ABSTRACT

The Preparation of Novel Modified Cyclodextrins and Their Application in Enantioseparations by Gas Chromatography.

Sheree N. Allen, Ph.D.

Mentor: Charles M. Garner, Ph.D.

Since biological processes depend on the stereospecific creation and conversion of chiral molecules, each enantiomer of a molecule can have a different biological effect. While numerous synthetic drugs are sold in racemic form for reasons of cost and convenience, in many cases one of the two enantiomers of a drug will cause undesirable and perhaps even devastating effects. Therefore, effective methods for the separation and quantification of enantiomers are of great importance.

Enantiomers can be separated only with the use of systems containing an optically active chiral selector. Modified cyclodextrins (CDs) are widely used as chiral selectors by virtue of their ability to form inclusion complexes. Because of the remarkably high efficiency, and sensitivity of chiral gas chromatography (GC), chiral separations by this method represent a preferred method for enantiomer analysis. The basic property of CDs that allows them to be successful for enantiomer separations is their ability to form selective inclusion complexes with a wide variety of organic molecules. The preparation and application of modified cyclodextrins for the GC separation of enantiomers is a focal point of this research.

In effort to identify useful new derivatives, modifications involving unique and separate reaction of the secondary hydroxyl groups and/or annulations bridging the secondary oxygens have been examined. Several new cyclodextrin derivatives, *e.g.* *per*(6-OTBS-2,3-*O*-diformyl)-β-CD, *per*(6-OTBS-2,3-*O*-cyclodimethylsilyl)-β-CD, *per*(6-deoxy-2,3-*O*-cyclodimethylsilyl)-β-CD and a mixed formyl/acetyl phase have been synthesized and evaluated as components of stationary phases for capillary GC. The efficiency of each new phase to separate enantiomers was evaluated against a 30 analyte panel to evaluate the influence of the different substituents on the selectivity. These enantioseparations were also compared to those observed on four commercially available chiral phases using the same 30 analyte panel. Overall, this work resulted in nine new CD derivatives and the discovery of a new chiral selector that is comparable in efficiency to what is currently commercially available.

The Preparation of Novel Modified Cyclodextrins and Their Application in Enantioseparations by Gas Chromatography.

by

Sheree N. Allen, B.S.

A Dissertation

Approved by the Department of Chemistry & Biochemistry

_____
Patrick J. Farmer, Ph.D., Chairperson

Submitted to the Graduate Faculty of
Baylor University in Partial Fulfillment of the
Requirements for the Degree
of
Doctor of Philosophy

Approved by the Dissertation Committee

_____
Charles M. Garner, Ph.D., Chairperson

_____
Kevin G. Pinney, Ph.D.

_____
Robert R. Kane, Ph.D.

_____
Darrin J. Bellert, Ph.D.

_____
Steve I. Dworkin, Ph.D.

Accepted by the Graduate School
May 2010

_____
J. Larry Lyon, Ph.D., Dean

Page bearing signatures is kept on file in the Graduate School.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF SCHEMES

LIST OF ABBREVIATIONS

Å	angstroms
Ac	acetyl
AIBN	azobisisobutyronitrile
b.p.	boiling point
t-Bu	tertiary butyl
cat.	catalytic
CD	cyclodextrin
CSP	chiral stationary phase
DMAP	dimethylaminopyridine
DMF	dimethylformamide
DMP	Dess-Martin periodinane
DMSO	dimethylsulfoxide
ee	enantiomeric excess
equiv	equivalents
ESI	electrospray ionization
Et	ethyl
Et_2O	diethyl ether
EtOAc	ethyl acetate
FID	flame ionization detector
g	grams
GC	gas chromatography

GC/MS	gas chromatography/mass spectrometry
h	hours
HPLC	high performance liquid chromatography
IBX	iodoxybenzoic acid
I.D.	internal diameter
L	liters
LC	liquid chromatography
m	meter
M	molarity
MALDI	matrix-assisted laser desorption/ionization
Me	methyl
min	minute
mL	milliliter
mm	millimeter
μm	micrometer
m.p.	melting point
MS	mass spectrometry
NBS	N-bromosuccinimide
NMR	nuclear magnetic resonance
Nu	nucleophile
OAc	acetate
OMe	methoxy
Ph	phenyl

ppm	parts per million
psi	pounds per square inch
sec	second
TBAF	tetrabutylammonium fluoride
TBS	*tert*-butyldimethylsilyl
THF	tetrahydrofuran
TLC	thin layer chromatography
TMS	trimethylsilyl
Tos	tosyl
VT	variable temperature

ACKNOWLEDGMENTS

This work would not have been possible without the support of many people. First, I would like to express extreme gratitude to my advisor, Dr. Charles Garner, for his invaluable guidance, his enthusiasm, and his useful suggestions throughout this research. His instructive comments, constant evaluation, and his encouragement to explore enabled me to not only complete this work successfully, but to grow from an inexperienced graduate student into a more creative and independent researcher. I am also deeply thankful to the members of my committee, Drs. Bob Kane, Kevin Pinney, Darrin Bellert, and Steve Dworkin for their time and indispensable ideas throughout this study. Each individual provided insight that has guided and challenged my thinking not only in the improvement of this work, but in my professional development as well.

I would also like to express sincere appreciation to all the members of the Baylor Chemistry Department whom have contributed to this project along the way. First and foremost, my group members and undergraduate researchers: Jason Bell, Nathan Duncan, Nelson Van Der Velde, Tiffany Turner Hayden, Adam Gann and Eric Bauch for their ideas, constant motivation, and support. Special thanks to Baylor University and the faculty and staff of the Chemistry & Biochemistry Department for making my journey as smooth and successful as possible.

I am especially indebted to my boyfriend, John W. Poston II, for his love, support, and understanding through the past four years. I also wish to thank my family: my parents, Mr. & Mrs. David Allen and Deborah Allen, and my siblings, Cliff, Sheena,

Shawna, Joshua, Ashley, and Hannah for their love, encouragement, and continuous support during my studies.

Finally, I would like to thank those people whose direction and insights have in one way or another encouraged my love of science. I am grateful for my undergraduate chemistry professors Dr. David Wazmund and Dr. John McClusky for constantly challenging me to think and encouraging me to come to graduate school. I am thankful for my junior high and high school science teachers, Mr. Otto Kollaus and Mrs. Marilyn Marshall for making science fun and for their encouragement of learning as a lifelong experience.

CHAPTER ONE

Introduction

Background

The extraordinary ability of Nature to produce and transform chiral compounds stereospecifically has always been fascinating to chemists and biologists alike. The chirality that results from these specific transformations plays an important role in many biological processes. Chiral interactions involving the discrimination between the enantiomers of a substance are especially important because each enantiomer can produce a dramatically different biological effect.[1] Examples of these effects can be observed when comparing optical isomers, demonstrated by the striking difference in the orange odor of (*R*)-(+)-limonene versus the lemon odor of (*S*)-(–)-limonene (Figure 1.1), and in the difference in tastes between the *R* and *S*-enantiomers of some common amino-acids.[2] More complex examples of chiral discrimination in biological processes occur in hormone regulation. L-Dopa, commonly used in the treatment of Parkinson's disease, yields the hormone, dopamine, after enzymatic decarboxylation which is subsequently converted to the correct configuration of epinephrine (adrenaline) through an enantioselective process.[3] Dopa-decarboxylase is completely stereospecific and acts only on the L-enantiomer. The importance of producing the correct configuration of epinephrine is evident from the fact that the natural (*R*)-(–)-epinephrine is at least twenty times as active as its enantiomer.[4]

Nearly all of the chiral amino acids found in proteins are of one enantiomer just as all the sugars found in nature are of one stereoisomer. Essentially, life is dependent on

only one enantiomer of chiral molecules, challenging synthetic chemists to mimic Nature's remarkable specificity when synthesizing chiral drugs.

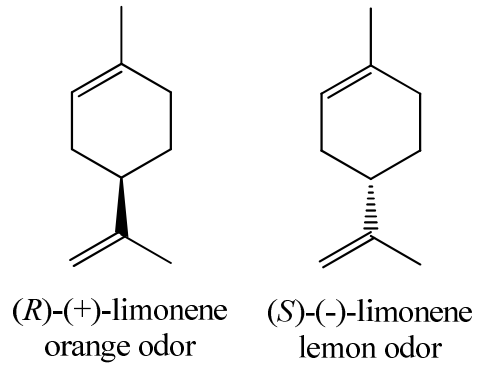
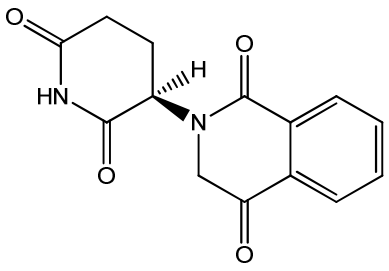


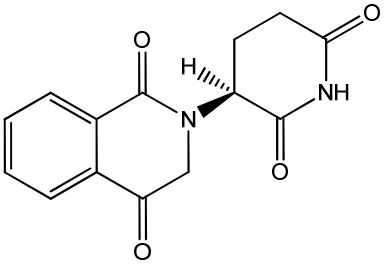
Figure 1.1. Odor and absolute configuration of limonene.

Since living systems rely on chirality to function, different enantiomers of a compound can produce different effects. Numerous synthetic drugs are racemic compounds that are used as such for reasons of cost and convenience. Although this is often adequate, there always exists the possibility that one of the two enantiomers will be undesirable and cause devastating effects. A tragic and notorious example is the use of thalidomide (Figure 1.2), a sedative and sleeping-drug used in the early 1960s to aid in morning sickness for pregnant women, which caused serious malformations in the newborns of women who were administered the drug.[3] In 1979 it was shown that the (*S*)-(–)-enantiomer of thalidomide was responsible for the teratogenic effects.[5]

With these examples highlighting the complexity of Nature, it should be evident that techniques used for the separation and identification of enantiomers are of great importance. In order to fully grasp the objectives of this present work one must know the basic definitions associated with chirality, as well as knowledge of the fundamentals and history of chiral separation techniques.



(*R*)-(+)-*N*-phthalylglutamic acid imide
(non-teratogenic form)



(*S*)-(−)-*N*-phthalylglutamic acid imide
(teratogenic form)

Figure 1.2. Stereoisomers of thalidomide.

Chirality

The property of nonsuperimposability of an object on its mirror image is called *chirality*, or handedness. The most classic example of chirality in nature is found in humans. A person's hands are *chiral*, nonsuperimposable mirror images of each other, and when placed next to each other, it is easy to see that there is no way to superimpose one hand on the other. A molecule is considered to be chiral if it is nonsuperimposable on its mirror image. For organic molecules, chirality is a necessary condition for optical activity. If a chiral molecule can rotate plane polarized light then it is termed *optically active*. Therefore, a non-racemic chiral compound will be optically active, that is it will be non-superimposable on its mirror image.[6] This condition provides evidence that the molecule and its mirror image are actually different molecules that differ only in their configuration, or left- and right-handedness, in three dimensional space (Figure 1.3).

Generally, organic molecules form chiral centers by the covalent bonding of a central carbon to four different atoms in a tetrahedral arrangement. The chiral molecule and its non-superimposable mirror image are called *enantiomers*.[6] Each enantiomer rotates plane polarized light to equal extents, but in opposite directions. The plane can be rotated either clockwise or counterclockwise depending on the nature of the optically

active compound. The isomer that requires a clockwise rotation of the plane, or to the right, is called dextro and designated (+), while the isomer that requires a counter-clockwise rotation, or to the left, is called levo and designated (–).[6]

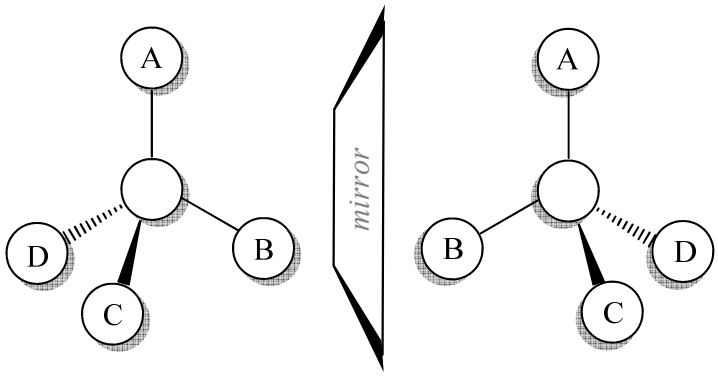


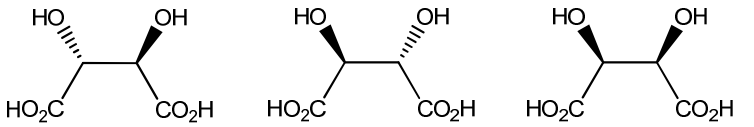
Figure 1.3. Schematic representation of nonsuperimposable mirror images.

Racemic mixtures of optically active chiral molecules contain equal amounts of each enantiomer and are optically inactive because the equal and opposite rotations cancel each other out. In general, enantiomers have identical physical and chemical properties rendering them indistinguishable by commonly used laboratory analysis. However, when placed in a chiral environment, each enantiomer generally interacts differently with other chiral compounds. This becomes particularly important in the development of chiral drugs, where one enantiomer possess the desired activity while the other will be less active, useless, or in some cases cause undesired effects.

Molecules can also have multiple chiral centers, each with its own configuration and assignment of (*R*) or (*S*) designated by the Cahn-Ingold-Prelog method.[6] When considering a chiral molecule containing two chiral centers there are a total of four possible stereoisomers, since each chiral center can be *R* or *S*. There are two pairs of enantiomers possible, (R,R)/(S,S) and (R,S)/(S,R); however, neither member of the first

pair is a mirror image of the second pair. Thus, stereoisomers that are not mirror images become possible. These are called *diastereomers*, and (in theory) do not have identical properties in any environment.

A molecule containing two or more chiral centers that also contains an internal plane of symmetry is called a *meso* isomer and is actually achiral. Achiral molecules are optically inactive. The most classic example of this behavior is tartaric acid. Tartaric acid contains only three isomers: a pair of enantiomers and an achiral meso isomer (Figure 1.4).



(+)-tartaric acid (−)-tartaric acid *meso*-tartaric acid

Figure 1.4. Stereoisomers of tartaric acid.

Chiral Resolution

Since Pasteur's epoch stereochemical resolution of tartaric acid in 1848 by mechanical separation, chemists have searched for improved methods to resolve and purify optically active compounds.[7] Obtaining one specific enantiomer of a substance is a very important, but difficult task. Unlike the chiral drugs and molecules found in nature that are of one enantiomer; chiral molecules obtained by total synthesis usually consist of mixtures of both enantiomers. Separation techniques, preparative and analytical, have advantages and disadvantages; however only the latter of these topics will be discussed in more detail over the course of this work.

Chiral resolution is performed on both the analytical and preparative scale, and the techniques used for each are quite different. A commonly used preparative method for chiral resolution of enantiomers is the formation of diastereomers. Unlike enantiomers, the properties of diastereomers are different and thus can be separated more easily by achiral techniques such as crystallization and chromatography. The majority of resolutions are accomplished on racemic mixtures of carboxylic acids with optically active amine bases to form diastereomeric salts, such as (*S*)-brucine. Although the acids are enantiomers, the salts formed are diastereomers and thus have different properties. The most common analytical method for chiral separation is chiral chromatography due in part to the fact that separation can be achieved without initial modification of the enantiomers. This method allows for the analytical separation of the enantiomers, which can then be detected separately and measured.[8] In many instances, developing and performing an enantioseparation is faster and more efficient than more traditional approaches for accessing enantiopurity.

Analysis of Enantiomers

Traditional Methods

One of the most important questions concerning studies of chiral compounds is how to determine their optical (or enantiomeric) purity. Of course, one of the oldest and most widely applied techniques for the determination of enantiomeric purity is *polarimetry*. This method makes use of the unique property of a chiral compound to rotate the plane of polarization of plane polarized light. Since enantiomers rotate the plane to a specific degree and each enantiomer has an optical rotation of equal magnitude

but opposite direction, this technique is useful for the rough determination of optical purity.[3] Optical purity is expressed in terms of *enantiomeric excess* or % ee, which is defined as the percent excess of one enantiomer over the racemate of a mixture. For example, a mixture that contains 75% one enantiomer and 25% of the other is said to have a 50% ee. This can be measured directly by optical rotation, [α], if the specific rotation, $[\alpha]^o$, of an enantiomerically pure sample has been determined accurately. The optical purity of a new sample would then be calculated according to the following:

$$\text{Enantiomeric excess} = \text{optical purity} = \frac{[\alpha]_{obs}}{[\alpha]^o} \text{ X } 100\ \%$$

The specific rotation is often highly solvent-, temperature-, and wavelength-dependent and can also be dependent upon the concentration used.[3] Thus, a sample that is enantiomerically pure may have a different observed rotation at a concentration that is not equal to the reported value for $[\alpha]^o$. Cases like this, where optical rotation is non-linearly related to enantiomeric purity, is usually due to aggregation or molecular associations in solution.[9] Impurities, achiral and chiral, can also affect the rotation of a given sample contributing to errors. Furthermore, the use of polarimetry for the purpose of optical purity determination requires knowledge of what the specific rotation of the enantiomerically pure compound would be which may not always be available, or accurate. Historically, there have been many erroneous determinations of enantiomeric purity using this method. Most other methods are considered more accurate and reliable.

A more straightforward technique useful in the analytical resolution of enantiomers is the use of optically active lanthanide-shift reagents in NMR spectroscopy. The chiral shift reagent is added directly to the mixture of enantiomers in a NMR tube.

The shift reagent moves the resonances that are associated with groups near Lewis-basic sites downfield, and often enantiomers to different degrees, which allows for the enantiomeric excess of the mixture to be determined. Upon coordination with the lanthanide reagent, chemical shifts as far downfield as 15 ppm are not unusual, though the differences between enantiomers will only be a tiny fraction of this.

A direct approach to the separation of enantiomers is by chiral chromatography. This process requires only small quantities making chiral chromatography a very useful analytical technique.

Chromatographic Separations

The concept of chromatography relies on the distribution of a compound between two phases, one of which (the mobile phase) is moving with respect to the other (stationary phase). The designations of LC (liquid) and GC (gas) prefer to the physical state of the mobile phase. As the mobile phase passes through the stationary phase, the components of the mixture equilibrate or partition between the two phases, resulting in differential migration rates through the system. Since all the molecules of a particular analyte do not behave the same, the peak for that analyte will have not only a particular retention time, but also finite width in the chromatogram.[11]

Liquid Chromatography. Liquid chromatography (LC) refers to any chromatographic procedure in which the moving phase is a liquid, in contrast to gas chromatography where the moving phase is a gas. The basic instrumental requirements for LC (and GC) are rather simple: a system to deliver the mobile phase through the

column, an injection device for application of the sample, a separation column, and a system for detection of the separated components.[12]

Liquid chromatography can be classified into four types: liquid-solid (LSC), liquid-liquid (LLC), ion exchange (IEC) and size exclusion (SEC).[12] Liquid-liquid chromatography involves a liquid stationary phase whose composition is different from the moving liquid phase. Sample molecules distribute between the two liquid phases just as in liquid-liquid extraction with the requirement that the liquids must be immiscible. Liquid-solid chromatography involves high surface area particles with retention of sample molecules occurring due to attraction to the surface of the particle. In ion-exchange chromatography the stationary phase contains fixed ionic groups such as SO_3^-, along with counter-ions of opposite charge. The samples also generally contain ions which are retained by the ion-exchange stationary phase. Finally, in size-exclusion chromatography the column packing is a porous material with pores of a certain size. Molecules that are too large are excluded from all the pores whereas molecules that are small enough will penetrate most of the pores. Thus, large molecules move through the system quickly while small molecules are retained in the packing.[12]

Aside from retention parameters and the differences in gas and liquids, the principles between LC and GC are very similar. Because LC is such an extensive field it is only briefly mentioned. On the other hand, gas chromatography will be discussed in greater detail as it is a central component in this research.

Gas Chromatography. In 1952, A. James and A. Martin introduced gas-liquid chromatography,[13] eleven years after the development of liquid-liquid chromatography by Nobel Prize awardees A. Martin and R. Synge.[14] As an analytical tool, GC was

developed for use in the direct separation and analysis of gaseous samples, volatile liquids, and solids.[15] The versatility of this technique became an attractive solution to many problems in various fields, including quality control of products for the pharmaceutical industry, and monitoring of metabolites in biological systems.

Gas chromatography has advantages and limitations compared to other analytical systems. GC is applicable to separations where only slight differences in boiling points exist, generally providing improved resolution compared to LC. Another advantage of GC is the sensitivity. Although the injected sample size is a mere 1 μL or less, the detection limits are in the picogram range.[15] A major limitation of GC is that the substances to be analyzed must be volatile (able to be vaporized without decomposition). In the case of organic molecules, volatility is rarely sufficient if the molecular weight of the compound exceeds 500 or if multiple polar functional groups are present. In such cases, the application of high temperatures (up to 300 °C) to enhance volatility, generally results in partial or complete decomposition of the analyte.

In GC, the mobile phase, generally nitrogen, helium or preferably hydrogen, enters a column through an injection port and leaves through a detector. The injector is usually heated to 150-250 ^{o}C which (hopefully) causes the volatile analytes to vaporize. The vaporized compounds are moved through the column by the carrier gas, all contained in a temperature-controlled oven. The solute travel through the column at a rate primarily determined by their physical properties, and the temperature and composition of the column.[15] The various solutes travel through the column at different rates. The fastest moving solute exits (elutes) the column first then is followed by the remaining

solutes in corresponding order. The column plays a central role in GC and considerable advancements have been made since the 1970's.[13]

Gas chromatography was first developed using packed columns containing a solid adsorbent comprised of small, uniform particles having large surface areas. It was theorized that improved performance would result from the use of such small particles packed into columns. Due to its very porous structure, diatomaceous earth was employed; however, treatment of the particles by acid washing or silanizing (reaction with surface hydroxyl groups) was required to lower the extremely high activity of this material. Active solids such as silica gel and alumina have also been used, but often resulted in undesirable tailing. To overcome this issue a thin layer of liquid was deposited on the inert solid support packed into a length of tubing, which was held in place using glass wool plugs. These were eventually replaced by immobilized and cross-linked stationary phases bound to the inner surface of capillary glass tubing, which was also considerably longer.[13] The introduction of fused-silica capillary columns represents one of the most significant advances in capillary gas chromatography. Capillary columns are open tubular columns containing a thin film of stationary phase uniformly coated on the inner walls of the tubing, unlike their packed-column counterparts, which contain more stationary phase thickly coated often having areas of thinner regions of stationary phase. The long length of capillary column, thin film thickness, and smaller inner diameter offers improved resolving power (large number of theoretical plates) compared to packed columns. Additionally, capillary columns provide narrower bandwidths compared to packed-columns as wells as better peak shapes (bands are sharper with less tailing).[13]

Capillary columns 0.25 mm or less in internal diameter often require sample loads of less than a microgram for their effective use due to their low sample capacity. It is extremely difficult to place samples of a microgram or less directly onto such columns, even with a micro-syringe. Thus, to achieve such small sample loads in practice, a split injection system can be employed. After vaporization of the sample, split injection (split flow) splits the gas stream, by a controllable restrictor, at a ratio of 40-100 to 1 or more, so that only 1 % (or less) of the sample passes into the capillary column, whereas the remainder passes to waste. Even with discarding so large a fraction of the sample, solution concentrations as low as 50 ppm are easily analyzed.

As each solute elutes from the column, it enters the detector, and an electronic signal is generated upon interaction with the solute. There are a variety of detection systems for GC, and several are shown in Table 1.1 along with their selectivities.

Table 1.1. Typical Characteristics for Common GC Detectors.

Detector	Selectivity	LOD
Thermal conductivity detector (TCD)	Responds if analyte thermal conductivity is different from carrier gas	1 ng/mL
Flame ionization detector (FID)	Organic compounds	1 pg C/s
Electron-capture detector (ECD)	Electron-capturing compounds (halogens)	10 fg/s
Nitrogen-Phosphorous detector (NPD)	N- and P-containing compounds	1 pg N/s 0.5 pg P/s
Flame photometric detector (FPD)	P and S-containing compounds	50 pg S/s 2 pg P/s
Photoionization detector (PID)	Aromatics	5 pg C/s

The limit of detection (LOD) refers to the quantity or concentration of solute, which generates a peak height (area) that corresponds to a S/N (signal-to-noise ratio) of 3. This is the minimum mass (or concentration) flow of the compound in the mobile phase required for the detector to give a response with certain probability (>99% for S/N of 3).[15] The FID detector is the most commonly employed detector for GC due the fact that it responds to virtually all organic molecules with favorable sensitivity.

The size of the signal generated by the detector is recorded by a data system and is plotted against elapsed time to produce a chromatogram. The profile of a chromatographic band, as registered by the detector, ideally should have a Gaussian distribution, resulting in a completely symmetrical peak (Figure 1.5).[11] The ideal chromatogram should have no overlap between closely spaced peaks. Any peaks that overlap are referred to as co-eluting.

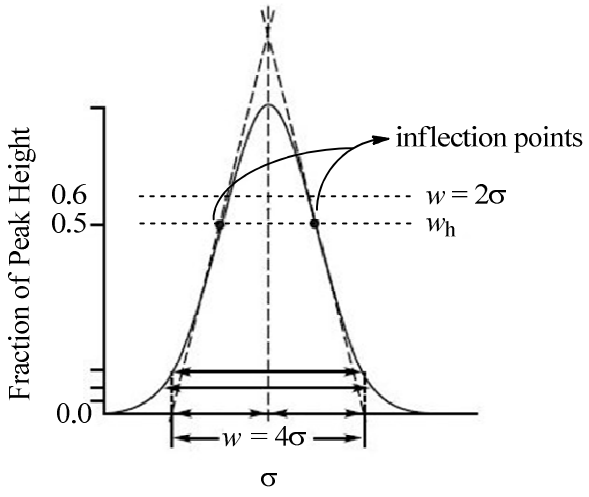


Figure 1.5. A normal Gaussian distribution. The inflection point occurs at 0.607 of the peak height. w_h is the width at 0.500 of the peak height and corresponds to 2.354σ. Modified with permission by John Wiley & Sons, Inc. (*Ref. 11).*

The retention time and area of a peak are important in that they are used to identify and measure the amount of the compound in the sample, and are of use in certain chromatographic calculations. The area of the resulting peak is proportional to the amount of the compound in the sample. A larger peak is obtained as the concentration of the corresponding compound increases; however, the proportionality typically varies for various compounds.

The ability of a chromatographic column to transport a compound with little peak broadening is referred to as its *efficiency*.[11] Column efficiency is expressed as plate height, (*H*), which for a GC column is the length equivalent to one theoretical plate and can be calculated from the chromatogram as follows:

$$N = 16\left(\frac{t_R}{w}\right)^2 = 5.54\left(\frac{t_R}{w_h}\right)^2 \qquad H = \frac{L}{N}$$

where, t_R is the retention time, w the baseline peak width, and L the length of the column. The peak width at 50% of the peak height is referred to as w_h. The narrow baseline peak width, w, observed with capillary columns is the main reason for why these columns are superior to packed columns and have been observed as narrow as 0.02 min (1.2 sec). If the peak is not symmetrical (uneven baseline) different values can be calculated for N because the width measurement does not follow a Gaussian distribution. Typically, N increases the higher up on the peak the width is measured. The most common measure of efficiency is the plate number, N. Because the concept originated from the analogy with distillation, it was originally called the number of theoretical plates contained in the chromatographic column. However, this is not particularly useful since the column does not contain true "plates." The theoretical plate number is a measure of relative peak

broadening (related to *w*) that has occurred while the analyte passed through the system.[11] Capillary columns range from 20 to 200m, and typically provide in theoretical plate numbers that are extremely high (N= 2x10^5 or more). Typically 30 m capillary columns have theoretical plate numbers in the 100,000 range. The retention of an analyte on a column can be expressed by its retention time (t_R), retention volume, or capacity ratio (k'). The capacity ratio is defined by:

$$k' = \frac{t_R - t_M}{t_M}$$

where, t_M refers to the dead volume (i.e. time required for the analyte to go through the "dead" space in the system without being sorbed). In GC, air or methane is often used to determine this volume and is measured by the retention time of the gas. Thus, the partition ratio, k, can then be determined as indicated above.

When considering the separation of two components, the ratio of their individual partition ratios or separation factor, α, is evaluated from the chromatogram by calculating the ratio of the net retention times (t_R) of the two components (Figure 1.6).[11]

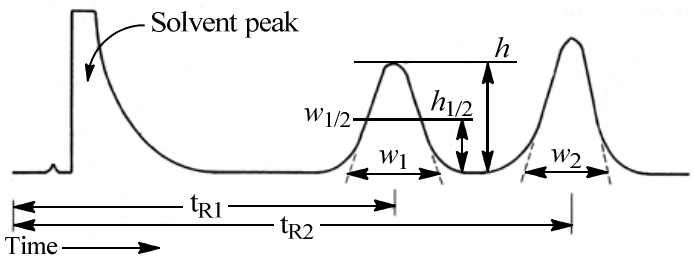


Figure 1.6. Separation and associated parameters of two resolved analytes. Modified with permission by John Wiley & Sons, Inc. (*Ref. 11*).

