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We are submitting herewith a dissertation written by Bin Jiang entitled "1. A Matrix ESR Study of Radical Cations of Acyclic Eneidyne Compounds; 2. A Solid-Phase Synthesis and Combinatorial Approach to 2,3-Dihydrobenzo[d]-isothiazole 1,1-Dioxide; 3. Synthesis of Hyaluronic Acid Disaccharide and Its C-Linked Analogue." We have examined the final copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Chemistry.



David C. Baker, Major Professor

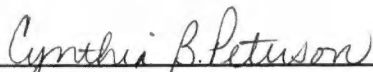


T. Ffranco Williams, Major Professor

We have read this dissertation
and recommend its acceptance:



Spiro D. Alexandratos



Cynthia B. Peterson

Accepted for the Council:



Associate Vice Chancellor and
Dean of The Graduate School

1. A MATRIX ESR STUDY OF RADICAL CATIONS
OF ACYCLIC ENEDIYNE COMPOUNDS
2. A SOLID-PHASE SYNTHESIS AND COMBINATORIAL
APPROACH TO 2,3-DIHYDROBENZO[d]ISOTHIAZOLE
1,1-DIOXIDE
3. SYNTHESIS OF HYALURONIC ACID DISACCHARIDE
AND ITS C-LINKED ANALOGUE

A Dissertation
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DEDICATION

This dissertation is dedicated to my parents

Mr. Zhao-Fu Jiang

and

Mrs. Ying-Hua Liu

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ABSTRACT

The enediyne moiety is the core structure of a family of anti-cancer agents with DNA cleavage ability. This ability comes from a highly reactive species, the 1,4-benzyne diradical, which is a cyclization product of enediyne. Described in Part One is our attempt to study the Bergman cyclization of enediyne radical cations in Freon matrixes at liquid nitrogen temperature. Unfortunately, in the radical cation state, the enediyne was less likely to cyclize, and all we have observed was the open form enediyne radical cation. Semi-empirical calculations and orbital symmetry consideration are used to explain this result. A comparison was made between a flexible (for example, 1,5-hexadiene) radical cation and a rigid (for example, enediyne) radical cation, and it is concluded that the rigidity of enediyne radical cation necessitates the orbital symmetry rule to be strictly observed in such systems.

Sultams are a family of cyclic sulfonamide compounds which have been recently discovered as potent HIV non-nucleoside reverse-transcriptase inhibitors. Described in Part Two is the successful design and implementation of a short, solid-phase synthesis route to sultams. In addition, solution-phase syntheses of sultams with N-side chain modification and trifluoromethylated derivative are described.

Hyaluronic acid is a high molecular weight aminoglycan that is a major component of the extracellular matrix. It appears to play an essential role in

cancer metastasis by interacting with the cancer cell's CD44 receptor. Our continuing efforts are to synthesize hyaluronic acid oligosaccharide mimics, which are smaller molecules containing CH_2 linkages strategically placed at enzymatically labile positions. Described in Part Three is the synthesis of two building blocks, hyaluronic acid disaccharide and corresponding C-disaccharide, which would serve in the future for blockwise synthesis of hyaluronic acid oligosaccharide mimics. Three out of eight possible synthetic routes are described.

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ABBREVIATIONS AND ACRONYMS

AcOH	Acetic acid
ADMA	Anisaldehyde dimethyl acetal
AIBN	Azobis(isobutyronitrile)
AlI	Allyl
Bn	Benzyl
CAN	Ammonium cerium(IV) nitrate
DIBAL-H	Diisobutylaluminum hydride
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
DMAP	4-Dimethylaminopyridine
DMF	Dimethyl formamide
DMP	Dess-Martin Periodinane
DMSO	Dimethyl sulfoxide
DP	Degree of polymerization
EC ₅₀	50% Effective concentration
EDTA	Ethylenediamine tetraacetate
EDCI	1-(3-Dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride
ESIMS	Electrospray-ionization mass spectrometry
ESR	Electron Spin Resonance
EtOAc	Ethyl acetate
Fmoc	9-Fluorenylmethoxycarbonyl
GC	Gas chromatography
HA	Hyaluronic acid

HMPA	Hexamethylphosphoramide
HPLC	High-pressure liquid chromatography
LAH	Lithium aluminum hydride
<i>m</i> CPBA	3-Chloroperoxybenzoic acid
MP	Methoxyphenyl
MS	Mass spectrometry
NBS	<i>N</i> -Bromosuccinimide
NMR	Nuclear magnetic resonance
NNRTI	Non-nucleoside reverse transcriptase inhibitor
NRTI	Nucleoside reverse transcriptase inhibitor
PCC	Pyridinium chlorochromate
Ph	Phenyl
PDC	Pyridinium dichromate
Phth	Phthalimido
PMB	<i>p</i> -Methoxybenzyl
SAR	Structural–activity relationship
TBAF	Tetrabutylammonium fluoride
TBS	<i>tert</i> -Butyldimethylsilyl
Tf	Trifluoromethanesulfonyl
TEA	Triethylamine
TFA	Trifluoroacetic acid
THF	Tetrahydrofuran
TLC	Thin-layer chromatography
TMS	Trimethylsilyl
TPAP	Tetrapropylammonium perruthenate

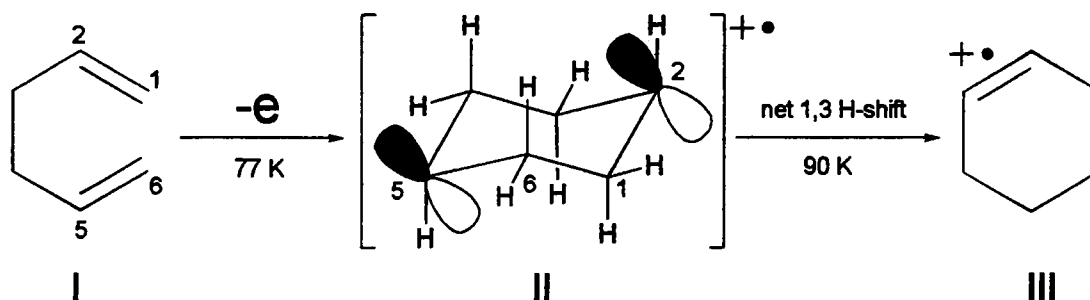
PART ONE

A MATRIX ESR STUDY OF RADICAL CATIONS OF ACYCLIC ENEDIYNE COMPOUNDS

1. INTRODUCTION

If a neutral molecule loses an electron by being one-electron oxidized, it forms a radical cation. In Williams' group, we use a Cobalt-60 γ source to irradiate neutral molecules in various Freon matrices (frozen at liquid nitrogen temperature, ca. 77 K) to form radical cations by radiolytic oxidation.¹ The ESR instrument was used with a variable temperature unit which allowed the spectrum to be recorded from 80 K to the softening point of the matrix (ca. 150 K). Matrix ESR experiments have proved to be a useful technique to study the transformation of the radical cations provided that a well-resolved spectrum can be obtained.

The research described in this part originated from a study of the Cope-like cyclization of the 1,5 hexadiene I radical cation. The Cope rearrangement of the neutral molecule I through a cyclic transition state has an activation energy of 34 kcal/mol, i.e., the reaction normally happens at 150–200 °C.² Our group's early experiments³ on the radiolytic oxidation of 1,5-hexadiene in CFCl_3 and other matrices indicated that cyclization had occurred at below 80 K to the cyclohexane-1,4-diyl radical cation II, and at 90 K to the cyclohexene radical cation III (Scheme 1.1). The ESR spectrum at 80 K consists of a seven-line binomial pattern with a coupling constant of 11.9 G, indicating hyperfine interaction with six nearly equivalent axial hydrogens as showed in structure II. This species decayed at 90 K with the concomitant growth of the cyclohexene radical cation III signals. There has been much interest centered on the detailed structure of the transition state of the Cope rearrangement. By regarding the cyclohexane-1,4-diyl radical cation

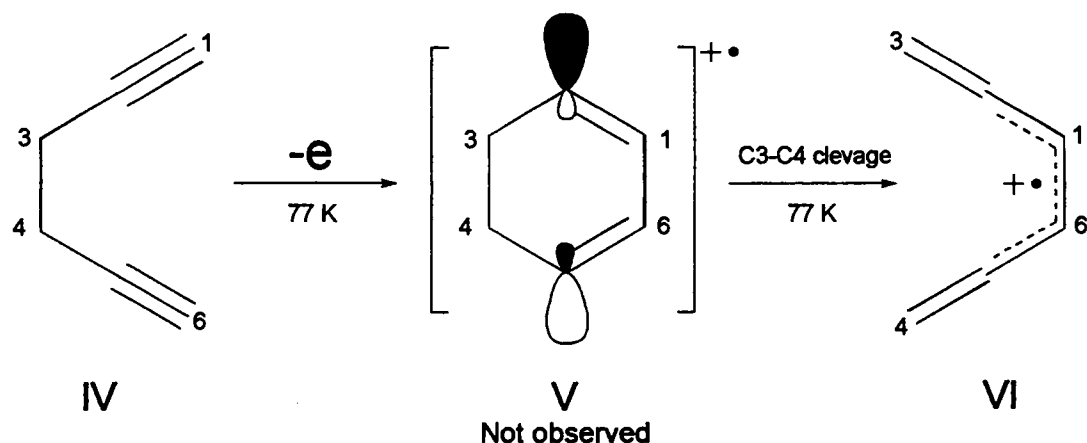


Scheme 1.1. Cyclization of the 1,5-hexadiene radical cation I.

II as corresponding to the "vertical" ionization of the Cope rearrangement transition state, this ESR study proved its highly symmetric structure. Interestingly, the cyclic intermediate II did not open again because of its greater stability⁴ and hence it represents a half-Cope reaction. However, complete Cope-like rearrangement has been observed for the *endo*-5-vinylbicyclo[2.2.1]hept-2-ene radical cation by means of an ESR study.⁵

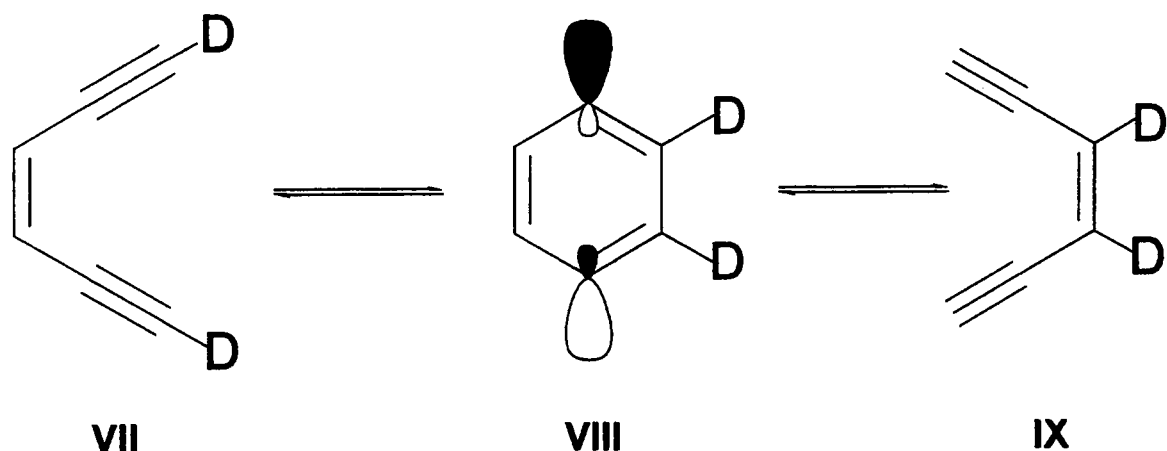
Even with two more π orbitals than I, the resulting hexa-1,5-diyne radical cation can also cyclize, but the difference is that the cyclic intermediate V can easily open again and form a Cope-like rearranged product VI (Scheme 1.2).⁶ The ESR studies established the rearrangement because the spectrum of the product corresponding to VI as a quintet [$A(4H) = 28.6$ G] of triplets [$A(2H) = 3.8$ G] with a g factor of 2.0024.

If the activation energy for a radical cation reaction is always lower than that



Scheme 1.2. Rearrangement of the hexa-1,5-diyne radical cation IV

of its neutral molecule reaction, a natural extension would be to study radical cations from enediyne systems. Bergman⁷ first reported a novel rearrangement of neutral enediyne systems (Scheme 1.3) about 27 years ago. The rearrangement of **VII** through the cyclic transition state **VIII** occurred at 160–200 °C and had an activation energy of 32 kcal/mol,⁸ which is slightly lower than that of the 1,5-hexadiene. As showed in Scheme 1.3, heating di-deuterated enediyne **VII** between 150 and 200 °C in the gas phase led to an equimolar mixture of starting enediyne **VII** and rearranged enediyne **IX**. The diradical character of a symmetric intermediate has to be invoked to rationalize all these observations. In the 80's, a class of highly potent antitumor agents⁹ were found with the *cis*-cyclic enediyne structure. The potency of these agents comes from the highly reactive biradical 1,4-didehydrobenzene structure (like **VIII**) which can abstract hydrogen atoms from the DNA backbone resulting in DNA cleavage. Calicheamicin γ_1^I is one of these



Scheme 1.3. Bergman cyclization of 3-hexene-1,5-diyne.

agents currently under phase II clinical trial and was recently totally synthesized.¹⁰ Interest in the 1,4-didehydrobenzene (*para*-benzyne) intermediate has therefore intensified since it was first proposed, and its structural characterization remains a fundamental problem in enediyne research.

2. STATEMENT OF THE PROBLEM

In regard to the present ESR study on enediyne radical cations, questions that need to be addressed are as follows: (1) What is the structure of the enediyne radical cation? Is it a closed or an open form? (2) If it is a closed form, are the positive charge and spin separated from each other (a distonic species)? (3) If it is an open form, how is the spin distributed? (4) What kind of reactions could this

enediyne radical cation have?

3. RESULTS

3.1. *cis*-3-Hexene-1,5-diyne (1) radical cation

Compound 1 was prepared and sent to us as dilute Freon solutions by Kamal Bharucha in Bergman's group. After γ irradiation, Dr. Wang in our group recorded its ESR spectrum in 1990. The best resolution came out at 156 K in $\text{CF}_2\text{ClCCl}_3$ matrix (Figure 1.1) and the same spectral profile was obtained in CFCl_3 and CF_3CCl_3 (Figure 1.2). In each case, the spectrum had a quintet appearance but since the three center lines were broader and had shoulders, it was analyzed as a triplet of triplets instead with the coupling constants of 9.3 G and 6.8 G, respectively. The g value is 2.0038.

Although these early spectra were quite well-resolved, a deuterium experiment would have been very helpful. But since Kamal Bharucha graduated in 1992, additional compounds were no longer available from Bergman's group. Finally, this project restarted after I entered the group in 1995.

3.2. *cis*-1,6-Bis(trimethylsilyl)-3-hexene-1,5-diyne (2) radical cation

Compound 2 was prepared by both the Heck reaction¹¹ (Scheme 1.4) and the carbenoid coupling reaction¹² (Scheme 1.5). The Heck reaction can only give one *cis*-enediyne starting from the *cis*-dichloro ethylene while the carbenoid coupling always gave a 2:1 ratio of *cis* and *trans* enediyne. The *cis* and *trans* enediyne can

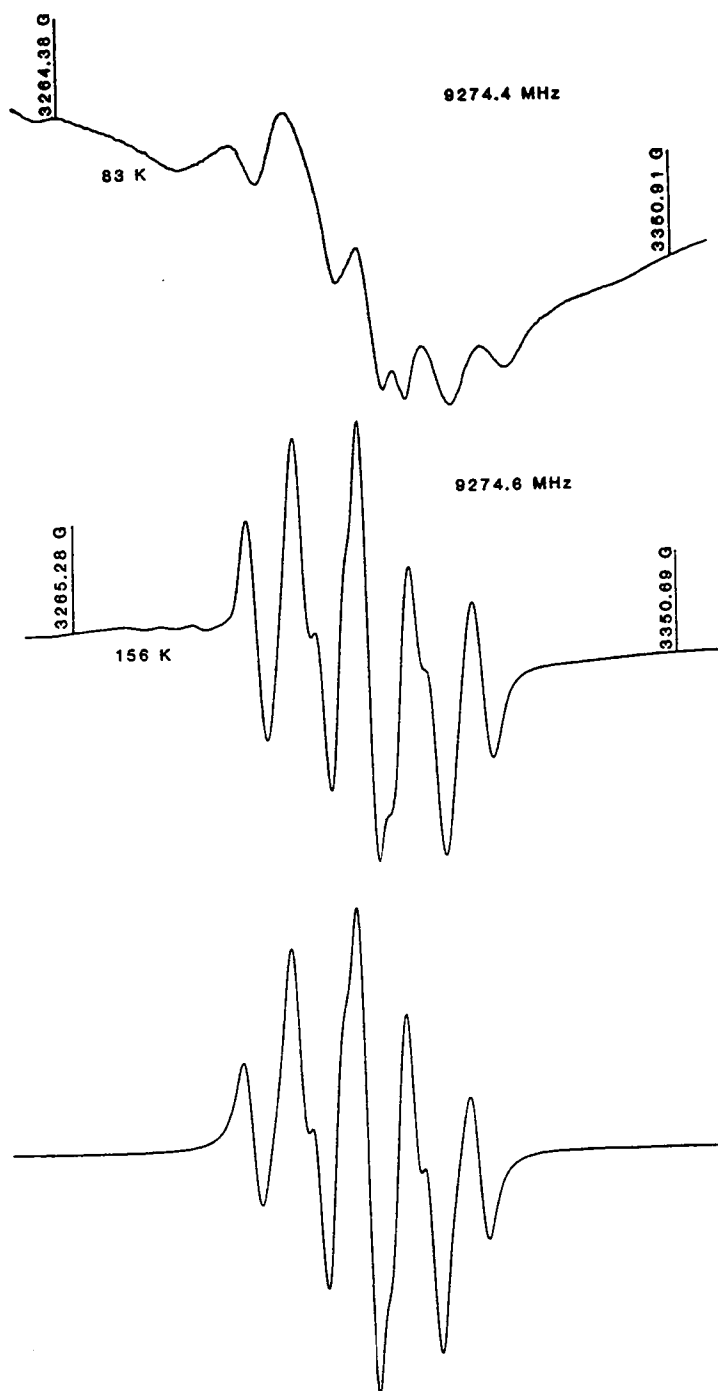


Figure 1.1. ESR spectrum of *cis*-enediynes **1** in $\text{CF}_2\text{ClCCl}_3$. Top: at 83 K; Middle: at 156 K; Bottom: simulated spectrum using $A(2\text{H}) = 9.3$ G, $A(2\text{H}) = 6.8$ G, line width = 2.05 G, and modulation = 2.0.

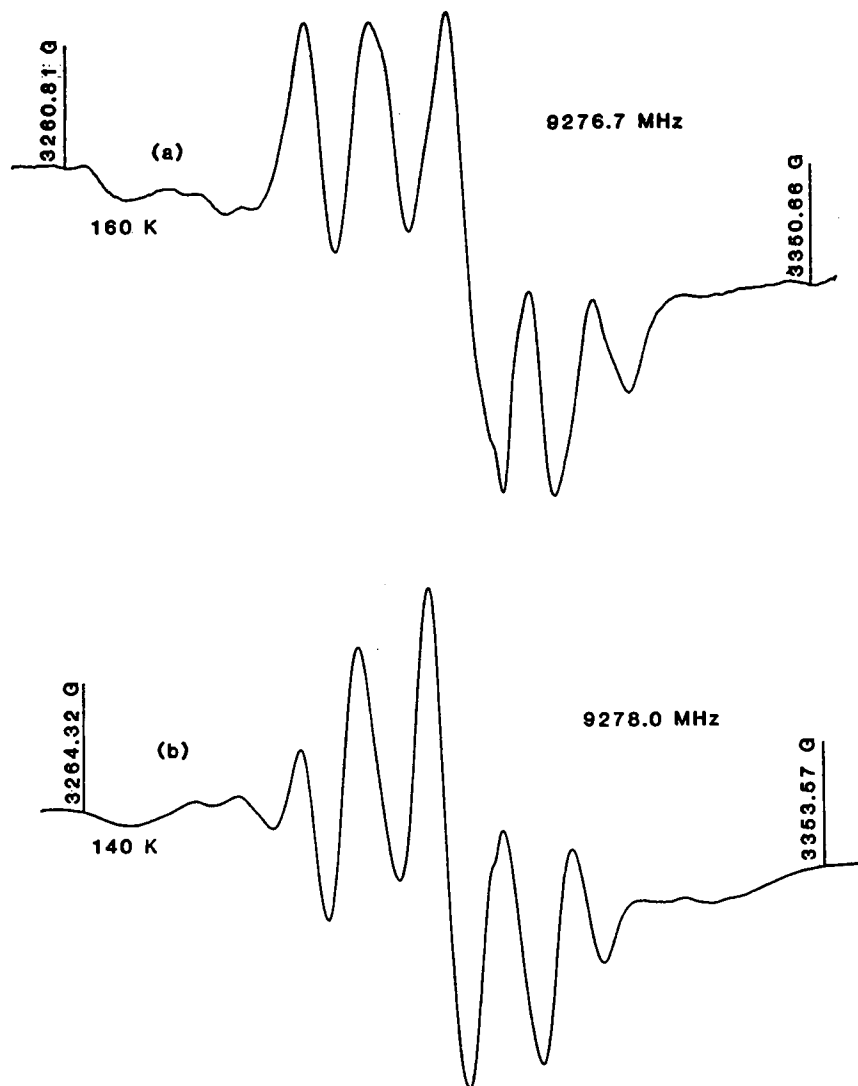
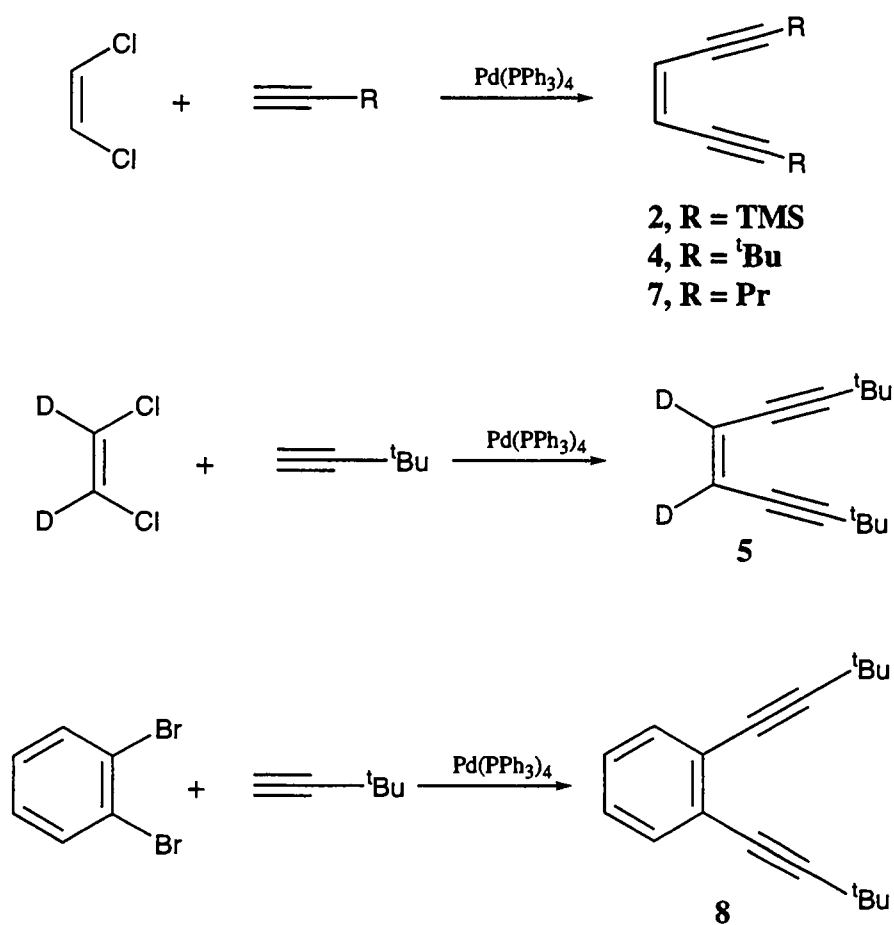
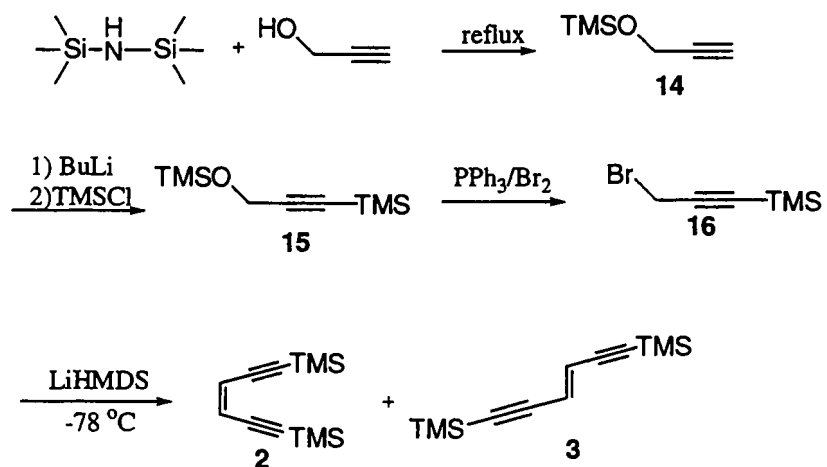


Figure 1.2. ESR spectrum of *cis*-enediyne 1. (a) in CFCl_3 at 160 K; (b) in CF_3CCl_3 at 140 K.



Scheme 1.4. Synthesis of *cis*-enediynes by Heck Reaction.



Scheme 1.5. Synthesis of *cis*- and *trans*-enediynes by carbenoid coupling.

be easily separated by flash column chromatography. Compound **2** was designed to have the trimethylsilyl groups capped at both yne positions to prevent the enediyne from polymerization since the parent *cis*-enediyne **1** very easily discolors at room temperature. After purification, enediyne **2** was a colorless liquid and its *trans* isomer **3** was a white solid. Both of them remained stable during the ESR sample preparation time. Upon exposure in air at room temperature for a while, the color of *cis* enediyne will change to light yellow. Another reason to cap the yne positions is the assumption that the introduction of the trimethylsilyl groups would be expected to increase the steric hindrance of the cyclization process, and hence it might be less likely to undergo cyclization than the parent enediyne radical cation.

The sample in the matrix had a weak yellow color ($\lambda_{\text{max}} \approx 435\text{--}480\text{ nm}$) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.3) showed a triplet signal from 85 K to 165 K with a coupling constant of ca. 6.4 G and $g = 2.0029$ at 165 K. The same profile was observed in the CF_3CCl_3 matrix.

3.3. *cis*-2,2,9,9-Tetramethyl-5-decene-3,7-diyne (**4**) radical cation

Compound **4** was prepared by the Heck reaction (Scheme 1.4). It was purified by flash column chromatography as a colorless liquid. It was stable during the ESR sample preparation time. Upon exposure in air at room temperature for a while, its color will change to light yellow. The experimental design idea here was similar to that of enediyne **2**. The sample in the matrix had a green color ($\lambda_{\text{max}} \approx 595\text{--}750\text{ nm}$) after γ irradiation. The ESR spectrum of this enediyne radical cation

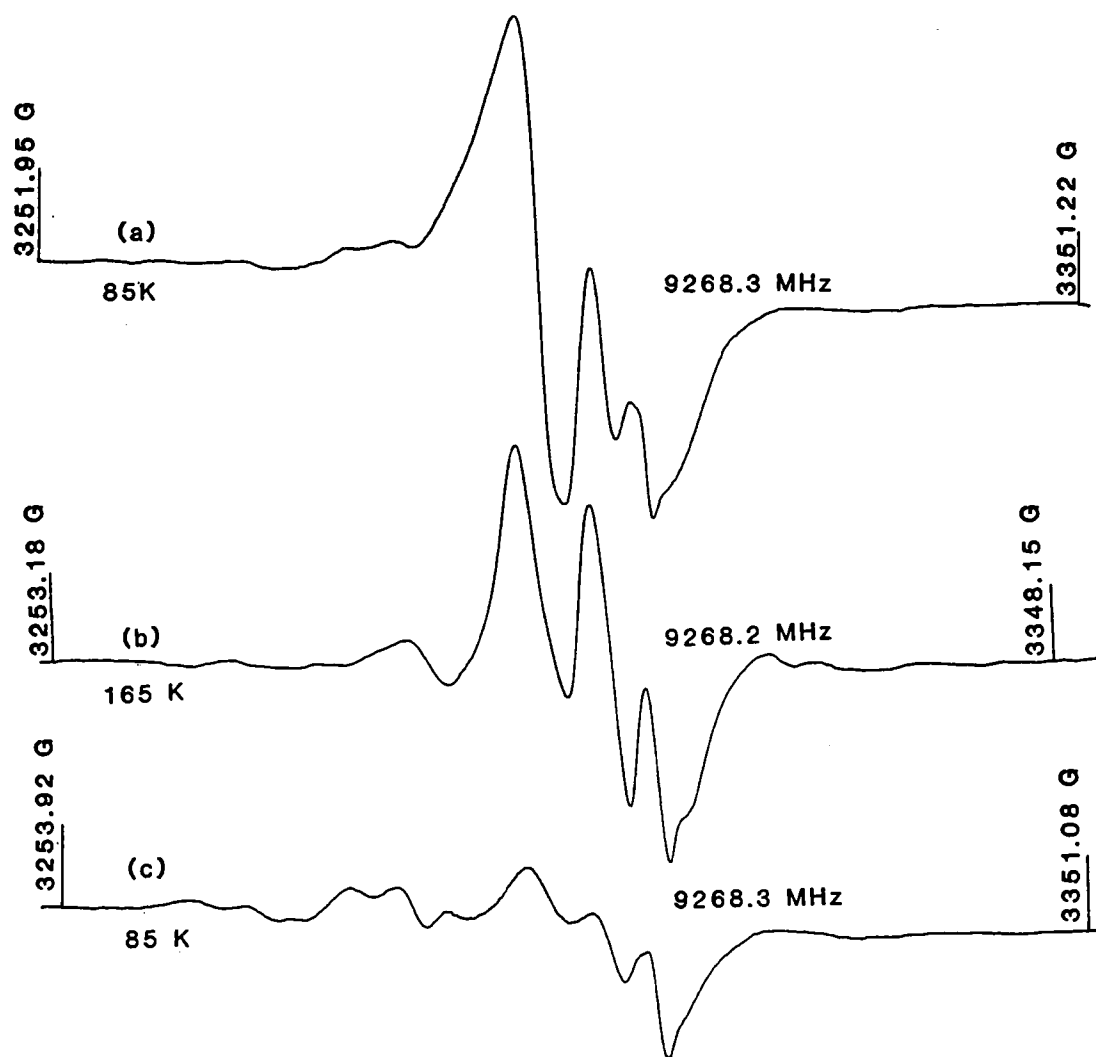


Figure 1.3. ESR spectrum of *cis*-enediyne 2 in CFCl_3 . (a) at 85 K; (b) at 165 K; (c) at 85 K (cooled back from 165 K).

in CFCl_3 (Figure 1.4) showed a triplet signal at 165 K with a coupling constant of ca. 6.4 G and $g = 2.0031$.

3.4. *cis*-5,6-Dideuterio-2,2,9,9-tetramethyl-5-decene-3,7-diyne (**5**) radical cation.

Compound **5** was prepared by the Heck reaction (Scheme 1.4). It was purified by flash column chromatography as a colorless liquid. It was stable during the ESR sample preparation time. Upon exposure in air at room temperature for a while, its color will change to light yellow. The design idea was to verify the assignments for radical cation **2** and **4**.

The sample in the matrix had a green color ($\lambda_{\text{max}} \approx 595\text{--}750\text{ nm}$) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.5) showed a clean singlet signal at 165 K with $g = 2.0036$.

3.5. *cis*-3-*tert*-Butyl-7,7-dimethyl-3-octene-1,5-diyne (**6**) radical cation.

Compound **6** was prepared by the McMurry coupling¹³ reaction (Scheme 1.6) followed by TBAF/THF desilylation. It was purified by preparative GC as a colorless liquid. It was stable during the ESR sample preparation time. Upon exposure in air at room temperature for a while, its color will change to brown. The design idea again was to verify the assignments for radical cation **2** and **4**.

The sample in the matrix had a green color ($\lambda_{\text{max}} \approx 595\text{--}750\text{ nm}$) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.6) showed a poorly resolved four-line pattern (comprising neither a quartet nor a

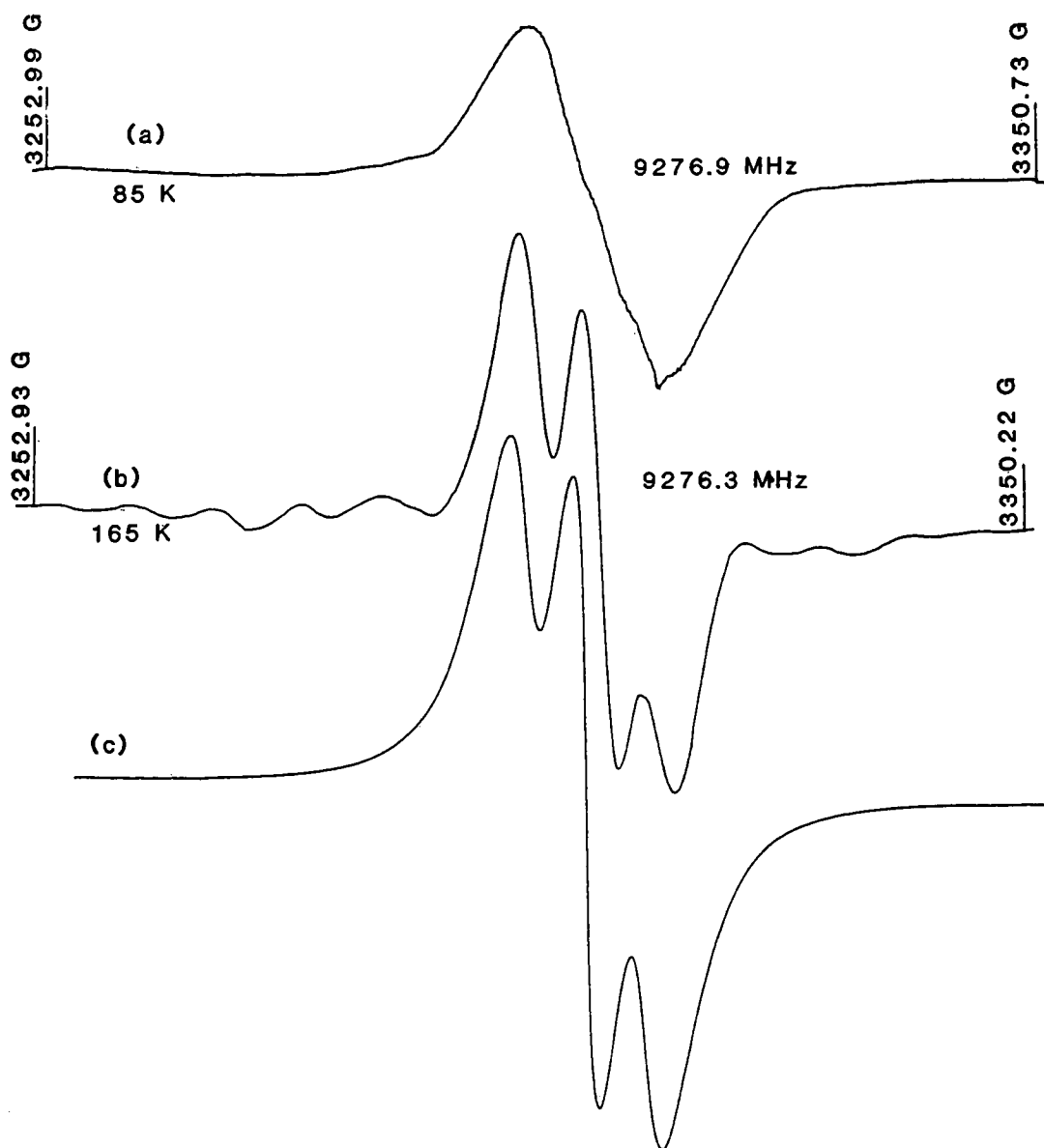


Figure 1.4. ESR spectrum of *cis*-enediynes **4** in CFCl_3 . (a) at 85 K; (b) at 165 K; (c) simulated spectrum using $A(2\text{H}) = 6.4$ G, line width = 2.05 G, and modulation = 2.0.

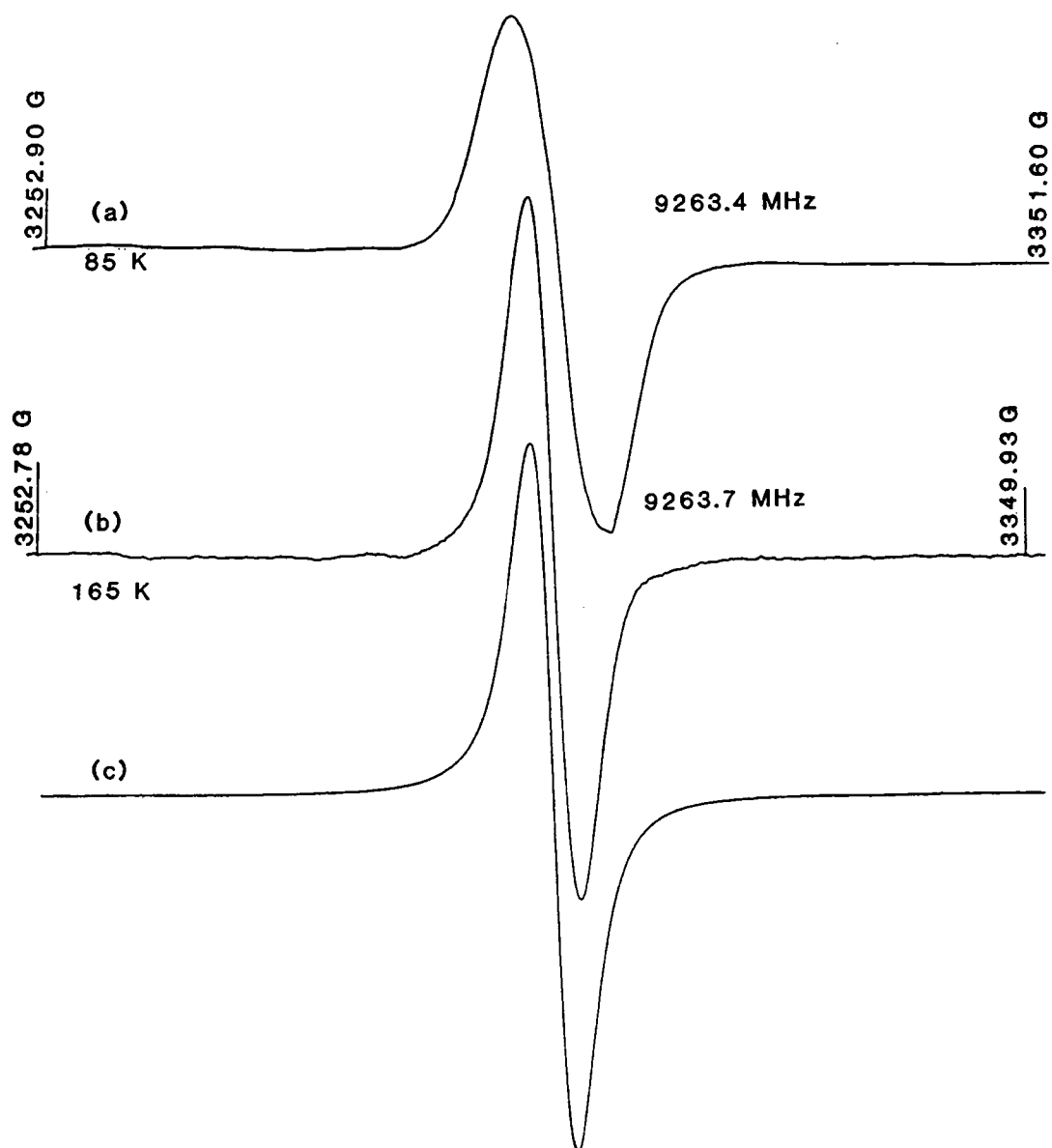


Figure 1.5. ESR spectrum of *cis*-enediyne **5** in CFCI_3 . (a) at 85 K; (b) at 165 K; (c) simulated spectrum using $A(2D) = 0.979$ G, line width = 2.05 G, and modulation = 2.0.



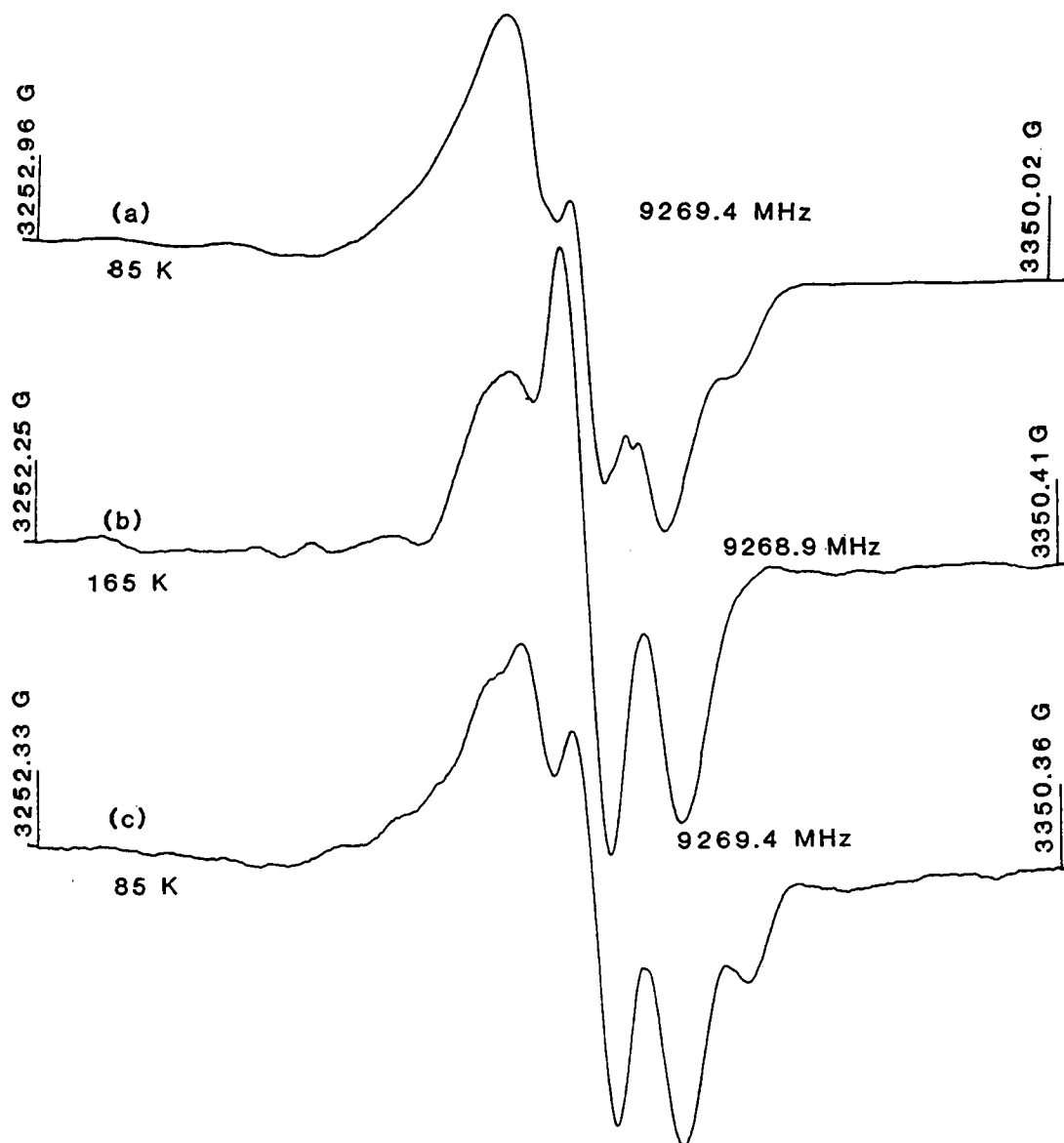


Figure 1.6. ESR spectrum of *cis*-enediyne 6 in CFCl_3 . (a) at 85 K; (b) at 165 K; (c) at 85 K (cooled back from 165 K).

doublet of doublets) at 85 K but more closely resembled a triplet at 165 K with a coupling constant of ca. 7.4 G and $g = 2.0036$.

3.6. *cis*-6-Dodecene-4,8-diyne (7) radical cation

Compound 7 was prepared by the Heck reaction (Scheme 1.4). It was purified by flash column chromatography as a light yellow oil. It was vacuum transferred into ESR tubes as colorless liquid. The design idea was to see whether the spin can delocalize to the four β hydrogens of the π system.

The sample in the matrix had a green color ($\lambda_{\text{max}} \approx 595\text{--}750$ nm) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.7) showed a complex hyperfine pattern even at 165 K and the spectrum could not be assigned except for $g = 2.0034$.

3.7. 1,2-Bis(3',3'-dimethylbutynyl)benzene (8) radical cation

Compound 8 was prepared by the Heck reaction (Scheme 1.4). It was obtained as a light yellow liquid after flash column chromatography. Both vacuum transfer and the TLC method were used to prepare ESR samples and they both gave the same result. The design idea was to see whether the benzene ring could facilitate the 1,5-cyclization.¹⁴

The sample in the matrix had a pink color ($\lambda_{\text{max}} \approx 490\text{--}500$ nm) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.8) showed a poorly resolved quintet or septet (since the spectrum was not well

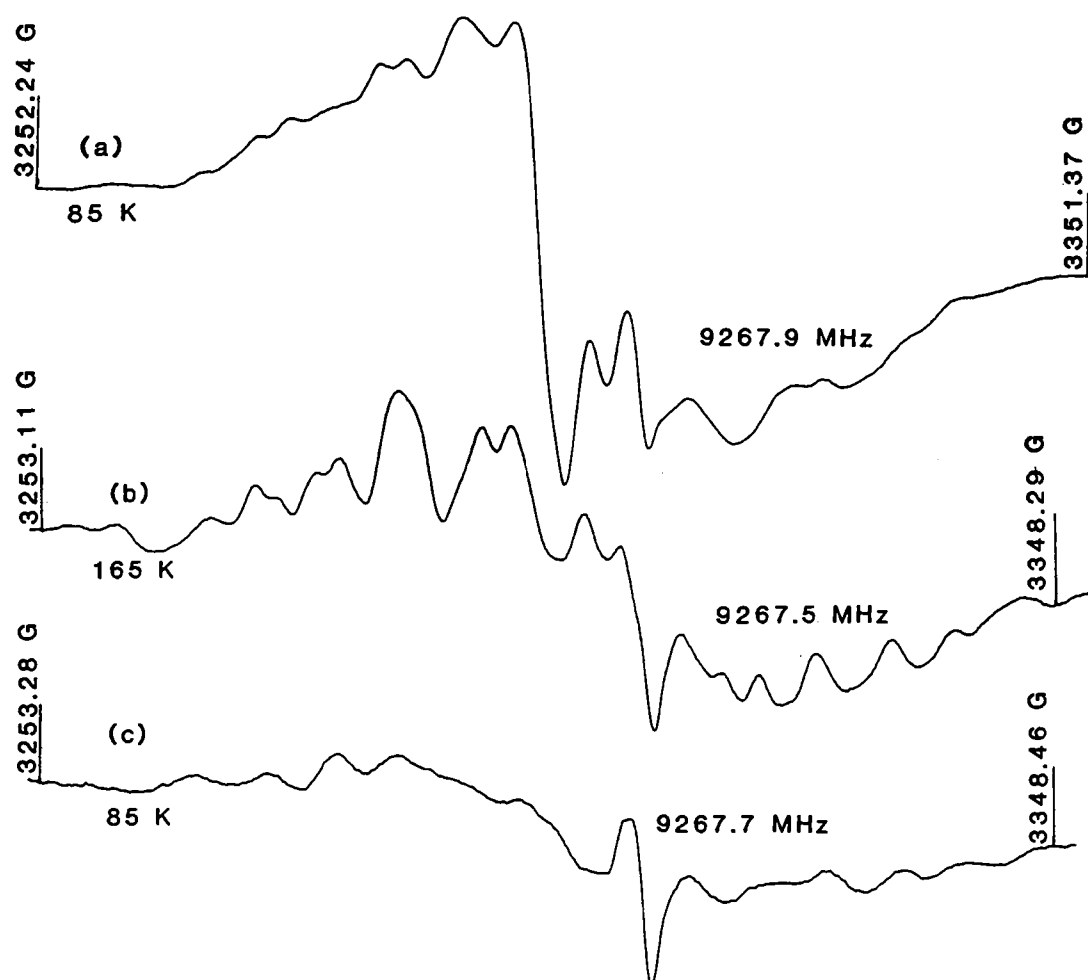


Figure 1.7. ESR spectrum of *cis*-enediyne 7 in CFCl_3 . (a) at 85 K; (b) at 165 K; (c) at 85 K (cooled back from 165 K).

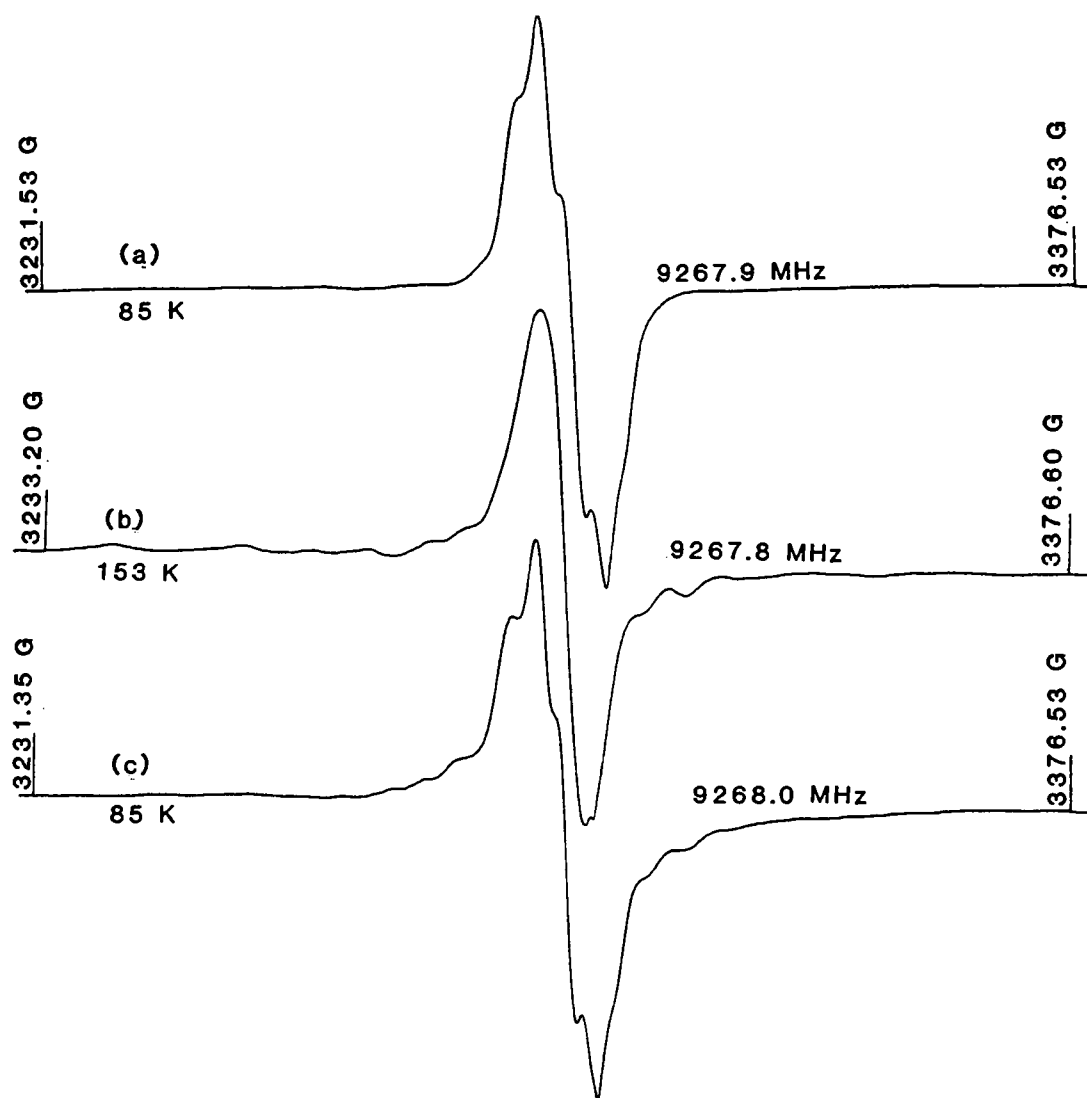


Figure 1.8. ESR spectrum of enediyne **8** in CFCI_3 . (a) at 85 K; (b) at 153 K; (c) at 85 K (cooled back from 165 K).

resolved, these two patterns gave almost the same spectrum when using coupling constant of 3.4 G with a line width of 3.4 G and 2.0 G modulation width in simulation) at 85 K but reversibly lost resolution at 153 K. The g factor is 2.0031.

3.8. *trans*-3-Hexene-1,5-diyne (**9**) radical cation.

Compound **9** was prepared and sent to us as dilute Freon solutions by Kamal Bharucha. Its radical cation spectrum (Figure 1.9), which was recorded by Dr. Wang in 1990, showed a five-line pattern (not a quintet since the intensity ratio is not binomial) with $g = 2.0043$ at 140 K. The line spacing of this pattern is comparable with that of figure 1.1.

3.9. *trans*-2,2,9,9-Tetramethyl-5-decene-3,7-diyne (**10**) radical cation

Compound **10** was prepared from its *cis* isomer **4** by UV photolysis (cf. experimental). It was obtained as white needles after flash column chromatography.

The sample in the matrix had a blue-green color ($\lambda_{\text{max}} \approx 605\text{--}750$ nm) after γ irradiation. The ESR spectrum of this enediyne radical cation in CFCl_3 (Figure 1.10) showed a broad singlet peak with some weak shoulders at 85 K. The spectrum reversibly changed to an apparent triplet with $A(2H) = 4.8$ G and $g = 2.0031$ at 165 K.

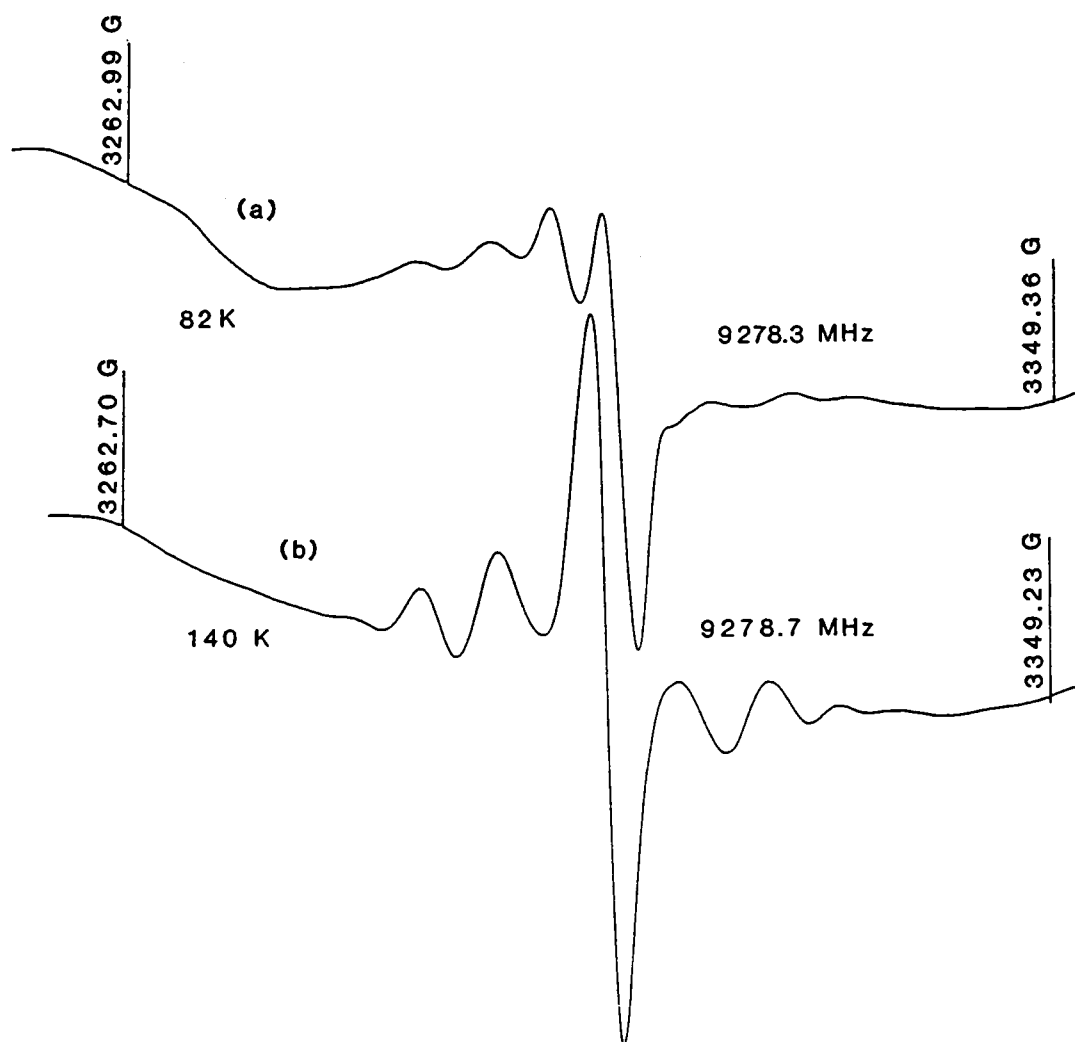


Figure 1.9. ESR spectrum of *trans*-enediyne **9** in CFCI_3 . (a) at 82 K; (b) at 140 K.

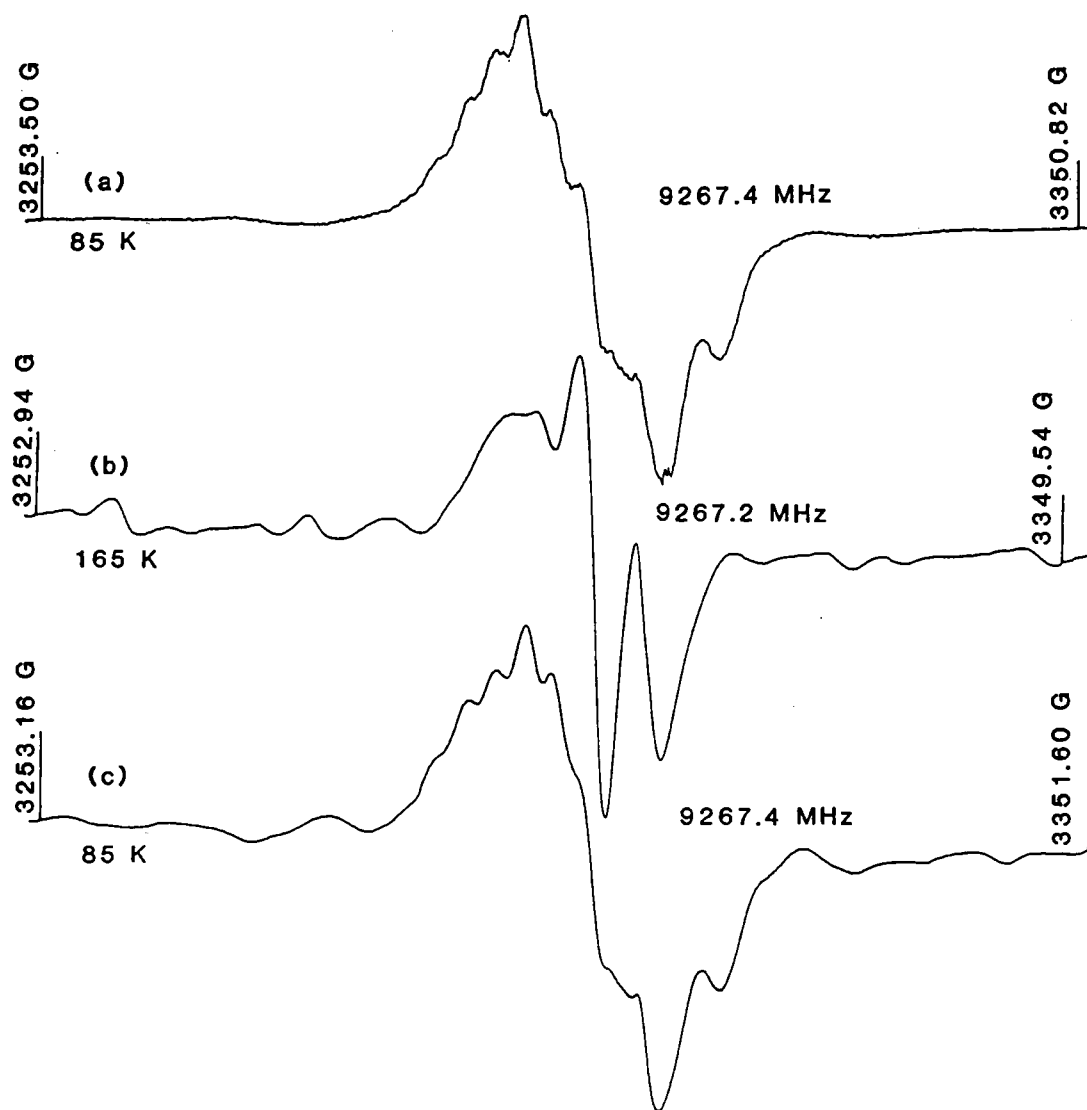


Figure 1.10. ESR spectrum of *trans*-enediynes **10** in CFCl_3 . (a) at 85 K; (b) at 165 K; (c) at 85 K (cooled back from 85 K).

4. DISCUSSION

By putting trimethylsilyl or *tert*-butyl groups at the yne positions of the *cis* parent enediyne **1**, the ESR spectrum (cf. Figure 1.1, 1.3 and 1.4) changed from a triplet of triplets [$A(2H) = 9.3$ G, $A(2H) = 6.8$ G] to a broad triplet [$A(2H) = 6.4$ G]. It indicated that, in radical cation **1**, the two acetylenic hydrogens had a coupling constant of 9.3 G, which is larger than 6.8 G of those two olefinic hydrogens. This is consistent with our AM1 calculations for an open form π radical cation whose s-spin population for acetylenic hydrogens is slightly larger (absolute value) than that of olefinic hydrogens (-0.02338 vs -0.02088, or -11.8 G vs -10.6 G by multiplying a factor of 506.7). The line-broadening effect in the spectra in Figure 1.3 and 1.4 may be due to the steric hindrance or bulk of the trimethylsilyl and *tert*-butyl groups which would be expected to undergo much slower molecular tumbling compared with radical cation **1**. The spectrum of radical cation **5** (cf. Figure 1.5) further supports this assignment since substituting two olefinic hydrogens by deuterium atoms in **4** results in the spectrum changing from a triplet to a singlet (two deuterium atoms would give a quintet pattern but since the coupling constant is reduced by a factor of 6.54 for deuterium relative to the value for hydrogen, this quintet is not resolved and appears as a singlet). If the above assignment is correct, radical cation **6** might give a doublet of doublet with coupling constants of about 9.0 and 6.4 G, respectively. But again because of the steric hindrance of the *tert*-butyl groups, the line-width of the spectrum may be larger than the difference between these two coupling constants so that the spectrum appears as a triplet (cf. Figure

1.6).

Assuming that the coupling constants for radical cation **1** are properly assigned, we now address the next question: Does this assignment correspond to an open or cyclic structure? It is well known that the isotropic coupling constants for the phenyl radical are $A_{\text{ortho}} = 17.4 \text{ G}$ and $A_{\text{meta}} = 5.9 \text{ G}$.¹⁵ If the radical cation has a closed-form structure without the spin and charge separation, its spectrum would be expected to be a quintet with a coupling constant of about 17 G. Also, if it is a closed form with spin and charge separation (a distonic species), its spectrum would be expected as a triplet of triplets with almost the same profile as that of the phenyl radical. Both of these possibilities were not observed, so the only remaining option is an open form structure, i.e., the radical cation of the parent enediyne did not cyclize at our experimental temperature (77–160 K). It is interesting to point out that the spectra for the *trans* enediyne radical cations **9** and **10** (cf. Figure 1.9 and 1.10) are very similar to those of the corresponding *cis* enediyne radical cations **1** and **4**. By recognizing that the *trans* enediyne is very unlikely to cyclize, and that the 6.8 G and 9.3 G couplings are well in the range of α -proton coupling constants for a π radical, we have to conclude that the structure of radical cation **1** corresponds to an open form. Another hint from the experiment was that all substituted enediyne radical cations, except for **2** with silyl groups which is light yellow and **8** with phenyl ring which is pink, have a green color.¹⁶ Although we were not able to run the electronic absorption (EA) spectra for these radical cations, we can still guess approximately that their maximum absorption wavelength is in the

range of 595–750 nm based on the color.¹⁷ This range actually contains one of the two absorption bands found in the EA spectrum of linear hexatriene radical cation¹⁸ [$\lambda_{\text{max}} = 642 \text{ nm}$ (weak band), 392 nm (strong band)].

So now we turn to the question of why the enediyne radical cation does not cyclize? We can rationalize this by consideration of both energetics and orbital symmetry theory.

The photodissociation of $\text{C}_6\text{H}_4^{+\cdot}$ generated from cyanobenzene and other precursors has been studied by van der Hart and co-workers.¹⁹ The kinetics were too complicated to be solved but the experiments did indicate that a major fraction of the $\text{C}_6\text{H}_4^{+\cdot}$ ions has a ring-opened structure. The semi-empirical and ab initio calculations in literature¹⁴ and by us (cf. Table 1.1) also predicted that the open form of the enediyne radical cation is more stable than the closed form. These theoretical results are in sharp contrast to those of 1,5-hexadiene and 1,5-hexadiyne radical cations. For the 1,5-hexadiene radical cation, the closed form 1,4-cyclohexanediyl radical cation is more stable than the open form by 34 kcal/mol according to an MINDO/3 calculation.⁴ However, more recent density functional theory calculations indicate that this cyclization is exothermic by only 3.7 kcal/mol.²⁰ For the 1,5-hexadiyne radical cation, the closed form 5,6-dihydro-1,4-benzyne radical cation was also more stable as predicted by AM1-UHF calculation.⁶ Hence the unfavorable energetics in the present case may explain why the parent enediyne radical cation cannot cyclize to form 1,4-benzyne radical cation.

Considerations of orbital symmetry via a correlation diagram might also be

Table 1.1. Theoretical calculations* on the open and closed form of the enediyne radical cation 1.

		Open Form	Closed Form
AM1	UHF (kcal/mol)	320.1 (320.2)	341.5 (341.5)
	RHF (kcal/mol)	323.60	351.07
MINDO/3	UHF (kcal/mol)	288.29	294.07
	RHF (kcal/mol)	292.09	298.41
	UHF/3-21G (hartree)	(-227.82730)	(-227.81961)
	PUHF/6-31G* (hartree)	(-229.12994)	(-129.07806)
	PMP2/6-31G* (hartree)	(-229.80966)	(-229.82352)

* Heat of Formation or Total Energy; Data in parenthesis are from reference 14; 1 hartree = 628.2245827 kcal/mol.

useful to understand the energetic uphill process of the radical-cation Bergman cyclization. Figure 1.11 shows the correlations between each of the five highest occupied orbitals of the open form and closed form structures. All orbitals are first drawn by MOPLOT²¹ using the output files of AM1-RHF calculations and then redrawn in Chemdraw by approximation in order to paste them into this thesis. In σ_{2v} symmetry, the σ_v symmetry plane which bisects the enediyne molecular plane is taken to be conserved throughout the reaction. The same correlation can also be obtained if the C_2 symmetry axis is conserved. Obviously, the correlation results in four crossings. The highest energy crossing indicates that for the open structure an electron needs to be excited from MO 12 to MO 14 to fulfill the state symmetry correlation. Hence the radical cation Bergman cyclization is both orbital and state symmetry forbidden.

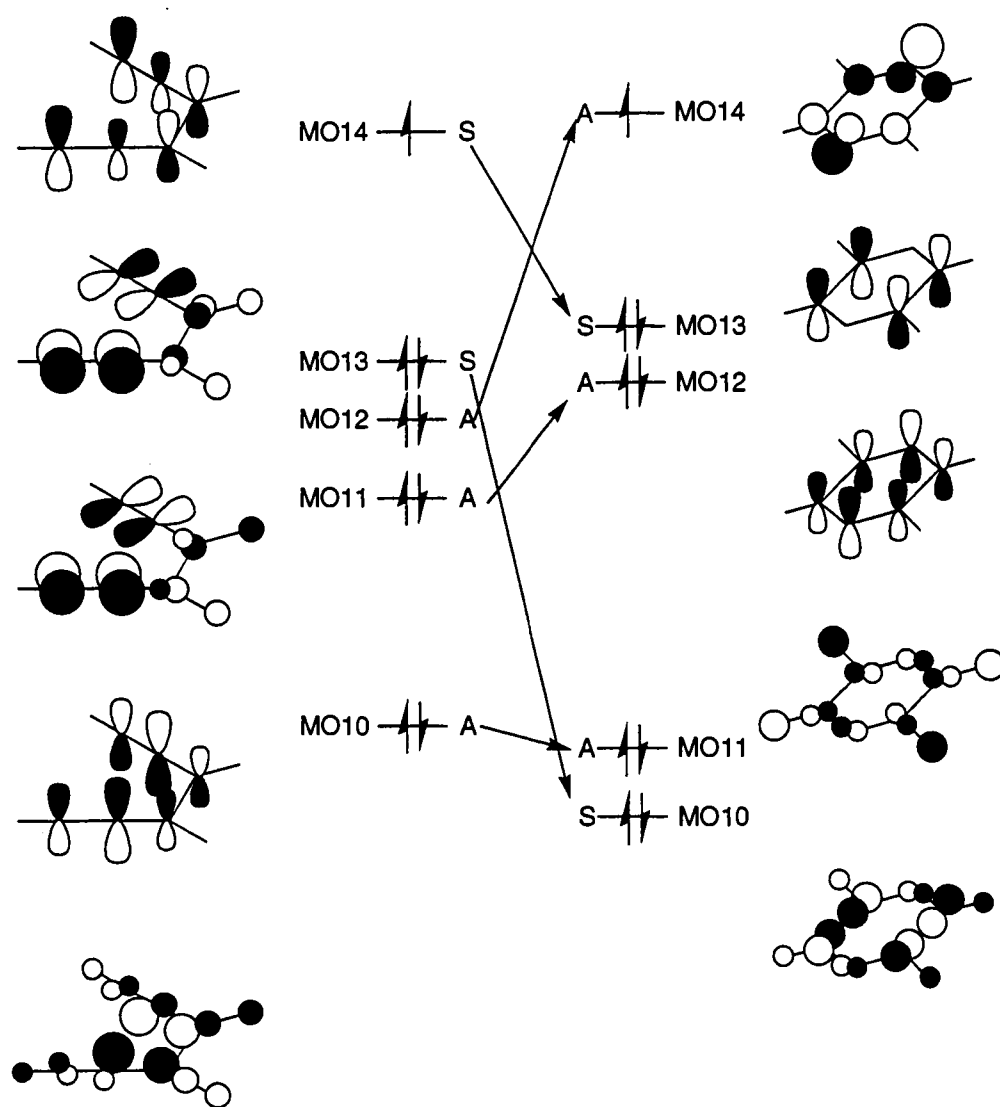


Figure 1.11. Orbital symmetry correlation diagram for the radical-cation Bergman cyclization.

It is interesting to point out that the correlation diagram for the 1,5-hexadiene radical cation cyclization (Figure 1.12) is also both orbital and state symmetry forbidden if we assume that C_2 symmetry holds throughout the reaction. In this case the theoretical prediction is contrast to the experimental result. Borden's unpublished ab initio calculations²² showed that the reaction may go through a localized model, i.e. the hole becomes localized in one double bond and then attacks the other one. In other words, the C_2 symmetry is broken instead of conserved in this radical cation reaction. We speculate that the multiple conformations of 1,5-hexadiene compared with the fixed conformation of enediyne may affect the path of the symmetry rule, i.e., make it easier to relax the symmetry. In the 1,5-hexadiene, the two π bonds are connected by a central σ bond which could have various conformations due to the single bond rotation.²³ The equilibrium conformation is probably an extended one instead of a chair-like one drawn in Figure 1.12, therefore we would expect this reaction would have both conformational and chemical barriers. Recent density function ab initio calculations²⁴ by Wu pointed out that the major barrier is a conformational one and no symmetry element is involved in this transformation. It is worthy to note that, for some of the radical cation reactions which are both orbital and state symmetry forbidden, lowering symmetry by the Jahn–Teller distortion might be a way to stabilize the transition state.²⁵

Attempts were tried to excite the ground state radical cation by photolysis using light from a 1000 W Tungsten lamp with and without 2424 Corning glass filter

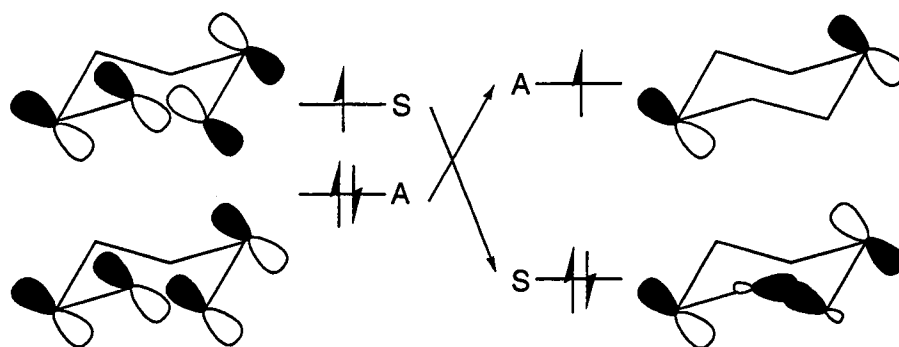


Figure 1.12. Orbital symmetry correlation of the 1,5-hexadiene radical cation cyclization.

(C.S.2-63, $\lambda > 580$ nm light will go through). Unfortunately, no reaction was observed. This might due to the weak efficiency of the absorption band.

5. CONCLUSION

Unlike radical cations of 1,5-hexadiene and 1,5-hexadiyne, radical cations of several enediynes prepared in this study can not cyclize. According to Woodward–Hoffmann theory, the cyclization is forbidden because the fixed conformation of enediyne necessitates a conservation of orbital symmetry.

6. EXPERIMENTAL

6.1. Synthetic procedures

All NMR spectra were recorded at ambient temperature on a Bruker AM-250 instrument. GC–MS was performed with a Hewlett-Packard 5890 chromatograph

equipped with a HP 5970 series mass selective detector. A 30 m capillary column containing DB-5 (95% methyl, 5% phenyl) as the stationary phase and helium carrier gas was used for separations. All GC separations were running under the following conditions unless otherwise noted: column temperature ramp 60 °C (3 min) → 280 °C @ 20 °C/min; injector temperature 250 °C; detector temperature 280 °C. Preparative GC was done on a Bendix GC 2300 equipped with a 1.5 m 20% SE30 column. Helium was used as carrier gas. The condition for the separation of compound **6** is following: column temperature 120 °C, injector temperature 160 °C, detector temperature 150 °C and the retention time is about 14 min. Flash column chromatography was accomplished with 230–400 mesh silica gel (SIP® Brand, Baxter cat.#C4582-87) as a solid support. THF, hexanes and benzene were distilled from sodium-benzophenone ketyl. All other reagents were purchased from commercial sources and used without further purification.

Preparation of *cis*-1,6-bis(trimethylsilyl)-3-hexene-1,5-diyne (**2**).

Method 1 (Heck reaction):¹¹ A 50 mL dried round-bottom flask was charged with Pd(PPh₃)₄ (910 mg, 0.06 equiv), CuI (200 mg, 0.08 equiv), benzene (20 mL) and BuNH₂ (4.1 mL, 40.7 mmol) under N₂ atmosphere at 0 °C. To the flask was added benzene (10 mL) solution of *cis*-dichloroethylene (1 mL, 13.3 mmol) and trimethylsilyl acetylene (4 mL, 2.1 equiv). The reaction was allowed to proceed at room temperature overnight. After removal of the solvent by rotary evaporator, the dark brown slurry was extracted with ether (3 × 15 mL) and the organic phase was

washed with 10% HCl, satd NaHCO₃ and brine, dried over MgSO₄ and concentrated. The resulting crude product was purified by flash column chromatography (hexanes) and the enediyne **2** ($R_f \approx 0.15$) was obtained as a colorless oil (2.42 g, 83%). ¹H NMR (CDCl₃): δ 5.84 (s, 2H), 0.22 (s, 18H); ¹³C NMR (CDCl₃): δ 120.59, 103.36, 101.88, -0.11; GC-MS: $t_R = 7.735$ min, m/z 220 (M⁺), 205 (base peak), 190, 177, 159, 145, 95, 73, 59, 53, 45. Lit²⁶. ¹H NMR (CDCl₃): δ 5.83 (s, 2H), 0.21 (s, 18H); ¹³C NMR (CDCl₃): δ 120.58, 103.38, 101.83, -0.13.

Method 2 (Carbenoid coupling reaction):¹² To hexamethyldisilazane (46.4 mL, 220 mmol) in a 200 mL round-bottom flask equipped with a water-jacketed condenser was added propargyl alcohol (25.7 mL, 2 equiv) dropwise with stirring. The reaction is exothermic. After addition, the mixture was left to stir at room temperature overnight. It was then gently heated to near reflux for 3 h, after which time the light-yellow mixture was poured (in small portions) onto crushed ice and washed with cold water repeatedly until the aqueous layer was neutral. Silyl ether **14** was obtained as a colorless oil by distillation at 102–110 °C (49.5 g, 87%). ¹H NMR (CDCl₃): δ 4.28 (d, 2H, $J = 2.4$ Hz), 2.40 (t, 1H, $J = 2.4$ Hz), 0.18 (s, 9H); GC-MS [ramp 30 °C (3 min) $\rightarrow$ 220 °C @ 20 °C/min]: $t_R = 3.460$ min, m/z 127 (M - 1), 113, 83 (base peak), 73, 59, 55, 45. Ether **14** (11.8 g, 91.7 mmol) was dissolved in THF (80 mL) and cooled to -20 °C. To this was added butyllithium (60 mL 1.6 M hexanes solution, 1.05 equiv) dropwise. After the addition, the reaction mixture was allowed to stir for a further 20 min at -20 °C before an excess of TMSCl (14 mL, 1.2 equiv) was added. The ethanol/dry ice bath was replaced with an ice bath and the

reaction mixture was stirred for 3 h. The reaction was quenched by the addition of ice-water (5 mL) and ice-cooled 3% HCl (5 mL). The mixture was extracted with ether and washed with NaHCO₃ solution and brine. The organic phase was dried over MgSO₄. Evaporation of solvent gave the bis-silylated product **15** which was then brominated without further purification. Some O-desilylation might occur during this workup but both OH and OTMS were readily brominated by the following procedure. To a solution of PPh₃ (26.0 g, 1.1 equiv) in CH₂Cl₂ (50 mL) at 0 °C was added bromine (5.1 mL, 1.1 equiv) dropwise and the resulting yellow suspension was allowed to stir at 0 °C for 0.5 h. To this was added **15** (91.7 mmol) dropwise at 0 °C. The yellow suspension became clear, dark brown solution. It was stirred at 0 °C for another 10 min before pentane (180 mL) was added and the resulting white suspension was filtered through a short bed of silica gel. The bed was washed with pentane (1 × 40 mL) and the combined organic phase was concentrated to give **16** as a black oil (15.0 g, 86%) which was used in the next step without further purification. ¹H NMR (CDCl₃): δ 3.90 (s, 2H), 0.18 (s, 9H); GC-MS: *t*_R = 4.863 min, *m/z* 192 (*M* + 2), 190 (*M*⁺), 177 (base peak), 175, 149, 147, 139, 137, 125, 123, 111, 96, 83, 66, 53, 43. To a THF (32 mL) solution of hexamethyldisilazane (3.7 mL, 1.1 equiv) at -10 °C was added butyllithium (20 mL, 0.97 M hexanes solution, 1.1 equiv) dropwise. After 1 h stirring, HMPA (3.07 mL, 1.1 equiv) was added and the solution color changed from light-yellow to brown. The resulting solution was allowed to stir for another 5 min before it was transferred over a period of 1 h to a THF (64 mL) solution of **16** (2.6 g, 15.9 mmol) at -78 °C.

Upon completion of the base addition, the solution was allowed to stir overnight and the temperature was allowed to rise slowly to room temperature. After about 15 h, the solution was poured into ice-cooled NH_4Cl solution. The mixture was extracted with ether and the combined organic phase was washed successively with 10% HCl , satd NaHCO_3 and brine. The organic phase was dried over Na_2SO_4 and concentrated. The *cis* and *trans* enediynes (0.98 g, 51%) were separated by flash column chromatography (hexanes). The *trans* isomer **3** eluted first ($R_f \approx 0.30$) from the column as a white solid. ^1H NMR (CDCl_3): δ 6.00 (s, 2H), 0.19 (s, 18H). The *cis* isomer **2** eluted second ($R_f \approx 0.15$) from the column as a colorless oil. The spectroscopic data for **2** were identical to those prepared from Heck reaction.

Preparation of ***cis*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (4)**. By using the Heck reaction as described in the preparation of **2**, enediyne **4** was obtained as a colorless oil (3.4 g, 65%) from the reaction of *cis*-dichloroethylene (2.68 g, 27.7 mmol) with 3,3-dimethylbutyne (5 g, 2.2 equiv). $R_f \approx 0.22$. ^1H NMR (CDCl_3): δ 5.71 (s, 2H), 1.29 (s, 18H); ^{13}C NMR (CDCl_3): δ 118.94, 105.74, 76.90, 31.00, 28.32; GC-MS, $t_R = 7.343$ min, m/z 188 (M^+), 173, 158, 143 (base peak), 131, 115, 105, 91, 77, 65, 51, 41. lit¹³. ^1H NMR (CDCl_3): δ 5.70 (s, 2H), 1.27 (s, 18H); ^{13}C NMR (CDCl_3): δ 118.96, 105.81, 76.84, 31.04, 28.32; GC-MS: m/z 188 (M^+) (100), 173 (58), 158 (37), 143 (79), 131 (62), 115 (41), 105 (30), 91 (64), 77 (43), 65 (20), 57 (30). Byproduct **2,2,7,7-tetramethyl-octa-3,5-diyne (12)** was obtained as white crystals. $R_f \approx 0.39$. ^1H NMR (CDCl_3): δ 1.23 (s, 18H); ^{13}C NMR (CDCl_3): δ 86.25,

63.80, 30.58, 28.00.

***cis*-2,2,9,9-Tetramethyl-5,6-Dideuterio-5-decene-3,7-diyne (5).** By using the same procedure as described in the preparation of **2** except that the starting material deuterated dichloroethylene (590 mg) was a *cis/trans* mixture (brought from C/D/N isotopes Inc., Canada, with a certificate of analysis of 99.86%-*d*₂, 51.8% *trans* and 47.8% *cis*), enediyne **5** was obtained as a pale-yellow oil (270 mg, 50%) after flash column chromatography. ¹H NMR (CDCl₃): δ 1.29 (s); ¹³C NMR (CDCl₃): δ 118.52 (t, ¹J_{C-D} = 25.6 Hz), 105.75, 76.75, 30.97, 28.27; GC-MS: *t*_R = 7.351 min, *m/z* 190 (M⁺), 175, 160, 145, 133, 130, 117, 107, 93, 92, 78, 65, 53, 41.

Preparation of ***cis*-3-*tert*-butyl-7,7-dimethyl-3-octene-1,5-diyne (6).**¹³ To a lime yellow pentane solution (64 mL) of AlCl₃ (9.88g, 1.3 equiv) at 0 °C was added a pentane solution (15 mL) of bis(trimethylsilyl)acetylene (10 g, 58.7 mmol) and a pentane solution (5 mL) of pivaloyl chloride (7.6 mL, 1.1 equiv). After the addition was complete, the mixture was stirred at 0 °C for 3 h, after which time the mixture was warmed up to room temperature and stirred overnight. The reaction was quenched by pouring the mixture onto crushed ice with vigorous stirring. The organic phase was washed with satd NaHCO₃ and brine, dried over MgSO₄, and concentrated to give **17** as a brown liquid (10.13 g, 94%). ¹H NMR (CDCl₃): δ 1.20 (s, 9H), 0.25 (s, 9H); ¹³C NMR (CDCl₃): δ 193.94, 100.14, 99.41, 44.49, 25.99, 1.91; GC-MS [ramp 45 °C (3 min) → 280 °C]: *t*_R = 7.147 min, *m/z* 167 (M - 15), 154, 139,

125, 97, 73, 57 (base peak), 43, 41. Lit²⁶. ¹H NMR (CCl₄): δ 1.16 (s, 9H), 0.25 (s, 9H); MS: *m/z* 182 (0.4), 167 (17), 139 (74), 125 (97), 97 (30), 75 (12), 73 (29), 57 (100). To a THF (86 mL) solution of TiCl₄ (7.8 mL, 1.3 equiv) under Ar atmosphere at 0 °C was added pyridine (3.9 mL) and Zinc dust (9.63 g, 2.6 equiv). The solution changed color from yellow to green then dark brown. After 5 min, a THF (8 mL) solution of **17** (10.1 g, 55.7 mmol) was added over a period of 15 min. The mixture was stirred at 0 °C for 1.5 h before the ice bath was removed. It was then heated to reflux overnight. The reaction was quenched by adding 10% K₂CO₃ solution into the ice-cooled mixture. Dark precipitate appeared. The slurry was extracted with pentane (6 × 50 mL). The yellow organic phase was washed with brine and dried over Na₂SO₄. After solvent evaporation, a yellow slurry was obtained (8.00 g, 87%). Further purification by flash column chromatography (hexanes) gave a white crystal (3.28 g, *R_f* ≈ 0.65). It was a mixture of enediynes **18**, **19** and **20** and could not be separated either by flash chromatography or sublimation. ¹H NMR (CDCl₃): δ 1.33 (s, C(CH₃)₃ of **20**), 1.31 (s, olefinic C(CH₃)₃ of **18**), 1.25 (s, acetylenic C(CH₃)₃ of **18**), 0.24 (s, olefinic Si(CH₃)₃ of **18**), 0.18 (s, both acetylenic Si(CH₃)₃ of **18** and **20**); ¹³C NMR (CDCl₃): δ 149.60 (of **18**), 138.99 (of **20**), 131.46 (of **18**), 117.09 (of **18**), 111.39 (of **18**), 107.11 (of **18**), 105.60 (of **20**), 103.75 (of **20**), 77.30 (of **18**), 37.27 (of **18**), 35.36 (of **18** and **20**), 30.68 (of **18**), 29.64 (of **18**), 29.43 (of **20**); GC-MS, **20**: *t_R* = 10.395 min, *m/z* 332 (M⁺), 259, 229, 187, 155, 151, 97, 73 (base peak), 57; **18** and **19**: *t_R* = 10.475 min, *m/z* 332 (M⁺), 245, 167, 156, 97, 73 (base peak), 57. To a solution of enediynes **18**, **19** and **20** (2.05 g, 6.2 mmol) from last step in

anhydrous methanol (60 mL) was added K_2CO_3 (0.54 g, 3.9 mmol) at room temperature. The reaction was stirred overnight, after which time it was quenched with water (50 mL). The solution was extracted with pentane (3×30 mL), and the organic phase was concentrated and distilled (0.02 torr) at ca. 75 °C to give **21** as a colorless liquid in a nitrogen trap. 1H NMR ($CDCl_3$): δ 3.80 (s, 2H), 1.36 (s, 18H); GC–MS: t_R = 7.236 min, m/z 188 (M^+), 173, 158, 143, 131, 115, 94, 79, 77, 65, 57 (base peak), 41. lit¹³. 1H NMR ($CDCl_3$): δ 3.79 (s, 2H), 1.35 (s, 18H); ^{13}C NMR ($CDCl_3$): δ 138.87, 93.78, 83.64, 35.60, 29.83; MS, m/z 188 (23), 173 (12), 158 (17), 143 (23), 131 (53), 115 (32), 94 (33), 91 (26), 77 (20), 65 (9), 57 (100). To the remained dark brown residue was added TBAF (3 mL 1M THF solution, 3 mmol). The reaction was run at room temperature for 3 h before quenched with water (10 mL). It was then extracted with pentane (3×10 mL), and the organic phase was concentrated into ca. 10 mL solution. By preparative GC, the pure *cis* enediyne **6** was obtained as a colorless liquid ($R_f \approx 0.22$). Its retention time on the preparative column was ca. 14 min. 1H NMR ($CDCl_3$): δ 5.79 (s, 1H), 3.35 (s, 1H), 1.28 (s, 9H), 1.15 (s, 9H); ^{13}C NMR ($CDCl_3$): δ 142.59, 113.91, 104.78, 84.52, 81.67, 77.30, 36.00, 31.03, 29.00, 28.30; GC–MS: t_R = 7.369 min, m/z 188 (M^+), 173, 158, 143, 131, 115, 105, 91, 77, 65, 57, 41 (base peak). lit¹³. 1H NMR ($CDCl_3$): δ 5.77 (s, 1H), 3.34 (s, 1H), 1.26 (s, 9H), 1.13 (s, 9H); ^{13}C NMR ($CDCl_3$): δ 142.63, 113.93, 104.80, 84.53, 81.68, 77.32, 36.03, 31.02, 28.98, 28.32; MS: m/z 188 (M^+) (81), 173 (83), 158 (64), 143 (100), 131 (69), 91 (53), 77 (34), 65 (14), 57 (41). Its *trans* isomer **24** has $R_f \approx 0.39$.

Preparation of ***cis*-6-dodecene-4,8-diyne (7)**. By using the same procedure as in the preparation of **2**, enediyne **7** was obtained as a brown oil (2.43 g, 83%) from the reaction of *cis*-dichloroethylene (1.77 g, 0.018 mmol) and 1-pentyne (3.8 mL, 2.1 equiv). $R_f \approx 0.15$ (hexanes). $^1\text{H NMR}(\text{CDCl}_3)$: δ 5.73 (s, 2H), 2.37 (t, 6H, $J = 6.9$ Hz), 1.57 (sextet, 4H, $J = 7.2$ Hz), 1.03 (t, 4H, $J = 7.3$ Hz); $^{13}\text{C NMR}(\text{CDCl}_3)$: δ 118.92, 97.62, 78.45, 22.05, 21.71, 13.36; GC-MS: $t_R = 7.849$ min, m/z 16 (M^+), 131, 115, 103, 91 (base peak), 77, 63, 51, 39.

Preparation of **1,2-bis(3',3'-dimethylbutynyl)benzene (8)**.²⁷ By using the same procedure as in the preparation of **2** except that after addition of the reagents, the reaction was run at room temperature for 2 days and refluxed for 1 day (with a dry ice condenser), enediyne **8** was obtained as a yellow liquid (0.930 g, 29%) from the reaction of 1,2-dibromobenzene (1.6 mL, 13.3 mmol) and 3,3-dimethylbutyne (3.5 mL, 2.1 equiv). $R_f \approx 0.10$ (hexanes). $^1\text{H NMR}(\text{CDCl}_3)$: δ 7.35 (dd, 2H, $J = 5.8$, 3.4 Hz), 7.14 (dd, 2H, $J = 5.8$, 3.4 Hz), 1.35 (s, 18H); $^{13}\text{C NMR}(\text{CDCl}_3)$: δ 131.92, 127.00, 126.13, 101.84, 78.12, 31.12, 28.14; GC-MS: $t_R = 8.416$ min, m/z 238 (M^+) (base peak), 223, 208, 193, 165, 152, 115, 103, 89, 57, 41.

Preparation of ***trans*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (10)**.¹⁴ Enediyne **4** (0.150 g, 0.798 mmol) and 2,4,6-triphenylpyrylium tetrafluoroborate ($\text{TPP}^+\text{BF}_4^-$, 0.03 g, 0.1 equiv) was dissolved in acetonitrile (10 mL) and isopropyl alcohol (5 mL). The solution was then transferred into a Pyrex tube (ca. 30 mL).

It was subsequently degassed three times by the standard freeze-pump-thaw cycle and then sealed. A 450 W Hanova UV lamp was used to irradiate the tube immersed in a water bath for 26.5 h. The highest temperature of the bath was recorded as 45 °C. The color of the solution changed from lemon yellow to golden yellow after the irradiation. GC–MS showed that about 50% of *cis* enediyne had been isomerized to the *trans*. Later irradiation experiments showed that even without $\text{TPP}^+\text{BF}_4^-$ the isomerization still occurred. The *trans* enediyne **10** ($R_f \approx 0.34$) was separated from the *cis* ($R_f \approx 0.22$) by flash column chromatography (hexanes). ^1H NMR (CDCl_3): δ 5.86 (s, 2H), 1.23 (s, 18H); ^{13}C NMR (CDCl_3): δ 120.04, 102.74, 30.90, 28.09; GC–MS: $t_R = 8.189$ min, m/z 188 (M^+), 173, 158, 143, 131, 115, 105, 91, 77, 65, 51, 41 (base peak). lit¹³. ^1H NMR (CDCl_3): δ 5.84 (s, 2H), 1.21 (s, 18H); ^{13}C NMR (CDCl_3): δ 120.08, 102.73, 77.74, 30.90, 28.12; GC–MS: m/z 188 (M^+) (100), 173 (56), 158 (31), 143 (67), 131 (46), 115 (30), 105 (21), 91 (45), 77 (30), 65 (14), 57 (17).

6.2. ESR sample preparation and instrumentation

The purification of the sample was done by one of the following three methods: flash column chromatography, preparative GC or TLC. Flash column chromatography was the most commonly used purification method. After the appropriate fraction was collected, the solvent was removed by rotary evaporator. The remaining solvent was then removed under high vacuum. Caution should be drawn that some enediynes like **4**, **5** and **6** were also easily removed under high

vacuum. The pure compound was then dissolved in CFCl_3 and then vacuum transferred into a ESR tube. The preparative GC was only used for the purification of enediyne 6. The colorless liquid in the collecting tube from the preparative GC was dissolved in CFCl_3 and then vacuum transferred into a ESR tube. The preparative TLC was used for both enediyne 5 and 6. The TLC plates were Whatman Ltd's PE SIL G/UV flexible plates with 250 mm layer. They were cut into ca. 65×40 mm pieces. A capillary tube was used to spot the pre-purified enediyne pentane solution onto the plate and it was developed in a pentane chamber. After developing was complete, the plate was taken out of the chamber and the pentane on the plate was evaporated by a heat gun for about 5 s. The enediyne area on the "dried" plate was detected under a UV (254 nm) lamp, and then peeled off into a vial, and extracted with CFCl_3 . The solution was then vacuum transferred into a ESR tube. It was observed that the ESR spectra of enediyne 6 were identical for samples prepared from either preparative GC or TLC.

All ESR samples were prepared in our home-made 4 mm O.D. ESR tubes. They all consist of, from the bottom, a 95 mm long quartz tube, either Spectrosil or Suprasil, a 35 mm long graded seal, a 80 mm long Pyrex tube and a 12/30 ground glass joint. There is a constriction in the Pyrex part where its distance from the bottom is about 165 mm. After the compound and Freon were transferred into the tube under vacuum, they were degassed three times by the standard freeze-pump-thaw cycle using liquid nitrogen and then sealed off at the constriction. The sample was allowed to reach room temperature and then shaken vigorously to assure

uniform mixing. The sample was then placed into a liquid nitrogen Dewar to be ready for the irradiation.

The radiation equipment was an Atomic Energy of Canada Ltd. Gammacell 200 using a ^{60}Co source ($t_{1/2} = 5.27$ years). On January 14, 1995, it had a calibrated dose rate of 21.52 krad/h. During the course of this work, the total irradiation dose for a sample was in the range of 0.15–0.35 Mrad.

The ESR instrument used in this work was a Bruker 200-D spectrometer installed in 1982. It was equipped with an ER 4111VT variable temperature unit and an ER 035M NMR gaussmeter. The center of the field was usually set at between 3295 and 3305 G.

