.9, 136.7, 134.8, 131.3, 129.9, 129.5, 128.3, 127.0, 126.9, 126.0, 125.5, 125.1,

124.8, 123.8, 122.3, 121.1, 120.6, 120.5, 120.1, 116.9, 115.7, 115.2, 112.5, 112.4, 108.9, 99.8, 86.0, 85.8, 72.9, 64.1, 59.6, 55.7, 55.6, 53.0, 44.1, 42.9, 37.1, 23.1; high resolution mass spectrum (FAB) *m/z* 708.2709 [calc'd for $C_{43}H_{38}N_3O_7$ (M+H) 708.2710].

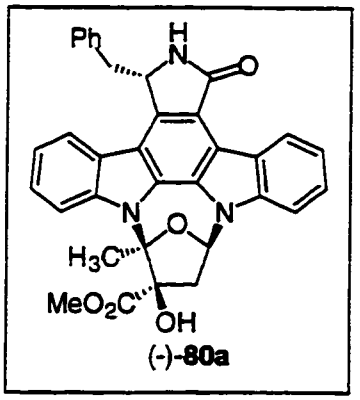
(+)-**78b:** mp 252-256 °C (dec.); $[\alpha]_D^{20}$ +89.73° (*c* 0.37, MeOH); IR (thin film/NaCl) 3369 (br w), 3008 (w), 2943 (w), 2843 (w), 1731 (m), 1671 (s), 1586 (w), 1506 (m), 1451 (s), 1386 (s), 1355 (m), 1315 (m), 1260 (s), 1195 (s), 1140 (s), 1025 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.35 (d, *J* = 7.8 Hz, 1H), 8.31 (d, *J* = 7.9 Hz, 1H), 8.05 (d, *J* = 8.5 Hz, 1H), 7.77 (d, *J* = 8.3 Hz, 1H), 7.49-7.45 (comp m, 2H), 7.36 (t, *J* = 7.5 Hz, 1H), 7.25 (t, *J* = 7.5 Hz, 1H), 7.14 (dd, *J* = 5.0, 7.4 Hz, 1H), 6.97-6.95 (comp m, 3H), 6.87-6.82 (comp m, 5H), 5.57 (dd, *J* = 3.7, 5.4 Hz, 1H), 5.42 (d, *J* = 15.1 Hz, 1H), 5.41 (s, 1H), 4.21 (d, *J* = 15.2 Hz, 1H), 4.02 (s, 3H), 3.89 (dd, *J* = 3.6, 14.5 Hz, 1H), 3.75 (s, 3H), 3.73 (s, 3H), 3.53 (dd, *J* = 7.4, 14.1 Hz, 1H), 3.45 (dd, *J* = 5.5, 14.5 Hz, 1H), 2.28 (dd, *J* = 5.0, 14.2 Hz, 1H), 2.22 (s, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 169.7, 150.3, 149.5, 141.5, 138.3, 137.0, 135.3, 131.6, 130.1, 129.6, 128.4, 127.2, 127.1, 127.0, 126.3, 125.6, 125.3, 124.8, 123.9, 122.7, 121.2, 120.9, 120.9, 120.2, 117.3, 115.7, 115.6, 112.7, 112.7, 109.0, 100.3, 86.1, 85.7, 60.0, 55.9, 53.4, 44.4, 43.1, 37.5, 30.0, 23.4; high resolution mass spectrum (FAB) *m/z* 708.2709 [calc'd for $C_{43}H_{38}N_3O_7$ (M+H) 708.2710].

(+)-**79a:** mp >280 °C (dec.); $[\alpha]_D^{20}$ +76.94° (*c* 0.36, MeOH); IR (thin film/NaCl) 3384 (br w), 3013 (w), 2928 (w), 2833 (w), 1731 (m), 1671 (s,), 1581 (m), 1506 (m), 1451 (s), 1396 (s), 1310 (s), 1250 (s), 1235 (s), 1140 (m), 1025 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.63 (d, *J* = 8.1 Hz, 1H), 8.32 (d, *J* = 7.9 Hz, 1H), 7.91 (t, *J* = 8.4 Hz, 2H), 7.54 (t, *J* = 7.7 Hz, 1H), 7.41-

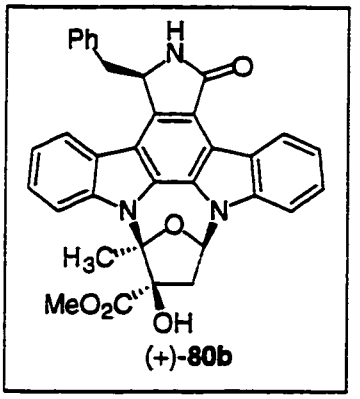
7.35 (m, 2H), 7.24 (t, J = 7.5 Hz, 1H), 7.18 (dd, J = 4.9, 7.4 Hz, 1H), 7.01 (m, 3H), 6.91 (m, 3H), 6.84 (m, 2H), 5.48 (m, 2H), 5.23 (s, 1H), 4.30 (d, J = 15.1 Hz, 1H), 4.00 (s, 3H), 3.93 (dd, J = 3.7, 14.5 Hz, 1H), 3.73 (s, 3H), 3.72 (s, 3H), 3.61 (dd, J = 5.0, 14.5 Hz, 1H), 3.49 (dd, J = 7.5, 14.1 Hz, 1H), 2.25-2.20 (comp m, 4H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.8, 170.3, 150.5, 149.8, 141.1, 138.5, 137.1, 135.8, 131.8, 130.3, 128.7, 127.8, 127.6, 127.3, 127.2, 126.3, 126.0, 125.9, 123.5, 123.2, 121.4, 121.1, 120.8, 120.4, 118.1, 115.2, 114.8, 113.1, 112.9, 110.3, 100.1, 86.5, 86.1, 59.7, 56.1, 56.0, 53.3, 44.5, 43.1, 43.1, 37.5, 23.5; high resolution mass spectrum (FAB) *m/z* 708.2709 [calc'd for $C_{43}H_{38}N_3O_7$ (M+H) 708.2710].

(+)-**79b**: mp 232-241 °C (dec.); $[\alpha]_D^{20}$ +71.34° (*c* 0.24, MeOH); IR (thin film/NaCl) 3384 (br w), 3013 (w), 2928 (w), 2833 (w), 1731 (m), 1671 (s,), 1581 (m), 1506 (m), 1451 (s), 1396 (s), 1310 (s), 1250 (s), 1235 (s), 1140 (m), 1025 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.64 (d, J = 8.1 Hz, 1H), 8.32 (d, J = 7.9 Hz, 1H), 7.91 (t, J = 8.3 Hz, 2H), 7.55 (t, J = 7.7 Hz, 1H), 7.41-7.33 (m, 2H), 7.24 (t, J = 7.5 Hz, 1H), 7.19 (dd, J = 4.9, 7.5 Hz, 1H), 7.01 (m, 3H), 6.90 (m, 3H), 6.84 (m, 2H), 5.49 (m, 2H), 5.23 (s, 1H), 4.29 (d, J = 15.1 Hz, 1H), 4.00 (s, 3H), 3.93 (dd, J = 3.7, 14.5 Hz, 1H), 3.73 (s, 3H), 3.72 (s, 3H), 3.61 (dd, J = 4.9, 14.5 Hz, 1H), 3.49 (dd, J = 7.5, 14.0 Hz, 1H), 2.26-2.22 (comp m, 4H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 170.0, 150.2, 149.6, 141.3, 138.4, 136.9, 135.2, 131.8, 130.2, 128.5, 127.4, 127.1, 127.0, 126.3, 125.7, 125.2, 124.1, 122.5, 121.5, 121.0, 117.2, 120.5, 116.0, 115.5, 113.2, 112.9, 109.1, 100.1, 86.3, 86.0, 73.3, 64.3, 59.9, 56.2, 56.1, 53.2, 44.7, 43.2, 37.4, 23.3; high resolution mass spectrum (FAB) *m/z* 708.2714 [calc'd for $C_{43}H_{38}N_3O_7$ (M+H) 708.2710].

Preparation of (-)-7-(*S*)-Benzyl K252a (80a) and (+)-7-(*R*)-Benzyl K252a (80b).



(-)-**80a**



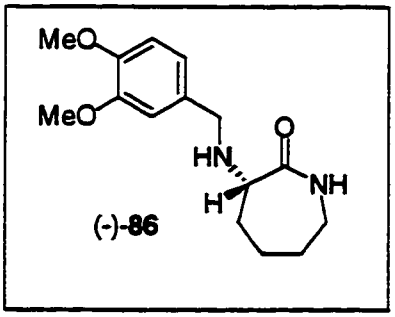
(+)-**80b**

Representative Procedure: (-)-7-(*S*)-Benzyl K252a (80a). A solution of protected amide (+)-**78a** (50 mg, 0.07 mmol, 1.0 equiv) and anisole (760 µL, 7 mmol, 100 equiv) in CH_2Cl_2 (2 mL) was treated dropwise with TFA (1 mL). After stirring at rt for 12 h, the reaction was quenched with 20% $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**80a** (32 mg, 81% yield) as a white film: $[\alpha]_D^{20}$ -26.30° (*c* 0.27, MeOH); IR (thin film/NaCl) 3260 (br w), 2923 (w), 2852 (w), 1731 (m), 1669 (s), 1582 (m), 1492 (w), 1451 (s), 1389 (w), 1314 (m), 1256 (s), 1138 (m), 1076 (s), 1017 (m), 742 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.32 (d, *J* = 8.1 Hz, 1H), 8.39 (d, *J* = 7.6 Hz, 1H), 8.06 (d, *J* = 8.4 Hz, 1H), 7.78 (d, *J* = 8.2 Hz, 1H), 7.52-7.41 (comp m, 4H), 7.28-7.18 (comp m, 5H), 7.17 (m, 2H), 5.67 (ddd, *J* = 1.0, 3.4, 7.9 Hz, 1H), 5.32 (s, 1H), 4.02 (s, 3H), 3.91 (dd, *J* = 3.3, 14.2 Hz, 1H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 3.07 (dd, *J* = 8.0, 14.1 Hz, 1H), 2.27 (s, 3H), 2.24 (dd, *J* = 4.9, 14.2 Hz, 1H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.8, 171.6, 141.4, 138.3, 138.2, 137.3, 130.3, 130.0, 129.0, 127.4, 127.3, 126.3, 125.8, 125.4, 125.3, 124.2, 122.7, 121.5, 120.8,

120.3, 117.4, 116.1, 115.7, 109.2, 100.2, 86.4, 86.1, 58.4, 58.3, 53.4, 43.3, 41.0, 30.1, 23.4; high resolution mass spectrum (FAB) *m/z* 558.2027 [calc'd for $C_{34}H_{28}N_3O_5$ (M+H) 558.2029].

(+)-**80b**: $[\alpha]_D^{20}$ +99.59° (*c* 0.25, MeOH); IR (thin film/NaCl) 3407 (br w), 3304 (br w), 2964 (w), 2928 (w), 2861 (w), 1738 (s), 1676 (s), 1588 (m), 1449 (s), 1393 (m), 1315 (m), 1202 (m), 1084 (m), 874 (w), 743 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.34 (d, *J* = 8.1 Hz, 1H), 8.41 (d, *J* = 7.8 Hz, 1H), 8.07 (d, *J* = 8.4 Hz, 1H), 7.79 (d, *J* = 8.3 Hz, 1H), 7.52-7.42 (comp m, 4H), 7.28-7.12 (comp m, 7H), 5.70 (dd, *J* = 3.0, 8.4 Hz, 1H), 5.42 (s, 1H), 4.02 (s, 3H), 3.92 (dd, *J* = 3.0, 14.1 Hz, 1H), 3.53 (dd, *J* = 7.5, 14.1 Hz, 1H), 2.91 (dd, *J* = 8.4, 14.1 Hz, 1H), 2.32 (dd, *J* = 5.0, 14.2 Hz, 1H), 2.24 (s, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 171.6, 141.5, 138.4, 138.3, 137.5, 130.1, 129.8, 128.9, 127.3, 127.2, 126.3, 125.7, 125.4, 125.0, 124.0, 122.8, 121.3, 120.8, 120.3, 117.5, 115.8, 109.0, 100.4, 86.1, 85.8, 58.5, 58.4, 53.4, 43.2, 43.2, 41.1, 41.1, 23.5; high resolution mass spectrum (FAB) *m/z* 558.2028 [calc'd for $C_{34}H_{28}N_3O_5$ (M+H) 558.2029].

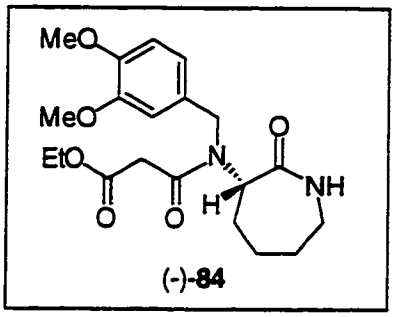
Preparation of Amine (-)-86.



(-)-**86**

Amine (-)-86. To a solution of L-(-)-α-amino-ε-caprolactam (**85**) (3.0 g, 23.4 mmol, 1.0 equiv), in MeOH (10 mL) was added 3,4-dimethoxybenzaldehyde (2.73 g, 16.4 mmol, 0.7 equiv) as a solution in EtOH (40 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in EtOH (60 mL) and $NaBH_4$ (890 mg, 23.4 mmol, 1.0 equiv) was added portionwise over a 15 minute period. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, the EtOH was removed under reduced pressure and the residue was dissolved in 1 N HCl (10 mL). A white precipitate quickly formed, which was removed by filtration. The white solid was then dissolved in 1 N aqueous NaOH (10 mL). The basic solution was extracted with CH_2Cl_2 (3 x 10 mL) and the combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield the protected amine (-)-**86** (3.93 g, 86% yield) as a white crystalline solid: mp 142-144 °C; $[\alpha]_D^{20}$ -9.26° (*c* 0.68, MeOH); IR (thin film/NaCl) 3299 (w), 2923 (m), 2838 (w), 1656 (s), 1511 (s), 1456 (m), 1411 (m), 1260 (s), 1225 (m), 1160 (m), 1130 (m), 1030 (m), 805 (m), 760 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.96 (m, 1H), 6.88 (m, 1H), 6.82 (m, 1H), 6.03 (s, 1H), 3.90 (s, 1H), 3.88 (s, 3H), 3.83 (d, *J* = 12.8 Hz, 1H), 3.69 (d, *J* = 12.8 Hz, 1H), 3.33 (d, *J* = 10.6 Hz, 1H), 3.21 (m, 2H), 2.00 (d, *J* = 11.0 Hz, 2H), 1.79 (m, 1H), 1.58 (comp m, 2H), 1.42 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 178.4, 148.8, 147.8, 132.8, 120.2, 111.3, 110.8, 59.4, 55.8, 55.7, 51.9, 41.8, 31.8, 28.9, 28.0; high resolution mass spectrum (EI) *m/z* 278.1630 [calc'd for $C_{15}H_{22}N_2O_3$(M+) 278.1630].

Preparation of Amide (-)-84.

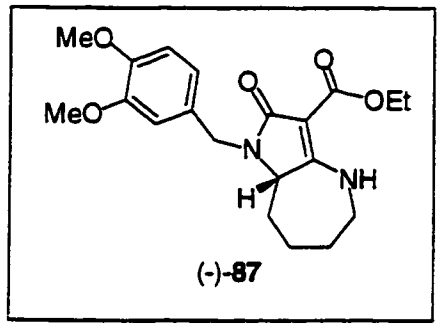


(-)-84

Amide (-)-84: A three-necked flask equipped with an addition funnel was charged with the amine (-)-**86** (7.1 g, 25.5 mmol, 1.0 equiv), ethyl hydrogen malonate (3.37 g, 25.5 mmol, 1.0 equiv), and CH_2Cl_2 (100 mL). The flask was cooled to 0°C and a solution of 1,3-dicyclohexylcarbodiimide (5.26 g, 25.5 mmol, 1.0 equiv) in CH_2Cl_2 (70 mL) was added dropwise through the addition funnel over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h at room temperature. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (250 mL). The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to yield amide (-)-**84** (9.7 g, 97% yield) as a clear, colorless oil: $[\alpha]_D^{20}$ -5.20° (*c* 0.25, MeOH); IR (thin film/NaCl) 3334 (br w), 2928 (m), 2853 (w), 1736 (s), 1676 (s), 1651 (s), 1511 (s), 1461 (m), 1421 (m), 1250 (s), 1145 (m), 1020 (m), 794 (w), 750 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.85-6.70 (comp m, 3H), 6.62 (s, 1H), 5.27 (m, 1H), 4.71 (d, *J* = 18.4 Hz, 1H), 4.46 (d, *J* = 18.4 Hz, 1H), 4.18-4.04 (comp m, 2H), 3.82 (s, 2.6H), 3.81 (s, 2.6H), 3.80 (s, 0.4H), 3.78 (s, 0.4H), 3.50-3.15 (comp m, 4H), 1.90 (m, 1H), 1.79 (m, 2H), 1.62 (m, 2H), 1.35 (m, 1H), 1.24 (t, *J* = 7.1 Hz, 0.8H), 1.18 (t, *J* = 7.1 Hz, 2.2H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 175.3, 174.8, 168.4, 167.7, 167.3, 149.3, 148.9, 148.0, 147.6, 130.5,

118.9, 117.6, 111.4, 111.0, 110.7, 109.0, 63.8, 63.6, 61.3, 61.0, 57.4, 55.8, 55.7, 55.7, 49.2, 42.3, 42.0, 41.7, 41.6, 41.2, 28.8, 28.6, 28.0, 27.9, 13.9, 13.8; high resolution mass spectrum (EI) *m/z* 392.1939 [calc'd for $C_{20}H_{28}N_2O_6$ (M+) 392.1947].

Preparation of Lactam (-)-87.

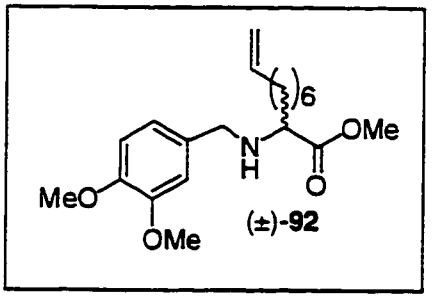


(-)-87

Lactam (-)-87. A three-necked flask was charged with EtOH (30 mL). Sodium metal (562 mg, 24.4 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester (-)-**84** (10.2 g, 26.0 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in EtOH (35 mL). The mixture was brought to reflux for 5 min and then allowed to cool to rt. The EtOH was removed under reduced pressure and the aqueous layer was extracted with EtOAc (3 x 100 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to a yellowish oil, which was subjected to flash chromatography (50% EtOAc/hexanes eluent) to furnish (-)-**87** (6.7 g, 70% yield) as a colorless oil: $[\alpha]_D^{20}$ -40.00° (*c* 1.25, MeOH); IR (thin film/NaCl) 175.6, 167.6, 166.9, 149.3, 148.3, 130.2, 120.2, 111.2, 111.0, 89.9, 59.7, 58.6, 56.0, 55.8, 45.4, 42.5, 29.2, 28.8, 27.9, 14.5 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 8.62 (br s, 1H), 6.78-6.73 (comp m, 3H), 5.07 (d, J = 15.2 Hz, 1H), 4.34-4.24 (comp m, 2H),

3.93 (d, J = 15.2 Hz, 1H), 3.84 (s, 3H), 3.83 (s, 3H), 3.48 (m, 1H), 3.14 (m, 1H), 2.22 (m, 1H), 2.06 (m, 1H), 1.84 (m, 1H), 1.46 (m, 2H), 1.36 (t, J = 7.1 Hz, 3H), 1.25 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 175.6, 167.6, 166.9, 149.3, 148.3, 130.2, 120.2, 111.2, 111.0, 89.9, 59.7, 58.6, 56.0, 55.8, 45.4, 42.5, 29.2, 28.8, 27.9, 14.5; high resolution mass spectrum (EI) *m/z* 374.1835 [calc'd for $C_{20}H_{26}N_2O_5$(M+) 374.1842].

Preparation of Amine (±)-92.

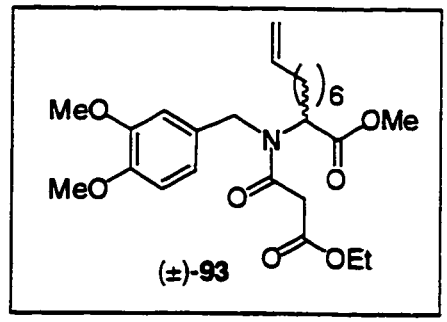


(±)-92

Amine (±)-92. To a solution of amine (±)-**91** (35.0 g, 176 mmol, 1.0 equiv), in EtOH (200 mL) was added 3,4-dimethoxybenzaldehyde (29.2 g, 176 mmol, 1.0 equiv) as a solution in EtOH (100 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in a 1:1 mixture of MeOH (295 mL) and THF (295 mL), and $NaBH_4$ (6.7 g, 176 mmol, 1.0 equiv) was added portionwise over a 15 minute period. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, solvent was removed under reduced pressure and the oily residue was taken up in CH_2Cl_2 (400 mL) and was washed with saturated NH_4Cl solution (400 mL). The aqueous layer was then extracted with additional CH_2Cl_2 (2 x 100 mL) and the combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield

the protected amine (±)-**92** (56.2 g, 92% yield) as an opaque pale yellow oil: IR (thin film/NaCl) 2933 (s), 2854 (m), 1737 (s), 1510 (m), 1450 (m), 1266 (m), 1236 (m), 1160 (m), 1024 (m), 909 (w), 810 (w), 755 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.90-6.79 (comp m, 3H), 5.79 (ddt, *J* = 6.7, 10.3, 17.0 Hz, 1H), 5.00-4.90 (comp m, 2H), 3.88 (s, 3H), 3.86 (s, 3H), 3.75 (d, *J* = 12.8 Hz, 1H), 3.72 (s, 3H), 3.56 (d, *J* = 12.8 Hz, 1H), 3.24 (t, *J* = 6.7 Hz, 1H), 2.04-2.00 (comp m, 2H), 1.78 (br s, 1H), 1.66-1.56 (comp m, 2H), 1.40-1.25 (comp m, 8H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 176.1, 148.9, 148.1, 139.1, 132.4, 120.4, 114.2, 111.4, 110.9, 60.5, 55.9, 55.8, 51.9, 51.6, 35.7, 33.5, 29.2, 28.9, 28.8, 25.8; high resolution mass spectrum (EI) *m/z* 349.2247 [calc'd for $C_{20}H_{31}NO_4$ (M+) 349.2253].

Preparation of Amide (±)-93.

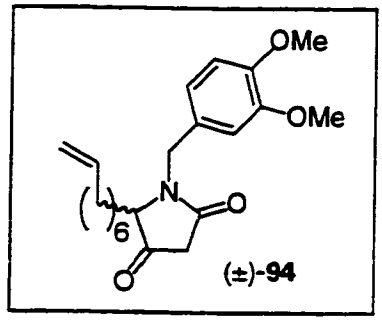


(±)-**93**

Amide (±)-93: A three-necked flask equipped with an addition funnel was charged with the amine (±)-**92** (6.5 g, 18.6 mmol, 1.0 equiv), ethyl hydrogen malonate (2.46 g, 18.6 mmol, 1.0 equiv), and CH_2Cl_2 (60 mL). The flask was cooled to 0°C and 1,3-dicyclohexylcarbodiimide (3.83 g, 18.6 mmol, 1.0 equiv) in CH_2Cl_2 (60 mL) was added dropwise through the addition funnel over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h

at room temperature. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (100 mL). The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to an oily residue. The residue was taken up in a minimum of acetone (10 mL) and filtered to remove the remaining urea by-product. Removal of the acetone under reduced pressure afforded amide (±)-**93** (7.2 g, 84% yield) as a clear, light yellow oil: IR (thin film/NaCl) 2928 (m), 2854 (w), 1739 (s), 1655 (s), 1514 (m), 1443 (m), 1260 (m), 1259 (m), 1149 (m), 1030 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.87-6.70 (comp m, 3H), 5.74 (ddt, J = 6.6, 10.2, 16.9 Hz, 1H), 4.95-4.86 (comp m, 2H), 4.61-4.55 (comp m, 2H), 4.46-4.41 (m, 1H), 4.24-4.11 (comp m, 2H), 3.84 (s, 1.8H), 3.83 (s, 1.8H), 3.82 (s, 1.2H), 3.80 (s, 1.2H), 3.59 (s, 1.8H), 3.49 (s, 1.2H), 3.43-3.35 (comp m, 2H), 2.00-1.92 (m, 3H), 1.72 (m, 1H), 1.30-1.15 (m, 11H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.2, 170.7, 167.2, 167.2, 149.2, 148.6, 148.4, 147.9, 138.8, 138.7, 130.1, 128.5, 120.1, 118.7, 114.1, 114.0, 111.4, 111.0, 110.5, 109.7, 61.4, 61.3, 60.6, 58.3, 55.8, 55.7, 55.7, 52.1, 51.9, 50.6, 46.2, 41.8, 41.4, 33.5, 33.4, 29.5, 29.2, 29.0, 28.9, 28.7, 28.6, 28.5, 26.3, 26.0, 13.9; high resolution mass spectrum (EI) *m/z* 463.2564 [calc'd for $C_{25}H_{37}NO_7$ (M+) 463.2570].

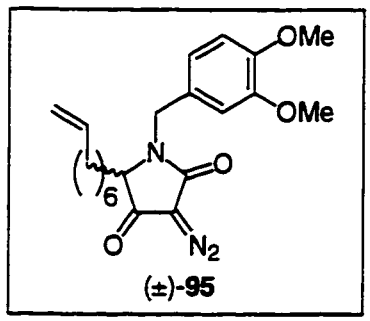
Preparation of Lactam (±)-94.



Lactam (±)-94. A three-necked flask was charged with EtOH (4 mL). Sodium metal (70 mg, 3.05 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester (±)-**93** (1.5 g, 3.24 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in EtOH (4 mL). The mixture was brought to reflux for 30 min and then allowed to cool to rt. The EtOH was removed under reduced pressure and the residue was dissolved in H_2O (5 mL). The aqueous layer was washed with EtOAc (5 mL), and acidified to a pH of 2 with 2 N HCl (2 mL). The acidic solution was extracted with CH_2Cl_2 (3 x 10 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield a yellow oil. A suspension of the oil in CH_3CN (255 mL) and H_2O (0.5 mL) was warmed to reflux open to the air for three hours. The mixture was cooled to rt and the CH_3CN/H_2O mixture was removed at reduced pressure to yield lactam (±)-**94** as a dark yellow oil (916 mg, 84% yield) which solidified upon standing to a dark yellow glassy solid: IR (thin film/NaCl) 3073 (w), 2928 (br s), 2853 (s), 1772 (s), 1690 (s), 1510 (s), 1410 (s), 1255 (s), 1140 (s), 1030 (s), 909 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.80 (m, 3H), 5.76 (m, 1H), 5.15 (d, J = 14.7 Hz, 1H), 4.94 (m, 2H), 3.92 (d, J = 14.7 Hz, 1H), 3.84 (m, 6H), 3.77 (br s, 1H), 3.03 (m, 2H), 2.00 (m, 2H), 1.83 (m, 1H), 1.68 (m, 1H), 1.33-1.04 (comp m,

8H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.9, 168.8, 149.3, 148.8, 138.7, 127.7, 120.8, 114.3, 111.4, 111.0, 65.7, 55.9, 55.8, 43.6, 41.4, 33.5, 29.2, 28.6, 28.6, 23.2; high resolution mass spectrum (EI) *m/z* 359.2099 [calc'd for $C_{21}H_{29}NO_4$ (M+) 359.2097].

Preparation of Diazo lactam (±)-95.

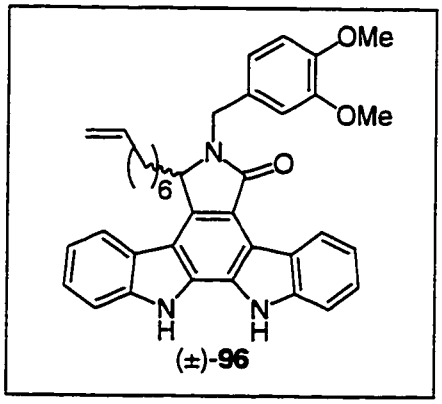


(±)-**95**

Diazo lactam (±)-95. A stirred solution of lactam (±)-**94** (18.4 g, 51.3 mmol, 1.0 equiv), *p*-ABSA (13.6 g, 56.4 mmol, 1.1 equiv), and CH_3CN (320 mL) was cooled to 0°C and Et_3N (20.8 mL, 149.4 mmol, 3.0 equiv) was added dropwise to the mixture. After gradually warming to rt, the mixture was stirred for an additional 3 h and the volatiles were removed under reduced pressure. The residue was subjected to flash chromatography (50% EtOAc/hexanes eluent) to provide diazo lactam (±)-**95** (16.7 g, 85% yield) as a bright yellow foam: IR (thin film/NaCl) 2923 (m), 2853 (w), 2116 (s), 1687 (s), 1510 (m), 1045 (s), 1355 (m), 1265 (m), 1235 (m), 1140 (m), 1024 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.79 (comp m, 3H), 5.80 (ddt, *J* = 6.7, 10.3, 17.0 Hz, 1H), 5.09 (d, *J* = 14.9 Hz, 1H), 5.02-4.93 (comp m, 2H), 3.98 (d, *J* = 14.9 Hz, 1H), 3.88 (s, 6H), 3.78 (dd, *J* = 3.1, 5.6 Hz, 1H), 2.06-2.00 (comp m, 2H), 1.90 (m, 1H), 1.74 (m, 1H), 1.40-1.16 (comp m, 8H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 189.3, 161.9, 149.4, 148.9, 138.9,

128.0, 120.7, 114.3, 111.3, 111.1, 65.5, 63.5, 56.0, 55.9, 44.3, 33.6, 29.3, 28.8, 28.7, 28.5, 22.8; high resolution mass spectrum (EI) *m/z* 385.1993 [calc'd for $C_{21}H_{27}N_3O_4$ (M+) 385.2002].

Preparation of Aglycon (±)-96.

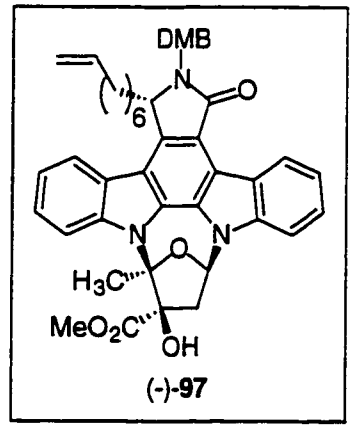


(±)-**96**

Aglycon (±)-96. To a three-necked flask equipped with a condenser were added the diazo lactam (±)-**95** (10.0 g, 26 mmol, 1.0 equiv), 2,2'-biindole (5.95 g, 26 mmol, 1.0 equiv), $Rh_2(OAc)_4$ (110 mg, 0.26 mmol, 0.01 equiv) and pinacolone (260 mL). The whole was degassed by bubbling a stream of N_2 through the solution for 2 h. The mixture was then warmed to reflux for an additional 8 h. The reaction mixture was concentrated to a dark brown foamy solid, which was adsorbed onto silica gel and subjected to flash chromatography (50% EtOAc/hexanes eluent), affording unreacted **11** (2.5 g, 42% yield) as a yellow powder, and aglycon (±)-**96** (4.8 g, 33% yield; 57% yield based on recovered **11**) was isolated as a pale yellow solid: mp 190-196 °C (dec.); IR (thin film/NaCl) 3348 (m), 3068 (w), 2933 (m), 2847 (w), 1641 (s), 1510 (m), 1461 (m), 1410 (m), 1321 (m), 1261 (m), 1140 (w), 1025 (w), 750 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 11.90 (s, 1H), 11.68 (s, 1H), 9.54 (d, J = 7.9 Hz, 1H), 8.42 (d, J = 7.8 Hz, 1H), 8.19 (d, J = 8.1

Hz, 1H), 8.07 (d, J = 8.1 Hz, 1H), 7.76 (t, J = 7.5 Hz, 2H), 7.52 (t, J = 7.2 Hz, 2H), 7.42 (m, 1H), 7.33 (m, 2H), 5.88 (ddt, J = 6.8, 10.1, 17.0 Hz, 1H), 5.52 (m, 1H), 5.48 (d, J = 15.1 Hz, 1H), 5.17 (m, 1H), 4.74 (d, J = 15.0 Hz, 1H), 4.03 (s, 3H), 4.01 (s, 3H), 2.58 (s, 3H), 2.30 (m, 2H), 1.26-1.04 (comp m, 7H), 0.62 (m, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 169.5, 148.8, 148.1, 139.4, 139.3, 138.5, 134.2, 130.8, 128.1, 125.6, 125.3, 125.2, 124.9, 122.7, 122.0, 121.7, 120.1, 119.9, 119.0, 118.6, 115.3, 114.3, 113.5, 112.0, 111.9, 111.4, 58.9, 55.5, 55.4, 43.2, 32.8, 28.4, 28.0, 27.9, 21.1; high resolution mass spectrum (EI) *m/z* 571.2829 [calc'd for $C_{37}H_{37}N_3O_3$(M+) 571.2835].

Preparation of Indolocarbazole (-)-97.

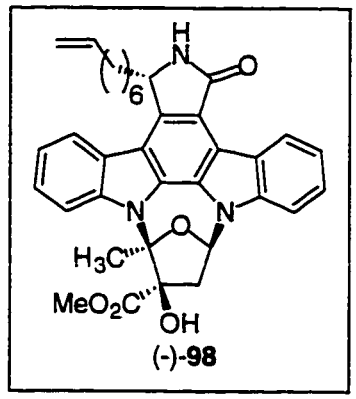


(-)-**97**

Indolocarbazole (-)-97. To a refluxing solution of aglycon (±)-**96** (325 mg, 0.57 mmol, 1.0 equiv) and camphorsulfonic acid (13 mg, 0.057 mmol, 0.1 equiv) in 1,2-dichloroethane (19 mL) was added, *via* addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)**17** (250 mg, 1.14 mmol, 2.0 equiv) in 1,2-dichloroethane (12 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with a 10% $NaHCO_3$ solution (10 mL). The aqueous layer was

washed with CH_2Cl_2 (3 x 10 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:2:1:1 mixture of indolocarbazoles (314 mg, 76% yield). Separation of the mixture was achieved *via* HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent) to provide the desired (-)-**97** (104 mg, 25% yield) as a white powder: mp >290 °C (dec.); $[\alpha]_D^{20}$ -3.67° (*c* 0.30, MeOH); IR (thin film/NaCl) 3298 (br w), 2930 (m), 2854 (w), 1733 (m), 1673 (s), 1646 (s), 1585 (m), 1515 (m), 1459 (s), 1392 (m), 1258 (s), 1199 (m), 1028 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.51 (d, *J* = 8.0 Hz, 1H), 8.09 (d, *J* = 7.9 Hz, 1H), 7.99 (d, *J* = 8.5 Hz, 1H), 7.81 (d, *J* = 8.2 Hz, 1H), 7.51 (ddd, *J* = 1.1, 7.1, 8.2 Hz, 1H), 7.43 (ddd, *J* = 1.0, 7.3, 8.3 Hz, 1H), 7.31 (m, 2H), 7.16 (dd, *J* = 4.9, 7.4 Hz, 1H), 7.10 (m, 1H), 7.00 (dd, *J* = 1.8, 8.2 Hz, 1H), 6.93 (s, 1H), 6.91 (s, 1H), 5.60 (tdd, *J* = 6.8, 10.2, 17.0 Hz, 1H), 5.33 (m, 2H), 5.24 (s, 1H), 4.78 (comp m, 2H), 4.42 (d, J = 15.2 Hz, 1H), 4.01 (s, 3H), 3.77 (s, 3H), 3.76 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.53 (m, 1H), 2.41 (m, 1H), 2.22 (comp m, 4H), 1.78 (m, 1H), 1.02 (m, 7H), 0.55 (m, 1H); ^{13}C NMR (125 MHz, CD_2Cl_2) δ 174.4, 170.0, 149.8, 148.9, 140.7, 139.4, 137.6, 135.1, 131.1, 129.3, 126.9, 126.0, 125.4, 124.9, 124.7, 123.7, 122.5, 120.9, 120.5, 120.4, 120.3, 115.1, 114.8, 114.1, 111.9, 111.8, 108.1, 99.1, 85.5, 85.4, 59.7, 56.2, 54.3, 44.0, 42.4, 33.9, 29.6, 29.5, 29.2, 29.1, 29.1, 23.2, 22.1, 21.9; high resolution mass spectrum (FAB) *m/z* 727.6851 [calc'd for $C_{44}H_{46}N_3O_7$ (M+H) 727.6848].

Preparation of (-)-7-(*S*)-Octenyl K252a (98).

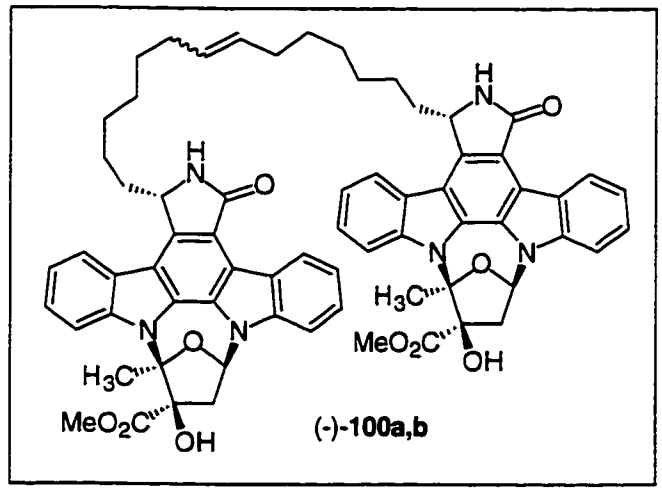


(-)-**98**

(-)-7-(*S*)-Octenyl K252a (98). A solution of protected amide (-)-**97** (30 mg, 0.041 mmol, 1.0 equiv) and anisole (446 mg, 4.1 mmol, 100 equiv) was treated with TFA (1.5 mL). After stirring at rt for 12 h, the reaction was quenched with a 20% $NaHCO_3$ solution (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**98** (5.0 mg, 25% yield) as a white film: mp 305-307 °C (dec.); $[\alpha]_D^{20}$ -3.67° (*c* 0.30, MeOH); IR (thin film/NaCl) 3355 (br w), 2980 (w), 2876 (m), 2851 (w), 1728 (m), 1681 (s), 1590 (w), 1463 (s), 1401 (m), 1392 (m), 1369 (m), 1297 (m), 1258 (m), 1224 (m), 1199 (m), 1019 (w), 743 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.54 (d, *J* = 7.8 Hz, 1H), 8.12 (d, *J* = 8.0 Hz, 1H), 7.97 (d, *J* = 8.4 Hz, 1H), 7.58 (s, 1H), 7.85 (d, *J* = 8.3 Hz, 1H), 7.49 (m, 2H), 7.41 (ddd, *J* = 1.0, 7.4, 8.0 Hz, 1H), 7.33 (ddd, *J* = 1.1, 7.4, 8.4 Hz, 1H), 7.17 (dd, *J* = 4.8, 7.5 Hz, 1H), 5.76 (tdd, *J* = 6.7, 10.0, 17.1 Hz, 1H), 5.48 (m, 2H), 4.85 (comp m, 2H), 4.02 (s, 3H), 3.54 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.55 (m, 1H), 2.44 (dd, *J* = 5.0, 14.2 Hz, 1H), 2.32 (s, 3H), 1.89-1.36 (comp m, 11H); ^{13}C NMR (125 MHz, CD_2Cl_2) δ 174.0, 172.1, 141.7, 139.3, 138.1, 130.6, 129.7, 129.1, 126.6, 125.8, 125.0,

124.9, 124.7, 123.7, 122.5, 120.9, 120.5, 120.4, 120.3, 115.6, 109.8, 100.1, 85.7, 85.6, 61.9, 52.0, 43.6, 38.8, 24.1, 22.2, 19.6, 19.4, 11.9; high resolution mass spectrum (FAB) *m/z* 578.6582 [calc'd for $C_{35}H_{36}N_3O_5$ (M+H) 578.6580].

Preparation of Dimer (-)-100a,b.



Dimer (-)-100a,b. A solution of olefin (-)-**98** (30 mg, 0.05 mmol, 1.0 equiv) in CH_2Cl_2 (1 mL) was treated with Grubb's catalyst **99** (10 mg, 0.01 mmol, 0.2 equiv). After stirring at rt for 24 h, the volatiles were removed at reduced pressure, affording a residue that was subjected to flash chromatography (50% EtOAc/hexanes eluent) to provide (-)-**100a,b** (15 mg, 52% yield) as a brownish film: mp >330 °C (dec.); $[\alpha]_D^{20}$ -52.20° (*c* 0.10, MeOH); IR (thin film/NaCl) 3227 (br w), 2861 (w), 2840 (w), 2816 (w), 1725 (m), 1672 (s), 1510 (w), 1449 (s), 1411 (w), 1386 (w), 1297 (w), 1232 (m), 1258 (m), 1197 (m), 1083 (m), 1005 (s), 748 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.41 (d, J = 8.1 Hz, 1H), 8.18 (d, J = 7.8 Hz, 1H), 8.01 (d, J = 8.5 Hz, 1H), 7.79 (m, 2H), 7.47 (m, 2H), 7.44 (ddd, J = 1.0, 7.3, 8.1 Hz, 1H), 7.34 (ddd, J = 1.1, 7.4, 8.0 Hz, 1H), 7.15 (dd, J = 4.7, 7.4

Hz, 1H), 5.38 (m, 2H), 5.29 (m, 1H), 4.01 (s, 3H), 3.50 (dd, J = 5.0, 14.1 Hz, 1H), 2.53 (m, 2H), 2.21 (m, 4H), 1.91-0.82 (comp m, 11H); ^{13}C NMR (125 MHz, CD_2Cl_2) δ 174.0, 170.1, 150.2, 149.7, 141.4, 138.2, 131.5, 129.7, 129.1, 126.6, 125.8, 125.0, 124.9, 124.7, 123.6, 122.0, 121.0, 120.9, 120.7, 120.5, 118.4, 117.1, 113.7, 109.9, 100.2, 85.5, 85.4, 59.7, 55.9, 51.8, 43.6, 42.4, 39.1, 24.1, 22.6, 22.5, 22.4, 21.9; high resolution mass spectrum (FAB) *m/z* 1127.9116 [calc'd for $C_{68}H_{68}N_6O_{10}$ (M+H) 1127.9112].

2.8 Notes and References.

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(16) The author is indebted to Roger W. Babbitt for the photomicrographs and CHOC assays.

(17) The author is indebted to Daniel Elbaum and David R. Stover of Kinetix Pharmaceuticals, Inc., 200 Boston Avenue, Suite 4700, Medford, MA 02155 for the kinase inhibition assays.

(18) The kinase inhibition assays were performed as follows: Kinase domains were expressed as Glutathione S-transferase (GST) fusion proteins in *Spodoptera frugiperda* (SF9) or High-Five cells, using the Bac-to-Bac expression system (Life Technologies, Paisley, UK). The proteins were then purified to near homogeneity by glutathione-affinity chromatography. The kinase was incubated with [^{33}P]-ATP in a 96-well plate coated with substrate (i.e. poly[Glu, Tyr]4:1). The kinase activity was then measured by a 96-well scintillation counter (i.e. Microbeta, Wallac or Top-Count, Packard). Inhibition was quantified by comparing the relative activity of kinase in the presence and absence of various concentrations of inhibitor. The IC_{50}

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APPENDIX ONE: SYNTHETIC SUMMARY FOR METHYL, BENZYL, OCTENYL AND DIMERIC K252a

Figure A.1.1 The Synthesis of (-)-**52a**, (+)-**52b**, (-)-**80a**, and (+)-**80b**.

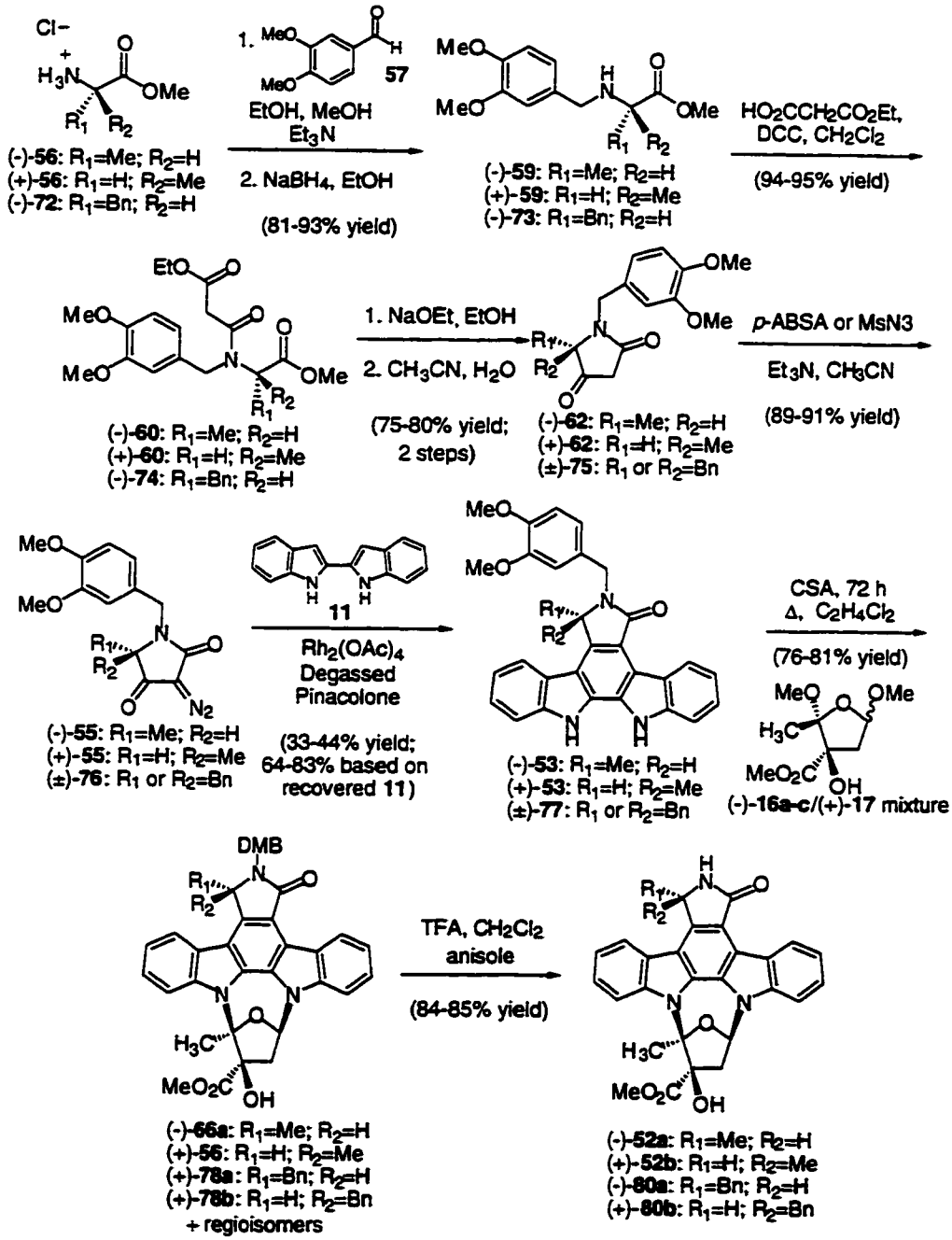
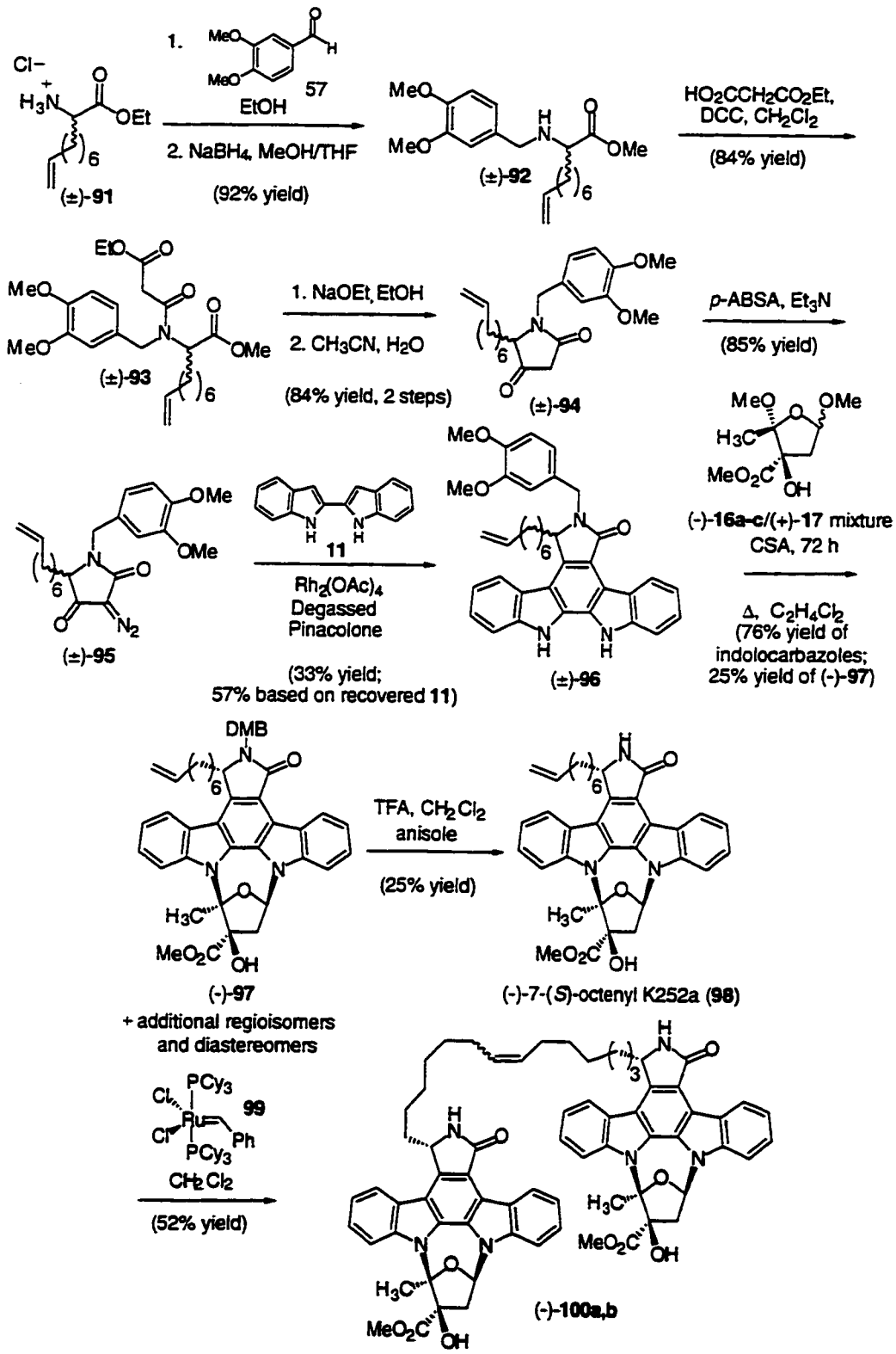


Figure A.1.2 The Synthesis of (-)-**98** and (-)-**100a,b**.



APPENDIX TWO: SPECTRA RELEVANT TO CHAPTER TWO

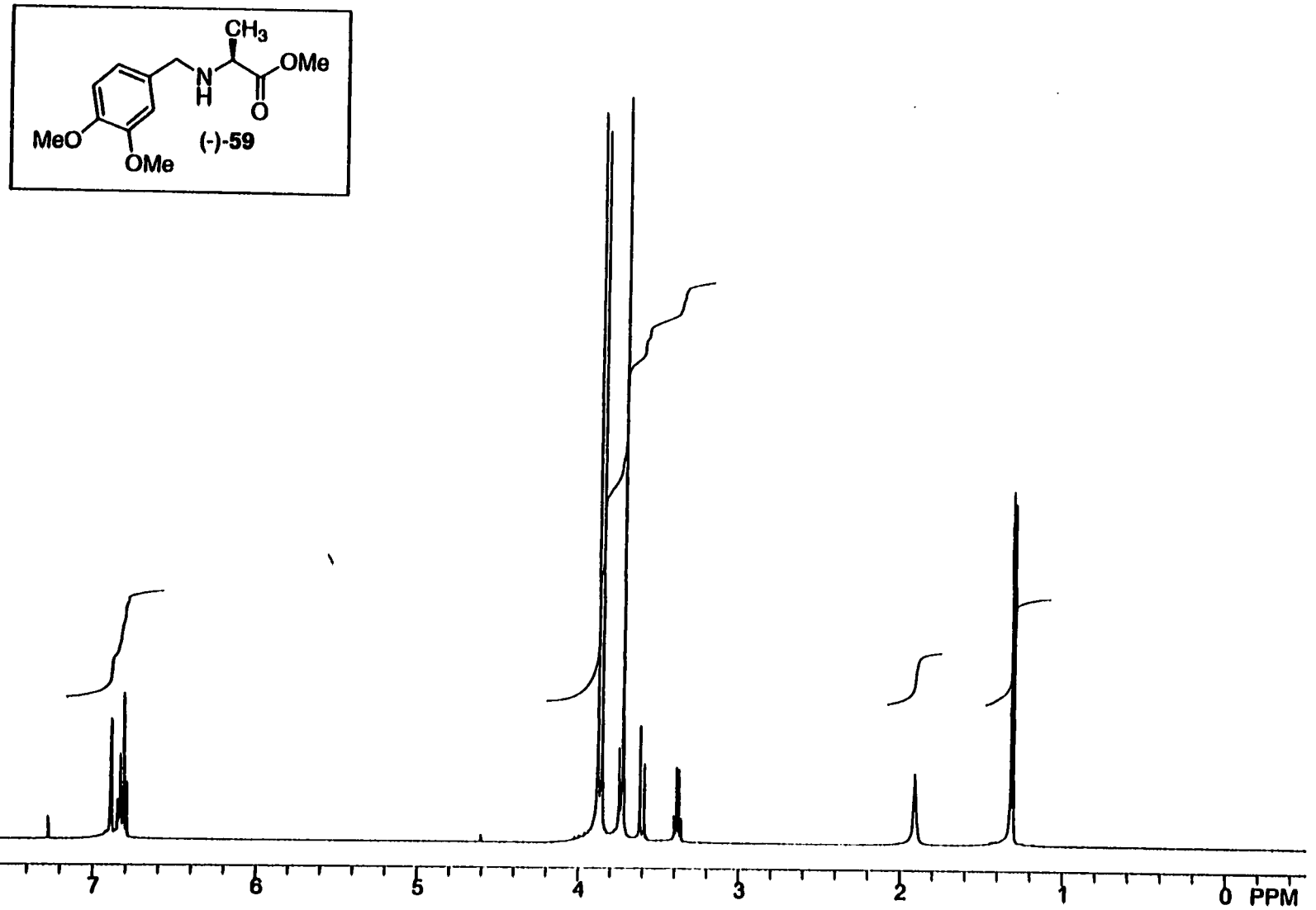


Figure A.2.1 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**59**.

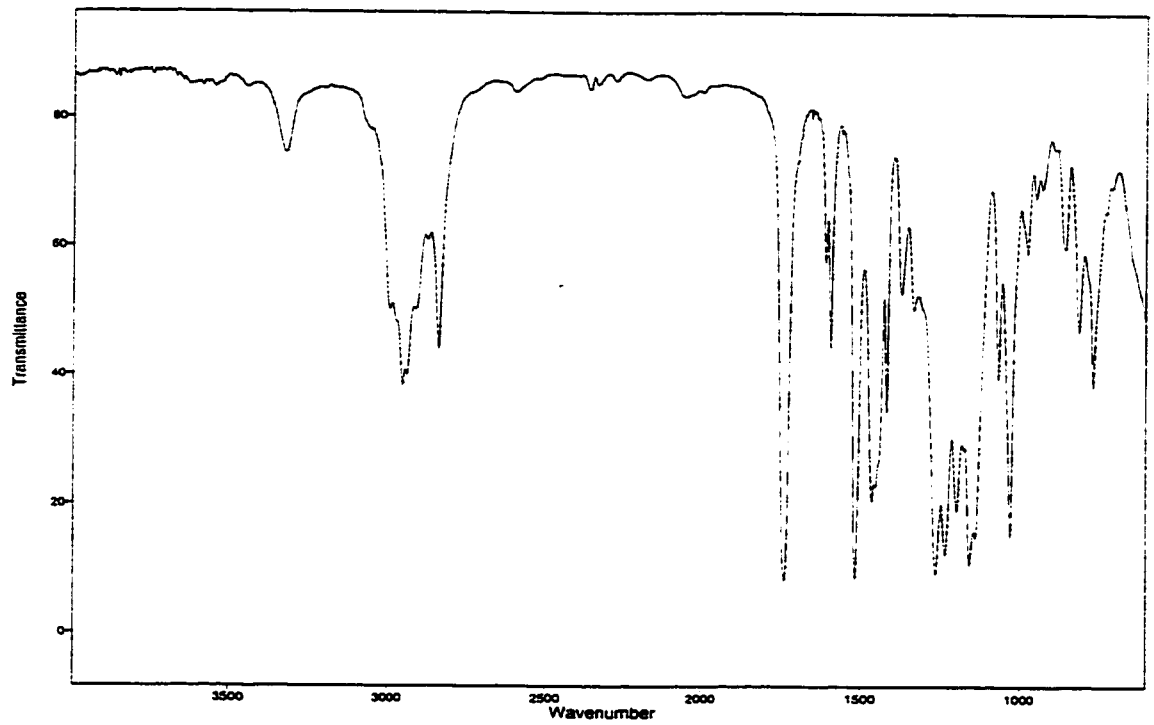


Figure A.2.2 Infrared Spectrum (thin film/NaCl) of compound (-)-**59**.

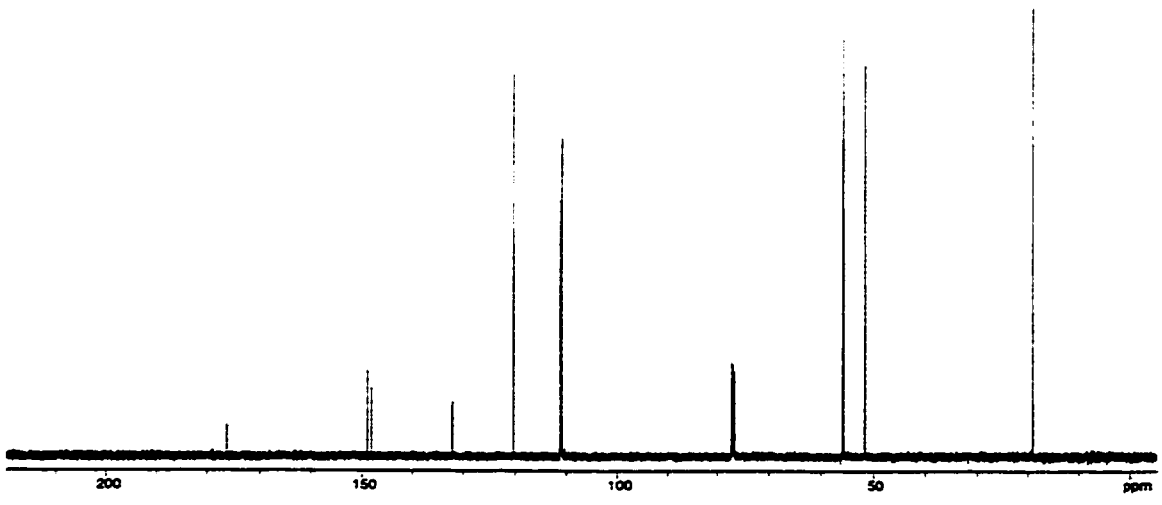


Figure A.2.3 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**59**.

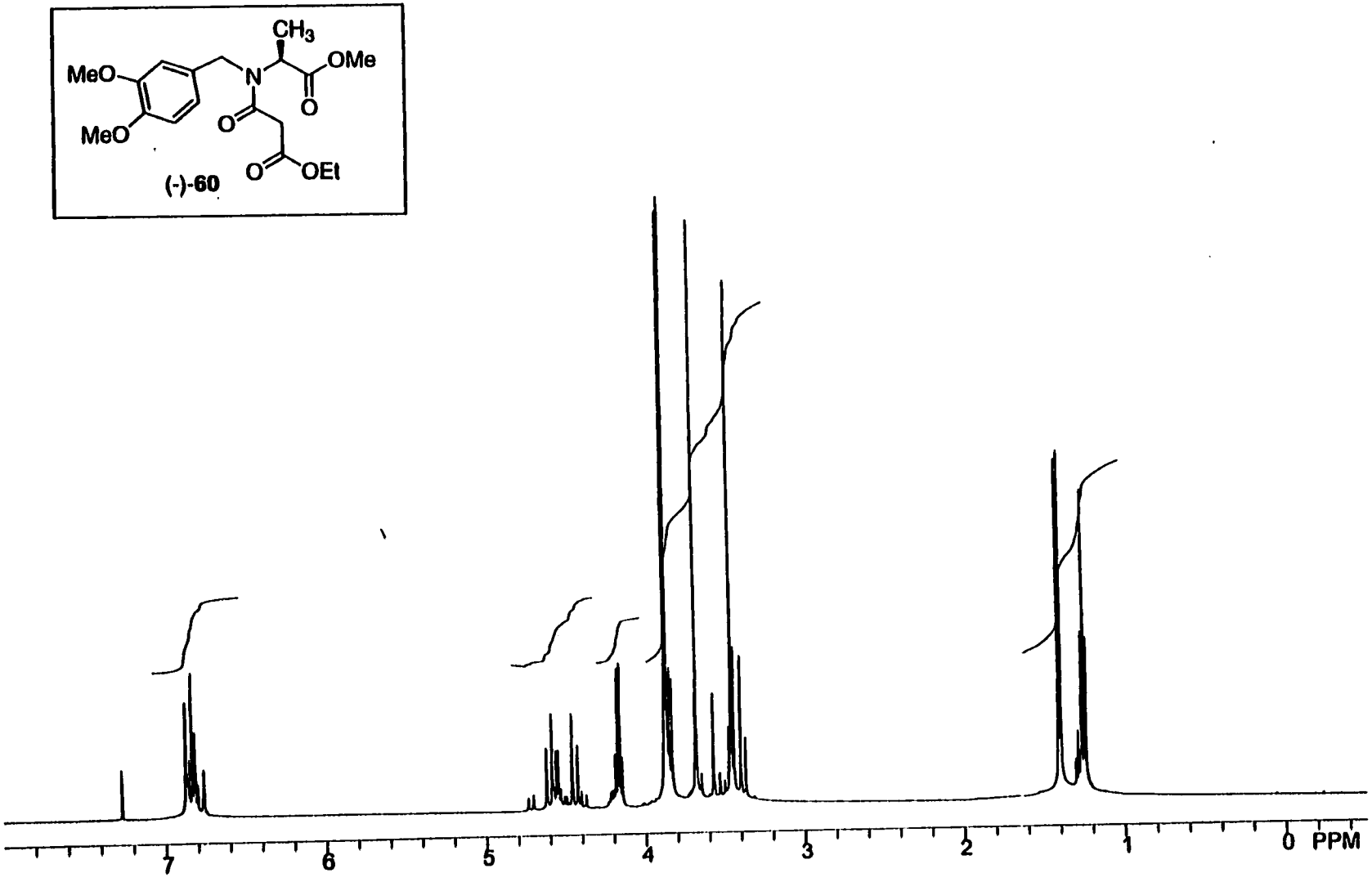


Figure A.2.4 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**60**.

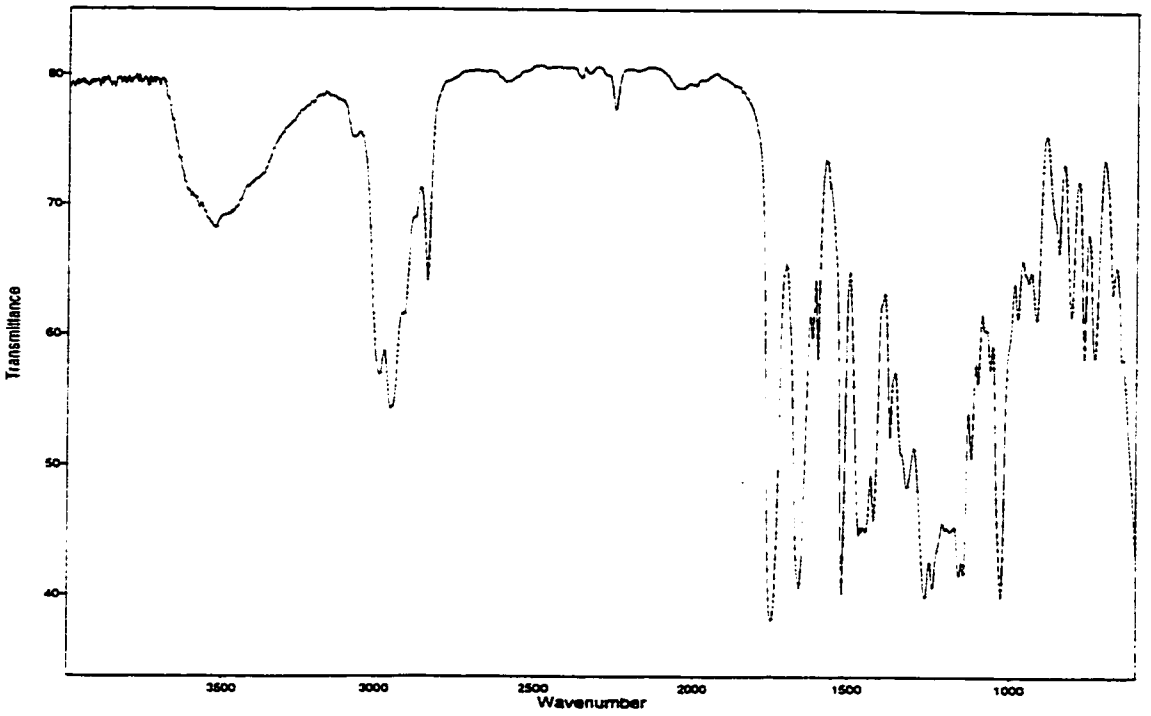


Figure A.2.5 Infrared Spectrum (thin film/NaCl) of compound (-)-**60**.

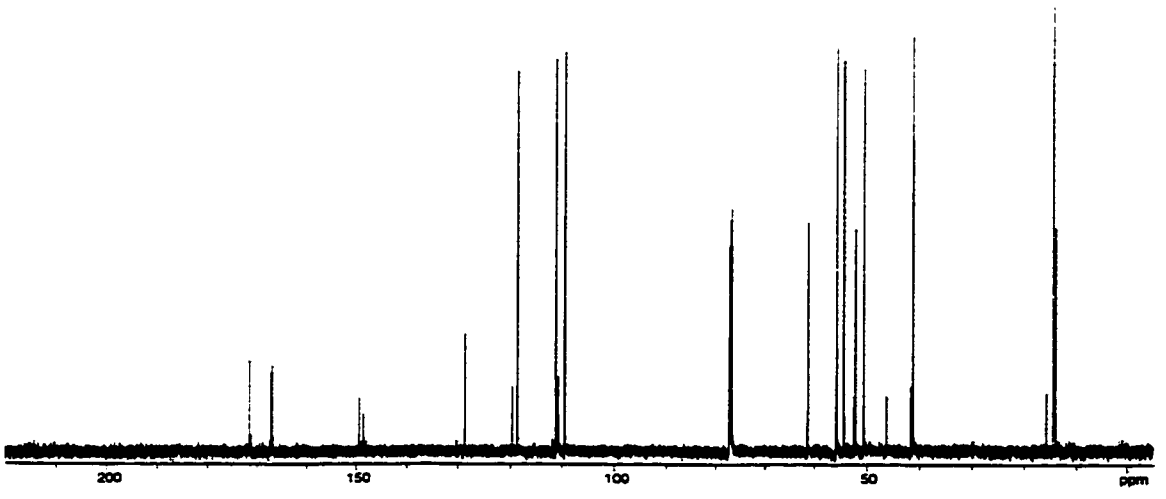


Figure A.2.6 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**60**.

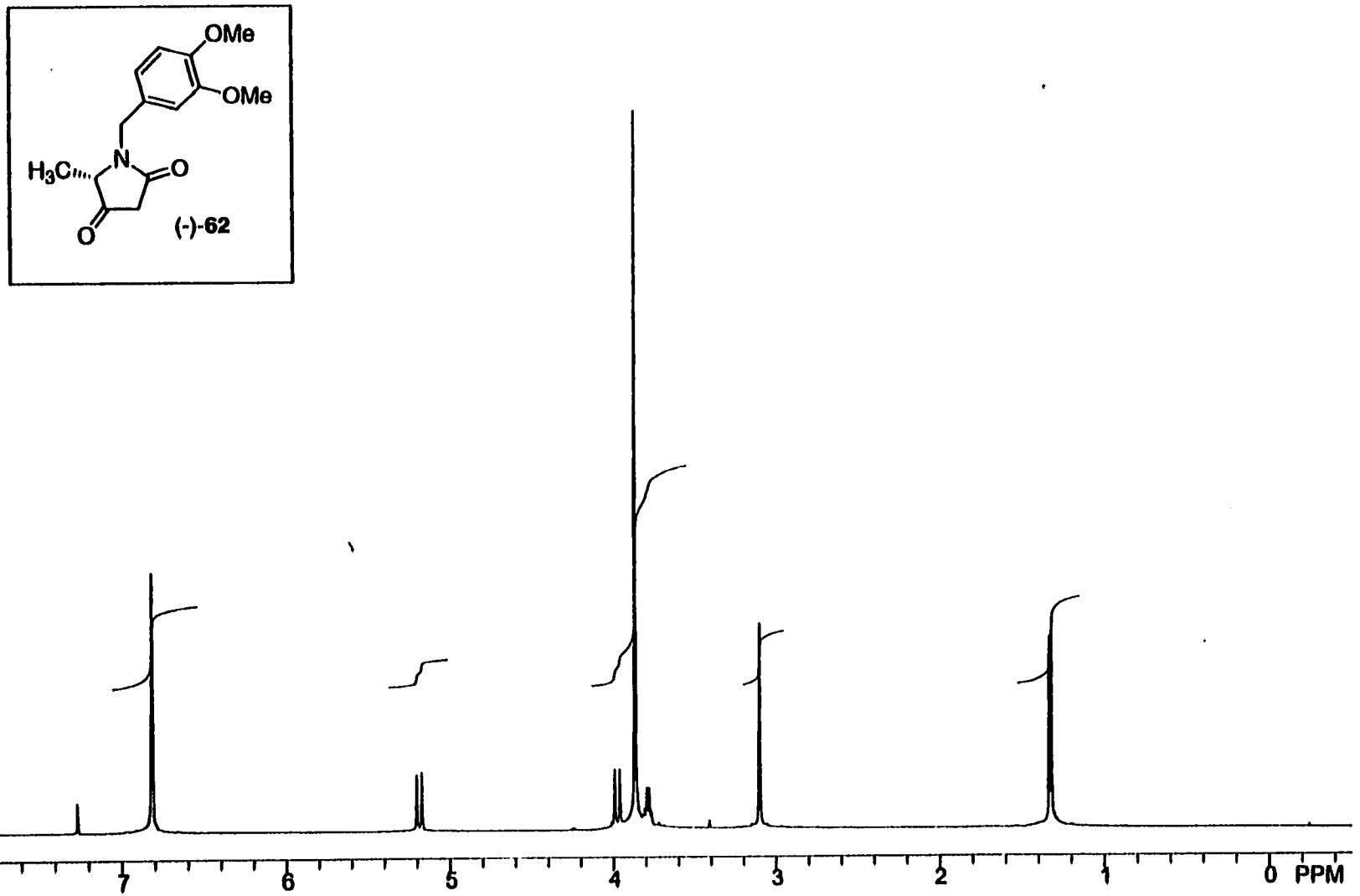


Figure A.2.7 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**62**.

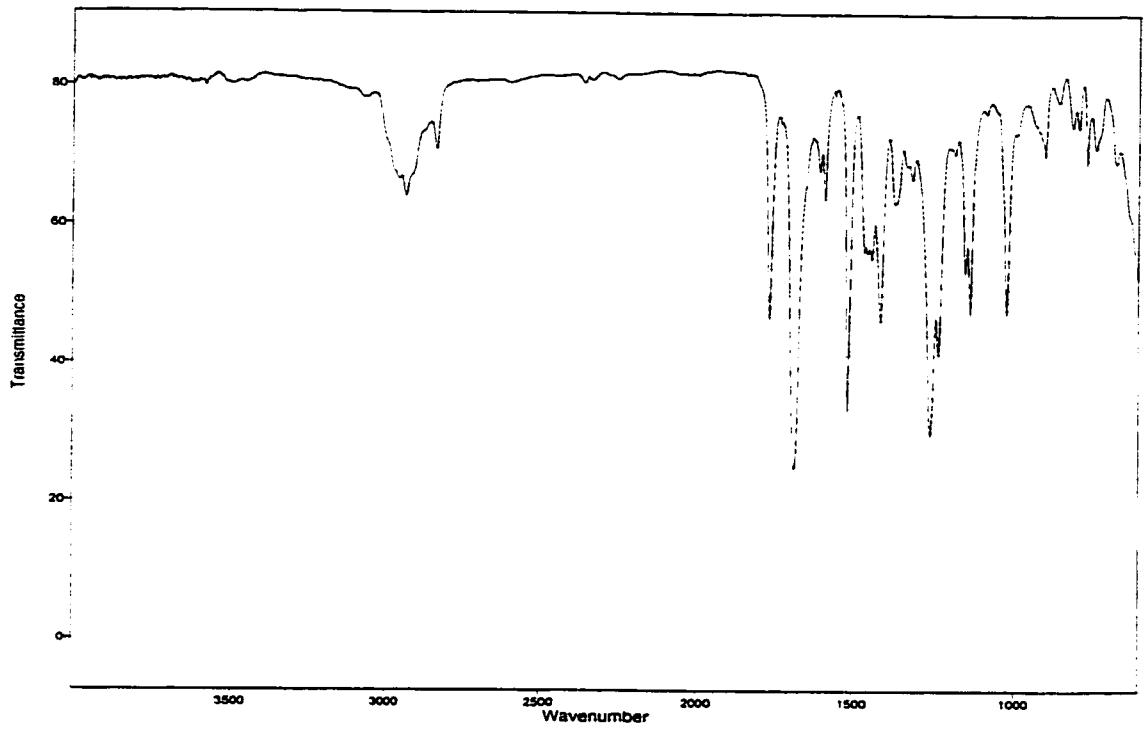


Figure A.2.8 Infrared Spectrum (thin film/NaCl) of compound (-)-**62**.

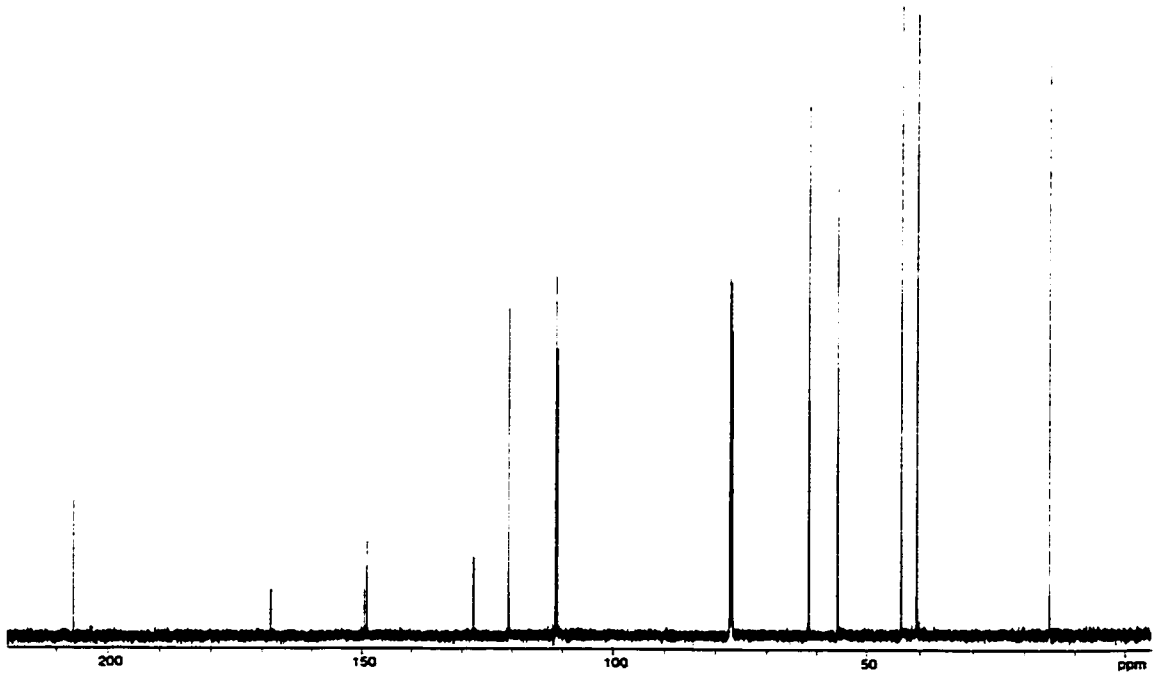
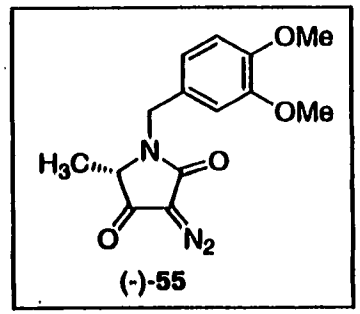


Figure A.2.9 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**62**.



(-)-55

Figure A.2.10 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**55**.

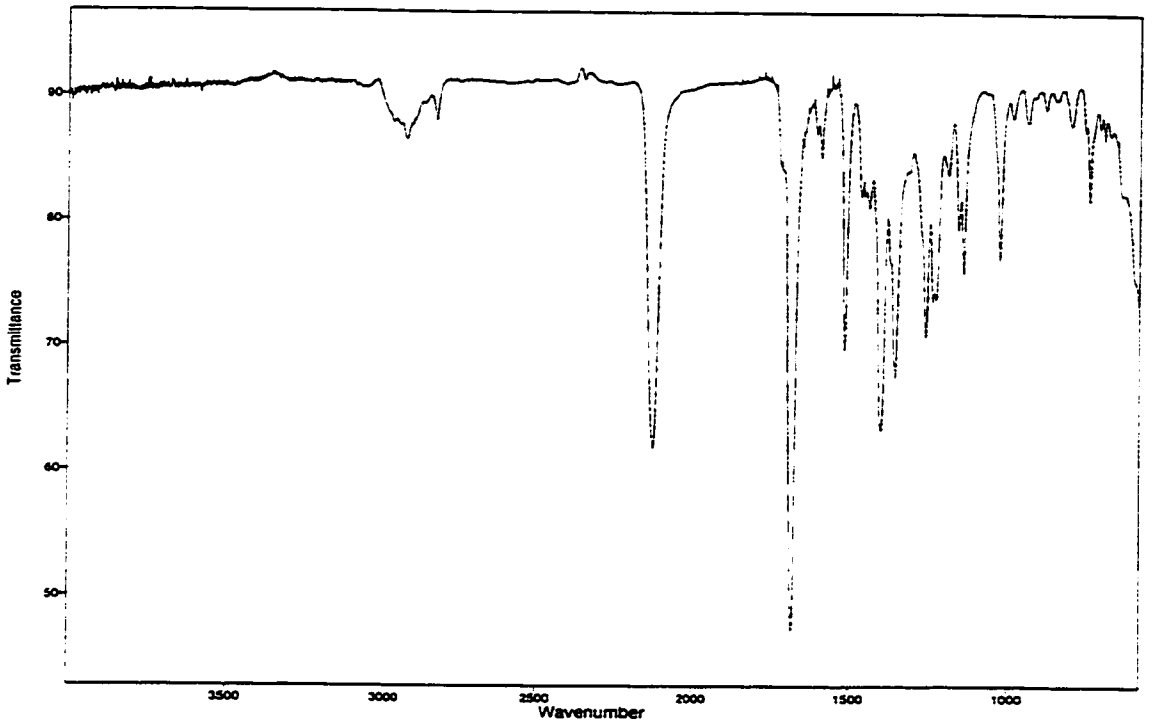


Figure A.2.11 Infrared Spectrum (thin film/NaCl) of compound (-)-**55**.

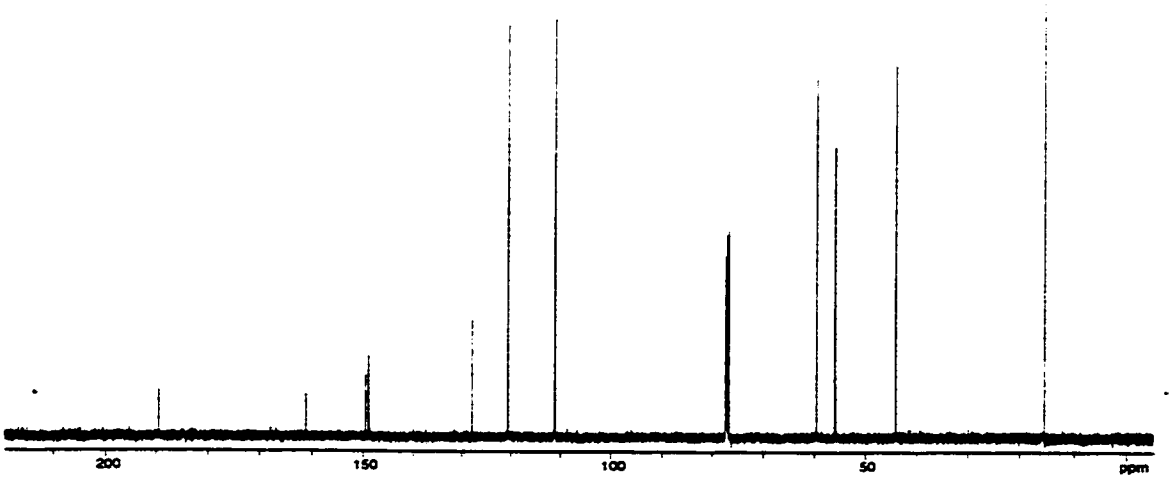


Figure A.2.12 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**55**.

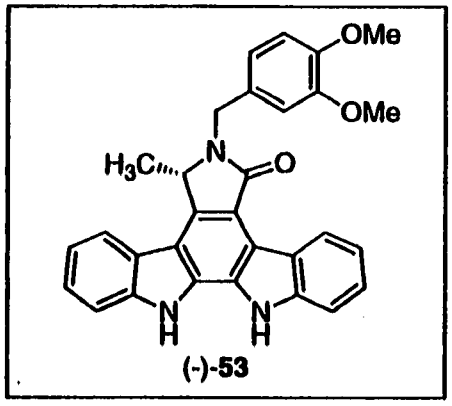


Figure A.2.13 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**53**.

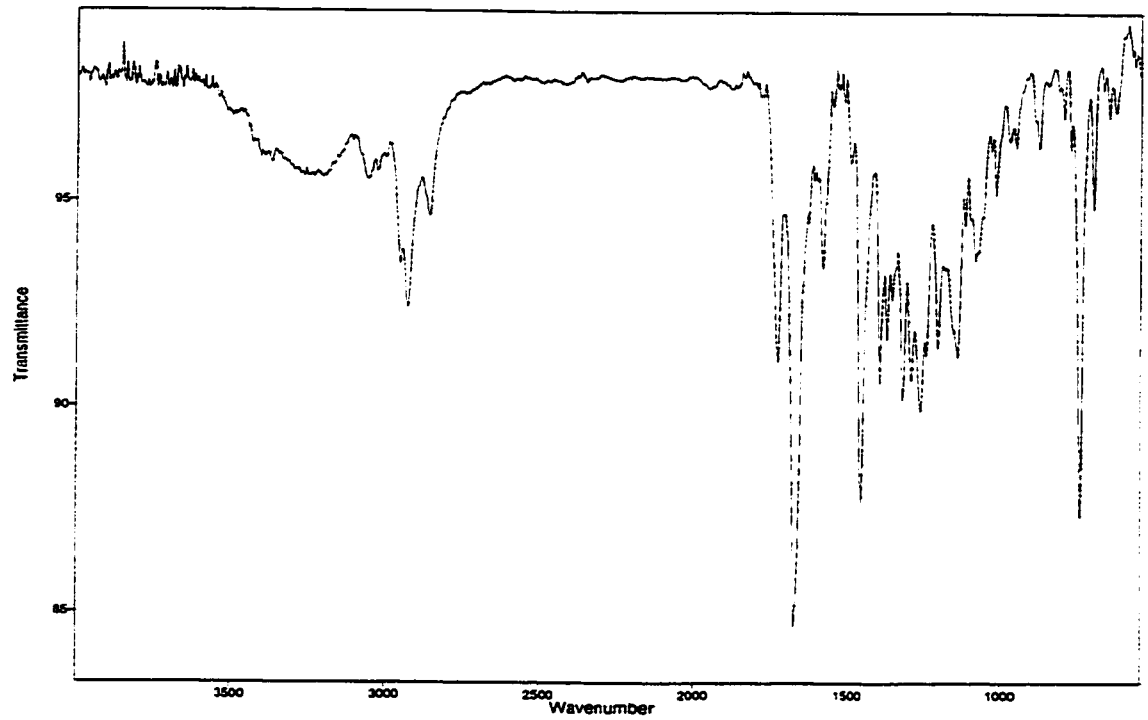


Figure A.2.14 Infrared Spectrum (thin film/NaCl) of compound (-)-**53**.

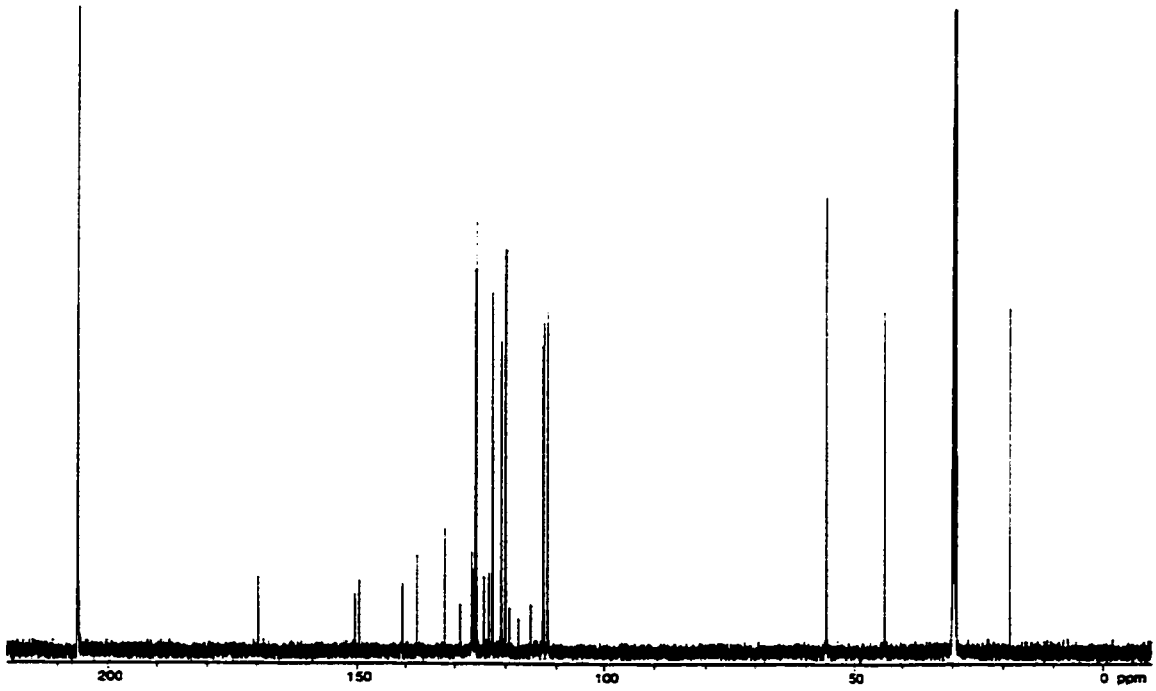


Figure A.2.15 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**53**.

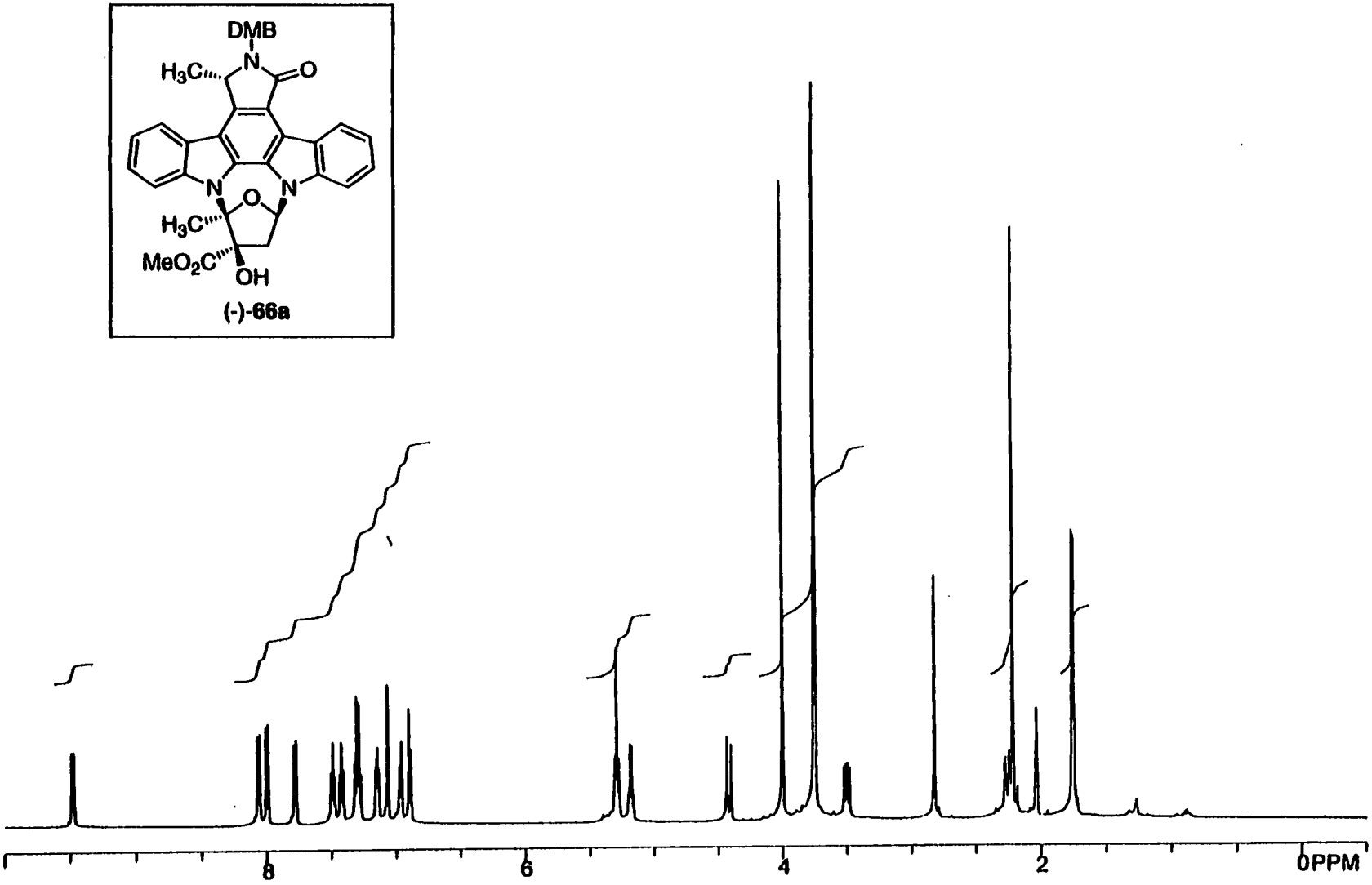


Figure A.2.16 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**66a**.