The separation factor is a measure of relative peak separation. In the case of a mixture of two components, it is usually defined so that its numerical value is >1.0, which means that analyte B is retained longer than analyte A, according to the following:

$$\alpha = \frac{k_B'}{k_A'}$$

A better measure of the efficiency is resolution (R_s). It defines the degree of separation of two analytes, or peaks, in a chromatographic system:

$$R_S = 2\frac{(t_{R2}-t_{R1})}{(w_1+w_2)} = 2\frac{\Delta t_R}{(w_1+w_2)}$$

As shown, R_s, is defined as the peak separation, divided by the sum of the mean values of the baseline peak widths.[11] The larger the resolution the better the separation. As the baseline peak width of a Gaussian band equals 4σ (where σ equals standard deviation), it follows that a difference in retention time equal to 4σ corresponds to $R_s = 1$, about a 2% peak overlap. A '6σ resolution' or $R_s = 1.5$ represents a complete baseline separation, whereas $R_s < 0.8$ is generally considered insufficient separation.[7]

The determination of enantiomeric purity can be achieved by GC in either of two ways: (1) conversion of the enantiomers into diastereomers with an appropriate asymmetrical (chiral) reagent, followed by separation of the diastereomers on an achiral GC column, or (2) separation of the enantiomers on an optically active stationary phase. The latter is known as *chiral chromatography*. When a mixture of enantiomers is placed on a chromatographic column for which the stationary phase contains chiral and non-racemic substances, then in principle the enantiomers could interact differently with such

a phase and move along the column at different rates. In this approach, enantiomers may be separable without having to be converted to diastereomers. When sufficient separability is achieved, chiral gas chromatography is the preferred analytical method, mainly due to its low detection limits; it is possible to detect at little as 0.01% of a minor enantiomer.

Chiral Stationary Phases for Gas Chromatography. Enantiomer separation by gas chromatography is mainly performed on three types of stationary phases (CSPs). The first successful results in the field were achieved using chiral stationary phases (CSPs) comprised of amino acids or oligopeptide derivatives.[8] Enantiomer separation was based largely on hydrogen bonding differences, was generally limited to amino acids, and required derivatization of the amino acids to increase volatility. A common problem associated with these phases was low temperature limits (110 ^{o}C). A breakthrough in this type of phase occurred with the development of polysiloxane-immobilized phases, which exhibited what was then unsurpassed thermal stability.

The second type of CSPs, was based on complexation to chiral coordination compounds, and was introduced by Schurig. A chiral β-dicarbonyl ligand complexed to a transition metal by the two oxygen's, constitutes the enantioselective component. One such dicarbonyl compound, dicarbonylrhodium(I)-3-trifluoroacetyl-(1*R*)-camphorate was employed for the resolution of 3-methylcyclopentene.[16] The metal complex is initially coated onto a capillary column as a solution in squalane ($C_{30}H_{62}$). A limiting factor of the coordination-type CSPs was the low temperature range of operation (25-120 ^{o}C). The preparation of immobilized polymeric CSPs (Chirasil-Metal) provided a solution having improved thermostabilities.

The third and most recent type of CSP is based on inclusion effects. It has been frequently demonstrated that cyclodextrins (CDs) are capable of forming inclusion complexes with a variety of guest molecules. Inclusion complex formation is stereoselective; therefore it can be used for the resolution of enantiomers. The first enantioseparations performed using a cyclodextrin stationary phase for gas chromatography was reported in 1983 for α- and β-pinene, and *cis*- and *trans*-pinane on packed columns employing α-cyclodextrin in formamide.[17, 18] It was later recognized that derivatized cyclodextrins could be used in high resolution capillary columns for enantiomer analysis when mixed with a polar solvent which acts as a liquid matrix.[17] A variety of cyclodextrin derivatives have been effectively employed as chiral selectors for stationary phases utilized in chiral chromatography. CD phases offer numerous advantages, including increased thermal stability and the ability to separate a much wider variety of chemical classes.[8]

Cyclodextrins

Cyclodextrins are macrocyclic oligosaccharides most commonly comprised of 6, 7, or 8 glucosidic units , referred to as α, β and γ-CDs, respectively (Figure 1.7).

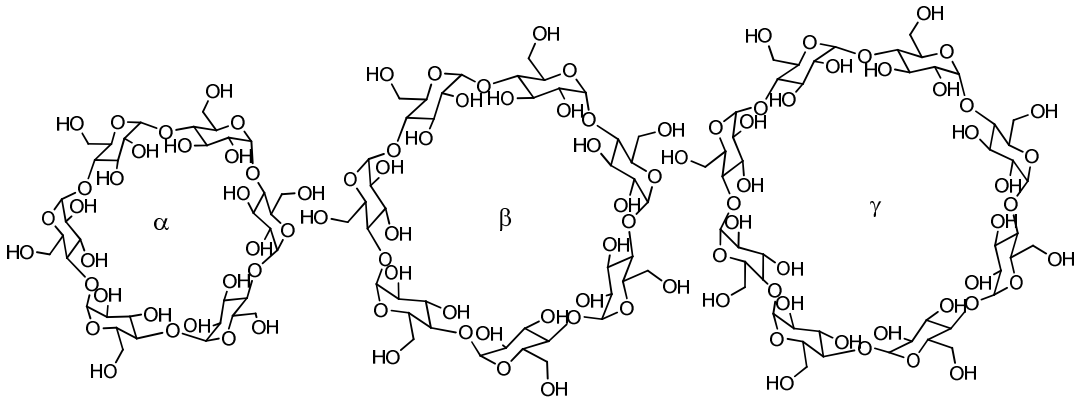


Figure 1.7. Schematic structures of α-, β- and γ-CDs.

Composed of D-glucose units linked in a 1,4' manner, these are chiral, non-racemic molecules. Smaller guest molecules can enter the cyclodextrin's cavity forming inclusion complexes. This phenomenon is called *molecular recognition*, while the selectivity in the formation of complexes with enantiomeric species as guests is called *chiral recognition*.[19]

Physical Properties of Cyclodextrins

Cyclodextrins are obtained by enzymatic degradation of starch by the enzyme CD glucosyltransferase, and are cyclic, non-reducing oligosaccharides consisting of D-glucopyranose units bonded through α-1,4 linkages. Unsubstituted CDs are crystalline, non-hygroscopic, homogeneous substances, which are torus-like macrocycles built up from glucose units. In CDs, the sugars adopt a $^4C_1$ chair conformation and orient themselves so that the molecule forms a toroidal truncated cone structure (Figure 1.8).

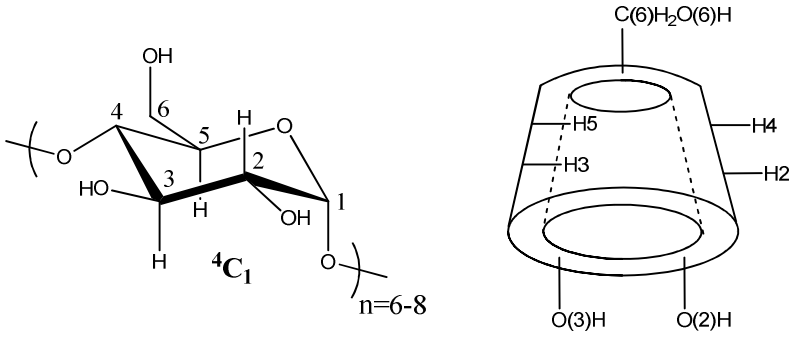


Figure 1.8. Notation of conformation and schematic view of the glucopyranoside ring.

CDs were originally thought to have rigid structures; however, this has been shown to be inconsistent with the CDs ability to selectively complex guests of various shapes.[19] CD complexes are held together by weak intermolecular forces which somewhat limit the mobility of the CD, but does not render it completely rigid. A completely rigid structure is also inconsistent with the ease of formation of inclusion

complexes of various shapes and sizes, since the above implies an efficient fit of the host and guest is required.[20] The cavity is lined by the hydrogen atoms H3 and H5 and glucosidic oxygen bridges connecting the glucose units. These oxygen bridges have electron pairs oriented toward the inside of the cavity lending it to some Lewis base character due to the high electron density. The C(2) hydroxyl group of a glucopyranose unit can form a hydrogen bond with the C(3') hydroxyl group of the neighboring glucopyranose unit. In β-CD a complete secondary belt is formed by these hydrogen bonds giving it a more rigid structure compared to α and γ-CD. This arrangement of hydrogen bonding can explain the observation that β-CD has the lowest solubility of all CDs.[21] The most important physical and chemical characteristics of the most common CDs can be compared in Table 1.2.[22]

Table 1.2. Some characteristics of α, β, and γ-CD.[22]

CD	α-CD	β-CD	γ-CD
Number of glucopyranose units	6	7	8
Molecular weight (anhydrous)	972.85	1134.99	1297.14
Solubility per dm^3 H_2O at 298.2 K	14.5	1.85	23.2
Annular diameter measured from C(5) hydrogens, Å	4.7	6	7.5
Annular diameter measured from C(3) hydrogens, Å	5.2	6.4	8.3
Annular depth from primary to secondary hydroxyl groups, Å	7.9 - 8.0	7.9 - 8.0	7.9 - 8.0
Annular volume, $Å^3$	174	262	472
Partial molar volumes, cm^3 mol^{-1}	311.4	703.8	801.2
pK_a O(2)H and O(3)H at 298.2 K	12.33	12.20	12.08

Other important properties of cyclodextrins include the following: (1) they are non-reducing sugars, (2) glucose is the only product of acid hydrolysis, (3) their molecular weights are always integral numbers of (162.1), the value of glucose, (4) they are non-toxic, and (5) they do not appreciably absorb UV or visible light.[22]

Chromatography is one of the most important methods for direct studies of molecular and chiral recognition by CDs today. The amazing sensitivity of CDs to the shapes of guest molecules is illustrated by the significant differences in retention times of very similar compounds. For illustration, complexes between isomers of 1,8-dimethylnaphthalene **2a-d** with β-CD (**1**) have been shown to exhibit large differences in retention times compared to the complex between naphthalene isomer **2e** and β-CD (**1**) determined by gas chromatography (Figure 1.9).[19] Although the large differences in retention times are most likely due to stoichiometric differences among the complexes,1:1 vs. 2:1(CD to guest), it serves to illustrate the versatility and selectivity of CDs as hosts.

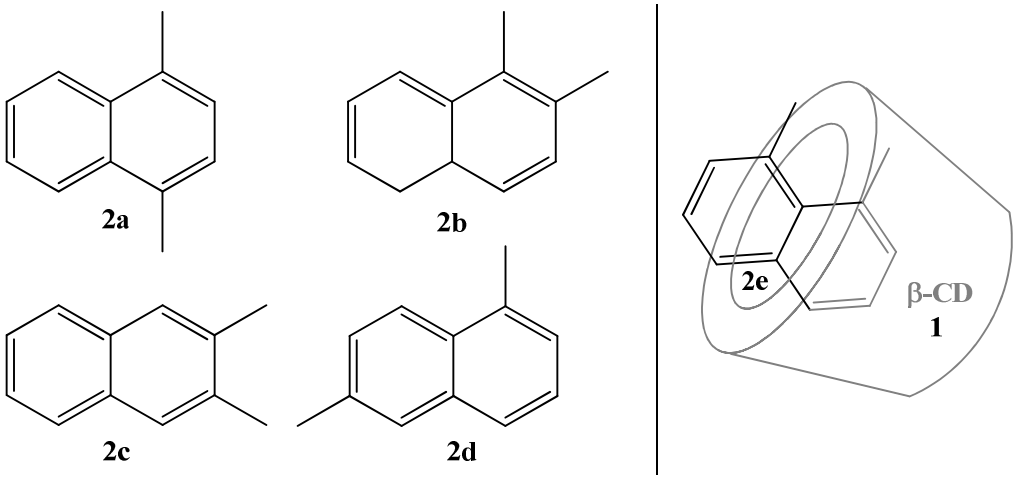


Figure 1.9. Schematic representation of the complexation between β-CD and select isomers of 1,8-dimethylnaphthalene.[19]

Complexation Behavior of Cyclodextrins

The complexation of a guest by cyclodextrin involves the partial or complete desolvation of the guest as it enters the CD cavity, displacement of the water from the

cavity and adjustment of the guest to the thermodynamically most favorable orientation within the cavity.[22]

The majority of CD complexation studies have been carried out in aqueous solution and considerable attention has been focused on the hydration of the CD and the guest during the complexation process.[22] Cyclodextrins form complexes with differing amounts of water, meaning CD cavities are seldom empty. Even if a cyclodextrins does not contain another guest there is usually solvent or water molecules occupying the cavity.[19] Hydrogen bonding between the guest and CD stabilizes complexes along with conformational changes by either the guest or cyclodextrin. Other effects demonstrated by CD complexation behavior include hydrophobic effects, release of high energy water from the CD annulus, relief of conformation strain in the uncomplexed CD, dipole-dipole interactions, and London dispersion forces.[22] Water occupying the CD annulus does not form typical hydrogen bonding interactions found in bulk water and is often referred to as high energy water. The entry of a guest into the CD annulus expels some or all of this high energy water, an important part of the complexation process.[22] The enthalpy and entropy of complexation follows a trend where ΔH^o is negative and ΔS^o is negative, and as a consequence CD complexation is said to be enthalpy driven. The inherent chirality and shape of CDs provide opportunities for both chiral and size discrimination by complexation.

The cavity of β-CD and its derivatives is wide enough to accommodate a naphthalene ring axially.[18] Predominately, analytes such as naphthalene enter the CD cavity through the secondary face first, shown by ^{1}H NMR studies, although examples of exceptions do exist. This is in agreement with the fact that 2,6-dimethyl-4-nitrophenol,

the corresponding phenolate anion and 4-nitrophenol and its -phenolate form stable complexes with α-CD, while 3,5-dimethyl-4-nitrophenol and -phenolate are inhibited from forming α-CD complexes most likely due to the steric hindrance formed from the nitro-substituted end entering the cavity. When complexes of this series form, neither the phenol or phenolate end of the guest enters the cavity first, and the phenolates are known to complex more strongly than the phenols.[22] The stereochemistry and stability of a CD complex can be profoundly affected by the structure and charge of the guest. Alignment of the dipole in the molecule, generally in a head-to-tail fashion, with the dipole moment of the cyclodextrin affects how the guest will enter and complex with the CD. In the case of 4-nitrophenol, the nitro group will align with the positive, primary face of the cyclodextrin due to the nitro group containing an adjacent negative charge. At the same time, inhibition of a guest complexing with cyclodextrin can occur where hydration of the guest is possible, as was the case with the phenol and phenolate groups, above.

Novel Modified Cyclodextrins as Chiral Selectors in Gas Chromatography

Basis for this Research

Introduction. As shown above, enantiomers can only be directly separated by systems that contain a chiral, non-racemic selector. Therefore, enantioseparations are generally carried out by chiral chromatography on a chiral stationary phase containing a resolving agent of high (but not necessarily complete) enantiomeric purity.[23] The use of cyclodextrins as chiral selectors in GC has provided a deeper insight into the mechanisms of chiral recognition and has proved optimum for the separation of a broad spectrum of optically active compounds which is of particular importance to the pharmaceutical

industry.[24] In this work, we will introduce the progress we have made toward the development of unique CSPs for the separation of enantiomers.

The majority of enantiomer separations have been carried out with use of β-CD and its various derivatives due to its commercial availability, and this is precisely the reason, along with cost, for its use in our research. The first GC enantioseparations employing β-cyclodextrins as chiral selectors was reported by the Sybilska's group in 1983, in which packed columns were employed, but resulted in short lifetimes and low peak efficiencies.[25] Contributions made by Schurig and co-workers not only increased the lifetimes of the columns, but provided a solution for the problem of the high melting point of CDs by dissolving them in moderately polar polysiloxanes.[26] Capillary columns prepared by coating their inner surface with polar derivatives of CDs were introduced by Armstrong and co-workers as very useful chiral GC columns.[27] The majority of modern commercially available chiral GC columns are prepared based on this technology.[28]

Since cyclodextrins have been so widely applied in the separation sciences, the evolution of cyclodextrins and its derivatives has been intense over the last two decades resulting in a number of different phases with different selectivities. For example, the Lipodex series which incorporate per-*n*-pentylated CDs in their CSPs are liquid at room temperature.[28] The Chiraldex series by Astec offers the widest variety of commercially available chiral GC columns, marketing eight different types of cyclodextrin derivatives, two of which are patented (Table 1.3). In addition, these eight types of derivatization are available on all three of the common cyclodextrins (α, β and γ). One of the Astec phases (DM) corresponds to that offered by J&W as CycloSil-B.

Novel Cyclodextrin Stationary Phases. Our search for new synthetic derivatives of CDs has been motivated by the relatively low thermal stability of the current commercially available columns as well as by the recognition that each CD derivative has different separation characteristics, i.e. there is no one best phase for all separations. As was the case with the Chiraldex series, out of 24 possible α-, β-, and γ-CD phases only eight of these exhibited separation characteristics and each of the eight phases has different selectivity.

Table 1.3. Several Currently Available Cyclodextrin-based CSPs. (*) denotes patented.

Phase	**Cyclodextrin**
TA*	Trifluoroacetyl (2,6-di-O-pentyl-3-trifluoroacetyl)
DM	Dimethyl (2,3-dimethoxy-6-O-*tert*-butyldimethylsilyl)
DP	Dipropionyl (2,3-di-O-propionyl-6-O-*tert*-butyldimethylsilyl)
DA	Dialkyl (2,6-di-O-pentyl-3-methoxy)
PN	Propionyl (2,6-di-O-pentyl-3-propionyl)
BP	Butyryl (2,6-di-O-pentyl-3-butyryl)
PH*	S-Hydroxypropyl ((S)-2-hydroxy propyl methyl ether)
PM	Permethyl (2,3,6-tri-O-methyl)

This means that only a limited number of chiral separations can be performed on any single CD stationary phase,[21] supporting the continual need for improved chiral selectors. There is no universal chiral stationary phase available, and selection and efficiency are often determined by trial and error. Our objectives are to synthesize novel cyclodextrins that are significantly different from what is currently used in existing CSPs

and to test their performance as chiral selectors in enantioseparations by gas chromatography. To accomplish this, our derivatization efforts will be primarily focused on the secondary face of β-cyclodextrin. It has been shown that the site of attachment can affect the extent of complexation and that the secondary face is particularly important in chiral discrimination.[21]

The only established approach to elaborate CDs is through reaction of their hydroxyl groups. In cyclodextrins, every glucopyranose unit has three free hydroxyl groups available for modification. For β-CD this results in twenty-one modifiable hydroxyl groups generating the potential for a complex mixture of fully substituted to partially substituted products. Two of the hydroxyl groups, C(2) and C(3), are secondary while the other, C(6), is primary (Figure 1.10).

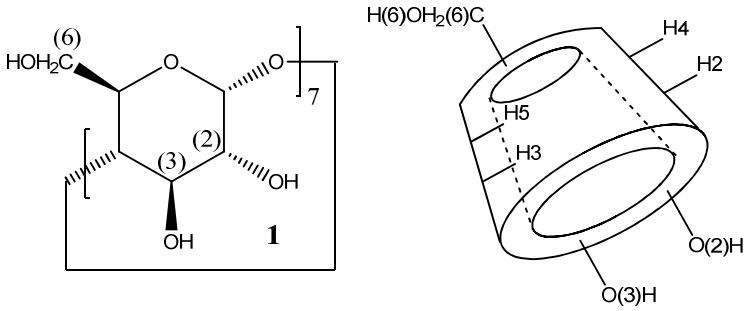


Figure 1.10. β-cyclodextrin (**1**).

Modification generally involves the substitution of the hydrogen atom or the hydroxyl group by a wide variety of substituents, though, reactions of this type generally results in a large number of positional isomers. It is possible to control, to some degree, the regioselectivity of the reactions, to substitution of either one or a specific combination of hydroxyl groups. The most commonly employed methodology includes the initial masking (protecting) of the primary C(6) hydroxyl groups by per-silylation with *tert*-

butyldimethylsilyl chloride in pyridine.[29] This methodology allows for sufficient reaction of the primary side, but leaves the secondary side available for subsequent reaction. By employing a robust group at the C(6) position, more intense modification at the secondary side can be achieved without fear of hindering the protected primary hydroxyl groups.

The unique functionality of the modified cyclodextrins that we have pursued is comprised of characteristics such as annulations bridging the C(2) and C(3) oxygen and/or containing significantly different functionality at the C(2) and/or C(3) positions. Annulations of this type have not yet been reported for cyclodextrin stationary phases, though annulations reactions have been reported for glucose derivatives.[30] By following the methodology developed for glucose, we believe that similar functionality can be realized on cyclodextrin and its derivatives. In addition, modification to the stereochemistry of the secondary hydroxyl groups and/or replacement of the primary hydroxyl group with smaller groups was also considered when designing our chiral selectors. Contributions made by Baer et al. in the development of per(6-deoxy)-β-cyclodextrins,[31] has provided us with an elegant approach to the synthesis of cyclodextrins bearing smaller functional groups (i.e., methyl) on the primary side. To our knowledge, derivatives of this type have not been explored for their ability to perform enantioseparations. We believe that β-cyclodextrin derivatives containing the described attributes could offer a potential change in the thermal stability and chiral recognition properties. Our approaches to new chiral selectors are described below, as well as a detailed account of the performance of our CSPs at separating enantiomers by chiral gas

chromatography. Some simpler CD derivatives that appear to have been overlooked to date were also prepared and tested.

Application in Gas Chromatography.

Since its introduction, chiral capillary gas chromatography has remained a very active field where interesting and new discoveries have occurred for over 25 years. As far as for the production of new chiral stationary phases, surprising achievements have been made enabling the accumulation of reported literature on enantioseparations by cyclodextrin CSPs. With increased knowledge, chemists have been able to fine tune the chiral discrimination by modification of the cyclodextrin chiral selector, and is the focal point of our group's research.

In the present work we have prepared several new cyclodextrin derivatives, and used them as CSPs for chiral capillary gas chromatography to investigate the influence that altering the functional groups, primarily at the secondary face, has on the selectivity. Because enantioselectivity depends largely on the groups fixed by derivatization of the hydroxyl groups on the CD rings,[32] further investigation on the influence of chemical modifications of the CDs on enantioseparations is still significant.

CHAPTER TWO

Initial Attempts at Modification of β-Cyclodextrin

Introduction

Through modification, cyclodextrins can be transformed into a wide variety of molecular hosts. This creates the possibility of tailoring a cyclodextrin host for a particular type of guest to improve characteristics such as chiral discrimination. The selectivity is often improved through modification of the CD by improving solubility, the ability of CD derivatives to form secondary bonds or by controlling the degree of hydrophobicity in the CD cavity.[23] The presence of a variety of different functionality could provide additional interactions between the analyte and the cyclodextrin increasing the number of racemates that can be separated. The key to obtaining such compounds involves the use of a strategic approach to control the regioselectivity and limit the extent of reaction, to substitution of either one or a specific combination of the hydroxyl groups. Investigations in this area have been performed in our laboratory and progress has been made towards the preparation of new modified cyclodextrins.

Reactivity of Hydroxyl Groups

The two major factors that should be considered when modifying cyclodextrins is the nucleophilicity of the C(2), C(3), and C(6)-hydroxyl groups, and the potential for the cyclodextrin to form a complex, at least in some part, with the reagents used. Among the three types of hydroxyl groups, the primary C(6)-OH groups are the most easily substituted, the C(2)-OH groups are most acidic, and the C(3)-OH groups are the most

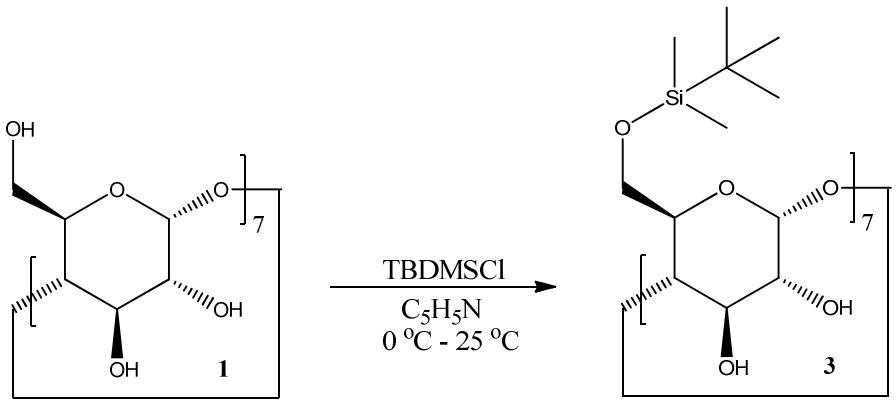
inaccessible.[19] Therefore electrophilic reagents will initially react with the C(6)-OH group. More reactive reagents will react with all of the hydroxyl groups in the order of decreasing reactivity: C(6)-OH > C(2)-OH > C(3)-OH.[19] The lack in reactivity of the C(3)-OH group can be attributed to its additional hydrogen bonding interaction with an adjacent C(2)-OH from a neighboring glucose unit.

Preparation of Modified Cyclodextrins

Modification of the C(6) Primary Hydroxyl Groups

Since each cyclodextrin contains numerous sites for modification, reaction of the hydroxyl groups can occur to different degrees resulting in a complex mixture of fully- and partially-substituted products. Instead, controlled modification of cyclodextrins can be used by exploiting the different reactivities of the hydroxyl groups, but there are serious limits. For example, when β-CD is reacted with *p*-toluenesulphonyl chloride in pyridine, a mixture of products differing in their degree of substitution is generated upon which the C(6) mono-tosylate can be obtained in only 30% yield only after chromatography.[22,33] Moreover, the use of excess *p*-toluenesulphonyl chloride in pyridine results in the per-tosylation of all the C(6) primary hydroxyl groups. The purification of the per-tosylated CD can be accomplished after column chromatography has removed other CDs in which all but one of those groups has reacted or in which some reaction of the secondary hydroxyl groups has occurred.[22] The major difference is that mono-tosylation involves competition between the primary hydroxyl groups, whereas with per-tosylation competing reactions of the secondary hydroxyl groups becomes more significant as the degree of substitutions increases.

Often it is desirable to mask (protect) all of the primary hydroxyl groups at one time. Consistent with Fugedi's methodology, (6-*O*-*tert*-butyldimethylsilyl)-β-CD (**3**) can be obtained, upon reaction of the parent β-CD (**1**) with *tert*-butyldimethylsilyl chloride in pyridine at 0°C in good yields (Scheme 2.1).[29] These reactions produce a mixture of compounds having different degrees of silylation from between four to seven of the primary hydroxyl groups, requiring tedious chromatography similar to the tosyl reactions, but without the more easily visualized aromatic group. Though tedious, protection of the primary hydroxyl groups is required for subsequent reaction of the secondary face. The advantages to using bulky silyl ethers on the primary face include increased solubility in common organic solvents and the stability of the protecting group in neutral and basic conditions.



Scheme 2.1.Protection of the primary hydroxyl groups via per-silylation of β-cyclodextrin.[29]

Direct Route to Cyclodextrins Modified at the Secondary Face

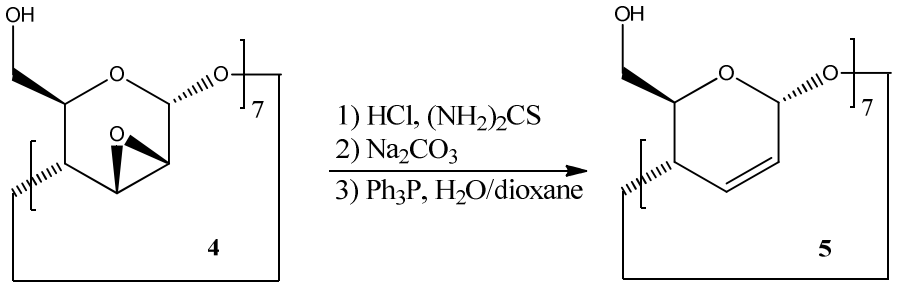
The secondary hydroxyl groups of β-CD are the most acidic, with pK_a values around 12.2.[34] They are located on the wider end pointing towards the cavity and under basic conditions will react readily as alkoxide nucleophiles. The inclusion of the reagents

in the cavity of the CD and the orientations of the reactive groups in the complexes affects the regioselectivity of reactions on CDs.[19] By simple adjustment of the reaction pH and size of the CD cavity, *O*-alkylation can alternate between the secondary and primary hydroxyl groups. While the primary hydroxyl groups are the most nucleophilic under neutral or basic conditions, above pH 10-11 the secondary hydroxyl groups are generally selectively modified. The size of the CD cavity has an effect on the strength of interactions and the orientation of the complex with the reagent. For example, in alkaline aqueous solutions, tosyl chloride reacts with α-CD to give C(2)-tosyl-α-CD, whereas when reacted with β-CD the C(6)-tosyl-β-CD is obtained.[35] Synthetic strategies used to access cyclodextrins derivatized on the secondary face generally proceed through nucleophilic displacement reactions of tosyl intermediates. A direct route to the modification at the secondary face would be an efficient alternative to these approaches and has been relatively unexplored.

Involving the Reduction of the C(2) and C(3) Hydroxyl Groups. Conversion of vicinal diols into olefins (alkenes) opens the possibilities for a variety of structural modifications and as a consequence, a number of reactions exist for this conversion.[36, 37, 38] With the formation of an olefin derivative, reactions such as the Diels Alder cycloadditions or catalytic additions of cyanide could, conceivably, provide cyclodextrins bearing larger more extended cavities and altered polarities resulting in the potential for increased interactions in inclusion complexation.