6.3. Semi-empirical calculations and simulations

AM1 and MINDO/3 calculation were performed by using MOPAC 6.0 in UNIX mainframe, which was allowed to used by Dr. Bartmess in our department. "VECTORS" and "RHF" were used as keywords when orbital pictures were needed. "UHF" was used when the spin populations were needed to calculate the ESR coupling constants. "SYMMETRY" was used to enforce symmetry.

Isotropic ESR spectral simulations were done by using an IBM EPRSRC.015 software package written for the Aspect 2000 Computer, which was interfaced with the Bruker instrument.

7. REFERENCES AND NOTES

1. For a review, see: Pappas, Robert S. Ph. D. Dissertation, The University of Tennessee, Knoxville, August 1995, p6.
2. Doering, W. von E.; Toscano, V. G.; Beasley, G. H. Kinetics of the Cope rearrangement of 1,1-dideuteriohexa-1,5-diene. *Tetrahedron* **1971**, *27*, 5299–5306.
3. (a) Guo, Q.-X.; Qin, X.-Z.; Wang, J. T.; Williams, F. Radical cation cyclizations of 1,5-hexadiene to cyclohexene via the cyclohexane-2,5-diyl radical cation intermediate. *J. Am. Chem. Soc.* **1988**, *110*, 1974–1976.
(b) Williams, F. Inverted potential-energy surfaces in the radical-cation Cope rearrangements of hexa-1,5-diene and semibullvalene. *J. Chem. Soc., Faraday Trans.* **1994**, *90*, 1681.
4. MINDO/3 calculation gave that the cleavage of C3–C4 bond in II required 34 kcal/mol. See: Bauld, N. L.; Bellville, D. J.; Pabon, R.; Chelsky, R.; Green, G. Theory of cation-radical pericyclic reactions. *J. Am. Chem. Soc.* **1983**, *105*, 2378–2382.
5. Chen, G.-F.; Williams, F. Cope rearrangement of the endo-5-vinylbicyclo[2.2.1]hept-2-ene (endo-5-vinylnorborn-2-ene) radical cation to the cis-bicyclo[4.3.0]nona-3,7-diene (cis-3a,4,7,7a-tetrahydroindene) radical cation at 100–150 K. A matrix-isolation ESR study. *J. Chem. Soc., Chem. Commun.* **1992**, 670–672.
6. Dai, S.; Pappas, R. S.; Chen, G.-F.; Guo, Q.-X.; Wang, J.-T.; Williams, F.

Radical cation Cope rearrangement of 1,5-hexadiyne to 1,2,4,5-hexatetraene (Bis(allene)) at 77 K. *J. Am. Chem. Soc.* **1989**, *111*, 8759–8761.

7. Jones, R. R.; Bergman, R. G. *p*-Benzyne. Generation as an intermediate in a thermal isomerization reaction and trapping evidence for the 1,4-benzenediyl structure. *J. Am. Chem. Soc.* **1972**, *94*, 660–661.
8. Bergman, R.G. Reactive 1,4-dehydroaromatics. *Acc. Chem. Res.* **1973**, *6*, 25–31.
9. For a general review see:
(a) Nicolaou, K. C.; Dai, W.-M. Chemistry and biology of enediyne-cytostatic antibiotics. *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 1387–1416.
(b) *Enediyne Antibiotics as Antitumor Agents*; Borders, D. B.; Doyle, T. W., Eds.; Marcel-Dekker: New York, 1995.
10. Nicolaou, K. C.; Hummel, C. W.; Pitsinos, E. N.; Nakada, M.; Smith, A. L.; Shibayama, K.; Saimoto, H. Total synthesis of calicheamicin γ_1^1 . *J. Am. Chem. Soc.* **1992**, *114*, 10082–10084.
11. Vollhardt, K. P. C.; Winn, L. S. Stereospecific synthesis of cis- and trans-1,6-bis(trimethylsilyl)hex-3-ene-1,5-diyne. *Tetrahedron Lett.* **1985**, *26*, 709–712.
12. Huber, R. S.; Jones, G. B. A mild and efficient route to the pharmacophore of the enediyne antitumor agents-(Z)-1,6-[bis(trimethylsilyl)]-hex-3-ene-1,5-diyne via a novel carbenoid coupling reaction. *Tetrahedron Lett.* **1994**, *35*,

2655–2658.

13. Roth, W. R.; Hopf, H.; Horn, C. *Chem. Ber.* Propargyl-stabilisierungsenergie. **1994**, 127, 1781–1795.
14. Ramkumar, D.; Kalpana, M.; Varghese, B.; Sankararaman, S.; Jagadeesh, M. N.; Chandrasekhar, J. Cyclization of enediyne radical cations through chemical, photochemical, and electrochemical oxidation: The role of state symmetry. *J. Org. Chem.* **1996**, 61, 2247–2250.
15. (a) Tolkachev, V. A.; Chkheidze, I. I.; Buben, N. Y. *Zhur. Strukt. Khim.* **1962**, 3, 709. This paper reported the coupling constants for phenyl radical recorded at 77 K are: $A_{\text{ortho}} = 18 \pm 2$ G, $A_{\text{meta}} = 7 \pm 2$ G.
(b) *Atlas of Electron Spin Resonance Spectra*, Bielski, B. H. J.; Gebicki, J. M., Eds.; Academic Press: New York, 1967, p466.
(c) Kasai, P. H.; Hedaya, E.; Whipple, E. B. *J. Am. Chem. Soc.* **1969**, 91, 4364. This paper reported the coupling constants for phenyl radical recorded at 4 K are: $A_{\text{ortho}} = +17.4 \pm 0.1$ G, $A_{\text{meta}} = +5.9 \pm 0.1$ G, $A_{\text{para}} = +1.9 \pm 0.1$ G.
16. Dr. Wang did not record the color for the parent enediyne radical cation **1** and **9** and the samples were not available to repeat the experiment.
17. *The Chemical Aspects of Light*, Bowen, E. J., Ed.; Oxford University Press: New York, 1946, p244.
18. For unsubstituted linear hexatriene radical cation, the transition energy for ${}^2A_u \rightarrow {}^2B_g^-$ (weak band) is 1.93 eV (642 nm) and that for ${}^2A_u \rightarrow {}^2B_g^+$ (strong

band) is 3.16 eV (392 nm): (a) Shida, T.; Kato, T.; Nosaka, Y. *J. Phys. Chem.* **1977**, *81*, 1095. (b) Bally, T.; Nitsche, S.; Roth, K.; Haselbach, E. *J. Am. Chem. Soc.* **1984**, *106*, 3927 and references cited therein.

For *tert*-butyl capped linear hexatriene radical cation, the two transition energies are 1.98 eV (626 nm) and 2.88 eV (430 nm) respectively: (c) Bally, T.; Roth, K.; Tang, W.; Schrock, R. R.; Knoll, K.; Park, L. Y. *J. Am. Chem. Soc.* **1992**, *114*, 2440.

19. van der Hart, W. J.; Oosterveld, E.; Molenaar-Langeveld, T. A.; Nibbering, N. M. M. Photodissociation and structures of $[C_6H_4]bul^+$ ions generated from cyanobenzene and other precursors. *Organic Mass Spectrometry* **1989**, *24*, 59.
20. Wu, Y.-D.; Wang, D.-P.; Guo, Q.-X.; Williams, F. to be submitted for publication.
21. MOPLOT (version 3.1) was sent to us as a gift from Professor Bally, Thomas, Institut de Chimie Physique, University of Fribourg, Perolles, CH-1700 Fribourg, Switzerland.
22. Adapted from Professor W. T. Borden's unpublished work and private communication with Professor T. F. Williams.
23. See general discussion, *J. Chem. Soc., Faraday Trans.*, **1994**, *90*(12), 1733.
24. Private communication between Professor Y.-D. Wu and Professor T. F. Williams.
25. Bischof, P. Forbidden Radical Rearrangements. Comparison with Potential

- Surfaces of Jahn–Teller Radicals. *J. Am. Chem. Soc.* **1977**, *99*, 8145–8149.
26. Birkofer, L.; Richtzenhain, K. Silyl-Derivate von Pyrazol, Isoxazol and 1,2,3-Triazol. *Chem. Ber.* **1979**, *112*, 2829–2836.
27. John, Jens Afred Hans. Ph. D. Dissertation, The University of South Carolina, 1994, p136.

PART TWO

A SOLID-PHASE SYNTHESIS AND COMBINATORIAL APPROACH TO 2,3-DIHYDROBENZO[d]ISOTHIAZOLE 1,1-DIOXIDES

1. INTRODUCTION

1.1. Therapeutic approaches to HIV

Human immunodeficiency virus (HIV) is a retrovirus that produces a fatal immunodeficiency condition by destroying the CD4⁺ T lymphocytes and thus causing *acquired immune deficiency syndrome* (AIDS). The battle against AIDS started two decades ago. At that time, HIV infection was regarded as a progressive, lethal disease for which no treatment was available. In 1987, AZT, a nucleoside reverse transcriptase inhibitor (NRTI), became the first AIDS drug. The virus responded quickly with an AZT-resistant strain, and it seems like the search for new drug might never catch up with the rapid mutation of the virus. Since late 1995, however, several related advances have led to an optimistic prospect for most patients who receive treatment today. The most significant one is the introduction of the revolutionary multidrug combination therapy consisting of more powerful protease inhibitors and non-nucleoside reverse transcriptase inhibitors (NNRTIs). The battle has started to be won on the side of the human because the death toll has declined in the developed countries. Nevertheless, the international picture for HIV is getting worse. According to the United Nations' AIDS program, the death toll of AIDS had reached 6.4 million and an additional 22.6 million people were infected with the virus by the end of 1996.¹ Much research has led to the following understanding of the life cycle of the AIDS virus: 1. The virus has two strands of RNA and some enzymes encased in a coating. 2. When the virus encounters a T cell, proteins on the virus coating bind to both CD4 and co-

receptors on the cell. 3. The virus then enters the cell and converts its RNA into DNA by an enzyme called reverse transcriptase (RT). 4. Next, an enzyme called integrase incorporates the virus' genetic code into the T cell's DNA. 5. The viral DNA uses the cell's machinery to replicate viral RNA and proteins. 6. An enzyme called protease cuts the viral proteins into shorter pieces so that they can be incorporated into new viruses. 7. The viruses bud off and attack other T cells and the cycle goes on. The life cycle of the virus presents a number of targets for drug action.² For example, the reverse transcriptase is the target of AZT. The triphosphate derivatives of AZT and other NRTIs, which are produced by endogenous kinases in vivo, act primarily through competitive binding and chain termination. NNRTIs, on the other hand, act noncompetitively at an allosteric site.³ NNRTIs also show a great diversity in chemical structure, but each elicits similar kinetic behavior in enzyme inhibition assays.⁴

HIV has two major population subtypes, e.g., HIV-1 and HIV-2, which differ in their genetic code. This may be related to the rapid mutation of the virus and its ability to evade the host's immune defense. While NRTIs show only modest selectivity between HIV-1 and HIV-2, NNRTIs are effective against HIV-1 only. In fact, the emergence of AZT-resistant^{5,6} and NNRTI-resistant^{7,8} HIV strains is the major concern of current chemotherapy. Because resistance can occur readily, and no single drug on the market is powerful enough to suppress HIV on its own, multidrug combinations that both maximize potency and reduce the likelihood of resistance are being used to solve the resistance problem. This so-called cocktail

treatment usually uses different combinations of NRTI, NNRTI and protease inhibitor, all targeting a different stage of the life cycle of the virus.

NRTIs, NNRTIs and protease inhibitors are three categories of drugs available for AIDS treatment (Table 2.1). As for the future development of new HIV drugs, two criteria should take priority over other considerations. First of all, higher potency and efficacy is desired. Most of the current drugs require dosage three times a day to achieve efficacy. If a patient skips one dose, the possibility will arise that the drug concentration in that patient's blood may drop to a level that the surviving virus can mutate to a resistant virus. Thus, once-daily drugs are preferred since they can maintain an effective concentration in blood for 24 h. Second, lower cost is needed. A recommended triple-drug combination therapy may cost around \$12,000 a year per patient. However, most HIV-infected patients, especially those in the developing countries, are indigent and they can not even afford to have single drug therapy. NNRTIs make up a category that may satisfy these two criteria because: 1) The first and the only once-daily AIDS drug is an NNRTI (Sustiva®); 2) NNRTIs have a great variety of structures, some of which allow some simple entities be developed into low-cost and high-efficacy drugs.

1.2. 2,3-Dihydrobenzo[d]isothiazole 1,1-dioxides (sultams) as non-nucleoside reverse transcriptase inhibitors

Sultams are cyclic sulfonamides consisting of two benzene rings and one heterocyclic five membered ring (see compounds **34**). Developing sultams as

Table 2.1. Available drugs for HIV treatment*

Brand Name	Generic/Common Names	Manufacturer (year)
Nucleoside Reverse Transcriptase Inhibitors (NRTIs)		
Retrovir®	zidovudine/AZT	Glaxo Wellcome (1987)
Videx®	didanosine/ddI	Bristol-Myers Squibb (1991)
HIVID®	zalcitabine/ddC	Roche Laboratories (1992)
Zerit®	stavudine/d4T	Bristol-Myers Squibb (1994)
Epivir®	lamivudine/3TC	Glaxo Wellcome (1995)
Combivir®	lamivudine/zidovudine	Glaxo Wellcome (1995)
Non-Nucleoside Reverse Transcriptase Inhibitors (NNRTIs)		
Viramune®	nevirapine	Roxane Laboratories (1995)
Rescriptor®	delavirdine	Pharmacia & Upjohn (1997)
Sustiva®	efavirenz	Dupont Pharmaceuticals (1998)
Protease Inhibitors (PIs)		
Invirase®	saquinavir	Roche Laboratories (1995)
Norvir®	ritonavir	Abbott Laboratories (1996)
Crixivan®	indinavir	Merck & Company (1996)
Viracept®	nelfinavir	Agouron Pharmaceuticals (1997)
Fortovase®	saquinavir (Gel capsule)	Roche Laboratories (1998)

*As of December 1998.

NNRTIs is a successful story of computer-assisted structure screening via pharmacophore mapping techniques. Compound **34z** ($R^1 = R^3 = R^4 = H$, $R^2 = Me$, NSC-108406) was chosen from a pool of compounds in the National Cancer Institute (NCI) database and predicted to be a potential NNRTI by these computer strategies. Compound **34z** was synthesized in our lab and showed to be active against HIV with an EC_{50} in the 10^{-6} M range.⁹ Because of their simple structure, sultams have potential to be developed into an inexpensive AIDS drug provided that more potent derivatives can be discovered.

2. STATEMENT OF THE PROBLEM

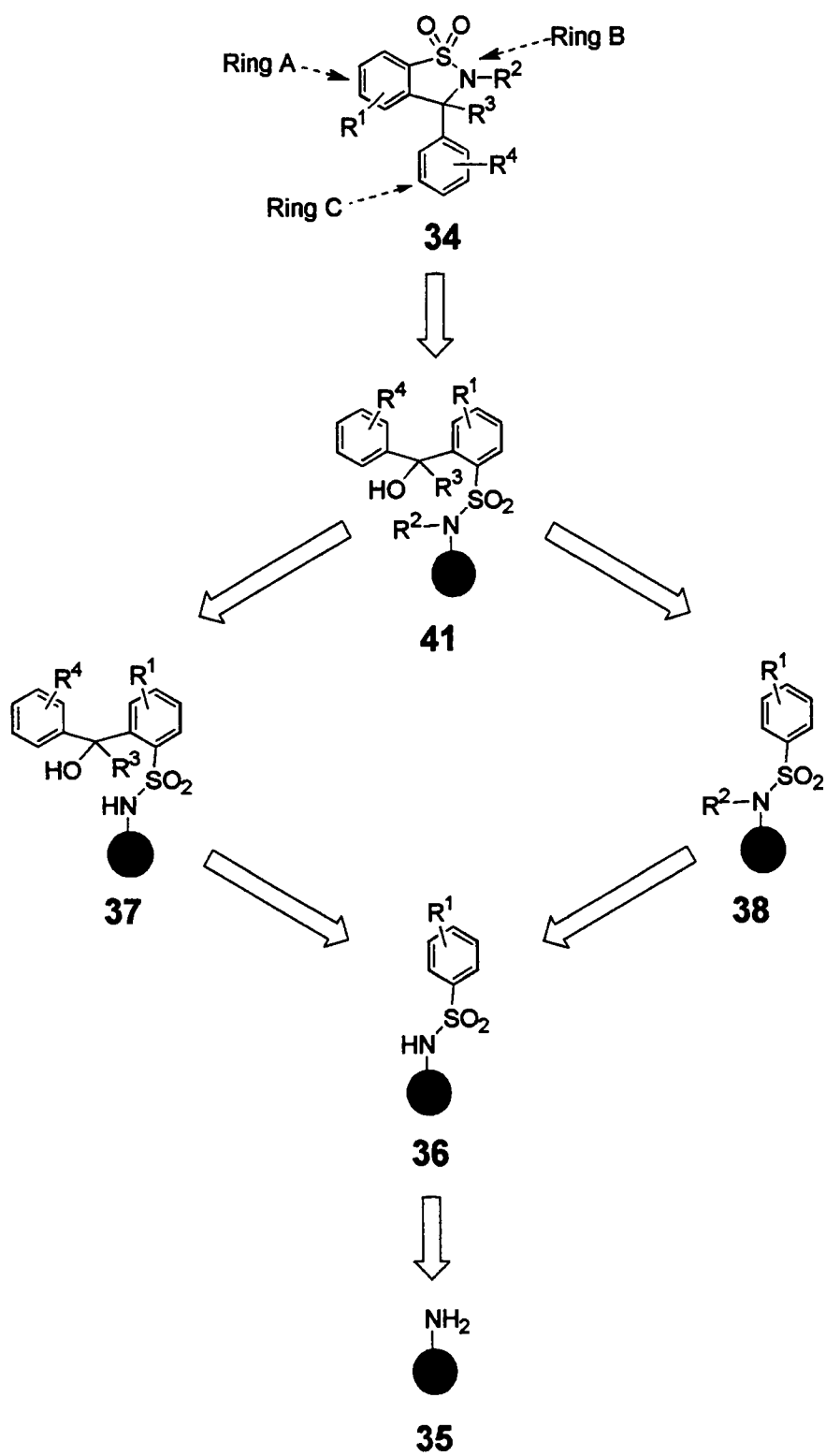
2.1. Objectives

This sultam project is an NCI research contract. With the knowledge that **34z** is active against HIV, our next goal is to optimize this lead by creating a library of sultam derivatives for screening and structure–activity relationship (SAR) studies. Conventional multistep solution-phase methods¹⁰ that require chromatographic purification at each step were initially employed for synthesis of **34z** and its analogues. This chapter describes how a general solid-phase method was developed to serve the same purpose with its advantages and limitations. Ring B modifications, i.e. changing R^2 and introducing a CF_3 at the chiral carbon atom, will also be discussed.

2.2. Research plan

At first, we were greatly influenced by Ellman's "traceless" solid-phase methods¹¹ wherein the polymer linker arm is removed, leaving a molecule unmodified at the point of former attachment to the linker arm. These methods that were based on a germanium linkage to ring A of the sultam seemed especially amenable to sultam synthesis. However, there was no commercially available Ge-linked resin at that time, and its synthesis required a rather lengthy process.¹² This forced us to rethink the whole problem. We proposed that the nitrogen atom of the sultams provides a possible handle for the linkage to a resin (see Scheme 2.1 for a retrosynthetic plan). We were fortunate to find that the Rink amide resin,¹³ which is a commercially available (NovaBiochem) modified Merrifield resin with Fmoc-protected amine groups, should well serve our needs. Our immediate goal was to further explore some new reactions of these sulfonamide resins and design a short, solid-phase synthetic route to the sultam library. One of the synthetic challenges in our original retrosynthetic analysis was the regiospecific deprotonation by butyllithium¹⁴ at the sulfonamide benzene ring rather than other benzene rings (i.e., the conversions of **36** → **38** and **37** → **41**). Another challenge was to succeed in the sultam ring closure and cleavage from the resin in a one-step process (i.e., the conversion of **41** → **34**). These two steps, which are so crucial to the whole synthetic plan, mandates that we prepare the solution-phase resin analogue **29** to test them.

2.3. Significance



Scheme 2.1. Retrosynthetic analysis of sultam solid-phase synthesis.

The most distinctive advantage of using a solid-phase method is its efficiency. By eliminating purification steps (e.g., column chromatography) and by use of robotic instrumentation, this process can be used to produce a greater range of modified compounds than is possible by normal solution-phase methods.

2.4. Anti-HIV drug testing system

All anti-HIV potency measurement were performed at the National Cancer Institute following the published procedure.¹⁵

1. Candidate agent is dissolved in DMSO (unless otherwise instructed) then diluted 1:100 in cell culture medium before preparing serial half-log₁₀ dilutions. T4 lymphocytes (CEM cell line) are added, and after a brief interval HIV-1 is added, resulting in a 1:200 final dilution of the compound. Uninfected cells with the compound serve as a toxicity control, and infected and uninfected cells without the compound serve as basic controls.
2. Cultures are incubated at 37 °C in a 5% carbon dioxide atmosphere for 6 days.
3. The tetrazolium salt, XTT, is added to all wells, and cultures are incubated to allow formazan color development by viable cells.
4. Individual wells are analyzed spectrophotometrically to quantitate formazan production, and in addition are viewed microscopically for detection of viable cells, a confirmation of protective activity.
5. Drug-treated virus-infected cells are compared with drug-treated noninfected cells and with other appropriate controls (untreated infected and untreated

noninfected cells, drug-containing wells without cells, etc.) on the same plate.

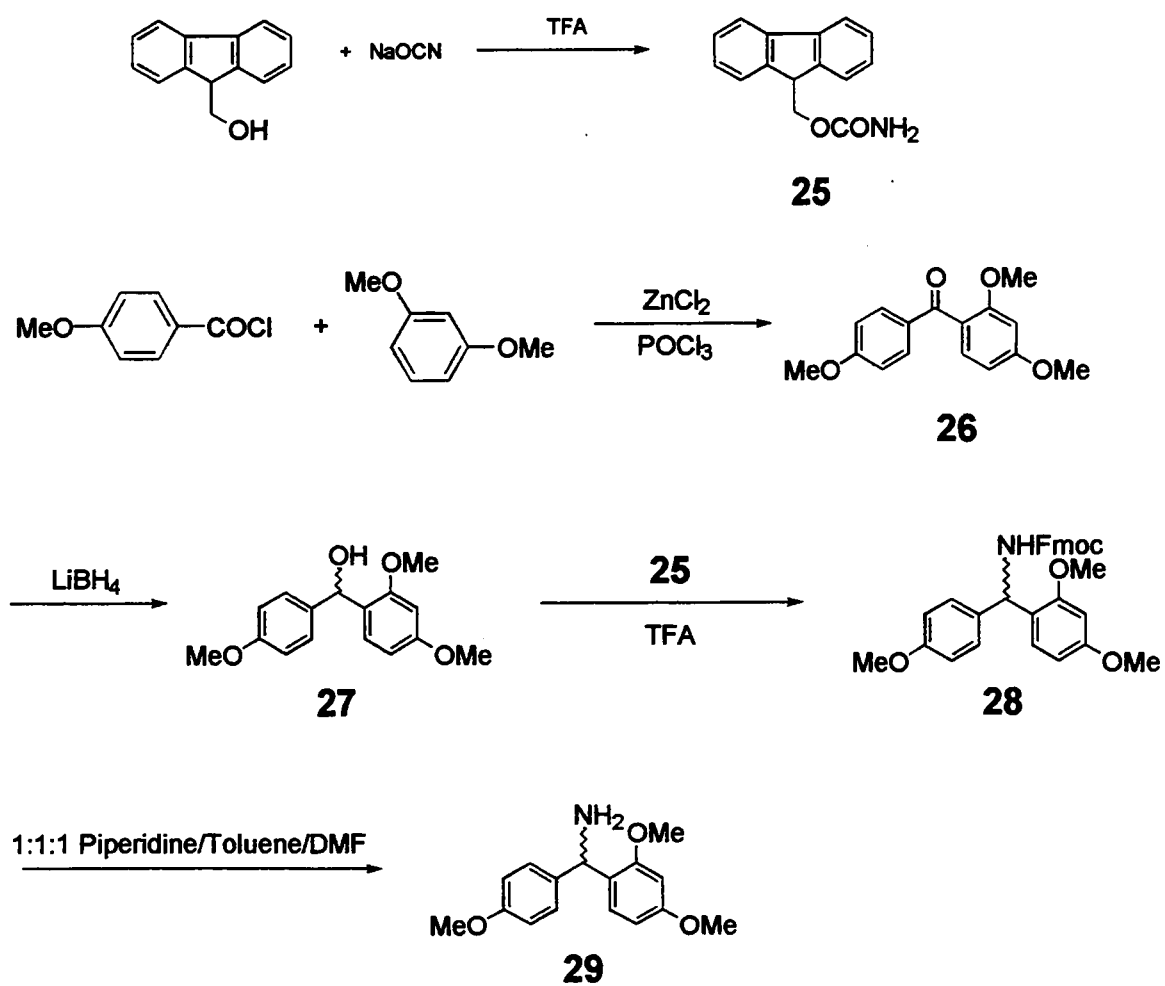
6. Data are reviewed in comparison with other tests done at the same time, and a determination about activity is made.

3. RESULTS AND DISCUSSION

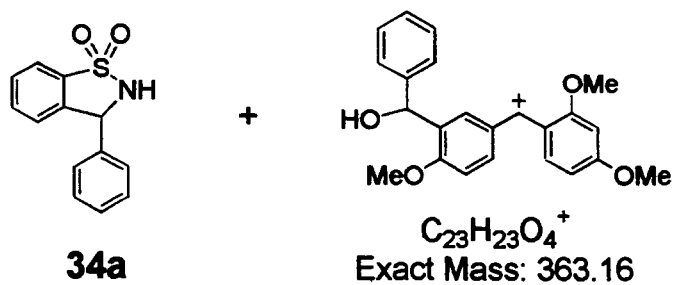
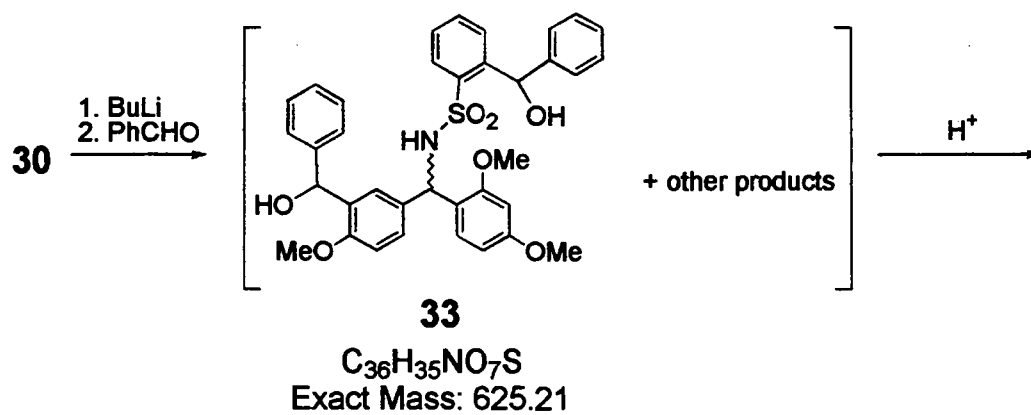
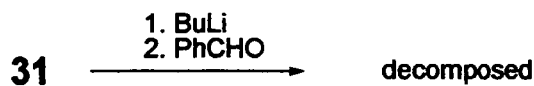
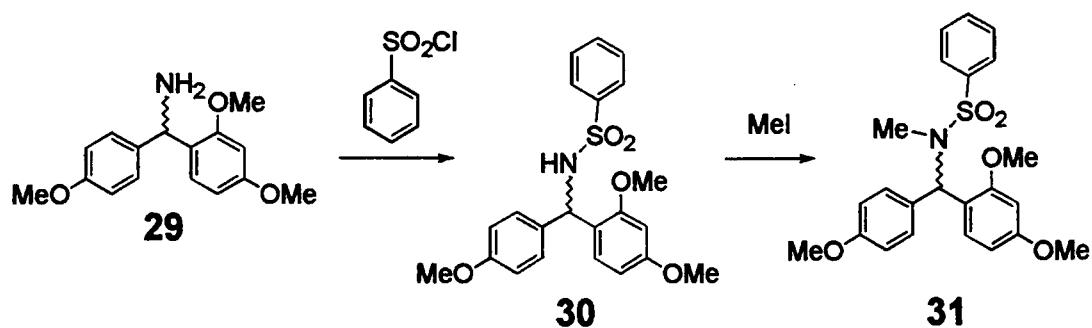
3.1. Development of solid-phase synthesis of sultams

The resin analogue **29** was prepared according to literature procedures (Scheme 2.2).¹⁶ Our initial trial was to first methylate the sulfonamide **30** to get the tertiary sulfonamide **31**, then deprotonate **31** and let the resulting anion react with benzaldehyde (Scheme 2.3). Unfortunately, the anion derived from **31** was found to be very unstable at 0 °C, resulting in rapid decomposition. The reaction sequence was then altered to deprotonate the secondary sulfonamide **30**, let the resulting anion react with benzaldehyde first, then with acid. As we carried out the reactions, the anion from **30** was found to be quite stable at room temperature, and its reaction with benzaldehyde went smoothly. But this also brought in a new problem: since an excess amount of benzaldehyde was used, the bis-adduct **33** and other unidentified products were obtained. Nonetheless, under acidic conditions, we demonstrated that the mixture can be converted to sultam **34a** (i.e., the NH sultam) directly in one step. This reaction validated our original proposal that cleavage and cyclization could be achieved in a one-step process.

Molecular ion peaks of **33** were detected as $M + Na^+$ (648) and $M + K^+$ (664) by ESIMS. The MS-MS fragmentation pattern (Figure 2.1) confirmed that the



Scheme 2.2. Synthesis of the solution-phase Rink amide resin analogue **29**.

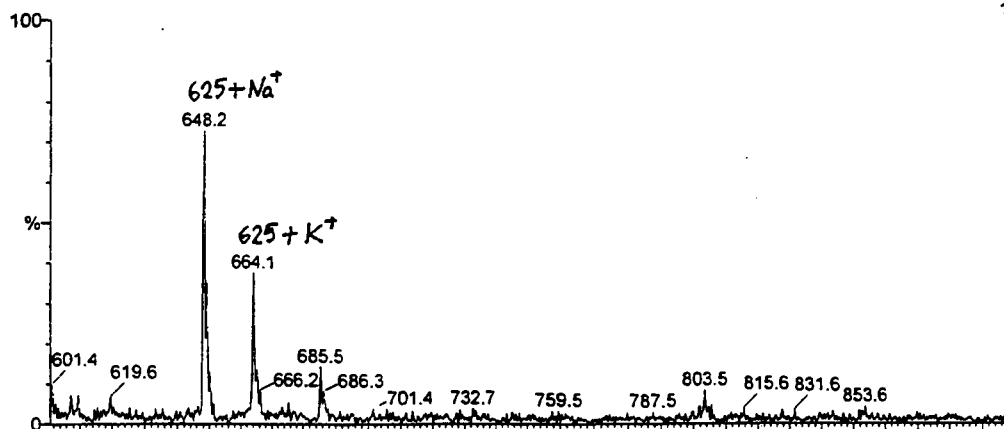


Scheme 2.3. Solution-phase trial of the resin analogue **29**.

Jiang
M Background for Bin Jiang
A663 1 (1.012) Sb (5.1.00); Sm (Mn, 2x0.70)

"Sin=" @ 5uL/min
cV 35

Scan ES+
1.03e6



530
MSMS 648 BJ sample
A663E 1 (1.022) Sb (5.1.00); Sm (Mn, 2x0.70)

"Sin="0808-1 @
CV = 150 ce = 25

Daughters of 648ES+
5.59e5

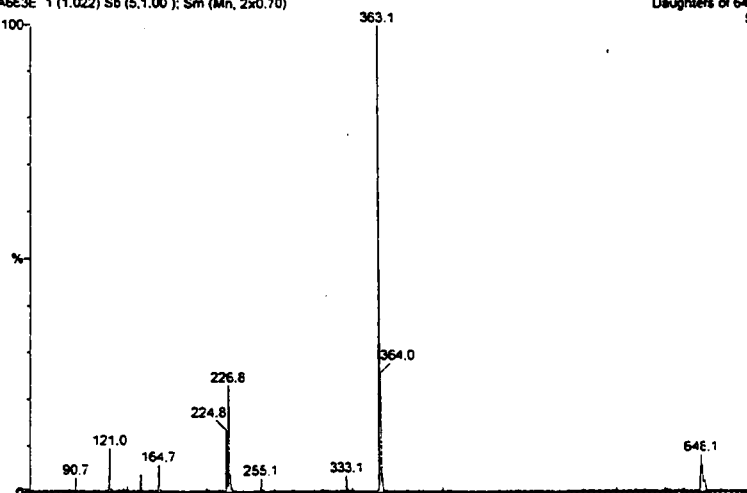
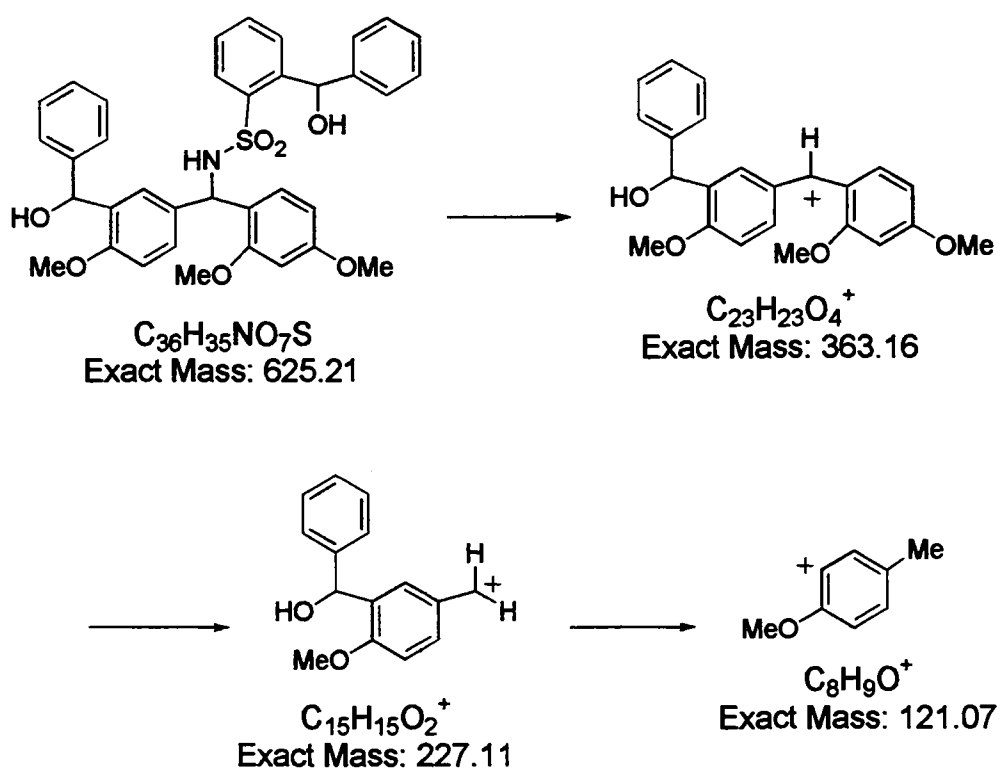


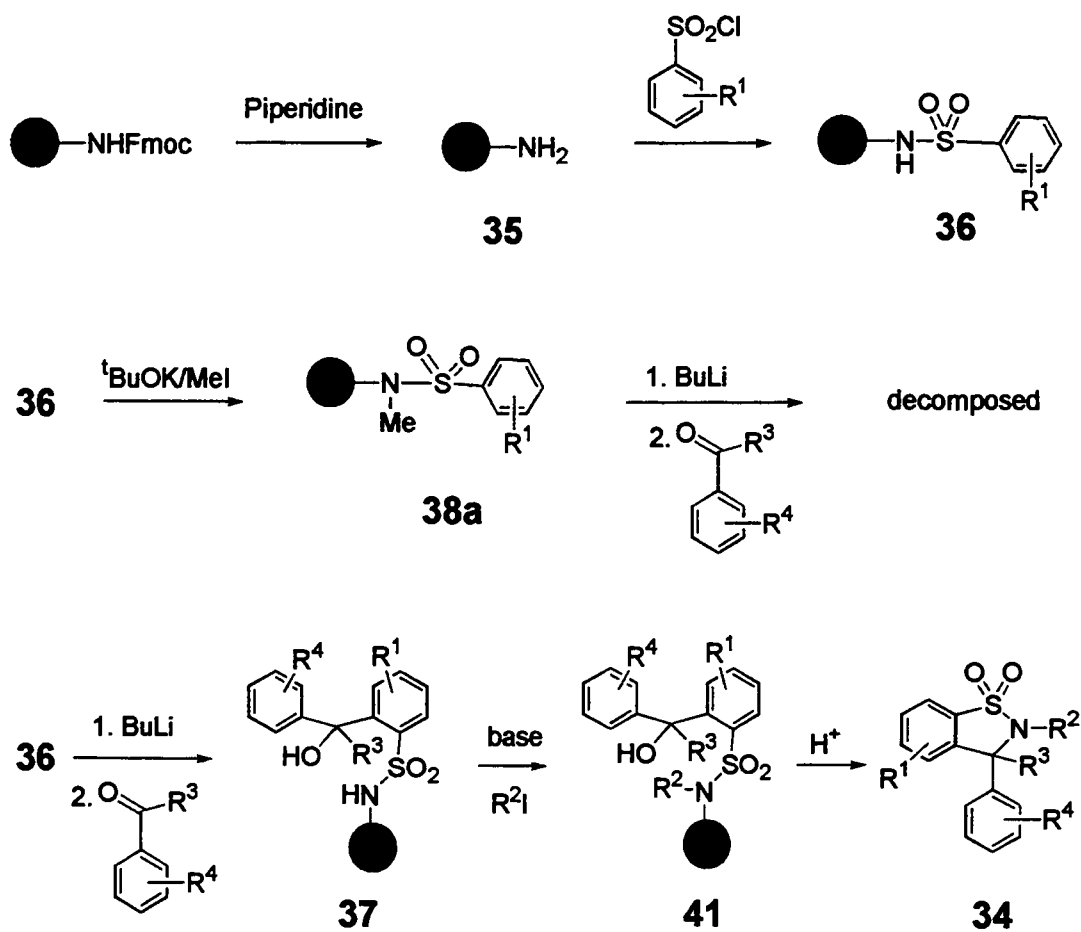
Figure 2.1. ESI-MS (top) and MS-MS (bottom) spectra of **33**.

second benzaldehyde unit is added on the mono-methoxy substituted benzene ring. The assignment was summarized in Scheme 2.4. Compound **33** is a result of Directed *ortho* Metalation (DoM) chemistry, which was mostly studied by Snieckus¹⁷ in solution-phase. Because both methoxy and sulfonamide are DoM groups, the lithiation occurs at both *ortho* positions of these two groups, hence the bis-adduct was obtained. Boehm and Showalter¹⁸ and Begtrup and co-workers¹⁹ have reported the directed *ortho*-lithiation on solid-phase. Based on our solution-phase trial, we believe that the same chemistry also occurs in our solid-phase reaction.

The advantages of using this Rink amide resin soon became apparent. These are listed as follows: (1) There is ample precedent to support the conversion of **35** → **36** → **38**. (2) The conjugate should ideally be stable under basic conditions, while cleavage only occurs under mildly acidic conditions (e.g., with 4:1 CH₂Cl₂-TFA). This makes it possible to carry out reactions with butyllithium (**36** → **37**) and *tert*-butoxide (**37** → **41**) without cleavage. (3) The resin is commercially available and widely used by peptide chemists, so it is very likely to facilitate automation of the whole process and allow ready construction of a combinatorial library. Conceivably, the automation could be carried out with a peptide synthesizer or in a multiple shake-flask reactor. Thus the Fmoc-protected amino resin was deprotected to give **35** that was subsequently used to prepare a secondary sulfonamide resin conjugate **36** (Scheme 2.5). It was reacted with butyllithium, followed by benzaldehyde, to give presumably **37**. It was then methylated to give **41**, which was easily cleaved and cyclized to sultam **34** under acidic conditions.



Scheme 2.4. MS-MS fragmentation pattern of **33**.



Scheme 2.5. Solid-phase synthesis of sultams **34**.

With a strong acid like H_2SO_4 or with 4:1 CH_2Cl_2 -TFA, this reaction was found to destroy the Rink amide resin, presumably giving some highly polar biphenyl products. On TLC (silica gel, 2:1 hexanes-ethyl acetate), all these impurities were showed to remain on the baseline. Our experiments also showed that most of these biphenyl products could be washed out by water. For example, **34y** is obtained in a relatively pure state without column chromatography (Figure 2.2).

To date several sultam molecules have been successfully prepared using this efficient, five-step, solid-phase method. At first, GC was used to monitor completion of each reaction by cleaving the product from the resin. Recently, NMR techniques²⁰ have been developed to allow direct determination of both ^1H and ^{13}C spectra of reactants immobilized on resins. In a set of examples submitted to the Bruker Applications Laboratory, we were able to observe conversions of the first two steps in the sequence by NMR spectroscopy of the intact polymer conjugates (Figure 2.3). The resonances that showed the most pronounced changes were in the region of δ 3.0–4.3. Trace A showed the Fmoc derivative of the commercial Rink amide resin. Its removal was showed in trace B. Conversion to the benzenesulfonamide **36** led to separation of the MeO signals (which were overlapped in traces A and B), giving a clean indication of formation of a new product. Both 1D and 2D (data not showed) experiments gave outstanding results on 10–20 mg samples swelled in CDCl_3 or $\text{DMF}-d_7$.

One may legitimately ask, “How versatile is this solid-phase process?” Studies to date indicate that almost any Ring A and Ring C variation is possible,

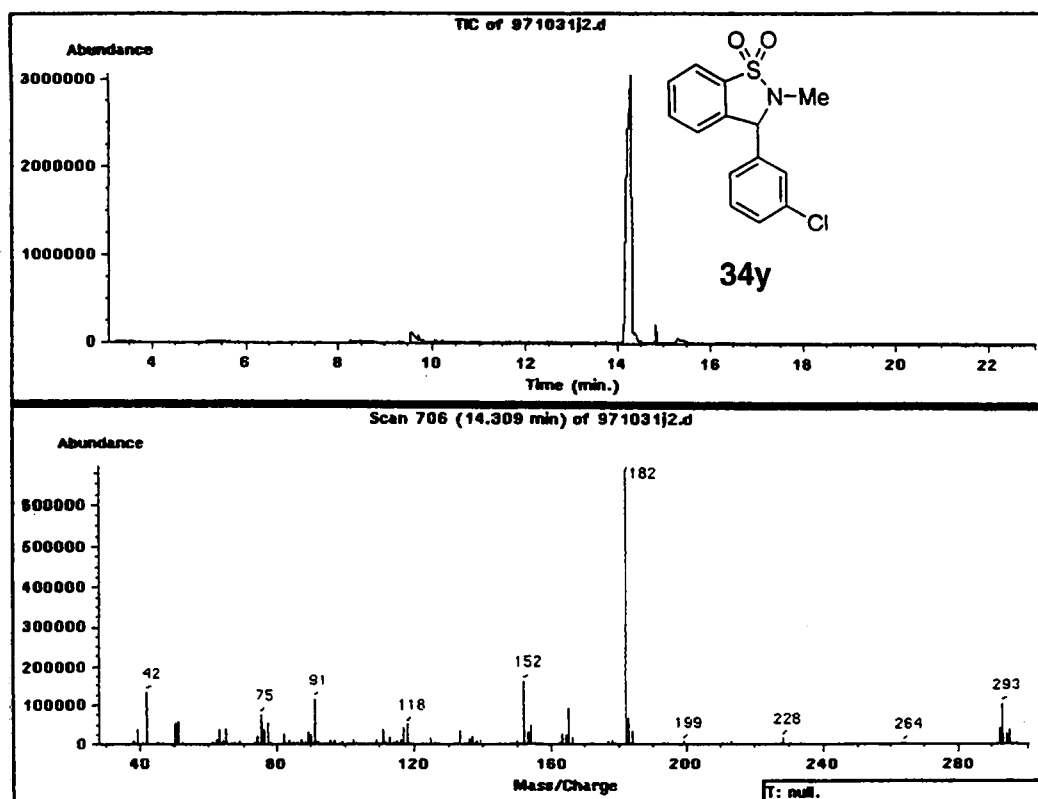


Figure 2.2. GC-MS spectra of crude sultam **34y** after water wash.

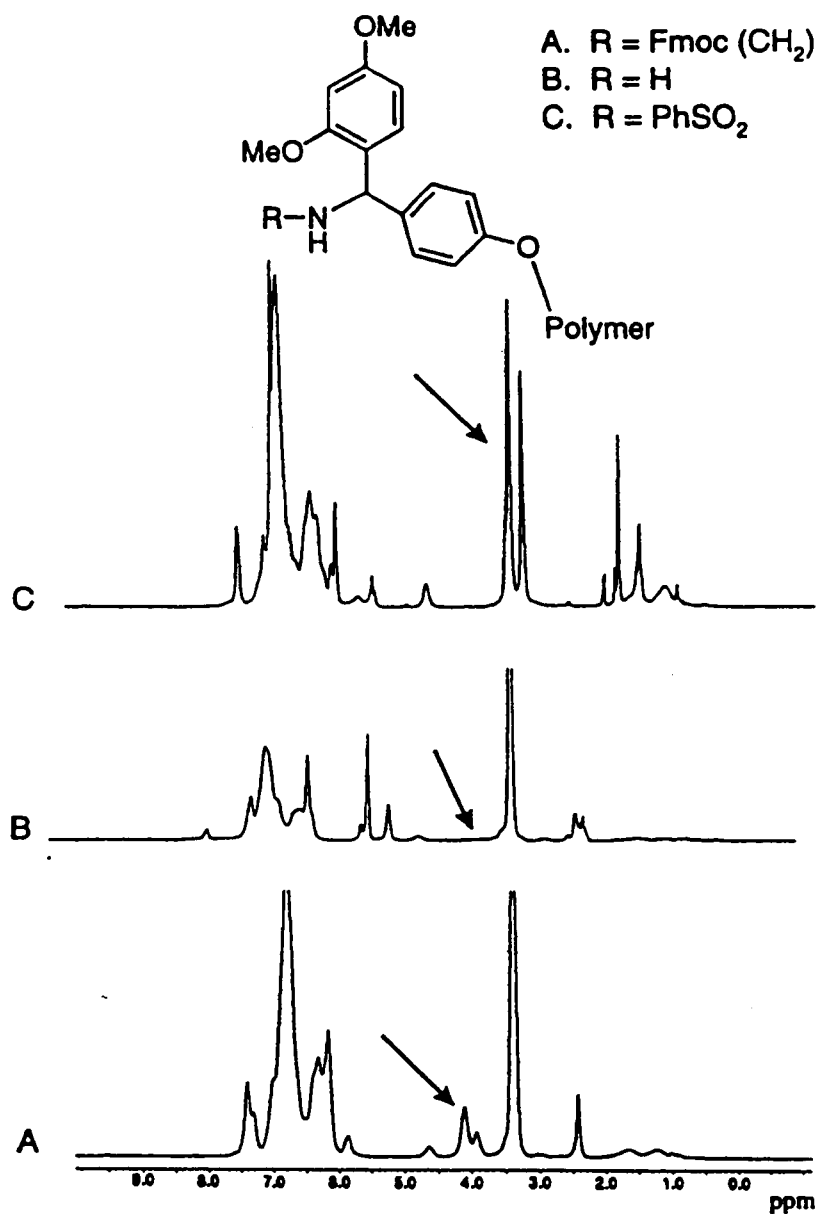
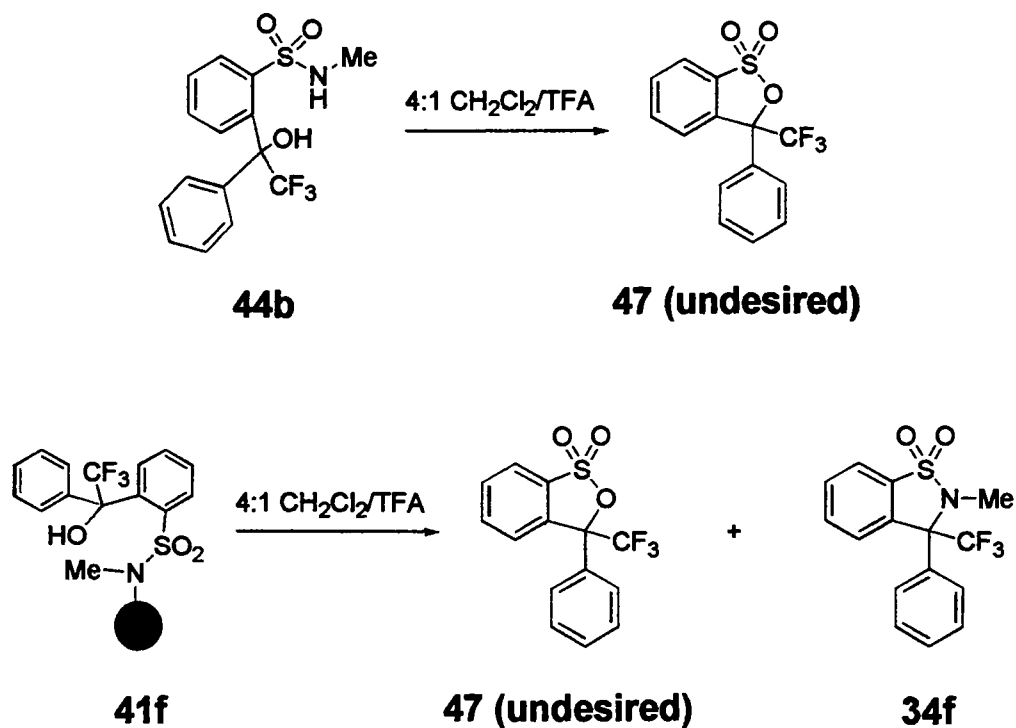


Figure 2.3. Solid-state 1H NMR (500 MHz) spectra for (A) the Fmoc-protected Rink amide resin, (B) the deprotected, free- NH_2 resin **35**, and (C) the $NH-SO_2Ar$ derivative **36**. Arrows denote sites that changed in the spectrum. The author would like to thank Dr. Kim Colson at the Bruker Applications Laboratory for recording the spectra.

provided that the aryl substituent is compatible with organolithium reagents used to generate the intermediate anion. Aldehydes and ketones both react, the latter a bit more slowly in the step of addition to the anion. As for N-substitution, problems have arisen with longer or branched-chain alkyls, as the elimination reaction ($X-CH_2CH_2CH_2-R \rightarrow H_2C=CHCH_2-R$) invariably predominates, giving little or none of the *N*-alkyl sultam. We have no solution for the elimination problem, which may be enhanced by the steric hindrance of the nitrogen attached to the polymer. However, it may be unnecessary to make variation on R^2 since anti-HIV assays showed that sultams with longer or branched alkyl chains on nitrogen all have diminished activity against HIV (see 3.2).

It is worthwhile to mention that when the synthesis of compound **34f** from **44b** was attempted by the conventional solution-phase method, the exclusive product was the sultone **47** (Scheme 2.6), which was formed via a competing reaction that favors N-protonation and nucleophilic attack by the OH group, due to the powerfully electron-withdrawing effect of the α -CF₃ group. When the solid-phase method was used, both **47** and **34f** were obtained as a 1:1 mixture, as determined by GC-MS (Figure 2.4). This result demonstrates that on the resin intermediate **41f**, the stereoelectronic environment is altered so that both the sulfonamide nitrogen and the hydroxyl oxygen can be protonated, which allows both nucleophilic reaction pathways to occur to give both products. Playing the steric hindrance on nitrogen to our advantage, we have managed to prepare the sultam **34f** exclusively by the solution-phase method (see 3.3).



Scheme 2.6. Solution-phase reaction vs solid-phase reaction: synthesis of the trifluoromethylated sultam **34f**.

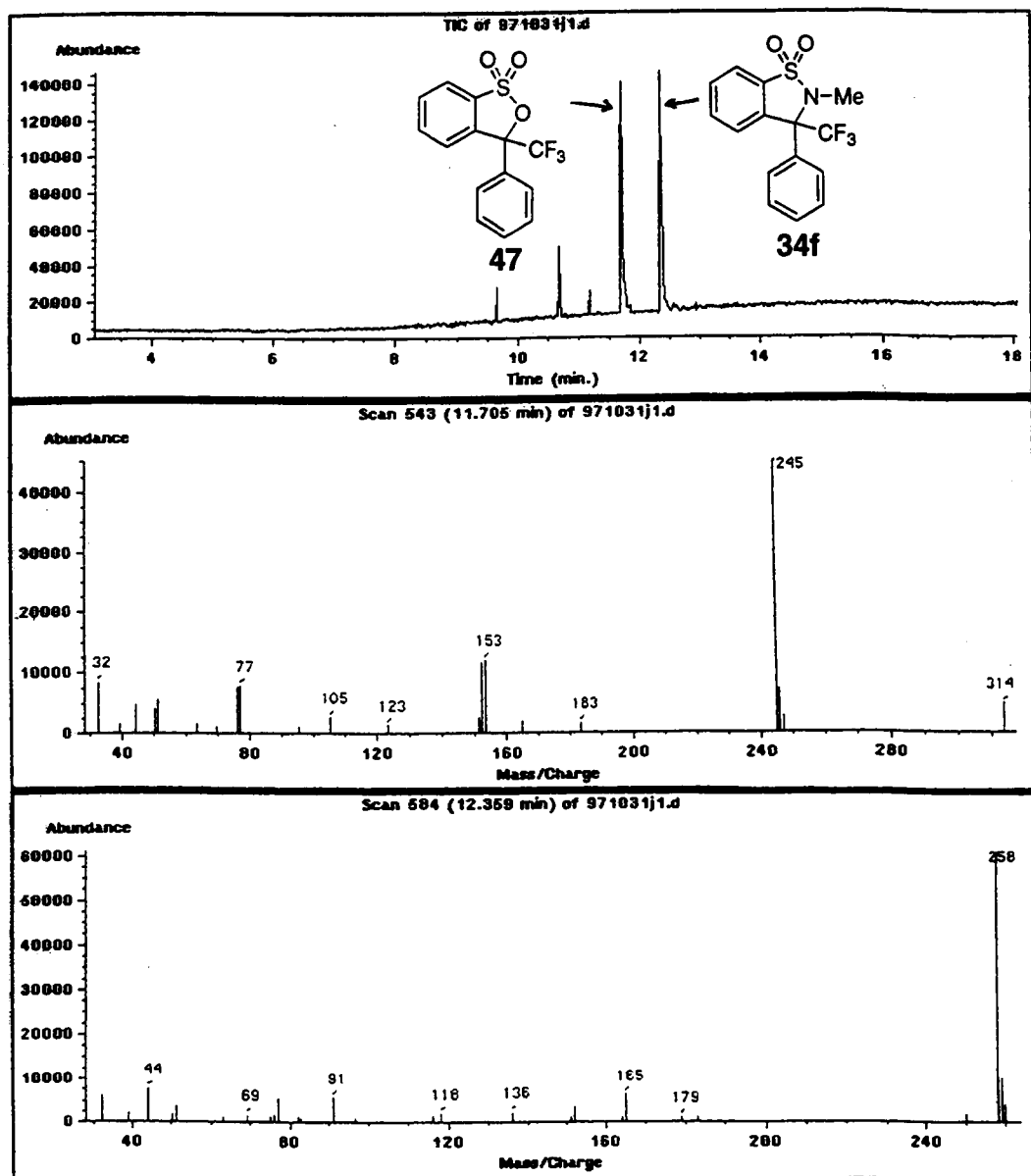


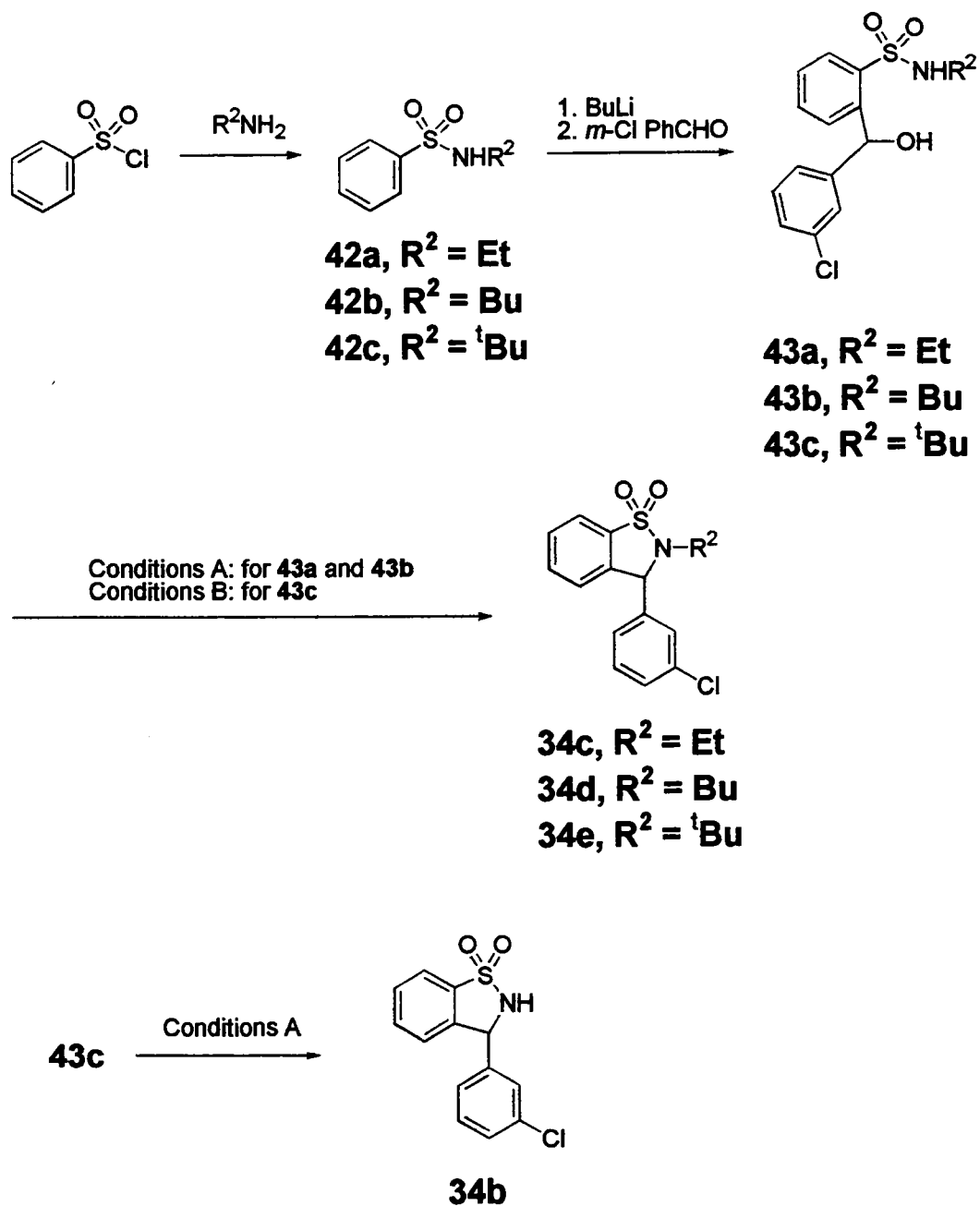
Figure 2.4. GC-MS spectra of solid-phase synthesis products **47** and **34f**.

3.2. Solution-phase synthesis of *N*-alkyl sultams and their structure-activity relationship

In the process of developing solid-phase approach to sultams, other members of Baker's group prepared sultams with different R^1 and R^4 groups by the conventional solution-phase method. Anti-HIV screening results from these sultams indicated that ring C modifications are most promising, as the *m*-Cl analogue (**34y**, NSC-698619, $R^1 = R^3 = H$, $R^2 = Me$, $R^4 = 3-Cl$) showed ten-fold enhanced activity ($EC_{50} = 1.19 \times 10^{-7} M$). It was therefore proposed that the ring C is the major binding region of the reverse-transcriptase (RT). We chose *m*-Cl sultams **34b–e** as our target molecules to study how ring B interacts with the RT enzyme (Scheme 2.7). Since the solid-phase method can not provide *N*-alkyl sultams except *N*-methyl one, we decided to make them by the conventional solution-phase method.¹⁰ Their anti-HIV activities are summarized in Table 2.2. The most interesting result is the *N*-ethyl analogue **34c**, which is about as active as the *N*-methyl compound **34y**. When the side chain becomes longer than two carbons or more bulky, the activity diminishes as showed by compounds **34d** and **34e**. Also interesting is the complete inactivity of the NH analog **34b**. These results may indicate that this portion of the binding pocket is lipophilic and very limited in space.

3.3. Solution-phase synthesis of the trifluoromethylated sultam (**34f**)

Fluorine has profound effects on the activity of many drugs. It would be interesting to see how fluorinated sultams behave in the testing. One such sultam

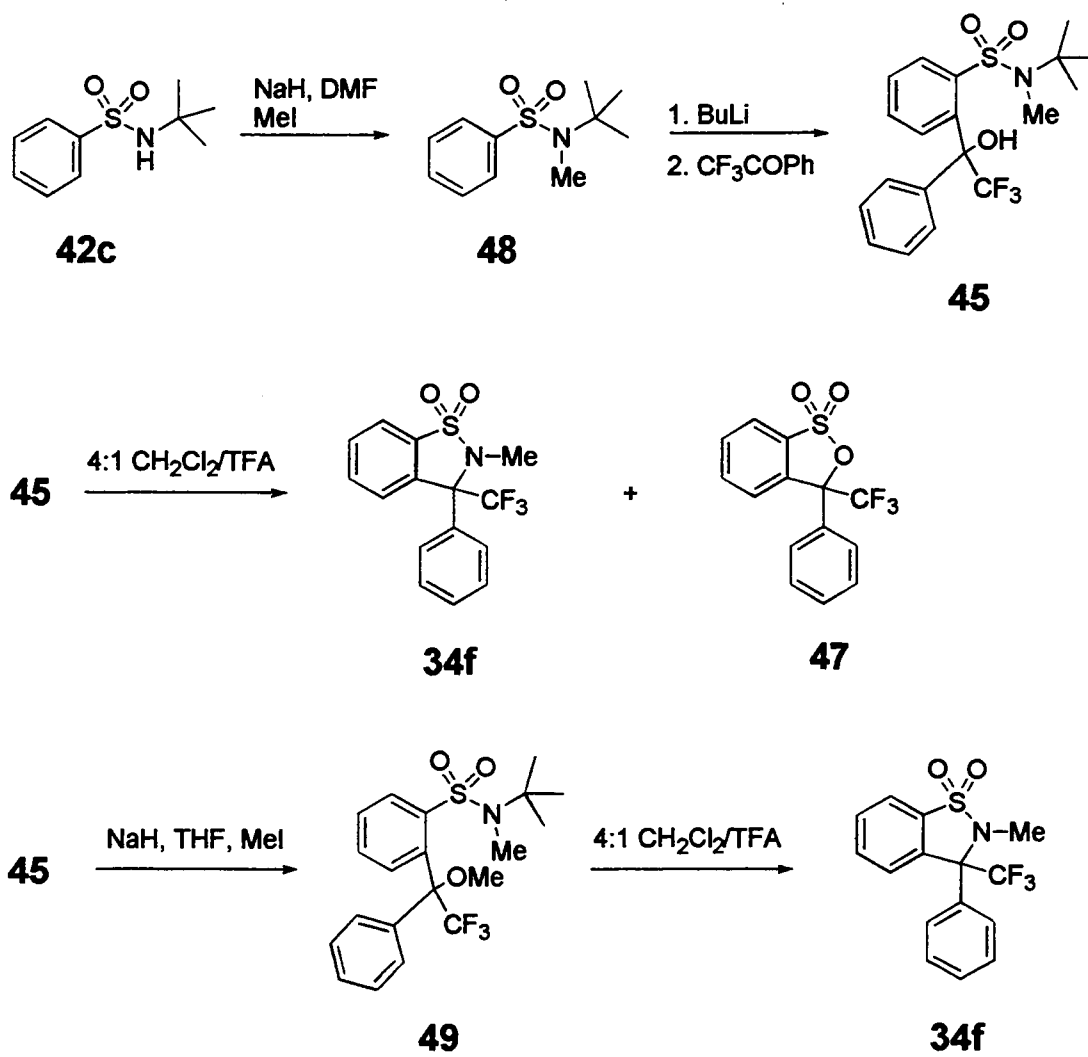


Scheme 2.7. Synthesis of four sultams with different R^2 substitutions.
 Conditions A: concd H_2SO_4 . Conditions B: 4:1 $\text{CH}_2\text{Cl}_2/\text{TFA}$.

Table 2.2. *N*-alkyl sultams and their activity

Compound	NSC#	Analogue	Mean EC ₅₀ (M)	Category
34b	702147	NH	-----	inactive
34c	702145	<i>N</i> -Et	2.29×10 ⁻⁷	active
34d	702144	<i>N</i> -Bu	1.97×10 ⁻⁶	moderate
34e	702146	<i>N</i> -tert-Bu	-----	inactive
34y	698619	<i>N</i> -Me	1.19×10 ⁻⁷	active

is the trifluoromethylated sultam **34f**. As mentioned previously, applying the solution-phase method to synthesize sultam **34f** from **44b** was not successful. Instead, sultone **47** was obtained as the exclusive product. One hypothesis to explain this result is that the trifluoromethyl group destabilized the benzyl cation intermediate and thus altered the reaction pathway from O-protonation to N-protonation to give the O-cyclization product. Interestingly, when the solid-phase method was used, both **47** and **34f** were obtained as a 1:1 mixture (Figure 2.4). This prompted us to make **45**, which is a solution-phase analog of resin **41f**. In fact, **45** gave 1.3:1 mixture (isolated) of **34f** and **47** under acidic conditions. It seems like a bulky substitution on the tertiary sulfonamide nitrogen favors N-cyclization. However, since O-protonation was still not favorable, sultone **47** was still produced (Scheme 2.8). Surprisingly, further methylating the hydroxy group in **45** gave **49**, which in fact gave exclusively N-cyclization product **34f** in 85% isolated yield.



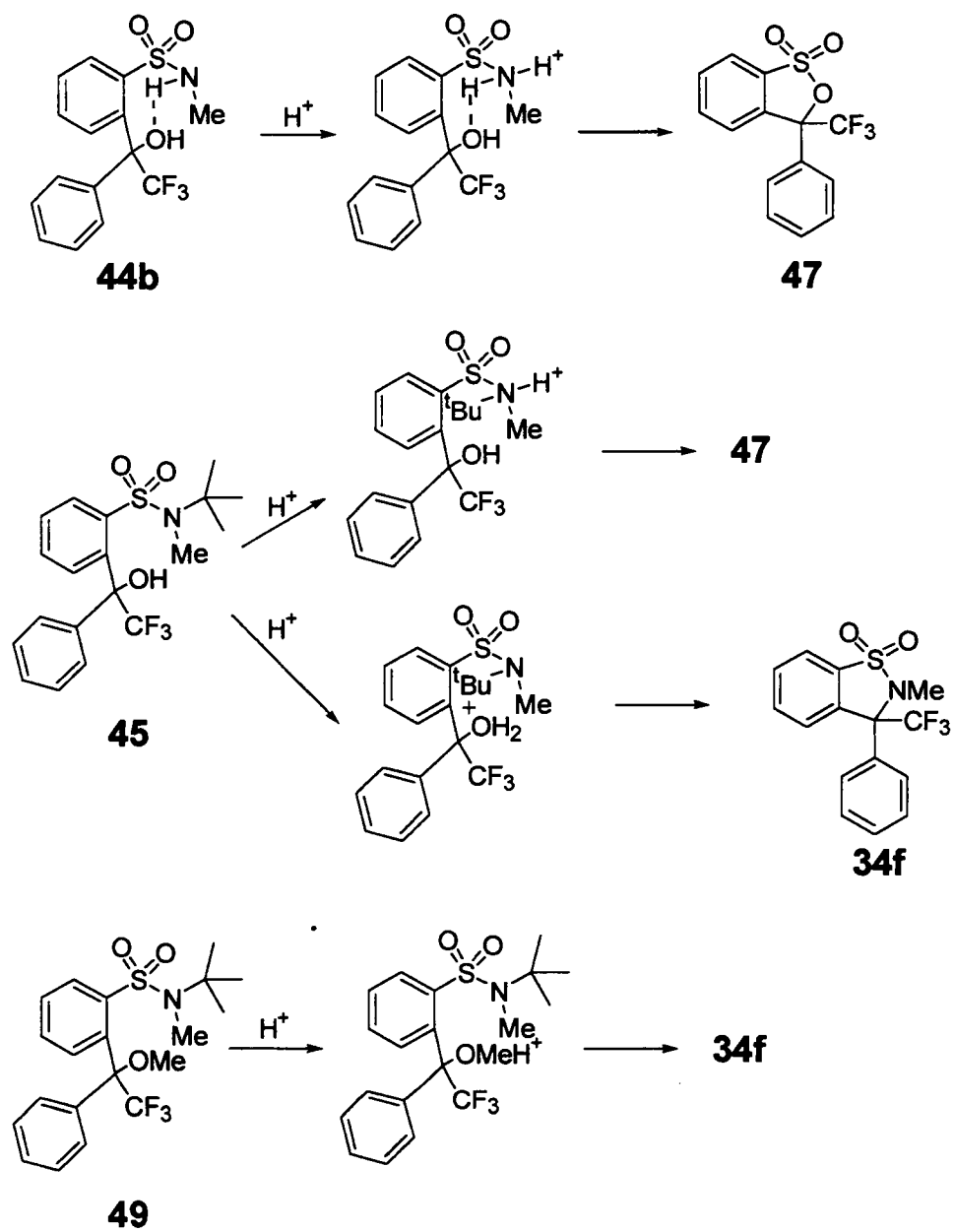
Scheme 2.8. Improved synthesis of **34f** by varying substrate structure.

The proposed mechanism (Scheme 2.9) for the formation of the O-cyclization product **47** is the predominance of the N-protonation process. There are two major factors that reduce the basicity of the hydroxyl oxygen atom in compound **44b** so that N-protonation predominate. One is the electron-withdrawing inductive effect of the trifluoromethyl group, and the other is the intramolecular hydrogen bond between the hydroxyl oxygen and the sulfonamide hydrogen. When the hydrogen was replaced by the *tert*-butyl group or a resin linker arm, the hydrogen bonding influence would be eliminated, as in compound **45** and resin **41f**. This would make two protonation processes competitive so that both cyclization products were formed. Methylating the hydroxyl oxygen, as in compound **49**, would mitigate the electron-withdrawing effect of the trifluoromethyl group so that the N-cyclization product was formed exclusively.

Sultam **34f** (NSC-702733) was submitted for testing and it was found to be active ($EC_{50} = 7.71 \times 10^{-7}$ M). Compared with the data from the methyl analogue (NSC-108408, $EC_{50} = 2.61 \times 10^{-6}$ – 6.44×10^{-6} M) made by our colleague, switching from CH_3 to CF_3 increases the potency by about ten-fold. Currently, my colleagues are using a similar approach to synthesize other sultams with a CF_3 group at the chiral carbon atom.

4. CONCLUSIONS

With respect to the objectives set forth in section 2.1., an efficient solid-phase synthesis of sultams with variations on rings A and C was developed. As for



Scheme 2.9. Proposed mechanism for the substrate-dependent product distribution between **47** and **34f**.

the variations on ring B, conventional solution-phase method were applied to synthesize *N*-alkyl sultams. In addition, a novel preparation of 3-CF₃ sultams was developed. EC₅₀ results show that a short alkyl chain (methyl or ethyl) is essential for maintaining anti-HIV activity. Also, a CF₃ substitution at the chiral carbon results in ten-fold increase of the potency. All of these provide a solid background for the further development of sultams as possible inexpensive drugs for HIV treatment.

5. EXPERIMENTAL

5.1. General Methods

All solution-phase reactions were monitored by thin-layer chromatography (TLC). Adsorption chromatography was carried out using E. Merck silica gel products: (a) TLC on 0.2 mm aluminum-backed plates, (b) flash chromatography using 230–400 mesh silica gel. The TLC plates were visualized under 254 nm UV light. All reagents were purchased from Aldrich Chemical Co. and used without further purification. All solvents were at least 99% pure and used without drying unless noted otherwise. Elemental analyses were furnished by Atlantic Microlab, Inc. of Atlanta, GA. ¹H and ¹³C NMR spectra were obtained by using a Bruker AC 250 spectrometer (250 MHz and 62.5 MHz, respectively) with CDCl₃ as the solvent and TMS as the internal standard. ¹H NMR chemical shifts are reported as (ppm) downfield from the TMS signal; multiplicities are first-order values in Hz: s, singlet; bs, broad singlet; d, doublet; t, triplet; pt, pseudo triplet; q, quartet, dd, doublet of doublet; m, multiplet. ¹³C NMR chemical shifts are reported as (ppm) relative to

CDCl_3 (77.00 ppm) as standard. All solid-phase reactions were monitored by the GC–MS method as described in the following: the resin intermediate (~5 mg) was mixed with 4:1 (v/v) CH_2Cl_2 –TFA solution for 1 h, and the resulting red solution was then filtered and concentrated. A wet-needle technique was used to inject the sample onto GC–MS. GC–MS was performed on a Hewlett–Packard 5890 chromatograph equipped with a HP 5970 series mass-selective detector. The GC injector temperature and detection temperature were kept at 250 °C, while the column temperature was ramped from 60 °C to 260 °C at the rate of 20 °C/min. A non-destructive solid-state NMR method was used to monitor the solid-phase reactions. Solid-phase ^1H and ^{13}C NMR spectra were obtained on a Bruker DRX 500 spectrometer (500.13 and 125.76 MHz, respectively), equipped with a 4-mm Bruker CCA HR-MAS probe. About 20 mg resin sample was loaded onto a ceramic rotor, suspended in a suitable deuterium solvent, and spun at the magic angle at 5 kHz. ^1H NMR spectra were obtained with a Carr–Purcell–Meiboom–Gill pulse sequence.

5.2. General procedure for solid-phase preparation of sultam

Loading of sulfonyl chlorides on the Rink resin was performed according to ref 13.

(±)-2-Methyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34g). Fmoc-protected Rink amide resin (Novabiochem; 0.45 g with a loading capacity of 0.47 mmol/g, 0.21 mmol) was treated with a mixture of piperidine

(5 mL), toluene (5 mL) and DMF (5 mL). After 1.5 h shaking on a mechanical shaker (Burrell "Wrist Action" model 15), the resin was washed with DMF (2 × 10 mL) and CH₂Cl₂ (2 × 10 mL). To the deprotected resin **35** was added pyridine (6 mL), CH₂Cl₂ (20 mL) and *p*-toluenesulfonyl chloride (1.15 g, 6.03 mmol). The reaction flask was shaken at room temperature for 4.5 h. The resin was then filtered and washed with CH₃OH (1 × 10 mL), DMF (2 × 10 mL) and THF (2 × 10 mL). The resin was dried in vacuum (0.1 torr) for 1 h at room temperature. To the dried sulfonyl group-loaded resin **36g** was added dry THF (20 mL, freshly distilled from Na–benzophenone ketyl), and the flask was cooled by liquid N₂ for about 20 s with hand shaking. To this was then added BuLi (1 mL of a 10.0 M solution in hexanes, 10 mmol) dropwise with hand shaking. After the reaction had completed, the resin color changed from pale yellow to brownish black. *m*-Chlorobenzaldehyde (1.2 mL, 10.6 mmol) was then added dropwise until the resin color turned back to pale yellow, while the solution color turned to brownish red. The flask was shaken for additional 10 min, then MeOH was added. After the resin had settled, the brown solution was removed with a pipet, and the resin was washed with MeOH (2 × 10 mL) and THF (2 × 10 mL). Again the pale-yellow resin was dried in vacuum (0.1 torr) for 1 h at room temperature. To the resin (assuming **37g**) was added THF (15 mL) and potassium *tert*-butoxide in THF (2 mL of a 1 M solution, 2 mmol) with hand shaking. After the resin color had turned to deep brown, iodomethane (2 mL, 32 mmol) was added, and the flask was shaken for 20 h. MeOH was then added to dissolve the precipitate. The resin was washed with MeOH (2 × 10 mL) and CH₂Cl₂

(2 × 10 mL) and then dried in vacuum for 1 h at room temperature to give a yellow resin. To a portion of this yellow, methylated resin **41g** (0.0424 g out of the 0.4562 g) was added 1:4 TFA-CH₂Cl₂ (4 mL), whereby the resin color instantly turned to blood red. After 19 h shaking, MeOH was added and the red resin was filtered off. The filtrate was concentrated to give a dark-brown syrup (5.3 mg, 88% overall yield) whose structure was demonstrated to be **34g** by GC-MS.

5.3. Solution-phase preparation of compound 25–49

Preparation of **9-fluorenylmethylcarbamate (25)**.²¹ This compound was prepared by my co-worker Michael S. Gaines according to the literature procedure. The NMR data were in agreement with those in the literature. ¹H NMR (DMSO-*d*₆): δ 4.24 (m, 3H, Ar₂CHCH₂O), 6.55 (bs, 2H, NH₂), 7.31–7.40 (m, 4H), 7.68 (d, *J* = 7.3 Hz, 2H), 7.88 (d, *J* = 7.4 Hz, 2H).

Preparation of **2,4-dimethoxy-4'-methoxybenzophenone (26)**.²² *p*-Methoxybenzoyl chloride (5.4 mL, 40 mmol), 1,3-dimethoxybenzene (6.8 mL, 1.3 equiv) and fused zinc chloride (12 g, 2.2 equiv) were dissolved in phosphorus oxychloride (28 mL). The resulting deep-red solution was heated in a 55 °C oil bath for 2 h. The solution was then slowly poured onto crushed ice, extracted with CH₂Cl₂, and the organic extract was dried over MgSO₄. After solvent evaporation, the crude amber syrup was purified by column chromatography (6:1 hexanes–ethyl acetate) to give **26** as a white solid (10.1 g, 93%). ¹H NMR: δ 3.72 (s, 3H, OMe),

3.87 (s, 6H, 2OMe), 6.52–6.56 (m, 2H), 6.90 (d, $J = 8.6$ Hz, 1H), 7.34 (d, $J = 8.1$ Hz, 1H), 7.78 (d, $J = 8.7$ Hz, 1H); ^{13}C NMR: δ 55.43 (OMe), 55.50 (OMe), 55.62 (OMe), 98.86, 104.47, 113.30, 122.09, 131.49, 131.56, 132.16, 159.15, 162.88, 163.18, 194.10.

Preparation of (2,4-dimethoxyphenyl)-(4-methoxyphenyl)methanol (27).¹⁶

To a dry THF (100 mL) solution of **26** (10.1 g, 37.1 mmol) was added LiBH_4 (1 g, 46.0 mmol) portionwise at room temperature. This solution was heated in a 70 °C oil bath for 3 h. An ice bath was used to cool the solution before brine was added dropwise to quench the reaction. The solution was extracted with ethyl acetate, and the organic extract was dried over MgSO_4 . After solvent evaporation, **27** was obtained as a white solid (10.2 g, 100%). ^1H NMR: δ 2.86 (d, $J = 5.1$ Hz, 1H, OH), 3.79 (s, 9H, 3OMe), 5.96 (d, $J = 4.7$ Hz, 1H), 6.42–6.46 (m, 2H), 6.86 (d, $J = 8.8$ Hz, 2H), 7.08 (d, $J = 8.1$ Hz, 1H), 7.28 (d, $J = 8.6$ Hz, 2H). ^{13}C NMR: δ 55.23 (OMe), 55.35 (OMe), 55.41 (OMe), 71.42, 98.83, 104.11, 113.51, 124.95, 127.73, 128.42, 135.71, 145.14, 157.79, 160.30.

Preparation of *N*-(2,4-dimethoxyphenyl-4'-methoxyphenyl)methyl-9-fluorenylmethyl carbamate (28).¹⁶ This compound was prepared by my co-worker Michael S. Gaines according to the literature procedure. ^1H NMR: δ 3.73 (s, 3H, OMe), 3.77 (s, 3H, OMe), 3.80 (s, 3H, OMe), 4.23 (m, 1H, CH), 4.41 (m, 2H), 5.80–5.91 (m, 1H), 6.04 (d, $J = 8.4$ Hz, 1H), 6.44–6.48 (m, 2H), 6.81 (d, $J = 8.5$ Hz,

2H), 7.12 (d, $J = 7.7$ Hz, 2H), 7.29–7.39 (m, 5H), 7.59 (d, $J = 6.6$ Hz, 2H), 7.75 (d, $J = 6.8$ Hz, 2H).

(2,4-Dimethoxyphenyl)-(4-methoxyphenyl)methylamine (29).¹⁶ This compound was prepared by my co-worker Michael S. Gaines according to the literature procedure. ^1H NMR: δ 1.97 (bs, 2H, NH_2), 3.77 (s, 9H, 3OMe), 5.38 (s, 1H), 6.40–6.43 (m, 2H), 6.83 (d, $J = 8.7$ Hz, 2H), 7.11 (d, $J = 9.1$ Hz, 1H), 7.28 (d, $J = 8.6$ Hz, 2H); ^{13}C NMR: δ 52.78, 55.14, 55.26, 98.65, 103.93, 113.42, 126.97, 127.91, 137.32, 157.63, 158.15, 159.66, 162.42.

***N*-[(2,4-Dimethoxyphenyl)-(4-methoxyphenyl)methyl]-benzenesulfonamide (30).** Compound **29** (0.77 g, 2.82 mmol), benzenesulfonyl chloride (0.36 mL, 2.82 mmol), and triethylamine (0.39 mL, 2.82 mmol) were dissolved in THF (20 mL) and stirred at room temperature. After 1 h, the solvent was evaporated, and the crude product was purified by flash column chromatography (2:1 hexanes–ethyl acetate) to give **30** as a white glassy solid (0.8 g, 69%). mp 102–103 °C; ^1H NMR: δ 7.63 (d, $J = 7.6$ Hz, 2H), 7.40–7.35 (m, 1H), 7.28–7.22 (m, 2H), 7.10 (d, $J = 8.5$ Hz, 2H), 6.87 (d, $J = 8.3$ Hz, 1H), 6.72 (d, $J = 8.5$ Hz, 2H), 6.26 (d, $J = 8.3$ Hz, 1H), 6.20 (s, 1H), 5.96 (d, $J = 8.9$ Hz, 1H), 5.61 (d, $J = 8.9$ Hz, 1H), 3.71 (s, 6H, 2OMe), 3.52 (s, 3H, OMe); ^{13}C NMR: δ 160.36, 158.45, 157.18, 140.50, 132.89, 131.80, 129.78, 128.28, 127.89, 126.62, 120.30, 113.36, 104.05, 98.80, 57.72 (Ar_2CH), 55.21 (OMe), 55.05 (OMe). Anal. Calcd for

C₂₂H₂₃NO₅S: C, 63.89; H, 5.62; N, 3.39. Found: C, 64.01; H, 5.64; N, 3.36.

***N*-[(2,4-Dimethoxyphenyl)-(4-methoxyphenyl)methyl]-*N*-methylbenzenesulfonamide (31).** To NaH (40.7 mg, 1.02 mmol, as a 60% mixture with mineral oil) was added a solution of **30** (0.154 g, 0.37 mmol) in THF (2 mL) at 0 °C. After 10 min, MeI (0.03 mL, 0.48 mmol) was added, and the reaction was allowed to progress at room temperature overnight. EtOAc (2 mL) was added to quench the reaction, followed by the addition of water (2 mL). After phase separation, the crude product was purified by flash column chromatography (2:1 hexanes–ethyl acetate) to give **31** as a pale-yellow solid (0.15 g, 94% yield). mp 149–150 °C ; ¹H NMR: δ 7.73 (d, *J* = 7.0 Hz, 2H), 7.53–7.38 (m, 3H), 6.99 (d, *J* = 9.4 Hz, 2H), 6.85–6.76 (m, 3H), 6.57 (s, 1H), 6.36–6.32 (m, 2H), 3.78 (s, 6H, 2OMe), 3.56 (s, 3H, OMe), 2.68 (s, 3H, NMe); ¹³C NMR: δ 160.51, 158.57, 158.19, 140.39, 131.86, 130.65, 129.03, 128.51, 127.71, 127.19, 119.94, 113.56, 103.80, 98.28, 58.30 (Ar₂CH), 55.30 (OMe), 55.21 (OMe), 55.11 (OMe), 32.05 (NMe). Anal. Calcd for C₂₃H₂₅NO₅S: C, 64.61; H, 5.91; N, 3.28. Found: C, 64.73; H, 6.00; N, 3.28.

(±)-3-Phenyl-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34a). To a THF solution of **30** (2 mL, 0.154 g, 0.37 mmol) at 0 °C was added BuLi (0.37 mL of a 2.5 M solution in hexanes, 0.93 mmol). The resulting solution was stirred at 0 °C for an additional 30 min, then benzaldehyde (0.06 mL, 0.59 mmol) was added

dropwise. The reaction was stirred at room temperature overnight. HCl (2 mL of a 5% aq solution) was then added dropwise with stirring, and the solution was extracted with CH₂Cl₂. After evaporation of the solvent, the crude product was purified by flash column chromatography (3:1 followed by 2:1 hexanes–ethyl acetate) to give **33** and other unidentified products as a white solid (0.128 g). To a portion of this white solid was added concd H₂SO₄ (2 mL), and the solution color instantly turned to deep red. The reaction was stirred at room temperature overnight. The red solution was then poured onto crushed ice and extracted with CH₂Cl₂. Both TLC (2:1 hexanes–ethyl acetate) and GC–MS showed that **34a** is the only product in the CH₂Cl₂ phase. The NMR data were in agreement with those from samples prepared by Dr. Mao. ¹H NMR: δ 5.73 (s, 1H, Ar₂CH), 7.13–7.16 (m, 1H), 7.38 (s, 5H), 7.54–7.57 (m, 2H), 7.82–7.86 (m, 1H); ¹³C NMR: δ 61.37, 121.12, 125.34, 127.58, 129.10, 129.25, 129.46, 133.26, 134.86, 136.71, 139.81; GC–MS: *t*_R = 13.77 min; *m/z* 245 (M), 168 (M - C₆H₅), 104 (base peak, 168 - SO₂), 77, 51.

(±)-3-(3-Chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34b).¹⁰ To **43c** (1.77 g, 5.01 mmol) was added concd H₂SO₄ (5 mL). The resulting dark brown solution was stirred at room temperature for 14 h. Crushed ice was then added to the solution, which was then extracted with CH₂Cl₂, and the organic extract was dried over MgSO₄. Evaporation of the solvent gave a crude product that was twice purified by column chromatography (2:1 hexanes–ethyl acetate in the first column, 20:1 CH₂Cl₂–MeOH in the second column) to give **34b** as a pale-

yellow solid (0.55 g, 33%). mp 142–143 °C; ^1H NMR: δ 5.30 (bs, 1H, NH), 5.71 (d, $J = 4.2$ Hz, 1H, Ar_2CH), 7.13–7.17 (m, 1H), 7.26–7.38 (m, 4H), 7.55–7.59 (m, 2H), 7.80–7.84 (m, 1H); ^{13}C NMR: δ 60.56 (Ar_2CH), 121.22, 125.25, 125.68, 127.59, 129.20, 129.71, 130.50, 133.46, 134.50, 135.05, 138.89, 140.77. Anal. Calcd for $\text{C}_{13}\text{H}_{10}\text{ClNO}_2\text{S}$: C, 55.82; H, 3.60; Cl, 12.67; N, 5.01; S, 11.46. Found: C, 56.01; H, 3.67; Cl, 12.55; N, 4.93; S, 11.38.

($\pm$)-2-Ethyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34c).¹⁰ To **43a** (1.02 g, 3.14 mmol) was added TFA– CH_2Cl_2 (1:4 v/v, 15 mL). The resulting dark brown solution was stirred at room temperature for 24 h. After the solvent was evaporated, the crude product was purified by column chromatography (CH_2Cl_2) and recrystallized (CH_2Cl_2) to give **34c** as white crystals (0.67 g, 70%). mp 100–101 °C; ^1H NMR: δ 1.23 (t, $J = 7.2$ Hz, 3H, CH_3), 3.28 (m, 2H, CH_2), 5.43 (s, 1H, Ar_2CH), 7.02–7.07 (m, 1H), 7.28–7.38 (m, 3H), 7.40 (s, 1H), 7.46–7.53 (m, 2H), 7.75–7.81 (m, 1H); ^{13}C NMR: δ 13.13 (CH_3), 36.78 (CH_2), 63.63 (Ar_2CH), 120.48, 124.70, 125.82, 127.43, 128.83, 129.19, 130.19, 132.71, 133.62, 134.59, 137.10, 139.47. Anal. Calcd for $\text{C}_{15}\text{H}_{14}\text{ClNO}_2\text{S}$: C, 58.53; H, 4.58; Cl, 11.52; N, 4.55; S, 10.42. Found: C, 58.64; H, 4.62; Cl, 11.59; N, 4.50; S, 10.50.

($\pm$)-2-Butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34d).¹⁰ To **43b** (2.43 g, 6.88 mmol) was added concd H_2SO_4 (10 mL). The resulting dark-brown solution was stirred at room temperature for 2 h. Crushed

ice was then added to the solution, which was extracted with CH_2Cl_2 , and the organic extract was dried over MgSO_4 . The crude product obtained upon evaporation of the solvent was twice purified by column chromatography (3:1 hexanes–ethyl acetate in the first column, CH_2Cl_2 in the second column) to give **34d** as white crystals (1.19 g, 52%). mp 116–117 °C; ^1H NMR: δ 0.81 (t, J = 7.3 Hz, 3H, CH_3), 1.29 (m, 2H), 1.61 (m, 2H), 3.09–3.31 (m, 2H, NCH_2), 5.39 (s, 1H, Ar_2CH), 7.02–7.07 (m, 1H), 7.27–7.35 (m, 3H), 7.39 (s, 1H), 7.46–7.53 (m, 2H), 7.76–7.82 (m, 1H); ^{13}C NMR: δ 13.17 (CH_3), 19.63 (CH_2), 29.57 (CH_2), 42.01 (CH_2), 64.53 (Ar_2CH), 120.61, 124.73, 125.89, 127.55, 128.88, 129.25, 130.19, 132.74, 133.68, 134.65, 137.20, 139.59. Anal. Calcd for $\text{C}_{17}\text{H}_{18}\text{ClNO}_2\text{S}$: C, 60.80; H, 5.40; Cl, 10.56; N, 4.17; S, 9.55. Found: C, 60.55; H, 5.32; Cl, 10.80; N, 4.06; S, 9.42.

(±)-2-*tert*-Butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34e).¹⁰ To **43c** (3.22 g, 9.12 mmol) was added TFA– CH_2Cl_2 (1:4 v/v, 40 mL). The resulting dark-brown solution was stirred at room temperature for 18 h. After solvent was evaporated, the crude products were separated by column chromatography (CH_2Cl_2) to give **34e** as white crystals (0.63 g, 21%) along with **34b** (0.59 g) and recovered **43c** (1.5 g). mp 162 °C; ^1H NMR: δ 1.45 (s, 9H, 3 CH_3), 5.60 (s, 1H, Ar_2CH), 7.05–7.09 (m, 1H), 7.25–7.31 (m, 2H), 7.36–7.40 (m, 1H), 7.45–7.49 (m, 3H), 7.74–7.78 (m, 1H); ^{13}C NMR: δ 29.05 (3 CH_3), 58.77 ($\text{N}\underline{\text{C}}\text{Me}_3$), 62.81(Ar_2CH), 120.88, 124.58, 124.70, 126.37, 128.34, 129.28, 130.47, 132.83, 134.47, 134.89, 137.08, 145.02. Anal. Calcd for $\text{C}_{17}\text{H}_{18}\text{ClNO}_2\text{S}$: C, 60.80; H, 5.40;

Cl, 10.56; N, 4.17; S, 9.55. Found: C, 60.89; H, 5.39; Cl, 10.49; N, 4.12; S, 9.64.

(±)-2-Methyl-3-phenyl-3-trifluoromethyl-2,3-dihydrobenzo[*d*]isothiazole 1,1-dioxide (34f).¹⁰ To **49** (0.78 g, 1.88 mmol) was added concd H₂SO₄ (6 mL). The resulting dark-brown solution was stirred at room temperature for 21 h. Crushed ice was then added to the solution, which was extracted with CH₂Cl₂, and the organic extract was dried over MgSO₄. The crude product that was obtained after solvent evaporation was purified by column chromatography (CH₂Cl₂) and recrystallized (ethyl acetate) to give **34f** as white crystals (0.51 g, 85%). mp 174 °C; ¹H NMR (DMSO-*d*₆): δ 2.82 (s, 3H, NCH₃), 7.27–7.30 (m, 2H), 7.42–7.54 (m, 4H), 7.75–7.85 (m, 2H), 8.16 (dd, *J* = 6.6, 2.2 Hz, 1H); ¹³C NMR (DMSO-*d*₆): δ 25.37 (NCH₃), 71.61 (q, ²*J*_{CF} = 28.6 Hz, Ar₂CCF₃), 121.56, 124.58 (q, ¹*J*_{CF} = 289 Hz, CF₃), 126.12, 127.06, 129.57, 129.68, 131.91, 132.17, 133.15, 133.57, 134.45. Anal. Calcd for C₁₅H₁₂F₃NO₂S: C, 55.04; H, 3.70; N, 4.28; S 9.79. Found: C 55.10, H 3.71, N 4.28, S 9.87.

Preparation of *N*-ethylbenzenesulfonamide (42a). To a well-stirred dry THF (50 mL) suspension of benzenesulfonyl chloride (20 mL, 0.16 mol) and ethylamine hydrochloride (16.2 g, 1.3 equiv) in a -10 °C bath was added dry triethylamine (55 mL, 2.5 equiv) dropwise. After the addition was complete, the suspension was slowly warmed up to room temperature and stirred overnight. The resulting white precipitate was filtered off, and the filtrate was washed with brine,

dried over MgSO_4 , and concentrated to give **42a** as pale yellow needles (24.4 g, 84%). ^1H NMR: δ 1.10 (t, $J = 7.3$ Hz, 3H, CH_3), 3.01 (quintet, $J = 6.9$ Hz, 2H, CH_2), 4.80 (bs, 1H, NH), 7.48–7.59 (m, 3H), 7.87–7.91 (m, 2H); ^{13}C NMR: δ 14.98(CH_3), 38.20 (CH_2), 128.99, 129.04, 132.53, 139.95 (quat.).

Preparation of *N*-butylbenzenesulfonamide (42b). To a well-stirred dry THF (20 mL) solution of butylamine (20 mL, 1 equiv) and triethylamine (29.3 mL, 1 equiv) in a -10 °C bath was added benzenesulfonyl chloride (26.7 mL, 0.21 mol) dropwise in 20 min. The resulting suspension was stirred at room temperature for 25 min. It was then poured into water and extracted with CH_2Cl_2 . The organic extract was dried over MgSO_4 and concentrated to give **42b** as white needles (43.3 g, 97%). ^1H NMR: δ 0.80 (t, $J = 7.2$ Hz, 3H, CH_3), 1.26 (sextet, $J = 7.3$ Hz, 2H), 1.43 (quintet, $J = 7.2$ Hz, 2H), 2.92 (q, $J = 6.6$ Hz, 2H), 5.55 (t, $J = 5.9$ Hz, 1H, NH), 7.46–7.60 (m, 3H), 7.89–7.93 (m, 2H); ^{13}C NMR: δ 13.20(CH_3), 19.35(CH_2), 31.16(CH_2), 42.62 (CH_2), 126.71, 128.77, 132.23, 139.69 (quat.).

Preparation of *N*-tert-butylbenzenesulfonamide (42c). To a well-stirred dry THF (100 mL) solution of *tert*-butylamine (16.22 g, 1.2 equiv) and triethylamine (33 mL, 1.1 equiv) in a 0 °C bath was added benzenesulfonyl chloride (23.6 mL, 0.18 mol) dropwise. The resulting suspension was stirred at room temperature overnight. After filtration, the filtrate was washed by brine, dried over MgSO_4 , and concentrated to give **42c** as a brown solid (39.4 g, 100%). ^1H NMR: δ 1.22 (s, 9H,

3CH₃), 4.82 (bs, 1H, NH), 7.46–7.54 (m, 3H), 7.89–7.93 (m, 2H); ¹³C NMR: δ 30.15 (3CH₃), 54.65 (NCMe₃), 128.92, 128.83, 132.10, 143.41 (quat.).