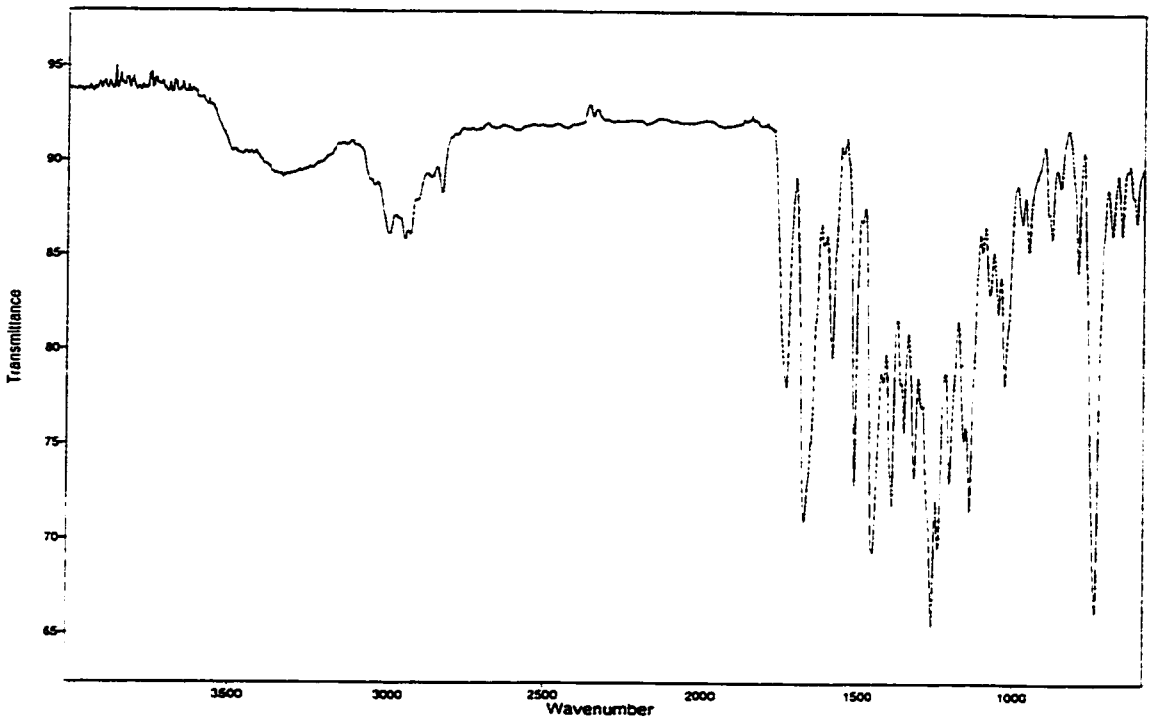


Figure A.2.17 Infrared Spectrum (thin film/NaCl) of compound (-)-**66a**.

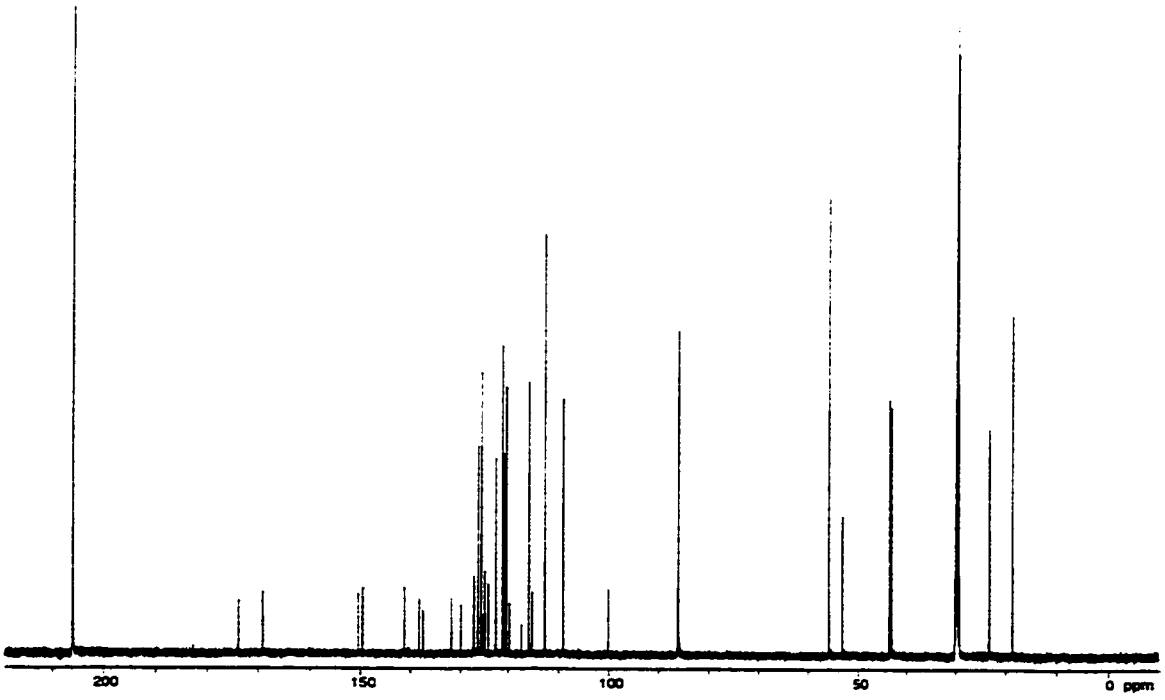


Figure A.2.18 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**66a**.

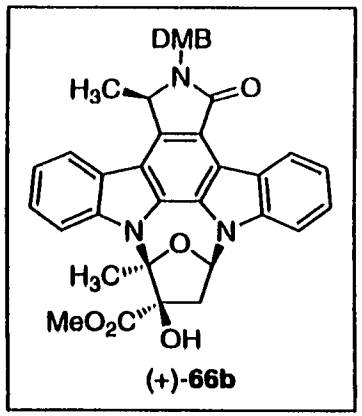


Figure A.2.19 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**66b**.

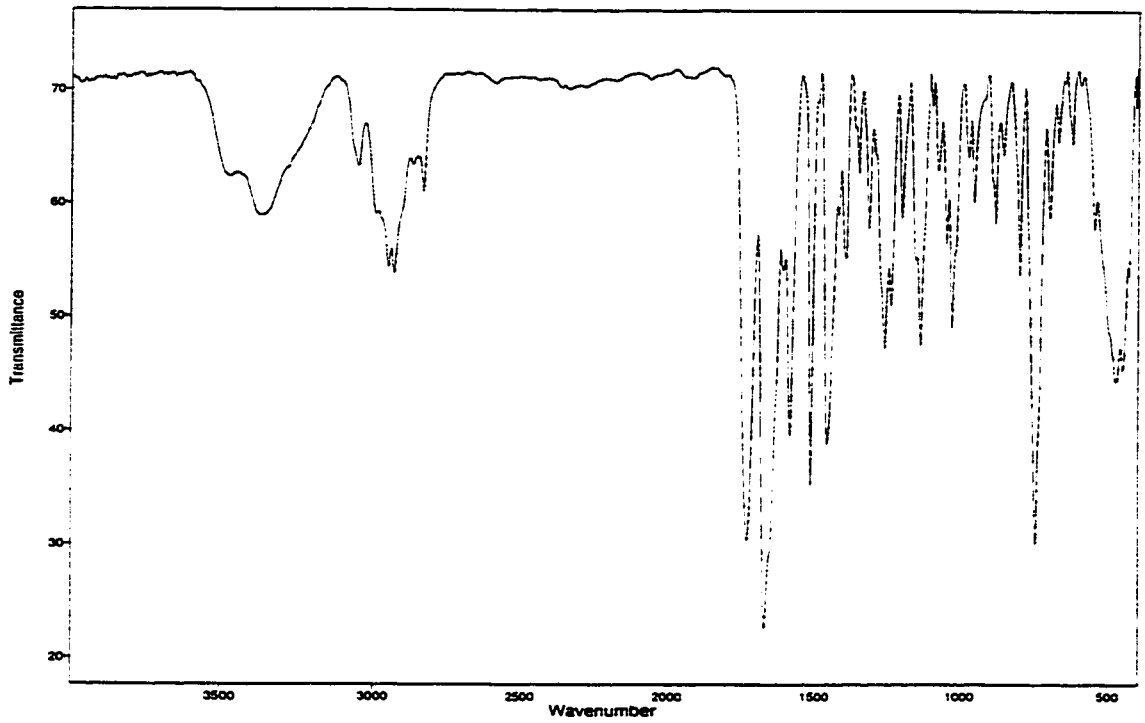


Figure A.2.20 Infrared Spectrum (thin film/NaCl) of compound (+)-**66b**.

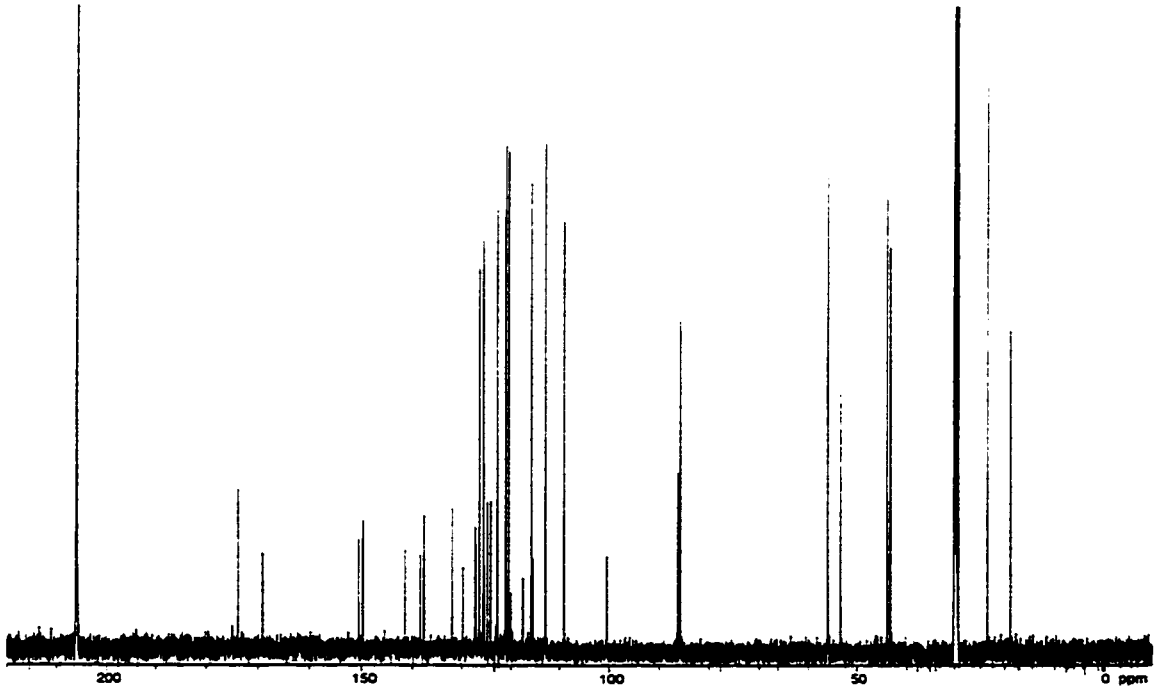


Figure A.2.21 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**66b**.

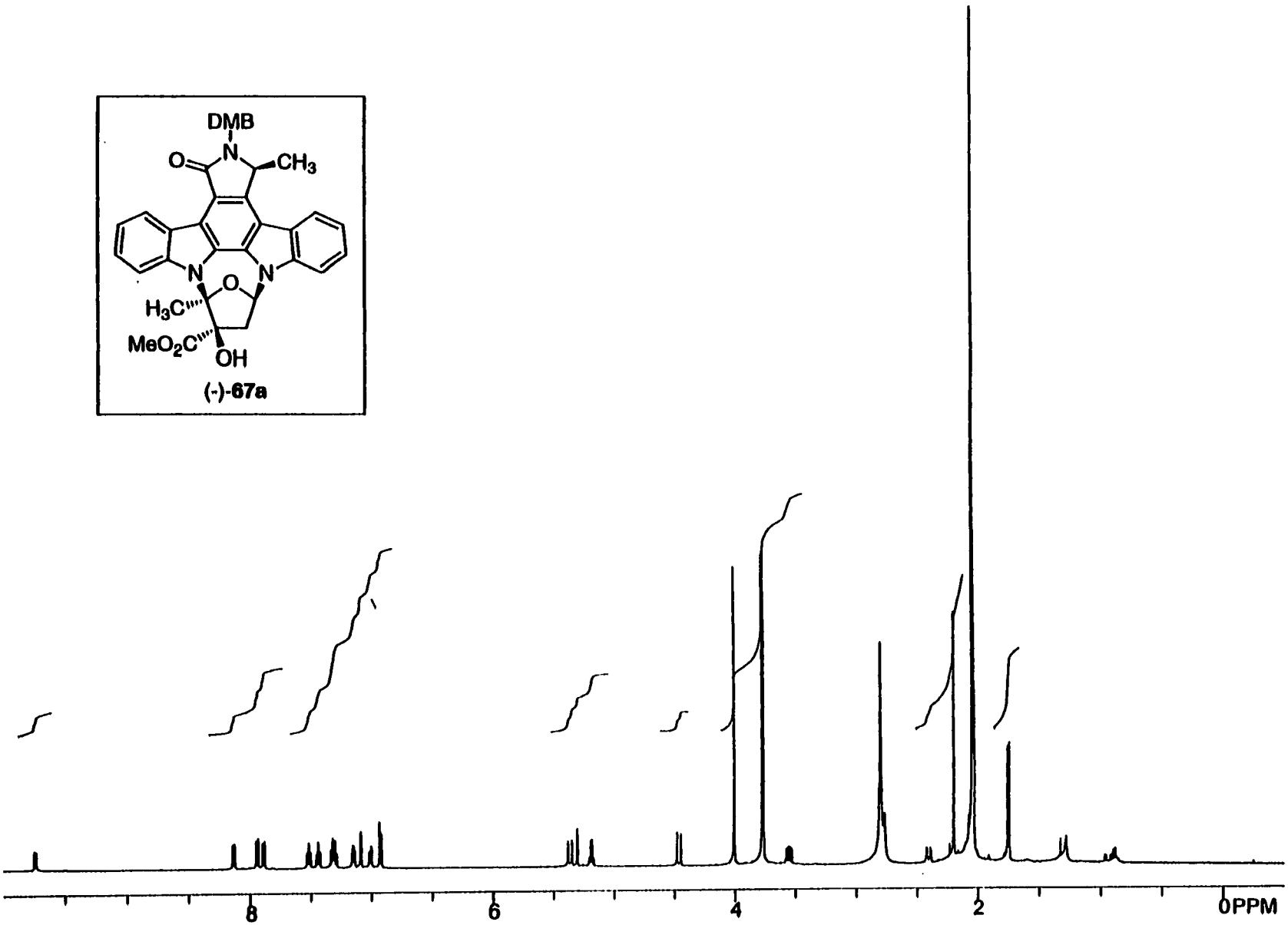


Figure A.2.22 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**67a**.

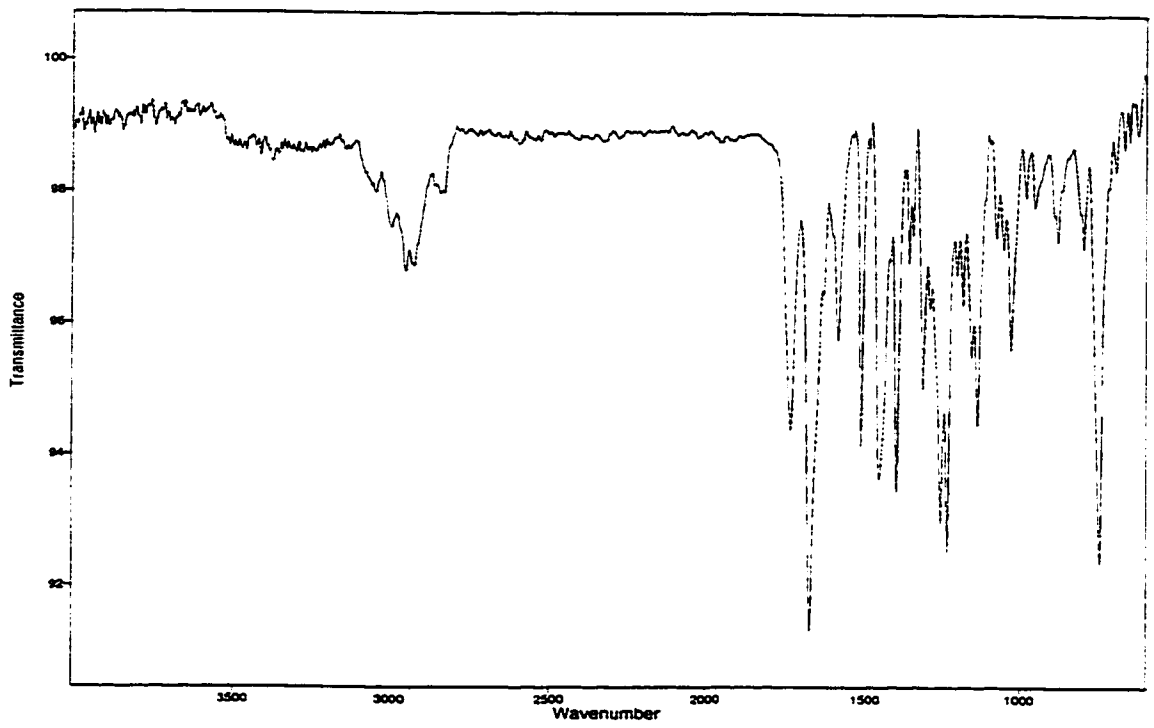


Figure A.2.23 Infrared Spectrum (thin film/NaCl) of compound (-)-**67a**.

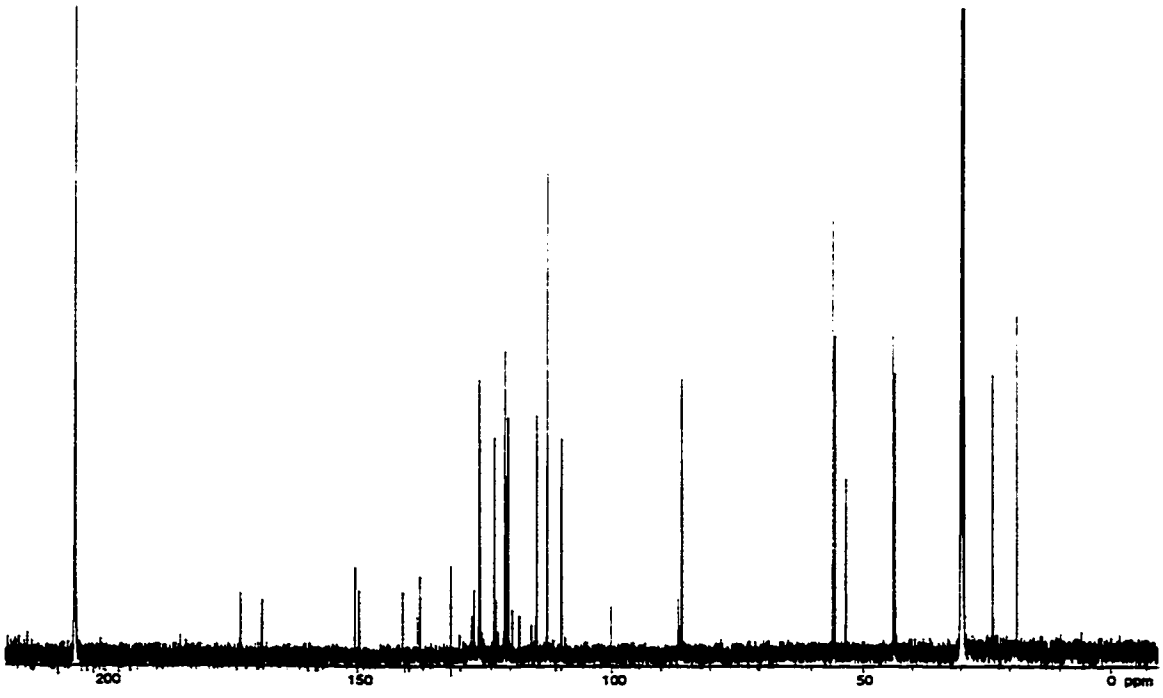


Figure A.2.24 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**67a**.

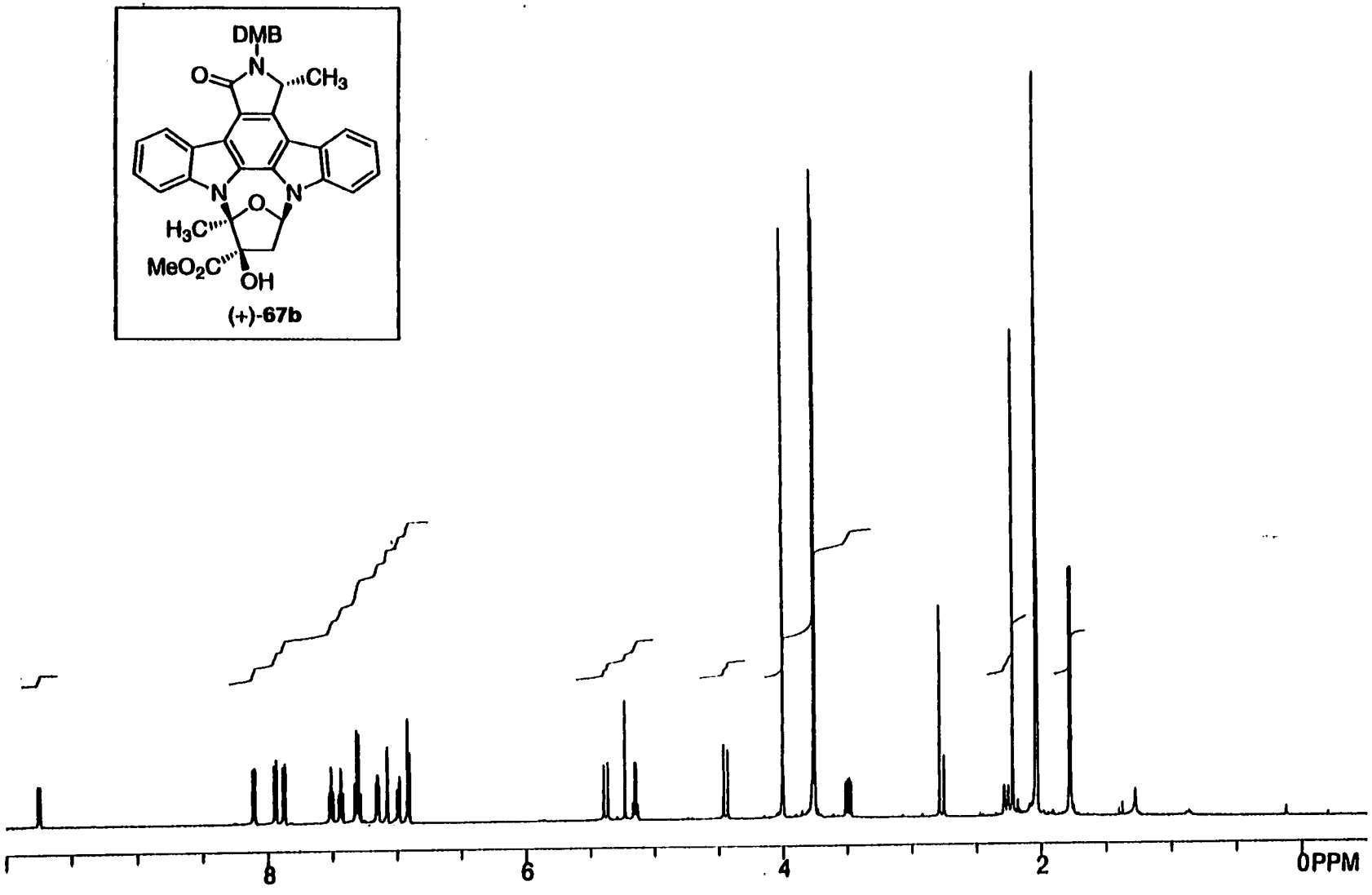


Figure A.2.25 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**67b**.

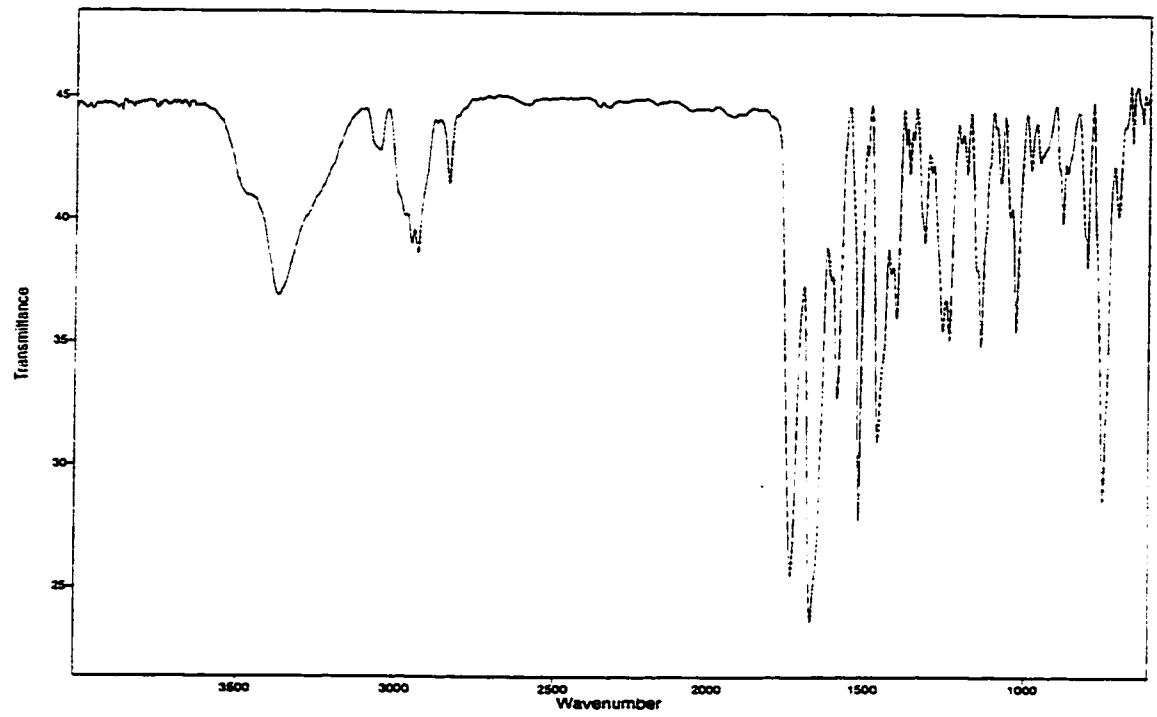


Figure A.2.26 Infrared Spectrum (thin film/NaCl) of compound (+)-**67b**.

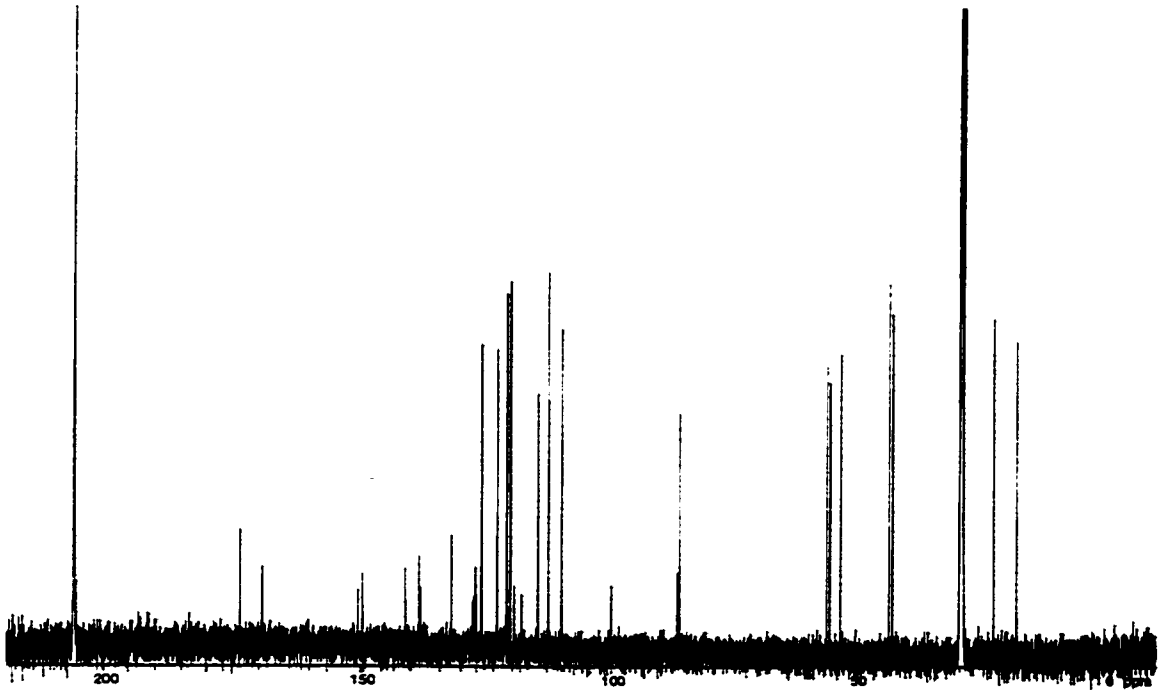


Figure A.2.27 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**67b**.

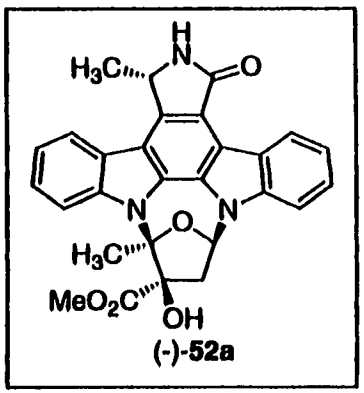


Figure A.2.28 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**52a**.

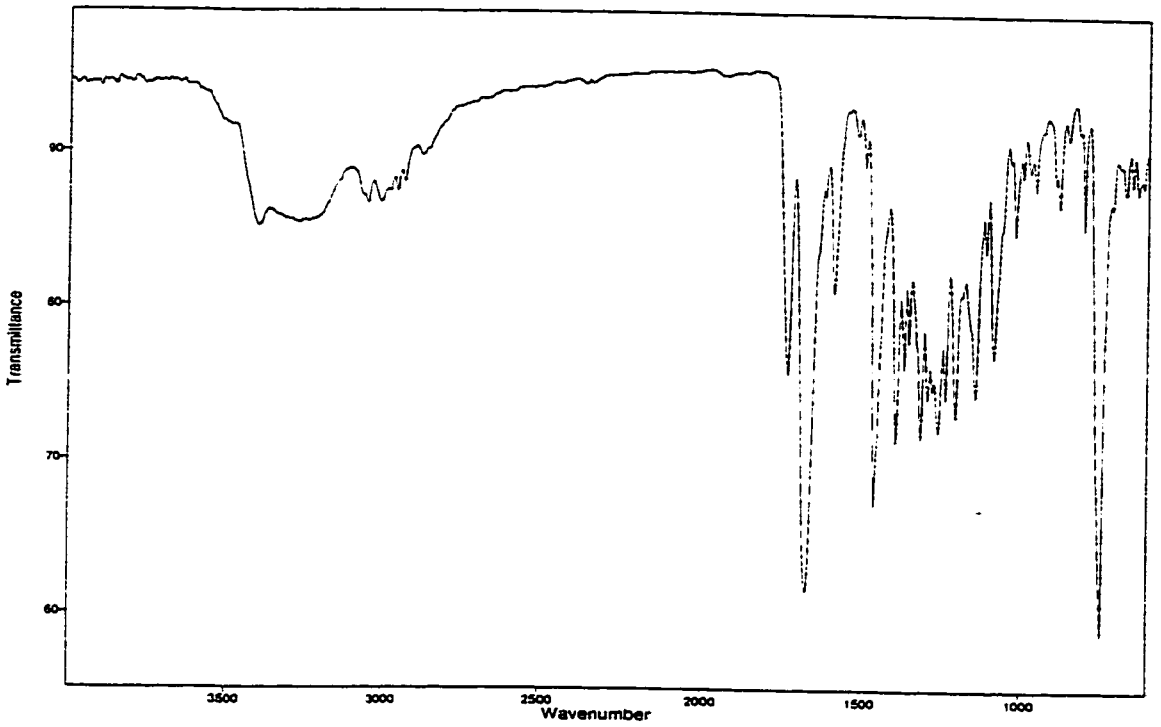


Figure A.2.29 Infrared Spectrum (thin film/NaCl) of compound (-)-**52a**.

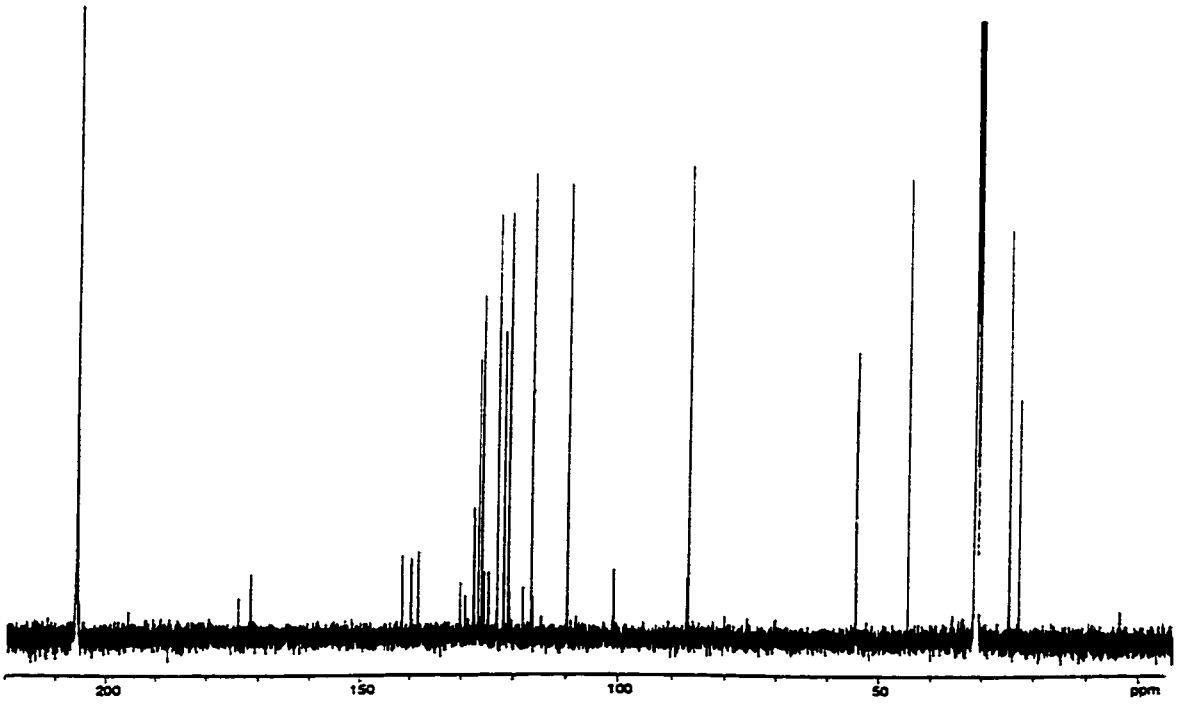


Figure A.2.30 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**52a**.

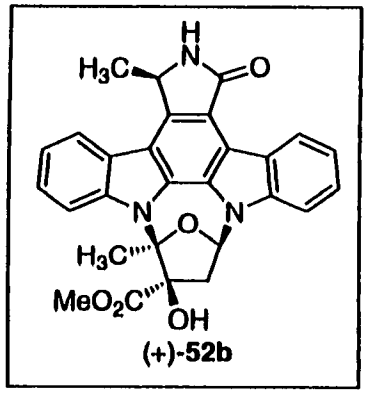


Figure A.2.31 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**52b**.

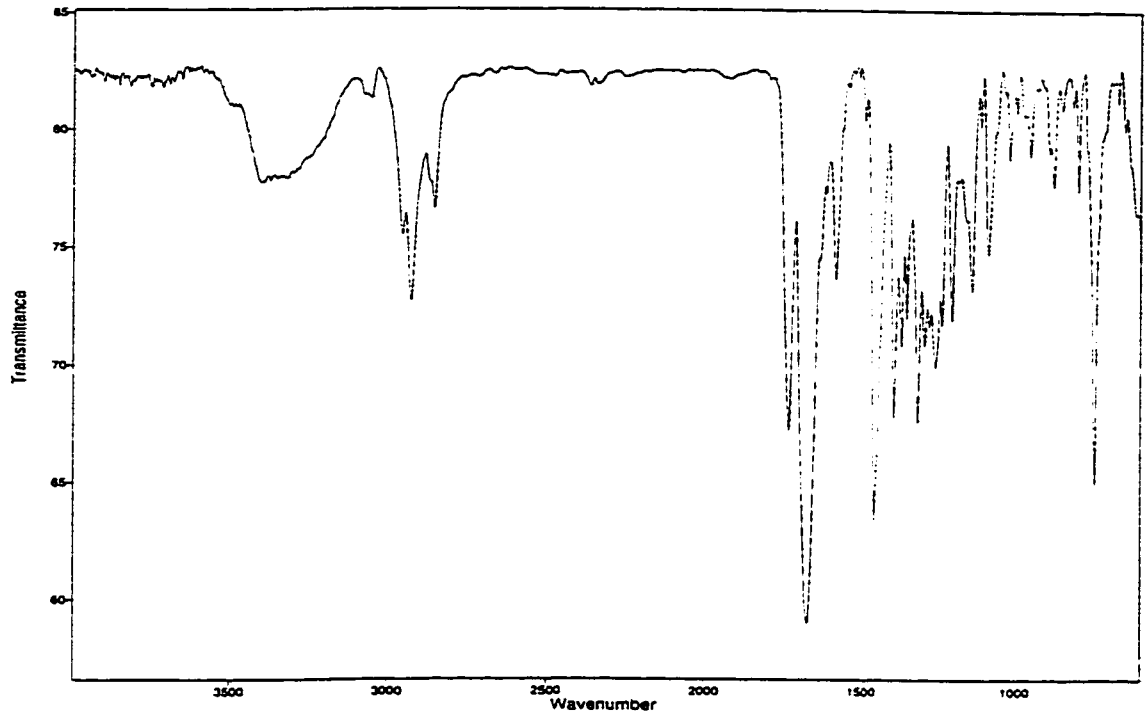


Figure A.2.32 Infrared Spectrum (thin film/NaCl) of compound (+)-**52b**.

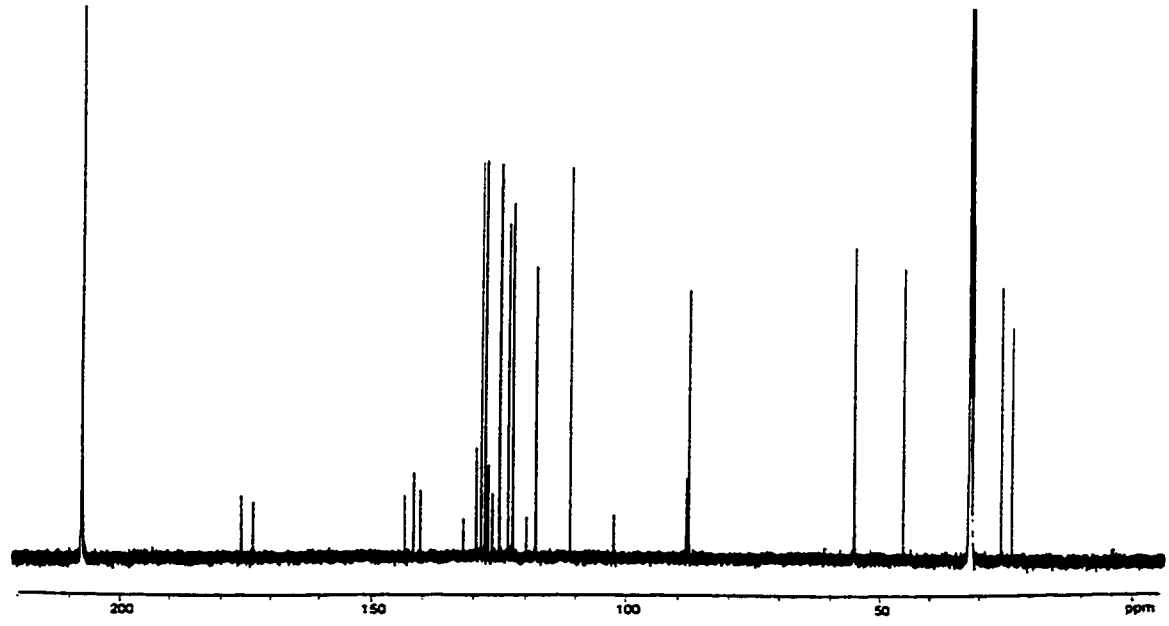


Figure A.2.33 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**52b**.

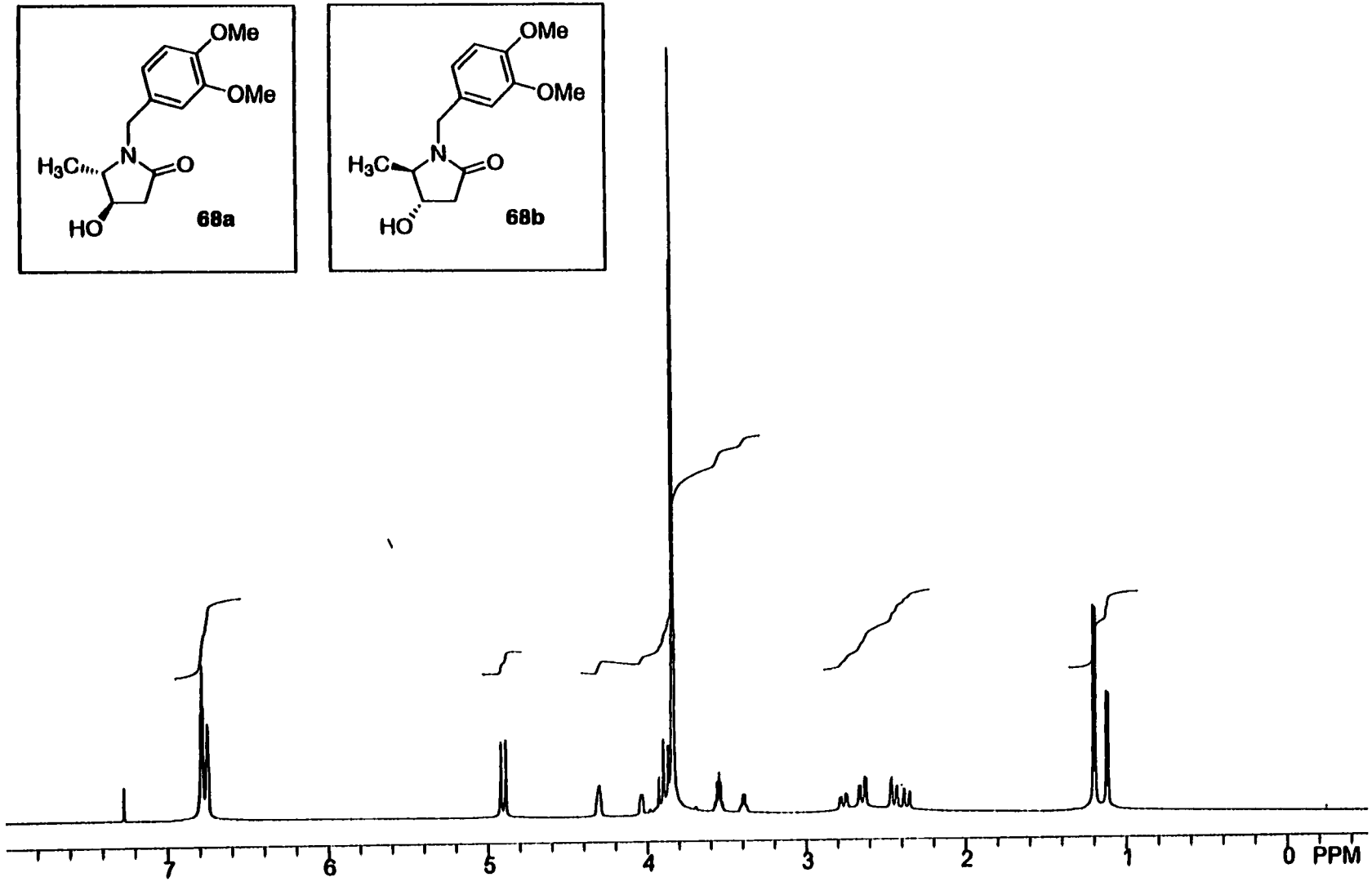


Figure A.2.34 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**68a,b**.

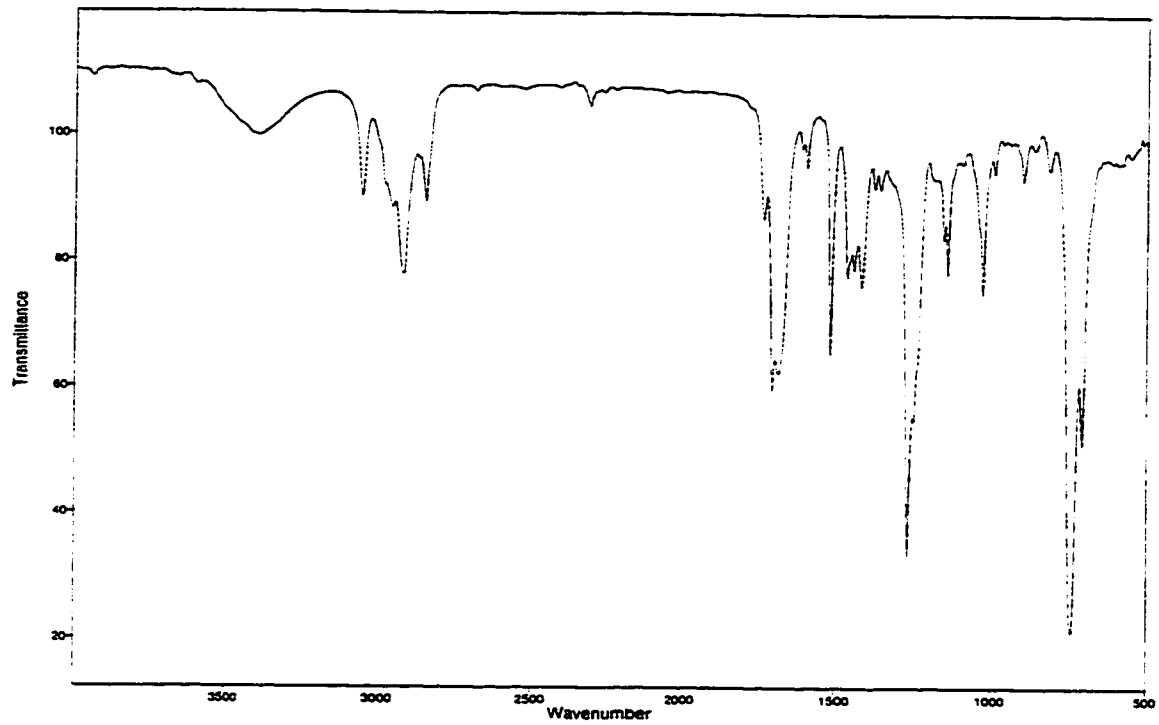


Figure A.2.35 Infrared Spectrum (thin film/NaCl) of compound (-)-**68a,b**.

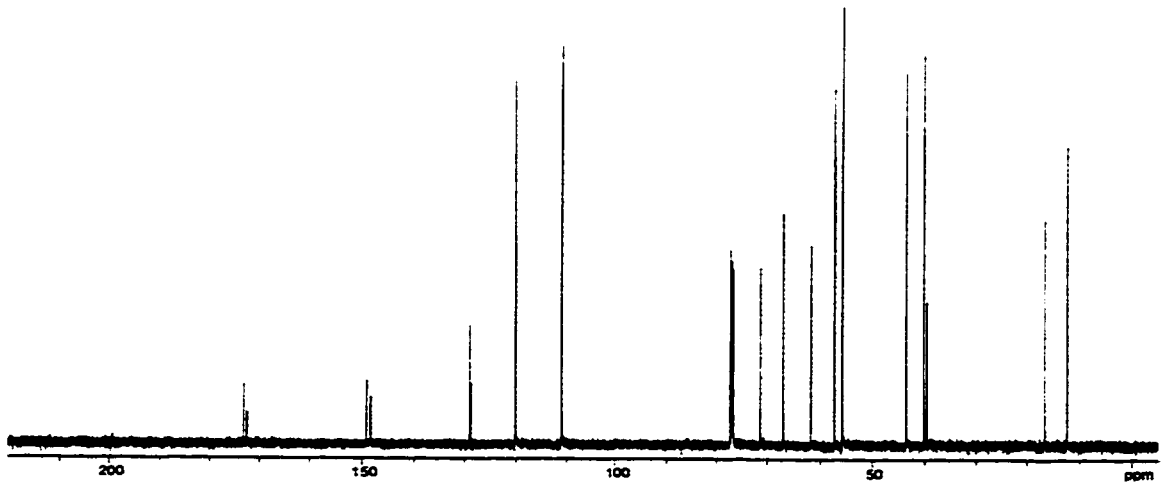


Figure A.2.36 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**68a,b**.

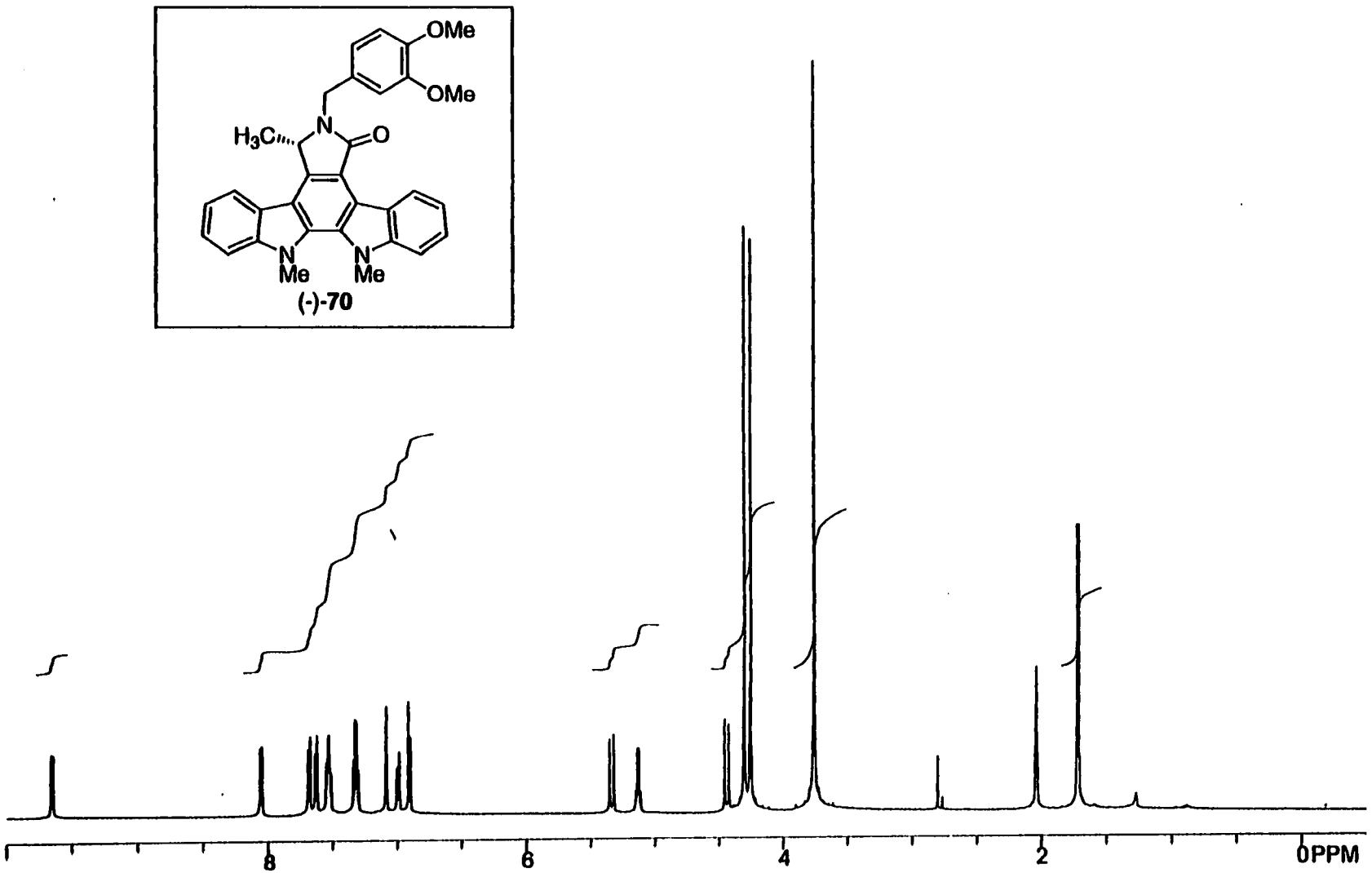


Figure A.2.37 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**70**.

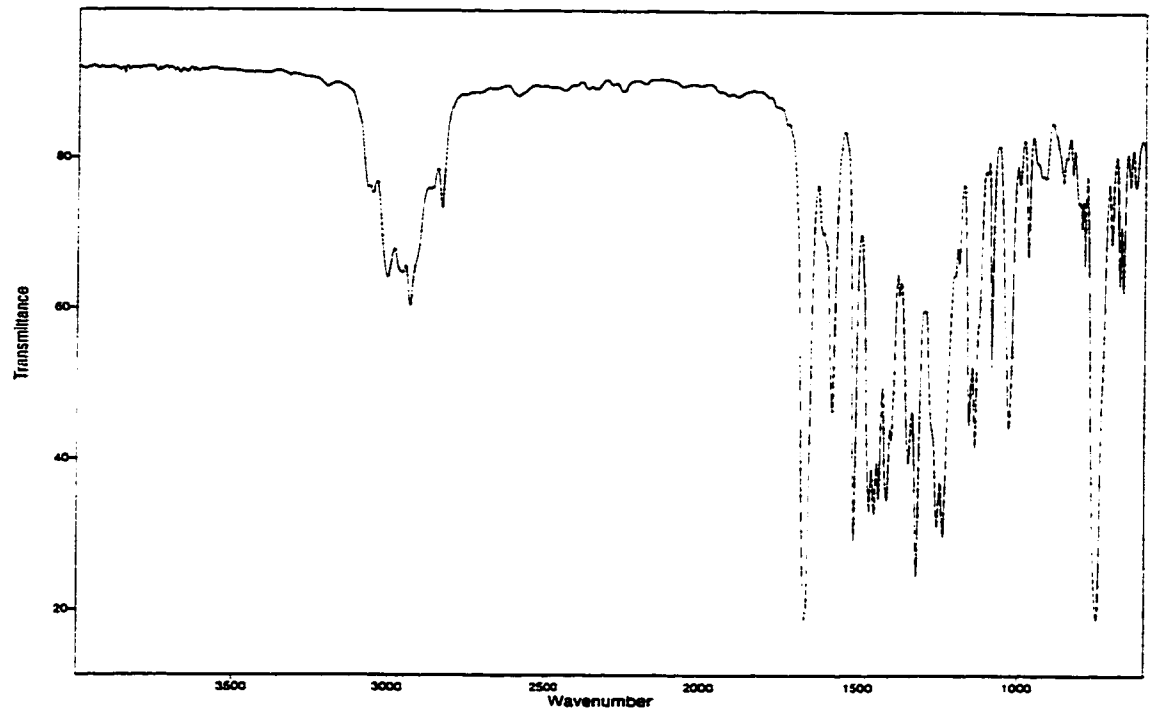


Figure A.2.38 Infrared Spectrum (thin film/NaCl) of compound (-)-**70**.

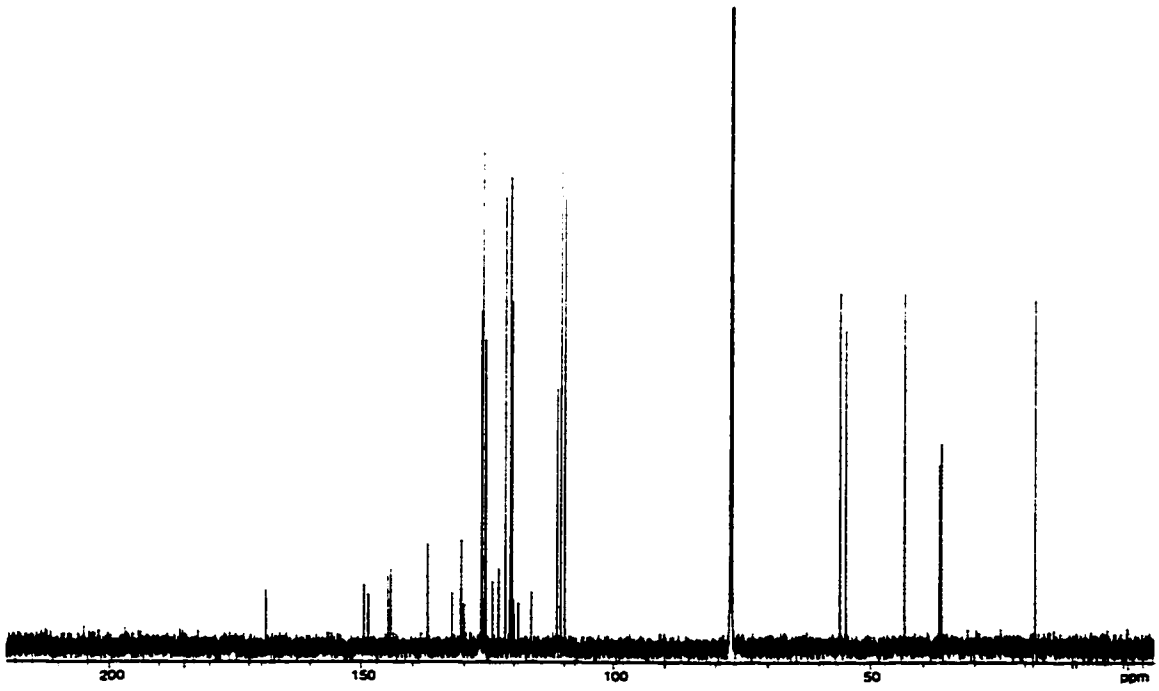


Figure A.2.39 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**70**.

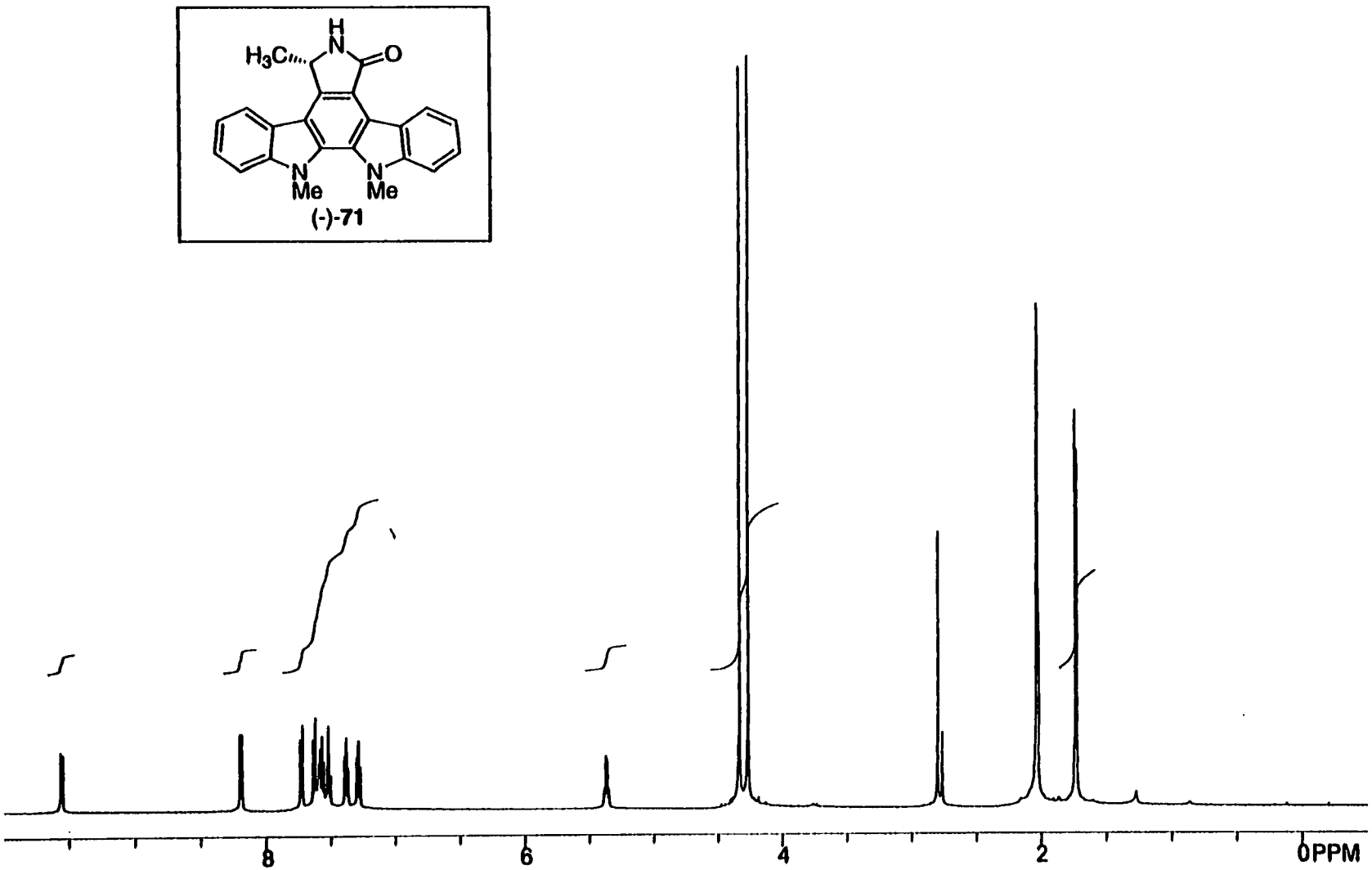


Figure A.2.40 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**71**.

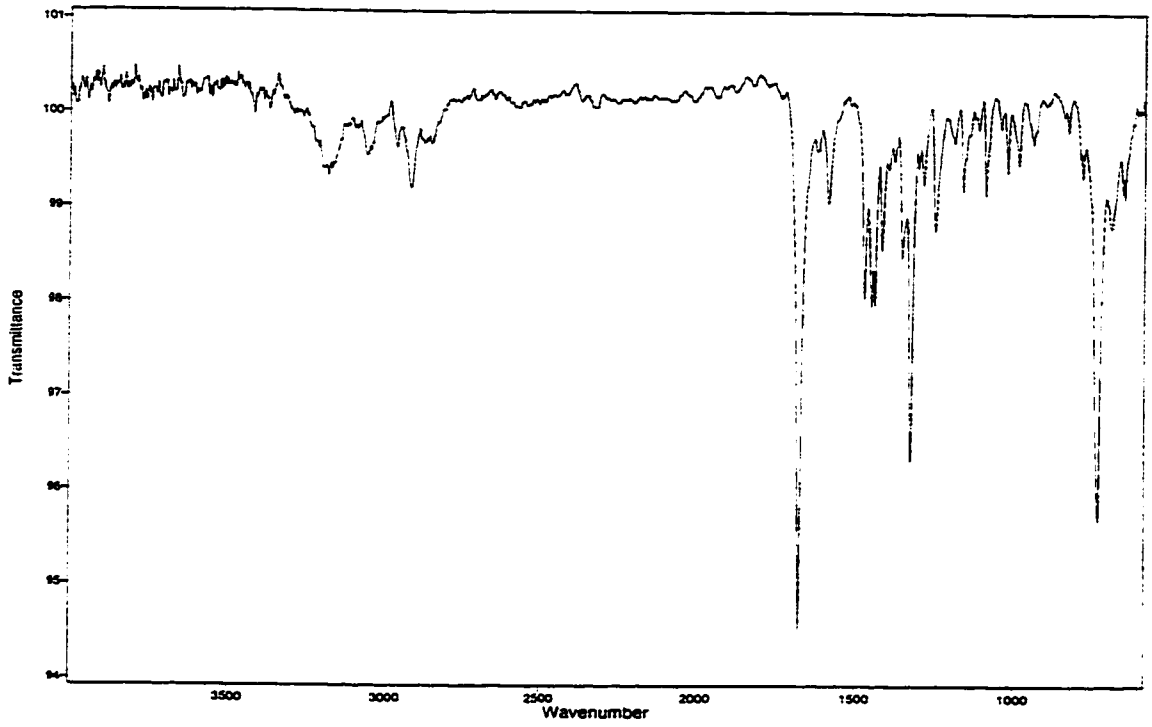


Figure A.2.41 Infrared Spectrum (thin film/NaCl) of compound (-)-**71**.

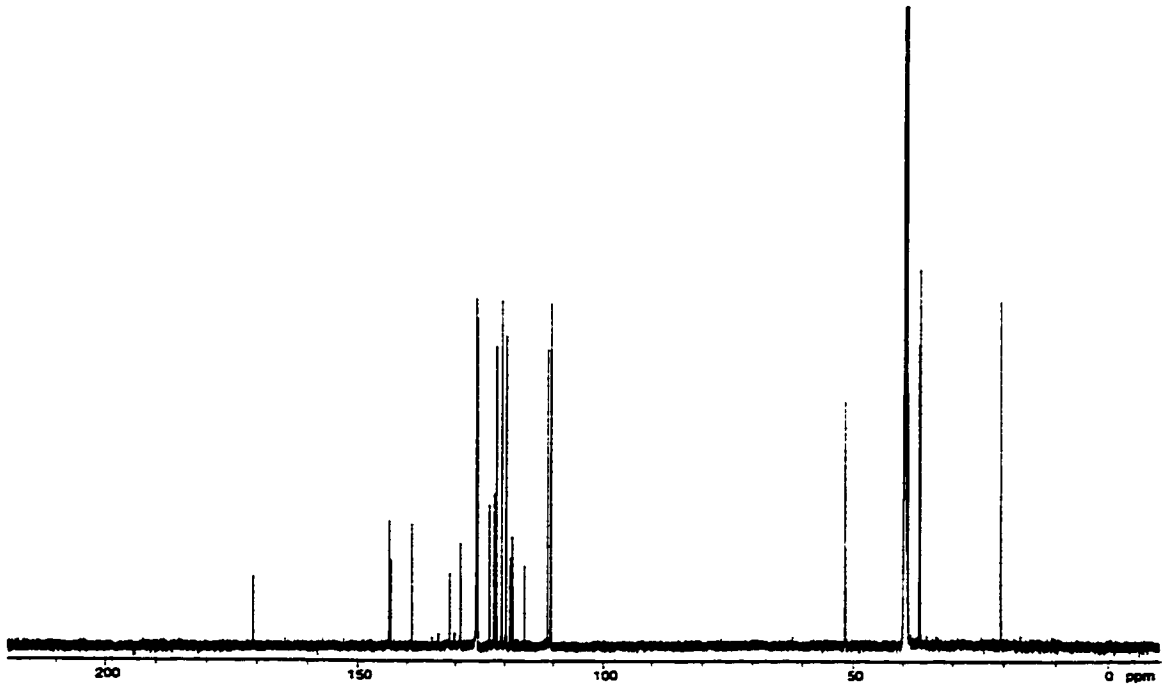


Figure A.2.42 ^{13}C NMR (125 MHz, DMSO-d_6) of compound (-)-**71**.

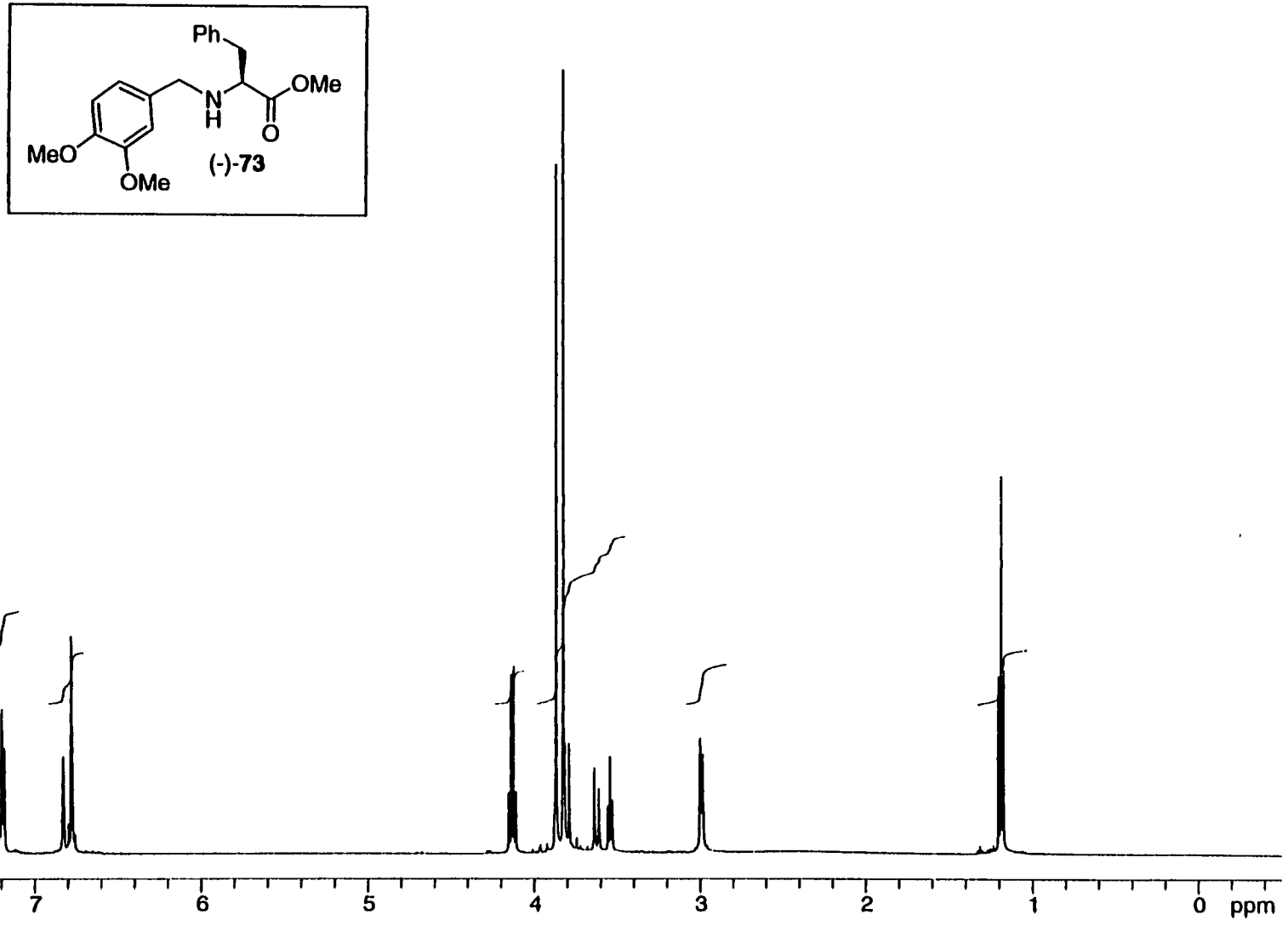


Figure A.2.43 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**73**.

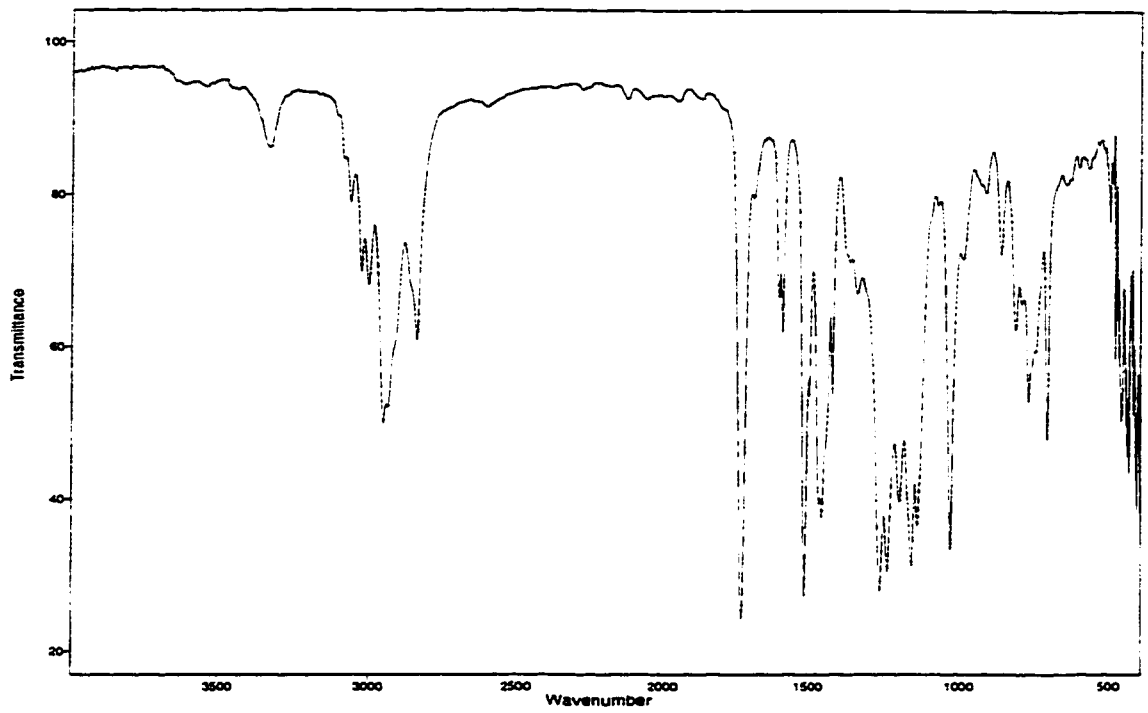


Figure A.2.44 Infrared Spectrum (thin film/NaCl) of compound (-)-**73**.

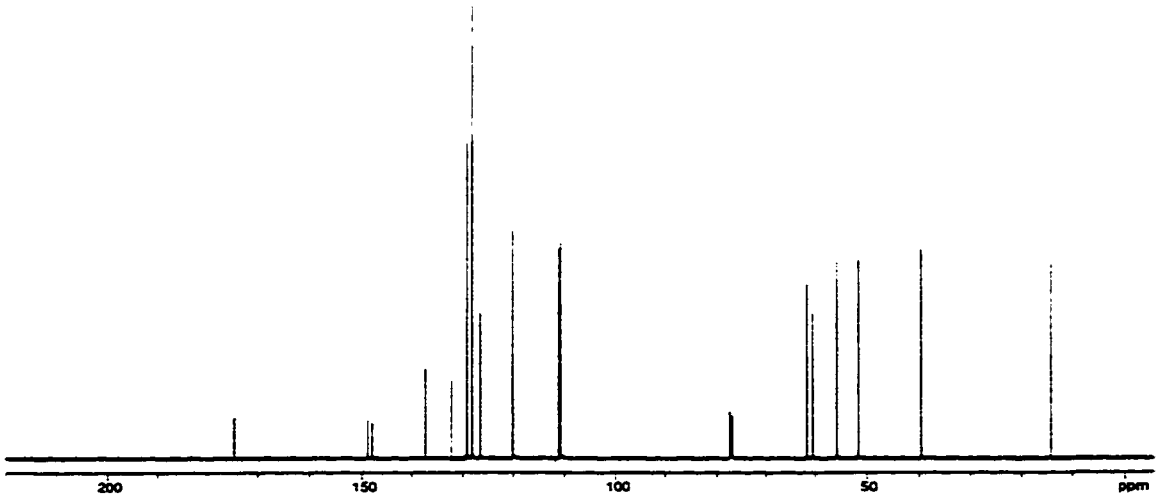


Figure A.2.45 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**73**.

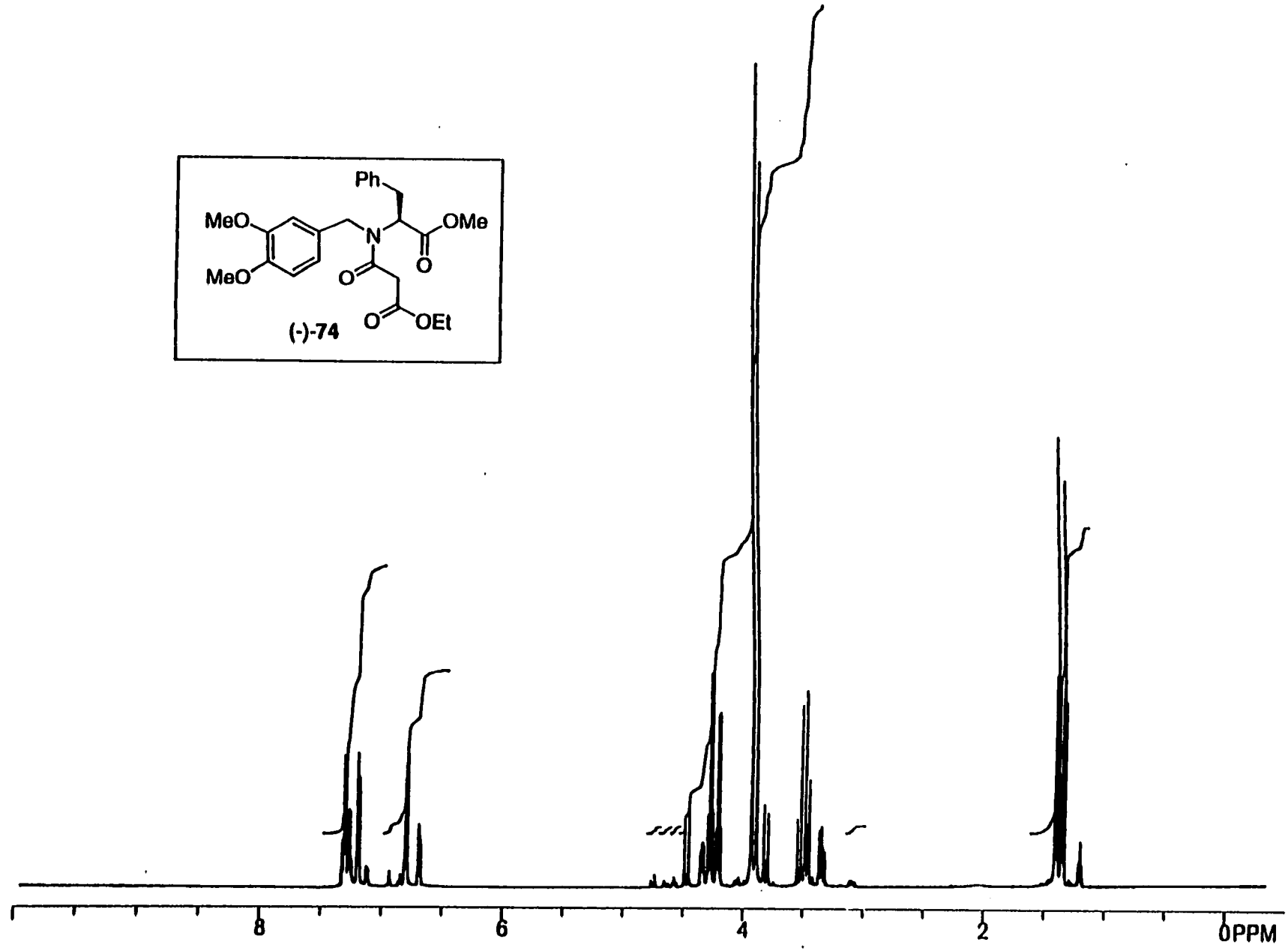


Figure A.2.46 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**74**.

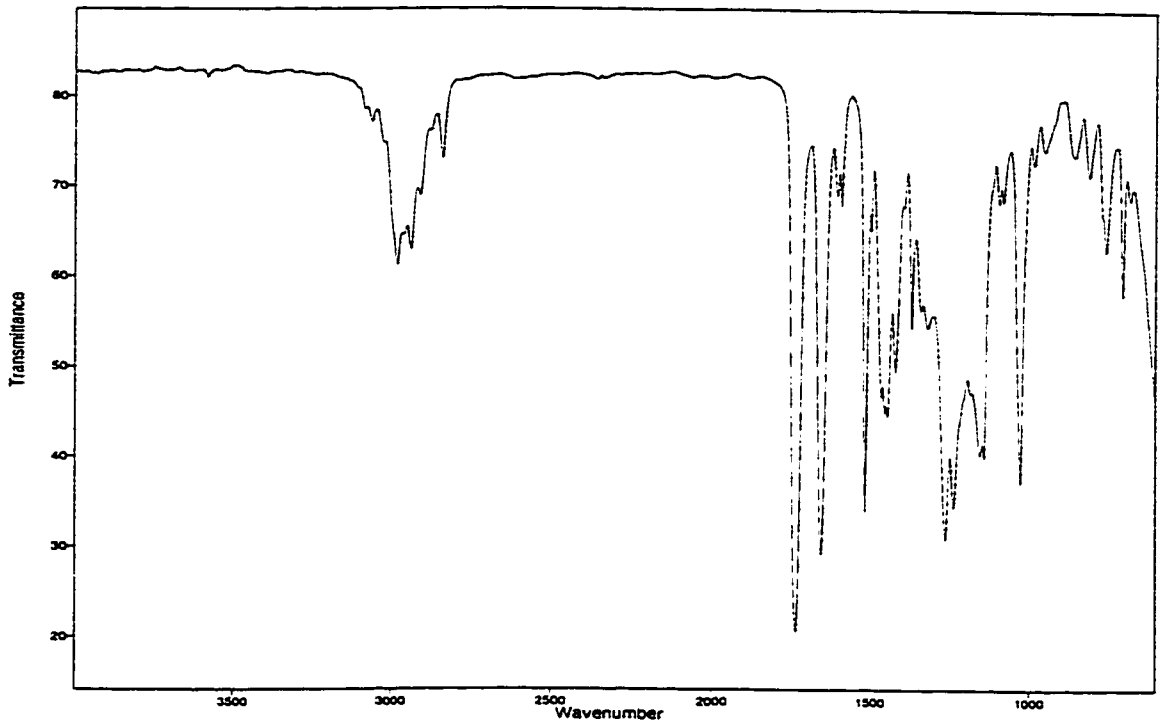


Figure A.2.47 Infrared Spectrum (thin film/NaCl) of compound (-)-**74**.

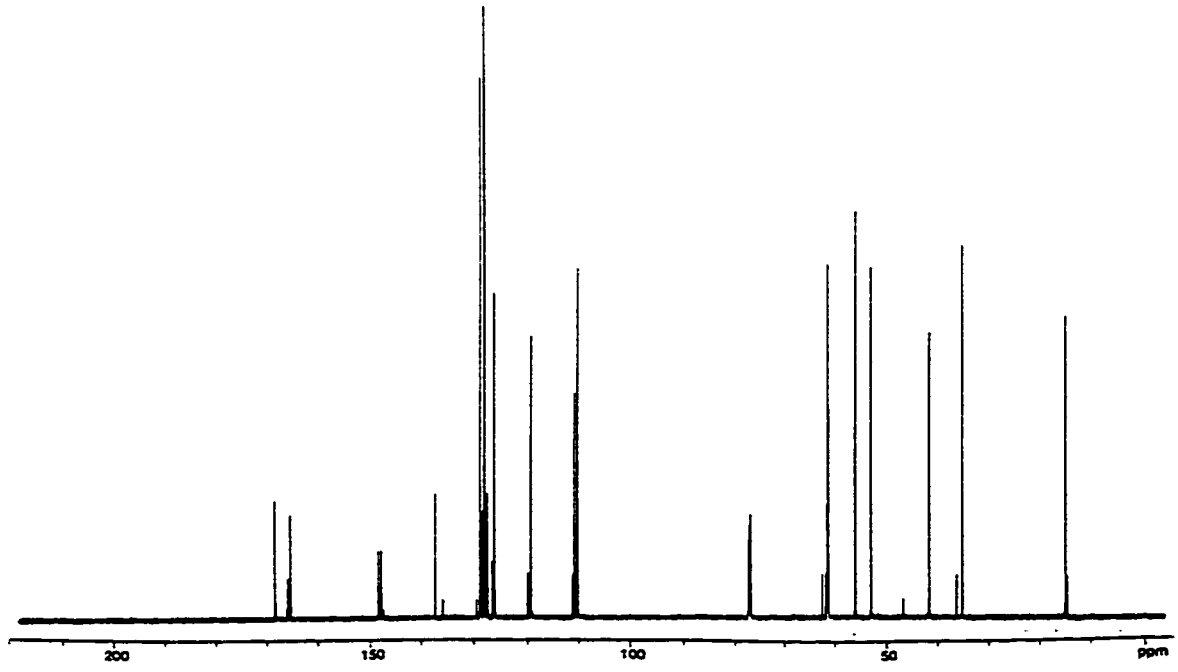


Figure A.2.48 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**74**.

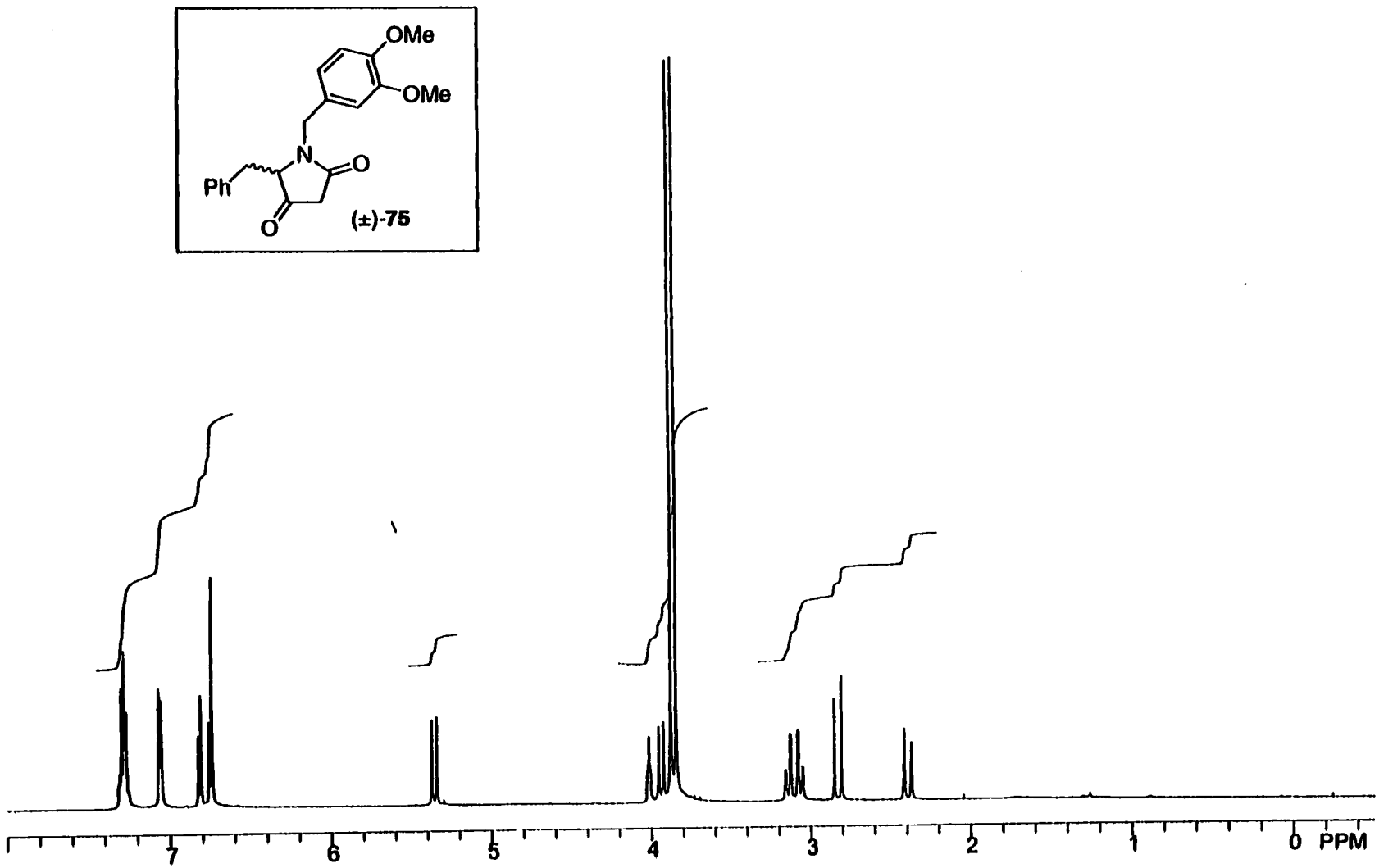


Figure A.2.49 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**75**.