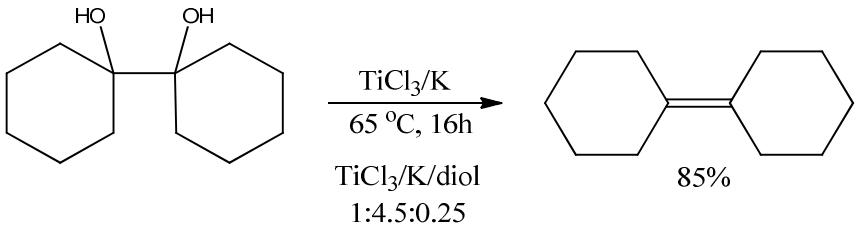
The only route used to access the olefinic cyclodextrin derivative (**5**) proceeds through an intermediate thiirane and does not employ a direct route. Fujita and coworkers have developed a one-pot preparation of this starting from the cyclomanno

derivative (**4**). The synthesis is simple and requires heating an aqueous acidic solution of (**4**) at 90 ^{o}C for 30 min followed by stirring at room temperature under alkaline conditions followed by reaction with triphenylphosphine (Scheme 2.2).[39] Interestingly enough, Fujita and coworkers were solely interested in the development of the thiirane intermediate and did not fully investigate the chemistry of the olefin (**5**). It was postulated that direct reduction of the secondary hydroxyl groups could be achieved without the need to proceed through a more reactive intermediate. Therefore, we sought to develop a direct route for the synthesis of (**5**) avoiding the use of an intermediate.



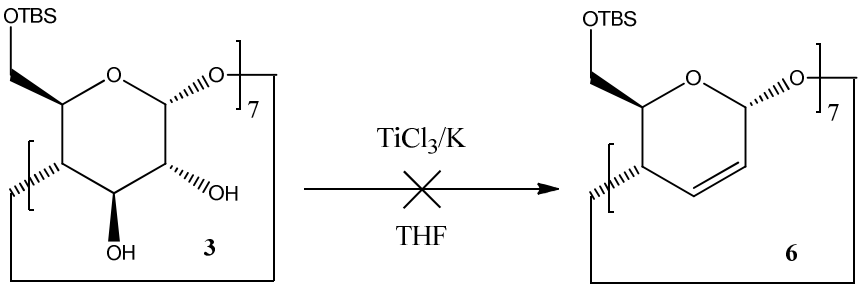
Scheme 2.2. Preparation of olefin (**5**) from cyclomannoepoxide (**4**).[39]

Progress made in the development of low-valent titanium reagents ($TiCl_4$/Zn,[40] $TiCl_3$/Mg,[41] $TiCl_3$/$LiAlH_4$[42]) has provided a straightforward and direct way to reductively dimerize ketones and aldehydes to olefins. It has also been established that the carbonyl coupling reaction described above proceeds through a pinacol dianion intermediate and therefore can be applied for the deoxygenation of 1,2-diols to olefins.[44] Rieke and coworkers have described a general method for the preparation of highly active metals, such as Ti^{o}, in a finely divided form by reduction of the metal halide with potassium in tetrahydrofuran (THF) (Scheme 2.3).[43]



Scheme 2.3. Deoxygenation of bicyclohexyl-1,1'-diol to cyclohexylidenecyclohexane with $TiCl_3$/K (*McMurry Reduction*).[43]

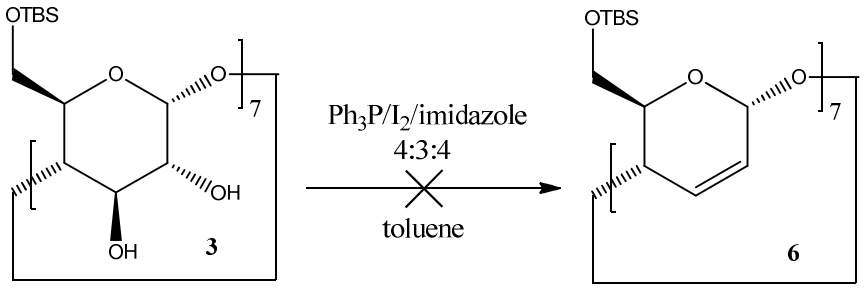
Utilizing Rieke's methodology, our first attempts at synthesizing an olefinic CD derivative began with the reduction of a slurry of $TiCl_3$ in THF with 4.5 equivalents of potassium upon which 0.25 equivalents of the starting diol (**3**) was added. A black slurry was observed, typically indicative of a successful reduction. The reaction resulted; however, in a slurry that no products could be extracted from, nor were there any resonances in the NMR spectrum to support the formation of the product (Scheme 2.4) In fact, reproduction of the reaction a second time only resulted in recovery of the starting material. Furthermore, titanium dioxide, a by-product of the reaction, was not isolated from the reaction mixture further supporting the lack of reactivity.



Scheme 2.4. Failed attempt at reduction of 6-*O*-*tert*-butyldimethylsilyl-β-CD (**3**) by the McMurry reduction.

After failures of the reduction via low-valent titanium, it was postulated that the cyclodextrin may not be able to react on the surface of the titanium and this may be

responsible for lack of reactivity. Therefore, an alternative synthesis for the reduction at the secondary face was sought. After searching the literature, to our satisfaction we stumbled upon a novel reagent system developed for carbohydrates employing the same conversion of *vic*-diols to olefins. The Garegg reduction successfully converts 1,2-diols into alkenes using a triphenylphosphine/iodine/imidazole system in the ratio 4:3:4.[45] This system has the main advantage of only a single reaction step and was noted to be particularly successful for *trans*-1,2-diols. With much anticipation, the starting diol (**3**) was subjected to the reagent system in refluxing toluene. (Scheme 2.5). However, this reaction proved to be unrewarding.



Scheme. 2.5. Failed Garegg reduction of (**3**).

Involving the Oxidation of the C(2) and C(3) Hydroxyl Groups. It was hypothesized that oxidation of the secondary face may provide a more facile route to modified cyclodextrins and could perhaps have some success where the reductions failed. Triacetoxyperiodinane (**7**), known as the Dess-Martin Periodinane (DMP), is a practical reagent used for the simple and efficient oxidation of secondary alcohols to ketones. As an oxidizing agent it is preferred over chromium- and dimethyl sulfoxide-based oxidizing agents due to shorter reaction times and the amount of reagent needed.[46] The Swern dimethyl sulfoxide-based reagents are selective and workup procedures are simple;

however, these reagents are sensitive and require immediate use after their preparation.[47] Though DMP has been utilized as a successful oxidizing agent, it is also known[48] to cleave the C-C bond of 1,2-diols making us reluctant to use it. Alternatively, *o*-iodoxybenzoic acid (IBX) (**8**), a DMP precursor, has been shown to be a valuable oxidant of functionalized alcohols, proceeding under milder reaction conditions, and has the ability to perform delicate transformations (Figure 2.1).

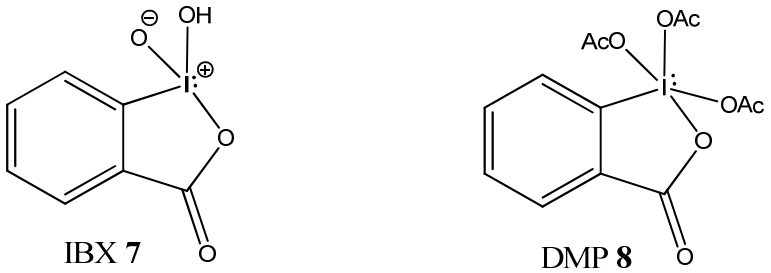
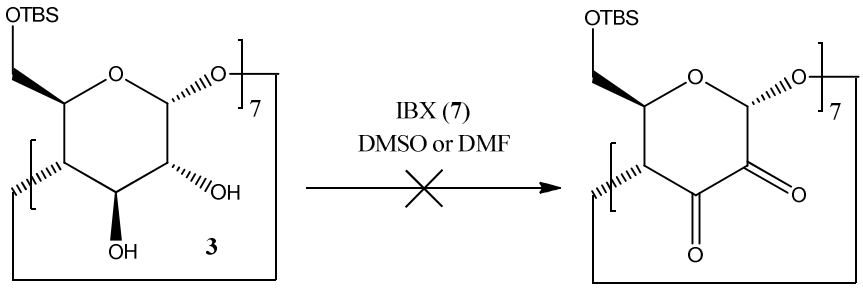


Figure 2.1. Dess-Martin Periodinane, DMP, and its precursor IBX.

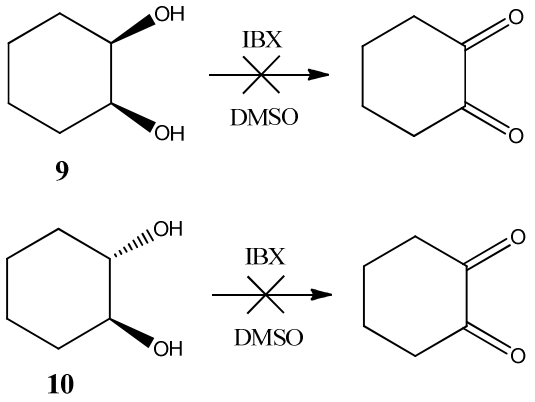
Based on our hope that IBX could oxidize the secondary alcohols of cyclodextrin to α-diketones without cleaving the C-C bond,[48] IBX was prepared according to literature[49] and then reacted with (6-*O*-TBS-2,3-dihydroxy)-β-CD (**3**). Reactions were run in both DMSO and DMF at room temperature and eventually heated to 60 ^{o}C. However, the reactions resulted in the recovery of starting materials (Scheme 2.6).



Scheme 2.6. Failed first attempts at the oxidation of (**3**) by IBX.

Cyclohexanediol Model Study

In order to better address whether the failed reactions were substrate specific, a model system using 1,2-cyclohexanediol was studied. Two separate reactions were run, one with *trans*-cyclohexanediol (**9**) and the other with *cis*-cyclohexanediol (**10**). In both cases the diol was added to a solution containing 3 equivalents of IBX dissolved in DMSO initially at room temperature and then heated to no greater than 60 ^{o}C (Scheme 2.7). No oxidized or partially oxidized products were observed by GC/MS, only starting materials.



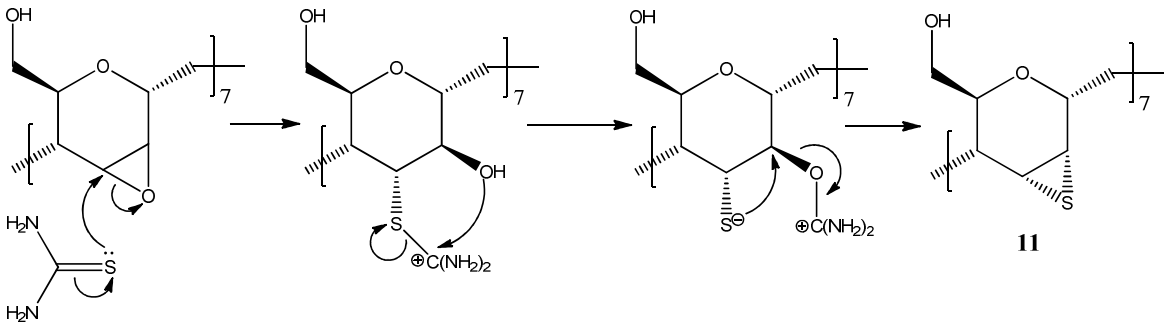
Scheme 2.7. 1,2-cyclohexanediol model system study.

The failure of the diol model system prompted us to investigate the purity of the prepared IBX. It has been reported that current procedures used to produce IBX often result in a reagent of poor quality. Fortuitously, Frigerio and coworkers have found that the use of Oxone ($2KHSO_5$-$KHSO_4$-K_2SO_4) provides a practical method for IBX synthesis. The oxidation is complete in 3 h in water at 70-75 ^{o}C, proceeding in high yield (79-81%) and good purity (≥95%).[49] The reactions were re-examined using IBX prepared from Oxone following the same reaction conditions as before. The reactions were monitored by GC/MS; however, no mass peak or mass fragment corresponding to

the correct product was observed. In both cases a molecular ion peak corresponding to the starting diol was observed at 116 along with isolation of unreacted IBX.

Discussion and Conclusions

It is quite surprising that the simple reaction conditions developed for the preparation of olefin (**5**) by Fujita and coworkers led to the complete conversion of all the epoxides. Not only does this require that the intermediate thiirane (**11**) remain stable to the acidic conditions of the reaction while each epoxide is converted, but also requires the inversion of configuration of each subunit of the cyclodextrin. A plausible reaction mechanism was proposed for this transformation and is depicted in Scheme 2.8.[38] We attempted to reproduce Fujita's methodology for obtaining olefin (**5**) in an effort to get a better understanding of the properties and chemistry involved in the production of the olefin. However, following literature conditions we failed to observe the olefin in the NMR spectrum despite the fact that Fujita and coworkers reported a total yield of 98%. It is was noted that each step of the literature preparation by Fujita et al. required tedious chromatography and was performed on a scale of 30 mg. However, no full paper has since been published on this chemistry.



Scheme 2.8. Plausible reaction mechanism for the formation of thiirane (**11**).[38]

In the McMurry reaction, the reduction of 1,2-diols does not require the pre-formation of the pinacol anions since free diols reduce directly, or presumably the anions are formed *in situ* by reaction with Ti^{o}.[44] Excess potassium was used because it was thought that it would provide initial deprotonation of the acidic hydroxyl protons to form the pinacol dianion intermediate. The failure of the reduction of 6-*O*-TBS-β-CD (**3**) to the corresponding olefin was unexpected due to the apparent ease of formation of this compound by Fujita and coworkers. Though the reaction proceeded through a reactive thiirane intermediate, it suggests that the cyclodextrin itself can adopt the necessary confirmation in order to incorporate the planarity of the double bond. It is been established that titanium forms aggregates of varying shapes and sizes and reactions can occur on the surface of the active titanium in a heterogeneous process.[44] It would be reasonable to conclude, then, that in order for the reduction to take place the deprotonated oxygens must bond to the surface of the titanium which may prove to be difficult for a complex molecule such as cyclodextrin; though, it is not fully understood (Figure 2.2). In addition, the unsuccessful conversion to the olefin could be due to the lack of initial formation of the pinacol intermediate by radical anions.

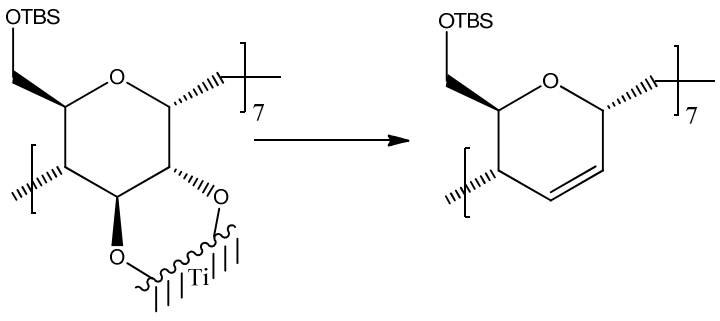


Figure 2.2. Illustration of reaction occurring on the surface of an active titanium particle in a heterogeneous process.

Alternatively, employment of the Garegg reduction for the same transformation proved to be fruitless. It has been suggested that imidazole has the dual function of forming a partially solvated complex with triphenylphosphine and iodine, as well as functioning as a base (Figure 2.3).[45] It was also expected that substitution of a single or both hydroxyl groups by iodine might be a possibility, according to the proposed mechanism, though this behavior was not observed (Figure 2.4). In the NMR spectrum, triphenylphosphine and triphenyphosphine oxide was observed; however, no peaks corresponding to the product were obvious. Either due to the large volume of by-product or the extremely low yield of the olefin, the reaction was deemed unsuccessful.

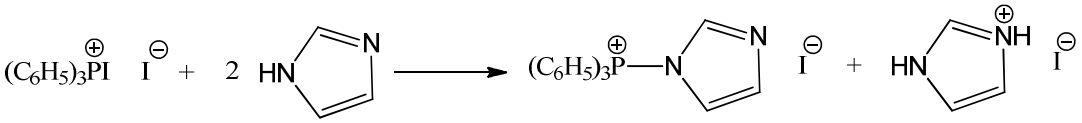


Figure 2.3. Complex formed in the Garegg reduction.

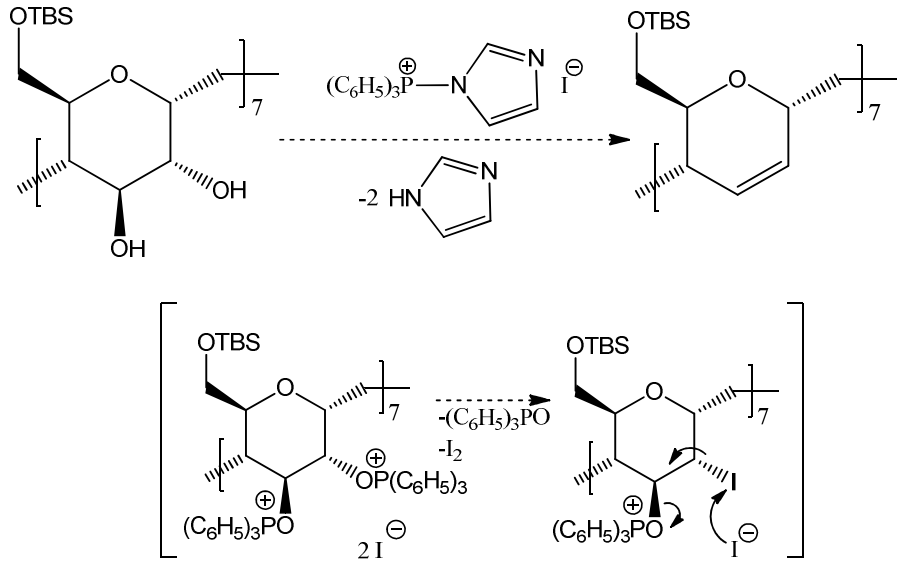


Figure 2.4. Plausible mechanism for Garegg reduction.

As for the oxidations of (**3**), in all cases no conversion to the oxidized or even partially oxidized product was observed. It is postulated that incorporation of two double bonds on to the glucose unit would create a great amount of strain and may explain the lack of reactivity towards oxidation. Simultaneously, efforts to develop a more robust route to modified cyclodextrins were proving to be more successful and will be discussed in greater detail in the following chapters.

CHAPTER THREE

Preparation and Reaction of Cyclomannoepoxides

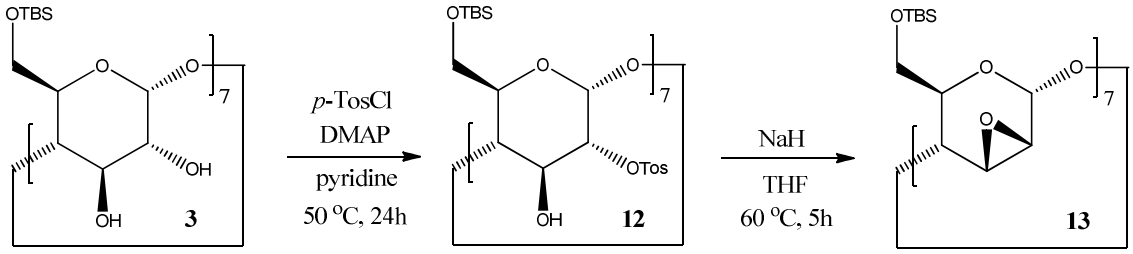
Introduction

Initial attempts at direct functionalization of the secondary face by oxidation or reduction were unsuccessful; however, routes proceeding through an epoxy intermediate have been reported to be successful for a variety of modifications.[50] We believe that per-epoxidation of β-CD could provide a valuable intermediate toward the synthesis of tailored cyclodextrins by simple nucleophilic ring-opening reactions. A variety of functionalizations could be achieved on the secondary face by varying the nucleophiles which would not only extend the chemistry, but also the potential utility for new cyclodextrins in enantioseparations.

Preparation of Cyclomannoepoxides

The general strategy we followed for the synthesis of *manno*-heptakis(2,3-epoxy)-β-CD (**13**) was developed by Coleman and Zhang,[51] which involves the initial protection of the primary hydroxyl groups via per-silylation.[52] Then the deprotonation of the hydroxyl groups at the C(2) position was accomplished using 15 equiv. of sodium hydride or potassium *tert*-butoxide in THF, leading to the corresponding oxygen anions. Subsequent addition of *para*-toluenesulfonyl chloride in the presence of a catalytic amount of DMAP to these anions yielded the known (6-*O*-TBS-2-*O*-tosyl)-β-CD (**12**) in 60% yield. This intermediate was either isolated by column chromatography or immediately converted to the corresponding known epoxide in a single step by use of

excess sodium hydride without isolating the tosylate intermediate (Scheme 3.1).[51] If the tosyl intermediate was purified by column chromatography, the yield of epoxide (**13**) could be improved from 75% yield to over 80% yield. If desired, the deprotection of the TBS protecting groups could be accomplished using boron trifluoride etherate in chloroform at room temperature in less than an hour to give the previously reported hydroxyepoxide in approximately 50% yield.



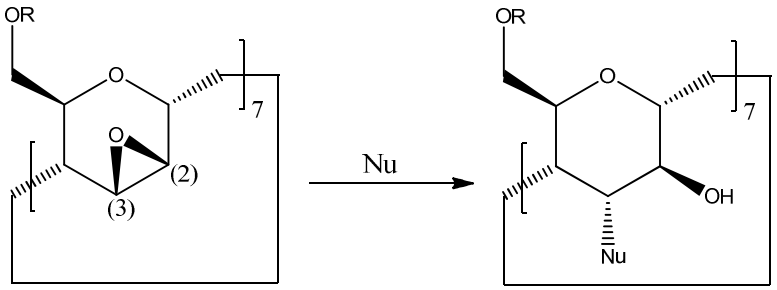
Scheme 3.1. Preparation of 6-*O*-*tert*-butyldimethylsilyl-2,3-*O*-epoxy-β-CD.[51]

Nucleophilic Ring-Opening of Cyclomannoepoxides

The secondary face of cyclodextrin has been shown to be more important in some applications of cyclodextrins than the primary face.[53] This prompted the investigation of functionalization via nucleophilic ring-opening of the CD cyclomannoepoxides by relatively small groups capable of forming an inclusion complex. We anticipated that the optimal substituent would provide an increase in the size and length of the CD cavity as well as alter the polarity, solubility, and thermal stability. The central point behind the ring-opening reactions is that by changing the nucleophile, one could alter the cyclodextrin properties important for application in enantioseparations.

The nucleophilic ring-opening of β-cyclodextrin 2,3-cyclomannoepoxide is known to give cyclodextrins functionalized at the C(3) position, almost exclusively (Scheme 3.2). In some cases the reaction selectivity can be reversed by use of an

appropriate nucleophile that prefers to access the C(2) position of the CD epoxide, as is the case with sodium sulfide.[53] The product distribution of the nucleophilic ring-opening reaction has been attributed to electronic and conformational effects.[54] The removal of a secondary hydroxyl group and inversion of C(2) could increase the hydrophobic nature of the cavity leading to an increase in compatibility between the cavity and some hydrophobic guest molecules.[52]



Scheme 3.2. Nucleophilic ring-opening of β-cyclodextrin cyclomannoepoxides.

The ring-opening of the CD epoxide requires the nucleophile to approach the reaction sites from inside the cavity,[53] therefore bulky groups were avoided in the selection of potential nucleophiles. Reactions of this type have employed small nucleophiles such as hydroxyl amine, ammonia, azide and imidazole type functionalitites,[50] to name a few. Examples of the ring-opening of cyclodextrin epoxides employing nitrile functionalities have not been described in the literature. Nitriles are small, linear, rigid nucleophiles that could alter the polarity and sterics involved in chiral discrimination. Therefore, we have examined the ring-opening of (**13**) with various cyanide reagents. Initial attempts involved the addition of 8 equivalents of solid NaCN dissolved in anhydrous DMSO, to thoroughly dried 6-*O*-TBS-2,3-epoxy-β-CD (**13**) under nitrogen atmosphere. After workup, the reaction resulted in an orange residue that

appeared to have identical NMR chemical shifts from the starting epoxide; this was confirmed by results obtained by MS, which indicated the presence of starting material and no product. Solid NaCN is only sparingly soluble in DMSO; therefore, the reaction was re-examined using DMF and MeCN as solvents. This resulted in an almost identical material with similar NMR behavior, once again indicative of no reaction of the epoxide. For comparison, KCN was used instead of NaCN, but the same results were obtained. After repeated failure, a literature search revealed a number of reagent systems that could be capable of completing the desired transformation. Because it was not obvious as to why the initial ring-opening reactions failed, we subjected cyclohexene oxide to various cyanating reagents in order to deduce the causes of failure.

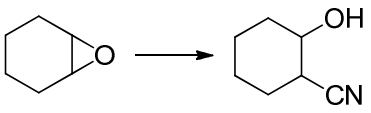
Cyclohexeneoxide Model Study

The results of the reaction of cyclohexene oxide with the various cyanating agents are shown in Table 3.1. In all the reactions studied, the conversion was monitored by GC/MS and NMR.

The epoxy cyclodextrin (**13**) was subjected to the reaction conditions in Table 3.1 including those that were not successful for cyclohexeneoxide. As noted in entry 1, when cyclohexene oxide was reacted under the conditions previously run on (**13**), similarly no product formation was observed. It was expected that the addition of a catalyst could increase the likelihood of obtaining the nitrile product. When (**13**) was reacted with an excess of a 1:1 mixture of $LiClO_4$/KCN in acetonitrile, the reaction resulted in the complete recovery of starting material as determined by the NMR spectrum. Furthermore, in the ^{13}C NMR spectrum a downfield shift corresponding to the nitrile carbon (~119 ppm) was not observed. Reaction of this same epoxide under the

conditions of entry 3 resulted in similar results as those conducted under the conditions of entry 2 where no appreciable amount of product was observed.

Table 3.1. Reaction of cyclohexene oxide with various cyananting agents.



entry	reagents	reaction time (h)	% yield
1	$\xrightarrow[CH_3CN]{NaCN}$	48	0
2	$\xrightarrow[CH_3CN]{LiClO_4/KCN}$	48	90.5
3	$\xrightarrow[CH_3CN]{NH_4Cl/KCN}$	24	90
4	$\xrightarrow[CH_3CN]{Et_4N^+CN}$	48	0
5	$\xrightarrow[CH_3CN]{KCN/18\text{-crown-6}}$	40	85

The ring-opening of (**13**) using KCN in the presence of a catalytic amount of 18-crown-6 resulted in a yellow oil. By NMR, the cyclodextrin epoxide appeared to be un-reacted; however, a large peak was observed overlapping the cyclodextrin protons in the spectrum along with a broadening of the CD multiplets. This could be explained by a 1:1:1 complex forming between the cyclodextrin, guest, and the crown ether. Complexes of this type have been observed.[56] Epoxide (**13**) was reacted with tetraethylammonium cyanide in anhydrous acetonitrile, but no reaction was observed under these conditions either.

We also investigated the addition reaction of Me_3SiCN (TMSCN) to epoxide (**13**). Due to the dual nature of TMSCN, its addition to epoxides can lead to β-hydroxy nitriles

or isonitriles, depending on the catalyst used. Based on HSAB theory, "hard" Lewis acid catalysts, such as $AlCl_3$, selectively lead to nitriles while "soft" Lewis acid catalysts, such as $ZnCl_2$, predominately lead to the isonitriles. Therefore, epoxide (**13**) was dissolved in anhydrous acetonitrile with a catalytic amount of the Lewis acid catalyst and 8 equiv. of TMSCN was added to the mixture which was heated to 60°C. We expected to see conversion of the epoxide to the *O*-trimethylsilyl protected β-hydroxy nitrile; however, no conversions to the nitriles or iso-nitriles were observed in the NMR spectrum. When $ZnCl_2$ was used as the catalyst only starting epoxide was recovered; alternatively, when $AlCl_3$ was employed, deprotection of the TBS protected primary hydroxyl groups was observed.

Results and Discussion

It is known that the 2,3-anhydropyranose unit in cyclodextrins, may adopt one of two half-chair conformations, ${}^{O}H_5$ or ${}^{5}H_O$ (Figure 3.2). These two conformations equilibrate with each other. According to energy minimizations by MM2 modeling, the mannoepoxide adopts the ${}^{O}H_5$ confirmation.[53] In this confirmation, the C(3') hydroxyl group of an adjacent glucosidic unit does not block the rear side of the C(2) carbons from the rear side of the epoxide as it does in the opposite conformation (not shown). In addition, the electronic repulsion between the nucleophile and the endo-O(5) and glucosidic oxygen appears to have a stronger repulsion in the ${}^{5}H_O$ conformation further supporting the preferred attack at C(3).[53]

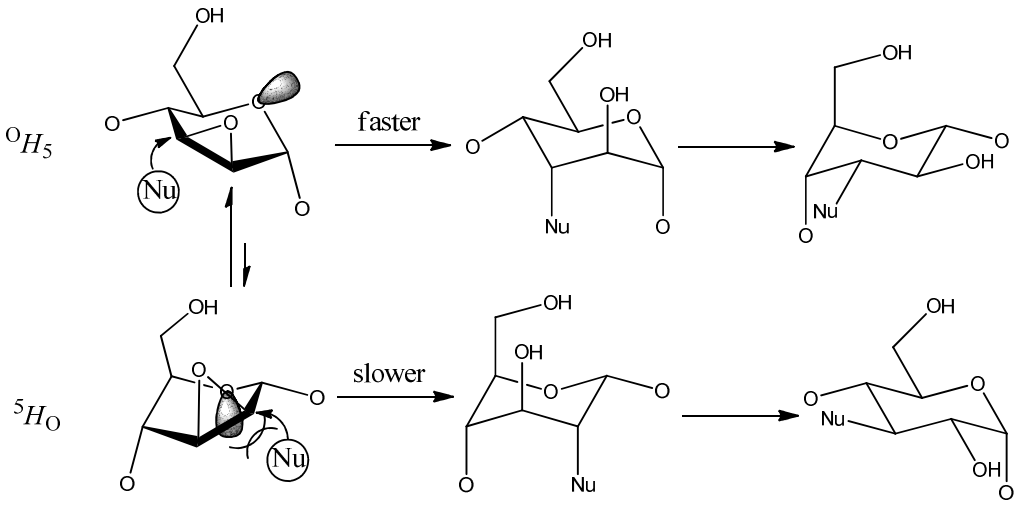


Figure 3.2. Mechanistic consideration of the ring-opening of CD *manno*epoxides.[53]

The epoxide (**13**) is reported to be unstable and to decompose at room temperature as well as under acidic conditions.[52] In our laboratory, the synthesized epoxide showed no appreciable decomposition after sitting for a week at room temperature as monitored by ^{1}H NMR. However, the compound did decompose in the presence of silica, supporting its sensitivity to acidic conditions. The instability of the (2,3-epoxy-6-*O*-*tert*-butyldimethylsilyl)-β-CD has been attributed to the additional strain caused by the bulky tert-butyldimethylsilyl groups on the primary face tilting slightly toward the cavity.[52] Because the epoxy-β-CD is known to react with water, the epoxide was pre-dried under reduced pressure in an Abderhalden drying pistol under refluxing toluene in the presence of P_2O_5. Nonetheless, an average yield of 65-75% of the epoxide (**13**) was suggesting this route might be conveniently used to access a wide variety of functionalized cyclodextrins.