***N*-tert-Butyl-2-[(3-chlorophenyl)hydroxymethyl]benzenesulfonamide (43c).**¹⁰ To a dry THF solution (50 mL) of **42c** (5.98 g, 28 mmol) in a 0 °C bath was added BuLi (7.5 mL of a 10.0 M hexanes solution, 2.7 equiv) dropwise. The resulting brown solution was stirred at 0 °C for 15 min before *m*-chlorobenzaldehyde (5.7 mL, 1.8 equiv) was added, after which time the solution changed color to green, black then finally amber. It was allowed to be slowly warmed up to room temperature over a period of 0.5 h, and aq HCl (5%, 11 mL) was added. The mixture was extracted with CH₂Cl₂, and the organic extract was dried over MgSO₄. The crude product obtained upon evaporation of solvent was then purified by column chromatography (CH₂Cl₂ followed by a CH₂Cl₂–MeOH gradient) to give **43c** as a brown glassy solid (7.98 g, 81 %). ¹H NMR: δ 1.21 (s, 9H, 3CH₃), 3.48 (s, 1H, OH), 4.65 (bs, 1H, NH), 6.72 (s, 1H, Ar₂CH), 7.35–7.50 (m, 7H), 8.07 (dd, *J* = 7.7, 1.4 Hz, 1H); ¹³C NMR: δ 30.12 (3CH₃), 55.50 (NCMe₃), 70.57 (Ar₂CH), 125.07, 126.98, 127.83, 128.19, 129.28, 129.77, 130.43, 132.93, 134.56, 140.96, 141.41, 144.02.

***N*-tert-Butyl-2-(2,2,2-trifluoro-1-hydroxy-1-phenylethyl)benzenesulfonamide (44a).** The preparation procedure was the same as that for **43c**, except using **42c** (2.02 g, 9.48 mmol) as starting material. The crude product was

purified by column chromatography (CH_2Cl_2) to give **44a** as a white solid (2.51 g, 68%). ^1H NMR: δ 1.05 (s, 9H, 3CH_3), 3.83 (s, 1H, OH), 6.67 (s, 1H, NH), 7.39–7.42 (m, 3H), 7.47–7.62 (m, 4H), 7.72–7.77 (m, 1H), 8.29 (dd, $J = 7.7, 1.6$ Hz, 1H); ^{13}C NMR: δ 30.03 (3CH_3), 55.84 ($\text{N}\underline{\text{C}}\text{Me}_3$), 81.04 (q, $^2J_{\text{CF}} = 27.9$ Hz, $\text{Ar}_2\underline{\text{C}}\text{CF}_3$), 124.87 (q, $^1J_{\text{CF}} = 288$ Hz, CF_3), 128.15, 128.53, 129.13, 129.25, 129.98, 130.04, 131.74, 137.17, 138.81, 142.35.

***N*-tert-Butyl-*N*-methyl-2-(2,2,2-trifluoro-1-hydroxy-1-phenylethyl)-benzenesulfonamide (45).** To a dry THF (5 mL) solution of **44a** (1.16 g, 3.0 mmol) at room temperature was added $^t\text{BuOK}$ (4.5 mL 1.0 M THF solution, 1.5 equiv), followed by MeI (1.7 mL, 9 equiv). The solution was stirred overnight. The resulting suspension was then poured onto crushed ice, extracted with CH_2Cl_2 , and the organic extract was dried over MgSO_4 . The crude product obtained upon solvent evaporation was separated by column chromatography (CH_2Cl_2). Compound **45** eluted first from the column, and the appropriate fraction was concentrated to give white needles (0.73 g, 61%) of pure **45**. mp 129–130 °C; ^1H NMR: δ 1.30 (s, 9H, 3CH_3), 2.92 (s, 3H, NCH_3), 7.09 (s, 1H, OH), 7.35 (s, 5H, C_6H_5), 7.52 (pt, $J = 7.6$ Hz, 1H), 7.62 (pt, $J = 8.4$ Hz, 1H), 7.82 (dd, $J = 7.8, 1.6$ Hz, 1H), 7.91 (dd, $J = 7.9, 1.5$ Hz, 1H); ^{13}C NMR: δ 28.83 (3CH_3), 32.51 (NCH_3), 59.59 ($\text{N}\underline{\text{C}}\text{Me}_3$), 80.79 (q, $^2J_{\text{CF}} = 28.0$ Hz, $\text{Ar}_2\underline{\text{C}}\text{CF}_3$), 124.92 (q, $^1J_{\text{CF}} = 286$ Hz, CF_3), 127.49, 128.46, 128.71, 128.77, 129.16, 130.25, 131.80, 138.32, 138.77, 143.53. Anal. Calcd for $\text{C}_{19}\text{H}_{22}\text{NSO}_3\text{F}_3$: C, 56.84; H, 5.53; N, 3.49. Found: C, 57.06; H, 5.45; N,

3.59.

***N*-tert-Butyl-2-(2,2,2-trifluoro-1-methoxy-1-phenylethyl)-benzenesulfonamide (46).** Compound **46** was obtained as a byproduct in the preparation of **45**. It eluted out of the column after **45** during the chromatographic separation. It crystallized as white block-like crystals (0.21 g, 18%). mp 163–164 °C; ¹H NMR: δ 1.27 (s, 9H, 3CH₃), 3.27 (s, 3H, OMe), 6.03 (s, 1H, NH), 7.15–7.19 (m, 1H), 7.34–7.46 (m, 7H), 8.44 (dd, *J* = 7.9, 1.7 Hz, 1H); ¹³C NMR: δ 30.10 (3CH₃), 55.17 (NCMe₃ and OMe), 128.62, 128.70, 129.31, 129.46, 130.31, 130.80, 131.98, 135.80, 137.03, 143.68. Anal. Calcd for C₁₉H₂₂NSO₃F₃: C, 56.84; H, 5.53; N, 3.49. Found: C, 56.92; H, 5.50; N, 3.43.

3-Phenyl-3-trifluoromethyl-3*H*-benzo[*c*][1,2]oxathiole 1,1-dioxide (47). This compound was obtained as a byproduct in the solid-phase synthesis of **34f**. It (*R_f* = 0.7) was separated from **34f** (*R_f* = 0.4) by column chromatography (CH₂Cl₂) as a white solid. The NMR data were in agreement with those from samples prepared by my co-worker James Miller.

***N*-tert-Butyl-*N*-methylbenzenesulfonamide (48).** To a suspension of **42c** (8.54 g, 40 mmol) and NaH (a 60% mixture with mineral oil, 3.7 g, 2.3 equiv) in DMF (200 mL) was added MeI (4.4 mL, 1.8 equiv) dropwise at room temperature. After 3 h stirring, DMF was evaporated. To the resulting yellow slurry was added ethyl

acetate and water to destroy excess NaH. The water phase was extracted twice with CH₂Cl₂. The combined organic phase was dried over MgSO₄, and the crude product obtained upon solvent evaporation was purified by column chromatography (CH₂Cl₂) to give **48** as white crystals (7.71 g, 85%). ¹H NMR: δ 1.36 (s, 9H, 3CH₃), 2.99 (s, 3H, NCH₃), 7.49–7.52 (m, 3H), 7.82–7.86 (dd, *J* = 7.7, 1.4 Hz, 2H); ¹³C NMR: δ 28.87 (3CH₃), 32.19 (NCH₃), 58.41 (NCMe₃), 126.52, 128.58, 131.74, 142.89.

***N*-tert-Butyl-*N*-methyl-2-(2,2,2-trifluoro-1-methoxy-1-phenyl-ethyl) benzenesulfonamide (49).** To a suspension of **45** (0.80 g, 2.0 mmol) and NaH (a 60% mixture with mineral oil, 0.44 g, 5.5 equiv) in dry THF (15 mL) was added MeI (1.9 ml, 15 equiv) dropwise at room temperature. After stirring overnight, water was slowly added to the solution to destroy excess NaH. The water phase was extracted once with CH₂Cl₂. The combined organic phase was dried over MgSO₄. Solvent evaporation gave a crude product that was purified by column chromatography (CH₂Cl₂) to give **49** as white crystals (0.70 g, 88%). ¹H NMR: δ 1.25 (s, 9H, 3CH₃), 3.03 (s, 3H, NCH₃), 3.55 (s, 3H, OMe), 7.32–7.34 (m, 3H), 7.41–7.42 (m, 2H), 7.61–7.65 (m, 2H), 7.87–7.90 (m, 1H), 8.04 (dd, *J* = 7.2, 1.5 Hz, 1H); ¹³C NMR: δ 28.53 (3CH₃), 32.31 (NCH₃), 54.85 (OMe), 58.37 (NCMe₃), 85.64 (q, ²*J*_{CF} = 25.1 Hz, Ar₂CCF₃), 125.01 (q, ¹*J*_{CF} = 293 Hz, CF₃), 126.94, 127.15, 127.68, 127.79, 128.86, 129.89, 130.58, 134.71, 138.54, 143.75.

7. References

1. Mann, J. M.; Tarantola, D. J. M. HIV 1998: The Global Picture. *Scientific American*, **1998**, July, page 82–83.
2. Young, S. D. Non-nucleoside Inhibitors of HIV-1 Reverse Transcriptase. *Perspectives in Drug Discovery and Design*, **1993**, 1, 181–192.
3. Tramontano, E.; Cheng, Y.-C. HIV-1 reverse transcriptase inhibition by a dipyrroldiazepinone derivative: BI-RG-587. *Biochem. Pharmacol.* **1992**, 43, 1371–1376.
4. Saunders, J.; Storer, R. *Drug News and Perspectives* **1992**, 5, 153.
5. Larder, B. A.; Darby, G.; Richman, D. D. HIV with reduced sensitivity to zidovudine (AZT) isolated during prolonged therapy. *Science* **1989**, 243, 1731–1734.
6. Larder, B. A.; Kemp, S. D. Multiple mutations in HIV-1 reverse transcriptase confer high-level resistance to zidovudine (AZT). *Science* **1989**, 246, 1155–1158.
7. Nunberg, J. H.; Schleif, W. A.; Boots, E. J.; O'Brien, J. A.; Quintero, J. C.; Hoffman, J. M.; Emini, E. A.; Goldman, M. E. Viral resistance to human immunodeficiency virus type 1-specific pyridinone reverse transcriptase inhibitors. *J. Virol.* **1991**, 65, 4887–4892.
8. Richman, D.; Shih, C. -K.; Lowy, I.; Rose, J.; Prodanovich, P.; Goff, S.; Griffin, J. Human immunodeficiency virus type 1 mutants resistant to

nonnucleoside inhibitors of reverse transcriptase arise in tissue culture.

Proc. Natl. Acad. Sci. USA **1991**, *88*(24), 11241–11245.

9. Mayasundari, A. Ph. D. dissertation, University of Tennessee, Knoxville, 1998.
10. Watanabe, H.; Gay, R. L.; Hauser, C. R. *ortho* Metalation of N-substituted benzenesulfonamide by excess *n*-butyllithium. Condensation with carbonyl compounds. Cyclization. *J. Org. Chem.* **1968**, *33*, 900–903.
11. Plunkett, M.; Ellman, J. A.; Germanium and Silicon linking strategies for traceless solid-phase synthesis. *J. Org. Chem.* **1997**, *62*, 2885–2893.
12. An improved preparation method for the Si-linked resin was later reported by Ellman's group: Woolard, F. X.; Paetsch, J.; Ellman, J. A. A Silicon linker for direct loading of aromatic compounds to supports. Traceless synthesis of pyridine-based tricyclics. *J. Org. Chem.* **1997**, *62*, 6102–6103.
13. Beaver, K. A.; Siegmund, A. C.; Spear, K. L. Application of the sulfonamide functional group as an anchor for solid phase organic synthesis (SPOS). *Tetrahedron Lett.* **1996**, *37*, 1145–1148.
14. We were aware of the possibility of getting lithiated benzene on the polystyrene backbone by butyllithium: Halm, C.; Evarts, J.; Kurth, M. J. A new attachment/cleavage strategy: Polymer-bound allylic sulfones in a solid-phase route to trisubstituted olefins. *Tetrahedron Lett.*, **1997**, *38*, 7709–7712.
15. Welslow, O. W.; Kiser, R.; Fine, D.; Bader, J.; Shoemaker, R. H.; Boyd, M.

- R. *J. Natl. Cancer Inst.*, **1989**, *81*, 577–586.
16. Rink, H. Synthetic Polystyrene Resin and its Use in Solid Phase Peptide Synthesis. US patent 4,859,736, 1989.
 17. Snieckus, V. Directed aromatic metalation: A continuing education in flatland chemistry. In *Chemical Synthesis*; Chatgililoglu, C. and Snieckus, V., Eds.; Kluwer Academic Publishers: Norwell, MA, 1996, pp 191–221.
 18. Boehm, T. L.; Showalter, H. D. H. Development of novel silyl ether linker for solid-phase organic synthesis. *J. Org. Chem.* **1996**, *61*, 6498–6499.
 19. Havez, S.; Begtrup, M.; and Vedsø, P. Directed *ortho*-lithiation on the solid phase; *J. Org. Chem.* **1998**, *63*, 7418–7420.
 20. Seeberger, P. H.; Beebe, X.; Sukenick, G. D.; Pochapsky S.; Danishefsky, S. J. Monitoring the progress of solid-phase oligosaccharide synthesis by high-resolution magic angle spinning NMR: observations of enhanced selectivity for β -glycoside formation from α -1,2-anhydrosugar donors in solid-phase couplings; *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 491–493.
 21. Carpino, L. A.; Mansour, E. M. E.; Cheng, C. H.; Williams, J. R.; MacDonald, R.; Knapczyk, J.; Carman, M. Polystyrene-based deblocking-scavenging agents for the 9-fluorenylmethyloxycarbonyl amino-protecting group. *J. Org. Chem.* **1983**, *48*, 661–665.
 22. Ray, S.; Grover, P. K.; and Anand, N. Synthesis of *cis*- and *trans*-3,4-diphenyldihydro-coumarins and *threo*- and *erythro*-2,3-diphenyl-3-*o*-hydroxyphenylpropionic Acids. *Indian J. Chem.* **1971**, *9*, 619–623.

PART THREE

SYNTHESIS OF HYALURONIC ACID DISACCHARIDE AND ITS C-LINK ANALOGUE

1. INTRODUCTION

1.1. Biological background

Hyaluronic acid (HA), a glycosaminoglycan with molecular weight up to ten million, is made up of the disaccharide repeating unit $[(-3)\text{-}\beta\text{-D-GlcNAc-(1}\rightarrow\text{4)-}\beta\text{-D-GlcA}]$. It is a constituent of the extracellular matrix.¹ Generally, the extracellular matrix consists of diverse proteins and carbohydrates in a dense space. This matrix acts as a filter wall that allows proteins and other molecules to be transported between cells but prevents the intermixing of cells. This function renders the extracellular matrix a natural barrier to tumor cell invasion. However, this barrier can be destroyed by an enzyme called metalloprotein secreted by malignant tumor cells.² One of the characteristics of a malignant tumor is that it can spread the malignant cells throughout the body (metastasis) so that secondary tumors are induced elsewhere. Metastasis is a complex series of events that requires the tumor cell to (1) detach from its primary cell (tumor) neighbors, (2) migrate through the extracellular matrix to reach a blood vessel, (3) cross the basal lamina and endothelium of the blood vessel to enter the general circulation, (4) arrive to a new location, (5) extravasate by crossing the basal lamina, and (6) migrate through the extracellular matrix to gain a new attachment point.³ Among these steps, HA plays a critical role in (2) and (6) by interacting with the tumor cell's CD44 receptor. CD44 is a widely distributed transmembrane glycoprotein that participates in cellular adhesion and migration, hyaluronate degradation, lymphocyte activation, lymph node homing, myelopoiesis and lymphopoiesis, angiogenesis, and release

of cytokines.⁴ The extracellular region of CD44 is variably spliced to give multiple CD44 isoforms. All these isoforms contain an amino-terminal domain which is important for HA binding.⁵ Based on several studies,⁶ a metastasis model related to the interaction between CD44 receptor and HA is summarized as follows: migrating tumor cells release signal molecules that induce adjacent host cells to produce increased amounts of extracellular HA; the cancer cells then make use of their CD44 receptors to bind to and migrate along the newly synthesized HA tracks into new tissues. Although this might be a simplified model for a series of complex events, there might be a possible therapeutic approach to the prevention of metastasis based on the introduction of an antagonist of the CD44 receptor that would effectively block a tumor cell's binding to HA. Such an approach would be significantly different from other approaches, as it would involve essentially cell-surface phenomena and would not make use of cytotoxic agents that involve the cell cycle.

1.2. HA oligosaccharides as antimetastatic agents

In 1997, our group submitted a grant application "Antimetastatic Oligosaccharides from Hyaluronic Acid" to the Medicinal Chemistry Study Section of NIH. We proposed to use small molecules derived from hyaluronic acid to interfere with CD44 receptors and to study their effects on metastasis. Our hypothesis is that an antagonist might be found for the CD44 receptor to prevent HA binding, and consequently limit metastasis. To this end, it was envisioned that

an oligosaccharide of unspecified length would be an ideal candidate as a probe for studying the role of CD44–HA binding, providing that it would (a) contain the requisite functionality to bind to the CD44 receptor, (b) be easily derived from HA by enzymatic or hydrolytic cleavage, and (c) would serve as a model for development of a hydrolytically stable, synthetic analogue. Furthermore, a synthetic analogue to this oligosaccharide, with an altered glycosidic linkage pattern, might be developed to provide a useful, stabilized probe for studies in vivo and possibly give insight into the development of an antimetastatic drug candidate. Exploratory structural and synthetic research was conducted by Ken Price for his Ph. D. research,⁷ and his excellent work led to the final granting of the NIH award.

In order to determine whether one might find an antagonist for the CD44 receptor from HA oligosaccharides, a set of experiments⁸ were carried out by our collaborator, Dr. Richard Cysyk (Head, Biochemistry Section, Laboratory of Medicinal Chemistry, National Cancer Institute), as follows: (1) HA was degraded with hyaluronate lyase (EC 4.2.2.1) from *Streptomyces hyalurolyticus*. (2) The resulting oligosaccharide mixture (1 mg/mL) was then incubated with B16F10 melanoma cells in vitro (10^5 cell count), and the mixture was injected i.v. (tail vein) into mice. (3) After 10 days, the lungs of the mice, along with a group of control animals that received only B16F10 cells, were excised, and the visible metastatic sites were counted. Our hopes were that we would see a reduction in metastasis in the mice whose B16F10 cells had been pre-treated with HA-oligosaccharide. To our surprise there was a near-total absence of metastatic colonies in the lungs of

the treated mice receiving the HA-oligosaccharide pretreated B16F10 cells! The result indicates that a relatively small molecule can function as an antimetastatic agent, which is essential to our proposed research. This experiment has been repeated with identical results using a variety of controls to ascertain that the result is genuine. B16F10 cell viability in the HA-oligosaccharide-treated sample was checked, as were various controls of column effluent, buffers, etc. Experiments using double and triple the amount of oligosaccharides (up to 3 mg/mL) were conducted to identify possible toxic effects of the digest on the melanoma cells—and no toxicity was found as determined by cell viability assays. Work is ongoing to determine activity vs. degree of polymerization (DP) relationship among the oligosaccharides. Preliminary studies show activity setting in with the 8-mer (DP = 8).

1.3. Design of hydrolase-resistant HA mimics

It is known that hyaluronan is degraded in the local tissues where it is produced, in the lymph nodes, and in the liver.⁹ In mammals, the degradation of HA is accomplished by the following three enzymes: hyaluronidase (EC 3.2.1.35), β -D-glucuronidase (EC 3.2.1.31), and 2-acetamido-2-deoxy-D-hexosaminidase (EC 3.2.1.30).^{9a,10} Hyaluronidase is an endoglycosidase specific for the glycosidic bond β -D-GlcNAc-(1-4)- β -D-GlcA, whereas the hexosaminidase and β -D-glucuronidase remove a GlcNAc or a GlcA residue, respectively, from the nonreducing end of the polymer. In other words, all β -(1-4) glycosidic bonds in HA are susceptible to

enzyme degradation. As a result, the β -(1 $\rightarrow$ 3) linked HA disaccharide can be made by degradation¹¹ of polymeric HA hence it is commercially available, but the β -(1 $\rightarrow$ 4) linked HA disaccharide cannot be obtained in this manner and has to be chemically synthesized.

In order to increase the in vivo lifetime of HA oligosaccharide, we proposed a "mixed" oligosaccharide that would contain C-linkages strategically placed at β -(1 $\rightarrow$ 4) bonds so that the oligosaccharide would be safe from enzymatic hydrolysis, while still contain regular glycosidic O-linkages at β -(1 $\rightarrow$ 3) bonds, mainly to facilitate ready synthesis, as the chemistry of glycosides is well-documented within carbohydrate chemistry.¹² C-linkages in this oligosaccharide would be placed, for example, on a octasaccharide at every linkage of the reducing end of a GlcNAc residue and at the first linkage of the nonreducing end of the oligomer, regardless of the residue (Figure 3.1).

The principal asset of this approach is its ready accessibility through a convergent "block synthesis" approach wherein multiple copies of a single C-linked disaccharide are linked n times via normal glycosidation techniques to produce oligomers of specified length (of either 4, 6, or 8 residues, etc.).

2. STATEMENT OF THE PROBLEM

2.1. Objectives

Among the three aspects (biological, structural and synthetic) of our HA project, the synthetic aspect is the focus of this dissertation. We felt that even

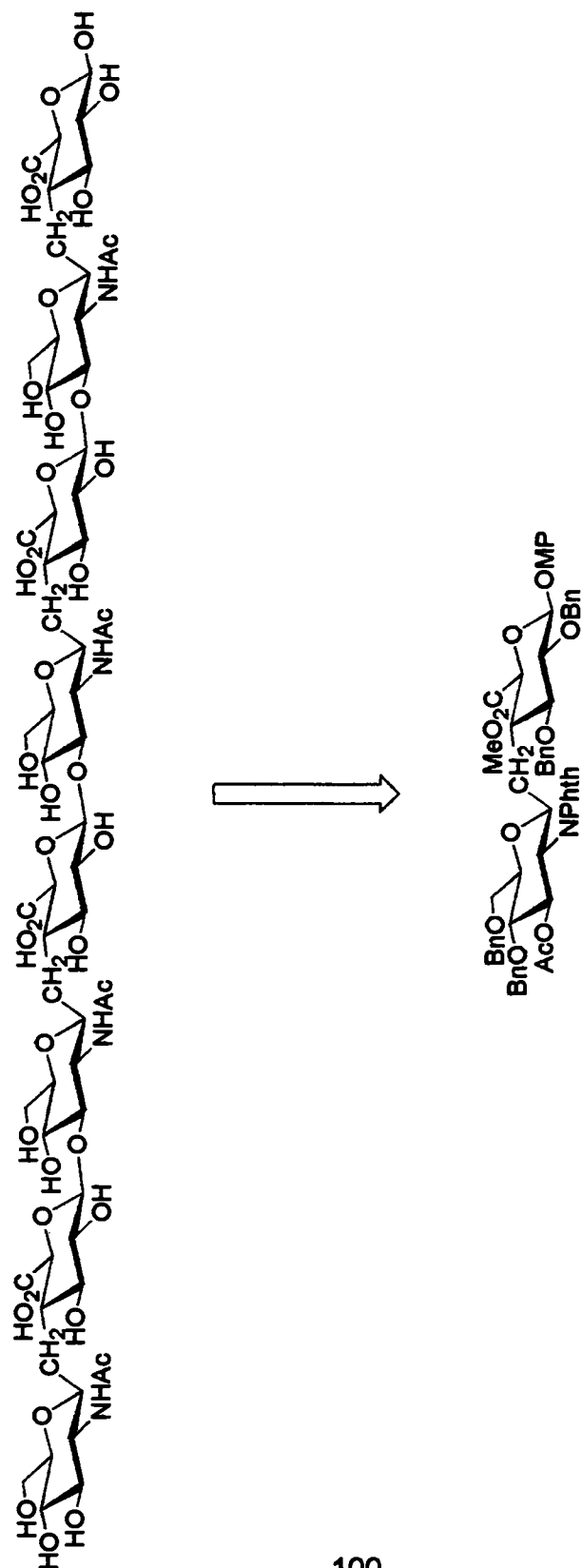


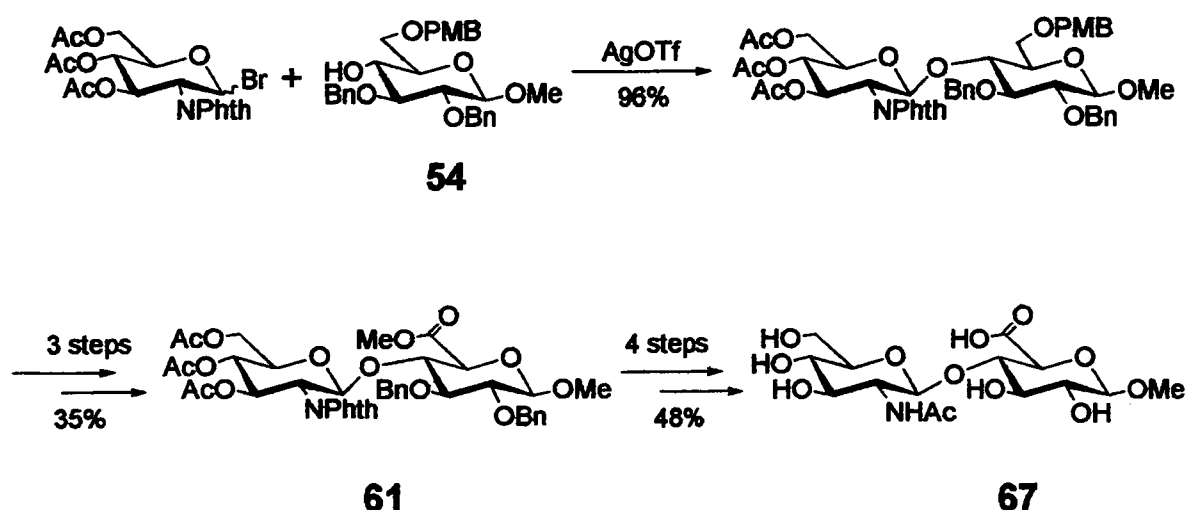
Figure 3.1. The design of a "mixed" octasaccharide and its C-disaccharide building block.

though the HA C-disaccharide building block is our major target, the HA disaccharide is also needed because it will provide us the material for NMR and molecular modeling studies as well as useful information for the C-disaccharide synthesis. Therefore, at this stage, both the HA disaccharide β -D-GlcNAc-(1 $\rightarrow$ 4)- β -D-GlcA and the HA C-disaccharide β -D-GlcNAc-CH₂(1 $\rightarrow$ 4)- β -D-GlcA are our synthesis targets. We chose the GlcNAc-(1 $\rightarrow$ 4)-GlcA block unit simply because this unit removes any vulnerability to glucuronidase activity.

2.2. Research plan

2.2.1. Synthesis plan for the β -D-GlcNAc-(1 $\rightarrow$ 4)- β -D-GlcA HA disaccharide

Lerner and co-workers¹³ reported a synthesis of the unprotected (1 $\rightarrow$ 4)-linked HA disaccharide **67** (Scheme 3.1). In their route, the *N*-phthalimido protected glycosyl bromide was coupled with the glycosyl acceptor **54** to give the

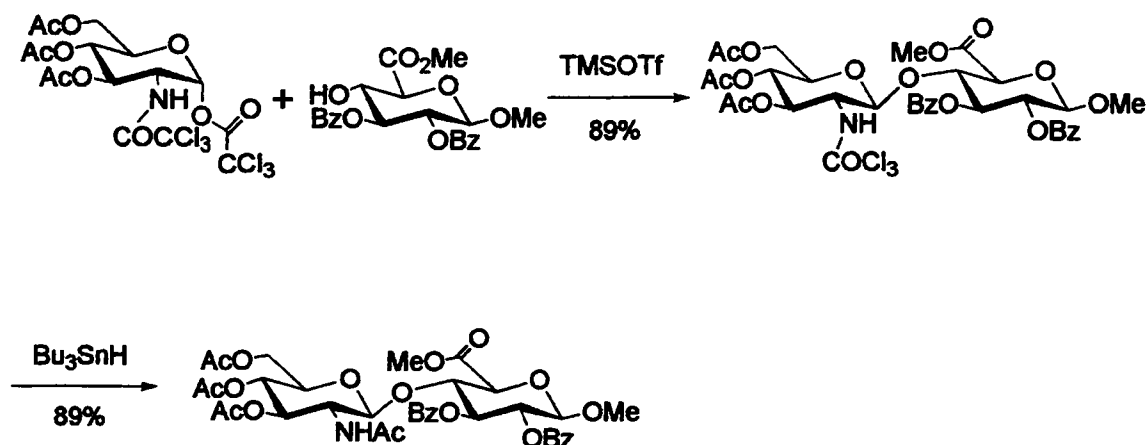


Scheme 3.1. Lerner's approach to HA disaccharide.

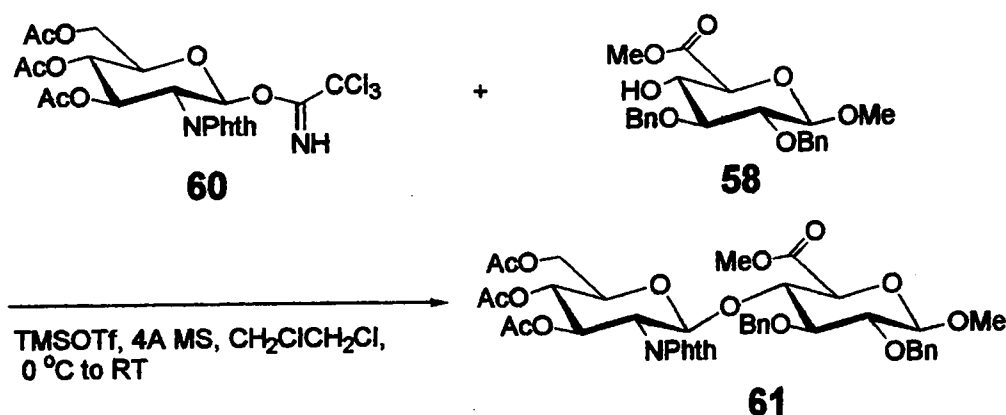
fully protected disaccharide, which required selective deprotection and oxidation at the C-6 of the reducing end and a four-step deprotection sequence to accomplish the unprotected HA disaccharide **67**. They have also successfully coupled the bromide with a glycosyl acceptor which already has C-6 oxidized as a *tert*-butyl ester, but failed to purify the acidic deprotection product derived from the ester.

Jacquinet and co-workers (Scheme 3.2)¹⁴ also reported a coupling between an *N*-trichloroacetamido protected trichloroacetimidate and a glycosyl acceptor with C-6 already oxidized to a methyl ester. This method has also been applied in syntheses of HA tetra-, hexa-, and octa-saccharides having a GlcA unit at the reducing end.¹⁵ The *N*-trichloroacetyl group in the coupling product can be transformed into *N*-acetyl group by Bu₃SnH reduction.

Because we would like to use common intermediates for both HA disaccharide and HA C-disaccharide, we decided to make the unprotected (1→4)-



Scheme 3.2. Jacquinet's approach to HA disaccharide.



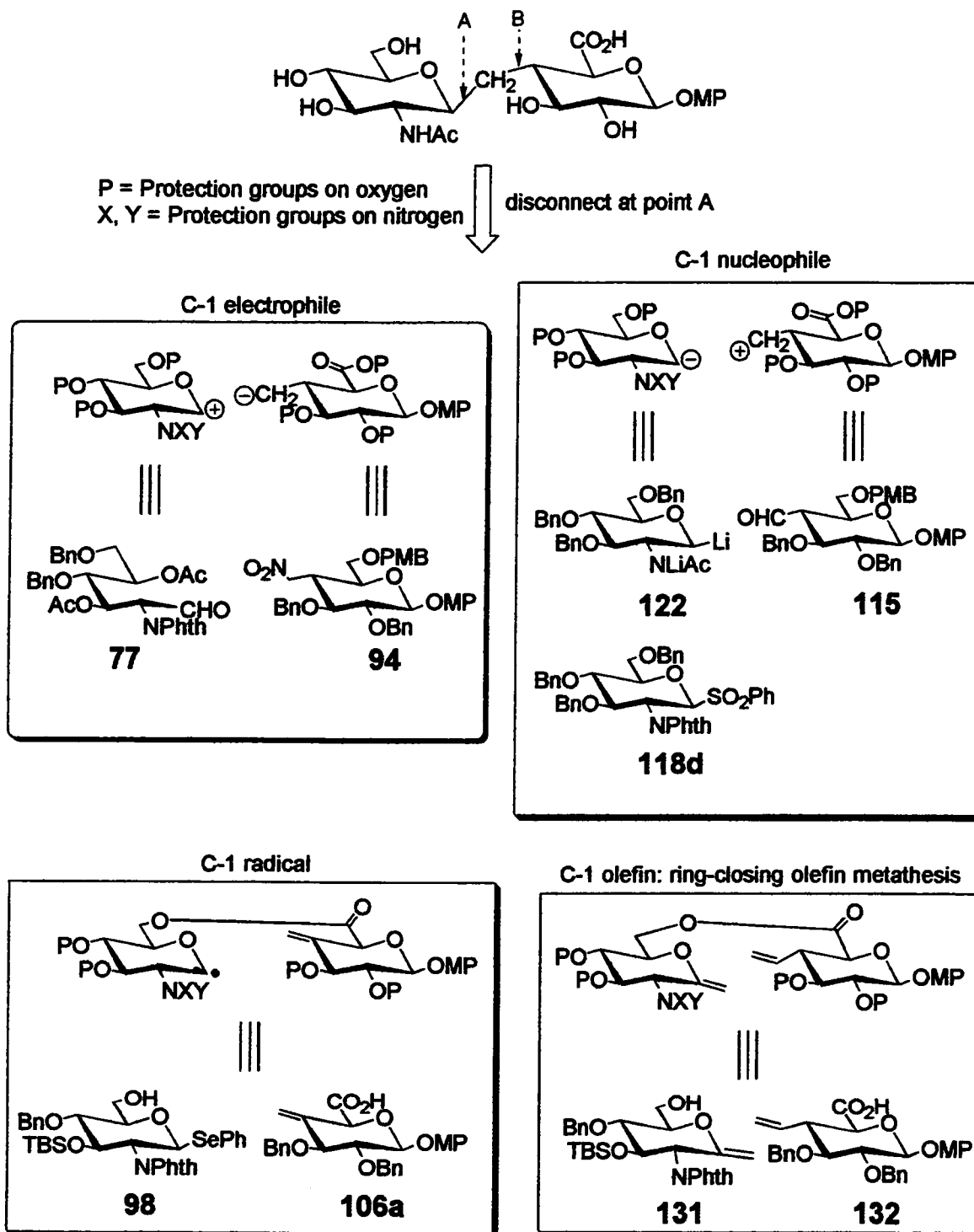
Scheme 3.3. Our synthesis plan for HA disaccharide

linked HA disaccharide **67** by coupling the *N*-phthalimido protected trichloroacetimidate **60** with the glycosyl acceptor **58** (Scheme 3.3). The deprotection would follow Lerner's four-step sequence.

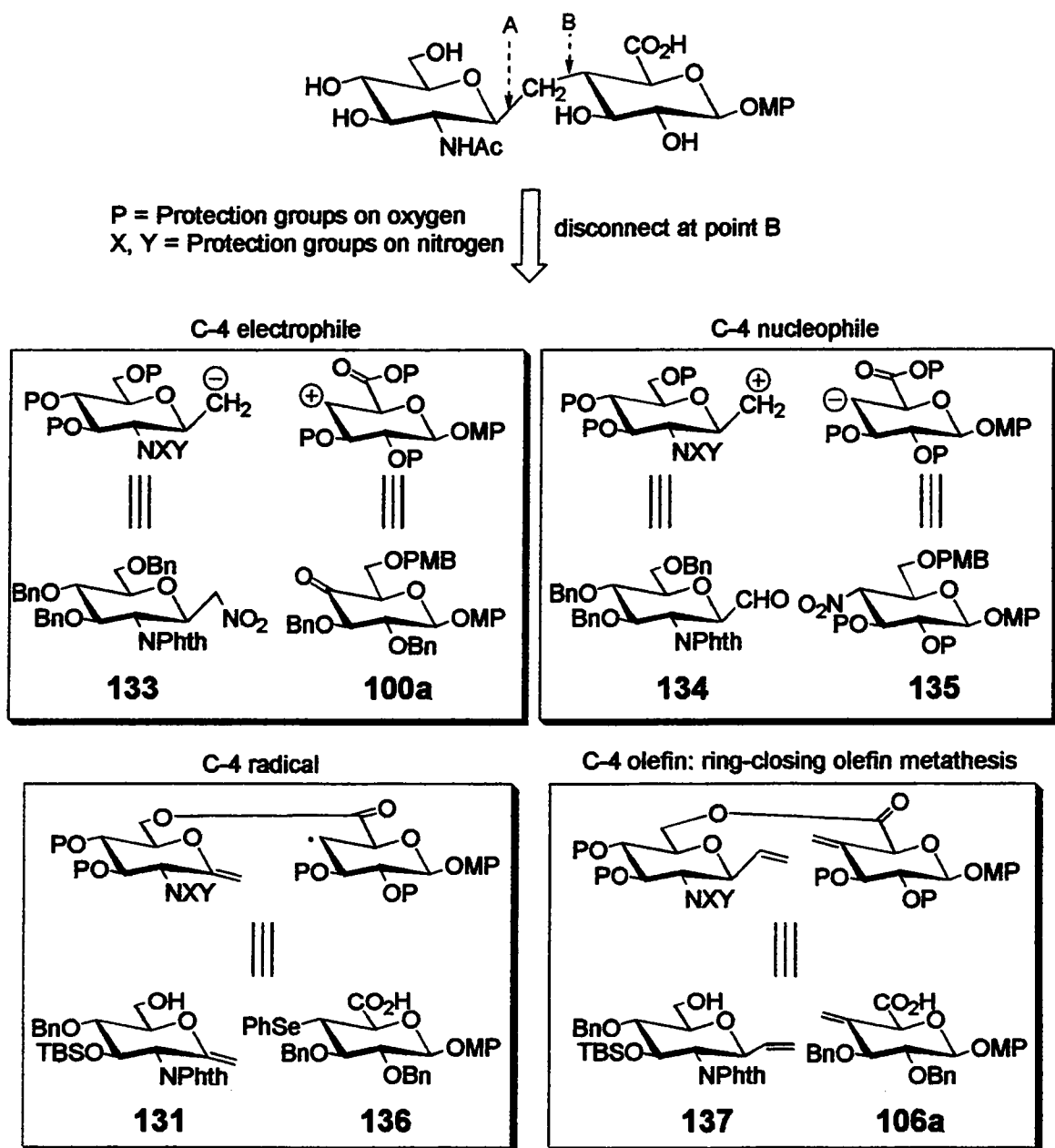
2.2.2. Retrosynthetic analysis of the β -D-GlcNAc-CH₂(1-4)- β -D-GlcA HA C-disaccharide

In general, there are two disconnection points in the retrosynthetic analysis of the target C-disaccharide, i.e., point A (between the CH₂ and the C-1 of the GlcNAc, Scheme 3.4) and point B (between the CH₂ and the C-4 of the GlcA, Scheme 3.5).

Point A disconnection would lead to four types of C-glycosyl donors, namely a C-1 electrophile (**77**), a C-1 nucleophile (**122** or **118d**), a C-1 radical (**98**), and a C-1 olefin (**131**). Among them, the ionic and radical coupling partners are very well reviewed in the area of C-glycoside synthesis.¹⁶ As for the ring-closing olefin



Scheme 3.4. Retrosynthetic analysis of HA C-disaccharide based on the disconnection at point A.

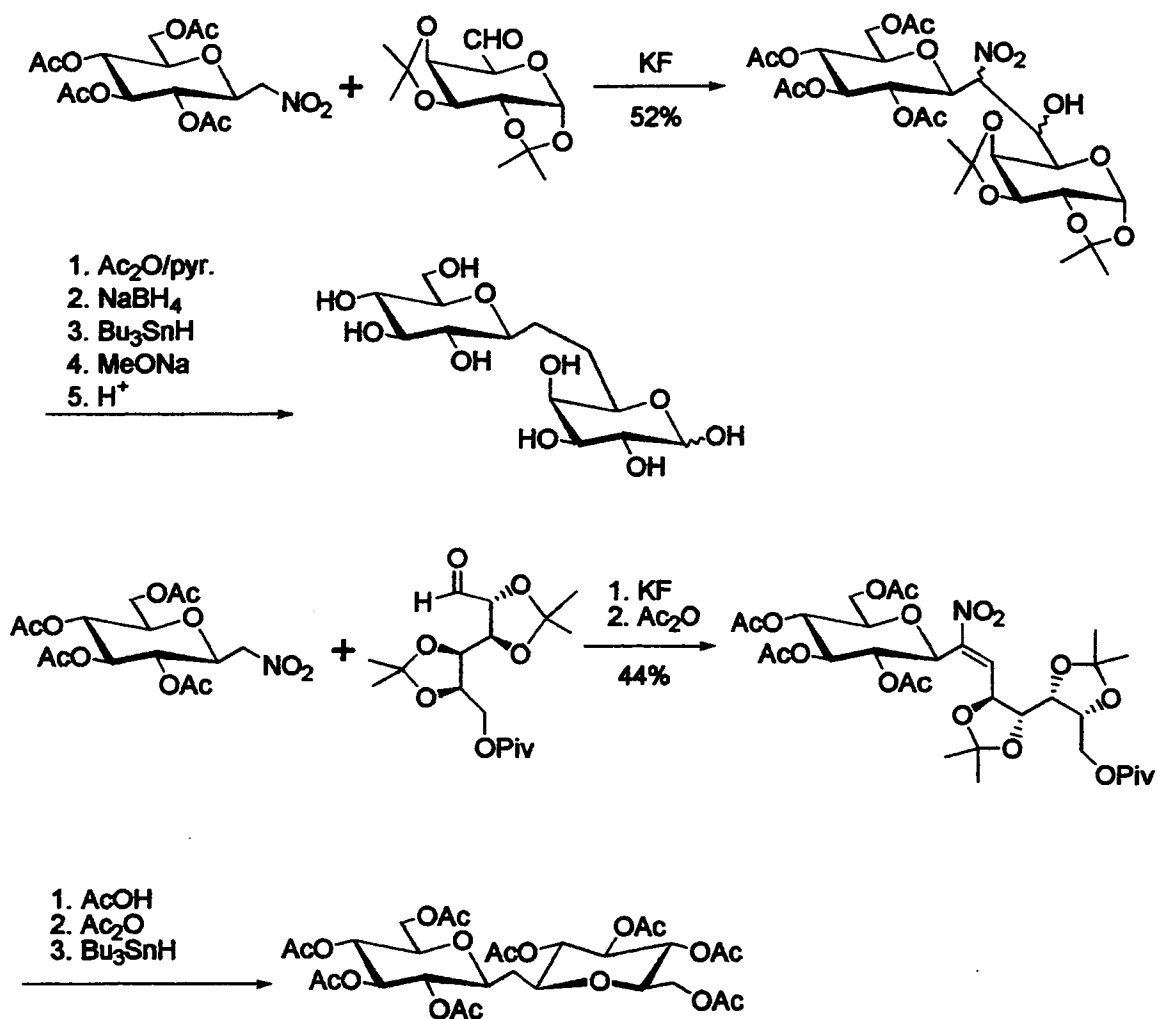


Scheme 3.5. Retrosynthetic analysis of HA C-disaccharide based on the disconnection at point B.

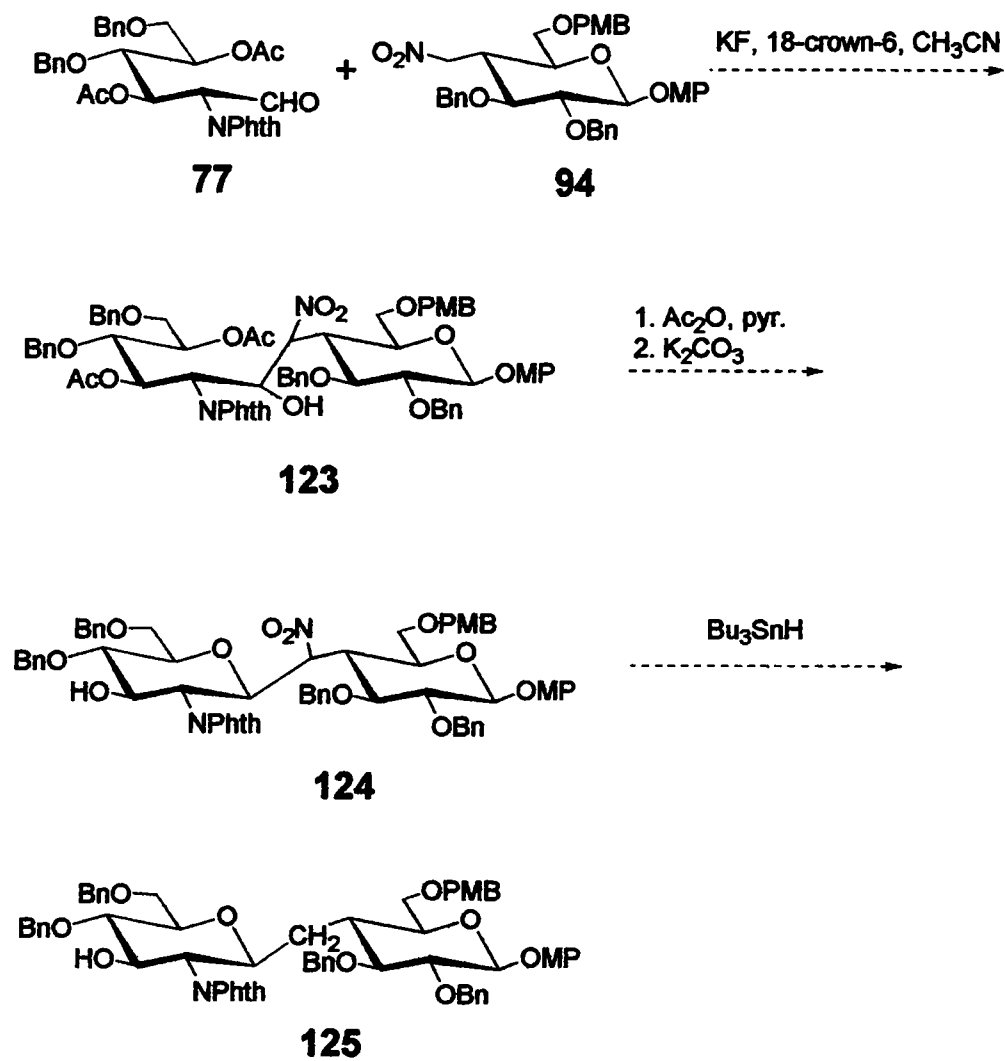
metathesis method,¹⁷ it has been very successfully applied in the synthesis of macrocyclic natural products, and its application in the C-disaccharide synthesis has just appeared in the literature recently.¹⁸ In point A disconnection, all C-glycosyl acceptors (**94**, **115** and **106a**) are one-carbon elongation products at the C-4 of the GlcA unit, of which the preparation methods are currently available. On the other hand, the point B disconnection would need one-carbon elongation (except **137**) at the C-1 of the GlcNAc unit (i.e. **133**, **134** and **131**). Limited by time and resources, we only tried three approaches based on the point A disconnection.

Our C-1 electrophile approach is closely related to that of Martin¹⁹ (Scheme 3.6). This approach was originally proposed in Ken Price's dissertation and our NIH grant application. In the plan (Scheme 3.7), coupling partners **77** and **94** would be synthesized from glucosamine hydrochloride in fifteen steps and galactose pentaacetate in nine steps, respectively. Martin's protocol would be used in the coupling of **77** and **94**, followed by pyran ring closure, and denitration. The resulting **125** would be transformed into the free HA C-disaccharide in five more steps. The advantage of this approach is that the stereochemistry of the C-disaccharide is well-defined, i.e., the 4-equatorial carbon atom is established by an S_N2 displacement of the 4-axial triflate from the galactose derivative **88**, and the β configuration of the C-glycosidic bond would be formed via an intramolecular Michael addition based on Martin's results. Obviously, the disadvantage is that it is a long synthesis.

In order to cut down on the number of the steps required for the C-



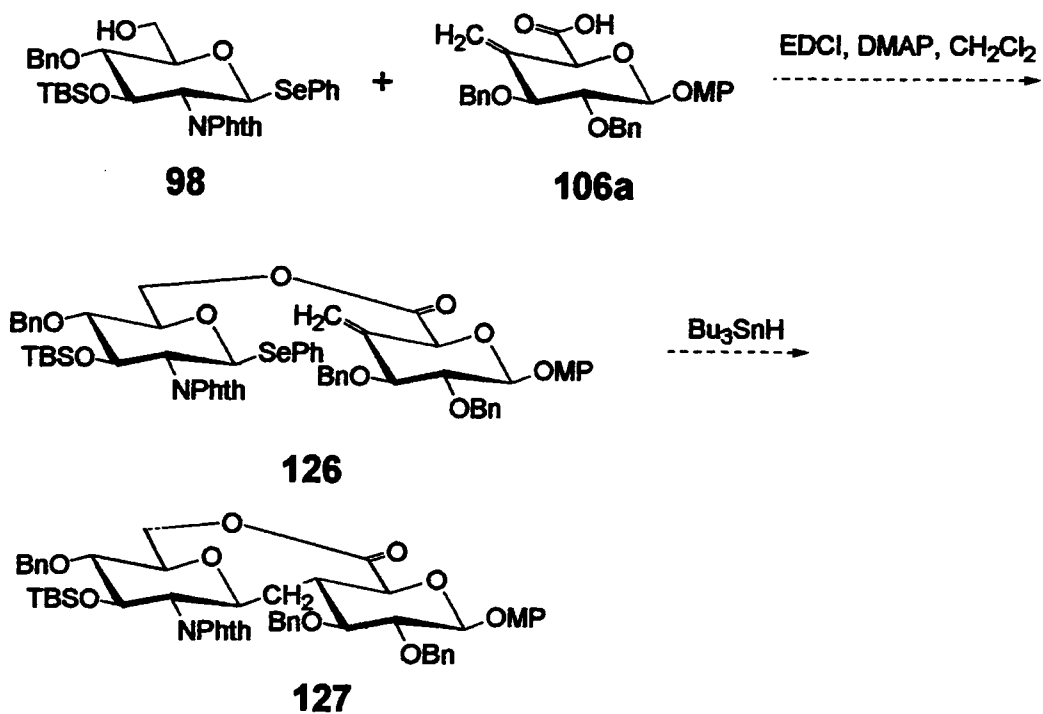
Scheme 3.6. Martin's nitrosugar approach to the β -C-(1-6)-linked and β,β -C-(1-1)-linked systems.



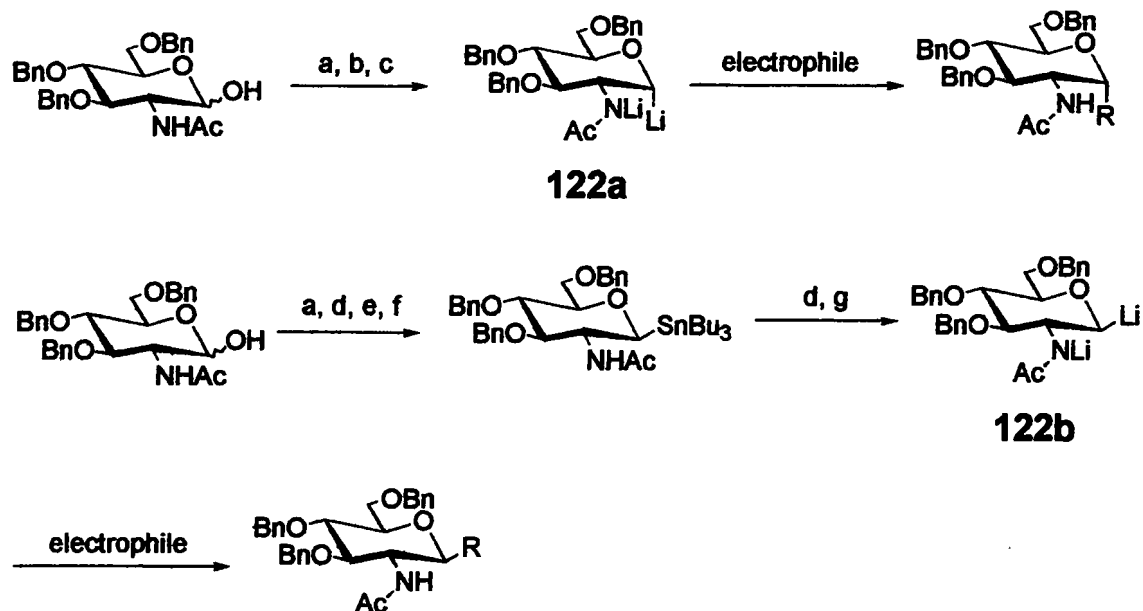
Scheme 3.7. Our nitrosugar approach to β -C-(1 $\rightarrow$ 4)-linked HA C-disaccharide.

disaccharides, an intramolecular radical coupling was proposed using C-1 radical (Scheme 3.8). The tethered sugar **126** is an ester from the condensation of phenylseleno aminosugar **98** and 4-*C*-methylene- β -D-xylo-hexopyranosiduronic acid **106a**. Sinaÿ et al. first used this tether strategy to make various C-disaccharides,²⁰ and it has also been successfully applied in O-glycosidic bond formation.²¹ The concept is to use a tether, with various lengths, to bring the two monosaccharide units close to each other so that the stereo- and regiochemistry of the disaccharide product can be controlled. More importantly, the tether enforces a radical addition to a non-activated double bond, and hence reduces the chance of competing with a hydrogen abstraction reaction (see section 5.3). Tethers have been used include methylketal,²² 1,1-dimethylketal,²³ silaketal,²⁴ α,α' -diether-*m*-xylene^{21b} and diester.^{21a} In our target C-disaccharide, an ester tether may well serve this purpose because the carboxylic acid of the GlcA unit can be established before the C-coupling. However, the C–O bond and the CO–C bond in an ester usually exist in the *s*-trans conformation,²⁵ and this might cause problems for a monoester tether (for example, **126**) in the nine-member ring-closing step. If that is the case, we might need to consider diester or other linkers to overcome this problem.

Another short route to the target is using C-1 nucleophiles. These include Kessler's 2-acetamido-2-deoxyglycosyl dianion²⁶ **122b** (Scheme 3.9) or Beau's phenyl 2-deoxy-2-phthalimidoglucopyranosyl sulfone C-1 anion²⁷ **119d** (Scheme 3.10). By varying lithiation conditions, it is anticipated that these anions could be obtained stereoselectively in either the α or β configuration. This would give us

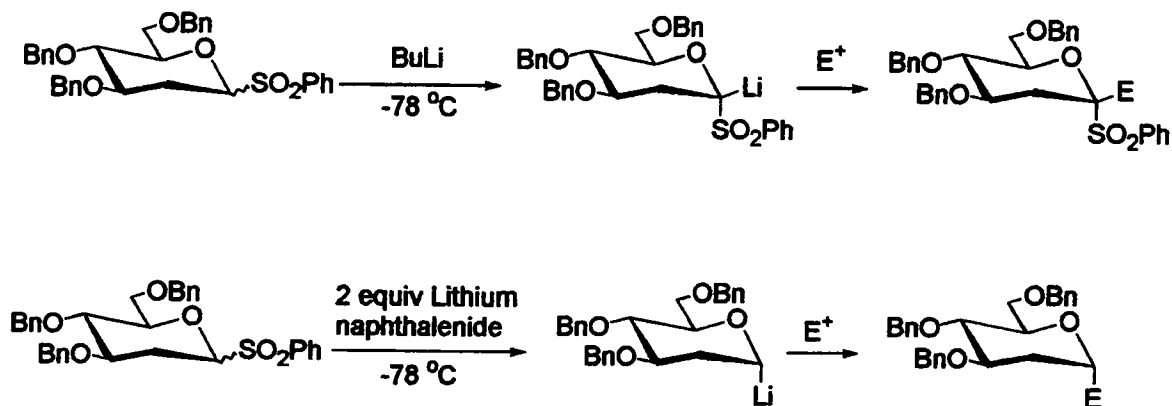


Scheme 3.8. Our intramolecular radical coupling approach to β -C-(1-4)-linked HA C-disaccharide.

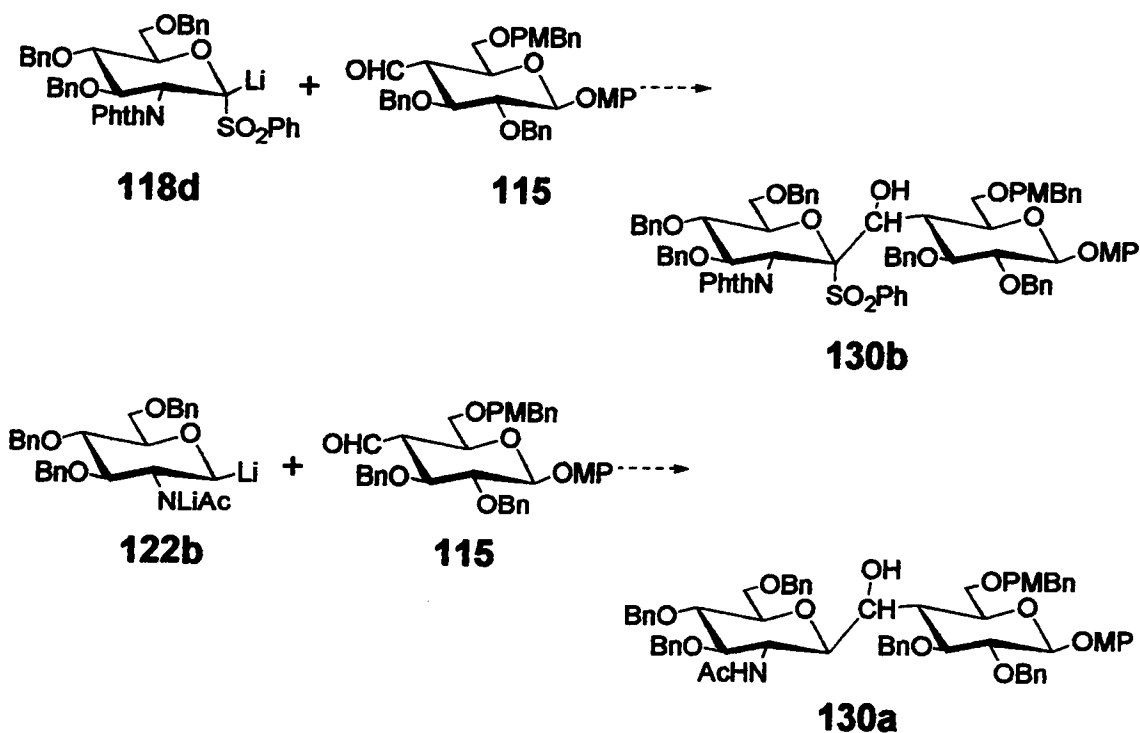


Keys: (a) SO_2Cl_2 ; (b) 1.0 equiv BuLi, -95°C ; (c) 2.1 equiv lithium naphthalenide, -95°C ; (d) 1.0 equiv BuLi, -78°C ; (e) 1.3 equiv Bu_3SnLi , -78°C ; (f) NH_4Cl ; (g) 1.0 to 1.5 equiv BuLi, -65°C .

Scheme 3.9. Kessler's stereoselective synthesis of 2-acetamido-2-deoxy-C-glucosides by using glycosyl dianions as key intermediates.



Scheme 3.10. Beau's stereoselective route to C-glycosides.



Scheme 3.11. Our C-1 nucleophile approaches to the HA C-disaccharide.

considerable advantage in the HA C-disaccharide synthesis since the β -C-glycosidic bond is desired (Scheme 3.11).

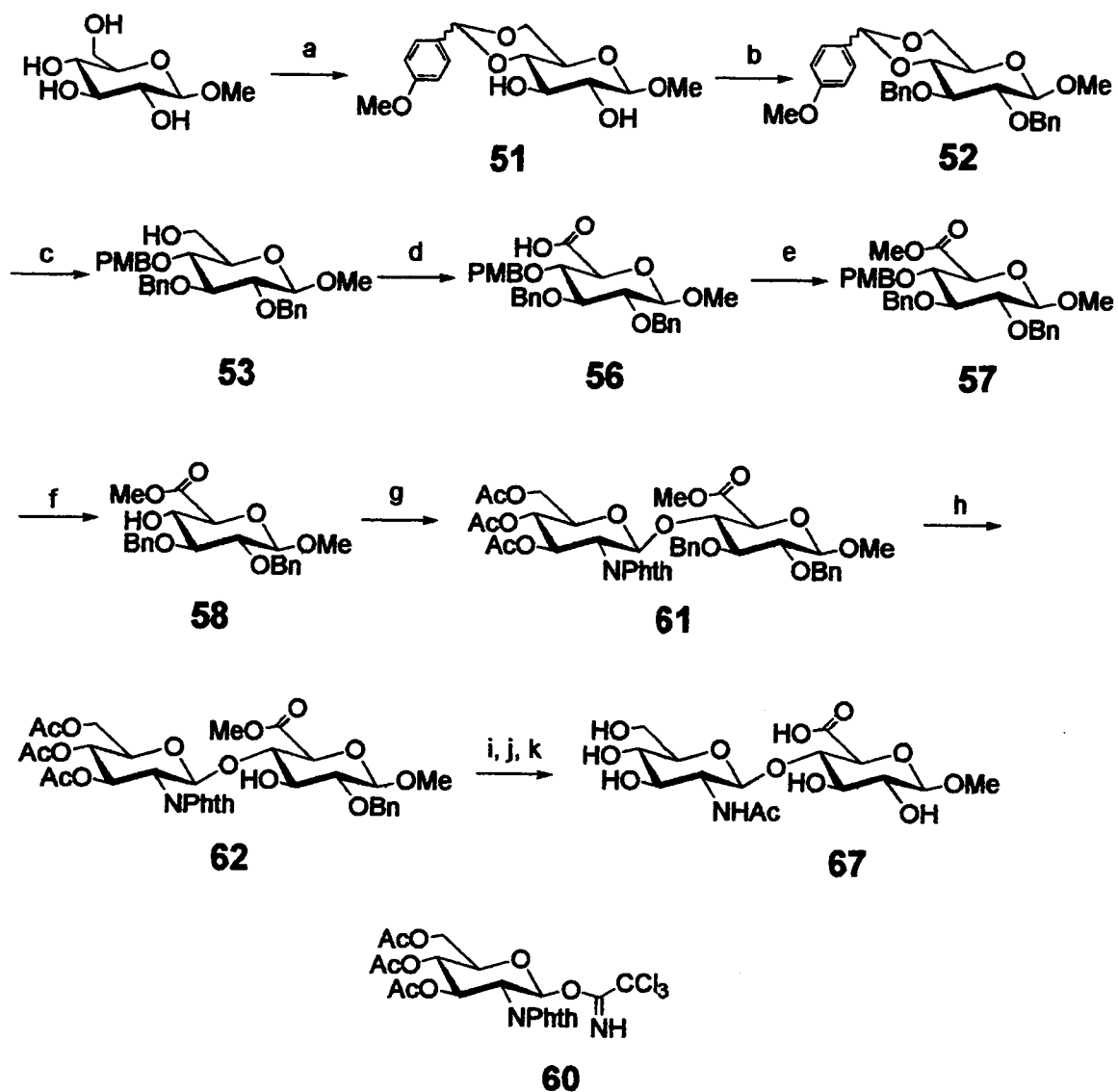
2.3. Significance

Synthesis of a (1 $\rightarrow$ 4)-linked HA disaccharide not only serves our immediate needs for the material for NMR and molecular modeling studies, but also provides useful information for the C-disaccharide synthesis. Synthesis of a (1 $\rightarrow$ 4)-linked HA C-disaccharide, which contains an *N*-acetylglucosamine unit at the nonreducing end and a glucuronic acid unit at the reducing end, is a challenging goal. The difficulty comes from the combination of *N*-acetylglucosamine unit and glucuronic acid unit in one molecule. Very few examples exist in literature for the synthesis of C-disaccharides containing a *N*-acetylglucosamine at the nonreducing end. The most closely related one is the synthesis of a (1 $\rightarrow$ 6)-linked α -C-disaccharide containing an *N*-acetylglucosamine unit at the nonreducing end by Anderson et al.²⁸ Furthermore, the C-6 oxidation has to be performed either before coupling, hence narrowing down the choice of the possible coupling reactions, or after the coupling, resulting in a less convergent synthesis.

3. RESULTS AND DISCUSSION

3.1. Synthesis of disaccharide β -D-GlcNAc-(1 $\rightarrow$ 4)- β -D-GlcA

The synthesis went exactly as planned (Scheme 3.12). Methyl α -D-glucopyranoside was heated with *p*-anisaldehyde dimethyl acetal and a catalytic



Keys: (a) $\text{TsOH} \cdot \text{H}_2\text{O}$, DMF, $p\text{-OMePhCH(OMe)}_2$, 65°C , 6 h; (b) NaH then BnBr, DMF, RT, 1 h; (c) NaCNBH₃, TMSCl, CH₃CN, 3A MS, 0°C to RT, 7 h; (d) TEMPO, aq NaClO (4%) Bu₄NBr, NaHCO₃, CH₂Cl₂-H₂O, pH 8-8.5, 0°C , 0.5 h; (e) CH₂N₂-ether, RT, 2 h; (f) CAN, RT, 5 min; (g) **60**, TMSOTf, 4A MS, CH₂ClCH₂Cl, 0°C to RT, 1 h; (h) Pd(OH)₂/C, H₂, RT, 50 psig, 21 h; (i) 1 N NaOH, 0°C to RT, 2 h; (j) NH₂NH₂, 85°C , 24 h; (k) Ac₂O, MeOH, 10 h.

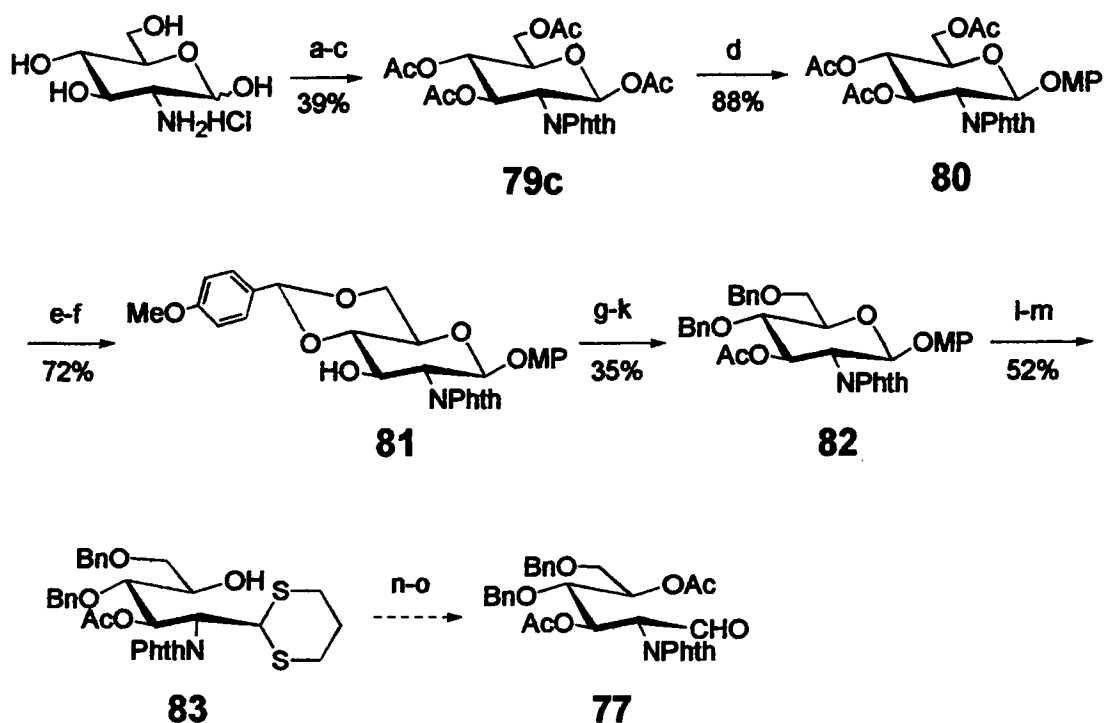
Scheme 3.12. Synthesis plan for HA disaccharide.

amount of *p*-toluenesulfonic acid in DMF to give the 4,6-protected sugar **51**.²⁹ The remaining hydroxyl groups were then protected by benzyl groups to give the fully protected sugar **52**. The benzylidene ring was then selectively opened³⁰ to give the 6-OH sugar **53**, which was oxidized by TEMPO/NaOCl³¹ and methylated by diazomethane to give methyl glucopyranosiduronate **57**. The PMB group was selectively removed by DDQ to give glycosyl acceptor **58**. The glycosyl donor **60** was prepared in three steps from the known 2-deoxy-2-phthalimido- α,β -D-glucosamine pentaacetate³². It was selectively deacetylated at the 1-position³³ and then converted to Schmidt's trichloroacetimidate **60**.³⁴ The coupling between **58** and **60** went smoothly to give disaccharide **61** (96%).¹⁴ Removal of the two benzyl groups took four days with 10% Pd-C in a Parr apparatus.¹³ An improvement was achieved by using Pearlman's catalyst [20% Pd(OH)₂-C], which took only 2 days under otherwise the same conditions. The resulting disaccharide was then hydrolyzed by NaOH, *N*-phthalimido deprotected by hydrazine, and *N*-acylated by acetic anhydride to give the final (1 $\rightarrow$ 4)-linked HA disaccharide **67**.¹³

3.2. Synthesis of HA C-disaccharide β -D-GlcNAc-CH₂-(1 $\rightarrow$ 4)- β -D-GlcA

3.2.1. Synthesis of the C-1 electrophile **77**

Preparation methods up to intermediate **82** was developed by Ken Price. The synthesis of **77** from **82** gave my co-worker a lot of trouble. Our original plan (Scheme 3.13) was to remove the *p*-methoxyphenyl group at the 1-position by CAN, open the sugar ring by Lewis acid, mask the resulting aldehyde by dithioacetal,

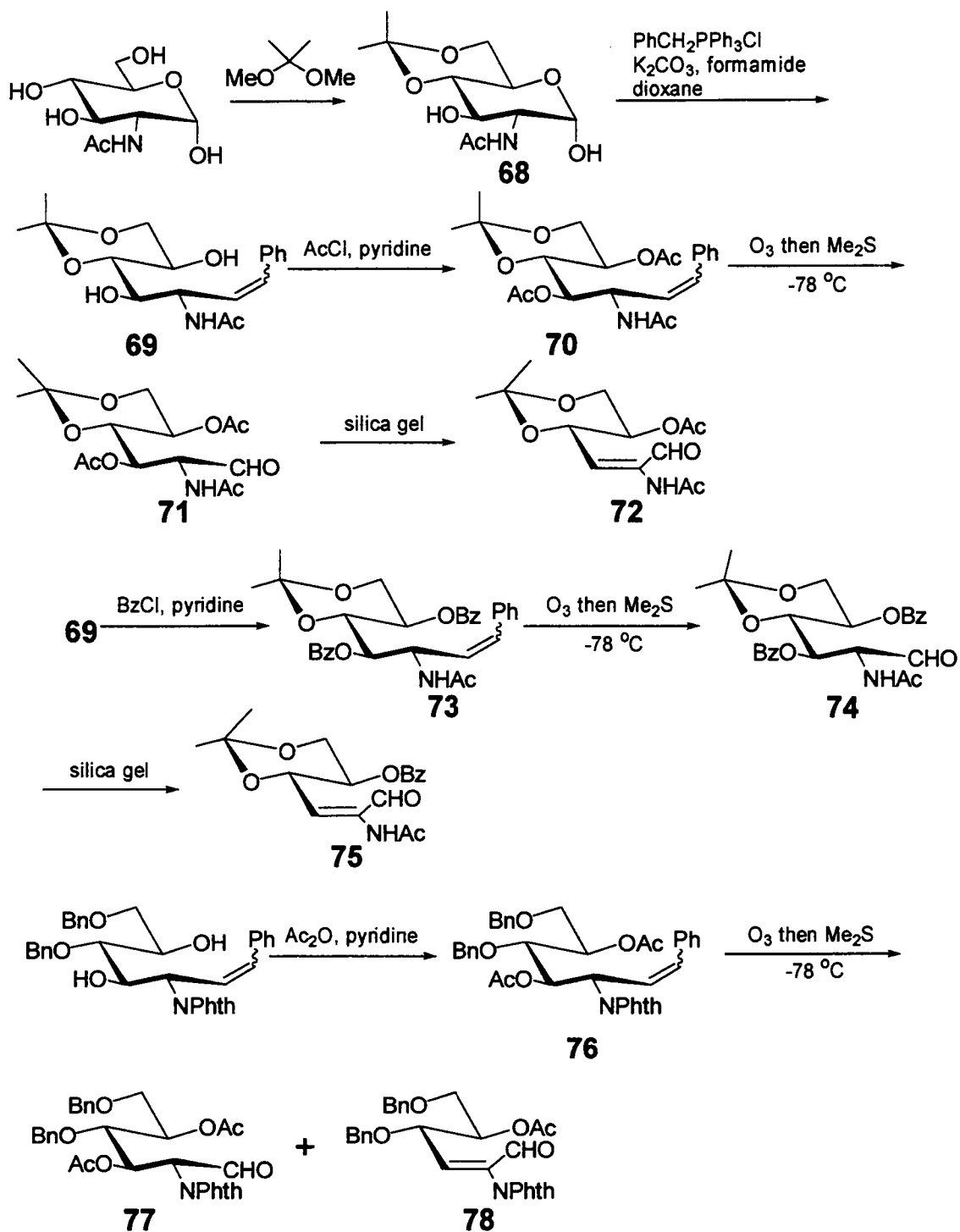


Keys: (a) Na, MeOH; (b) Phthalic anhydride, NEt_3 ; (c) Ac_2O , pyridine; (d) *p*-Methoxyphenol, TMSOTf, CH_2Cl_2 ; (e) NaOMe/MeOH; (f) Anisaldehyde dimethyl acetal, TsOH, DMF; (g) AlBr, NaH, DMF; (h) 88% formic acid; (i) BnBr, NaH, DMF; (j) $\text{Ir}[(\text{COD})(\text{PMePh}_2)_2]\text{PF}_6$ then I_2 , THF- H_2O -pyridine; (k) Ac_2O , pyridine; (l) CAN, $\text{CH}_3\text{CN-H}_2\text{O}$; (m) TiCl_4 , 1,3 propane dithiol, CH_2Cl_2 ; (n) Ac_2O , pyridine; (o) HgCl_2 , CdCO_3

Scheme 3.13. Original synthesis of the acyclic aldehyde sugar **77**.

protect the 5-OH as acetate, and deprotect the dithioacetal to give the aldehyde sugar **77**. Unfortunately, the removal of the dithioacetal **83** never gave detectable amount of **77**, which might due to the intrinsic instability of this α -phthalimido aldehyde. The conditions tried included $\text{HgCl}_2\text{--CdCO}_3$, $\text{HgCl}_2\text{--HgO}$ (yellow), HgClO_4 , $\text{BF}_3\text{Et}_2\text{O--HgO}$ (red)–aq THF³⁵, NBS³⁶, and bis-(trifluoroacetoxy)-iodobenzene.³⁷

The instability of 2-acylamino-2-deoxy-*aldehyde*-sugars in weakly basic solutions (for example, NaHCO_3) has been reported by Miljković et al.³⁶ In order to completely avoid acidic or basic conditions, an alternative method (Scheme 3.14) was pursued. Thus 2-acetamido-2-deoxy- α -D-glucose was reacted with 2,2-dimethoxy propane and a catalytic amount of *p*-toluenesulfonic acid to give the 4,6-protected sugar **68**. It was then refluxed with benzyltriphenylphosphonium chloride, potassium carbonate, and a catalytic amount of formamide in dioxane to give the expected Wittig olefin **69**.³⁸ Protection of the two free hydroxy groups by acetyl or benzoyl groups gave **70** or **73**, respectively. Subsequent ozonolysis of the olefins gave aldehydes **71** or **74**. They are relatively stable in CH_2Cl_2 or CHCl_3 solution at room temperature. However, when they were in contact with silica gel, considerable decomposition was detected due to the formation of the conjugated aldehydes **72** or **75** by β -elimination. The same method was used to prepare **77**. It turned out that **77** underwent β -elimination much faster than either **71** or **74**. A similar observation was also found in Martin's study:^{19b} 5-O-Acetyl-2,3,4,6-tetra-O-benzyl-*aldehyde*-D-glucose underwent extensive β -elimination under the mildly



Scheme 3.14. Modified approach to the acyclic aldehyde sugars **71**, **74** and **77**.

basic (KF) coupling conditions. However, with O-2 and O-3 incorporated into an isopropylidene ring, the *aldehydo*-hexose was much more resistant to β -elimination. Because we were not able to obtain pure **77**, **71** was used in the trial of the nitro-aldol coupling reaction.

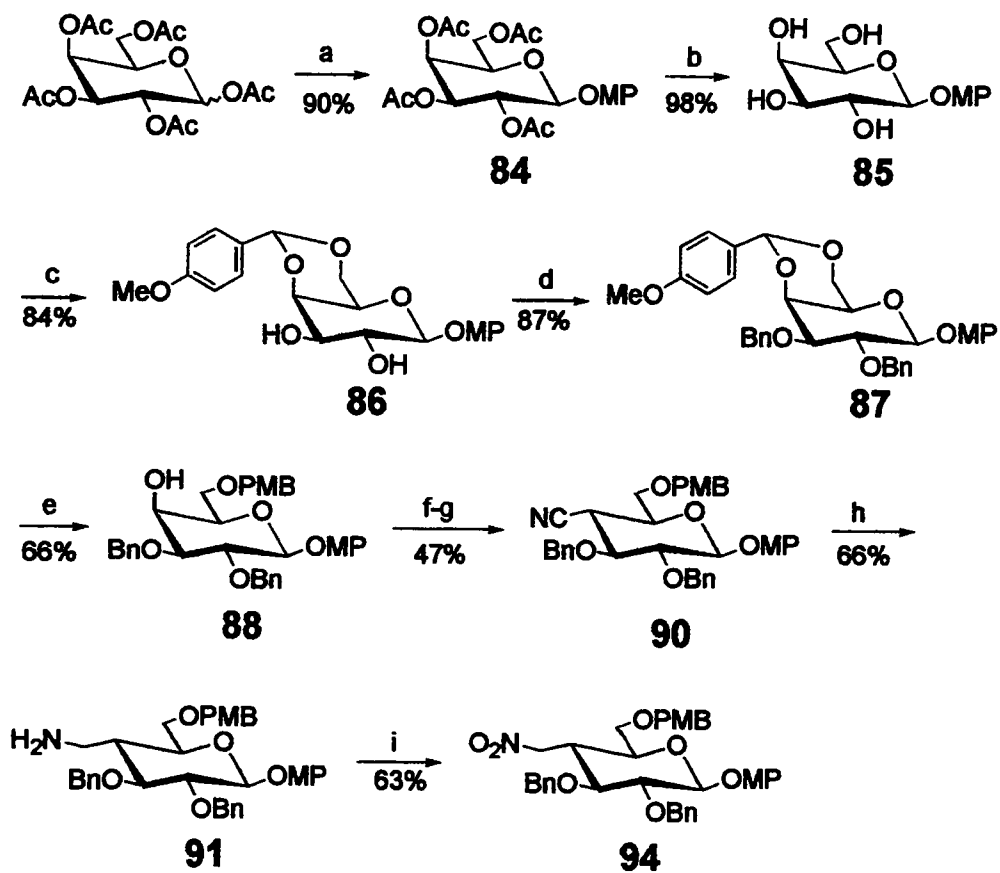
3.2.2. Synthesis of the nitrosugar **94** (Scheme 3.15).

The crucial step of converting the *galacto*-configuration in **88** to the *gluco*-configuration in **90** was developed by Ken Price using S_N2 chemistry. Compound **88** was converted to its triflate in quantitative yield, then the triflate was displaced by tetrabutylammonium cyanide in yields ranging from 20% to 50%. The main byproducts were presumably E2 elimination products, which are slightly less polar than the substitution product on TLC. In order to obtain more reproducible yields from this reaction, an experiment closely monitored by TLC (12:4:4:2:1 hexanes–toluene–methylene chloride–ethyl acetate–acetonitrile) was conducted from $-45\text{ }^{\circ}\text{C}$ to room temperature, and it showed that the substitution occurred at $-15\text{ }^{\circ}\text{C}$, and the elimination became significant at $0\text{ }^{\circ}\text{C}$. By controlling the temperature closely within this range, a reproducible yield of 50% was obtained.

An attempt to prepare **90** from ulose **100a** using tosylmethylisocyanide (TosMIC)³⁹ gave no desired product.

LAH reduction⁴⁰ of **90** gave the 4-*C*-methylamine sugar **91**, which was oxidized by dimethyldioxirane⁴¹ or *m*-CPBA⁴² to give the desired nitrosugar **94**.

3.2.3. Attempted coupling of the *aldehydo* sugar **71** with the nitrosugar **94** (Scheme 3.16)



Keys: (a) *p*-Methoxyphenol, TMSOTf, CH₂Cl₂; (b) NaOMe/MeOH; (c) Anisaldehyde dimethylacetal, TsOH, DMF; (d) BnBr, NaH, DMF; (e) NaBH₃CN, TFA, DMF, 0 °C to rt; (f) Tf₂O, pyridine, CH₂Cl₂, -10 to 0 °C; (g) Bu₄NCN, THF, -10 to 0 °C; (h) LiAlH₄, ether, rt; (i) dimethyldioxirane, acetone, rt.

Scheme 3.15. Synthesis of nitrosugar **94**.

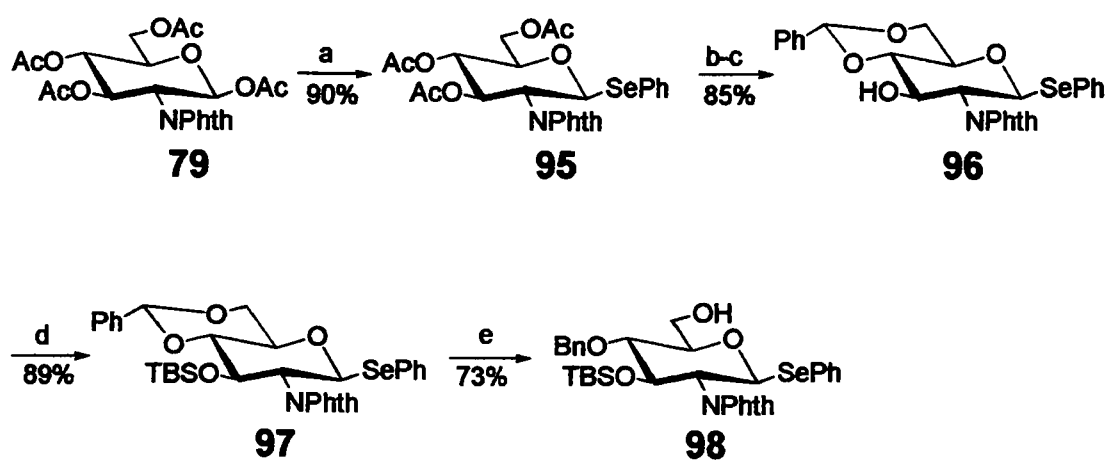
The KF-promoted nitro-aldol reaction of **71** with **94** was tried by Martin's procedure, but the nitrosugar **94** was not consumed, even after two days reaction time. Similarly, attempts to react either 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido- β -D-glucose or 2-acetamido-2-deoxy-4,6-*O*-isopropylidene- α -D-glucose **68** with the nitromethane anion⁴³ gave no coupling product. These might indicate that the β -*N*-acetamido and β -*N*-phthalimido aldehydes are less reactive compared with a β -alkoxyaldehyde, possibly due to β -participation. The incompatibility⁴⁴ between the *N*-protection groups and the nucleophile forced us to seek other coupling possibilities.

3.2.4. Synthesis of the phenylseleno sugar **98**

The phenylseleno sugar **98** was synthesized by Karen Welch, one of my co-workers (Scheme 3.17). Compound **79c** was treated with phenylselenol and trifluoroboron etherate to give phenylseleno sugar **95**. The three acetyl groups were removed by sodium methoxide, and the resulting triol was reacted with benzaldehyde and a catalytic amount of *p*-toluenesulfonic acid to give the 4,6-benzylidene sugar **96**. The remaining hydroxy group was protected by TBS, and the 4,6-*O*-benzylidene group was selectively opened by borane-THF⁴⁵ and TMSOTf to give the 6-OH phenylseleno sugar **98**.

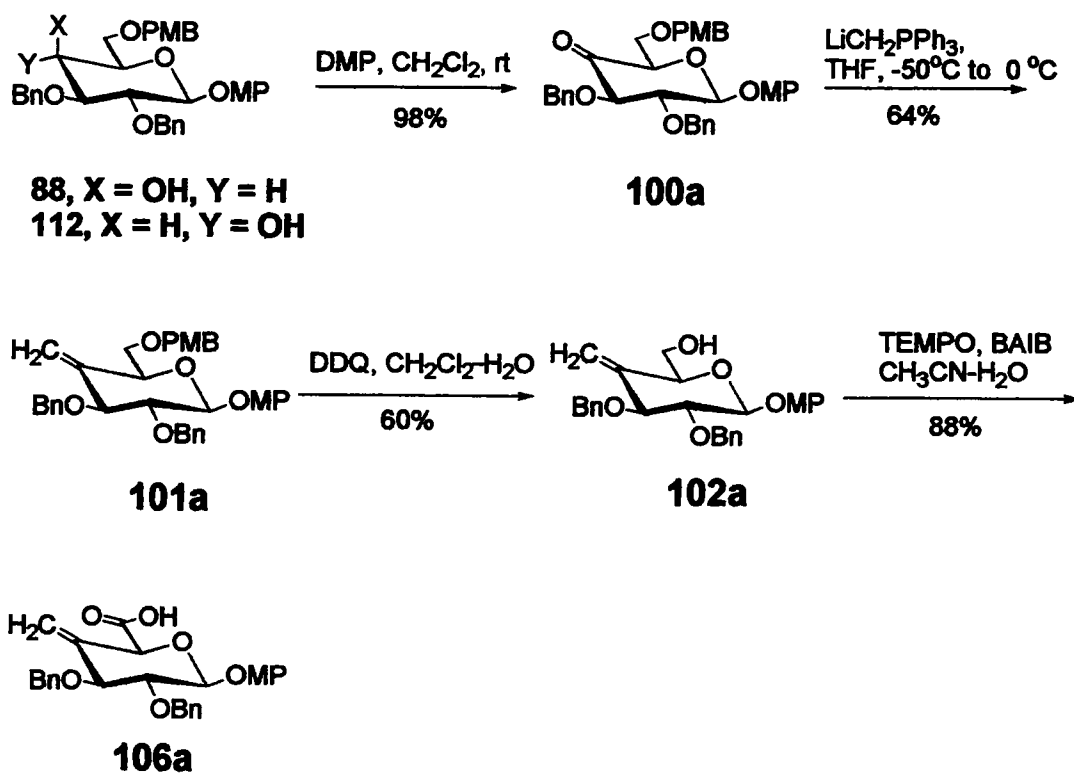
3.2.5. Synthesis of the 4-*C*-methylene-4-deoxy-xylo-hexopyranosid-uronic acid **106a**

The 4-*C*-methylene-4-deoxy-xylo-hexopyranosiduronic acid **106a** was prepared from **88** or **112** (Scheme 3.18). Oxidation of the 4-OH by PCC, Swern and



Keys: (a) PhSeH, $\text{BF}_3\cdot\text{OEt}_2$; (b) NaOMe/MeOH; (c) Benzaldehyde dimethyl acetal, TsOH, DMF; (d) TBSCl, imidazole, DMAP, DMF; (e) $\text{BH}_3\cdot\text{THF}$, TMSOTf.

Scheme 3.17. Synthesis of phenyl 1-seleno sugar **98**.

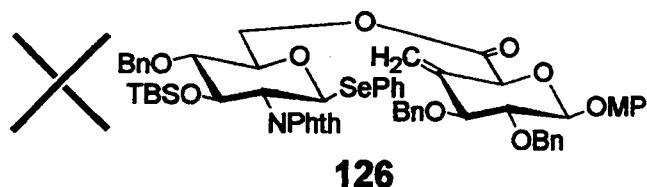
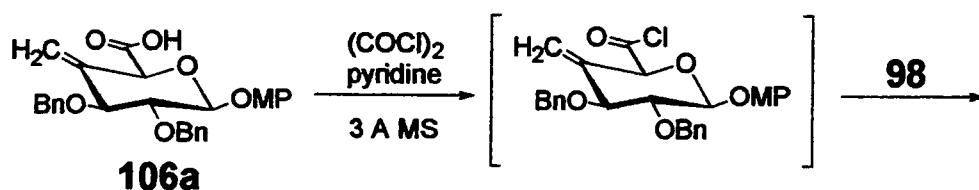
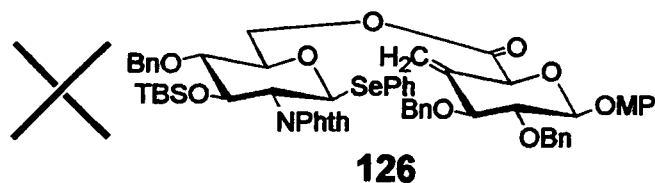
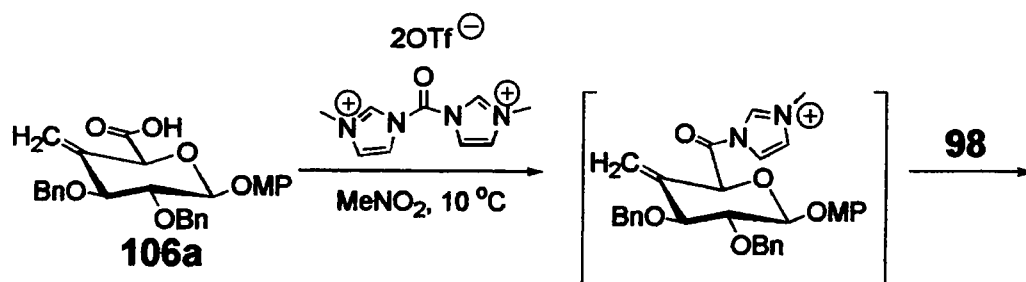
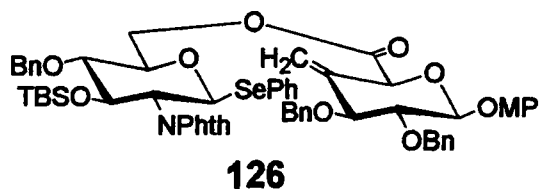
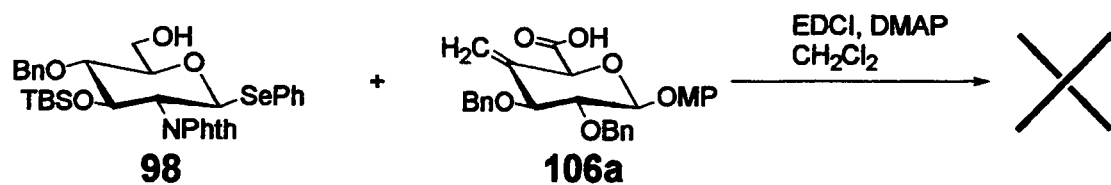


Scheme 3.18. Synthesis of the 4-deoxy-4-C-methylene-xyloronic acid **106a**.

Dess–Martin⁴⁶ methods were tried. By using PCC, the equatorial hydroxyl group in **112** cannot be oxidized while the axial hydroxyl group in **88** can be oxidized cleanly in 79% yield. Alternatively, both **88** and **112** can be oxidized cleanly either by the Swern oxidation or by the Dess–Martin method. The Dess–Martin oxidation has one advantage, i.e., it requires simpler reaction conditions (air is tolerated and moisture is even beneficial^{46c}) and has easier workup (CH₂Cl₂ evaporation). The resulting ulose **100a** was reacted with a methylene ylide⁴⁷ to give 4-C-methylene-*gluco*-hexopyranoside **101a**, which was selectively deprotected at the 6-position to give **102a**. Several attempts have been tried to oxidize the primary hydroxyl group in the presence of a double bond. These include TPAP oxidation,⁴⁸ PDC in DMF, sequential oxidation by Dess–Martin and NaClO₂/2-methyl-2-butene,⁴⁹ and TEMPO/NaOCl. None of these gave a satisfactory yield. Very recently, TEMPO/BAIB⁵⁰ was reported in the preparation of nucleoside-5'-carboxylic acids. This method was found to give the desired oxidation product **106a** in decent yield (88%).

3.2.6. Attempted coupling of **98** with **106a** (Scheme 3.19)

After having both coupling partners **98** and **106a** in hand, we have made several attempts to make the ester **126** (Scheme 3.19). Both EDCI/DMAP/CH₂Cl₂ and 1,1'-carbonylbis(3-methylimidazolium) bis(trifluoromethanesulfonate) were tried, but they all led to the decomposition of the **106a**. We also tried to react **106a** with (COCl)₂/pyridine⁵¹ to make the acyl chloride *in situ* and trapped it by **98**. This again resulted in the decomposition of both partners. It seems like these processes

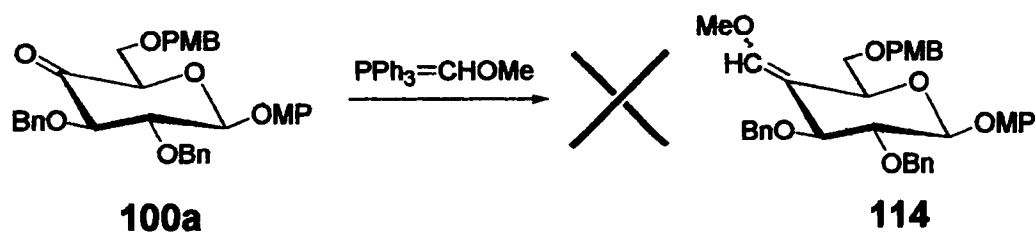


Scheme 3.19. Attempted coupling of **98** with **106a** under various carboxy group activation conditions.

involving carboxy group activation⁵² are not compatible with the double bond presented in **106a**. Further experiments need to be done to assess other methods, for example, the hydroxy group activation, in this ester formation.

3.2.7. Synthesis of 4-deoxy-4-C-formyl sugar **115**

4-Deoxy-4-C-formyl sugars could be synthesized from 4-uloses in four steps (Wittig olefination, hydroboration–oxidation, oxidation of the primary alcohols to aldehydes, and epimerization of the axial aldehyde to the equatorial aldehyde).⁵³ In our lab, **115** was first made by Ken Price through DIBAL reduction of the cyano sugar **90**. It was later isolated as a byproduct from the partial LAH reduction of **90** in the preparation of aminosugar **91**. Since the 4-ulose **100a** could be obtained in large quantity, the carbonyl homologation of **100a** with methoxymethylenetriphenylphosphorane was tried (Scheme 3.20). Unfortunately, this led to the decomposition of the 4-ulose without forming any product.



Scheme 3.20. Attempted carbonyl homologation by a Wittig reaction

4. CONCLUSIONS

With respect to the objectives set forth in section 2.1, the unprotected HA disaccharide β -GlcNAc-(1 $\rightarrow$ 4)-GlcA was synthesized. This provided not only the material for the NMR and molecular modeling studies, but also useful experience for the C-disaccharide synthesis. As for the HA C-disaccharide synthesis, key coupling partners *aldehydo* sugar **77**, nitrosugar **94**, 4-deoxy-4-C-methylene-xylo-hexopyranosiduronic acid **106a**, and 4-deoxy-4-C-formyl-glucopyranoside **115** were synthesized. Nitroaldol reaction between **77** and **94**, as well as tether formation between **106a** and **98**, were tried, but both gave negative results. Consequently, the objective of synthesis of the HA C-disaccharide was not achieved during the time-frame of this study. However, the dianion coupling between **122** and **115**, as well as the sulfone anion coupling between **118d** and **115**, were achieved by co-workers, and these developments made the possibility of obtaining the unprotected HA C-disaccharide and mixed CH_2O oligosaccharides in the near future look promising.