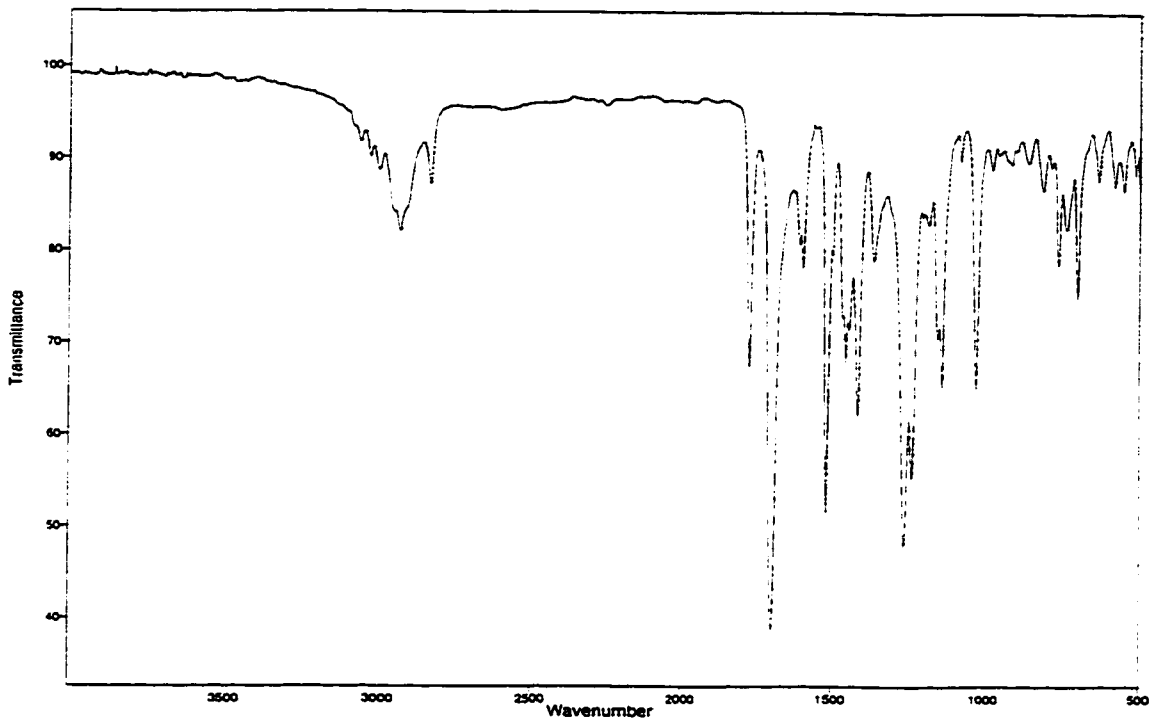


Figure A.2.50 Infrared Spectrum (thin film/NaCl) of compound (±)-**75**.

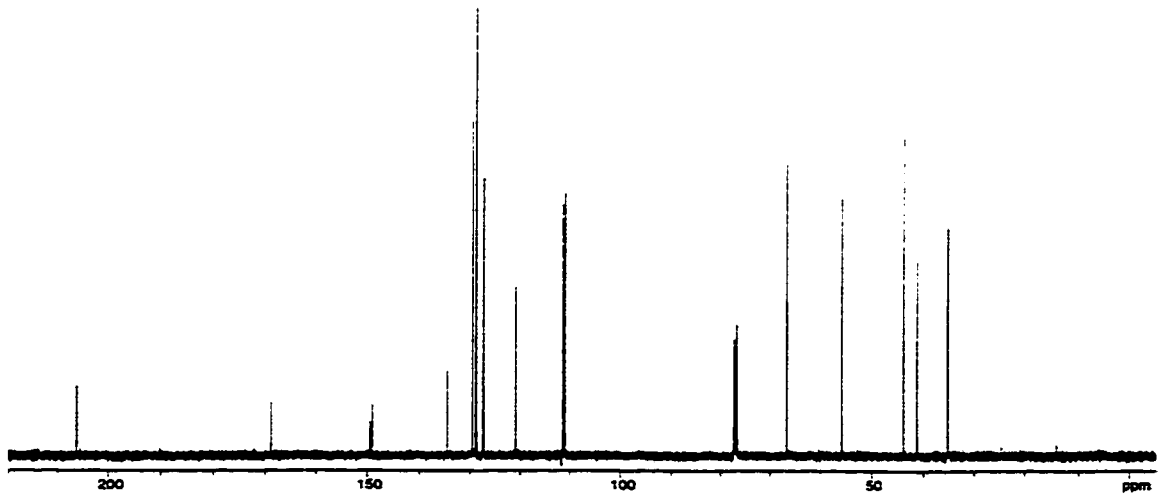


Figure A.2.51 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**75**.

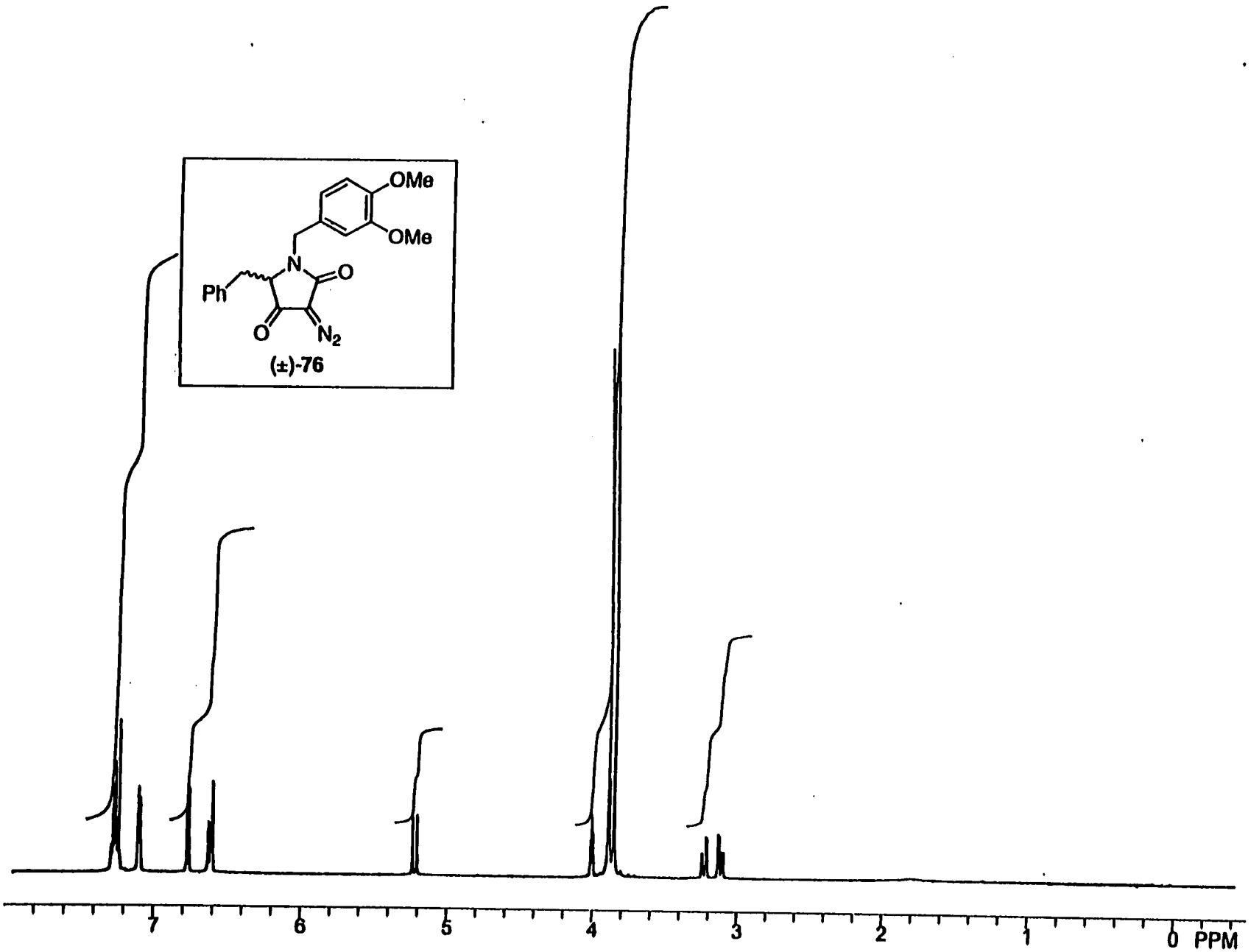


Figure A.2.52 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**76**.

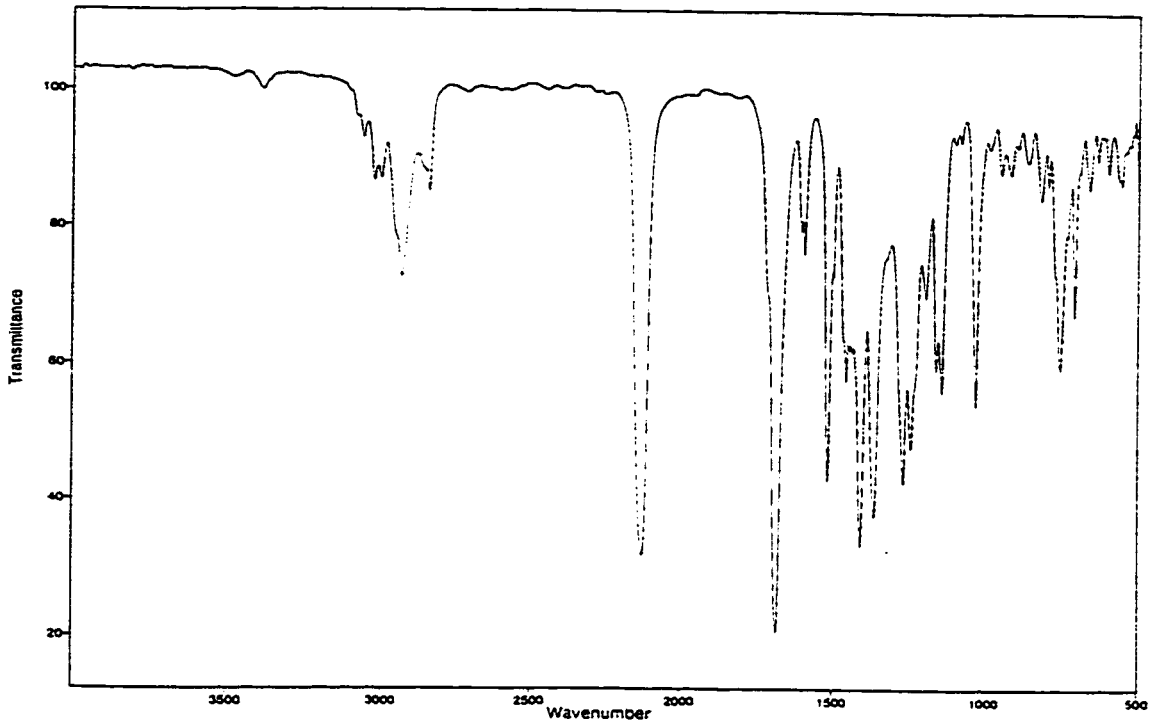


Figure A.2.53 Infrared Spectrum (thin film/NaCl) of compound (±)-**76**.

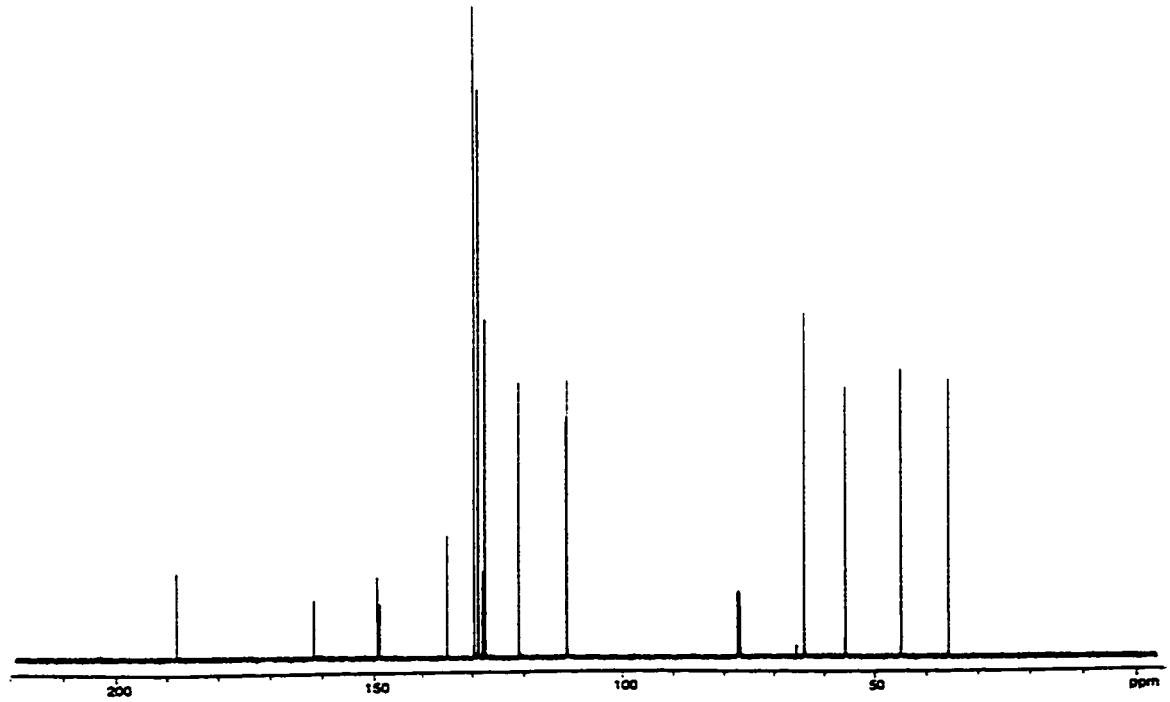


Figure A.2.54 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**76**.

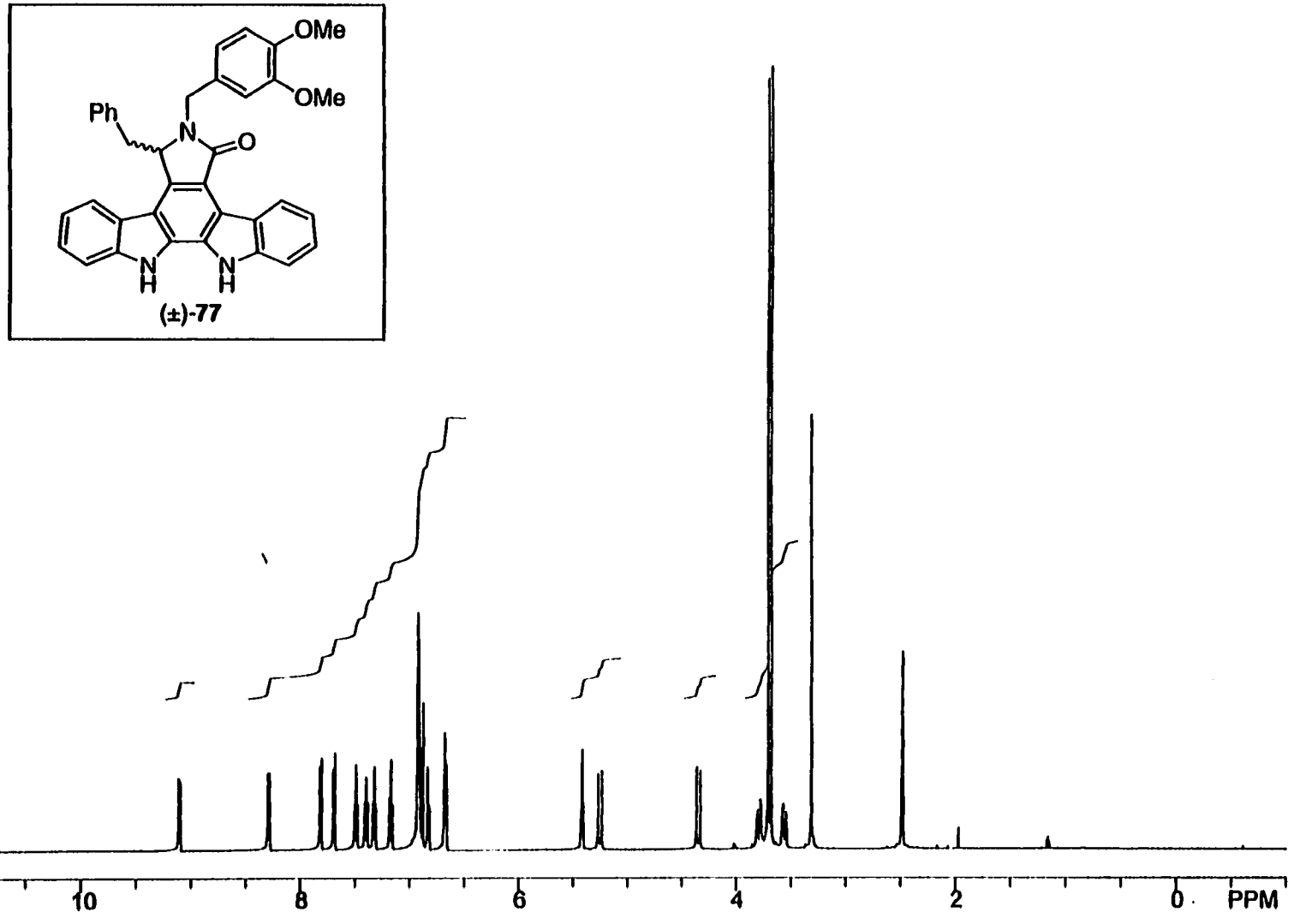


Figure A.2.55 ^{1}H NMR (500 MHz, DMSO-d_6) of compound (±)-**77**.

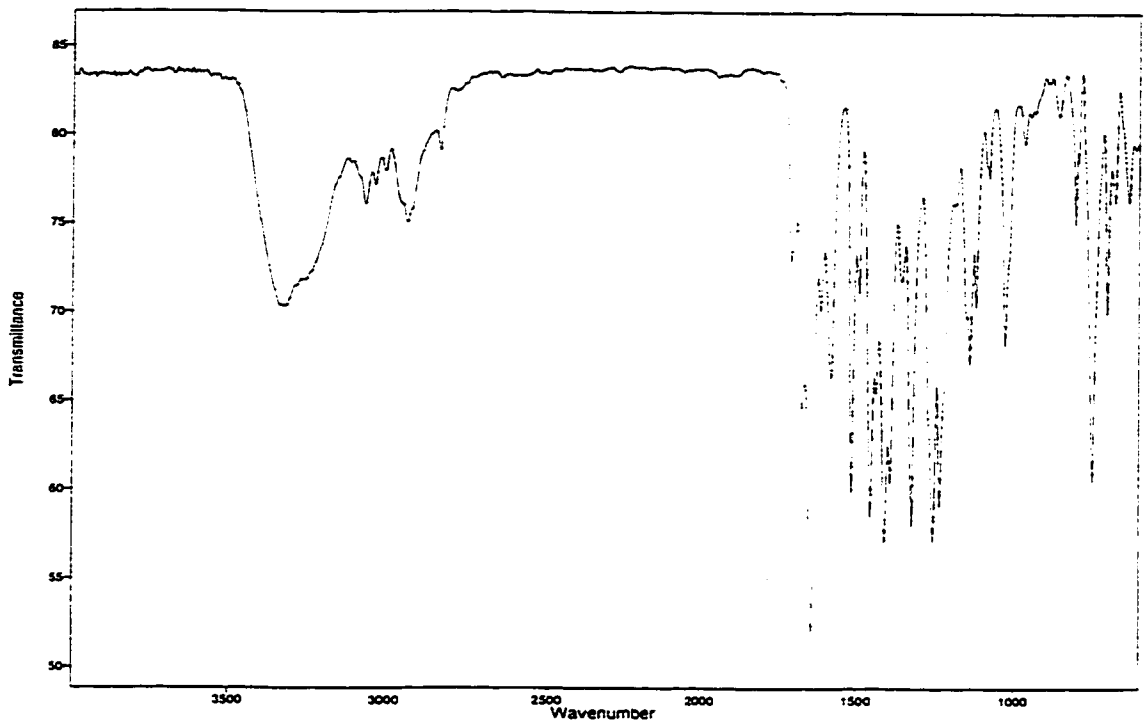


Figure A.2.56 Infrared Spectrum (thin film/NaCl) of compound (±)-**77**.

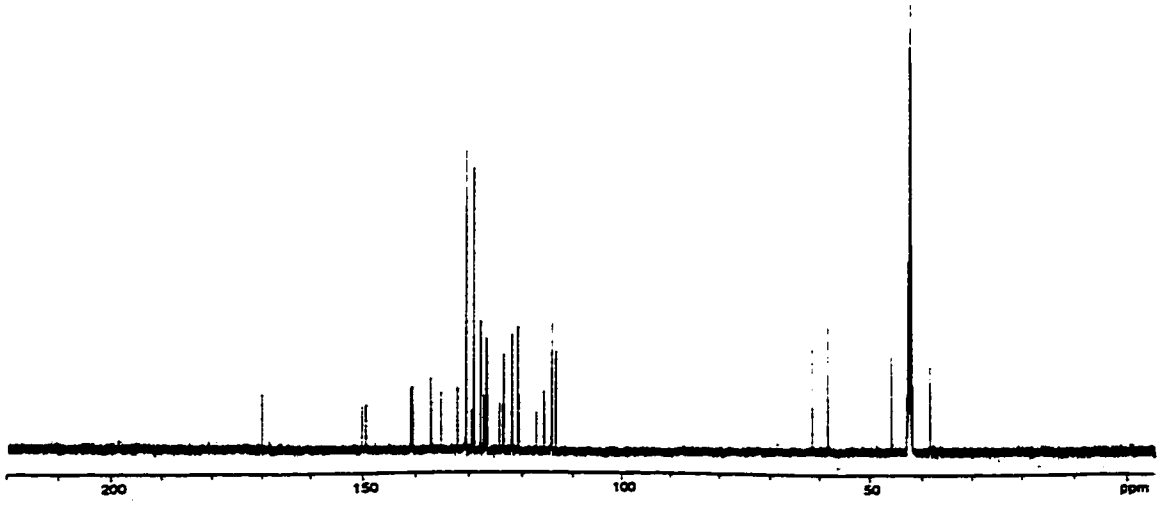


Figure A.2.57 ^{13}C NMR (125 MHz, DMSO-d_6) of compound (±)-**77**.

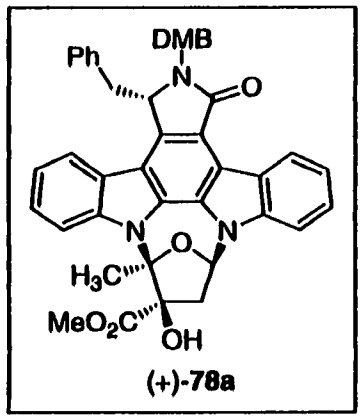


Figure A.2.58 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**78a**.

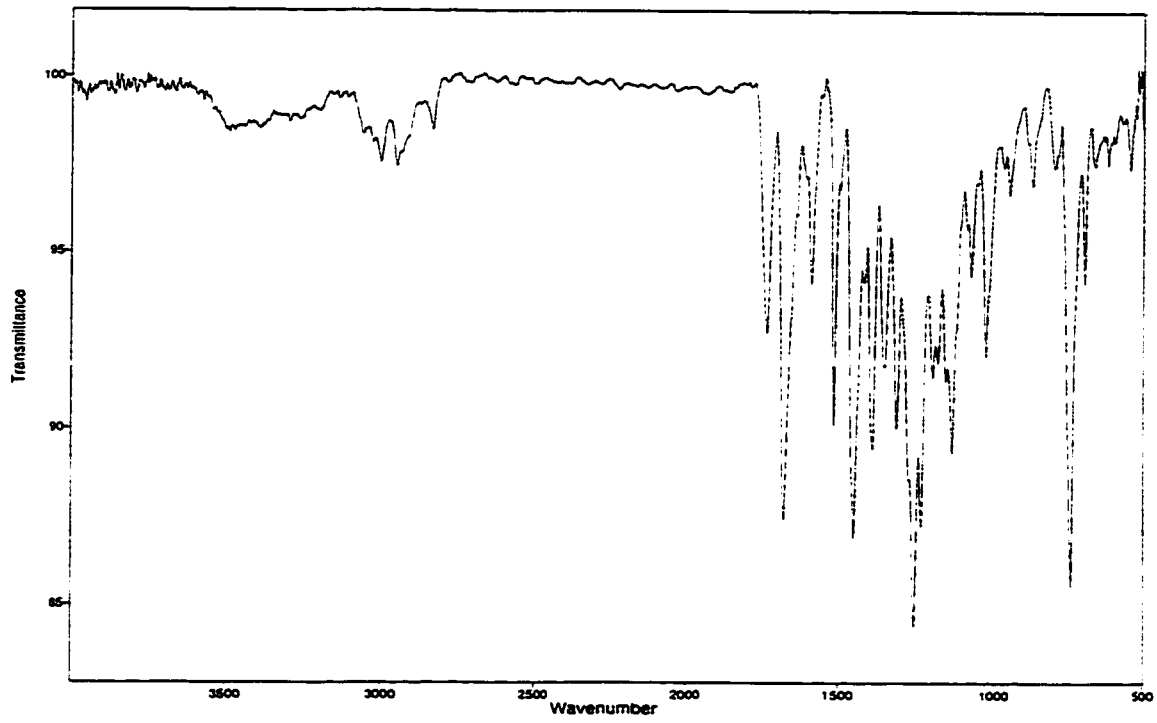


Figure A.2.59 Infrared Spectrum (thin film/NaCl) of compound (+)-**78a**.

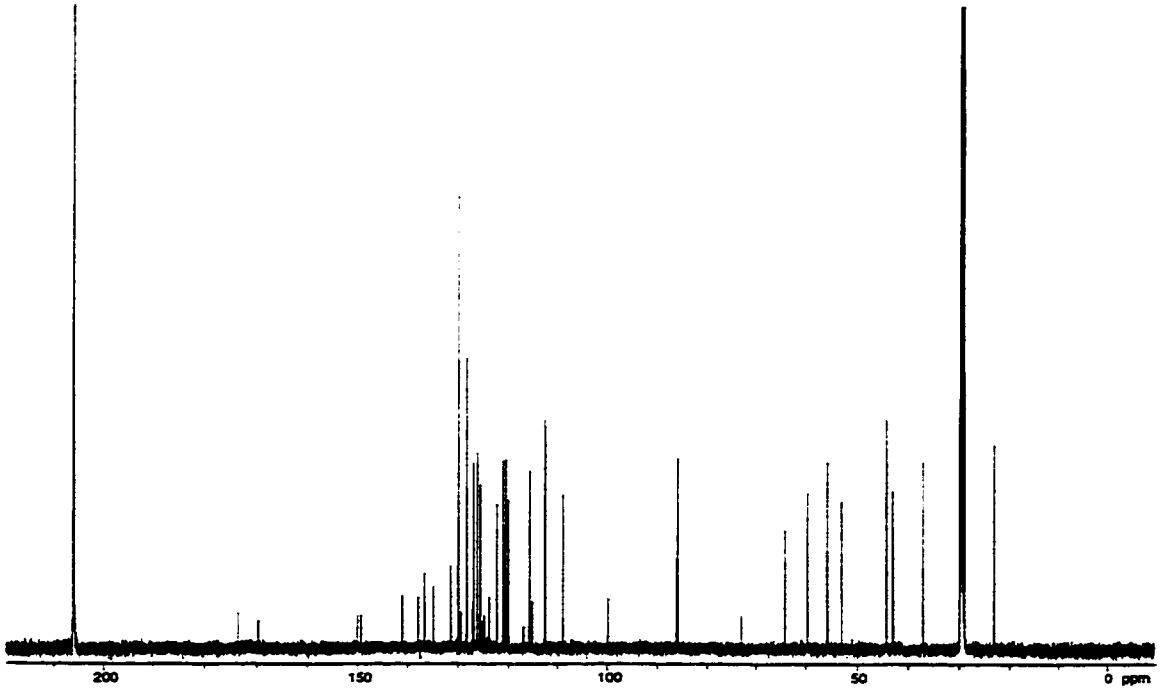


Figure A.2.60 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**78a**.

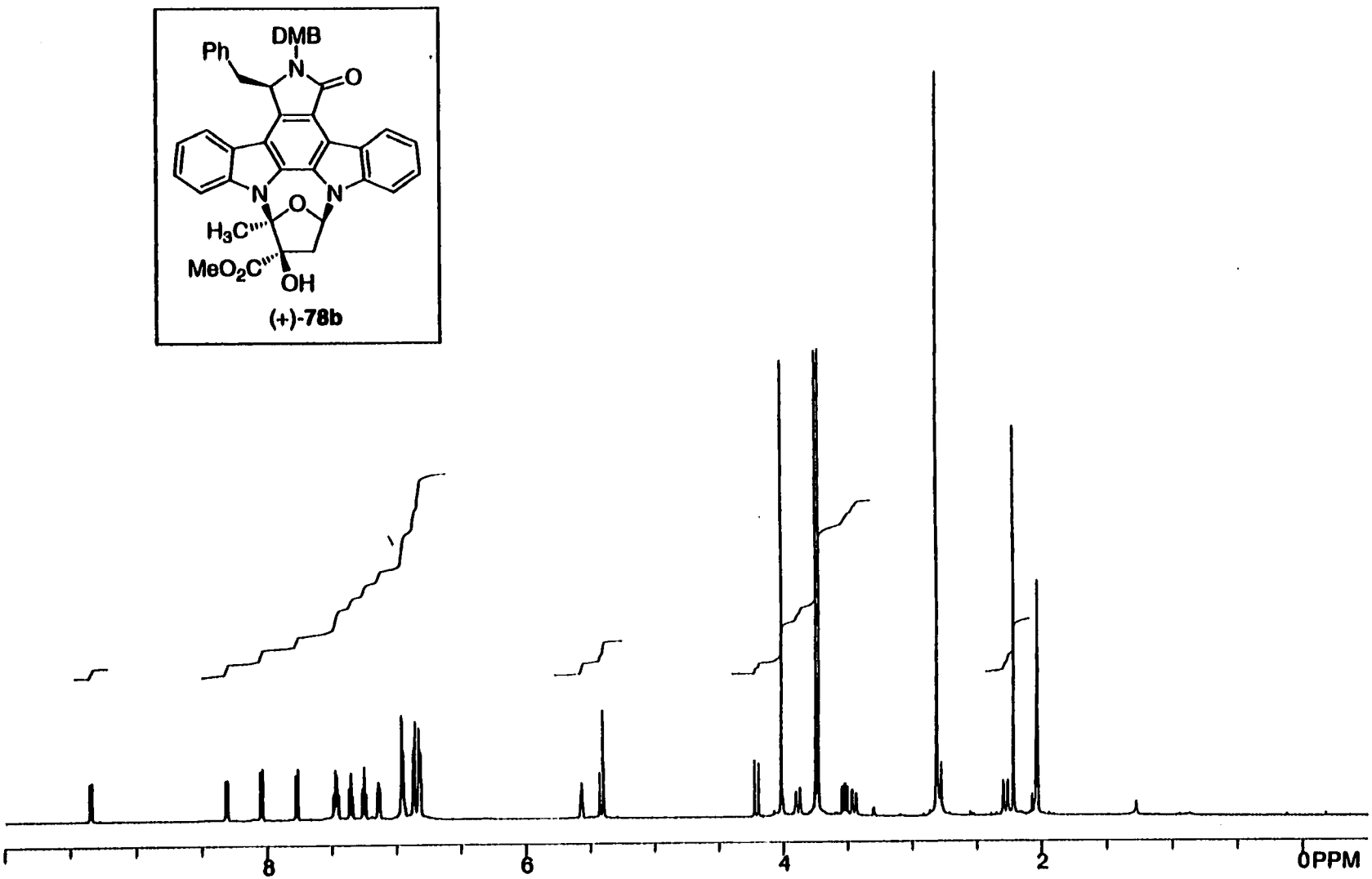


Figure A.2.61 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**78b**.

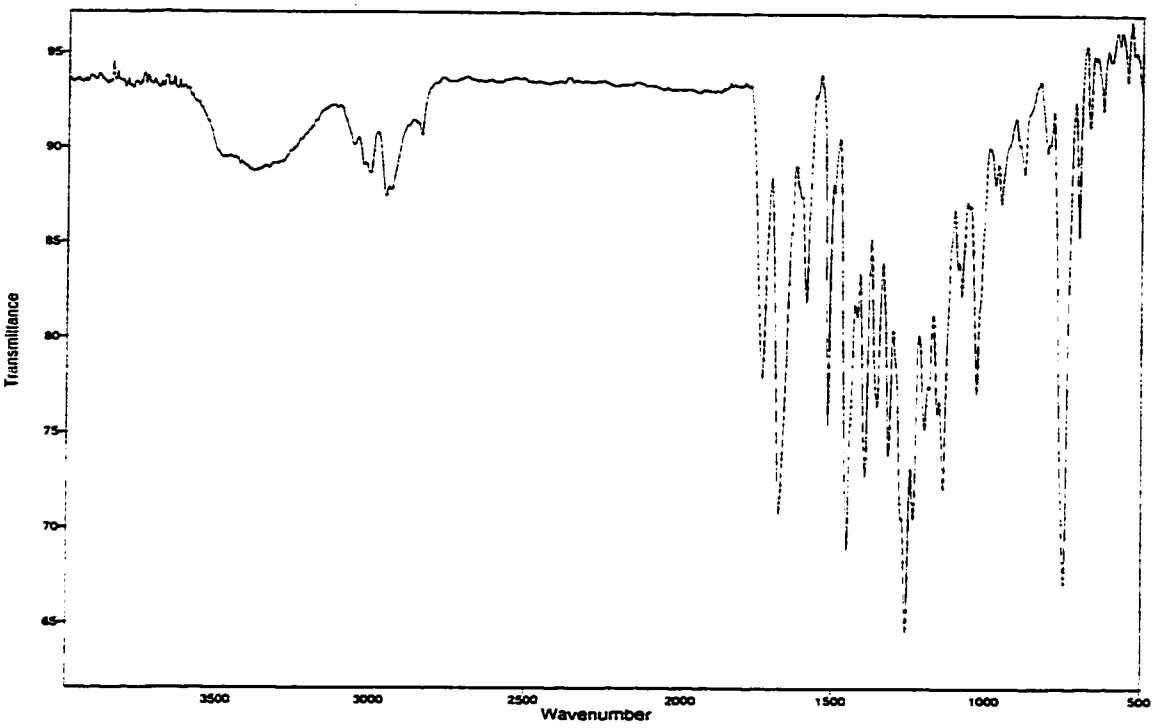


Figure A.2.62 Infrared Spectrum (thin film/NaCl) of compound (+)-**78b**.

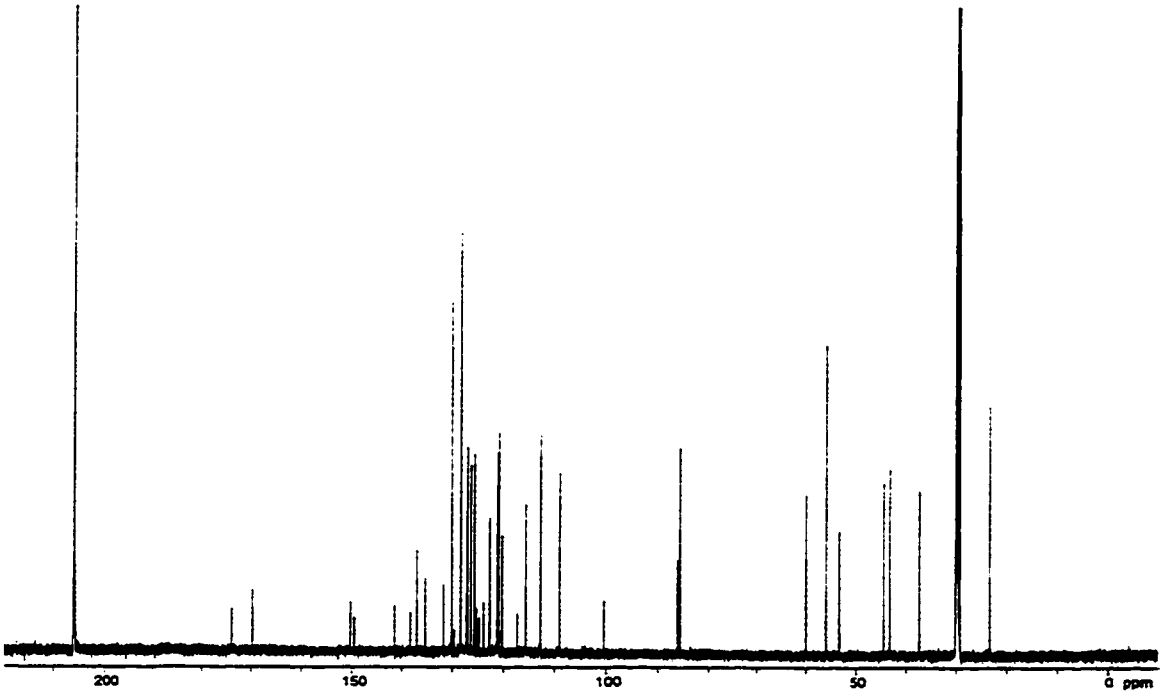


Figure A.2.63 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**78b**.

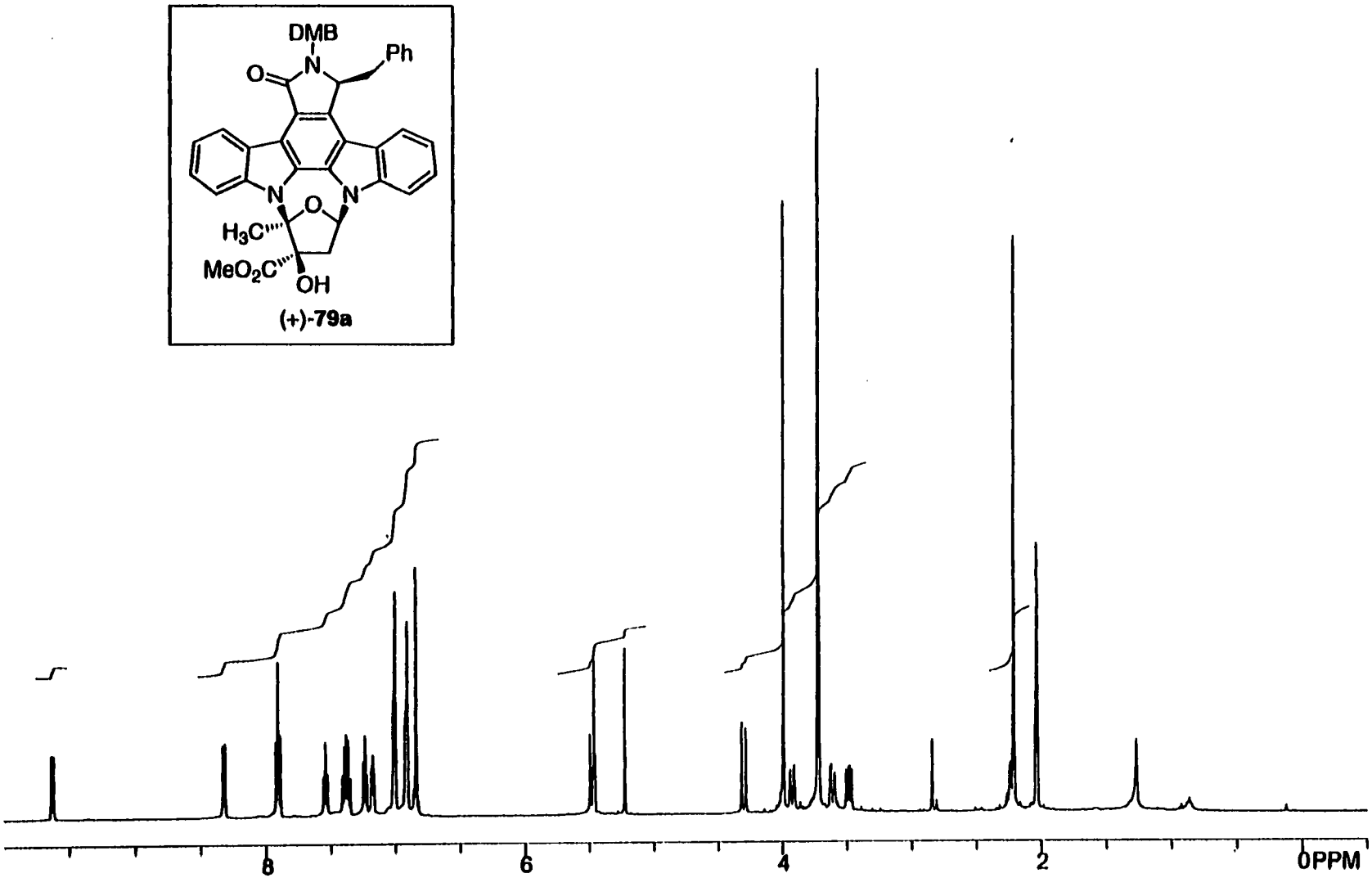


Figure A.2.64 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**79a**.

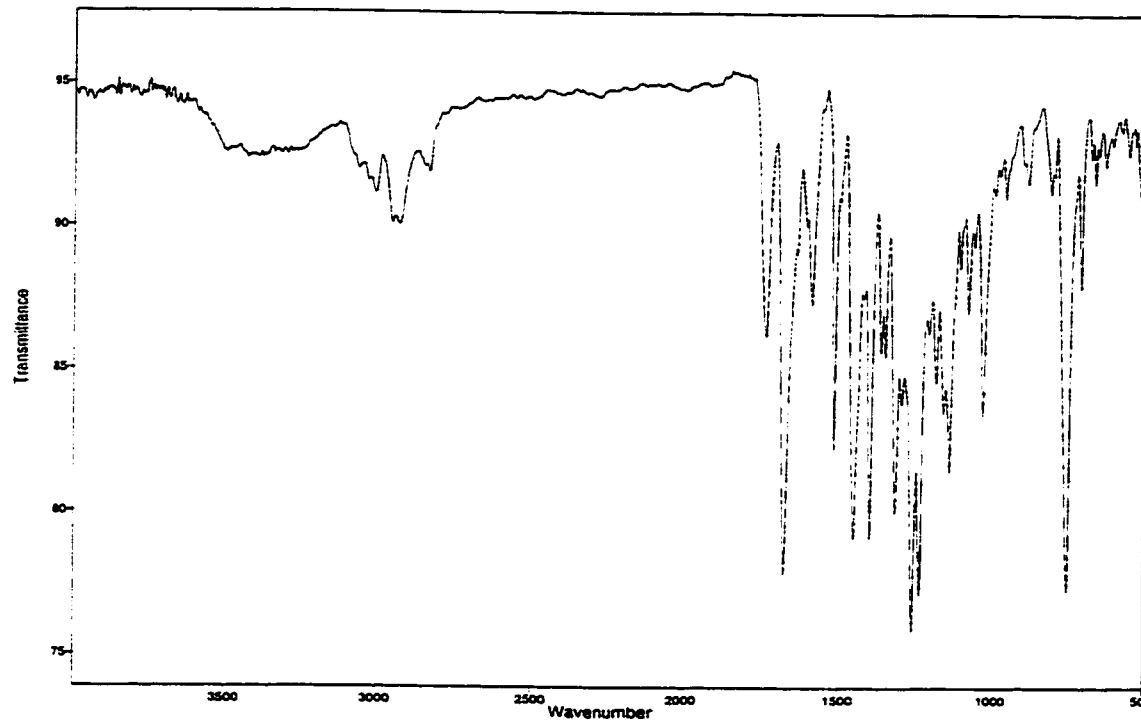


Figure A.2.65 Infrared Spectrum (thin film/NaCl) of compound (+)-**79a**.

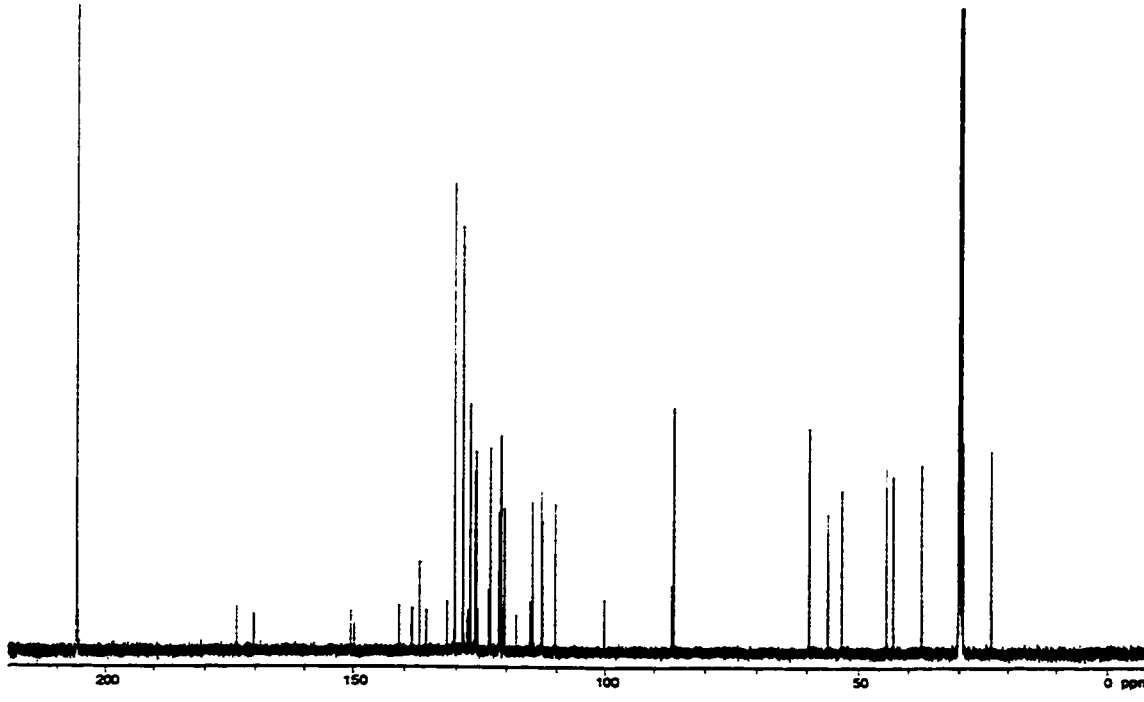


Figure A.2.66 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**79a**.

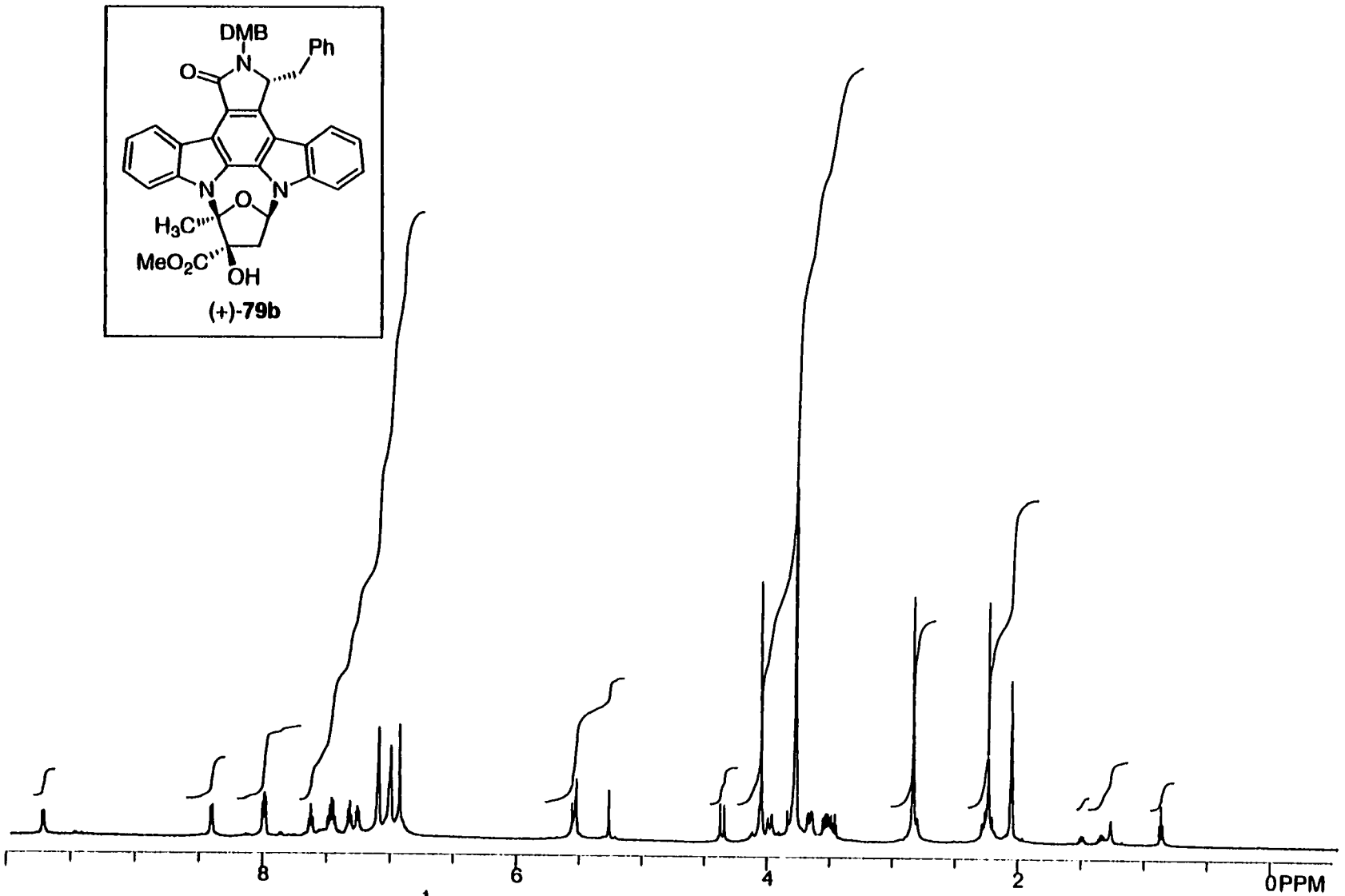


Figure A.2.67 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**79b**.

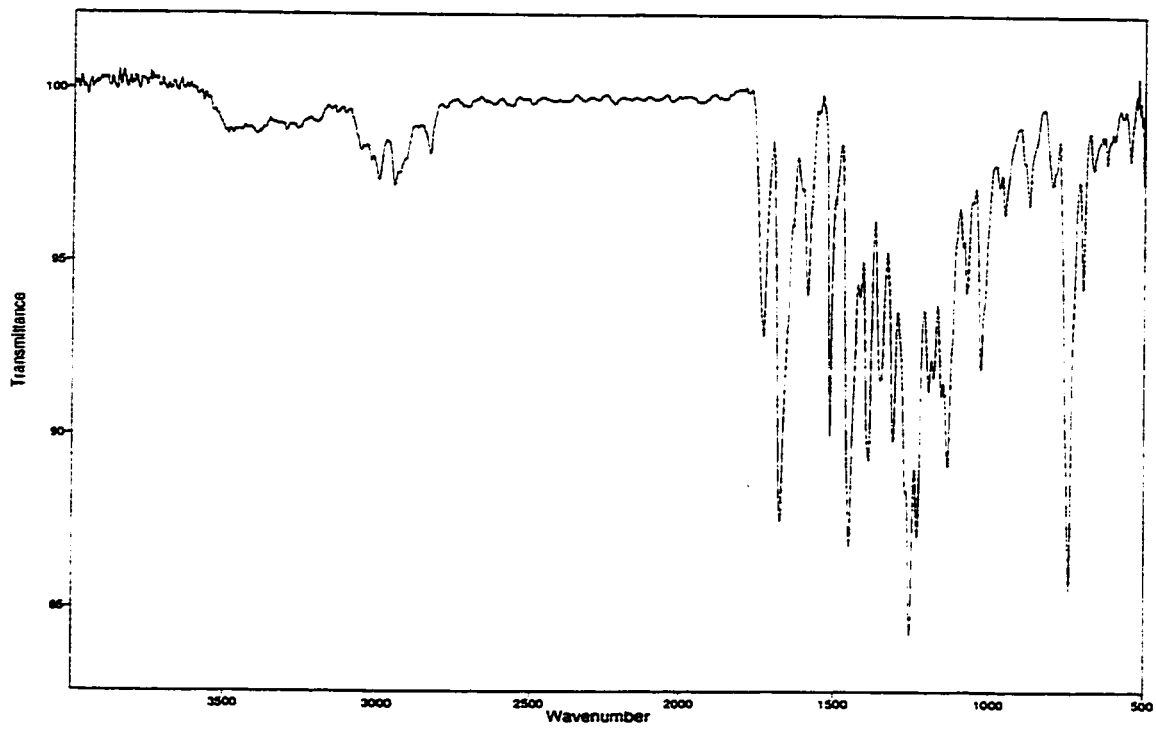


Figure A.2.68 Infrared Spectrum (thin film/NaCl) of compound (+)-**79b**.

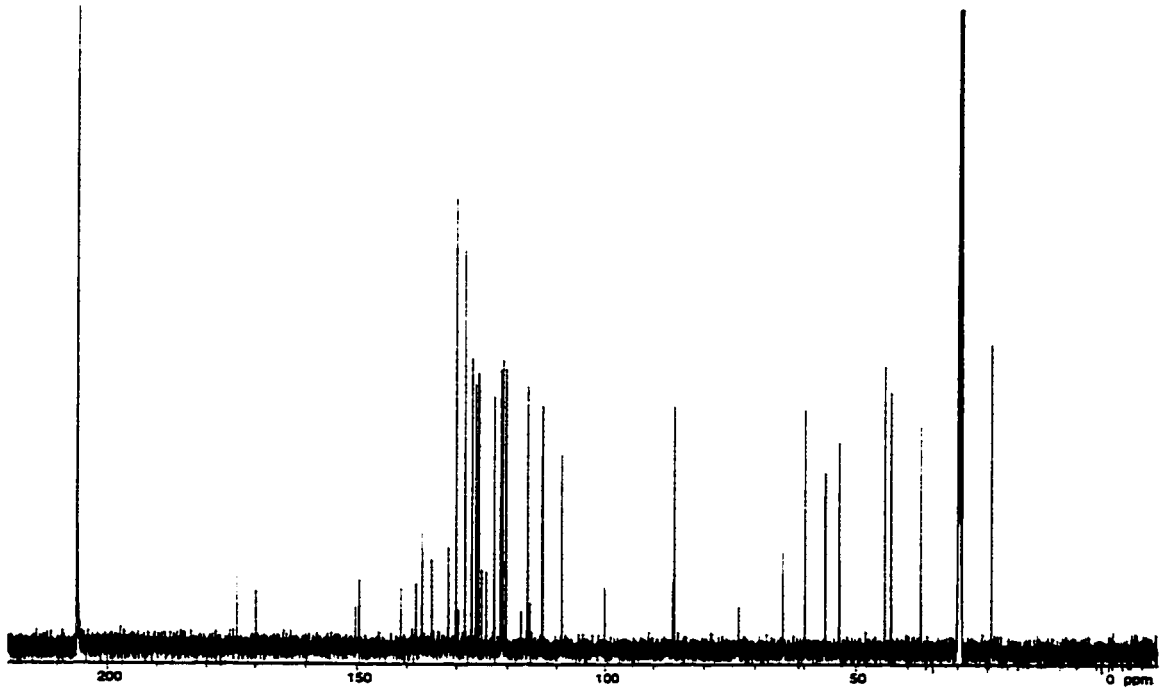


Figure A.2.69 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**79b**.

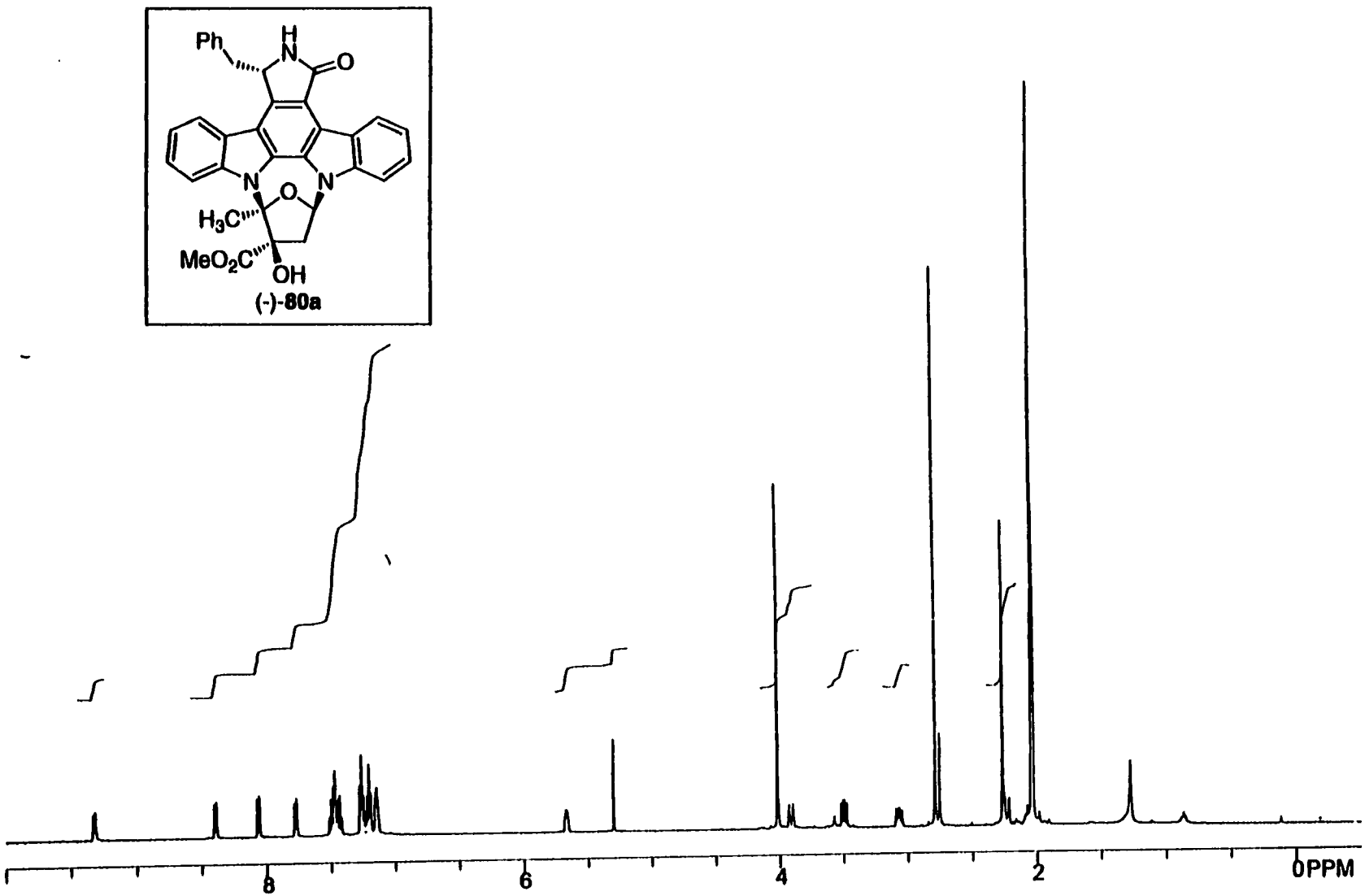


Figure A.2.70 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**80a**.

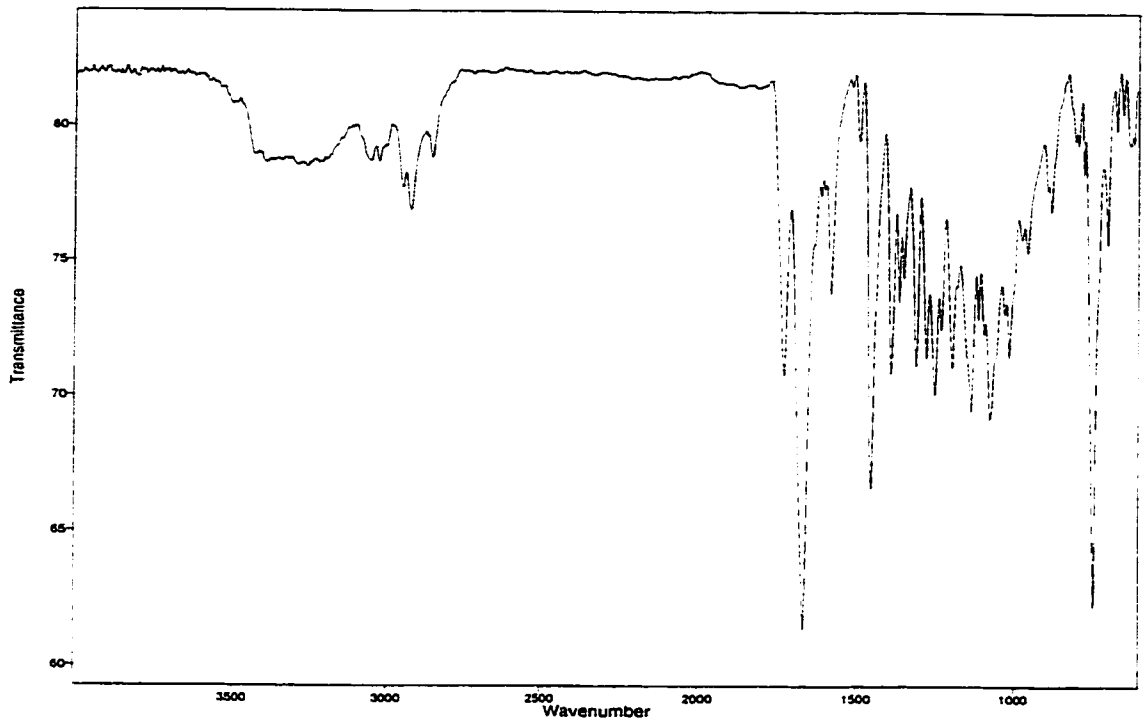


Figure A.2.71 Infrared Spectrum (thin film/NaCl) of compound (-)-**80a**.

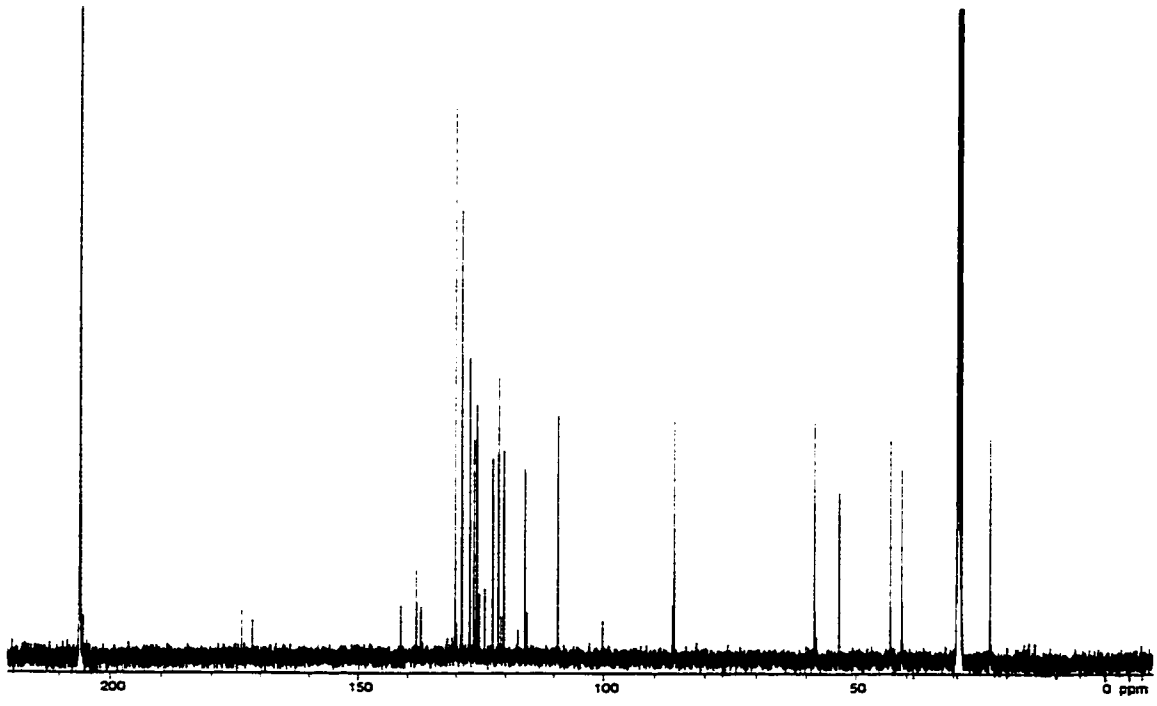


Figure A.2.72 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**80a**.

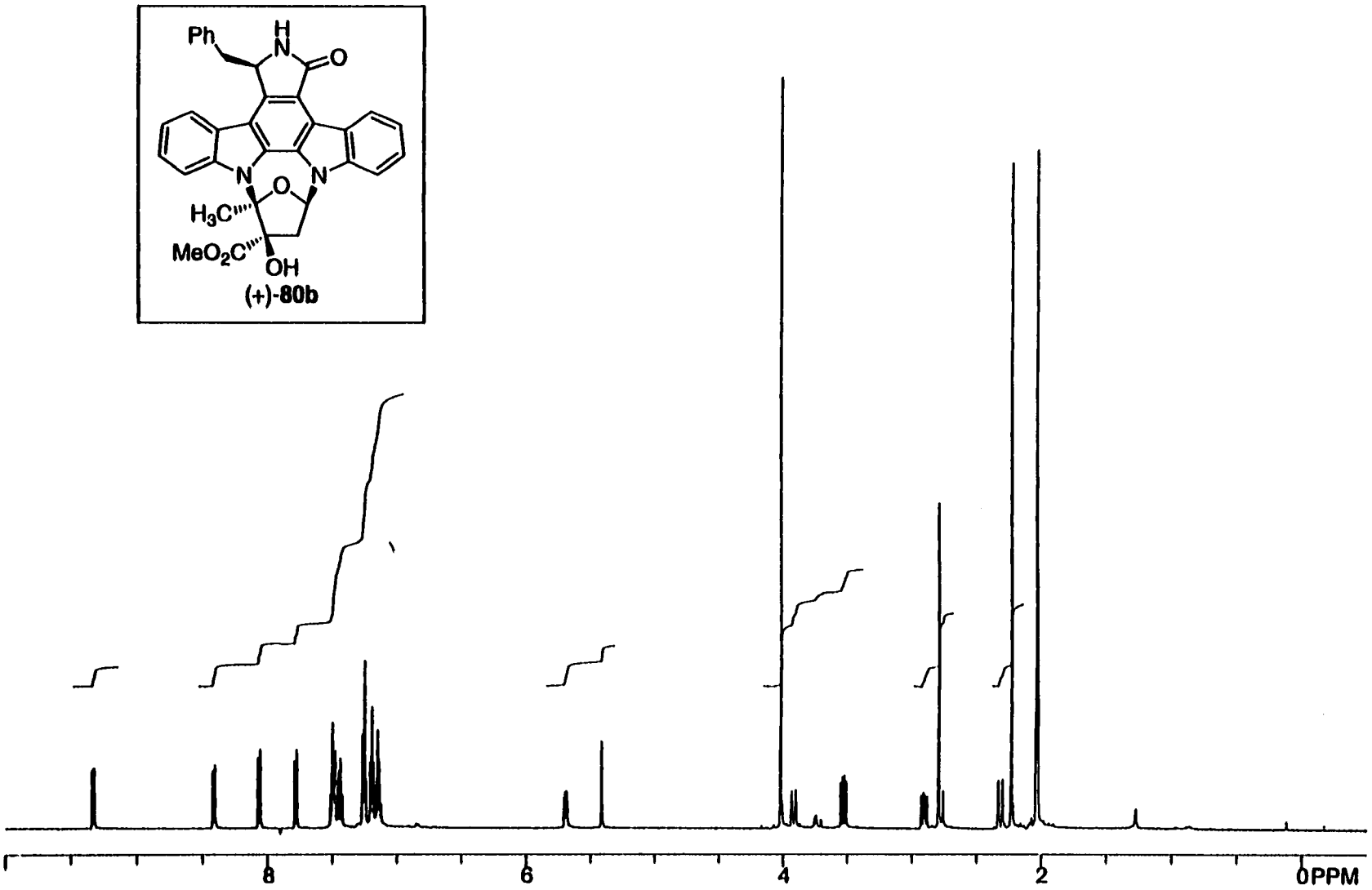


Figure A.2.73 ^{1}H NMR (500 MHz, acetone-d_6) of compound (+)-**80b**.

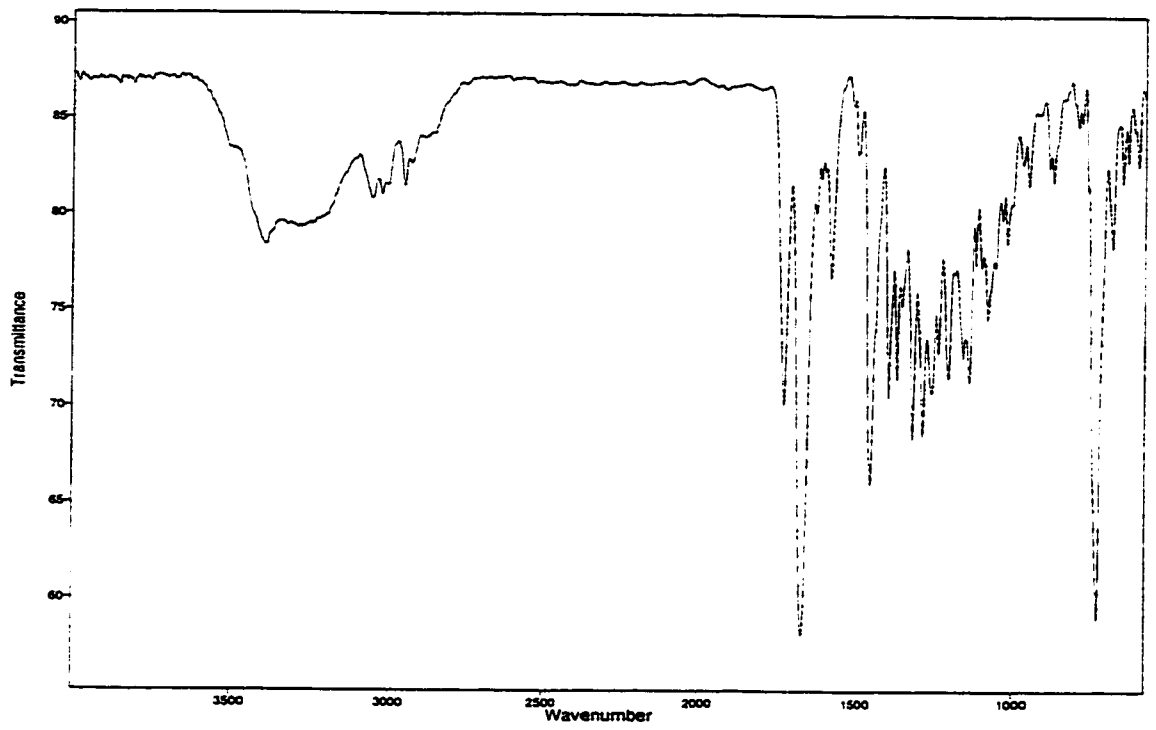


Figure A.2.74 Infrared Spectrum (thin film/NaCl) of compound (+)-**80b**.

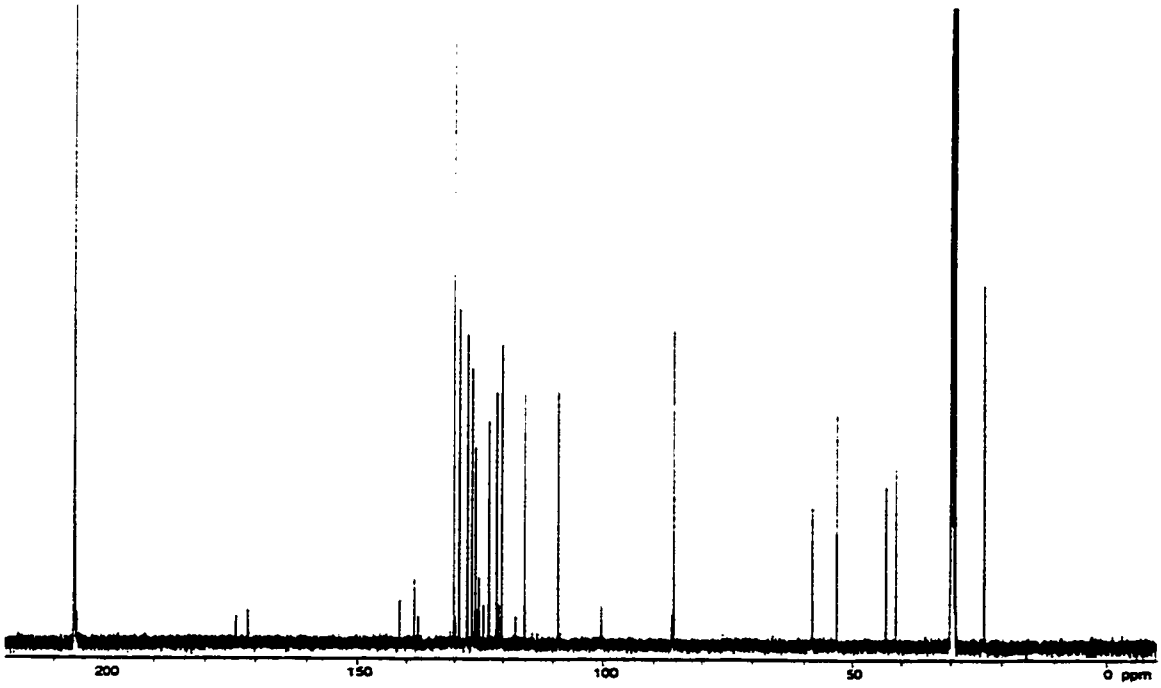


Figure A.2.75 ^{13}C NMR (125 MHz, acetone-d_6) of compound (+)-**80b**.

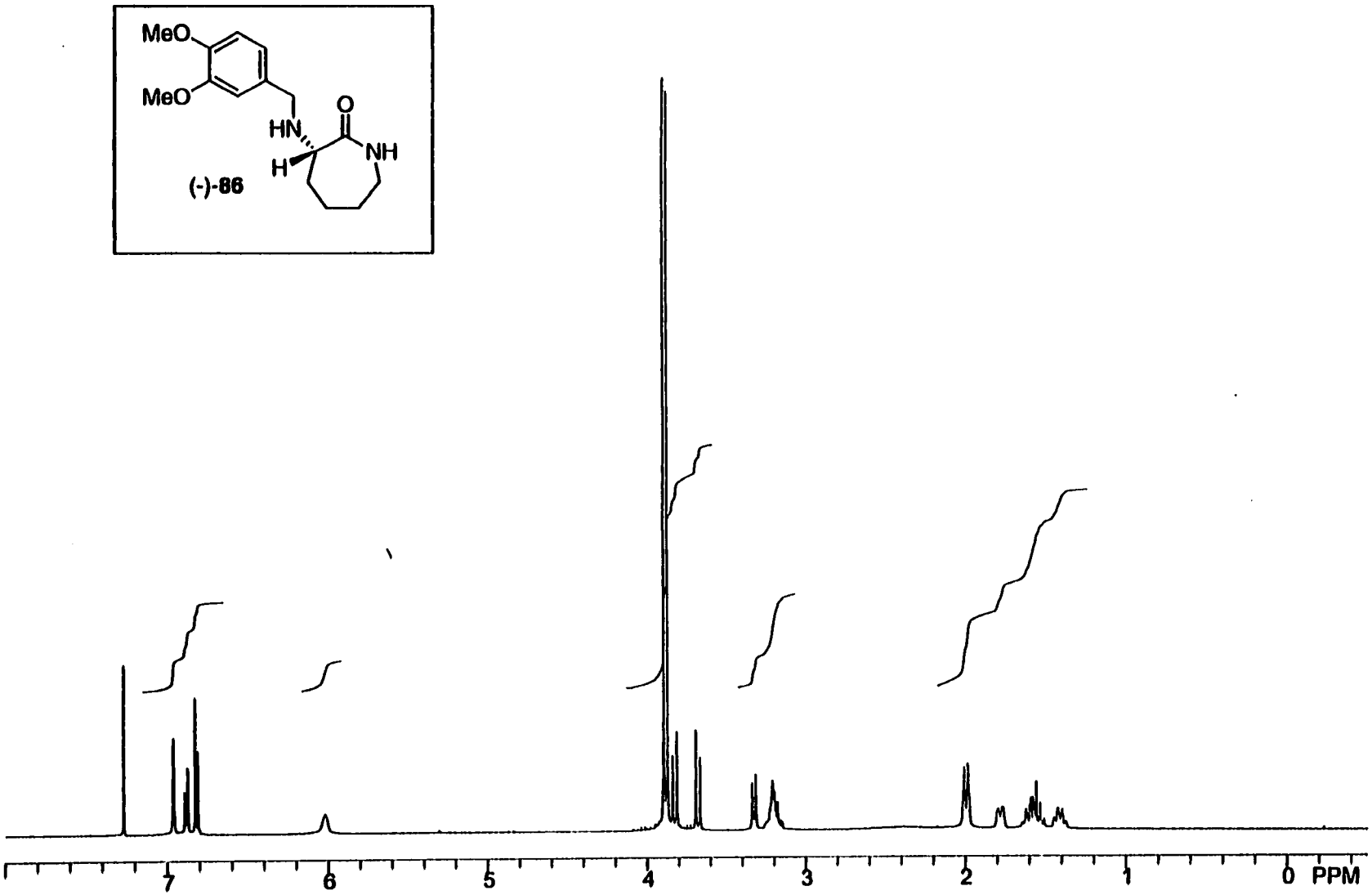


Figure A.2.76 ^{1}H NMR (500 MHz,$CDCl_3$) of compound (-)-**86**.

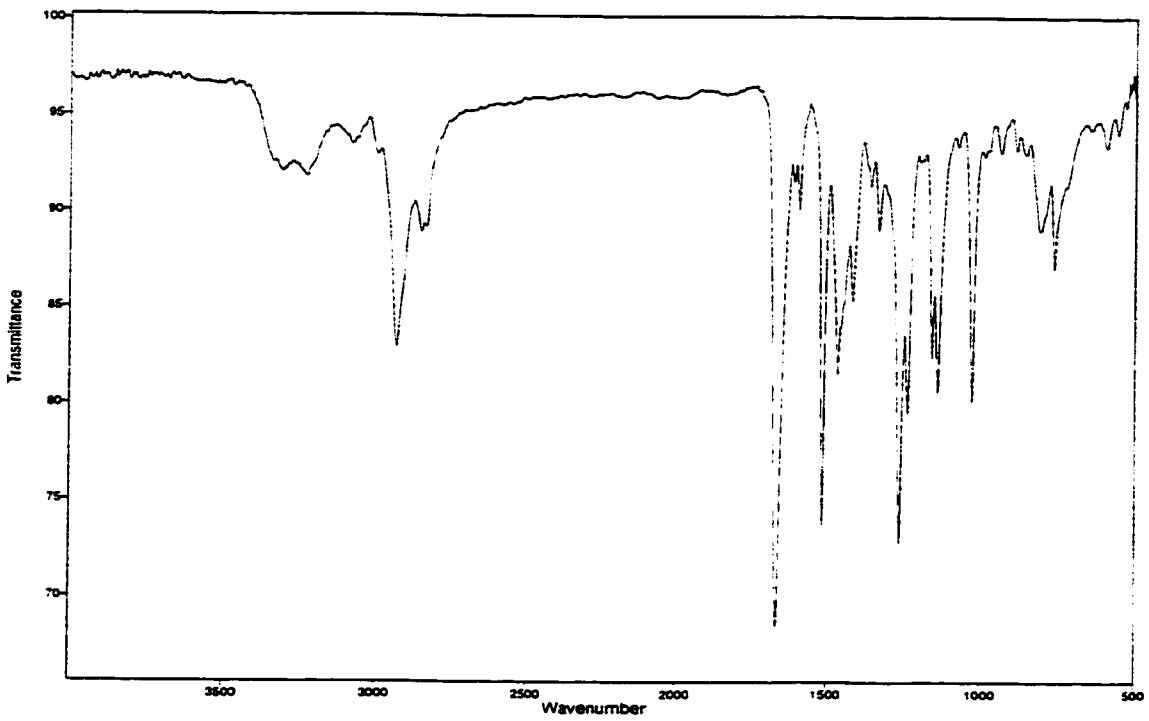


Figure A.2.77 Infrared Spectrum (thin film/NaCl) of compound (-)-**86**.

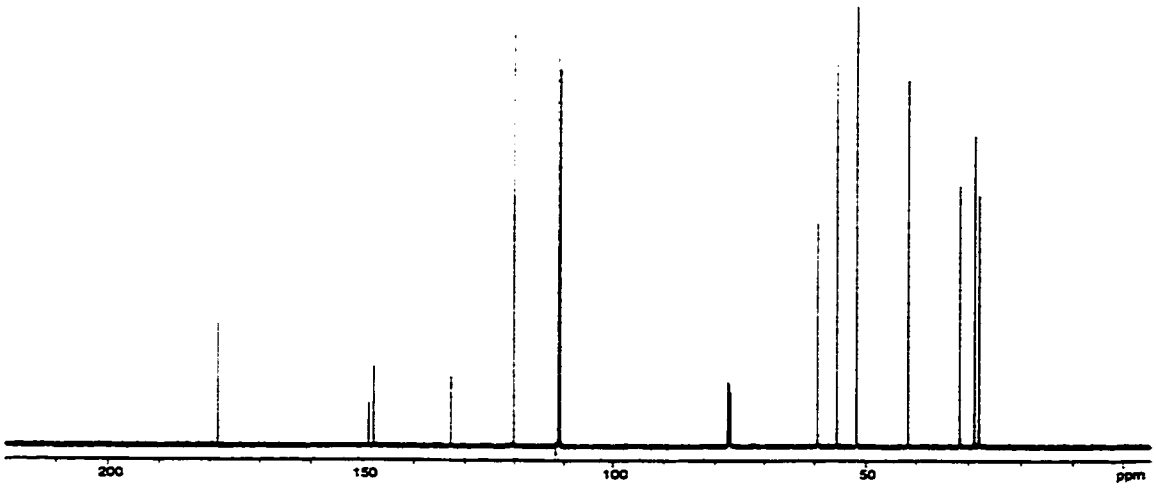


Figure A.2.78 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**86**.

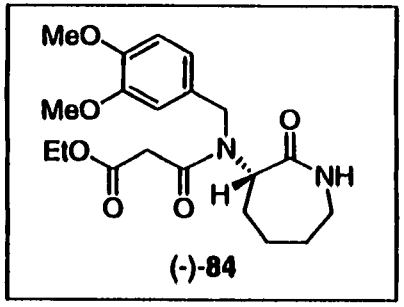


Figure A.2.79 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**84**.

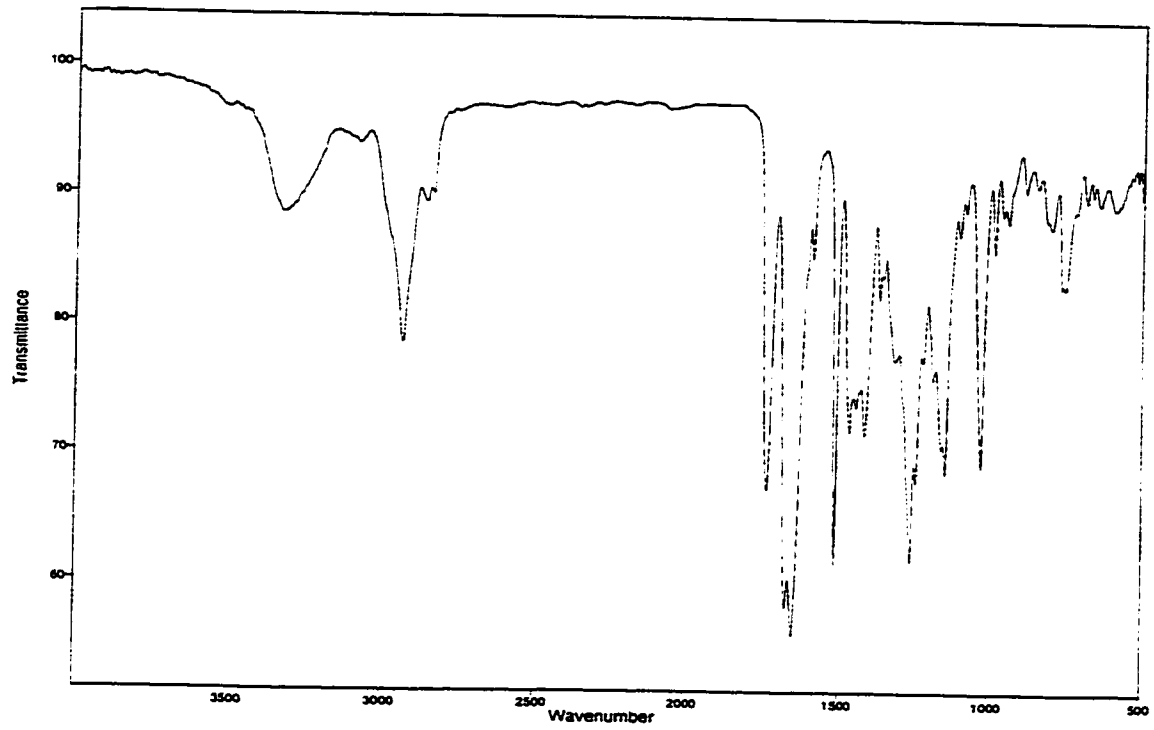


Figure A.2.80 Infrared Spectrum (thin film/NaCl) of compound (-)-**84**.

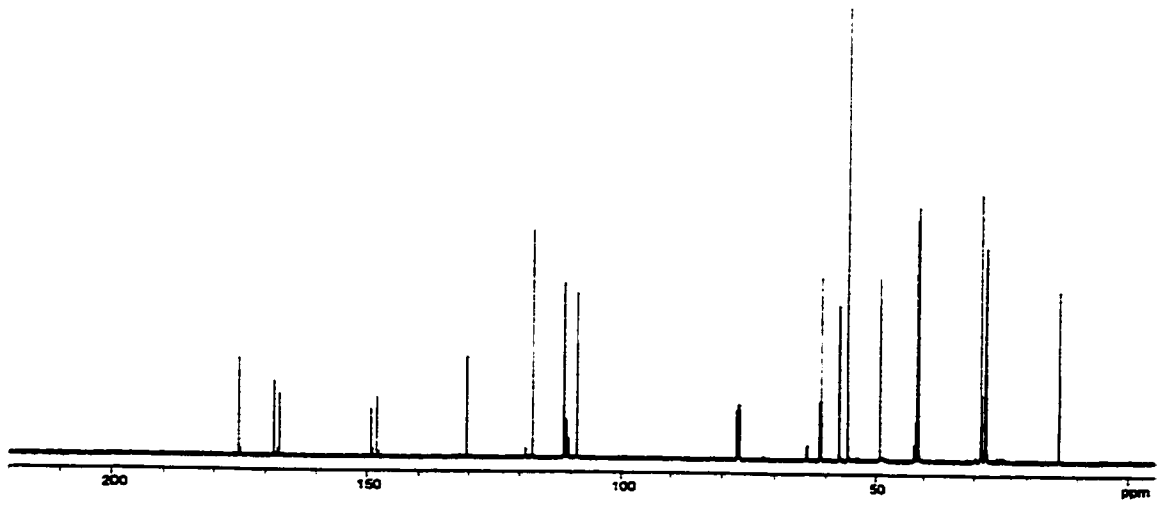


Figure A.2.81 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**84**.

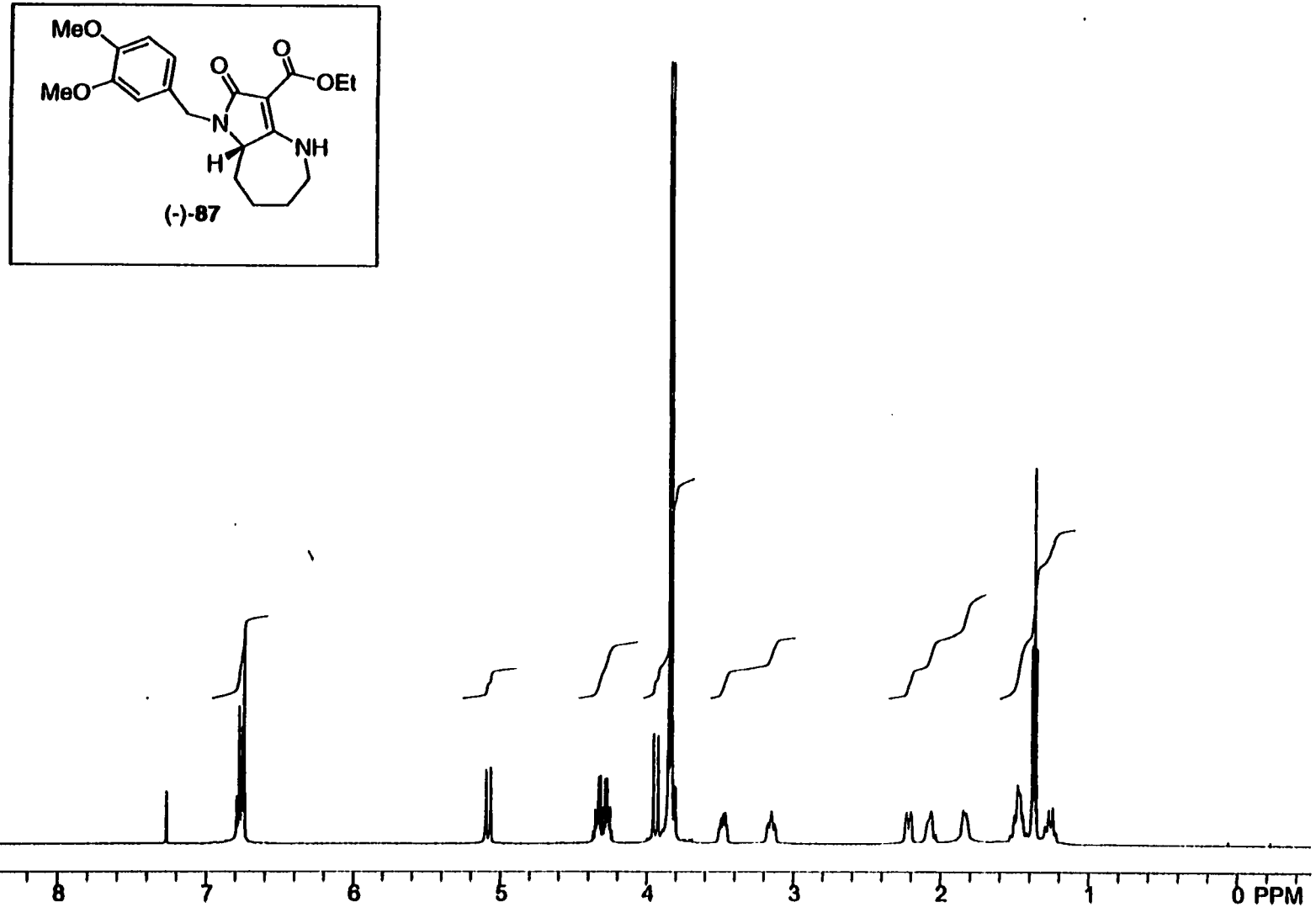


Figure A.2.82 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**87**.