The ring-opening of (**13**) using sodium cyanide in DMSO or DMF resulted in no product formation as discussed earlier. Though these results were initially surprising, subjecting cyclohexene oxide to the same reaction conditions resulted in no ring-opening

product either. Similarly, Macchia and coworkers reported that no ring-opened product was obtained when cyclohexene oxide was reacted with 2.2 equiv. of KCN in refluxing acetonitrile.[57] As show in Table 3.1, in all cases where the desired product was obtained an acidic medium or catalyst was added. $LiClO_4$ turned out to be more effective as a catalyst under the reaction conditions than the classical aqueous acidic conditions and due to the sensitive nature of the epoxide, acidic conditions were avoided. The metal salt catalyst most likely aides in the ring-opening reaction by coordinating with the epoxide oxygen. It should be mentioned that the reaction of cyclohexene oxide with tetraethylammonium cyanide (entry 4) was monitored by ^{1}H NMR. The reactants were added directly to an NMR tube and monitored over a two week period in both dimethyl sulfoxide-d_6 and acetonitrile-d_3 under sonication with the temperature maintained at 55 ^oC. However, no noticeable amount of product was observed. The straightforward routes toward β-hydroxy nitriles discussed in this chapter, were satisfactory only when the simple cyclohexene oxide was employed. When more complex epoxides are used, the literature documents that only low yields of the β-hydroxy nitriles were obtained even with forced reaction conditions.[57a] Therefore it is not completely surprising that reaction of a more complex epoxide such as (**13**) was completely ineffective under the proposed methodologies.

Interestingly, it was thought that direct nucleophilic displacement of the tosylate intermediate (**12**) could also provide us with the cyano-functionalized CD. The reaction was attempted using a slight excess of potassium cyanide dissolved in acetonitrile heated to 60 ^oC. The reaction failed to give any product, indicated by the presence of an intact tosyl group by ^{1}H NMR. The reaction was re-examined using methylene chloride as the

solvent to aid in better dissolution of the cyanide salt; however, this reaction also failed to produce the cyanide product.

CHAPTER FOUR

Preparation of Novel β-Cyclodextrin Derivatives

Modification of the C(6)-Position

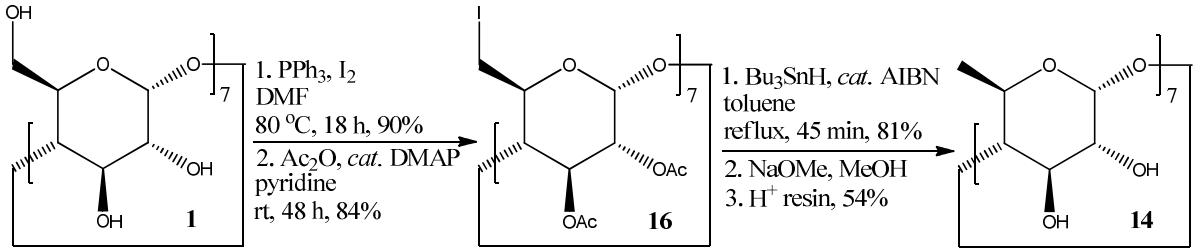
Generally, the C(6) carbon on the primary face is substituted with non-polar groups such as alkyl or silyl groups as introduced in Chapter Two. These substitutions are performed to prevent competition in the alkylation of the C(2) and C(3) hydroxyl groups, and to increase the solubility, whereas substituents on the secondary face, such as alkyl and acyl groups, are, typically introduced to alter the selectivity of the cyclodextrin. However, it has sometimes been argued that varying the size of the substituent at the C(6) position can also effect the selectivity due to the different shapes created by the various substituents.[58] For example, Kobor et al. found that the shape differences of the (6-*O*-methoxy-2,3-dimethoxy)- and the (6-*O*-TBS-2,3-dimethoxy)-β-CD could result in differences in their enantioselectivities.[59] Similarly, Vigh and coworkers reported observing different enantioselectivities when varying the C(6)-*O*-substituents in cyclodextrin-based chiral selectors. They agreed with Kobor, that the shape differences between the large and small groups at C(6) effected the selectivity, but added, that large non-polar groups at the C(6)-position were more advantageous than smaller groups only due to increased solubility.[58] While the smaller groups provide increased access to the smaller end of the cavity, the polarity, generally also increased. With these observations in mind, we have primarily focused the synthesis of new derivatives on those bearing a large TBS group or a small methyl group at the C(6) position in order to explore the effect, if any, of the size and shape difference on the selectivity.

By Deoxygenation

As modified cyclodextrins, per(6-deoxy)-β-CDs are of interest for several reasons. First, there is no group at the C(6)-position that can complicate synthetic efforts at the C(2) and C(3) positions. Second, deoxygenation at C(6) would be expected to change the polarity and perhaps inclusion complex-forming characteristics. The placement of a small group at the primary face might potentially create a larger opening and increased access to the smaller end of the cavity.

Using the improved preparation by Baer et al., the known (6-deoxy) compound (**14**) was obtained in four steps in 28% overall yield (Scheme 4.1).[60] This involved the initial halogenation of the C(6)-position by the addition of β-CD (**1**) to a mixture of iodine with PPh_3 in DMF at 80 ^{o}C for 18h to give the known (6-deoxy-6-iodo)-β-CD (**15**) in 90% yield. Compound (**15**) was then conventionally acetylated over 48 h to give the known diacetylated derivative (**16**) (84%). The reduction of (**16**) was accomplished using tributyltin hydride with catalytic AIBN in refluxing toluene. The reduction was complete in 45 min, and after purification by column chromatography, yielding 75-81% of known (6-deoxy-2,3-diacetoxy)-β-CD (**17**). This compound could then be diacetylated to give the desired (6-deoxy)-β-CD (**14**). In attempt to maximize yield and efficiency, a modified preparation for the deacetylation was developed that did not require exposure to water and the subsequent difficulty in extraction of the product in pure form. A solution of (**17**) was dissolved in dry methanol and was made alkaline with ~4M NaOMe in methanol (pH 8-9) to afford the poly-sodium salt of (**14**). After distillation of the methanol (which ensures complete deacetylation), the polyanion was protonated by passing a solution of the salt in methanol through Dowex 50-W 8x (H^+) cation-exchange

resin, and evaporated to give the sodium-free (6-deoxy)-β-CD (**14**) in moderate yield (54%). This derivative was soluble in H_2O, methanol and DMSO, but not in ethyl acetate, methylene chloride, or hexanes.

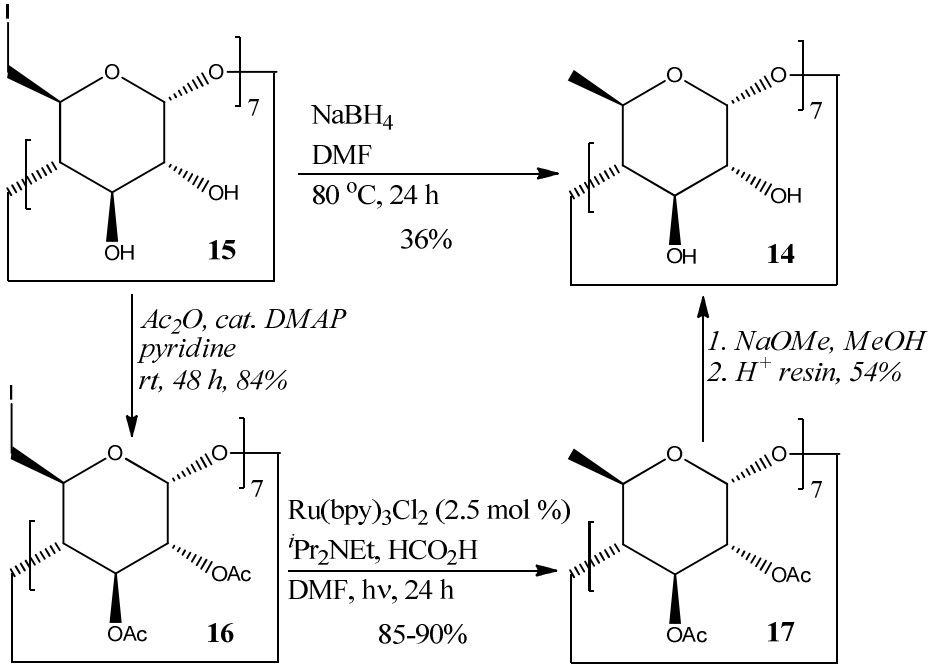


Scheme. 4.1. Synthesis of (6-deoxy)-β-CD (**14**) from (**1**).[60]

Finding an alternate route to access the 6-deoxy product while avoiding the toxicity, odor, and purification issues associated with tin hydrides,[62a,b] would be a desirable advantage. The direct reductive dehalogenation of (6-iodo)-β-CD (**16**) could be accomplished as previously described[61] by treatment with $NaBH_4$ in DMF; however, practical difficulties were encountered attempting this methodology. The 6-deoxy derivative was very soluble in water thus, it was necessary to aqueous workup specified in borohydride route. Also, attempts to purify the compound by column chromatography were time-consuming and unsuccessful, presumably due to the troublesome borate salt produced in the reaction. To overcome this problem, the salt was passed through acid resin and approximately 36% of impure (**14**) was isolated as a white solid.

A timely communication by Stephenson and coworkers provided an elegant tin-free reductive dehalogenation reaction.[63] Following their conditions, the 6-iodo derivative (**16**) was exposed to a 14W fluorescent lamp with $Ru(bpy)_3Cl_2$ (2.5 mol %) as a catalyst in the presence of 10 equiv. of $^{i}Pr_2NEt$ and 10 equiv. of formic acid in DMF for 24 h (Scheme 4.2). The desired product (**17**) was obtained after purification by column

chromatography in good yield (85-90%). This new methodology represents an elegant, tin-free method for the preparation of (6-deoxy-2,3-diacetoxy)-β-CD (**17**), and an improved route to the final (6-deoxy)-CD (**14**), as well.



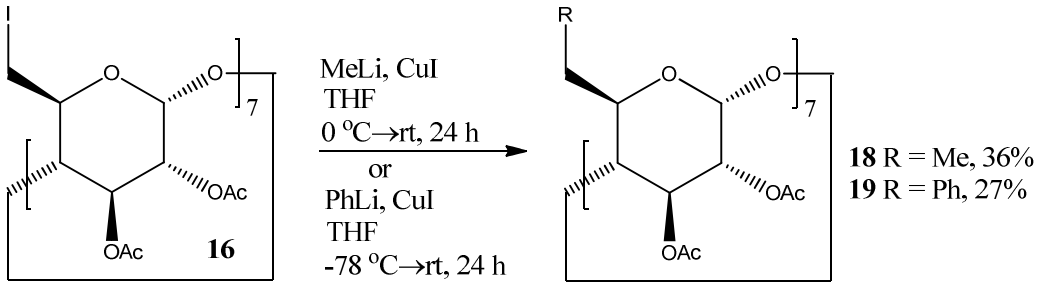
Scheme 4.2. Dehalogenation routes used to access (6-deoxy)-β-CD (**14**).

By Organocuprates

The synthesis of (6-deoxy)-β-CD also prompted an investigation into the synthesis of 6-deoxy compounds bearing alkyl or phenyl groups. Mentioned earlier, large non-polar groups at the C(6)-position would have the main advantage of increasing the solubility, a common problem we encountered working with these relatively polar (6-deoxy) derivatives. Also, information might be gained about how the shape and selectivity of the modified cyclodextrin changes as the size of the group changes. In the process of preparing (**14**), we realized that organocuprates might effectively be used for the alkylation and arylation of the previously prepared (6-iodo-2,3-diacetoxy)-β-CD (**16**).

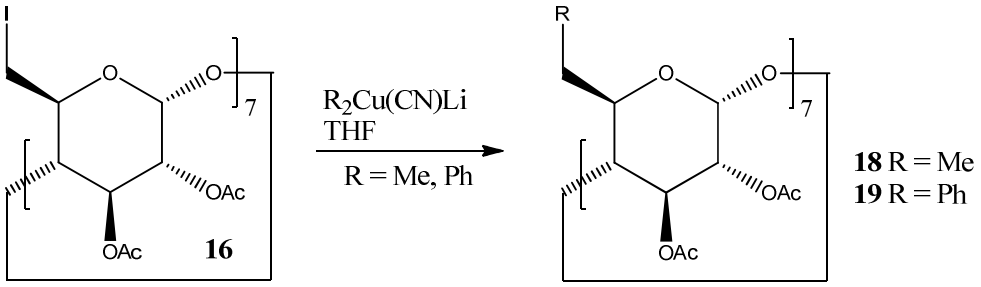
Cuprates react well with primary iodides are stable toward ester functionalities, making (**16**) an attractive substrate. To our knowledge, the reaction of CDs with cuprates has not been studied and no examples could be found in the literature.

Therefore, initial attempts to alkylate/arylate (**16**) were based on the general protocol typically employed in organocopper reactions.[64] The addition of Me_2CuLi to a solution of (**16**) in THF at 0 ^oC provided the alkylated product (6-deoxy-6-methyl-2,3-diacetoxy)-β-CD (**18**) in 36% yield after column chromatography. Similarly, addition of Ph_2CuLi resulted in the arylated derivative (**19**), but in a lower yield (27%) (Scheme 4.3).



Scheme 4.3. Alkylation and arylation of (**16**) with organocuprates.

Addition of $Ph_2Cu(CN)Li$ gave similar yields of the alkylated product and slightly better yields of the arylated product. (33%, Scheme 4.4). In contrast to CuI, the CuCN is not hygroscopic or light sensitive and does not require purification before use, making this the preferred reagent. The alkylated and arylated products were confirmed by ^1H , ^{13}C NMR and ESI-MS. After removal of the copper species by aqueous workup, both products could be easily purified by column chromatography. Derivatives (**18**) and (**19**) could (in theory) be deacetylated under the conditions described above to yield the corresponding hydroxyl compounds; however, we have not yet carried these materials on to new derivatives.



Scheme 4.4. Alkylation and arylation of (**16**) employing CuCN.

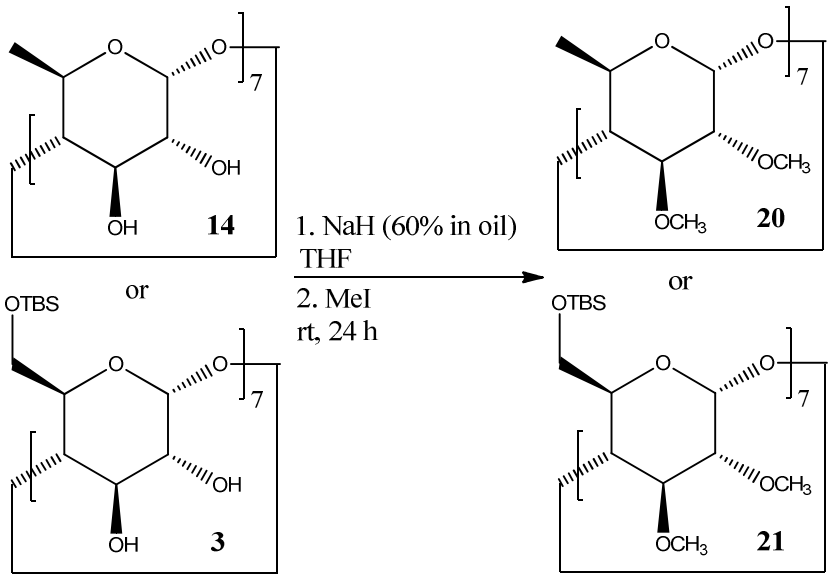
Involving Separate Reaction of the C(2) and C(3) Hydroxyl Groups

Since the ultimate goal of this research is the synthesis of new cyclodextrin derivatives for use as chiral selectors, the modifications described in the following sections were performed on the previously described (6-*O*-TBS-2,3-dihydroxyl)-β-CD (**3**) and (6-deoxy-2,3-dihydroxy)-β-CD (**14**). These two compounds, having similar reactivity, but different shapes, represent the building blocks for subsequent reaction of the secondary hydroxyl groups and the development of new chiral selectors. Furthermore, the 6-deoxy derivatives will represent the first of its type to be synthesized and employed in CSPs, providing a very different functionality than the most commonly used 6-OTBS group.

By Alkylation

The synthesis of the dimethoxy derivative of (**14**) would provide a promising start because this derivative could be used to make direct comparisons to stationary phases that are currently commercially available such as J&W's CycloSil B, which contains a dimethoxy functionality. To complete the synthesis, compound (**14**) and 20 equiv. of NaH were reacted in THF under inert nitrogen atmosphere to which 15 equiv. of MeI was added and reacted for 24 h. The reaction is most likely completed within 5 h; however,

to ensure complete conversion it was allowed to stir overnight. The product could be purified by column chromatography in 100% EtOAc to give (6-deoxy-2,3-dimethoxy)-β-CD (**20**) in 76% yield (Scheme 4.5). This methodology can also be applied to the 6-*O*-TBS derivative (**3**) to obtain the known (6-*O*-TBS-2,3-dimethoxy)-β-CD (**21**) in similar yield.



Scheme 4.5. Synthesis of (6-deoxy)- and (6-*O*-TBS-2,3-dimethoxy)-β-CD.

We have also prepared the methyl-deuterated version of (**21**) in anticipation that this derivative might have slightly increased selectivity and possibly increased thermal stability due to the increased strength of the carbon-deuterium bond. This methyl-deuterated derivative was accomplished by the addition of CD_3I instead of CH_3I when following the above methodology to give (**22**) in good yield (78%) and purity (Figure 4.1).

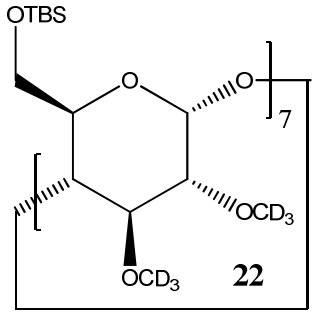
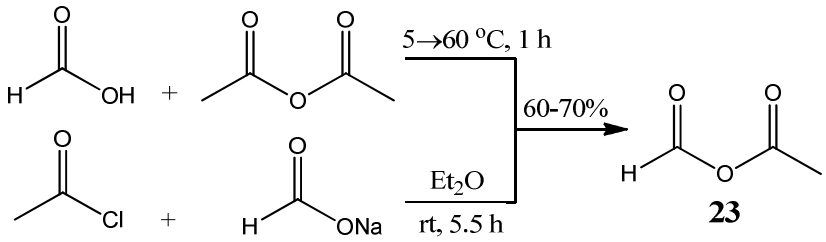


Figure 4.1. Structure of (6-*O*-TBS-2,3-bis-($^2H_3$)methoxy)-β-CD (**22**).

By Formylation

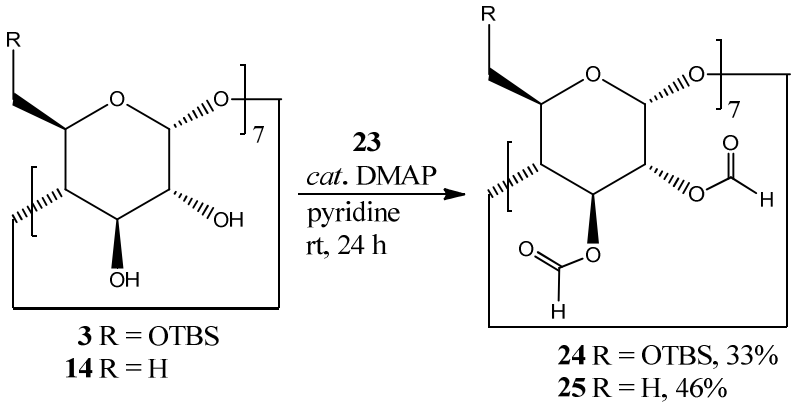
As mentioned earlier, acetyl groups are often used for the functionalization of the secondary face due to their ability to influence selectivity. Conceivably, a formyl group in the same position could have a similar or improved effect on the selectivity. The per(2,3-di-formyl) derivative has not been previously reported for any CDs. The formylation of (**3**) and (**14**) was achieved using acetic-formic anhydride which can be prepared in two ways following literature preparations[62a,b] (Scheme 4.6).



Scheme 4.6. Preparation of formic-acetic anhydride (**23**).[62a,b]

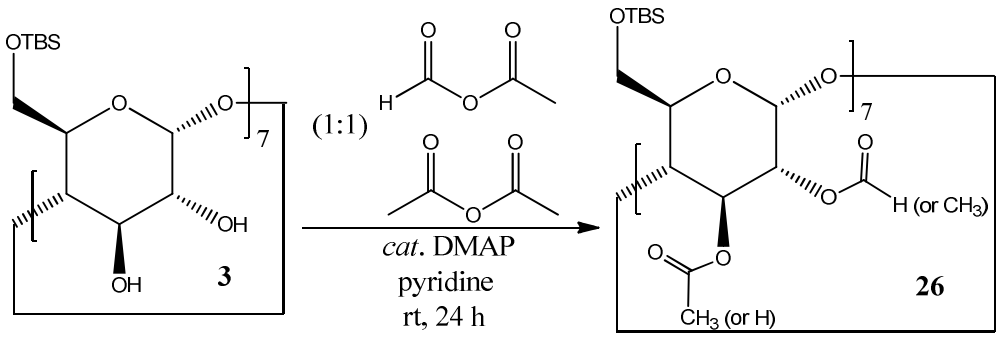
The formylation reactions were carried out in pyridine in the presence of a catalytic amount of DMAP to give (6-*O*-TBS-2,3-di-*O*-formyl)- and (6-deoxy-2,3-di-*O*-formyl)-β-CD, (**24**) and (**25**), respectively (Scheme 4.7). Column chromatography was used for the

purification of the compounds; however, the separations were preformed as quickly as possible to avoid potential hydrolysis of the formyl groups, though this was not apparent.



Scheme 4.7. Formylation of (6-*O*-TBS)- and (6-deoxy)-β-CD (**3**) and (**14**), respectively.

We also postulated the possibility of creating mixed-CD stationary phases containing a random distribution of two different functional groups. To accomplish this transformation, formic-acetic anhydride (**23**) and acetic anhydride were added dropwise in a 1:1 ratio to (6-*O*-TBS)-CD (**3**) in pyridine with *cat*. DMAP. Enough equivalents of the anhydrides were added to provide a random distribution of acetyl and formyl groups, while avoiding the possibility for per-acetylation or per-formylation (Scheme 4.8). Starting on a 540 mg scale, the reaction provided ~200 mg of the desired mixed-derivative (**26**) as a light yellow solid. The solid was quickly purified by column chromatography to separate only the polar and nonpolar compounds visualized by TLC, resulting in a mixed-phase containing a random distribution of approximately 45% formyl groups and 55% acetyl groups as indicated by integration of the NMR spectra.



Scheme 4.8. Preparation of a mixed-β-CD (**26**).

Involving Annulations Across the C(2) and C(3) Hydroxyl Groups

A primary goal of this work was the synthesis of cyclodextrin derivatives annulated to bridge the C(2) and C(3) hydroxyl groups. Derivatives of this type have not been described in literature for CDs and as such have never been employed as chiral selectors for chiral gas chromatography. It was hypothesized that these unique derivatives may exhibit an extended more organized cavity. It was envisioned that annulation could be accomplished by the reaction of compounds (**3**) and (**14**) with bis-electrophiles in the presence of a strong base. By varying the size and functionality of the electrophile a variety of annulated derivatives might be obtained.

Potential Electrophiles for Annulation Reactions

The goal with the selection of potential electrophiles is to pre-organize and extend the cavity length, width, and polarity. A number of potential electrophiles were selected that could generate five-, six-, and seven-membered rings when bridged across the secondary hydroxyl groups (Figure 4.2). The electrophiles were all purchased from commercial sources with the exception of bis(chloromethyl) ether (**37**), which was prepared according to a literature procedure.[65]

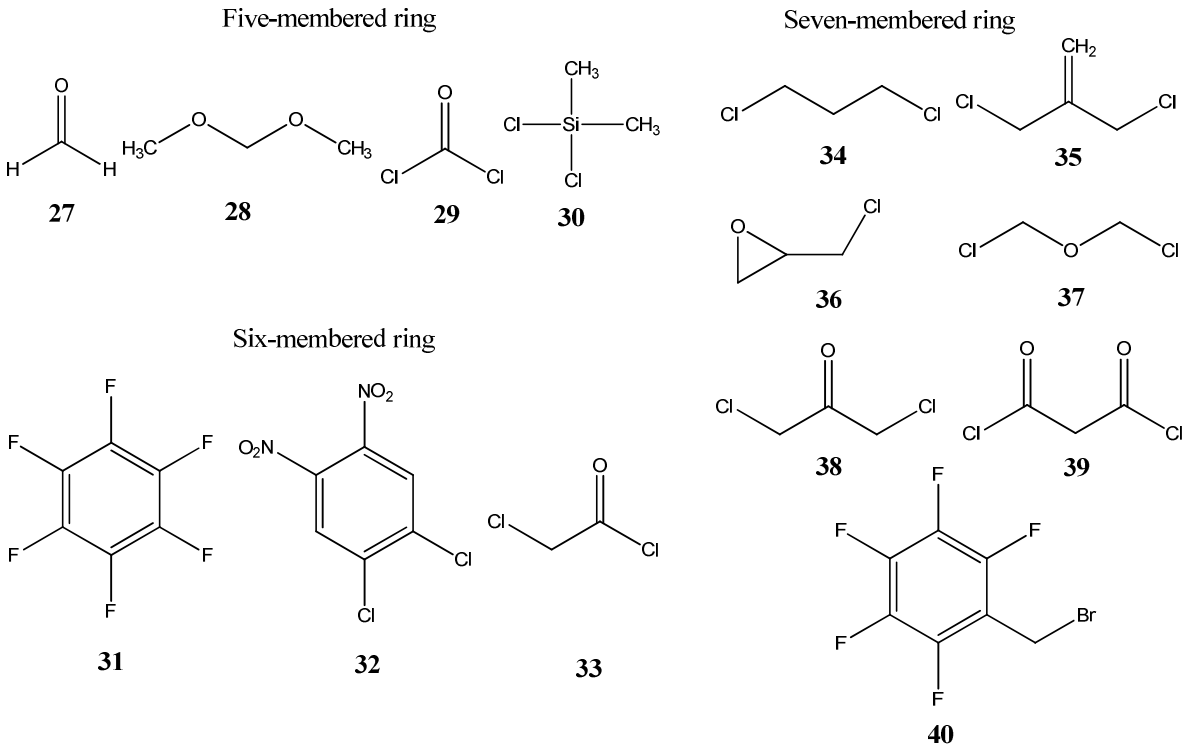


Figure 4.2. Potential electrophiles categorized by the size of the cyclized product after annulation.

Base and Solvent Considerations

The selection of base, solvent, and addition rate could play an important role in the success of these electrophilic reactions. For the majority of these, we have used NaH as a base in THF or DMF, with the exception of the acyl chloride electrophiles (**29**, **30**, **33**, **38**, and, **39**) in which pyridine was employed as the base. In some cases catalytic in DMAP was employed. While the use of NaH has been successfully employed, the workups have been problematic following reactions with electrophiles due to the sodium salts formed with use of this base. Often, aqueous workup was avoided due to the undetermined polarity of the products, requiring the reaction mixtures to be immediately subjected to chromatography. Without the initial extraction of the products and removal of salts, purification by chromatography was time-consuming and unrewarding. It was

thought that the use of a liquid superbase would ensure the deprotonation of the CDs in a homogeneous reaction, avoid the exposure to air that NaH required, allow the convenient slow addition of the base by syringe pump, and possibly yield more nucleophilic anions.

Superbases are strong, cation-free neutral phosphazene bases, which allow the deprotonation of a wide range of weak acids, to form highly reactive "naked" anions.[66] We were especially interested in these phosphazene bases because they can be used in nonpolar organic solvents, are reported to provide cleaner reactions and easier workups compared to other nitrogen bases, and have a low sensitivity to oxygen and moisture. The phosphazene bases are categorized by P_1-P_4 (Figure 4.2) or higher depending on the number of phosphorous atoms in the base. The bulkier the phosphazene base, the more "naked" the anion is for subsequent reaction. The pK_{BH} of phosphazene bases, P_1-P_4, range from 27-43 (acetonitrile). The basicity increases with increasing electron withdrawing character and bulkiness of the substituents.[66] Figure 4.3 shows a select few of these organic superbases including the commercially available phosphazene base (**41**) selected for use in this research. This particular phosphazene base, P_1-*t*-Bu-tris(tetramethylene) (**41**) was chosen because it is a liquid form and less expensive than other phosphazene bases.