5. EXPERIMENTAL

5.1. General

All reactions were monitored by thin-layer chromatography (TLC). Adsorption chromatography was carried out using E. Merck silica gel products: (a) TLC on 0.2 mm aluminum-backed plates, (b) flash chromatography using 230–400 mesh silica gel. The TLC plates were visualized by spray–heat development using

a *p*-anisaldehyde–sulfuric acid reagent. All reagents were purchased from Aldrich Chemical Co. Anhydrous solvents were prepared as follows: CH₂Cl₂, pyridine, and toluene were distilled from CaH₂; DMF was distilled at reduced pressure from CaH₂; THF was refluxed with Na–benzophenone ketyl and distilled. All reactions were carried out under a nitrogen atmosphere unless otherwise indicated. Elemental analyses were furnished by Atlantic Microlab, Inc. of Atlanta, GA. ¹H and ¹³C NMR spectra were obtained by using a Bruker AC 250 spectrometer (250 MHz and 62.5 MHz, respectively) with CDCl₃ as the solvent and TMS as an internal standard. ¹H NMR chemical shifts are reported as δ (ppm) downfield from the TMS signal; multiplicities are first-order values in Hz: s, singlet; bs, broad singlet; d, doublet; t, triplet; pt, pseudo triplet; q, quartet; dd, doublet of doublet; m, multiplet. ¹³C NMR chemical shifts are reported as δ (ppm) relative to CDCl₃ (77.00 ppm) as an internal standard. Optical rotations were measured with a Perkin–Elmer Model 243 automatic polarimeter equipped with sodium and mercury lamps for solutions in a 1-dm cell at the indicated temperature. The mass spectral analyses were obtained on a Micromass VG Quattro II electrospray instrument at the University of Tennessee. IR were recorded with a Bomem instrument by preparing samples as neat oil films.

5.2. Synthesis of compounds 51–115

Preparation of methyl 4,6-*O*-*p*-methoxybenzylidene-β-D-glucopyranoside (51).¹³ Methyl β-D-glucopyranoside (14.88 g, 76.6 mmol), *p*-toluenesulfonic acid

monohydrate (4.21 g, 0.29 equiv) and *p*-anisaldehyde dimethyl acetal (15 mL, 1.15 equiv) were dissolved in DMF (115 mL), and the flask was put onto a rotary evaporator at ~20 torr. The reaction was allowed to run at 65 °C for 6 h, at the end of which time TLC (9:1 CH₂Cl₂–CH₃OH) showed that it was complete. TEA was added, and the DMF was removed by coevaporating with toluene. The crude product was purified by flash column chromatography (5% CH₃OH–CH₂Cl₂) to give a white solid (22.0 g, 92%). The NMR data were in agreement with those in the literature.¹³ ¹³C NMR (DMSO-*d*₆): δ 159.52, 130.19, 127.65, 113.28, 104.50 (C1), 100.63 (CHAr), 80.59, 74.26, 72.83, 67.92 (C6), 65.79, 58.36, 55.09.

Preparation of **methyl 2,3-di-*O*-benzyl-4,6-*O*-*p*-methoxybenzylidene-β-D-glucopyranoside (52)**.¹³ To a suspension of NaH (60 %, 3.77 g, 2.6 equiv) in 50 mL of dry DMF was added dropwise at room temperature a solution of **51** (11.38 g, 36.4 mmol) in DMF (90 mL). After 15 min, BnBr (9.1 mL, 2.1 equiv) was added dropwise, and the reaction was stirred overnight at room temperature. Water was added dropwise to quench the reaction and a white solid precipitated. After filtration, the solid was redissolved in CH₂Cl₂ and dried over MgSO₄. Solvent evaporation gave **52** as an off-white solid (17.1 g, 95%). The NMR data were in agreement with those in the literature.¹³ ¹H NMR (CDCl₃): δ 6.88, 6.92, 7.39, 7.43 (AB, 4H, *J* = 10.0 Hz, ArOMe), 7.25–7.37 (m, 10H, Ar), 5.53 (s, 1H, ArCH), 4.73, 4.76, 4.89, 4.92 (AB, 2H, *J* = 7.5 Hz, CH₂Ph), 4.77, 4.81, 4.85, 4.88 (AB, 2H, *J* = 7.5 Hz, CH₂Ph), 4.42 (d, 1H, *J* = 7.5 Hz, H1), 4.34 (dd, 1H, *J* = 10.0, 5.0 Hz, H4), 3.81

(s, 3H, ArOMe), 3.62–3.77 (m, 3H, H6a, H6b, H3), 3.58 (s, 1H, OMe), 3.39–3.47 (m, 2H, H2, H5); ^{13}C NMR (CDCl_3): δ 160.01, 138.52, 138.41, 129.83, 128.30, 128.02, 127.77, 127.65, 127.59, 127.31, 113.58, 105.20 (C1), 101.12 ($\underline{\text{CHAr}}$), 82.17, 81.46, 80.81, 77.52 (CH_2Ph), 77.01 (CH_2Ph), 68.74 (C6), 65.98, 57.43, 55.29.

Preparation of methyl 2,3-di-O-benzyl-4-O-*p*-methoxybenzyl- β -D-glucopyranoside (53).⁵⁴ To a solution of **52** (11.30 g, 23.0 mmol) and NaCNBH_3 (8.65 g, 6 equiv) in dry acetonitrile (500 mL) maintained over 3 Å molecular sieves (19.5 g) in an ice-bath was added ice-cooled TMSCl (18 mL, 6.2 equiv) dropwise over a period of 25 min. After the addition was completed, the ice-bath was removed, and the reaction was allowed to progress at room temperature for 7 h. The resulting mixture was then filtered through Celite. The filtrate was washed by satd NaHCO_3 , extracted with CH_2Cl_2 , and the organic extract was dried over MgSO_4 , and concentrated. The crude product was purified by column chromatography (3:1 hexanes–ethyl acetate) to give **53** (6.93 g, 61%), **54** (1.21 g, 11%) and **55** (1.50 g), respectively. The physical data for **53** was as follows: ^1H NMR (CDCl_3): δ 7.25–7.32 (m, 10H, Ar), 6.82, 6.85, 7.18, 7.21 (AB, 4H, $J = 7.5$ Hz), 4.55–4.95 (m, 6H), 4.34 (d, 1H, $J = 7.5$ Hz, H1), 3.76–3.89 (m, 1H, H6a), 3.76 (s, 3H, ArOMe), 3.57–3.74 (m, 3H), 3.56 (s, 3H, OMe), 3.36–3.42 (m, 2H); ^{13}C NMR (CDCl_3): δ 159.34, 138.55, 138.40, 130.10, 129.70, 128.30, 127.98, 127.74, 127.58, 127.54, 113.65, 104.75 (C1), 84.40, 82.31, 77.23, 75.57 (CH_2Ph), 74.99, 74.73 (CH_2Ph), 74.67 (CH_2Ph), 61.93 (C6), 57.20, 55.18. The NMR data were in

agreement with those in the literature.⁵⁴

Preparation of methyl 2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl- β -D-glucopyranoside (**54**). Compound **54** was isolated as a byproduct in the preparation of compound **53**. The NMR data were in agreement with those in the literature.¹³ ¹H NMR (CDCl₃): δ 7.22–7.33 (m, 12H, Ar), 6.83–6.86 (pd, 2H, Ar), 4.66, 4.71, 4.89, 4.93 (AB, 2H, J = 10.0 Hz, CH₂Ph), 4.71, 4.76, 4.89, 4.93 (AB, 2H, J = 10.0 Hz, CH₂Ph), 4.49 (s, 2H, CH₂ArOMe), 4.30 (d, 1H, J = 6.0 Hz, H1), 3.74 (s, 3H, ArOMe), 3.57–3.72 (m, 3H, H6a, H6b, H4), 3.55 (s, 3H, OMe), 3.33–3.50 (m, 3H, H5, H2, H3); ¹³C NMR (CDCl₃): δ 159.11, 138.55, 138.38, 129.86, 129.16, 128.28, 128.16, 127.89, 127.73, 127.55, 127.46, 113.66, 104.57 (C1), 83.88, 81.61, 75.03 (CH₂Ph), 74.46 (CH₂Ph), 73.99, 73.12 (CH₂Ph), 71.46, 69.78 (C6), 56.89, 55.04.

Methyl (methyl 2,3-di-*O*-benzyl-4-*O*-*p*-methoxybenzyl- β -D-glucopyranosid)uronate (**57**).³¹ To a biphasic system of **53** (7.47 g, 15.1 mmol), TEMPO (36 mg, 0.015 equiv), satd NaHCO₃ (36 mL), Bu₄NBr (0.49 g, 0.1 equiv) and CH₂Cl₂ (50 mL) in an ice bath was added a mixture of bleach solution (5% NaClO, 75 mL), satd NaHCO₃ (18 mL) and satd NaCl (36 mL) dropwise via a dropping funnel. The pH of the reaction mixture was maintained within 7.70–8.40 by addition of 10% NaOH. The reaction was allowed to progress at 0 °C for 30 min. Then it was quenched by adding 1 N HCl until pH = 6.6, extracted with ethyl

acetate, and the organic extract was dried over MgSO_4 and concentrated to give **56** as a yellow syrup (6.85 g, 89%). The crude product was used in the next step without further purification. To an ether (10 mL) solution of **56** (2.53 g, 4.97 mmol) was added a solution of diazomethane [prepared by mixing of 40% NaOH (15 mL, 150 mmol), ether (50 mL) and nitrosomethylurea (5 g, 48.5 mmol) at 0 °C] until the solution stopped bubbling and a yellow tint persisted. The resulting solution was stirred for 20 min before acetic acid was added to react with the excess CH_2N_2 . The mixture was then concentrated, and the crude product was purified by column chromatography (2:1 hexanes–ethyl acetate) to give **57** as white crystals (2.19 g, 84%). ^1H NMR (CDCl_3): δ 7.29 (bs, 10H, Ar), 6.78, 6.82, 7.12, 7.16 (AB, 4H, ArOMe), 4.50–4.91 (m, 6H), 4.34 (d, 1H, $J = 7.5$ Hz, H1), 3.61–3.93 (m, 9H), 3.42–3.58 (m, 4H); ^{13}C NMR (CDCl_3): δ 168.75, 159.04, 138.21, 138.10, 129.71, 129.34, 128.01, 127.71, 127.46, 127.31, 113.45, 104.65 (C1), 83.43, 81.46, 78.73, 75.25 (CH_2Ph), 74.34 (CH_2Ph), 74.13, 56.93, 54.80, 52.04.

Preparation of methyl (methyl 2,3-di-O-benzyl- β -D-glucopyranosid)-uronate (58).^{14a} To a solution of **57** (2.63 g, 5.03 mmol) in a 4:1 acetonitrile–water (50 mL) was added CAN (13.8 g, 5 equiv) at room temperature. After 5 min, satd NaHCO_3 was added to precipitate the cerium salt. The suspension was filtered through Celite, and the clear, light-yellow filtrate was extracted with ethyl acetate. The organic extract was dried over MgSO_4 and concentrated. The crude product was purified by column chromatography (3:1 hexanes–ethyl acetate) to give **58** as

a colorless syrup (2.0 g, 99%). The NMR data were in agreement with those in the literature.^{14a} ¹H NMR (CDCl₃): δ 7.23–7.28 (m, 10H, Ar), 4.63–4.88 (m, 4H), 4.31 (d, 1H, *J* = 7.5 Hz, H1), 3.75–3.85 (m, 2H), 3.70 (s, 3H), 3.37–3.50 (m, 6H); ¹³C NMR (CDCl₃): δ 169.28, 138.23, 138.02, 127.93, 127.90, 127.61, 127.43, 127.22, 104.49 (C1), 82.78, 80.79, 74.79 (CH₂Ph), 74.24 (CH₂Ph), 74.12, 71.32, 56.85, 52.14.

Preparation of **3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-β-D-glucose (59)**.³³ To a solution of **80** (5.23 g, 9.66 mmol) in 4:1 acetonitrile–water (50 mL) was added CAN (26.5 g, 5 equiv). After 10 min, satd NaHCO₃ was added to precipitate the cesium salt. The suspension was filtered through Celite, and the clear, light-yellow filtrate was extracted with ethyl acetate. The organic extract was dried over MgSO₄ and concentrated. The crude product was purified by column chromatography (1:1 hexanes–ethyl acetate) to give **59** as a brown solid (3.63 g, 86%). ¹H NMR (CDCl₃): δ 7.72–7.88 (m, 4H, Phth), 5.86 (dd, 1H, *J* = 10.5, 9.2 Hz), 5.64 (d, 1H, *J* = 8.4 Hz), 5.18 (pt, 1H, *J* = 9.6 Hz), 4.22–4.32 (m, 4H), 3.90–4.00 (m, 1H, H6a), 2.12 (s, 3H, OAc), 2.04 (s, 3H, OAc), 1.87 (s, 3H, OAc); ¹³C NMR (CDCl₃): δ 170.78, 170.10, 169.55, 167.80, 134.38, 131.40, 123.67, 92.66 (C1), 72.11, 70.49, 68.93, 62.07 (C6), 56.09 (C2), 20.79, 20.64, 20.49.

Preparation of **3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl trichloroacetimidate (60)**.⁵⁵ To a solution of **59** (3.63 g, 8.33

mmol) and trichloroacetonitrile (2.51 mL, 3 equiv) in dry CH_2Cl_2 (10 mL) was added K_2CO_3 (1.93 g, 1.68 equiv). After 4 h, the suspension was filtered through Celite. The filtrate was concentrated, and the crude product was purified by column chromatography (3:2 hexanes–ethyl acetate) to give **60** as a white glassy solid (3.23 g, 67%). ^1H NMR (CDCl_3): δ 8.68 (s, 1H, NH), 7.72–7.86 (m, 4H, Phth), 6.63 (d, 1H, J = 8.9 Hz), 5.93 (dd, 1H, J = 10.4, 9.3 Hz), 5.28 (pt, 1H, J = 9.6 Hz), 4.64 (dd, 1H, J = 10.5, 9.1 Hz), 4.37–4.44 (m, 1H, H6a), 4.06–4.24 (m, 3H), 2.12 (s, 3H, OAc), 2.05 (s, 3H, OAc), 1.90 (s, 3H, OAc); ^{13}C NMR (CDCl_3): δ 170.62, 170.01, 169.37, 167.33, 160.47, 134.41, 131.14, 123.63, 93.52 (C1), 90.07 (CCl_3), 72.74, 70.36, 68.38, 61.48 (C6), 53.50 (C2), 20.68, 20.55, 20.37.

Preparation of methyl [(3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido- β -D-glucopyranosyl)-(1→4)-(methyl 2,3-di-*O*-benzyl- β -D-glucopyranosid)]uronate (**61**).¹³ A solution of **60** (1.42 g, 1.18 equiv) and **58** (0.84 g, 2.07 mmol) in dry $\text{CH}_2\text{ClCH}_2\text{Cl}$ (20 mL) maintained over powdered 4 Å molecular sieves (2.2 g) was stirred at room temperature for 1 h. The solution was then cooled by an ice-bath. To this was added TMSOTf (0.08 mL, 0.2 equiv). After 1.5 h the reaction was complete. TEA was then added to the brownish-red solution. The resulting light-yellow solution was filtered through a short bed of silica gel. The filtrate was concentrated, and the crude product was purified by column chromatography (2:1 hexanes–ethyl acetate) to give disaccharide **61** as a white glassy solid (1.63 g, 96%). The NMR data were in agreement with those in the literature.¹³ ^1H NMR

(CDCl₃): δ 7.85 (bs, 2H, Phth), 7.72–7.77 (m, 2H, Phth), 7.18–7.40 (m, 10H, Ar), 5.77 (pt, 1H, J = 10.0 Hz, N3), 5.56 (d, 1H, 8.4 Hz, N1), 5.11 (pt, J = 9.6 Hz, N4), 4.85, 4.90, 4.97, 5.01 (AB, 2H, J = 11.8 Hz, CH₂Ph), 4.55, 4.59, 4.75, 4.79 (AB, 2H, J = 10.0 Hz, CH₂Ph), 4.25 (d, 1H, J = 7.5 Hz, U1), 4.08–4.19 (m, 3H, N2, N6a, U4), 3.88 (d, 1H, J = 12.5 Hz, N6b), 3.69 (d, 1H, J = 10 Hz, U5), 3.54–3.65 (m, 5H, N5, U3, CO₂Me), 3.35–3.41 (m, 4H, U2, OMe), 1.95 (s, 6H, 2Ac), 1.80 (s, 3H, Ac); ¹³C NMR (CDCl₃): δ 170.1, 169.6, 169.0, 167.9, 138.6, 137.8, 133.9, 131.3 (br), 127.9, 127.6, 127.2, 126.9, 126.4, 123.0, 104.4 (U1), 97.2 (N1), 81.7, 80.9, 77.2, 74.2 (CH₂Ph), 73.7 (CH₂Ph), 71.3, 70.2, 68.2, 61.1 (N6), 56.8 (CO₂CH₃), 54.7 (OMe), 52.2 (N2), 20.2, 20.0.

Preparation of methyl [(3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido- β -D-glucopyranosyl)-(1 $\rightarrow$ 4)-(methyl β -D-glucopyranosid)]uronate (62).¹³ To a hydrogenolysis flask was added **61** (1.00g 1.22 mmol), Pearlman's catalyst [20% Pd(OH)₂ on carbon, 0.4 g], ethyl acetate (10 mL) and 95% ethanol (20 mL). The reaction was stirred at room temperature for 21 h under a hydrogen-pressurized (51.5~56.5 psig) environment. After releasing the pressure, the black suspension was filtered through a short bed of silica gel. The filtrate was concentrated, and the crude product was purified by column chromatography (100:1 CH₂Cl₂–MeOH) to give **62** as a white glassy solid (0.70 g, 90%). The NMR data were in agreement with those in the literature.¹³ ¹H NMR (CDCl₃): δ 7.89 (bs, 2H, Phth), 7.75–7.80 (m, 2H, Phth), 5.89 (pt, 1H, J = 10.0 Hz, N3), 5.45 (d, 1H, J = 8.4 Hz, N1), 5.13 (pt, 1H,

$J = 10$ Hz, N4), 4.28–4.35 (m, 5H), 3.60–3.81 (m, 3H), 3.49 (s, 3H, CO₂Me), 3.39–3.44 (m, 1H), 3.14 (s, 3H, OMe), 2.13 (s, 3H, Ac), 2.05 (s, 3H, Ac), 1.86 (s, 3H, Ac); ¹³C NMR (CDCl₃): δ 170.5, 169.8, 169.3, 167.2, 134.0, 123.4, 103.6 (U1), 98.6 (N1), 81.7, 74.3, 73.2, 72.6, 71.7, 70.0, 68.6, 61.7 (N6), 57.2 (CO₂CH₃), 54.2 (OMe), 51.7 (N2), 20.5, 20.2.

Preparation of methyl (2-acetamido-2-deoxy- β -D-glucopyranosyl-(1 $\rightarrow$ 4)- β -D-glucopyranosid)uronic acid (67**).¹³** To a solution of **62** (0.7 g, 1.09 mmol) in MeOH (30 mL) was added 1 N NaOH (33 mL, 33 equiv) dropwise at 0 °C. After the addition was completed, the reaction was slowly warmed up to room temperature and stirred overnight. The reaction was quenched by adding Amberlite IR-120 (H⁺) resin. After solvent was evaporated, a mixture of **63** and **64** (characterized by ESI-MS) was obtained as a white solid (0.5 g, 91%). A portion of this solid (0.45 g) was dissolved in 95% ethanol (50 mL) and hydrazine monohydrate (distilled colorless, 30 mL). The solution was refluxed for 24 h until ESI-MS showed that all of **63** and **64** was converted to **65** and **66**. The solvent was removed by co-evaporating with toluene. The resulting white solid (1.15 g) was dissolved in dry MeOH (25 mL). To this solution was added Ac₂O (5 mL) at 0 °C. After 10 h, ESI-MS showed that all **65** was converted to **67**. The solution was then concentrated, and the crude product was purified by column chromatography (1:1 CHCl₃–MeOH) to give **67** as a white solid (96 mg, 21% in three steps). The NMR data were in agreement with those in the literature.¹³ ¹H NMR [D₂O, the acetamido methyl proton (δ 2.04 ppm)

was used as the internal reference]: δ 2.04 (s, 3H, Ac), 3.31 (t, J = 8.6 Hz, 1H, U2), 3.41–3.48 (m, 3H, N4, N5 and N3), 3.52–3.61 (m, 4H, OMe and U3), 3.69–3.78 (m, 4H, N2, U5, U4 and N6a), 3.89–3.94 (m, 1H, N6b), 4.36 (d, J = 7.9 Hz, 1H, U1), 4.53 (d, J = 8.3 Hz, 1H, N1); ^{13}C NMR [D_2O , the acetamido methyl carbon (δ 23.3 ppm) was used as the internal reference]: δ 23.29 (CH_3CO), 56.24 (OCH_3), 58.20 (N2), 61.43 (N6), 70.60 (N4/N5), 73.61 (U2), 74.75 (U3 and N3), 76.76 (N5/N4), 77.55 (U5), 80.84 (U4), 101.64 (N1), 104.27 (U1), 175.21 (CO), 175.77 (CO).

Preparation of 2-acetamido-2-deoxy-4,6-O-isopropylidene- α -D-glucopyranose (68).⁵⁶ To a solution of 2-deoxy-2-acetamido- α -D-glucose (9.63 g, 43.5 mmol) and *p*-toluenesulfonic acid monohydrate (0.122 g, 0.015 equiv) in DMF (46 mL) was added 2,2-dimethoxypropane (20 mL, 3.7 equiv) at room temperature. The mixture was stirred for 5.5 h, and TEA (1 mL) was added to quench the reaction. The white suspension was filtered to recover unreacted starting glucose as white needle crystals (3.78 g). The filtrate was concentrated to give **68** as a white solid (5.65 g, 82%). ^1H NMR (CD_3OD): δ 1.28 (s, 3H), 1.42 (s, 3H), 1.89 (s, 3H, Ac), 3.20–3.30 (m, 1H), 3.46 (t, J = 8.4 Hz, 1H, H2), 3.64–3.74 (m, 3H), 3.80–3.86 (m, 1H, H6a), 4.50 (bs, 1H, NH), 4.97 (d, J = 3.6 Hz, 1H, H1); ^{13}C NMR (CD_3OD): δ 19.40 (CH_3), 22.61 (CH_3), 29.53 (CH_3CO), 56.40 (C2), 63.64 (C6), 64.47, 69.96, 76.41, 93.24 (C1), 100.91 (CMe_2), 173.67 (CO).

(*EZ*)-3-Acetamido-1,2,3-trideoxy-5,7-O-isopropylidene-1-phenyl-D-glucopyranose (69).

hept-1-enitol (69). Compound **68** (3.62 g, 13.8 mmol), benzyltriphenylphosphonium chloride (10.76 g, 2 equiv), flame-dried K_2CO_3 (3.83 g, 2 equiv) and formamide (dried over 3 Å MS, 3.3 mL, 6 equiv) were dissolved in dry dioxane (80 mL). The solution was refluxed for 12 h, then concentrated. The crude product was purified by column chromatography (2:1:0.5 hexanes–ethyl acetate–acetonitrile) to give **69** as a colorless syrup (3.90 g, 84%). 1H NMR ($CDCl_3$): δ 1.30–1.43 (m, 5.00H), 1.89 (s, 1.30H), 2.00 (s, 1.66H), 3.54–3.63 (m, 1.11H), 3.72–3.86 (m, 3.62H), 3.94–4.03 (m, 1.66H), 4.85–4.88 (m, 0.48H), 5.20–5.27 (m, 0.50H), 5.57 (dd, J = 11.3, 9.5 Hz, 0.33H), 6.14 (dd, J = 15.9, 5.8 Hz, 0.49H), 6.46–6.59 (m, 1.33H), 6.81 (d, J = 8.5 Hz, 0.35H), 7.03 (d, J = 8.7 Hz, 0.53H), 7.19–7.36 (m, 6H); ^{13}C NMR ($CDCl_3$): δ 19.17 (CH_3), 23.30 (CH_3), 28.45 (CH_3CO), 50.68, 54.62, 61.75, 64.47 (C6), 69.37, 69.60, 73.95, 74.14, 98.57 (CMe_2), 125.76, 126.25, 127.21, 127.37, 127.53, 128.26, 128.41, 131.13, 132.28, 136.06, 136.35, 170.28, 170.56.

(*E/Z*)-3-Acetamido-4,6-di-O-acetyl-1,2,3-trideoxy-5,7-O-isopropylidene-1-phenyl-D-glucO-hept-1-enitol (70). To a solution of **69** (0.25 g, 0.75 mmol) in pyridine (15 mL) was added acetyl chloride (0.21 mL, 4 equiv) at room temperature. After 13 h stirring, the resulting brown solution was poured into a mixture of crushed ice and satd $NaHCO_3$. The mixture was extracted with ethyl acetate, and the extract was dried over $MgSO_4$. The crude product upon solvent evaporation was purified by column chromatography (2:1 hexanes–ethyl acetate) to give **70** as a white glassy solid (0.19 g, 61%). 1H NMR ($CDCl_3$): δ 1.44–1.53 (m, 7.70H), 1.92 (s, 1.75H), 1.95

(s, 1.84H), 2.09 (s, 2.24H), 3.63 (t, $J = 8.9$ Hz, 0.97H), 3.93–4.01 (m, 1.02H), 4.17–4.21 (m, 0.78H), 4.80–4.85 (m, 0.93H), 5.08–5.12 (m, 1.69H), 5.44–5.52 (m, 0.77H), 6.09 (dd, $J = 15.9, 5.7$ Hz, 0.34H), 6.54–6.62 (m, 1.21H), 6.72 (dd, $J = 8.2$ Hz, 0.38H), 6.89 (d, $J = 8.1$ Hz, 0.45H), 7.22–7.47 (m, 6H); ^{13}C NMR (CDCl_3): δ 19.20 (CH_3), 19.42 (CH_3), 20.39 (CH_3), 20.70 (CH_3), 23.04 (OAc), 23.15 (OAc), 27.43 (NHAc), 47.38, 51.74, 61.53, 61.59, 64.03, 64.11, 69.27, 69.43, 69.87, 70.38, 99.41, 99.50, 124.95, 126.13, 126.97, 127.28, 127.62, 128.14, 128.35, 128.53, 131.63, 132.49, 135.49, 136.02, 169.00, 169.34, 169.49, 170.05; ESI-MS (positive-ion mode): m/z 419.9 ($\text{M} + \text{H}^+$), 441.8 ($\text{M} + \text{Na}^+$), 457.9 ($\text{M} + \text{K}^+$).

2-Acetamido-3,5-di-O-acetyl-2-deoxy-4,6-O-isopropylidene-aldehydo-D-glucose (71). Compound **70** (0.15 g, 0.35 mmol) was dissolved in a solution of 2:1 CH_2Cl_2 –MeOH (9 mL) and cooled to -78°C , at which time a stream of oxygen and ozone was passed through the solution until a bluish color appeared that indicated an excess of ozone. After removal of excess ozone by a nitrogen stream, methyl sulfide (0.1 mL) was added. The solution was allowed to warm up to room temperature and stirred overnight. An aliquot of this solution was taken to run ESI-MS (positive-ion mode), which indicated that no elimination product was formed. The solution was then concentrated, and the crude product was purified by a fast (within 2 h) column chromatography (CHCl_3 then 100:1 CHCl_3 –MeOH) to give **71** as a colorless syrup (0.050 g, 41%). ^1H NMR (CDCl_3): δ 1.46 (s, 3H), 1.52 (s, 3H), 2.04 (s, 3H), 2.05 (s, 3H), 2.08 (s, 3H), 3.40 (d, $J = 6.0$ Hz, 1H), 3.68 (t, $J = 10.0$ Hz,

2H), 3.95–4.02 (m, 1H), 4.28 (d, $J = 9.7$ Hz, 1H), 4.78 (t, $J = 7.2$ Hz, 1H), 4.88 (m, 1H), 9.70 (s, 1H, CHO); ^{13}C NMR (CDCl_3): δ 19.46, 20.36, 20.59, 22.77, 60.11, 61.45 (CH_2), 63.80, 70.10, 71.47, 99.77 (CMe_2), 169.80, 170.17, 170.25, 196.66 (CHO); ESI-MS (positive-ion mode): m/z 346 ($\text{M} + \text{H}^+$), 368 ($\text{M} + \text{Na}^+$), 386 ($\text{M} + \text{H}_2\text{O} + \text{Na}^+$), 400 ($\text{M} + \text{CH}_3\text{OH} + \text{Na}^+$).

2-Acetamido-5-O-acetyl-2,3-dideoxy-4,6-O-isopropylidene-*aldehydo*- D -erythro-hex-2-enose (72). When **71** was in contact with silica gel for more than 2 h, it decomposed to give **72** as a colorless syrup. ^1H NMR (CDCl_3): δ 1.43 (s, 3H), 1.55 (s, 3H), 2.05 (s, 3H, Ac), 2.16 (s, 3H, Ac), 3.74 (dd, $J = 11.6, 8.0$ Hz, 1H, H6a), 4.03 (dd, $J = 11.6, 5.3$ Hz, 1H, H6b), 4.78–4.87 (m, 1H), 5.10–5.17 (m, 1H), 6.02 (d, $J = 6.3$ Hz, 1H, H2), 9.29 (s, 1H, CHO); ^{13}C NMR (CDCl_3): δ 19.98, 20.79, 23.49 (OAc), 27.85 (NAc), 61.60 (CH_2), 68.45, 68.84, 99.59 (CMe_2), 134.14 (C3), 135.73, 167.83, 169.93, 189.05 (CHO).

(*E/Z*)-3-Acetamido-4,6-di-O-benzoyl-5,7-O-isopropylidene-1-phenyl-1,2,3-trideoxy- D -gluco-hept-1-enitol (73). To a solution of **69** (0.25 g, 0.75 mmol) in pyridine (15 mL) was added benzoyl chloride (0.35 mL, 4 equiv) at room temperature. After 13 h stirring, the resulting brown solution was poured into a mixture of crushed ice and satd NaHCO_3 . The mixture was extracted with ethyl acetate, and the organic extract was dried over MgSO_4 . The crude product was purified by column chromatography (2:1 hexanes–ethyl acetate) to give **73** as a

colorless syrup (0.24 g, 58%). ^1H NMR (CDCl_3): δ 1.49 (s, 1.81H), 1.54 (s, 3.62H), 1.91 (s, 1.66H), 2.01 (s, 1.31H), 3.72–3.81 (m, 0.74H), 4.08–4.17 (m, 1.04H), 4.43–4.50 (m, 0.75H), 5.06–5.35 (m, 0.78H), 5.40–5.58 (m, 1.30H), 5.71–5.82 (m, 0.37H), 6.15–6.25 (m, 0.12H), 6.52–6.66 (m, 1.34H), 6.94 (dd, $J = 8.1$ Hz, 0.16H), 7.25–7.54 (m, 11.25H), 7.86–8.03 (m, 3.69H); ^{13}C NMR (CDCl_3): δ 20.08, 23.25, 27.28 (NAc), 47.95, 52.32, 61.95 (CH_2), 65.72, 65.89, 70.31, 71.02, 99.83 (CMe_2), 125.14, 126.37, 127.18, 127.43, 127.73, 128.22, 128.31, 128.46, 128.71, 129.13, 129.21, 129.34, 129.56, 129.70, 132.14, 132.87, 133.09, 133.20, 135.68, 165.23, 165.58, 165.64, 169.16, 169.47; ESI-MS (positive-ion mode): m/z 544 ($\text{M} + \text{H}^+$), 566 ($\text{M} + \text{Na}^+$), 582 ($\text{M} + \text{K}^+$).

2-Acetamido-3,5-di-O-benzoyl-2-deoxy-4,6-O-isopropylidene-aldehydo-D-glucose (74). Compound **73** (0.24 g, 0.44 mmol) was dissolved in a solution of 2:1 CH_2Cl_2 –MeOH (15 mL) and cooled to -78°C , at which time a stream of oxygen and ozone was passed through the solution until a bluish color appeared that indicated an excess of ozone. After removal of excess ozone by a nitrogen stream, a solution of methyl sulfide (0.1 mL) was added. The solution was allowed to warm up to room temperature and stirred overnight. An aliquot of this solution was taken to run ESI-MS (positive-ion mode), which indicated that no elimination product was formed. The solution was then concentrated, and the crude product was purified by a fast (within 2 h) column chromatography (CHCl_3 then 100:1 CHCl_3 –MeOH) to give **74** as a colorless syrup. ^1H NMR (CDCl_3): δ 1.53 (s, 3H), 1.62 (s, 3H), 2.02

(s, 3H, Ac), 3.83 (dd, $J = 11.7, 7.8$ Hz, 2H), 4.18 (dd, $J = 11.7, 5.4$ Hz, 1H), 4.60 (d, $J = 9.5$ Hz, 1H), 5.04–5.13 (m, 2H), 7.39–7.60 (m, 10H), 9.86 (s, 1H, CHO); ^{13}C NMR (CDCl_3): δ 20.13, 22.67, 27.59 (NAc), 60.87, 61.79 (CH_2), 65.25, 71.75, 72.00, 100.08 (CMe_2), 128.44, 128.49, 128.70, 129.10, 129.74, 129.83, 133.37, 133.59, 165.34 (PhCO), 165.64 (PhCO), 170.41 (NHCO), 196.85 (CHO); ESI-MS (positive-ion mode): m/z 470 ($\text{M} + \text{H}^+$), 487 ($\text{M} + \text{NH}_4^+$), 492 ($\text{M} + \text{Na}^+$), 510 ($\text{M} + \text{H}_2\text{O} + \text{Na}^+$), 524 ($\text{M} + \text{CH}_3\text{OH} + \text{Na}^+$), 540 ($\text{M} + \text{CH}_3\text{OH} + \text{K}^+$).

2-Acetamido-5-O-benzoyl-2,3-dideoxy-4,6-O-isopropylidene- α -erythro-hex-2-enose (75). When **74** was in contact with silica gel for more than 2 h, it decomposed to give **75** as a colorless syrup. ^1H NMR (CDCl_3): δ 1.47 (s, 3H), 1.62 (s, 3H), 2.05 (s, 3H, Ac), 3.89 (dd, $J = 11.6, 8.1$ Hz, 1H, H6a), 4.14 (dd, $J = 11.6, 5.8$ Hz, 1H, H6b), 5.08–5.15 (m, 1H, H5), 5.45 (dd, $J = 9.5, 6.4$ Hz, 1H, H4), 6.05 (d, $J = 6.4$ Hz, 1H, H2), 7.45 (t, $J = 7.6$ Hz, 2H), 7.59 (t, $J = 7.4$ Hz, 1H), 7.99 (d, $J = 7.2$ Hz, 2H), 9.23 (s, 1H, CHO); ^{13}C NMR (CDCl_3): δ 20.13, 23.63, 27.79 (NAc), 61.86 (CH_2), 68.78, 69.21, 99.72 (CMe_2), 128.48, 129.19, 129.64, 133.50, 134.57, 135.23, 165.42, 167.43, 189.09 (CHO).

(Z)-4,6-Di-O-acetyl-5,7-di-O-benzyl-1-phenyl-3-phthalimido-1,2,3-trideoxy- α -gluco-hept-1-enitol (76). To a solution of (Z/E)-5,7-di-O-benzyl-1-phenyl-3-phthalimido-1,2,3-trideoxy- α -gluco-hept-1-enitol (47.9 mg, 0.085 mmol, prepared by co-worker Sean K. Hamilton in a similar procedure as that for

compound **69**) in Ac₂O (2 mL) was added pyridine (0.014 mL, 2 equiv) at room temperature. After 5 h stirring, the solvent was evaporated, and the crude product was purified by column chromatography (2:1:0.3 hexanes–ethyl acetate–acetonitrile) to give (*Z/E*)-**76** as a colorless syrup (33 mg, 60%). Further separation of *Z* and *E* isomers was achieved by using 4:1:1 hexanes–acetone–toluene as eluting solvent to give pure *Z*-**76** as a colorless syrup. ¹H NMR (CDCl₃): δ 1.74 (s, 3H, OAc), 1.98 (s, 3H, OAc), 3.65–4.00 (m, 5H, H4, H5, H6, H7a and H7b), 4.38 (dd, *J* = 10.5, 8.9 Hz, 1H, H2), 4.49–4.55 (m, 3H, 1.5 OCH₂Ph), 4.70 (d, *J* = 12.0 Hz, 1H, 0.5 OCH₂Ph), 5.84 (dd, *J* = 10.5, 8.7 Hz, 1H, H3), 6.52 (d, *J* = 8.9 Hz, 1H, H1), 7.12–7.37 (m, 15H), 7.70–7.86 (m, 4H, Phth); ¹³C NMR (CDCl₃): δ 20.42 (CH₃), 20.80 (CH₃), 53.99 (C3), 67.78 (CH₂), 72.91, 73.56 (CH₂), 74.51 (CH₂), 75.31, 75.92, 89.89, 123.57, 125.24, 127.74, 127.98, 128.16, 128.36, 128.97, 131.31, 134.23, 137.80, 167.51, 168.74, 169.98; ESI-MS (positive-ion mode): *m/z* 648 (*M* + H⁺).

5-O-Acetyl-2,3-dideoxy-4,6-O-isopropylidene-2-phthalimido-*erythro*-hex-2-enose (78).³⁸ Compound **76** (0.462 g, 0.71 mmol) was submitted to the same ozonolysis procedure as that described in the preparation of compound **71**. However, the column chromatography (2:1 hexanes–ethyl acetate) took more than 2 h to obtain **78**. ¹H NMR (CDCl₃): δ 2.05 (s, 3H, OAc), 3.86 (m, 2H, H6a and H6b), 4.42 (s, 2H, OCH₂Ph), 4.49–4.58 (m, 2H, H4 and 0.5 OCH₂Ph), 4.70 (d, *J* = 11.8 Hz, 0.5 OCH₂Ph), 5.24 (m, 1H, H5), 7.04 (d, *J* = 8.6 Hz, H3), 7.12–7.33 (m,

10H), 7.72–7.89 (m, 4H, Phth), 9.46 (s, 3H, CHO); ^{13}C NMR (CDCl_3): δ 20.96 (OAc), 67.32 (C6), 72.12 (CH_2), 72.62, 72.97 (CH_2), 73.97, 123.82 (C3), 127.31, 127.52, 127.79, 128.10, 128.25, 128.40, 128.58, 131.92, 134.37, 134.80, 137.35, 137.62, 151.94, 170.28, 184.17, 186.45 (CHO).

***p*-Methoxyphenyl 4-C-aminomethyl-2,3-di-*O*-benzyl-4-deoxy-6-*O*-*p*-methoxy benzyl- β -D-glucopyranoside (91).**⁴⁰ To a solution of **90** (0.454 g, 0.76 mmol) in dry ether (10 mL) was added LiAlH_4 (0.0719 g, 1.89 mmol) portionwise at room temperature. After 3 h stirring, ethyl acetate was added dropwise followed by careful addition of water. The white suspension was filtered through Celite. The filtrate was dried over NaSO_4 and concentrated. The crude product was purified by preparative thin-layer chromatography (1:1 hexanes-ethyl acetate) to give **91** as a yellow syrup (0.304 g, 66%). IR (KBr, neat thin film): 3406.87, 3385.98 (s) cm^{-1} ; $[\alpha]_{\text{D}}^{20} = -24.1^\circ$ (c 0.37, CHCl_3); ^1H NMR (CDCl_3): δ 1.76 (m, 1H, H4), 2.82 (bd, NH_2), 3.61–3.79 (m, 13H, 2ArOCH₃, H2, H3, H5, CH_2NH_2 , CH_2ArOMe), 4.42, 4.46, 4.49, 4.54 (AB, 2H, $J = 11.3$ Hz, H6a, H6b), 4.86 (d, 1H, $J = 10.0$ Hz, H1), 4.66, 4.71, 4.93, 4.98 (AB, 2H, $J = 12.5$ Hz, CH_2Ph), 4.80, 4.84, 5.07, 5.11 (AB, 2H, $J = 10.0$ Hz, CH_2Ph), 6.82, 6.86, 7.00, 7.04 (AB, 4H, $J = 10.0$ Hz, ArOMe), 6.79, 6.82, 7.20, 7.24 (AB, 4H, $J = 10.0$ Hz, ArOMe), 7.27–7.39 (m, 10H, Ar); ^{13}C NMR (CDCl_3): δ 37.94 (weak CH_2NH_2), 45.73 (weak, C4), 55.17 (OCH₃), 55.55 (OCH₃), 70.63 (C6), 73.13 (CH_2), 73.39, 74.70 (CH_2), 74.79 (CH_2), 78.01, 83.57, 102.72 (C-1), 113.69, 114.47, 118.17, 127.63, 127.75, 128.13, 128.22, 128.34, 128.40, 129.28,

129.40, 130.11 (quat.), 138.39 (quat.), 151.56 (quat.), 155.07 (quat.), 159.15 (quat.).

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-4-deoxy-6-*O*-*p*-methoxybenzyl 4-*C*-nitromethyl- β -D-glucopyranoside (**94**).** To the neat **91** (0.100 g, 0.167 mmol) was added freshly prepared dimethyldioxirane⁴¹ solution in acetone prepared as follows: To a solution of NaHCO₃ (12 g) in 20:13 H₂O–acetone (33 mL) was added Oxone® (25 g, 245 equiv) with vigorous stirring at room temperature and under reduced pressure (water aspirator). The product, dimethyldioxirane, was collected in a dry ice–acetone trap within an hour. The resulting yellow solution was stirred at room temperature for 3 h. Solvent evaporation gave **94** as a light yellow syrup (0.066 g, 63%). Pure **94** was obtained by column chromatography (200:1 CH₂Cl₂–MeOH) as a colorless syrup. IR (KBr, neat thin film): 1553.03 (vs), 1379.2 (m) cm⁻¹; [α]_D²⁰ = -22.7° (c 0.715, CHCl₃); ¹H NMR (CDCl₃): δ 2.47 (bs, 1H, H4), 3.67–3.76 (m, 4H, H2, H3, H5 and H6a), 3.78 (s, 3H), 3.79 (s, 3H), 3.87–3.93 (m, 1H, H6b), 4.31–4.38 (m, 2H, 0.5 OCH₂ and 0.5 CH₂NO₂), 4.52 (d, *J* = 11.5 Hz, 1H, 0.5 OCH₂), 4.61–4.72 (m, 2H, 0.5 OCH₂ and 0.5 CH₂NO₂), 4.81 (d, *J* = 10.9 Hz, 1H, 0.5 OCH₂), 4.87 (d, *J* = 7.3 Hz, 1H, H1), 4.99 (d, *J* = 10.9 Hz, 1H, 0.5 OCH₂), 5.09 (d, *J* = 10.8 Hz, 1H, 0.5 OCH₂), 6.82 (d, *J* = 9.0 Hz, 2H), 6.85 (d, *J* = 7.5 Hz, 2H), , 7.00 (d, *J* = 6.9 Hz, 2H), 7.20 (d, *J* = 8.6 Hz, 2H), 7.25–7.37 (m, 10H); ¹³C NMR (CDCl₃): δ 42.43 (C4), 55.25 (OCH₃), 55.85 (OCH₃), 69.53 (CH₂), 72.08 (CH₂), 72.36, 73.18 (CH₂), 74.87 (CH₂), 75.37 (CH₂), 78.03, 83.39, 102.86 (C1), 113.82,

114.58, 118.40, 127.86, 127.95, 128.10, 128.21, 128.47, 128.52, 129.56, 129.88 (quat.), 137.89 (quat.), 138.02 (quat.), 151.30 (quat.), 155.39 (quat.), 159.36 (quat.).

Preparation of phenyl 3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-1-seleno- β -D-glucopyranoside (95).⁵⁷ This compound was prepared by my co-worker Karen Welch according to the literature procedure. ¹H NMR (CDCl₃): δ 7.65–7.76 (m, 4H, Phth), 7.42–7.45 (m, 2H, Ar), 7.12–7.25 (m, 3H, Ar), 5.83 (d, 1H, J = 10.6 Hz, H1), 5.71 (pt, 1H, J = 9.7 Hz, H3), 5.05 (pt, 1H, J = 9.7 Hz, H4), 4.31 (pt, 1H, J = 10.4 Hz, H2), 4.15–4.24 (m, 2H, H6a, H6b), 3.78–3.85 (m, 1H, H5), 2.02 (s, 3H, OAc), 1.94 (s, 3H, OAc), 1.75 (s, 3H, OAc); ¹³C NMR (CDCl₃): δ 170.40, 169.88, 169.28, 167.61, 166.73, 135.25, 134.23, 131.39, 130.98, 128.80, 128.39, 125.95, 123.52, 78.25, 76.77, 71.27, 68.49, 61.98 (C6), 54.52 (C2), 20.59, 20.45, 20.25.

Phenyl 4-O-benzyl-3-O-(*tert*-butyldimethylsilyl)-2-deoxy-2-phthalimido-1-seleno- β -D-glucopyranoside (98). This compound was prepared by my co-worker Karen Welch according to the literature procedure.⁴⁵ ¹H NMR (CDCl₃): δ 7.72–7.84 (m, 4H, Phth), 7.16–7.48 (m, 10H, Ar), 5.83 (d, 1H, J = 10.5 Hz, H1), 4.63, 4.68, 4.80, 4.85 (AB, 2H, J = 12.5 Hz, CH₂Ph), 4.50 (dd, 1H, J = 9.7, 7.9 Hz, H3), 4.31 (pt, 1H, J = 10.1 Hz, H2), 3.87–3.93 (m, 1H, H6a), 3.48–3.72 (m, 3H, H4, H5, H6b), 0.72 (s, 9H, Me₃CSiMe₂), -0.05 (s, 3H, Me), -0.42 (s, 3H, Me); ¹³C NMR (CDCl₃): δ 168.25, 167.89, 138.01, 134.70, 134.19, 131.79, 128.96, 128.30, 128.13, 127.49, 127.07, 123.36, 60.67, 79.41 (C1), 74.61 (CH₂Ph), 73.12, 61.87 (C6), 57.88 (C2),

25.60 ($\text{Me}_3\text{CSiMe}_2$), 17.58 ($\text{Me}_3\text{CSiMe}_2$), -0.69 ($\text{Me}_3\text{CSiMe}_2$), -1.24 ($\text{Me}_3\text{CSiMe}_2$).

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl- β -D-xylo-hexopyranosid-4-ulose (100a).** To a solution of compound **115** (6.12 g, 10.4 mmol) in CH_2Cl_2 (50 mL) was added Dess–Martin Periodinane (5.75 g, 1.3 equiv) at room temperature. The reaction was stirred with the flask open to the air for 6 h, then it was quenched by a mixture of satd NaHCO_3 (75 mL) and 1% $\text{Na}_2\text{S}_2\text{O}_3$ (175 mL). After phase separation, the organic phase was dried over Na_2SO_4 , and the crude product obtained upon solvent evaporation was purified by column chromatography to give **100a** (5.02 g, 82% yield) as white needles. mp 74–75 °C; $[\alpha]_{\text{D}}^{20} = +1.2$ (c 1.05, CHCl_3); ^1H NMR (CDCl_3): δ 3.60–3.67 (m, 1H, H6b), 3.70 (s, 3H), 3.71 (s, 3H), 3.90 (dd, $J = 10.9, 3.5$ Hz, 1H, H6a), 3.99 (dd, $J = 9.1, 6.4$ Hz, 1H, H2), 4.18–4.22 (m, 2H, H3 and H5), 4.37 (s, 2H, OCH_2), 4.59 (d, $J = 11.6$ Hz, 1H, 0.5 OCH_2), 4.77 (d, $J = 11.2$ Hz, 1H, 0.5 OCH_2), 4.82 (d, $J = 12.6$ Hz, 1H, 0.5 OCH_2), 4.90 (d, $J = 11.3$ Hz, 1H, 0.5 OCH_2), 5.27 (d, $J = 6.3$ Hz, 1H, H1), 6.78 (d, $J = 9.1$ Hz, 2H), 6.79 (d, $J = 8.6$ Hz, 2H), 7.03 (d, $J = 9.0$ Hz, 2H), 7.15 (d, $J = 8.5$ Hz, 2H), 7.26–7.40 (m, 10H); ^{13}C NMR (CDCl_3): δ 54.92 (OCH_3), 55.29 (OCH_3), 67.96 (CH_2), 73.00 (CH_2), 73.55 (CH_2), 74.37 (CH_2), 77.28, 82.73, 82.98, 101.65 (C1), 113.48, 114.39, 118.00, 127.62, 127.70, 127.84, 128.17, 129.13, 129.73 (quat.), 137.26 (quat.), 137.62 (quat.), 150.78 (quat.), 155.18 (quat.), 159.00 (quat.), 201.51 (CO). Anal. Calcd for $\text{C}_{35}\text{H}_{36}\text{O}_8$: C, 71.89; H, 6.22. Found: C, 71.88; H, 6.14.

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-4-*C*-methylene- β -D-xylo-hexopyranoside (101a).** To a suspension of methyltriphenylphosphonium bromide (3.14 g, 1.5 equiv) in freshly distilled THF (30 mL) was added BuLi (2.5 M in hexanes, 3.52 mL, 1.5 equiv) dropwise in a -53 °C bath. The resulting yellow solution was stirred at the same temperature for 0.5 h, then a solution of **100a** (3.43 g, 5.86 mmol) in freshly distilled THF (30 mL) was transferred into completely via a N₂ pressurized cannula all at once. The solution was slowly warmed up to 0 °C over a period of 2 h followed by quenching with brine (50 mL). The organic phase was dried over Na₂SO₄ and concentrated. The crude product was purified by column chromatography (3:1 hexanes–ethyl acetate) to give **101a** as white needles. mp 67–68 °C; $[\alpha]_D^{20} = +0.1^\circ$ (c 0.705, CHCl₃); ¹H NMR (CDCl₃): δ 3.65 (t, *J* = 8.0 Hz, 1H, H₂), 3.70 (s, 3H, OCH₃), 3.72 (s, 3H, OCH₃), 3.76 (m, 1H, H_{6b}), 3.84–3.88 (m, 1H, H_{6a}), 4.03 (d, *J* = 8.4 Hz, 1H, H₃), 4.13 (m, 1H, H₅), 4.40, 4.45, 4.48, 4.52 (AB, *J* = 11.4 Hz, 2H, OCH₂Ar), 4.70 (s, 2H, OCH₂Ar), 4.78, 4.82, 4.92, 4.96 (AB, *J* = 11.1 Hz, 2H, OCH₂Ar), 5.03 (d, *J* = 7.1 Hz, 1H, H₁), 5.08 (s, 1H, =CH), 5.39 (s, 1H, =CH), 6.76, 6.79, 7.03, 7.06 (AB, *J* = 8.8 Hz, 4H, OC₆H₄OMe), 6.81, 6.84, 7.20, 7.23 (AB, *J* = 8.5 Hz, 4H, OCH₂C₆H₄OMe), 7.26–7.33 (m, 10H, 2OC₆H₅); ¹³C NMR (CDCl₃): δ 54.98 (OMe), 55.34 (OMe), 69.72 (C₆), 72.70 (C₅), 73.06 (CH₂), 73.24 (CH₂), 74.51 (CH₂), 81.01 (C₃), 83.59 (C₂), 102.35 (C₁), 109.05 (=CH₂), 113.57, 114.31, 118.04, 127.51, 127.90, 128.18, 129.22, 129.90 (quat.), 137.99 (quat.), 138.17 (quat.), 141.08 (quat.), 151.35 (quat.), 154.94 (quat.), 159.06 (quat.). Anal. Calcd for C₃₆H₃₈O₇: C, 74.19; H, 6.59. Found: C, 74.12; H, 6.52.

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-4-*C*-methylene- β -D-xylo-hexopyranoside (102a).** To a solution of **101a** (1.29 g, 2.21 mmol) in 10:1 CH₂Cl₂–H₂O (66 mL) was added DDQ (1.5 g, 3 equiv) at room temperature. After 1 h stirring, the solution was poured into aq K₂CO₃ (0.4 M, 100 mL). The mixture was shaken vigorously and then separated into two phases. The bottom organic phase, which was a clear, light yellow-green color, was collected, dried over NaSO₄ and concentrated. The crude product was purified by column chromatography (3:1 hexanes–ethyl acetate) to give **102a** as pale yellow needles (0.85 g, 83% yield). A purer sample was obtained by recrystallization in hexanes–ethyl acetate to give **102a** as a white needle crystal. mp 79–80 °C; $[\alpha]_D^{20} = +6.1^\circ$ (c 0.575, CHCl₃); ¹H NMR (CDCl₃): δ 3.67 (t, *J* = 7.5 Hz, 1H, H2), 3.73 (s, 3H, OCH₃), 3.83–3.89 (m, 2H, H6a and H6b), 4.02–4.09 (m, 2H, H3 and H5), 4.71 (s, 2H, OCH₂Ar), 4.76, 4.80, 4.88, 4.92 (AB, *J* = 11.2 Hz, 2H, OCH₂Ar), 5.05 (s, 1H, =CH), 5.07 (d, *J* = 7.0 Hz, 2H, H1), 5.38 (s, 1H, =CH), 6.79, 6.82, 6.96, 7.00 (AB, *J* = 9.0 Hz, 4H, OC₆H₄OMe), 7.26–7.25 (m, 10 H, 2 OC₆H₅); ¹³C NMR (CDCl₃): δ 55.50 (OCH₃), 63.02 (C6), 73.18 (CH₂), 74.41 (C5), 74.42 (CH₂), 80.73 (C3), 83.22 (C2), 102.08 (C1), 109.99 (=CH₂), 114.57, 117.75, 127.67, 127.98, 128.25, 128.31, 137.89 (quat.), 138.07 (quat.), 140.47 (quat.), 151.05 (quat.), 155.09 (quat.). Anal. Calcd for C₂₈H₃₀O₆: C, 72.70; H, 6.55. Found: C, 72.48; H, 6.60.

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-4-*C*-methylene- β -D-xylo-hexopyranosiduronic acid (106a).**⁵⁰ To a solution of **102a** (0.134 g, 0.290 mmol)

in 1:1 CH₃CN–H₂O (2 mL) at room temperature was added bis(acetoxy)iodo benzene (0.205 g, 2.2 equiv) and TEMPO (0.009 g, 0.2 equiv). The resulting yellow mixture was stirred for 8 h. After phase separation, the water phase was once extracted with CH₂Cl₂. The combined organic phase was dried over Na₂SO₄ and concentrated. The crude product was purified by column chromatography (gradient elution starting with 2:1 ethyl acetate–hexanes up to pure ethyl acetate followed by 10:1 ethyl acetate–acetic acid) to give pure **106a** as a white solid (0.0651 g) along with a yellow syrup that proved to be impure **106a** (0.0557 g, total 0.1208 g, 88% yield). mp 129–130 °C; $[\alpha]_D^{20} = +18.3$ (c 0.295, CHCl₃); ¹H NMR (CDCl₃): δ 3.76 (s, 3H, OCH₃), 3.80 (t, *J* = 6.1 Hz, 1H, H₂), 4.03 (d, *J* = 5.5 Hz, 1H, H₃), 4.47, 4.52, 4.64, 4.69 (AB, *J* = 11.7 Hz, 2H, OCH₂Ar), 4.70, 4.74, 4.78, 4.83 (AB, *J* = 12.0 Hz, 2H, OCH₂Ar), 4.78 (s, 1H, H₅), 5.06 (d, *J* = 6.7 Hz, 1H, H₁), 5.42 (s, 1H, =CH), 5.49 (s, 1H, =CH), 6.80, 6.84, 6.96, 7.00 (AB, *J* = 8.9 Hz, 4H, OC₆H₄OMe), 7.31–7.33 (m, 10H, 2 OC₆H₅); ¹³C NMR (CDCl₃): δ 55.62 (OCH₃), 72.08 (CH₂), 73.83 (CH₂), 74.88 (C₅), 80.17 (C₃), 81.92 (C₂), 101.96 (C₁), 114.66, 116.01 (=CH₂), 118.64, 127.94, 128.01, 128.41, 128.46, 136.51 (=C₄), 137.29 (quat.), 137.65 (quat.), 150.76 (quat.), 155.69 (quat.), 169.64 (CO₂H). Anal. Calcd for C₂₈H₂₈O₇: C, 70.56; H, 5.93. Found: C, 70.42; H, 6.05.

Preparation of *p*-methoxyphenyl 2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranoside (108).⁵⁸ To a light yellow solution of β-D-glucose pentaacetate (30 g, 76.86 mmol) and *p*-methoxyphenol (14 g, 1.5 equiv) in dry CH₂Cl₂ (300 mL)

at 0 °C was added trimethylsilyl trifluoromethanesulfonate (TMSOTf, 2.2 mL, 0.15 equiv) dropwise. The brownish-red reaction mixture was allowed to stirred at 0 °C for 5 h, at the end of which time the reaction was quenched with TEA (2 mL), and the color of the mixture changed back to light yellow. After the addition of satd NaHCO₃ (200 mL), the reaction mixture was allowed to stir for 5 min. The suspension was then extracted with CH₂Cl₂ (2 × 100 mL). The organic extract was dried over MgSO₄ and concentrated. The crude product was purified by column chromatography (4:1 then 2:1 hexanes–ethyl acetate) and recrystallized (CH₂Cl₂–MeOH) to give **108** as white needles (30.47 g, 88%). The NMR data were in agreement with those in the literature.⁵⁸ ¹H NMR (CDCl₃): δ 2.04 (s, 6H, 2OAc), 2.09 (s, 6H, 2OAc), 3.78 (s, 3H, OMe), 3.82 (m, 1H), 4.14–4.33 (m, 2H), 4.94–4.99 (d, *J* = 7.1 Hz, 1H, H1), 5.13–5.29 (m, 3H), 6.80, 6.84, 6.93, 6.97 (AB, *J* = 9.0 Hz, 4H, OC₆H₄OMe); ¹³C NMR (CDCl₃): δ 20.62 (4OAc), 55.63 (OMe), 61.93 (C6), 68.30, 71.22, 71.93, 72.74, 100.30 (C1), 114.54, 118.70, 150.87 (quat.), 155.79 (quat.), 169.29 (COMe), 169.38 (COMe), 170.25 (COMe), 170.58.

Preparation of *p*-methoxyphenyl β-D-glucopyranoside (109).⁵⁸ To a solution of compound **108** (2.33 g, 5.16 mmol) in CH₂Cl₂ (1.6 mL) and methanol (31.4 mL) at room temperature was added NaOMe in methanol (2.0 M, 0.13 mL, 0.05 equiv). The reaction was allowed to stir overnight then neutralized with Dowex 50 × 2, 100 mesh (H⁺). The suspension was then filtered, and the filtrate was concentrated to give **109** as a white solid (1.54 g, quantitative). It was used in

the next step without further purification.

***p*-Methoxyphenyl 4,6-*O*-*p*-methoxybenzylidene- β -D-glucopyranoside (110).** A flask with a solution of compound **109** (28.47 g, 99.45 mmol), TsOH·H₂O (3.7 g, 0.22 equiv) and *p*-anisaldehyde dimethyl acetal (ADMA, 16.4 mL, 1.1 equiv) in DMF (300 mL) was placed on the rotary evaporator at 50 °C and under <20 torr. After 7 h, TLC showed that the reaction was completed, and TEA (3.7 mL) was then added. The DMF was evaporated, and the resulting yellow sticky solid was purified by column chromatography (CH₂Cl₂) to give **110** as an off-white solid (21.16 g, 53%). ¹³C NMR (DMSO-*d*₆): δ 55.10, 55.33, 65.73, 67.82, 72.76, 74.13, 80.35, 100.62, 101.86, 113.31, 114.43, 117.77, 127.83, 130.11, 151.04, 154.50, 159.52.

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-4,6-*O*-*p*-methoxybenzylidene- β -D-glucopyranoside (111).** A suspension of NaH (60% in mineral oil, 3 equiv) in DMF (50 mL) was cooled to 0 °C. To this was then added dropwise a DMF (250 mL) solution of **110** (23.32 g, 57.67 mmol). Upon completion of the addition, the reaction mixture was allowed to reach room temperature over a period of 15 min and was then recooled to 0 °C. Benzyl bromide (15 mL, 2.2 equiv) was added dropwise, and the reaction was allowed to stir overnight at room temperature. The reaction was then quenched with satd NH₄Cl (100 mL) and water (400 mL) at 0 °C. Compound **111** precipitated as a white solid. After filtration, the solid was washed by water (1000 mL) and redissolved in CH₂Cl₂ (400 mL). A separatory funnel was

used to separate the organic phase. The organic phase was then dried over MgSO_4 , and concentrated to give **111** as light-yellow gel-like solid (28.9 g, 86%) that was used in the next step without further purification.

***p*-Methoxyphenyl 2,3-di-O-benzyl-6-O-*p*-methoxybenzyl- β -D-glucopyranoside (112).** A suspension of compound **111** (9.41 g, 16.1 mmol) and NaBH_3CN (5.5 g, 5.4 equiv) in DMF (91 mL) maintained over 3Å molecular sieves (12.67 g) was stirred at 0 °C for 10 min. TFA (15 mL, 12 equiv) was added dropwise. The reaction was allowed to reach room temperature and stir overnight, after which time the reaction was filtered through Celite. The filtrate was combined with a solution of ice-cold satd NaHCO_3 . The mixture was stirred for 30 min and then extracted with CH_2Cl_2 . The organic extract was dried over MgSO_4 , and concentrated. The crude product was purified by column chromatography (20:1 CH_2Cl_2 – CH_3CN) to give **112** as white needles (4.07 g, 43%). ^1H NMR (CDCl_3): δ 2.57 (bs, 1H, OH), 3.48–3.56 (m, 2H), 3.63–3.71 (m, 4H), 3.77 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 4.50 (s, 2H, OCH_2), 4.76 (d, J = 11.5 Hz, 1H, 0.5 OCH_2), 4.82 (d, J = 10.9 Hz, 1H, 0.5 OCH_2), 4.90 (d, J = 7.7 Hz, 1H, H1), 4.95 (d, J = 11.4 Hz, 1H, 0.5 OCH_2), 5.05 (d, J = 11.0 Hz, 1H, 0.5 OCH_2), 6.79–6.87 (m, 4H), 7.02 (d, J = 7.0 Hz, 2H), 7.23 (d, J = 7.6 Hz, 2H), 7.26–7.39 (m, 10H); ^{13}C NMR (CDCl_3): δ 55.26 (OCH_3), 55.62 (OCH_3), 69.92 (CH_2), 71.58, 73.31 (CH_2), 74.21, 74.94 (CH_2), 75.33 (CH_2), 81.52, 84.04, 102.81 (C1), 113.79, 114.55, 118.40, 127.79, 127.86, 127.95, 128.22, 128.40, 128.55, 129.31, 129.98 (quat.), 138.22 (quat.), 138.56 (quat.),

151.42 (quat.), 155.33 (quat.), 159.27 (quat.). The 6-hydroxy regioisomer, *p*-Methoxyphenyl 2,3-di-*O*-benzyl-4-*O*-*p*-methoxybenzyl- β -D-glucopyranoside (**113**), eluted out of the column after compound **112**, and it was concentrated to give white needles (0.44 g, 4.6%). ^1H NMR (CDCl_3): δ 1.93 (bs, 1H, OH), 3.40–3.47 (m, 1H), 3.58–3.67 (m, 2H), 3.70–3.73 (m, 2H), 3.76 (s, 3H, OMe), 3.77 (s, 3H, OMe), 3.83–3.89 (m, 1H), 4.56, 4.61, 5.00, 5.05 (AB, J = 10.9 Hz, 2H), 4.78–4.87 (m, 3H), 4.93–5.05 (m, 3H), 6.81–6.86 (m, 4H), 6.96–7.00 (AB, J = 9.0 Hz, 2H), 7.19–7.22 (AB, J = 8.7 Hz, 2H), 7.27–7.33 (m, 10H); ^{13}C NMR (CDCl_3): δ 55.22 (OMe), 55.59 (OMe), 61.96 (C6), 74.73 (CH_2), 75.03, 75.28 (CH_2), 75.86 (CH_2), 77.09, 82.04, 84.44, 102.49 (C1), 113.88, 114.64, 118.05, 127.62, 127.76, 128.14, 128.36, 129.74, 130.03, 138.14, 138.48, 151.18, 155.33, 159.39.

***p*-Methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-4-*C*-formyl-6-*O*-*p*-methoxybenzyl- β -D-glucopyranoside (**115**).** Compound **115** was isolated as a by-product of LiAlH_4 reduction of compound **90**. ^1H NMR (CDCl_3): δ 3.03 (dt, J = 5.2 and 2.6 Hz, 1H, H4), 3.61 (m, 2H, H6a and H6b), 3.71 (dd, J = 8.8 and 6.1 Hz, 1H, H2), 3.80 (s, 3H, OMe), 3.81 (s, 3H, OMe), 3.83 (m, 1H, H5), 4.02 (dd, J = 10.7 and 9.0 Hz, 1H, H3), 4.43 (AB, J = 11.4 Hz, 2H, OCH_2Ar), 4.54, 4.59, 5.04, 5.08 (AB, J = 10.9 Hz, 2H, OCH_2Ar), 4.79, 4.83, 4.87, 4.90 (AB, J = 10.6 Hz, 2H, OCH_2Ar), 4.89 (d, J = 7.5 Hz, 1H, H1), 6.80, 6.83, 6.98, 7.01 (AB, 4H, Ar), 6.84, 6.88, 7.18, 7.21 (AB, 4H, Ar), 7.27–7.36 (m, 10H, Ar), 9.68 (d, J = 2.5 Hz, 1H, CHO); ^{13}C NMR (CDCl_3): δ 55.24 (OCH_3), 55.62 (OCH_3), 57.16 (C4), 70.18 (CH_2), 71.58, 73.15 (CH_2), 74.97

(CH₂), 75.16 (CH₂), 78.76, 82.47, 102.71 (C1), 113.82, 114.57, 118.39, 127.84, 128.01, 128.16, 128.37, 128.43, 129.40, 129.61 (quat.), 137.84 (quat.), 138.08 (quat.), 151.27 (quat.), 155.39 (quat.), 159.30 (quat.), 200.06 (CHO).

5.3. Unsuccessful synthesis

Nitroaldol reaction of 71 with 94: Compounds **94** (0.042 g, 0.066 mmol) and **71** (0.055 g, 2 equiv) were dissolved in 8:3 ether–acetonitrile solution (5.5 mL). To the solution was added powder 4 Å molecular sieve (0.17 g). The suspension was stirred at room temperature for 40 min, at which time KF (0.003 g, 8 equiv) and 18-crown-6 (0.002 g, 9 equiv) were added. TLC monitoring of the reaction up to 24 h showed no consumption of **94**.

Attempt of intermolecular radical coupling between 95 and 1-methylenecyclohexane: Bu₃SnH was added to a toluene solution of compound **95**, 1-methylenecyclohexane and AIBN at room temperature under N₂. The reaction was allowed to reflux overnight. TLC (10 : 1 CH₂Cl₂–MeOH) indicated all starting material consumed. The isolated product was confirmed by NMR as **3,4,6-tri-O-acetyl-1,5-anhydro-2-deoxy-2-phthalimido-D-glucitol (107)**.

***p*-Methoxyphenyl 2,3-di-O-benzyl-4-deoxy-4-C-formyl-6-O-*p*-methoxybenzyl-β-D-glucopyranoside (115).** First attempt: To a suspension of methoxymethyltriphenylphosphonium chloride (0.65 g, 2 equiv) in THF (20 mL) was added

KHMDS (0.5 M toluene solution, 3.8 mL, 2 equiv) at -78 °C over the period of 20 min under N₂. To the resulting deep orange-red solution was added a THF (20 mL) solution of **100a** (0.54 g, 0.92 mmol). The bath temperature was then slowly raised to 0 °C, and the reaction was allowed to proceed for 4.5 h at this temperature. TLC (1:1:0.5 hexanes—ethyl acetate—methanol) showed that **100a** decomposed to a very polar product which was on the baseline. The reaction was then quenched with satd NH₄Cl. The organic layer was dried over Na₂SO₄ and concentrated to give a brown syrup. Acid treatment of this syrup did not show any of the desired product, as determined by comparing with the authentic sample prepared from the DIBAL reduction of compound **90**. Second attempt:⁵⁹ To a suspension of methoxymethyltriphenylphosphonium chloride (0.13 g, 2.3 equiv) in THF (20 mL) was added *tert*-BuLi (1.5 M pentane solution, 0.25 mL, 2.3 equiv) at -78 °C under N₂. The bath temperature was slowly raised to -25 °C over a period of 3 h. The reaction mixture was then cooled back to -78 °C. To this pale-yellow solution was added a THF (20 mL) solution of **100a** (0.097g, 0.166 mmol). The bath temperature was allowed to slowly warm to room temperature over a period of 6 h. The following workup was the same as that of the first attempt. TLC still showed that the very polar product formed.

6. REFERENCES

1. Stryer, L. *Biochemistry*, 4th ed.; W.H. Freeman: New York, 1995, pp 474–5.
2. Liotta, L. A. Cancer Cell Invasion and Metastasis. *Scientific American*, **1992**, February, 54–63.
3. (a) Fidler, I.; Hart, I. Biological diversity in metastatic neoplasms: origins and implications. *Science* **1982**, 217, 998–1003.

(b) Frixen, U. H.; Behrens, J.; Sachs, M.; Eberle, G.; Voss, B.; Warda, A.; Lochner, D.; Birchmeier, W. E-Cadherin-mediated cell-cell adhesion prevents invasiveness of human carcinoma cells. *J. Cell. Biol.* **1991**, 113, 173–185.

(c) Stetler-Stevenson, W.; Aznavoorian, S.; Liotta, L. Tumor cell interactions with the extracellular matrix during invasion and metastasis. *Annu. Rev. Cell Biol.* **1993**, 9, 541–574.

(d) Takeichi, M. Cadherins in cancer: implications for invasion and metastasis. *Curr. Opin. Cell Biol.* **1993**, 5, 806–811.
4. Sneath, R. J. S.; Mangham, D. C. The Normal Structure and Function of CD44 and its Role in Neoplasia. *Mol. Pathol.* **1998**, 51, 191–200.
5. Jackson, D. G.; Buckley, J.; Bell, J. I. Multiple Variants of the Human Lymphocyte Homing Receptor CD44 Generated by Insertions at a Single Site in the Extracellular Domain. *J. Biol. Chem.* **1992**, 267, 4732–4739.
6. (a) Knudsen, W. The role and regulation of tumor-associated hyaluronan. In *The Biology of Hyaluronan*; Evered D.; Whelan, J., Eds.; Wiley:

- Chichester (Ciba Foundation Symposium, 143), 1989, pp 159–186.
- (b) Underhill, C. *ibid*, pp 87–106.
- (c) Toole, B. *ibid*, pp138–149.
- (d) Underhill, C. CD44: the hyaluronan receptor. *J. Cell. Sci.* **1992**, *103*, 293–298.
- (e) Cysyk, R.; August, M.; Duncan, K.; Malinowski, N. Inhibition of fibroblast hyaluronic acid production by suramin. *Oncol. Res.* **1994**, *5*, 1–9.
- (f) Cysyk, R. The National Cancer Institute, unpublished results.
7. Price, K. N. Ph. D. Dissertation, University of Tennessee, Knoxville, 1997.
 8. Cysyk, R. L. and co-workers, unpublished results.
 9. (a) Rodén, L.; Campbell, P.; Fraser, J.; Laurent, T.; Pertoft, H.; Thompson, J. Enzymic Pathways of Hyaluronan Catabolism. In *The Biology of Hyaluronan*; Evered, D., Whelan, J., Eds.; Wiley: Chichester (Ciba Foundation Symposium, 143), 1989; pp 60–86.
 - (b) Fraser, J.; Laurent, T. turnover and metabolism of hyaluronan, *ibid*, pp 41–59.
 10. Kresse, H.; Glössl, J. Glycosaminoglycan degradation. *Adv. Enzymol.* **1987**, *60*, 217–311.
 11. (a) Sicińska, W.; Adams, B. R.; Lerner, L. E. A detailed proton and carbon 13 NMR study of a repeating disaccharide of hyaluronan: the effects of temperature and counterion type. *Carbohydr. Res.* **1993**, *242*, 29–51;
 - (b) Inoue, Y.; Nagasawa, K. Preparation of methyl glycosides of di- and

- higher oligosaccharides from glycosaminoglycuronans by solvolysis with dimethyl sulfoxide containing methanol. *ibid*, **1980**, 85, 107–119.
12. Schmidt, R. R.; Kinzy, W. Anomeric-Oxygen Activation for Glycoside Synthesis: The Trichloroacetimidate Method. *Adv. Carbohydr. Chem. Biochem.* **1994**, 50, 21–123, and references cited therein. (This review, although devoted mainly to the trichloroacetimidate method, gives an excellent overview of the field of glycosidation.)
 13. Carter, M. B.; Petillo, P. A.; Anderson, L.; Lerner, L. E. The 1,4-Linked Disaccharide of Hyaluronan: Synthesis of Methyl 2-acetamido-2-deoxy- β -D-glucopyranosyl-(1-4)- β -D-glucopyranosiduronic Acid. *Carbohydr. Res.* **1994**, 258, 299–306.
 14. (a) Blatter, G.; Beau, J.-M.; Jacquinet, J.-C. The Use of 2-deoxy-2-trichloroacetamido-D-glucopyranose Derivatives in Syntheses of Oligosaccharides. *Carbohydr. Res.* **1994**, 260, 189–202;
(b) Coutant, C.; Jacquinet, J. -C. 2-Deoxy-2-trichloroacetamido-D-glucopyranose Derivatives in Oligosaccharide Synthesis: From Hyaluronic Acid to Chondroitin 4-sulfate Trisaccharides. *J. Chem. Soc., Perkin Trans. 1* **1995**, 1573–1581.
 15. Blatter, G.; Jacquinet, J.-C. 2-Deoxy-2-trichloroacetamido-D-glucopyranose Derivatives in Syntheses of Hyaluronic acid-related Tetra-, Hexa-, and Octasaccharides Having a Methyl β -D-glucopyranosiduronic Acid at the Reducing End. *Carbohydr. Res.* **1996**, 288, 109–125.

16. (a) Postema, M. H. D. *C-Glycoside Synthesis*; CRC press: London, 1995.
(b) Levy, D. E.; Tang, C. *The Chemistry of C-Glycosides*; Pergamon Press: Elmsford, NY, 1995.
17. (a) Schuster, M.; Blechert, S. Olefin Metathesis in Organic Chemistry. *Angew. Chem. Int. Ed. Engl.* **1997**, *36*, 2036–2056;
(b) Grubbs, R. H. and Chang, S. Recent Advances in Olefin Metathesis and Its Application in Organic Synthesis. *Tetrahedron* **1998**, *54*, 4413–4450.
18. (a) Postema, M. H. D.; Calimente, D. Convergent preparation of 1,6-linked C-disaccharides via olefin metathesis. *Tetrahedron Lett.* **1999**, *40*, 4755–4759.
(b) Calimente, D.; Postema, M. H. D. Preparation of C-1 glycals via olefin metathesis. A convergent and flexible approach to C-glycoside synthesis. *J. Org. Chem.* **1999**, *64*, 1770–1771.
19. (a) Martin, O.; Lai, W. A Concise Approach to β -(1–6)- and β,β -(1–1)-linked C-disaccharides. The Synthesis of C- β,β -tetrahalose Peracetate. *J. Org. Chem.* **1990**, *55*, 5188–5190;
(b) Martin, O.; Lai, W. Synthesis and conformational studies of β -(1–6)- and β,β -(1–1)-linked C-disaccharides. *J. Org. Chem.* **1993**, *58*, 176–185.
20. For a review, see Sinaÿ, P. Synthesis of oligosaccharide mimetics. *Pure Appl. Chem.* **1997**, *69*, 459–463.
21. (a) Ziegler, T.; Lemanski, G. Synthesis of β -Mannosides via Prearranged Glycosides. *Angew. Chem. Int. Ed. Engl.* **1998**, *37*, 3129–3132;

- (b) Huchel, U.; Schmidt, R. R. Intramolecular Glycoside Bond Formation—
a Rigid Spacer Concept for the Diastereoselective Linkage Between
Glycosyl Donor and Acceptor. *Tetrahedron Lett.* **1998**, 39, 7693–7694.
22. Hashimoto, H.; Endo, T.; Kajihara, Y. Synthesis of the First Tricomponent
Bisubstrate Analogue That Exhibits Potent Inhibition Against GlcNAc:β-1,4-
galactosyltransferase. *J. Org. Chem.* **1997**, 62, 1914–1915.
23. Vauzeilles, B.; Cravo, D.; Mallet, J.-M.; Sinaÿ, P. An Expeditious Synthesis
of a C-disaccharide Using a Temporary Ketal Connection. *Synlett.* **1993**,
522–524.
24. (a) Xin, Y. C.; Mallet, J.-M.; Sinaÿ, P. An Expeditious Synthesis of a C-
disaccharide Using a Temporary Silaketal Connection. *J. Chem. Soc.,
Chem. Commun.* **1993**, 864;
(b) Chénédé, A.; Perrin, E.; Rekaï, E. D.; Sinaÿ, P. Synthesis of C-
disaccharides by a Samarium Diiodide Induced Intramolecular Coupling
Reaction of Tethered Sugars. *Synlett*, **1994**, 420–422.
25. A discussion with Professor Amos Smith's group members during a visit in
May 1998.
26. (a) Hoffmann, M.; Kessler, H. A stereoselective synthesis of 2-acetamido-2-
deoxy-C-glucosides: Glycosyl dianions as key intermediates. *Tetrahedron
Lett.* **1994**, 35, 6067–6070;
(b) Hoffmann, M.; Kessler, H. Competition of Deprotonation and Tin-lithium
Exchange in the Generation of a Glycosyl Dianion. *ibid*, **1997**, 38,

1903–1906.

27. (a) Beau, J.-M.; Sinaÿ, P. Preparation and Reductive Lithiation of 2-deoxy-D-glucopyranosyl Phenylsulfones: a Highly Stereoselective Route to C-glycosides. *Tetrahedron Lett.* **1985**, 26, 6185–6188.
- (b) Beau, J.-M.; Sinaÿ, P. D-Glycopyranosyl phenylsulfones: their use in a stereocontrolled synthesis of *cis*-2,6-disubstituted tetrahydropyrans (β -D-C-glycosides). *ibid.* **1985**, 26, 6189–6192.
28. Anderson, L.; Mikkelsen L. M.; Beau J.-M.; Skrydstrup, T. Stereoselective Synthesis of α -C-Glucosamines via Anomeric Organosamarium Reagents. *Synlett.* **1998**, 1393–1395.
29. Calinaud, P.; Gelas, J. Synthesis of Isopropylidene, Benzylidene, and Related Acetals. In *Preparative Carbohydrate Chemistry*, Hanessian, S., Ed.; Marcel Dekker: New York, 1997, pp 3–34.
30. Johansson, R.; Samuelsson, B. Regioselective Reductive Ring-opening of 4-Methoxybenzylidene Acetals of Hexopyranosides. Access to a Noval Protecting-group Strategy. Part 1. *J. Chem. Soc., Perkin Trans. 1* **1984**, 2371–2374.
31. Timmers, C. M.; Turner, J. J.; Ward, C. M.; van der Marel, G. A.; Kouwijzer, M. L. C. E.; Grootenhuys, P. D. J.; van Boom, J. H. Synthesis and Biological Evaluation of a Backbone-modified Phytoalexin Elicitor. *Chem. Eur. J.* **1997**, 3, 920–929.
32. Lemieux, R. U.; Takeda, T.; and Chung, B. Y. Synthesis of 2-Amino-2-

- deoxy- β -D-glucopyranosides. Properties and use of 2-deoxy-2-phthalimidoglycosyl halides. ACS Symp. Ser. (Synth. Methods Carbohydr., Symp.) **1976**, *39*, 90–115.
33. Sim, M. M.; Kondo, H.; Wong, C.-H. Synthesis and Use of Glycosyl Phosphites: An Effective Route to Glycosyl Phosphates, Sugar Nucleotides, and Glycosides. *J. Am. Chem. Soc.* **1993**, *115*, 2260–2267.
34. Schmidt, R. R.; Jung, K.-H. Oligosaccharide Synthesis with Trichloroacetimidates. In *Preparative Carbohydrate Chemistry*; Hanessian, S., Ed.; Marcel Dekker: New York, 1997, pp 283–312.
35. Vedejs, E.; Fuchs, P. L. An Improved Aldehyde Synthesis from 1,3-Dithianes. *J. Org. Chem.* **1971**, *36*, 366–367.
36. Miljković, M.; Dropkin, D.; Hagel, P.; Habash-Marino, M. Removal of Sugar Dithioacetal Group with *N*-Bromosuccinimide. *Carbohydr. Res.* **1984**, *128*, 11–20.
37. Stork, G.; Zhao, K. A Simple Method of Dethioacetalization. *Tetrahedron Lett.* **1989**, *30*, 287–290.
38. Giannis, A.; Münster, P.; Sandhoff, K.; Steglich, W. Fragmentation and Wittig Olefination of Glucosamine Derivatives-A Simple Route to Open Chain Amino Sugars and Chiral Glycerols. *Tetrahedron Lett.* **1988**, *44*, 7177–7180.
39. Oldenziel, O. H.; van Leusen, D.; van Leusen, A. M. A General One-Step Synthesis of Nitriles from Ketones Using Tosylmethyl Isocyanide.

- Introduction of a One-Carbon unit. *J. Org. Chem.* **1977**, *42*, 3114–3118.
40. López, M.-T. G.; De las Heras, F.; Félix, A. S. Cyanosugars. IV. Synthesis of α -D-Glucopyranosyl and α -D-Galactopyranosyl Cyanides and Related 1,2-*cis* C-Glycosides. *J. Carbohydr. Chem.* **1987**, *6*, 273–279.
 41. Adam, W.; Chan, Y. -Y.; Cremer, D.; Gauss, J.; Scheutzw, D.; Schindler, M. Spectral and Chemical Properties of Dimethyldioxirane as Determined by Experiment and Ab Initio Calculations. *J. Org. Chem.* **1987**, *52*, 2800–2803.
 42. Vega-Pérez, J. M.; Candela, J. I.; Iglesias-Guerra, F. A Facile Synthesis of Saturated 2-Nitrosugar Derivatives. *J. Org. Chem.* **1997**, *62*, 6608–6611.
 43. Drew, K. N.; Gross, P. H. C-glycoside Syntheses II: Henry Condensations of 4,6-O-alkylidene pyranoses with a 1,3-Proton Transfer Catalyst-A Route to Blocked Aminomethyl-C-glycosides. *Tetrahedron* **1991**, *47*, 6113–6126.
 44. A 4-C-lithiomethyl intermediate might strong enough to add onto the deactivated aldehyde, but protection groups on the nitrogen seem incompatible. For 4-C-lithiomethyl intermediate, see: Khan, A.T.; Sharma, P.; Schmidt, R. R. Synthesis of C-disaccharides. An Unusual Ring Closure Reaction. *J. Carbohydr. Chem.* **1995**, *14*, 1353–1367.
 45. Jiang, L.; Chan, T.-H. Borane/Bu₂BOTf: A Mild Reagent for the Regioselective Reductive Ring Opening of Benzylidene Acetals in Carbohydrates. *Tetrahedron Lett.* **1998**, *39*, 355–358.
 46. (a) Dess, D. B.; J. Martin, C. Readily Accessible 12-I-5 Oxidant for the Conversion of Primary and Secondary Alcohols to Aldehydes and Ketones.

- J. Org. Chem.* **1983**, *48*, 4155–4156;
- (b) Ireland, R. E.; Liu, L. An Improved Procedure for the Preparation of the Dess-Martin Periodinane. *ibid*, **1993**, *58*, 2899;
- (c) Meyer, S. D.; Schreiber, S. L. Acceleration of the Dess-Martin Oxidation by Water. *ibid*, **1994**, *59*, 7549–7552.
47. Molino, B. F.; Cusmano, J.; Mootoo, D. R.; Faghih, R.; Fraser-Reid, B. Stereoselectivity in the Hydroboration of C4-C-Methylene Groups. *J. Carbohydr. Chem.* **1987**, *6*, 479–493.
48. For general review, see: (a) Ley, S. V.; Norman, J.; Griffith, W. P.; Marsden, S. P. Tetrapropylammonium perruthenate, $\text{Pr}_4\text{N}^+\text{RuO}_4^-$, TPAP: A Catalytic Oxidant for Organic Synthesis. *Synthesis* **1994**, 639–666.
- For oxidation of an alcohol to a carboxylic acid in the presence of an olefin, see: (b) Xu, Z.; Johannes, C. W.; Houri, A. F.; La, D. S.; Cogan, D. A.; Hofilena, G. E.; Hoveyda, A. H. Application of Zr-catalyzed Carbomagnesation and Mo-catalyzed Macrocyclic Ring Closing Metathesis in Asymmetric Synthesis. Enantioselective Total Synthesis of Sch 38516 (Fluvirucin B1). *J. Am. Chem. Soc.* **1997**, *119*, 10302–10316.
49. (a) Bal, B. S.; Childers, W. E. Jr.; Pinnick, H. W. Oxidation of α,β -unsaturated Aldehydes. *Tetrahedron* **1981**, *37*, 2091–2096;
- (b) Marshall, J. A.; Johns, B. A. Total Synthesis of (+)-discordermolide. *J. Org. Chem.* **1998**, *63*, 7885–7892.
50. Epp, J. B.; Widlanski, T. S. Facile Preparation of Nucleoside-5'-carboxylic

Acids. *J. Org. Chem.* **1999**, *64*, 293–295.

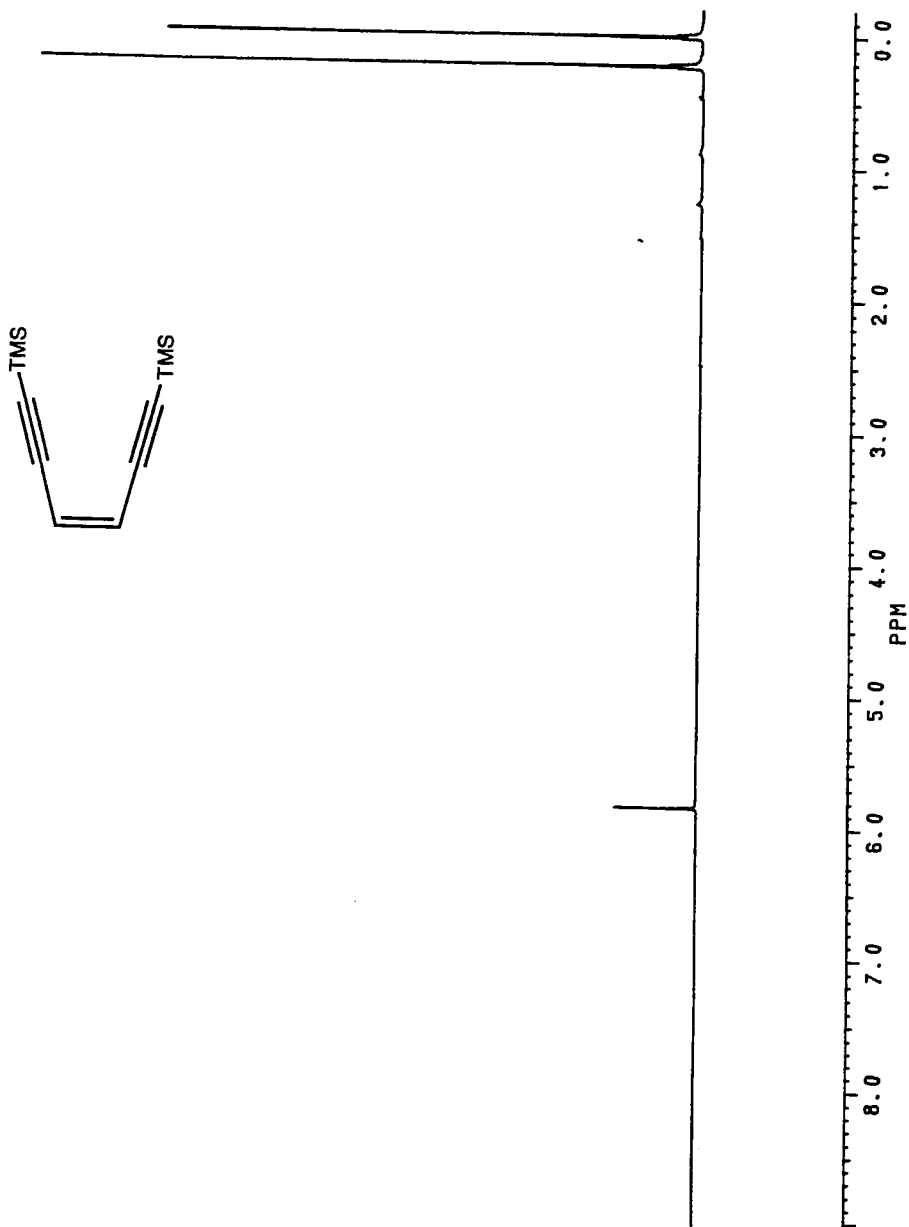
51. $(\text{COCl})_2/\text{CH}_2\text{Cl}_2$ /catalytic amount of DMF might work. However, It was not tried during the time-frame of this dissertation.
52. Mulzer, J. Synthesis of Esters, Activated Esters and Lactones. In *Comprehensive Organic Synthesis*, B. M. Trost, Ed., Pergamon Press, 1991, Vol. 6, pp 323–380.
53. (a) Schmidt, R. R.; Preuss, R. Synthesis of Carbon Bridged C-disaccharides. *Tetrahedron Lett.* **1989**, *30*, 3409–3412;
(b) Daly, S. M.; Armstrong, R. W. Stereoselective Synthesis of “Linear” C-disaccharides. *Tetrahedron Lett.* **1989**, *30*, 5713–5716.
54. Nilsson, M.; Svahn, C. M.; Westman, J. Synthesis of the methyl glycosides of a tri- and a tetra-saccharide related to heparin and heparan sulfate. *Carbohydr. Res.* **1993**, *246*, 161–172.
55. Slaghek, T. M.; Hypponen, T. K.; Ogawa, T.; Kamerling, J. P.; Vliegthart, J. F. G. Synthesis of hyaluronic acid related di- and tetra-saccharides having a glucuronic acid at the reducing end. *Tetrahedron. Asymmetry* **1994**, *5*, 2291–2301.
56. Hasegawa, A.; Fletcher, H. G., Jr. Behavior of some aldoses with 2,2-dimethoxypropane-*N,N*-dimethylformamide-*p*-toluenesulfonic acid. I. At room temperature (25. Deg.). *Carbohydr. Res.* **1970**, *29*, 209–222.
57. Mehta, S.; Pinto, B. M. Phenylselenoglycosides as novel, versatile glycosyl donors. Selective activations over thioglycosides. *Tetrahedron Lett.* **1991**,

32, 4435–4438.

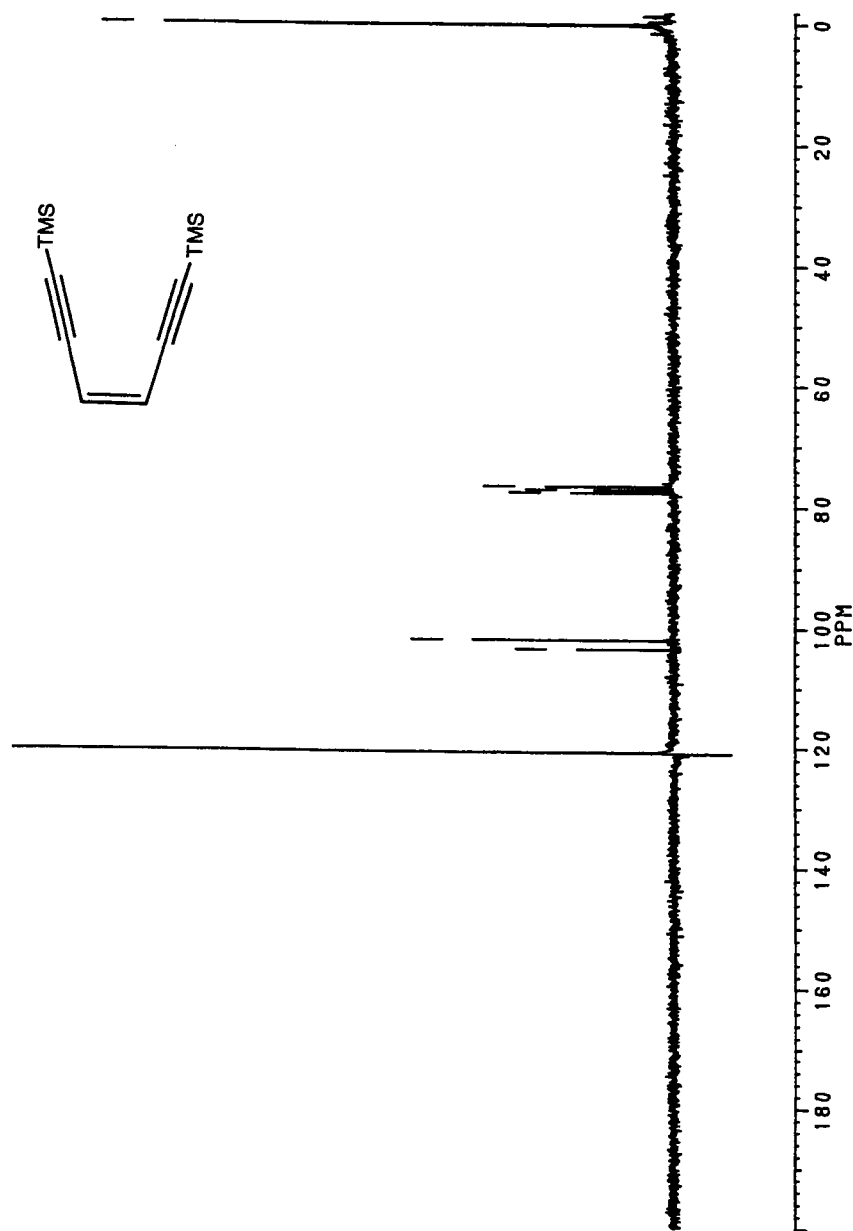
58. Tietze, L. F.; Fischer, R.; Guder, H. J. Glycoside syntheses. 4. Simple and stereoselective synthesis of α - and β -phenyl glycosides. *Tetrahedron Lett.* **1982**, 23, 4661–4664.
59. Fish, P. V.; Johnson, W. S. The First Examples of Nonenzymic, Biomimetic Polyene Pentacyclizations. Total Synthesis of the Pentacyclic Triterpenoid Sophoradiol. *J. Org. Chem.* **1994**, 59, 2324–2335.