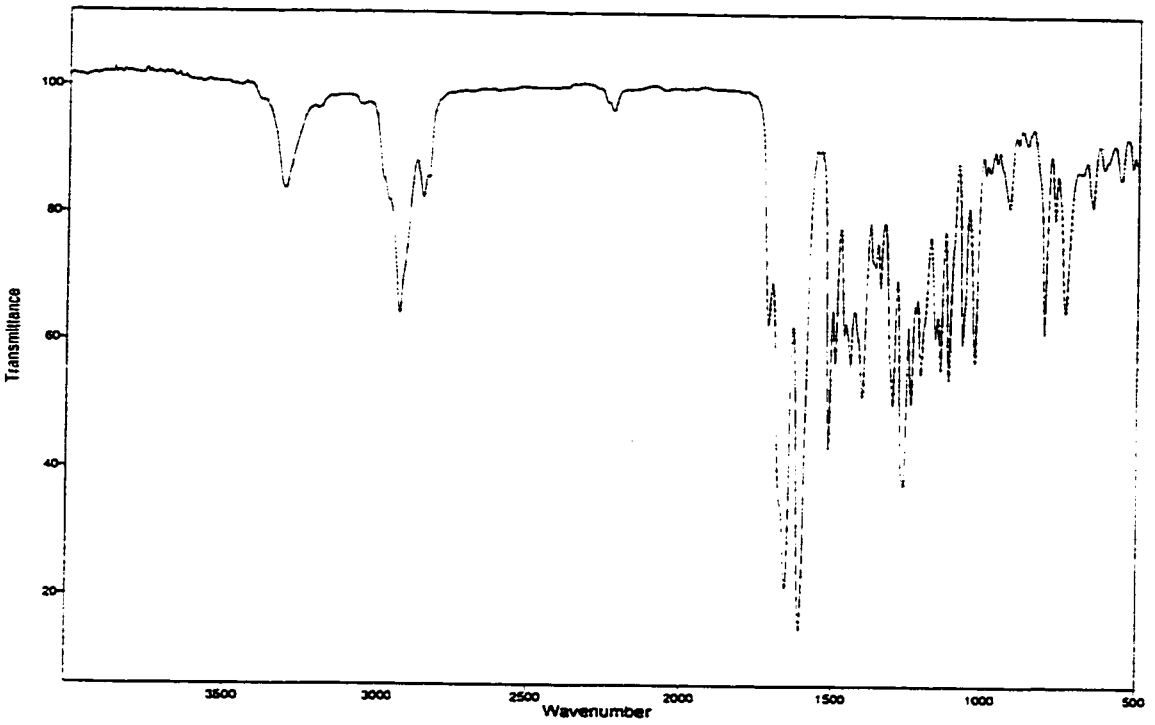


Figure A.2.83 Infrared Spectrum (thin film/NaCl) of compound (-)-**87**.

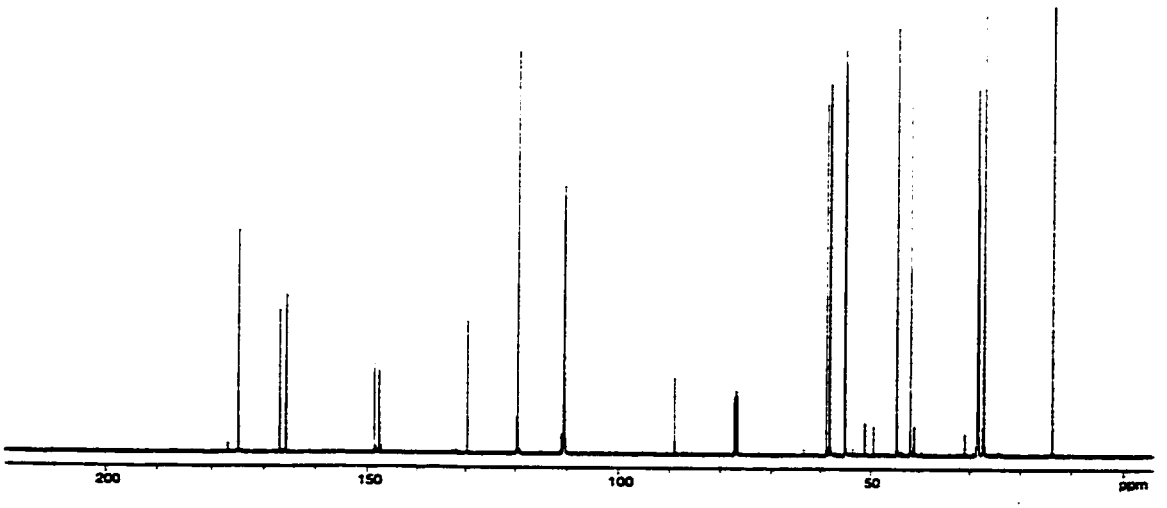


Figure A.2.84 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**87**.

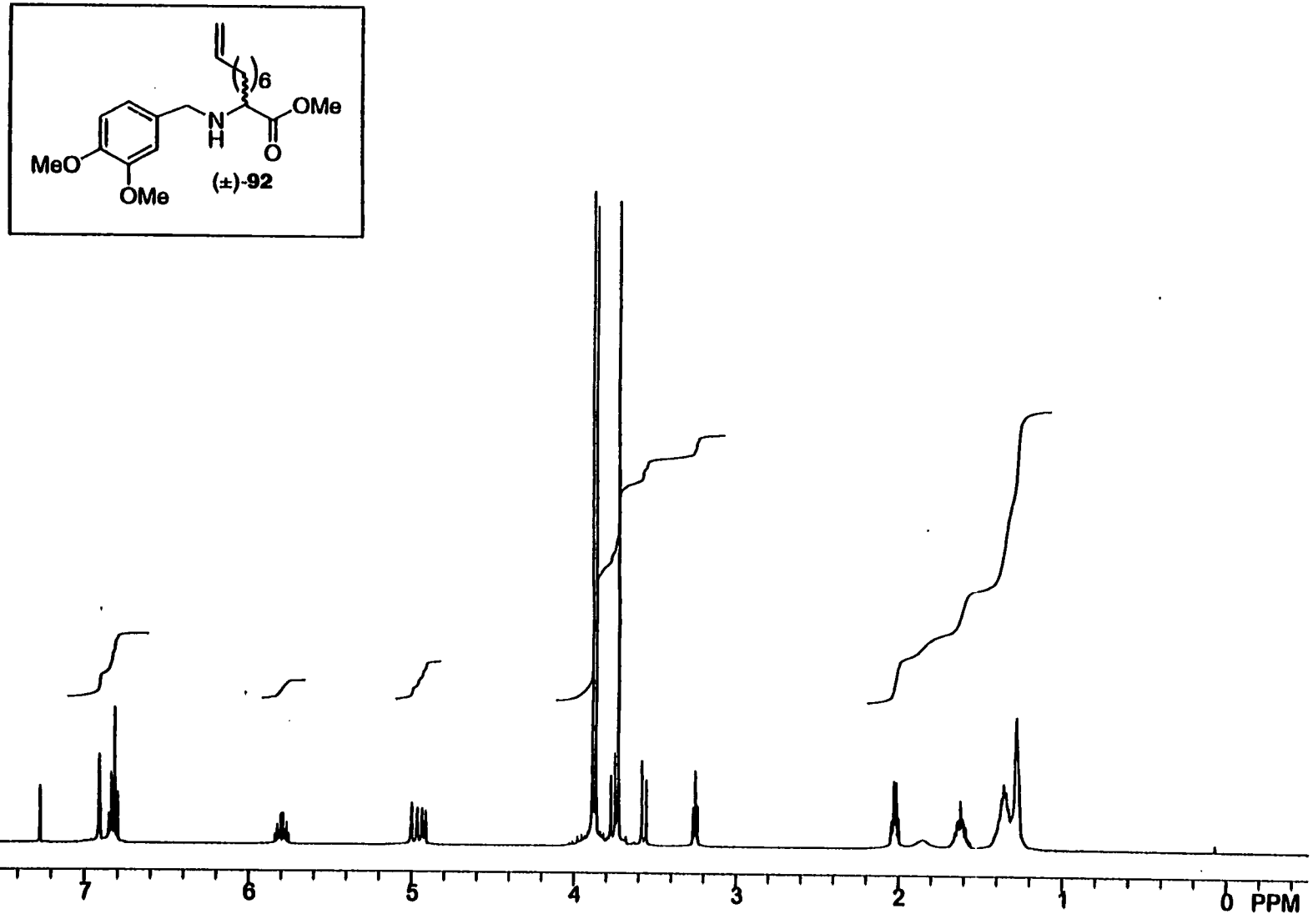


Figure A.2.85 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**92**.

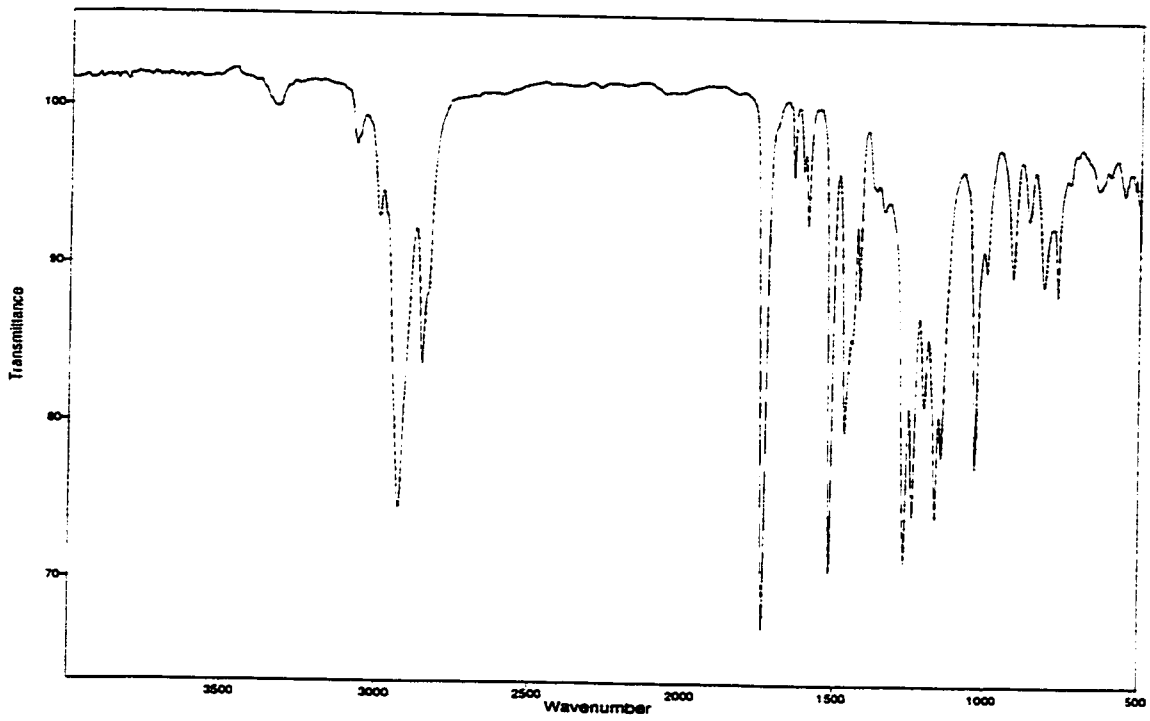


Figure A.2.86 Infrared Spectrum (thin film/NaCl) of compound (±)-**92**.

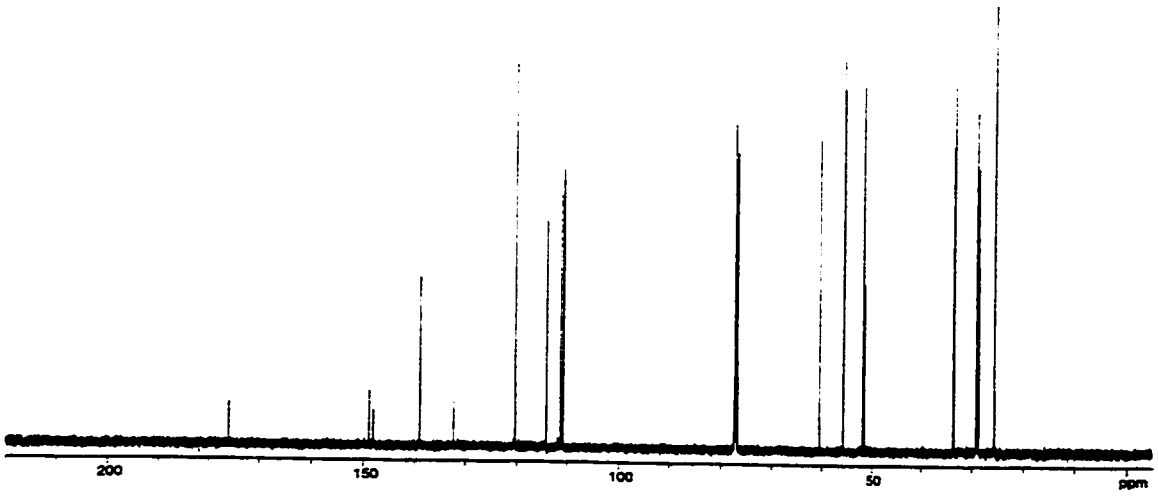


Figure A.2.87 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**92**.

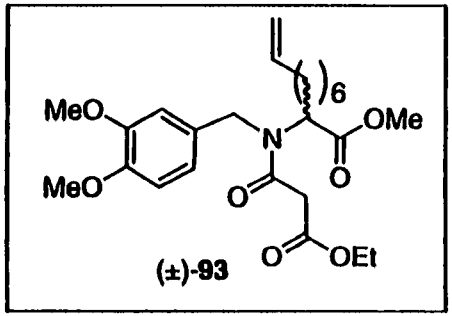


Figure A.2.88 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**93**.

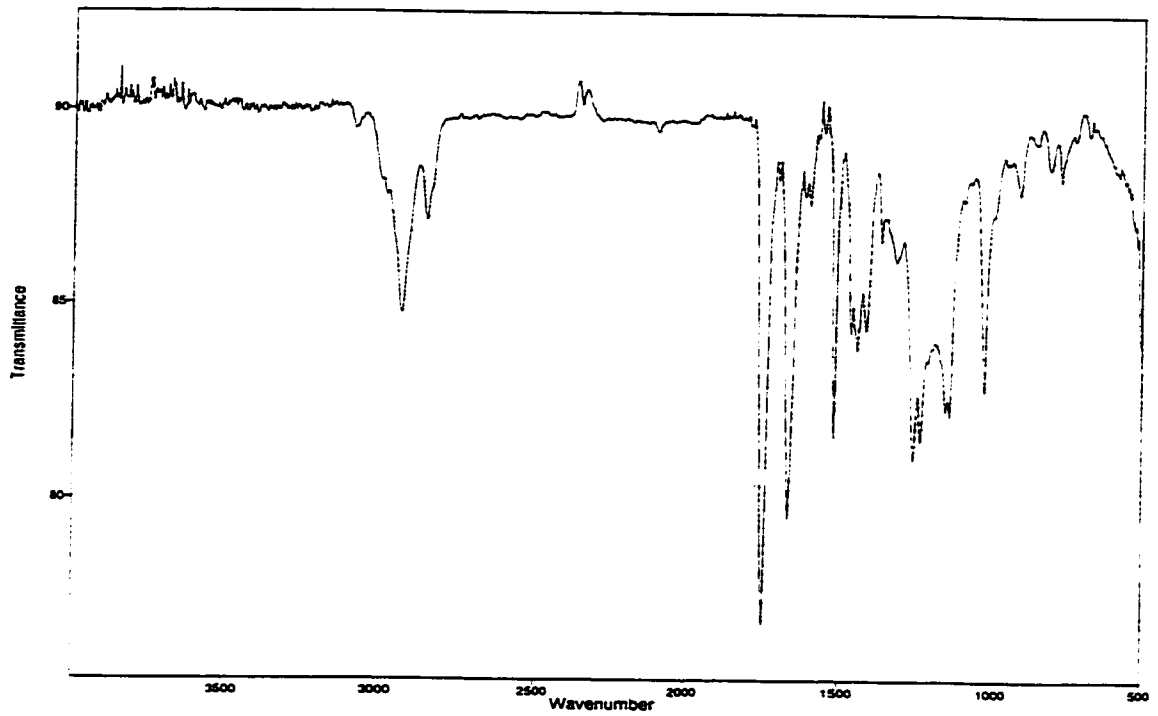


Figure A.2.89 Infrared Spectrum (thin film/NaCl) of compound (±)-**93**.

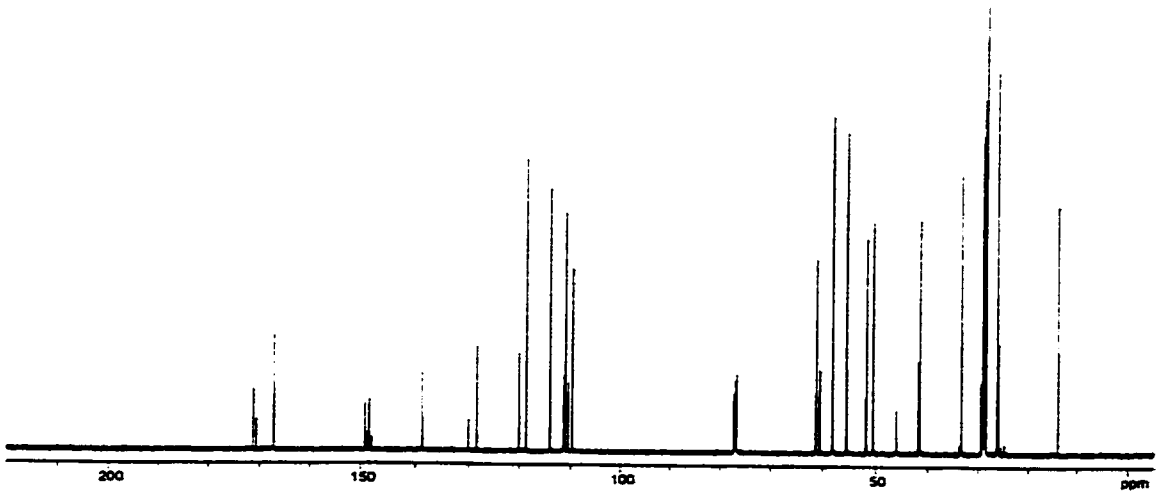


Figure A.2.90 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**93**.

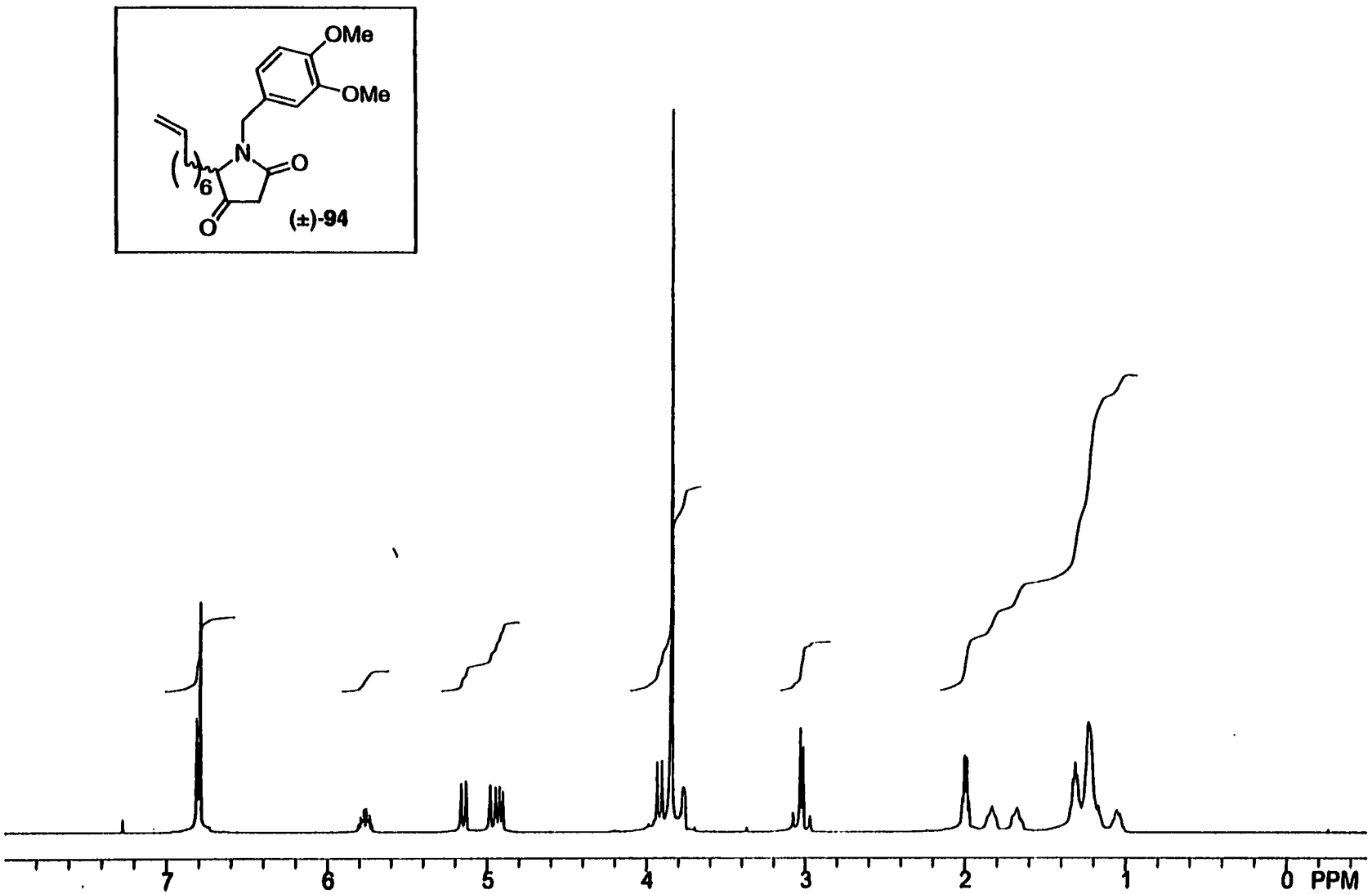


Figure A.2.91 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**94**.

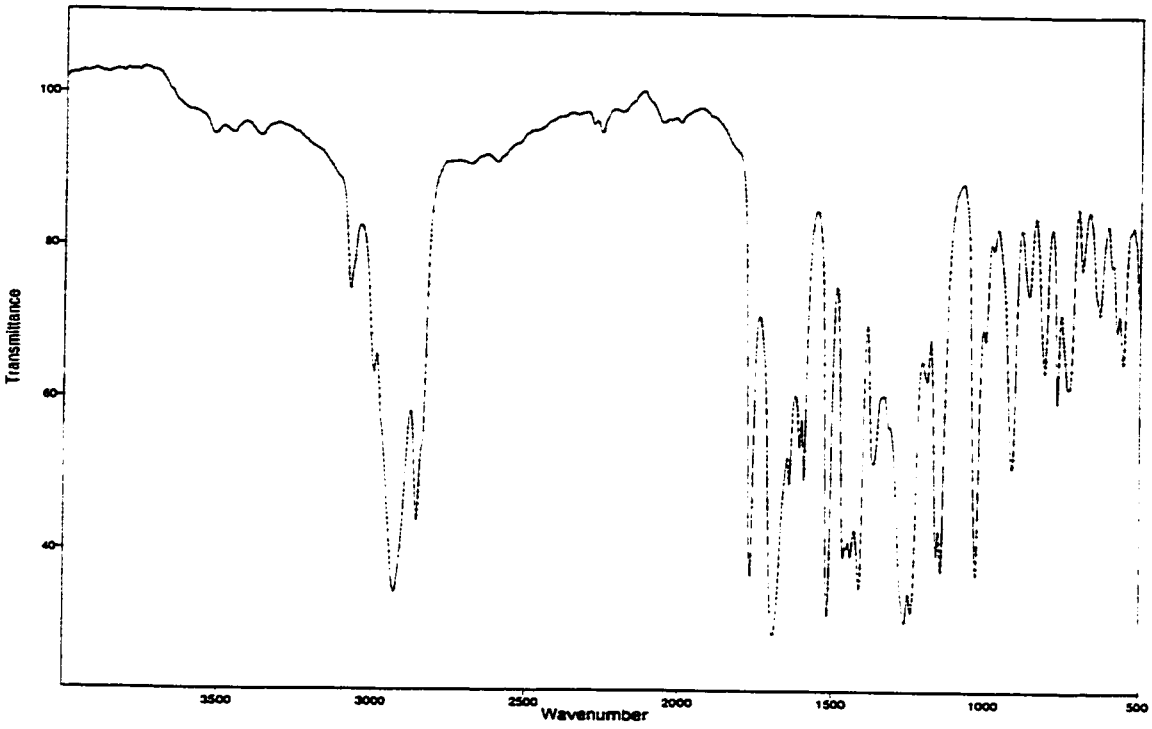


Figure A.2.92 Infrared Spectrum (thin film/NaCl) of compound (±)-**94**.

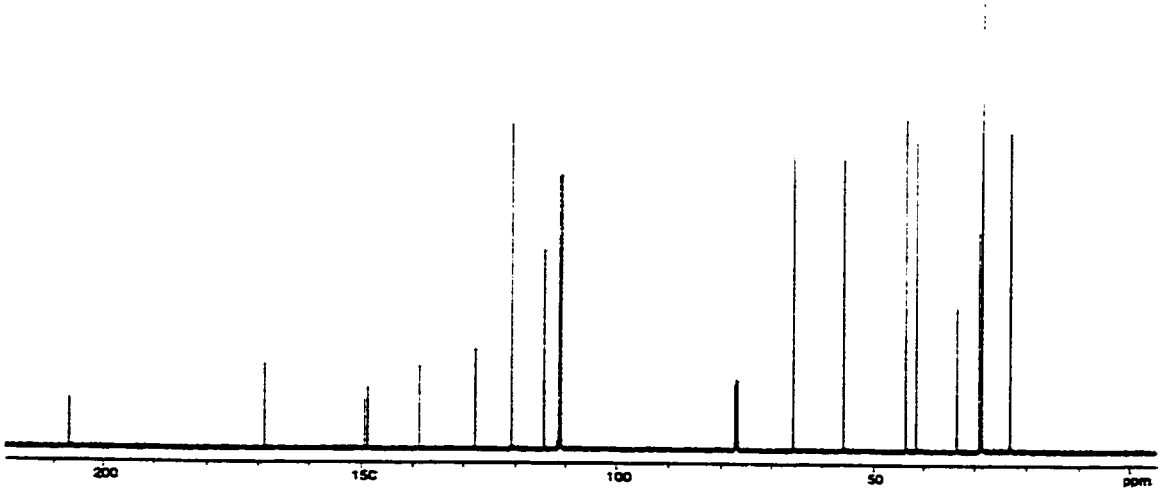


Figure A.2.93 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**94**.

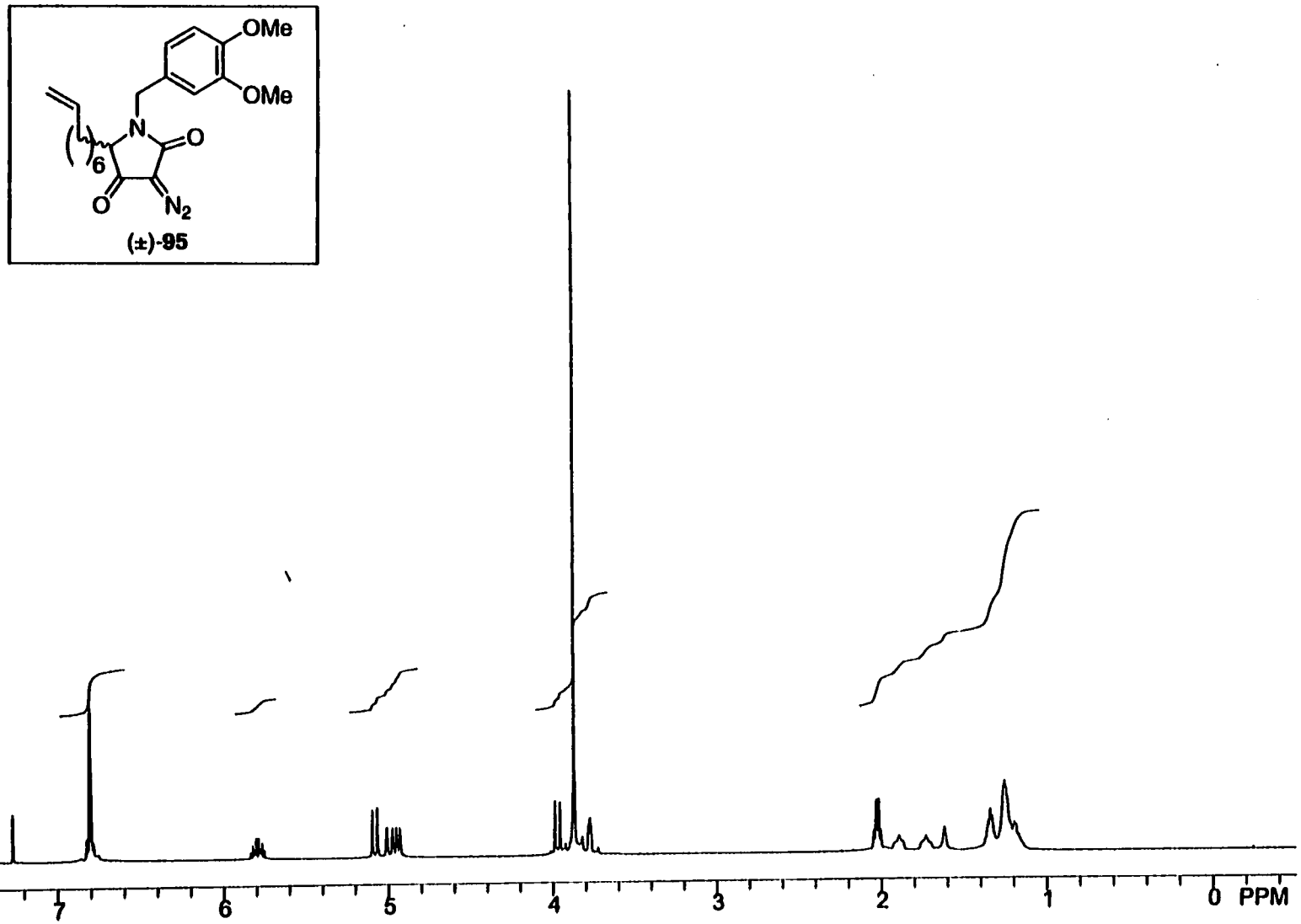


Figure A.2.94 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**95**.

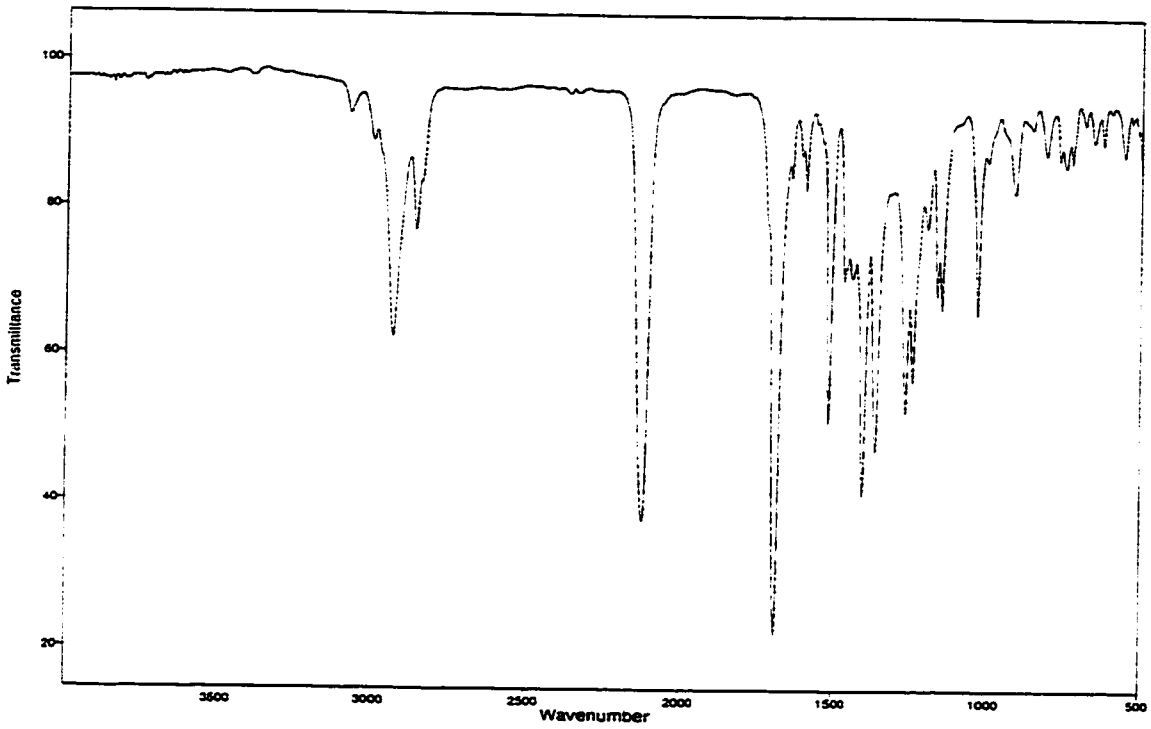


Figure A.2.95 Infrared Spectrum (thin film/NaCl) of compound (±)-**95**.

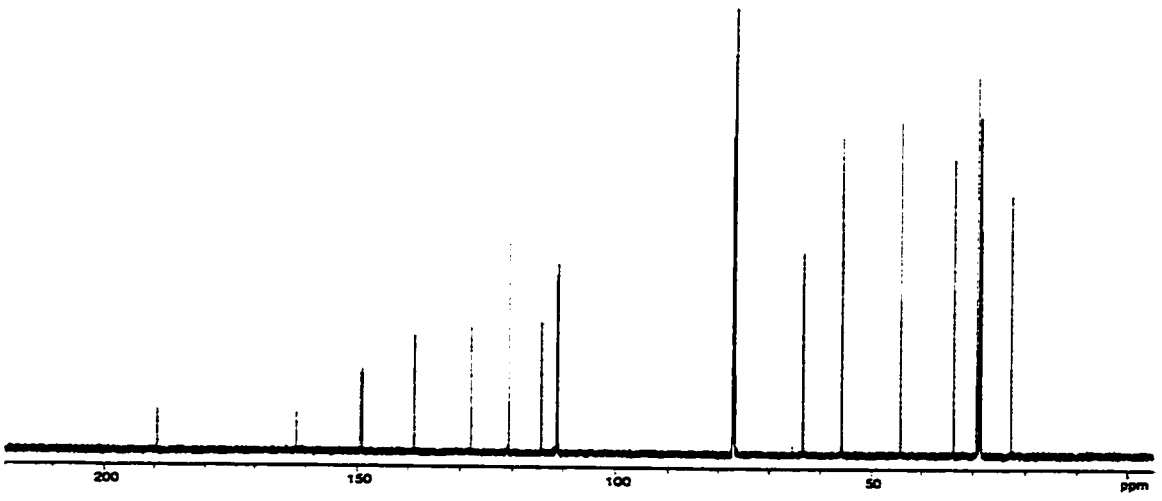


Figure A.2.96 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**95**.

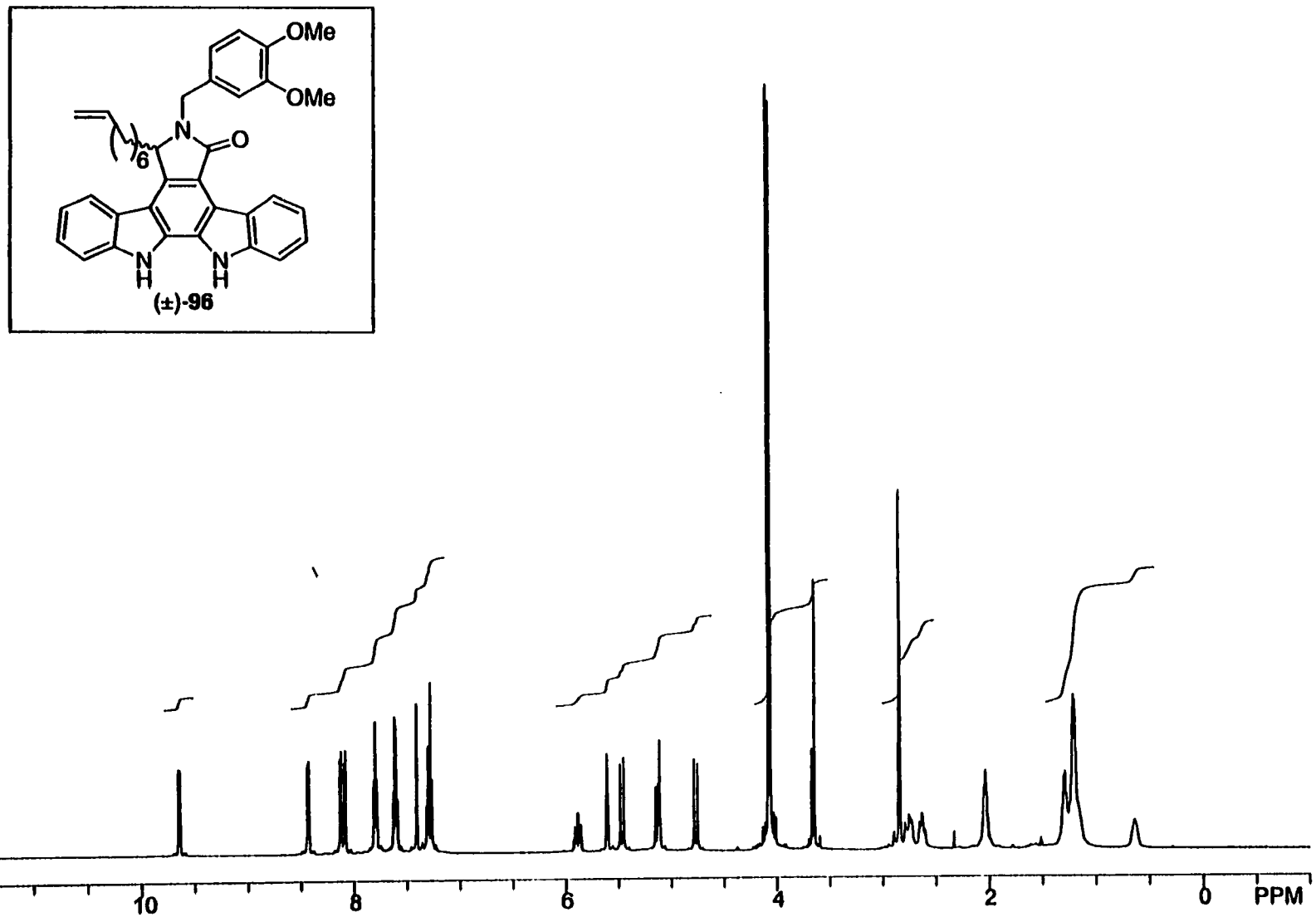


Figure A.2.97 ^{1}H NMR (500 MHz, acetone-d_6) of compound (±)-**96**.

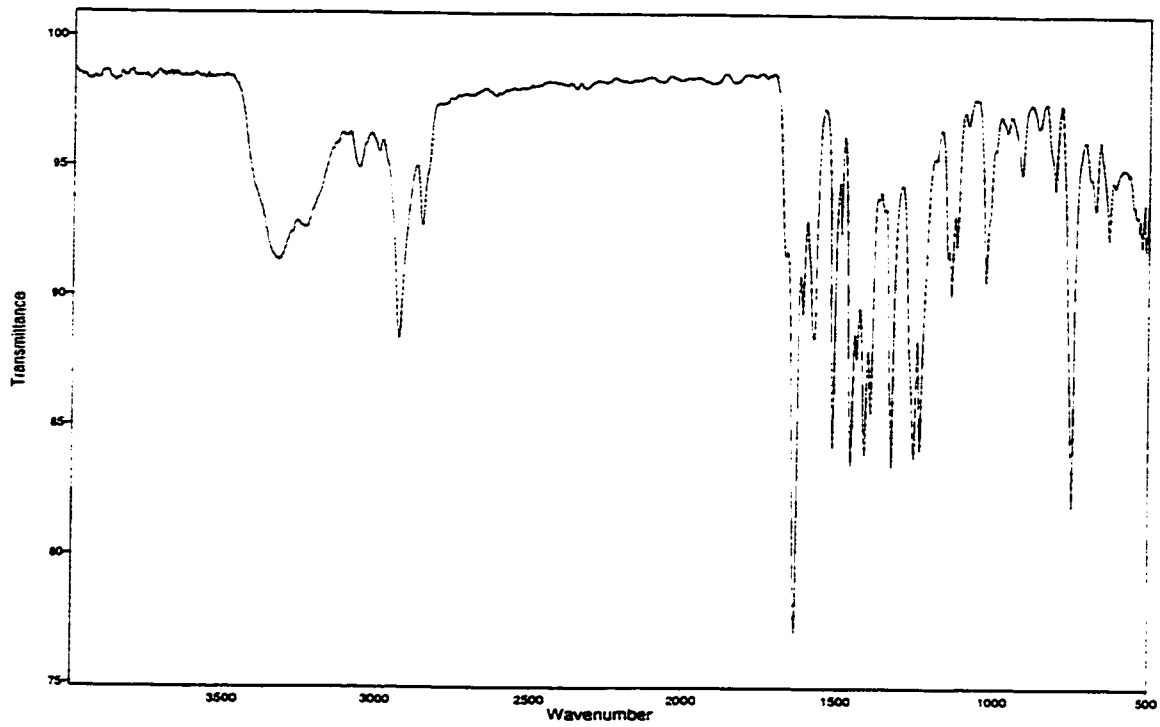


Figure A.2.98 Infrared Spectrum (thin film/NaCl) of compound (±)-**96**.

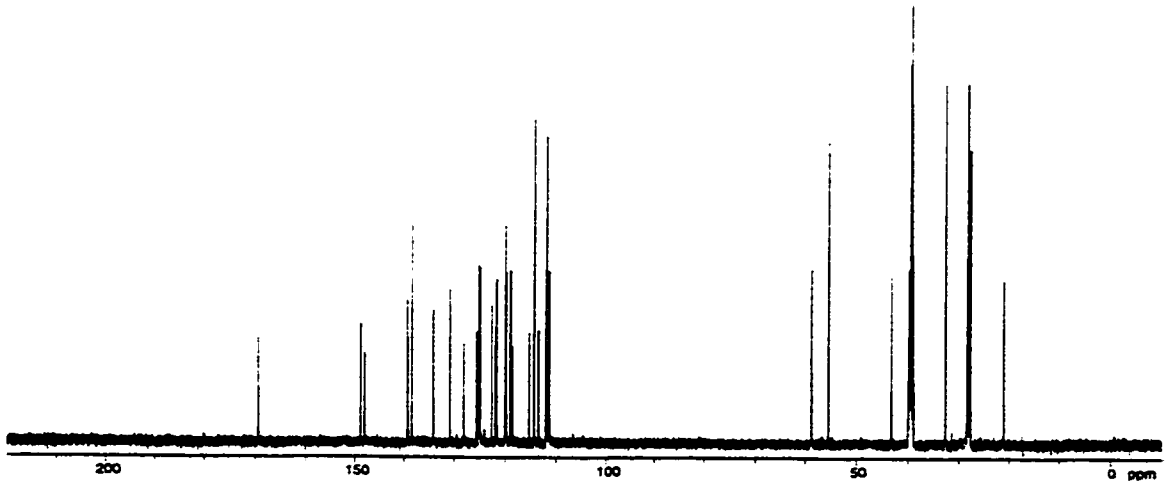
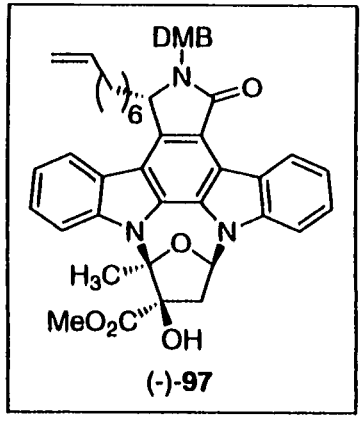


Figure A.2.99 ^{13}C NMR (125 MHz, DMSO-d_6) of compound (±)-**96**.



(-)-**97**

Figure A.2.100 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**97**.

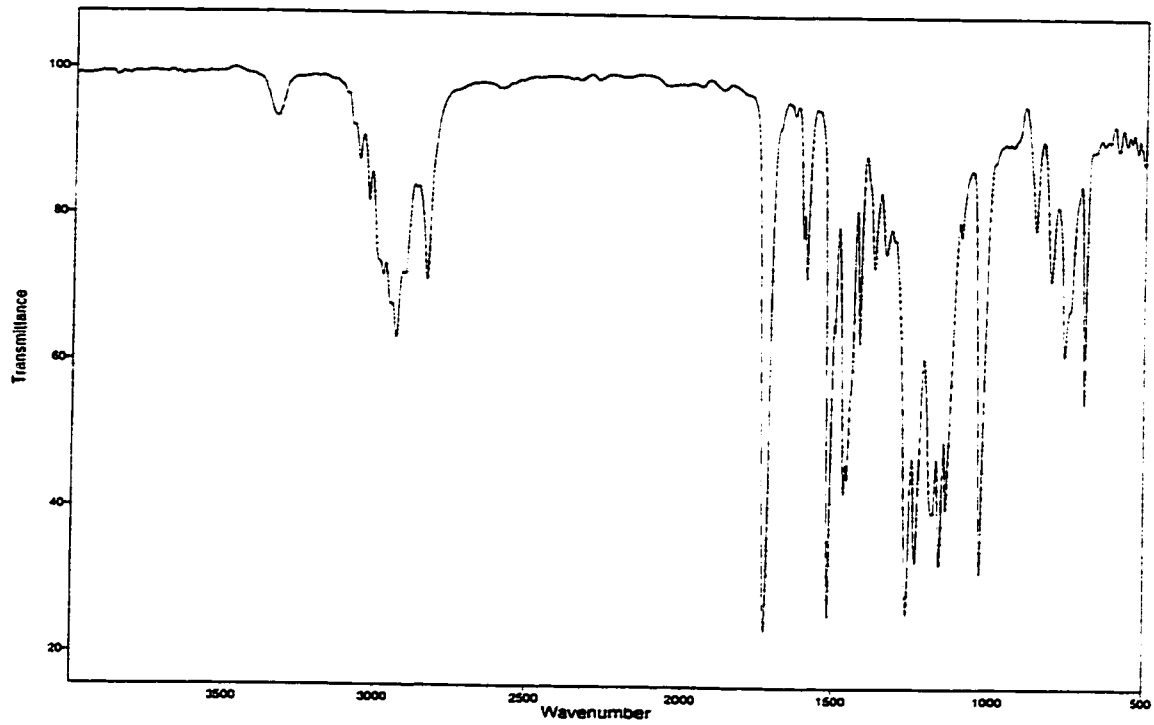


Figure A.2.101 Infrared Spectrum (thin film/NaCl) of compound (-)-**97**.

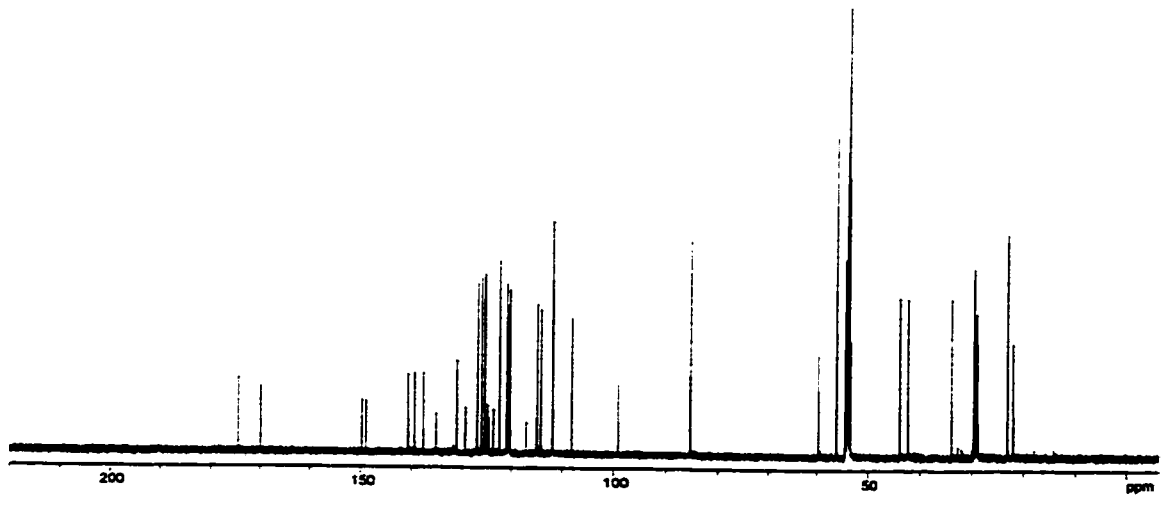
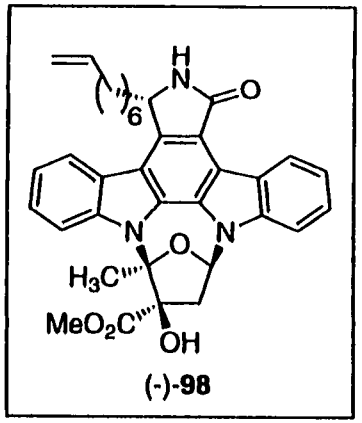


Figure A.2.102 ^{13}C NMR (125 MHz, CD_2Cl_2) of compound (-)-**97**.



(-)-**98**

Figure A.2.103 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**98**.

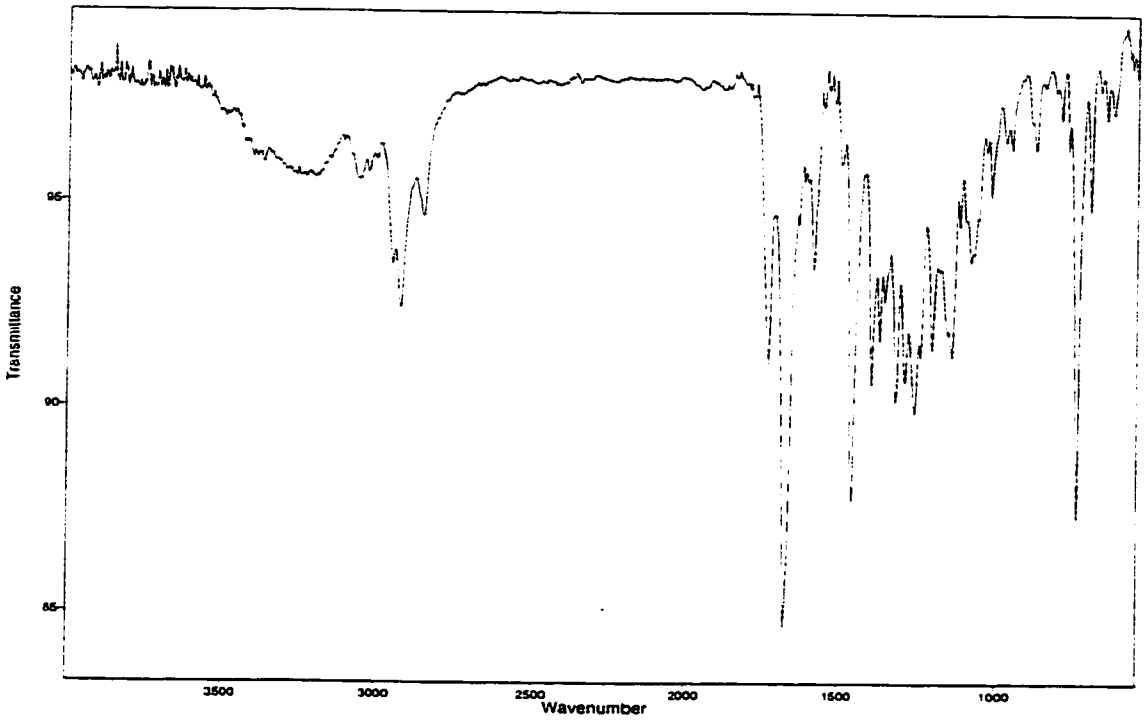


Figure A.2.101 Infrared Spectrum (thin film/NaCl) of compound (-)-**98**.

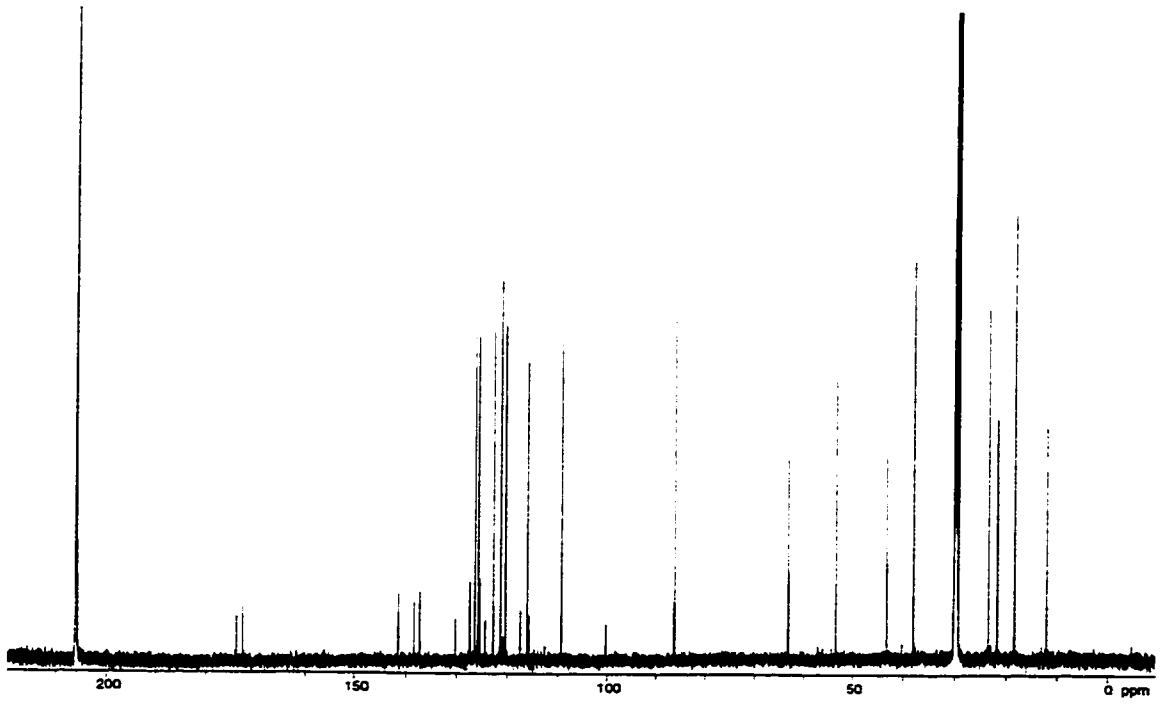


Figure A.2.102 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**98**.

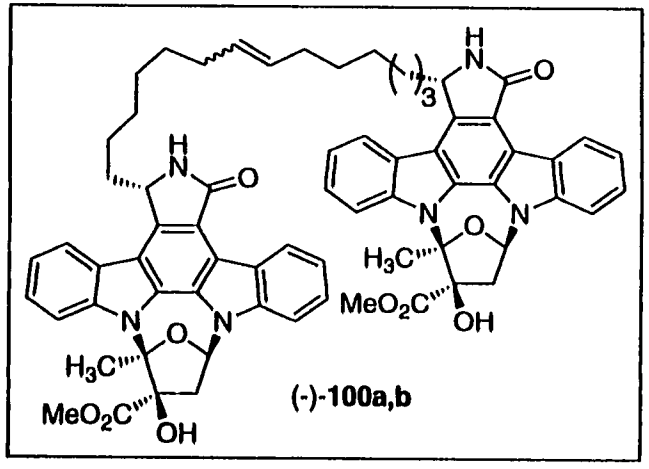


Figure A.2.106 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**100a,b**.

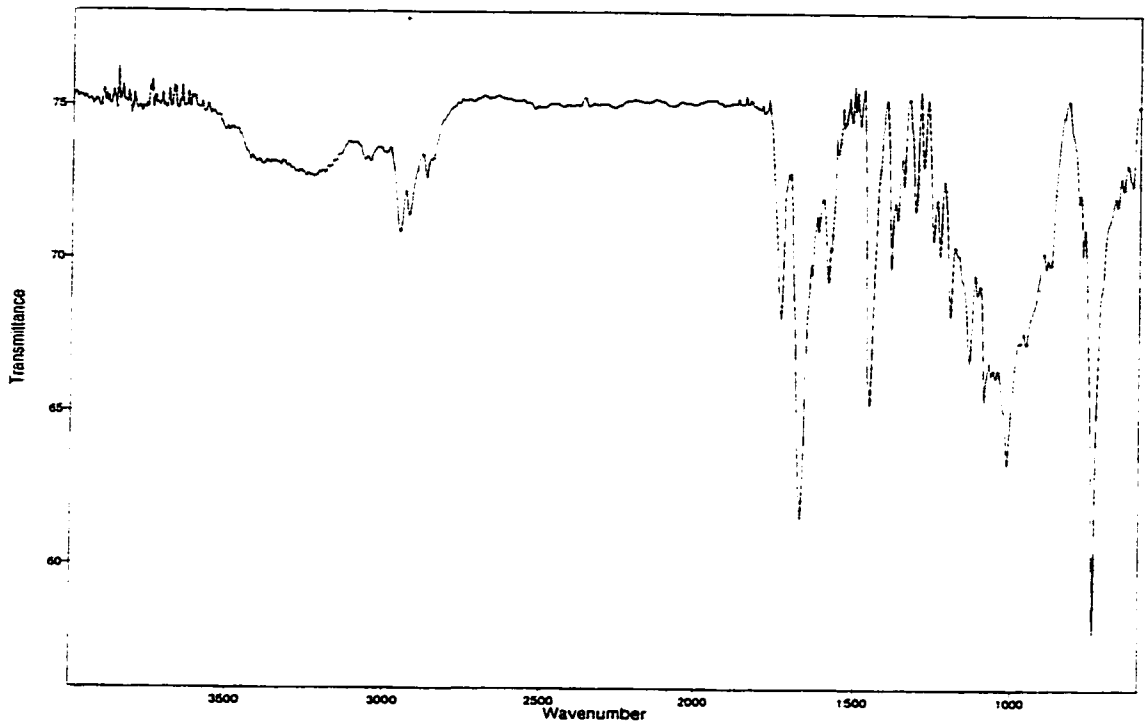


Figure A.2.101 Infrared Spectrum (thin film/NaCl) of compound (-)-**100a,b**.

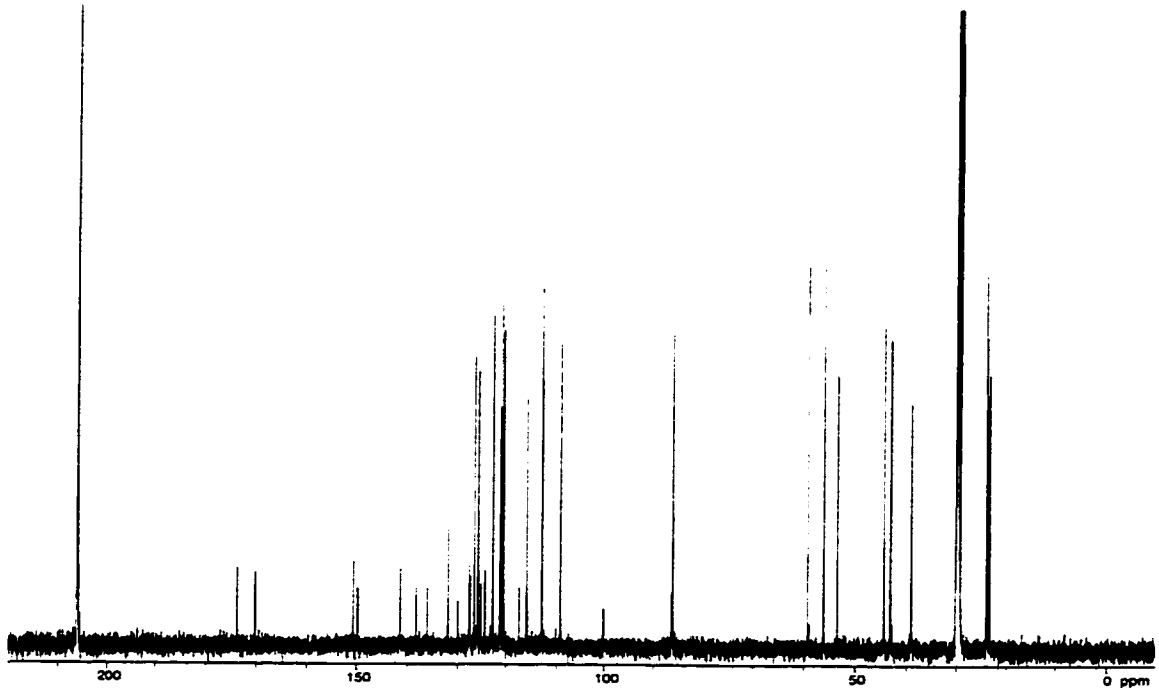


Figure A.2.102 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**100a,b**.

CHAPTER THREE

The Design and Synthesis of C(7) Alkyl Analogs of (+)-K252a and Their Application to Selective Inhibition of Engineered Protein Kinases

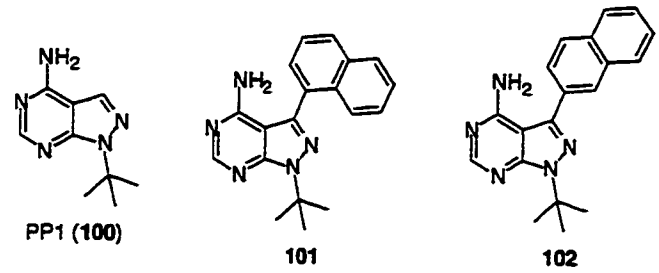
3.1 Background.

3.1.1 Introduction.

Although selective protein kinase inhibitors are highly useful as tools for the study of cellular signal transduction cascades, few have been identified due to the high conservation of kinase catalytic domains. Phosphorylation can be described as a chemical switch that allows the cell to transmit signals from the membrane to the nucleus and ultimately control cell processes in a highly regulated manner. Therefore, highly selective kinase inhibitors would allow for better understanding of the cellular function of individual kinases.[1]

The Shokat group at Princeton University has recently devised an approach combining chemistry and genetics to develop highly specific cell-permeable inhibitors of the oncogenic tyrosine kinase v-Src.[2] More specifically, the Shokat group's strategy employs a point mutation to create a unique pocket in the ATP binding site of the kinase of interest. This empty cavity does not occur in any other protein kinase, therefore a specific inhibitor of the engineered kinase can be synthesized by modifying a known inhibitor with a bulky group designed to fit the novel active site pocket.[3,4]

Figure 3.1.1



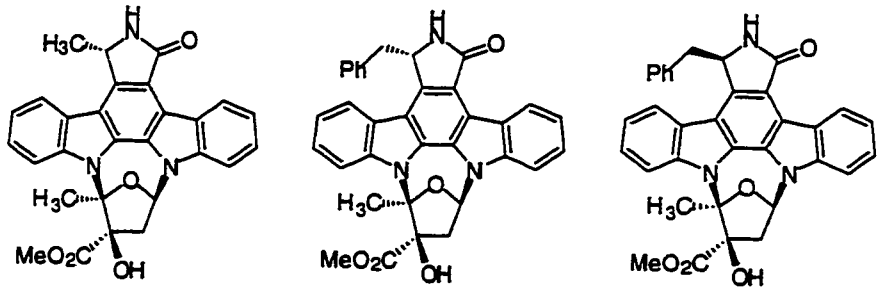
PP1 (**100**) **101** **102**

This selective inhibition was achieved by making a functionally silent active-site mutation in the target kinase enabling a selective inhibitor to distinguish it from other kinases in the cell. It was then possible to design and synthesize a derivative of the known Src-specific inhibitor PP1 (**100**) that could be a tight-binding inhibitor of the engineered kinase and would not inhibit wild-type kinases. These efforts resulted in two highly potent (IC_{50} = 1.5 nM) and uniquely specific inhibitors (**101** and **102**) of a mutated version of v-Src (see Figure 3.1.1).[5,6]

3.1.2 Evaluating K252a Analogs as Inhibitors of Mutant Protein Kinases.

We were intrigued by the idea that known kinase inhibitors modified with bulky alkyl groups could be both potent and selective inhibitors of engineered kinases. Since K252a (**1**) was well precedented as a broad-based kinase inhibitor, we turned to the Shokat group with three of our K252a analogs (-)-**52a**, (-)-**80a**, and (+)-**80b** (see Figure 3.1.2).[7] Preliminary results from assays against wild-type and mutant kinases were very promising.[8] In Table 3.1.1, IC_{50} values are shown for the inhibition of wild type protein kinases v-Src, c-Fyn, c-Abl, CDK2 and CAMKIIa as well as five engineered versions of these kinases. The mutant kinases are designated in the form of an abbreviated code. In the case of the v-Src mutant, I338G indicates mutation of isoleucine residue 338 in the active site to a glycine, while the Fyn mutant contains a glycine rather than a tyrosine at residue 339.

Figure 3.1.2



(-)-7-(*S*)-Methyl K252a (**52a**) (-)-7-(*S*)-Benzyl K252a (**80a**) (+)-7-(*R*)-Benzyl K252a (**80b**)

Methyl derivative (-)-**52a** was a somewhat potent inhibitor (IC_{50} = 0.74 - >11 μM) as we had previously observed,[9] but the alkyl group was not large enough to confer any selectivity for the engineered kinases on this analog. Benzyl derivatives (-)-**80a** and (+)-**80b** on the other hand showed some striking differences. The benzyl group in (+)-**80b** was not oriented correctly to take full advantage of the pocket in the mutant kinase. Therefore, although (+)-**80b** showed modest (<100-fold: **1a** vs **1b** or **2a** vs **2b**; Table 3.1.1) selectivity for the v-Src and c-Fyn mutants, in the other cases no appreciable selectivity was observed. Conversely, derivative (-)-**80a** was very potent and showed very high selectivity (3000 - >16,000-fold: **1a** vs **1b** or **2a** vs **2b**; Table 3.1.1) for the mutant versions of the kinases v-Src and Fyn. In the case of Abl none of the three analogs were potent inhibitors, probably due to the fact that the natural product is itself a poor inhibitor (IC_{50} = 2.2 μM).

Interestingly, both benzyl derivatives (-)-**80a** and (+)-**80b** showed a preference for the wild-type CAMKIIa kinase (**5a** vs **5b**; Table 3.1.1). This could be attributed to a favorable interaction of the C(7) benzyl group with phenylalanine 89 in the kinase. Since F89 is the side chain that has been excised in the mutant version, loss of this interaction might account for the higher IC_{50} values in the case of F89G CAMKIIa.

Table 3.1.1

Kinase	IC_{50} (μM)			
	(+)-**1**	(-)-**52a**	(-)-**80a**	(+)-**80b**
1a v-Src	0.020	2.5	3.6	11
1b I338G v-Src	--------	2.1	0.0012	0.019
2a c-Fyn	0.17	3.8	30	>33
2b T339G Fyn	--------	>3.7	0.0018	0.057
3a c-Abl	2.2	>11	>33	>33
3b T314A c-Abl	--------	>11	>11	>11
4a CDK2	0.20	1.0	1.0	10
4b F80G CDK2	--------	0.54	0.15	10
5a CAMKIIa	0.0054	0.88	0.13	0.49
5b F89G CAMKIIa	--------	0.95	0.39	>1.2

The most suprising result of these assays was that although K252a is known as a broad-based, unselective kinase inhibitor, both (-)-**52a** and (-)-**80a** showed very low selectivity (2 - 6 fold: **4a** vs **4b**; Table 3.1.1) for F80G CDK2. A possible reason for this is that the pocket created by the mutation in the Src family (here Src and Fyn) is effectively larger than that created in the mutation of CDK2 or CAMKIIa. We postulated that the benzyl group was either too large, or else not flexible enough to take advantage of the pocket in other families of protein kinases. To test this hypothesis we prepared a number of additional C(7) K252a analogs with smaller and more flexible alkyl groups.

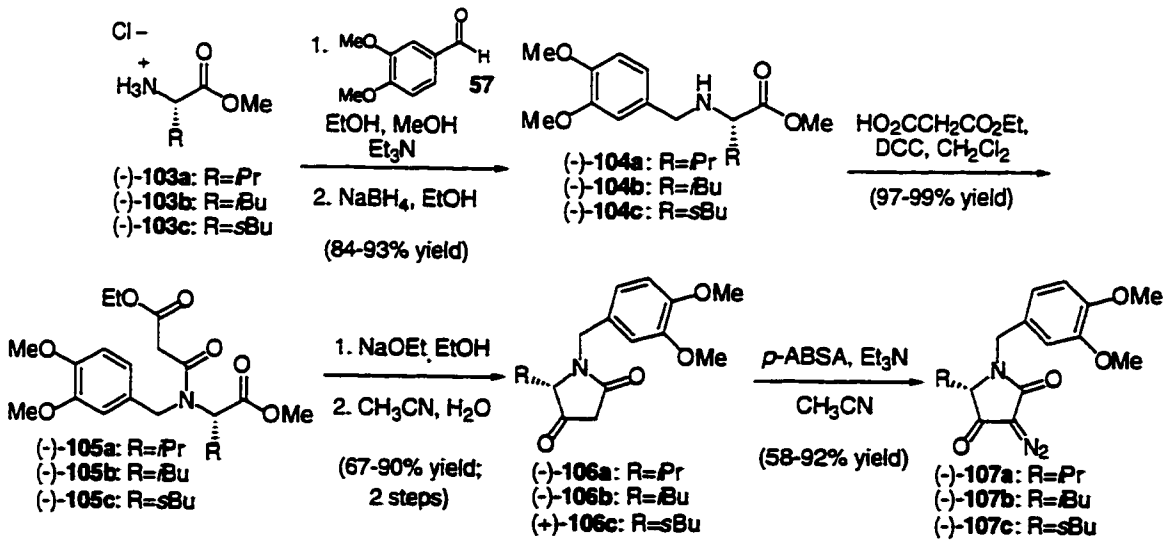
3.2 Synthesis of K252a Analogs Containing Smaller Alkyl Groups.

3.2.1 Preparation of C(7) Isopropyl, Isobutyl and *Sec*-butyl Analogs.

To facilitate preparation, we focused on commercially available amino acids in our new series of derivatives with smaller C(7) groups targeting the remaining natural amino acids possessing an alkyl side chain; valine, leucine and isoleucine.

In a manner analogous to our methyl series amino acid methyl ester hydrochlorides (**103a-c**, Scheme 3.2.1) were monoprotected as the dimethoxybenzyl derivatives giving rise to the protected amines in 84-93% yield. The protected amines (-)-**104a-c** were then coupled to ethyl hydrogen malonate affording esters (-)-**105a-c** in 97-99% yield.

Scheme 3.2.1

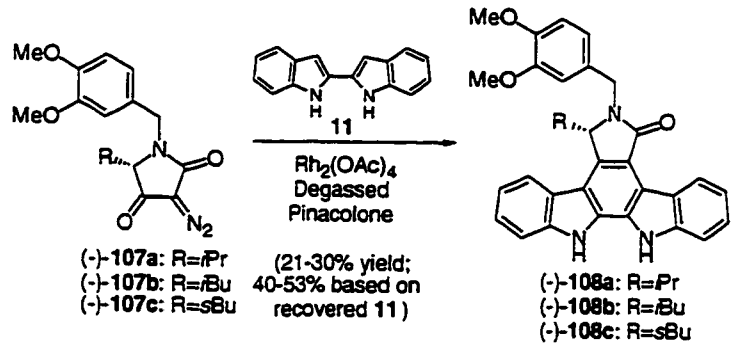


Dieckmann cyclization of (-)-**105a-c** was effected with slightly less than one equivalent of NaOEt in EtOH. After being held at reflux for 5 minutes affording the desired lactams which, without further purification, were subjected to

decarboethoxylation in a refluxing mixture of CH_3CN and H_2O to provide ketolactams (-)-**106a,b** and (+)-**106c** in 67-90% yield over two steps. Diazo transfer from *p*-ABSA gave rise to the desired diazo lactams (-)-**107a-c** in 58-92% yield.

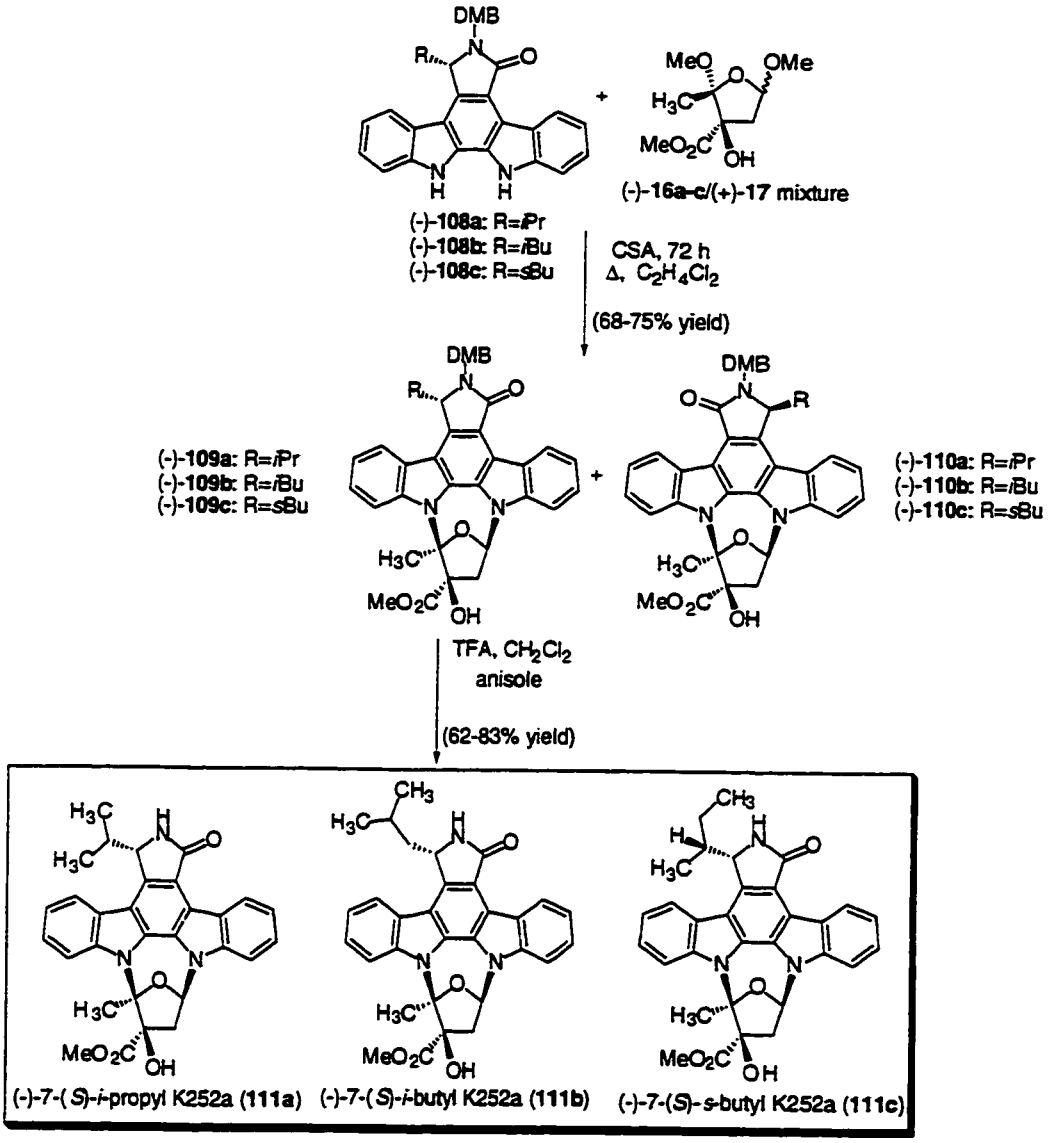
Finally, treating diazo lactams (-)-**107a-c** with 2,2'-biindole (**11**) in the presence of 1 mol % $Rh_2(OAc)_4$ provided aglycons (-)-**108a-c** in 40-53% yield based on recovered **11** (see Scheme 3.2.2).

Scheme 3.2.2



With the aglycons in hand the analog syntheses were completed by subjecting (-)-**108a-c** to cycloglycosidation with the carbohydrate mixture (-)-**16a-c**/(+)-**17** in the presence of a catalytic amount of CSA in refluxing 1,2-dichloroethane which produced the expected 2:1 mixtures of regioisomers (-)-**109a-c** and (-)-**110a-c** in 68-75% yield. Chromatographic separation of the mixtures, followed by deprotection of dimethyoxybenzyl amides (-)-**109a-c** with TFA/CH_2Cl_2 in the presence of anisole afforded the desired analogs (-)-**111a-c** in 62-83% yield (see Scheme 3.2.3).

Scheme 3.2.3

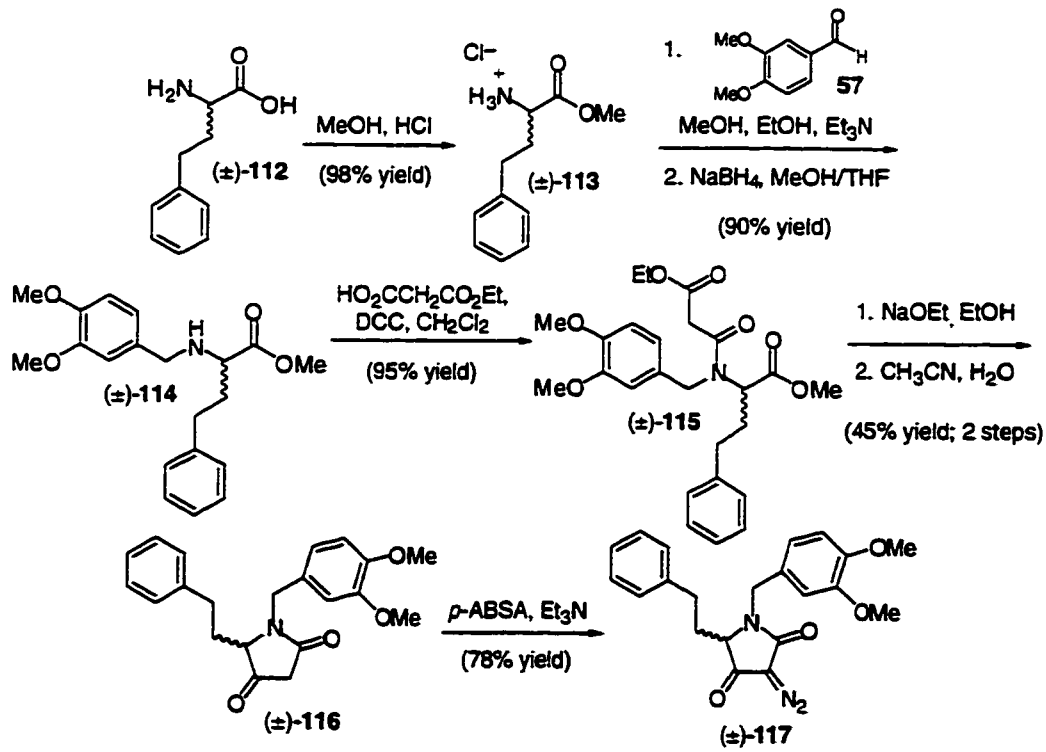

DMB
(-)-108a: R=iPr
(-)-108b: R=iBu
(-)-108c: R=sBu
(-)-16a-c/(+)-17 mixture
CSA, 72 h
Δ, C2H4Cl2
(68-75% yield)
(-)-109a: R=iPr
(-)-109b: R=iBu
(-)-109c: R=sBu
(-)-110a: R=iPr
(-)-110b: R=iBu
(-)-110c: R=sBu
TFA, CH2Cl2
anisole
(62-83% yield)
(-)-7-(S)-i-propyl K252a (111a)
(-)-7-(S)-i-butyl K252a (111b)
(-)-7-(S)-s-butyl K252a (111c)

3.3 Synthesis of a K252a Analog With a More Flexible Alkyl Group.

3.3.1 Synthesis of a Homophenylalanine-Derived K252a Analog.

To address the issue of a C(7) alkyl group which would retain the size of the benzyl, yet have more flexibility, we turned to an unnatural amino acid with one additional methylene unit in the amino acid side chain. We began the synthesis by esterifying commercially available DL-(±)-homophenylalanine (**112**) under Fisher conditions which afforded the amino acid ester hydrochloride (±)-**113** in excellent yield. Amine (±)-**113** could be protected as dimethoxybenzyl derivative (±)-**114** in 90% yield (see Scheme 3.3.1).

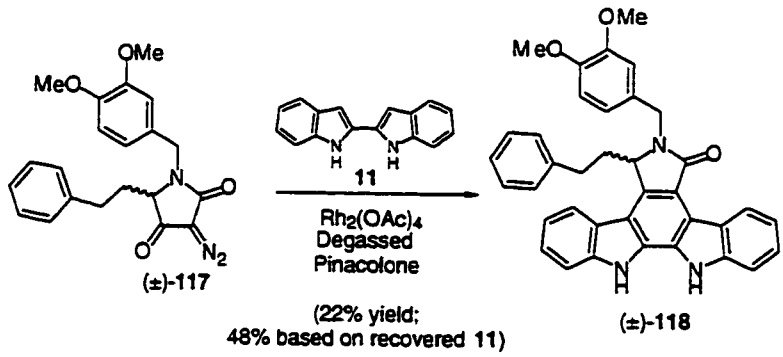
Scheme 3.3.1



Protected amine (±)-**114** was then coupled to ethyl hydrogen malonate affording the Dieckmann cyclization precursor (±)-**115** in 95% yield. Ester (±)-**115** was warmed to reflux in an ethanolic solution of NaOEt and then decarboethoxylated in a refluxing mixture of CH_3CN and H_2O to provide ketolactam (±)-**116** in 45% yield over two steps. Finally, diazo transfer from *p*-ABSA furnished diazo lactam (±)-**117** in 78% yield.

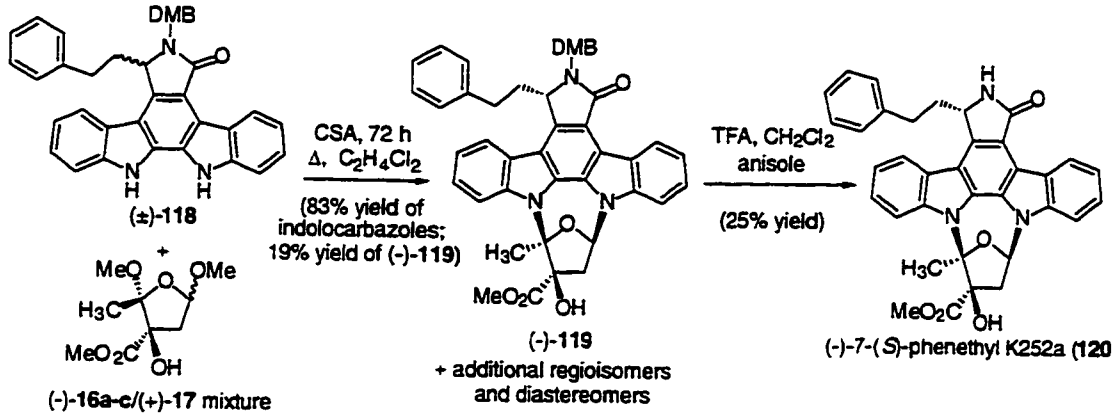
Aglycon (±)-**118** was produced by coupling equimolar amounts of diazo lactam (±)-**117** with 2,2'-biindole (**11**) in the presence of 1 mol % $Rh_2(OAc)_4$ providing (±)-**118** (48% yield based on recovered **11**; see Scheme 3.3.2).

Scheme 3.3.2



In accord with our previous approach to benzyl K252a analogs, aglycon (±)-**118** was subjected to cycloglycosidation with the carbohydrate mixture (-)-**16a-c**/(+)-**17** in the presence of a catalytic amount of CSA in refluxing 1,2-dichloroethane to give an expected 2:2:1:1 mixture of regioisomers and diastereomers. Chromatographic separation of the mixture, followed by deprotection of amide (-)-**119** with TFA/CH_2Cl_2 in the presence of anisole afforded the desired analog (-)-**120** in 25% yield (see Scheme 3.3.3).[10]

Scheme 3.3.3


DMB
(±)-118
MeO
OMe
H3C
MeO2C
OH
(-)-16a-c/(+)-17 mixture
CSA, 72 h
Δ, C2H4Cl2
(83% yield of indolocarbazoles; 19% yield of (-)-119)
DMB
H3C
MeO2C
OH
(-)-119
+ additional regioisomers and diastereomers
TFA, CH2Cl2
anisole
(25% yield)
H
H3C
MeO2C
OH
(-)-7-(S)-phenethyl K252a (120)

3.4 Inhibition of Engineered Protein Kinases Using the Second Generation Inhibitors.

3.4.1 Evaluation of Biological Activity.

With the four additional C(7) derivatives in hand, we again turned to the Shokat group for inhibition assays against their engineered kinases. Once again the derivatives showed striking selectivity for I338G v-Src and T339G Fyn. Although these analogs were still poor inhibitors of c-Abl and T314A c-Abl, some improvements were observed in the cases of CDK2 and CAMKIIa mutants (see Table 3.4.1).

Table 3.4.1

Kinase	IC_{50} (μM)				
	(+)-**1**	(-)-**111a**	(-)-**111b**	(-)-**111c**	(-)-**120**
1c v-Src	0.020	>33	>33	26	12
1d I338G v-Src	--------	0.044	0.00023	0.0092	0.0021
2c c-Fyn	0.17	>33	>33	>33	>33
2d T339G Fyn	--------	0.12	0.00055	0.030	0.0043
3c c-Abl	2.2	>33	>33	>33	>33
3d T314A c-Abl	--------	>11	3.1	>11	9.6
4c CDK2	0.20	29	22	5.8	3.7
4d F80G CDK2	--------	1.9	0.070	>3.7	0.037
5c CAMKIIa	0.0054	2.7	1.7	0.82	0.54
5d F89G CAMKIIa	--------	2.3	0.95	>3.7	0.0040

Isobutyl derivative (-)-**111b** showed 314-fold selectivity (**4c** vs **4d**; Table 3.4.1) for F80G CDK2 confirming our hypothesis that a smaller alkyl group might improve the selectivity for a smaller binding pocket in CDK and CAMKIIa mutants. In addition, phenethyl derivative (-)-**120** showed selectivity for F89G CAMKIIa (135-fold: **5c** vs **5d**; Table 3.4.1), unlike its benzyl congener, demonstrating that a more flexible side chain did lead to increased selectivity.

Importantly, improvements were made in the inhibition of the Src family kinases as well. Isobutyl derivative (-)-**111b** was an extremely potent inhibitor of both I338G v-Src and T339G Fyn (IC_{50} = 230 - 550 pM) and to our knowledge is the most potent inhibitor of Src family mutants which has been reported to date. Selectivity was also increased with (-)-**111b** to 60,000 - >140,000-fold (**1c** vs **1d** or **2c** vs **2d**; Table 3.4.1) for the engineered Src family kinases.

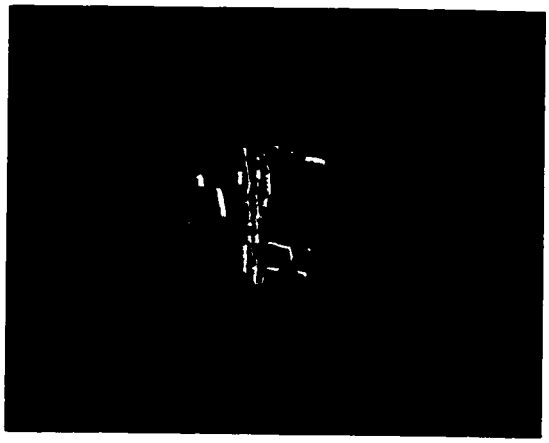
Isopropyl and *sec*-butyl analogs (-)-**111a** and (-)-**111c** were much less selective (275 - 2800-fold: **1c** vs **1d** or **2c** vs **2d**; Table 3.4.1) for I338G v-Src and T339G c-Fyn than the benzyl derivatives (-)-**80a** and (+)-**80b**, but showed improvement over methyl analog (-)-**52a**. Homophenylalanine-derived inhibitor (-)-**120** was also somewhat less selective (5700 - 7600-fold: **1c** vs **1d** or **2c** vs **2d**; Table 3.4.1) than (-)-**80a** or (-)-**111b**, probably due to its slightly larger size.

3.4.2 Structural Rationale.

The protein kinases studied were engineered by mutation of a key residue in the binding site, thereby creating a unique binding pocket. These kinases were inhibited by C(7) alkyl derivatives of K252a with various degrees of selectivity and potency. In order to achieve both selectivity and potency, a very close match between the size of the binding pocket and size of the alkyl group was neccessary. It was found that the size of the pocket created by the mutation varied among

kinases making it difficult to identify a single compound that would be a broad-based inhibitor of similarly engineered kinases from widely diverse families. We again turned to crystal structure data for staurosporine (**6**) bound to CDK2 as a model for K252a analog binding. The view of the C(7) position is again shown in Figure 3.4.1 with the bulky isopropyl and benzyl groups creating a confined steric environment around C(7).

Figure 3.4.1



In Figure 3.4.2, the benzyl side chain of phenylalanine 80 has been excised to create a model for the engineered kinase F80G CDK2. It is clear that there is now additional space in the binding site that could accomodate an alkyl group.

Figure 3.4.2

3.5 Conclusion.

The total syntheses of four new K252a analogs were achieved. Isopropyl, isobutyl, *sec*-butyl analogs were prepared in order to ascertain the effect of different sized alkyl groups on binding and selectivity for engineered protein tyrosine kinases (I338G v-Src, T339G c-Fyn, T314A c-Abl, F80G CDK2 and F89G CAMKIIa). Isobutyl derivative (-)-**111b** showed extremely potent inhibition of Src and Fyn mutants (IC_{50} = 230 to 550 pM) and also showed increased selectivity as high as >140,000-fold. In addition, modest selectivities were observed for F80G CDK2 and F89G CAMKIIa by inhibitors (-)-**111b** and (-)-**120** respectively.

3.6 Experimental Section.

3.6.1 Material and Methods.

Unless stated otherwise, reactions were performed in flame dried glassware under a nitrogen atmosphere, using freshly distilled solvents. Diethyl ether (Et_2O) and tetrahydrofuran (THF) were distilled from sodium/benzophenone ketyl. Methylene chloride (CH_2Cl_2), benzene, and triethylamine (Et_3N) were distilled from calcium hydride. Boron trifluoride etherate ($BF_3 \bullet OEt_2$), 1,2-dichloroethane and pinacolone were purchased from the Aldrich Chemical Co. in Sure/Seal™ containers and used without further purification. All other commercially obtained reagents were used as received.