1,2-Cyclohexanediol Model Study

In order to determine the optimal reaction conditions, *trans*-cyclohexanediol was chosen as a model system. To compare the efficiency of the phosphazene base (**41**) with that of NaH, in different solvents (THF, DMF), 2.2 equivalents of each base was reacted with the diol followed by addition of 1.1 equiv. of the electrophile, C_6F_6 (**31**) (Scheme 4.9). For those reactions where the phosphazene base was employed, the rate of addition

of the base and electrophile was controlled via a digital syringe pump set to add the reagents over an 8 h period. We found that the slow addition of this electrophile was important for obtaining cyclization and to minimize the amount of mono- and di-substituted products (Figure 4.4).

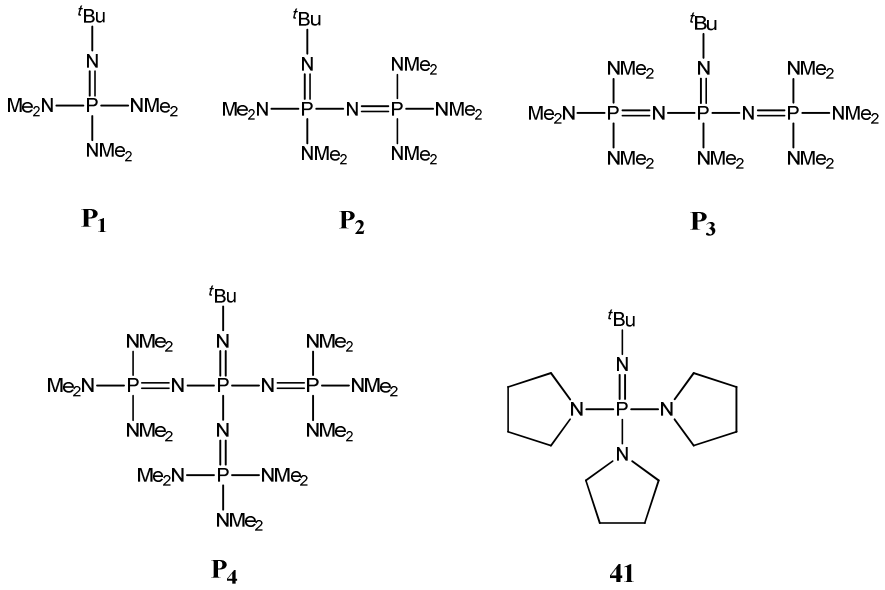
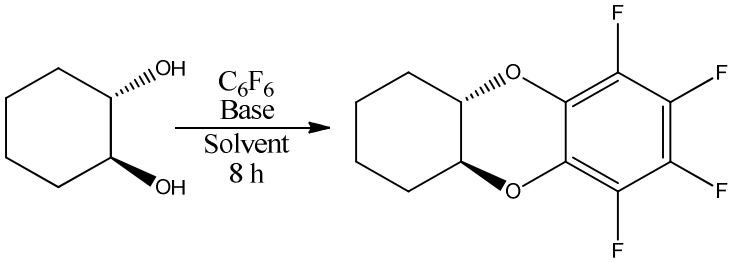


Figure 4.3. Select phosphazene bases.



Scheme 4.9. Annulation of *trans*-cyclohexanediol with hexafluorobenzene.

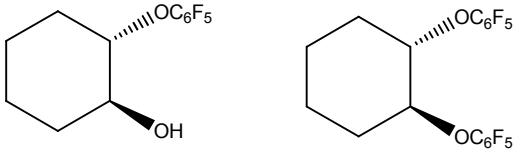


Figure 4.4. Possible mono- and di-substituted products.

For those reactions where NaH was employed, the base was added at the onset of the reaction and the electrophile was added over an 8 h period. The effectiveness of the phosphazene base as compared to NaH, and the ideal solvent was determined by comparing the results of the reactions as monitored by GC/MS. The results suggested that a combination of NaH in DMF, and the addition of the electrophile via slow addition were optimal conditions providing the cyclized product in approximately 93% yield. The other conditions did provide the desired product, but in much reduced yields (relative), for example, 6% for NaH/THF and 3% for phosphazene base (**41**)/THF.

Preparation of Annulated Derivatives

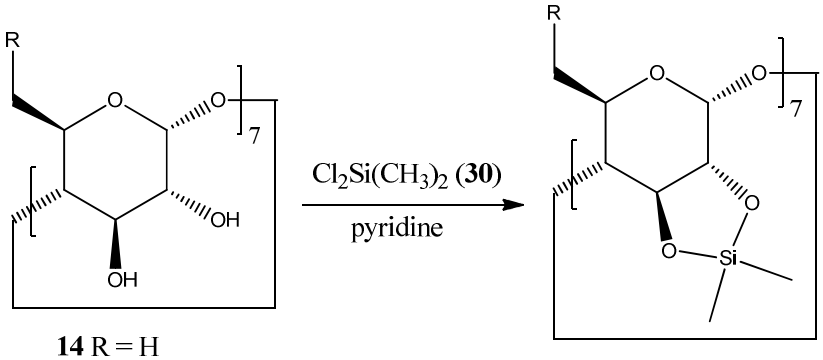
The preparation of annulated derivatives might be accomplished in the same manner as for C_6F_6 by reaction of the (6-deoxy)-β-CD (**14**) with the other bis-electrophiles (Table 4.1) in the presence of either NaH or the phosphazene base (**41**). This was tested using electrophiles (**27-40**) under the reaction conditions (NaH/DMF/8 h) with the exception of those where pyridine was employed. To verify our conclusions that NaH is a superior base, a few reactions were also examined using the phosphazene base. If the reaction resulted in successful isolation of the desired product, the reaction was also examined using (6-*O*-TBS)-β-CD (**3**) under the same reaction conditions.

In general, we were typically able to get reasonable yields with the *trans*-cyclohexanediol model system, but unable to accomplish the poly-annulation using cyclodextrins. The majority of attempts to annulate (6-deoxy)-β-CD (**14**) with the selected electrophiles were unsuccessful. The reaction of (**14**) with phosgene (**29**), hexafluorobenzene (**31**), 3-chloro-2chloromethyl-1-propene (**35**), and pentafluorobenzyl bromide (**40**) resulted in complex mixtures which the products could not be verified by

NMR, nor be isolated by column chromatography. The ^{1}H and ^{13}C NMR of these particular electrophiles could be interpreted as confirmation of a successful reaction; however, the results could not be confirmed by ESI-MS. For example, when phosgene was used as the electrophile, the ^{13}C NMR spectrum indicated the presence of a carbonyl peak around 150 ppm; however, due to the complex product mixture the desired derivative could not be isolated. Similar results were observed for the reaction of 3-chloro-2-methyl-1-propene with (**14**), resulting in a complex mixture that was difficult to analyze. The ^{1}H NMR spectra displayed downfield peaks possibly representative of the vinyl hydrogens at 5.49 and 5.29 ppm. A peak was also observed at 3.85 ppm that could be indicative of the CH_2 groups deshielded by the vinyl hydrogens. In the case of the fluoro-benzene electrophiles (**31**) and (**40**), the product mixtures were very difficult to analyze. Both NMR spectra had severe peak broadening making it impossible to assign any protons on the CD ring. We attempted ESI-MS on these derivatives as well, though; no molecular ion peak or fragmentation pattern was supportive of the formation of the desired products, requiring perhaps further study of these derivatives. Because it was unknown as to whether these molecules would even ionize in the MS, we attempted several methods to increase the chances of ionization by ESI MS. For CDs containing free hydroxyl groups, the CDs might be successfully ionized by dissolving them in a solution of methanol containing 10% formic acid; however, derivatized CDs could not be ionized by this preparation. Additionally, samples of the annulated derivatives in question were sent to UC Riverside and subjected to MALDI MS. We were hoping that the technique MALDI, which is often successful for proteins, sugars, and large

macromolecules, would be successful where ESI MS failed; however, this analysis also failed to provide indication of successful annulation for any of the annulations.

However, when (6-deoxy)-CD (**14**) was reacted with dichlorodimethylsilane (**30**) in pyridine the previously unknown annulated derivative (**42**) was obtained as an impure solid (Scheme 4.10) It was also observed that the same results could be obtained whether the electrophile was added by slow addition or added to the reaction all at once. The addition of heat (50 ^{o}C) to the reaction increased the yield slightly. The (6-deoxy-2,3-*O*-cyclodimethylsilyl)-β-CD (**42**) was confirmed by NMR and MS. ^{1}H NMR showed two resonances upfield for the methyl protons adjacent to the silyl group at (0.14 and 0.15 ppm). Analysis of (**42**) by MS, revealed that the molecule breaks apart one silyl group at a time indicated by the decrease in ~60 mass units for each silyl group as it is removed until the starting compound (**14**) remains (m/z = 1023). Additionally, reaction with dichlorodiphenylsilane gives the corresponding phenyl derivative (previously synthesized in our group), as expected. Compound (**42**) is soluble in methylene chloride and could be purified by column chromatography in the presence of 1% Et_3N. However, even with the added triethylamine, a significant amount of product was lost during column chromatography, presumably due to the cleaving of the silyl group by the acidic silica gel, resulting in only 50% yield. Similar results were obtained with the TBS-protected compound (**3**) leading to the corresponding annulated derivative (**43**) (Scheme 4.10), which was previously synthesized by Tiffany Turner-Hayden, a member of our group, following a similar procedure.



14 R = H
3 R = OTBS

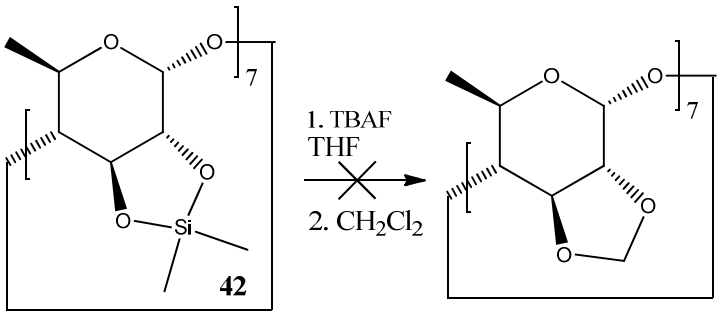
42 R = H
43 R = OTBS

Scheme 4.10. Annulation of (6-deoxy)-β-CD (**14**).

Because it was unknown whether the annulated group would block access to the cavity when employing β-CD, we synthesized the gamma-analogue (**BU 4**) of the annulated derivative (**14**). Due to the larger size of γ-CD, we hypothesized that the blockage by the bridging groups would be greatly diminished (if occurring in the beta-analogue).

With the newly synthesized compound (**42**) in hand, we decided to examine the possibility that highly nucleophilic anions could be generated by treatment with anhydrous fluoride sources.[67] Ideally, reaction of (**42**) in the presence of anhydrous tetrabutylammonium fluoride (TBAF) in THF would create an alkoxide nucleophile that would subsequently react with anhydrous methylene chloride to provide an annulated derivative; however, attempts to annulate by this route failed to produce the desired product (Scheme 4.11). Interestingly, the TBAF failed to desilylate the (6-deoxy)-dimethylsilyl group on the starting derivative, which it is well known to do. This conclusion was drawn based on observation of the intact dimethylsilyl group in the NMR spectrum. Furthermore, methylene chloride is not the ideal electrophile; therefore, future

experiments should employ more reactive electrophiles, as well as ensuring the condition of the anhydrous TBAF.



Scheme 4.11. Attempt at reaction of (**42**) with anhydrous fluoride salts.

With the preparation of the above described new cyclodextrin derivatives, their testing as chiral stationary phases was made possible by an agreement with Agilent Technologies, Inc., to prepare coated capillary columns. The performance of each stationary phase could then be tested chromatographically to determine their ability to perform enantioseparations. These results will be discussed in greater detail in the next chapter.

Results and Discussion

An improved route to the previously reported[31] (6-deoxy)-β-CD (**14**) has been established. This route circumvents the toxicity and purification problems encountered with tin hydrides while providing high yields and easier purification. The reaction involves a catalytic photoredox reduction using a combination of $Ru(bpy)_3Cl_2$, an amine, and a hydrogen source.[63] The photoredox light source can be purchased for less than $5 from any local hardware store, providing a cost effective and efficient synthesis of

previously reported (6-deoxy-2,3-diacetoxy)-β-CD. The product obtained by this route showed identical NMR values to those reported in literature.

Additionally, a route for the efficient synthesis of alkylated and arylated cyclodextrins by organocuprates has been accomplished. This reaction has been successfully applied for the synthesis of (6-methyl-2,3-diacetoxy)- and (6-phenyl-2,3-diacetoxy)-β-CD from (6-iodo-2,3-diacetoxy)-β-CD (**17**),with methyl and phenyl cuprates. Though, the reactions were successful the reaction conditions and yields (27-36%) were not optimized. The products were confirmed by NMR and MS. The 6-methyl showed seven-fold symmetry in the ^{13}C NMR with definitive chemical shifts which was further supported by the molecular ion peak (plus Na^+) at 1731 by ESI MS. The 6-phenyl derivative; however, exhibited significant broadening in both NMR spectrums, though the chemical shifts are suggestive of a successful reaction (i.e., peaks around 7.3 for the phenyl substituent by ^1H NMR and the lack of the iodine substituted carbon by ^{13}C NMR). By ESI-MS, a molecular ion peak at 2167 (Na^+) was observed; however, the product could not be isolated in its pure form. While there was some indication of the presence of phenyl groups and seven-fold symmetry, the splitting patterns could not be accurately assigned. It was thought that either a guest occupying the cavity could cause the observed broadening or the phenyl groups were exhibiting dynamic behavior not observable in the NMR time-scale. In attempt to remedy this, the compound was dried overnight in an Abderhalden drying pistol under reduced pressure with refluxing toluene. If a guest, it could be driven out by subjection to the drying pistol. However, drying the compound did not improve the broadening observed in the NMR spectrum. Consequently, we performed a quick variable temperature (VT) NMR

experiment (-20 ^{o}C – 130 ^{o}C) to hopefully reach a fast average over all confirmations that would help give a sharp spectrum, but no peak changes were observed. Several NMR solvents, including $CDCl_3$, DMSO-d_6, and DMF-d_6 were studied by VT NMR (not to exceed their boiling points), but these attempts also failed to sharpen the signals. A more extensive VT experiment could be necessary over a wider range of temperatures. It was not completely surprising that the arylation reaction resulted in lower yields compared to the alkylation reaction. We anticipated that the lower reaction temperatures required for formation of the phenyl cuprate may result in lower yields due to the slower rate of reaction with the (6-iodo)-derivative. The isolation of biphenyl in the reaction is also indicative of a lower yielding reaction. Though optimization and further study is merited, this reaction represents the first successful utilization of organocuprates for the alkylation/arylation of CDs.

Aside from the above described derivatives, this work also resulted in an additional seven new cyclodextrin derivatives. The methoxy derivative (**20**) was accomplished by simple alkylation with iodomethane of the starting (6-deoxy)-CD. The (6-*O*-TBS-2,3-bis-($^{2}H_3$)methoxy)-β-CD derivative (**23**) was prepared on the basis that the deuterium may offer a change in the thermal stability of the chiral stationary phase and a change in selectivity due to their smaller size. The methyl-deuterated version of (**20**) was easily accomplished by alkylation with iodomethane-d_3. The deuterated compound was confirmed by ^{1}H and ^{13}C NMR, indicated by the lack of methoxy hydrogens that typically appear around 3.2 ppm for these compounds. The product was also confirmed by the appearance of two singlets (2.92 and 3.06 ppm) by ^{2}D NMR corresponding to the slightly different deuterated methoxy groups. The formylation of (6-deoxy)-β-CD (**14**)

and (6-O-TBS)-β-CD (**3**) by formic-acetic anhydride, proceeds smoothly to give the formyl derivatives (**25**) and (**26**) indicated by the evidence of two peaks at ~7.98 ppm and ~8.09 ppm corresponding to the formyl protons at O(2) and O(3). When a (1:1) mixture of formic-acetic anhydride and acetic anhydride is employed, mixed-derivatives can be obtained, in this case with a random distribution of 55% acetyl- and 45 % formyl-functionality. The ^{1}H NMR of the mixed-derivative confirmed the presence of formyl protons (7.98 and 8.09 ppm) and acetyl protons (2.07 ppm); however, the peaks widths were unusually large. Attempts to sharpen the peaks by VT NMR were unsuccessful. The mixed-derivative was purified by column chromatography whereby the all the formyl/acetyl derivatives were kept separated from partially substituted derivatives and unreacted CDs. Hydrolysis did not seem to be a concern during purification of this derivative.

The annulation reactions proved only to be successful when a five-membered ring incorporating a silyl group was employed. Attempts to generate five-membered rings incorporating carbon atoms were either unsuccessful or inconclusive. It could be reasoned that electrophiles containing more than one carbon do not lead to successful cyclization based on these initial attempts. It is possible that cyclization may be occurring across adjacent glucose units and in the case of electrophiles containing at least three carbons, annulations across the cavity of the cyclodextrin could be possible. Without the aid of analytical tools, such as HPLC and MALDI MS, the complex mixtures obtained from the annulation reactions are overwhelming and inconclusive.

The reactions with bis-electrophiles (**28**, **33**, **34**, **36-39**) resulted in product mixtures in which the desired product could not be isolated nor confirmed by TLC or

NMR. Product mixtures obtained from the reactions with all acid chlorides (**33**, **38**, **39**), turned black immediately upon addition of the electrophile resulting in a dark brown tar and the formation of desired products could not be confirmed. The NMR spectra of several of the annulation reactions did often show integrations that could be interpreted as arising from the correct product; however, they could not be confirmed by MS. Based on these initial attempts of the annulations of CDs, it would appear that more experimentation should be conducted to gather more conclusive evidence for or against the formation of cyclized products according to the described methodology.

It has been determined that both strong and superbases can be effective for deprotonation reactions of CDs. NaH in DMF in combination with slow addition of the electrophile provided the annulation of *trans*-cyclohexanediol in the highest yield. In the reaction of 1,2-cyclohexanediol with C_6F_6 and NaH in THF, only 6% of cyclized product was formed, the remainder was the mono-substituted product and starting material. In contrast, when the phosphazene base was used with THF, again the majority of isolated product was mono-substituted and starting material, and only a very small amount of cyclized product was observed. Changing the solvent from THF to DMF, facilitated the formation of the annulated compound, supporting the hypothesis that the solvent can play a pivotal role in the annulation reactions of CDs.

CHAPTER FIVE

Chromatographic Testing of Unique Chiral Stationary Phases: Collaboration with Agilent Technologies

Chiral Stationary Phases Developed for Gas Chromatography

Capillary GC columns containing chiral stationary phases (CSPs) incorporating the chiral selectors developed in our group, were collaboratively provided by Agilent Technologies, a leading company in the development and manufacturing of capillary columns. The efficiency of these CSPs can be determined by evaluating their ability to perform enantiomer separations against a selected set of analytes. The information gathered can be used to gain insight about the inclusion characteristics and selectivity of each new CSP. The compiled results can also be used to make clear comparisons to current, commercially available CSPs.

New Chiral Stationary Phases

Since each chiral selector is functionally different, we hypothesized that each CSP may display selectivity towards particular analytes. The cyclodextrin structures of the chiral stationary phases that were tested are shown in Figure 5.1. Depicted this way, the structures in the figure do not accurately represent the overall shape and volume of the cyclodextrin. Thus, Figure 5.2 is intended to serve as a three-dimensional representation of these molecules, shown here with **BU 1**.

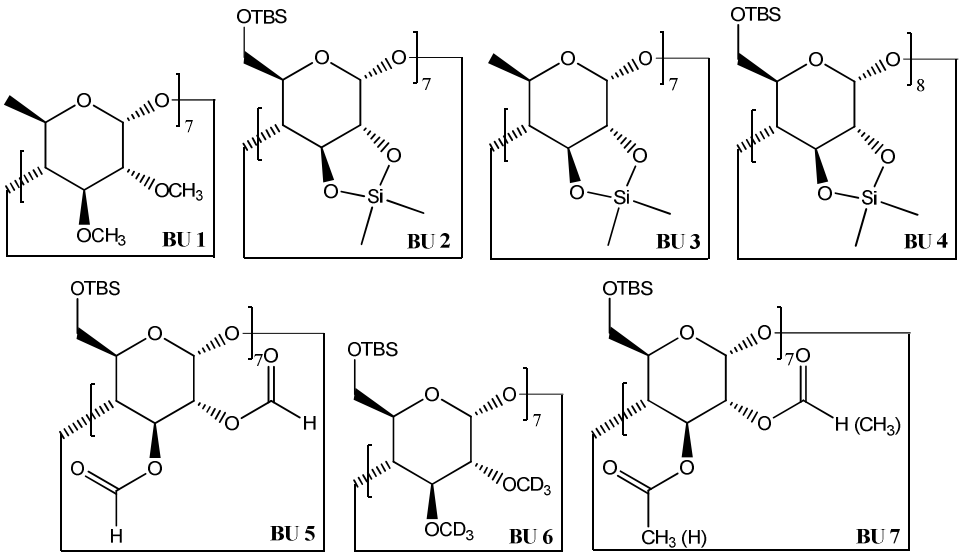


Figure 5.1. Structures of new β-CD-based CSPs for GC.

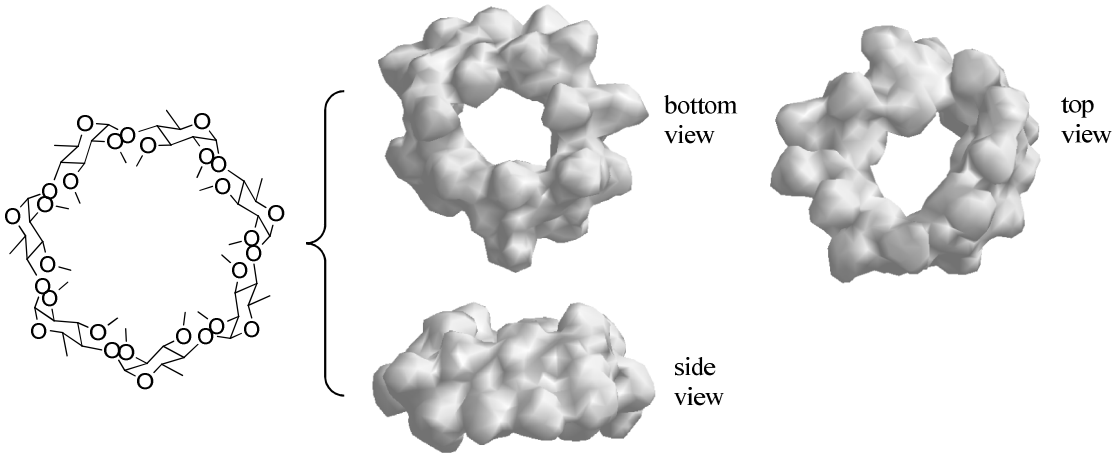


Figure 5.2. Structure and surface map of **BU 1**.

The majority of these phases contain functionality that is significantly different from what is currently used in existing CSPs. As in **BU 1** and **3**, we varied the size of the group at the 6-position. For **BU 1** this allows for both ends of the cavity to be fully accessible by virtue of the 6-deoxy functionality. It was unknown whether the small methyl group would be advantageous since studies have pointed to the 6-*tert*-

butyldimethylsilyl group as being preferred. To our knowledge, this phase represents the first of its kind to be tested in gas chromatography.

BU 2-4 are unique derivatives bearing an annulating group bridging the C(2) and C(3) positions. These groups serve to extend the cavity, alter the polarity and possibly improve the thermostability. To our knowledge, no annulated cyclodextrins have been reported in literature, and our expectation is that such functionality will impose a greater degree of enantioselectivity by virtue of the increased organization of the cavity.

The size of the CD is also important, thus, we developed **BU 4**, the γ-CD analogue of **BU 2**. This phase should allow us to draw conclusions about how the size of the CD effects the separation. In addition, if the bridging silyl group "chokes" the cavity in BU 2, then perhaps the analytes would interact more freely with the larger γ-CD cavity size.

BU 5 is an interesting phase in that it contains small formyl groups at C(2) and C(3). Since acetyl groups are commonly used in cyclodextrin CSPs, our initial speculation was that formyl groups, being very similar to acetyl groups might exhibit similar, but distinct and possibly improved chiral discrimination.

Studies have suggested that the thermal stability of CDs derivatives depend on the size, type, and location of any substituents.[18] The degradation process involves the opening the CD rings followed by a chemical decomposition similar to that of cellulose with loss of the glucosidic structure and hydroxyl groups. One of the main limitations to current CSPs is the relatively low maximum allowable operating temperatures (typically $< 250\,^{o}C$). Our thought was that incorporation of deuterium might provide a phase with increased thermal stability. Deuterium is both smaller and heavier than hydrogen and

when bonded to carbon, the C-D bond is stronger than the carbon-hydrogen bond. This could results in two effects. First, since more energy (higher temperature) is needed to break the bond between carbon and deuterium, higher thermal stability might result. Though a single deuterium might not exhibit a signification change, the incorporation of forty-two deuterium in the β-CD derivative might influence the thermal stability. This would be particularly true if the rate determining step in the decomposition involved breakage of the C-D bond. Second, deuterium- carbon bonds are slightly shorter than C-H bonds, making CD_3 groups. This difference in size could prove advantageous in chiral discrimination processes. Thus, **BU 6** was developed on the basis that the incorporation of deuterium in the molecule would provide increased thermal stability and/or enhanced enantioseparation behavior.

The mixed-phase **BU-7** contains a random distribution of acetyl and formyl groups. We anticipate the results of this phase to be interesting in that the selectivity could be greatly improved, reduced, or possibly reversed. Cyclodextrin derivatives are invariably solids; however, GC requires a liquid interface, requiring the chiral selectors to be dissolved in a fluid siloxane for column preparation. Solubility is sometimes a problem, and given that mixtures have decreased melting points, the mixture of many species from semi-random derivation could be advantageous.

Commercially Available Chiral Stationary Phases

In order to fully assess the applicability of our CSPs for enantioseparations in GC, we compared the results from the chromatographic testing to commercially available columns. Four such phases are shown in Figure 5.3. These phases were thought to represent some of the most efficient CD phases currently available.

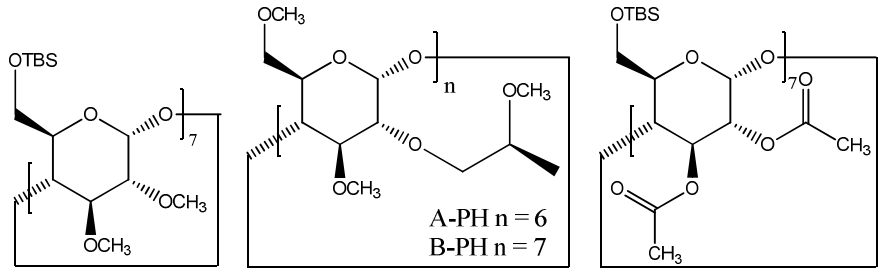


Figure 5.3. Structures of commercially available CD-based chiral stationary phases.

Analytes

Approximately 30 different analytes were chosen based on marginal separability on commercially available phases and availability. The Advanced Separation Technologies catalog gives information on the separation of nearly 400 chiral compounds.[68] Although approximately 13 phases are referred to at various places, the separation for any given compound is generally given on only one stationary phase, implying that the phase listed is the best phase for the separation of a given compound. The separations are characterized by their α values, which is the ratio of the retention times of each enantiomer after correcting for column dead volume. Analytes were selected that exhibited an α value of 1.02 or less (i.e., no more than a 2% difference in retention times) and that could be purchased or easily made in racemic (50:50) form. (An α value of 1.00 means no separation was observed). Given that Astec offers nearly every commercially available chiral phase, if a new stationary phase we make exhibits better α values than those reported, we can tentatively conclude that we have a stationary phase better than any that are commercially available, at least with respect to analytes that are marginal on Astec columns. In a few cases, analytes were chosen based on their use in the Garner group. The set of analytes selected for enantioseparation testing are shown in

Figure 5.4. Several of these compounds had to be synthesized before testing could begin; this was generally accomplished following literature preparation.