APPENDIX

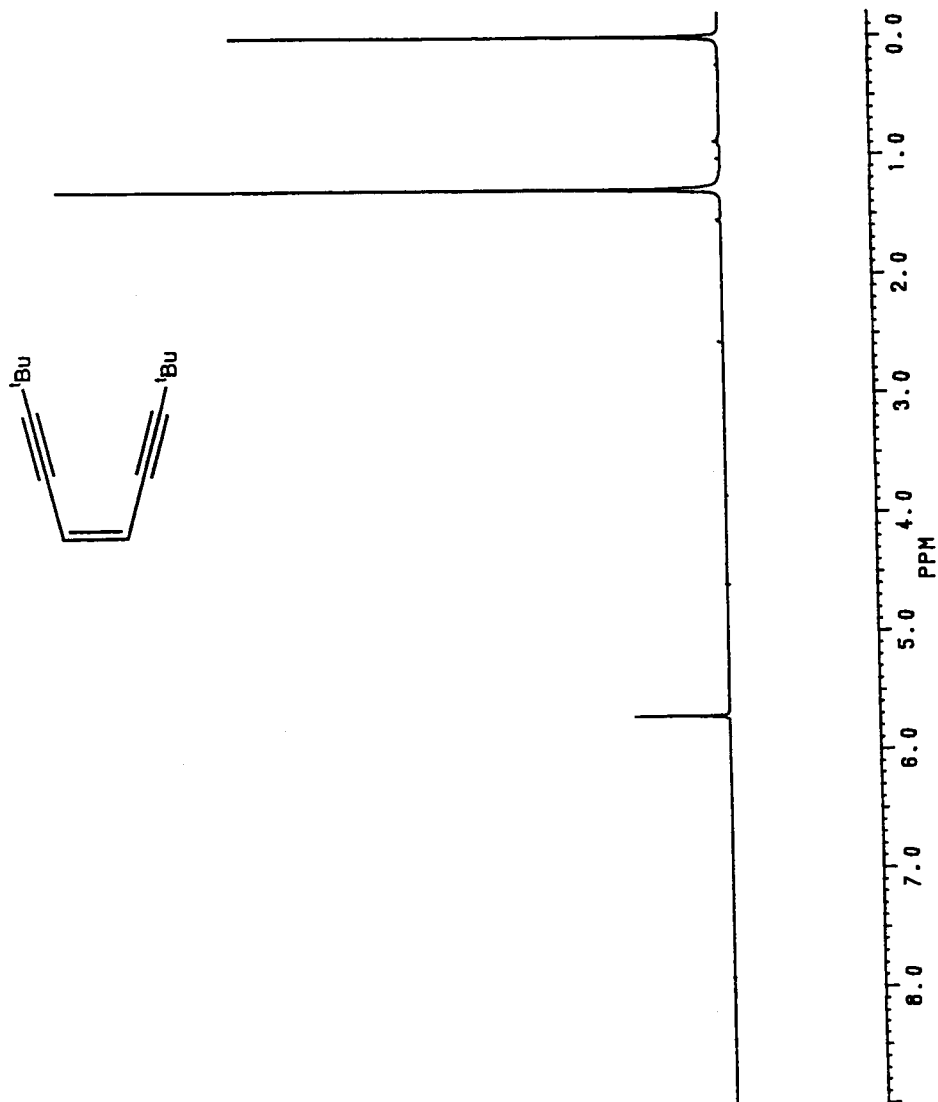
^1H and ^{13}C NMR SPECTRA OF SELECTED COMPOUNDS



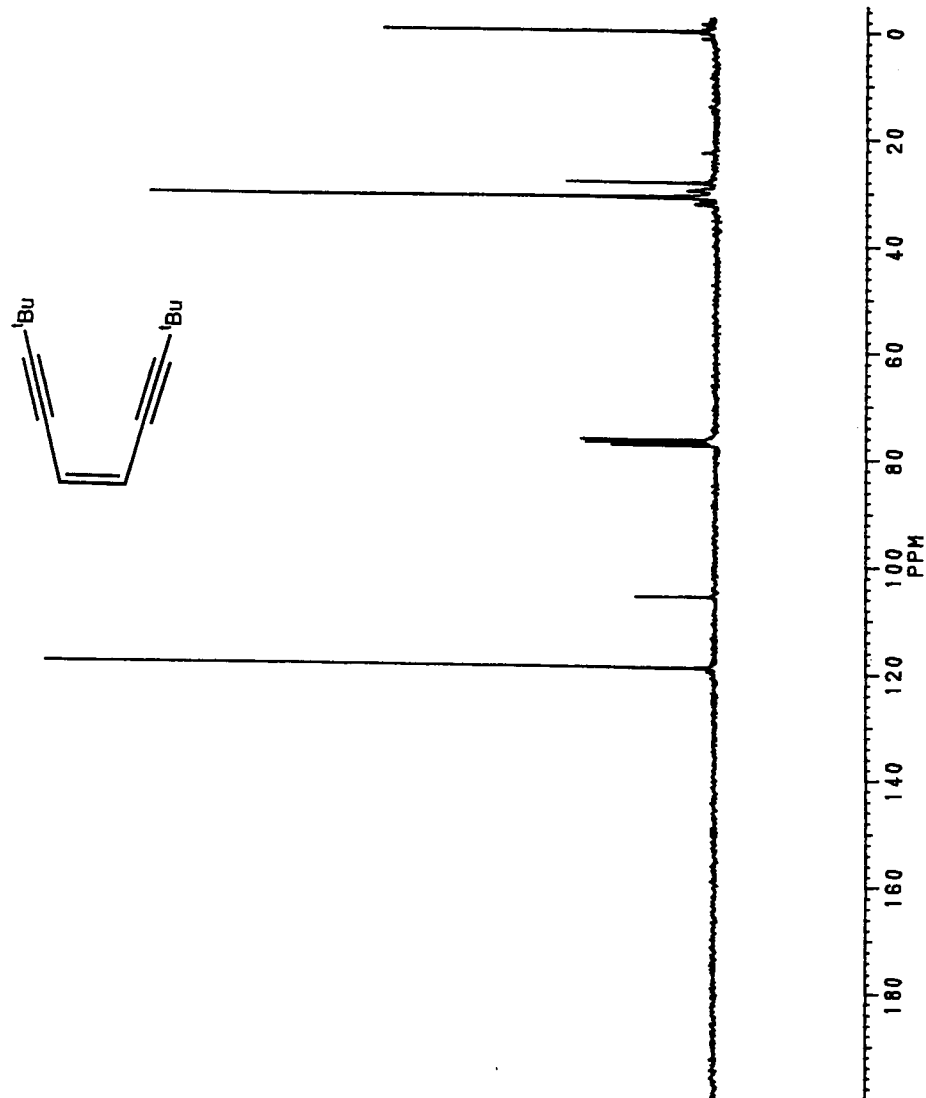
^1H NMR (250 MHz, CDCl_3) spectrum of *cis*-1,6-bis(trimethylsilyl)-3-hexene-1,5-diyne (2).



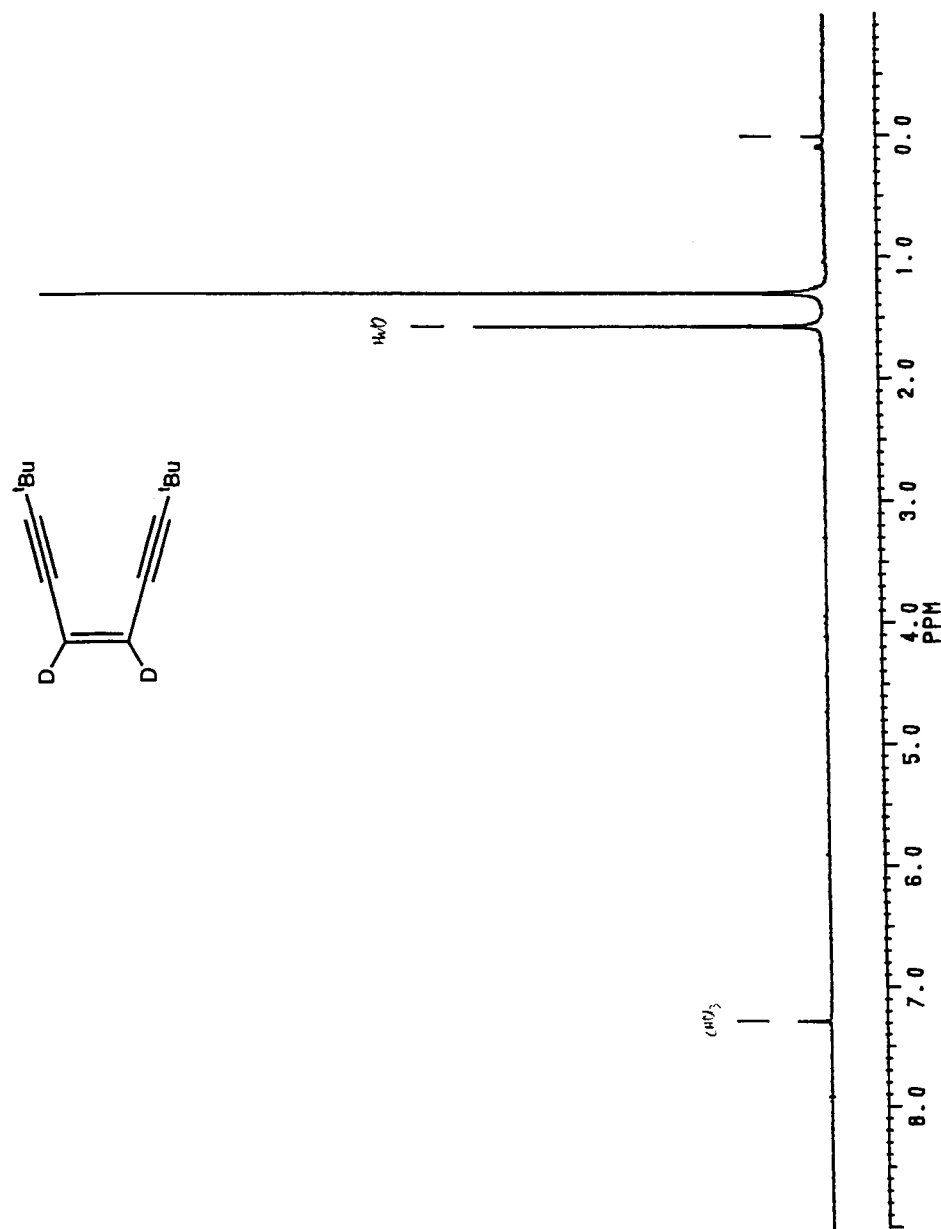
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *cis*-1,6-bis(trimethylsilyl)-3-hexene-1,5-diyne (2)



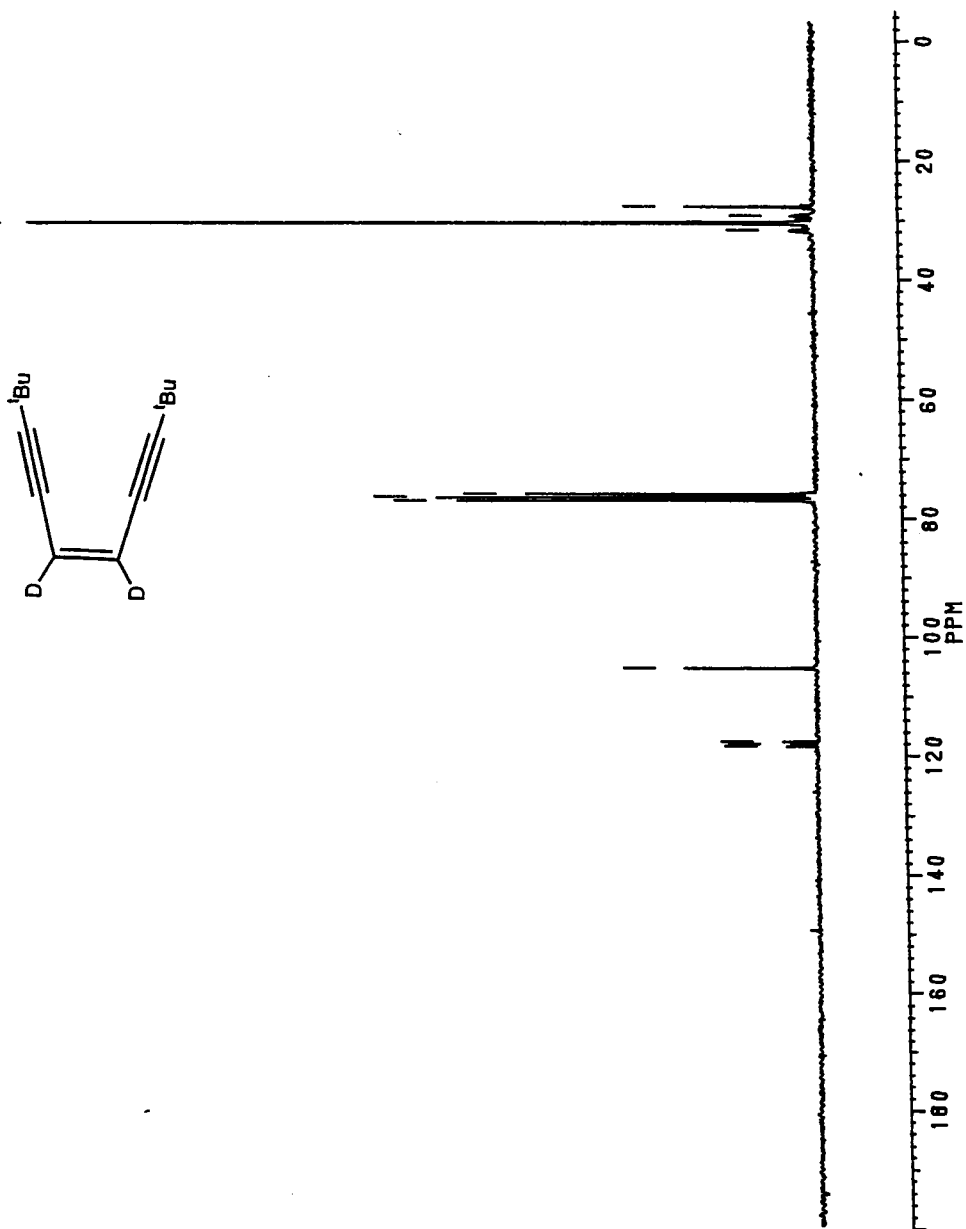
¹H NMR (250 MHz, CDCl₃) spectrum of *cis*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (4).



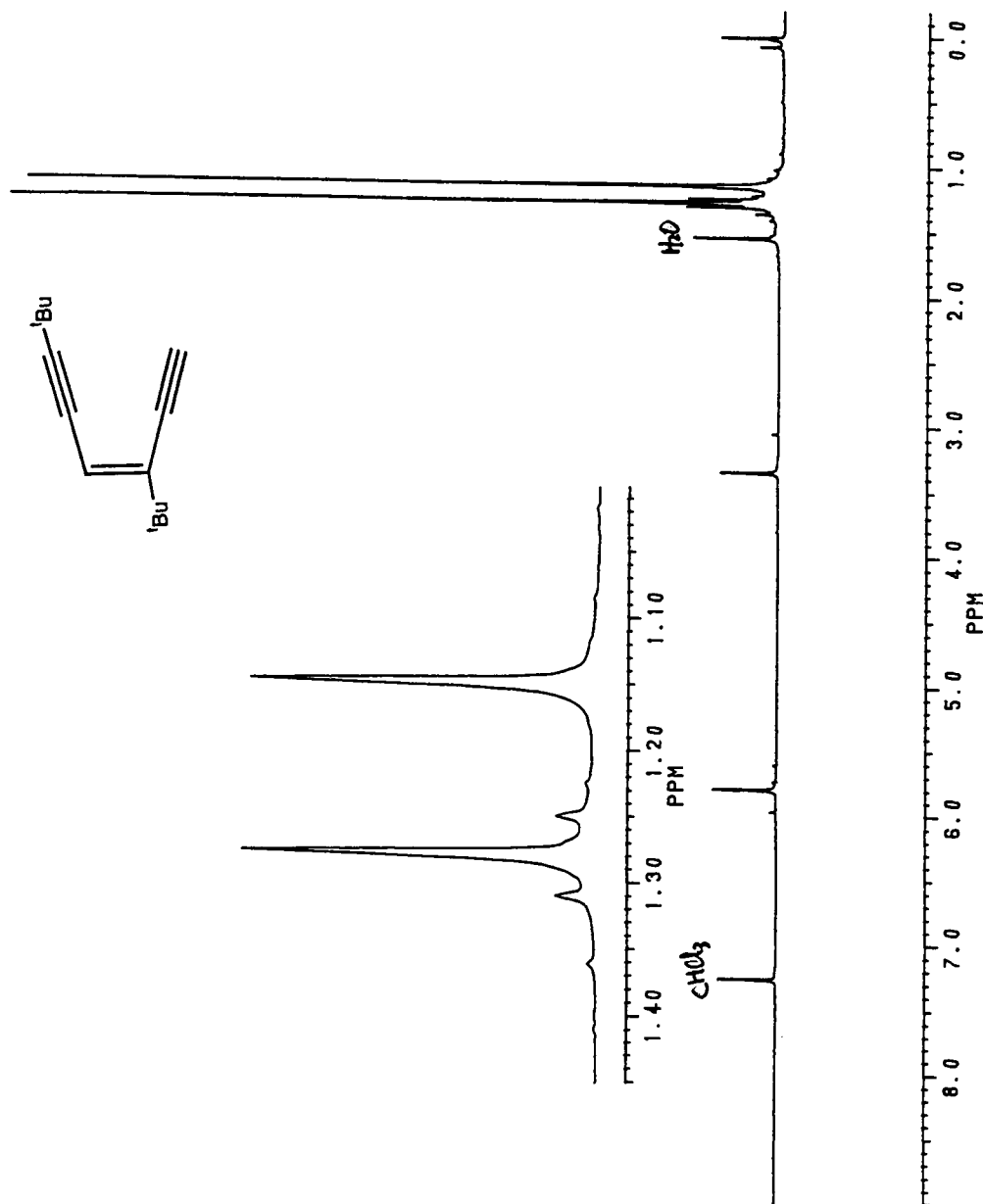
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *cis*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (4).



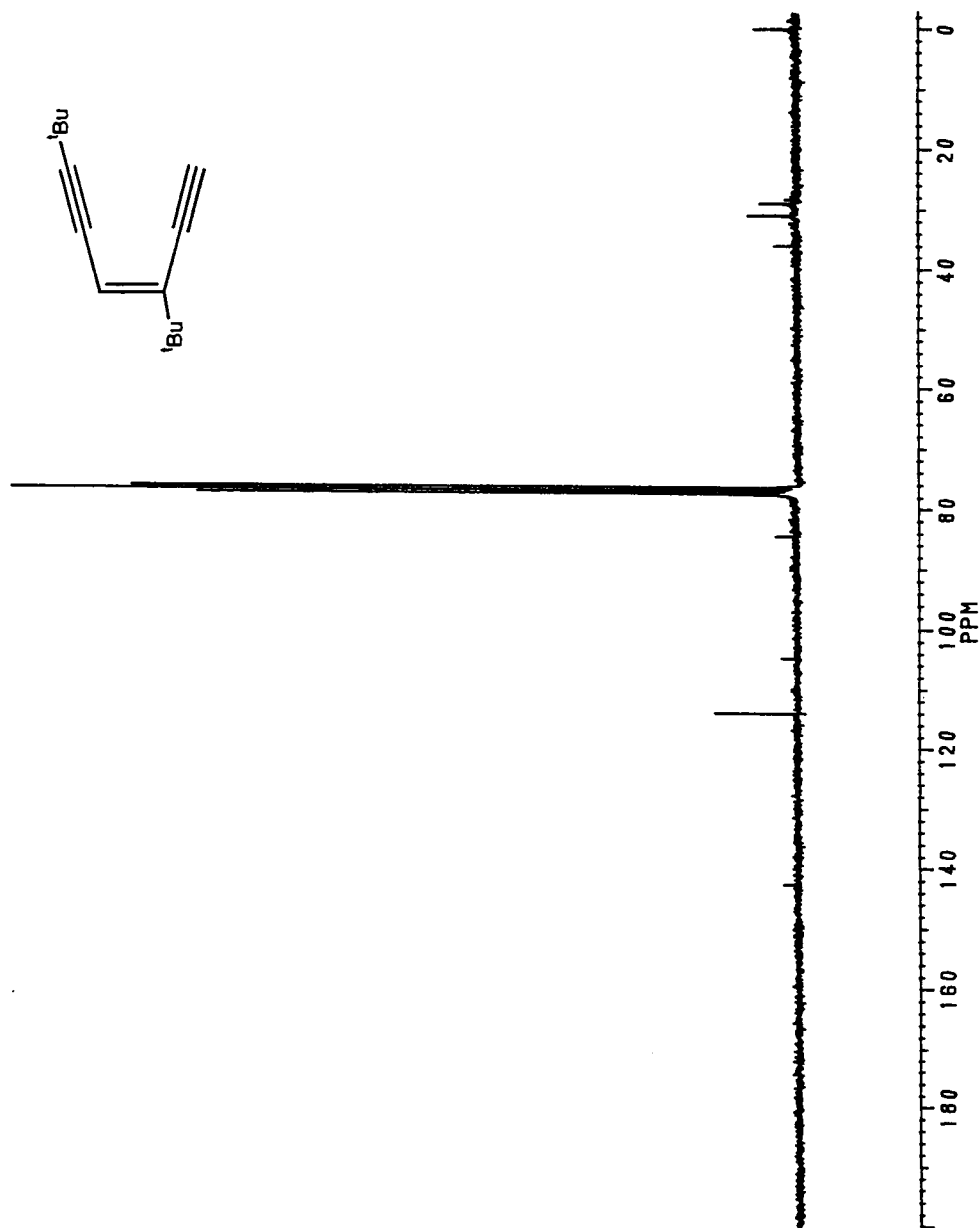
¹H NMR (250 MHz, CDCl₃) spectrum of *cis*-5,6-dideuterio-2,2,9,9-tetramethyl-5-decene-3,7-diyne (**5**).



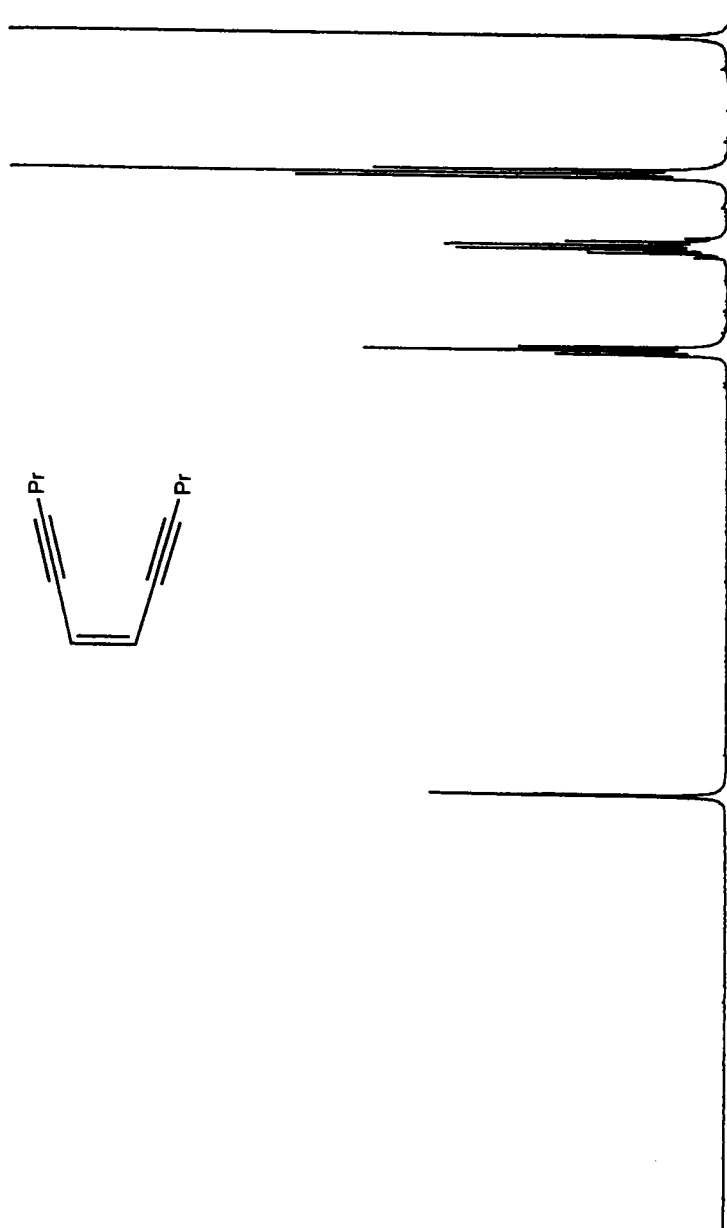
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *cis*-5,6-dideuterio-2,2,9,9-tetramethyl-5-decene-3,7-diyne (5).



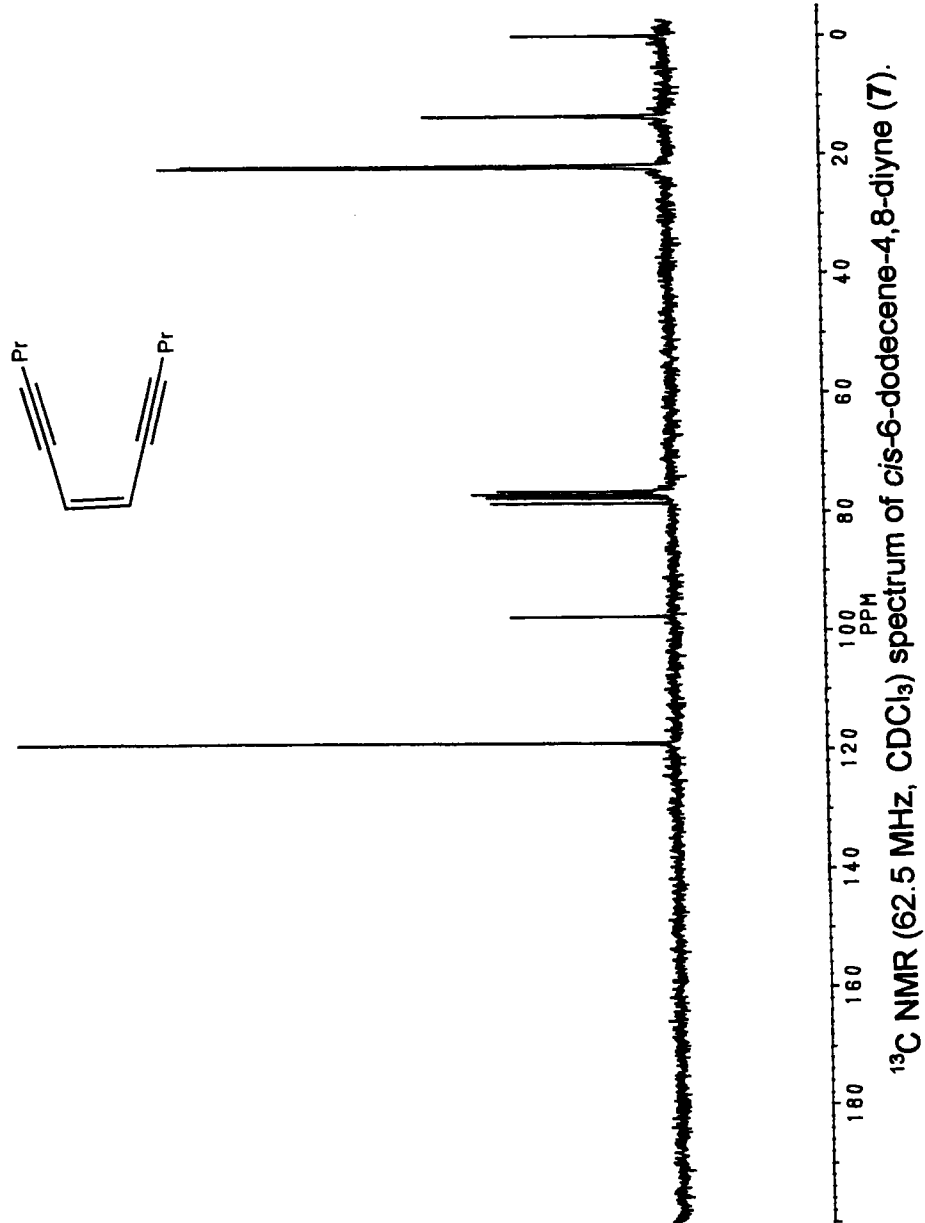
^1H NMR (250 MHz, CDCl_3) spectrum of *cis*-3-*tert*-butyl-7,7-dimethyl-1,5-diyne (6).

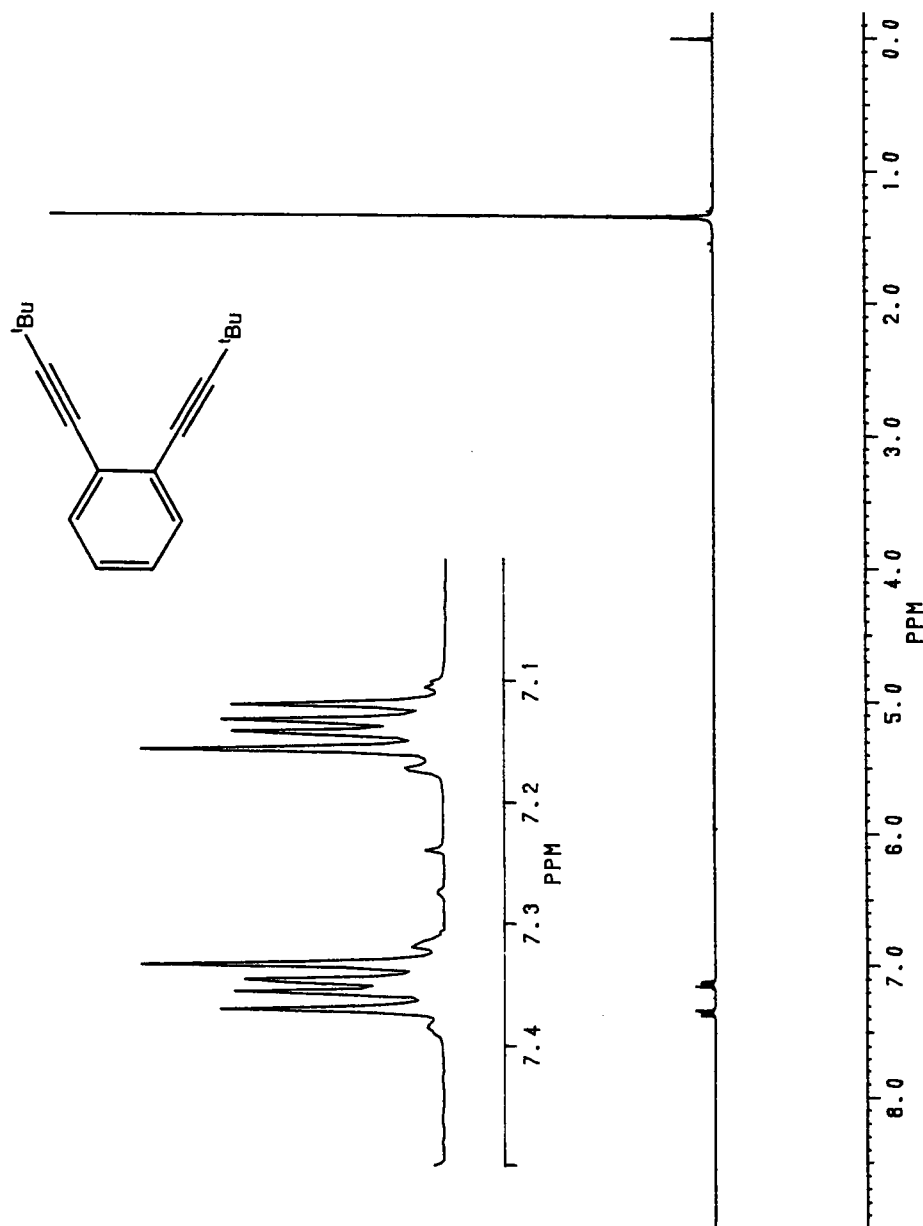


^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *cis*-3-*tert*-butyl-7,7-dimethyl-1,5-diyne (**6**).

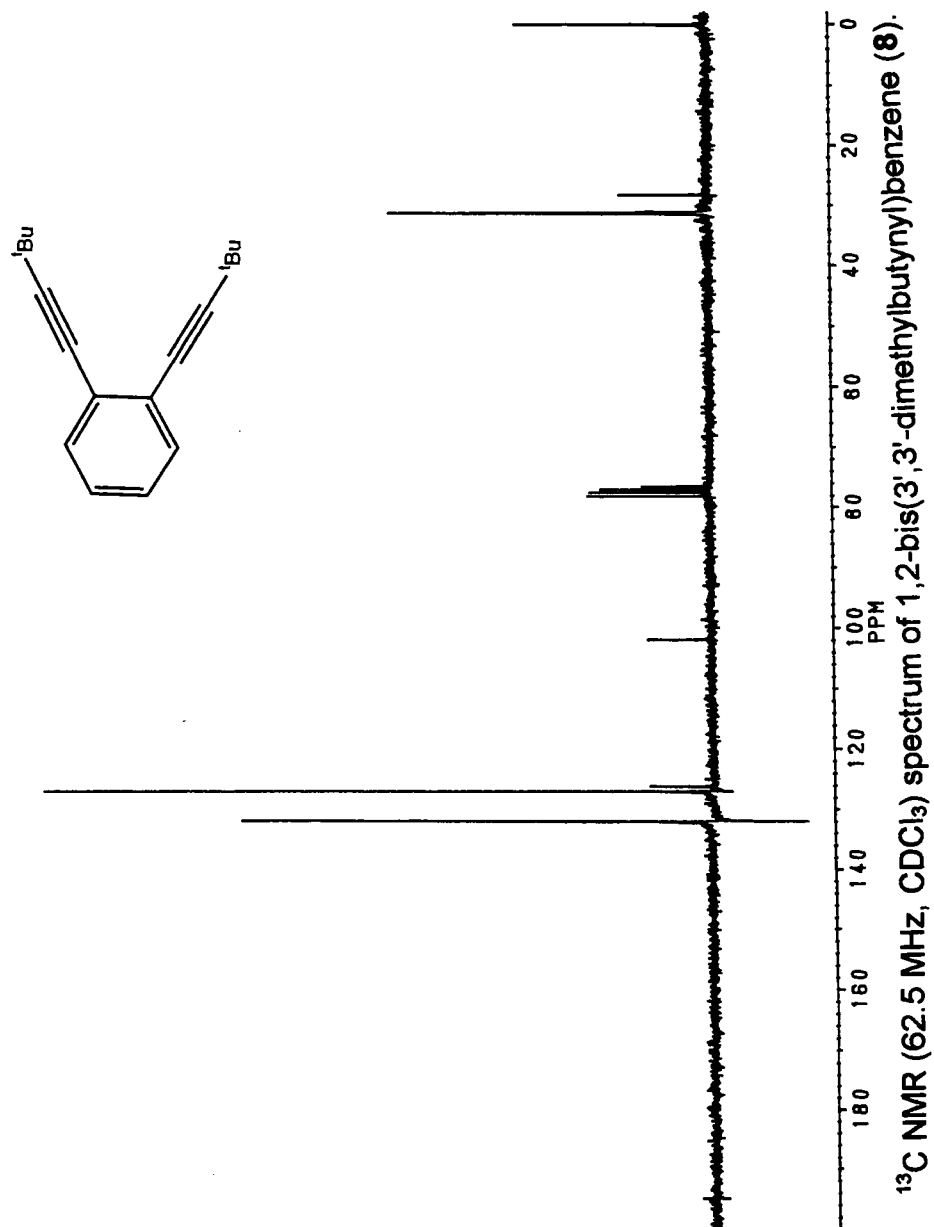


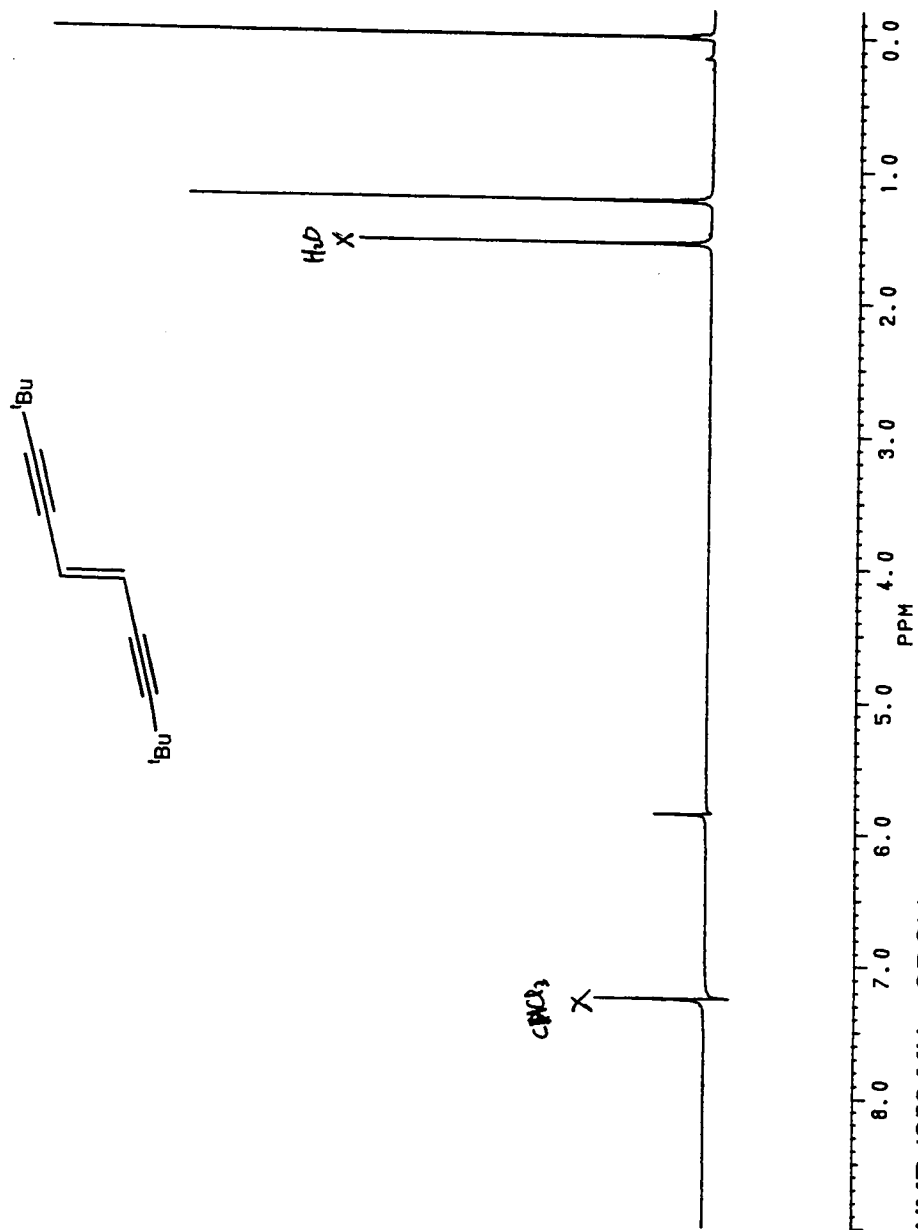
^1H NMR (250 MHz, CDCl_3) spectrum of *cis*-6-dodecene-4,8-diyne (7).



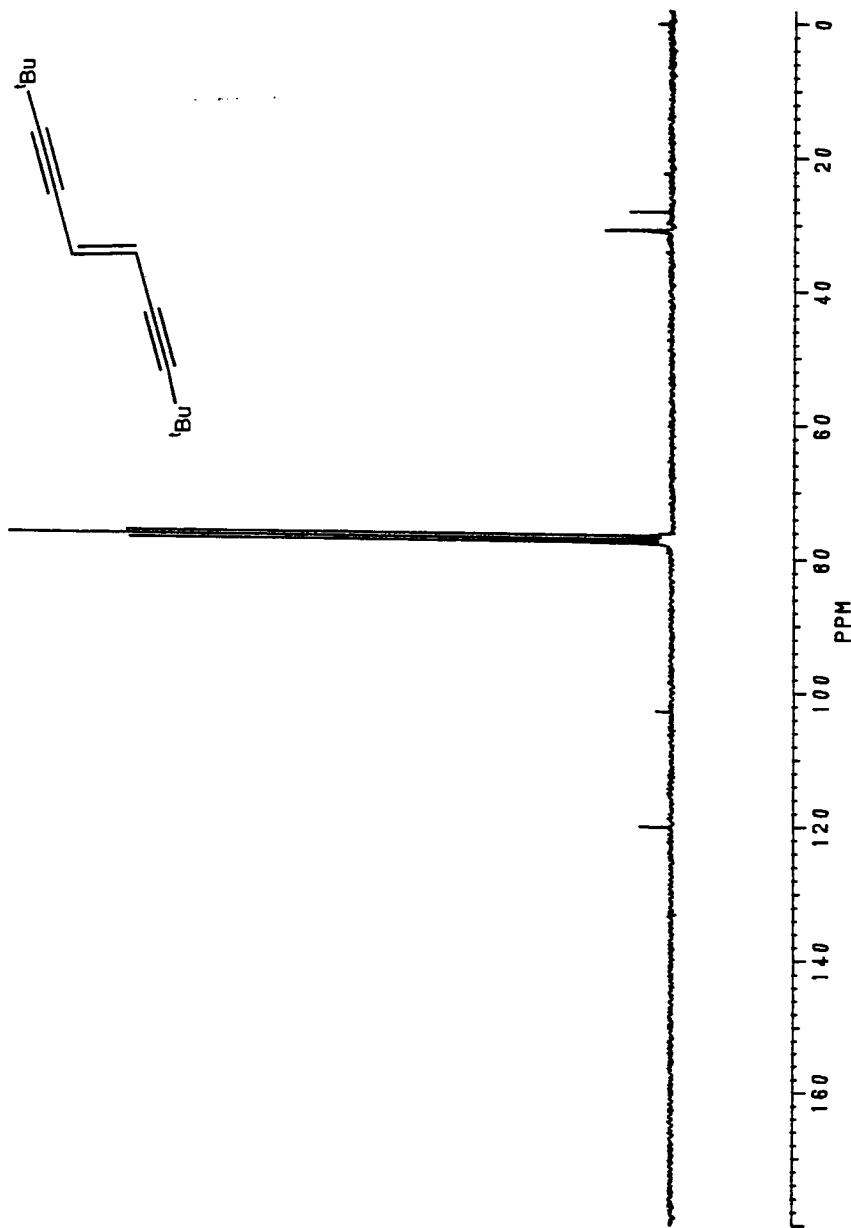


^1H NMR (250 MHz, CDCl_3) spectrum of 1,2-bis(3',3'-dimethylbutynyl)benzene (**8**).

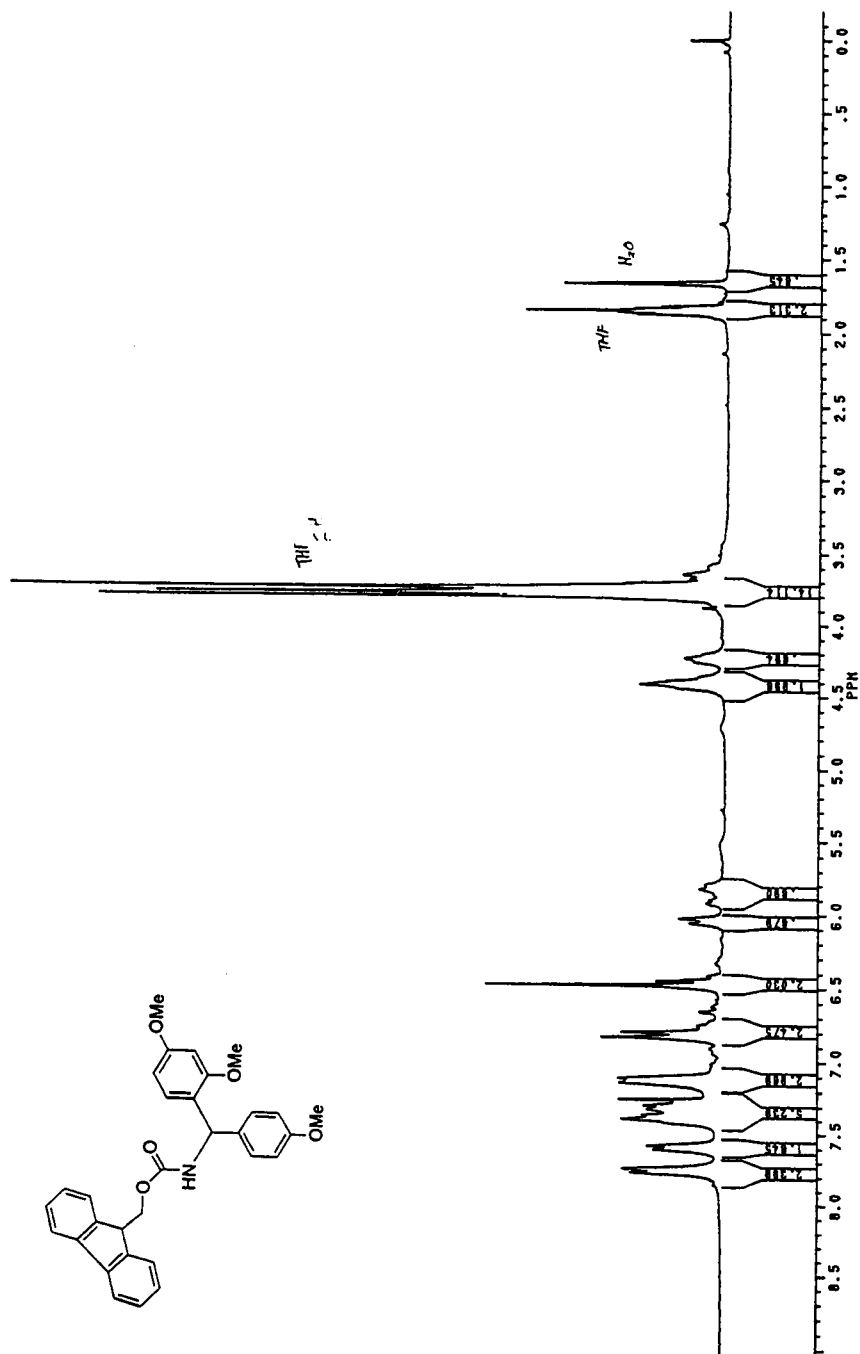




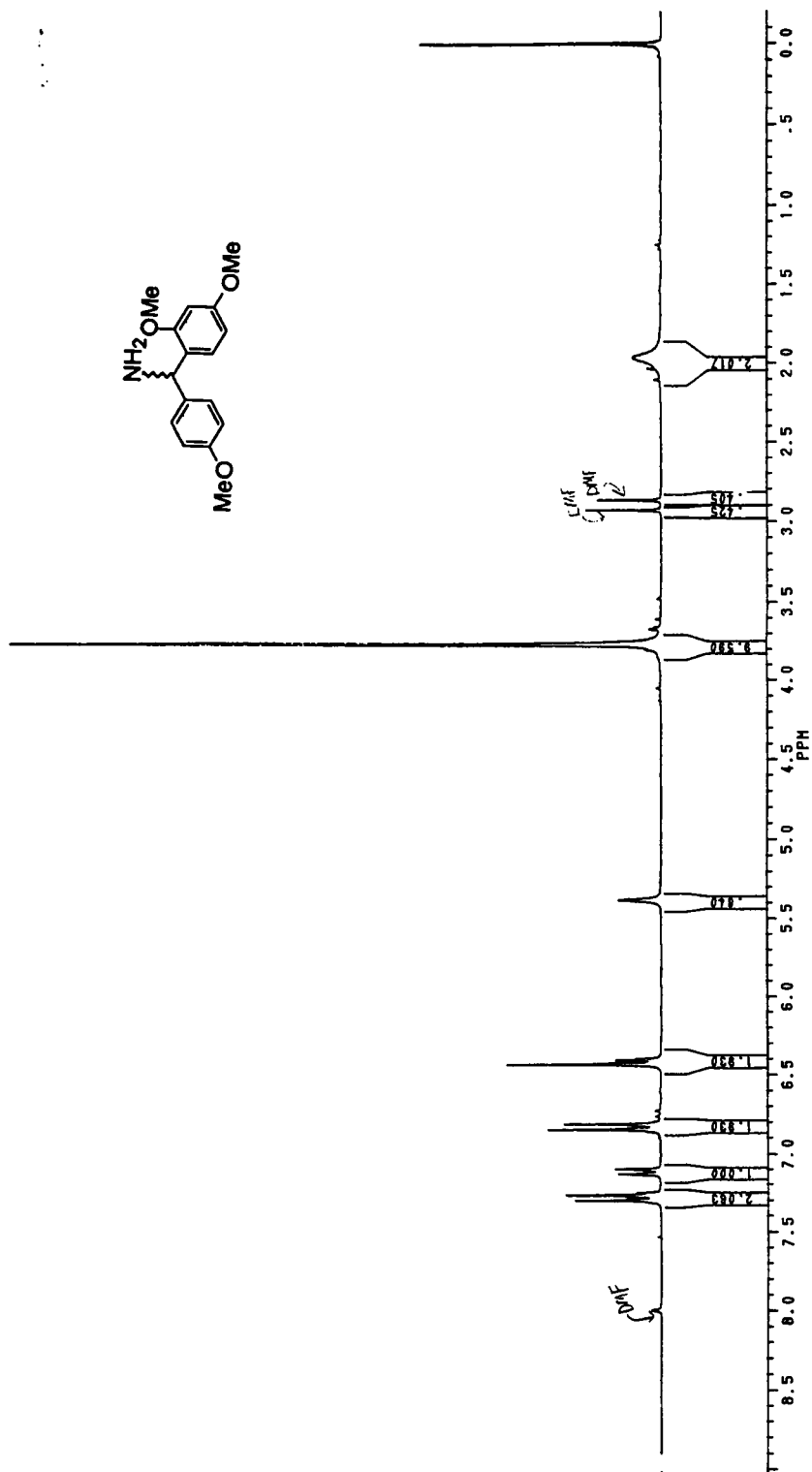
^1H NMR (250 MHz, CDCl_3) spectrum of *trans*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (10).



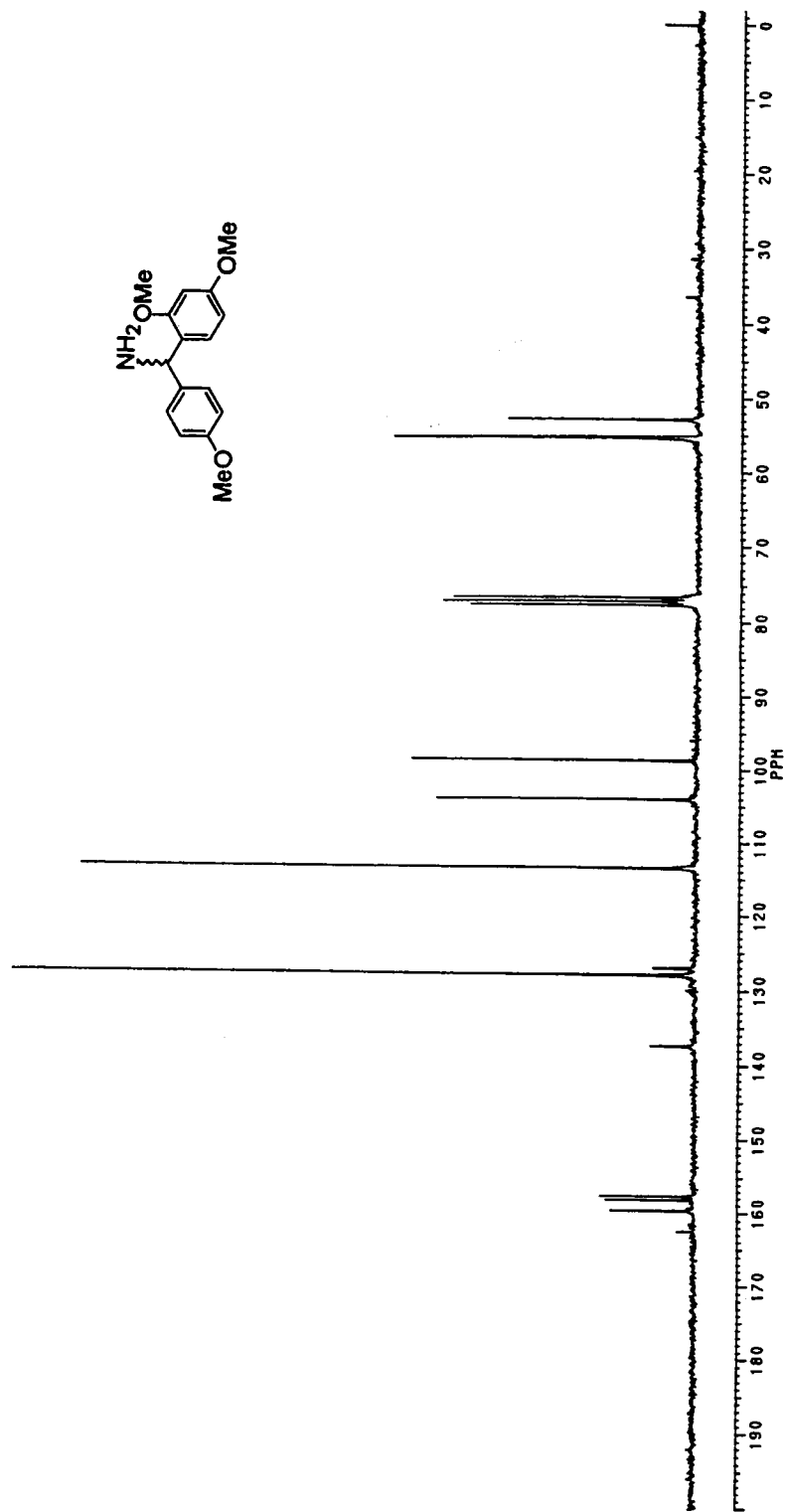
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *trans*-2,2,9,9-tetramethyl-5-decene-3,7-diyne (10).



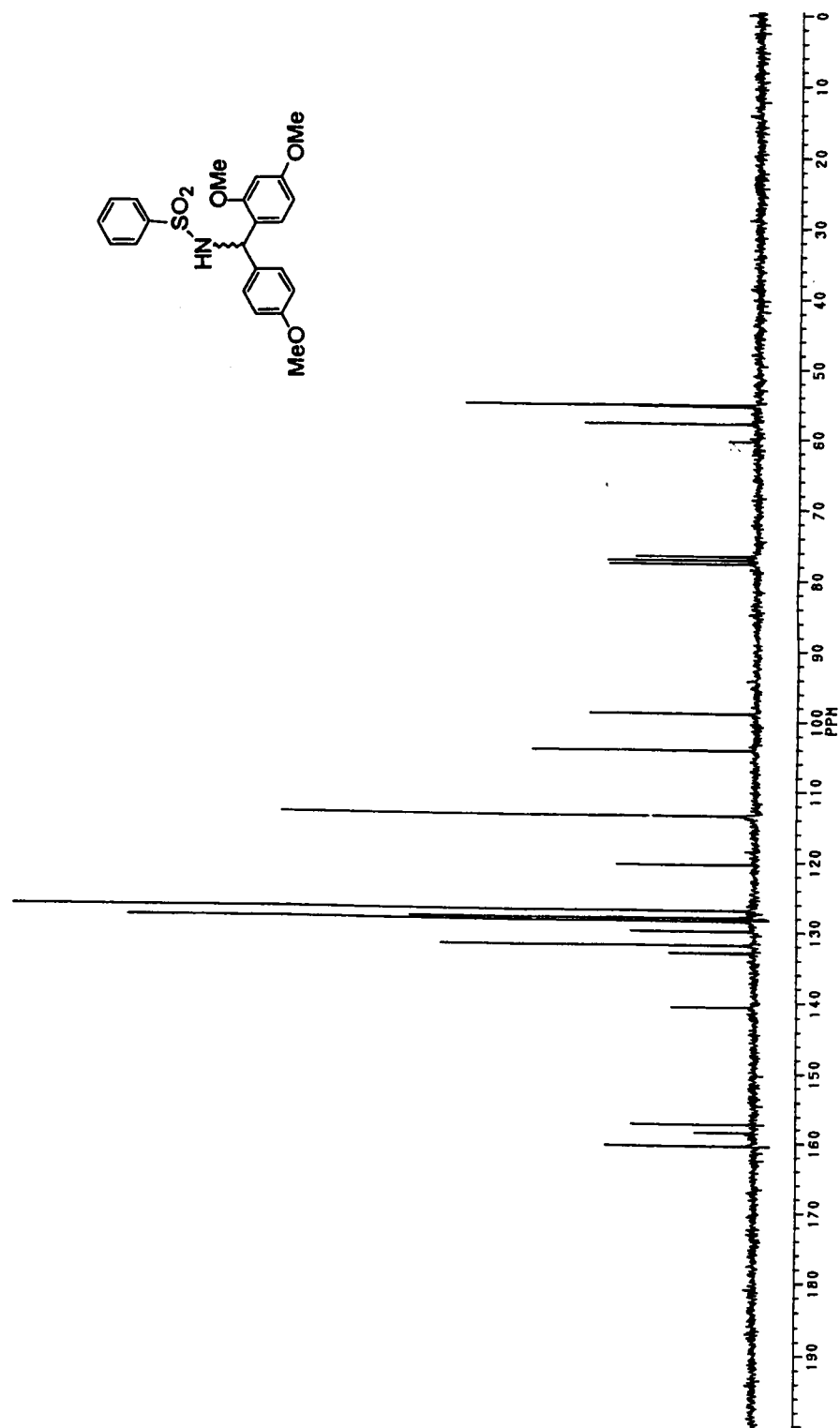
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-(2,4-dimethoxyphenyl)-4'-methoxyphenyl)methyl-9-fluorenylmethylcarbamate (**28**).



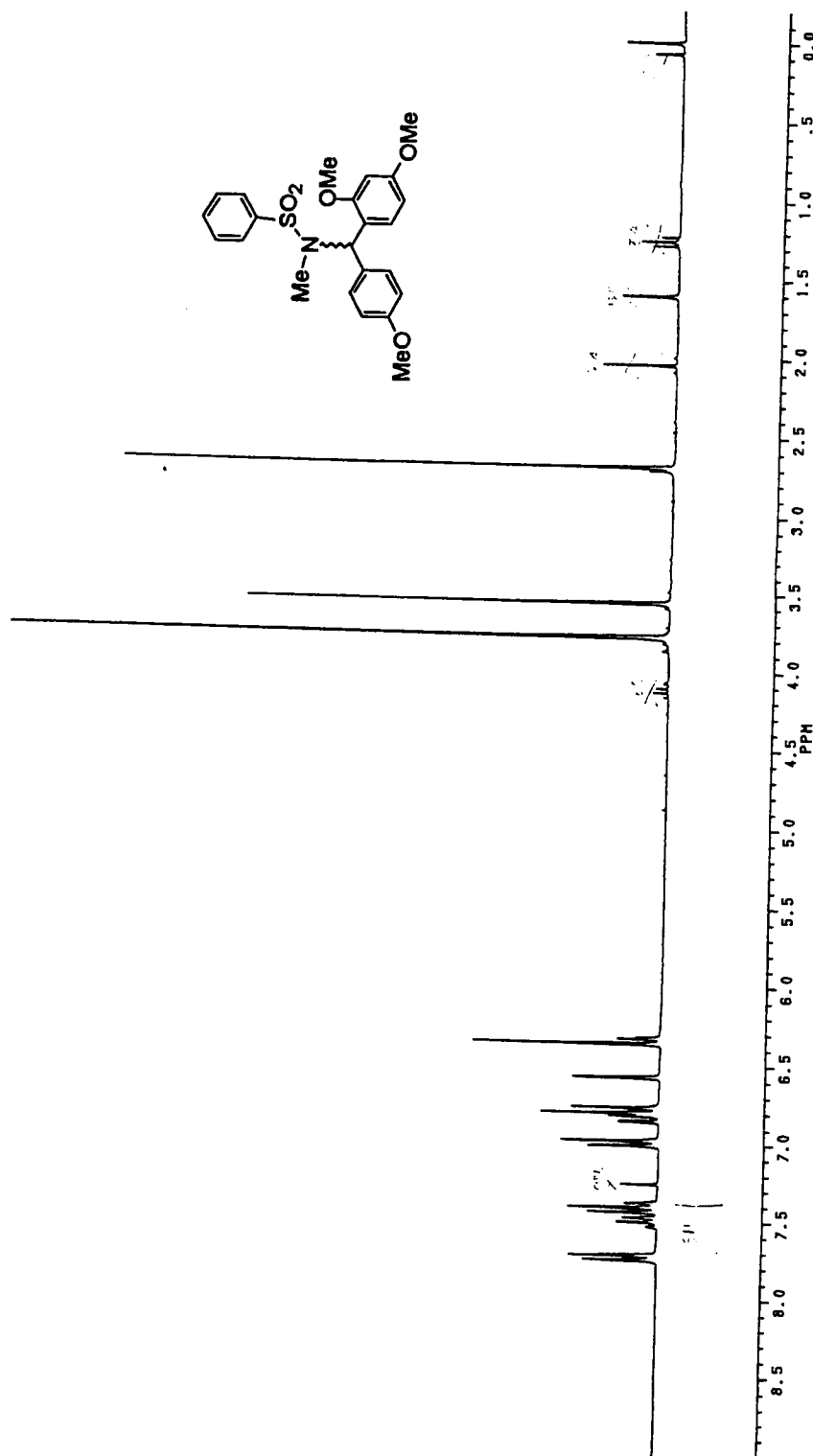
¹H NMR (250 MHz, CDCl₃) spectrum of (2,4-dimethoxyphenyl)-(4-methoxyphenyl)methylamine (29).



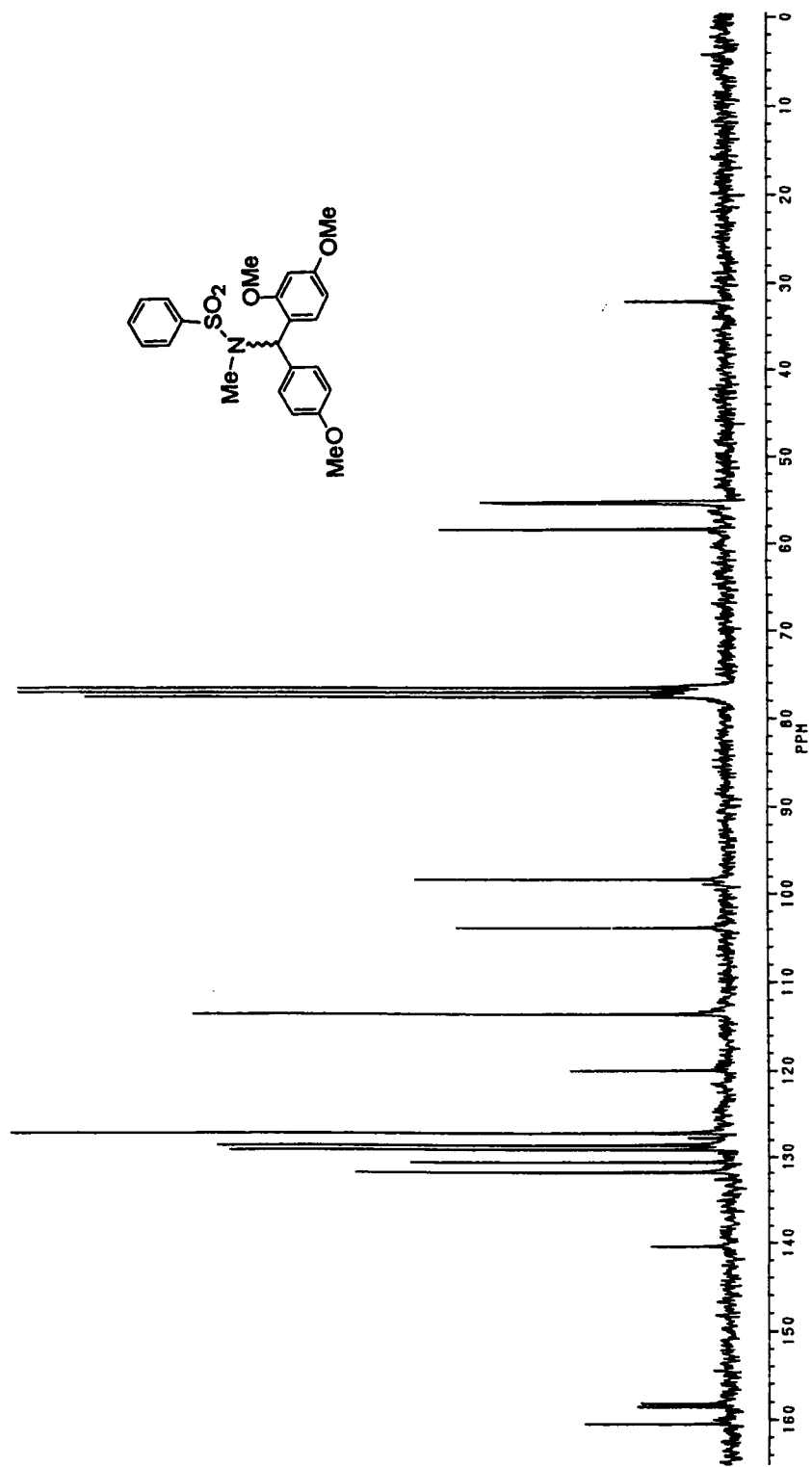
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of (2,4-dimethoxyphenyl)-(4-methoxyphenyl)methylamine (29).



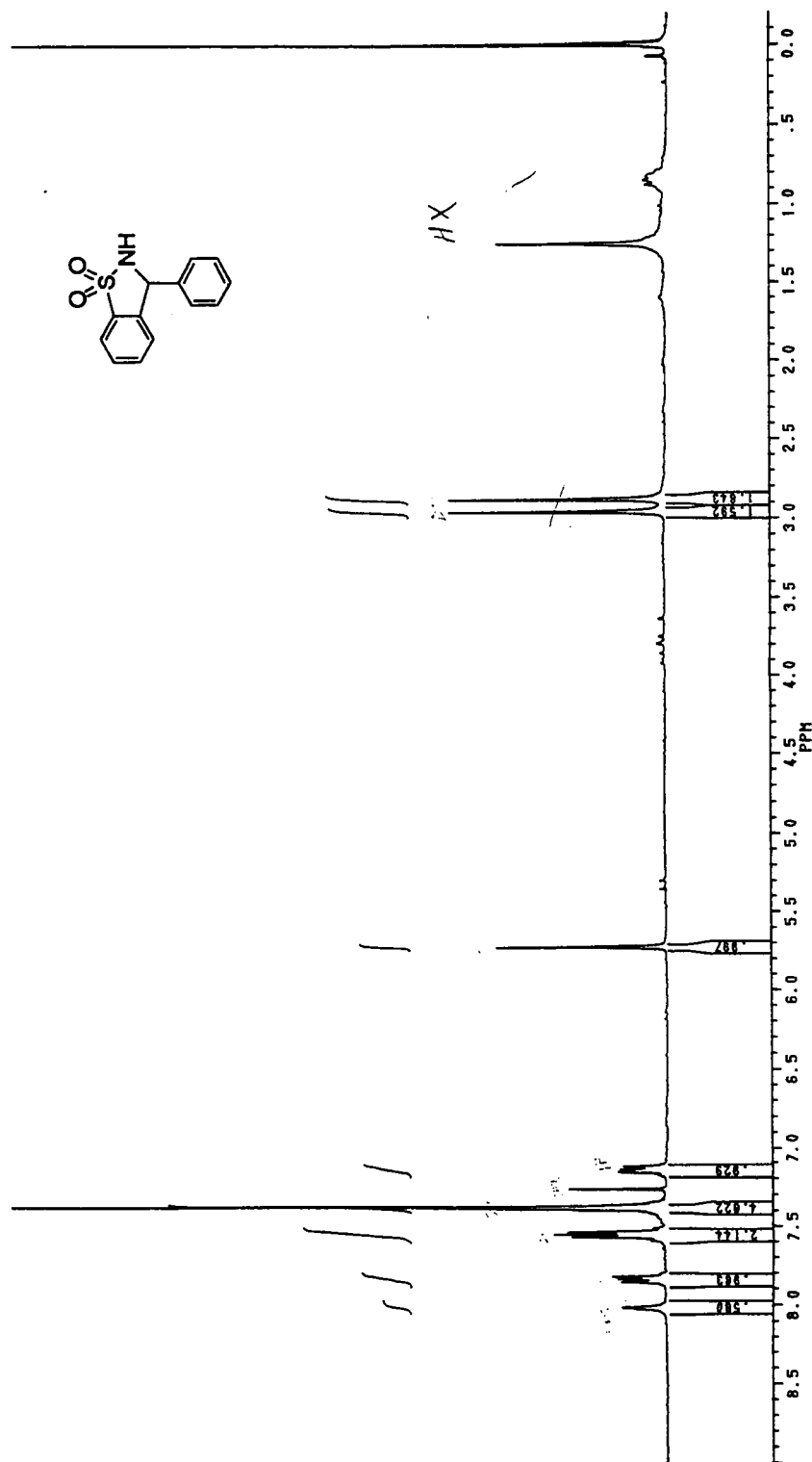
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *N*-[(2,4-dimethoxyphenyl)-(4'-methoxyphenyl)methyl]benzenesulfonamide (30).



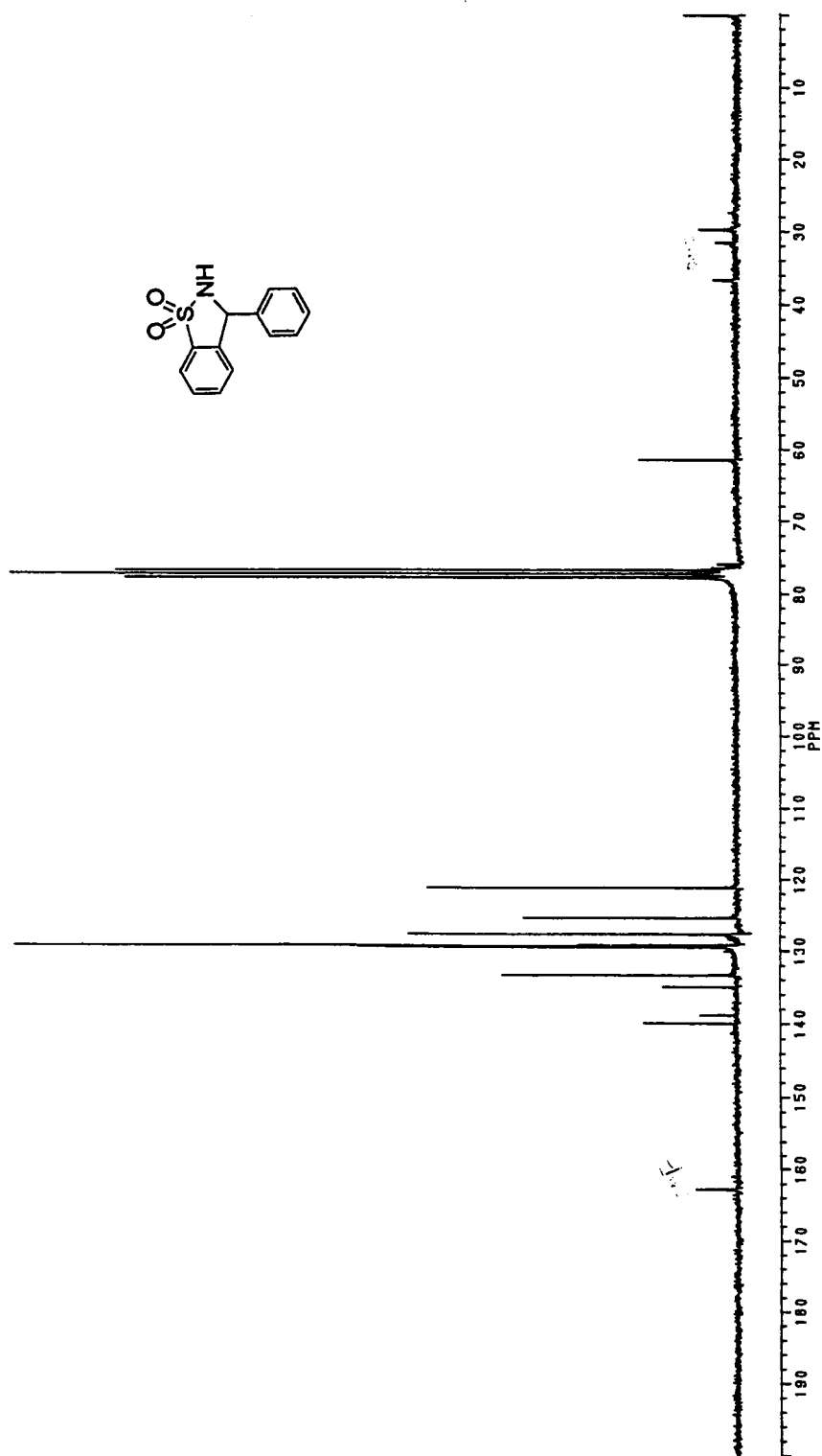
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-[(2,4-dimethoxyphenyl)-(4'-methoxyphenyl)methyl]-*N*-methylbenzenesulfonamide (**31**).



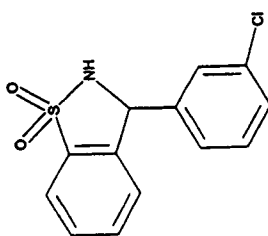
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *N*-[2,4-dimethoxyphenyl]-(4'-methoxyphenyl)methyl]-*N*-methylbenzenesulfonamide (**31**).



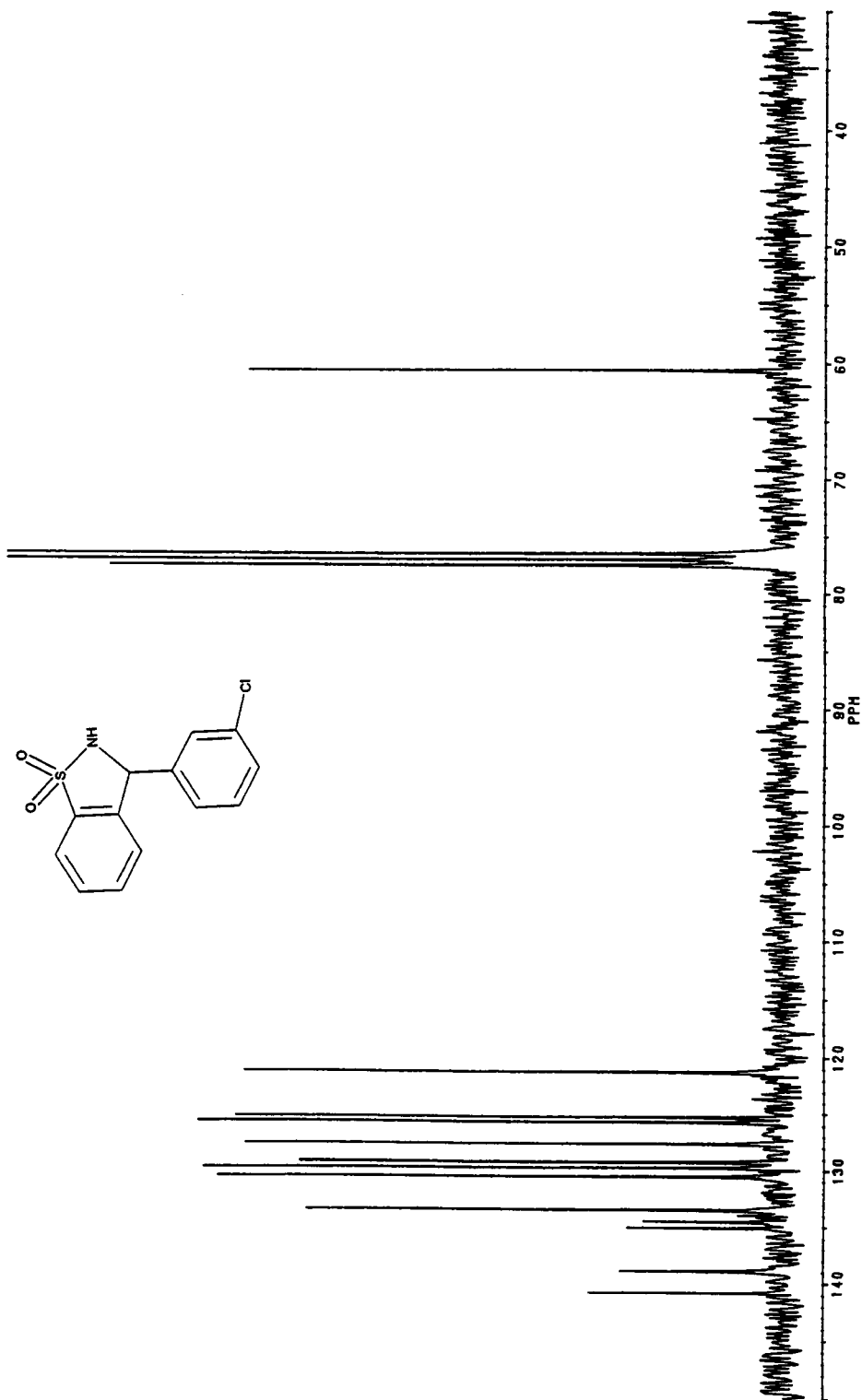
¹H NMR (250 MHz, CDCl₃) spectrum of (±)-3-phenyl-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34a).



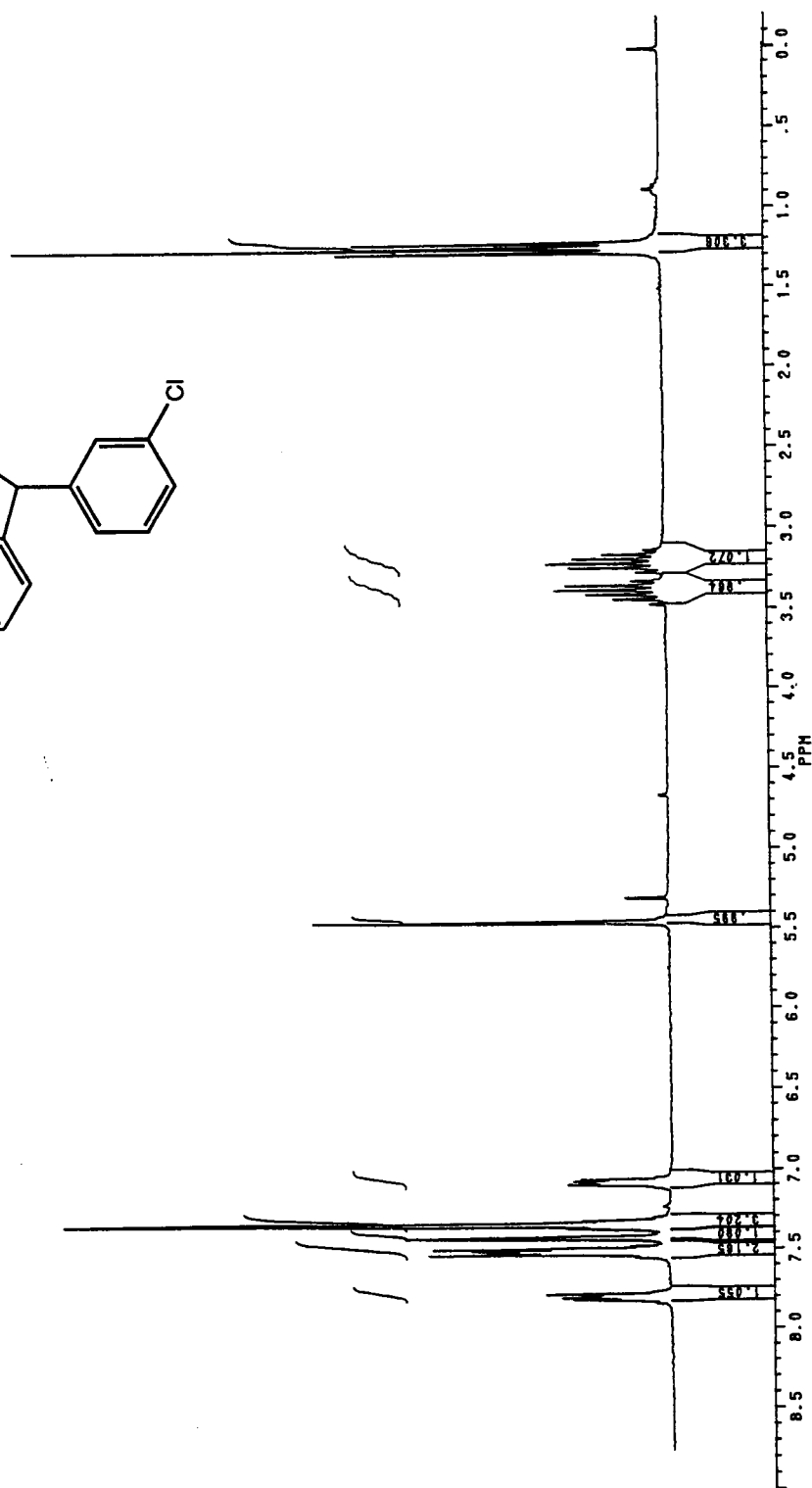
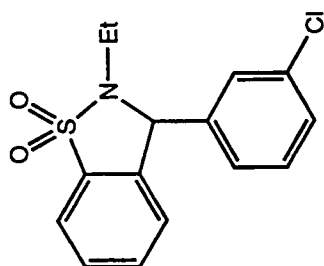
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of (±)-3-phenyl-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34a).



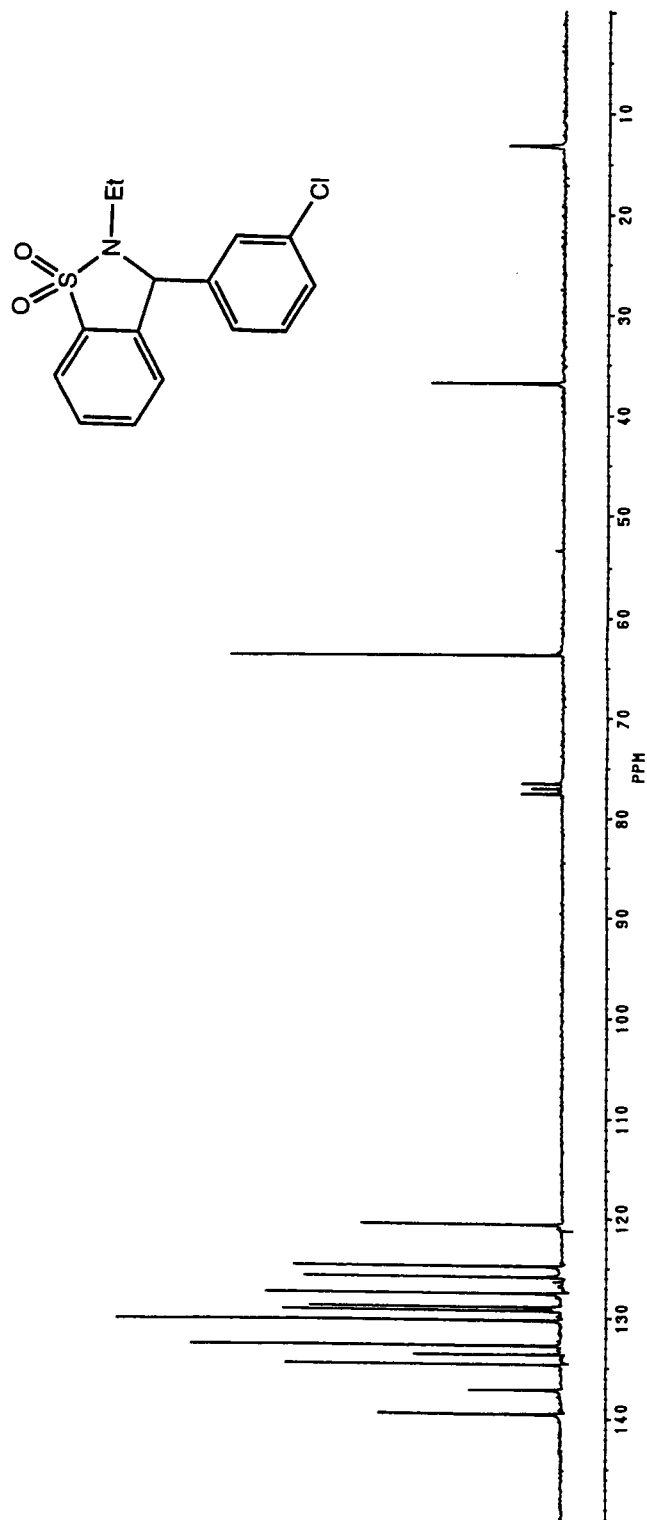
^1H NMR (250 MHz, CDCl_3) spectrum of ($\pm$)-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34b**).



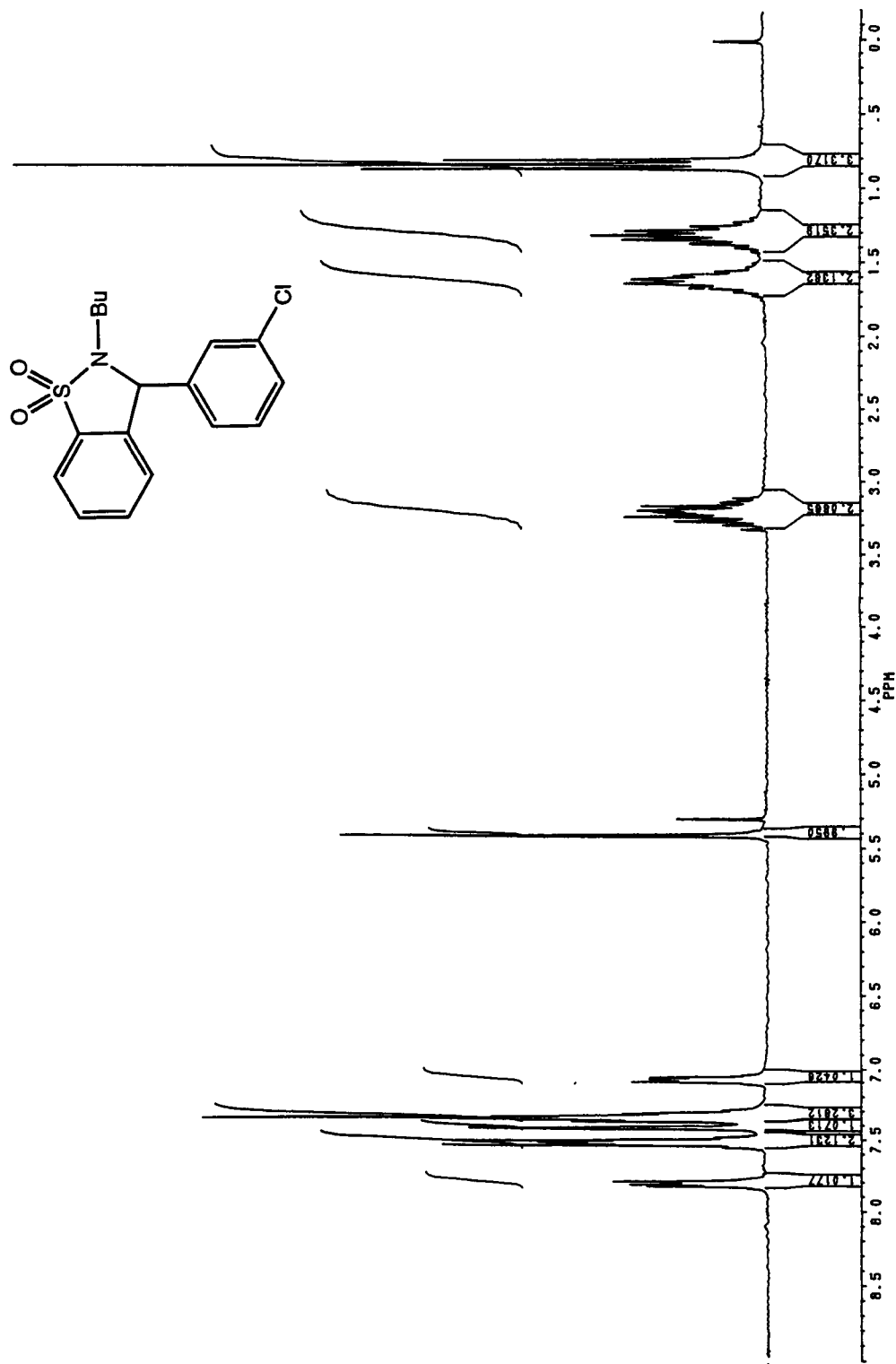
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of (±)-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34b**).



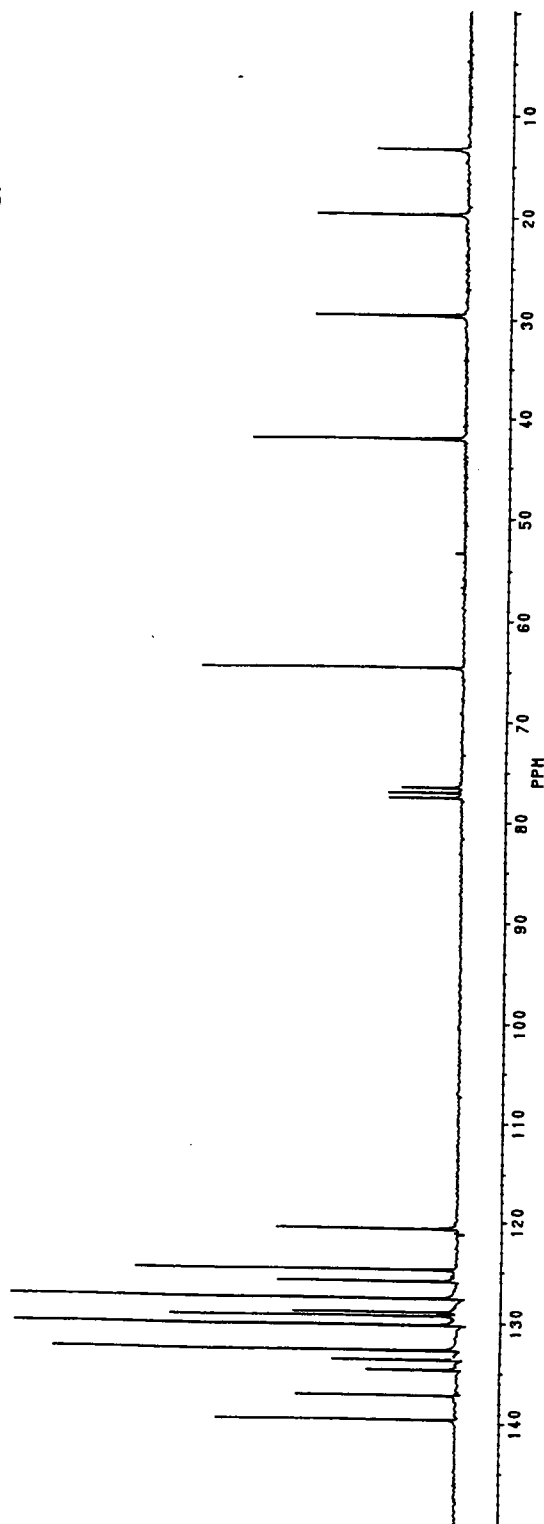
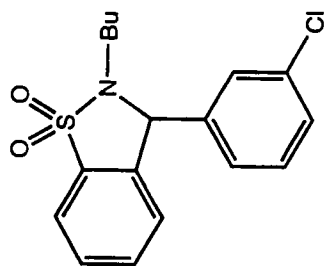
¹H NMR (250 MHz, CDCl₃) spectrum of (±)-2-ethyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34c**).



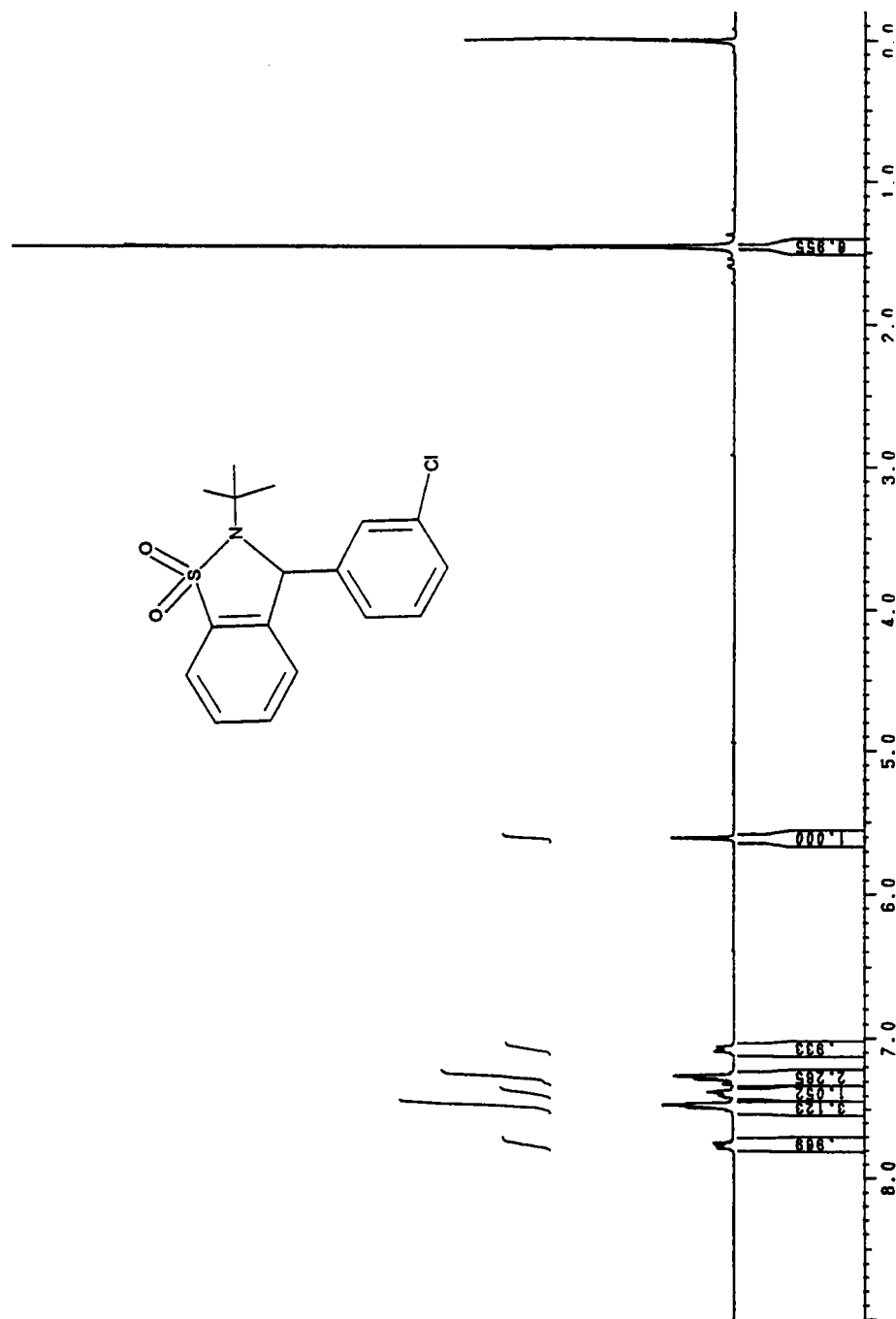
¹³C NMR (62.5 MHz, CDCl₃) spectrum of (±)-2-ethyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34c**).



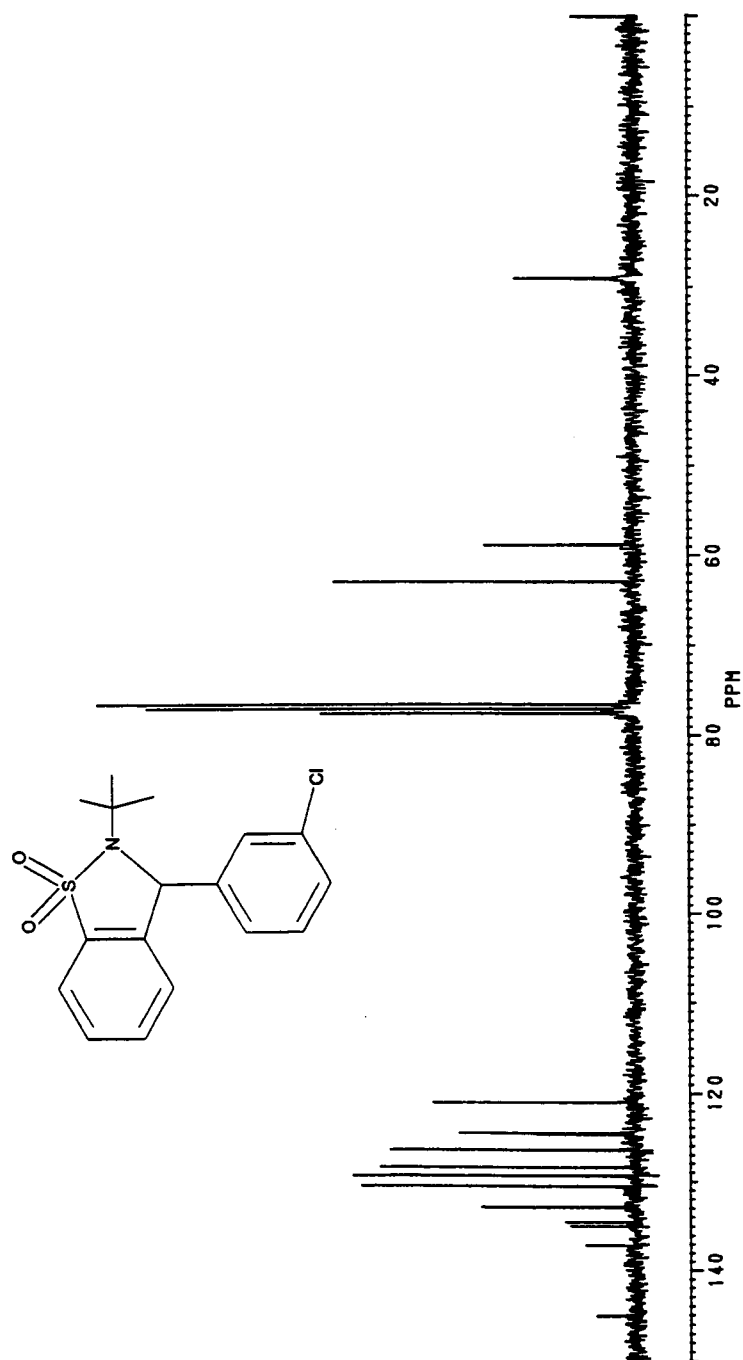
¹H NMR (250 MHz, CDCl₃) spectrum of (±)-2-butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34d).