Unless stated otherwise all reactions were magnetically stirred and monitored by thin-layer chromatography (TLC) using E. Merck silica gel 60 F254 pre-coated plates (0.25 mm). Preparative TLC was also performed using E. Merck silica gel 60 F254 pre-coated plates (0.25 mm). Column or flash chromatography (silica) was performed with the indicated solvents using silica gel (particle size 0.032-0.063 mm) purchased from Fisher Scientific. In general, the chromatography guidelines reported by Still were followed.[11]

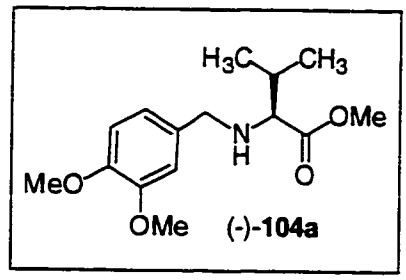
All melting points were obtained on a Haacke-Buchler variable temperature melting point apparatus (model: MFB 595 8020) and are uncorrected. Infrared spectra were recorded on a Midac M-1200 FTIR. ^1H and ^{13}C NMR spectra were recorded on Bruker AM-500 or a Varian 800 spectrometer. Chemical shifts are reported relative to solvent residuals: chloroform (^1H, δ 7.27 ppm, ^{13}C, δ 77.0 ppm), acetone (^1H, δ 2.04 ppm, ^{13}C, δ 206.0 ppm). High resolution mass spectral analyses were performed at The University of Illinois Mass Spectrometry Center. High performance liquid chromatography (HPLC) was performed on a

Waters model 510 system using a Rainin Microsorb 80-199-C5 column, or a Rainin Dynamax SD-200 system with a Rainin Microsorb 80-120-C5 column. Optical rotations were measured on a Perkin-Elmer 241 polarimeter.

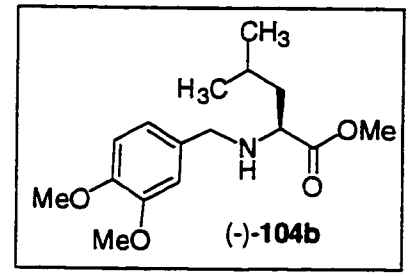
In some compounds containing a tertiary amine, spectra indicate a mixture of rotamers. Although these rotamers could be observed to coalesce at high temperatures, characterization spectra were obtained at room temperature.

3.6.2 Preparative Procedures.

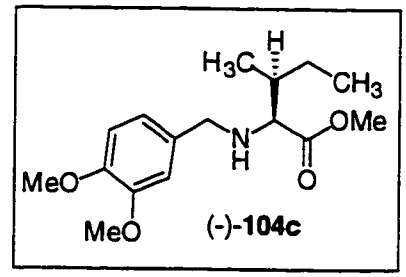
Preparation of Amines (-)-104a-c.



(-)-**104a**



(-)-**104b**



(-)-**104c**

Representative Procedure: Amine (-)-104a. To a solution of L-(-)-valine methyl ester hydrochloride (**103a**; 5.0 g, 29.8 mmol, 1.0 equiv), in MeOH (11 mL) was added Et_3N (4.2 mL, 29.8 mmol, 1.0 equiv) followed by addition of 3,4-dimethoxybenzaldehyde (3.5 g, 20.9 mmol, 0.7 equiv) as a solution in EtOH (53 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in EtOH (70 mL) and $NaBH_4$ (1.1 g, 29.8 mmol, 1.0 equiv) was added portionwise over a 15 minute period. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, the EtOH was removed under reduced pressure and the residue was dissolved in 1 N HCl (50 mL). The acid solution was washed with EtOAc (50 mL) and then brought to pH 10 using 10 N aqueous NaOH (5 mL). The basic solution was extracted with CH_2Cl_2 (3 x 30 mL) and the combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield protected amine (-)-**104a** (5.5 g, 93%

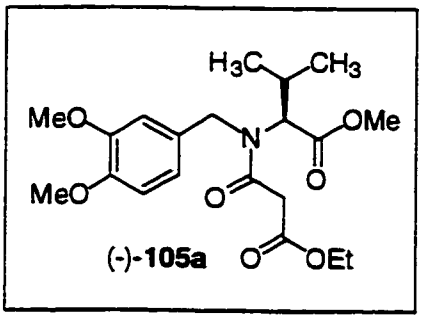
yield) as clear, colorless oil: $[\alpha]_D^{20}$ -44.32° (*c* 0.37, MeOH); IR (thin film/NaCl) 2957 (br m), 2872 (w), 2834 (w), 1731 (s), 1665 (w), 1515 (s), 1464 (m), 1262 (s), 1235 (s), 1195 (m), 1153 (m), 1020 (m), 764 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.94-6.80 (comp m, 3H), 3.89 (s, 3H), 3.88 (s, 3H), 3.80 (d, *J* = 13.0 Hz, 1H), 3.73 (s, 3H), 3.53 (d, *J* = 13.0 Hz, 1H), 3.01 (d, *J* = 6.1 Hz, 1H), 1.92 (tq, *J* = 6.7, 13.4 Hz, 1H), 1.70 (br s, 1H), 0.96 (d, *J* = 6.8 Hz, 3H), 0.94 (d, *J* = 6.8 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 175.8, 148.9, 148.1, 132.7, 120.3, 111.5, 111.0, 66.3, 55.9, 55.8, 52.2, 51.4, 31.7, 19.3, 18.6; high resolution mass spectrum (EI) *m/z* 281.1622 [calc'd for $C_{15}H_{23}NO_4$ (M+) 281.1627].

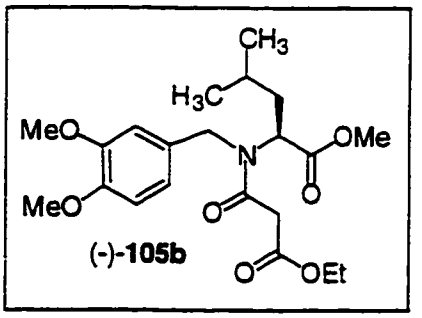
(-)-**104b**: The above procedure was followed using L-(-)-leucine methyl ester hydrochloride (**103b**; 10.0 g, 55.0 mmol), Et_3N (7.7 mL, 55.0 mmol), 3,4-dimethoxybenzaldehyde (6.4 g, 38.5 mmol) and $NaBH_4$ (2.1 g, 55.0 mmol) to afford amine (-)-**104b** (9.5 g, 84% yield) as a clear, colorless oil: $[\alpha]_D^{20}$ -37.00° (*c* 0.30, MeOH); IR (thin film/NaCl) 3333 (br w), 2953 (m), 2870 (w), 2839 (w), 1734 (s), 1592 (w), 1514 (s), 1461 (m), 1264 (s), 1235 (s), 1195 (m), 1154 (s), 1029 (s), 764 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.92-6.80 (comp m, 3H), 3.89 (s, 3H), 3.88 (s, 3H), 3.77 (d, *J* = 12.9 Hz, 1H), 3.73 (s, 3H), 3.56 (d, *J* = 12.8 Hz, 1H), 3.30 (t, *J* = 7.3 Hz, 1H), 1.78 (tt, *J* = 6.7, 13.5 Hz, 1H), 1.67 (br s, 1H), 1.48 (m, 2H), 0.93 (d, *J* = 6.7 Hz, 3H), 0.86 (d, *J* = 6.6 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 176.5, 149.0, 148.2, 132.6, 120.4, 111.6, 111.1, 59.1, 56.0, 55.8, 51.9, 51.5, 42.9, 24.9, 22.8, 22.2; high resolution mass spectrum (EI) *m/z* 295.1779 [calc'd for $C_{16}H_{25}NO_4$ (M+) 295.1784].

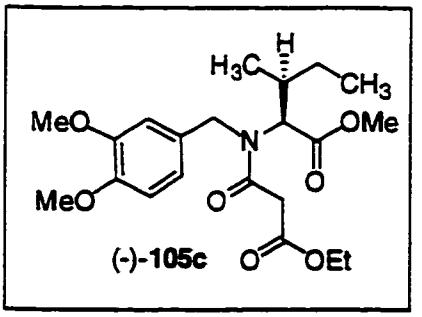
(-)-**104c**: The above procedure was followed using L-(-)-isoleucine methyl ester hydrochloride (**103c**; 5.0 g, 27.5 mmol), Et_3N (3.8 mL, 27.5 mmol), 3,4-dimethoxybenzaldehyde (3.2 g, 19.3 mmol) and $NaBH_4$ (1.0 g, 27.5 mmol) to

afford amine (-)-**104c** (4.94 g, 87% yield) as a clear, colorless oil: $[\alpha]_D^{20}$ -39.76° (*c* 0.42, MeOH); IR (thin film/NaCl) 3345 (w), 2961 (m), 2835 (w), 1732 (s), 1592 (w), 1515 (s), 1463 (m), 1418 (w), 1264 (m), 1236 (m), 1156 (m), 1140 (m), 1030 (m), 764 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.93-6.80 (comp m, 3H), 3.89 (s, 3H), 3.87 (s, 3H), 3.78 (d, *J* = 13.0 Hz, 1H), 3.73 (s, 3H), 3.53 (d, *J* = 13.0 Hz, 1H), 3.10 (d, *J* = 6.2 Hz, 1H), 1.74 (br s, 1H), 1.69 (m, 1H), 1.58 (dqd, *J* = 4.4, 7.5, 14.2 Hz, 1H), 1.20 (m, 1H), 0.90 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 175.7, 148.9, 148.1, 132.6, 120.4, 111.5, 111.0, 65.2, 55.9, 55.8, 52.2, 51.3, 38.3, 25.5, 15.7, 11.4; high resolution mass spectrum (EI) *m/z* 295.1783 [calc'd for $C_{16}H_{25}NO_4$ (M+) 295.1784].

Preparation of Amides (-)-105a-c.







Representative Procedure: Amide (-)-105a: A three-necked flask equipped with an addition funnel was charged with the amine (-)-**104a** (5.5 g, 19.5

mmol, 1.0 equiv), ethyl hydrogen malonate (2.6 g, 19.5 mmol, 1.0 equiv), and CH_2Cl_2 (60 mL). The flask was cooled to 0°C and a 1 M solution of 1,3-dicyclohexylcarbodiimide (19.5 mL, 19.5 mmol, 1.0 equiv) in CH_2Cl_2 (40 mL) was added dropwise *via* addition funnel over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h at room temperature. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (100 mL). The organic layer was separated, and the aqueous layer was extracted with CH_2Cl_2 (3 x 100 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to yield amide (-)-**105a** (7.5 g, 97% yield) as a clear, light yellow oil: $[\alpha]_D^{20}$ -37.84° (*c* 0.37, MeOH); IR (thin film/NaCl) 2962 (br w), 1739 (s), 1655 (s), 1517 (m), 1465 (w), 1416 (w), 1259 (s), 1238 (m), 1141 (s), 1027 (s), 768 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.91-6.69 (comp m 3), 4.86 (d, *J* = 10.4 Hz, 0.77H), 4.62 (m, 2H), 4.34 (d, *J* = 15.2 Hz, 0.23H), 4.24 (q, *J* = 7.1 Hz, 0.46H), 4.18 (q, *J* = 7.1 Hz, 1.54H), 3.87 (s, 4.62H), 3.85 (s, 1.38H), 3.50 (s, 2.31H), 3.42 (s, 0.69H), 3.46-3.34 (comp m, 2H), 2.36 (m, 0.77H), 2.21 (m, 0.23H), 1.31 (t, J=7.1 Hz, 0.69H), 1.26 (t, J=7.1 Hz, 2.31H), 1.00 (d, *J* = 6.5 Hz, 2.31H), 0.98 (d, *J* = 6.9 Hz, 2.31H), 0.95 (d, *J* = 6.9 Hz, 0.69H), 0.82 (d, *J* = 6.7 Hz, 0.69H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.6, 167.6, 167.3, 149.3, 148.4, 128.8, 120.3, 117.9, 111.7, 111.3, 110.7, 109.3, 66.6, 62.3, 61.3, 55.9, 55.8, 52.1, 51.8, 51.6, 48.7, 45.9, 42.0, 41.5, 28.1, 27.6, 19.8, 18.6, 18.6, 14.0; high resolution mass spectrum (EI) *m/z* 395.1953 [calc'd for $C_{20}H_{29}NO_7$ (M+) 395.1944].

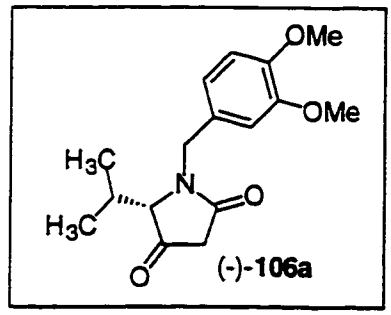
(-)-**105b**: The above procedure was followed using (-)-**104b** (4.2 g, 14.2 mmol), ethyl hydrogen malonate (1.9 g, 14.2 mmol), and a 1 M solution of 1,3-dicyclohexylcarbodiimide (14.2 mL, 14.2 mmol) to afford amine (-)-**105b** (5.6 g, 97% yield) as a clear, light yellow oil: $[\alpha]_D^{20}$ -31.30° (*c* 1.55, MeOH); IR (thin

film/NaCl) 2955 (br m), 1740 (s), 1655 (s), 1517 (s), 1465 (m), 1416 (m), 1368 (w), 1262 (s), 1238 (m), 1142 (m), 1027 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.91-6.76 (comp m, 3H), 4.81 (m, 0.7H), 4.69 (d, *J* = 15.3 Hz, 0.3H), 4.62 (d, *J* = 17.2 Hz, 0.7H), 4.48 (d, *J* = 15.3 Hz, 0.3H), 4.46 (d, *J* = 17.2 Hz, 0.7H), 4.32 (t, *J* = 7.2 Hz, 0.3H), 4.21 (m, 2H), 3.89 (s, 2.1H), 3.89 (s, 2.1H), 3.87 (s, 0.9H), 3.86 (s, 0.9H), 3.64 (s, 2.1H), 3.56 (s, 0.9H), 3.47-3.35 (comp m, 2H), 1.89 (m, 1H), 1.63 (m, 2H), 1.29 (m, 3H), 0.92 (d, *J* = 6.4 Hz, 2.1H), 0.88 (d, *J* = 6.6 Hz, 0.9H), 0.87 (d, *J* = 6.4 Hz, 2.1H), 0.75 (d, *J* = 6.6 Hz, 0.9H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.7, 167.4, 167.3, 149.5, 148.7, 128.9, 120.2, 118.9, 111.6, 111.5, 111.0, 110.0, 61.5, 61.4, 58.9, 56.4, 56.0, 56.0, 52.3, 52.0, 50.4, 46.6, 41.8, 41.7, 38.7, 38.5, 29.7, 25.1, 24.7, 22.6, 22.3, 22.3, 22.2, 14.1; high resolution mass spectrum (EI) *m/z* 409.2101 [calc'd for $C_{21}H_{31}N_2O_7$ (M+) 409.2101].

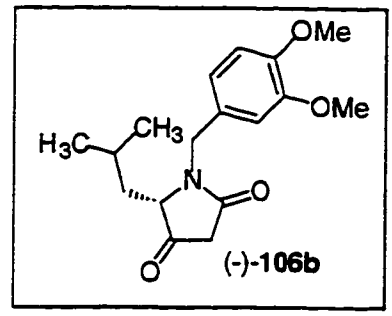
(-)-**105c**: The above procedure was followed using (-)-**104c** (4.5 g, 15.3 mmol), ethyl hydrogen malonate (2.0 g, 15.3 mmol) and a 1 M solution of 1,3-dicyclohexylcarbodiimide (15.3 mL, 15.3 mmol) to afford amine (-)-**105b** (6.2 g, 99% yield) as a clear, light yellow oil: $[\alpha]_D^{20}$ -53.60° (*c* 1.72, MeOH); IR (thin film/NaCl) 2964 (br m), 2877 (w), 2837 (w), 1740 (s), 1656 (s), 1517 (s), 1464 (m), 1415 (m), 1367 (m), 1259 (s), 1239 (s), 1142 (s), 1028 (s), 809 (w), 768 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.90-6.65 (comp m, 3H), 4.97 (d, *J* = 10.2 Hz, 0.7H), 4.60 (m, 2H), 4.21 (q, *J* = 7.1 Hz, 0.6H), 4.16 (q, *J* = 7.1 Hz, 1.4H), 3.90 (d, *J* = 10.8 Hz, 0.3H), 3.85 (s, 4.2H), 3.83 (s, 1.8H), 3.62 (d, *J* = 3.7 Hz, 1H), 3.49 (s, 2.1H), 3.46 (s, 0.9H), 3.37 (m, 1H), 2.05 (m, 1H), 1.53 (m, 0.7H), 1.36 (m, 0.3H), 1.29 (t, *J* = 7.1 Hz, 0.9H), 1.24 (t, *J* = 7.1 Hz, 2.1H), 1.12 (m, 1H), 0.93 (d, *J* = 6.5 Hz, 2.1H), 0.89 (d, *J* = 6.2 Hz, 0.9H), 0.87 (t, *J* = 7.4 Hz, 2.1H), 0.70 (t, *J* = 7.3 Hz, 0.9H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.8, 169.9, 167.8, 167.6, 167.3, 167.2, 149.3, 148.7, 148.3, 148.0, 130.2, 128.9, 120.2, 117.7, 111.5, 111.3,

110.7, 109.0, 66.0, 61.5, 61.4, 61.0, 55.9, 55.8, 51.9, 51.7, 48.3, 46.0, 42.0, 41.6, 34.2, 34.0, 25.0, 24.7, 16.0, 15.8, 14.0, 14.0, 11.0, 10.9; high resolution mass spectrum (EI) *m/z* 409.2103 [calc'd for $C_{21}H_{31}NO_7$ (M+) 409.2101].

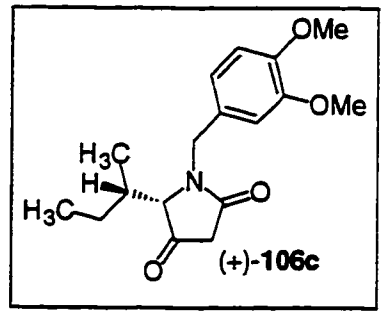
Preparation of Lactams (-)-106a,b and (+)-106c.



(-)-**106a**



(-)-**106b**



(+)-**106c**

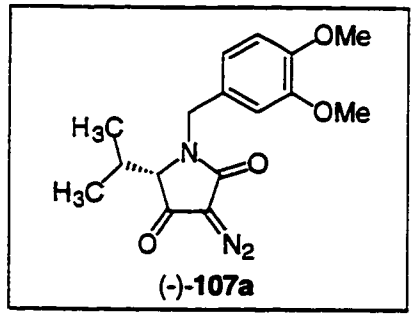
Representative Procedure: Lactam (-)-106a. A three-necked flask was charged with EtOH (10 mL). Sodium metal (240 mg, 10.5 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester(-)-**105a** (4.4 g, 11.1 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in EtOH (20 mL). The mixture was brought to reflux for 5 min and then allowed to cool to rt. The EtOH was removed under reduced pressure and the residue was dissolved in H_2O (50 mL). The aqueous layer was washed with EtOAc (50 mL), and acidified to a pH of 2 with 2 N HCl (10 mL). The acidic solution was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried

over anhydrous $MgSO_4$, filtered, and concentrated to yield a yellow oil. A suspension of the oil in CH_3CN (200 mL) and H_2O (1 mL) was warmed to reflux open to the air for three hours. The mixture was cooled to rt and the CH_3CN/H_2O mixture was removed at reduced pressure to yield lactam (-)-**106a** as a dark yellow oil (2.9 g, 90%) which solidified upon standing to a yellow glassy solid: $[\alpha]_D^{20}$ -33.86° (*c* 0.70, MeOH); IR (thin film/NaCl) 2961 (m), 1765 (m), 1694 (s), 1516 (s), 1416 (m), 1261 (s), 1238 (s), 1155 (m), 1047 (m), 1020 (m), 766 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.78 (comp m, 3H), 5.23 (d, *J* = 14.9 Hz, 1H), 3.95 (d, *J* = 14.8 Hz, 1H), 3.87 (s, 3H), 3.86 (s, 3H), 3.60 (d, *J* = 3.3 Hz, 1H), 3.01 (s, 2H), 2.21 (m, 1H), 1.07 (d, *J* = 7.0 Hz, 3H), 0.88 (d, *J* = 7.0 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.2, 169.1, 149.4, 148.9, 127.7, 120.8, 111.5, 111.2, 70.2, 56.0, 55.9, 43.5, 42.4, 28.5, 17.9, 16.0; high resolution mass spectrum (EI) *m/z* 291.1475 [calc'd for $C_{16}H_{21}NO_4$ (M+) 291.1471].

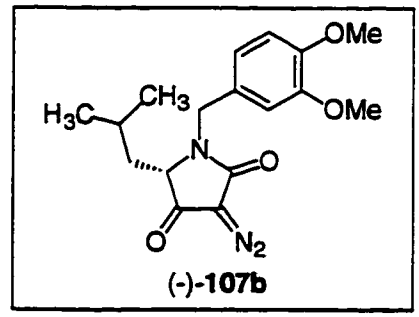
(-)-**106b**: The above procedure was followed using (-)-**105b** (5.6 g, 13.7 mmol), and sodium metal (295 mg, 12.8 mmol) to afford lactam (-)-**106b** as a yellow oil (3.2 g, 82% yield) which solidified upon standing to a dark yellow glassy solid: $[\alpha]_D^{20}$ -12.50° (*c* 0.28, MeOH); IR (thin film/NaCl) 2958 (m), 2928 (m), 1766 (w), 1686 (s), 1516 (s), 1456 (m), 1411 (m), 1260 (s), 1230 (m), 1020 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.80 (comp m, 3H), 5.28 (d, *J* = 14.7 Hz, 1H), 3.90-3.87 (comp m, 7H), 3.74 (m, 1H), 3.16-3.03 (comp m, 2H), 1.82 (m, 1H), 1.65 (m, 2H), 0.92 (d, *J* = 6.7 Hz, 3H), 0.83 (d, *J* = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.7, 168.6, 149.4, 149.0, 127.8, 120.9, 111.6, 111.2, 64.1, 56.0, 55.9, 43.6, 41.1, 38.2, 24.3, 23.5, 22.3; high resolution mass spectrum (EI) *m/z* 305.1628 [calc'd for $C_{17}H_{23}NO_4$ (M+) 305.1627].

(+)-**106c**: The above procedure was followed using (-)-**105c** (4.0 g, 9.78 mmol), and sodium metal (211 mg, 9.19 mmol) to afford lactam (+)-**106c** as a yellow oil (2.0 g, 67% yield) which solidified upon standing to a dark yellow glassy solid: $[\alpha]_D^{20}$ +20.45° (*c* 1.11, MeOH); IR (thin film/NaCl) 2963 (s), 2936 (m), 1766 (m), 1690 (s), 1516 (s), 1462 (m), 1418 (m), 1264 (s), 1237 (s), 1155 (m), 1127 (m), 1041 (m), 733 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.78 (comp m, 3H), 5.26 (d, *J* = 14.7 Hz, 1H), 3.90 (d, *J* = 14.9 Hz, 1H), 3.88 (s, 3H), 3.86 (s, 3H), 3.71 (d, *J* = 3.1 Hz, 1H), 3.01 (s, 2H), 1.94 (m, 1H), 1.50 (m, 2H), 0.89 (t, *J* = 7.4 Hz, 3H), 0.84 (d, *J* = 7.0 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.4, 169.1, 149.4, 148.9, 127.7, 121.0, 111.6, 111.2, 68.6, 56.0, 55.9, 43.3, 42.5, 35.2, 25.2, 13.6, 12.1; high resolution mass spectrum (EI) *m/z* 305.1622 [calc'd for $C_{17}H_{23}NO_4$ (M+) 305.1627].

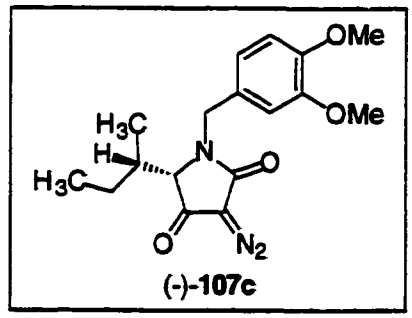
Preparation of Diazo lactams (-)-107a-c.



(-)-**107a**



(-)-**107b**



(-)-**107c**

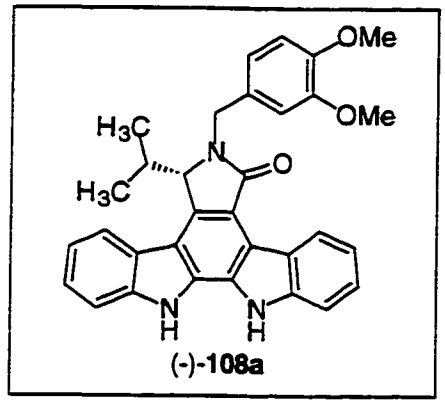
Representative Procedure: Diazo lactam (-)-107a. A stirred solution of lactam (-)-**106a** (4.0 g, 13.7 mmol, 1.0 equiv), *p*-ABSA (3.6 g, 15.1 mmol, 1.1 equiv), and CH_3CN (90 mL) was cooled to 0°C and Et_3N (5.7 mL, 41.1 mmol, 3.0 equiv) was added dropwise to the mixture. After gradually warming to rt, the mixture was stirred for an additional 3 h and the volatiles were removed under reduced pressure. The dark red residue was subjected to flash chromatography (50% EtOAc/hexanes eluent) to afford diazo lactam (-)-**107a** (3.9 g, 89% yield) as a bright yellow foam: $[\alpha]_D^{20}$ -109.18° (*c* 0.49, MeOH); IR (thin film/NaCl) 2961 (br w), 2934 (w), 2123 (s), 1683 (s), 1515 (m), 1405 (m), 1361 (m), 1261 (m), 1237 (w), 1219 (w), 1156 (w), 1027 (w), 795 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.77 (comp m, 3H), 5.17 (d, *J* = 15.0 Hz, 1H), 3.97 (d, *J* = 15.0 Hz, 1H), 3.88 (s, 3H), 3.87 (s, 3H), 3.62 (d, *J* = 3.2 Hz, 1H), 2.26 (ttd, *J* = 3.2, 7.0, 14.0 Hz, 1H), 1.12 (d, *J* = 7.0 Hz, 3H), 0.92 (d, *J* = 7.0 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 188.9, 162.2, 149.5, 148.9, 128.0, 120.7, 111.3, 111.2, 67.8, 56.0, 55.9, 44.4, 28.7, 17.9, 15.9; high resolution mass spectrum (EI) *m/z* 317.1371 [calc'd for $C_{16}H_{19}N_3O_4$ (M+) 317.1376].

(-)-**107b**: The above procedure was followed using (-)-**106b** (3.0 g, 9.8 mmol), *p*-ABSA (2.6 mg, 10.8 mmol) and Et_3N (4.1 mL, 29.4 mmol to afford diazo lactam (-)-**107b** (1.9 g, 58% yield) as a bright yellow oil: $[\alpha]_D^{20}$ -108.3° (*c* 0.7, MeOH); IR (thin film/NaCl) 2958 (m), 2923 (m), 2848 (w), 2122 (s), 1681 (s), 1511 (m), 1400 (m), 1361 (m), 1260 (m), 1235 (m), 1140 (w), 1030 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.83-6.78 (comp m, 3H), 5.20 (d, *J* = 14.9 Hz, 1H), 3.91 (d, *J* = 14.9 Hz, 1H), 3.88 (s, 3H), 3.87 (s, 3H), 3.72 (dd, *J* = 4.0, 7.4 Hz, 1H), 1.90 (m, 1H), 1.69 (m, 2H), 0.92 (d, *J* = 6.7 Hz, 3H), 0.86 (d, *J* = 6.5 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 189.5, 161.6, 149.4, 148.9, 127.9, 120.6, 111.3,

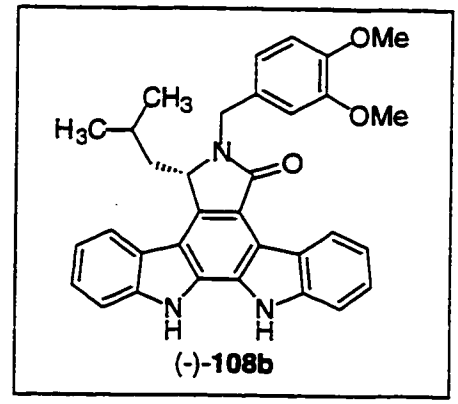
111.1, 61.9, 55.9, 55.8, 44.2, 38.0, 24.0, 23.4, 22.5; high resolution mass spectrum (EI) *m/z* 331.1534 [calc'd for $C_{17}H_{21}N_3O_4$ (M+) 331.1532].

(-)-**107c**: The above procedure was followed using (-)-**106c** (2.8 g, 9.2 mmol), *p*-ABSA (2.32 g, 9.7 mmol) and Et_3N (3.9 mL, 27.6 mmol) to afford diazo lactam (-)-**107c** (2.8 g, 92% yield) as a bright yellow oil: $[\alpha]_D^{20}$ -141.2° (*c* 1.8, MeOH); IR (thin film/NaCl) 2968 (s), 2943 (s), 2837 (m), 2121 (s), 1680 (s), 1591 (m), 1400 (s), 1260 (s), 1030 (m), 940 (w), 755 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.81-6.75 (comp m, 3H), 5.16 (d, *J* = 14.9 Hz, 1H), 3.90 (d, *J* = 14.9 Hz, 1H), 3.86 (s, 3H), 3.85 (s, 3H), 3.68 (d, *J* = 3.1 Hz, 1H), 1.94 (dqd, *J* = 3.1, 7.1, 14.4 Hz, 1H), 1.54 (m, 2H), 0.87 (t, *J* = 7.4 Hz, 3H), 0.85 (d, *J* = 6.8 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 188.8, 162.1, 149.4, 148.8, 127.9, 120.7, 111.3, 111.1, 66.2, 55.9, 55.8, 44.0, 35.3, 25.1, 13.2, 12.1; high resolution mass spectrum (EI) *m/z* 331.1540 [calc'd for $C_{17}H_{21}N_3O_4$ (M+) 331.1532].

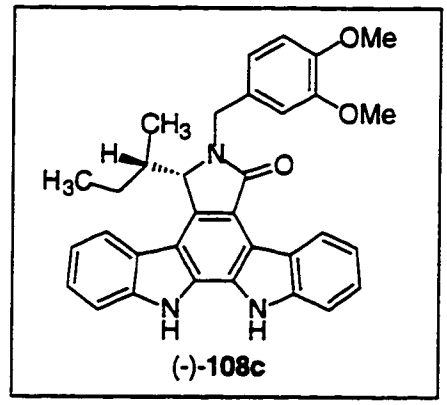
Preparation of Aglycons (-)-108a-c.



(-)-**108a**



(-)-**108b**



(-)-**108c**

Representative Procedure: Aglycon (-)-108a. To a three-necked flask equipped with a condenser were added the diazo lactam (-)-**107a** (3.0 g, 9.46 mmol, 1.0 equiv), 2,2'-biindole (**11**) (2.2 g, 9.46 mmol, 1.0 equiv), $Rh_2(OAc)_4$ (42 mg, 0.095 mmol, 0.01 equiv) and pinacolone (95 mL). The whole was degassed by bubbling a stream of N_2 through the solution for 2 h. The mixture was then warmed to reflux for an additional 8 h. The reaction mixture was concentrated to a dark brown foamy solid, which was adsorbed onto silica gel and subjected to flash chromatography (50% EtOAc/hexanes eluent), affording unreacted **11** (1.1 g, 50% yield) as a white powder, and aglycon (-)-**108a** (1.2 g, 25% yield; 50% based on recovered **11**) was isolated as a pale yellow solid: mp >290 °C (dec.) $[\alpha]_D^{20}$ -39.17° (*c* 0.24, MeOH); IR (thin film/NaCl) 3337 (br w), 2960 (w), 2929 (w), 1640

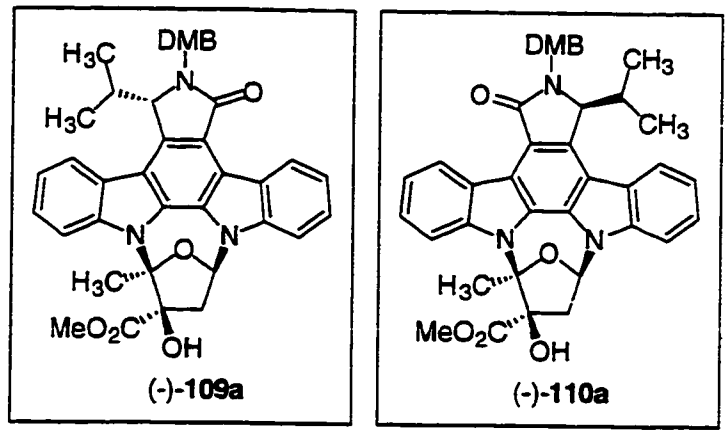
(s), 1574 (m), 1514 (m), 1459 (m), 1408 (m), 1393 (m), 1326 (m), 1260 (m), 1237 (m), 1139 (m), 1027 (m), 746 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 10.89 (s, 1H), 10.66 (s, 1H), 9.52 (d, *J* = 7.8 Hz, 1H), 7.97 (d, *J* = 7.9 Hz, 1H), 7.67 (d, *J* = 8.1 Hz, 1H), 7.62 (d, *J* = 8.2 Hz, 1H), 7.42 (m, 1H), 7.27 (m, 2H), 7.01 (d, *J* = 1.6 Hz, 1H), 6.92-6.86 (comp m, 3H), 5.60 (d, *J* = 15.4 Hz, 1H), 5.15 (d, *J* = 1.0 Hz, 1H), 4.44 (d, *J* = 15.5 Hz, 1H), 3.74 (s, 3H), 3.70 (s, 3H), 2.97 (m, 1H), 1.54 (d, *J* = 7.4 Hz, 3H), 0.52 (d, *J* = 6.8 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 171.8, 150.6, 149.7, 140.9, 140.8, 136.6, 132.0, 129.4, 127.0, 126.8, 126.8, 126.3, 125.9, 124.4, 123.5, 122.7, 121.0, 120.9, 120.2, 120.0, 117.3, 115.0, 112.9, 112.6, 111.8, 64.9, 56.1, 56.1, 46.4, 32.1, 22.6, 14.7; high resolution mass spectrum (FAB) *m/z* 504.2287 [calc'd for $C_{32}H_{30}N_3O_3$ (M+H) 504.2287].

(-)-**108b**: The above procedure was followed using (-)-**107b** (2.7 g, 8.2 mmol), 2,2'-biindole (**11**; 1.89 g, 8.2 mmol) and $Rh_2(OAc)_4$ (36 mg, 0.082 mmol) to afford aglycon unreacted **11** (1.0 g, 53% yield) as a white powder, and aglycon (-)-**108b** (900 mg, 21% yield; 40% yield based on recovered **11**) was isolated as a pale yellow solid: mp 256-260 °C (dec.); $[\alpha]_D^{20}$ -30.00° (*c* 0.27, MeOH); IR (thin film/NaCl) 3335 (br w), 2953 (w), 2931 (w), 1640 (s), 1577 (w), 1514 (m), 1456 (m), 1413 (m), 1394 (m), 1327 (s), 1258 (s), 1237 (s), 1139 (m), 1026 (w), 747 (m) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 10.90 (s, 1H), 10.69 (s, 1H), 9.54 (d, *J* = 7.9 Hz, 1H), 8.12 (d, *J* = 7.9 Hz, 1H), 7.68 (d, *J* = 8.1 Hz, 1H), 7.63 (d, *J* = 8.1 Hz, 1H), 7.43 (t, *J* = 7.6 Hz, 2H), 7.28 (t, *J* = 7.6 Hz, 2H), 7.09 (m, 1H), 6.99 (m, 1H), 6.90 (m, 1H), 5.45 (d, *J* = 15.2 Hz, 1H), 5.28 (t, *J* = 4.0 Hz, 1H), 4.39 (d, *J* = 15.2 Hz, 1H), 3.76 (s, 3H), 3.74 (s, 3H), 2.41 (m, 2H), 1.39 (tt, *J* = 6.5, 13.0 Hz, 1H), 0.84 (d, *J* = 6.7 Hz, 3H), 0.20 (d, *J* = 6.6 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 170.7, 150.3, 149.4, 140.5, 140.5, 135.9, 131.7, 128.9, 126.7, 126.5, 125.9, 125.6, 124.1, 123.2, 122.5, 120.7, 120.6, 119.9, 119.9, 117.2,

115.3, 112.7, 112.6, 112.2, 111.5, 59.2, 55.8, 55.8, 44.3, 38.7, 23.8, 23.7; high resolution mass spectrum (EI) *m/z* 517.2364 [calc'd for $C_{33}H_{31}N_3O_3$ (M+) 517.2365].

(-)-**108c**: The above procedure was followed using (-)-**107c** (2.8 g, 8.5 mmol), 2,2'-biindole (**11**; 1.96 g, 8.5 mmol) and $Rh_2(OAc)_4$ (37 mg, 0.085 mmol) to afford unreacted **11** (860 mg, 44% yield) as a white powder and aglycon (-)-**108c** (1.31 g, 30% yield; 53% yield based on recovered **11**) was isolated as a pale yellow solid: mp >310 °C (dec.); $[\alpha]_D^{20}$ -47.89° (*c* 0.19, MeOH); IR (thin film/NaCl) 3331 (br w), 3055 (w), 2961 (w), 2926 (m), 1639 (s), 1576 (w), 1515 (m), 1460 (m), 1414 (m), 1328 (m), 1264 (s), 1142 (w), 740 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 11.01 (s, 1H), 10.78 (s, 1H), 9.50 (d, *J* = 8.0 Hz, 1H), 7.96 (d, *J* = 7.9 Hz, 1H), 7.69 (d, *J* = 8.1 Hz, 1H), 7.64 (d, *J* = 8.1 Hz, 1H), 7.43 (m, 2H), 7.27 (t, *J* = 7.4 Hz, 2H), 7.00 (s, 1H), 6.89 (s, 2H), 5.61 (d, *J* = 15.4 Hz, 1H), 5.14 (s, 1H), 4.40 (d, *J* = 15.4 Hz, 1H), 3.75 (s, 3H), 3.71 (s, 3H), 2.68 (m, 1H), 1.56 (d, *J* = 7.3 Hz, 3H), 1.07 (m, 1H), 0.88 (m, 1H), 0.54 (t, *J* = 7.4 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 171.7, 150.5, 149.6, 140.9, 140.7, 136.4, 131.9, 127.0, 126.8, 126.3, 125.9, 124.3, 123.4, 122.7, 120.9, 120.9, 120.2, 120.1, 117.3, 115.0, 112.9, 112.8, 112.6, 112.5, 111.8, 65.2, 56.1, 56.0, 46.3, 38.9, 22.5, 19.7, 12.4; high resolution mass spectrum (FAB) *m/z* 518.2446 [calc'd for $C_{33}H_{32}N_3O_3$ (M+H) 518.2444].

Preparation of Indolocarbazoles (-)-109a and (-)-110a.



Indolocarbazoles (-)-109a and (-)-110a. To a refluxing solution of aglycon (-)-**108a** (600 mg, 1.2 mmol, 1.0 equiv) and camphorsulfonic acid (29 mg, 0.12 mmol, 0.1.0 equiv) in 1,2-dichloroethane (30 mL) was added, *via* addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)-**17** (525 mg, 2.4 mmol, 2 equiv) in 1,2-dichloroethane (25 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% aqueous $NaHCO_3$ (20 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 20 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:1 mixture of regioisomers (-)-**109a** and (-)-**110a** (535 mg, 68% yield) as a white film. Separation of the regioisomers was achieved using HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent).

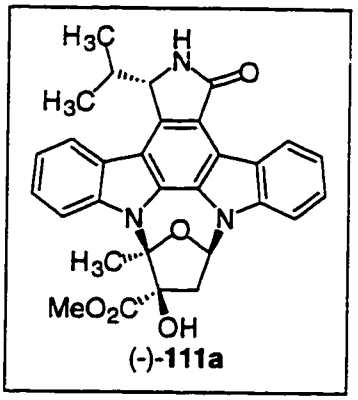
(-)-**109a**: $[\alpha]_D^{20}$ +16.00° (*c* 0.25, MeOH); IR (thin film/NaCl) 3363 (br w), 2957 (m), 2926 (m), 1725 (s), 1674 (s), 1584 (m), 1514 (m), 1459 (s), 1451 (s), 1391 (m), 1259 (s), 1235 (s), 1138 (s), 1097 (m), 1027 (m), 747 (m) cm^{-1}; ^1H

NMR (500 MHz, acetone-d_6) δ 9.50 (d, J = 7.9 Hz, 1H), 7.99 (d, J = 8.5 Hz, 1H), 7.94 (d, J = 7.8 Hz, 1H), 7.81 (d, J = 8.3 Hz, 1H), 7.51 (ddd, J = 1.1, 7.1, 8.2 Hz, 1H), 7.42 (ddd, J = 1.1, 7.3, 8.4 Hz, 1H), 7.31 (m, 2H), 7.16 (dd, J = 4.8, 7.4 Hz, 1H), 7.01 (s, 1H), 6.88 (m, 2H), 5.63 (d, J = 15.5 Hz, 1H), 5.24 (s, 1H), 5.16 (d, J = 1.0 Hz, 1H), 4.43 (d, J = 15.5 Hz, 1H), 4.01 (s, 3H), 3.75 (s, 3H), 3.72 (s, 3H), 3.50 (dd, J = 7.4, 14.1 Hz, 1H), 2.93 (m, 1H), 2.23 (m, 4H), 1.56 (d, J = 7.4 Hz, 3H), 0.57 (d, J = 6.8 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 171.2, 150.6, 149.8, 141.4, 138.4, 136.3, 131.8, 130.2, 127.5, 127.3, 126.5, 125.8, 125.5, 125.2, 124.3, 122.6, 121.4, 120.9, 120.6, 120.5, 117.3, 116.0, 115.3, 113.0, 109.3, 100.3, 86.5, 86.2, 64.8, 56.2, 56.1, 53.4, 46.4, 43.4, 33.6, 23.4, 22.5, 14.8; high resolution mass spectrum (FAB) *m/z* 660.2701 [calc'd for $C_{39}H_{38}N_3O_7$ (M+H) 660.2710].

(-)-**110a**: $[\alpha]_D^{20}$ -18.91° (*c* 0.2, MeOH); IR (thin film/NaCl) 3363 (br w), 2957 (m), 2926 (m), 1725 (s), 1674 (s), 1584 (m), 1514 (m), 1459 (s), 1451 (s), 1391 (m), 1259 (s), 1235 (s), 1138 (s), 1097 (m), 1027 (m), 747 (m) cm^{-1}; ^{1}H NMR (500 MHz, acetone-d_6) δ 9.70 (d, J = 8.1 Hz, 1H), 8.01 (d, J = 7.8 Hz, 1H), 7.95 (d, J = 8.5 Hz, 1H), 7.89 (d, J = 8.2 Hz, 1H), 7.51 (ddd, J = 1.0, 7.2, 8.1 Hz, 1H), 7.42 (ddd, J = 1.4, 7.0, 8.4 Hz, 1H), 7.32 (m, 2H), 7.16 (dd, J = 5.0, 7.4 Hz, 1H), 7.00 (s, 1H), 6.90 (m, 2H), 5.63 (d, J = 15.5 Hz, 1H), 5.40 (s, 1H), 5.17 (d, J = 1.1 Hz, 1H), 4.43 (d, J = 15.5 Hz, 1H), 4.00 (s, 3H), 3.76 (s, 3H), 3.71 (s, 3H), 3.56 (dd, J = 7.5, 14.1 Hz, 1H), 2.97 (m, 1H), 2.43 (dd, J = 5.0, 14.1 Hz, 1H), 2.23 (m, 4H), 1.56 (d, J = 7.4 Hz, 3H), 0.49 (d, J = 6.7 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 171.0, 149.3, 148.3, 140.1, 137.5, 136.2, 130.4, 127.1, 126.9, 126.7, 125.8, 125.4, 125.3, 122.6, 120.7, 120.4, 120.2, 119.6, 114.3, 113.0, 111.2, 111.0, 108.4, 98.7, 85.2, 84.4, 63.7, 55.9, 55.9, 54.0, 45.9, 42.2,

31.2, 29.7, 22.9, 14.6; high resolution mass spectrum (FAB) *m/z* 660.2701 [calc'd for $C_{39}H_{38}N_3O_7$ (M+H) 660.2710].

Preparation of (-)-7-(*S*)-Isopropyl K252a 111a.

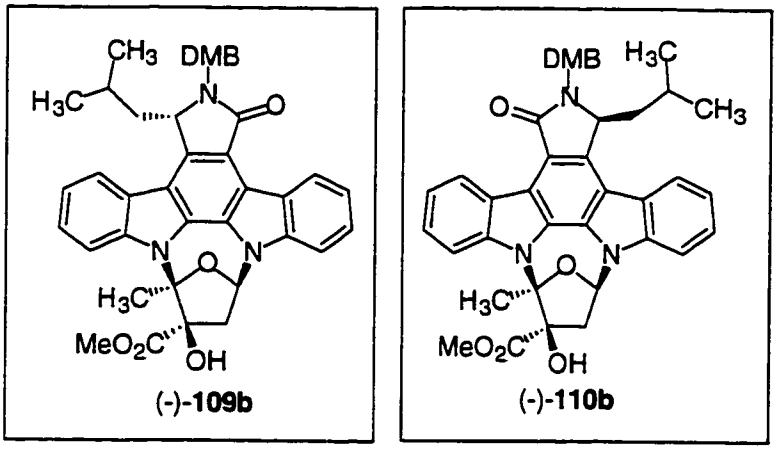


(-)-**111a**

(-)-7-(*S*)-Isopropyl K252a 111a. A solution of protected amide (-)-**109a** (20 mg, 0.03 mmol, 1.0 equiv) and anisole (325 μL, 3.0 mmol, 100 equiv) in CH_2Cl_2 (1 mL) was treated dropwise with TFA (1 mL). After stirring at rt for 12 h, the reaction was quenched with 20% aqueous $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and concentrated to a residue. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**111a** (9.6 mg, 62% yield) as a pale yellow film: $[\alpha]_D^{20}$ -34.90° (*c* 0.51, MeOH); IR (thin film/NaCl) 3263 (br w), 2957 (w), 2925 (w), 2852 (w), 1731 (m), 1668 (s), 1582 (m), 1457 (s), 1391 (m), 1312 (m), 1256 (m), 1200 (m), 1139 (m), 1091 (s), 1017 (s), 741 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.39 (d, *J* = 7.9 Hz, 1H), 8.14 (d, *J* = 7.8 Hz, 1H), 8.02 (d, *J* = 8.5 Hz, 1H), 7.78 (d, *J* = 8.2 Hz, 1H), 7.72 (s, 1H), 7.47 (m, 2H), 7.35 (t, *J* = 7.4 Hz, 1H), 7.27 (t, *J* = 7.5 Hz, 1H), 7.15 (dd, *J* = 4.9, 7.4 Hz, 1H), 5.43 (s, 1H), 5.33 (s, 1H), 4.01 (s, 3H), 3.50 (dd, *J* = 7.5, 14.1 Hz, 1H), 2.98 (m, 1H), 2.24 (m, 4H), 1.47

(d, J = 7.0 Hz, 3H), 0.43 (d, J = 6.7 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 172.8, 141.4, 138.3, 137.5, 130.2, 127.5, 126.4, 125.7, 125.4, 124.3, 122.7, 121.3, 121.0, 120.4, 117.4, 116.0, 115.6, 109.2, 100.2, 86.5, 86.2, 62.7, 62.6, 53.4, 43.3, 31.1, 23.4, 21.6, 14.1; high resolution mass spectrum (FAB) *m/z* 510.1036 [calc'd for $C_{30}H_{28}N_3O_5$ (M+H) 510.1042].

Preparation of Indolocarbazoles (-)-109b and (-)-110b.



Indolocarbazoles (-)-109b and (-)-110b. To a refluxing solution of aglycon (-)-**108b** (600 mg, 1.16 mmol, 1.0 equiv) and camphorsulfonic acid (27 mg, 0.12 mmol, 0.1 equiv) in 1,2-dichloroethane (30 mL) was added, *via* addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)-**17** (511 mg, 2.32 mmol, 2 equiv) in 1,2-dichloroethane (20 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% aqueous $NaHCO_3$ (20 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 20 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography

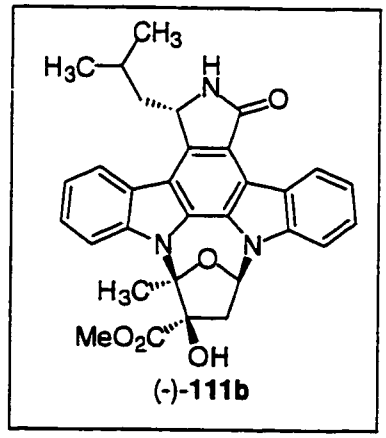
(50% EtOAc/hexanes eluent) to provide a 2:1 mixture of regioisomers (-)-**109b** and (-)-**110b** (586 mg, 75% yield) as a white film. Separation of the regioisomers was achieved using HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent).

(-)-**109b**: $[\alpha]_D^{20}$ -22.72° (*c* 0.55, MeOH); IR (thin film/NaCl) 3368 (br w), 2953 (m), 2867 (w), 1732 (m), 1673 (s), 1584 (w), 1514 (s), 1451 (s), 1392 (m), 1360 (m), 1258 (s), 1238 (s), 1199 (m), 1138 (s), 1082 (w), 1028 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.52 (d, *J* = 7.7 Hz, 1H), 8.08 (d, *J* = 7.7 Hz, 1H), 8.00 (d, *J* = 8.4 Hz, 1H), 7.82 (d, *J* = 7.8 Hz, 1H), 7.51 (ddd, *J* = 1.1, 7.1, 8.2 Hz, 1H), 7.43 (ddd, *J* = 1.1, 7.2, 8.3 Hz, 1H), 7.32 (t, *J* = 7.5 Hz, 1H), 7.30 (t, *J* = 7.5 Hz, 1H), 7.17 (dd, *J* = 4.8, 7.4 Hz, 1H), 7.07-6.89 (comp m, 3H), 5.47 (d, *J* = 15.3 Hz, 1H), 5.31 (m, 2H), 4.39 (d, *J* = 15.3 Hz, 1H), 4.00 (s, 3H), 3.75 (s, 3H), 3.74 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.39 (m, 1H), 2.23 (s, 3H), 2.20 (dd, *J* = 5.0, 14.3 Hz, 1H), 1.45 (m, 1H), 0.88 (d, *J* = 6.7 Hz, 3H), 0.28 (d, *J* = 6.6 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.8, 170.2, 150.6, 149.7, 141.4, 138.3, 136.0, 131.9, 129.9, 127.5, 127.3, 126.4, 125.7, 125.5, 125.1, 124.3, 122.7, 121.2, 120.9, 120.8, 120.5, 117.5, 115.9, 113.0, 113.0, 109.2, 100.2, 86.4, 86.2, 59.2, 56.1, 56.1, 53.4, 44.5, 43.3, 39.2, 24.2, 24.1, 23.8, 23.4; high resolution mass spectrum (FAB) *m/z* 674.2867 [calc'd for $C_{40}H_{40}N_3O_7$ (M+H) 674.2866].

(-)-**110b**: $[\alpha]_D^{20}$ -47.62° (*c* 0.10, MeOH); IR (thin film/NaCl) 3498 (br w), 3010 (w), 2988 (m), 2950 (m), 1736 (s), 1675 (s), 1596 (m), 1525 (m), 1460 (s), 1392 (s), 1262 (s), 1152 (m), 1082 (m), 1025 (m), 755 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.78 (d, *J* = 7.8 Hz, 1H), 8.10 (d, *J* = 7.9 Hz, 1H), 8.03 (d, *J* = 8.2 Hz, 1H), 7.98 (d, *J* = 8.1 Hz, 1H), 7.58 (ddd, *J* = 1.2, 7.1, 8.0 Hz, 1H), 7.52 (ddd, *J* = 1.0, 7.0, 8.1 Hz, 1H), 7.45 (m, 2H), 7.15 (dd, *J* = 4.6, 7.4 Hz, 1H), 7.31

(m, 1H), 7.03 (m, 1H), 6.99 (m, 1H), 5.52 (d, J = 15.3 Hz, 1H), 5.37 (m, 1H), 5.35 (s, 1H), 4.48 (d, J = 15.4 Hz, 1H), 4.04 (s, 3H), 3.78 (s, 3H), 3.76 (s, 3H), 3.50 (dd, J = 7.4, 14.0 Hz, 1H), 2.44 (m, 2H), 2.26 (m, 4H), 1.71 (m, 1H), 0.84 (d, J = 7.4 Hz, 3H), 0.50 (d, J = 7.6 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.8, 170.9, 150.2, 149.8, 140.8, 139.0, 136.8, 130.9, 130.0, 127.9, 127.7, 126.5, 125.8, 125.7, 125.3, 124.9, 122.6, 121.6, 121.4, 120.9, 120.7, 117.7, 116.1, 115.2, 112.7, 109.8, 100.1, 86.8, 86.7, 65.5, 56.4, 56.3, 52.6, 46.7, 43.8, 39.5, 23.4, 22.5, 19.9, 12.2; high resolution mass spectrum (FAB) *m/z* 674.2864 [calc'd for $C_{40}H_{40}N_3O_7$ (M+H) 674.2866].

Preparation of (-)-7-(*S*)-Isobutyl K252a 111b.

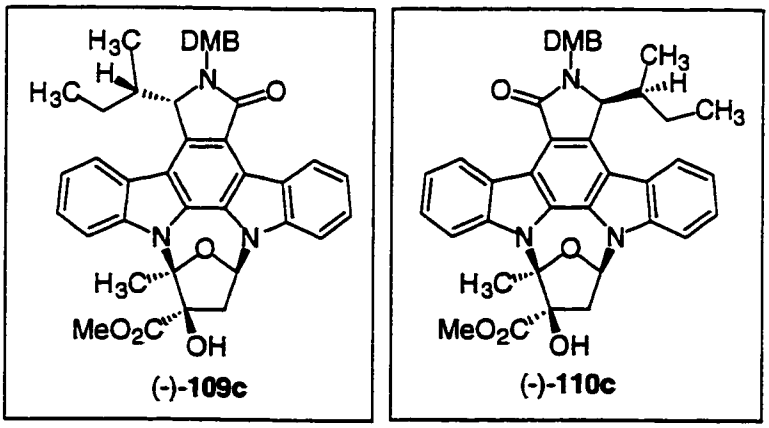


(-)-**111b**

(-)-7-(*S*)-Isobutyl K252a 111b. A solution of protected amide (-)-**109b** (25 mg, 0.04 mmol, 1.0 equiv) and anisole (870 µL, 8.0 mmol, 200 equiv) in CH_2Cl_2 (1 mL) was treated dropwise with TFA (1 mL). After stirring at rt for 12 h, the reaction was quenched with 20% aqueous $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50%

EtOAc/hexanes eluent) afforded (-)-**111b** (17.5 mg, 83% yield) as a yellow film: $[\alpha]_D^{20}$ -27.96° (*c* 1.01, MeOH); IR (thin film/NaCl) 3243 (br w), 2953 (m), 2868 (w), 1732 (m), 1669 (s), 1582 (m), 1458 (s), 1391 (m), 1366 (m), 1314 (m), 1282 (m), 1256 (s), 1200 (s), 1141 (s), 1017 (w), 743 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.37 (d, *J* = 7.9 Hz, 1H), 8.12 (m, 2H), 8.03 (d, *J* = 8.4 Hz, 1H), 7.77 (d, *J* = 8.2 Hz, 1H), 7.49 (m, 2H), 7.38 (t, *J* = 7.4 Hz, 1H), 7.26 (t, *J* = 7.5 Hz, 1H), 7.14 (dd, *J* = 5.2, 7.2 Hz, 1H), 5.38 (m, 2H), 4.01 (s, 3H), 3.51 (dd, *J* = 7.5, 14.1 Hz, 1H), 2.40 (t, *J* = 11.5 Hz, 1H), 2.31 (dd, *J* = 4.9, 14.1 Hz, 1H), 2.21 (m, 4H), 1.43 (m, 1H), 1.33 (d, *J* = 6.5 Hz, 3H), 0.95 (d, *J* = 6.6 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 172.3, 141.5, 139.0, 138.4, 130.2, 127.4, 127.3, 126.4, 125.7, 125.4, 124.3, 122.7, 121.3, 120.9, 118.4, 116.1, 115.5, 109.2, 100.3, 86.5, 86.2, 56.2, 56.1, 53.5, 45.3, 43.4, 26.8, 24.4, 23.4, 22.0; high resolution mass spectrum (FAB) *m/z* 524.2183 [calc'd for $C_{31}H_{30}N_3O_5$ (M+H) 524.2185].

Preparation of Indolocarbazoles (-)-109c and (-)-110c.



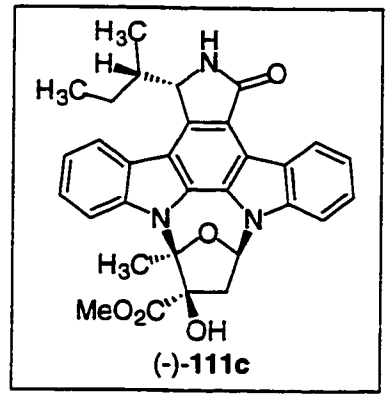
Indolocarbazoles (-)-109c and (-)-110c. To a refluxing solution of aglycon (-)-**108c** (220 mg, 0.43 mmol, 1.0 equiv) and camphorsulfonic acid (10 mg, 0.04 mmol, 0.1 equiv) in 1,2-dichloroethane (14 mL) was added, *via* addition funnel over 24 h, a solution of alcohols (-)-**16a-c**/(+)-**17** (189 mg, 0.86 mmol, 2.0 equiv) in 1,2-dichloroethane (10 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% aqueous $NaHCO_3$ (10 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 10 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:1 mixture of regioisomers (-)-**109c** and (-)-**110c** (210 mg, 73% yield) as a yellow film. Separation of the regioisomers was achieved using HPLC (190:10:1 CH_2Cl_2:EtOAc:MeOH eluent).

(-)-**109c**: $[\alpha]_D^{20}$ -32.93° (*c* 0.91, MeOH); IR (thin film/NaCl) 3492 (br w), 3005 (w), 2960 (m), 2933 (w), 1732 (m), 1671 (s), 1584 (m), 1515 (m), 1459 (s), 1392 (s), 1258 (s), 1138 (m), 1080 (m), 1027 (m), 747 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.50 (d, *J* = 7.7 Hz, 1H), 7.99 (d, *J* = 7.8 Hz, 1H), 7.93 (d, *J* = 7.6 Hz, 1H), 7.81 (d, *J* = 8.3 Hz, 1H), 7.51 (ddd, *J* = 1.2, 7.1, 8.2 Hz, 1H), 7.42 (ddd, *J* = 1.2, 7.2, 8.4 Hz, 1H), 7.31 (m, 2H), 7.16 (dd, *J* = 4.8, 7.4 Hz, 1H), 7.00 (m, 1H), 6.84 (m, 2H), 5.63 (d, *J* = 15.5 Hz, 1H), 5.25 (s, 1H), 5.16 (s, 1H), 4.39 (d, *J* = 15.5 Hz, 1H), 4.01 (s, 3H), 3.74 (s, 3H), 3.71 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.65 (m, 1H), 2.22 (m, 4H), 1.56 (d, *J* = 7.4 Hz, 3H), 1.09 (m, 1H), 0.94 (m, 1H), 0.59 (t, *J* = 7.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 171.2, 150.7, 149.8, 141.5, 138.6, 136.2, 131.9, 130.4, 127.6, 127.5, 126.7, 125.9, 125.7, 125.3, 124.4, 122.7, 121.5, 121.0, 120.7, 120.7, 117.4, 116.2, 115.5, 113.1, 109.5, 100.4, 86.6, 86.4, 65.2, 56.3, 56.2, 53.6, 46.3, 43.5, 39.2,

23.6, 22.6, 19.7, 12.4; high resolution mass spectrum (FAB) *m/z* 674.2858 [calc'd for $C_{40}H_{40}N_3O_7$ (M+H) 674.2866].

(-)-**110c**: $[\alpha]_D^{20}$ -60.94° (*c* 0.10, MeOH); IR (thin film/NaCl) 3498 (br w), 3010 (w), 2988 (m), 2950 (m), 1736 (s), 1675 (s), 1596 (m), 1525 (m), 1460 (s), 1392 (s), 1262 (s), 1152 (m), 1082 (m), 1025 (m), 755 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.65 (d, *J* = 8.0 Hz, 1H), 8.08 (d, *J* = 7.8 Hz, 1H), 7.99 (d, *J* = 7.9 Hz, 1H), 7.91 (d, *J* = 8.2 Hz, 1H), 7.53 (ddd, *J* = 1.1, 7.1, 8.1 Hz, 1H), 7.48 (ddd, *J* = 1.2, 7.0, 8.3 Hz, 1H), 7.36 (m, 2H), 7.14 (dd, *J* = 4.7, 7.4 Hz, 1H), 7.01 (m, 1H), 6.91 (m, 2H), 5.56 (d, *J* = 15.5 Hz, 1H), 5.25 (s, 1H), 5.16 (s, 1H), 4.39 (d, *J* = 15.5 Hz, 1H), 4.01 (s, 3H), 3.74 (s, 3H), 3.71 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.65 (m, 1H), 2.22 (m, 4H), 1.56 (d, *J* = 7.4 Hz, 3H), 1.09 (m, 1H), 0.94 (m, 1H), 0.59 (t, *J* = 7.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 171.2, 150.7, 149.8, 141.5, 138.6, 136.2, 131.9, 130.4, 127.6, 127.5, 126.7, 125.9, 125.7, 125.3, 124.4, 122.7, 121.5, 121.0, 120.7, 120.7, 117.4, 116.2, 115.5, 113.1, 109.5, 100.4, 86.6, 86.4, 65.2, 56.3, 56.2, 53.6, 46.3, 43.5, 39.2, 23.6, 22.6, 19.7, 12.4; high resolution mass spectrum (FAB) *m/z* 674.2858 [calc'd for $C_{40}H_{40}N_3O_7$ (M+H) 674.2866].

Preparation of (-)-7-(*S*)-Secbutyl K252a 111c.

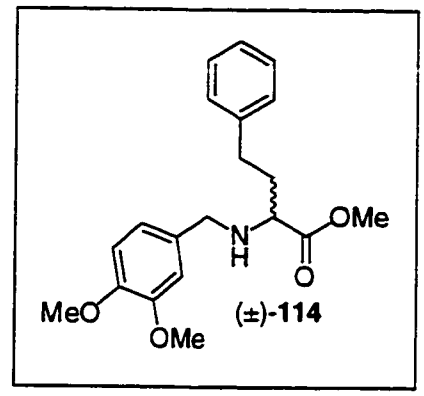


(-)-**111c**

(-)-7-(*S*)-Secbutyl K252a 111c. A solution of protected amide (-)-**109c** (26 mg, 0.04 mmol, 1.0 equiv) and anisole (430 mg, 4.0 mmol, 100 equiv) in CH_2Cl_2 (1 mL) was treated dropwise with TFA (1 mL). After stirring at rt for 12 h, the reaction was quenched with 20% aqueous $NaHCO_3$ (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**111c** (15 mg, 71% yield) as a pale yellow solid: $[\alpha]_D^{20}$ -31.76° (*c* 0.34, MeOH); IR (thin film/NaCl) 3329 (br w), 2957 (w), 2928 (w), 1729 (m), 1666 (s), 1632 (m), 1452 (s), 1391 (m), 1349 (w), 1200 (m), 1139 (m), 1016 (s), 952 (s), 741 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.39 (d, J = 7.9 Hz, 1H), 8.13 (d, J = 7.7 Hz, 1H), 8.02 (d, J = 8.5 Hz, 1H), 7.78 (d, J = 8.3 Hz, 1H), 7.72 (s, 1H), 7.47 (m, 2H), 7.35 (t, J = 7.5 Hz, 1H), 7.27 (t, J = 7.5 Hz, 1H), 7.15 (dd, J = 4.9, 7.4 Hz, 1H), 5.37 (s, 1H), 5.30 (s, 1H), 4.01 (s, 3H), 3.49 (dd, J = 7.5, 14.1 Hz, 1H), 2.70 (m, 1H), 2.24 (s, 4H), 1.48 (d, J = 7.0 Hz, 3H), 1.00 (m, 1H), 0.77 (m, 1H), 0.55 (t, J = 7.5 Hz, 3H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 172.6, 141.5, 138.4, 137.2, 130.2, 127.4, 126.4, 125.7, 125.4, 124.3, 122.8, 121.3, 120.9, 120.4, 117.4, 116.0, 115.7, 109.2, 100.3,

86.5, 86.2, 63.3, 63.2, 53.4, 43.4, 38.1, 23.4, 21.7, 18.4, 12.1; high resolution mass spectrum (FAB) *m/z* 524.2178 [calc'd for $C_{31}H_{30}N_3O_5$ (M+H) 524.2185].

Preparation of Amine (±)-114.

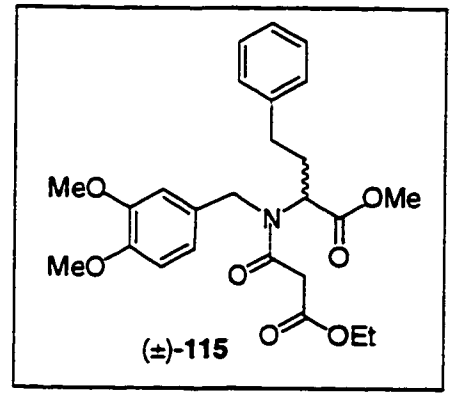


(±)-**114**

Amine (±)-114. To a solution of homophenylalanine methyl ester (±)-**113** (6.1 g, 26.6 mmol, 1.0 equiv), in MeOH (10 mL) was added Et_3N (3.7 mL, 26.6 mmol, 1.0 equiv) followed by 3,4-dimethoxybenzaldehyde (3.1 g, 18.7 mmol, 0.7 equiv) as a solution in EtOH (50 mL). The mixture was concentrated under reduced pressure to yield the crude imine as a white solid. The imine was suspended in EtOH (65 mL) and $NaBH_4$ (1.0 g, 26.6 mmol, 1.0 equiv) was added portionwise over a 15 minute period. The heterogeneous mixture was stirred for 24 h at rt. The heterogeneous mixture was stirred for 24 h at rt. At the end of this period, the EtOH was removed under reduced pressure and the residue was dissolved in 1 N HCl (50 mL). The acid solution was washed with EtOAc (50 mL) and then brought to pH 10 using 10 N aqueous NaOH (5 mL). The basic solution was extracted with CH_2Cl_2 (3 x 50 mL) and the combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to yield the protected amine (±)-**114** (5.8 g, 90% yield) as an opaque pale yellow oil: IR (thin film/NaCl) 3330 (br w), 2950 (m),

2835 (m), 1733 (s), 1592 (w), 1515 (s), 1454 (m), 1264 (s), 1237 (s), 1158 (s), 1029 (s), 764 (m), 701 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.27 (m, 2H), 7.17 (m, 3H), 6.92-6.81 (comp m, 3H), 3.90 (s, 3H), 3.89 (s, 3H), 3.78 (d, J = 12.8 Hz, 1H), 3.73 (s, 3H), 3.59 (d, J = 12.8 Hz, 1H), 3.30 (dd, J = 6.0, 7.4 Hz, 1H), 1.53 (m, 2H), 2.04-1.88 (comp m, 2H), 1.80 (br s, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 175.7, 149.1, 148.3, 141.3, 132.5, 128.4, 128.3, 125.9, 120.4, 111.8, 111.3, 60.0, 55.9, 55.8, 51.8, 51.6, 35.0, 32.0; high resolution mass spectrum (EI) *m/z* 343.1778 [calc'd for $C_{20}H_{25}NO_4$ (M+) 343.1784].

Preparation of Amide (±)-115.

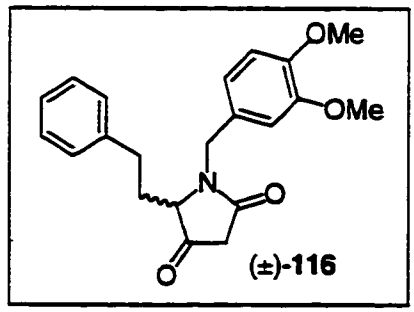


(±)-115

Amide (±)-115: A three-necked flask equipped with an addition funnel was charged with the amine (±)-**114** (4.5 g, 13.1 mmol, 1.0 equiv), ethyl hydrogen malonate (1.7 g, 13.1 mmol, 1.0 equiv), and CH_2Cl_2 (80 mL). The flask was cooled to 0°C and 1,3-dicyclohexylcarbodiimide (2.7 g, 13.1 mmol, 1.0 equiv) in CH_2Cl_2 (50 mL) was added dropwise through the addition funnel over a 15 min period. The ice bath was removed and the mixture was stirred for an additional 3 h at room temperature. The mixture was filtered to remove the urea by-product and the filtrate was washed with H_2O (100 mL). The organic layer was separated, and

the aqueous layer was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and concentrated to an oily residue. The residue was taken up in a minimum of acetone (10 mL) and filtered to remove the remaining urea by-product. Removal of the acetone under reduced pressure afforded amide (±)-**115** (5.7 g, 95% yield) as a clear, light yellow oil: IR (thin film/NaCl) 2938 (br m), 2838 (w), 1739 (s), 1651 (s), 1516 (s), 1455 (s), 1418 (s), 1262 (s), 1158 (s), 1094 (w), 1028 (s), 810 (w), 767 (w), 702 (m) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.32-7.24 (comp m, 3H), 7.18 (m, 2H), 6.80-6.76 (comp m, 2H), 6.61 (m, 1H), 4.39 (d, *J* = 16.6 Hz, 1H), 4.28 (dd, *J* = 5.4, 9.5 Hz, 1H), 4.21 (q, *J* = 7, 1 Hz, 2H), 3.87 (s, 3H), 3.83 (s, 3H), 3.72 (d, *J* = 16.6 Hz, 1H), 3.69 (s, 3H), 3.47-3.37 (comp m, 3H), 3.28 (m, 1H), 1.30 (t, *J* = 7.1 Hz, 3H), 1.30-1.26 (comp m, 2H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.0, 170.7, 167.5, 167.3, 167.2, 149.3, 148.7, 148.3, 140.9, 139.9, 130.2, 128.6, 128.4, 128.3, 128.3, 126.4, 126.0, 120.5, 119.1, 111.7, 111.2, 110.8, 110.1, 61.5, 59.7, 58.1, 55.9, 55.9, 52.3, 52.1, 51.2, 46.5, 41.6, 41.5, 41.3, 32.6, 32.0, 31.1, 30.9, 14.1; high resolution mass spectrum (EI) *m/z* 457.2108 [calc'd for $C_{25}H_{31}NO_7$ (M+) 457.2101].

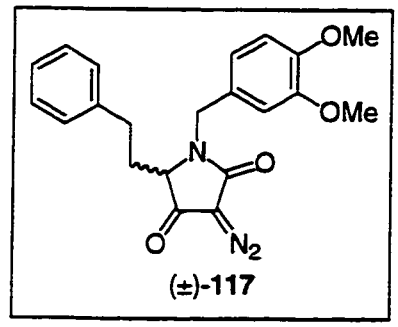
Preparation of Lactam (±)-116.



(±)-**116**

Lactam (±)-116. A three-necked flask was charged with EtOH (15 mL). Sodium metal (270 mg, 11.7 mmol, 0.94 equiv) was then carefully added in one portion. When the sodium had completely reacted, ester (±)-**115** (5.7 g, 12.5 mmol, 1.0 equiv) was added dropwise to the mixture as a solution in EtOH (15 mL). The mixture was brought to reflux for 30 min and then allowed to cool to rt. The EtOH was removed under reduced pressure and the residue was dissolved in H_2O (50 mL). The aqueous layer was washed with EtOAc (50 mL), and acidified to a pH of 2 with 2 N HCl (20 mL). The acidic solution was extracted with CH_2Cl_2 (3 x 50 mL). The combined organic layers were dried over anhydrous $MgSO_4$, filtered, and concentrated to furnish a yellow oil. A suspension of the oil in CH_3CN (60 mL) and H_2O (0.1 mL) was warmed to reflux open to the air for three hours. The mixture was cooled to rt and the CH_3CN/H_2O mixture was removed at reduced pressure to yield lactam (±)-**116** as a dark yellow oil (2.0 g, 45% yield) which solidified upon standing to a dark yellow glassy solid: IR (thin film/NaCl) 2934 (br m), 2837 (w), 1769 (m), 1693 (s), 1593 (w), 1516 (s), 1455 (m), 1415 (m), 1261 (s), 1238 (s), 1154 (m), 1141 (m), 1027 (m), 737 (w), 701 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.25 (comp m, 3H), 7.08 (d, J = 7.1 Hz, 2H), 6.78 (comp m, 3H), 5.16 (d, J = 14.6 Hz, 1H), 3.99 (d, J = 14.6 Hz, 1H), 3.87 (s, 3H), 3.85 (s, 3H), 3.83 (m, 1H), 3.00 (m, 2H), 2.62 (m, 1H), 2.47 (m, 1H), 2.25 (m, 1H), 1.99 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 206.4, 168.7, 149.2, 148.7, 139.7, 128.3, 128.2, 127.6, 126.2, 120.7, 111.4, 110.9, 65.0, 55.7, 55.6, 43.6, 41.0, 30.1, 29.5; high resolution mass spectrum (EI) *m/z* 353.1636 [calc'd for $C_{21}H_{23}NO_4$ (M+) 353.1627].

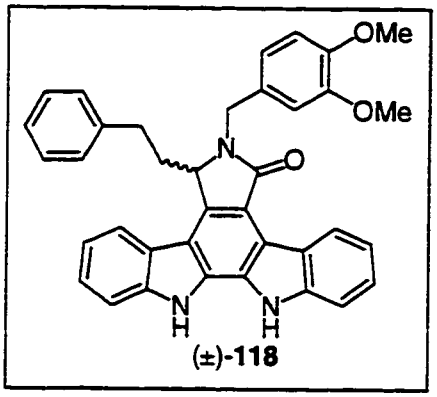
Preparation of Diazo lactam (±)-117.



(±)-117

Diazo lactam (±)-117. A stirred solution of lactam (±)-**116** (1.9 g, 5.4 mmol, 1.0 equiv), *p*-ABSA (1.5 g, 5.9 mmol, 1.1 equiv), and CH_3CN (40 mL) was cooled to 0°C and Et_3N (2.3 mL, 16.2 mmol, 3.0 equiv) was added dropwise to the mixture. After gradually warming to rt, the mixture was stirred for an additional 3 h and the volatiles were removed under reduced pressure. The residue was subjected to flash chromatography (50% EtOAc/hexanes eluent) to provide diazo lactam (±)-**117** (1.6 g, 78% yield) as a bright yellow foam: IR (thin film/NaCl) 3025 (w), 2932 (br m), 2936 (w), 2124 (s), 1684 (s), 1593 (w), 1515 (m), 1403 (m), 1358 (m), 1260 (m), 1237 (m), 1138 (m), 749 (w) cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 7.28 (t, *J* = 7.4 Hz, 1H), 7.21 (t, *J* = 7.4 Hz, 1H), 7.09 (d, *J* = 7.2 Hz, 1H), 6.82-6.75 (comp m, 5H), 5.03 (d, *J* = 14.9 Hz, 1H), 4.05 (d, *J* = 14.9 Hz, 1H), 3.88 (s, 3H), 3.86 (s, 3H), 3.84 (dd, *J* = 2.9, 6.2 Hz, 1H), 2.58 (m, 2H), 2.27 (m, 1H), 2.04 (m, 1H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 188.9, 161.9, 149.5, 149.0, 140.2, 128.4, 128.3, 128.0, 126.3, 120.8, 111.4, 111.2, 65.6, 63.1, 56.0, 55.9, 44.5, 30.3, 29.3; high resolution mass spectrum (EI) *m/z* 379.1538 [calc'd for $C_{21}H_{21}N_3O_4$ (M+) 379.1532].

Preparation of Aglycon (±)-118.

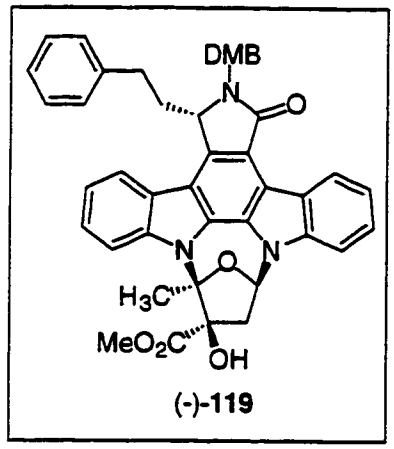


(±)-**118**

Aglycon (±)-118. To a three-necked flask equipped with a condenser were added the diazo lactam (±)-**117** (400 mg, 1.1 mmol, 1.0 equiv), 2,2'-biindole (245 mg, 1.1 mmol, 1.0 equiv), $Rh_2(OAc)_4$ (5.0 mg, 0.01 mmol, 0.01 equiv) and pinacolone (11 mL). The whole was degassed by bubbling a stream of N_2 through the solution for 2 h. The mixture was then warmed to reflux for an additional 8 h. The reaction mixture was concentrated to a dark brown foamy solid, which was adsorbed onto silica gel and subjected to flash chromatography (50% EtOAc/hexanes eluent), affording unreacted **11** (130 mg, 53% yield) as a yellow powder, and aglycon (±)-**118** (134 mg, 22% yield; 48% based on recovered **11**) was isolated as a pale yellow solid: mp >330 °C (dec.); IR (thin film/NaCl) 3325 (br m), 3058 (w), 2930 (w), 1641 (s), 1569 (m), 1514 (s), 1455 (s), 1412 (s), 1326 (s), 1258 (s), 1237 (s), 1139 (m), 1027 (m), 745 (s) cm^{-1}; ^1H NMR (500 MHz, DMSO-d_6) δ 11.59 (s, 1H), 11.35 (s, 1H), 9.31 (d, J = 7.9 Hz, 1H), 8.09 (d, J = 7.9 Hz, 1H), 7.79 (d, J = 8.1 Hz, 1H), 7.75 (d, J = 8.1 Hz, 1H), 7.45 (t, J = 7.5 Hz, 2H), 7.25 (q, J = 7.7 Hz, 2H), 7.11 (s, 1H), 7.06-6.93 (comp m, 5H), 6.70 (d, J = 7.1 Hz, 2H), 5.37 (s, 1H), 5.14 (d, J = 15.3 Hz, 1H), 4.56 (d, J = 15.2 Hz, 1H), 3.72 (s, 6H), 2.63 (m, 2H), 2.14 (m, 1H), 1.60 (m, 1H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 169.5, 148.1,

141.0, 139.4, 139.2, 133.8, 130.9, 128.2, 127.9, 127.7, 125.6, 125.4, 125.1, 124.9, 122.6, 121.8, 121.6, 120.1, 119.8, 118.9, 118.6, 115.3, 113.4, 112.2, 111.9, 111.3, 58.9, 55.6, 43.4, 31.0, 27.8; high resolution mass spectrum (FAB) *m/z* 566.2450 [calc'd for $C_{37}H_{32}N_3O_3$ (M+H) 566.2444].

Preparation of Indolocarbazole (-)-119.

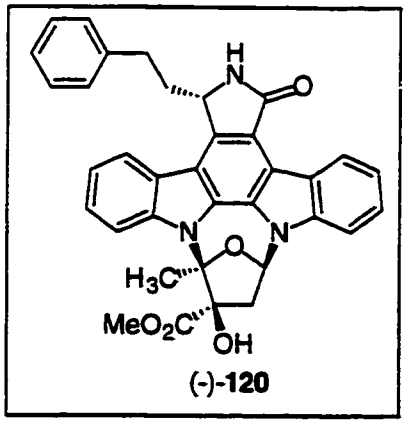


(-)-119

Indolocarbazole (-)-119. To a refluxing solution of aglycon (±)-**118** (130 mg, 0.24 mmol, 1.0 equiv) and camphorsulfonic acid (5.0 mg, 0.024 mmol, 0.1 equiv) in 1,2-dichloroethane (8 mL) was added, *via* addition funnel over 24 h, a solution of alcohols(-)-**16a-c**/(+)-**17** (100 mg, 0.47 mmol, 2 equiv) in 1,2-dichloroethane (4 mL). Reflux was then continued for an additional 48 h. The crude reaction mixture was washed with 10% aqueous $NaHCO_3$ (5 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 10 mL) and the combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was taken up in a minimum of CH_2Cl_2 and purified by flash chromatography (50% EtOAc/hexanes eluent) to provide a 2:2:1:1 mixture of indolocarbazoles (143 mg, 83% yield). Separation of the mixture was achieved using HPLC (190:10:1

CH_2Cl_2:EtOAc:MeOH eluent) to provide the desired (-)-**119** (33 mg, 19% yield) as a white film: $[\alpha]_D^{20}$ -9.44° (*c* 0.36, MeOH); IR (thin film/NaCl) 3343 (br w), 2926 (w), 2853 (w), 1733 (m), 1669 (m), 1586 (w), 1510 (m), 1451 (s), 1394 (m), 1313 (w), 1252 (s), 1142 (m), 1025 (m), 749 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.54 (d, *J* = 8.0 Hz, 1H), 8.09 (d, *J* = 7.8 Hz, 1H), 8.00 (d, *J* = 8.5 Hz, 1H), 7.82 (d, *J* = 8.3 Hz, 1H), 7.52 (t, *J* = 7.7 Hz, 1H), 7.42 (t, *J* = 7.6 Hz, 1H), 7.34 (t, *J* = 7.6 Hz, 1H), 7.27 (t, *J* = 7.5 Hz, 1H), 7.19-6.77 (comp m, 9H), 5.44 (t, *J* = 3.5 Hz, 1H), 5.34 (d, *J* = 15.3 Hz, 1H), 5.25 (s, 1H), 4.57 (d, *J* = 15.2 Hz, 1H), 4.01 (s, 3H), 3.78 (s, 3H), 3.75 (s, 3H), 3.50 (dd, *J* = 7.4, 14.1 Hz, 1H), 2.77 (m, 2H), 2.33 (m, 1H), 2.24 (s, 3H), 2.22 (m, 1H), 1.88 (m, 1H); ^{13}C NMR (125 MHz, acetone-d_6) δ 173.9, 170.3, 150.8, 150.0, 142.4, 141.5, 138.4, 135.0, 132.2, 130.2, 128.9, 128.8, 127.5, 127.4, 126.5, 126.4, 125.8, 125.7, 125.2, 124.4, 122.7, 121.3, 121.1, 120.6, 117.5, 116.0, 115.6, 113.4, 113.2, 109.3, 100.3, 86.5, 86.2, 60.0, 56.3, 56.2, 53.4, 44.6, 43.4, 43.4, 23.5; high resolution mass spectrum (FAB) *m/z* 722.2857 [calc'd for $C_{44}H_{40}N_3O_7$ (M+H) 722.2866].

Preparation of (-)-7-(*S*)-Phenethyl K252a 120.



(-)-**120**

(-)-7-(*S*)-Phenethyl K252a 120. A solution of protected amide (-)-**119** (10 mg, 0.014 mmol, 1.0 equiv) and anisole (300 mg, 2.8 mmol, 200 equiv) was treated with TFA (1 mL). After stirring at rt for 12 h, the reaction was diluted with CH_2Cl_2 (5 mL) and quenched with 20% aqueous $NaHCO_3$ solution (2 mL). The aqueous layer was washed with CH_2Cl_2 (3 x 5 mL) and the combined organic layers were dried over Na_2SO_4, filtered and adsorbed onto SiO_2. Flash chromatography (50% EtOAc/hexanes eluent) afforded (-)-**120** (2.0 mg, 25% yield) as a white film: $[\alpha]_D^{20}$ -39.09° (*c* 0.22, $CHCl_3$); IR (thin film/NaCl) 3254 (br w), 2950 (w), 2924 (m), 2854 (w), 1731 (m), 1677 (s), 1583 (m), 1453 (s), 1392 (m), 1314 (m), 1257 (m), 1138 (m), 1081 (w), 743 (s) cm^{-1}; ^1H NMR (500 MHz, acetone-d_6) δ 9.41 (d, *J* = 7.8 Hz, 1H), 7.99 (m, 2H), 7.86 (d, *J* = 7.8 Hz, 1H), 7.79 (d, *J* = 8.2 Hz, 1H), 7.49 (ddd, *J* = 1.0, 7.1, 8.1 Hz, 1H), 7.43 (ddd, *J* = 1.1, 7.3, 8.4 Hz, 1H), 7.33-7.17 (comp m, 7H), 7.14 (dd, *J* = 4.9, 7.4 Hz, 1H), 5.35 (d, *J* = 8.2 Hz, 1H), 5.26 (s, 1H), 4.01 (s, 3H), 3.49 (dd, *J* = 7.4, 14.1 Hz, 1H), 3.03 (m, 1H), 2.87 (m, 2H), 2.22 (m, 4H); ^{13}C NMR (125 MHz, acetone-d_6) δ 174.0, 172.1, 142.4, 141.2, 138.2, 137.9, 129.4, 129.1, 127.2, 126.7, 126.3, 125.6, 125.4, 125.1, 124.2, 122.5, 121.1, 120.3, 117.4, 115.8, 115.4, 109.1, 99.8, 86.3, 86.0, 56.7, 53.3, 43.2, 37.6, 32.6, 29.1, 23.2; high resolution mass spectrum (FAB) *m/z* 571.1130 [calc'd for $C_{35}H_{30}N_3O_5$ (M+H) 571.1126].

3.7 Notes and References.

(1) Shokat, K. M. *Chem. Biol.* **1995**, *2*, 509.

(2) Shah, K.; Liu, Y. Deirmengian, C. Shokat, K. M. *Proc. Natl. Sci. USA* **1997**, *94*, 3565.

(3) Liu, Y.; Shah, K.; Yang, F.; Witucki, L.; Shokat, K. M. *Chem. Biol.* **1998**, *5*, 91.

(4) Liu, Y.; Shah, K.; Yang, F.; Witucki, L.; Shokat, K. M. *Bioorg. Med. Chem. Lett.* **1998**, *6* 1219.

(5) Bishop, A. C.; Shah, K.; Liu, Y.; Witucki, L.; Kung, C.; Shokat, K. M. *Curr. Biol.* **1998**, *8*, 257.

(6) Bishop, A. C.; Kung, C.; Shah, K.; Witucki, L.; Shokat, K. M.; Liu, Y. *J. Am. Chem. Soc.* **1999**, *121*, 627.

(7) The author is indebted to Anthony Bishop and Kevan Shokat for both the mutant and wild type kinase assays as well as helpful discussions and suggestions.

(8) The kinase inhibition assays were performed as follows: IC_{50}'s for putative kinase inhibitors were determined by measuring the counts per minute (cpm) of ^{32}P transferred to a peptide substrate for that particular kinase. Various

concentrations of inhibitor were incubated with 50 mM Tris (pH 8.0), 10 mM $MgCl_2$, 1.6 mM glutathione, 1 mg/mL of BSA, 100 μM peptide, 3.3% DMSO, the appropriate kinase, and 11 nM (2 μCi) [γ-^{32}P]ATP (6000 Ci/mmol, NEN) in a total volume of 30 μL for 30 min. Reaction mixtures (25 μL) were spotted onto a phosphocellulose disk, immersed in 10% HOAc and washed with 0.5% H_3PO_4. The transfer of ^{32}P was measured by standard scintillation counting. IC_{50} was defined to be the concentration of inhibitor at which the cpm was 50% of the control disk. When the IC_{50} fell between two measured concentrations, it was calculated based on the assumption of an inversely proportional relationship between inhibitor concentration and cpm between the two data points.

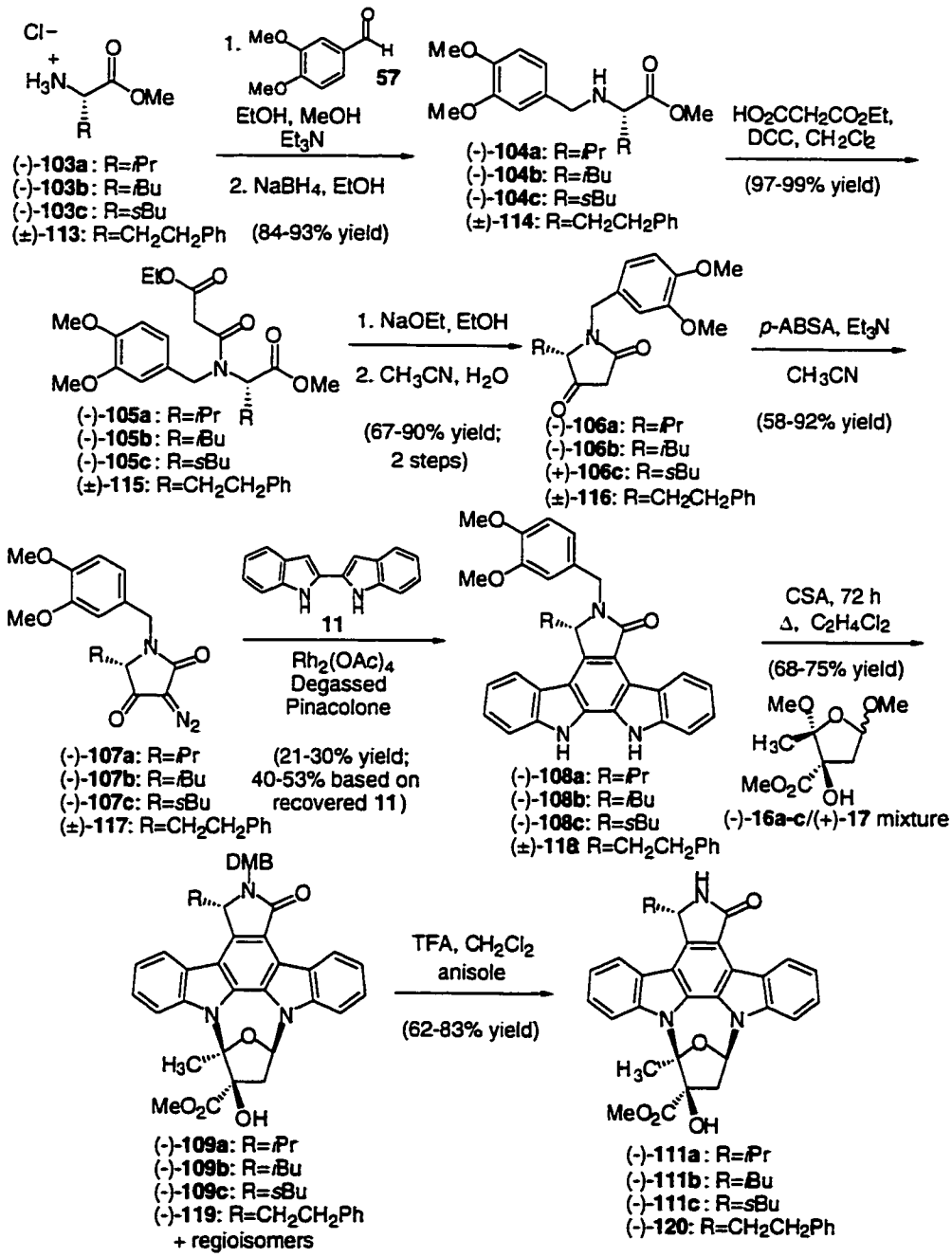
(9) For protein tyrosine kinase assays of (-)-**52a** and (+)-**80a** see Chapter 2.