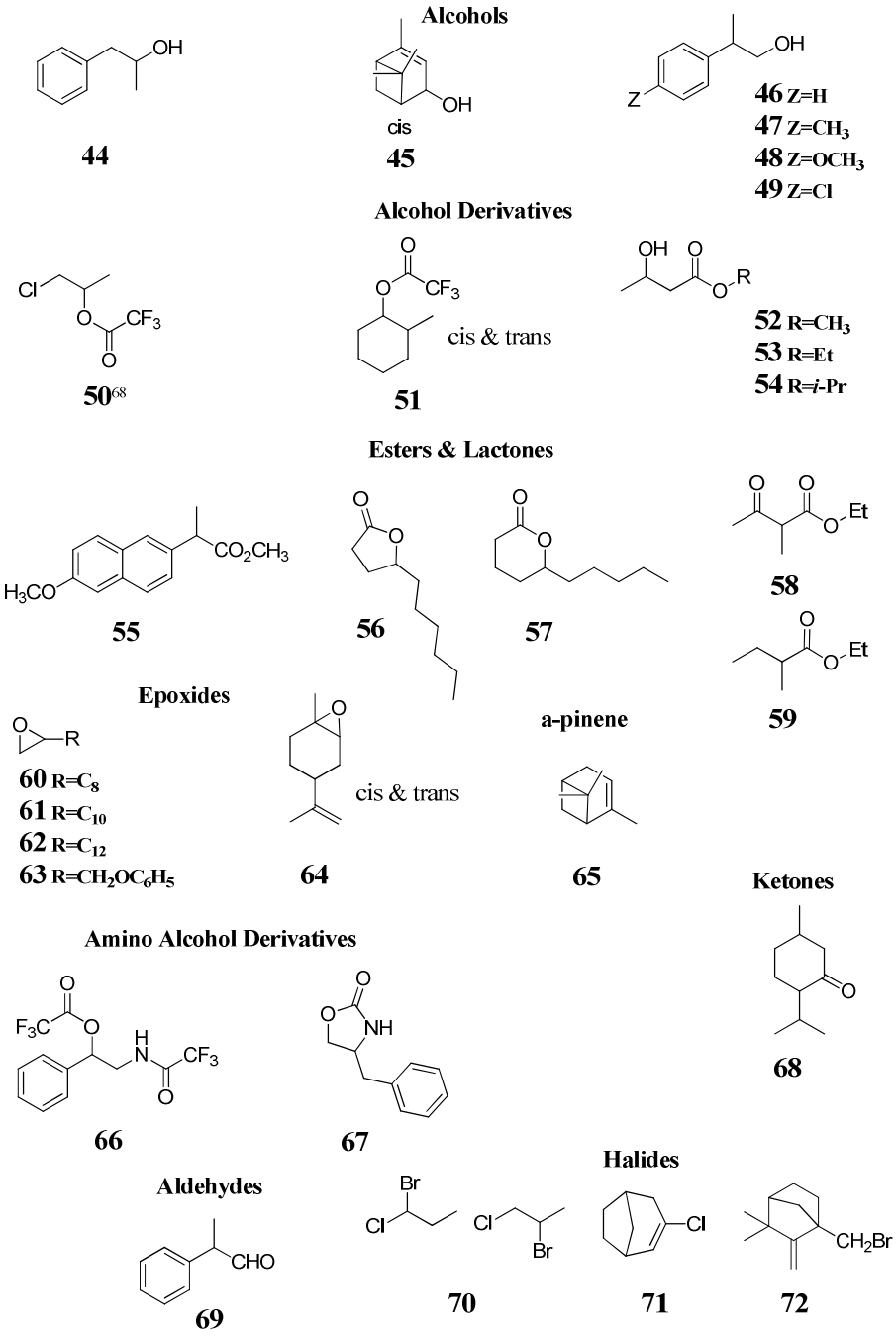


Figure 5.4. Structures and types of selected analytes.

GC Parameters and Pertinent Information

Column testing was conducted on a Hewlett Packard 5890 series II GC with a flame ionization detector. Injection volumes were maintained at ½ μL with a split flow of >100:1. The stationary phase compositions and specifications can be found in Appendix B:Tables (B.5.1-11). All columns tested were of the same dimensions, 30 m X 0.25 mm I.D. Each column was operated under hydrogen carrier gas with a linear velocity of 40 cm/sec, determined using methane at 80 ^{o}C. For each analyte, the initial temperature and rate was determined such that a retention time of 8-15 min was observed. In Appendix B: Table B.5.12-13, the ability of each column to separate enantiomers was evaluated and recorded in the form of α, k', and R_s values, based on the equations introduced in Chapter One. Those analytes showing any separation, having an α value ≥1.000 and a resolution value of 0.5, were recorded. In all cases where enantiomer separation was observed, a theoretical plate calculation (*N*) was included. For each column tested, column efficiency (*N*) was also determined using dodecane, with an initial temperature and rate such that a retention time of 8-15 min was obtained.

The samples were prepared by dissolving 1 mg of the racemic analyte in 1 mL of 2,2-dimethylbutane, except in the case of analytes **55**, **66**, and **67** which required distilled methylene chloride. We found 2,2-dimethylbutane to be an optimal solvent due to the fact that it gives a single, fast-eluting and sharp solvent peak. This could be attributed to its low boiling point (50 ^{o}C) and possibly steric hindrance, preventing inclusion into the cyclodextrin cavity. In contrast, hexanes gave multiple solvent peaks spread over up to 1.6 minutes, depending on choice of initial column temperature.

Comparison and Chromatographic Testing of Commercially Available Phases

J & W CycloSil B. CycloSil-B contains 30% (2,3-*O*-dimethyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin dissolved in OV-1701. Given that the **BU 1** and CycloSil-B differ only in functionalization at the 6-position, where CycloSil-B contains a larger *tert*-butyldimethylsilyl group and **BU 1** incorporates a small methyl group, we felt that a direct comparison would address the structural value of these size differences. To summarize, **BU 1** separated 35% of the analytes we used, giving an average α of 1.01 ± 0.004, and an average R_s of 0.93 ± 0.411 for compounds that exhibited any separation. Comparatively, CycloSil B separated 60% of the analytes, giving an average α of 1.02 ± 0.014, and an average R_s of 2.220 ± 1.356. Overall, this phase separated a broad range of analytes. Of the analytes separated, most were baseline resolved; however, on a few occasions the separations were marginal (Figure 5.5).

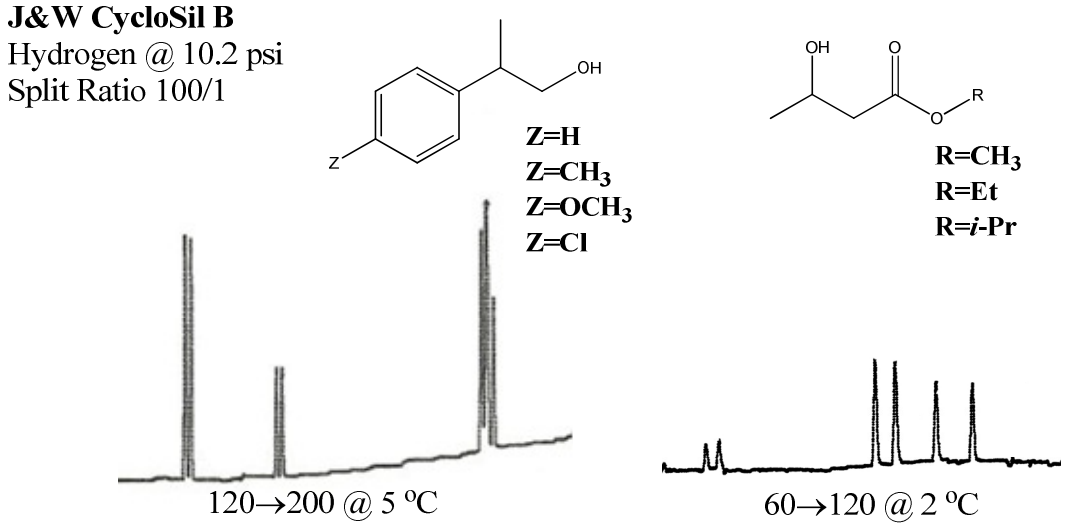


Figure 5.5. Separation of analytes (**44**) and (**52-54**) by CycloSil B.

CycloSil B gave better separations for all the analytes compared to all the BU phases. Based on these observations, it would seem reasonable to conclude that the larger *tert*-butyldimethylsilyl group is advantageous at the 6 position.

Chiraldex A-PH. The Supleco/Astec patented PH phases are unique in that they contain an additional chiral center by virtue of alkylation of the 2-hydroxyl group with (*S*)-propylene oxide, followed by permethylation. In our hands, the A-PH column (based on alpha cyclodextrin) separated about 40% of the analytes we tested, but in most cases the separation was quite marginal and unlikely to be useful (Figure 5.6).

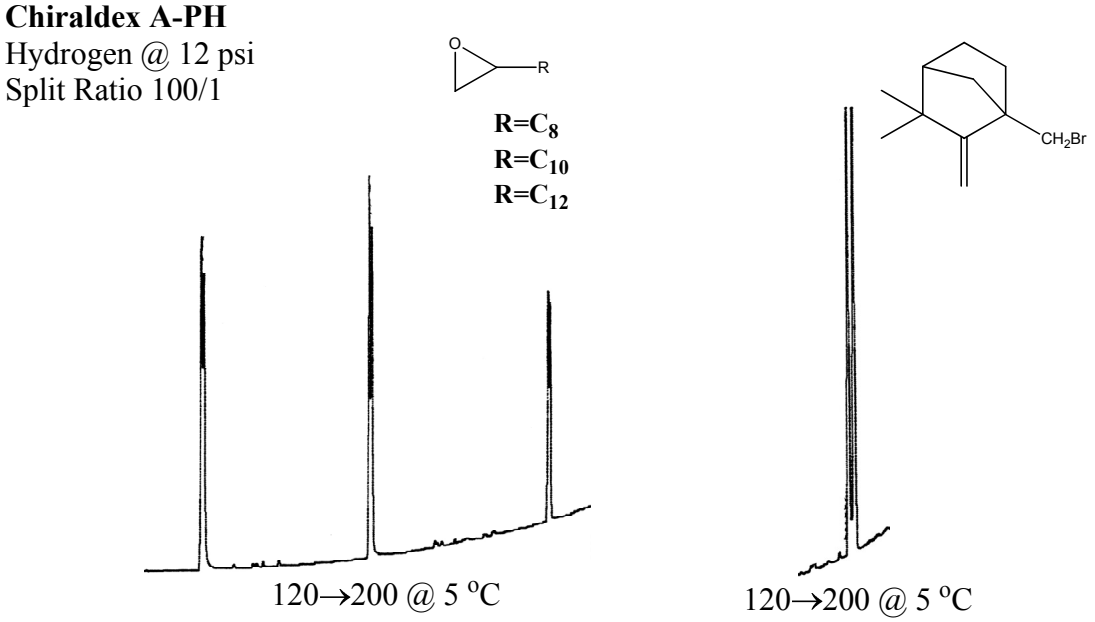


Figure 5.6. Separation of analytes (**60-62**) and (**72**) by A-PH.

The average α value for the 13 observed separations (out of 31 analytes) was only 1.009 ± 0.008, and the average resolution was 1.34 ± 0.7, both the lowest for any of the four columns we tested. Only three of the separations were baseline resolved ($R_s > 1.5$). However, the A-PH column was the only one of the four tested that successfully

separated the chlorobicyclooctene enantiomers (**71**). This phase can be considered as a general purpose column as it did separate at least one compound from each of the types of analytes, excluding ketones. In most cases, k' values were lower for the A-PH phases, which may suggest a reduced influence of inclusion complexing. This column's high price (~$1250) appears to be unjustified, and based more on the patented exclusivity of its manufacture than on its performance.

Chiraldex B-PH. B-PH is the β-CD analogue of the PH phase columns While it separated a far smaller fraction of analytes (16%) than did the A-PH, the separations were better, the average α was 1.02 ± 0.01, and the average R_s = 2.6 ± 1.8. B-PH was more selective for aliphatic and olefinic analytes in general and showed a preference for lightly functionalized analytes. In the case of the cis-verbenol (**45**), bromomethylene fenchone (**73**), and the benzyloxazolidinone (**68**) analytes, this phase showed superior performance (Figure 5.7). However, the cost of this column (~$1250) appears to be unjustified by its performance.

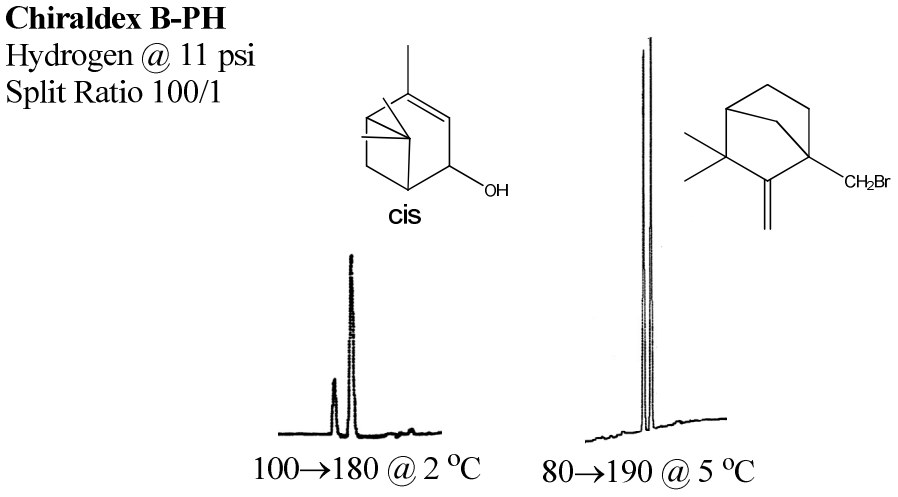


Figure 5.7. Separation of analytes (**45**) and (**72**) by B-PH.

Restek Rt-βDEXsa. The Restek Rt-βDEXsa, which is a simple (6-OTBS-2,3-diacetoxy)-β-CD derivative, separated 70% of the analytes we used, and relatively well. The average α value was 1.03 ± 0.01, and the average R_s was 3.27 ± 1.6, both the highest for any of the four columns we tested. This phase showed the highest selectivity for alcohol derivatives, esters, and lactones, amino alcohol derivatives, and aldehydes. Interestingly, all aromatic analytes were separated with this phase except the largest aromatic analyte, naprosyn methyl ester (**56**). Based on the results obtained, it would be difficult to conclude the size of the cavity influences the separating ability of this phase. As the R_s values indicate (Appendix B: Table 5.12-13), this stationary phase was superior at separating the analytes of interest except in a few cases (e.g., both amino alcohol derivatives (**67**) and (**68**), and bromomethylene fenchone (**73**)). We did note one slightly disturbing feature of this column: it tends to give wider peaks (average half-height widths = 0.106 min) than any of the others tested (range 0.052 – 0.077 min), hence the lower theoretical plate calculation for this column (Figure 5.8).

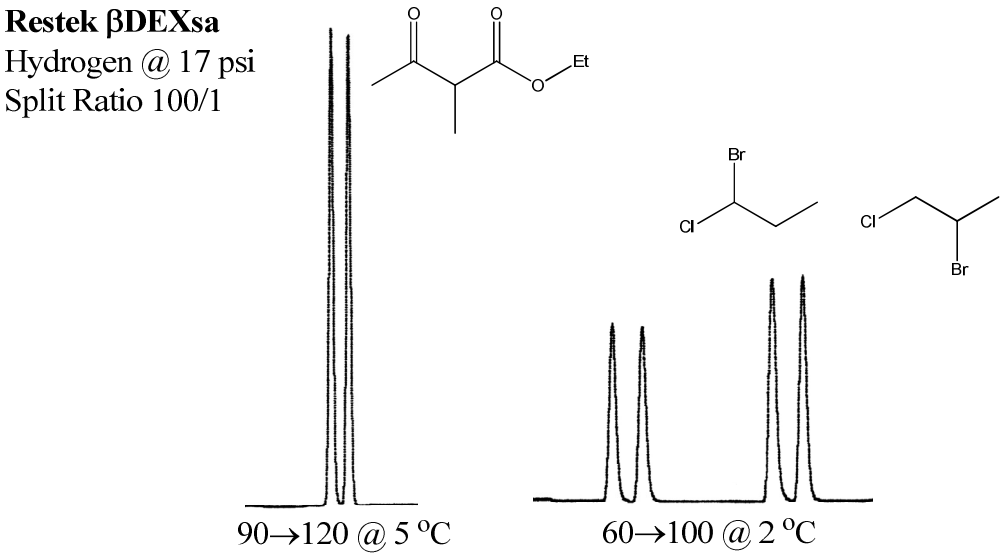


Figure 5.8. Separation of analytes (**58**) and (**70**) by Rt-βDEXsa.

*New Chiral Stationary Phases **BU 1 -7**.* **BU 1** separated 35% of the analytes we used, giving an average α of 1.01 ± 0.004, and an average R_s of 1.58 ± 0.7. Based on the values provided by Agilent, it also offers the higher maximum allowable temperature (see Appendix B: Tables (B.5.12)) compared to the other columns tested. This phase often showed the best selectivity for alcohols, lactones, and esters. In some cases **BU 1** showed better selectivity than the PH phases; however, the Rt-βDEXsa remained superior in most cases. In the cases where **BU 1** was superior to the PH phases, the majority of the analytes contained more polar functional groups. In two cases, 2-ethylbutyric acid ethyl ester (**60**) and trifluoroaetic acid, 1,-phenyl-2-(2,2,2-trifluoroacetylamino)-ethyl ester (**67**), **BU 1** showed better resolution compared to all the commercially available columns tested. Figure 5.9 shows two separations representative of what was typically observed for the majority of analytes by this phase.

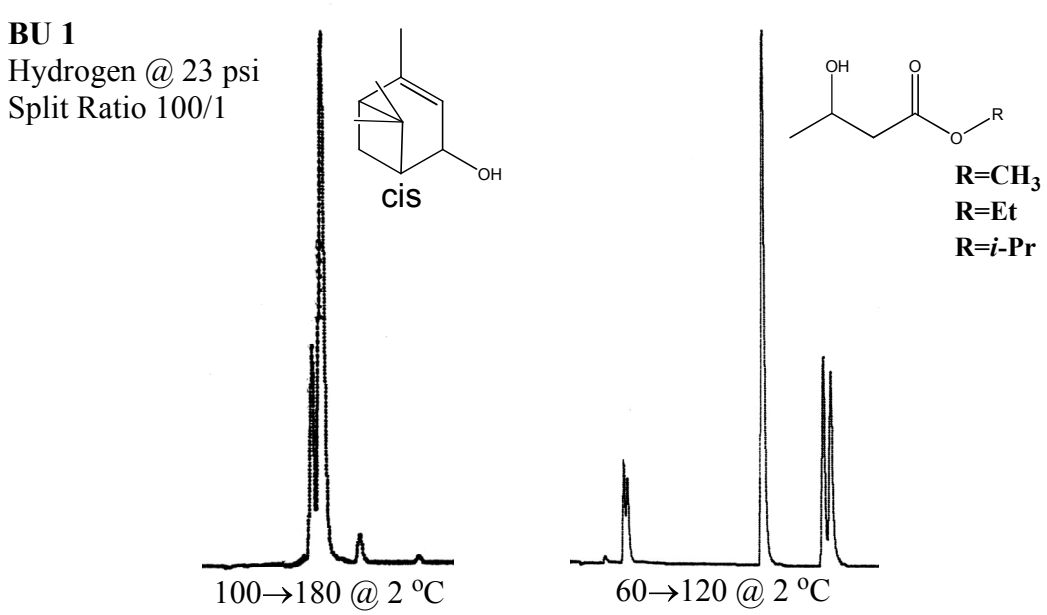


Figure 5.9. Separation of analytes (**45**) and (**52-54**) by **BU 1**.

Unfortunately, no separations were observed for the **BU 2** chiral selector; however, all of the analytes showed some retention in this phase. Of the select analytes, only α-pinene exhibited some separation ($\alpha = 1.01$, $R_s = 0.741$) as shown in Figure 5.10. It was our expectation that **BU 2**, which is the first derivative tested containing an annulation between the C(2) and C(3) oxygens, would show improved results compared to currently available phases; however, this was not the case. A plausible explanation for the lack of selectivity might be due to the annulated silyl group "chokes" the cavity.

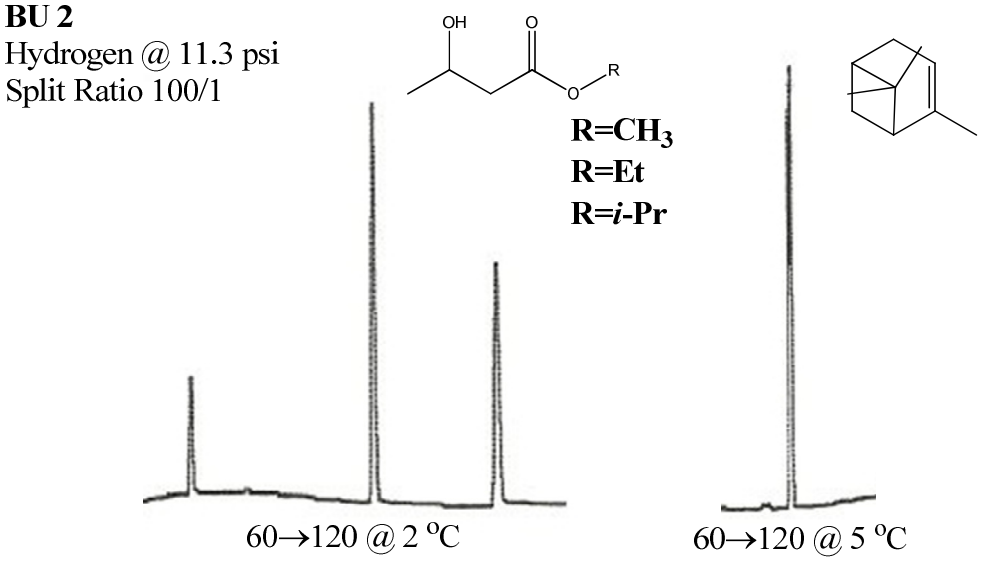


Figure 5.10. Separation of analytes (**52-54**) and (**65**) by **BU 2**.

As a chiral selector, **BU 3** exhibited poor behavior during testing. No separations were observed for any of the selected analytes and in many cases the analytes showed no retention with this phase. For those analytes that exhibited some retention, the peaks were severely broad with tailing. Two examples of this behavior are shown in Figure 5.11, to give an idea of what was observed for the majority of analytes with this phase. The comparison between **BU 2** and **3**; and CycloSil B and **BU 1**, suggests the *tert*-

butyldimethylsilyl group at the 6 position is preferred. This material was noted to have poor solubility in the siloxane matrix during column preparation, which may have contributed to the poor results.

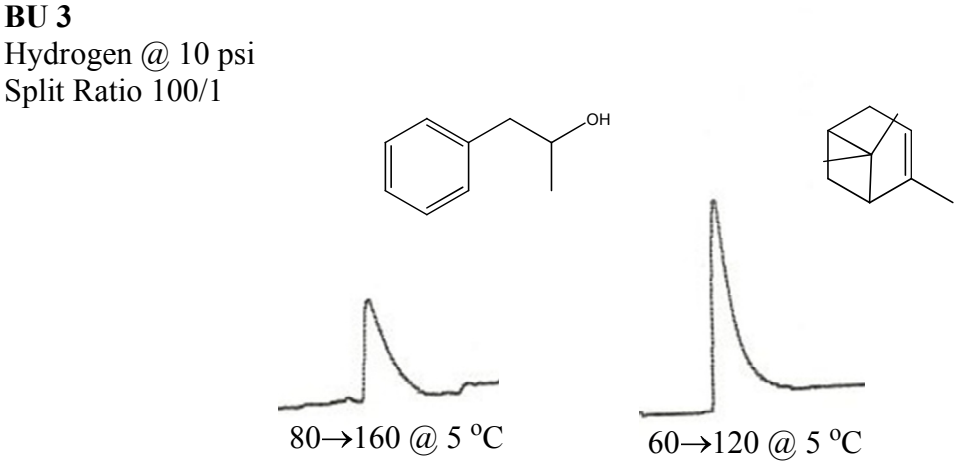


Figure 5.11. Separation of analytes (**44**) and (**65**) by **BU 3**.

Comparatively, **BU 4** showed similar results to **BU 3**, with no separations. The only major difference between the two phases is the group at the C(6) position, where **BU 4** contains the larger *tert*-butyldimethylsilyl group. Unlike, **BU 3**, every analyte was retained on the column at a retention time comparable to the other columns tested, suggesting that interaction is occurring without complexation. These results further support the argument that the tert-butyldimethylsilyl group results in improved phases; therefore it is not completely surprising that **BU 4** would exhibit improved behavior. Additionally, the annulated dimethylsilyl group does not seem to influence the selectivity. Figure 5.12 shows two representative examples of the behavior observed for the majority of separations by this phase.

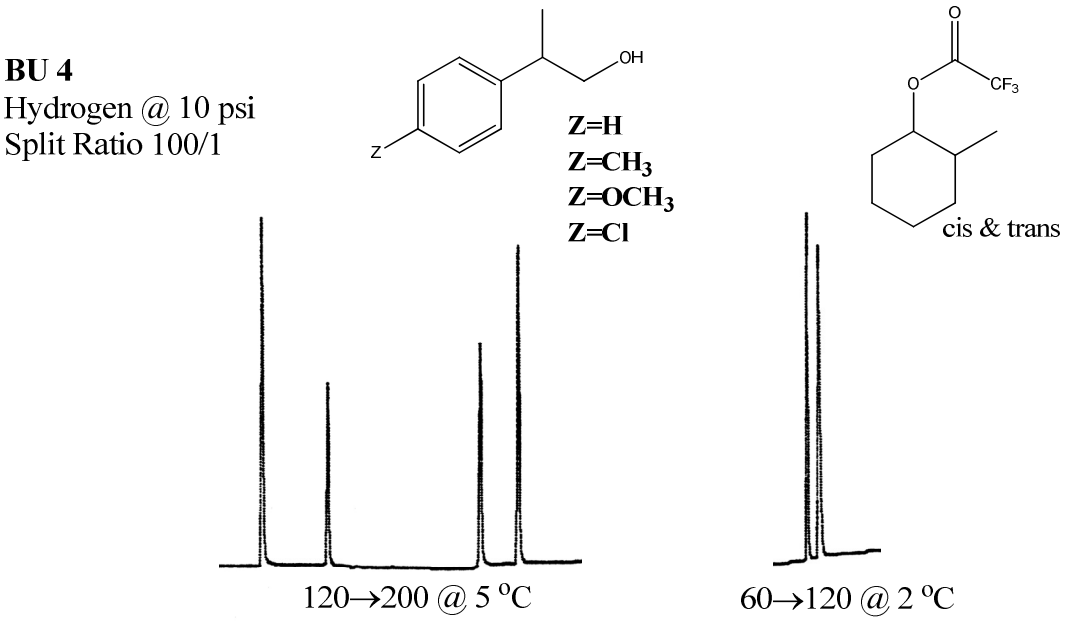


Figure 5.12. Separation of analytes (**46-49**) and (**51**) by **BU 4**.

BU 5 exhibited very surprising behavior. We observed no separations with this phase despite the fact that it contains groups that are very similar to what is currently employed in CSPs, such as acetyl groups. The peaks obtained were severely broad with tailing just as observed with **BU 3**. Figure 5.13, depicts this behavior.

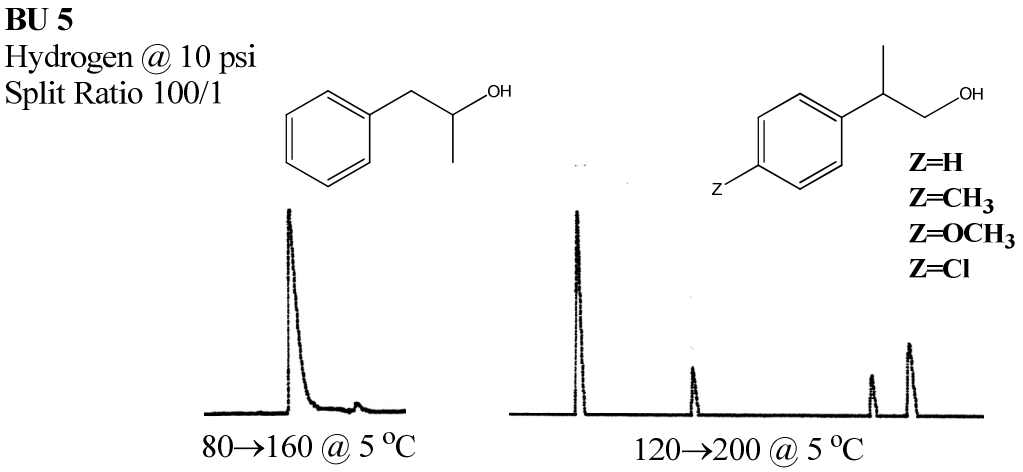


Figure 5.13. Separation of analytes (**44**) and (**46-49**) by CycloSil B

BU 6 separated 52% of the analytes with an average α of 1.016 ± 0.01 and R_s values averaging at 1.393 ± 0.837. This phase appeared to be selective for aliphatic compounds, alcohols, and aldehydes. This phase contains deuterated methoxy groups at the C(2) and C(3) positions. Figure 5.14 provides two examples of typical separations observed by this phase. Our initial thought was that perhaps the deuterium would provide the added steric interactions necessary to affect an increase in the thermal stability of the phase compared to its non-deuterated analogue. Deuterium is heavier than hydrogen and smaller and it has been suggested that given the right steric environment, the deuterium might provide increased thermal stability.

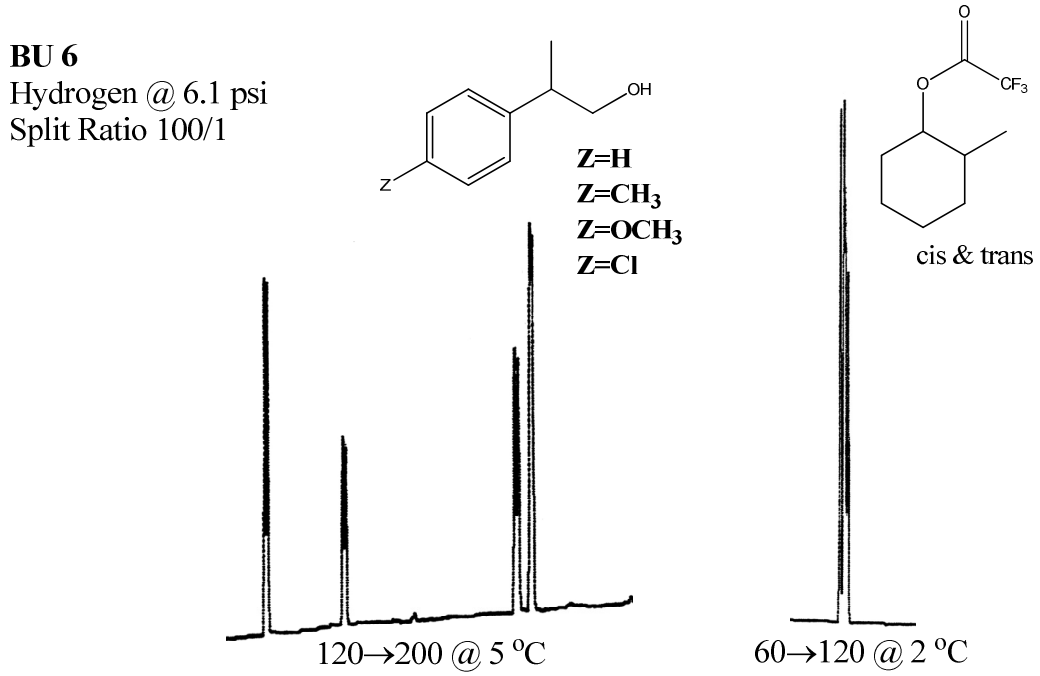


Figure 5.14. Separation of analytes (**44**) and (**51**) by **BU 6**.