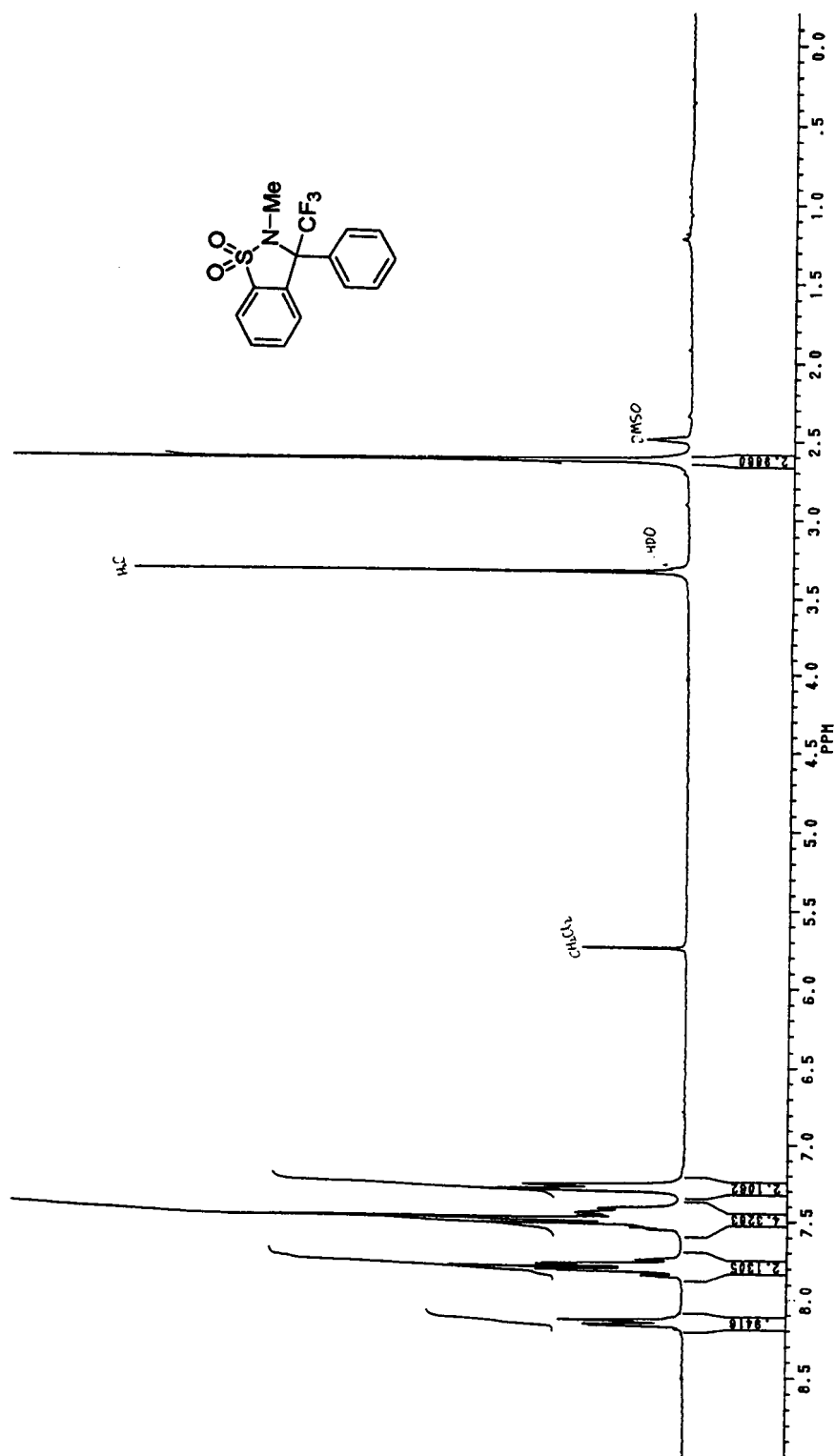
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of (±)-2-butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34d**).



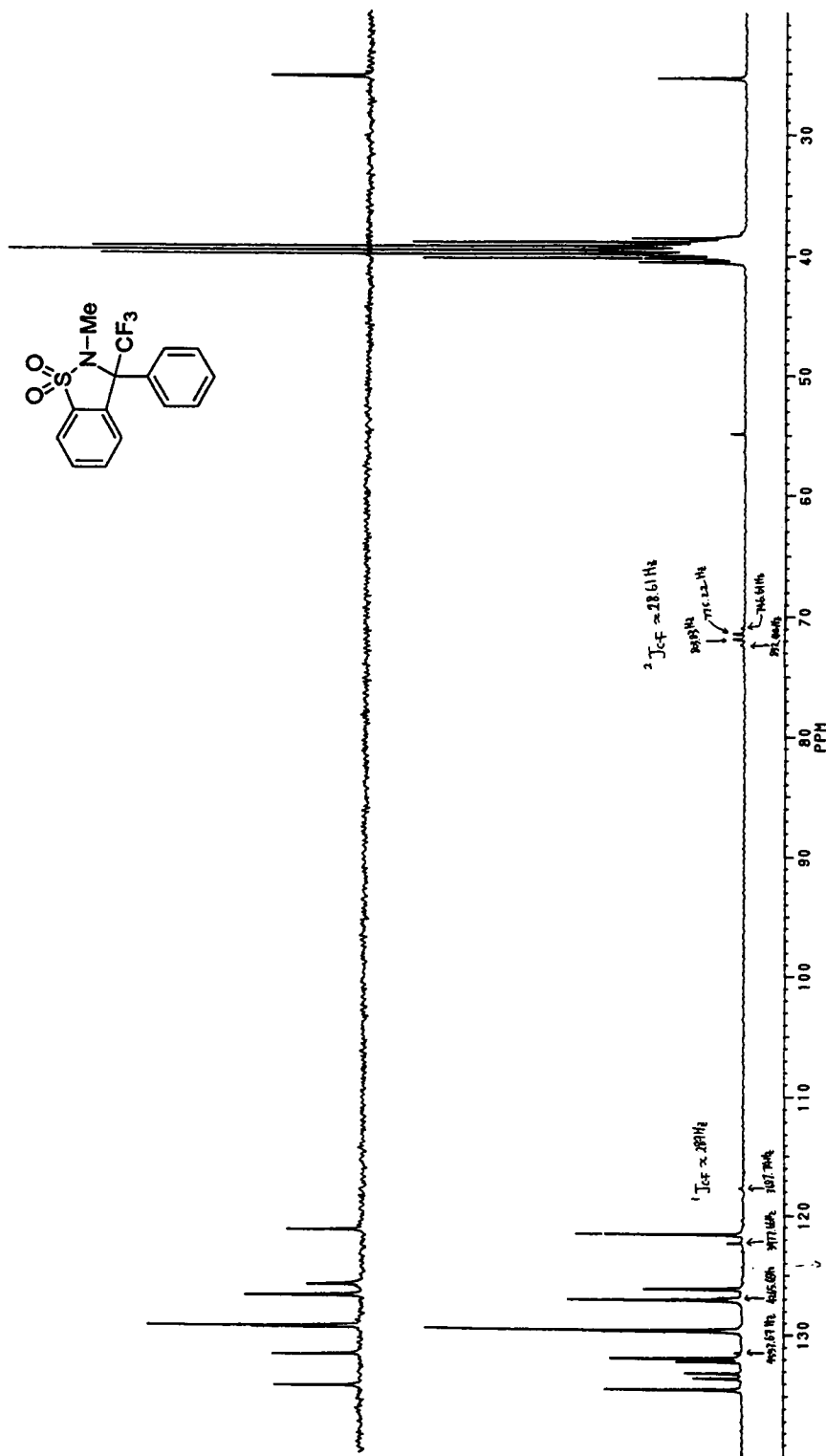
¹H NMR (250 MHz, CDCl₃) spectrum of (±)-2-tert-butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34e).



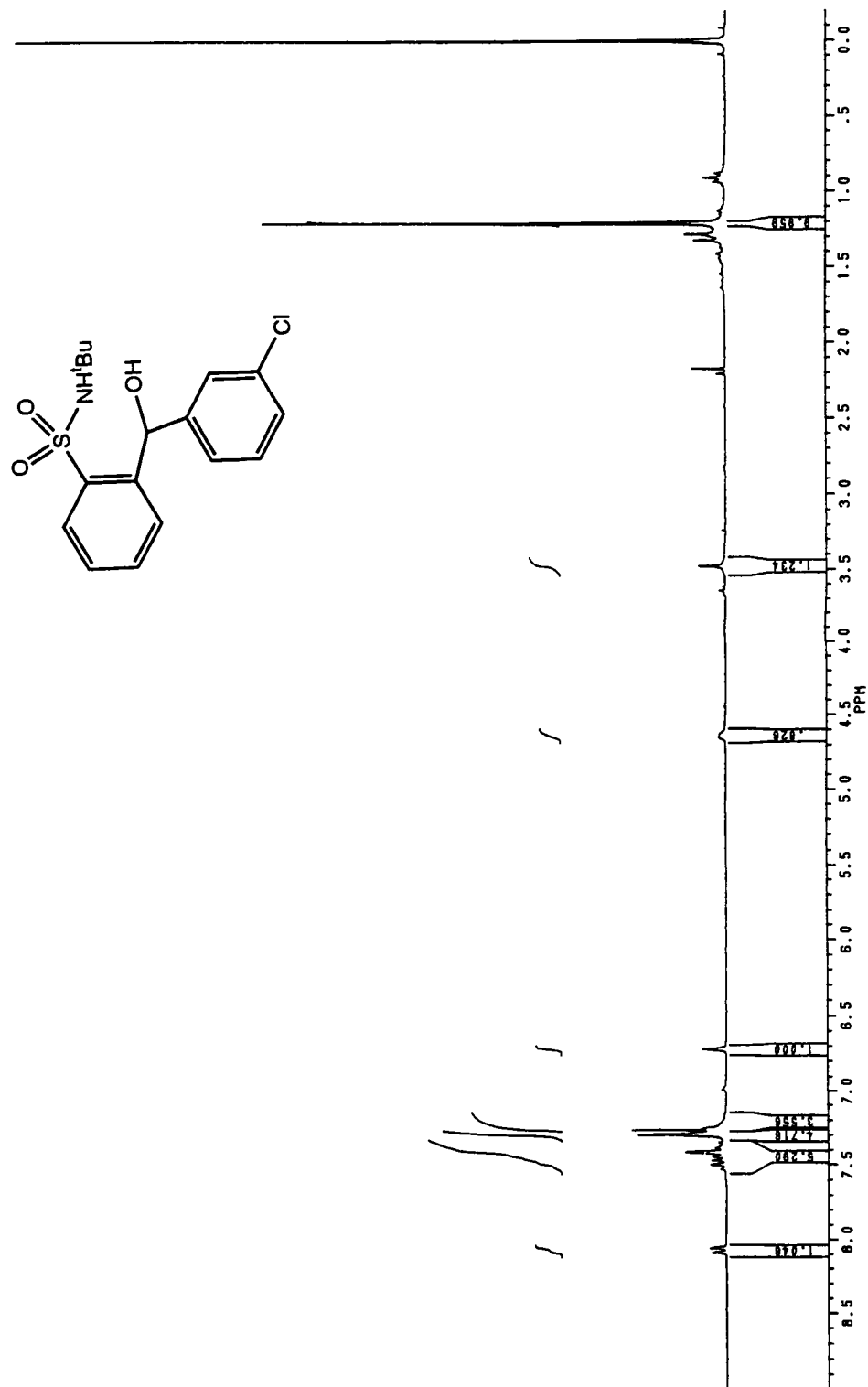
¹³C NMR (62.5 MHz, CDCl₃) spectrum of (±)-2-*tert*-butyl-3-(3-chlorophenyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (**34e**).



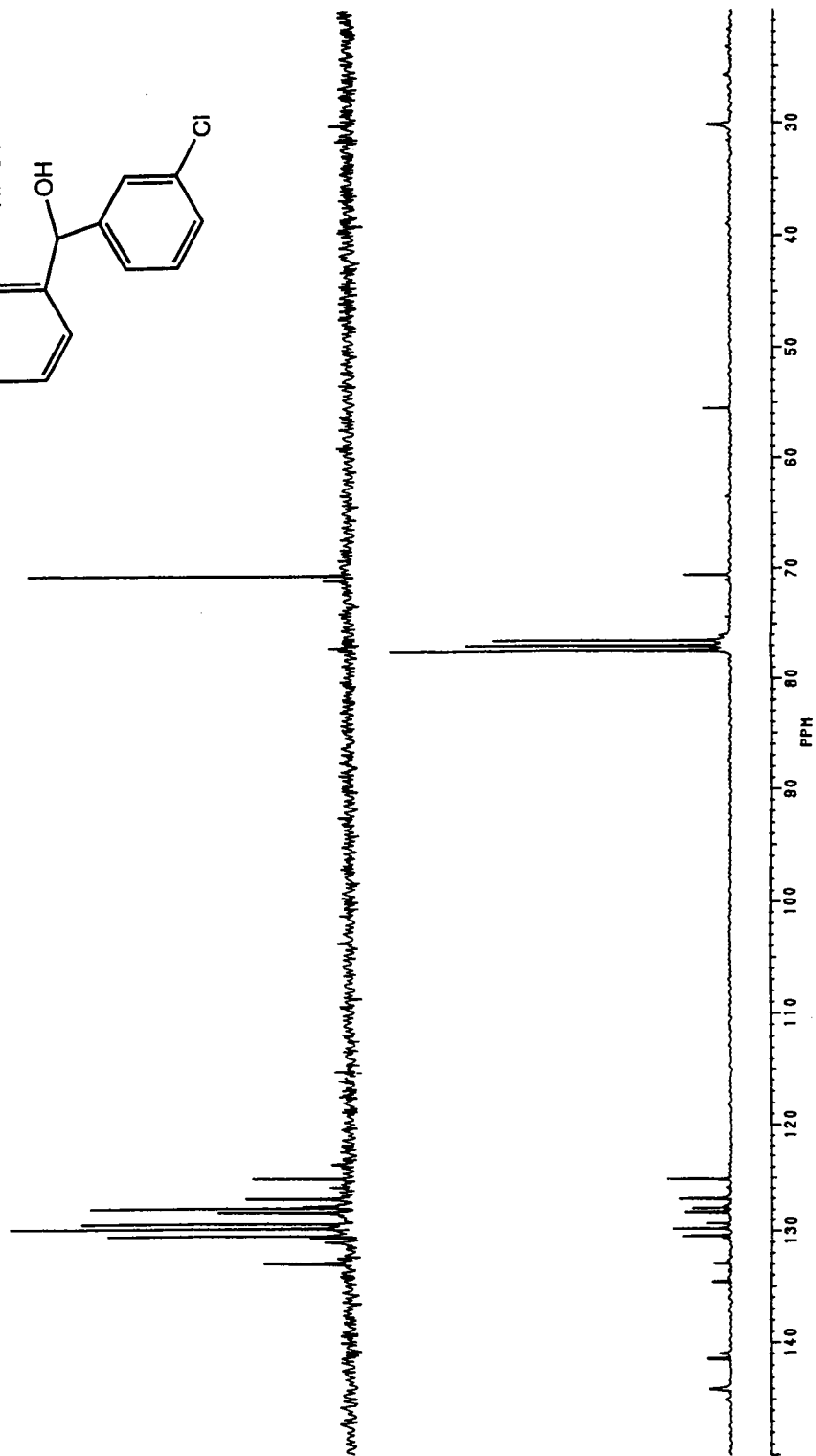
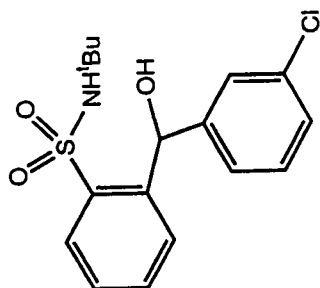
¹H NMR (250 MHz, DMSO-*d*₆) spectrum of (±)-2-methyl-3-phenyl-3-(trifluoromethyl)-2,3-dihydrobenzo[d]isothiazole 1,1-dioxide (34f).



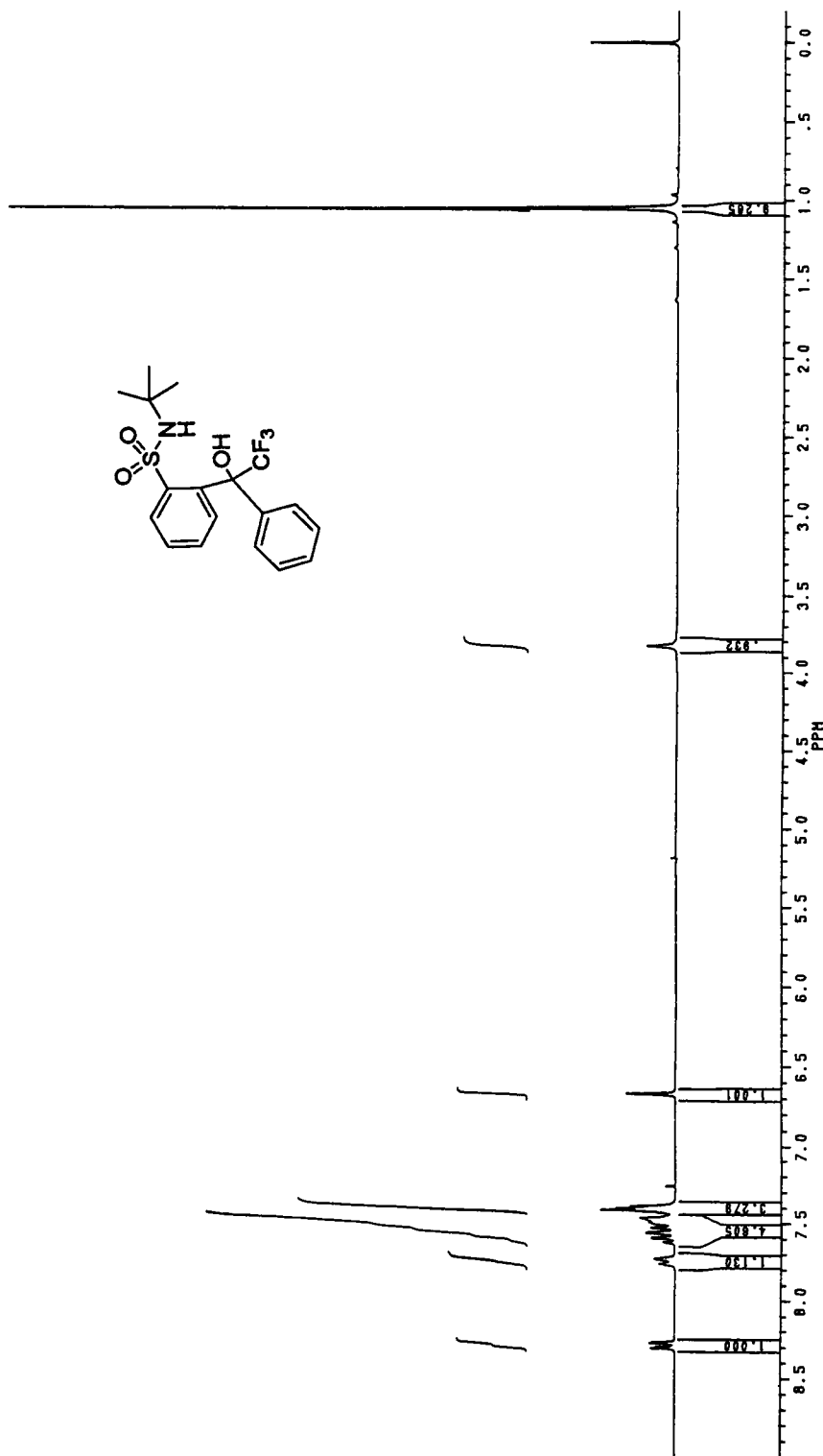
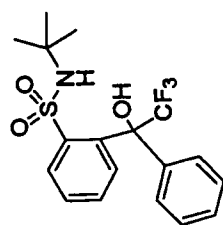
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, DMSO- d_6) spectra of (±)-2-methyl-3-phenyl-3-(trifluoromethyl)-2,3-dihydro-benzodisothiazole 1,1-dioxide (34f).



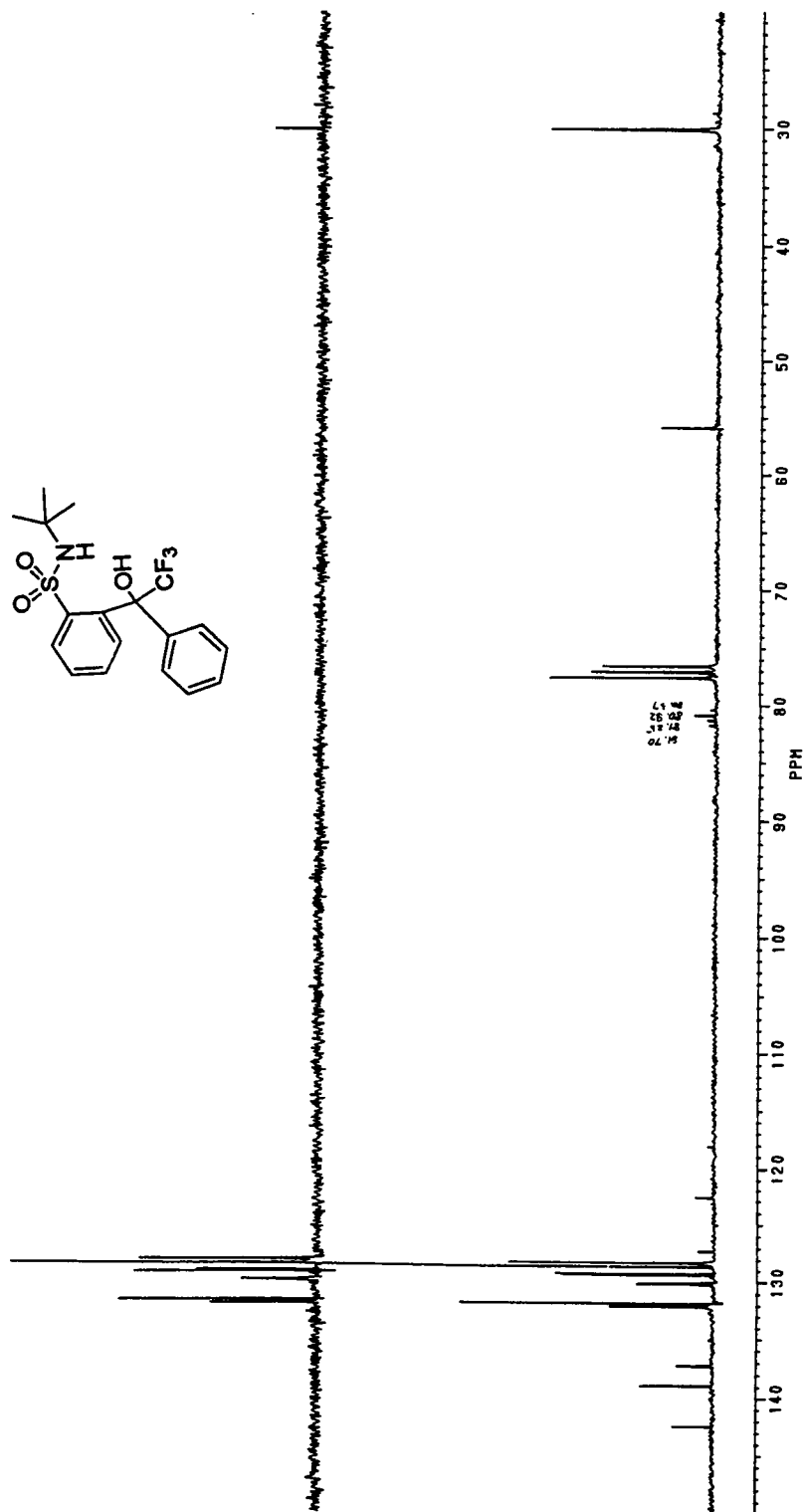
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-*tert*-butyl-2-[(3-chlorophenyl)hydroxymethyl]benzenesulfonamide (**43c**).



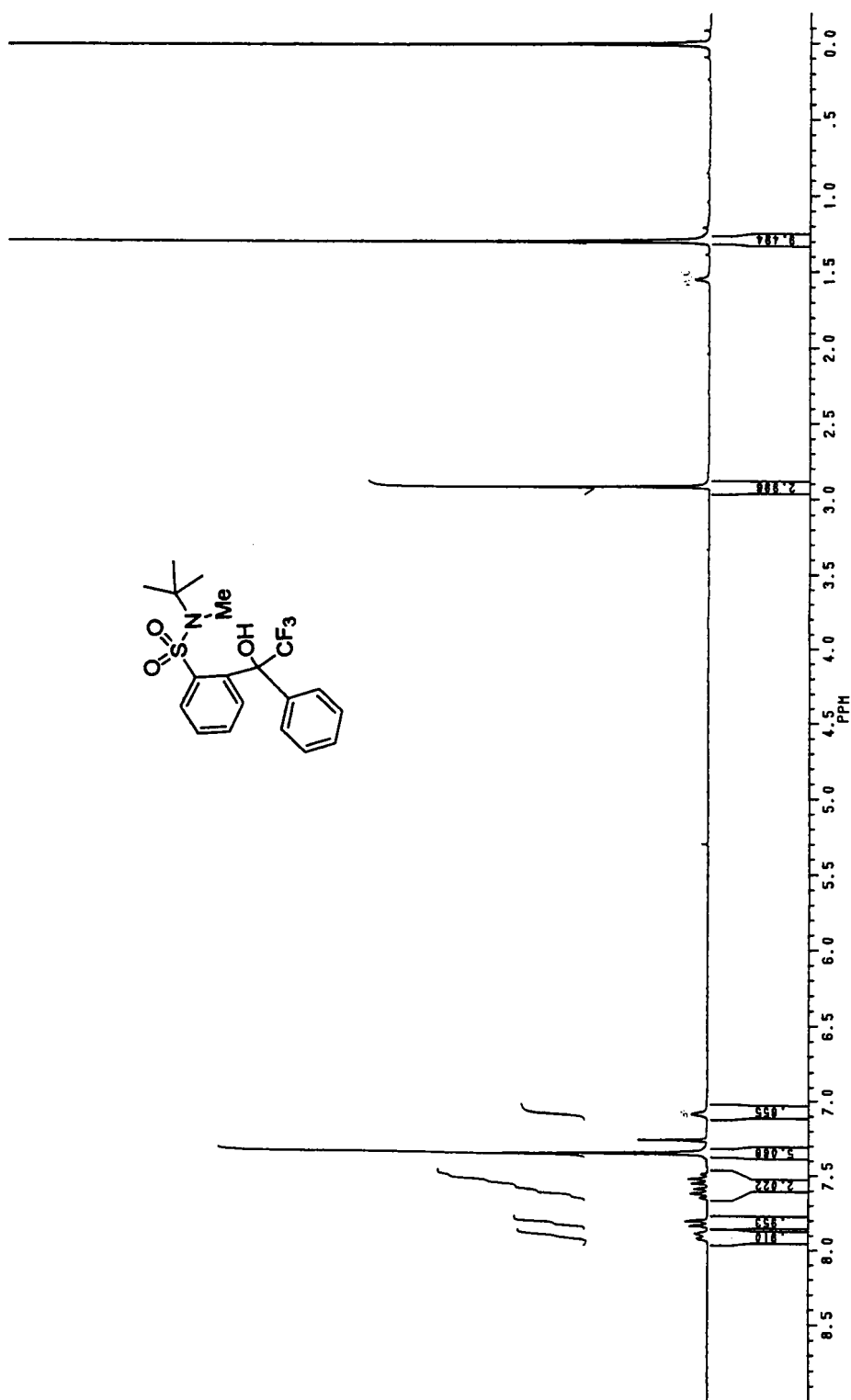
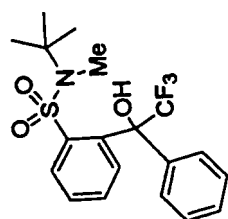
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of *N*-tert-butyl-2-[(3-chlorophenyl)hydroxymethyl]-benzenesulfonamide (**43c**).



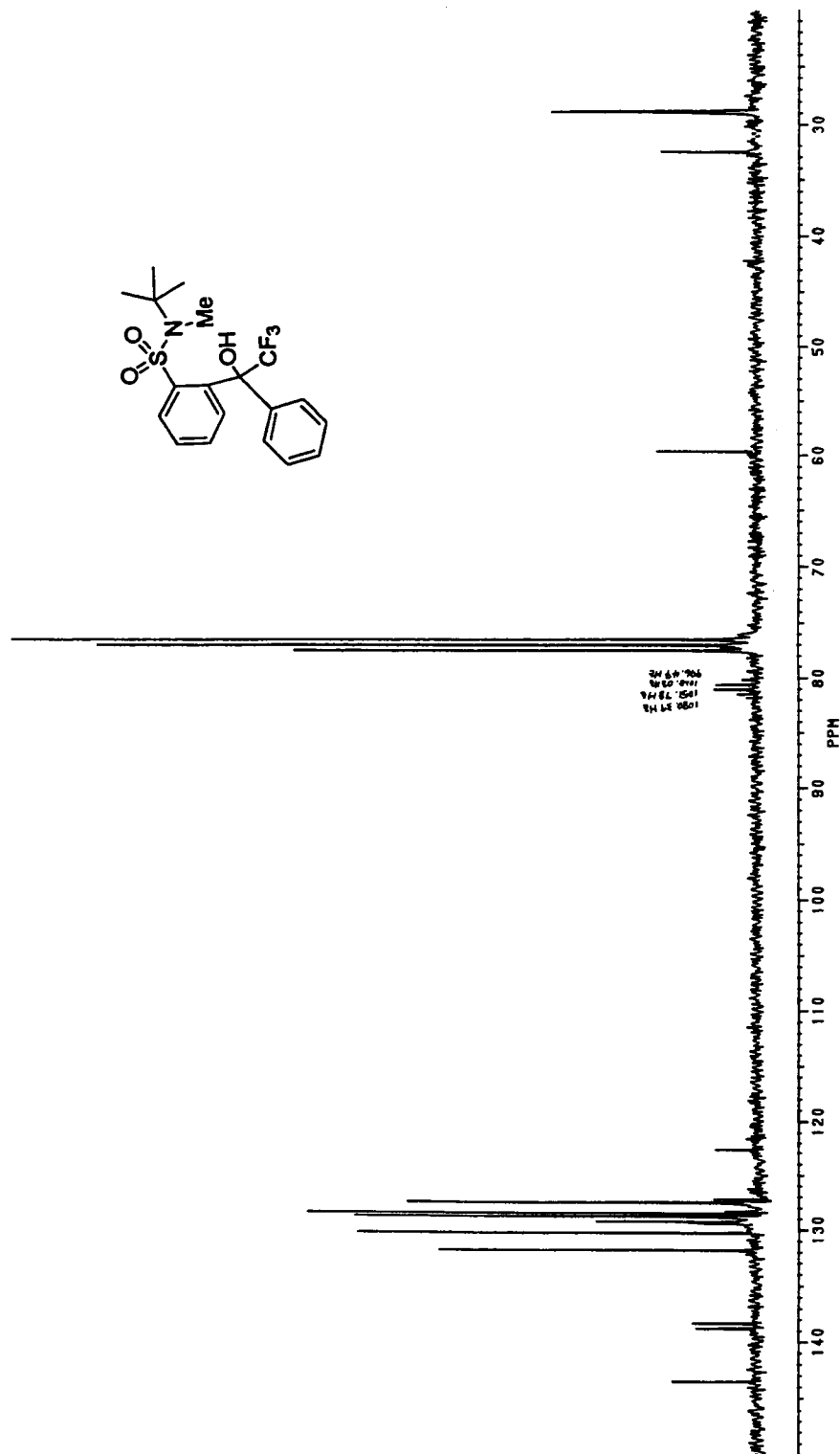
^1H NMR (250 MHz, CDCl_3) spectrum of *N*-tert-butyl-2-(2,2,2-trifluoro-1-hydroxy-1-phenylethyl)benzenesulfonamide (44a).



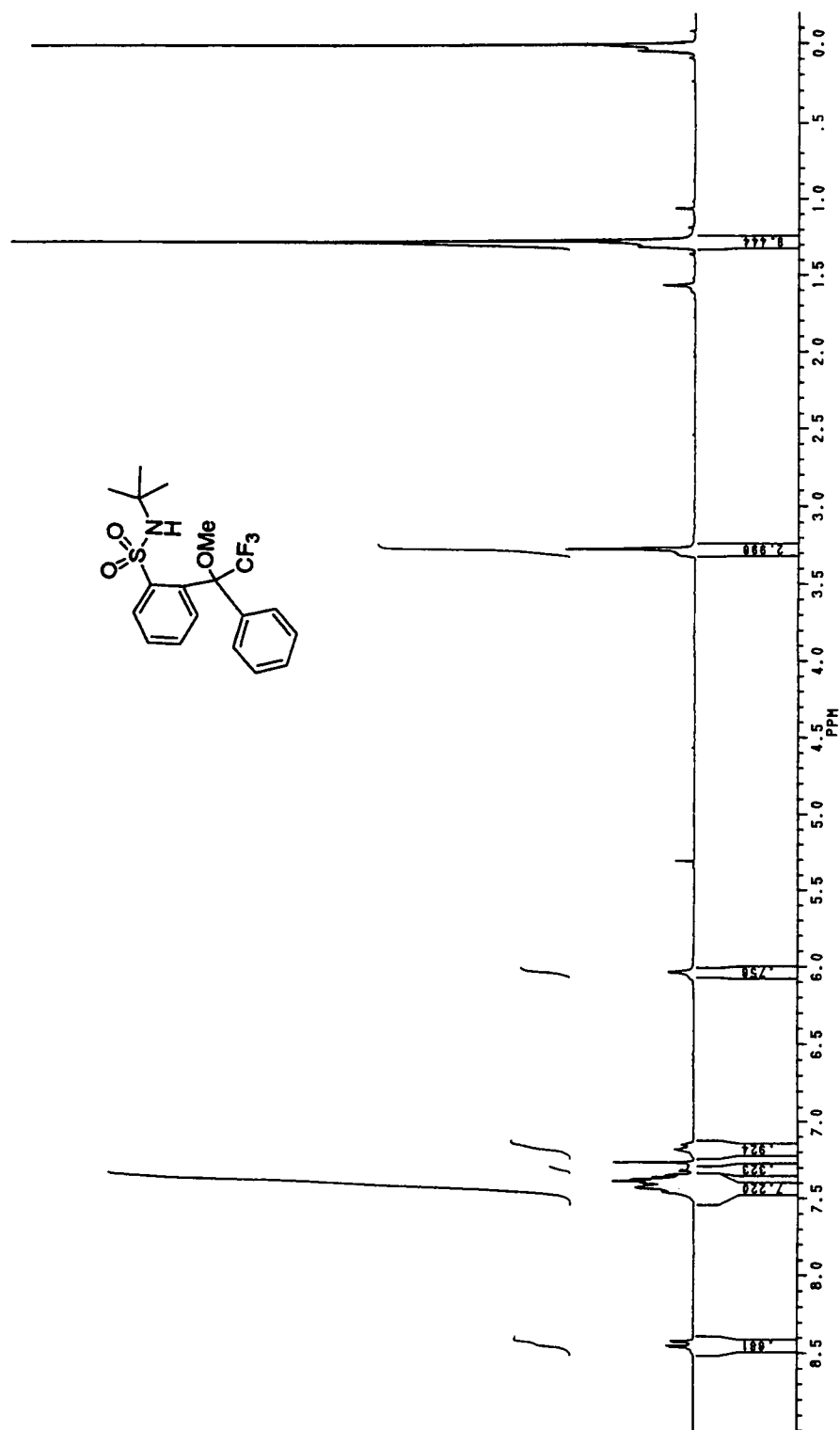
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *N*-tert-butyl-2-(2,2,2-trifluoro-1-phenyl-1-phenylethyl)benzenesulfonamide (**44a**).



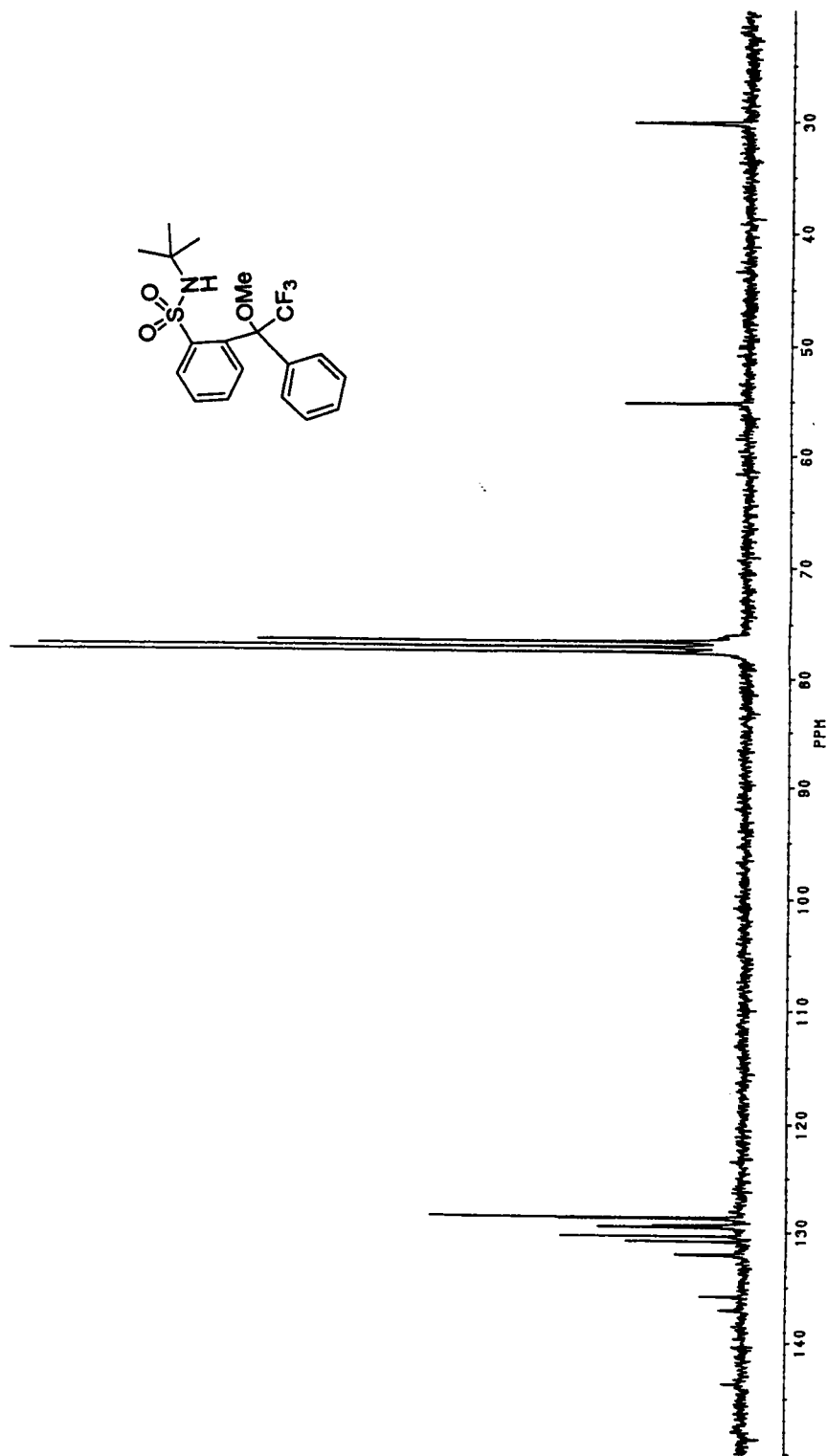
^1H NMR (250 MHz, CDCl_3) spectrum of *N*-tert-butyl-*N*-methyl-2-(2,2,2-trifluoro-1-phenylethyl)-1-phenylethanesulfonamide (**45**).



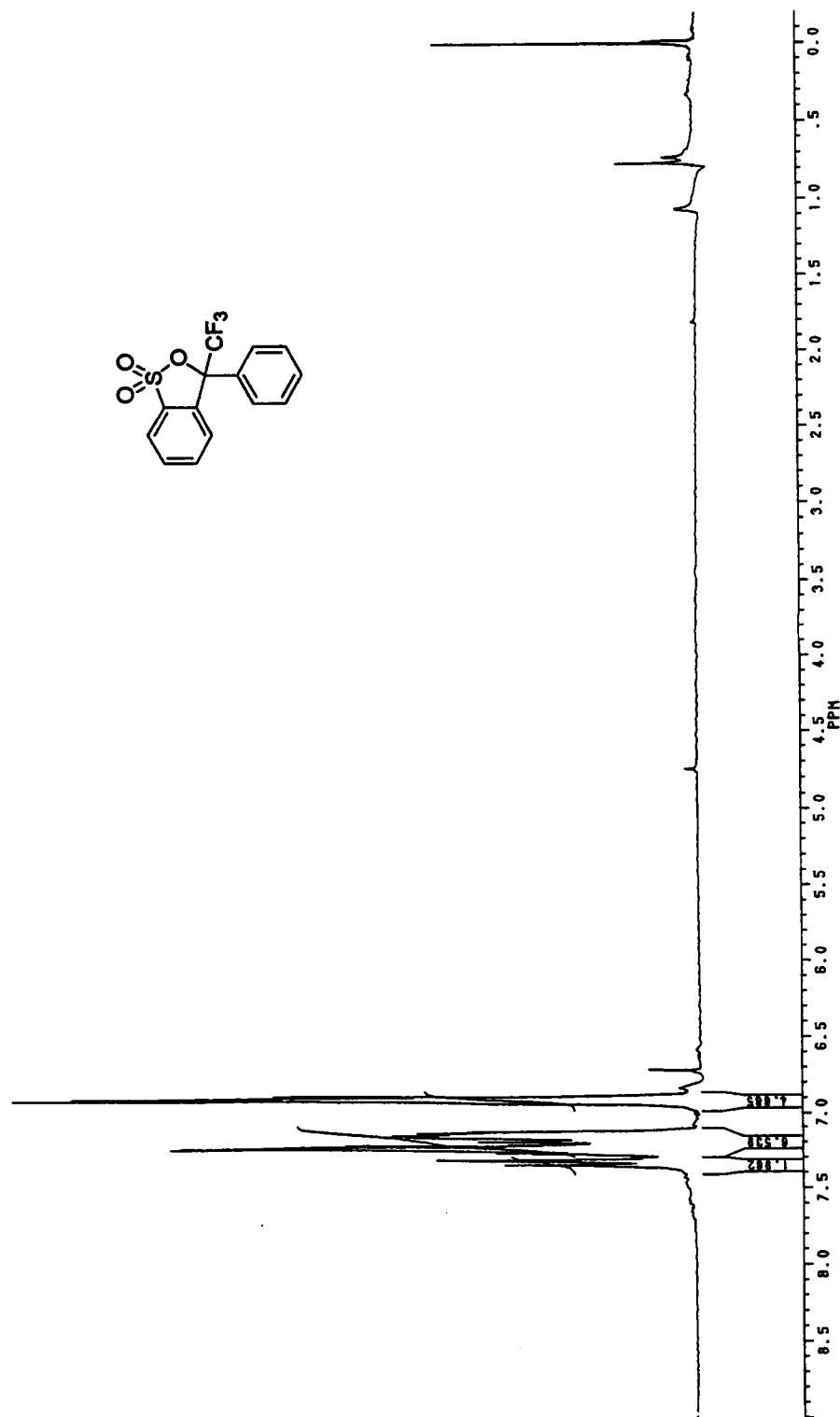
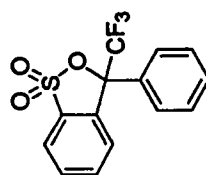
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of *N*-tert-butyl-*N*-methyl-2-(2,2,2-trifluoro-1-phenylethyl)benzenesulfonamide (45).



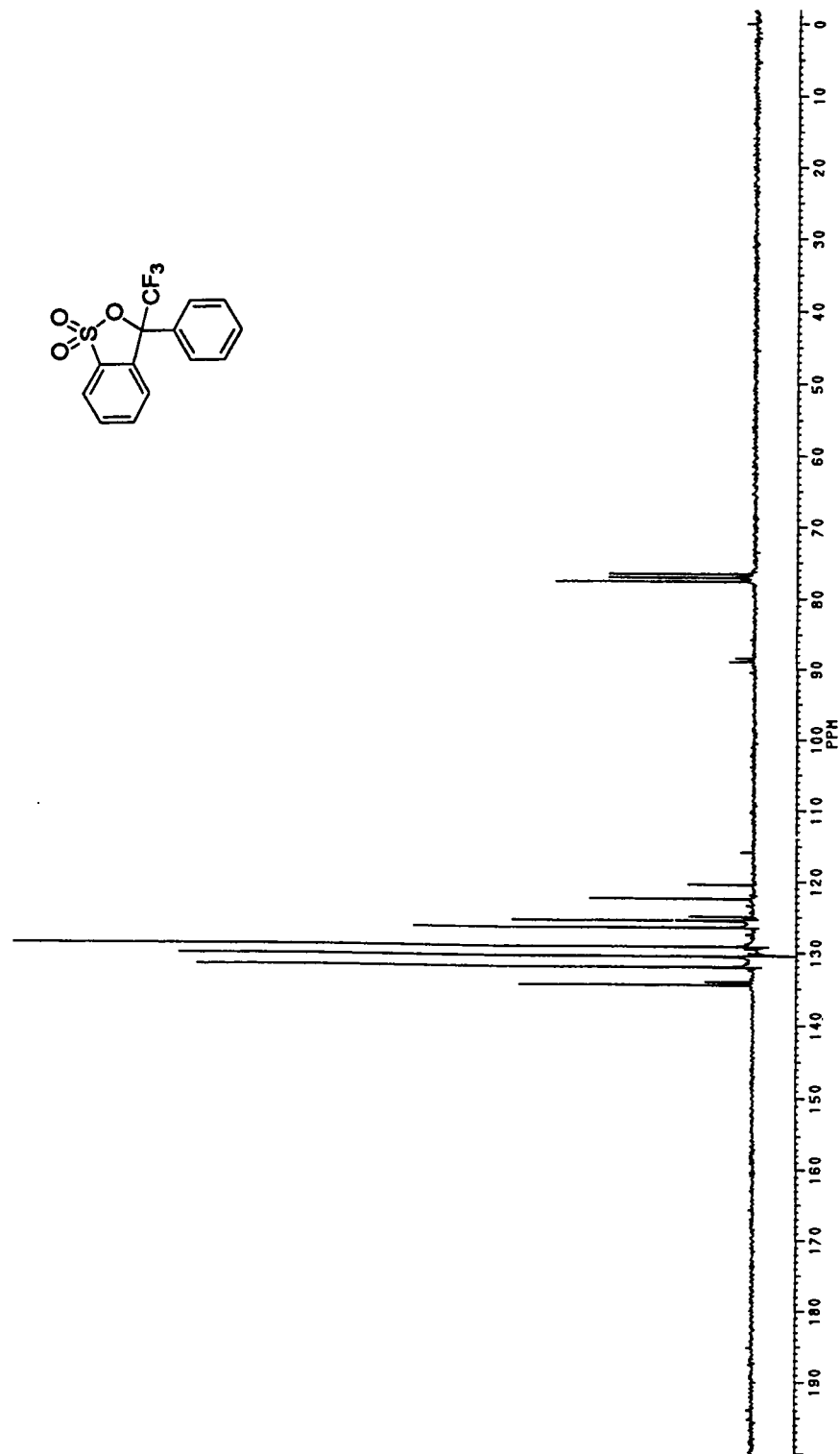
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-tert-butyl-2-(2,2,2-trifluoro-1-methoxy-1-phenylethyl)benzenesulfonamide (**46**).



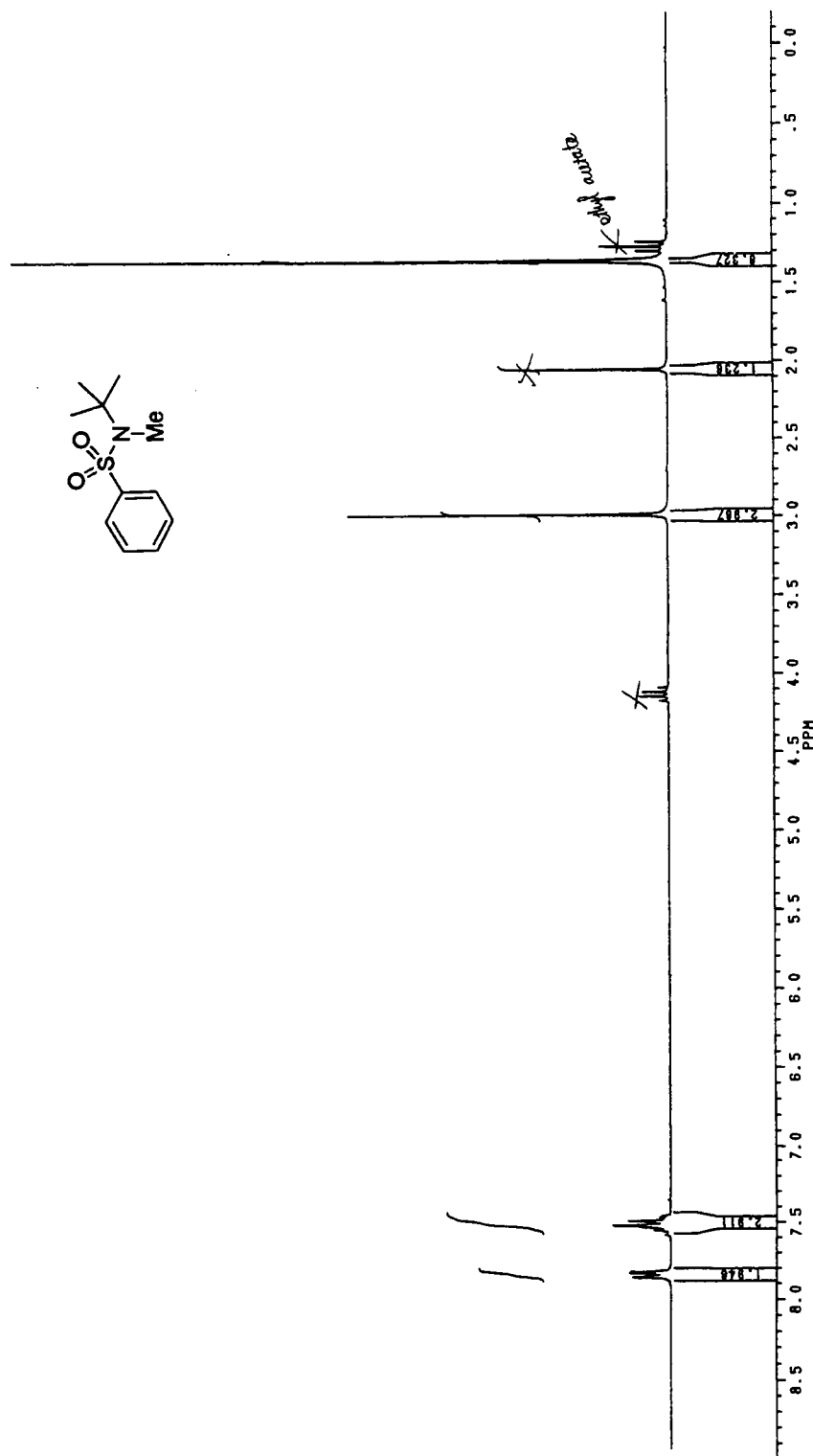
¹³C NMR (62.5 MHz, CDCl₃) spectrum of *N*-tert-butyl-2-(2,2,2-trifluoro-1-methoxy-1-phenylethyl)benzenesulfonamide (46).



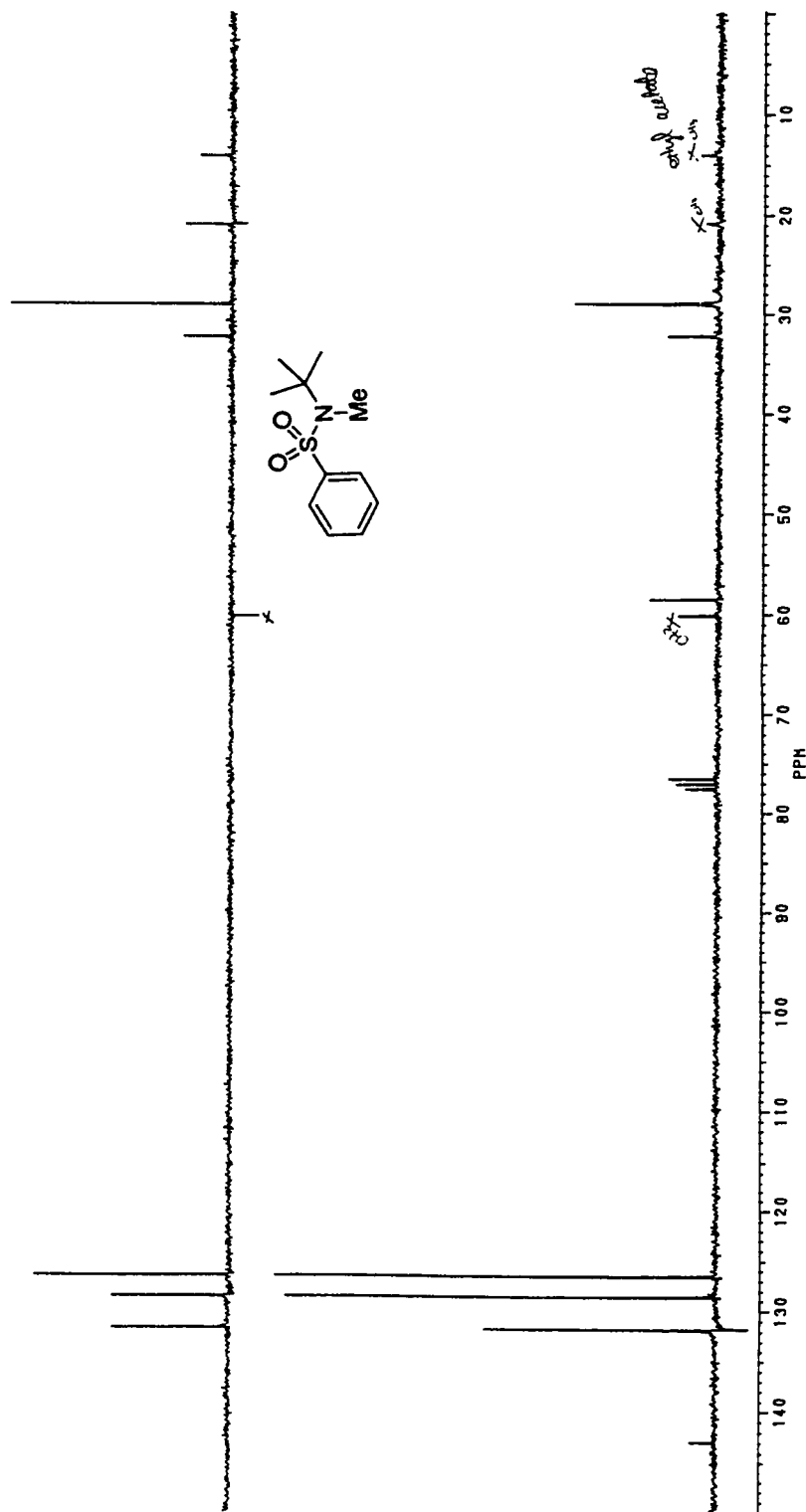
¹H NMR (250 MHz, CDCl₃) spectrum of 3-phenyl-3-(trifluoromethyl)-1,2-dioxole 1,1-dioxide (**47**).



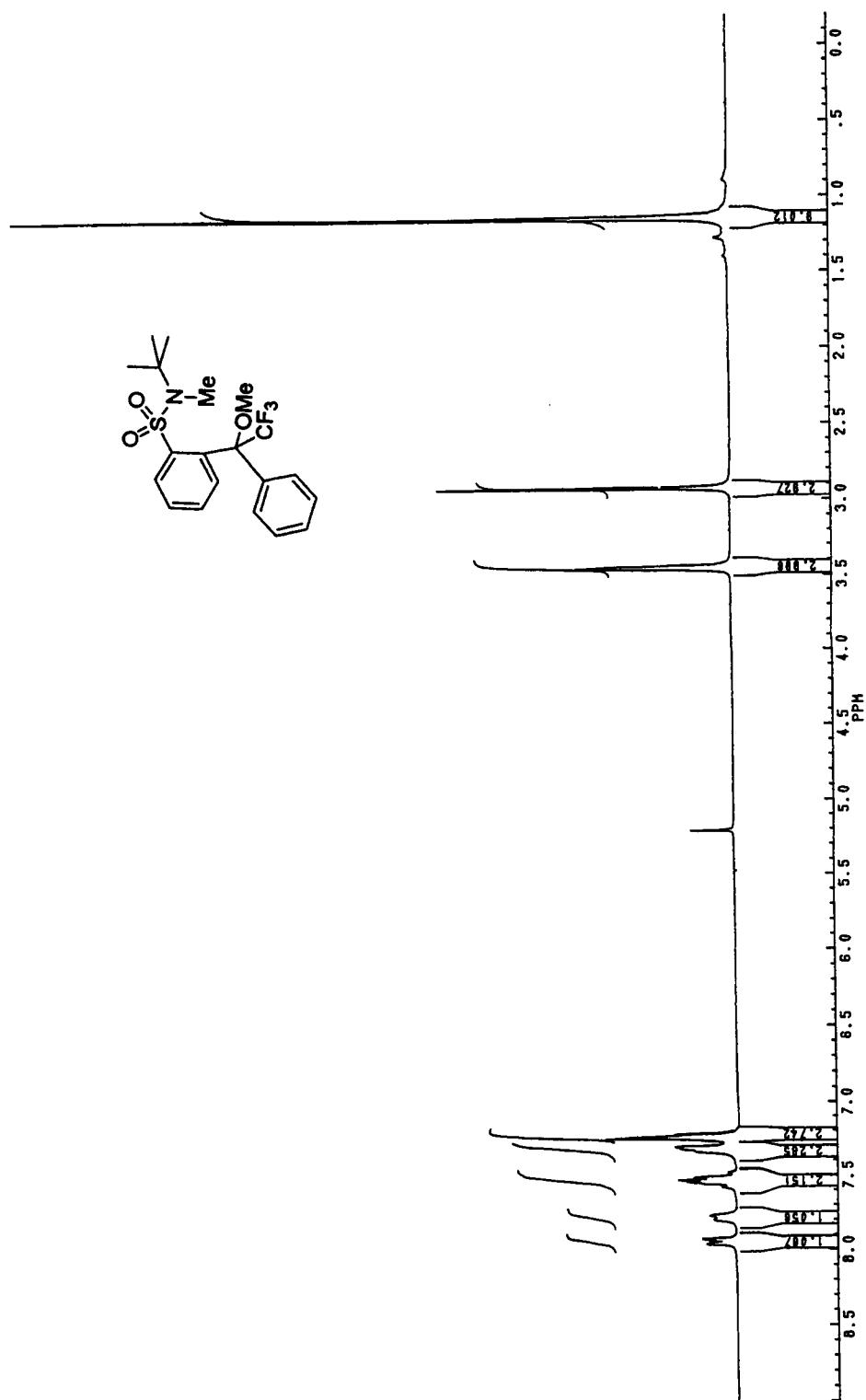
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of 3-phenyl-3-trifluoromethyl-1,2-oxathiole 1,1-dioxide (47).



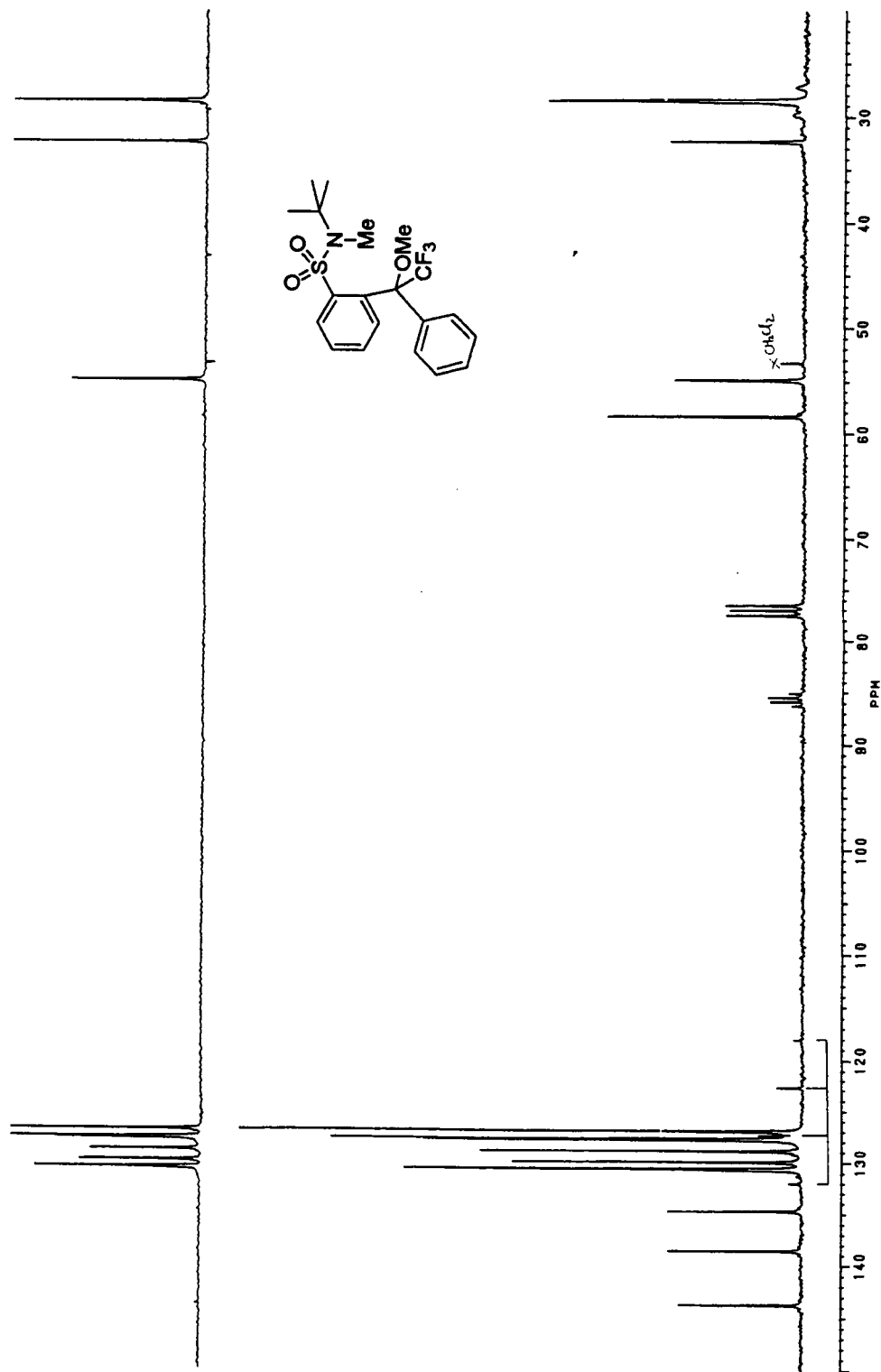
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-*tert*-butyl-*N*-methylbenzenesulfonamide (48).



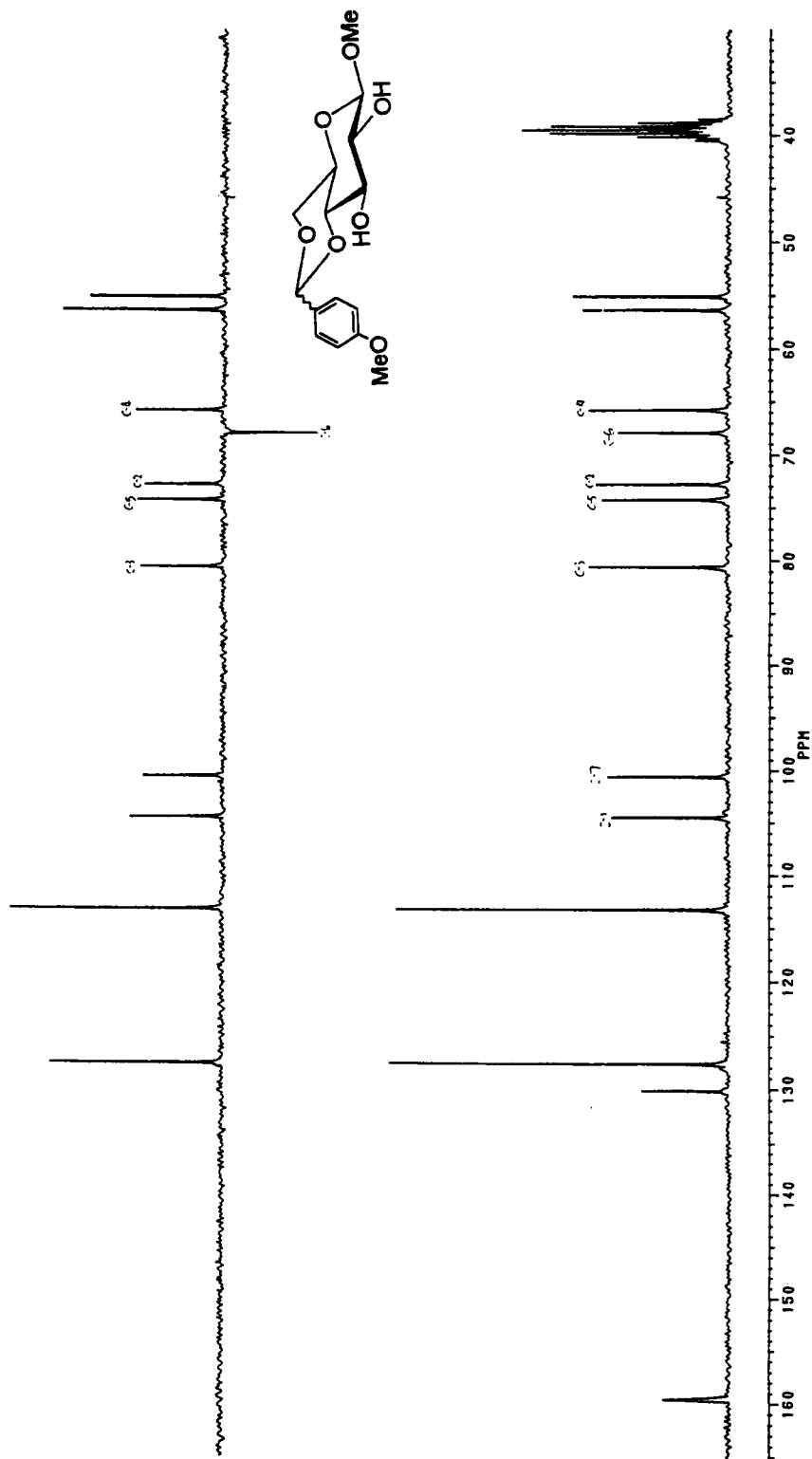
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *N*-tert-butyl-*N*-methylbenzenesulfonamide (48).



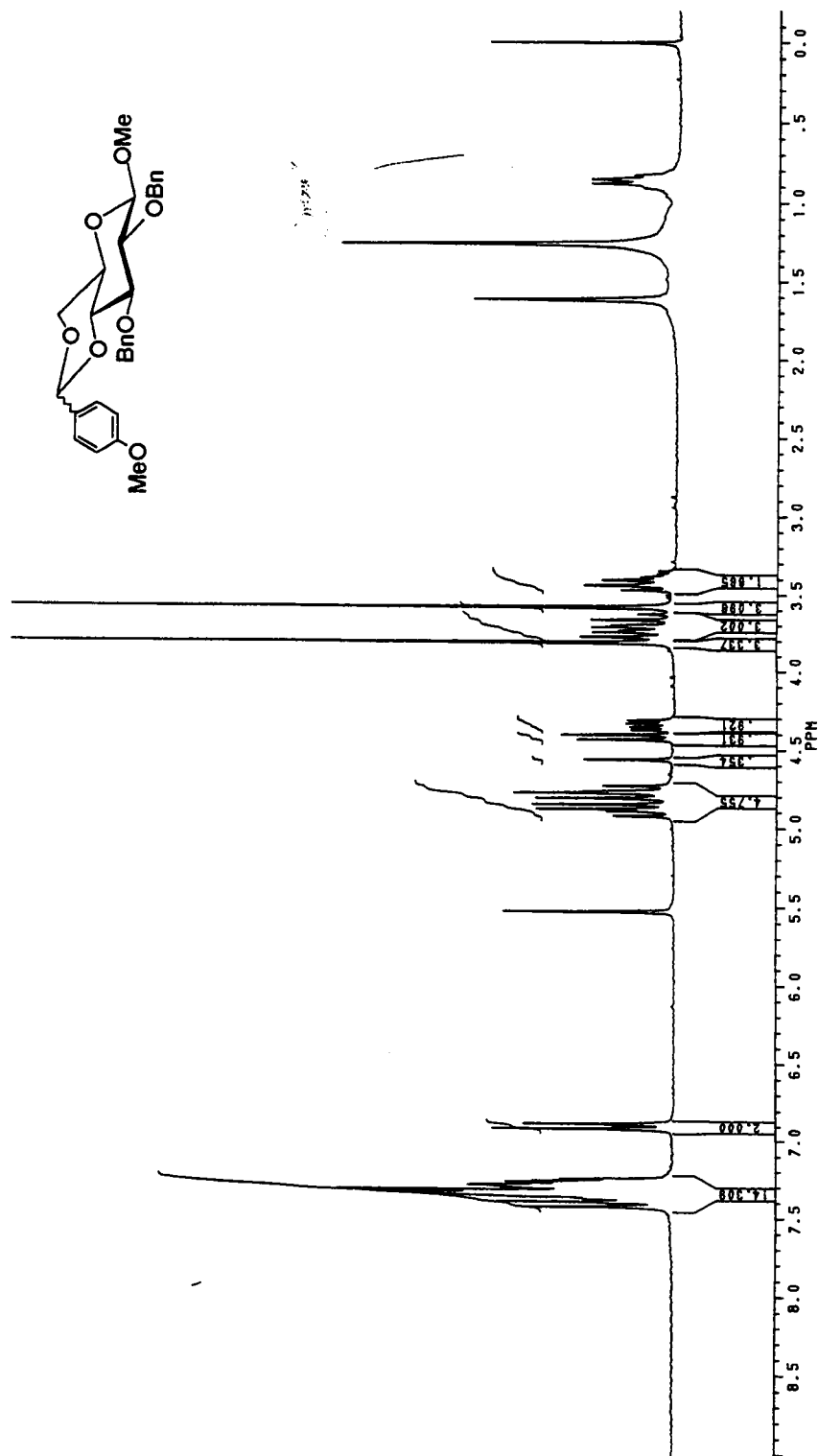
¹H NMR (250 MHz, CDCl₃) spectrum of *N*-*tert*-Butyl-2-(2,2,2-trifluoro-1-methoxy-1-phenylethyl)benzenesulfonamide (**49**).



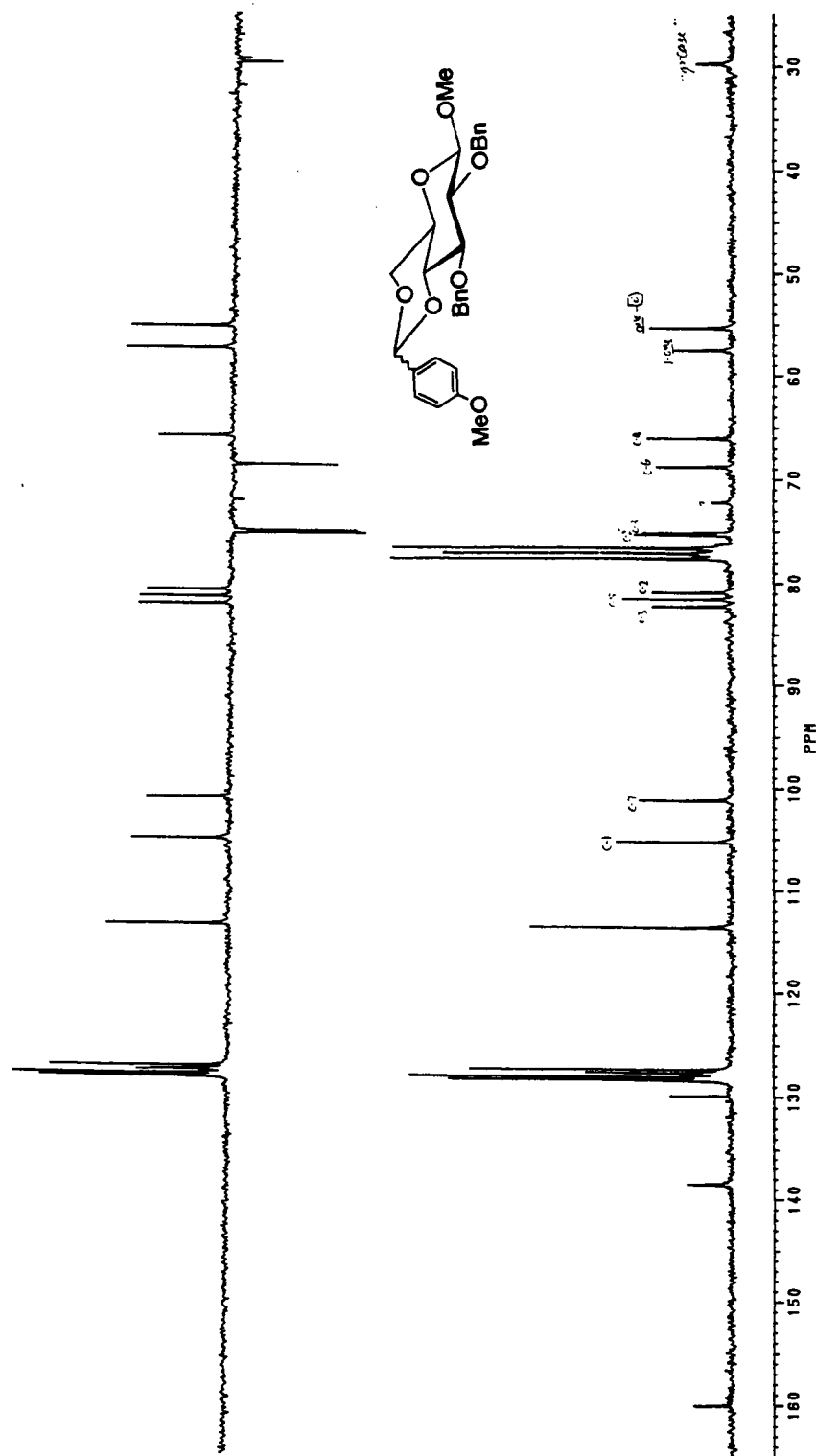
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of *N*-*tert*-Butyl-*N*-methyl-2-(2,2,2-trifluoro-1-methoxy-1-phenylethyl)benzenesulfonamide (**49**).



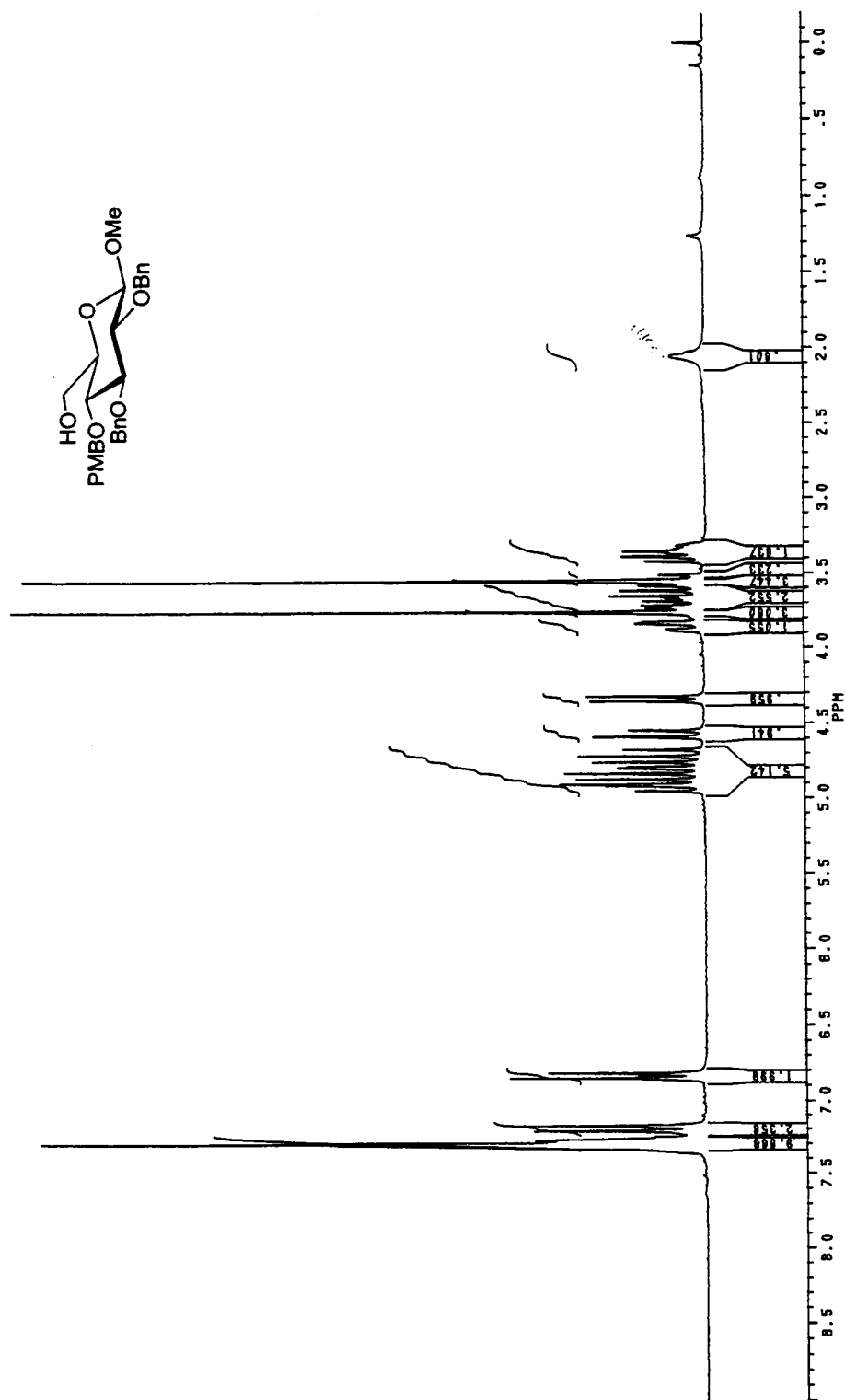
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, $\text{DMSO-}d_6$) spectra of methyl 4,6-O-p-methoxybenzylidene- β -D-glucopyranoside (51).



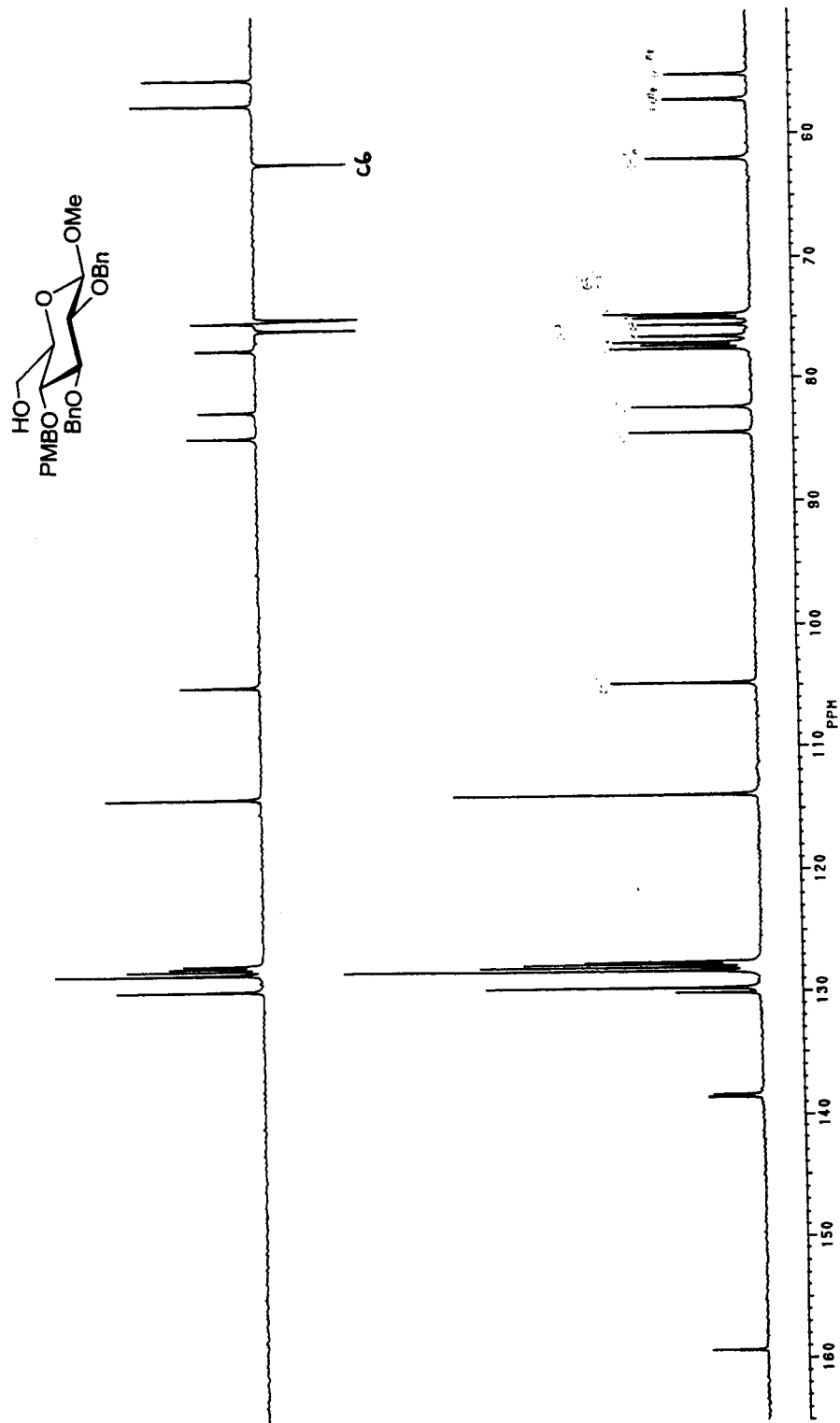
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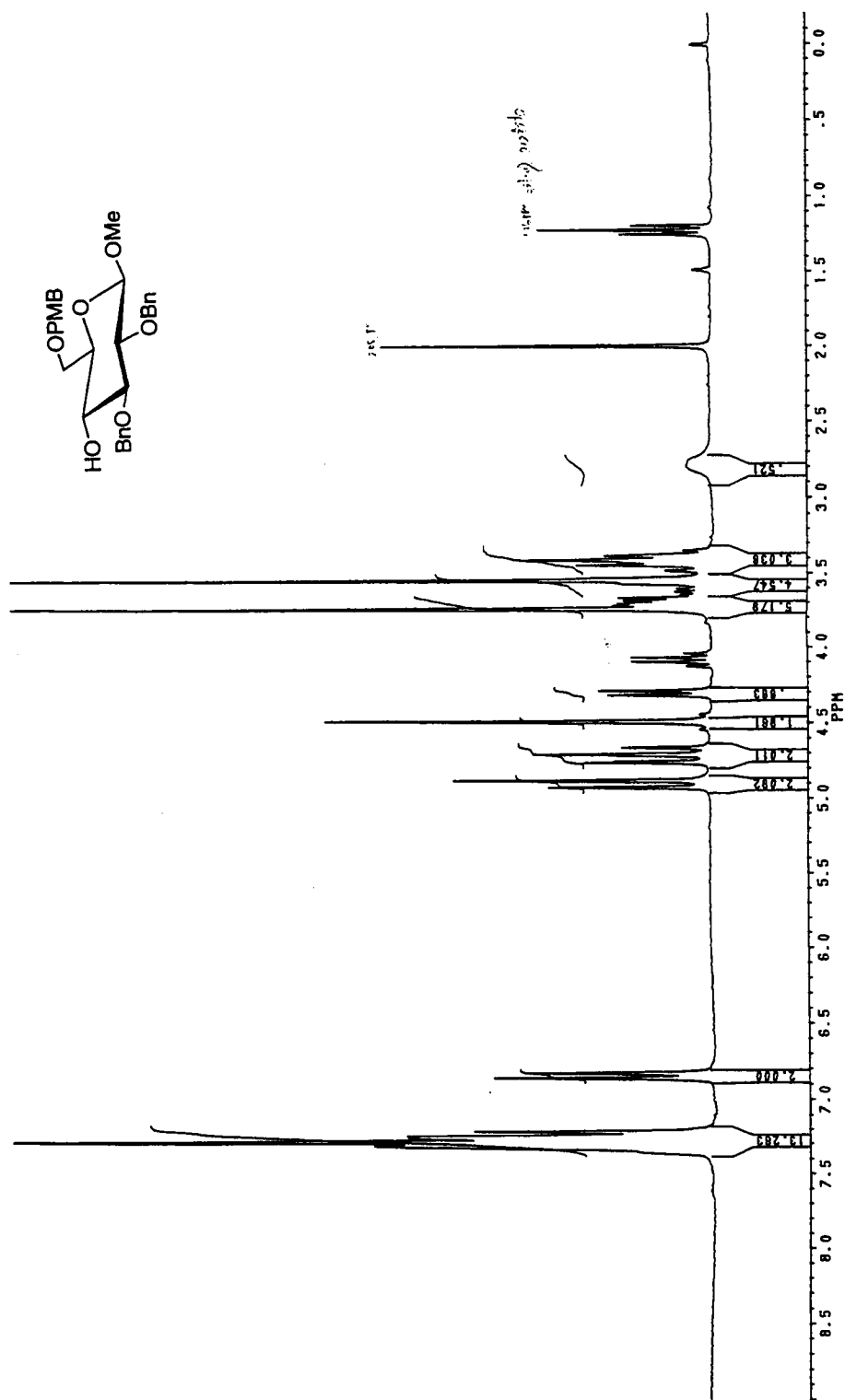
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of methyl 2,3-di-O-benzyl-4,6-O-p-methoxybenzylidene-β-D-glucopyranoside (**52**).



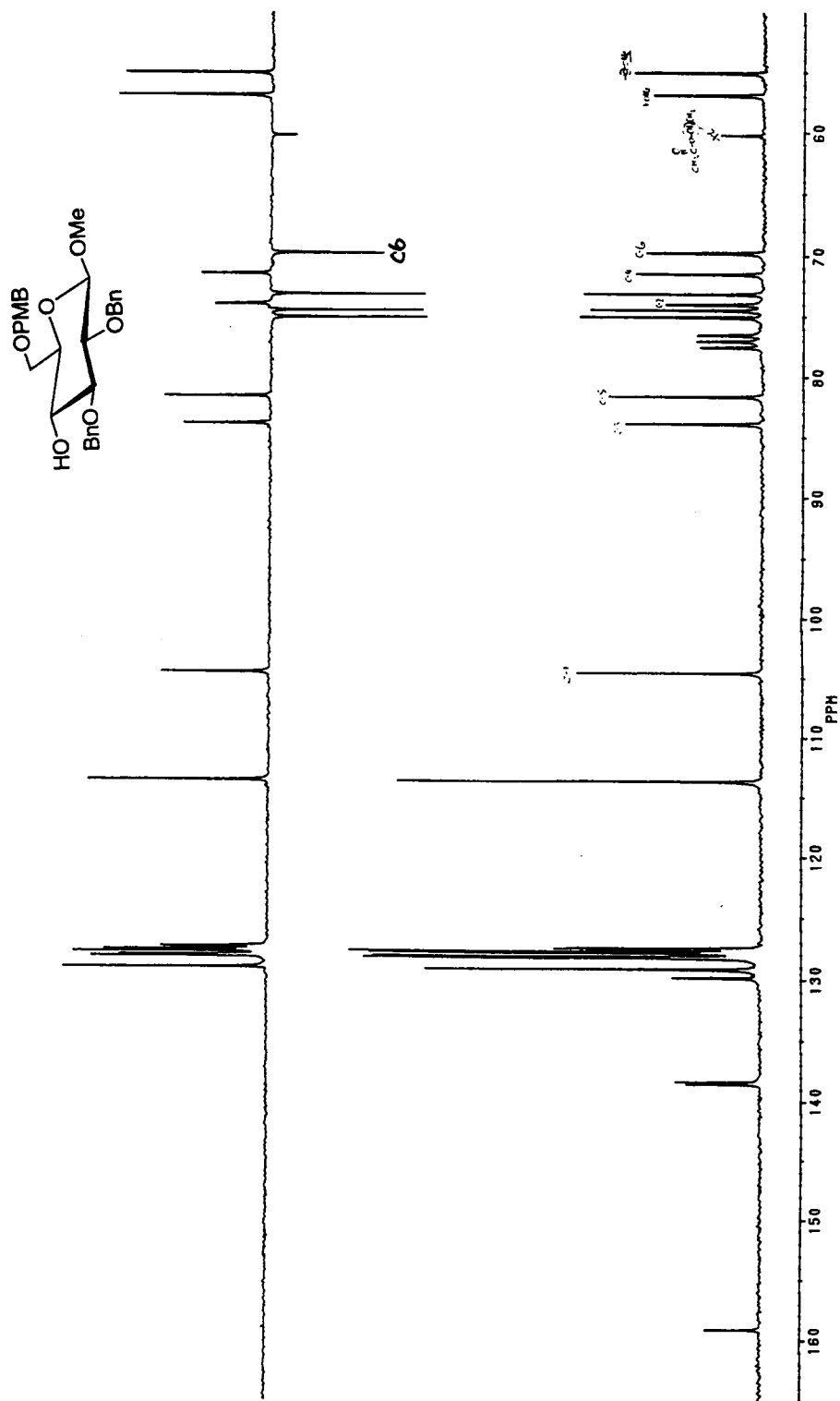
^1H NMR (250 MHz, CDCl_3) spectrum of methyl 2,3-di-O-benzyl-4-O-p-methoxybenzyl- β -D-glucopyranoside (53).

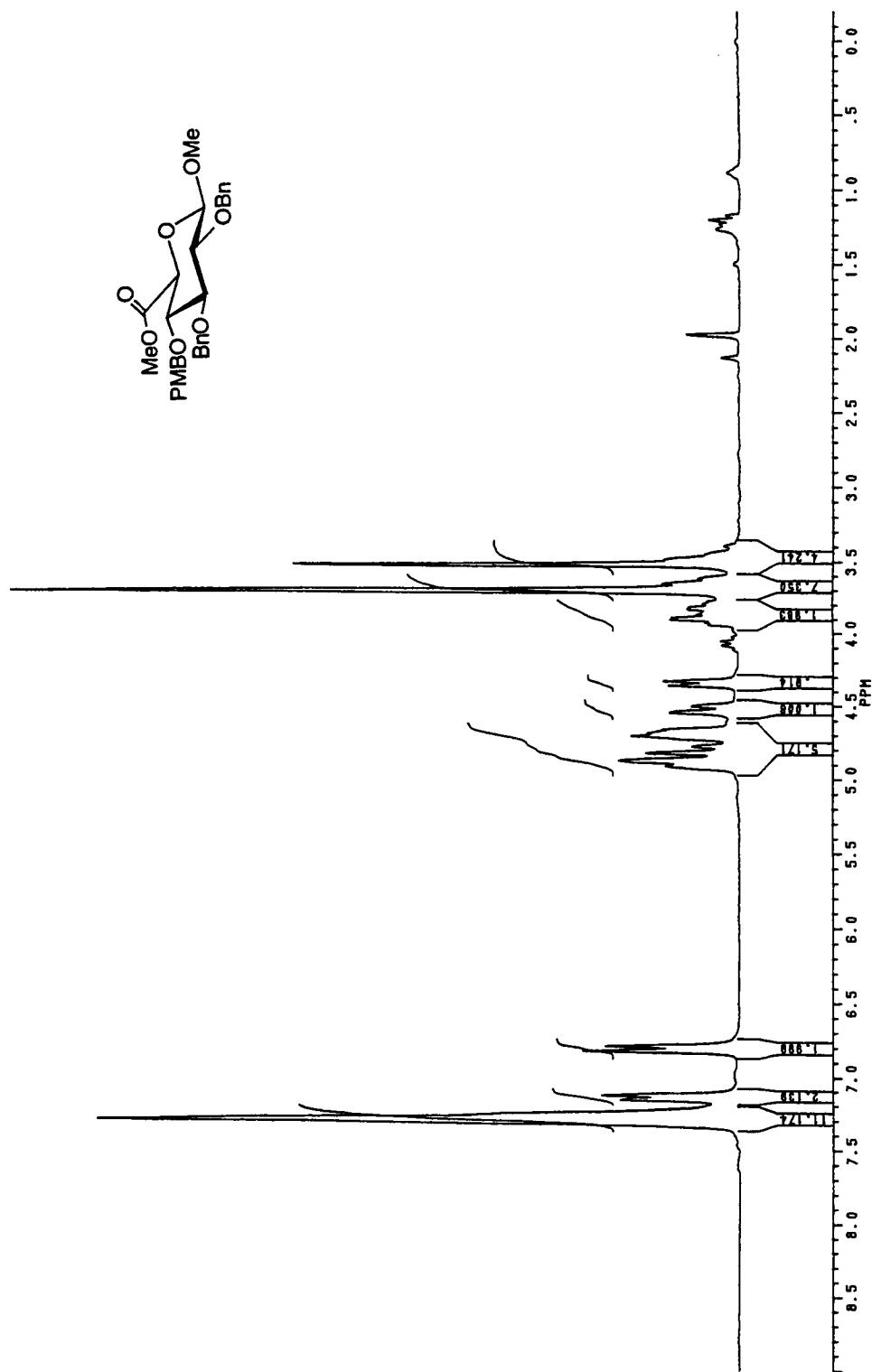


DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of methyl 2,3-di-O-benzyl-4-O-p-methoxybenzyl-β-D-glucopyranoside (**53**).