(10) Stereochemistry at the C(7) position was determined as in the benzyl case (see Chapter 2; Figure 2.3.1).

(11) Still, W. C.; Kahn, M.; Nitra, A. *J. Org. Chem.* **1978**, *43*, 2923.

APPENDIX THREE: SYNTHETIC SUMMARY FOR ISOPROPYL, ISOBUTYL, SECBUTYL AND PHENETHYL K252a

Figure A.3.1 The Synthesis of (-)-**111a-c** and (-)-**120**.



APPENDIX FOUR: SPECTRA RELEVANT TO CHAPTER THREE

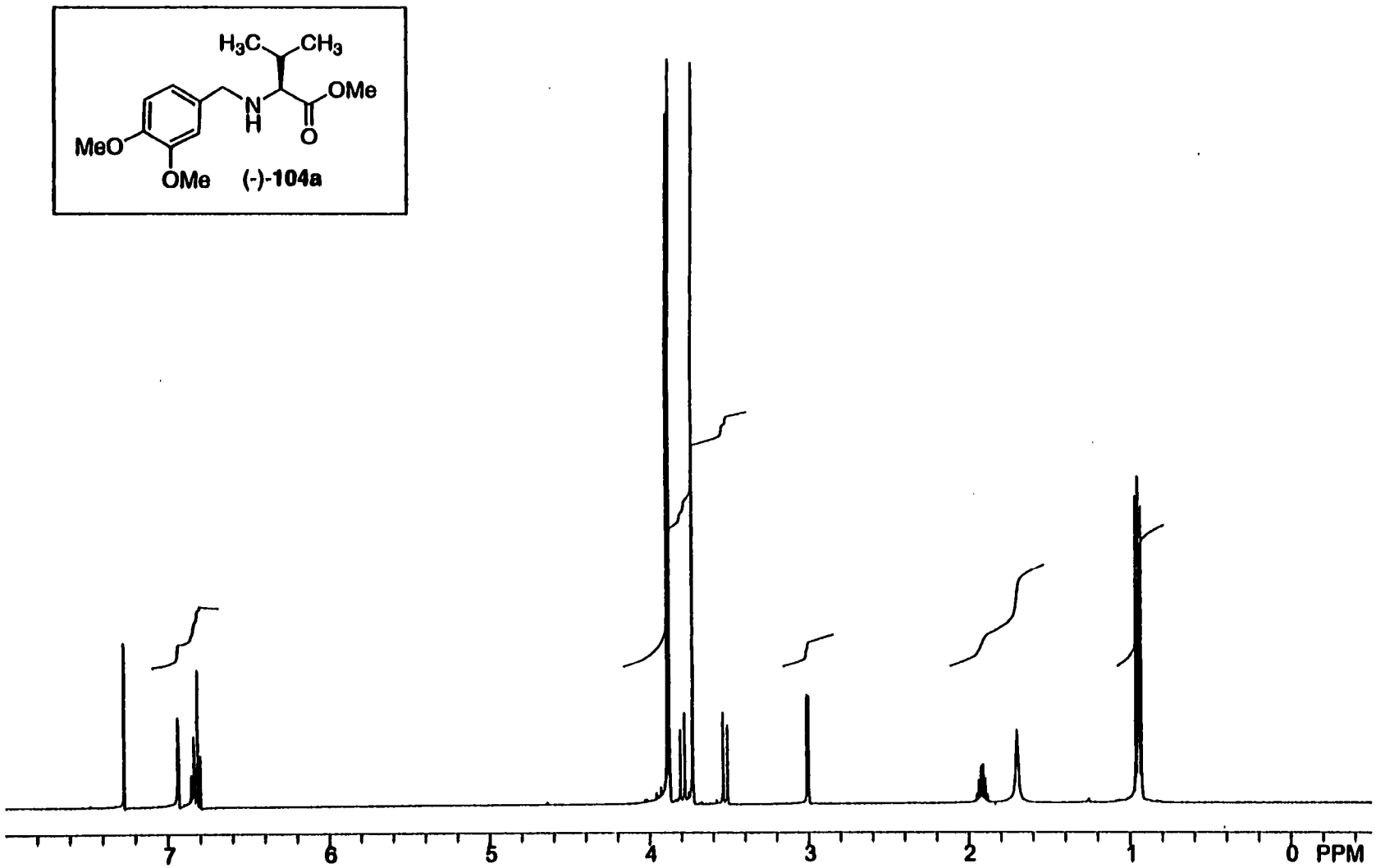


Figure A.4.1 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**104a**.

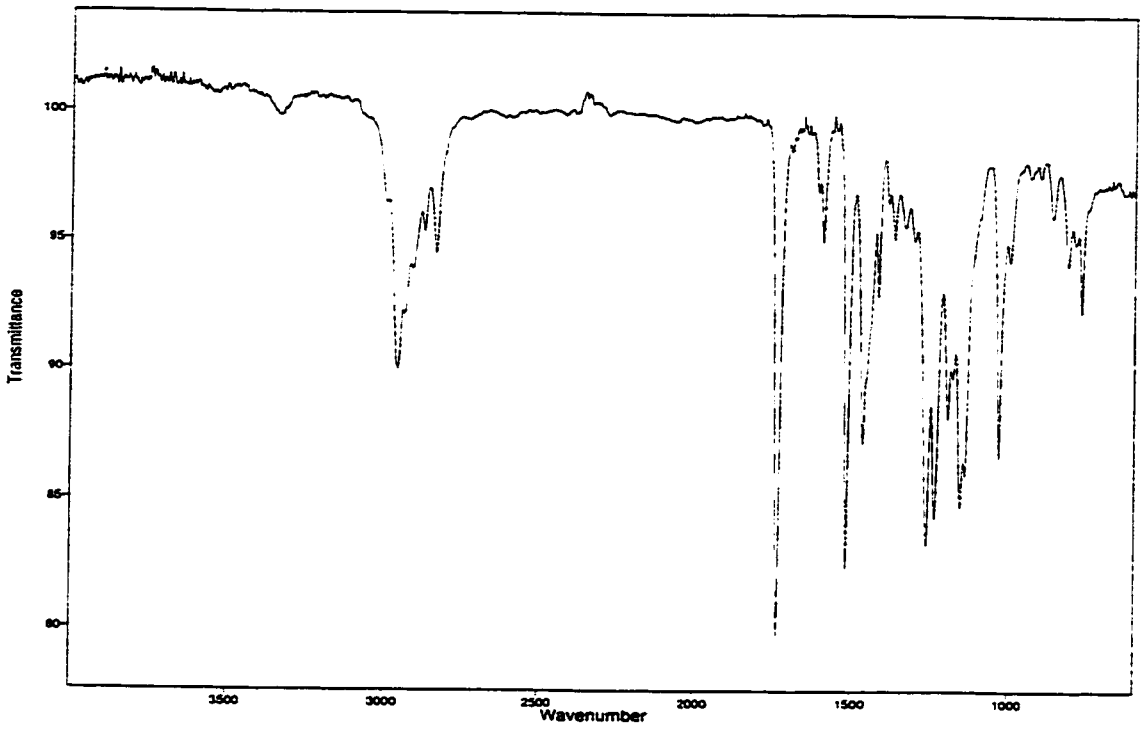


Figure A.4.2 Infrared Spectrum (thin film/NaCl) of compound (-)-**104a**.

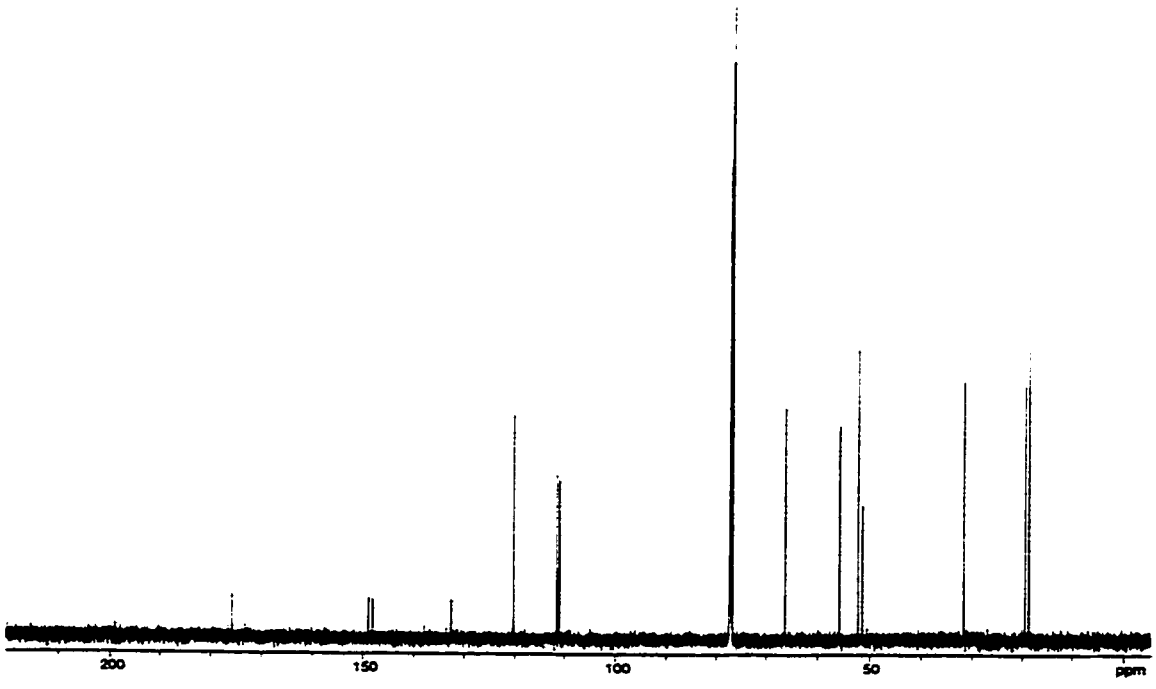


Figure A.4.3 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**104a**.

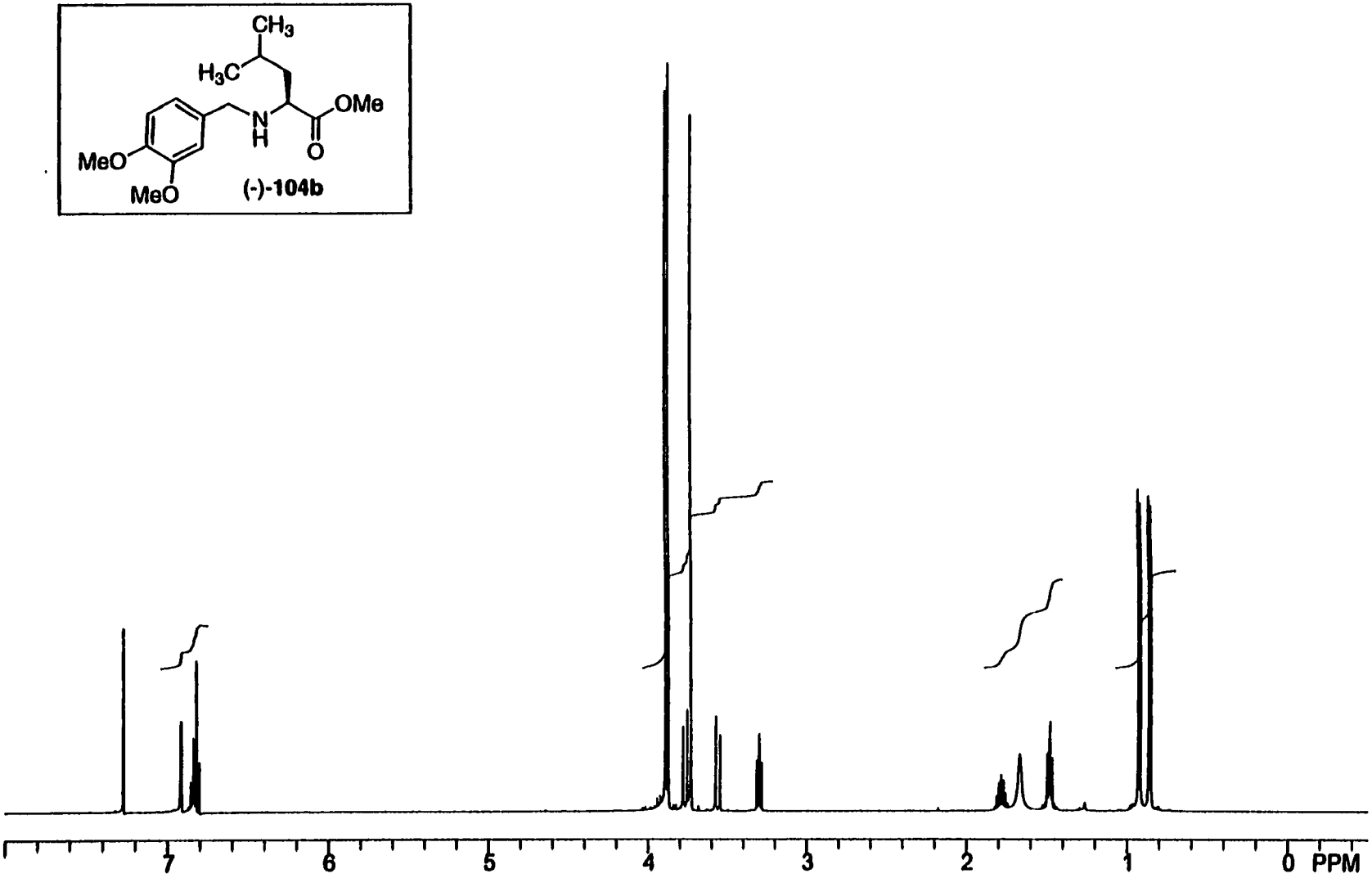


Figure A.4.4 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**104b**.

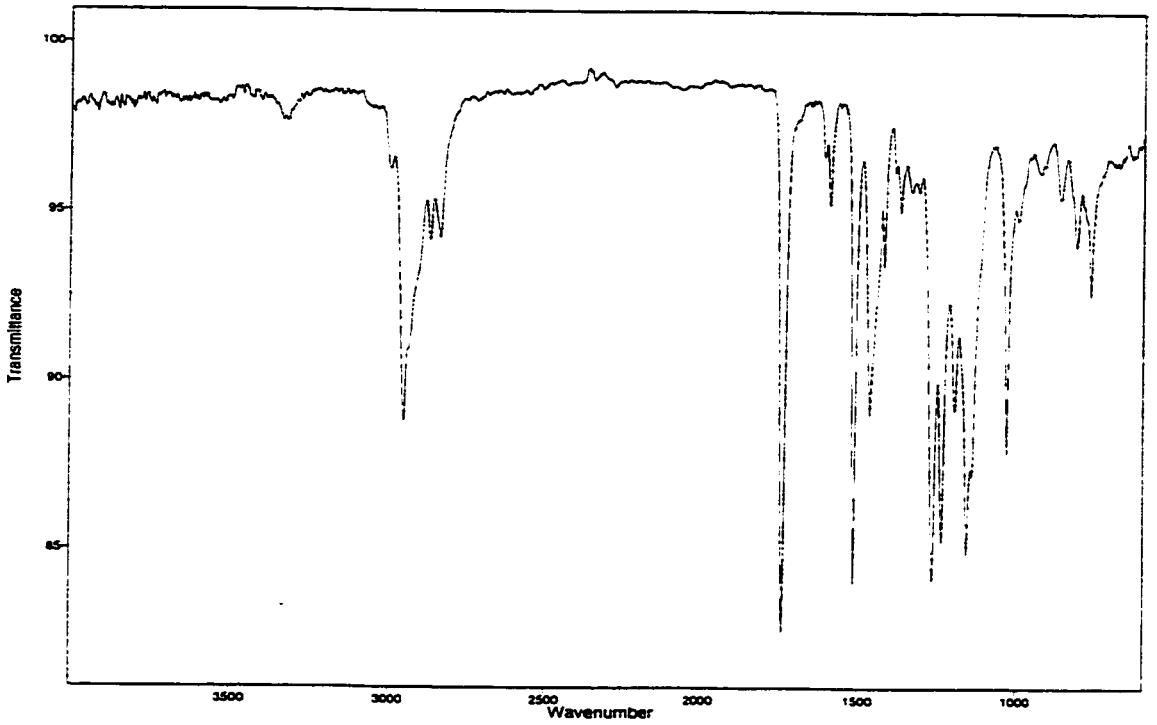


Figure A.4.5 Infrared Spectrum (thin film/NaCl) of compound (-)-**104b**.

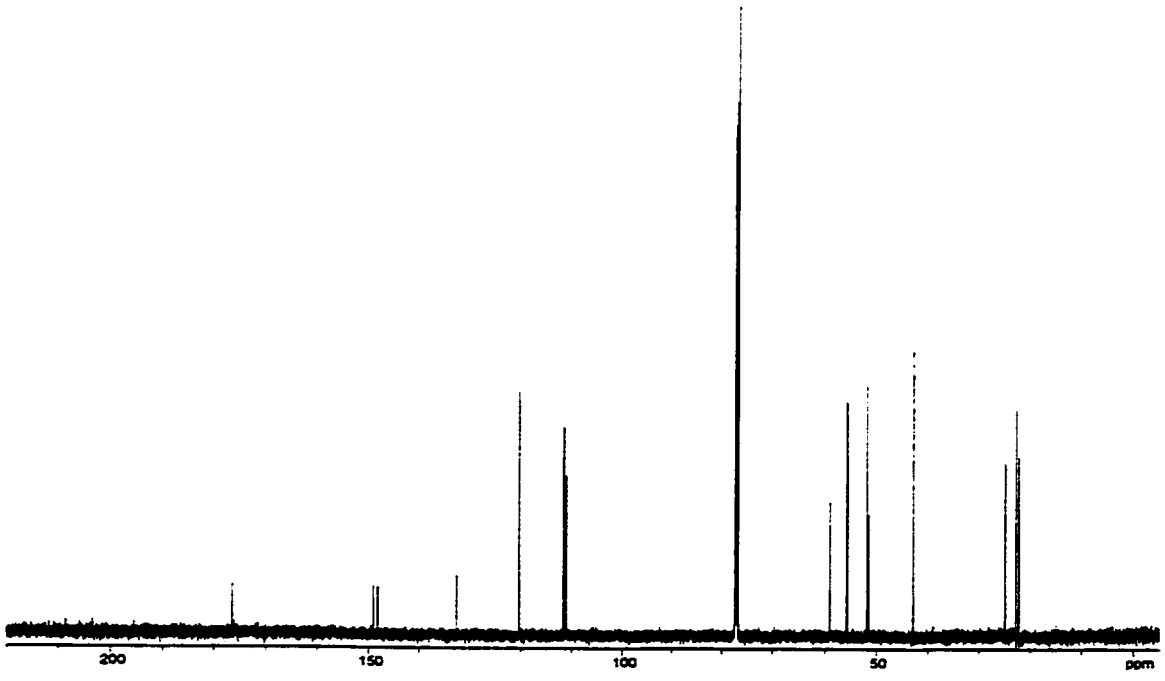


Figure A.4.6 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**104b**.

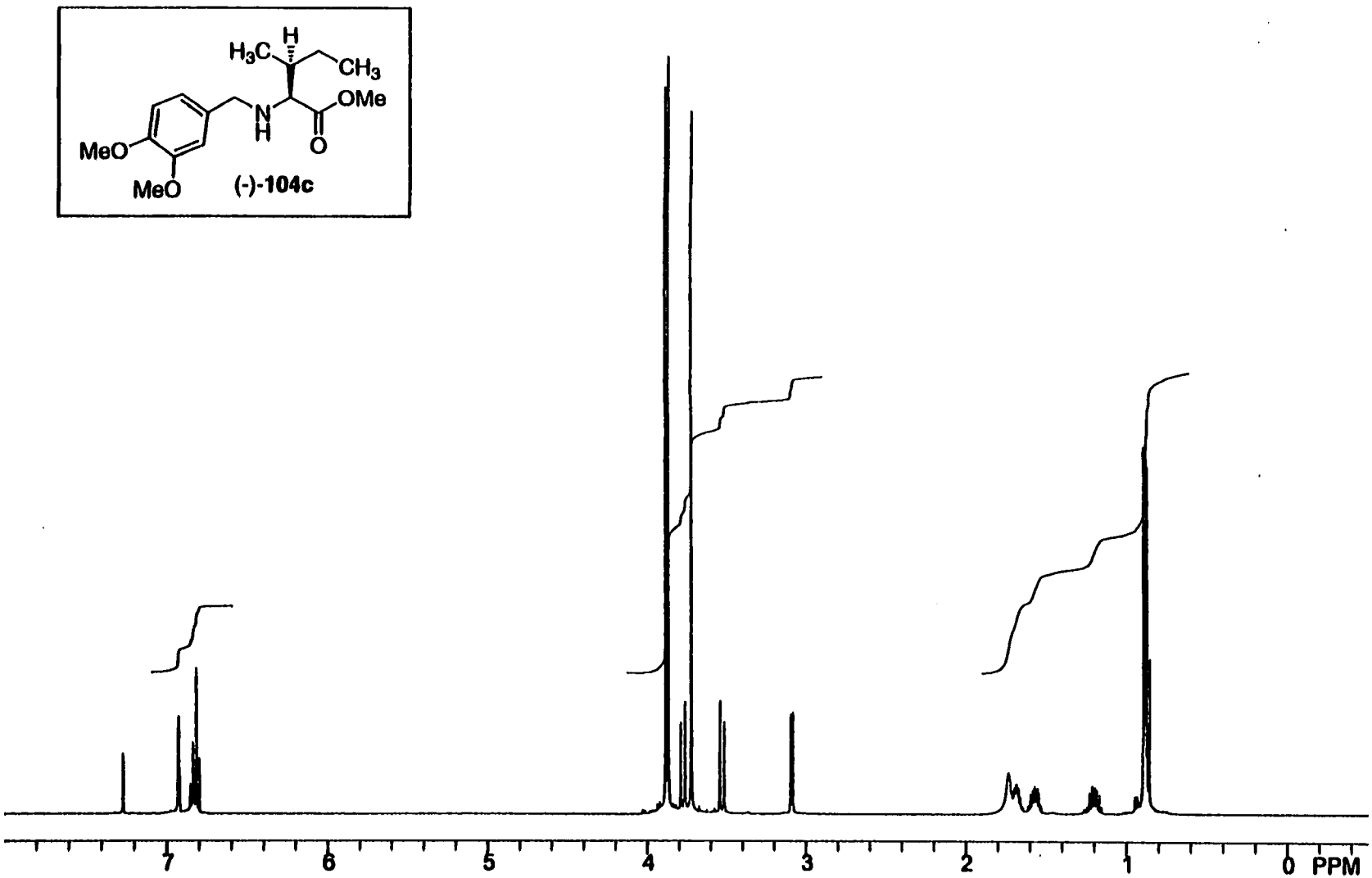


Figure A.4.7 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**104c**.

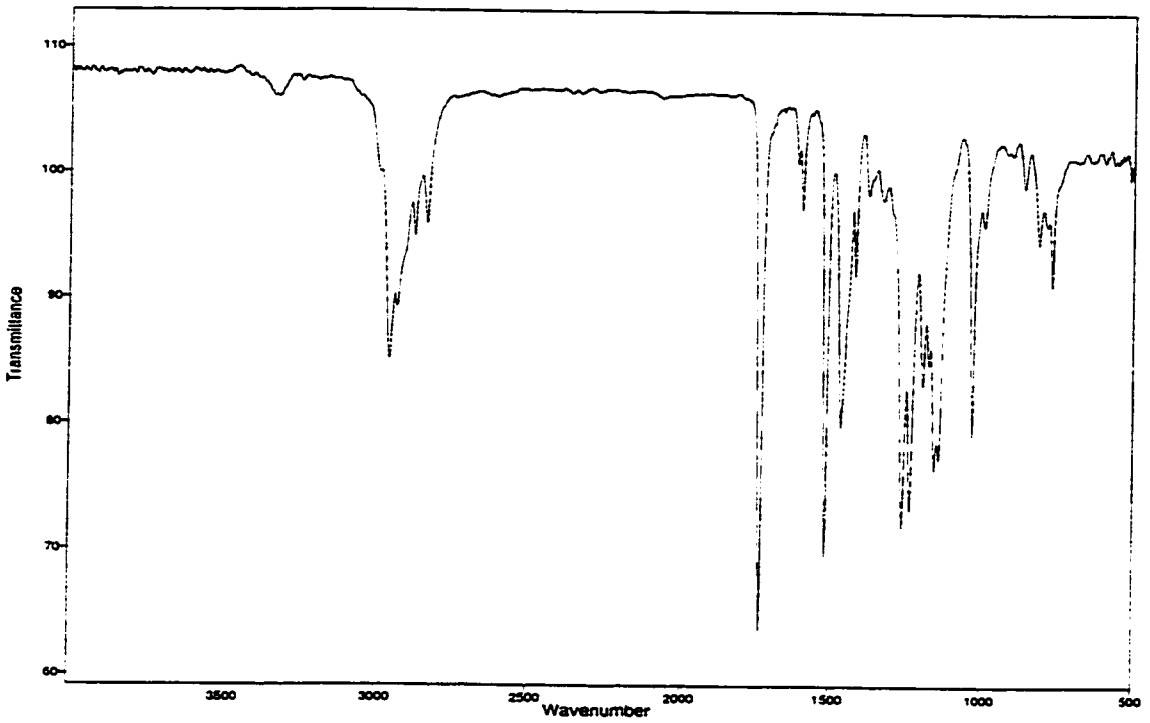


Figure A.4.8 Infrared Spectrum (thin film/NaCl) of compound (-)-**104c**.

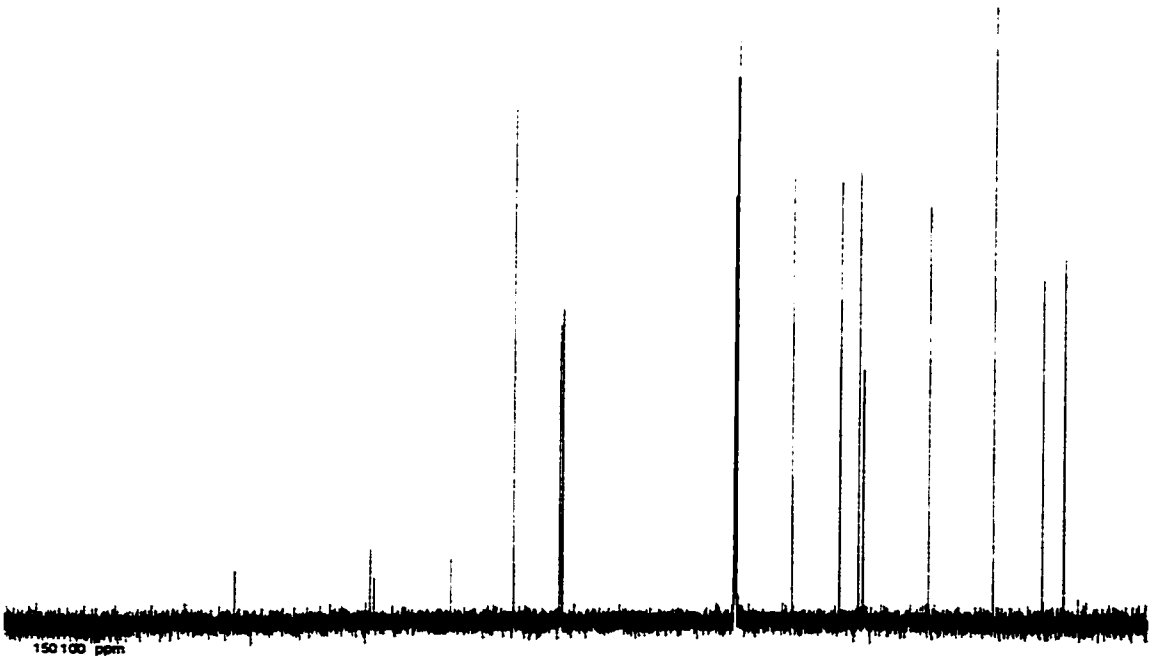


Figure A.4.9 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**104c**.

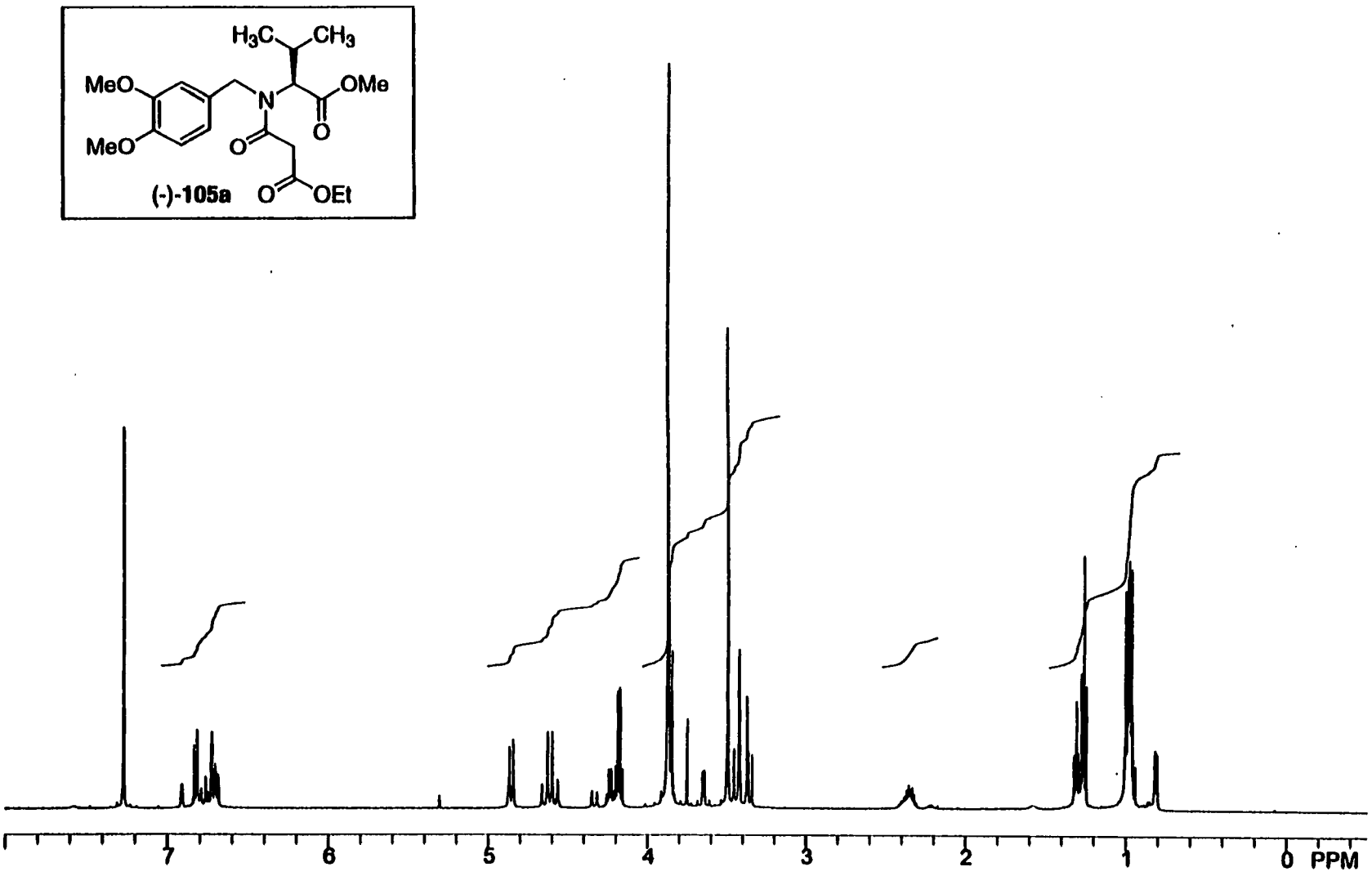


Figure A.4.10 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**105a**.

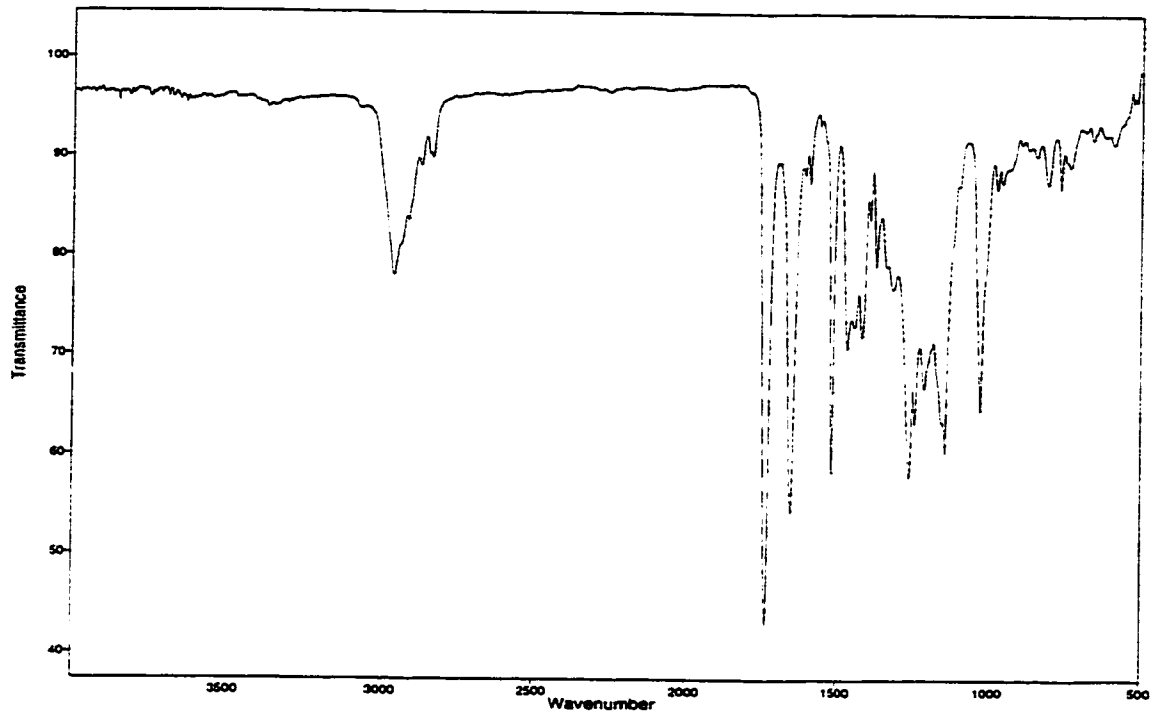


Figure A.4.11 Infrared Spectrum (thin film/NaCl) of compound (-)-**105a.**

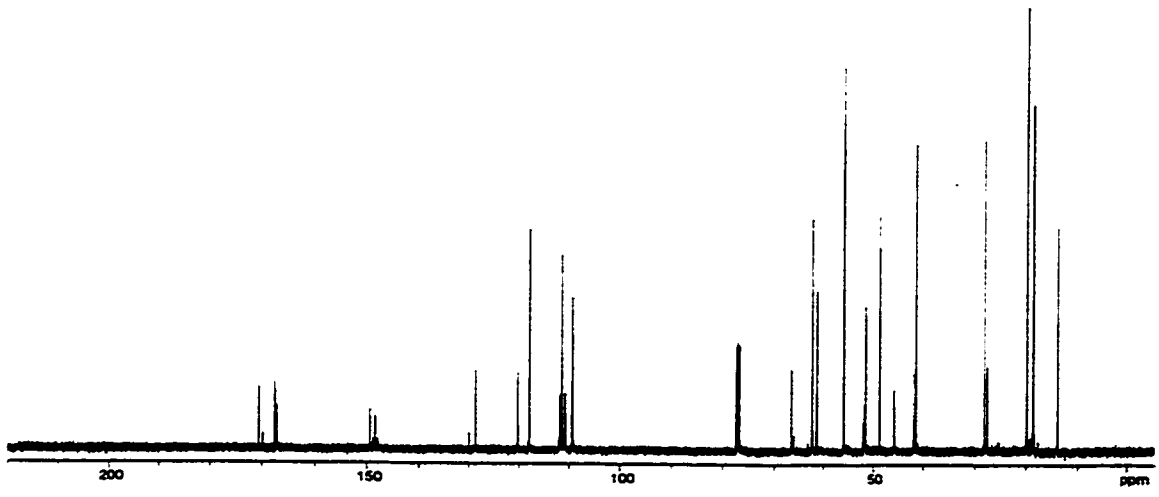


Figure A.4.12 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**105a.**

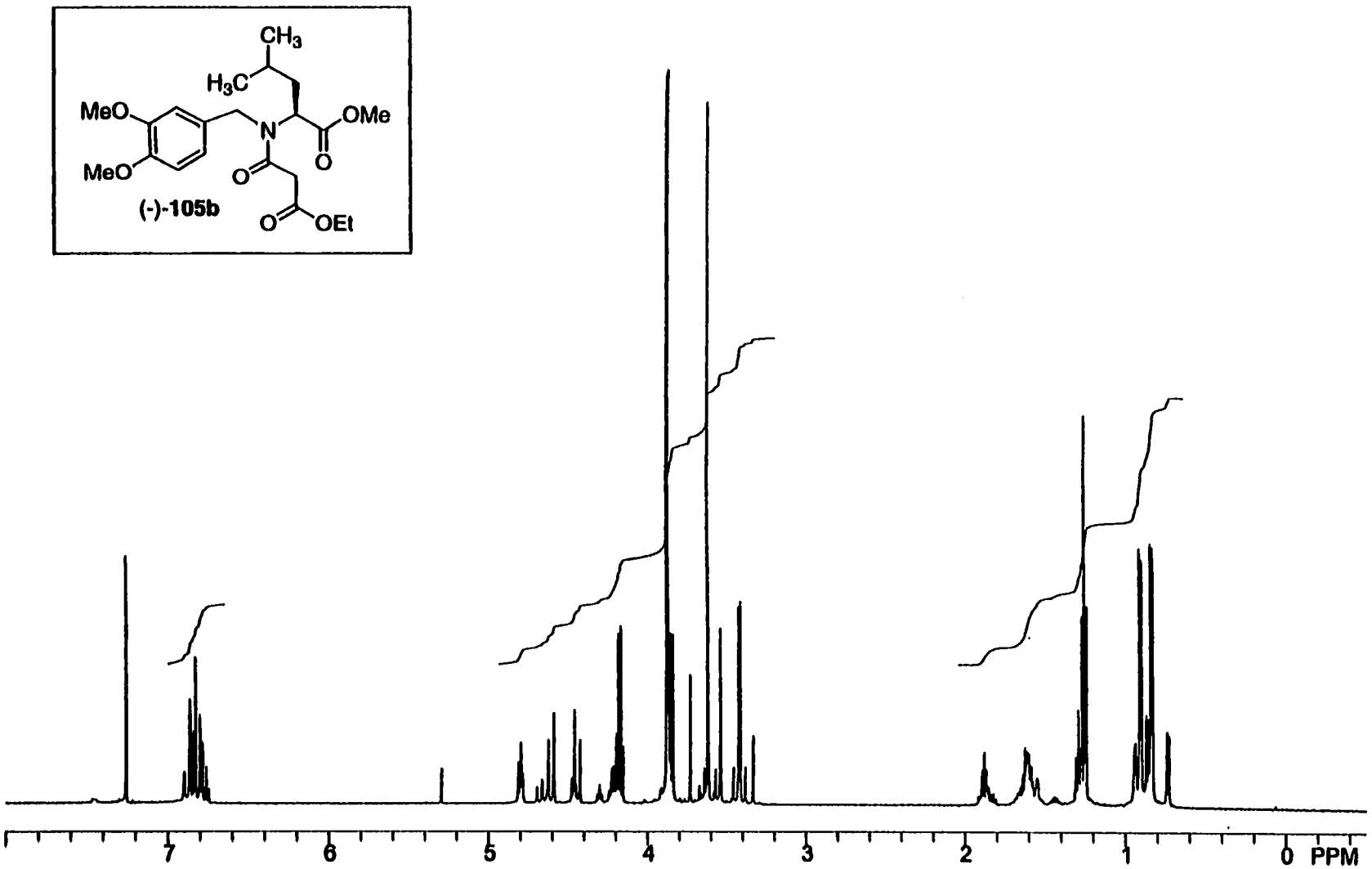


Figure A.4.13 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**105b**.

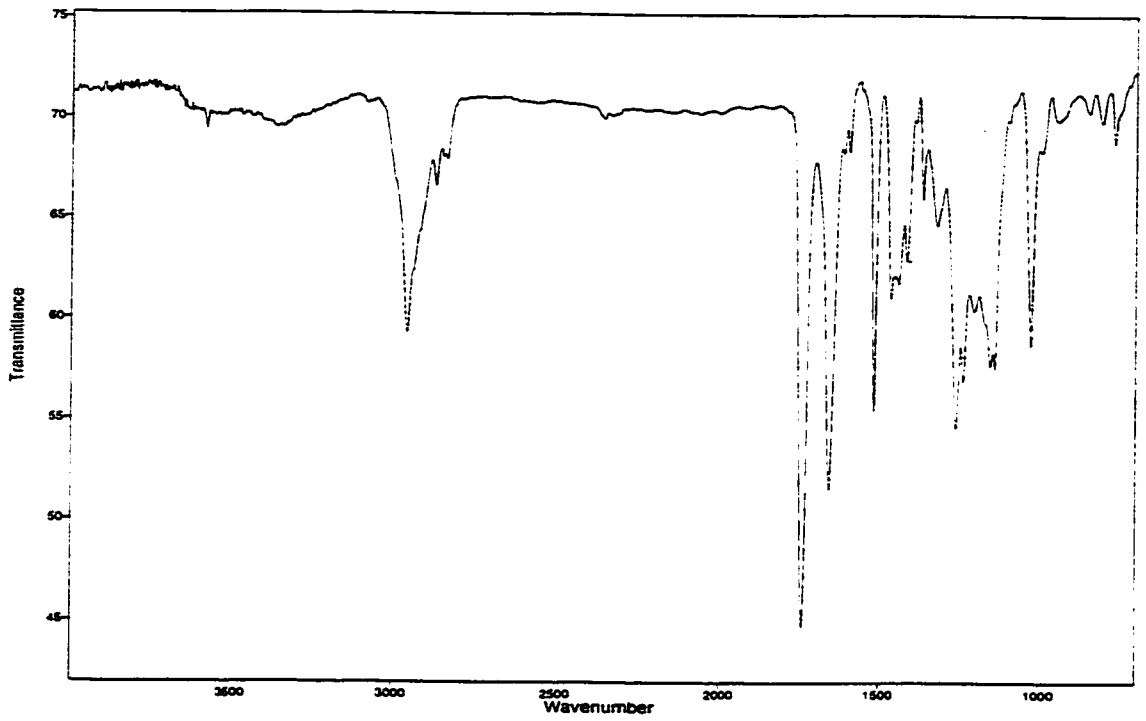


Figure A.4.14 Infrared Spectrum (thin film/NaCl) of compound (-)-**105b**.

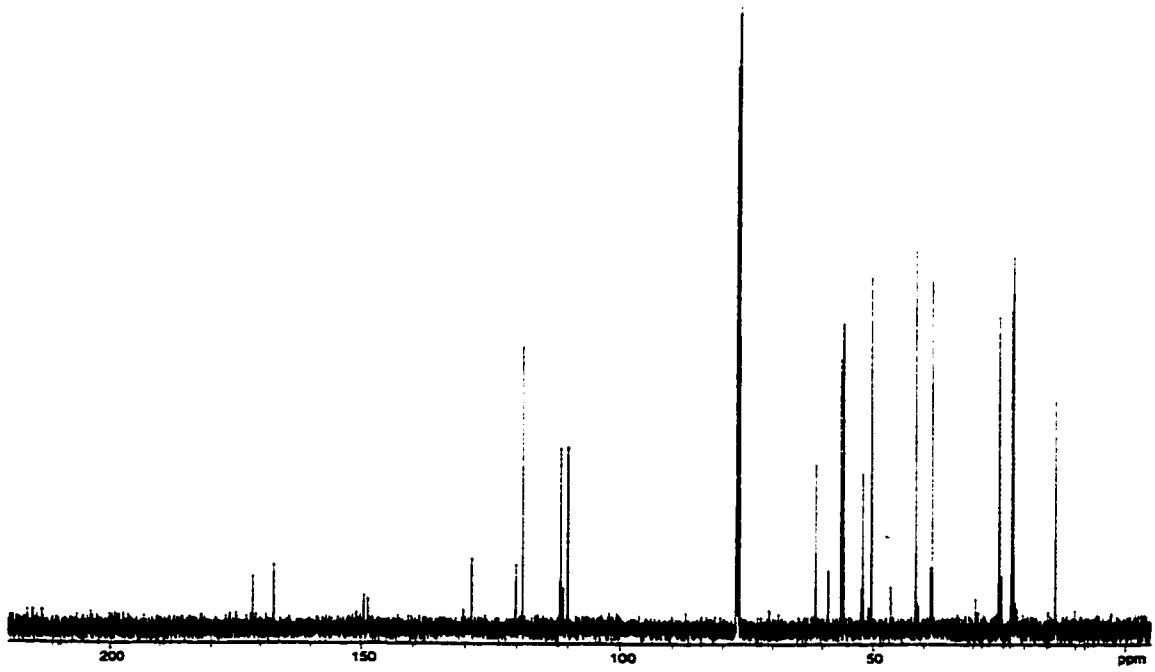


Figure A.4.15 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**105b**.

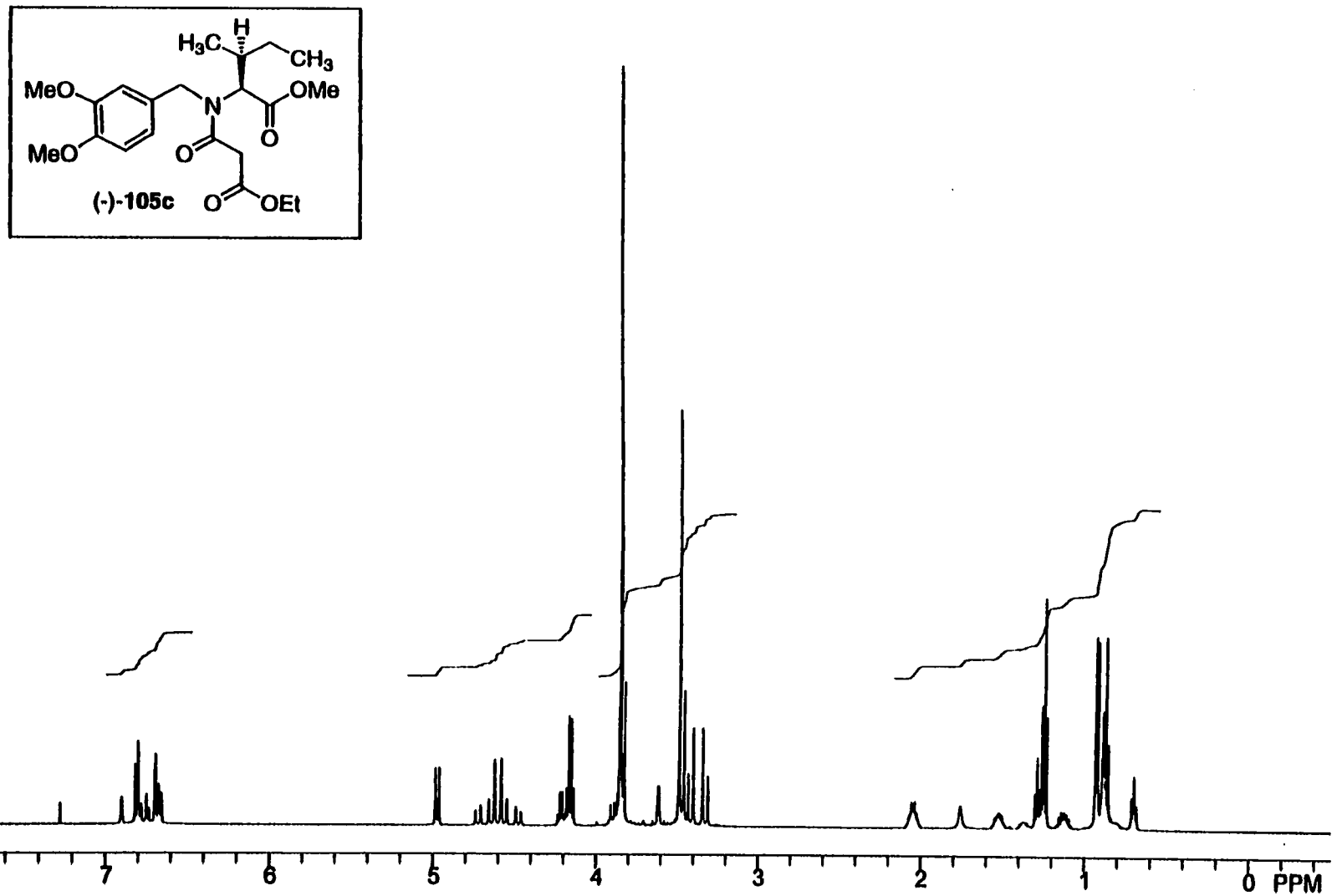


Figure A.4.16 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**105c**.

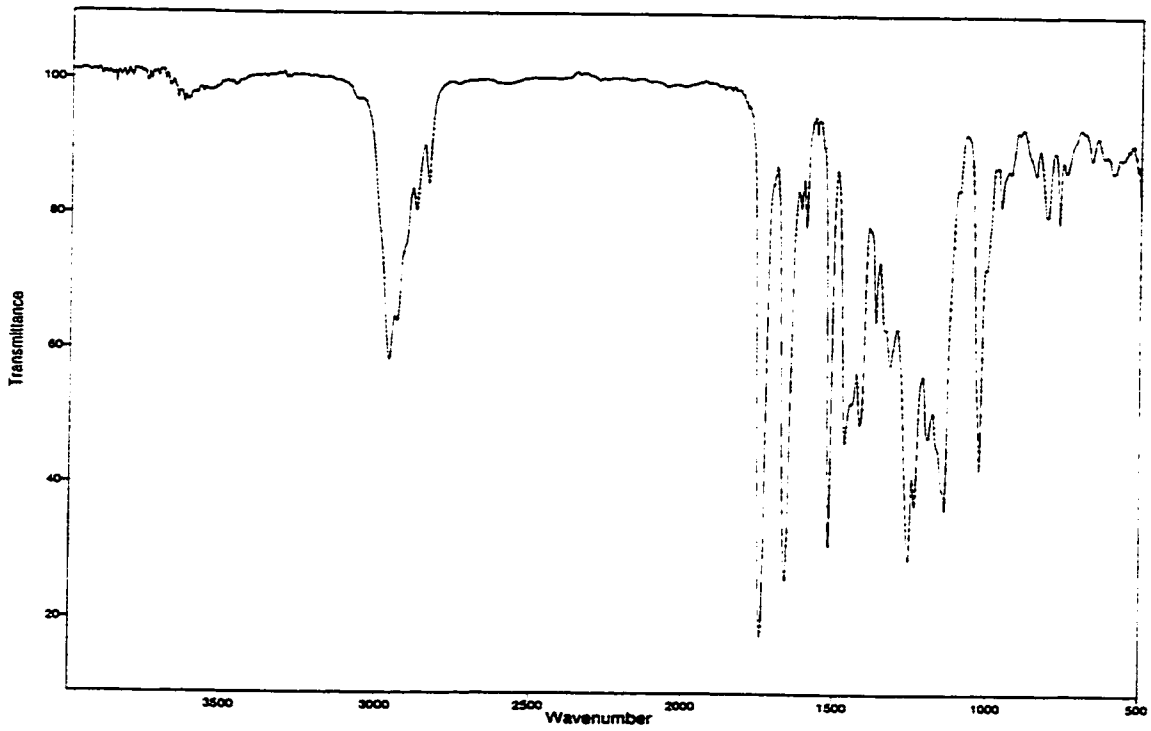


Figure A.4.17 Infrared Spectrum (thin film/NaCl) of compound (-)-**105c.**

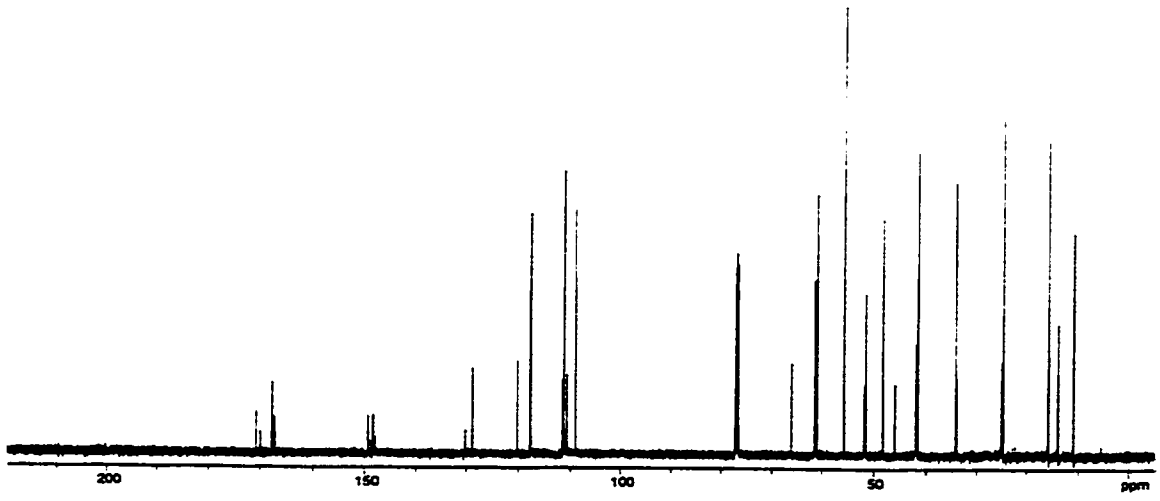


Figure A.4.18 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**105c.**

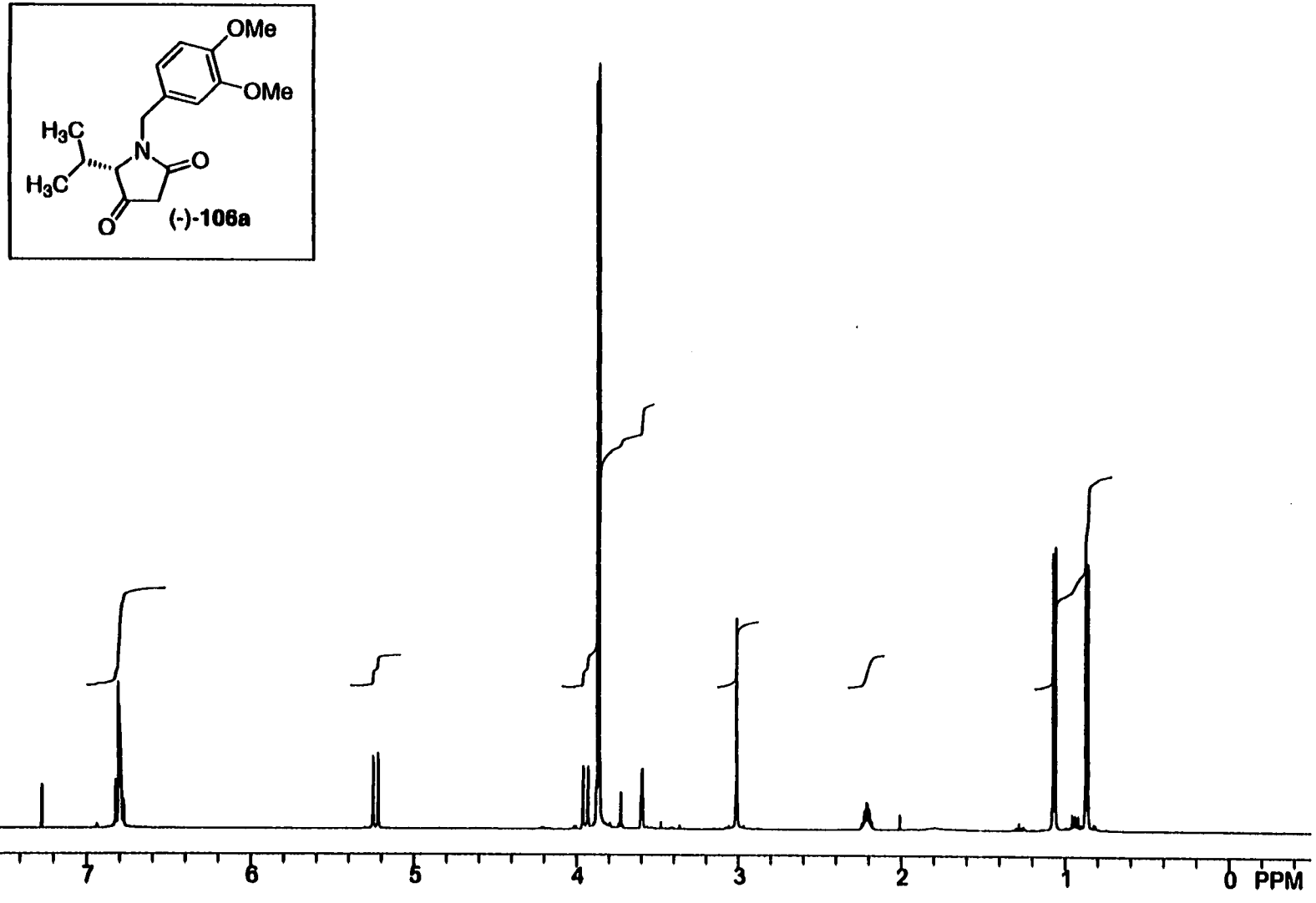


Figure A.4.19 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**106a**.

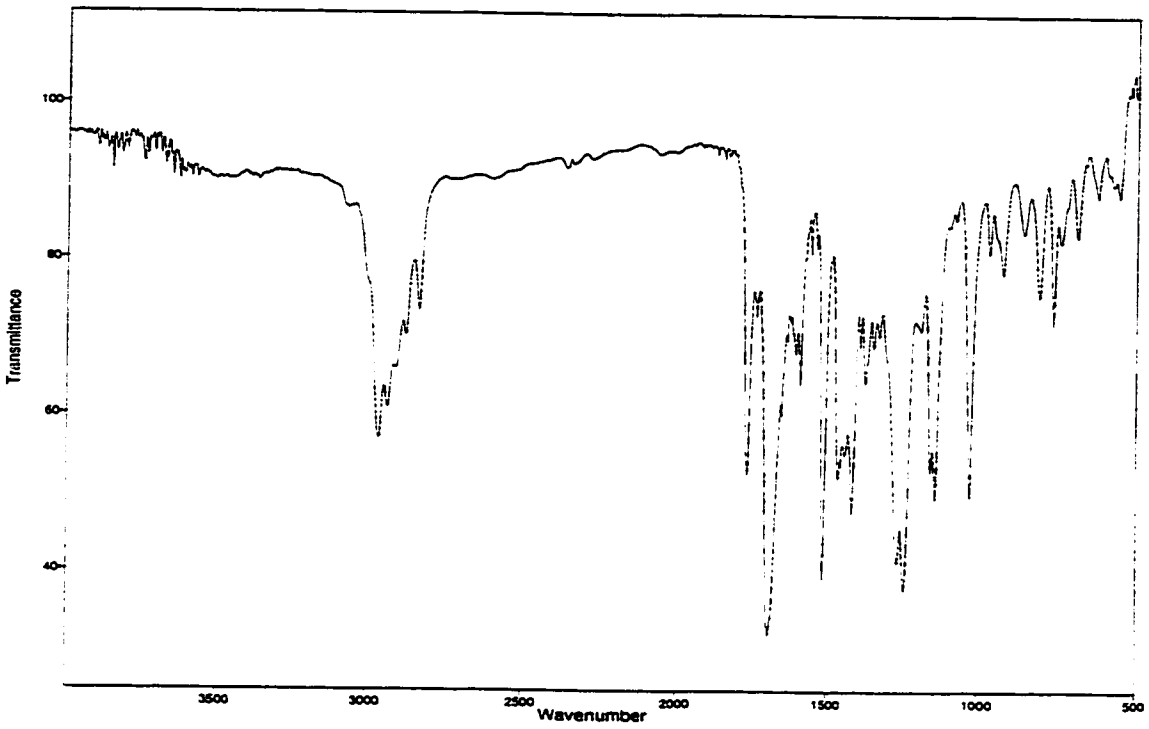


Figure A.4.20 Infrared Spectrum (thin film/NaCl) of compound (-)-**106a**.

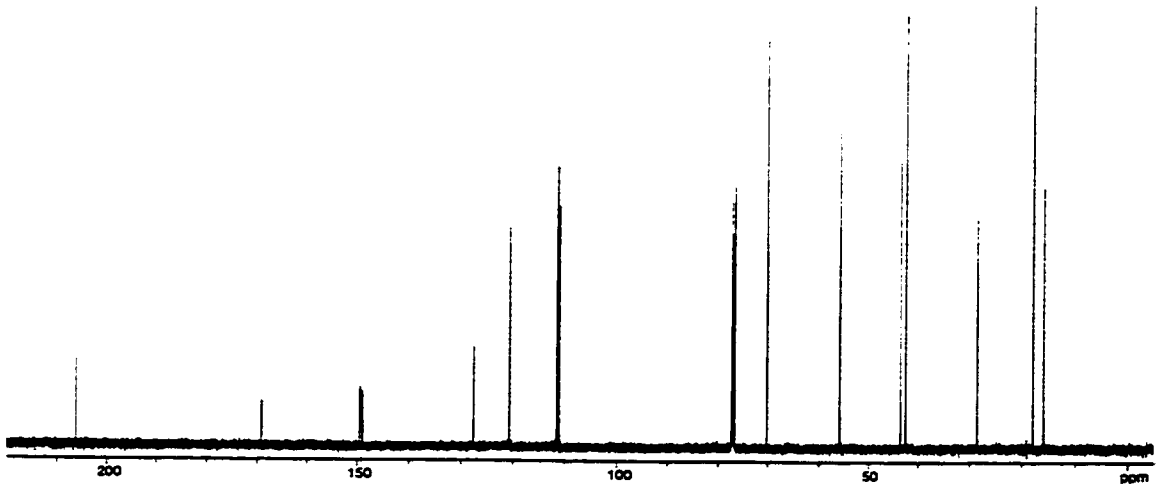


Figure A.4.21 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**106a**.

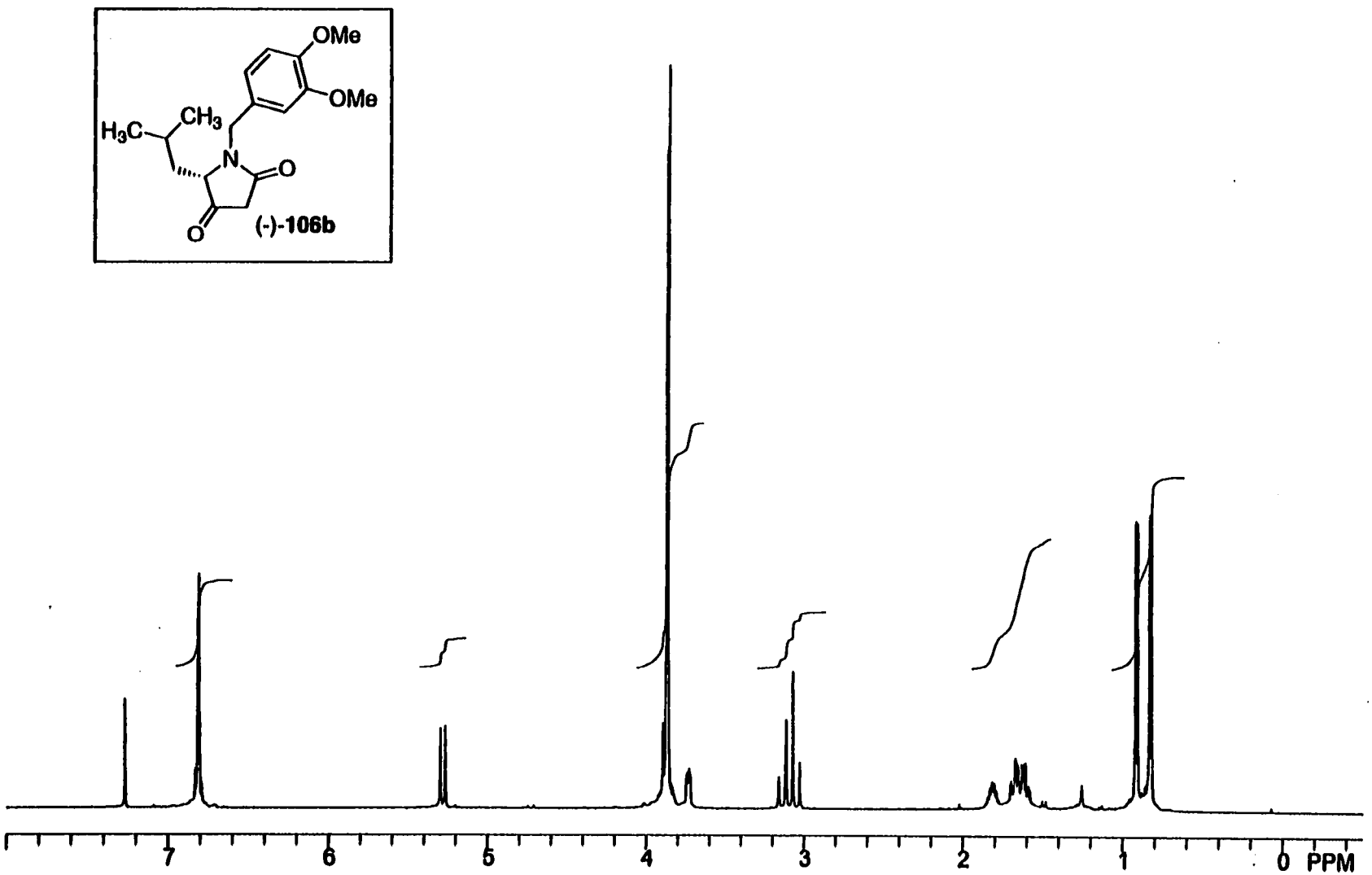


Figure A.4.22 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**106b**.

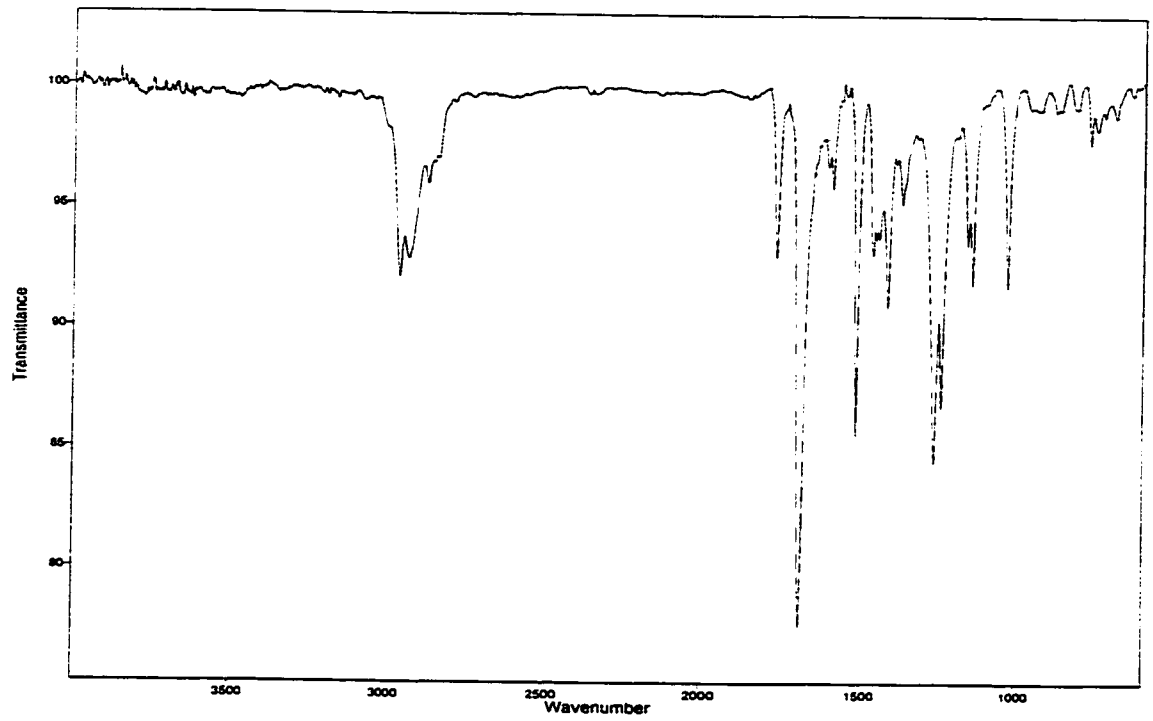


Figure A.4.23 Infrared Spectrum (thin film/NaCl) of compound (-)-**106b**.

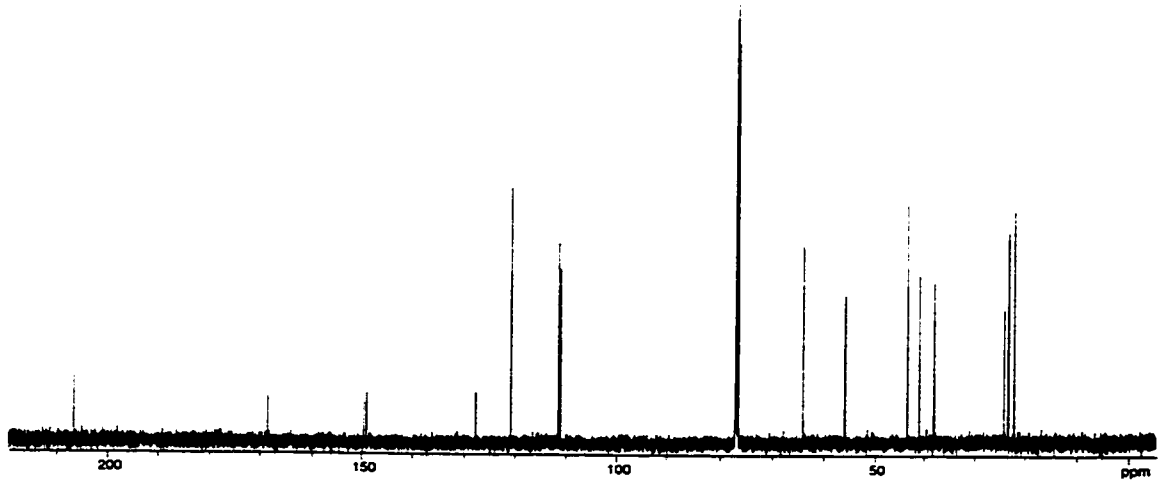


Figure A.4.24 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**106b**.

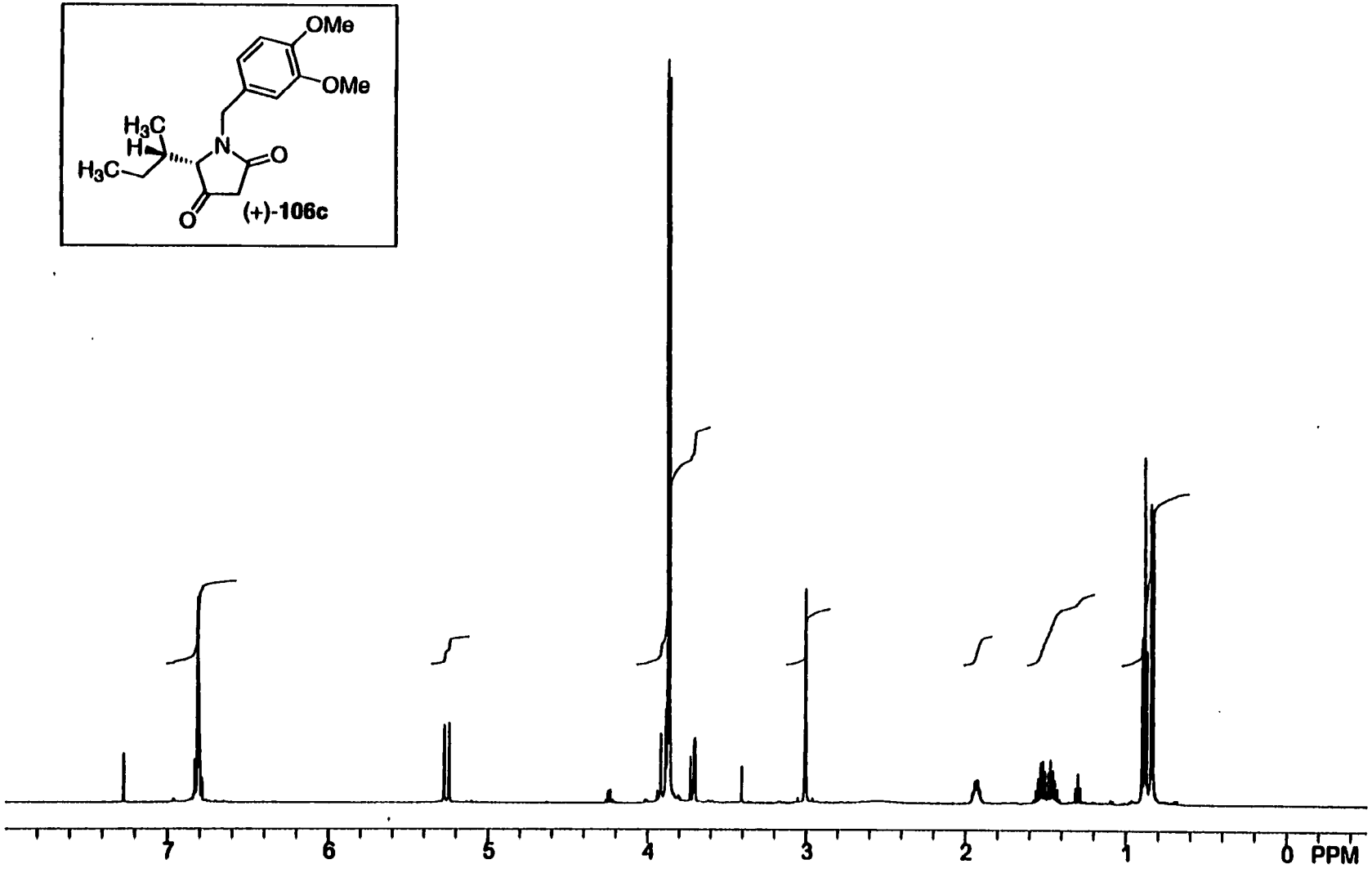


Figure A.4.25 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**106c**.

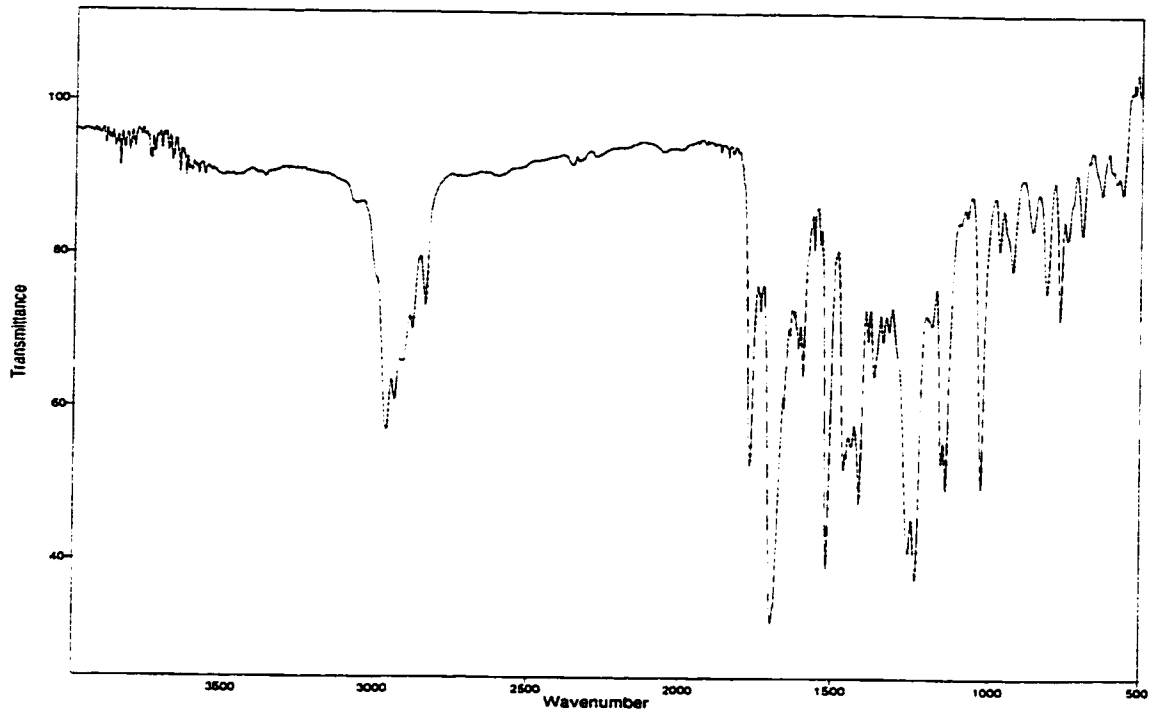


Figure A.4.26 Infrared Spectrum (thin film/NaCl) of compound (-)-**106c**.

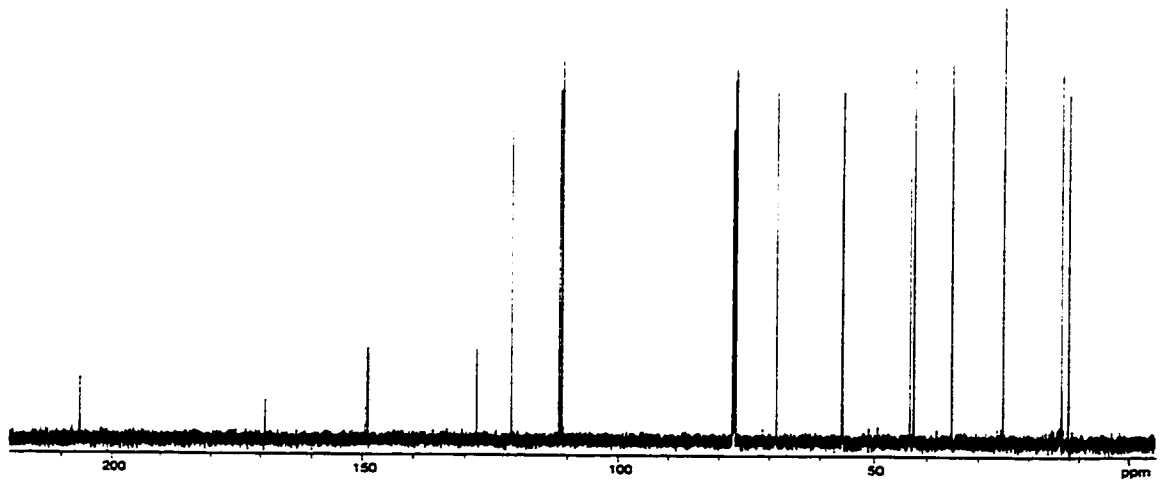


Figure A.4.27 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**106c**.

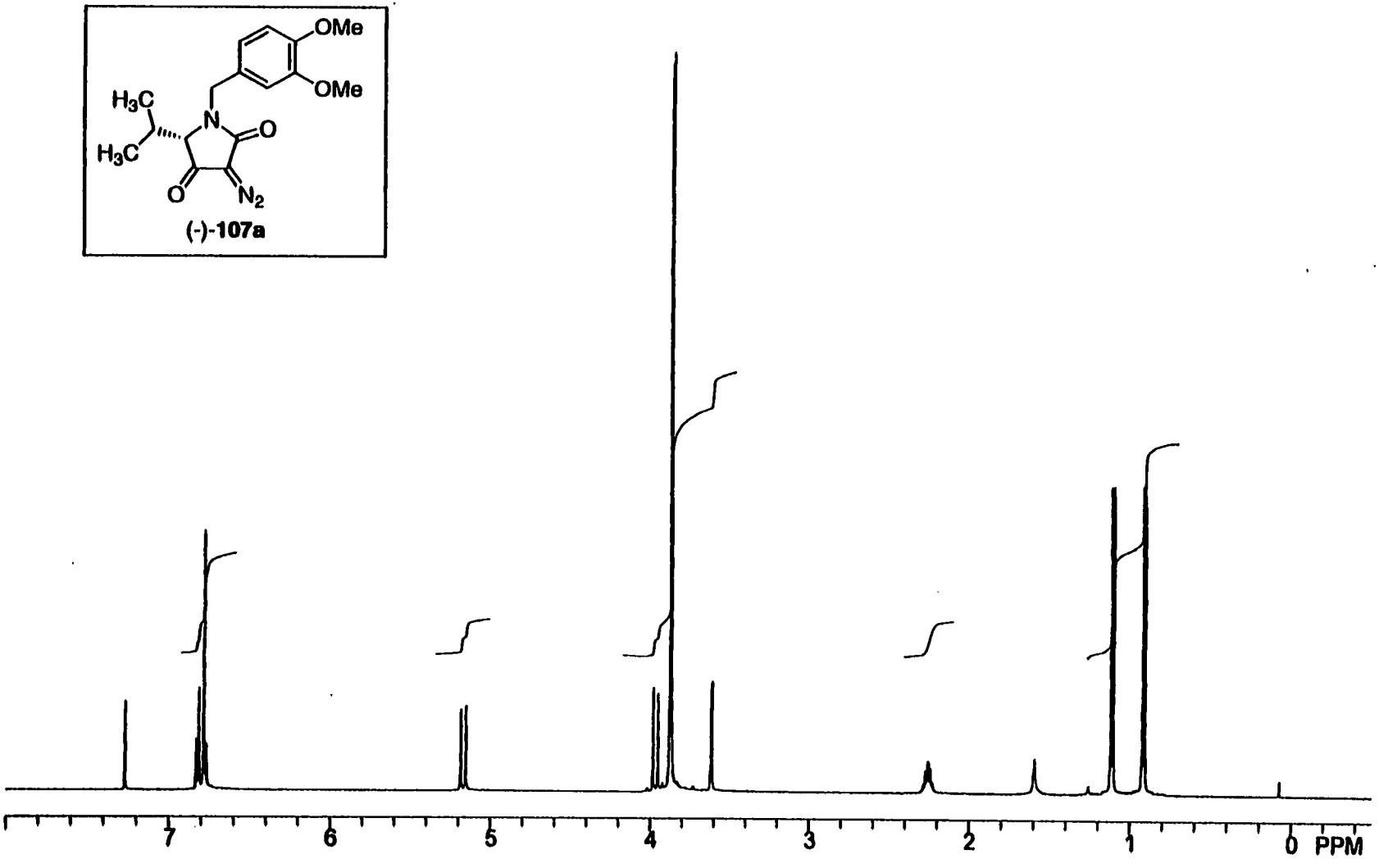


Figure A.4.28 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**107a**.

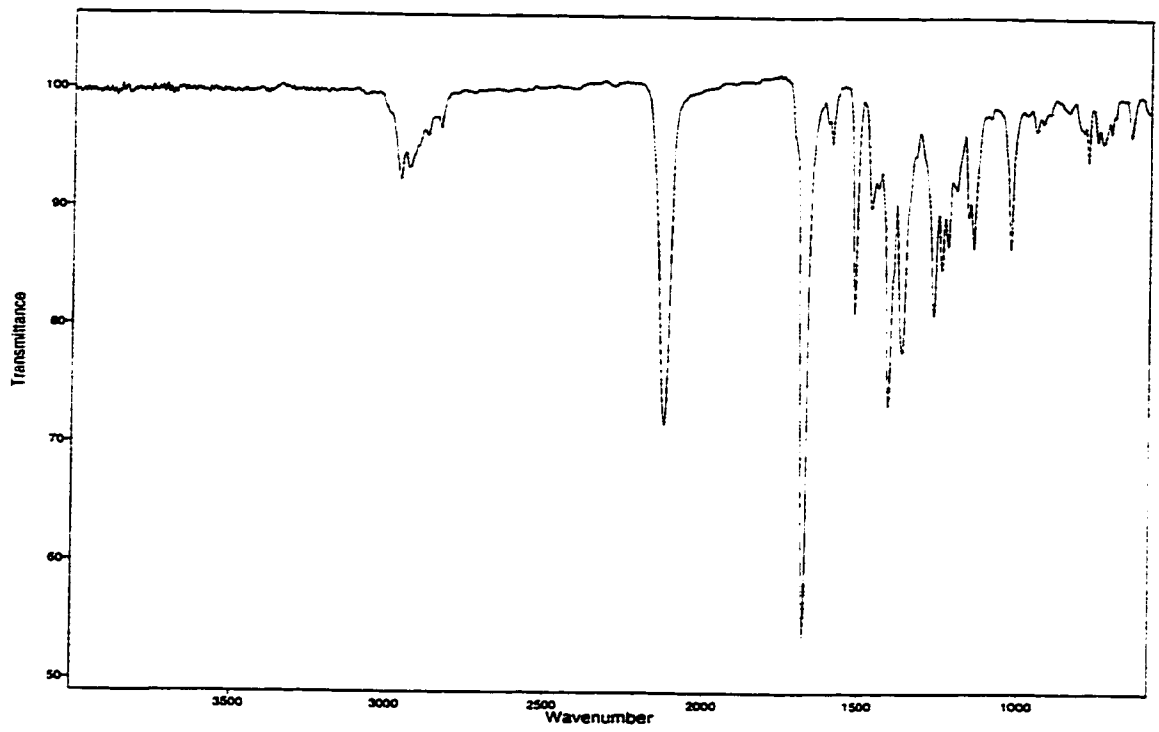


Figure A.4.29 Infrared Spectrum (thin film/NaCl) of compound (-)-**107a**.

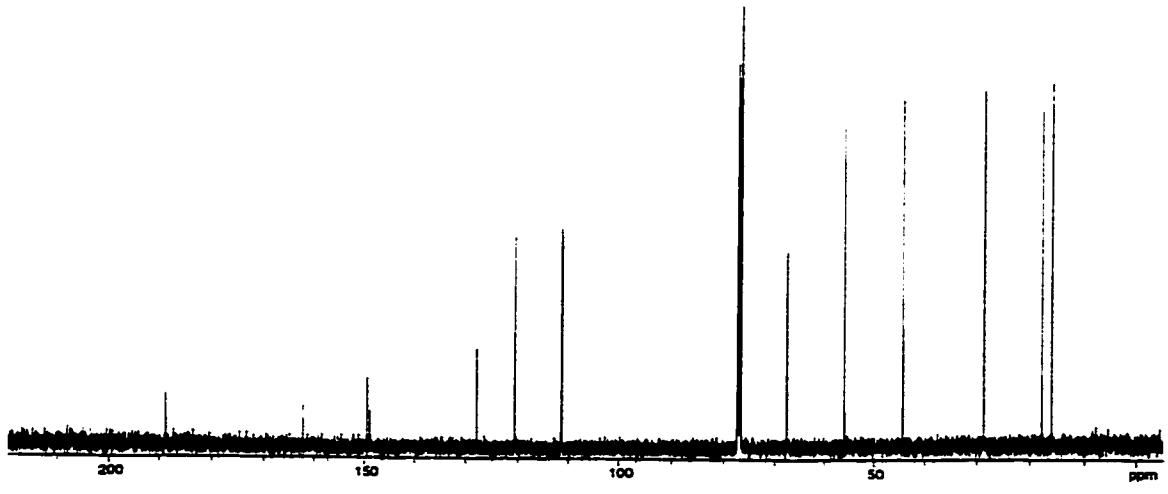


Figure A.4.30 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**107a**.

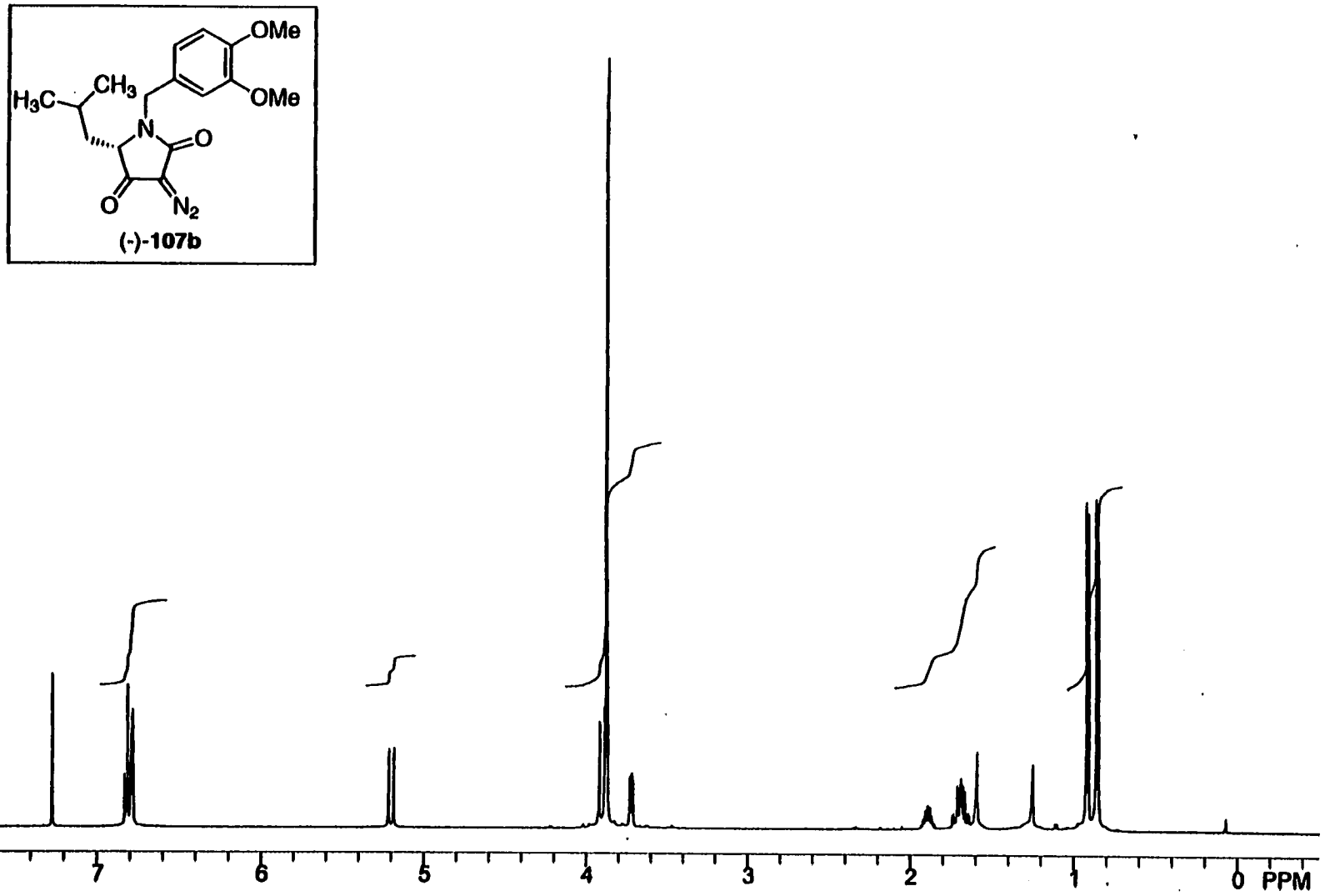


Figure A.4.31 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**107b**.