To test this hypothesis, BU 6 and a commercially available column CycloSil B was subjected to thermal increases at 15 °C intervals over the range 230 °C to 330 °C. At each interval the temperature was held constant for one hour, and then cooled down. The

separation of a chosen analyte (α-pinene (**65**)) was tested and the R_s (resolution) recorded. Figure 5.15, compares the resolution values for analyte (**65**) for BU 6 and CycloSil B after 1 h at each temperature. The results suggest that the incorporation of deuterium into the cyclodextrin does not influence the thermal stability according to this thermal testing method.

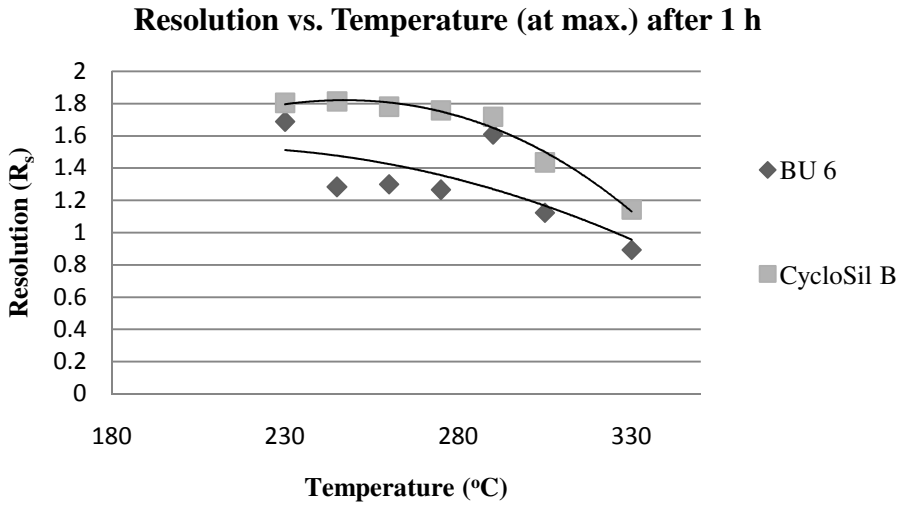


Figure 5.15. Plot of resolution values versus max. temp. after 1 h for BU 6 and J&W CycloSil B.

BU 7. The first example of a mixed phase, separated only separated 24% of the analytes. Only one analyte, aldehyde (**69**) was baseline resolved (R_s = 1.99). The average α value was 0.98 ± 0.13 and the average R_s was 1.18 ± 0.38. Shown in Figure 5.16, cis-verbenol (**45**) as well as both amino-acid derivatives (**66**) and (**67**) were separated on this column.

Overall, the Restek Rt-β-DEXsa phase gave better separations for the selected analytes compared to the other commercially available columns and **BU 1-7**. The Supleco/Astec PH phases were substandard in that for A-PH and B-PH combined only in

seven cases (or 11% of analytes) were the separations baseline resolved. **BU 1** was comparable to the PH phases and in many cases gave better separation.

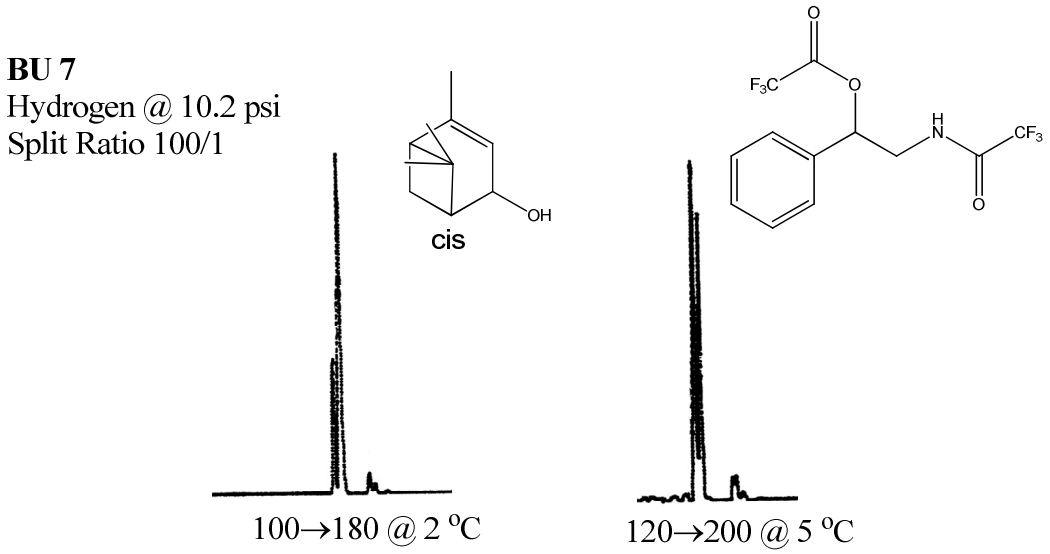


Figure 5.16. Separation of analytes (**45**) and (**66**) by CycloSil B

The commercially available CycloSil B and the Restek Rt-βDEXsa proved to be superior phases during our testing. The Restek phase separated slightly more of the selected analytes than the CycloSil B phase. The **BU 2**, **3**, **4**, **5, and 7** phases behaved poorly compared to **BU 1**. **BU 6**, exhibited comparable to slightly worse separations than the methoxy analogue of this phases, CycloSil B. These results are further illustrated in Figures 5.17 and 5.18 which show a comparison of resolution and separation values (R_s and α, resp.) for two of the analytes (**44**) and (**65**), each having different functionality, against all of the columns tested.

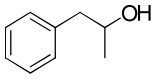
Compound	J&W CycloSil B	Chiraldex A-PH	Chiraldex B-PH	Restek Rt-βDEXsa	BU 1	BU 2	BU 3	BU 4	BU 5	BU 6	BU 7
					α						
	1.014	1.003	-	1.008	1.007	-	-	-	-	1.010	-
	1.02	-	1.017	-	1.019	1.014	-	-	-	1.017	-

Figure 5.17. Separation factor, α, values for analytes (**44**) and (**65**) for all columns tested.

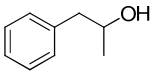
Compound	J&W CycloSil B	Chiraldex A-PH	Chiraldex B-PH	Restek Rt-βDEXsa	BU 1	BU 2	BU 3	BU 4	BU 5	BU 6	BU 7
					R_s						
	2.413	0.465	-	2.063	1.140	-	-	-	-	1.436	-
	1.74	-	0.528	-	1.257	0.672	-	-	-	1.279	-

Figure 5.18. Resolution, R_s, values for analytes (**44**) and (**65**) for all columns tested.

We conclude that (a) bridging 2,3-*O*-dimethylsilyl groups block the larger opening too much to allow analytes access; (b) 6-*O*-TBS groups are superior to the 6-deoxy derivative both in terms of performance and in terms of solubility in the siloxane matrix; (c) assuming solubility issues were not responsible for the observed behavior, the 6-deoxy derivative does not allow analytes access to the smaller end of the cavity or such access is ineffectual in enantiomer discrimination.

CHAPTER SIX

Materials and Methods

General Section

All reactions which required the use of air or water sensitive reagents were carried out in flame-or oven-dried glassware under nitrogen atmosphere, unless otherwise stated. Cannulas and metal luer needles were oven-dried and stored in the oven prior to use and disposable needles were dried with a stream of dry air and stored at room temperature prior to use. Methanol (MeOH), ethanol (EtOH), acetone, diethyl ether (Et_2O), and *N,N*-dimethylformamide (DMF) were obtained from Aldrich Chemical Company, VWR, Acros Chemical, or Fischer Scientific and used as obtained. Hexanes, ethyl acetate (EtOAc), and methylene chloride (CH_2Cl_2), were obtained from these same sources and distilled prior to use. Solvents that required special drying such as pyridine and tetrahydrofuran (THF) were distilled from calcium hydride and potassium, respectively.

Unless otherwise stated, all reactions were monitored by either thin layer chromatography (TLC), or nuclear magnetic resonance (NMR) and in some cases electron-ionization mass spectrometry (ESI-MS). Chiral gas chromatography was carried out on a Hewlett Packard 5890 series II GC with a flame ionization detector. Gas chromatography/mass spectroscopy was carried out on either a Hewlett Packard GCD 1800a with electron impact ionization or a Thermo GC/MS with electron impact ionization. ^{1}H and ^{13}C NMR spectra were obtained using a Varian Innova 500 MHz NMR operating at 500 MHz for proton and 125 MHz for carbon, or a Brüker Advance 300 MHz NMR operating at 300 MHz for proton and 75 MHz for carbon. Chemical

shifts are expressed in ppm, and peaks are reported as singlets (s), doublets (d), triplets (t), quartets (q), pentets (pent), multiplets (m), or any combination of these with coupling constants (J) reported in Hz. All carbon spectra are proton-decoupled. Slow additions were accomplished using a KdS 100 digital syringe pump. Concentration *in vacuo* was accomplished using a rotory evaporator followed by house vacuum (between 5 and 25 torr) and further concentrated by use of a mechanical pump (~ 0.1 torr) if necessary. Unless otherwise stated, isolated yields are reported. All aqueous solutions were prepared using deionized (DI) water. All commercially available chemicals were obtained from Aldrich, Acros, VWR, Fischer, and TCI and were used as obtained without further purification, unless otherwise stated. Analytes and electrophiles were obtained from similar commercial sources as stated above and used as obtained unless otherwise noted. Drying of β-cyclodextrin and derivatives was accomplished by subjecting the commercially available material to an Abderhalden drying pistol under reduced pressure with refluxing toluene in the presence of P_2O_5.

Preparation of Modified Cyclodextrins

Per(6-O-tert-butyldimethylsilyl-2,3-dihydroxy)-β-cyclodextrin (**3**):[29] Per(6-*O*-TBS-2,3-*O*-dihyroxy)-β-CD was synthesized according to the method of Fugedi[29] and is prepared as follows: Dry β-CD (**1**) (9.04 g, 7.96 mmol) was dissolved under vigorous stirring in dry pyridine (100 mL). The solution was cooled in an ice bath to 0 ^{o}C, producing a thick gel. A solution of TBDMSCl (14.5 g, 96.2 mmol) in dry pyridine (100 mL) was then added dropwise via an addition funnel to the cooled reaction vessel over 3.5 h. During this time the gel liquified. Cooling was continued for an additional 3 h

before the solution was allowed to warm to room temperature. After 18 h, the solvent was removed under reduced pressure to give a brown solid, which was taken up in CH_2Cl_2 (100 mL). The CH_2Cl_2 layer was washed with $KHSO_4$ (100 mL, 1 M) to remove any residual pyridine, followed by saturated aqueous NaCl solution. The CH_2Cl_2 was separated, dried with anhydrous Na_2SO_4, and evaporated to dryness. Column chromatography afforded (**3**) (12.9 g, 84%) as a light yellow solid. Rf 0.24 ($MeOH/CH_2Cl_2$, 10:90); ^{1}H-NMR (300 MHz, $CDCl_3$): 6.72 (s, 7H), 5.26 (s, 7H), 4.88 (d, *J* = 3 Hz, 7H), 4.03 (dd, *J* = 9.5, 9.5 Hz, 7H), 3.89 (dd, *J* = 11, 2 Hz, 7H), 3.70 (bd, *J* = 11 Hz, 7H), 3.65 (dd, *J* = 9.5, 3 Hz, 7H), 3.59 (bs, 7H), 3.55 (dd, *J* = 9.5, 9.5 Hz, 7H), 0.86 (s, 63H), 0.04 (s, 21H), 0.03 (s, 21H); ^{13}C NMR (75 MHz, $CDCl_3$): 102.1 (C-1), 81.8 (C-4), 73.7, 73.4, 72.6 (C-2,3,5), 61.7 (C-6), 25.9, 18.3, -5,1, -5.2..

Preparation of Cyclomannoepoxides

Per(6-O-tert-butyldimethylsilyl-2-O-tosyl)-β-cyclodextrin (**12**):[51] Per(6-*O*-TBS-2-OTos)-β-CD was synthesized according to the method of Coleman et al.[51] and is prepared as follows: To a solution of dry β-CD (**1**) (1 g, 0.88 mmol) in dry pyridine (50 mL) was added 4-dimethylaminopyridine (1.3 g, 10.6 mmol) and para-toluenesulphonyl chloride (2.1g, 11 mmol), and the mixture was stirred for 24 h at 50 ^{o}C. Water (10 mL) was added, the mixture was concentrate under reduced pressure, and the residue was extracted with ethyl acetate (2 x 50 mL). The combined extracts were washed with HCl (2N) (2 x 25 mL), saturated aqueous $NaHCO_3$ (25 mL) and water (2 x 25 mL), then dried and concentrated. Column chromatography of the residue gave (**12**) (58%). Rf 0.23 ($CHCl_3/(CH)_3CO$, 90:10); ^{1}H-NMR (500 MHz, $CDCl_3$): 5.19 (d, *J* = 3.5, 7H, H-1), 4.26

(dd, *J* = 9.7, 3.5 Hz, 7H, H-2), 3.08 (d, *J* = 3.2 Hz, 7H, 3-OH), 7.80 (d, *J* = 8.3 Hz, 14H, tosyl), 7.33 (d, *J* = 8.3 Hz, 7H, tosyl), 2.45 (s, 21H, tosyl), 0.86 (s,63H), 0.00 (s, 42H); ^{13}C NMR (125 MHz, $CDCl_3$): 98.84(C-1), 80.02, 79.85 (C-2, C-4), 72.62, 69.93 (C-3, C-5), 62.63 (C-6), 25.81, 18.23, -3.23, -3.41, 145.03, 133.00, 129.55, 128.25, 21.70 (tosyl).

Per(6-O-tert-butyldimethylsilyl-2,3-cyclomannoepoxide)-β-cyclodextrin (**13**): *From (**12**):*[51] Per(6-*O*-TBS-2,3-epoxy)-β-CD was synthesized according to the method of Coleman et al.[51] and is prepared as follows: To a solution of dry (6-*O*-TBS-2-*O*-tosyl)-β-CD (**1**) (150 mg) in dry THF (10 mL) was added NaH (30 mg, 60% in oil). The mixture was stirred for 3 h at 60 ^{o}C and methanol (10 mL) was added. The pH was adjusted to7 using HCl (2N), and then concentrated under reduced pressure. The residue was extracted with ethyl acetate (2 x 25 mL) and then dried and concentrated Column chromatography of the residue gave (**13**) (70 mg, 75%). Rf 0.41 (EtOAc/CH_2Cl_2, 20:80); ^{1}H-NMR (500 MHz, $CDCl_3$): 5.16 (s, 7H, H-1), 4.13 (d, *J* = 9.0 Hz, 7H, H-4), 3.91 (dd, *J* = 11.3, 3.0 Hz, 7H, H-6), 3.67 (d, *J* = 11.3 Hz, 7H, H-6'), 3.54 (m, 7H, H-5), 3.33 (d, *J* = 3.5 Hz, 7H, H-2), 3.12 (d, *J* = 3.5 Hz, 7H, H-3), 0.88(s, 63H); ^{13}C NMR (125 MHz, $CDCl_3$): 96.91(C-1), 69.80, (C-5), 68.84(C-4), 62.84 (C-6), 53.86 (C-2), 49.35 (C-3), 25.88, 25.56, 18.38, -5.10, -5.28.

Nucleophilic Ring-Opening of Cyclomannoepoxides

Cyclohexene oxide Model Study

trans-2-Hydroxy-cyclohexanecarbonitrile: From ref. 57(a): A solution of cyclohexene oxide (5.0 mmol) in acetonitrile (5 mL) was treated with anhydrous $LiClO_4$ (7.5 mmol) and KCN (7.5 mmol). The resulting reaction mixture was stirred at 70 ^{o}C for

24 h, then cooled to room temperature, diluted with water and extracted with diethyl ether. Evaporation of the washed (water) ether extracts afforded a crude reaction mixture that was analyzed by GC/MS. (91% yield), MS: 125 [M^+], 96, 82, 69, 54.

From ref. 57(b). To an oven-dried flask, KCN (6.44 g, 0.98 mol), and NH_4Cl (2.34 g, 0.04 mol) is mixed together in a MeOH/H_2O (8:1) solution (10 mL), upon which acetonitrile (10 mL) is added. Cyclohexene oxide (2 mL, 1.94g, 0.02 mol) is added via syringe to the mixture, which was allowed to stir at reflux for 24h. The reaction mixture was cooled to room temperature and extracted twice with EtOAc, filtered, dried and concentrated to obtain an orange oil. (90 % yield), MS: 125 [M^+], 96, 82, 69, 54.

From ref. 57(c). To a magnetically stirred solution of KCN (9.65 g, 0.15mol) in acetonitrile (60 mL) and H_2O (200 mL), was added cyclohexene oxide (4.85 g, 0.05 mol) along with a catalytic amount of 18-crown-6. The reaction was allowed to stir for 40 h at 50 ^oC. The mixture was extracted with CH_2Cl_2 (4 x), dried, and concentrated under reduced pressure to obtain a light brown oil (85% yield), MS: 125 [M^+], 96, 82, 69, 54.

Preparation of Novel Modified β-Cyclodextrin Derivatives

Per(6-iodo-2,3-dihydroxy)-β-cyclodextrin (**15**):[31] Per(6-deoxy)-β-CD was synthesized according to the method of Baer et al.[31] and is prepared as follows: To a stirred solution of dessicator-dried PPh_3 (21 g) in dry DMF (80 mL) was added I_2 (20.5 g) in small portions, followed after 30 min by dry β-CD (**1**) (4.32 g, 3.8 mmol). The mixture was stirred for 18 h at 80 ^oC under nitrogen atmosphere, and then concentrated at reduced pressure to half its volume, cooled to 5 ^oC, made alkaline with NaOMe in MeOH (~4 M) to pH 9-10, and kept at room temperature for 30 min. The solution was then

poured into vigorously stirred ice-water (1.5 L), and the beige-colored precipitate was collected by filtration. The product was washed well with water, dried in the air, and suspended in CH_2Cl_2 (1 L). After thorough agitation of the suspension the undissolved material was filtered off, washed several times with CH_2Cl_2, dissolved in DMF (100 mL), and reprecipitated by pouring the solution into stirred ice-water. The dried product was freed from some remnant, discoloring impurity by trituration with a small amount of MeOH, to give colorless (**15**) (6.49 g, 89.5%) Rf 0.4 (EtOAc/Hexanes, ^{13}C NMR (125 MHz, DMSO-d_6): 102.5(C-1), 85.7 (C-4), 72.1, 71.9, 70.9 (C-2,3,5), 9.4 (C-6).

Per(6-iodo-2,3-diacetoxy)-β-cyclodextrin (**16**):[31] A solution of (6-iodo)-β-CD (**15**) (2.0 g) in Ac_2O (15 mL) and pyridine (10 mL) containing a catalytic amount of 4-dimethylaminopyridine was kept for 48 h at room temperature. The mixture was quenched by the slow addition of MeOH (30 mL), and coevaporation of the solvent with additional MeOH and several portions of toluene. The crude product was purified by column chromatography to give (**16**) (2.2 g, 84%). Rf 0.4 (EtOAc/Hexanes, 1:1); ^{1}H-NMR (500 MHz, $CDCl_3$): 5.31 (dd, *J* = 9.9, 8.3 Hz, 7H, H-3), 5.17 (d, *J* = 3.8 Hz, 7H, H-1), 4.80 (dd, *J* = 9.9, 8.3 Hz, 7H, H-2), 3.8-3.5 (complex m, 28H, H-4,5,6,6'), 2.06 and 2.03 (s, 6H); ^{13}C NMR (125 MHz, $CDCl_3$): 170.7, 169.5 (2 CO), 95.9 (C-1), 79.5, (C-4),70.3, 70.1, 69.9, (C-2,3,5), 20.5 (2 $COCH_3$), 7.7 (C-6).

Per(6-deoxy-2,3-diacetoxy)-β-cyclodextrin (**17**): *(a) From* (**16**):[31] A solution of (**16**) (2.29 g), Bu_3SnH (6 mL), and a catalytic amount of AIBN in toluene (100 mL) was refluxed for 45 min under nitrogen atmosphere. The solvent was evaporated, and the residue, dissolved in CH_2Cl_2 (100 mL), washed with H_2O (2 x 60 mL). The dried

(Na_2SO_4) organic phase was concentrated and the product purified by column chromatography to give (**17**). (80 % yield). Rf 0.10 (ether, 200mL, followed by EtOAc/Hexanes, 3:1); ^{1}H-NMR (500 MHz, $CDCl_3$): 5.25 (dd, *J* = 9.7, 8.4 Hz, H-3), 4.96 (d, *J* = 3.9 Hz, H-1), 4.74 (dd, *J* = 9.9, 3.3 Hz, H-2), 4.04 (m, H-5), 3.31(t, *J* = 9.3, 8.4 Hz, H-4), 2.04, 2.01(2 s, 6H, 2 OAc), 1.35 (d, *J* = 6.2 Hz, 3H, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): 170.7, 169.3(2 CO), 96.4 (C-1), 82.4 (C-4), 71.01, 70.97 (C-2,3), 67.0 (C-5), 20.7 (2 $COCH_3$), and 17.8 (C-6).

(b) New improved preparation from (**16**): To an oven dried flask with stir bar was added $Ru(bipy)_3Cl_2.6H_2O$ (0.025 equiv.), and (**17**) (250 mg). The reaction vessel was purged with N_2, then *i*Pr_2NEt (0.2 mL, 10 eq.) and formic acid (0.04 mL, 10 equiv.) was added. The bright orange mixture was degassed with N_2 for 20 min and then place ~ 10 cm from a 14 W fluorescent bulb. The reaction mixture was diluted with EtOAc and poured into a separatory funnel containing EtOAc (10 mL) and H_2O (10 mL). The aqueous layer was re-extracted with EtOAc and the combined organic extracts were dried, and concentrated to give (**17**). (85-90%). Rf 0.26 (EtOAc/hexanes, 80:20). The ^{1}H-NMR and ^{13}C NMR spectra were identical with those previously reported.[31]

Per(6-deoxy)-β-cyclodextrin (**14**): *(a) From* (**15**):[31] Sodium borohydride (1.0 g, 26 mmol) was added portion wise to a solution of (**15**) (4.5 g, 2.85 mmol) in DMSO (100 mL) at 100 ^{o}C, and the mixture was stirred for 24 . After being cooled to 0 ^{o}C, it was diluted with MeOH (2 x 50 mL), and the solvents were evaporated under reduced pressure. The solid residue was dissolved in H_2O (100 mL), and 2 % AcOH was carefully added to reach pH 7. The collected solid was passed through Dowex 50-W 8x

(H^+) cation-exchange resin to give (**17**) (36%), ^{13}C NMR (125 MHz, DMSO-d_6): 102.3 (C-1), 88.1 (C-4), 73.2 (C-3), 72.6 (C-2), 66.7 (C-5) and 17.4 (C-6).

(b) *From* (**17**):[31] Baer et al. reported subjecting a solution of (**17**) in dry MeOH to a solution of NaOMe until alkaline and then passing the solution through Amberlite IR-120 (H^+) cation-exchange resin, and evaporated to give sodium-free (**14**). Our modified method is described as follows: To an oven-dried flask with stir bar (6-deoxy-2,3-diacetoxy) (**17**) (3.52 g, 2.18 mmol) was added anhydrous MeOH (50 mL). Followed by a 10% excess of NaOMe (7.7 mL, 33.6 mmol, 15.4 equiv.) (25 wt% solution in MeOH, ~4.36 M), a white precipitate formed immediately. The flask was equipped with a distillation head and heated to slowly distill away the MeOH until a thick slurry remained. After cooling to room temperature, MeOH (40 mL) was added to make a stirable suspension. Then Dowex 50-W 8x cation-exchange resin (H^+) (10.5 g) was added to the reaction vessel. Between 0.5-1.5 h, the solution became slightly cloudy yellow, indicating neutral or slightly acidic pH. The cloudy solution was filtered through Hirsch funnel with no filter paper, and evaporated to dryness to obtain (**14**) (54% yield). ^1H-NMR (500 MHz, DMSO-d_6): 5.7 (br, 2-OH), 4.80 (d, J = 3.5 Hz, H-1), 3.71 (dq, J = 9.4, 6.2 Hz, H-5), 3.55 (t, J_{tot} = 18 Hz, H-3), 3.22 (dd, J = 9.5, 3.5 Hz, H-2), 2.99 (t, J_{tot} = 18.3 Hz, H-4), and 1.18 (d, J = 6.2 Hz, 3H, CH_3); ^{13}C NMR (125 MHz, DMSO-d_6): 102.3 (C-1), 88.1 (C-4), 73.2 (C-3), 72.6 (C-2), 66.7 (C-5) and 17.4 (C-6).

Alkylation/Arylation By Organocuprates

Per(6-methyl-2,3-diacetoxy)-β-cyclodextrin (**18**): To an oven dried flask purged with N_2 containing CuX (X = I, CN) (8 equiv.) is added THF (15 mL). The reaction

vessel is cooled to 0°C, and MeLi (15 equiv.) is added and allowed to stir for 10 min. The (6-iodo)-β-CD derivative (**17**) (1.7 g, 0.68 mmol) was taken up in THF (5 mL) and added slowly via cannulation to the methyl cuprate. The reaction was kept at 0°C and then slowly allowed to warm to room temperature overnight. After 24 h the reaction was warmed to 50°C and allowed to stir an additional 2 h. The reaction was poured into a saturated aqueous solution of ammonium chloride, buffered to pH 8-9 by addition of small volume of con. aq. ammonia (10% v/v). The resulting solution was poured over a pad of Celite® followed with several washes of CH_2Cl_2. The reaction was further extracted with CH_2Cl_2, washed with H_2O, sat. aq. NaCl, dried and concentrated to give (**18**). (35-36%). Rf 0.23 (EtOAc/hexanes, 80:20); ^{1}H NMR (500 MHz, $CDCl_3$): 5.31 (t, H-1), 5.17 (d, H-2), 4.80 (dd, H-3), 3.78 (m, H-5), 3.58 (t, H-4), 2.08, and 2.05 (2 s,6H, 2 OAc), 1.54 (m, 14H, H-6), 0.98 (s, CH_3); ^{13}C NMR (125 MHz, $CDCl_3$): 170.5, 169.3 (2 CO), 96.4 (C-1), 80.4 (C-4), 70.3, 70.1, 69.9 (C-3, 2, 5), 20.6 (C-6), 7.9 (6-CH_3).

Per(6-phenyl-2,3-diacetoxy)-β-cyclodextrin (**19**): To an oven dried flask purged with N_2 containing CuCN (8 equiv.) is added THF (15 mL). The reaction vessel is cooled to -78°C, and PhLi (15 equiv.) is added and allowed to stir for 10 min. The (6-iodo)-β-CD derivative (**17**) was taken up in THF (5 mL) and added slowly via cannulation to the phenyl cuprate. The reaction was kept at -78°C and then slowly allowed to warm to room temperature overnight. After 24 h the reaction was warmed to 50°C and allowed to stir an additional 2 h. The reaction was poured into a saturated aqueous solution of ammonium chloride, buffered to pH 8-9 by addition of small volume of con. aq. ammonia (10% v/v). The resulting solution was poured over a pad of Celite® followed with several washes of CH_2Cl_2. The reaction was further extracted with CH_2Cl_2,

and then washed with H_2O, sat. aq. NaCl, dried and concentrated to give (**19**). (27-33%). Rf 0.5 (EtOAc/hexanes, 20:80); ^{1}H NMR (500 MHz, $CDCl_3$): 7.31 (m, 5H), 5.31 (br.s, 7H, H-1), 4.80 (m, H-2, 3), 3.80-3.66 (m, H-5, 6), 2.34 (m, 6-CH_2), 2.04, and 2.01 (2 s, 2 OAc); ^{13}C NMR (125 MHz, $CDCl_3$): 170.5, 169.6 (2 CO), 137.3, 129.0, 128.7, 127.5 (phenyl) 96.5 (C-1), 72.3, 70.4, 69.1 (C2-5), 35.3 (C-6), 21.9, 20.6 (2 OAc).

Per(6-deoxy-2,3-dimethoxy)-β-cyclodextrin (**20**): To an oven-dried flask NaH (20 equiv.), and (6-deoxy)-β-CD (**14**) (540 mg, 0.53 mmol) was reacted in THF (40 mL). To help dissolve (**14**), DMF (5 mL) can be added. MeI (0.65 mL, 20 equiv.) is added to the reaction mixture and then it is allowed to stir overnight. The reaction mixture is quenched slowly with MeOH and H_2O (5 mL) is added, after which it is extracted with EtOAc, dried, and concentrated to give (**20**) (76%), Rf 0.25 (EtOAc); ^{1}H-NMR (500 MHz, DMSO-d_6): 5.27 (d, H-1), 4.09 (m, H-5), 3.60 (dd, H-2), 3.55 (dd, H-3), 3.26 (dd, H-4), 3.24 (2 s, 2 OCH_3), 1.21 (d, CH_3); ^{13}C NMR (125 MHz, DMSO-d_6): 97.8 (C-1), 76.8 (C-4), 75.0 (C-2), 69.4 (C-3), 62.5 (CH_3) and 15.2 (C-6).