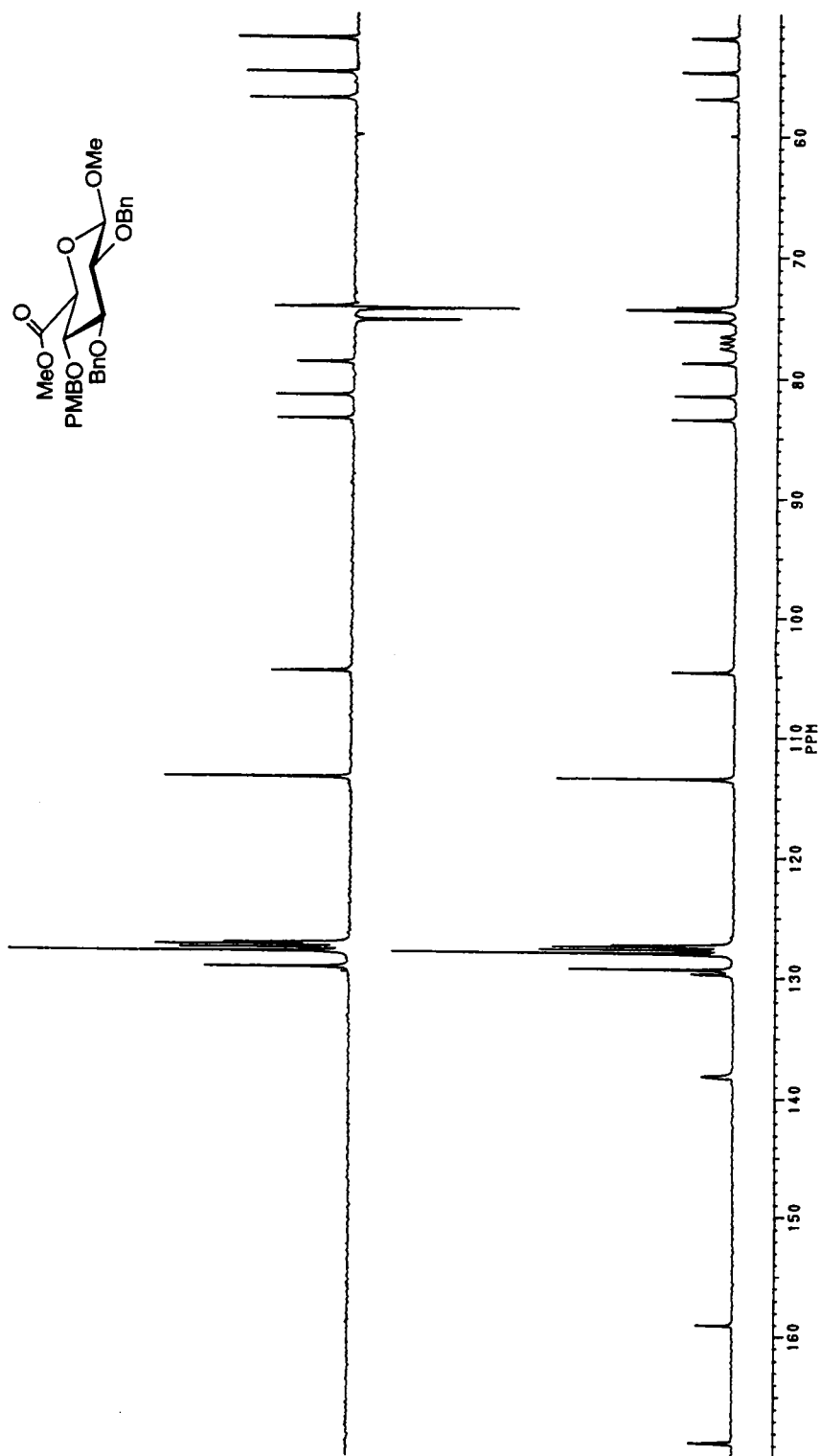


¹H NMR (250 MHz, CDCl₃) spectrum of methyl 2,3-di-O-benzyl-6-O-p-methoxybenzyl-β-D-glucopyranoside (54).

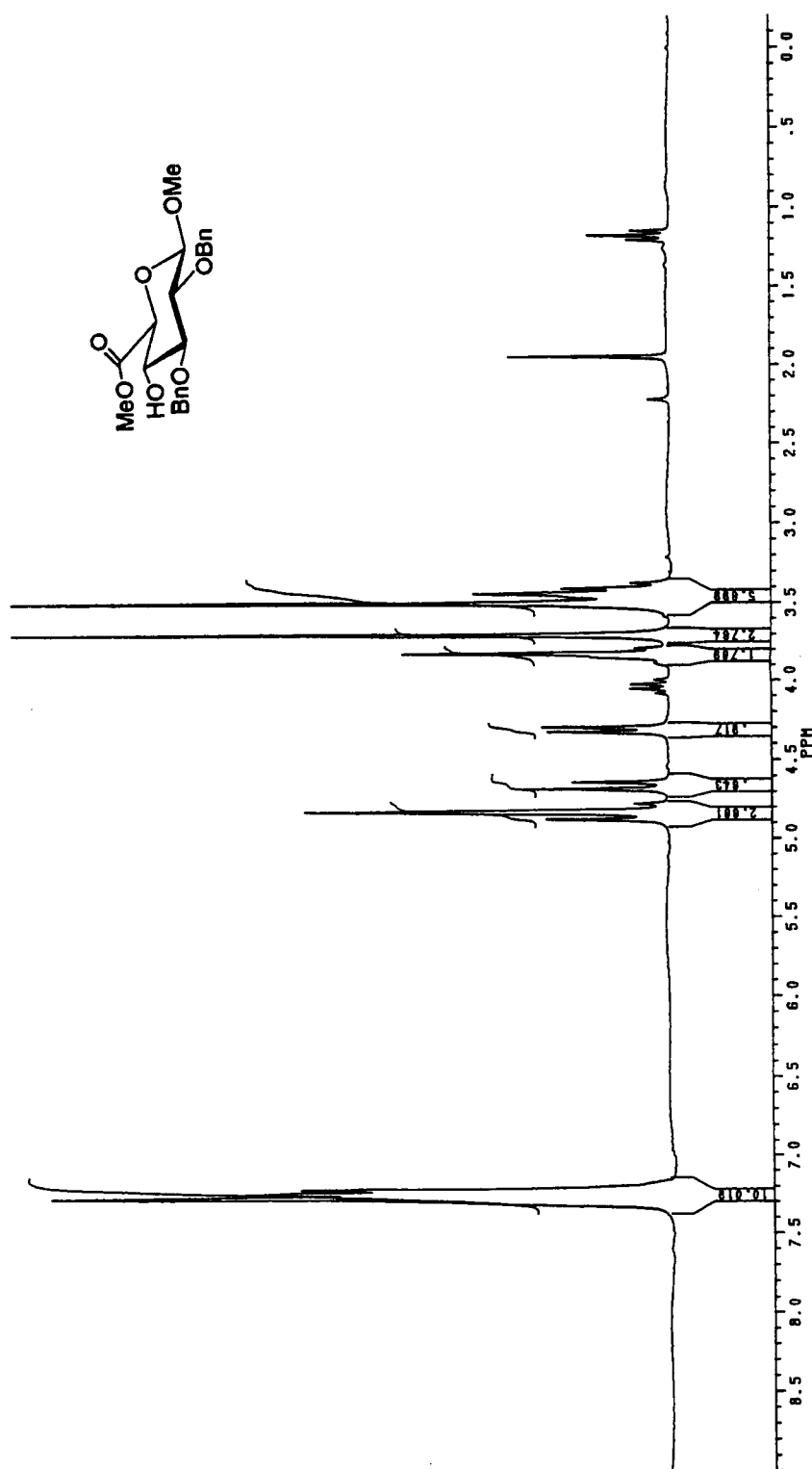




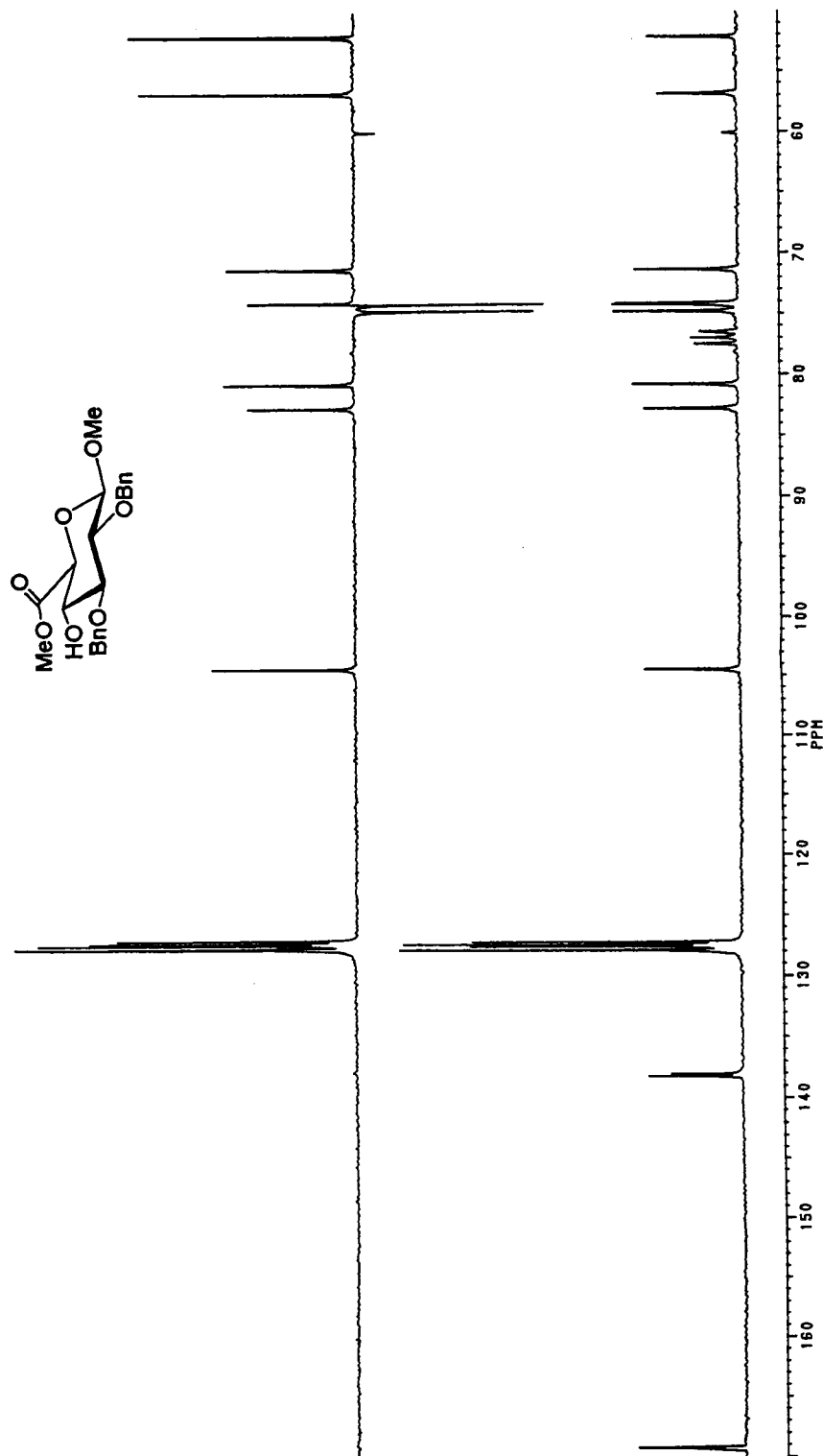
¹H NMR (250 MHz, CDCl₃) spectrum of methyl 2,3-di-O-benzyl-4-O-p-methoxybenzyl-β-D-glucopyranosiduronate (57).

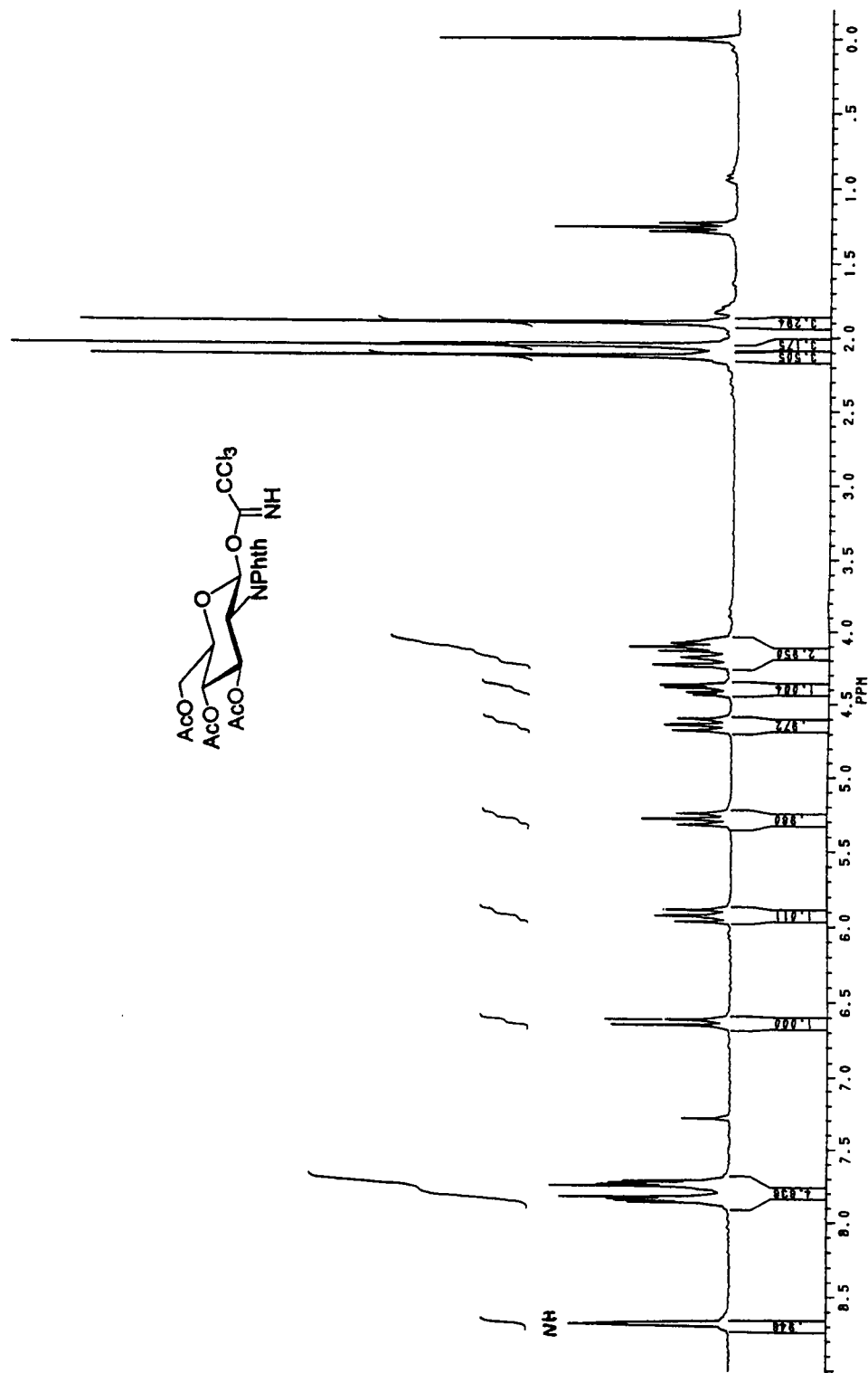


DEPT-135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of methyl 2,3-di-O-benzyl-4-O-p-methoxybenzyl-β-D-glucopyranoside (57).



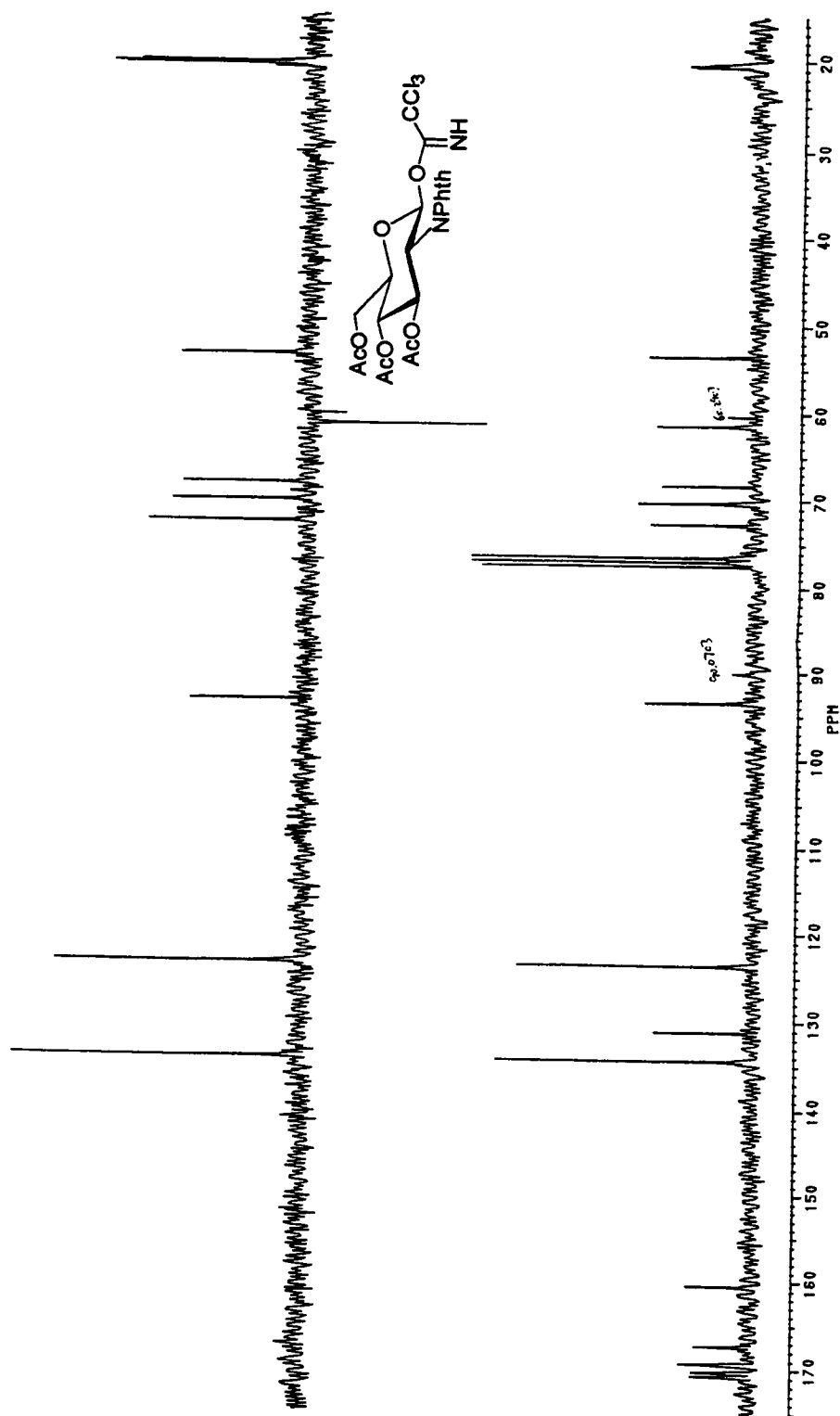
^1H NMR (250 MHz, CDCl_3) spectrum of methyl 2,3-di-O-benzyl- β -D-glucopyranosiduronate (58).



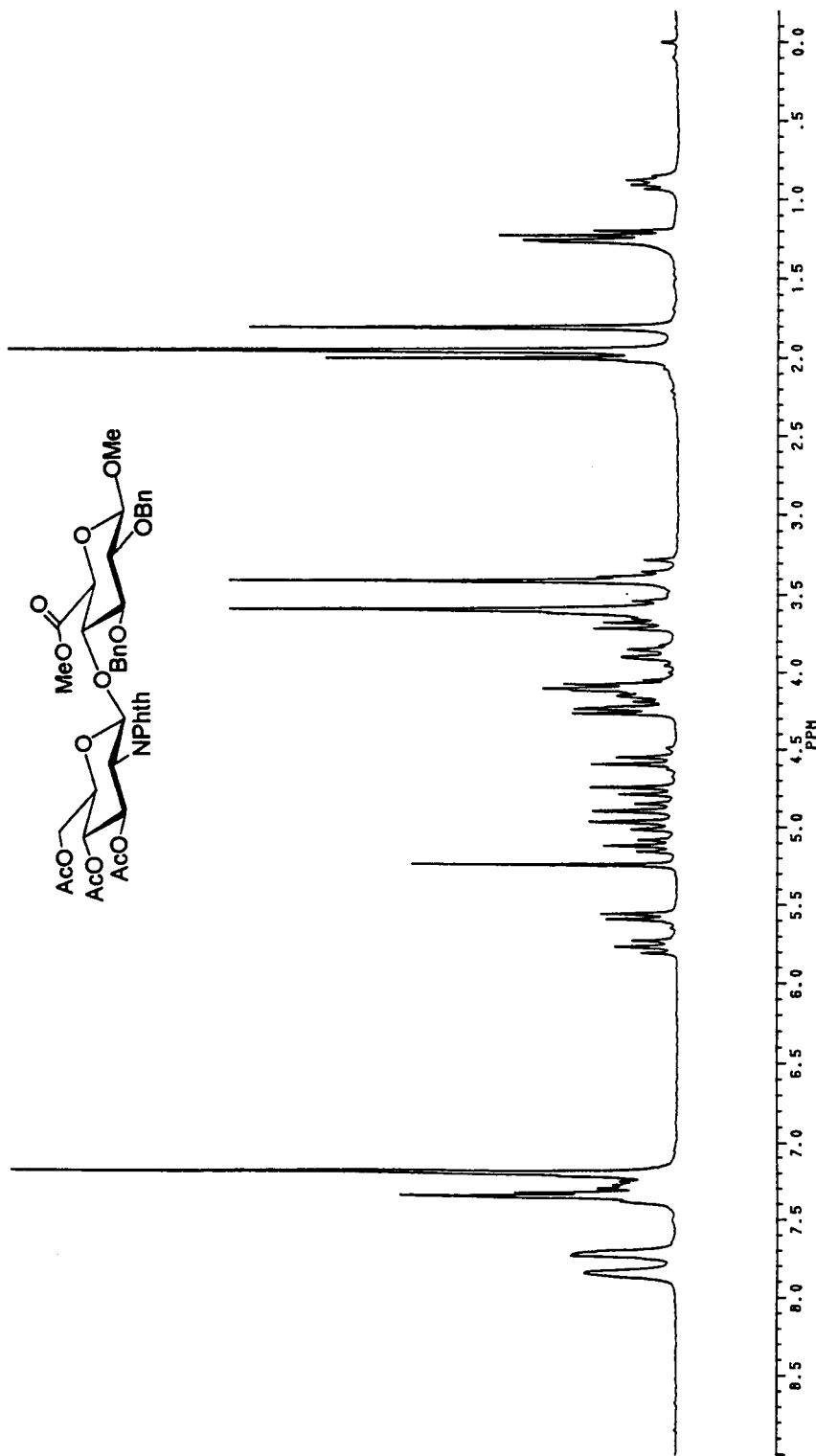


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¹H NMR (250 MHz, CDCl₃) spectrum of 3,4,6-tri-O-acetyl-2-phthalimido-β-D-glucopyranosyl trichloroacetimidate (60).



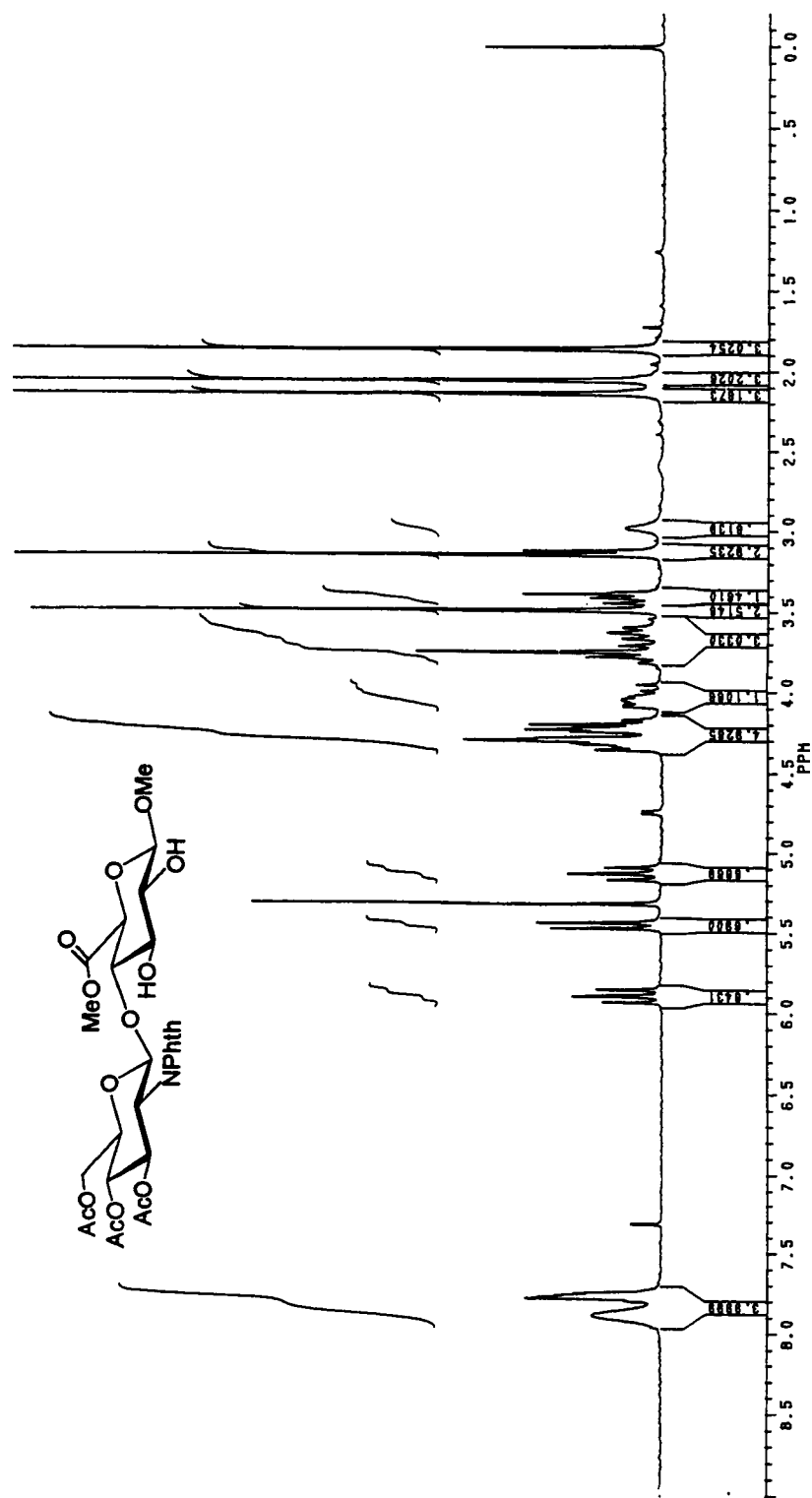
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of 3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl trichloroacetimidate (60).



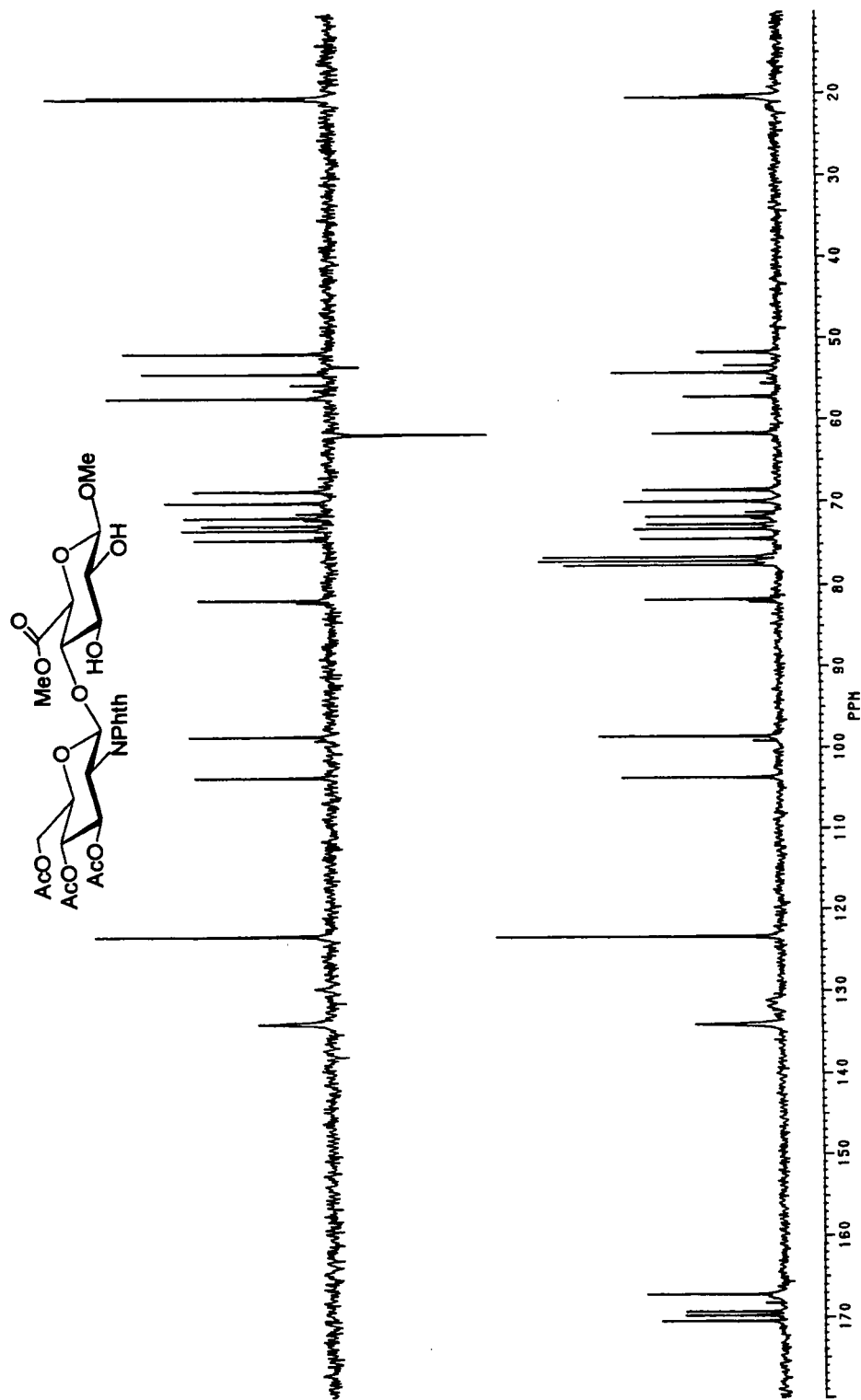
^1H NMR (250 MHz, CDCl_3) spectrum of methyl [(3,4,6-tri-O-acetyl-2-phthalimido-β-D-glucopyranosyl)-(1→4)-(methyl 2,3-di-O-benzyl-β-D-glucopyranosid)]uronate (**61**).



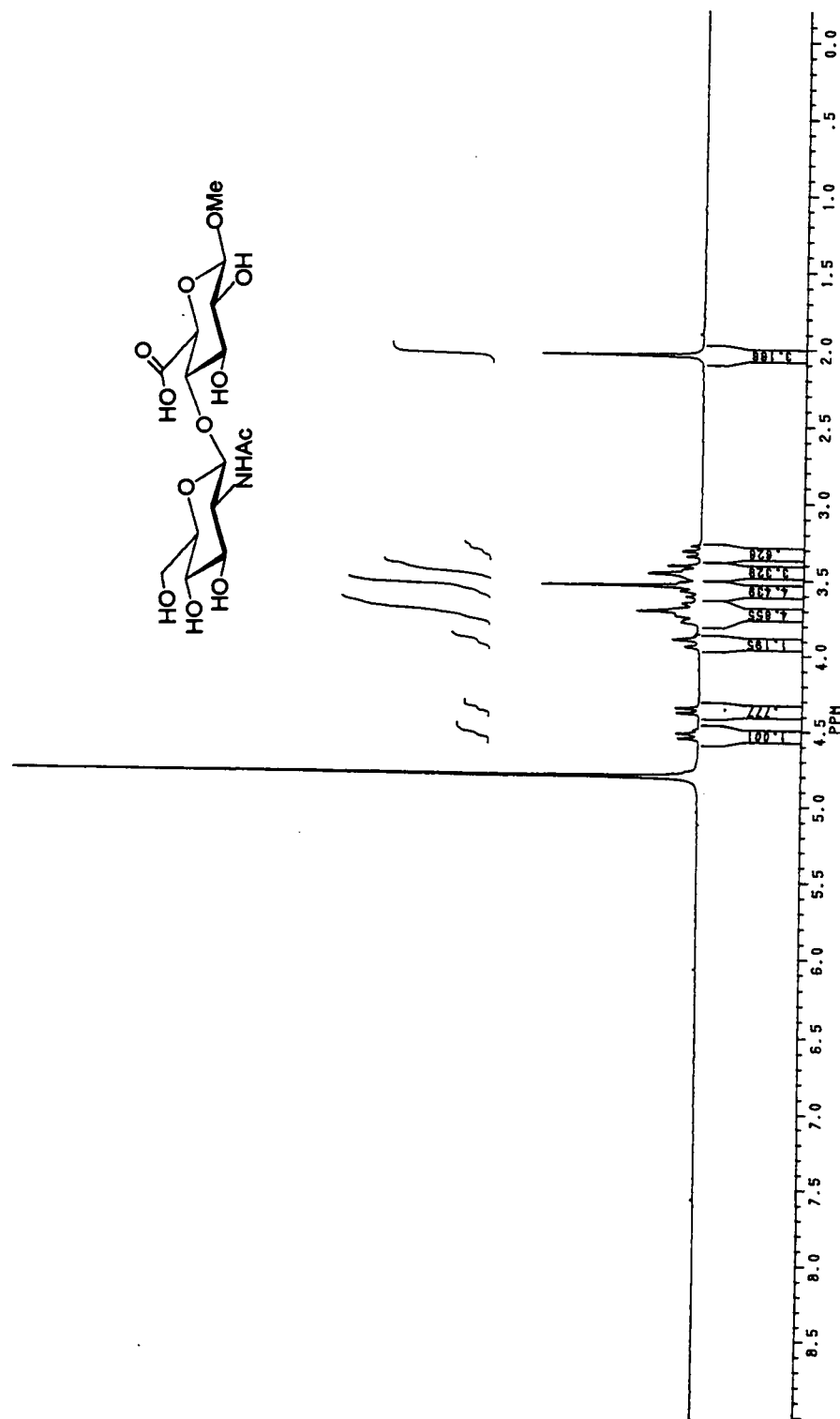
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of methyl [(3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido- β -D-glucopyranosyl)-(1 $\rightarrow$ 4)-(methyl 2,3-di-O-benzyl- β -D-glucopyranosid)]uronate (**61**).



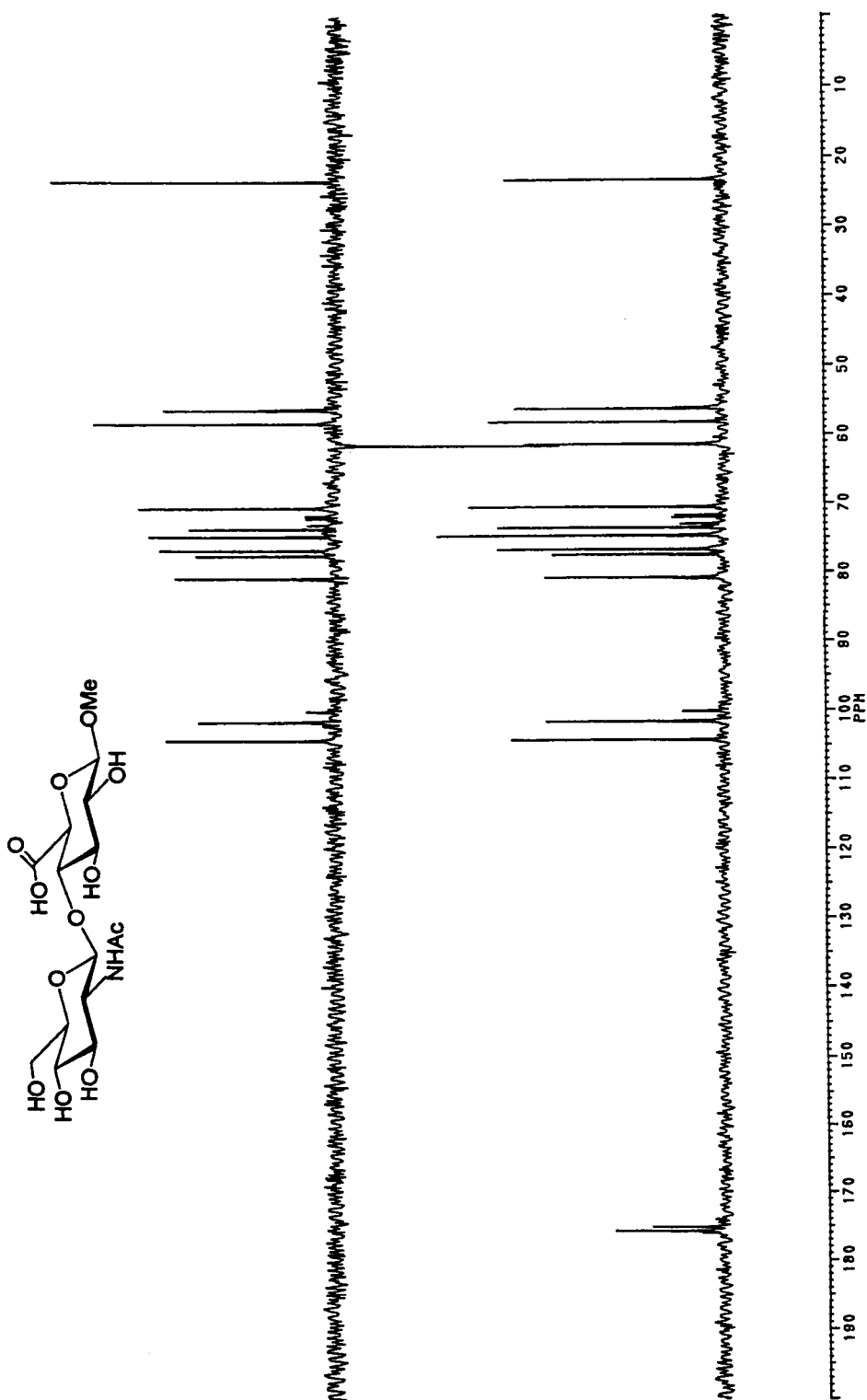
^1H NMR (250 MHz, CDCl_3) spectrum of methyl [(3,4,6-tri-O-acetyl-2-phthalimido- β -D-glucopyranosyl)-(1 $\rightarrow$ 4)-(methyl β -D-glucopyranosid)]uronate (**62**).



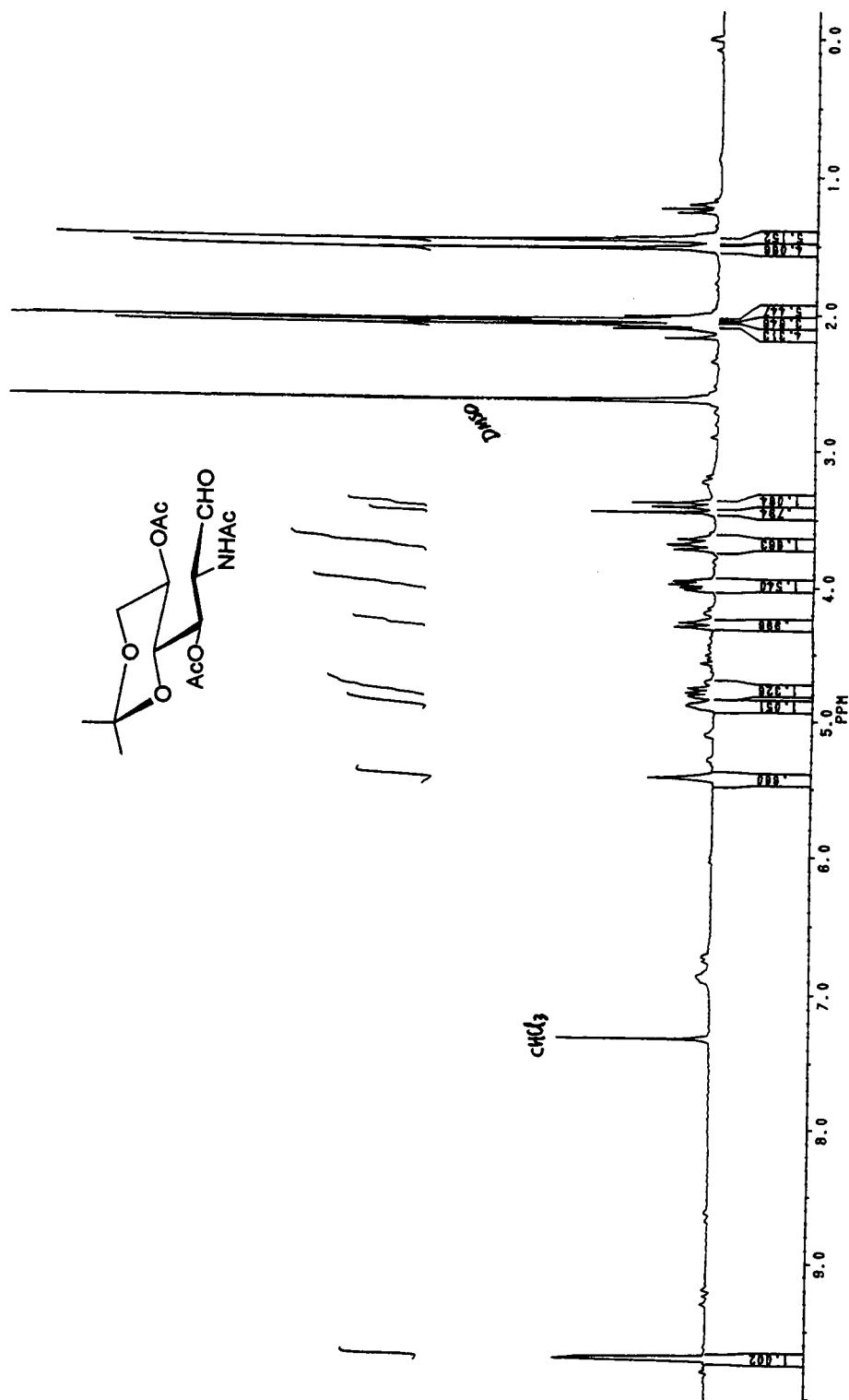
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of methyl [(3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-β-D-glucopyranosyl)-(1→4)-(methyl β-D-glucopyranosid)]uronate (62).



¹H NMR (250 MHz, D₂O) spectrum of methyl (2-deoxy-2-acetamido-β-D-glucopyranosyl-(1→4)-β-D-glucopyranosid)uronic acid (67).

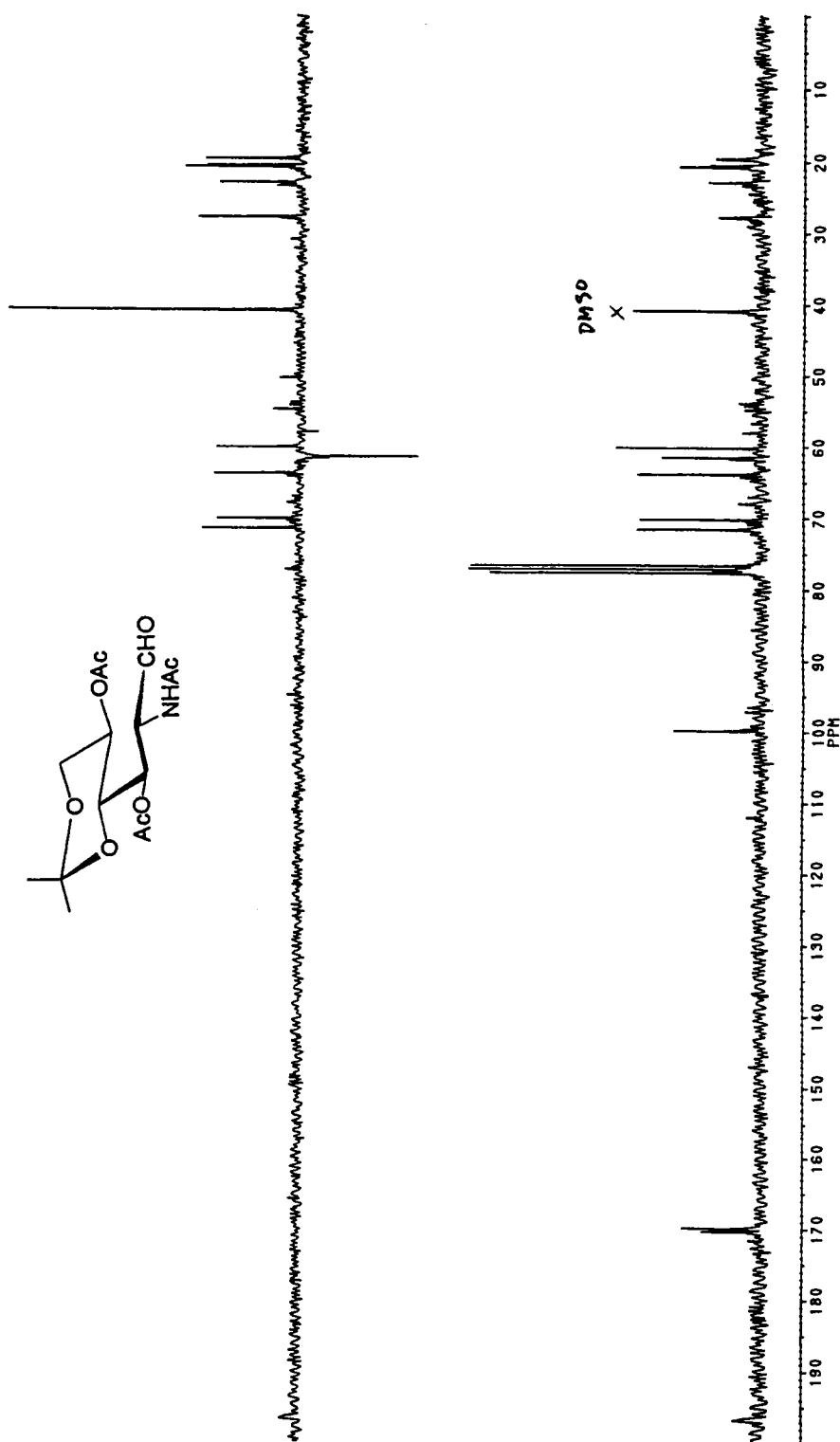


DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, D_2O) spectra of methyl (2-deoxy-2-acetamido- β -D-glucopyranosyl)-(1 $\rightarrow$ 4)- β -D-glucopyranosiduronic acid (**67**).



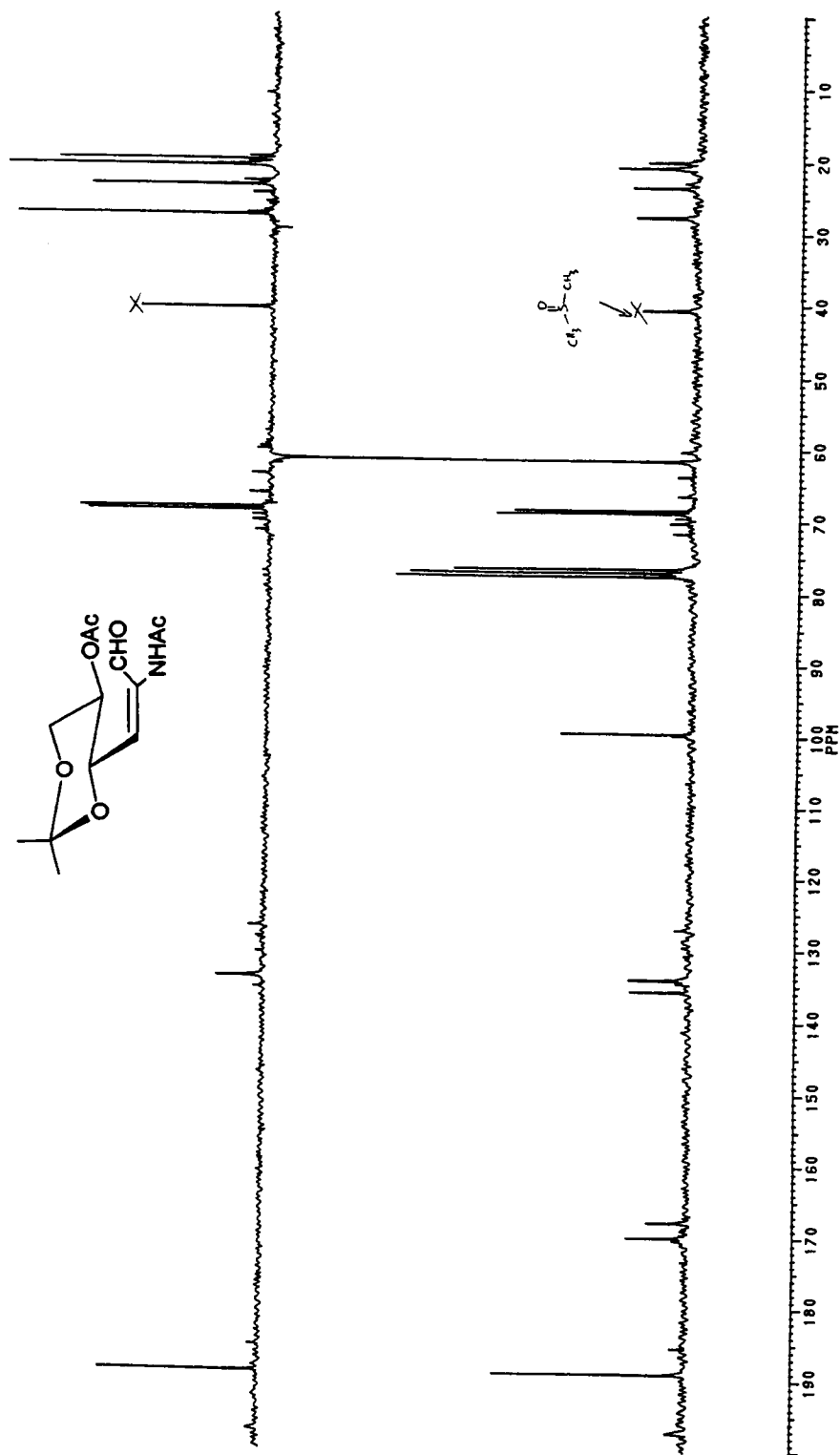
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¹H NMR (250 MHz, CDCl₃) spectrum of 2-acetamido-3,5-di-O-acetyl-4,6-O-isopropylidene-D-glucose (71).

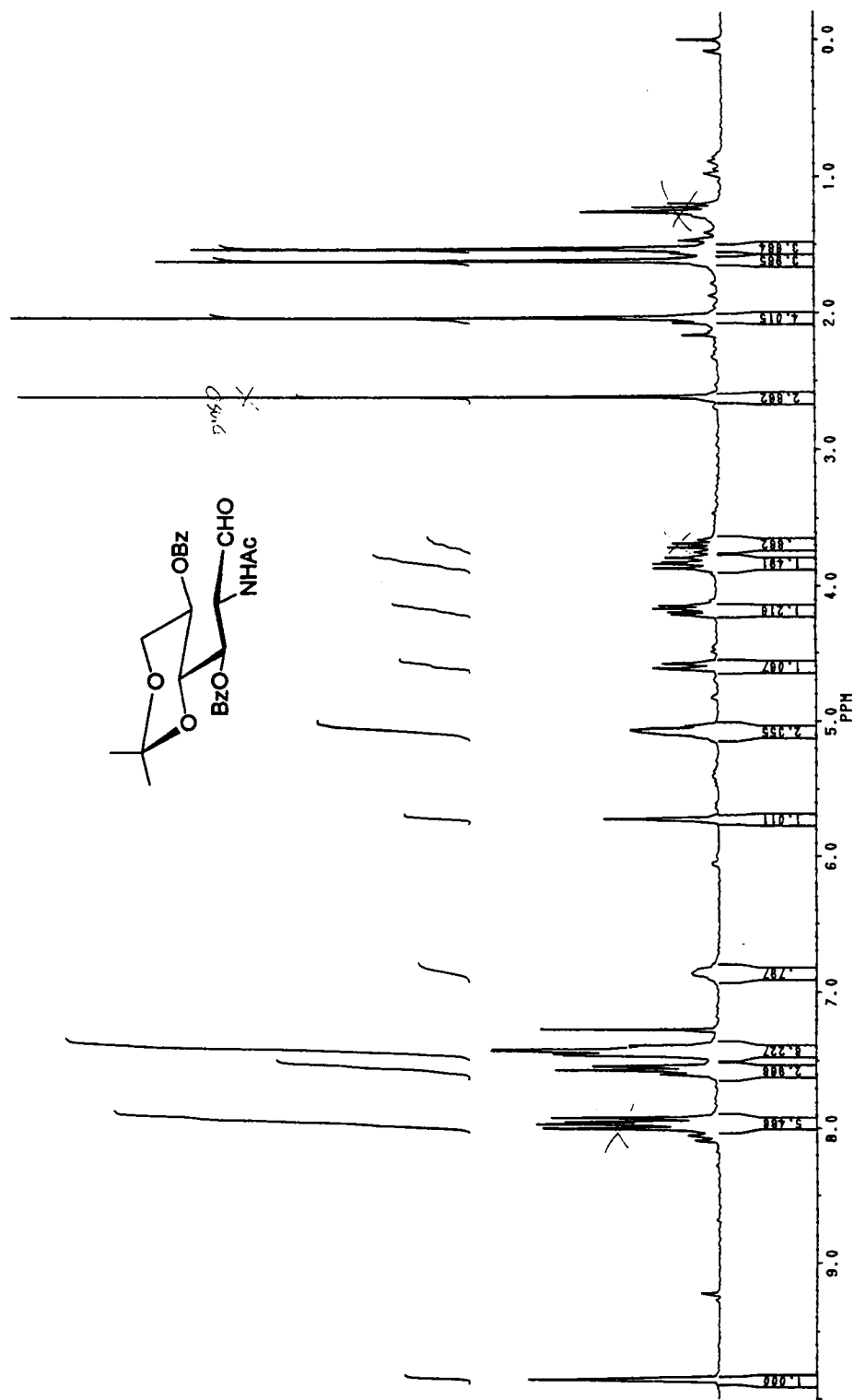


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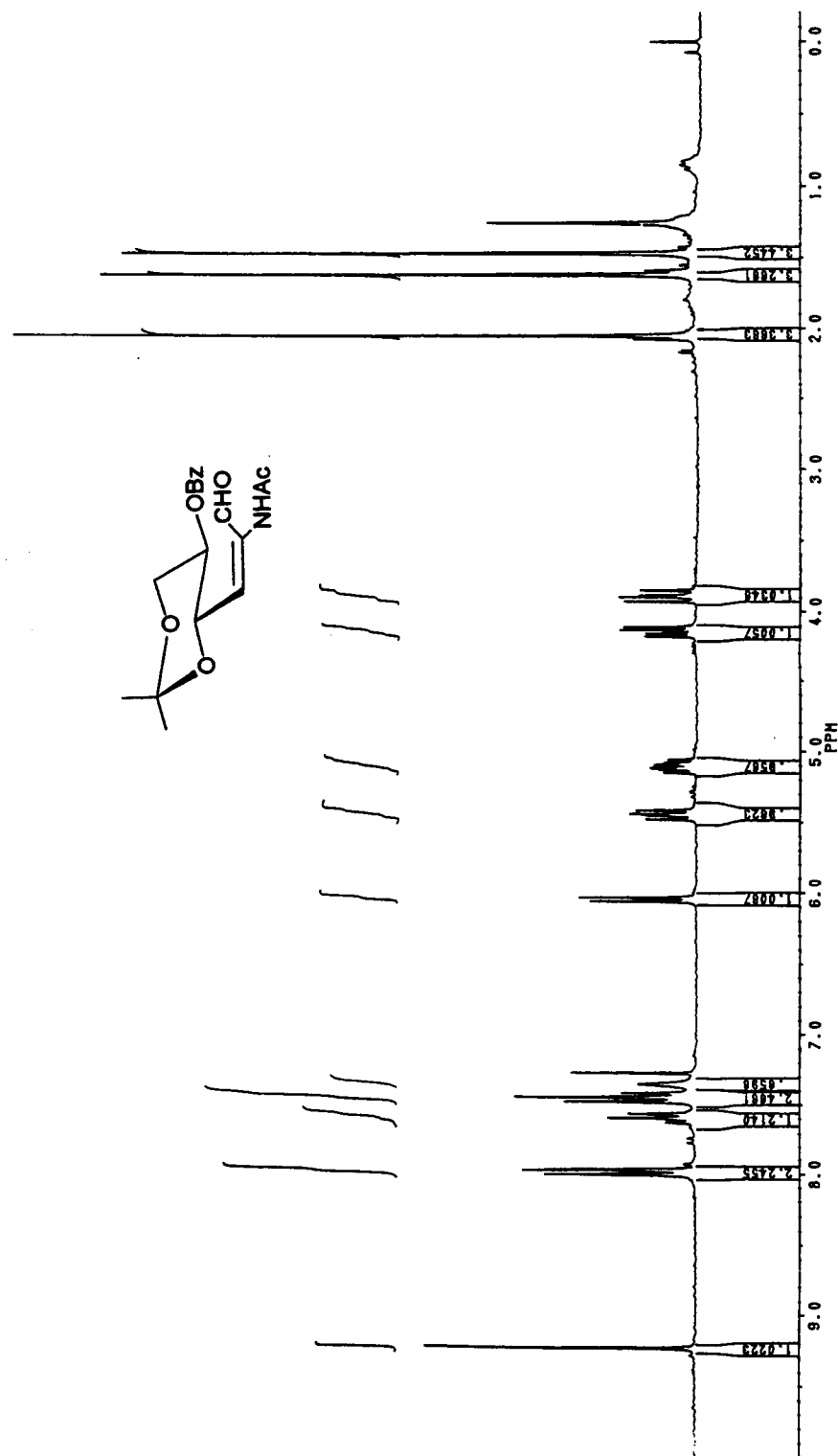
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of 2-acetamido-3,5-di-O-acetyl-2-deoxy-4,6-O-isopropylidene-D-glucose (71).



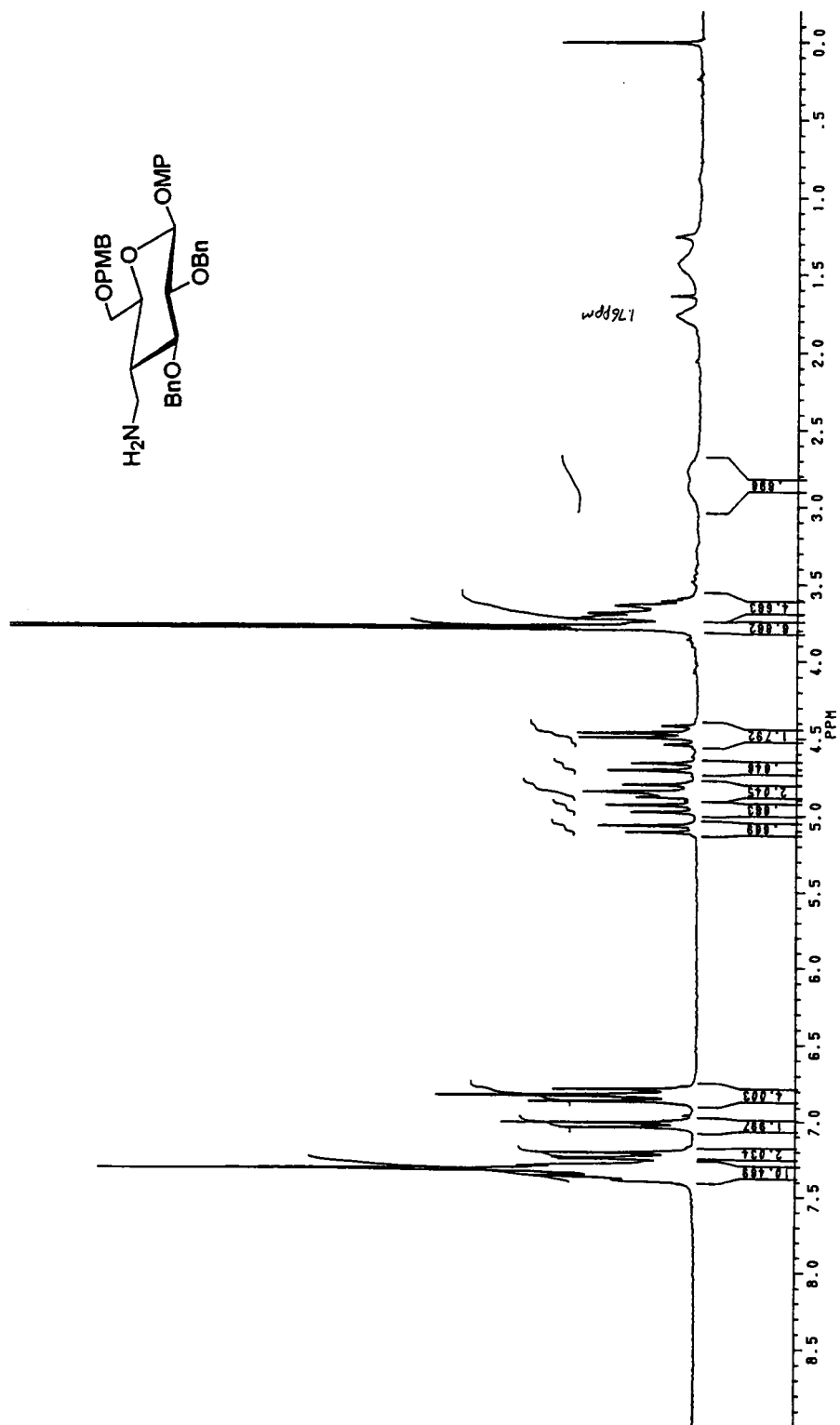
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of 2-acetamido-5-O-acetyl-2,3-dideoxy-4,6-O-isopropylidene- α -D-erythro-hex-2-enose (72).



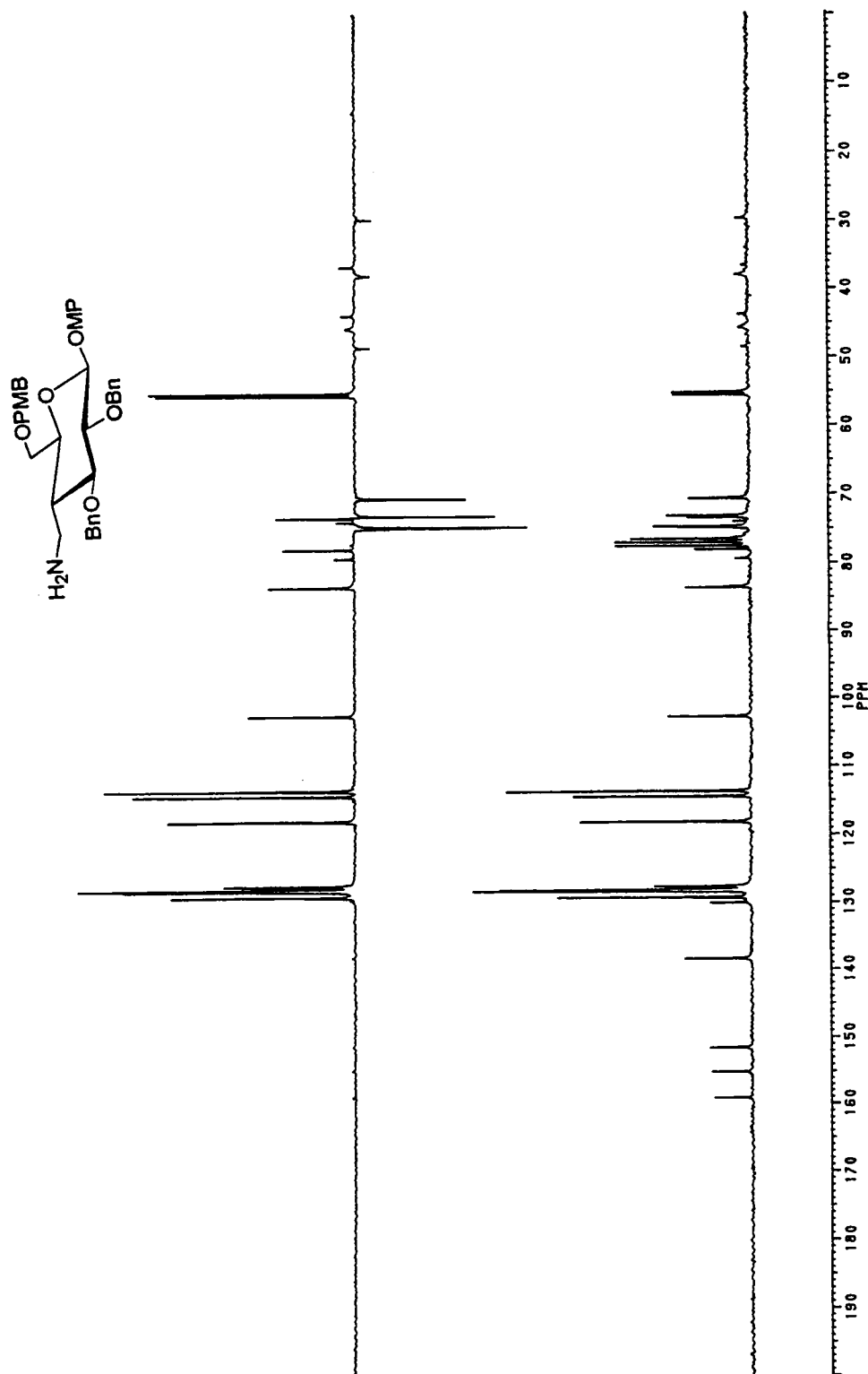
¹H NMR (250 MHz, CDCl₃) spectrum of 2-acetamido-3,5-di-O-benzoyl-2-deoxy-4,6-O-isopropylidene-D-glucose (74).



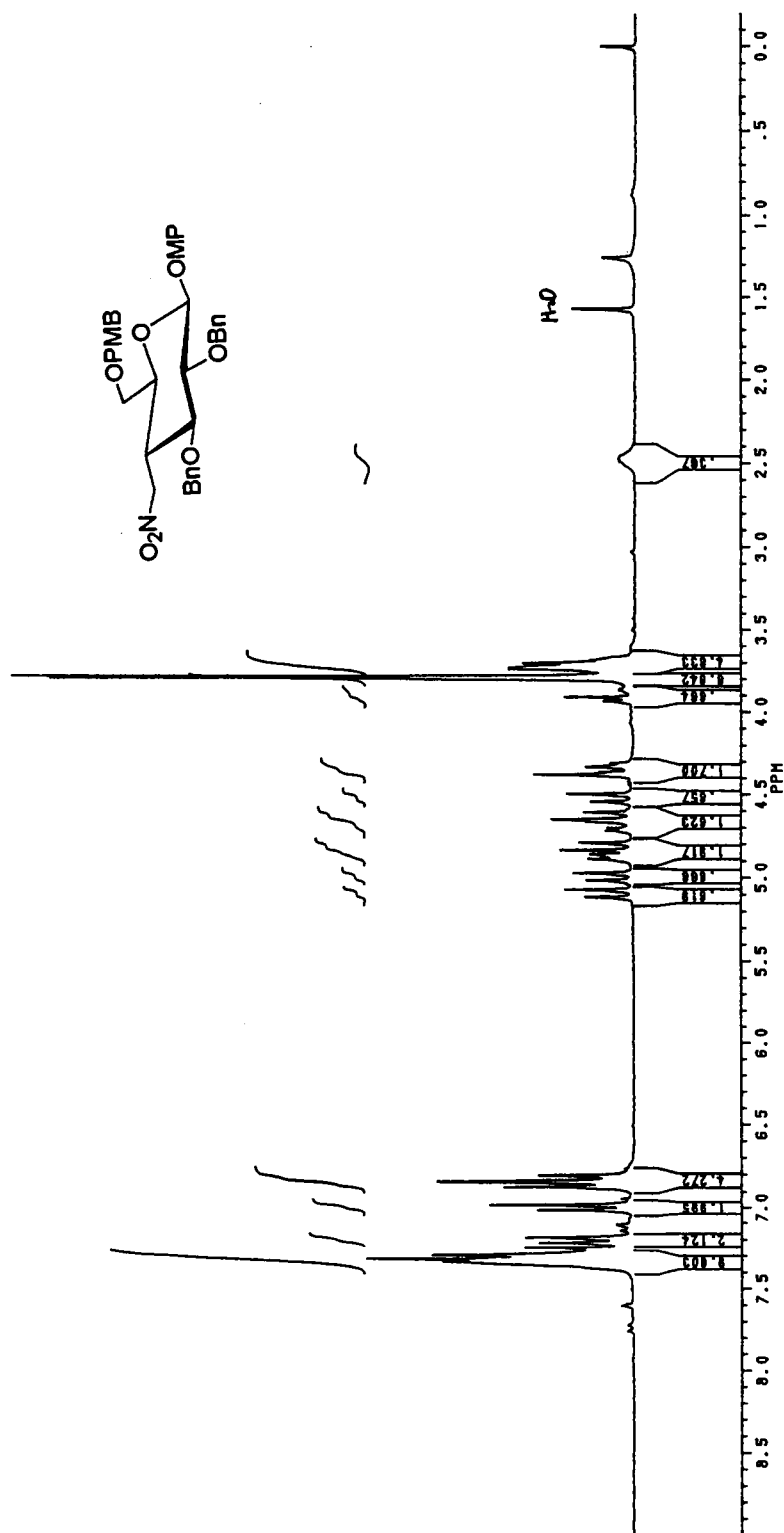
¹H NMR (250 MHz, CDCl₃) spectrum of 2-acetamido-5-O-benzoyl-2,3-dideoxy-4,6-O-isopropylidene-aldehyde-D-erythro-hex-2-enose (**75**).



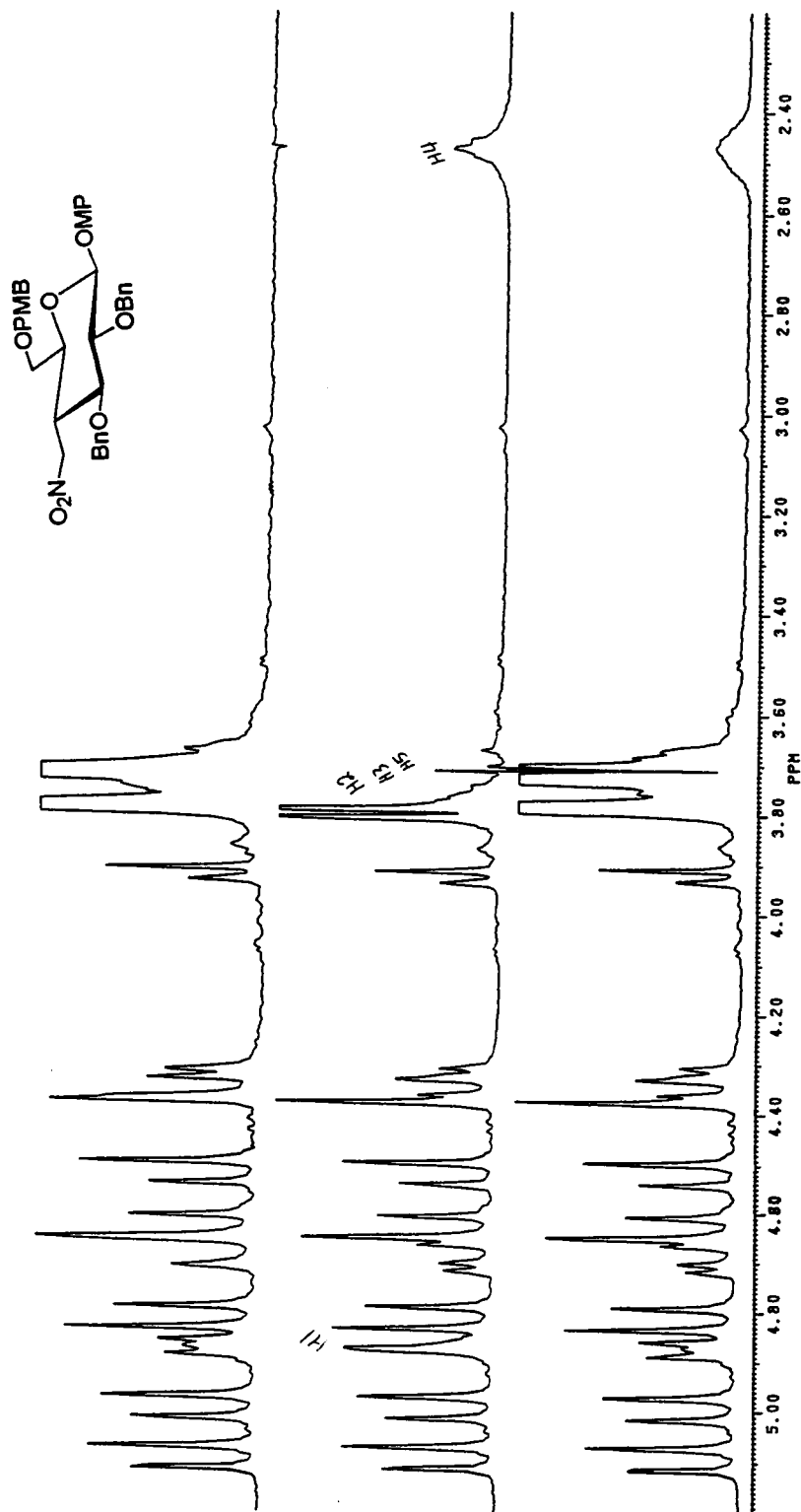
¹H NMR (250 MHz, CDCl₃) spectrum of *p*-methoxyphenyl 4-C-aminomethyl-2,3-di-O-benzyl-4-deoxy-6-O-*p*-methoxybenzyl-β-D-glucopyranoside (**91**).



DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of *p*-methoxyphenyl 4-*C*-aminomethyl-2,3-di-*O*-benzyl-4-deoxy-6-*O*-*p*-methoxybenzyl- β -D-glucopyranoside (91).

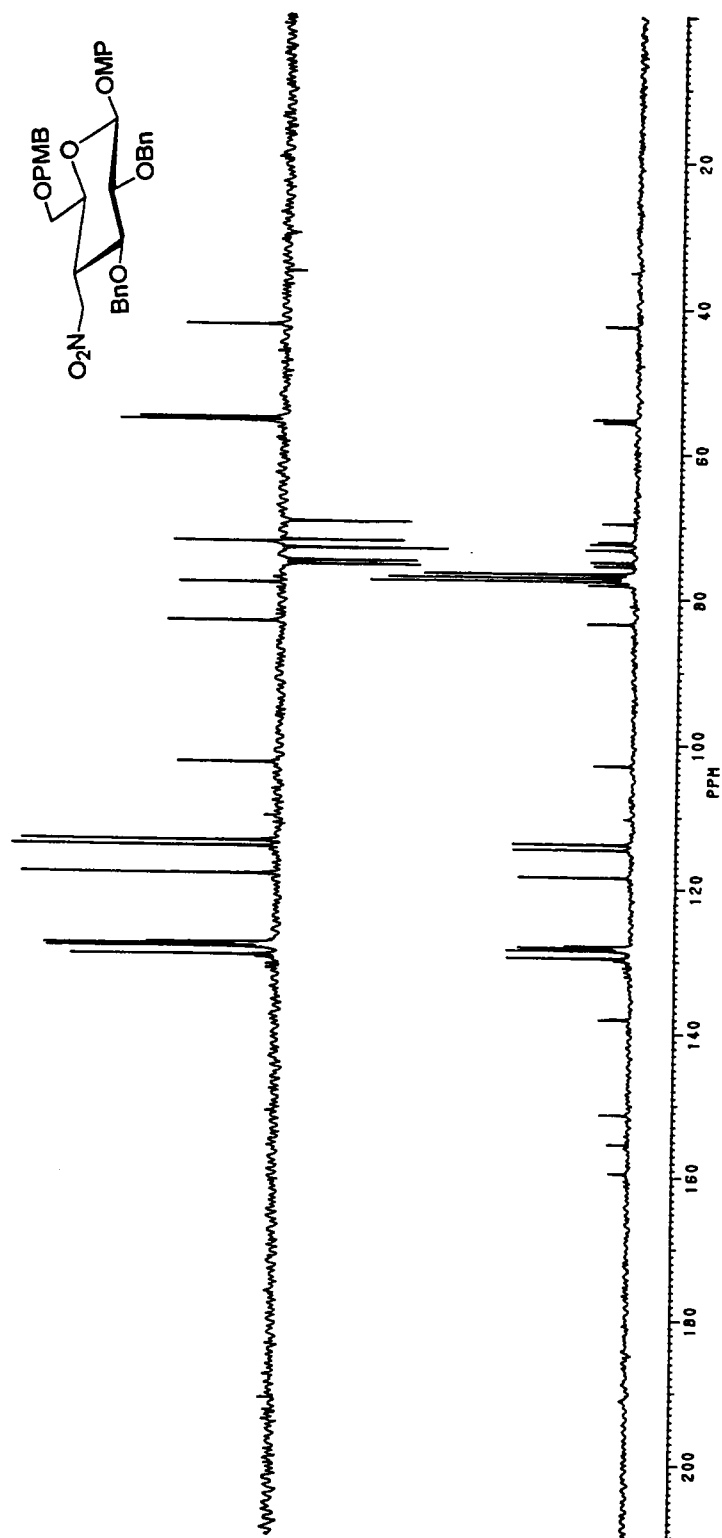


¹H NMR (250 MHz, CDCl₃) spectrum of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-6-O-*p*-methoxybenzyl-4-C-nitromethyl-β-D-glucopyranoside (94).

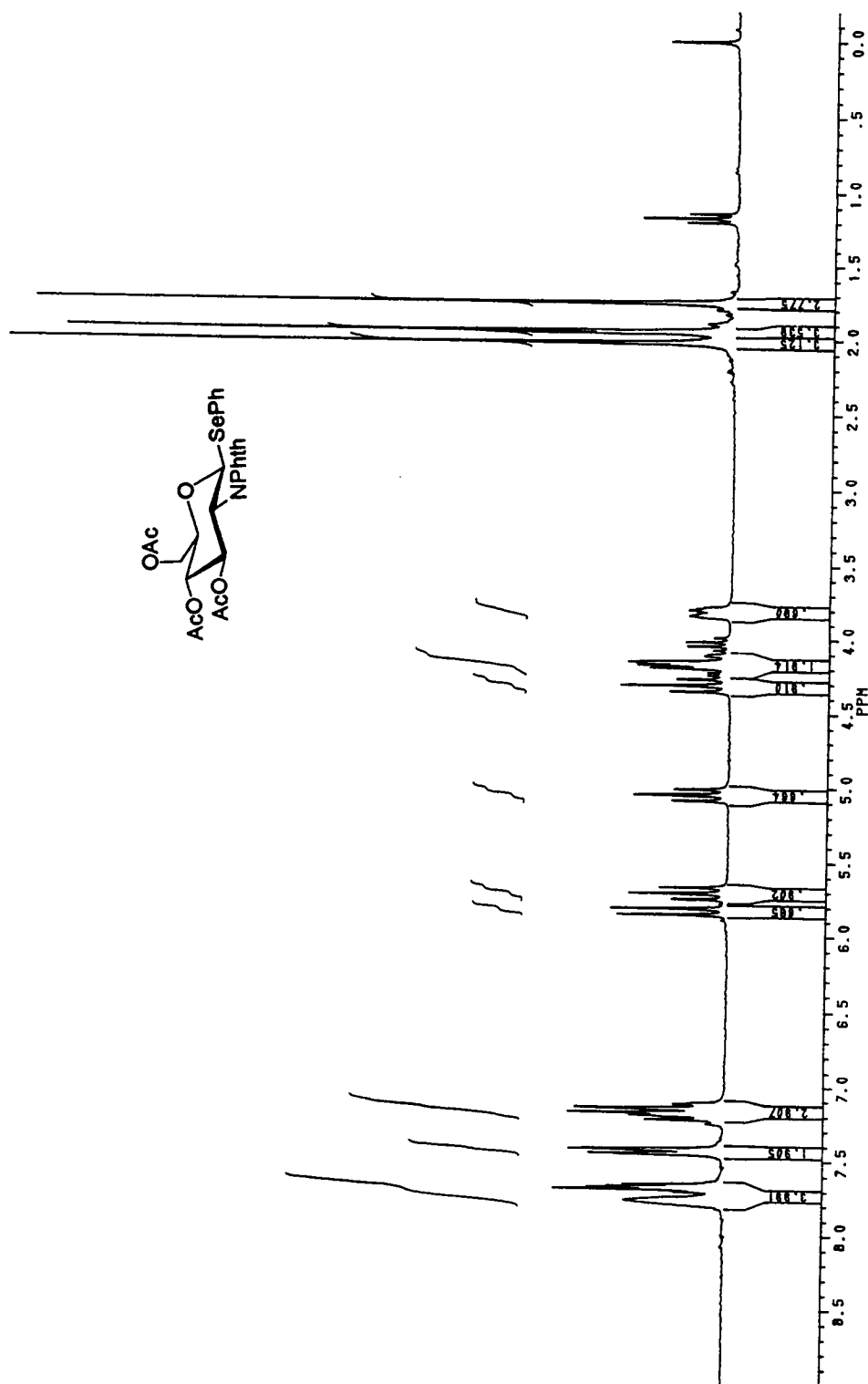


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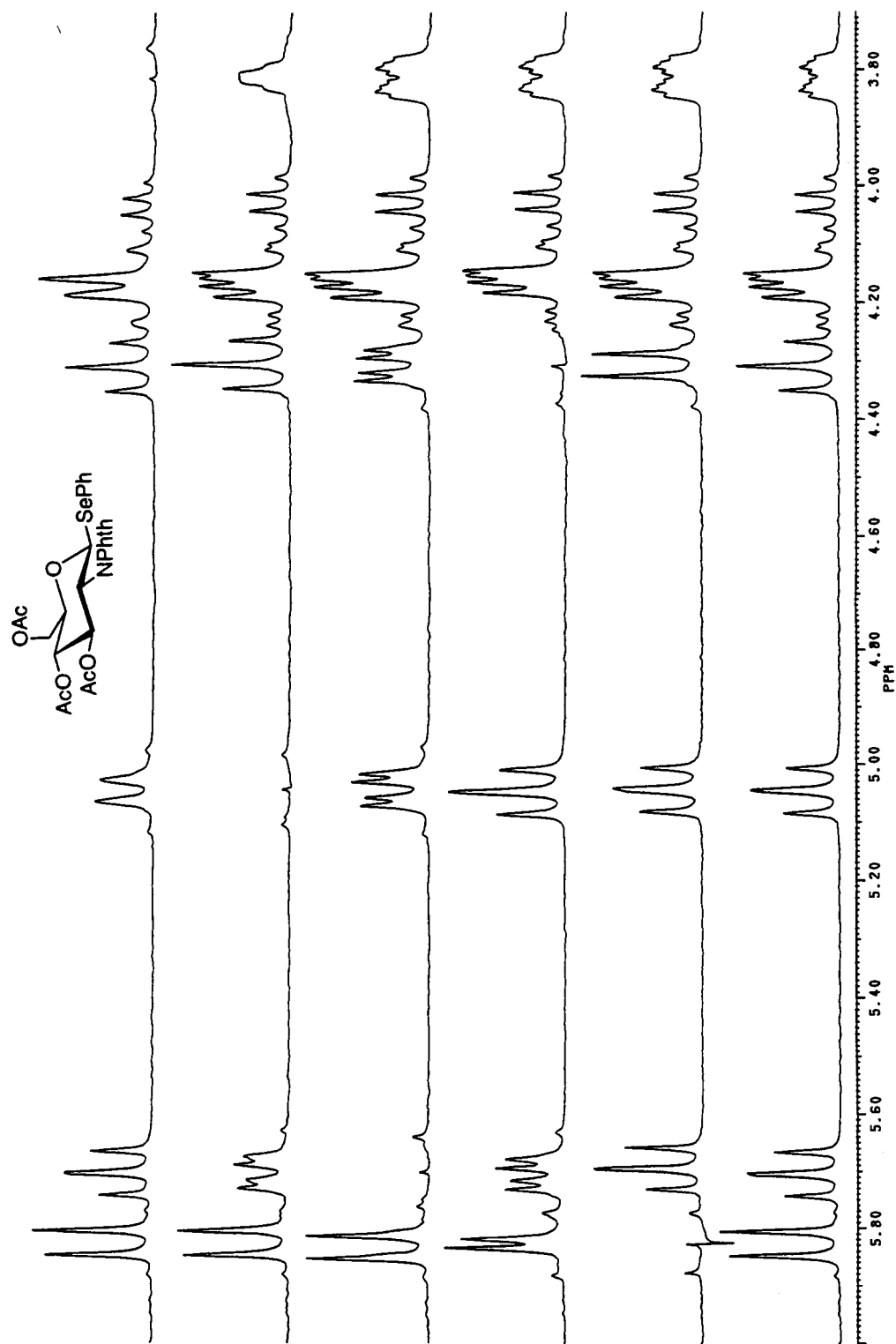
^1H double resonance spectrum of *p*-methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-6-*O*-*p*-methoxybenzyl-4-*C*-nitromethyl- β -D-glucopyranoside (**94**).

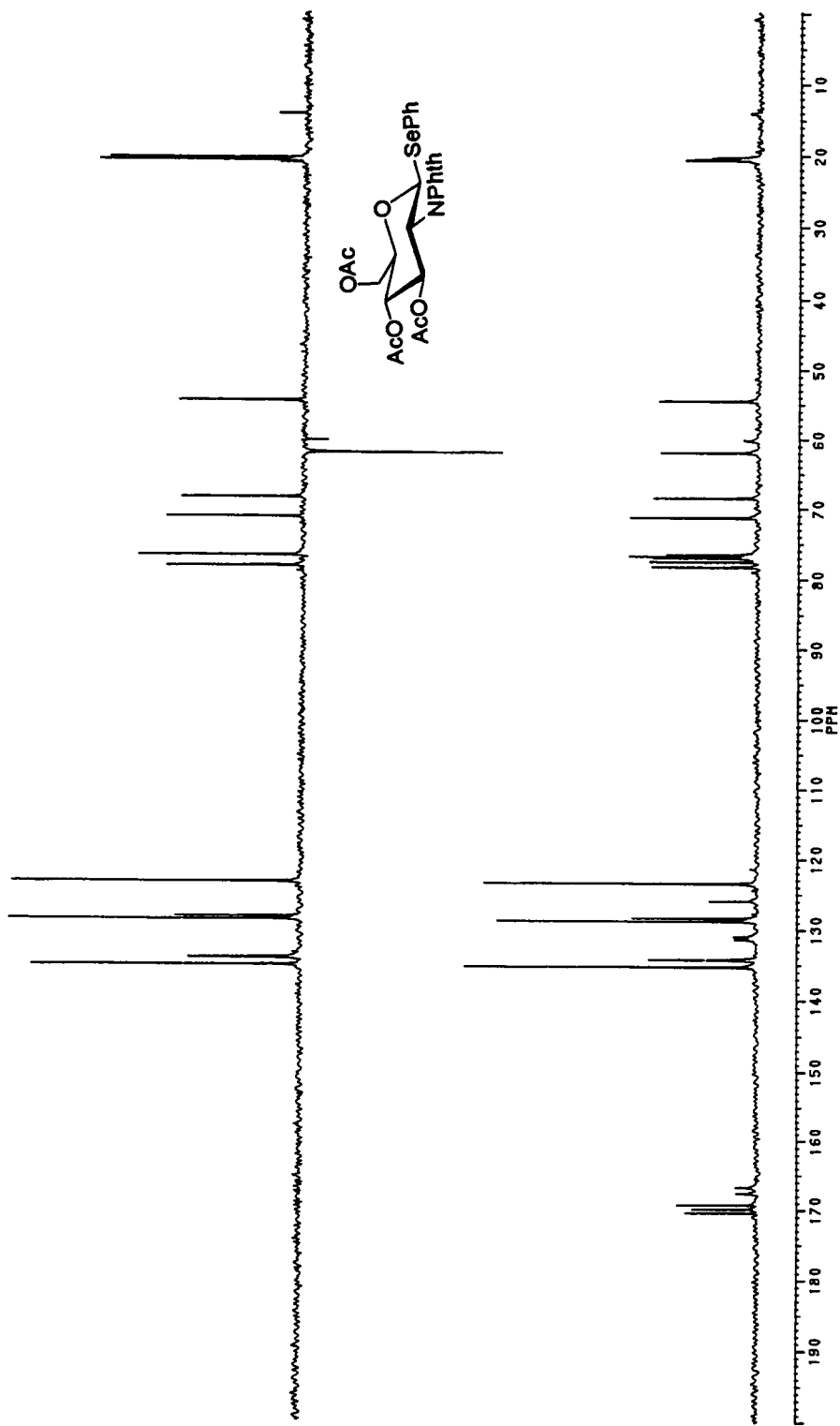


DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-6-O-*p*-methoxybenzyl-4-C-nitromethyl-β-D-glucopyranoside (**94**).

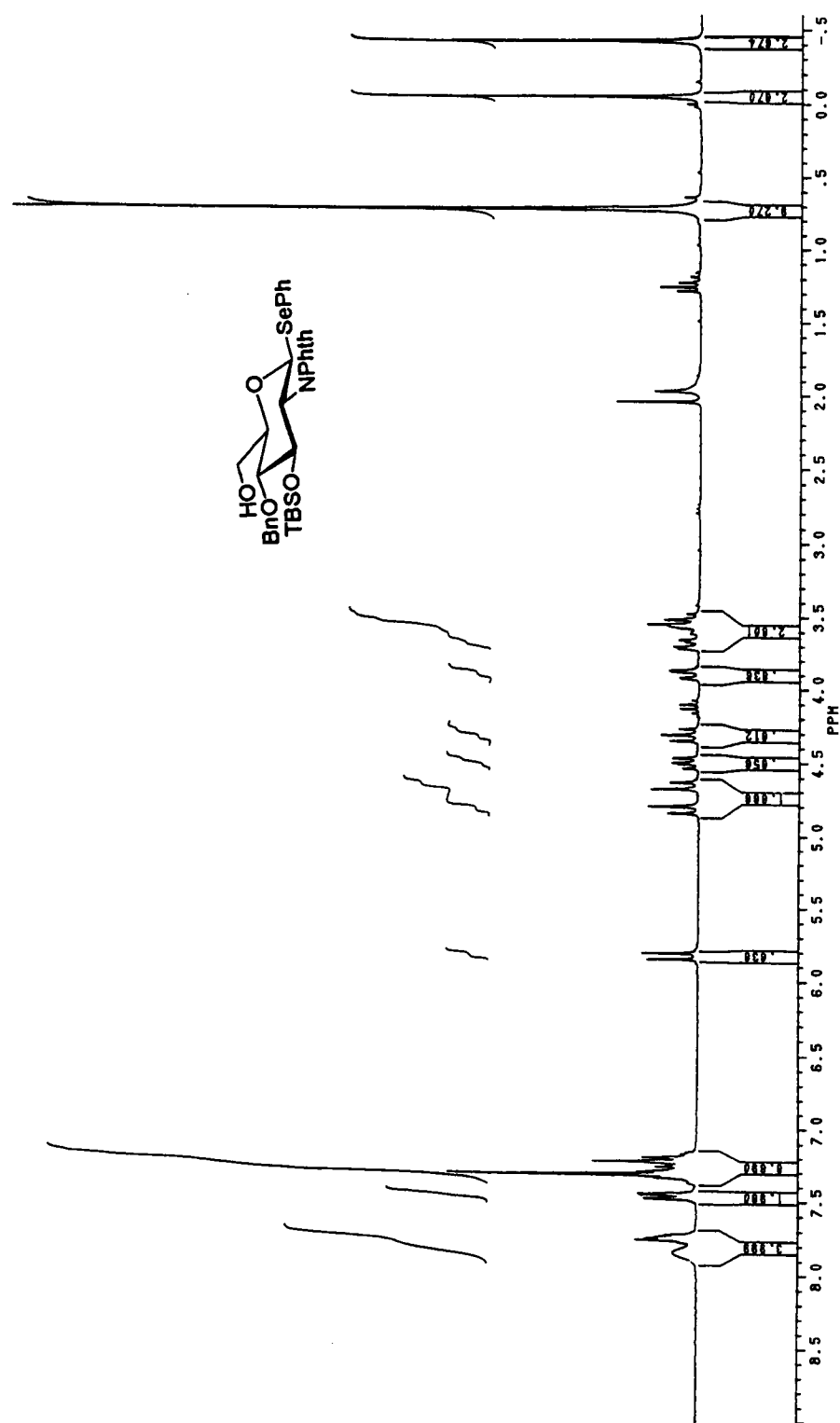


¹H NMR (250 MHz, CDCl₃) spectrum of phenyl 3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-1-seleno- β -D-glucopyranoside (**95**).

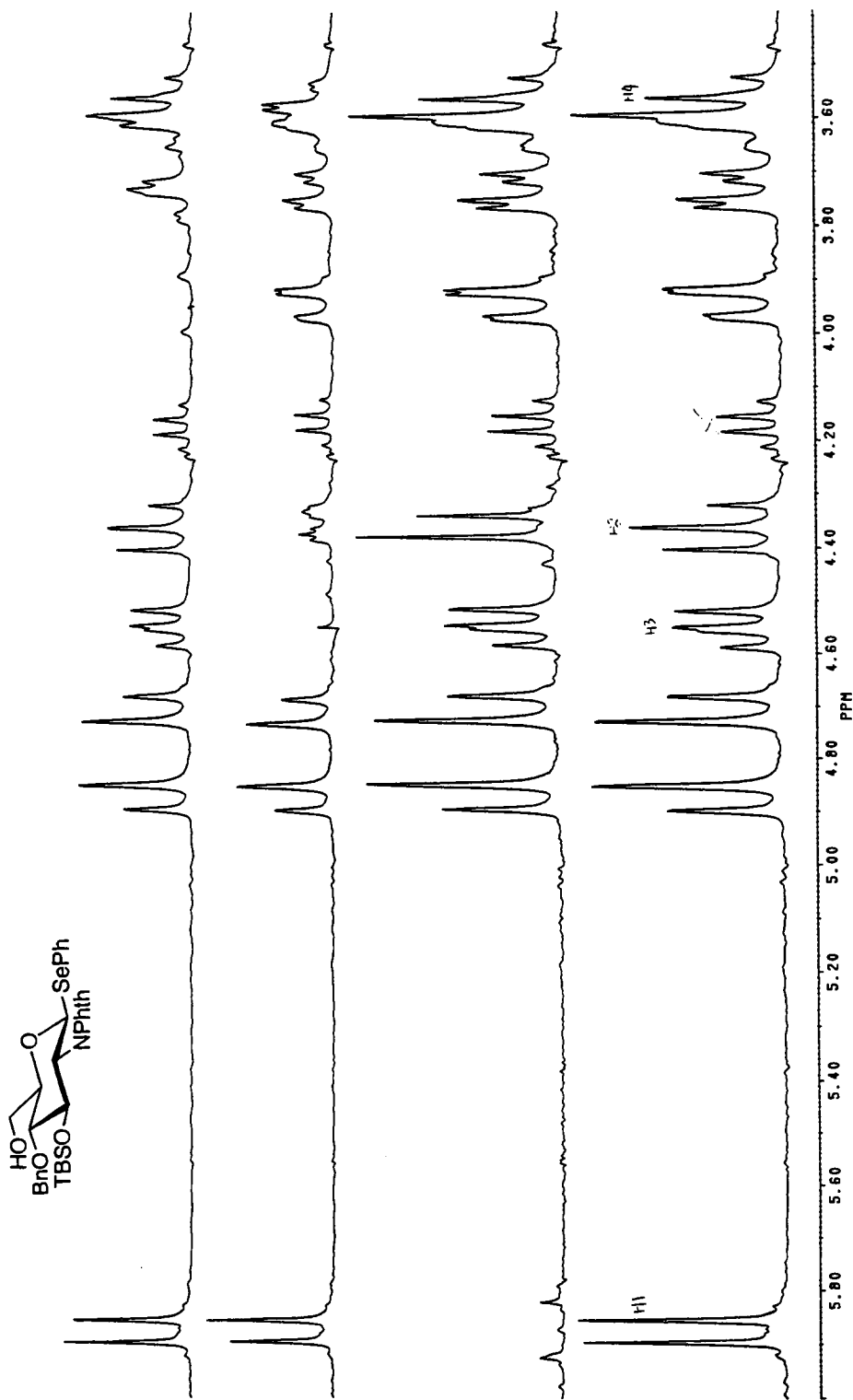


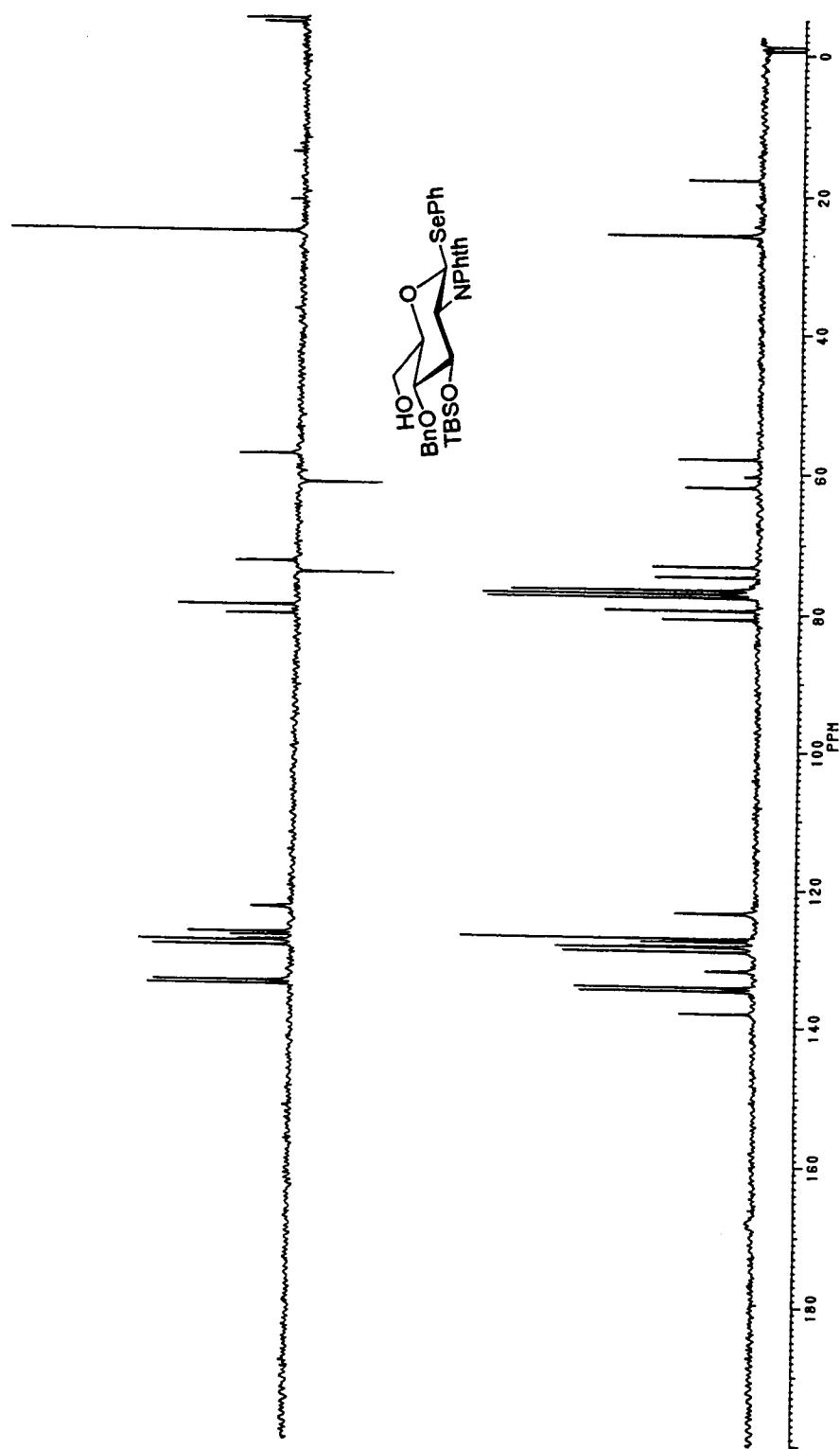


DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of phenyl 3,4,6-tri-O-acetyl-2-deoxy-2-phthalimido-1-seleno-β-D-glucopyranoside (**95**).