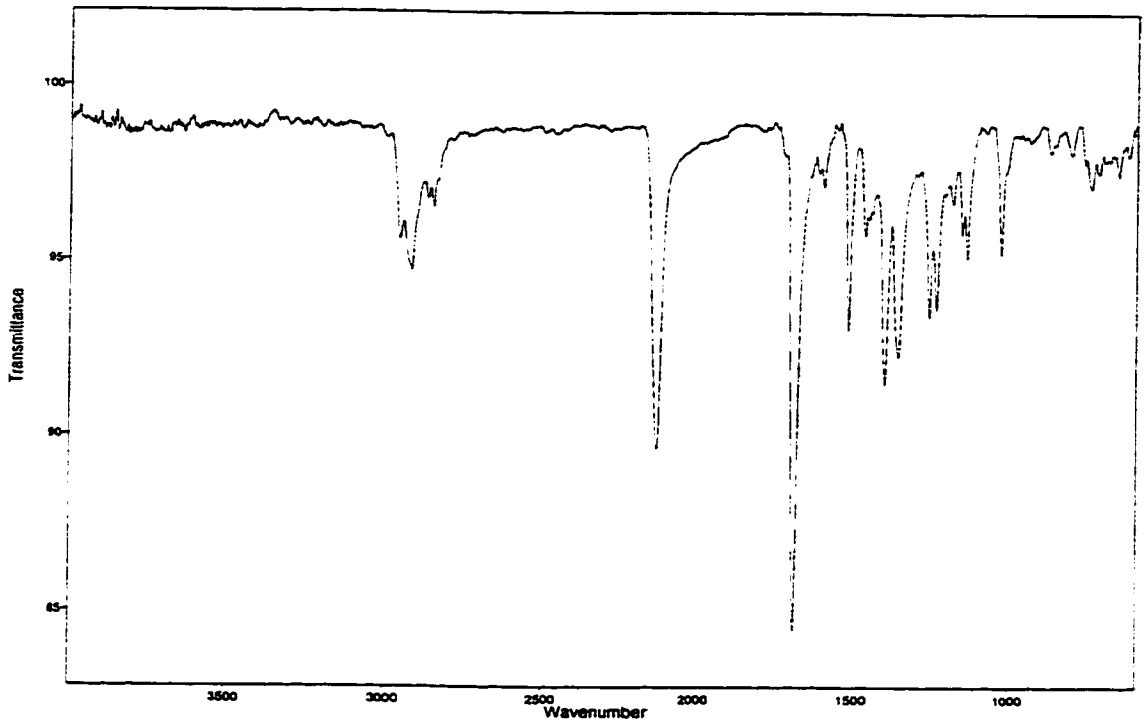


Figure A.4.32 Infrared Spectrum (thin film/NaCl) of compound (-)-**107b**.

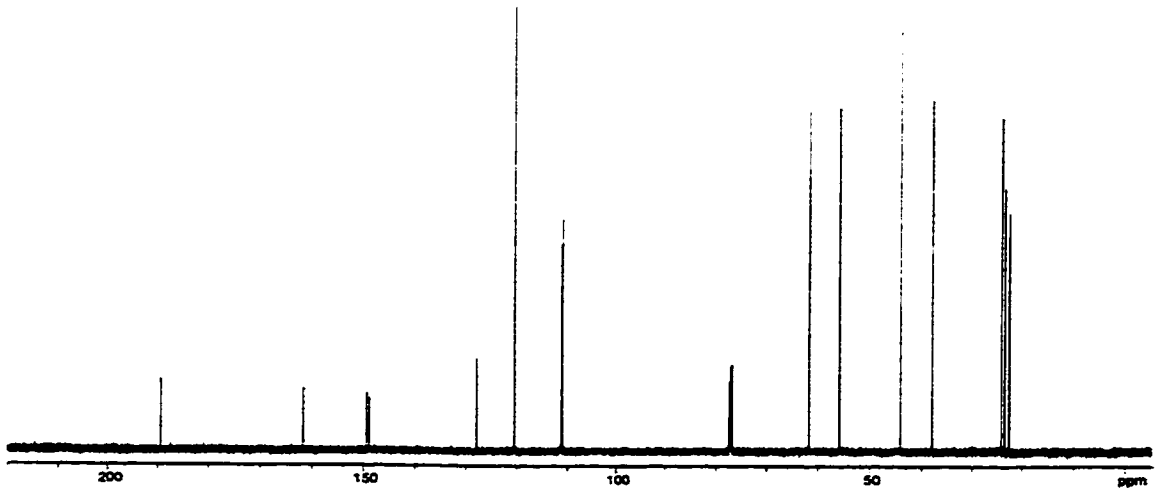


Figure A.4.33 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**107b**.

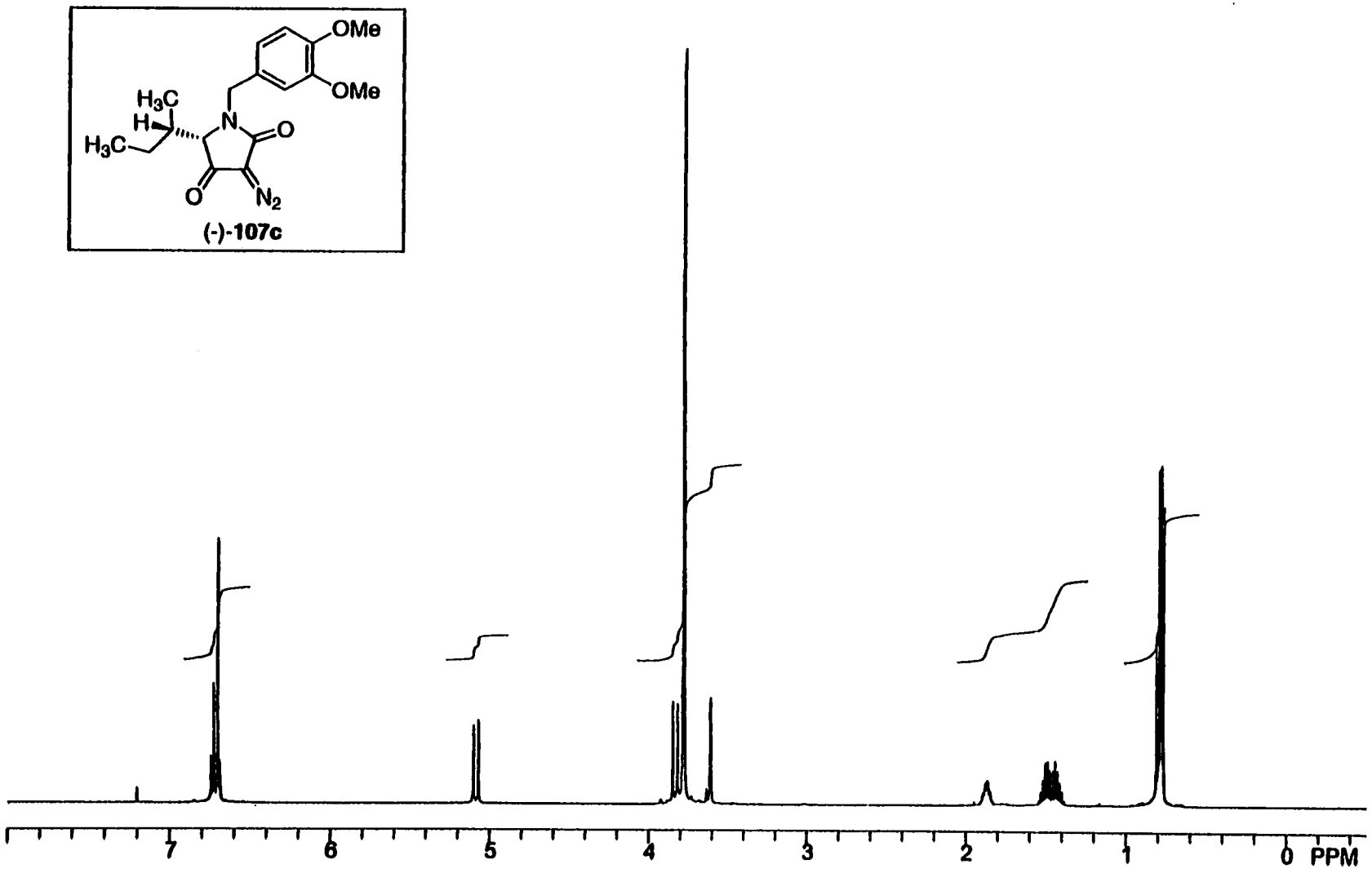


Figure A.4.34 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (-)-**107c**.

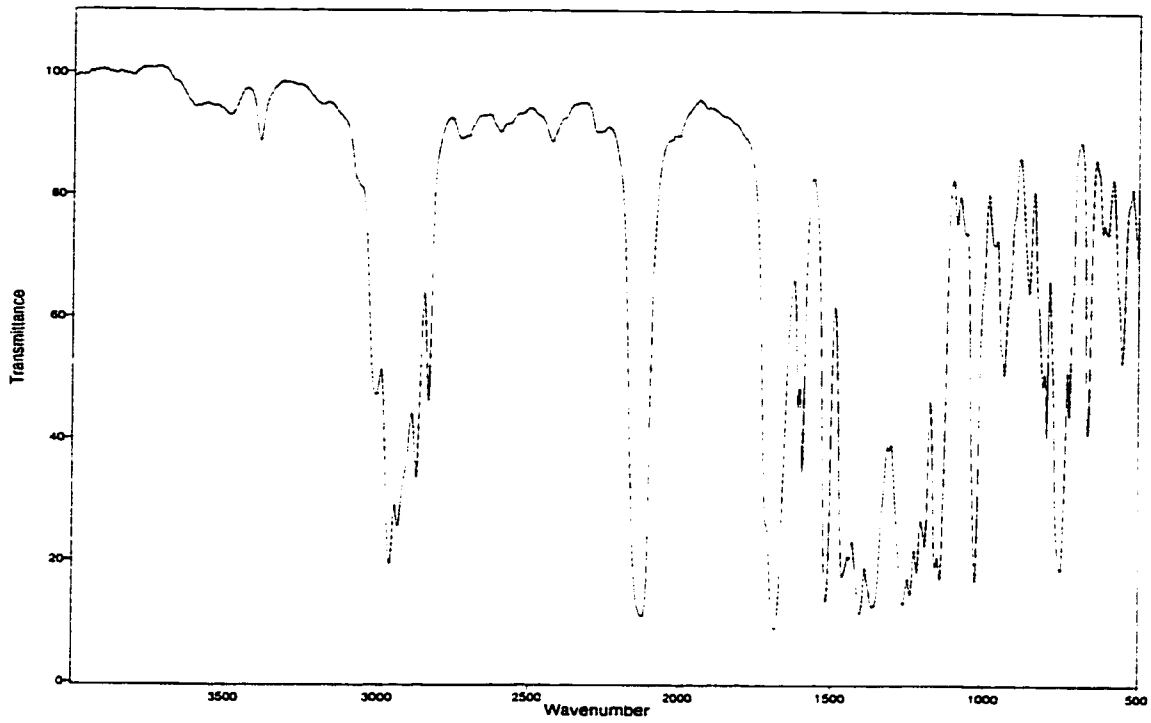


Figure A.4.35 Infrared Spectrum (thin film/NaCl) of compound (-)-**107c**.

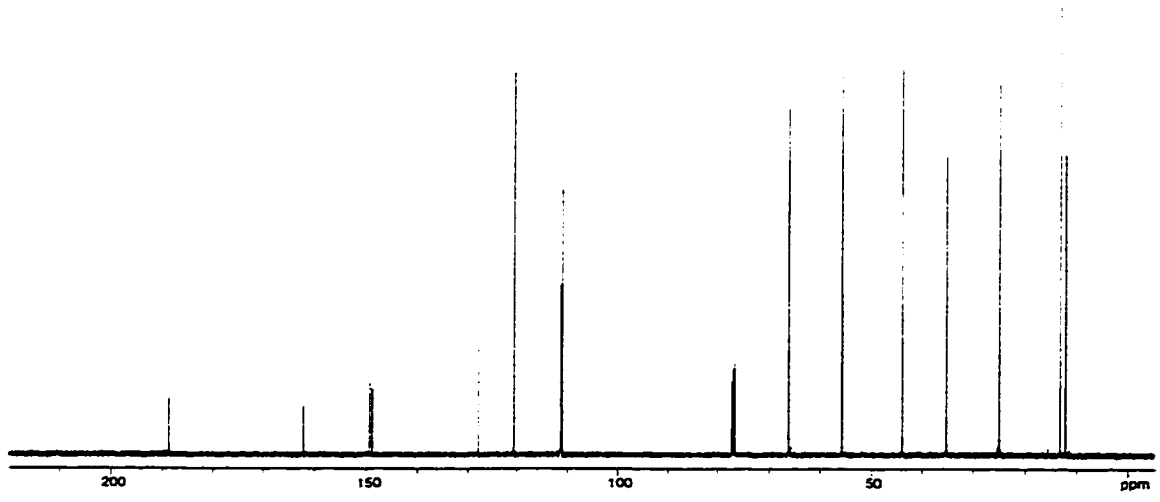


Figure A.4.36 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (-)-**107c**.

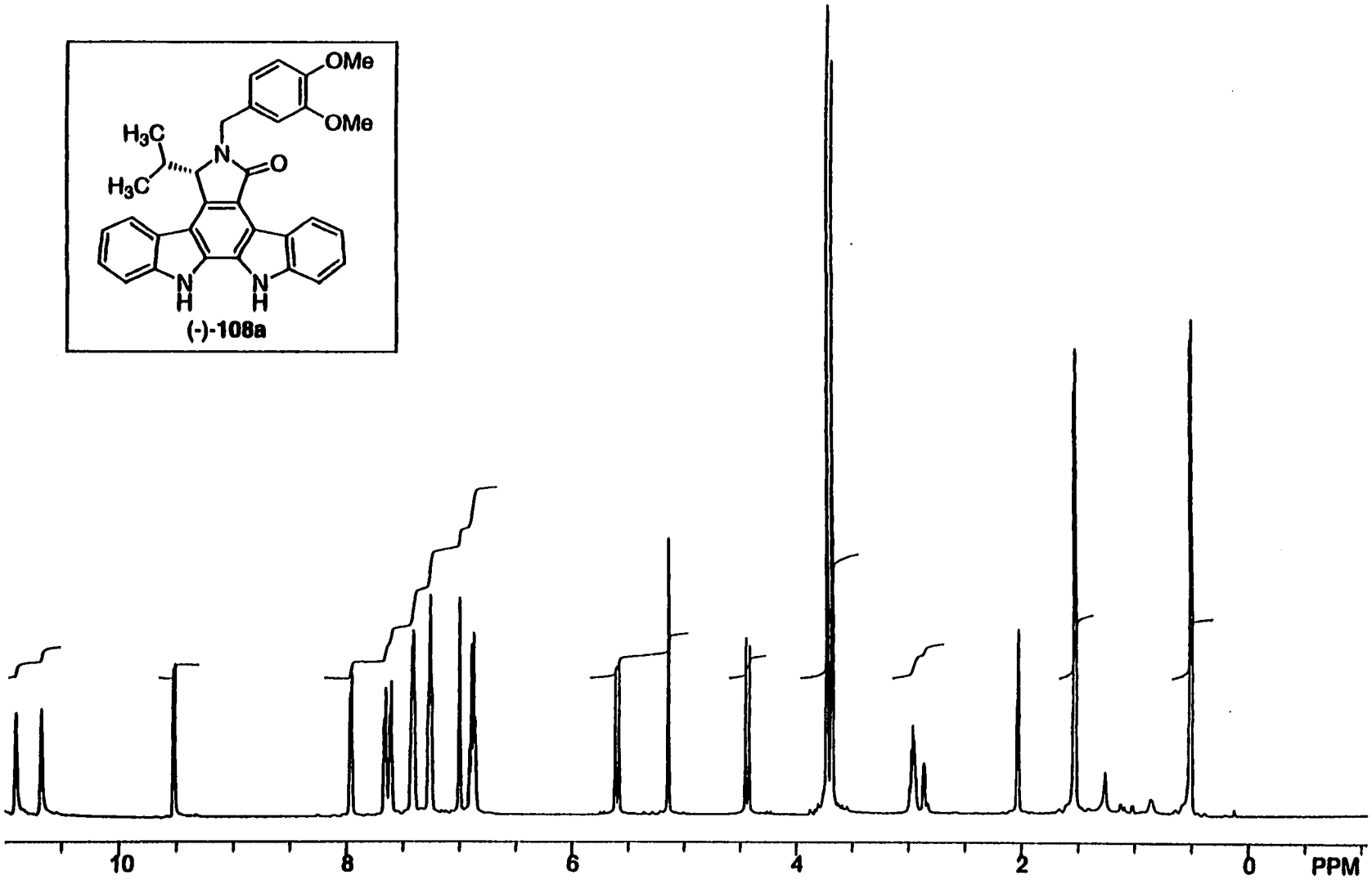


Figure A.4.37 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**108a**.

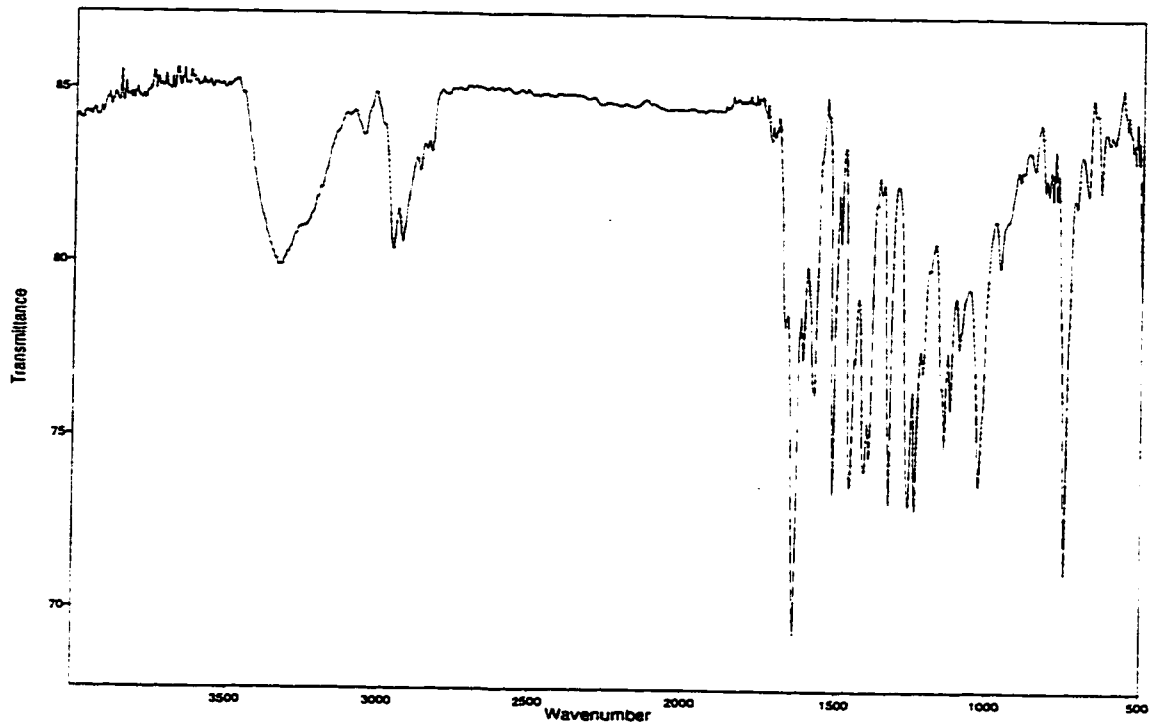


Figure A.4.38 Infrared Spectrum (thin film/NaCl) of compound (-)-**108a**.

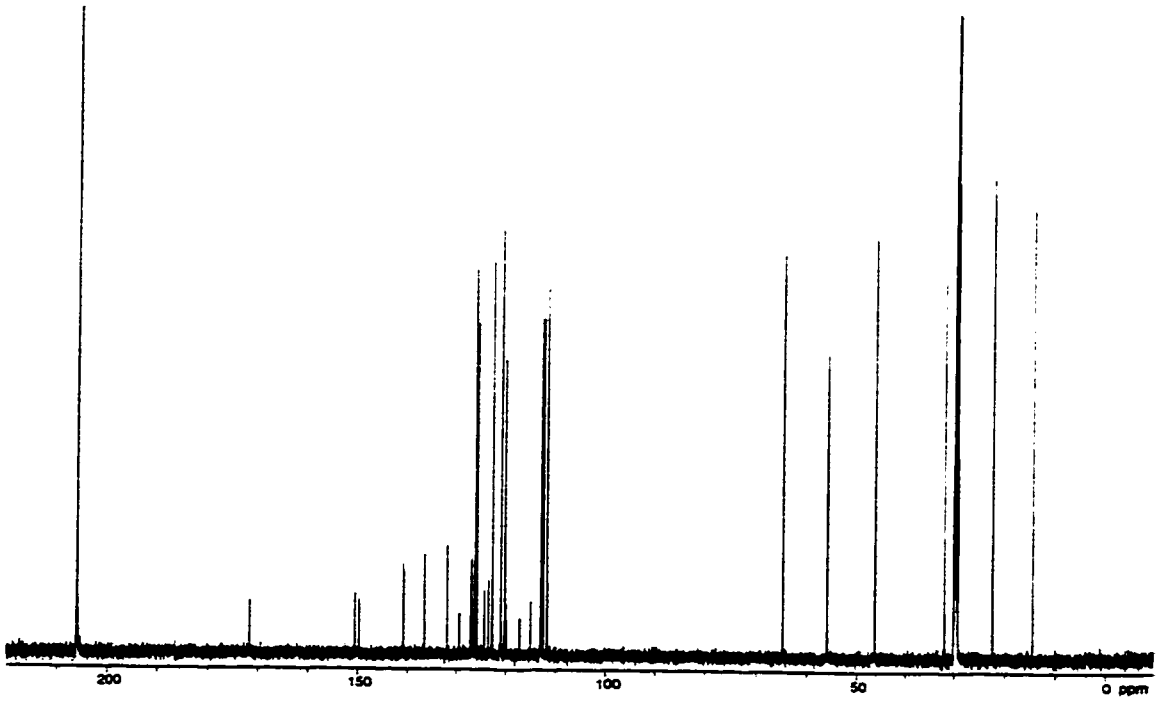


Figure A.4.39 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**108a**.

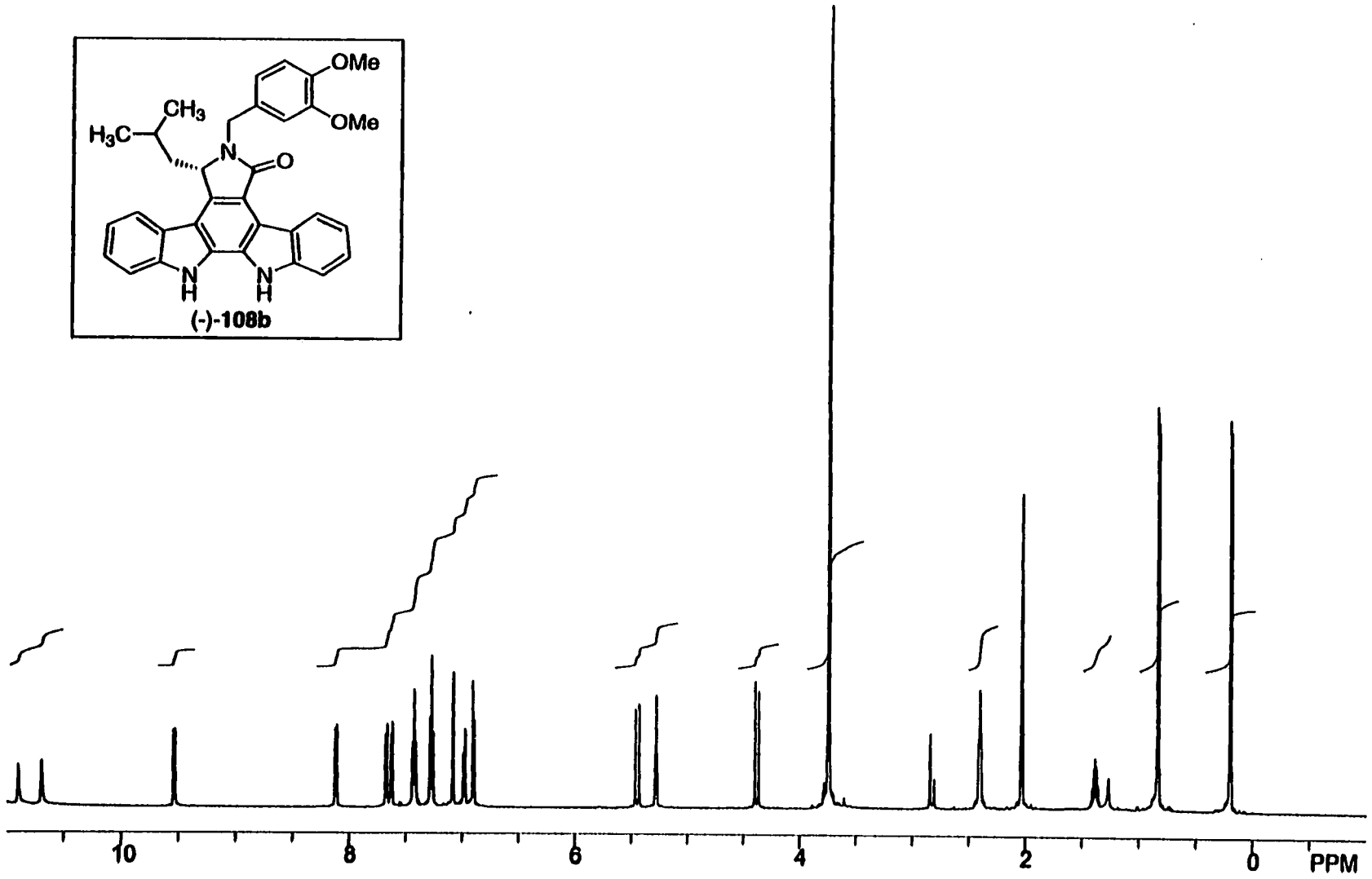


Figure A.4.40 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**108b**.

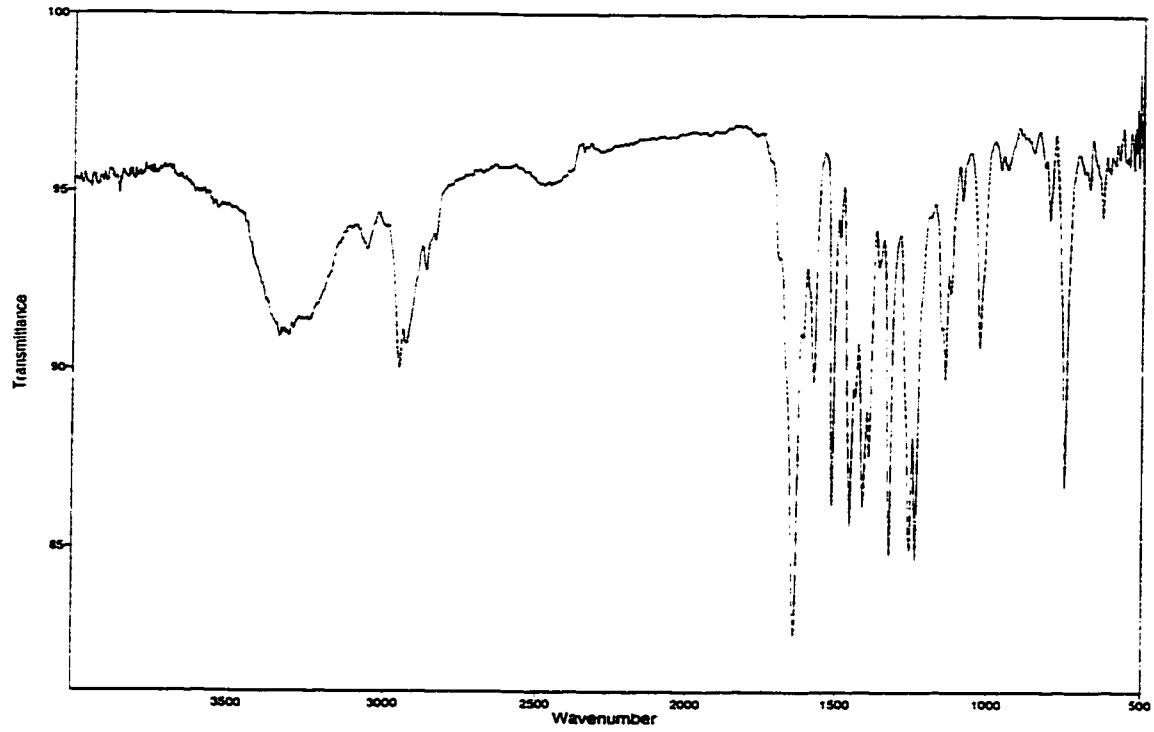


Figure A.4.41 Infrared Spectrum (thin film/NaCl) of compound (-)-**108b**.

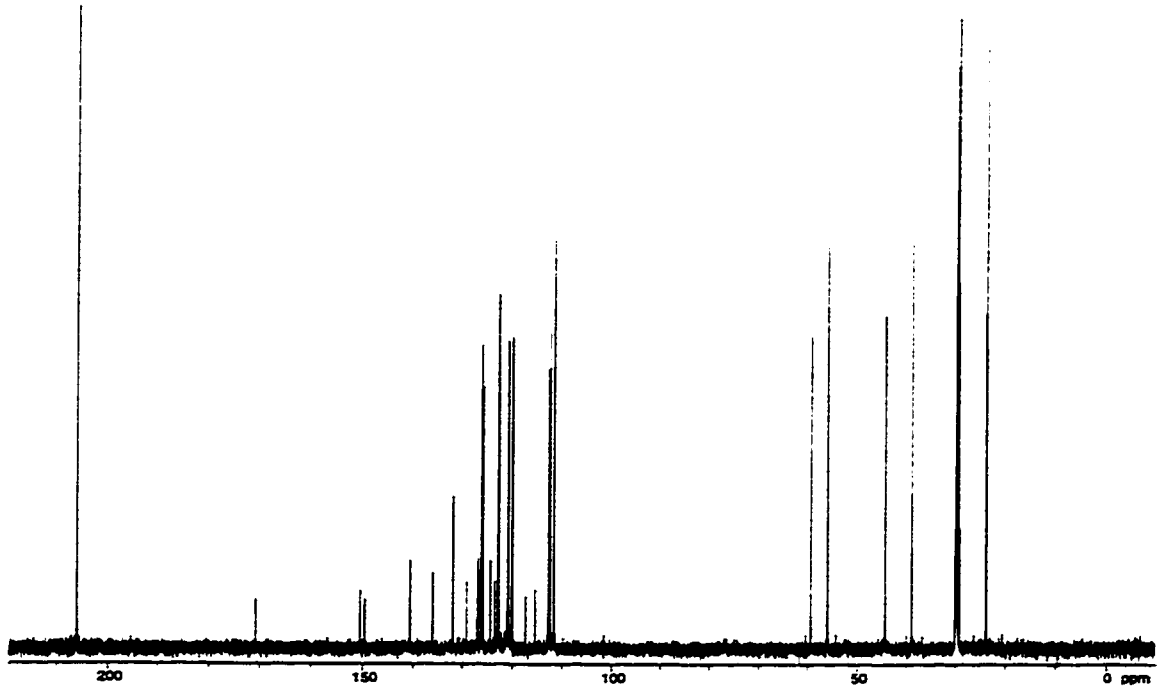


Figure A.4.42 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**108b**.

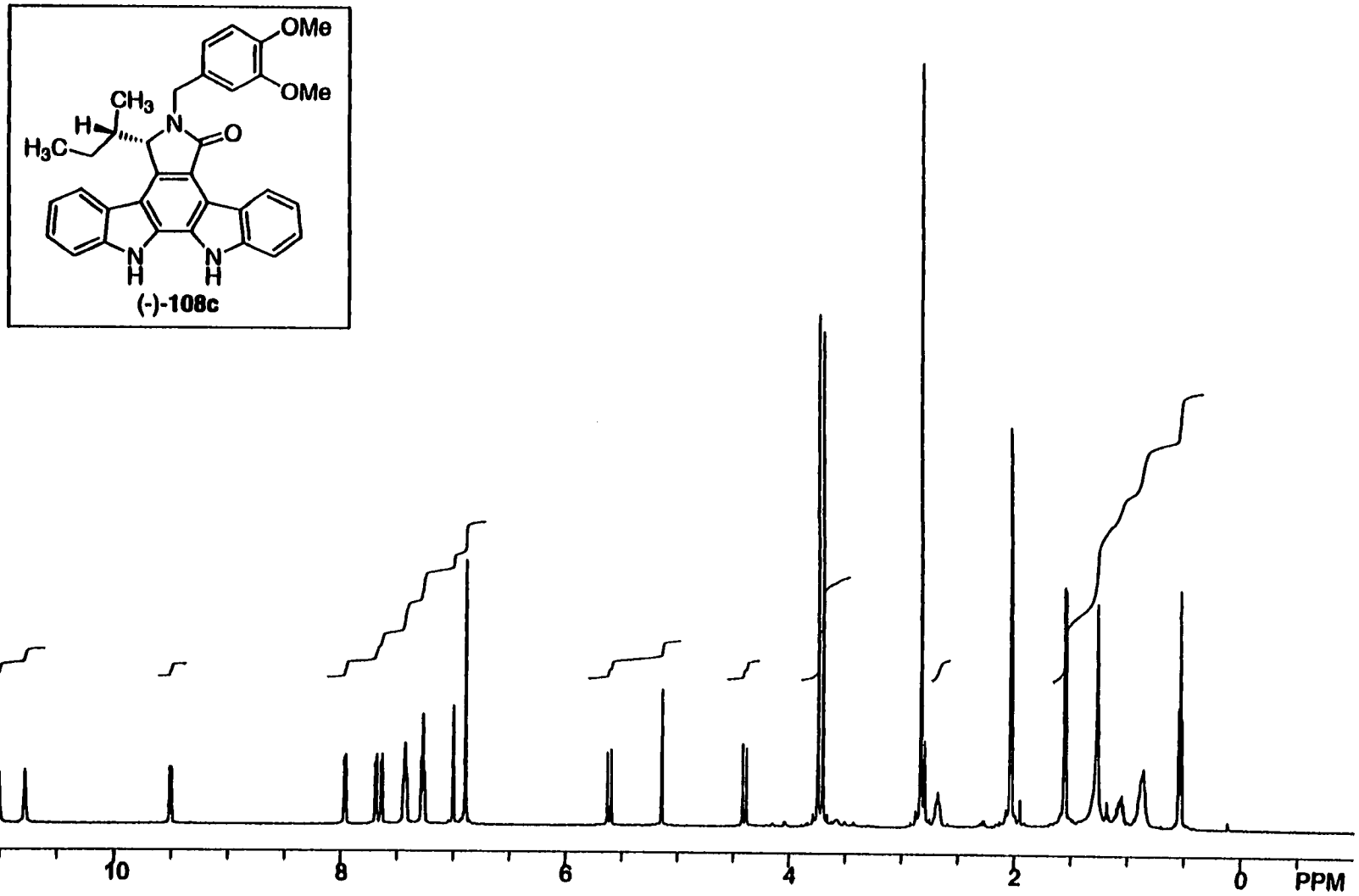


Figure A.4.43 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**108c**.

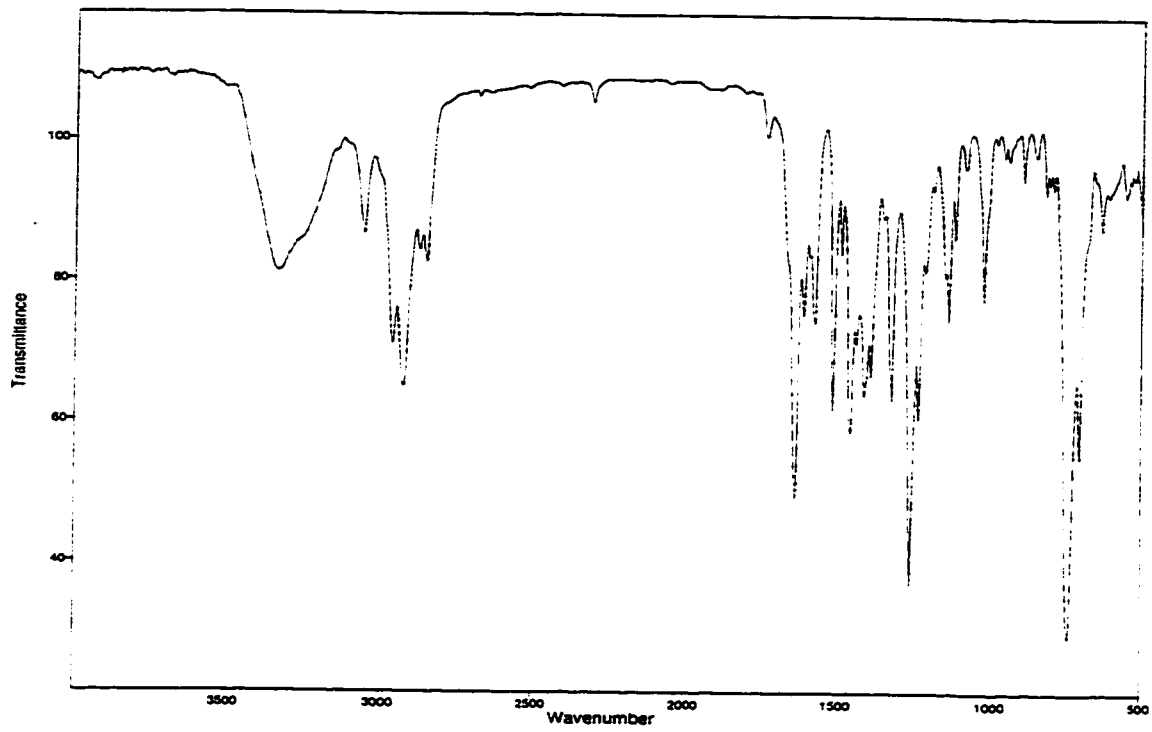


Figure A.4.44 Infrared Spectrum (thin film/NaCl) of compound (-)-**108c**.

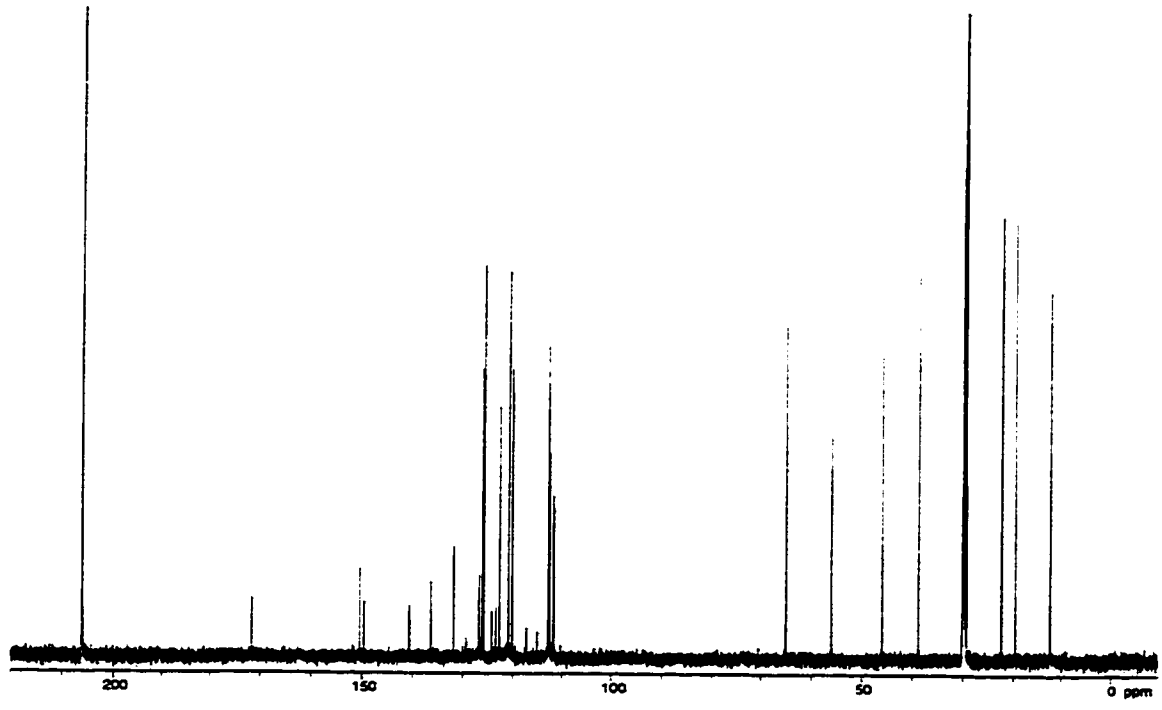
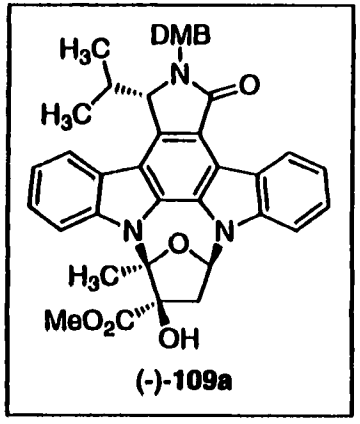


Figure A.4.45 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**108c**.



(-)-**109a**

Figure A.4.46 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**109a**.

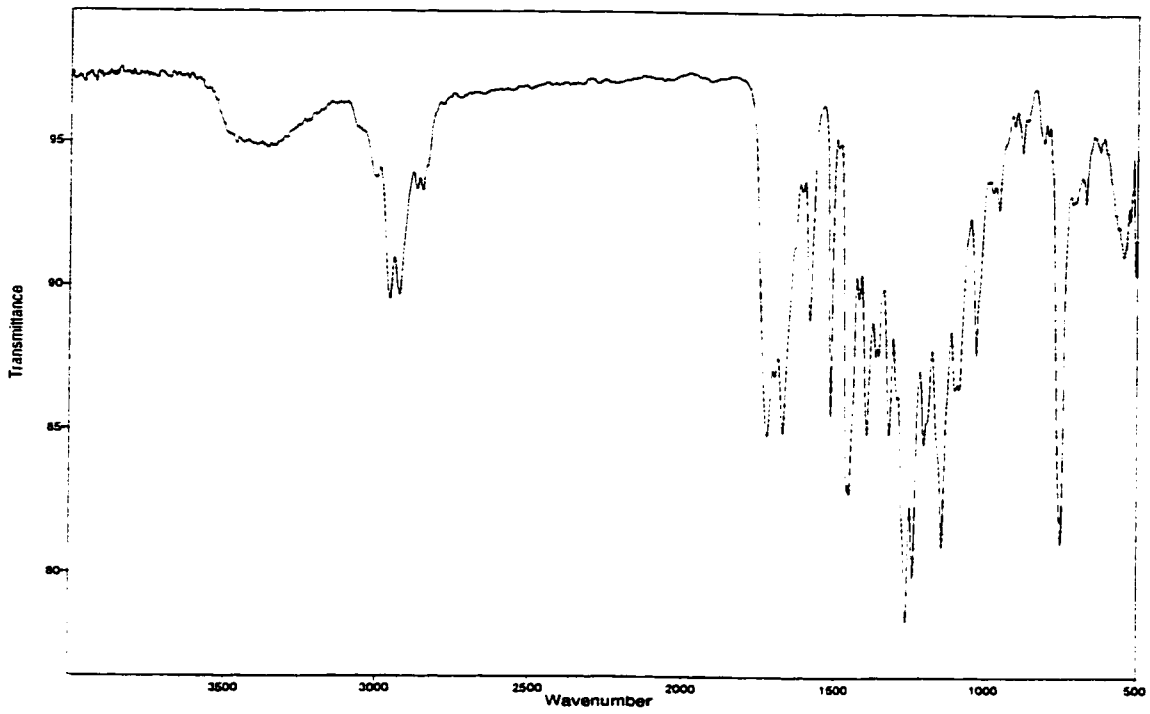


Figure A.4.47 Infrared Spectrum (thin film/NaCl) of compound (-)-**109a.**

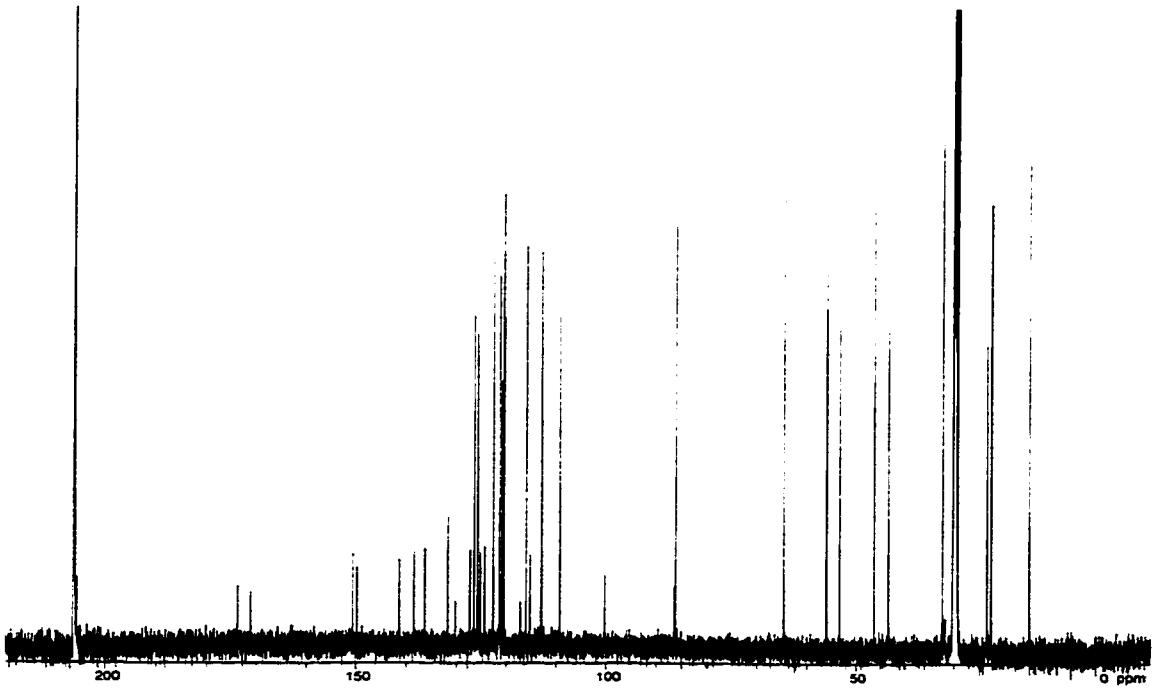


Figure A.4.48 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**109a.**

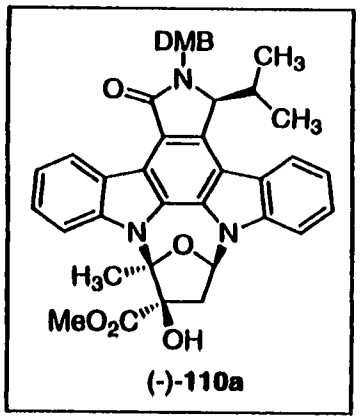


Figure A.4.49 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**110a**.

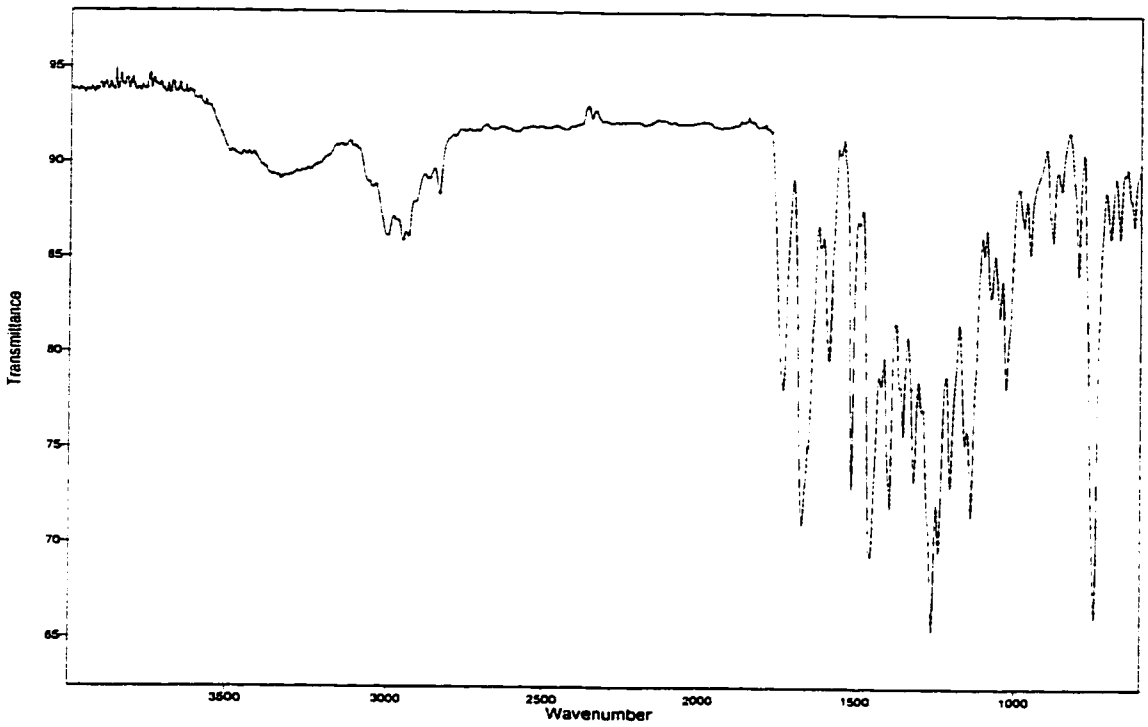


Figure A.4.50 Infrared Spectrum (thin film/NaCl) of compound (-)-**110a**.

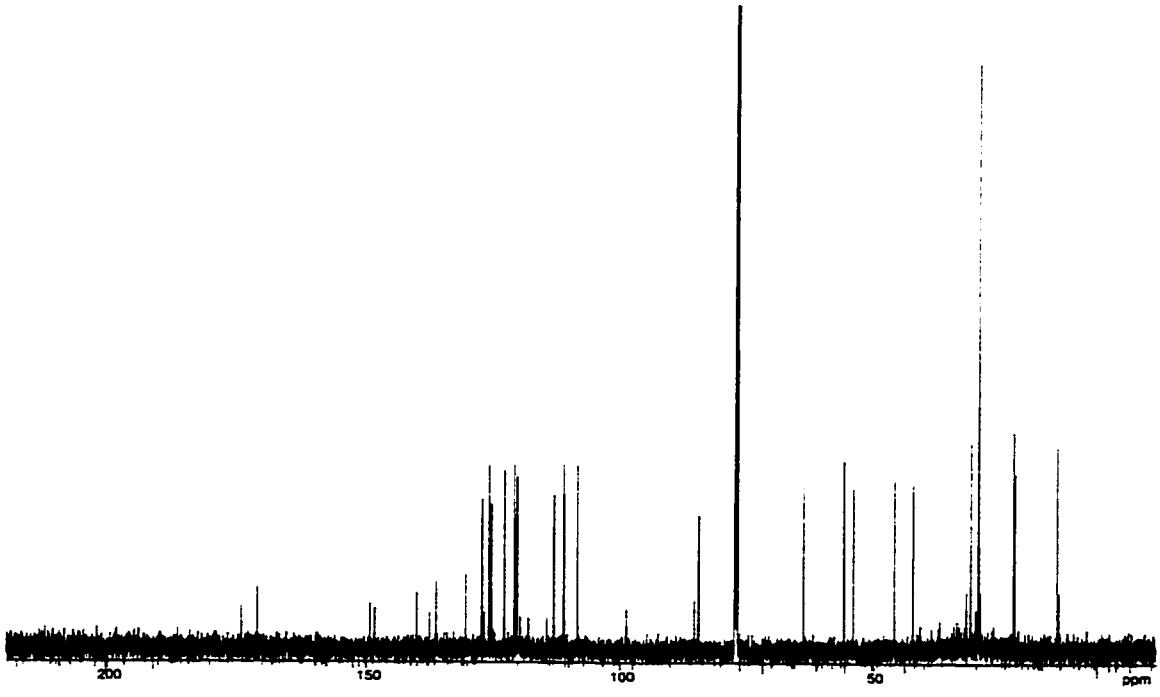


Figure A.4.51 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**110a**.

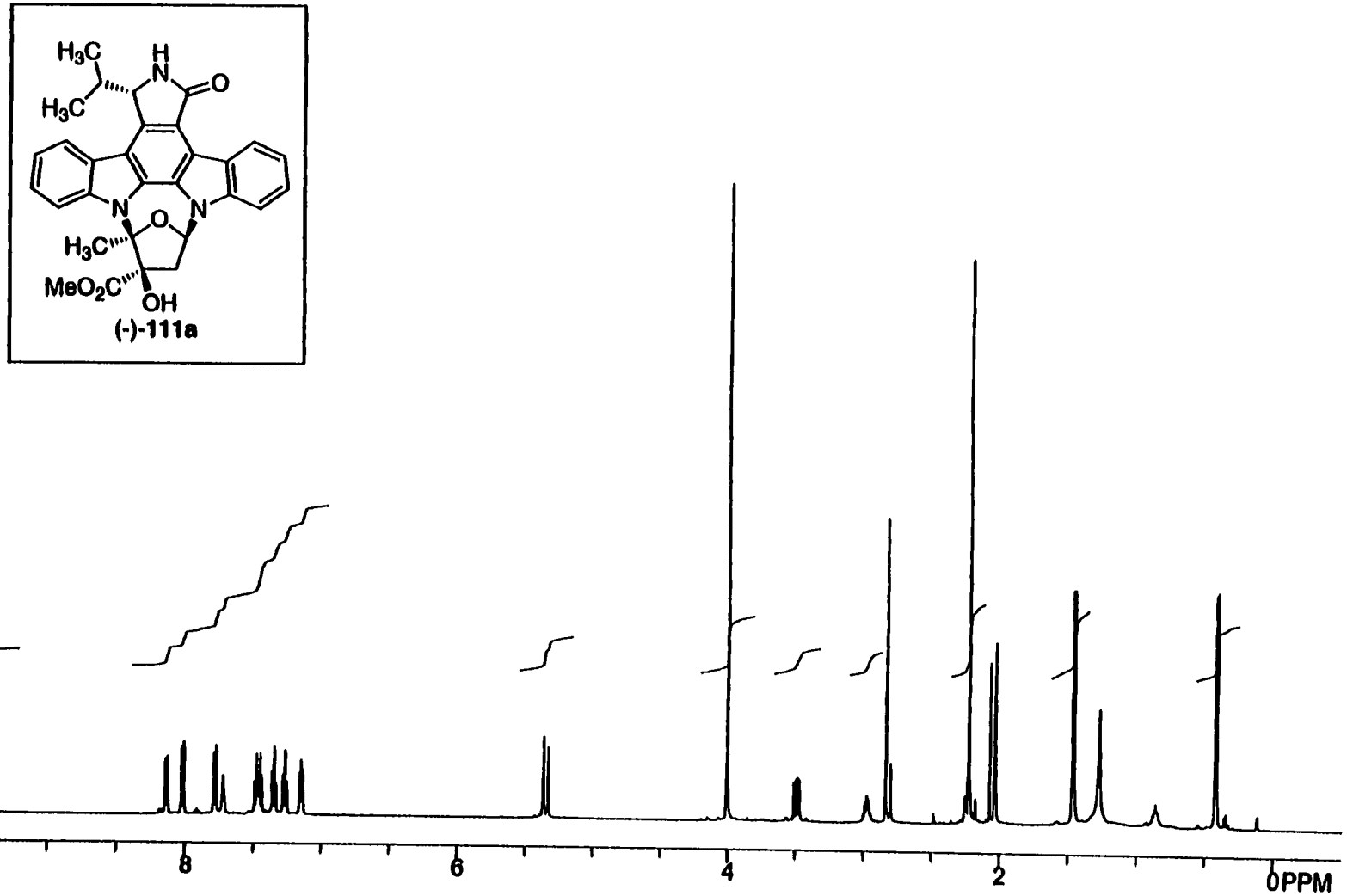


Figure A.4.52 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**111a**.

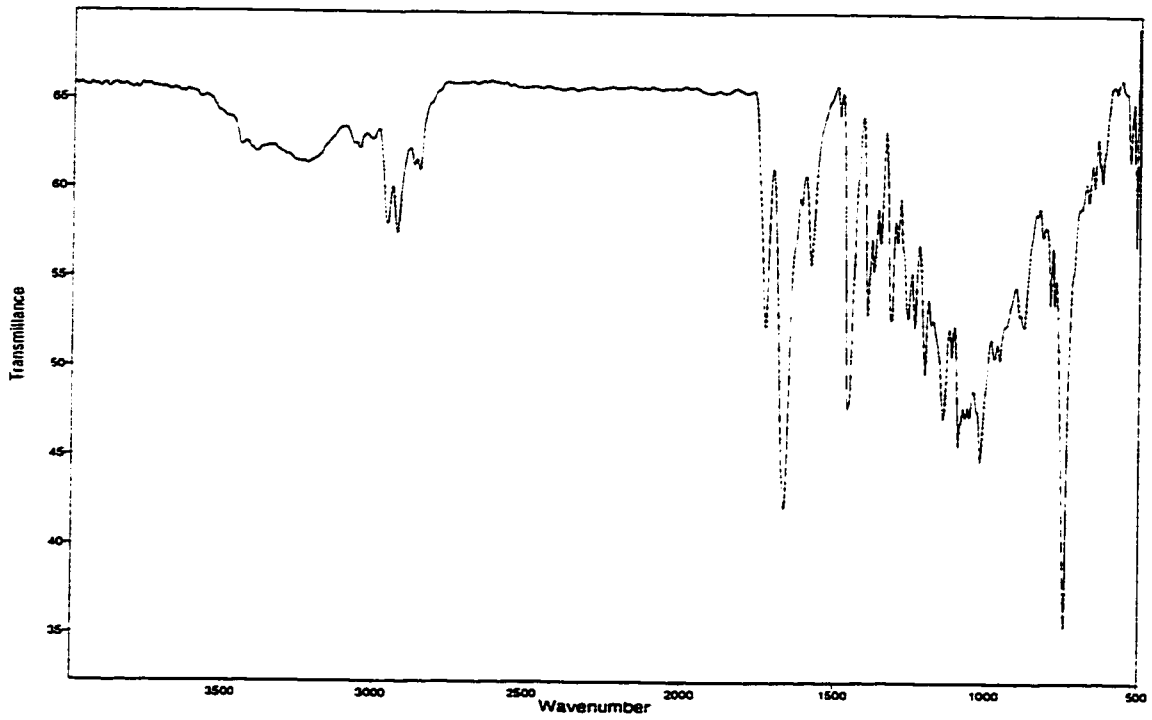


Figure A.4.53 Infrared Spectrum (thin film/NaCl) of compound (-)-**111a**.

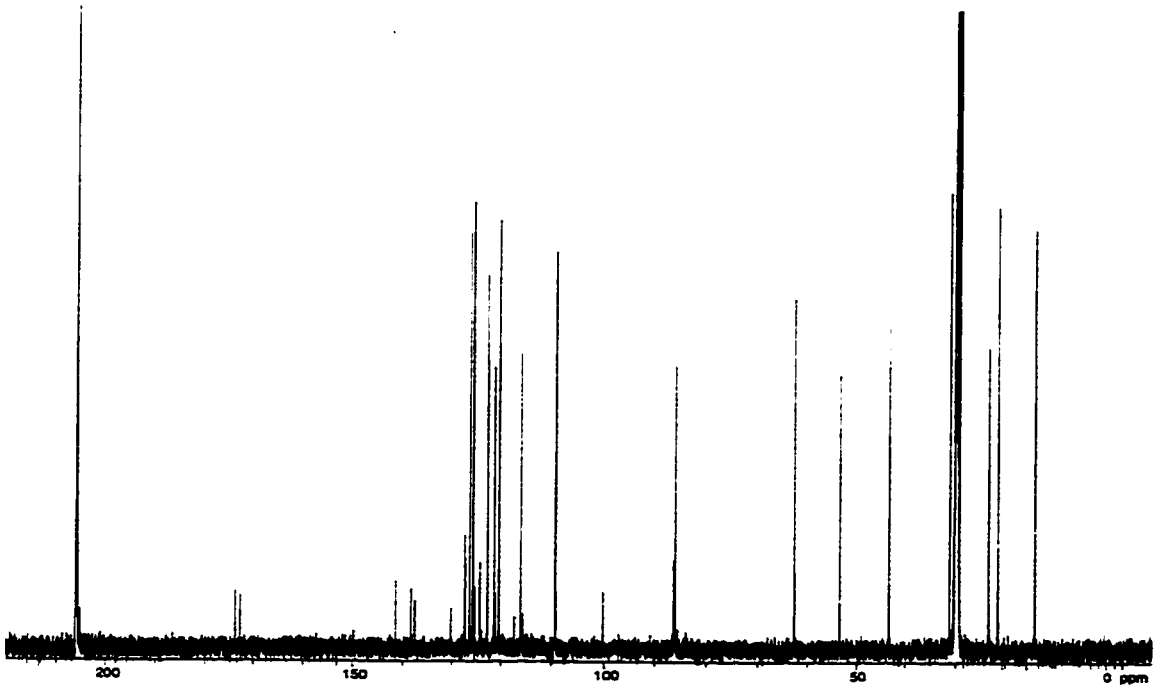


Figure A.4.54 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**111a**.

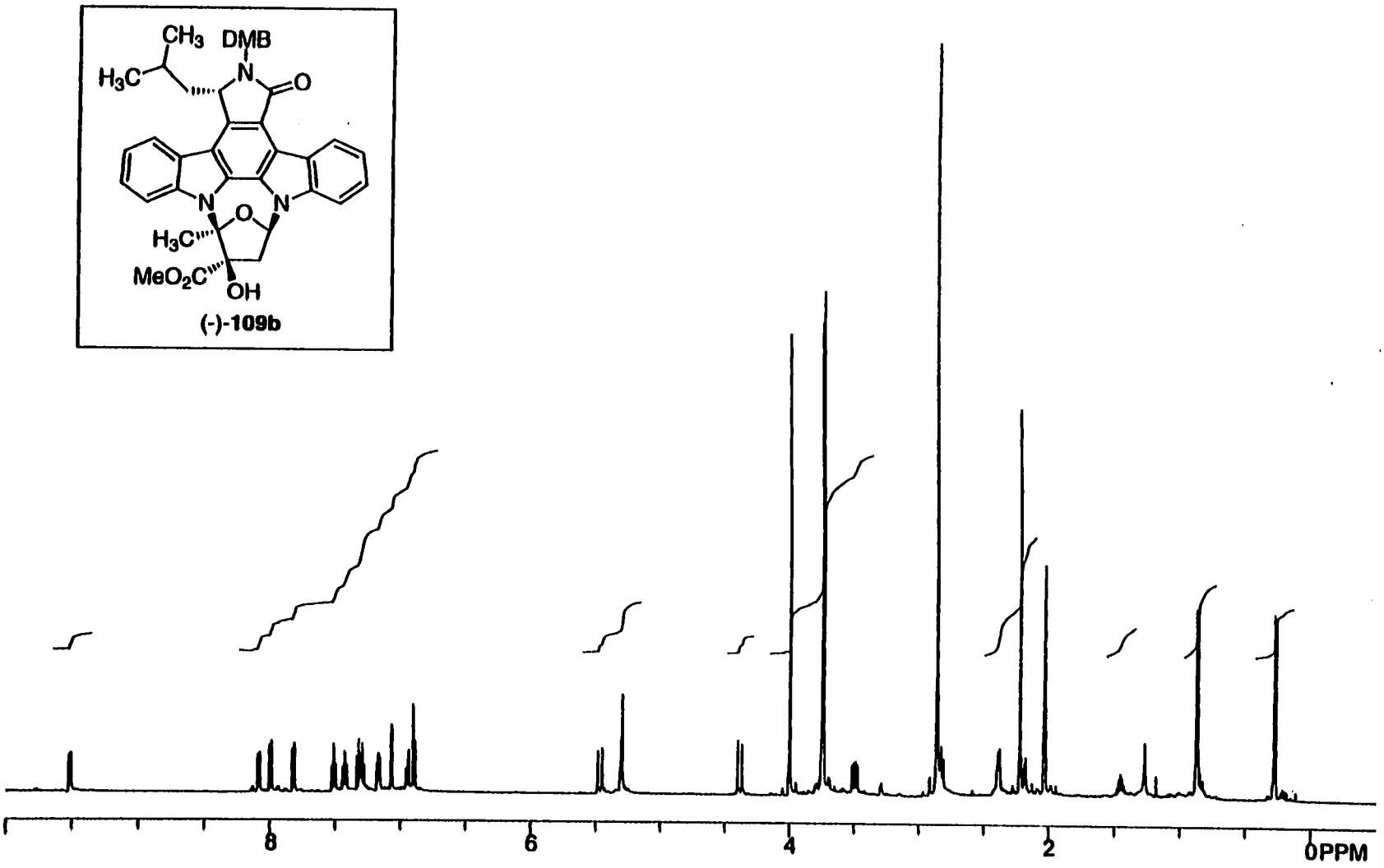


Figure A.4.55 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**109b**.

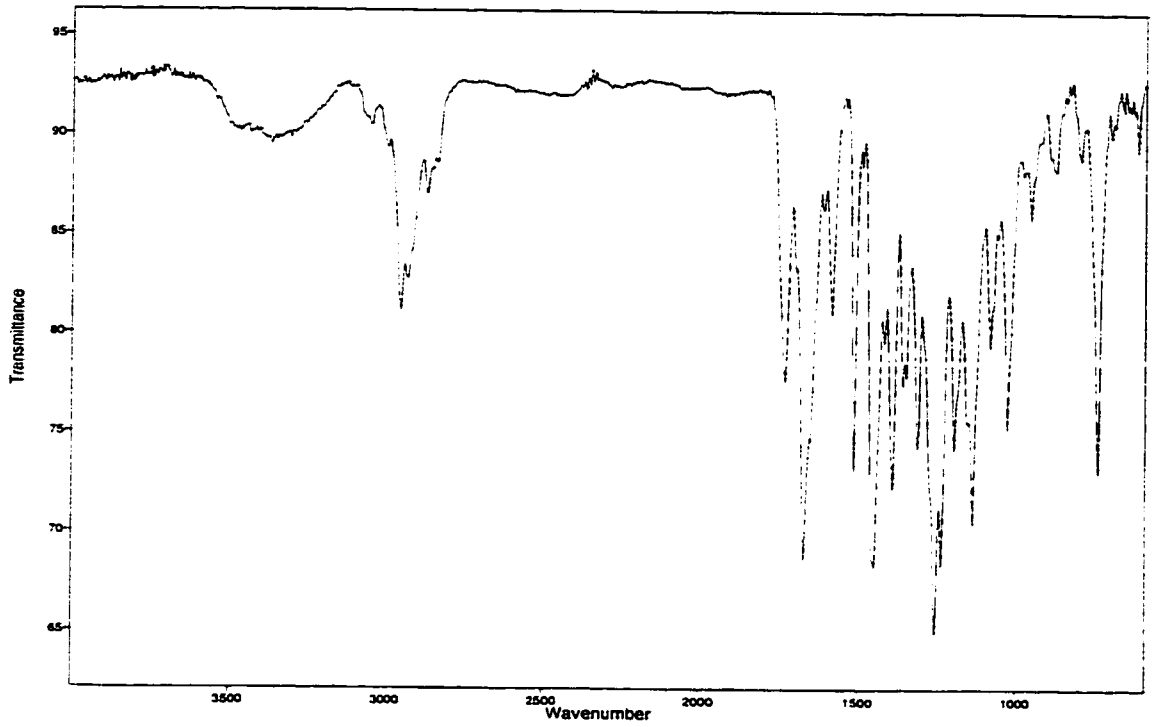


Figure A.4.56 Infrared Spectrum (thin film/NaCl) of compound (-)-**109b**.

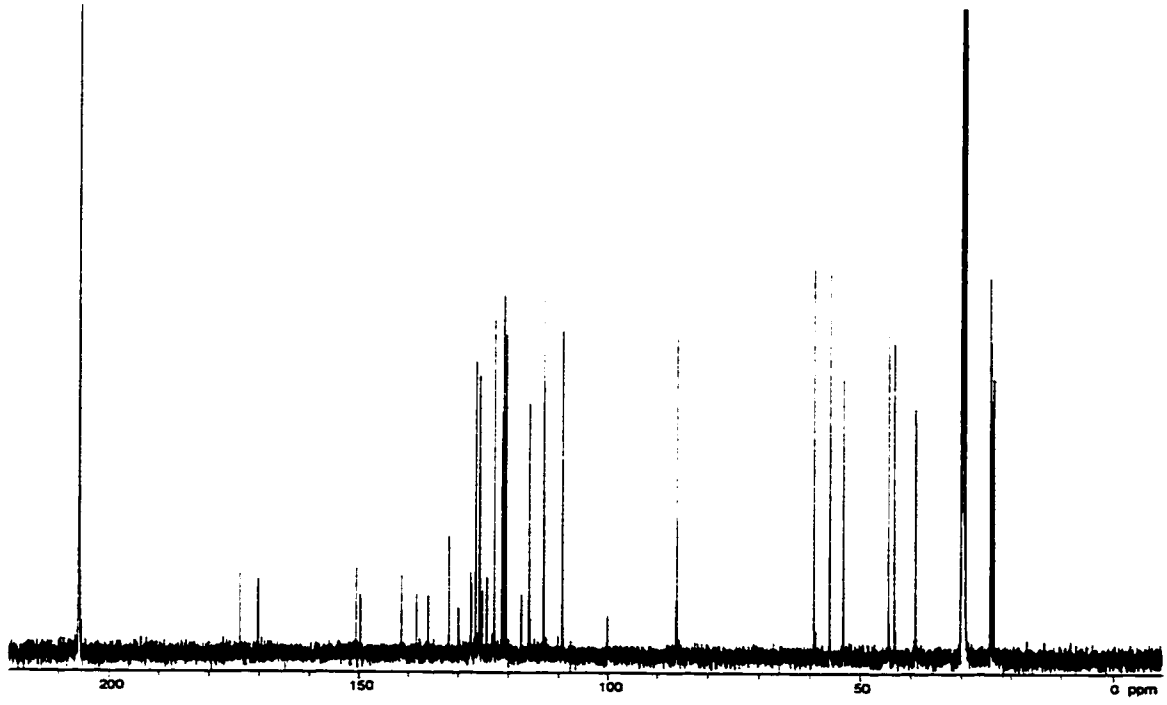
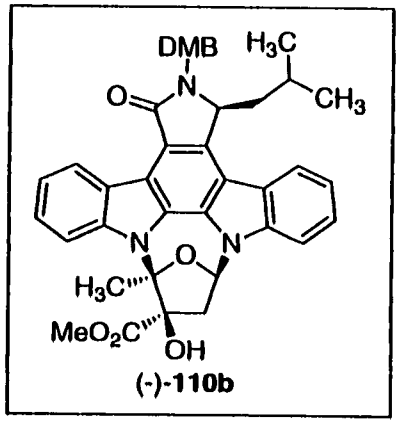


Figure A.4.57 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**109b**.



(-)-**110b**

Figure A.4.58 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**110b**.

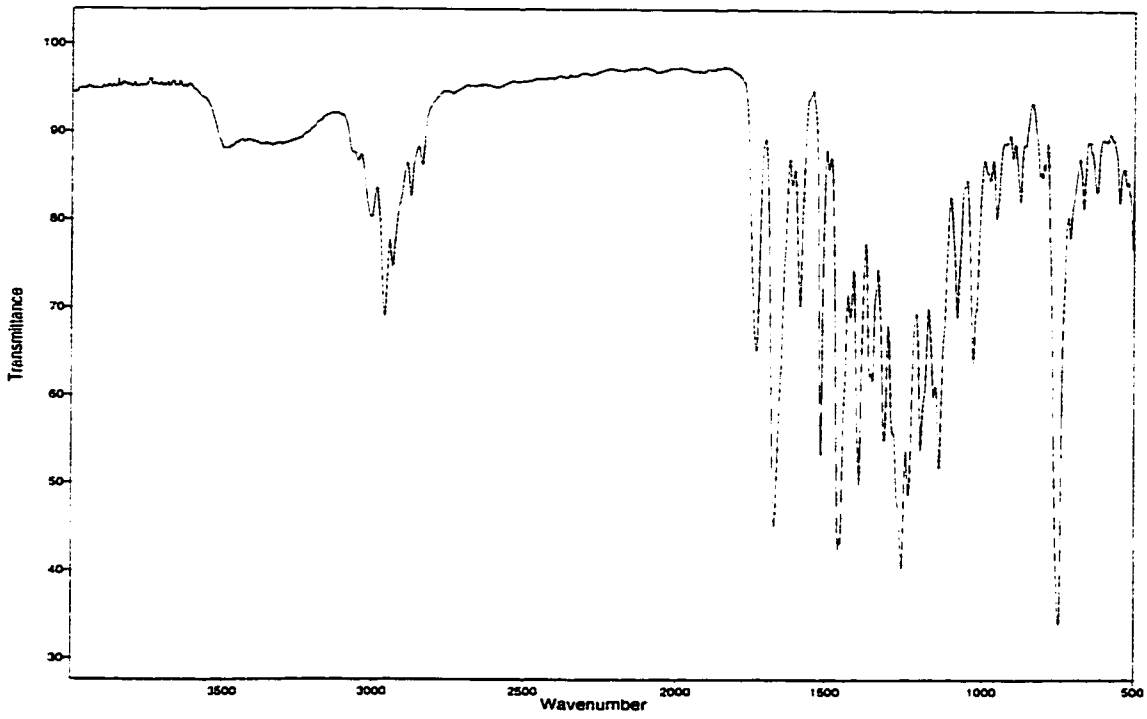


Figure A.4.59 Infrared Spectrum (thin film/NaCl) of compound (-)-**110b**.

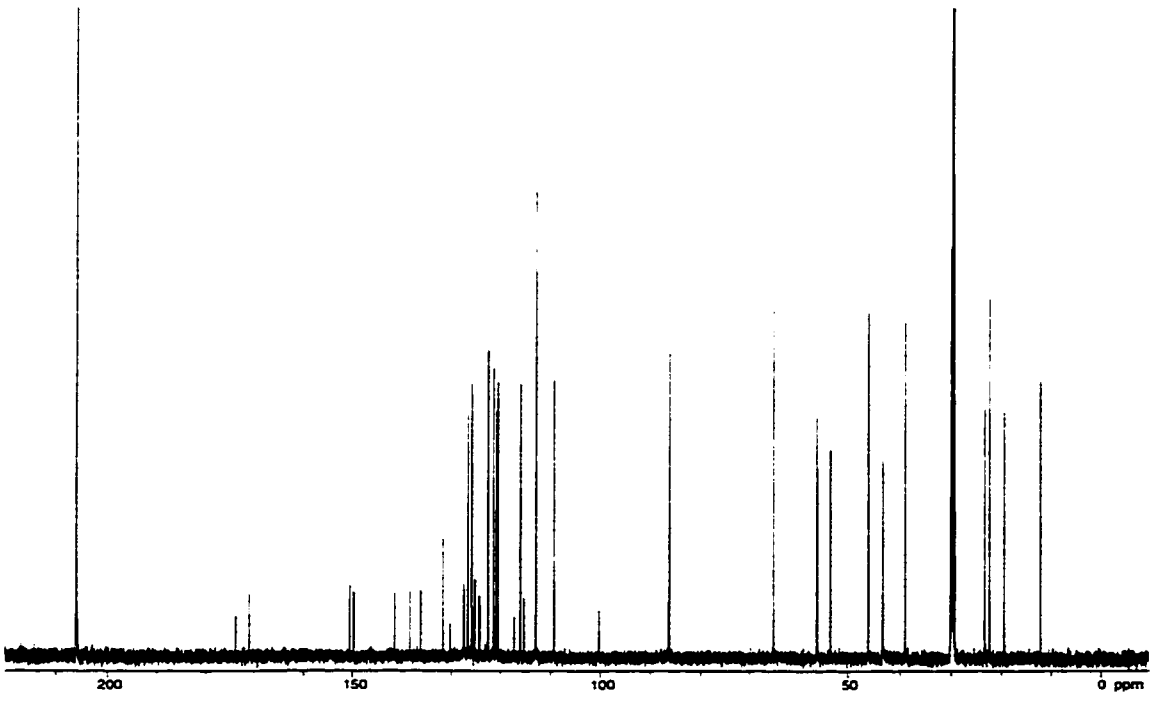


Figure A.4.60 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**110b**.

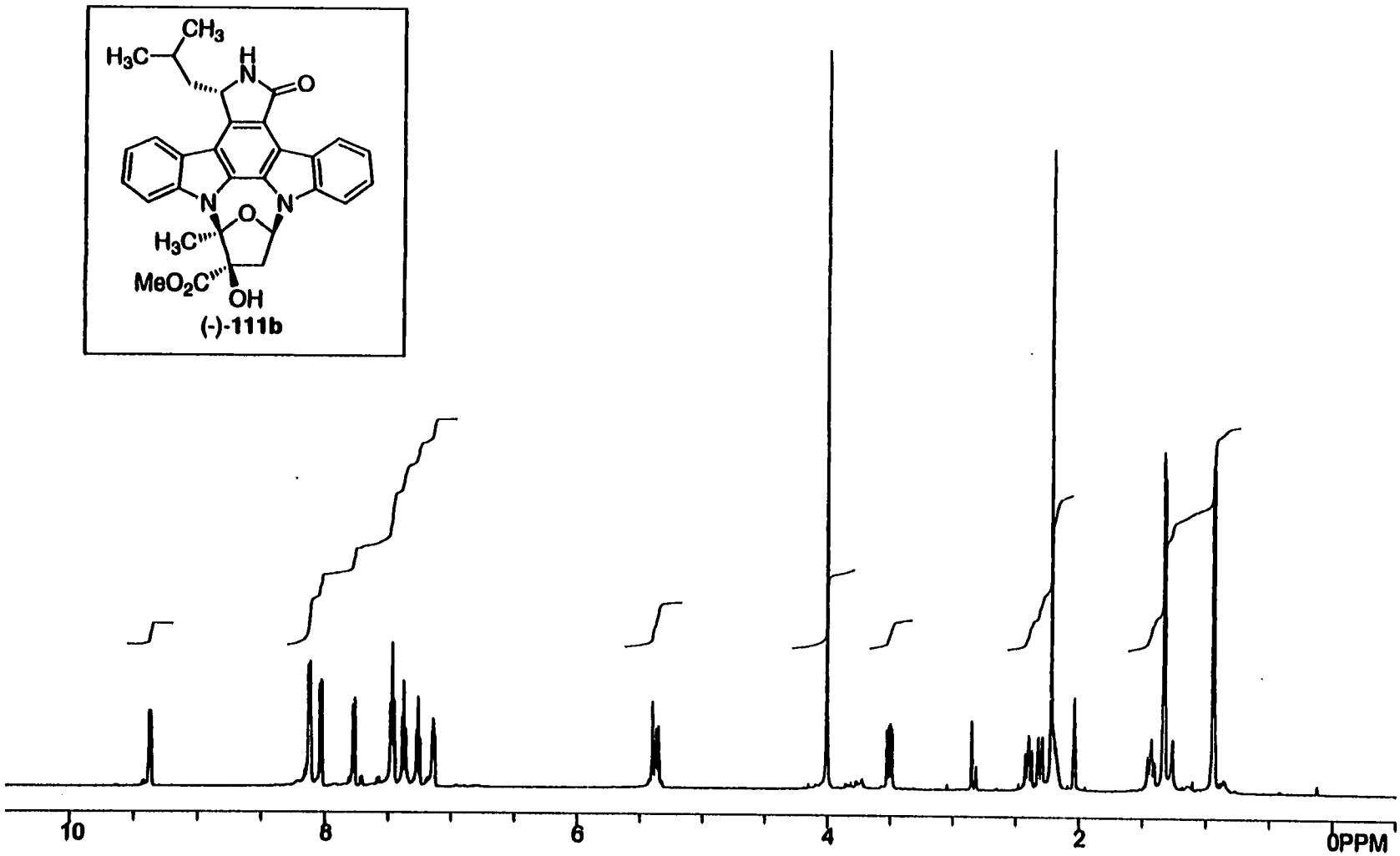


Figure A.4.61 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**111b**.

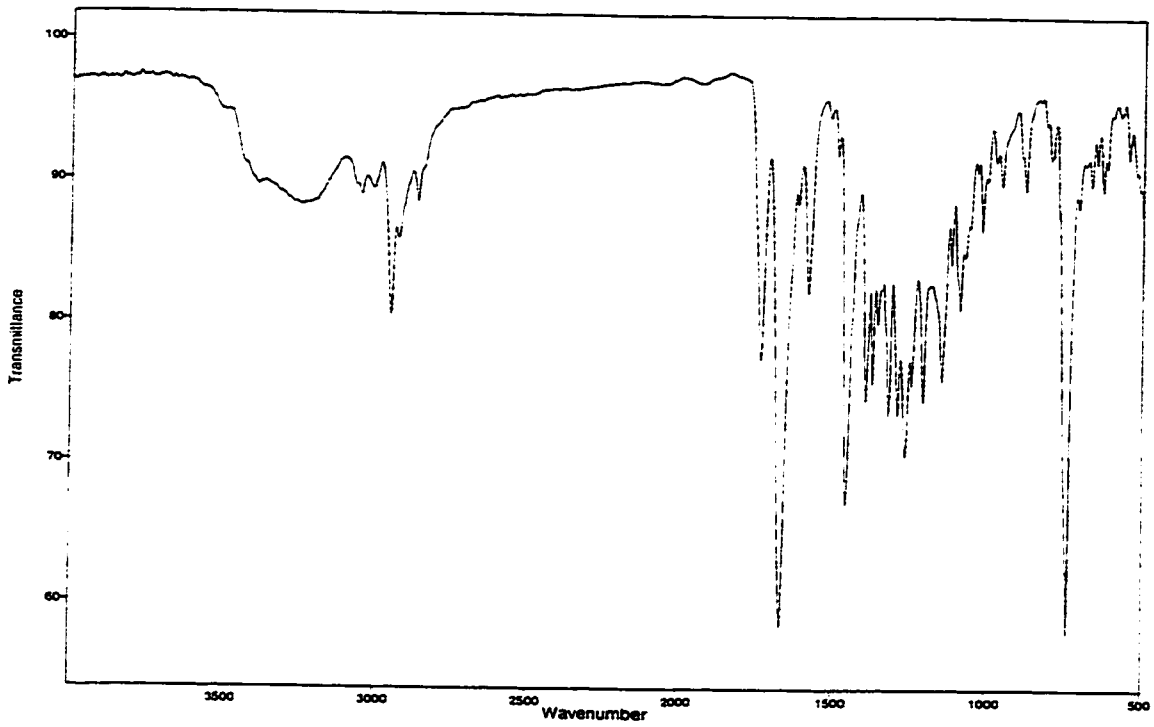


Figure A.4.62 Infrared Spectrum (thin film/NaCl) of compound (-)-**111b**.

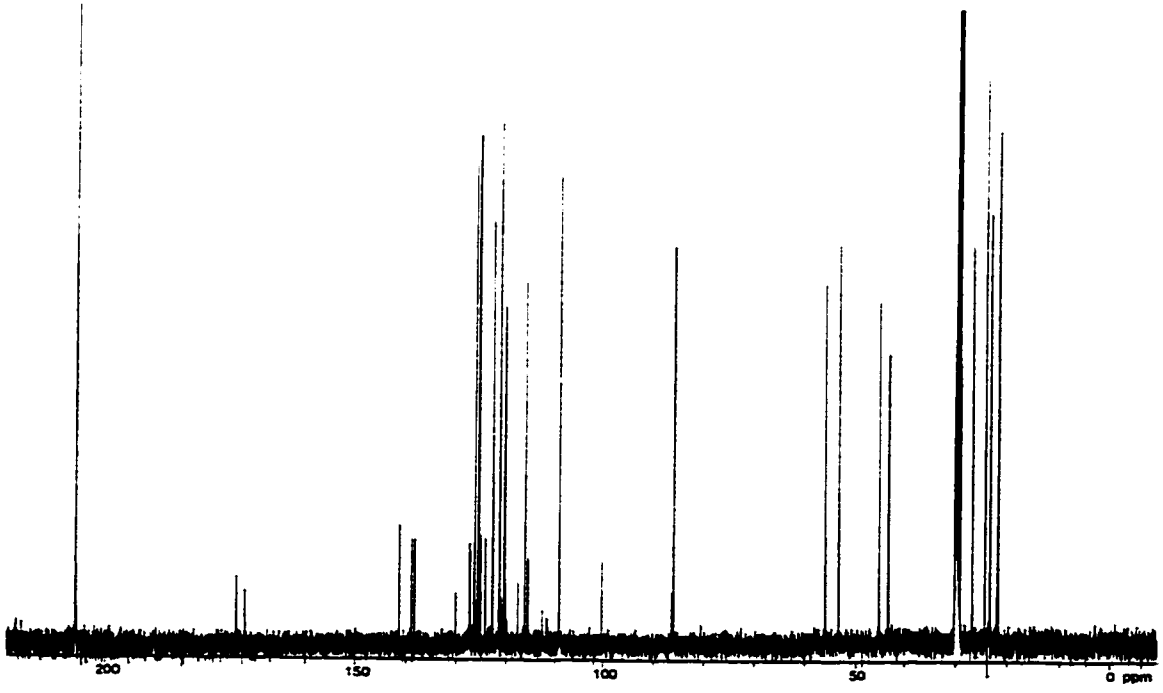


Figure A.4.63 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**111b**.

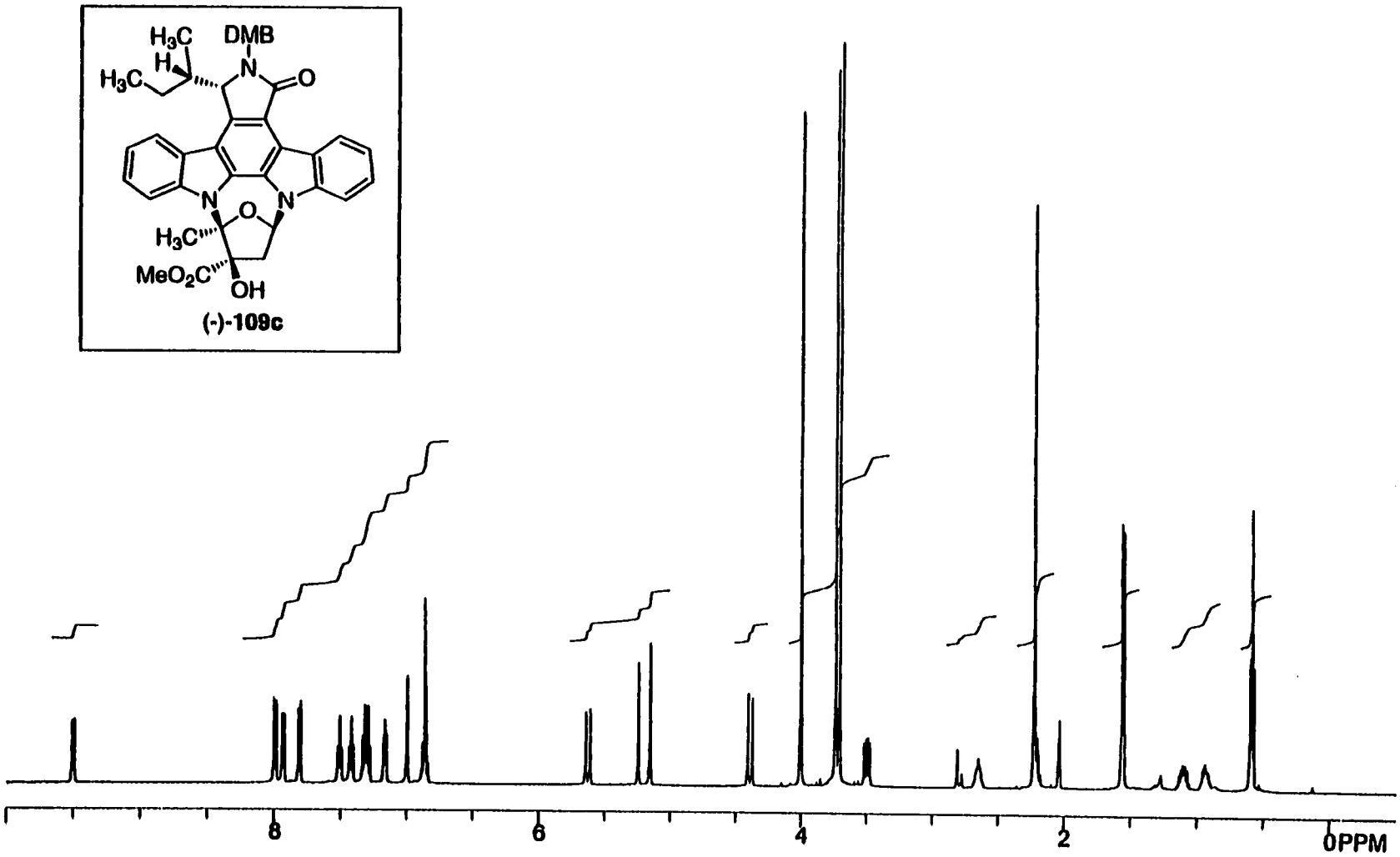


Figure A.4.64 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**109c**.