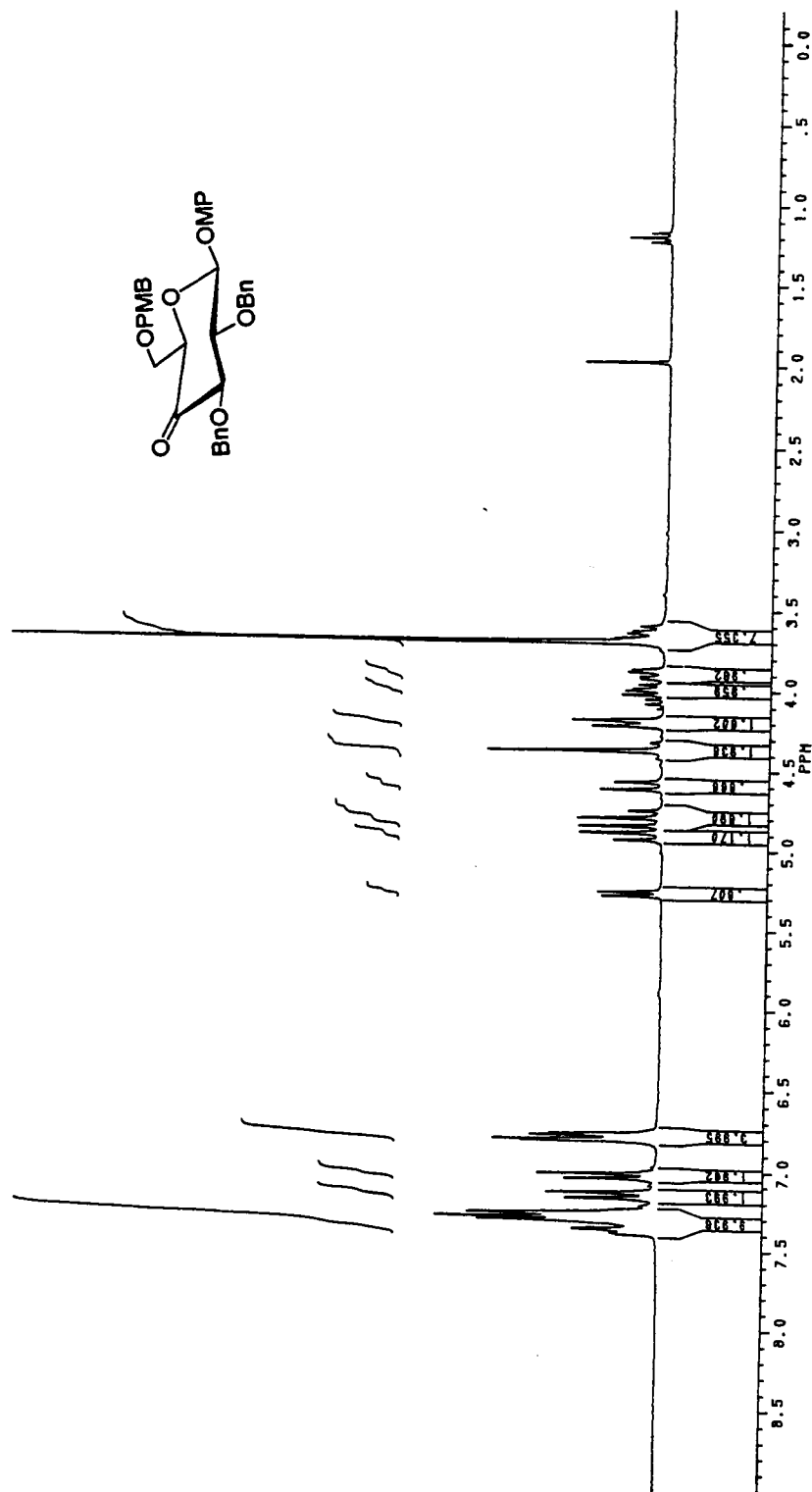
^1H NMR (250 MHz, CDCl_3) spectrum of phenyl 4-O-benzyl-3-(*tert*-butyldimethylsilyl)-2-deoxy-2-phthalimido-1-seleno- β -D-glucopyranoside (**98**).



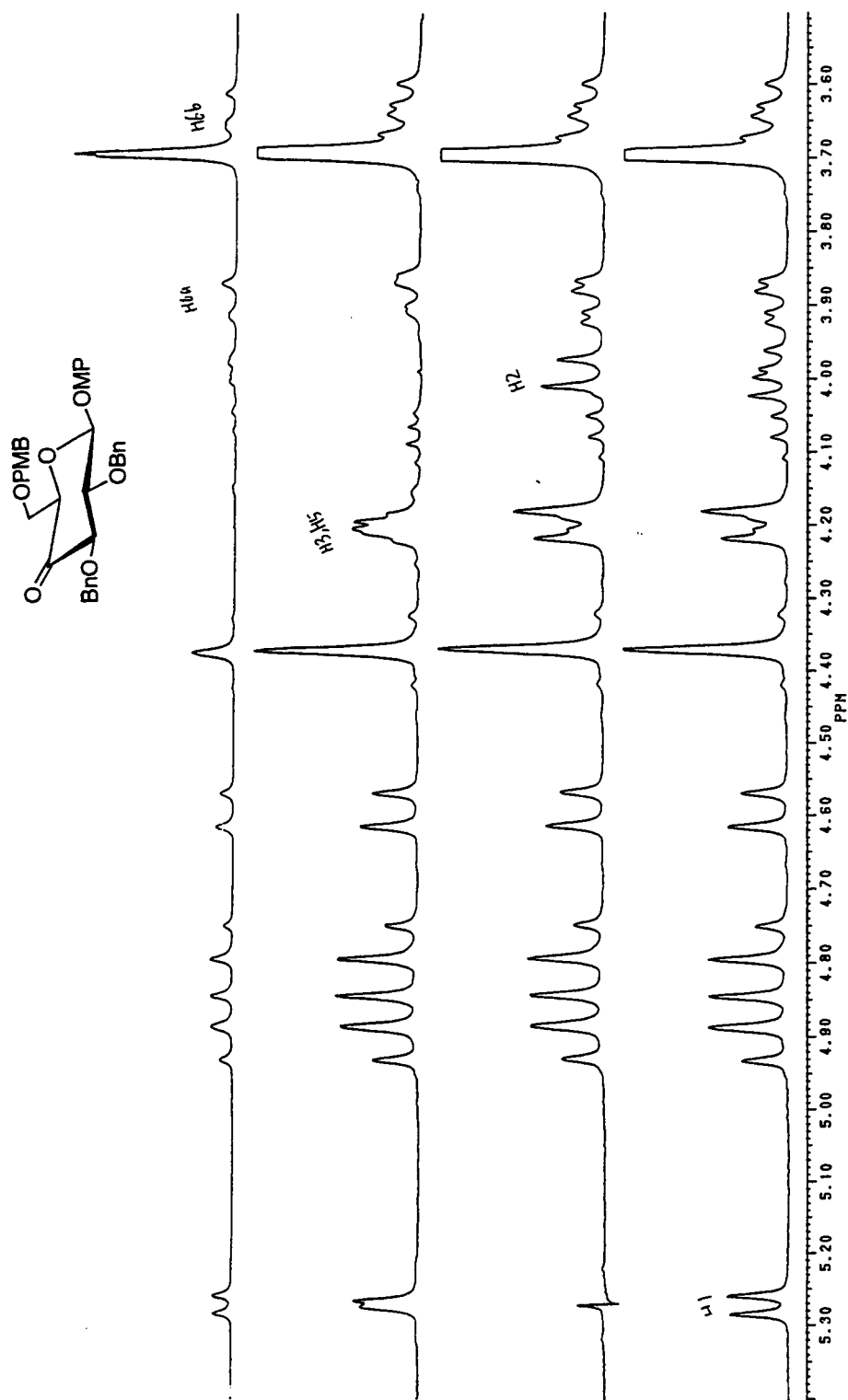


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DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of phenyl 4-O-benzyl-3-(*tert*-butyldimethylsilyl)-2-deoxy-2-phthalimido-1-seleno- β -D-glucopyranoside (**98**).

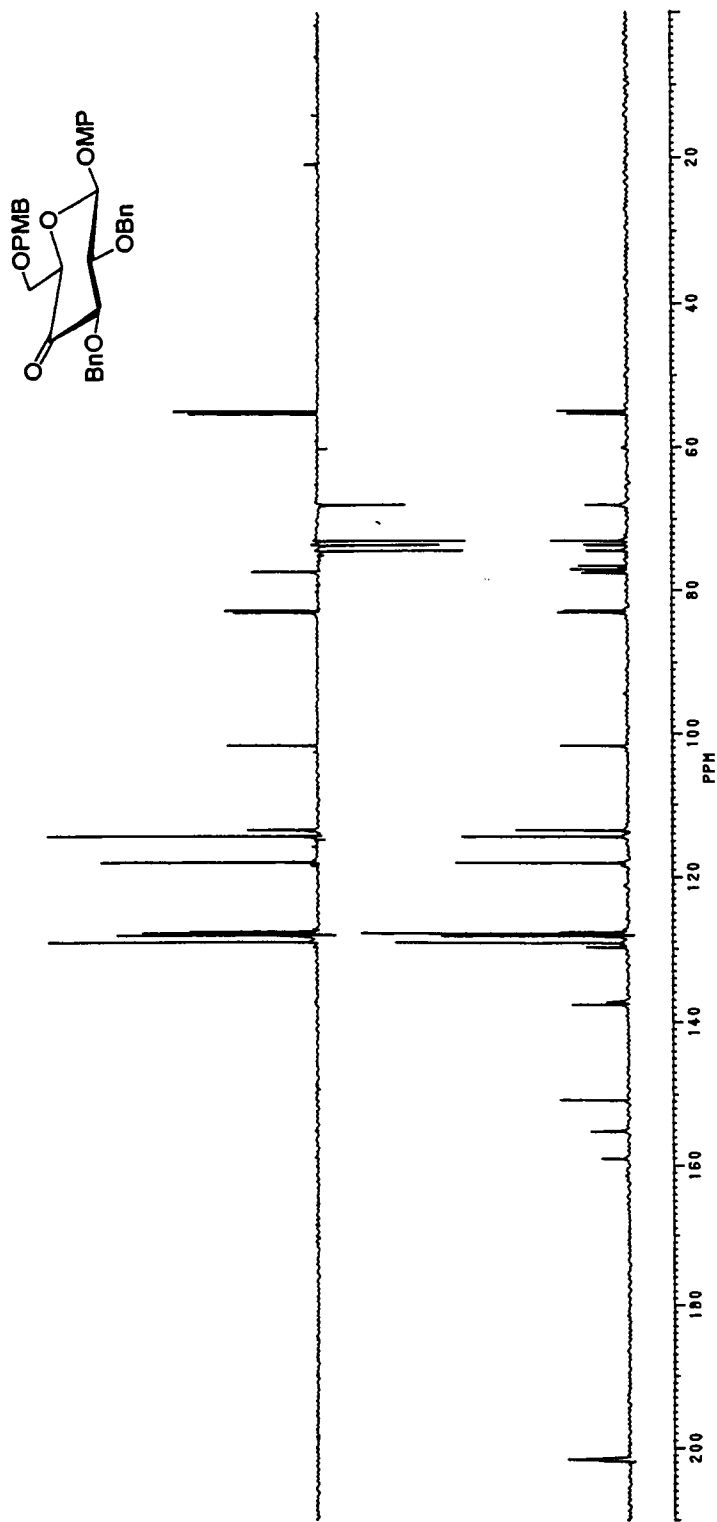


¹H NMR (250 MHz, CDCl₃) spectrum of *p*-methoxybenzyl-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl-β-*D*-xylo-hexopyranoside (100a).

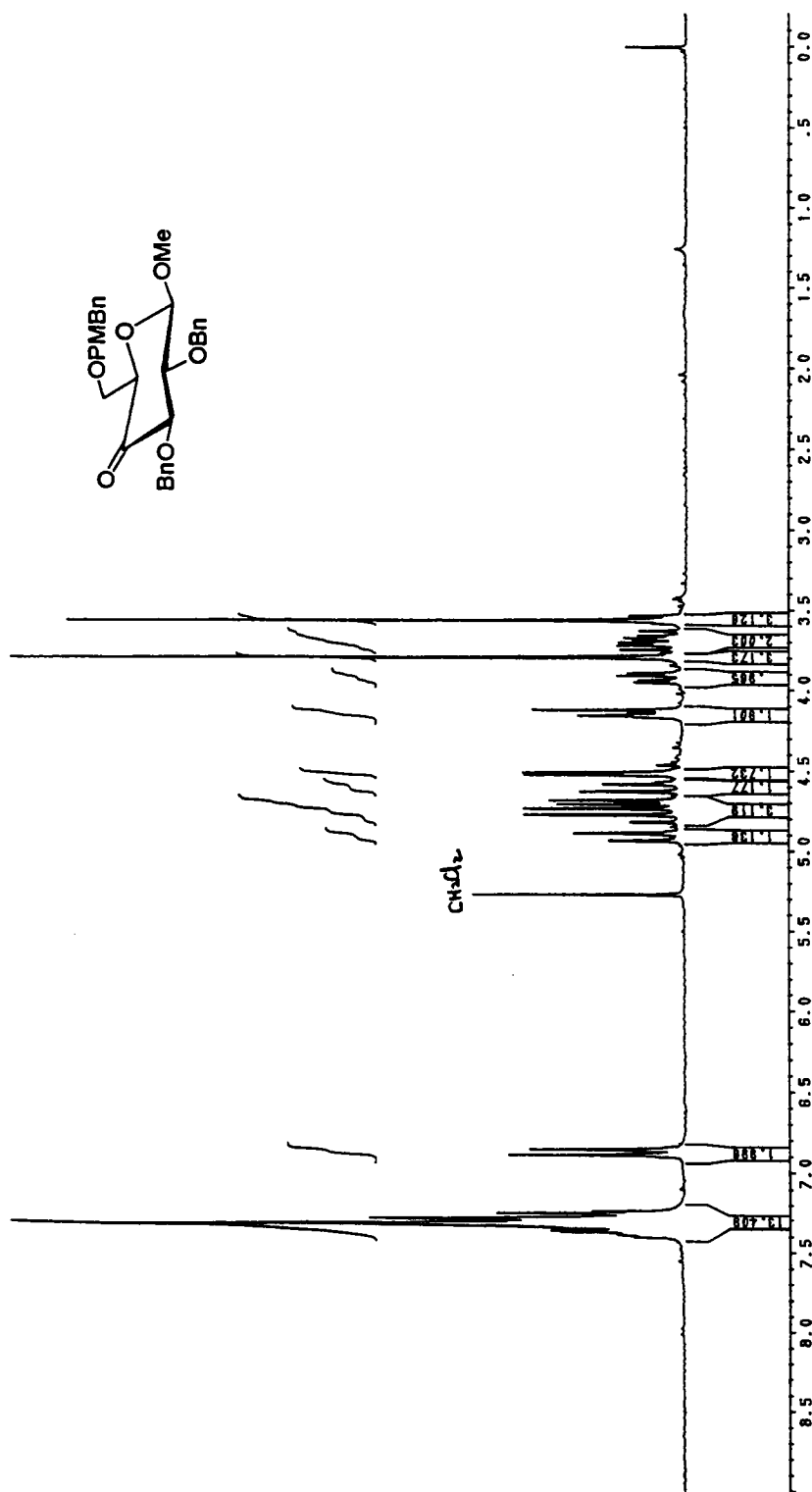


256

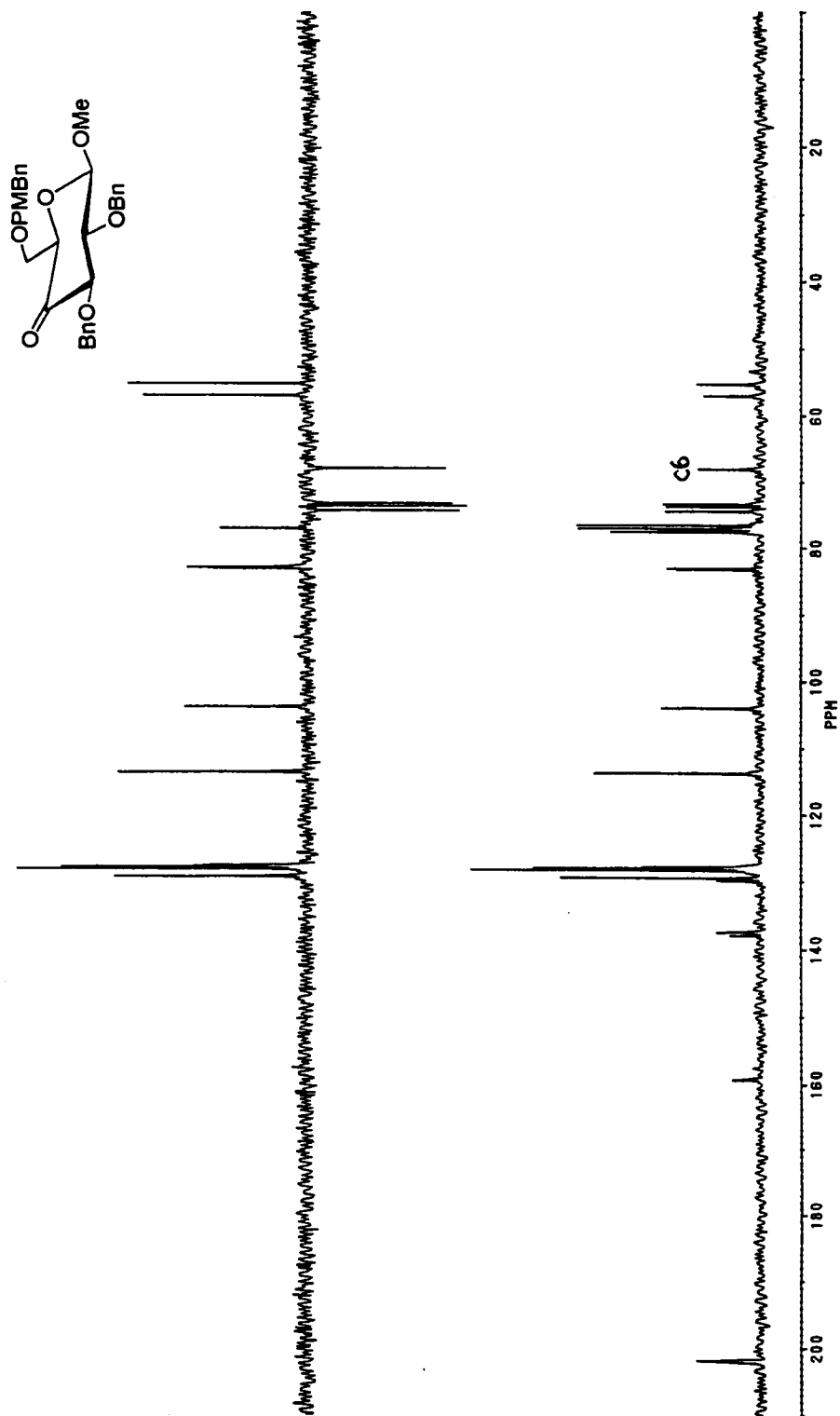
^1H double resonance spectrum of *p*-methoxyphenyl-2,3-di-*O*-benzyl-6-*O*-*p*-methoxybenzyl- β -D-xylo-hexopyranoside (**100a**).



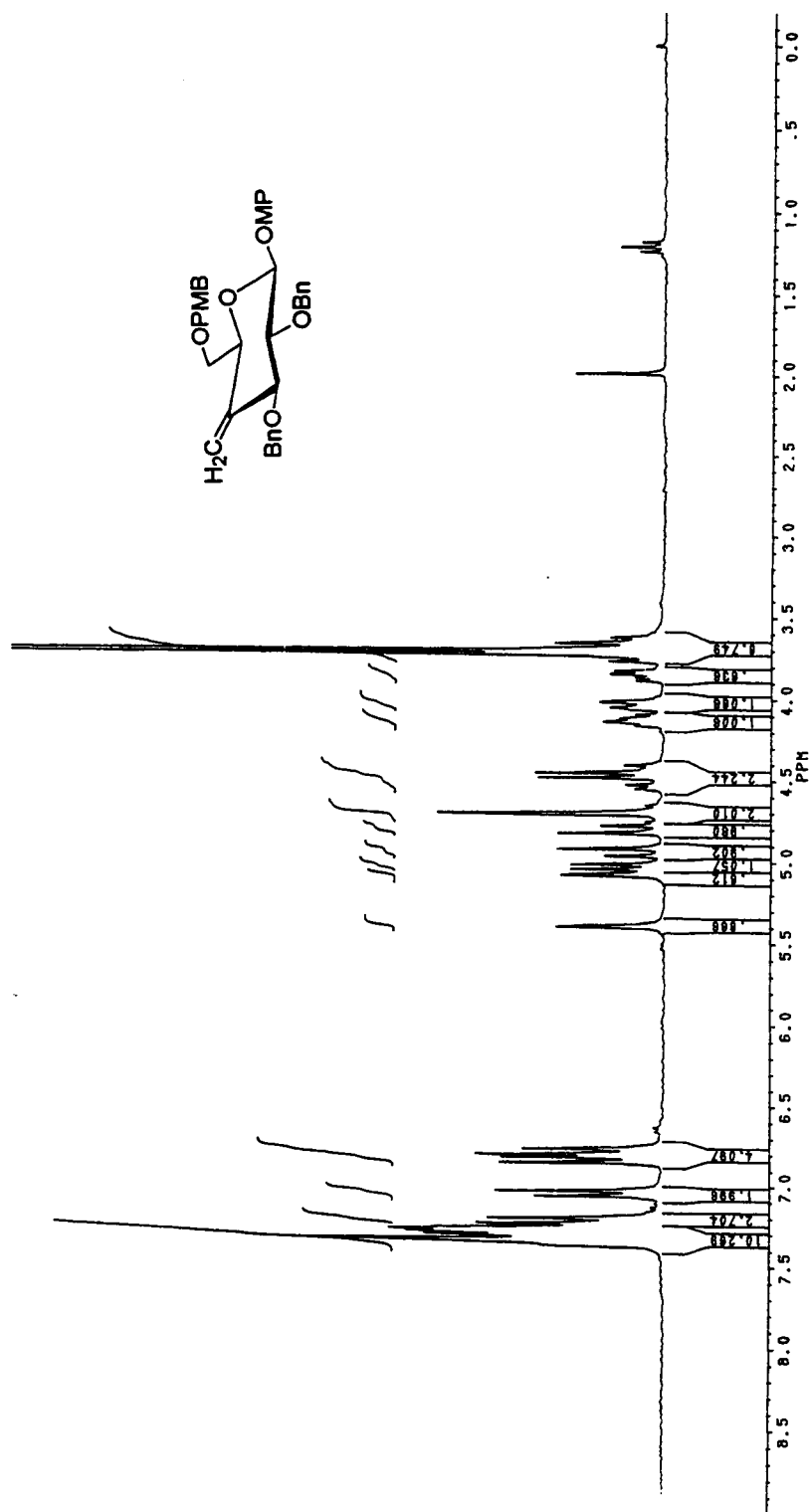
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *p*-methoxyphenyl-2,3-di-O-benzyl-6-O-*p*-methoxybenzyl-β-D-xylo-hexopyranosid-4-ulose (**100a**).



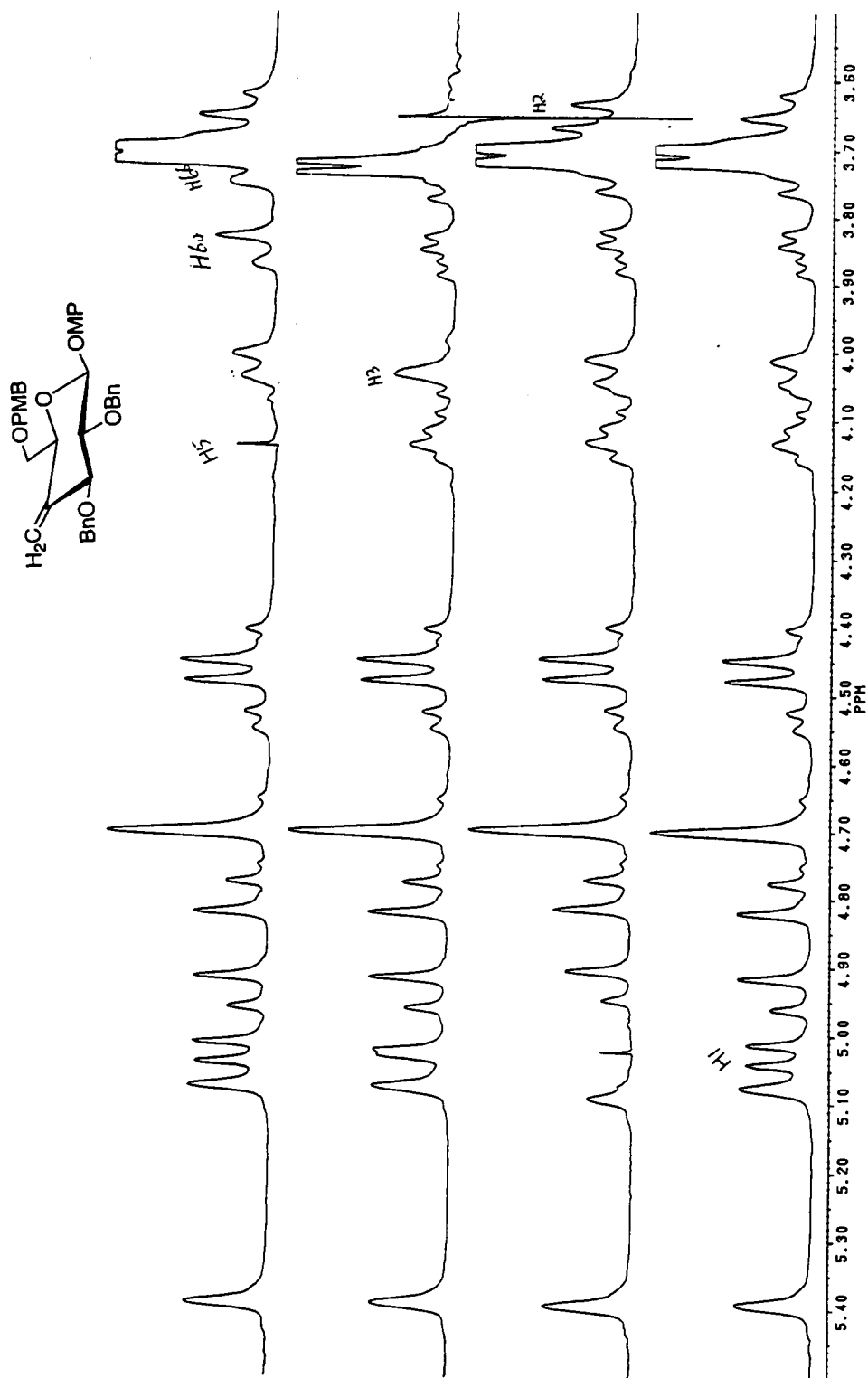
¹H NMR (250 MHz, CDCl₃) spectrum of methyl 2,3-di-O-benzyl-6-O-p-methoxybenzyl-β-D-xylo-hexopyranoside (100b).



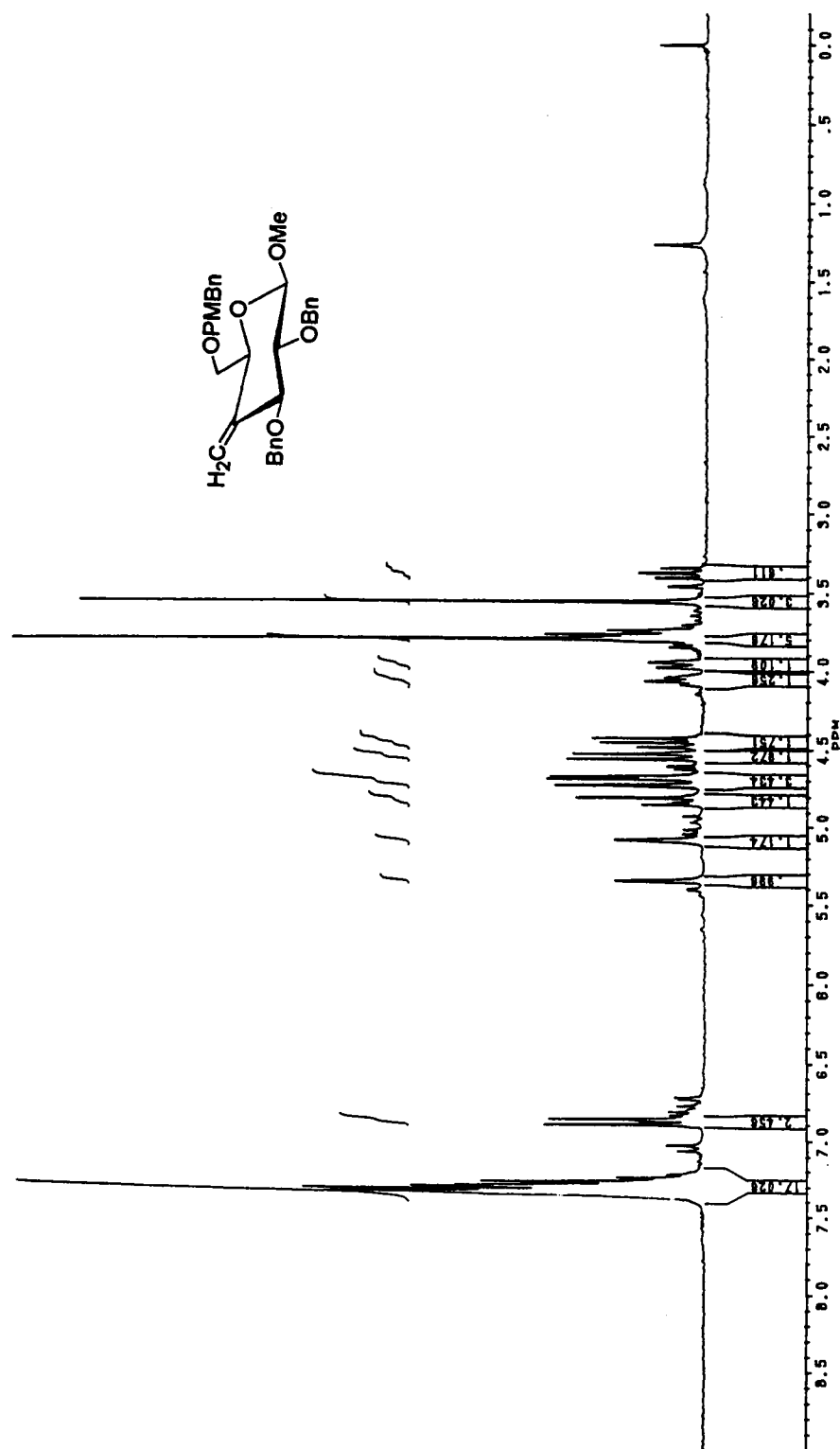
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of methyl 2,3-di-O-benzyl-6-O-*p*-methoxybenzyl-β-D-xylo-hexopyranosid-4-ulose (**100b**).



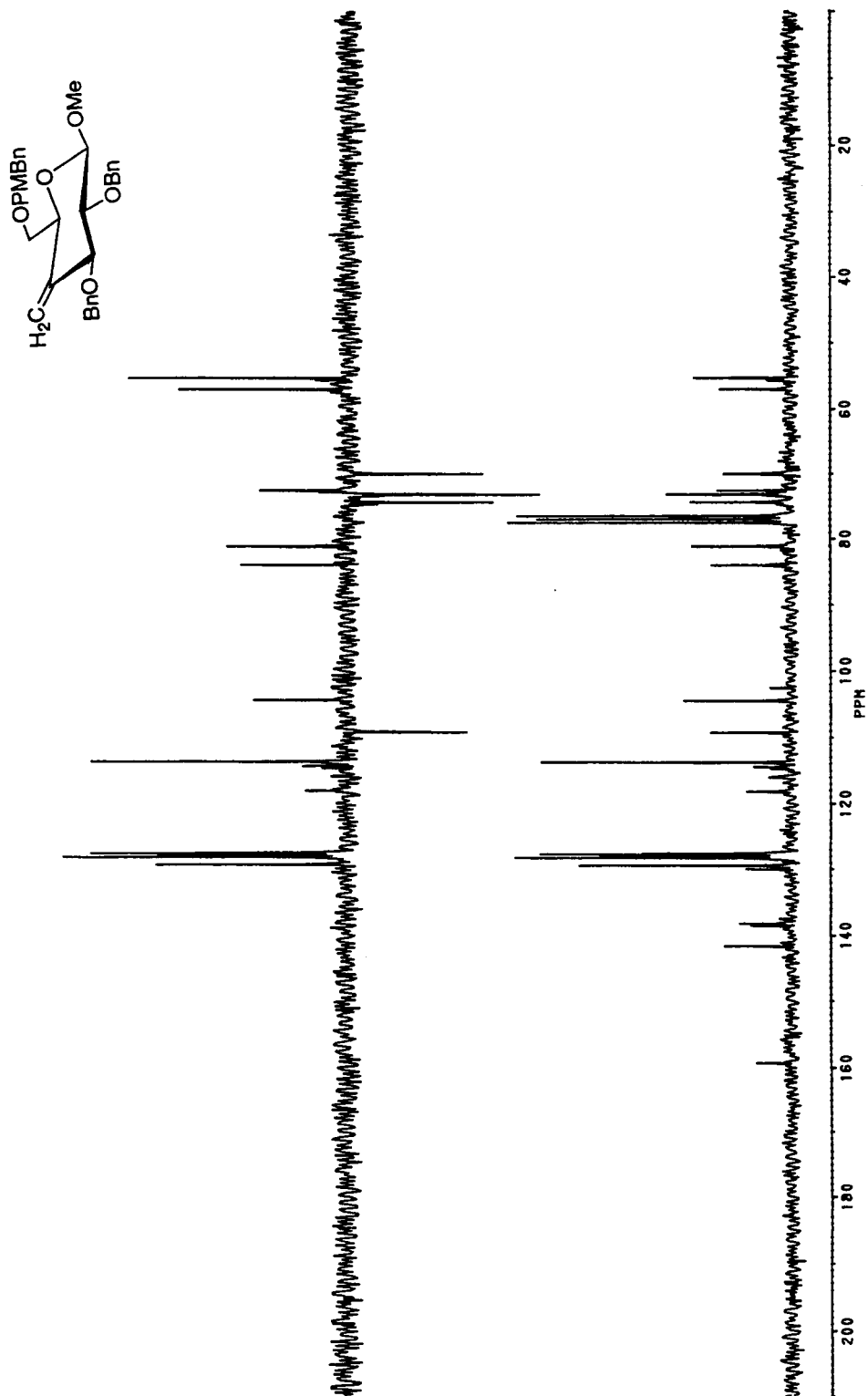
^1H NMR (250 MHz, CDCl_3) spectrum of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-6-*p*-methoxybenzyl-4-C-methylene- β -D-xylo-hexopyranoside (101a).



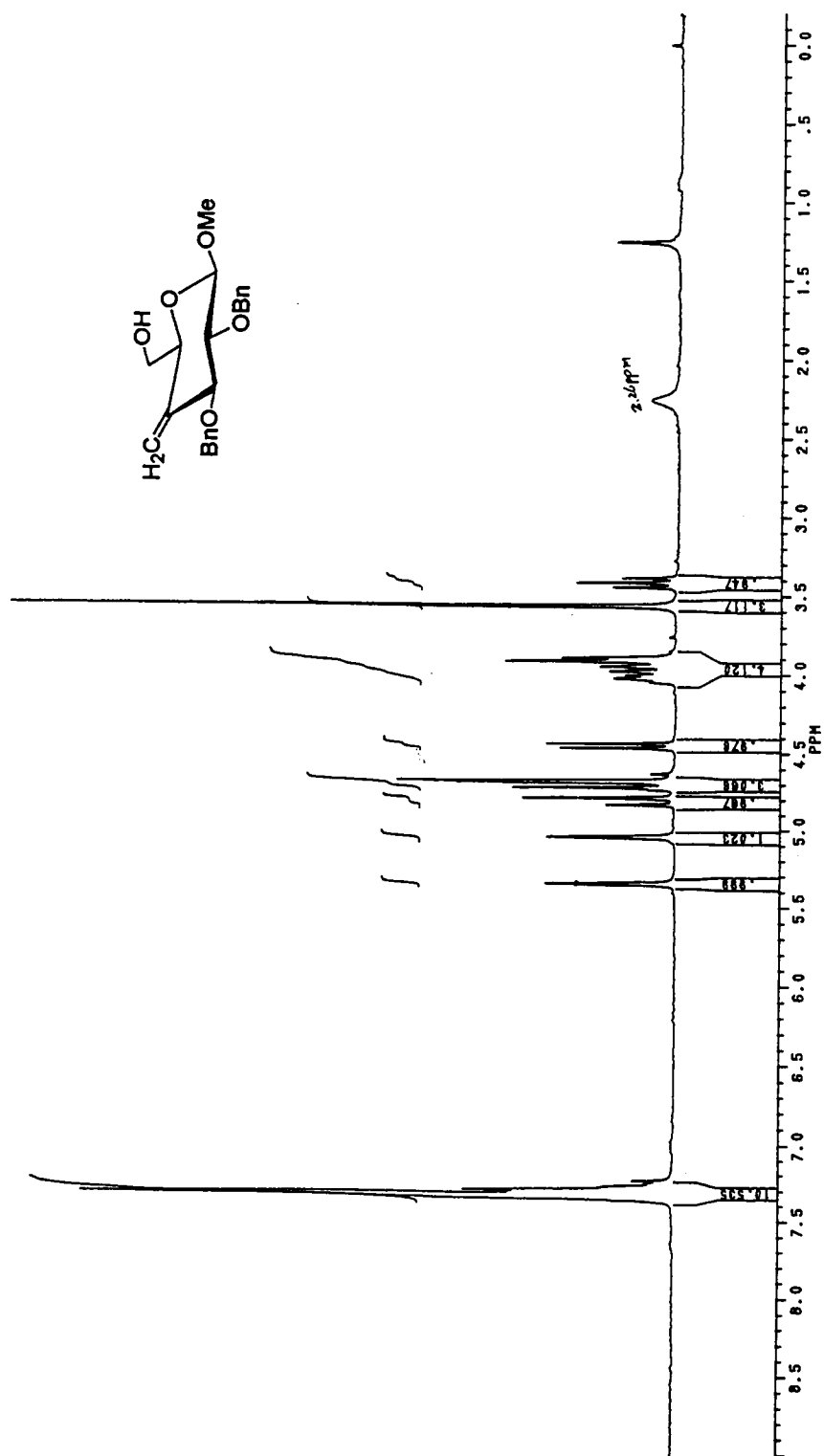
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-6-*p*-methoxybenzyl-4-C-methylene- β -D-xylo-hexopyranoside (**101a**).



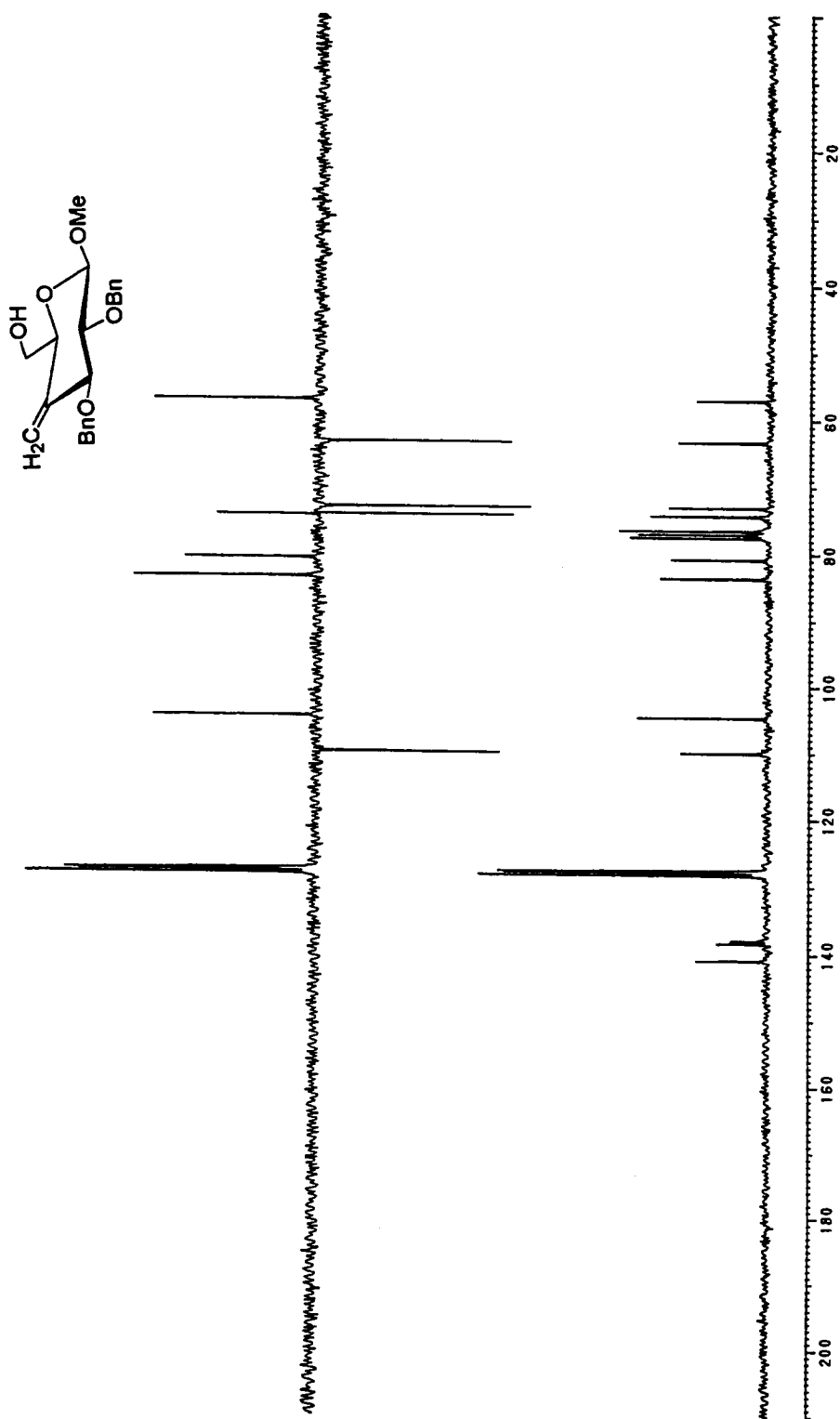
^1H NMR (250 MHz, CDCl_3) spectrum of methyl 2,3-di-O-benzyl-4-deoxy-6-p-methoxybenzyl-4-C-methylene- β -D-xylohexopyranoside(101b).



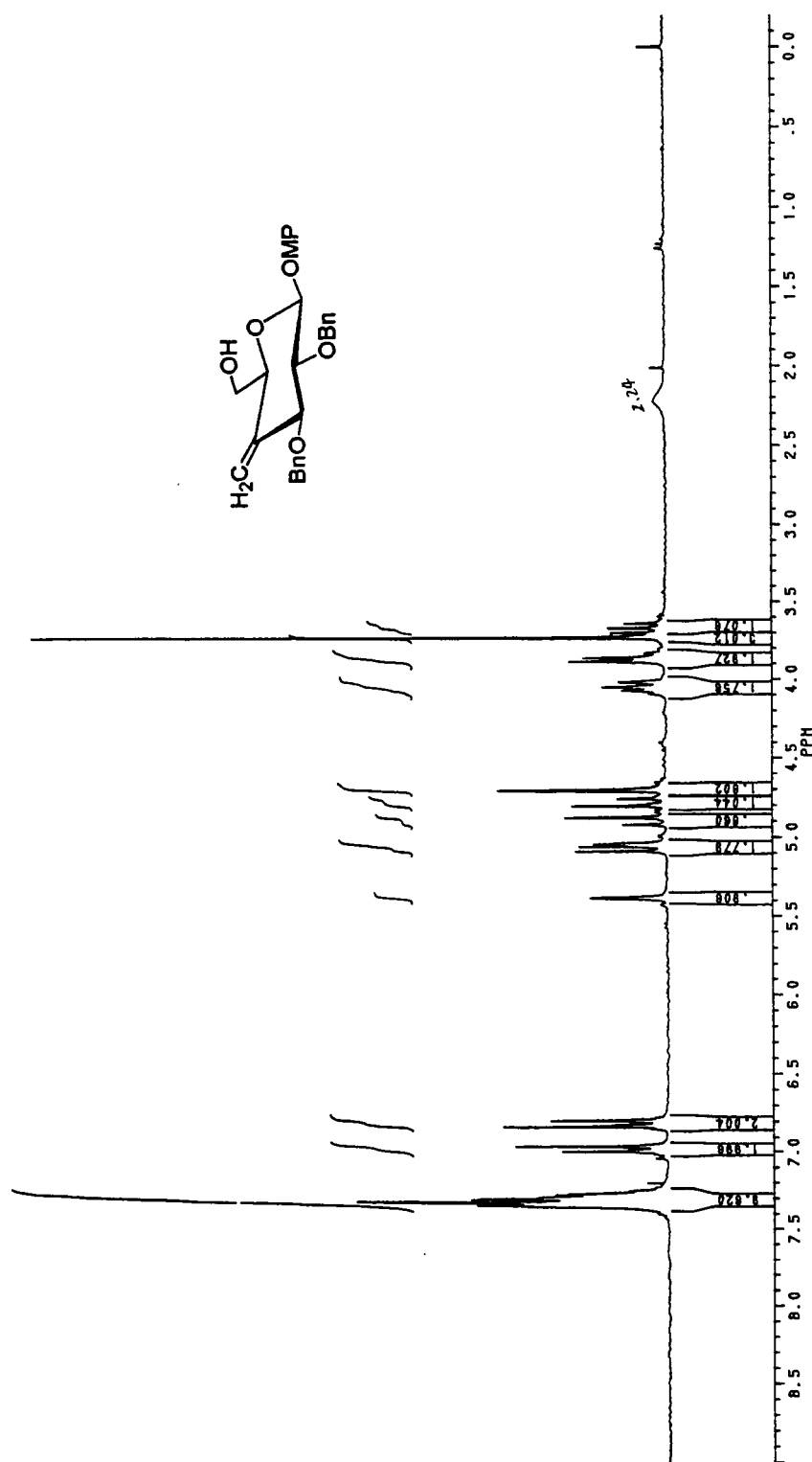
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of methyl 2,3-di-O-benzyl-4-deoxy-6-*p*-methoxybenzyl-4-C-methylene-β-D-xylo-hexopyranoside(**101b**).



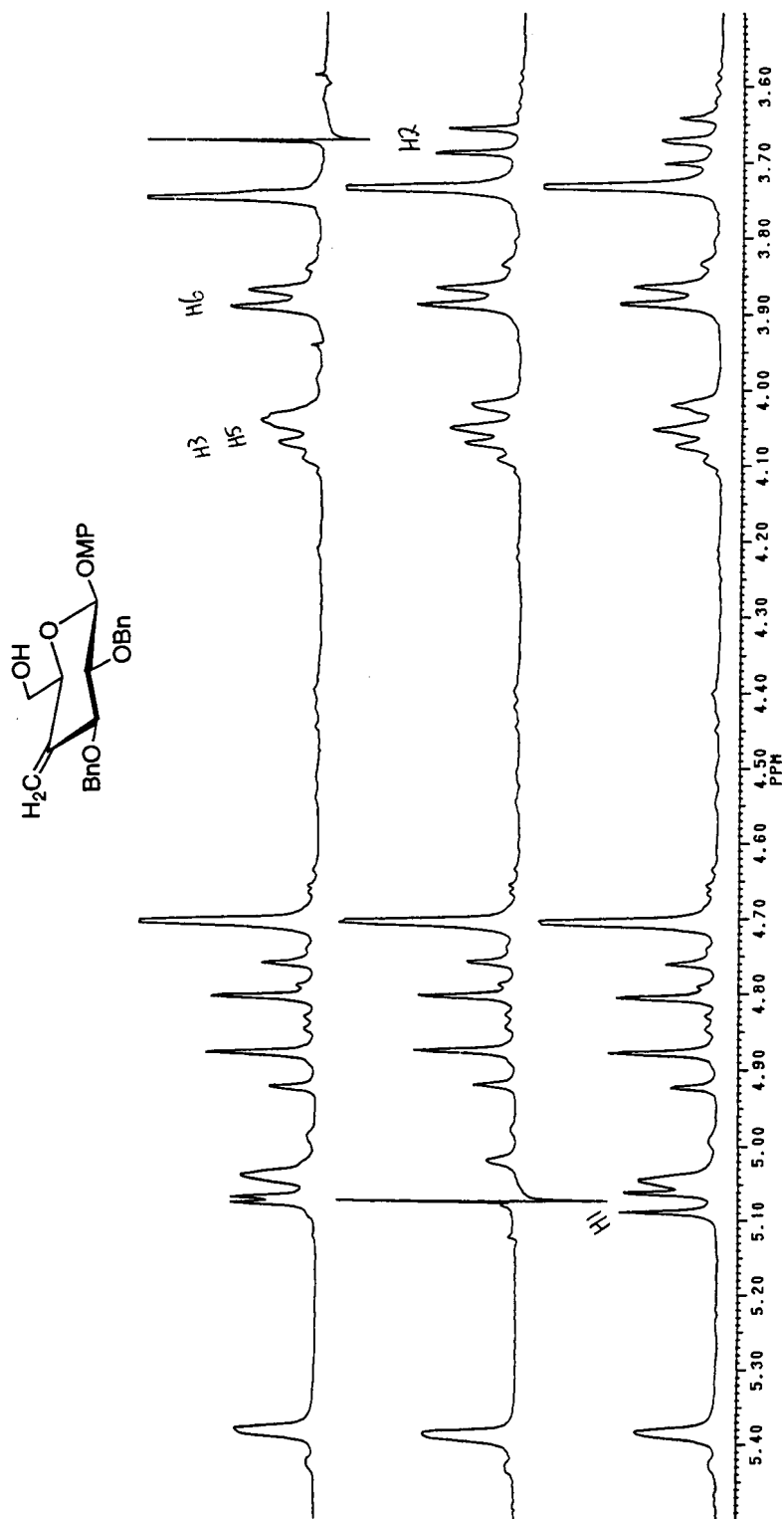
¹H NMR (250 MHz, CDCl₃) spectrum of methyl 2,3-di-O-benzyl-4-deoxy-4-C-methylene-β-D-xylo-hexopyranoside (102b).



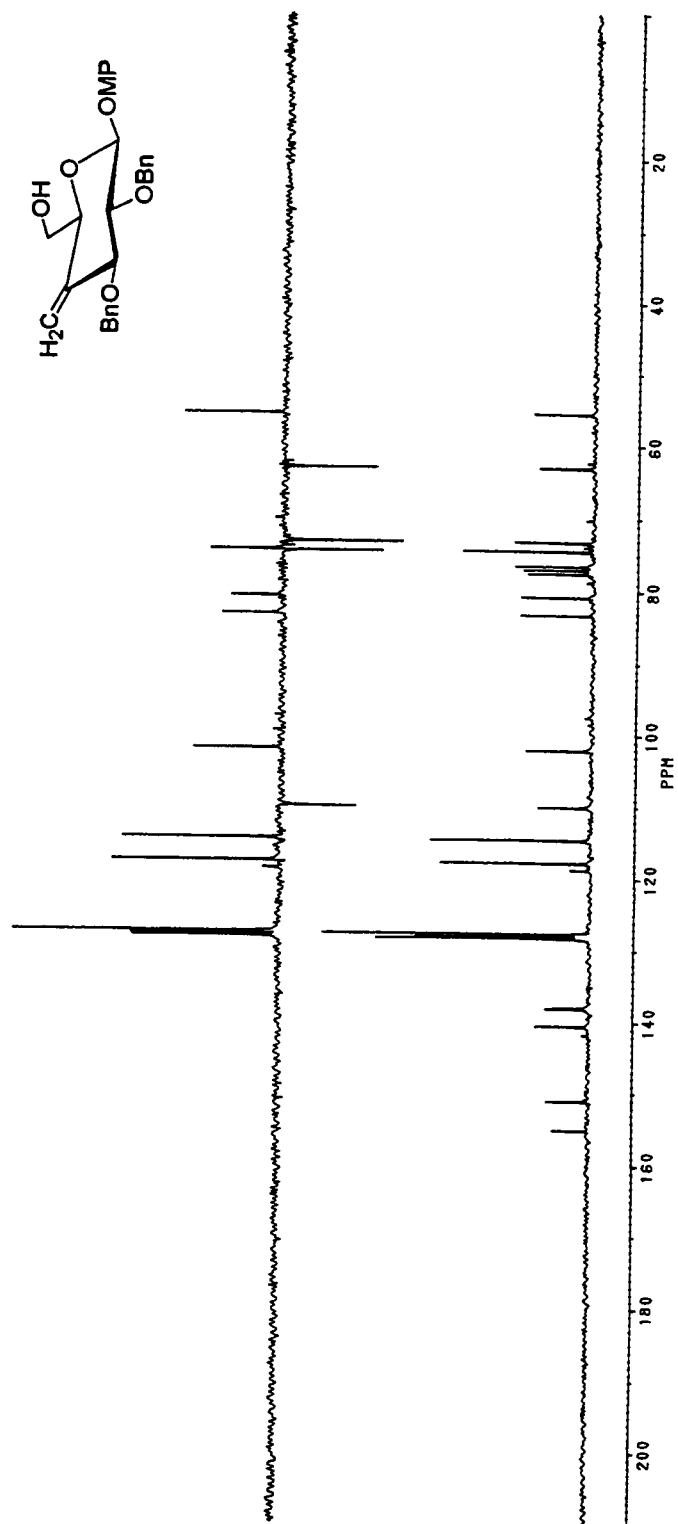
DEPT-135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of methyl 2,3-di-O-benzyl-4-deoxy-4-C-methylene- β -D-xyl-o-hexopyranoside (**102b**).



^1H NMR (250 MHz, CDCl_3) spectrum of 2,3-di-O-benzyl-4-deoxy-4-C-methylene- β -D-xylohexopyranoside (102a).

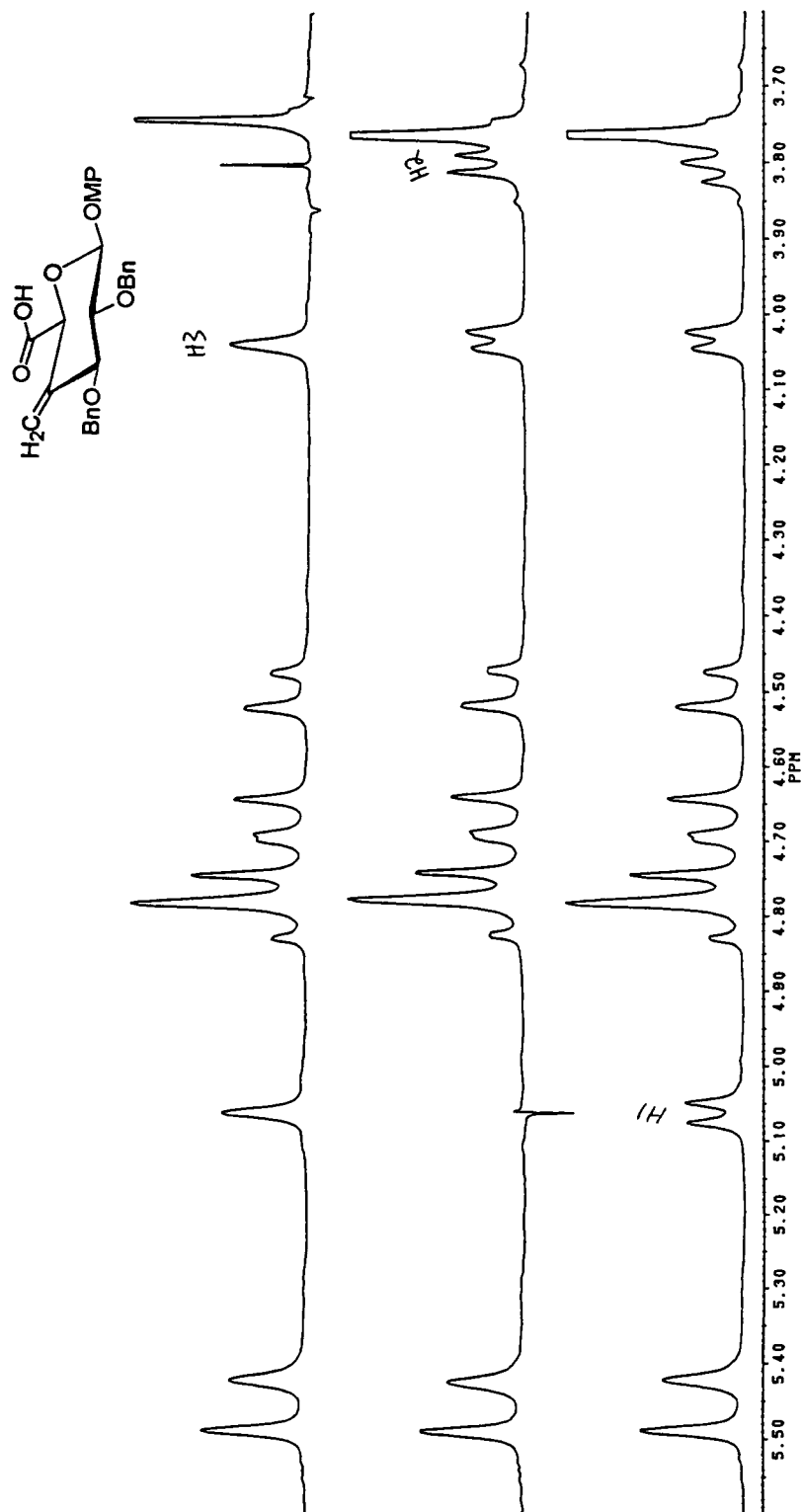


^1H double resonance spectrum of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-4-C-methylene- β -D-xylohexopyranoside(102a).

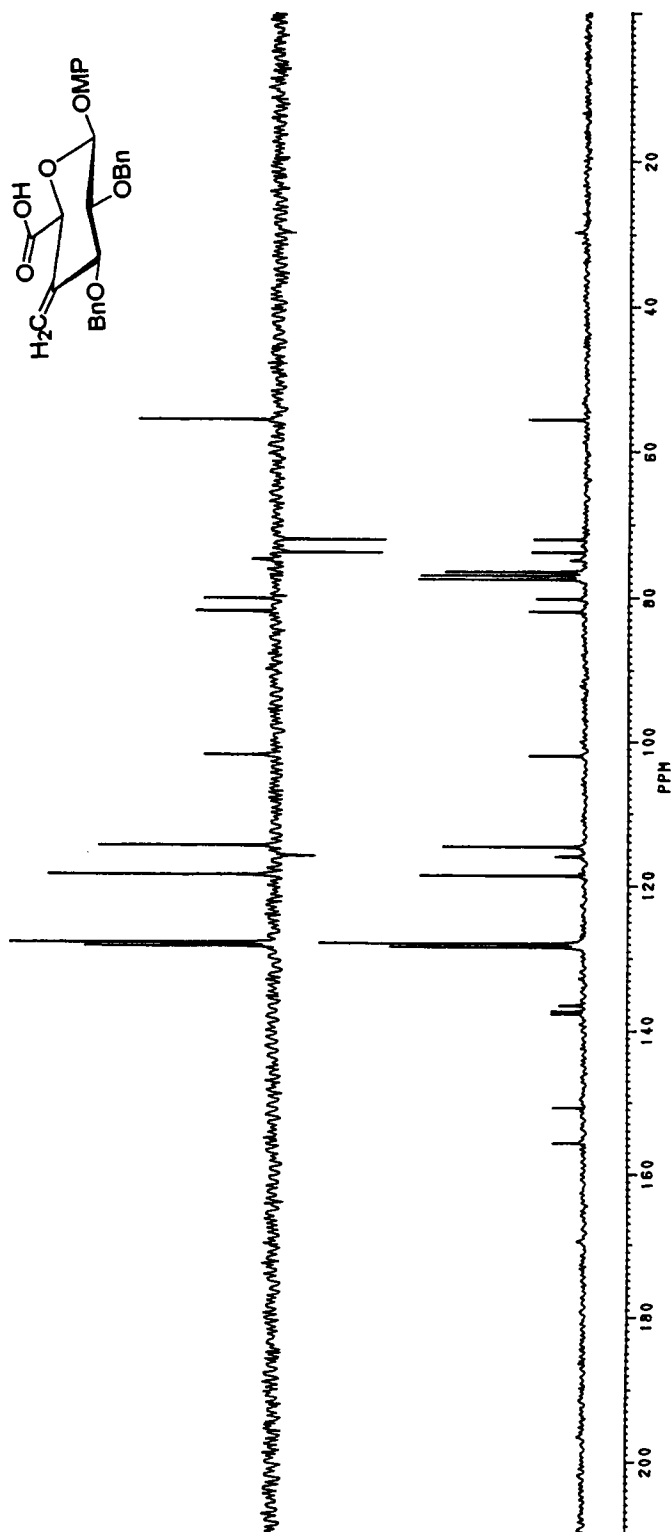


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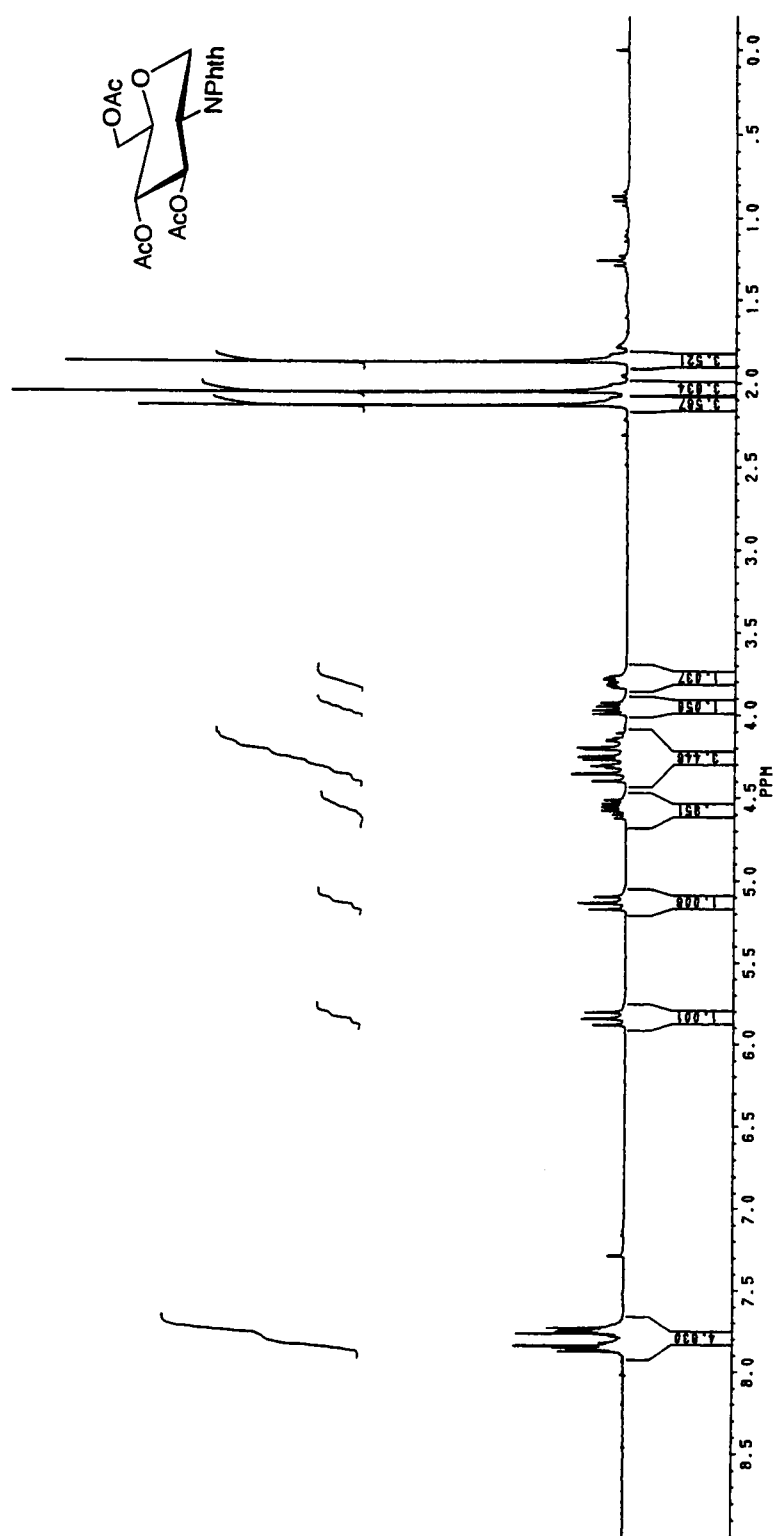
DEPT135 (top) and ^{13}C (bottom) NMR (62.5 MHz, CDCl_3) spectra of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-4-C-methylene- β -D-xylo-hexopyranoside (**102a**).



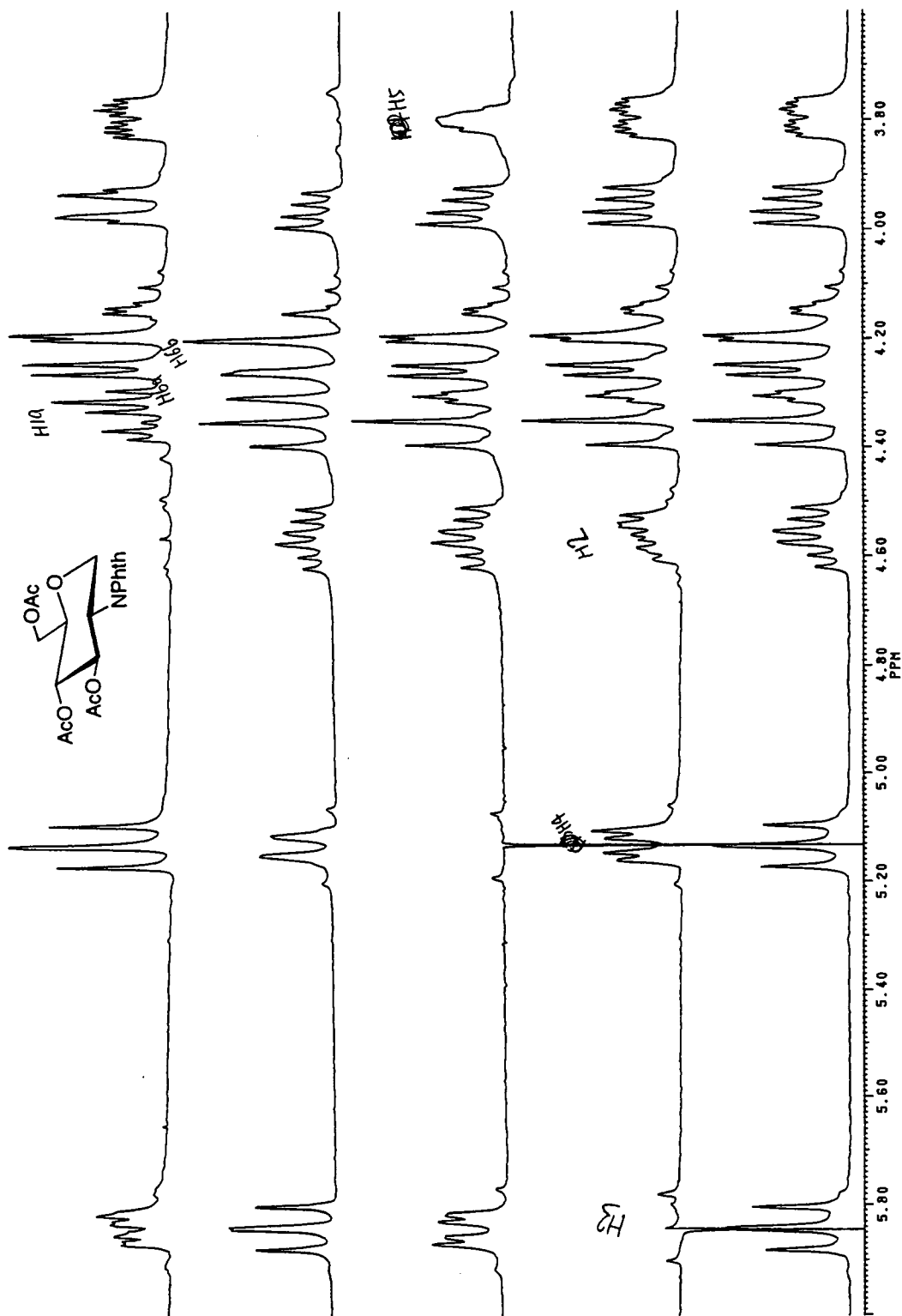
^1H double resonance spectrum of *p*-methoxyphenyl 2,3-di-*O*-benzyl-4-deoxy-4-*C*-methylene- β -D-xylo-hexopyranosiduronic acid (**106a**).



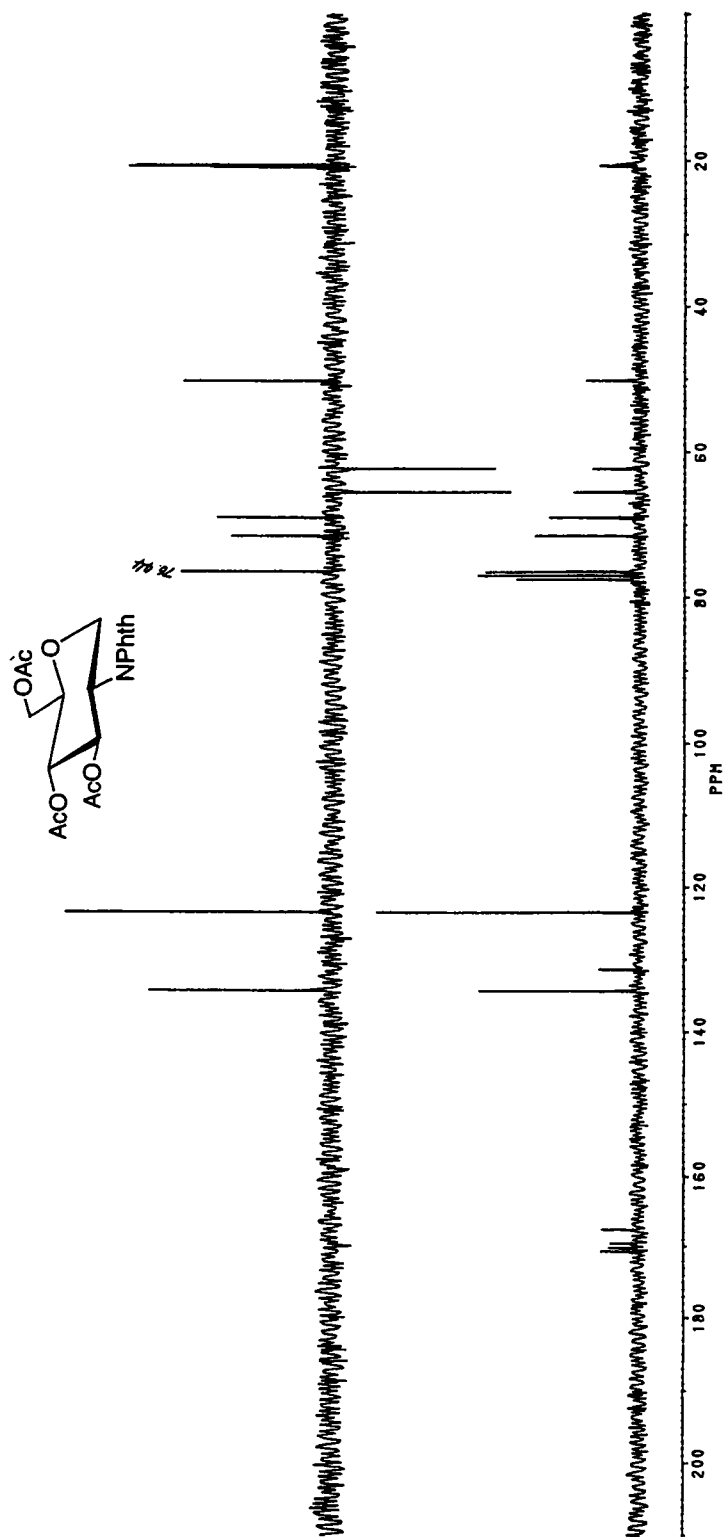
DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-4-C-methylene-β-D-xyllo-hexopyranosiduronic acid (**106a**).

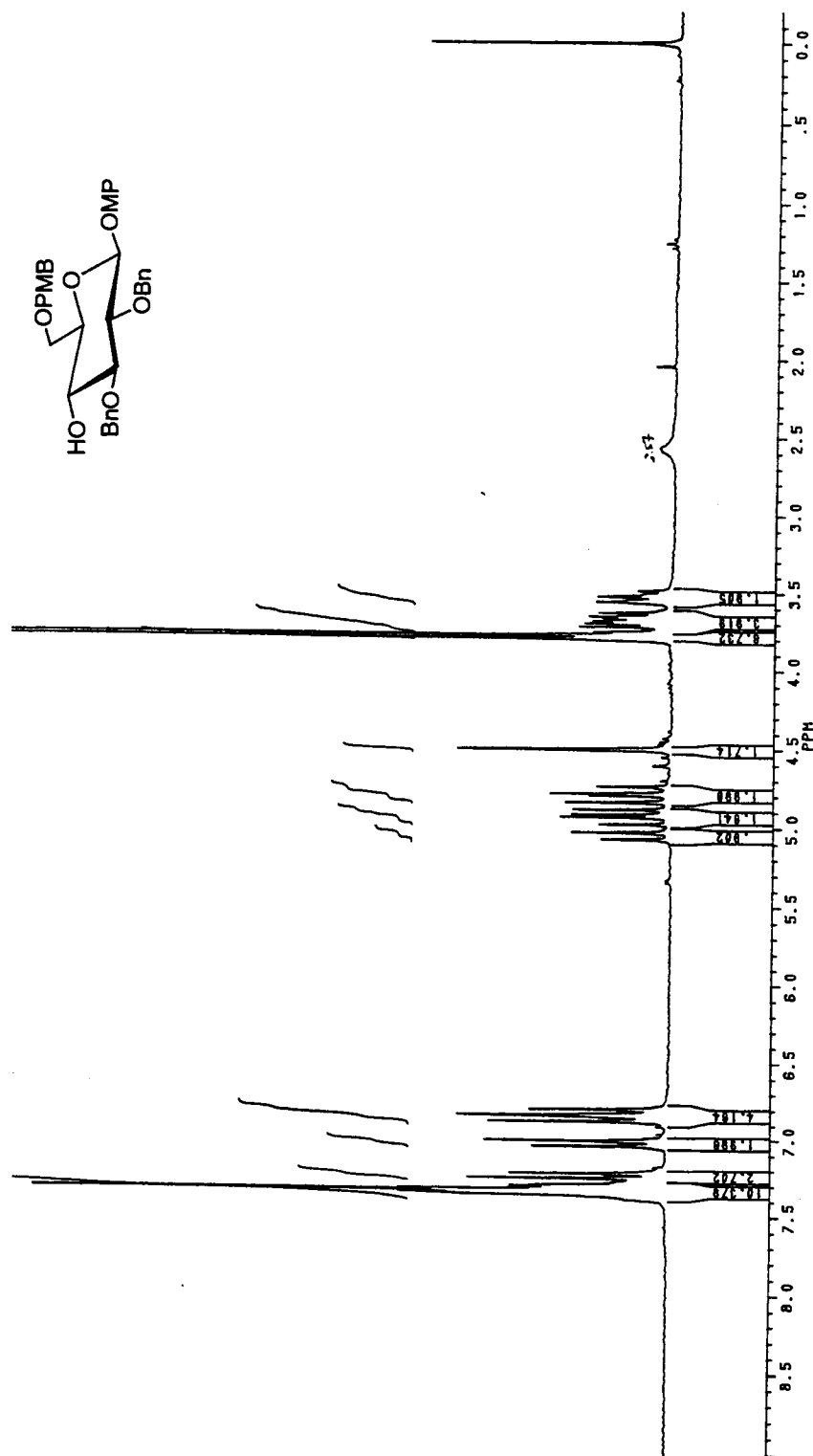


¹H NMR (250 MHz, CDCl₃) spectrum of 3,4,6-tri-O-acetyl-1,5-anhydro-2-phthalimido-D-glucitol (**107**).

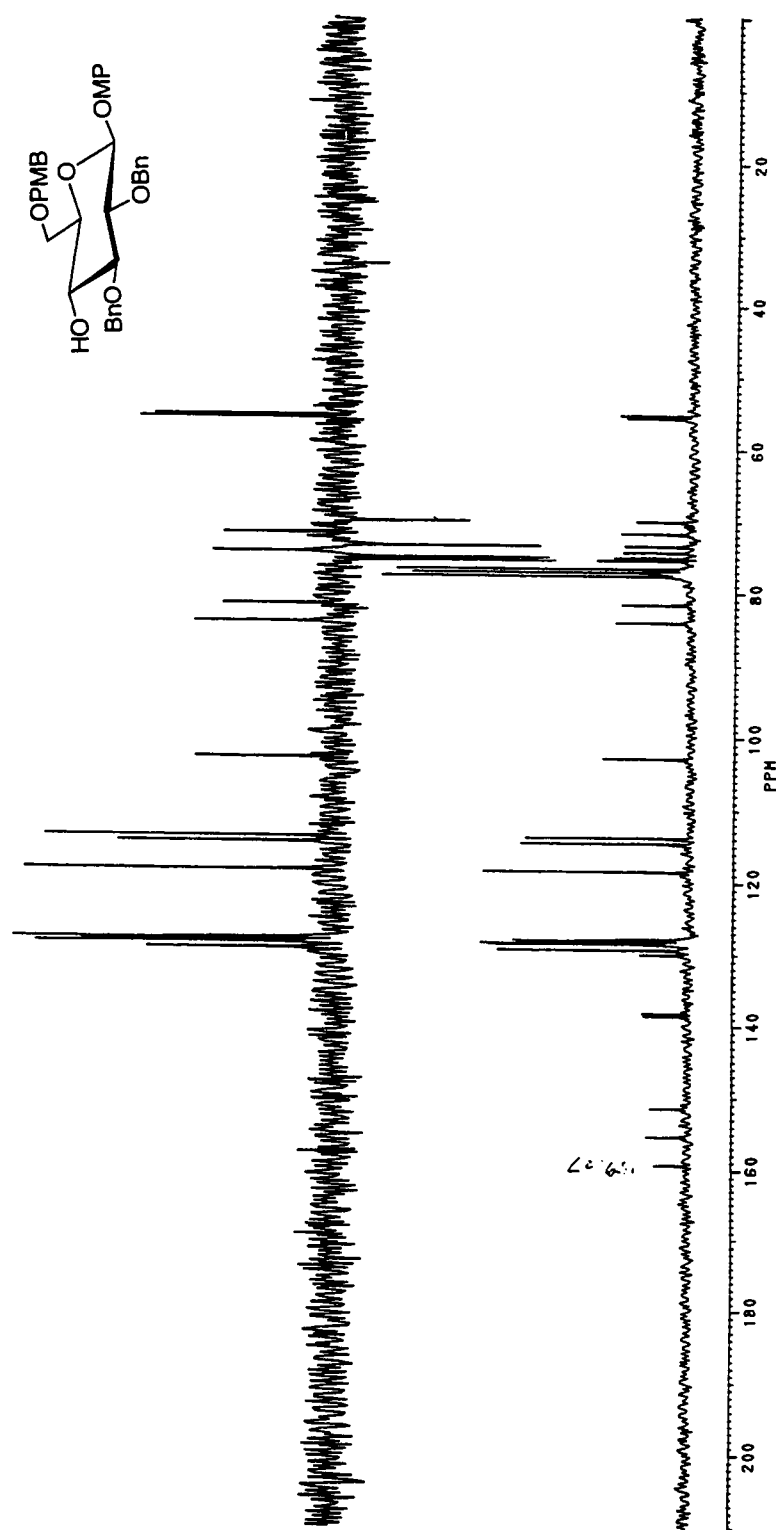


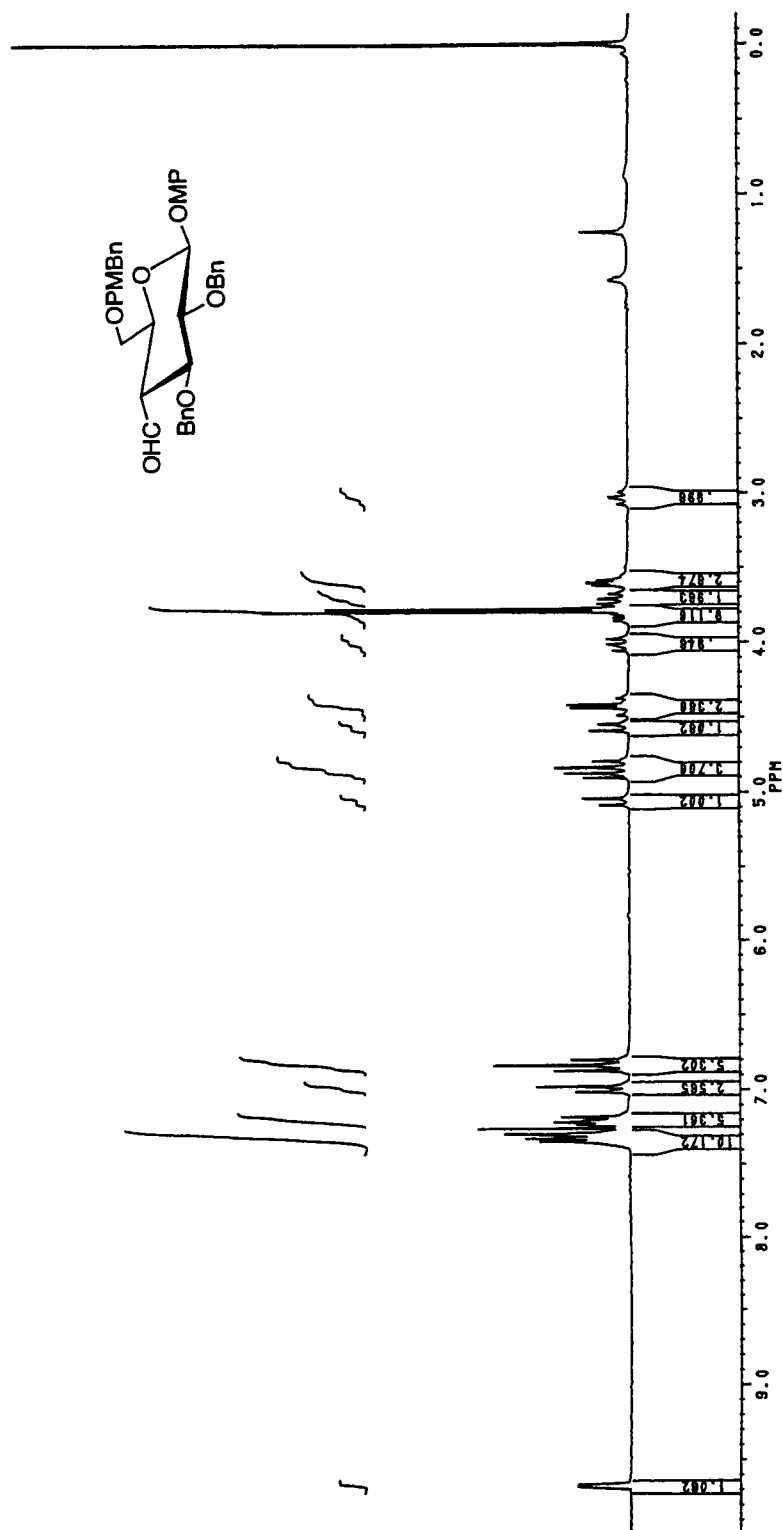
^{13}C NMR (62.5 MHz, CDCl_3) spectrum of 3,4,6-tri-O-acetyl-1,5-anhydro-2-phthalimido-D-glucitol (107).

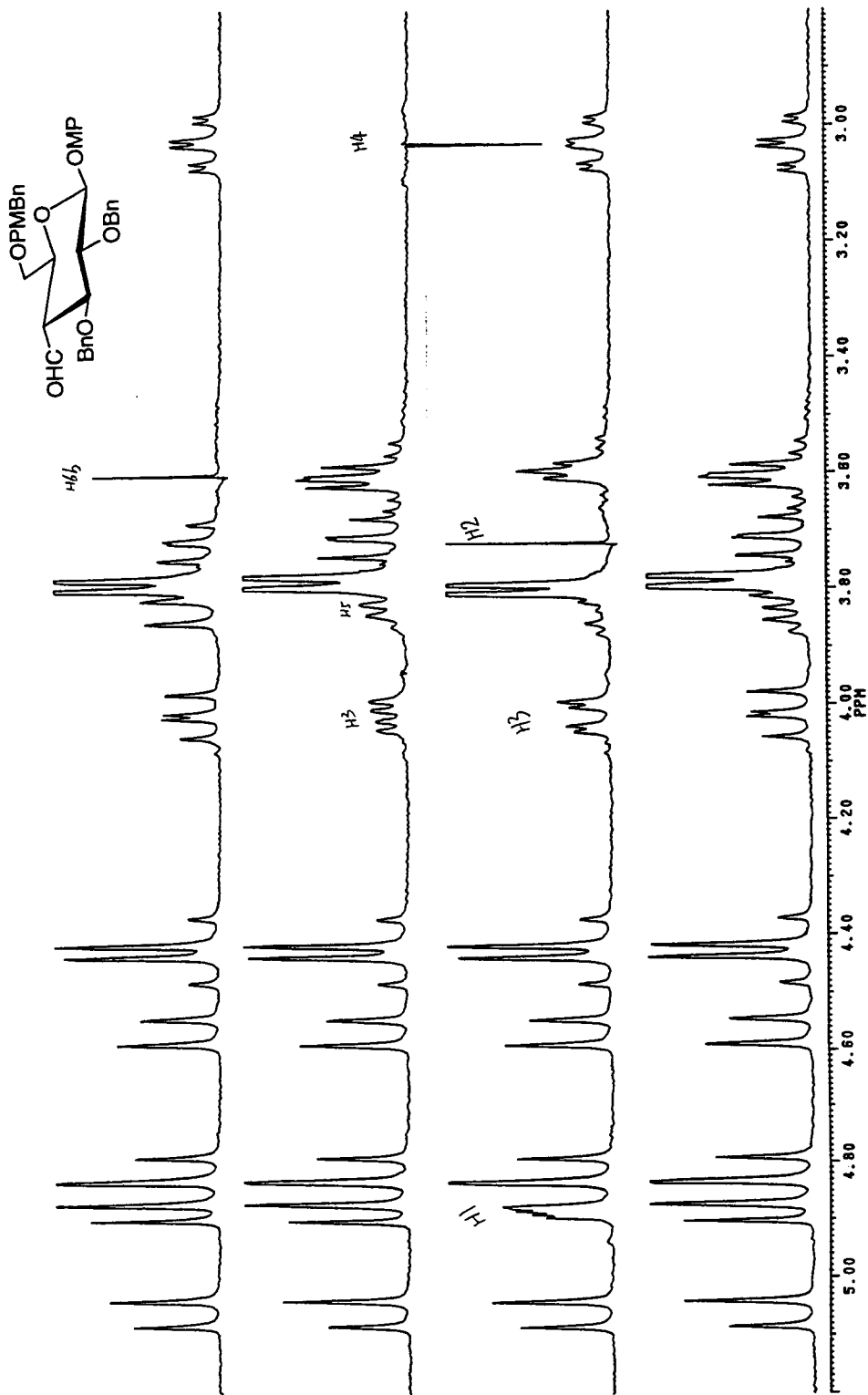




¹H NMR (250 MHz, CDCl₃) spectrum of *p*-methoxybenzyl 2,3-di-O-benzyl-6-O-*p*-methoxybenzyl- β -D-glucopyranoside (112).

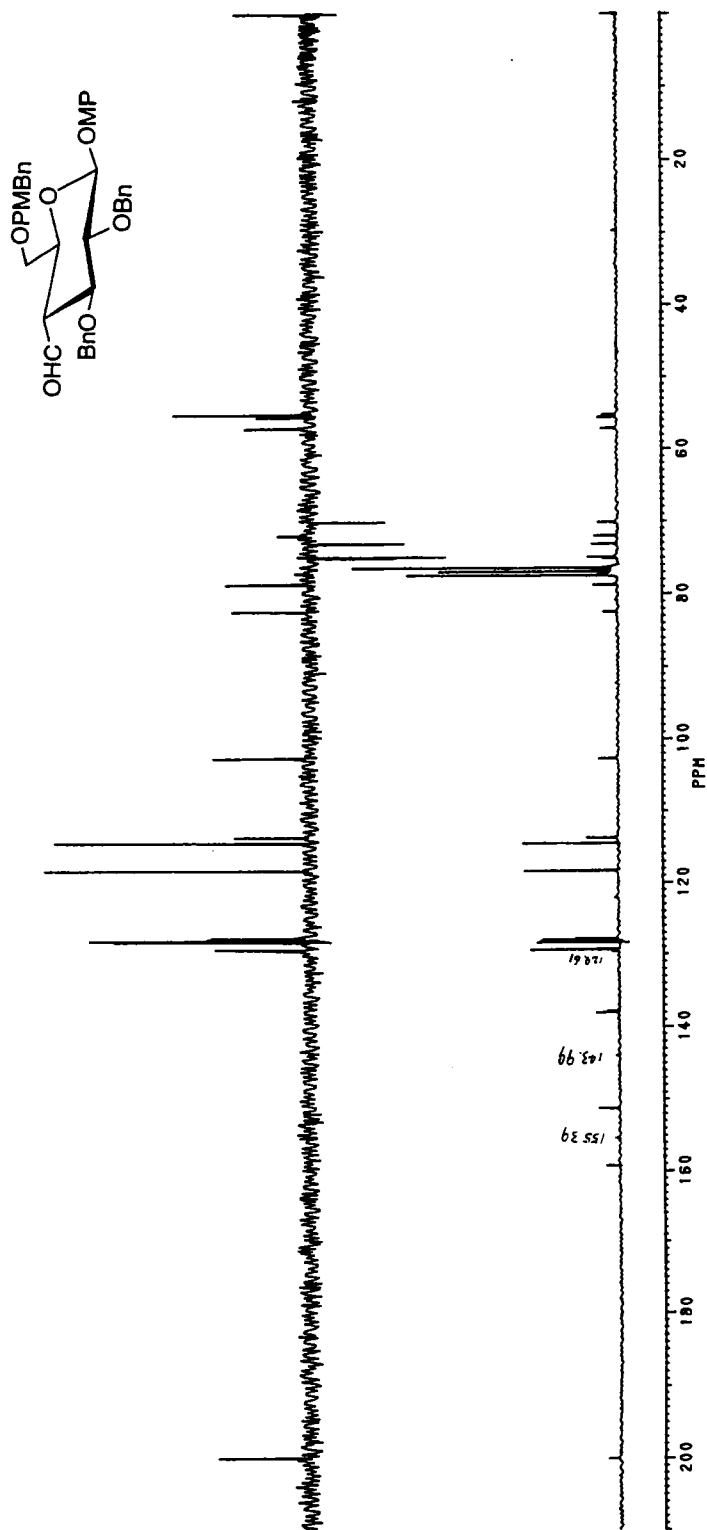






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^1H double resonance spectrum of *p*-methoxyphenyl 2,3-di-O-benzyl-4-deoxy-4-C-formyl-6-O-*p*-methoxybenzyl- β -D-glucopyranoside (115).



DEPT135 (top) and ¹³C (bottom) NMR (62.5 MHz, CDCl₃) spectra of *p*-Methoxyphenyl 2,3-di-O-benzyl-4-C-formyl-6-O-*p*-methoxybenzyl-β-D-glucopyranoside (115).

VITA

Bin Jiang was born in Beijing, China in 1968. After graduating from Zhixin High School in Guangzhou in 1986, he attended Zhongshan University, where he graduated with a Bachelor's Degree in Chemistry. From 1990 to 1993 he entered the graduate school of Shanghai Institute of Organic Chemistry, Chinese Academic of Sciences, conducted research with Professors Xi-Kui Jiang and Yu-Huang Zhang, and earned the Master's degree in Organic Chemistry. After working as an instrument sales engineer in LECO Corporation's Shanghai representative office for a brief period, he began doctorate study in chemistry at the University of Tennessee in 1994. After two and a half years in Professor T. Ffrancon Williams' group conducting ESR research and two years in Professor David C. Baker's group conducting solid-phase synthesis and carbohydrate synthesis, he accepted a position in Chemical and Physical Sciences Department at the DuPont Pharmaceuticals Company in Wilmington, Delaware. His doctorate research led to one US patent application and at least three manuscripts for publication.