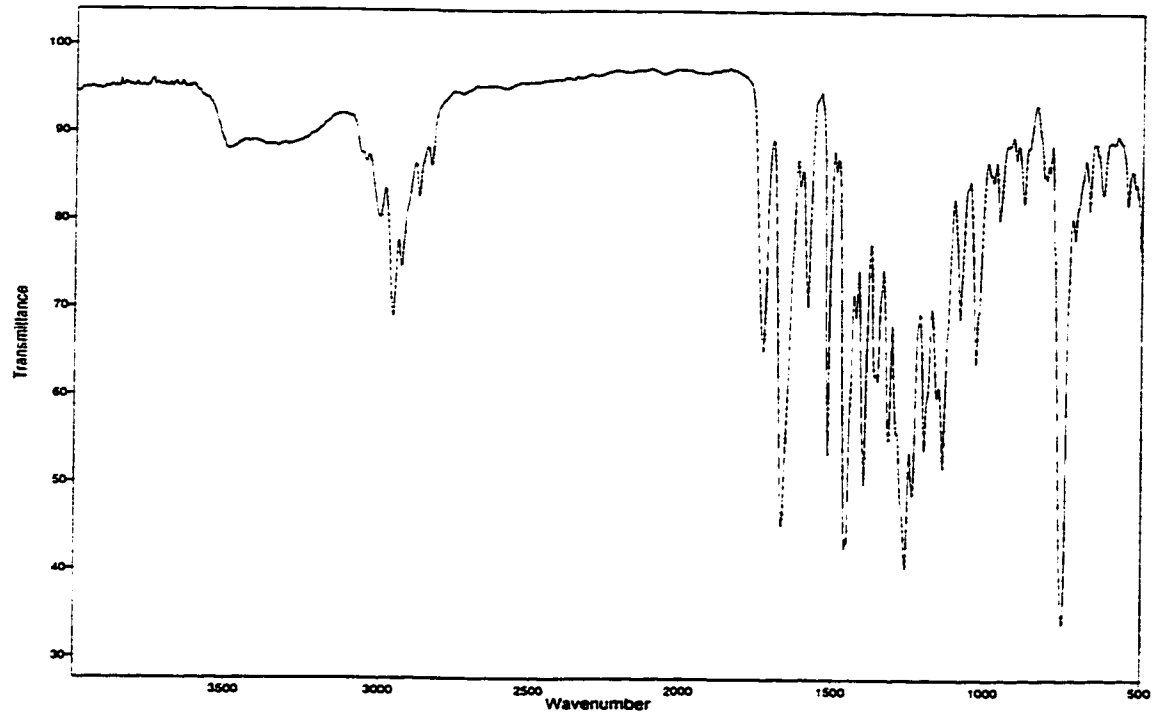


Figure A.4.65 Infrared Spectrum (thin film/NaCl) of compound (-)-**109c**.

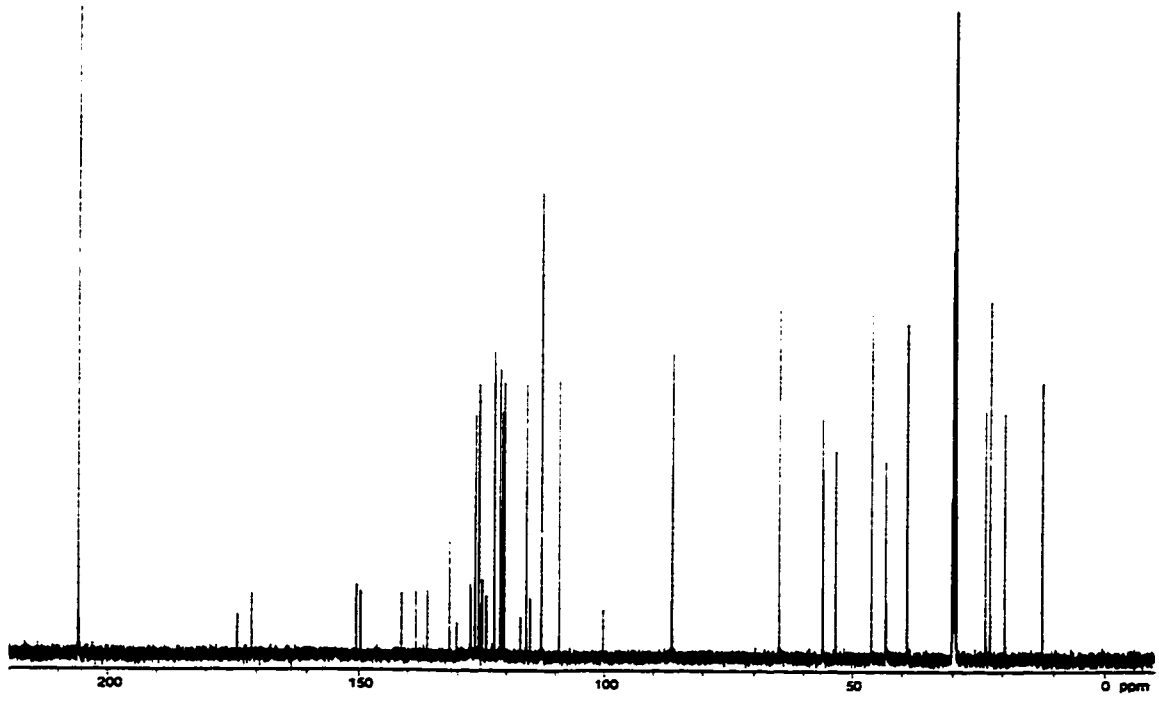
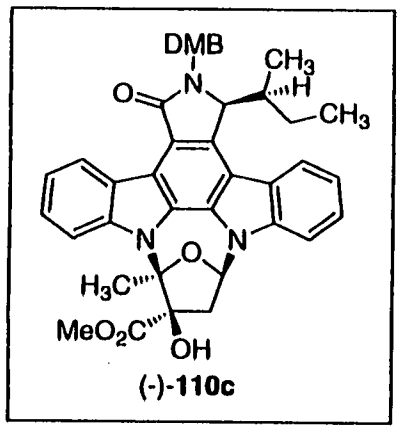


Figure A.4.66 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**109c**.



(-)-**110c**

Figure A.4.67 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**110c**.

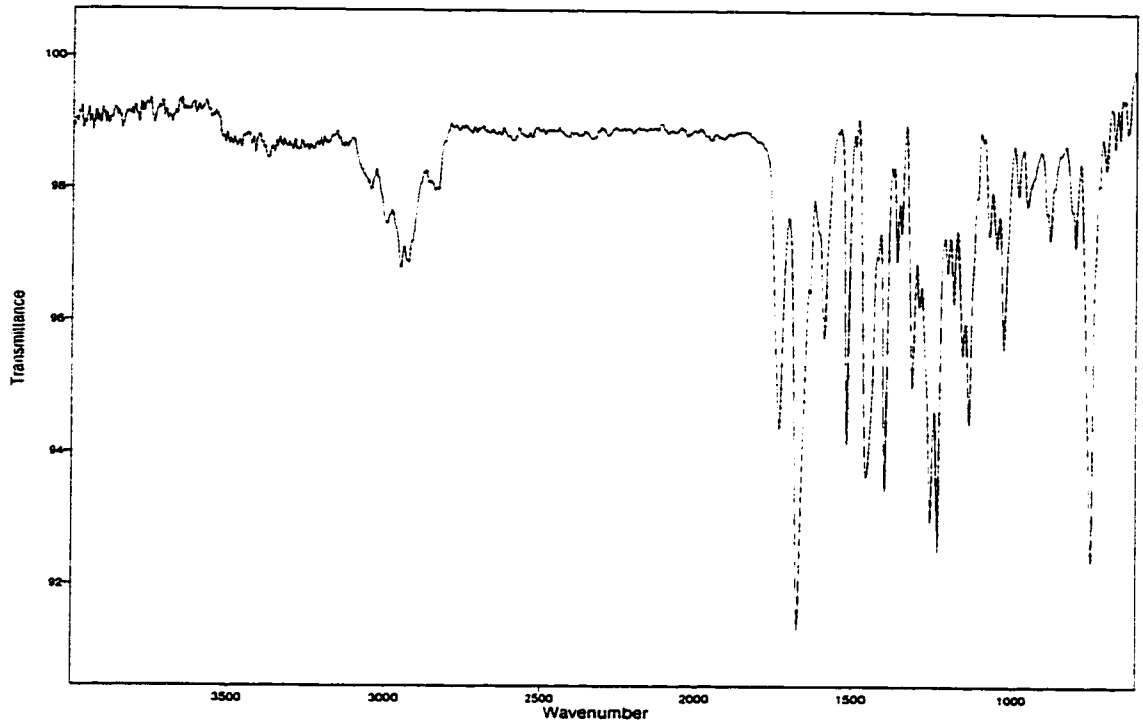


Figure A.4.68 Infrared Spectrum (thin film/NaCl) of compound (-)-**110c**.

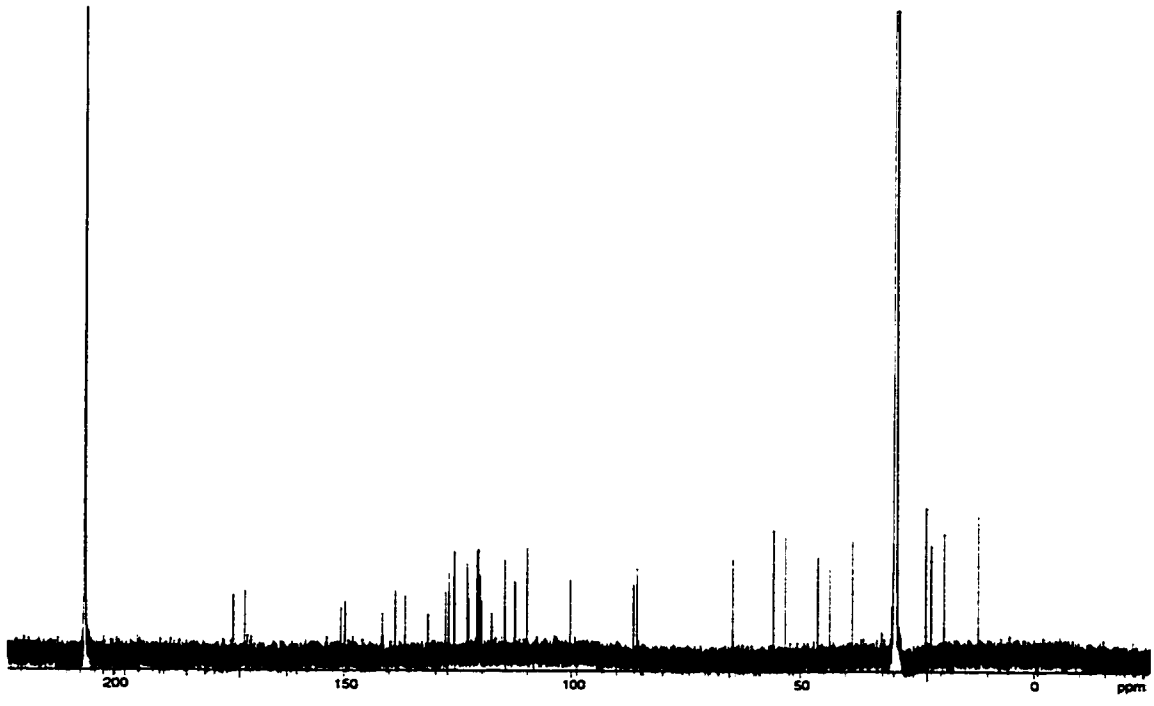


Figure A.4.69 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**110c**.

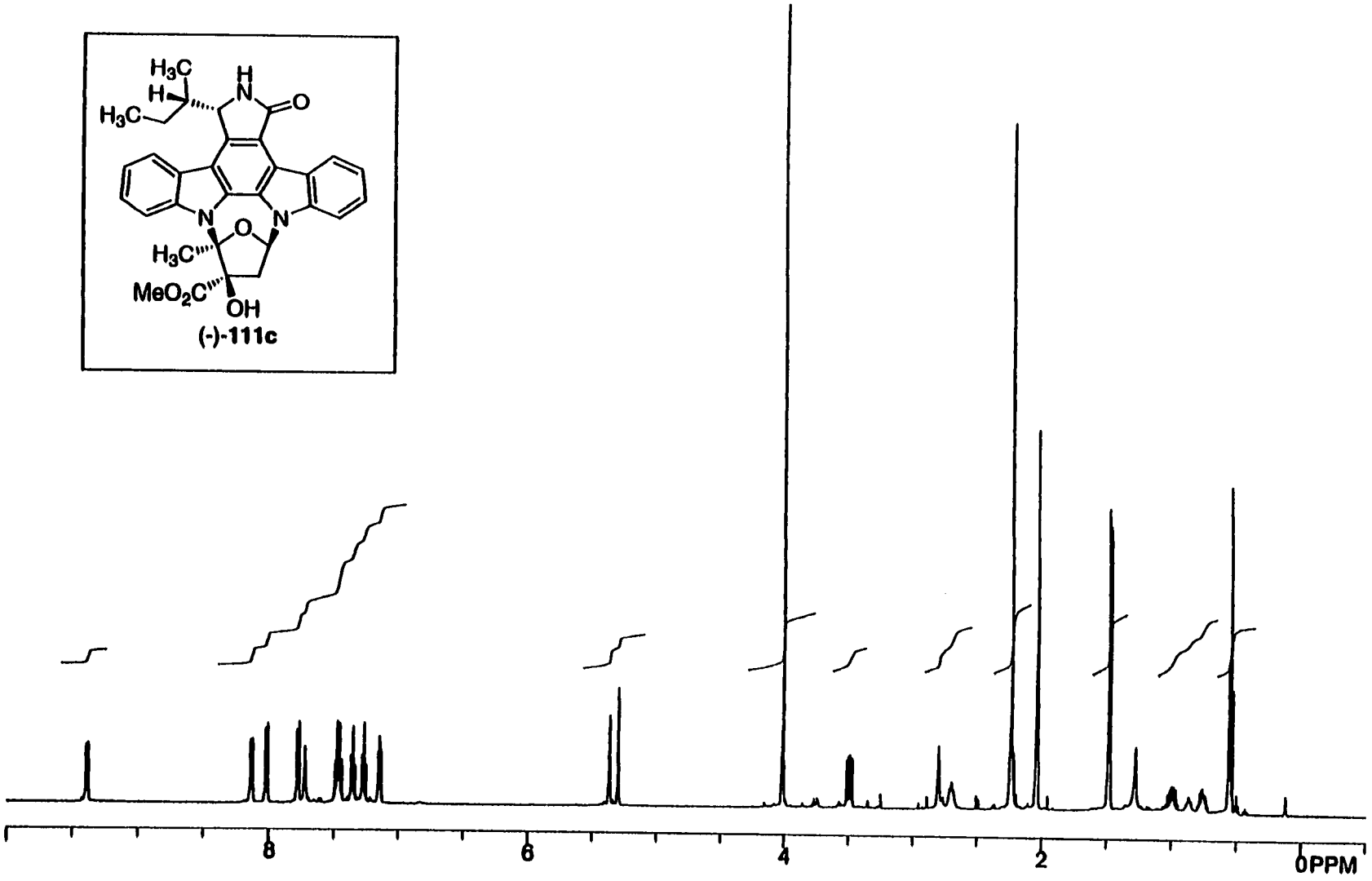


Figure A.4.70 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**111c**.

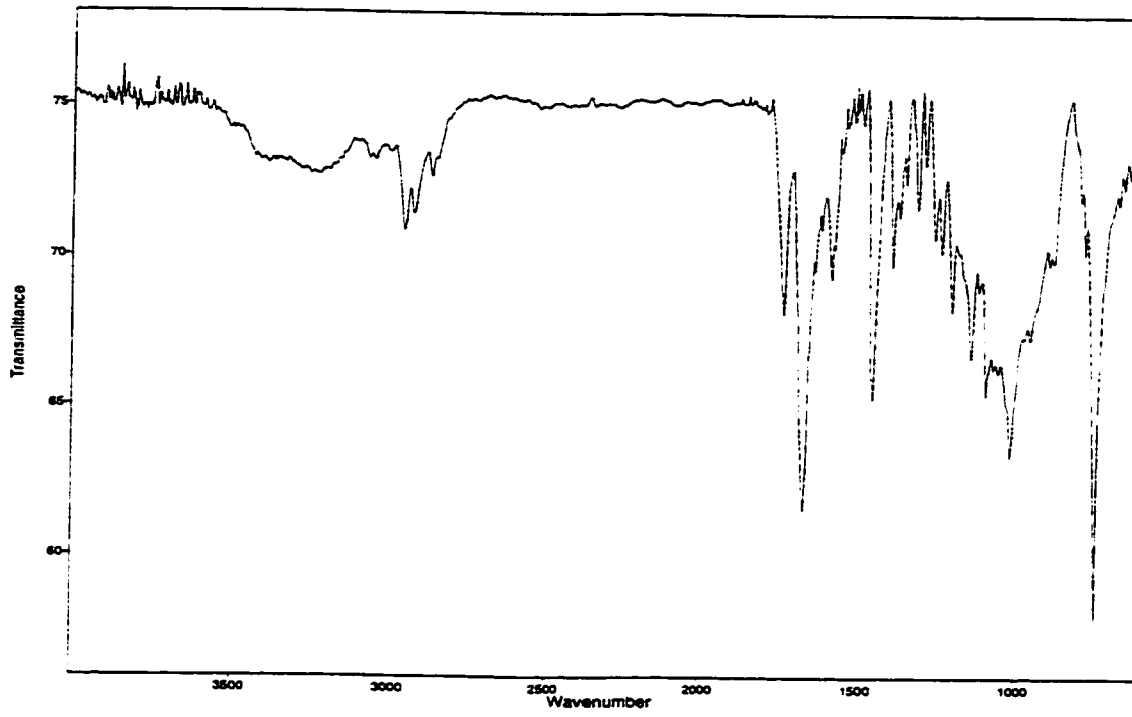


Figure A.4.71 Infrared Spectrum (thin film/NaCl) of compound (-)-**111c**.

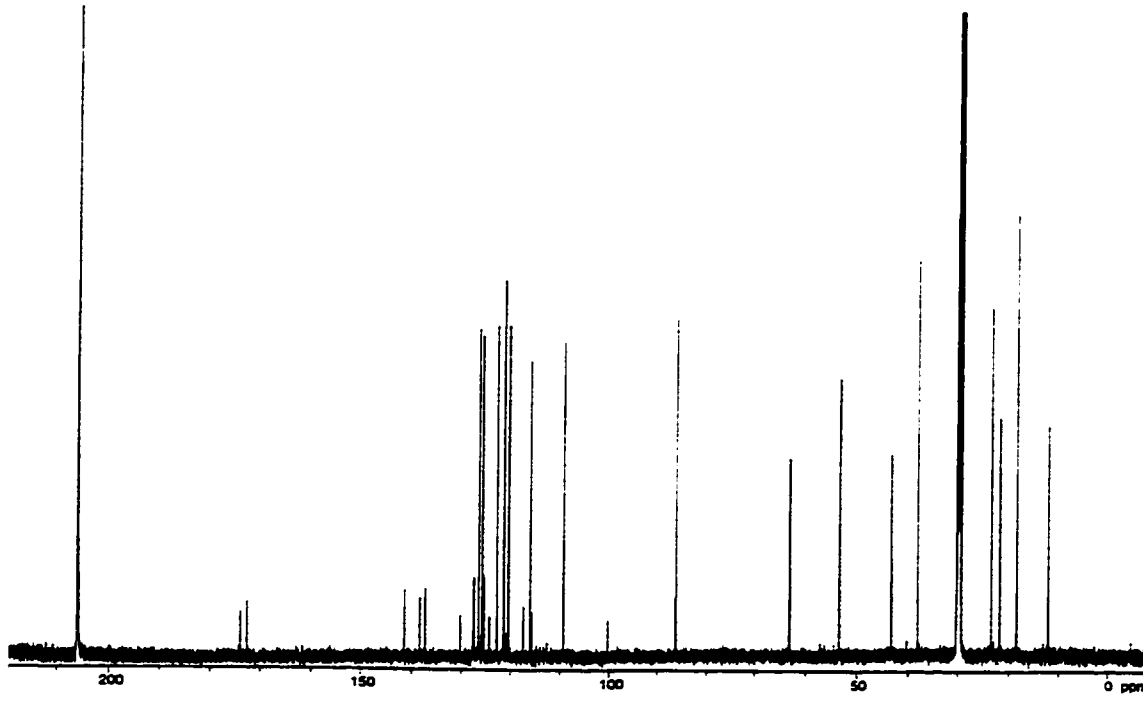


Figure A.4.72 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**111c**.

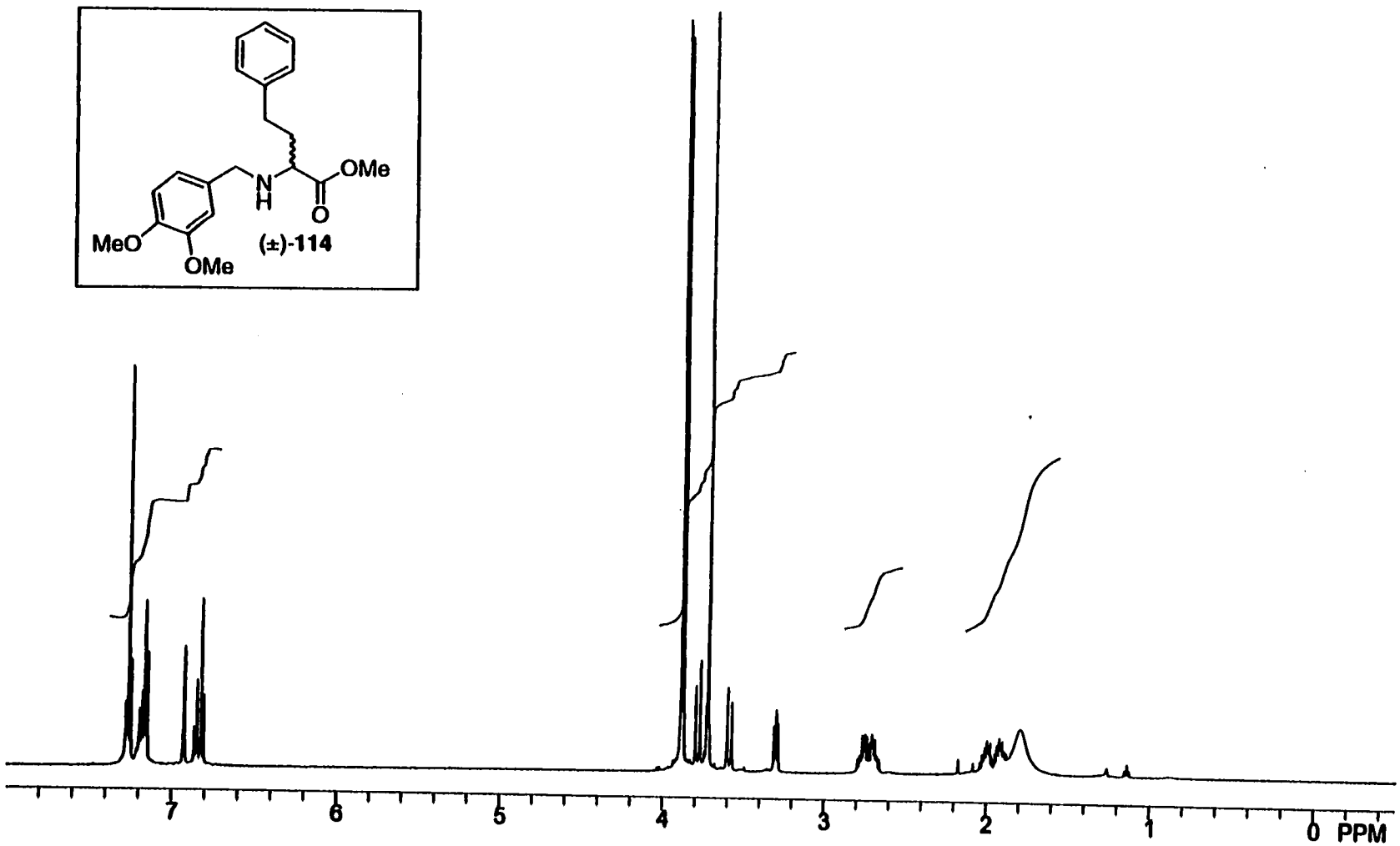


Figure A.4.73 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**114**.

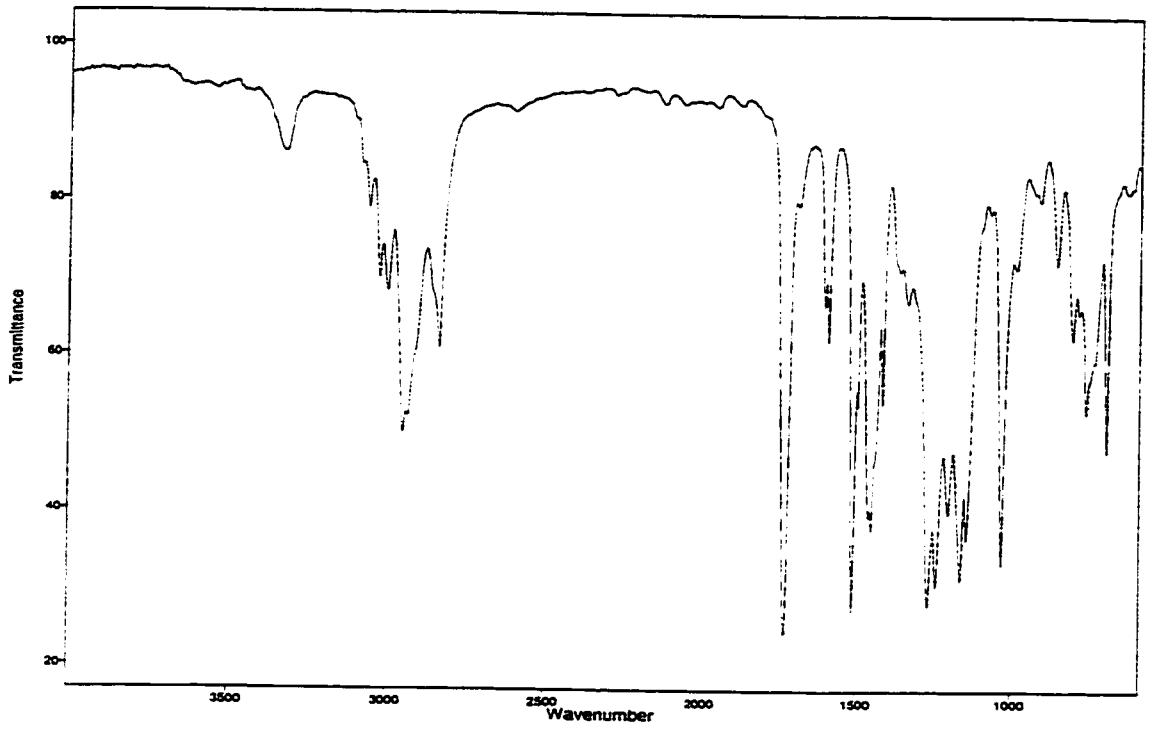


Figure A.4.74 Infrared Spectrum (thin film/NaCl) of compound (±)-**114**.

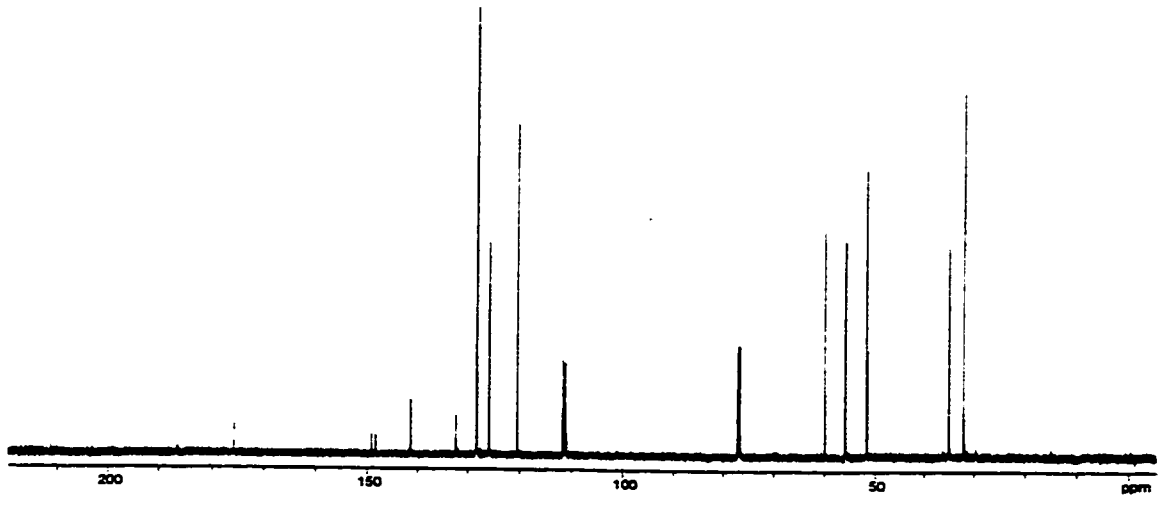


Figure A.4.75 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**114**.

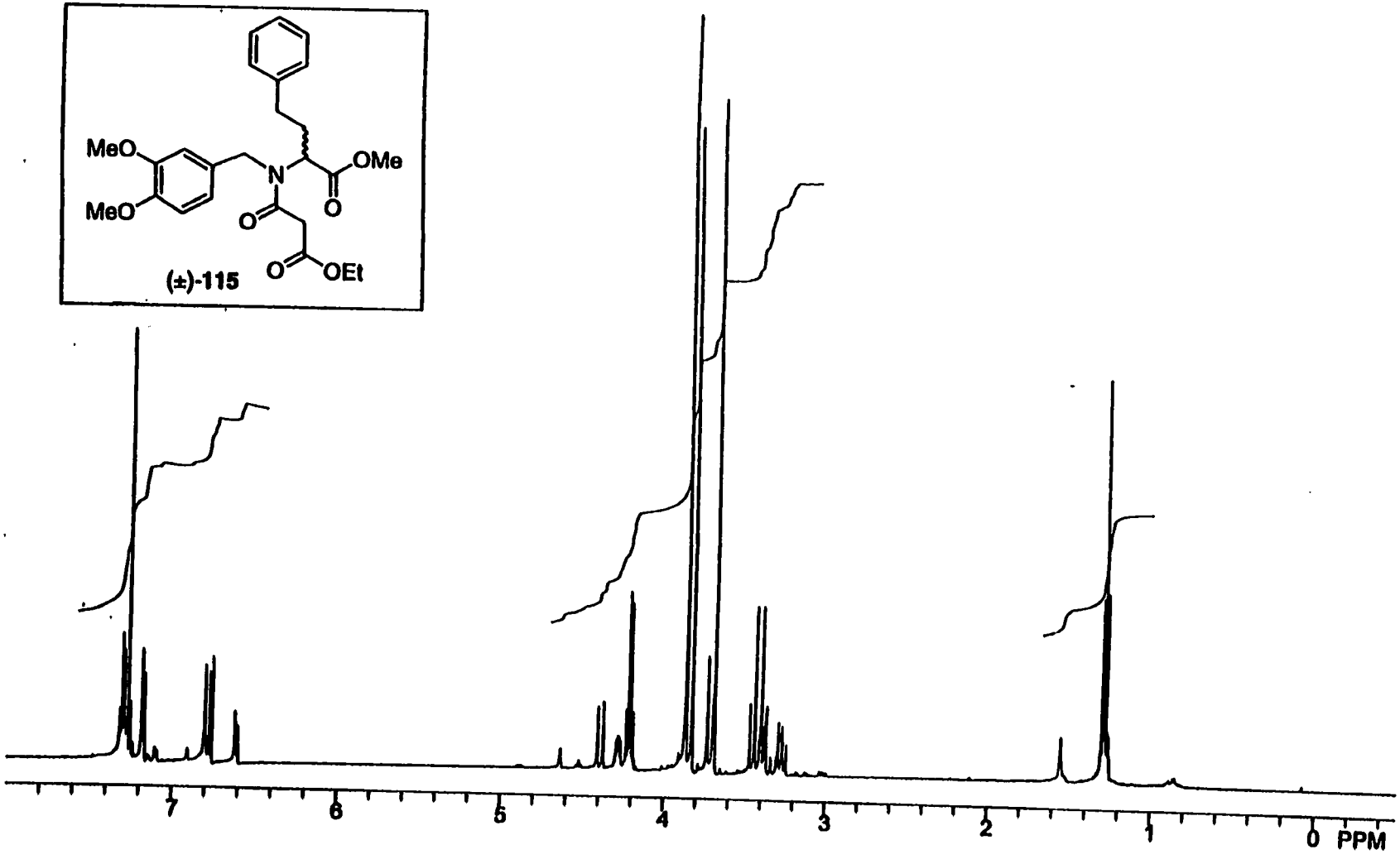


Figure A.4.76 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-115.

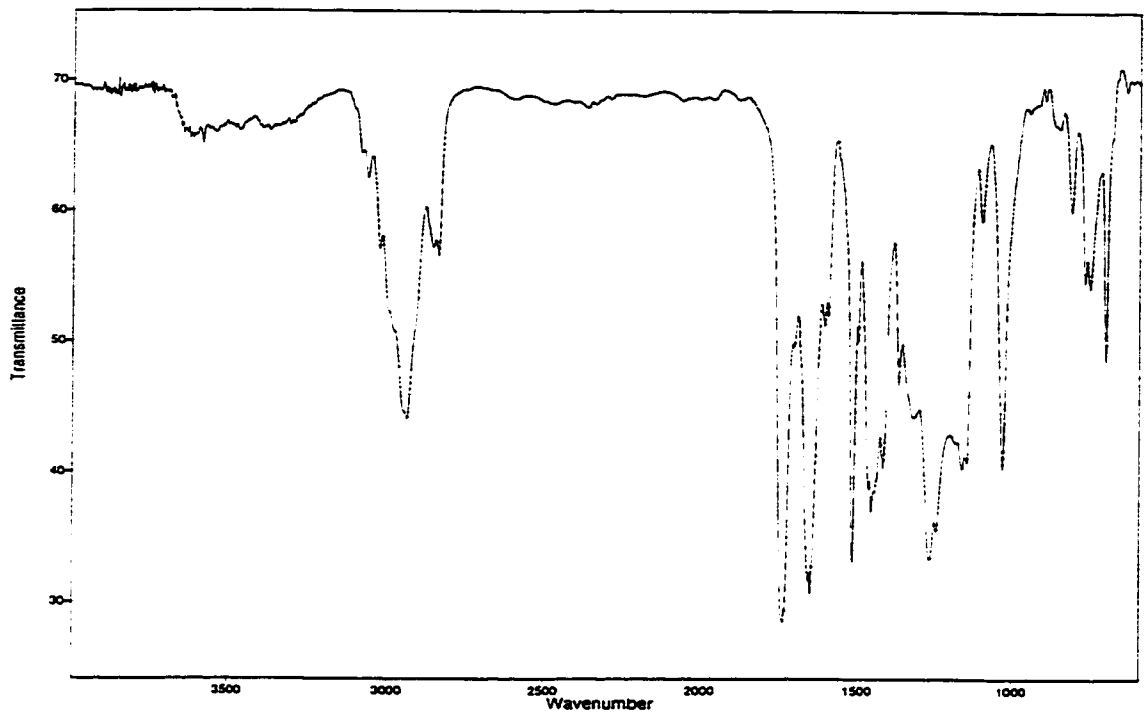


Figure A.4.77 Infrared Spectrum (thin film/NaCl) of compound (±)-**115**.

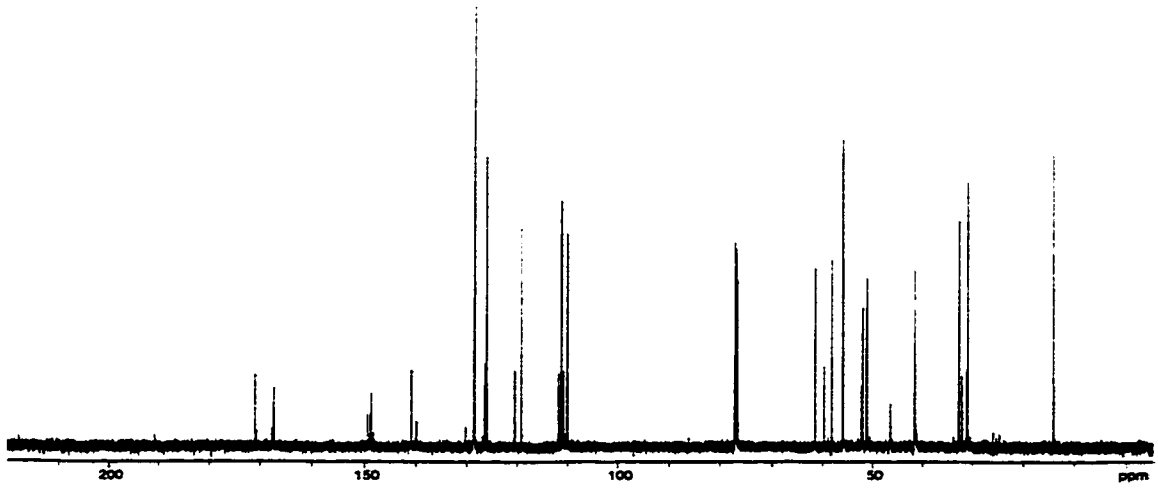


Figure A.4.78 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**115**.

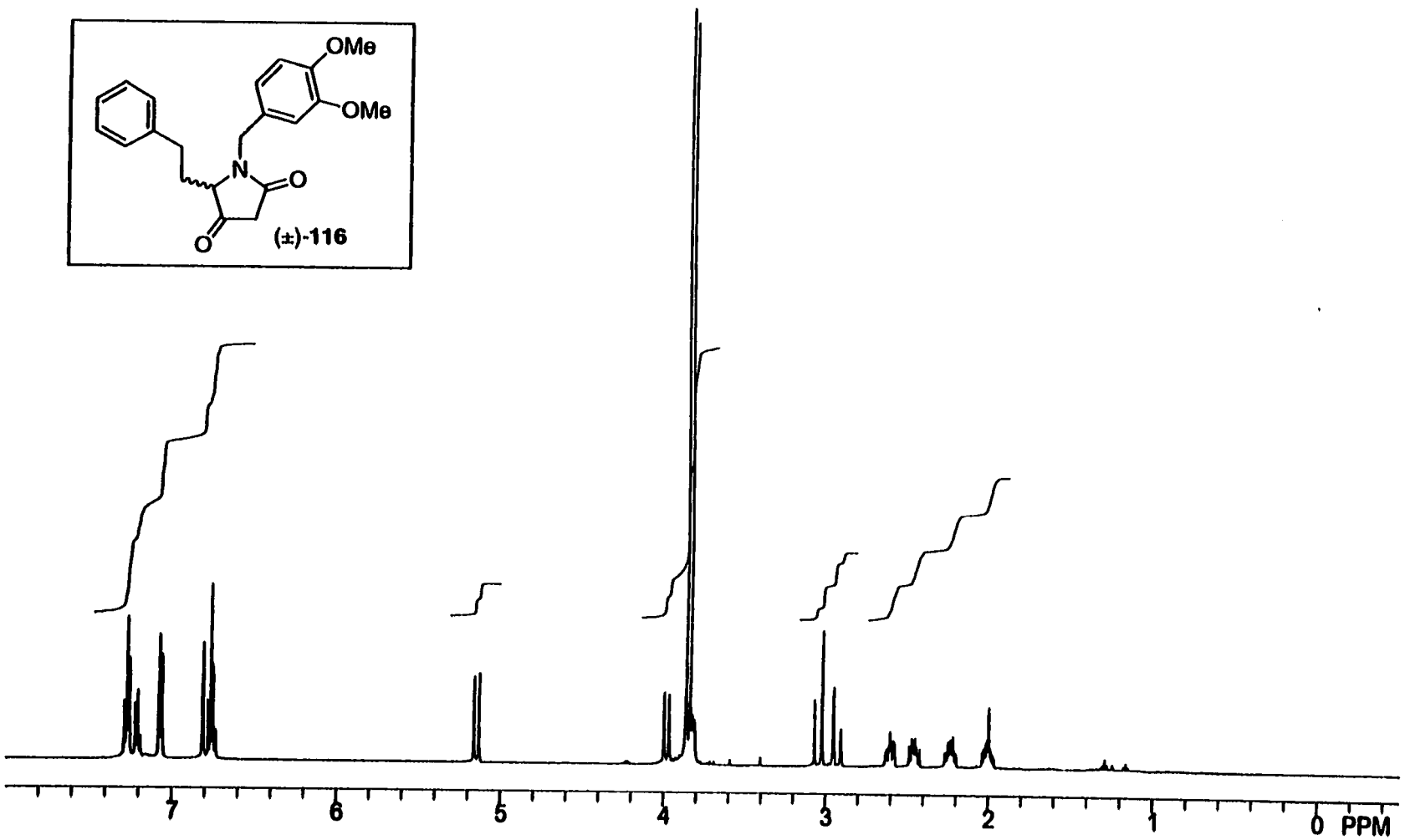


Figure A.4.79 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**116**.

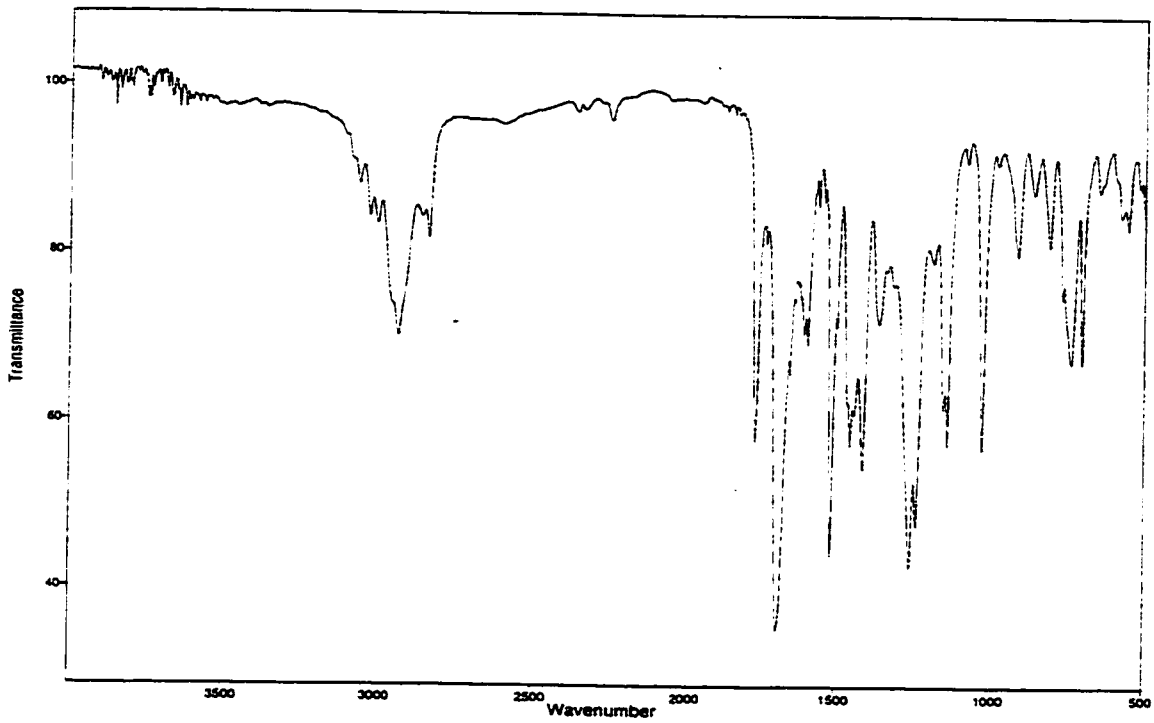


Figure A.4.80 Infrared Spectrum (thin film/NaCl) of compound (±)-**116**.

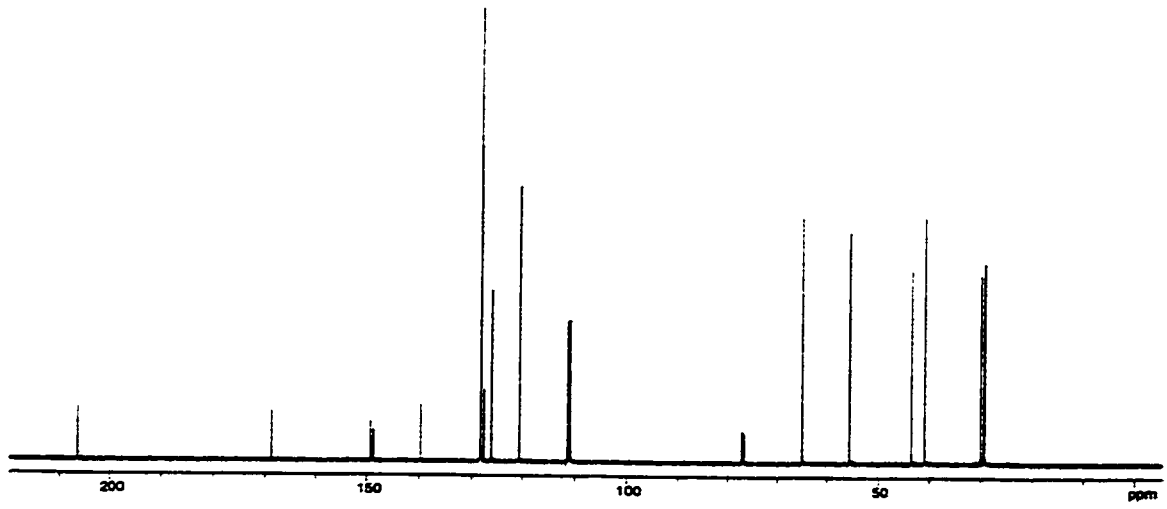


Figure A.4.81 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**116**.

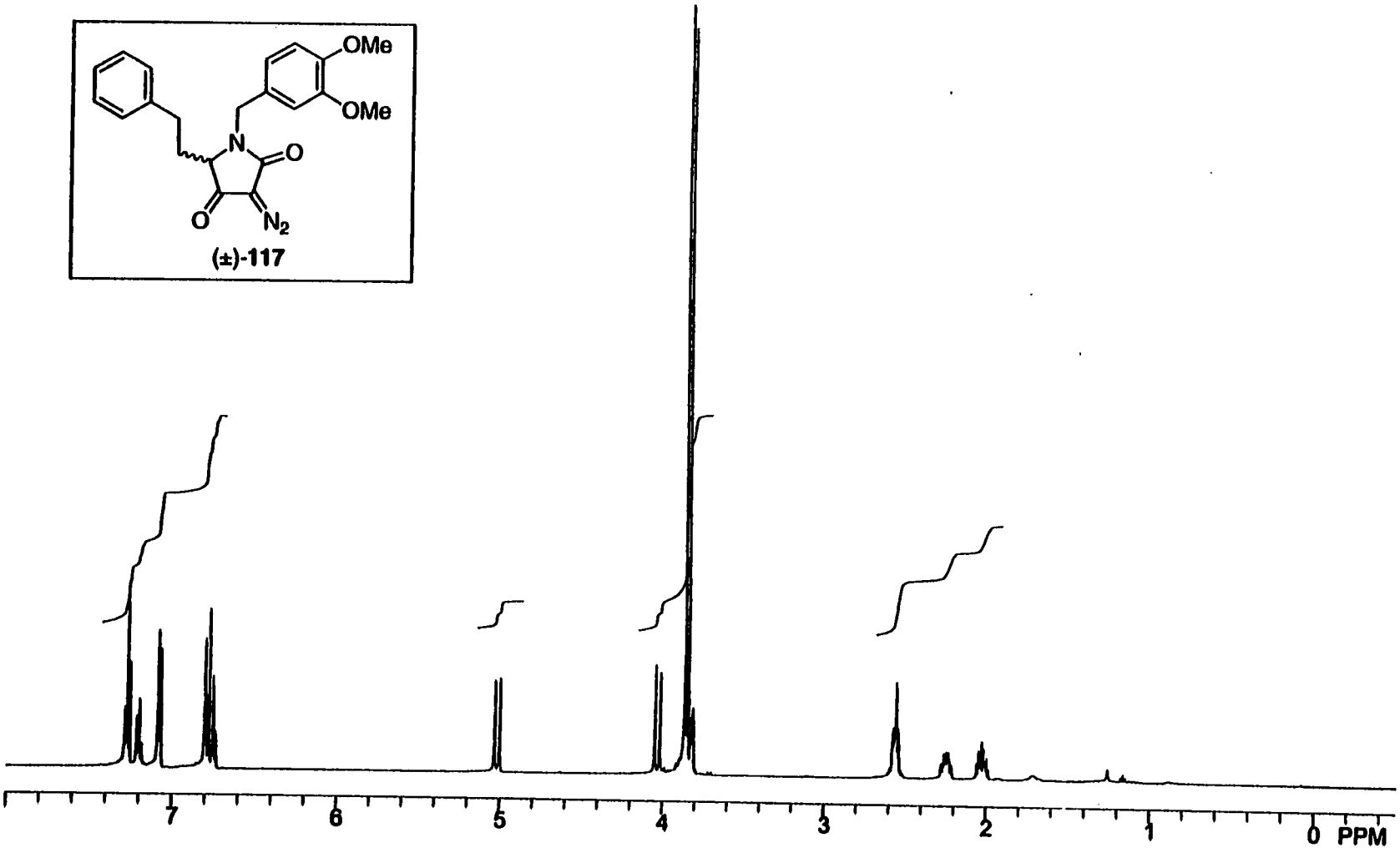


Figure A.4.82 ^{1}H NMR (500 MHz, $CDCl_3$) of compound (±)-**117**.

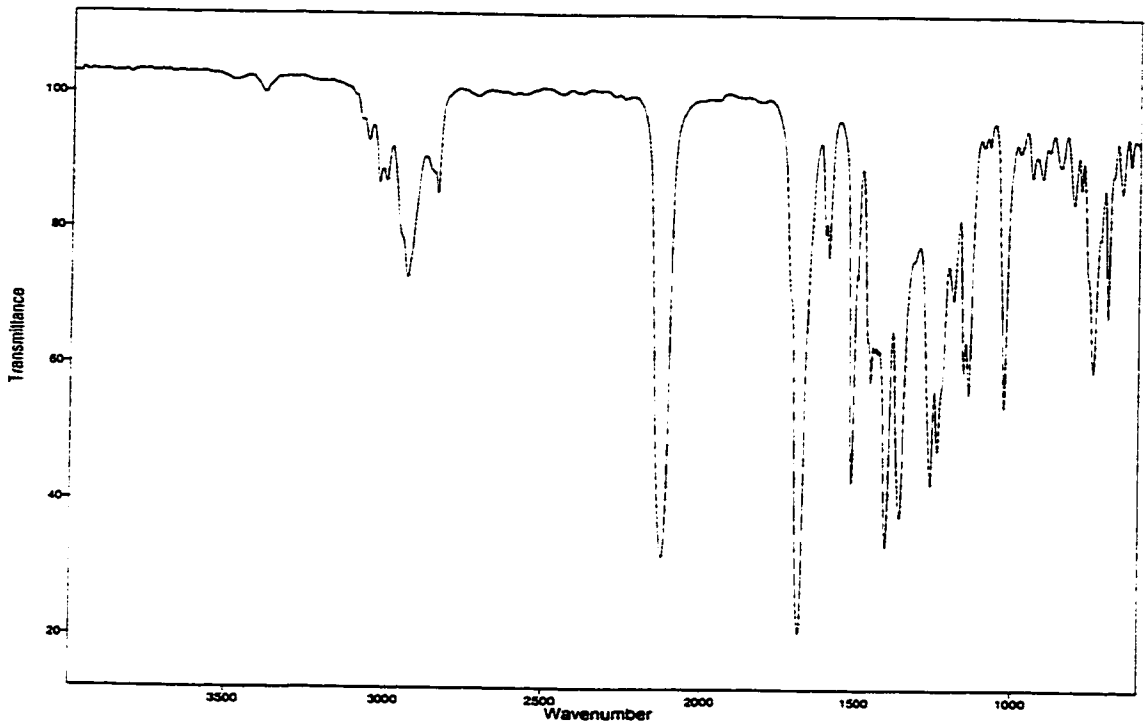


Figure A.4.83 Infrared Spectrum (thin film/NaCl) of compound (±)-**117**.

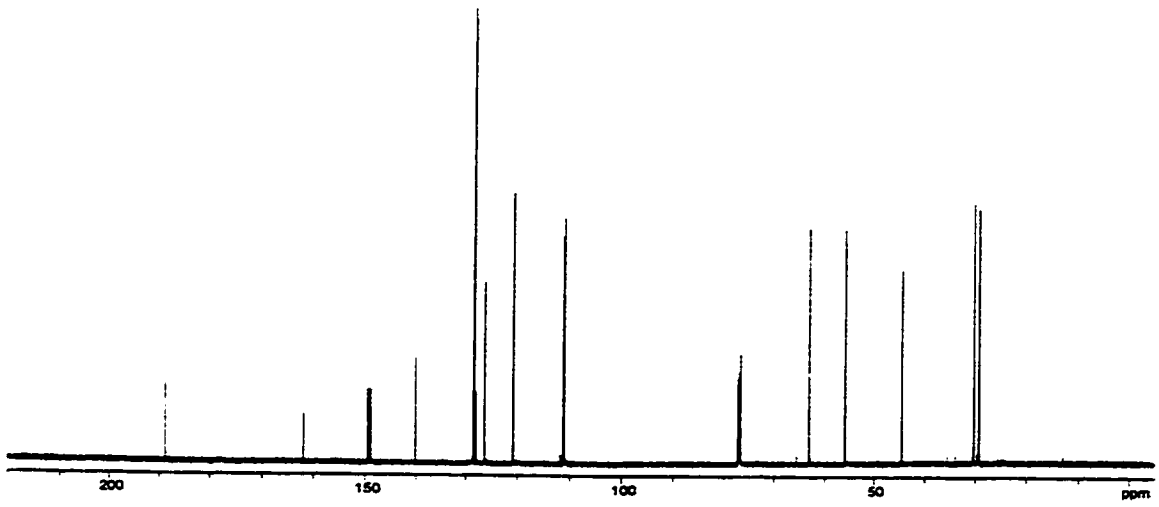


Figure A.4.84 ^{13}C NMR (125 MHz, $CDCl_3$) of compound (±)-**117**.

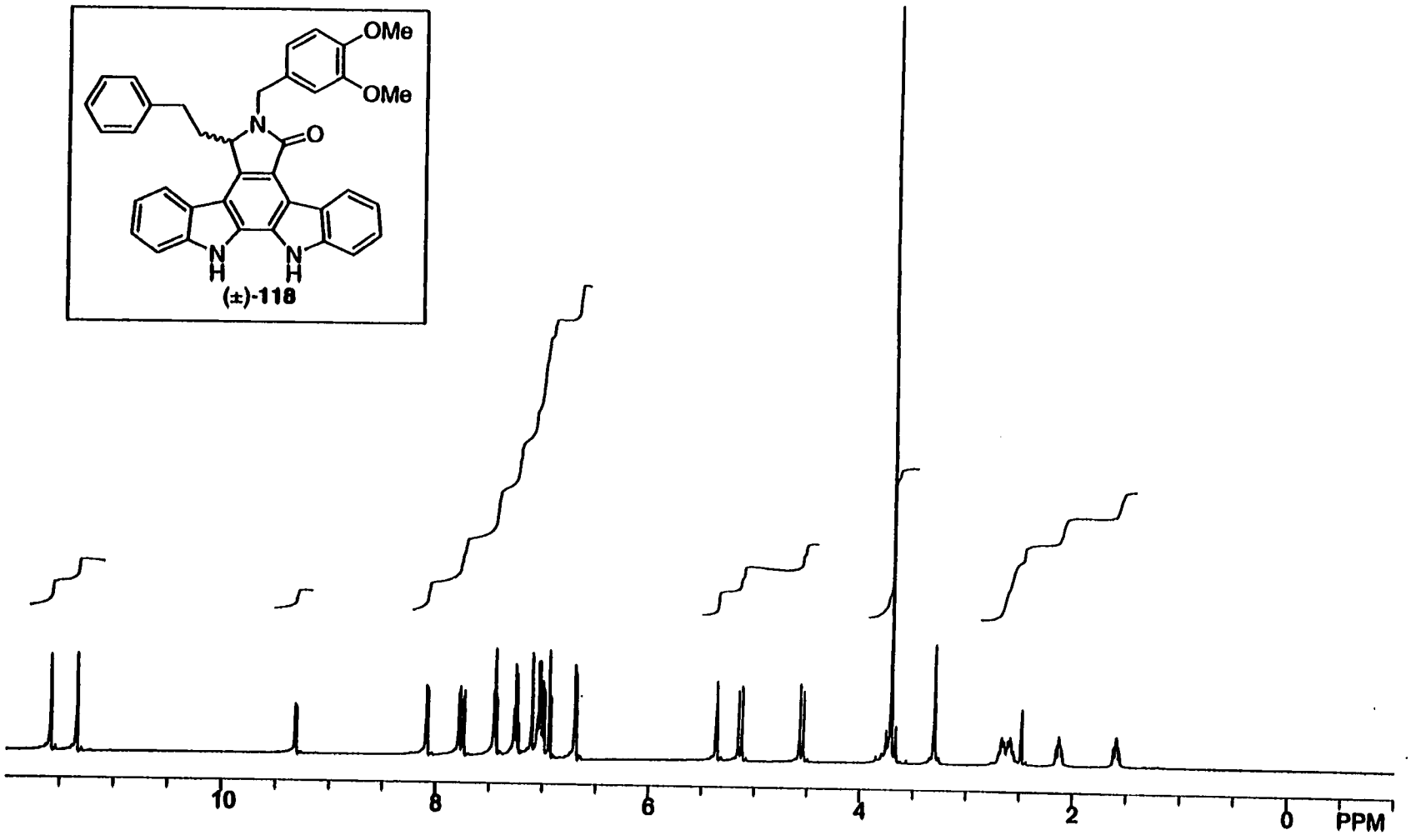


Figure A.4.85 ^{1}H NMR (500 MHz, DMSO-d_6) of compound (±)-**118**.

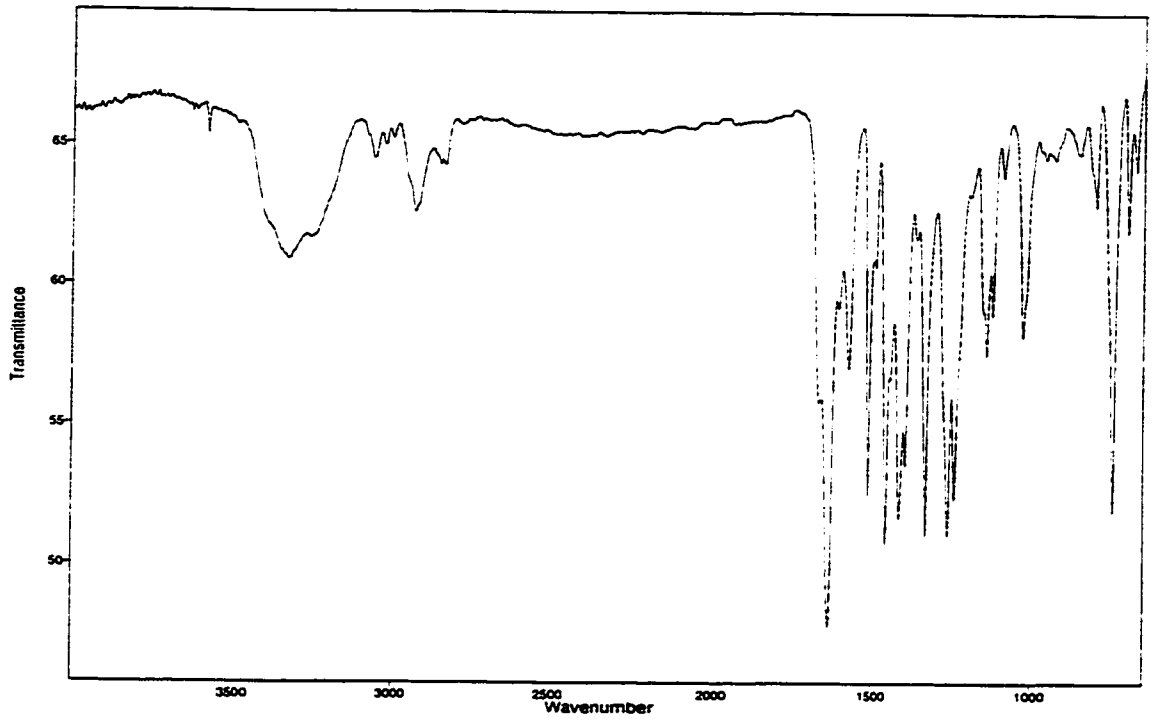


Figure A.4.86 Infrared Spectrum (thin film/NaCl) of compound (±)-**118**.

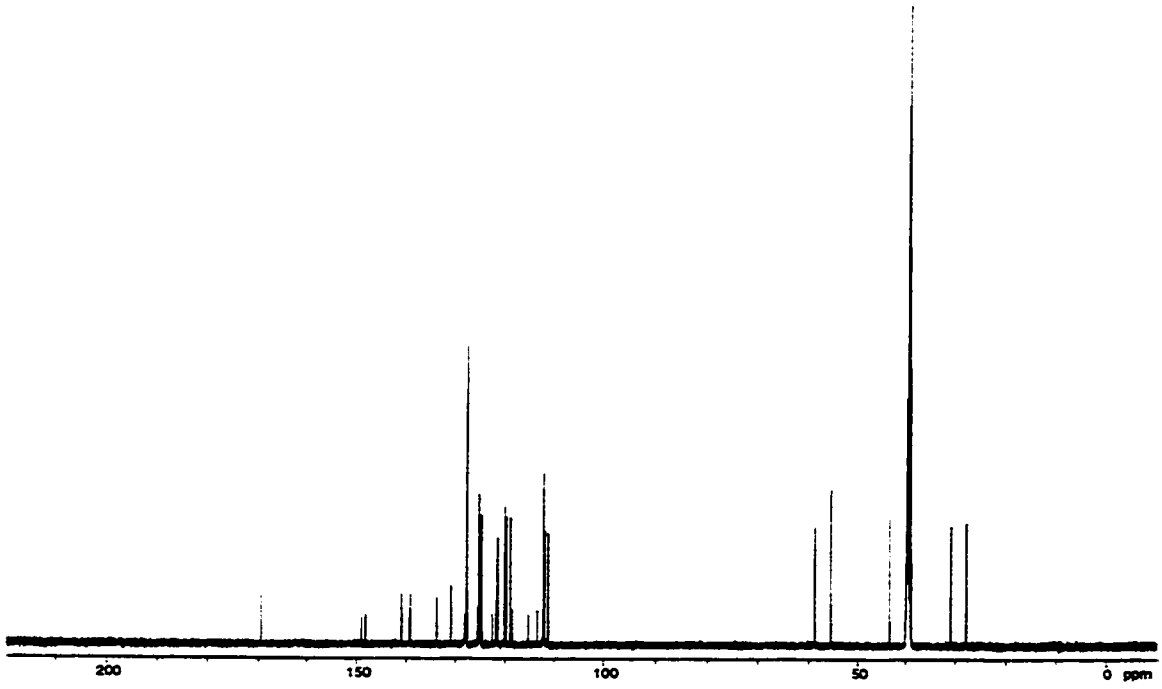


Figure A.4.87 ^{13}C NMR (125 MHz, DMSO-d_6) of compound (±)-**118**.

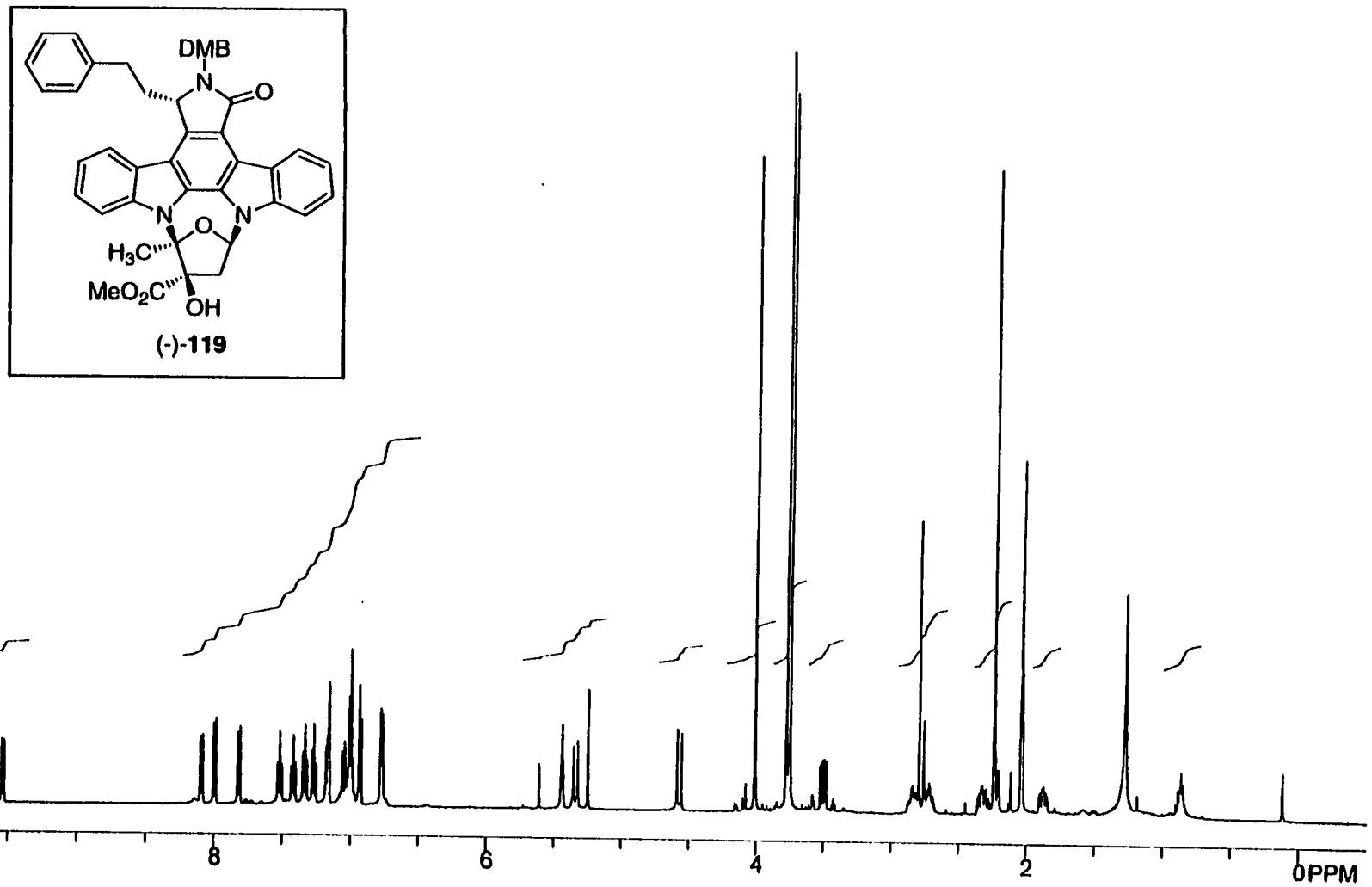


Figure A.4.88 ^{1}H NMR (500 MHz, DMSO-d_6) of compound (-)-**119**.

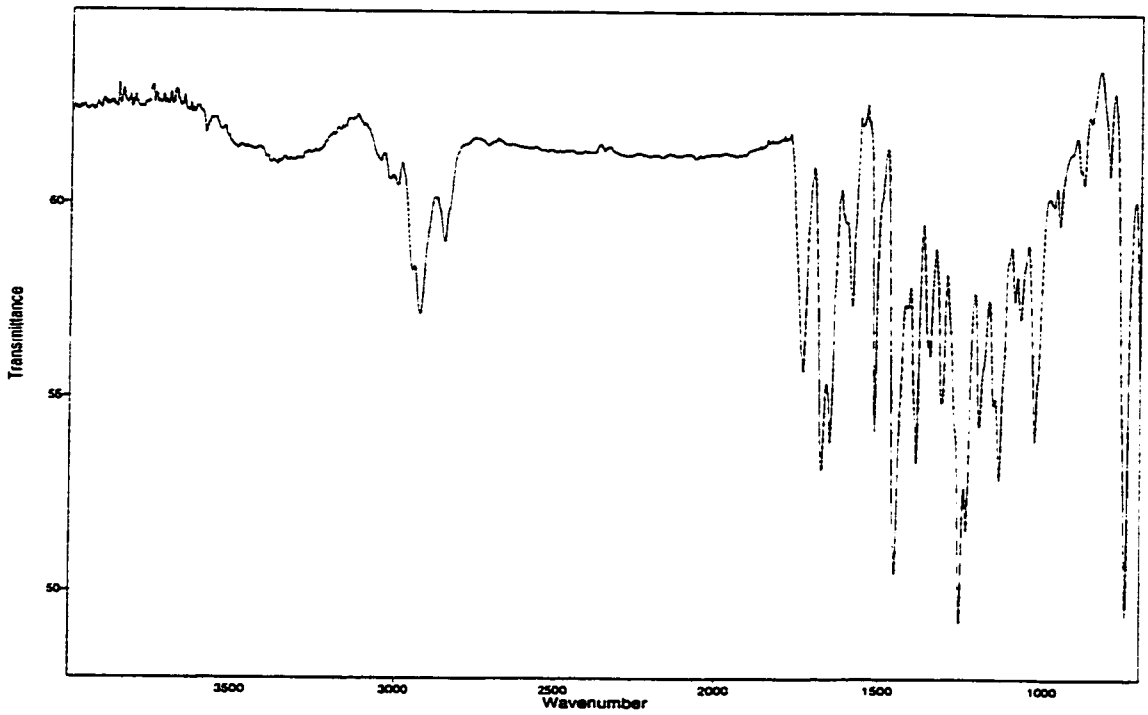


Figure A.4.89 Infrared Spectrum (thin film/NaCl) of compound (-)-**119**.

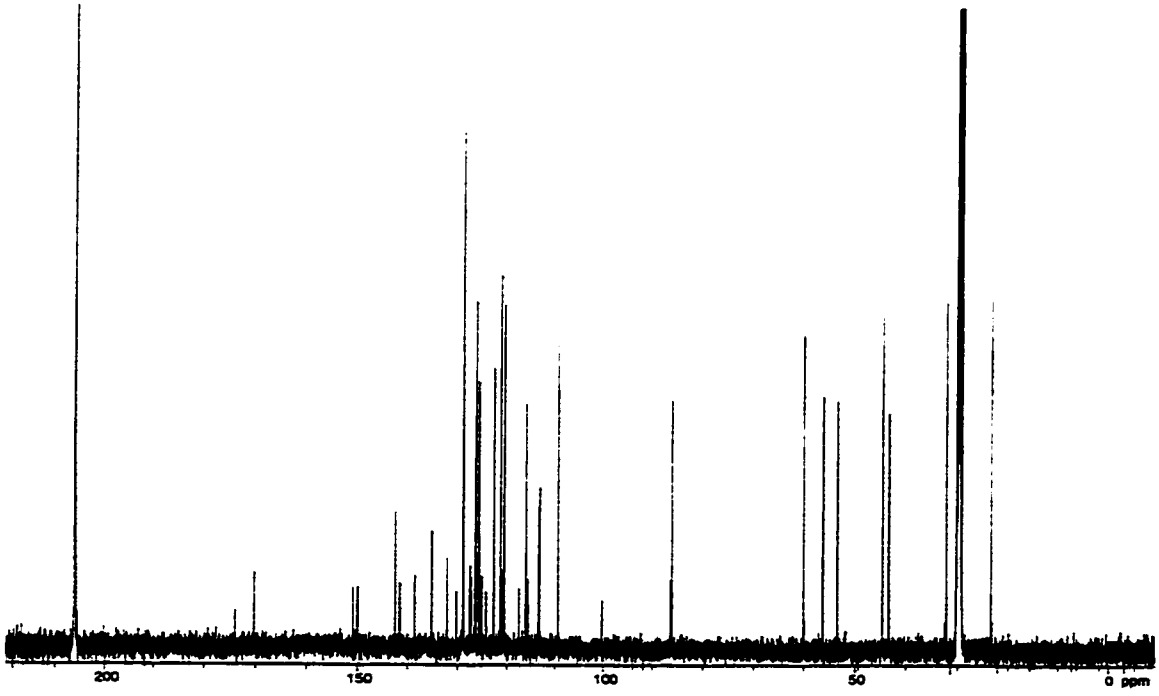


Figure A.4.90 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**119**.

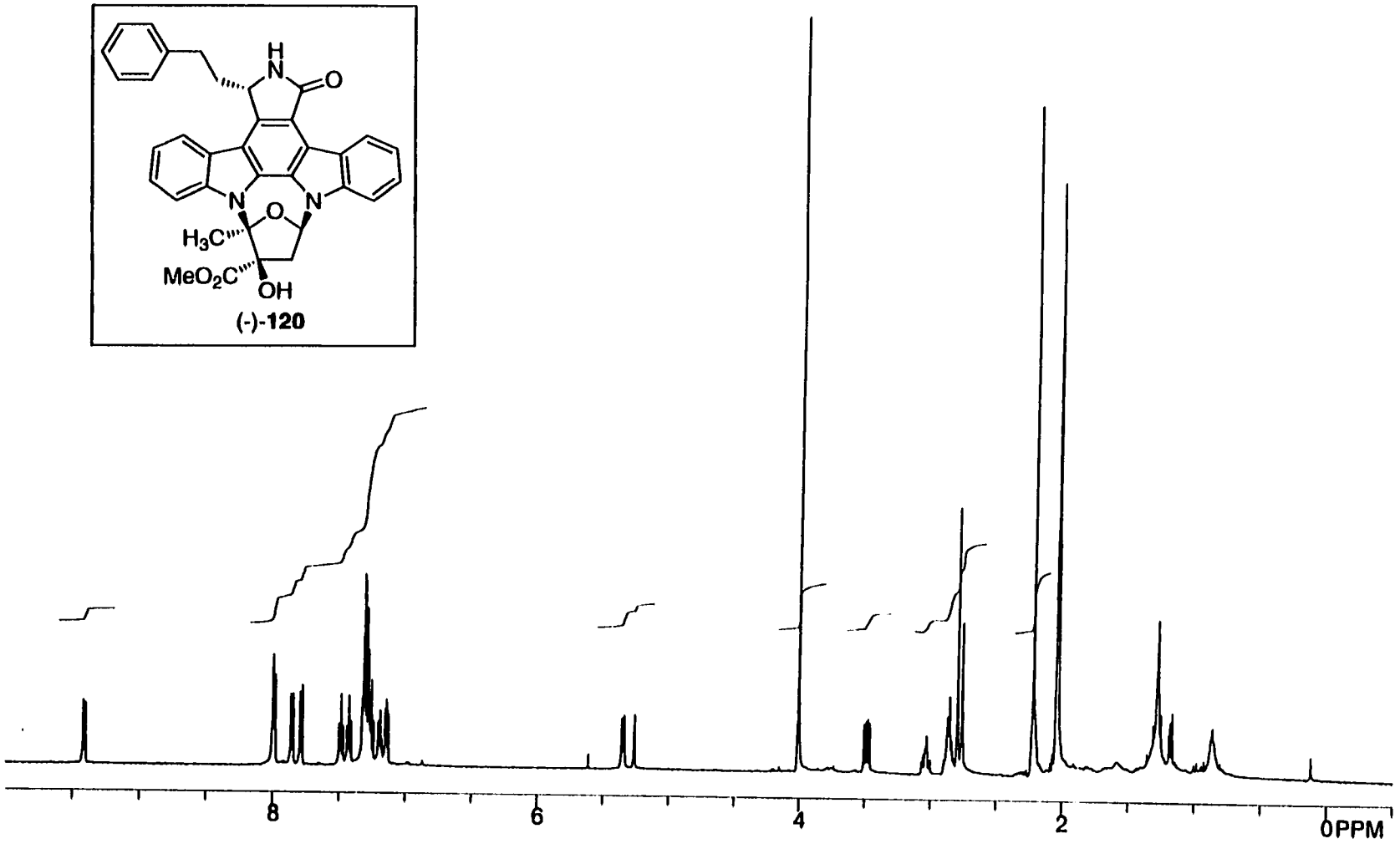


Figure A.4.91 ^{1}H NMR (500 MHz, acetone-d_6) of compound (-)-**120**.

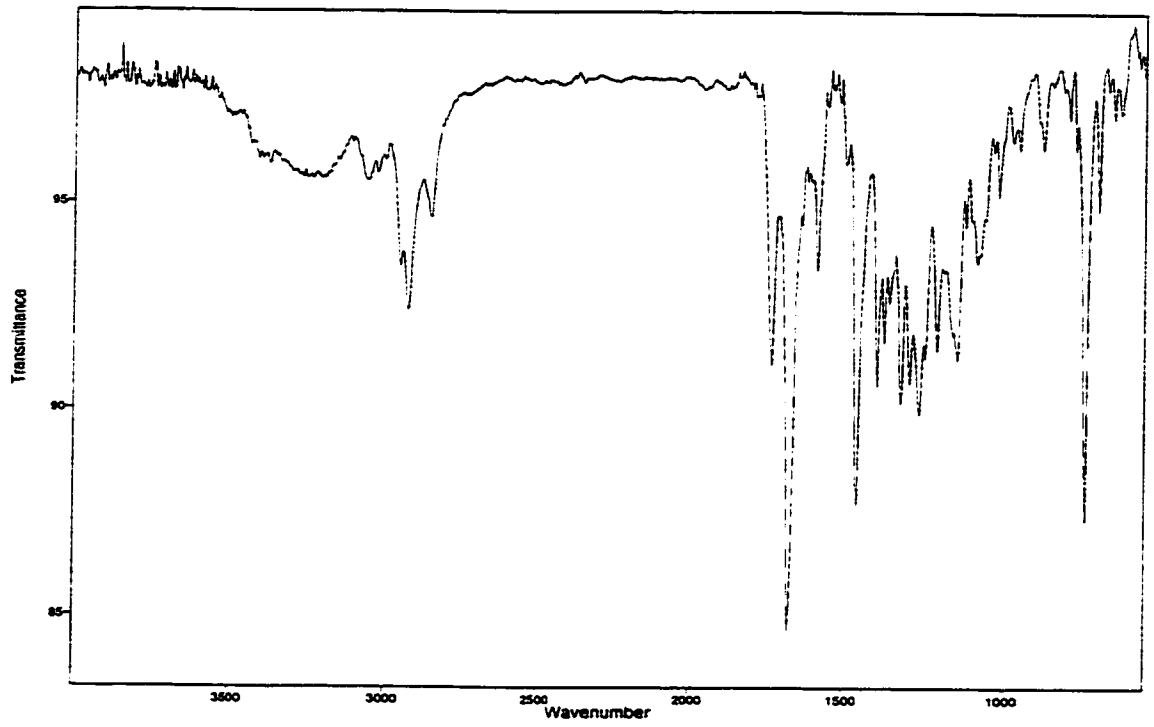


Figure A.4.92 Infrared Spectrum (thin film/NaCl) of compound (-)-**120**.

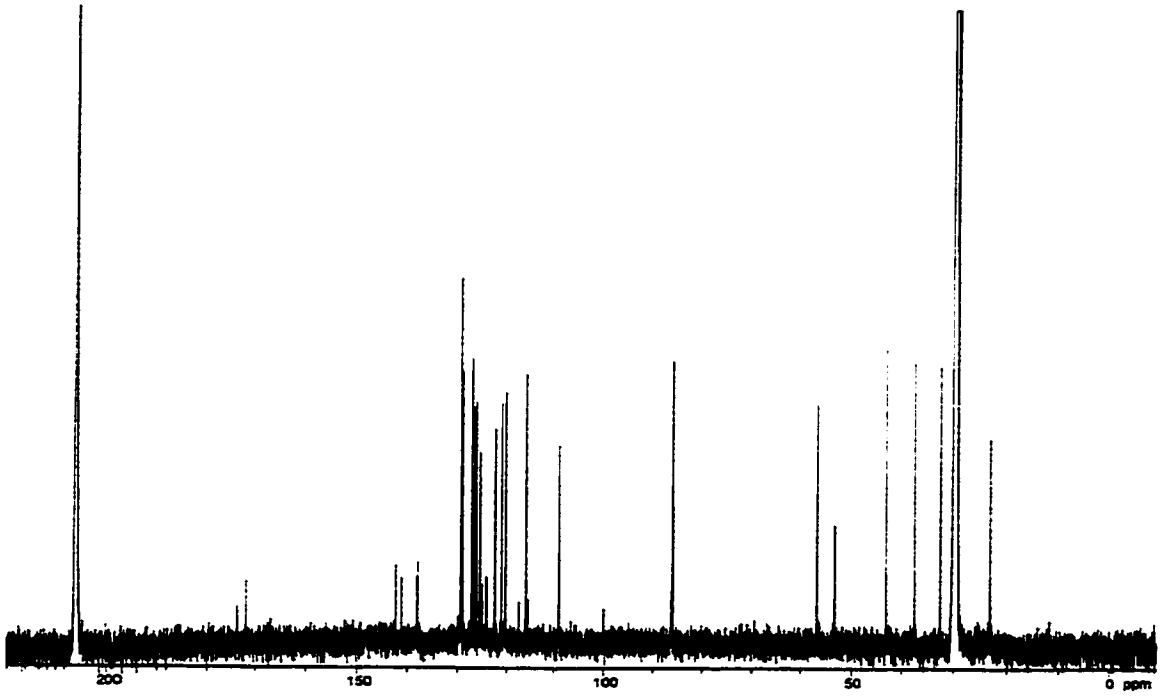


Figure A.4.93 ^{13}C NMR (125 MHz, acetone-d_6) of compound (-)-**120**.

APPENDIX FIVE: NOTEBOOK CROSS-REFERENCE

NOTEBOOK CROSS-REFERENCE

The following notebook cross-reference has been included to facilitate access to the original spectroscopic data obtained for the compounds presented in this thesis. For each compound a folder name is given (i.e., DTP2A-222) which corresponds to an archived characterization folder hard copy, as well as a folder stored on a ZIP disk. For each spectrum a notebook number (i.e., DTP2), a spectrum letter (i.e., A), and a page number (i.e., 222) is given. All notebooks, spectral data, and diskettes are stored in the Wood Group archives.

Compounds Appearing in Chapter Two

Compound	Folder	^{1}H NMR	^{13}C NMR	IR
(-)-**59**	DTPIA.269	DTPIAP.269	DTPIAC.269	DTPIA001
(-)-**60**	DTPIA.254	DTPIAP.254	DTPIAC.254	DTPIA002
(-)-**62**	DTPIIIA.113	DTPIIIAP.113	DTPIIIAC.113	DTPIA004
(-)-**55**	DTPIIA.081	DTPIIAP.081	DTPIIAC.081	DTPIA.005
(-)-**53**	DTPIIA.103	DTPIIAP.103	DTPIIAC.103	DTP2A103
(-)-**66a**	DTPIIA.159	DTPIIAP.159	DTPIIAC.159	DTP2A159
(+)-**66a**	DTPIIA.169	DTPIIAP.169	DTPIIAC.169	DTP2A169
(-)-**67a**	DTPIIB.159	DTPIIBP.159	DTPIIBC.159	DTP2B159
(+)-**67b**	DTPIIB.169	DTPIIBP.169	DTPIIBC.169	DTP2B169
(-)-**52b**	DTPIA.274	DTPIAP.274	DTPIAC.274	DTPIA274
(+)-**52b**	DTPIIA.071	DTPIIAP.071	DTPIIAC.071	DTP2A071
(-)-**68a,b**	DTPIIIA.157	DTPIIIAP.157	DTPIIIAC.157	DTP3A157
(-)-**70**	DTPIIIA.269	DTPIIIAP.269	DTPIIIAC.269	DTP3A269

Compound	Folder	^{1}H NMR	^{13}C NMR	IR
(-)-**71**	DTPIIIA.227	DTPIIIAP.227	DTPIIIAC.227	DTP3A227
(-)-**73**	DTPVIA.239	DTPVIAP.239	DTPVIAC.239	DTPVIA239
(-)-**74**	DTPVIA.263	DTPVIAP.263	DTPVIAC.263	DTPVIA263
(±)-**75**	DTPVIA.277	DTPVIAP.277	DTPVIAC.277	DTPVIA277
(±)-**76**	DTPVIA.261	DTPVIAP.261	DTPVIAC.261	DTPVIA261
(±)-**77**	DTPVIA.267	DTPVIAP.267	DTPVIAC.267	DTPVIA267
(+)-**78a**	DTPVIB.269	DTPVIBP.269	DTPVIB3.269	DTPVIB269
(+)-**78b**	DTPVIC.269	DTPVICP.269	DTPVICC2.269	DTPVIC269
(+)-**79a**	DTPVID.269	DTPVIDP.269	DTPVIDC.269	DTPVID269
(+)-**79b**	DTPVIE.269	DTPVIEP.269	DTPVIEC.269	DTPVIE269
(-)-**80a**	DTPVIIA.035	DTPVIIAP.035	DTPVIIAC.035	DTPVIIA035
(+)-**80b**	DTPVIIA.037	DTPVIIAP.037	DTPVIIAC.037	DTPVIIA037
(-)-**86**	DTPIIA.175	DTPIIAP.175	DTPIIAC.175	DTPIIA175
(-)-**84**	DTPIIA.185	DTPIIAP.185	DTPIIAC.185	DTPIIA185
(-)-**87**	DTPIIIA.285	DTPIIIAP.285	DTPIIIAC.285	DTPIIIA285
(±)-**92**	DTPVIA.031	DTPVIAP.031	DTPVIAC.031	DTPVIA031
(±)-**93**	DTPVIA.039	DTPVIAP.039	DTPVIAC.039	DTPVIA039
(±)-**94**	DTPVIA.047	DTPVIAP.047	DTPVIAC.047	DTPVI047A
(±)-**95**	DTPIVA.251	DTPIVAP.251	DTPIVAC.251	DTPIV251A
(±)-**96**	DTPIVA.209	DTPIVAP.209	DTPIVAC.209	DTPIV209A
(-)-**97**	DTPIVA.257	DTPIVAP.257	DTPIVAC.257	DTPIVA257
(-)-**98**	DTPIVA.279	DTPIVAP.279	DTPIVAC.279	DTPIVA279
(-)-**100a,b**	DTPIVA.277	DTPIVAP.277	DTPIVAC.277	DTPIVA277

Compounds Appearing in Chapter Three

Compound	Folder	^{1}H NMR	^{13}C NMR	IR
(-)-**104a**	DTPVIIA.193	DTPVIIAP.193	DTPVIIAC.193	DTPVIIA193
(-)-**104b**	DTPVIIA.195	DTPVIIAP.195	DTPVIIAC.195	DTPVIIA195
(-)-**104c**	DTPVIIA.215	DTPVIIAP.215	DTPVIIAC.215	DTPVIIA215
(-)-**105a**	DTPVIIA.197	DTPVIIAP.197	DTPVIIAC.197	DTPVIIA197
(-)-**105b**	DTPVIIA.199	DTPVIIAP.199	DTPVIIAC.199	DTPVIIA199
(-)-**105c**	DTPVIIA.227	DTPVIIAP.227	DTPVIIAC.227	DTPVIIA227
(-)-**106a**	DTPVIIA.205	DTPVIIAP.205	DTPVIIAC.205	DTPVIIA205
(-)-**106b**	DTPVIIA.245	DTPVIIAP.245	DTPVIIAC.245	DTPVIIA245
(-)-**107c**	DTPVIIA.207	DTPVIIAP.207	DTPVIIAC.207	DTPVIIA207
(-)-**107a**	DTPVIIA.209	DTPVIIAP.209	DTPVIIAC.209	DTPVIIA209
(-)-**107b**	DTPVIIA.211	DTPVIIAP.211	DTPVIIAC.211	DTPVIIA211
(-)-**107c**	DTPVIIA.247	DTPVIIAP.247	DTPVIIAC.247	DTPVIIA247
(-)-**108a**	DTP08A.037	DTP08AP.037	DTP08AC.037	DTP08A037
(-)-**108b**	DTP08A.033	DTP08AP.033	DTP08AC.033	DTP08A033
(-)-**108c**	DTP08A.031	DTP08AP.031	DTP08AC.031	DTP08A031
(-)-**109a**	DTP08A.087	DTP08AP.087	DTP08AC.087	DTP08A087
(-)-**110a**	DTP08B.087	DTP08BP.087	DTP08BC.087	DTP08B087
(-)-**111a**	DTP08A.105	DTP08AP.105	DTP08AC.105	DTP08A105
(-)-**109b**	DTPVIIA.281	DTPVIIAP.281	DTPVIIAC.281	DTPVIIA281
(-)-**110b**	DTPVIIB.281	DTPVIIBP.281	DTPVIIBC.281	DTPVIIB281
(-)-**111b**	DTP08A.089	DTP08AP.089	DTP08AC.089	DTP08A089
(-)-**109c**	DTP08A.091	DTP08AP.091	DTP08AC.091	DTP08A091
(-)-**110c**	DTP08B.091	DTP08BP.091	DTP08BC.091	DTP08B091

(-)-**111c**	DTP08A.101	DTP08AP.101	DTP08AC.101	DTP08A101
(±)-**114**	DTPVIIA.239	DTPVIIAP.239	DTPVIIAC.239	DTPVIIA239
(±)-**115**	DTPVIIA.243	DTPVIIAP.243	DTPVIIAC.243	DTPVIIA243
(±)-**116**	DTPVIIA.269	DTPVIIAP.269	DTPVIIAC.269	DTPVIIA269
(±)-**117**	DTPVIIA.275	DTPVIIAP.275	DTPVIIAC.275	DTPVIIA275
(±)-**118**	DTP08A.035	DTP08AP.035	DTP08AC.035	DTP08A035
(-)-**119**	DTP08A.137	DTP08AP.137	DTP08AC.137	DTP08A137
(-)-**120**	DTP08A.103	DTP08AP.103	DTP08AC.103	DTP08A103

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ABOUT THE AUTHOR

The author, Dejah T. Petsch, was born on February 2, 1973 in Allentown, Pennsylvania. She was the first daughter of Drs. Douglas and Joanne Follweiler and was joined eighteen months later by her sister Jaelieth. The Follweiler family lived in Springtown, PA where Dejah attended Springfield Elementary School and Palisades Jr./Sr. High School. Following high school graduation, Dejah attended college at The Pennsylvania State University at University Park where she followed in her parents footsteps and decided to major in chemistry. At University Park, Dejah had the opportunity to work in the laboratories of Professor Katherine H. Freeman performing GC and GC/MS analyses of sediment samples.

During her tenure at Penn State, Dejah was a member of the University Scholars Program, and it was during freshman honors classes that she met her future husband Steven Petsch. Dejah found that she shared an interest in chemistry with Steve, who was majoring in geochemistry. Steve and Dejah also found that they shared common interests in dogs, reading, and travel. After graduation from Penn State with bachelor's degrees in chemistry and geology, respectively, Dejah and Steve were married and began to pursue their common interests. First they spent their honeymoon in Germany, Switzerland, Italy and Austria. They have since traveled together to Newfoundland, Iceland, Yellowstone/Grand Teton National Parks, and all over the Northeastern United States. Also during the summer of 1994, Steve and Dejah acquired their first Newfoundland dog and named him "Dejah's Oberon", known as Obie. Steve and Dejah then moved to Connecticut to attend Yale University in the fall of 1994, taking Obie along with them. In 1997 a Landseer Newfoundland "Kiredor's The Mighty Quinn", known as Quinner, was added to the family.

Dejah earned her Ph.D. from Yale University under the direction of Professor John L. Wood. In September of 1999, Dejah will move a few miles north to Wallingford, where she has accepted the position of Research Investigator in the Process Research Division of the Bristol-Myers Squibb Pharmaceutical Research Institute.