Per(6-O-tert-butyldimethylsilyl-2,3-dimethoxy)-β-cyclodextrin (**21**): Our modified methodology for this previously reported compound, was prepared from (**3**) (800 mg), as previously detailed for (**20**), the procedure being analogous to the subsequent preparation of (**22**) and (**23**). The yield of (**21**) was (74%), Rf 0.48 (EtOAc/hexanes, 20:80); ^{13}C NMR (125 MHz, $CDCl_3$): 97.9 (C-1), 81.8 (C-4), 78.5 (C-3), 72.0 (C-2), 62.2 (C-5), 25.8, 18.1, -4.99, -5.34.

Per(6-O-tert-butyldimethylsilyl-2,3-bis-($^2H_3$)methoxy)-β-cyclodextrin (**22**): To an oven-dried flask NaH (20 equiv.), and (6-*O*-TBS)-β-CD (**3**) (800 mg, 0.41 mmol) was

reacted in THF (40 mL). CD_3I (0.65 mL, 20 equiv.) is added to the reaction mixture and then it is allowed to stir overnight. The reaction mixture is quenched slowly with MeOH and H_2O (5 mL) is added, after which it is extracted with EOAc, dried, and concentrated to give (**22**) (78%), Rf 0.48 (EtOAc/hexanes, 20:80); ^{1}H NMR (500 MHz, $CDCl_3$): 5.10 (d, 7H, H-1), 4.11 (d, 7H), 3.5-3.8 (complex m, H-6), 3.43 (d, H-3), 3.06 (d, OCH_3), 0.88 (s, 63H), and 0.03 (s, 42H); ^{13}C NMR (125 MHz, $CDCl_3$): 98.1 (C-1), 82.2 (C-4), 72.2 (C-3), 61.5 (C-2), 58.6 (C-5), 29.7, 18.3, 14.1, 0.00, -5.2.

Formic-acetic anhydride (**23**): *(a) From ref. 64a*: Formic-acetic anhydride was prepared following literature preparation[64a] and is described as follows: To an oven-dried flask with stir bar, equipped with a reflux condenser and addition funnel, all maintained under nitrogen atmosphere, is added sodium formate (28.3 g, 0.42 mol). Diethyl ether (100 mL, anhydrous) is added to the flask while vigorously stirring. To this stirred solution the additional funnel, charged with acetyl chloride (25 mL, 0.32 mol) was added as quickly as possible over 5 min with maintaining the temperature at 25-27^{o}C. After the addition is complete, the mixture is stirred for 5.5 h at room temperature. (NOTE: Work-up must be carried out as quickly as possible). The mixture was then filtered, and the residue washed with 100 mL diethyl ether, and these washings were added back to the original filtrate. The ether was removed by distillation (short path) at reduced pressure to yield (**23**) (70g, 64%), of colorless formic-acetic anhydride, b.p. 27-28^o (10 mm.), identical to reported values.

(b) From ref 64b: Formic-acetic anhydride was prepared following literature preparation[64b] and is described as follows: Formic acid (85%) (5 mL) was added slowly to cold acetic anhydride (10 mL) kept at 5^{o}C in an oven-dried flask. The mixture was

then heated to 60°C for 1h. The formic-acetic anhydride (24) (50%) was either removed by distillation analogous to *(a)* above, or immediately used.

Per(6-deoxy-2,3-O-diformyl)-β-cyclodextrin (**24**): To an oven-dried flask (6-deoxy)-β-CD (**14**) (200 mg, 0.196 mmol) with a catalytic amount of 4-dimethylaminopyridine was dissolved in pyridine (10 mL). The reaction was cooled to 5°C and previously prepared formic-acetic anhydride (20 equiv.) was added drop wise. The reaction was allowed to stir overnight, after which H_2O (2 mL) was added. The reaction mixture was washed with sat aq. $NaHCO_3$ and H_2O. The organic layer was separated, washed with brine, dried, concentrated, and purified to give (**24**), Rf 0.4 (EtOAc/hexane, 60:40); ^{1}H-NMR (500 MHz, DMSO-d_6): 8.26 (bs., 2 CHO), 5.26 (bs, 7H), 4.98 (br. m, 14H, H), 4.14 (m, 7H), 3.67 (m, 7H), 1.23 (H-6).

Per(6-O-tert-butyldimethylsilyl-2,3-O-diformyl)-β-cyclodextrin (**25**): Derivative (**25**) was prepared from (**3**) (558 mg), as previously detailed for (**24**); ^{1}H-NMR (500 MHz, $CDCl_3$): 8.23, 8.30 (2 s, 2H, CHO), 5.23 (d, 7H, H-1), 4.90 (m, H-2), 4.70 (m, H-3), 4.52, (m, H-5), 4.05 (m, 7H), 3.5-3.8 (complex m, H-,6), 3.43 (m, H-3), 3.06 (d, OCH_3), 0.88 (s, 63H), and 0.03 (s, 42H).

(6-O-tert-butyldimethylsilyl-2,3-di-O-(%formyl/%acetyl)mixed)-β-cyclodextrin (**BU 7**): To an oven-dried flask, (6-O-TBS)-β-CD(3) (540 mg, 0.28 mmol) and a catalytic amount of 4-dimethylaminopyridine was dissolved in anhydrous pyridine (5 mL). Formic-acetic anhydride (0.17 mL, 7 equiv.) and acetic anhydride (0.18 mL, 7 equiv.) is added simultaneously dropwise. The reaction was allowed to stir overnight at room temperature. A small amount of formic-acetic anhydride was added to ensure

complete reaction. The reaction mixture was quenched with sat. aq. $NaHCO_3$, extracted with EtOAc, dried and concentrated to give the mixed-CD; ^{1}H-NMR (500 MHz, $CDCl_3$): 4.89 (bs, 7H, H-1), 3.97 (d, 7H, H-5), 3.90 (d, 7H, H-6), 3.48-3.71 (complex m, H-2,3,4), 0.84 (bs, 72H), 0.132 (s, silyl CH_3), 0.039 (s, silyl CH_3), and 0.01 (s, 48H).

Synthesis of Potential Electrophiles for Annulation Reactions

Bischloromethyl ether (**37**): Bischloromethyl ether was prepared according to literature conditions[65] and are described as follows: In an oven-dried flask immersed in an ice bath, equipped with a stir bar and addition funnel is added con. HCl (28 mL, 33 g, 37-38%) and *para*-formaldehyde (40 g) (effectively 1.3 moles formaldehyde). While the temperature is maintained below 10^{o}C, chlorosulfonic acid (2.3 mol) is added drop wise at such a rate that HCl (g) is not lost from the reaction mixture, about 5.5 h. The mixture is stirred for 4 h in the melting ice-bath and comes to room temperature. It may be allowed to stand overnight. The layers are separated and the product is washed twice with ice-water. Ice is added to the product, and 40% NaOH (85 mL) is then added to the mixture slowly with vigorous agitation until the aqueous phase is strongly alkaline. Careful attention must be taken to ensure overheating does not occur due to rapid decomposition. The product is separated and dried rapidly over potassium carbonate and then over potassium hydroxide, keeping the product cold during drying. After, quick separation of the drying agent by filtration, 58 g of (37), sufficiently pure is obtained (76% yield). After distillation of the bischloromethyl ether, b.p. 100-104^{o}C, provides 72% yield. MS: 114 [M^+], 79 [M^+]-Cl, 49 [M^+]-CH_2OCH_2Cl.

1,2-Cyclohexanediol Model Study

Annulated hexafluorobenzene. To an oven-dried flask, NaH (205 mg, 2.2 equiv., 60% in oil) was added and the system purged with N_2. *trans*-Cyclohexane diol (270 mg, 2.32 mmol) and DMF (5 mL) were added to the reaction vessel with stirring. Hexafluorobenzene (0.3 mL, 2.32 mmol) was added to the stirred solution via a digital syringe pump set at a rate of 33.5 μL/h. After 8 h, the reaction mixture was cautiously quenched with water, extracted with EtOAc, dried and concentrated (93%), MS: 262 $[M]^+$, 81.

Per(6-deoxy-2,3-di-O-cyclodimethysilyl)-β-cyclodextrin (**42**): To an oven-dried flask, dry (6-deoxy)-β-CD (**14**) (620 mg, 0.61 mmol) was dissolved in anhydrous pyridine (5 mL) to which dichlorodimethylsilane (94 μL, ~8 equiv.) was added via a digital syringe pump set at a rate of 11.3μL/h. The reaction was quenched with H_2O (5 mL) and the product was extracted with CH_2Cl_2, dried, concentrated under reduced pressure to give (**42**) (540 mg, 62.5%) after column chromatography (EtOAc/ hexanes, 20:80). ^{1}H-NMR (500 MHz, $CDCl_3$): 4.85 (bs, H-1), 4.00 (m, H-5), 3.79-3.82 (m, H-2,3), 2.96 (bd, H-4),1.29(bs, 3H, CH_3), 0.35 (s, silyl), 0.11 (bs, silyl) 0.06 (bs, silyl); ^{13}C NMR (125 MHz, DMSO-d_6): 103.8 (C-1), 89.5,76.3, 74.9, 66.9 (C-2,3,4,5), 17.5 (C-6), 1.028 (silyl CH_3) and -0.46(silyl CH_3).

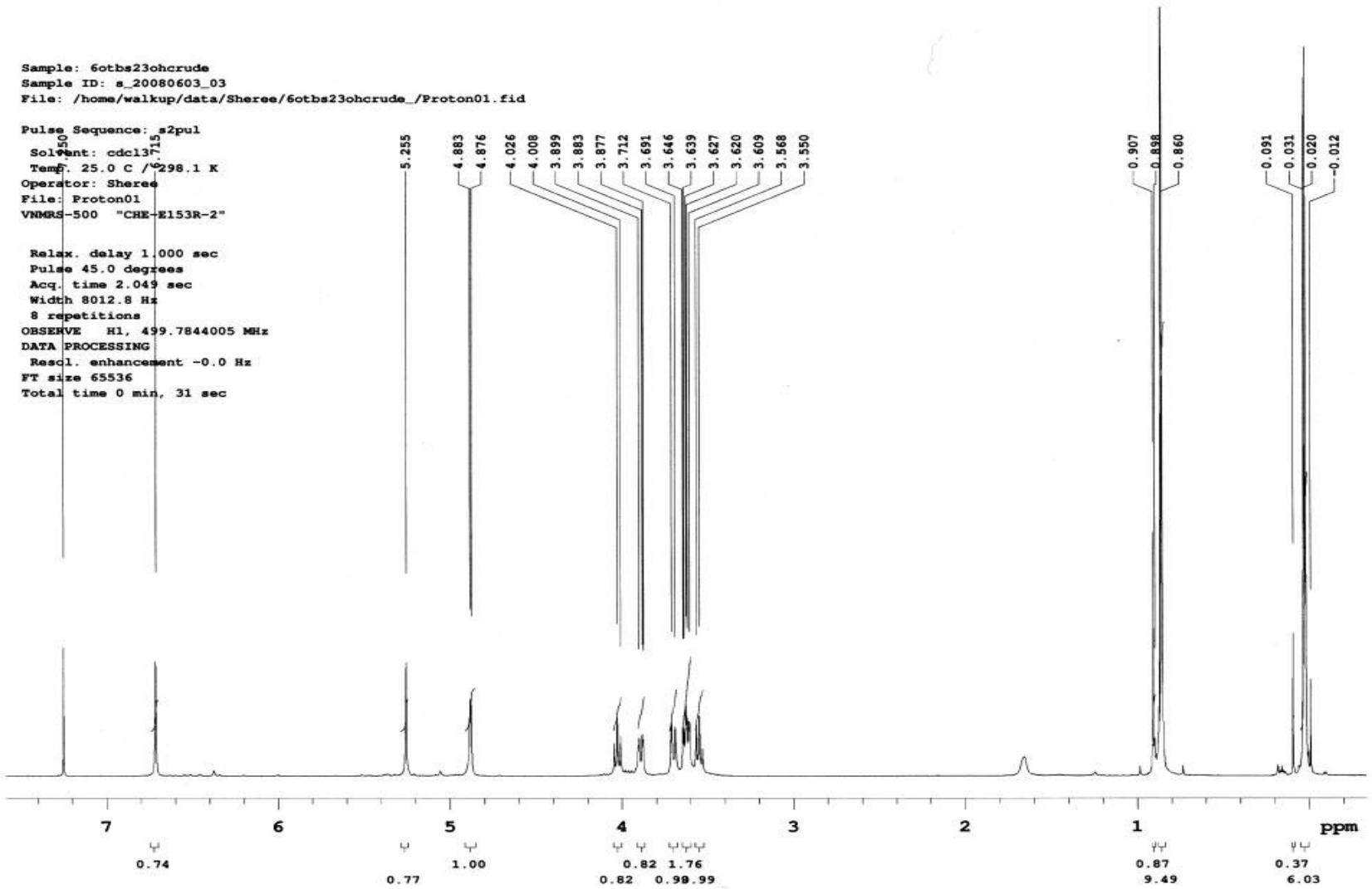
Per(6-O-tert-butyldimethylsilyl-2,3-di-O-cyclodimethysilyl)-β-cyclodextrin (**43**): This compound was previously synthesized in our group following an analogous procedure as previously described for (**42**) and was tested as received.

(6-O-tert-butyldimethylsilyl-2,3-di-O-cyclodimethysilyl)-γ-cyclodextrin (**BU-4**): This gamma derivative was prepared from γ-cyclodextrin (210 mg, 0.095 mmol), as previously described for (**42**), (100 mg, 40%), ^{1}H-NMR (500 MHz, $CDCl_3$): 4.89 (bs, 7H, H-1), 3.97 (d, 7H, H-5), 3.90 (d, 7H, H-6), 3.48-3.71 (complex m, H-2,3,4), 0.84 (bs, 72H), 0.132 (s, silyl CH_3), 0.039 (s, silyl CH_3), and 0.01 (s, 48H); ^{13}C NMR (125 MHz, $CDCl_3$): 102.4(C-1), 82.1 (C-4), 73.9, 72.9, 72.6 (C-2,3,5), 66.98 (C-6), 25.9 (OTBS), 18.3 (OTBS), -4.99 and -5.14 (silyl CH_3).

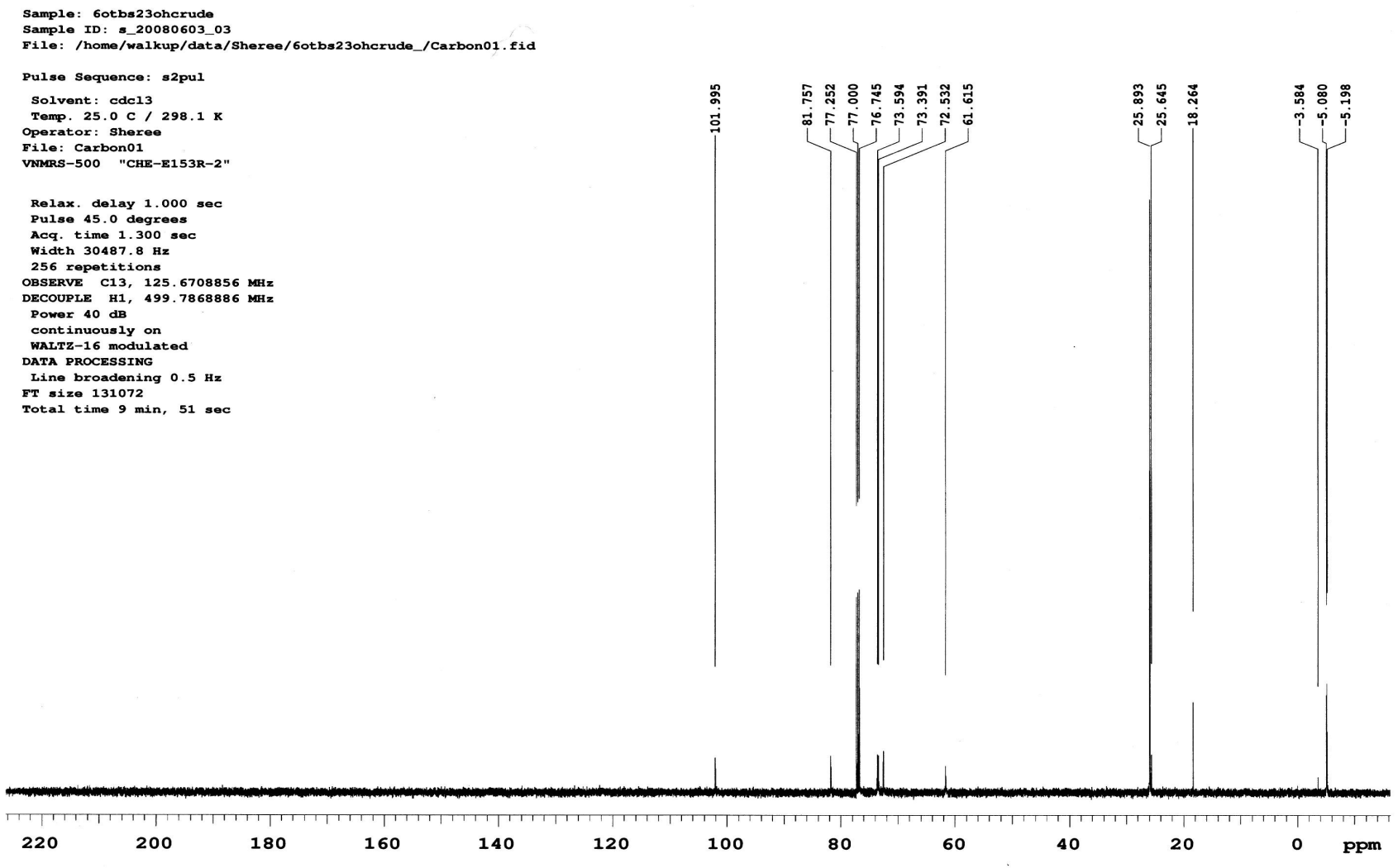
APPENDICES

APPENDIX A

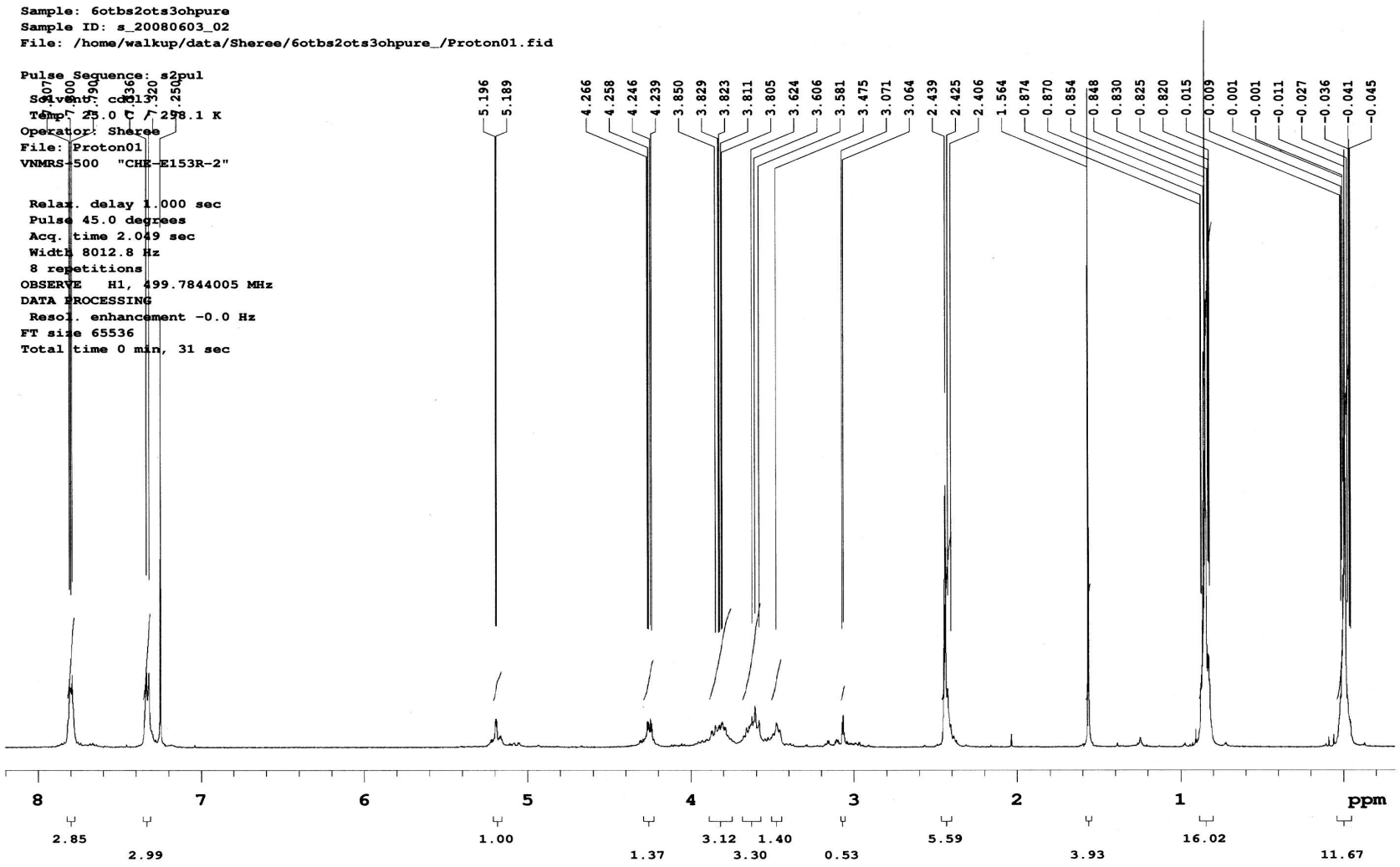
Selected NMR Spectra



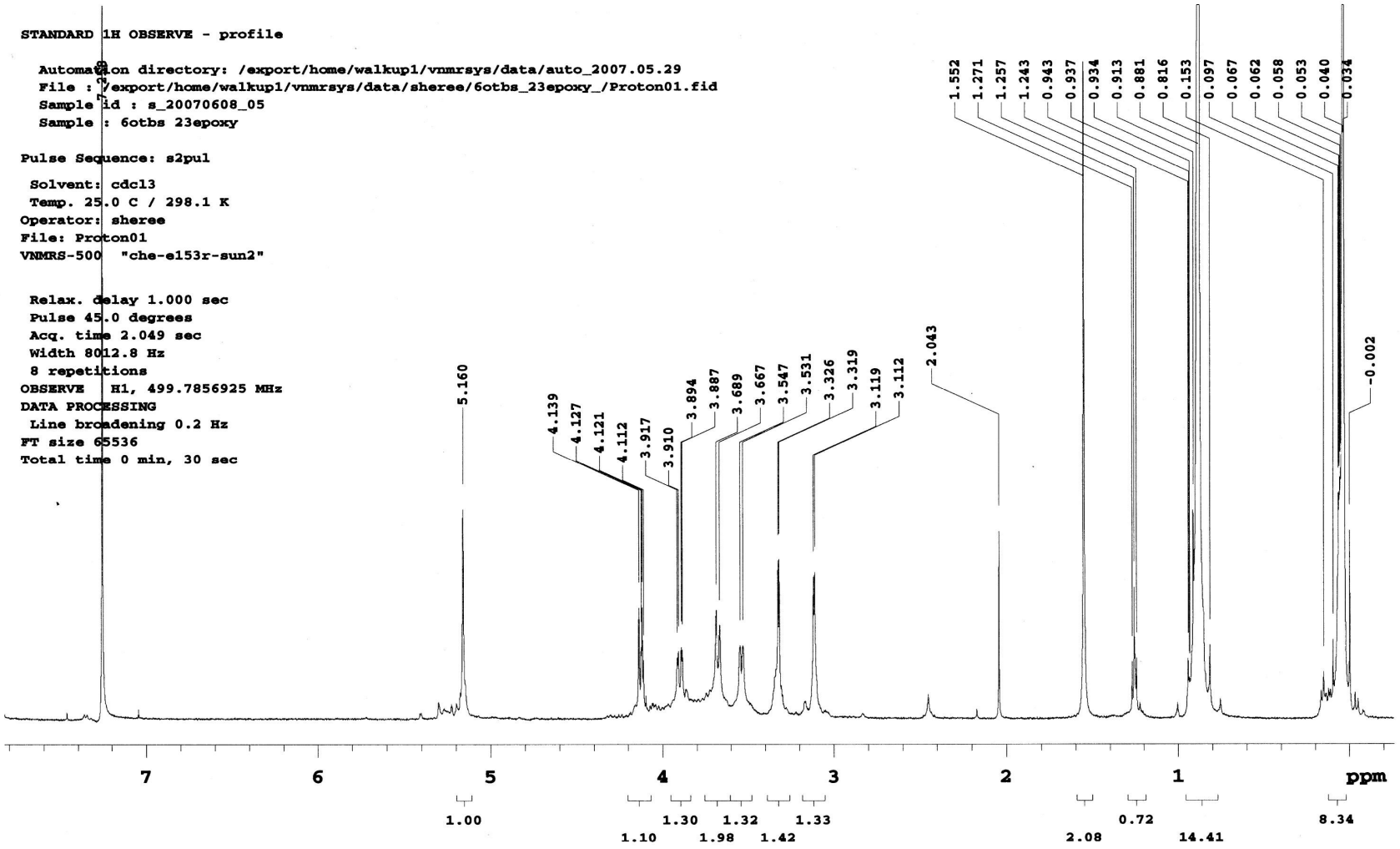
A.2.1 ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-dihydroxy)-β-CD (**3**).



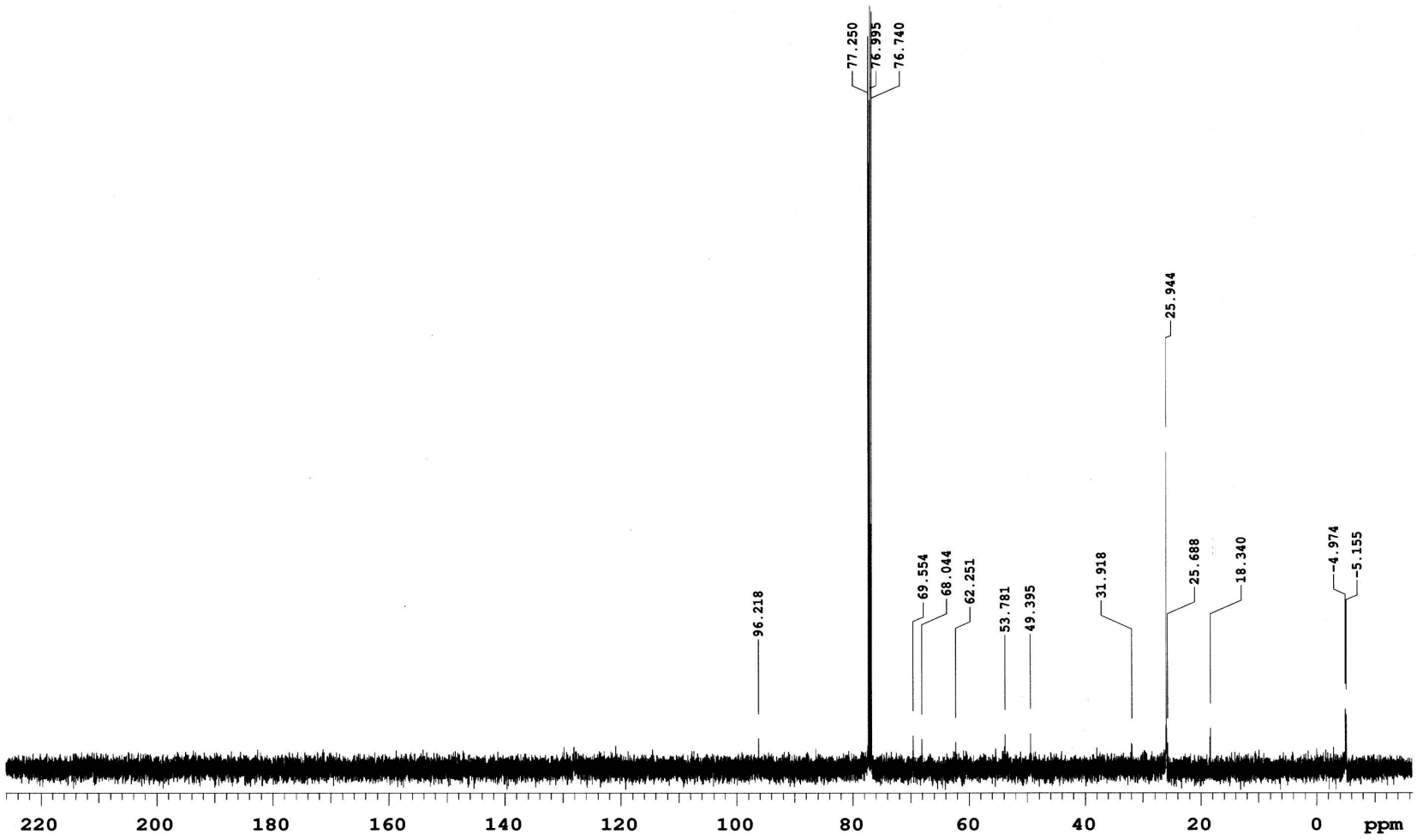
A.2.2. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-dihydroxy)-β-CD (**3**).



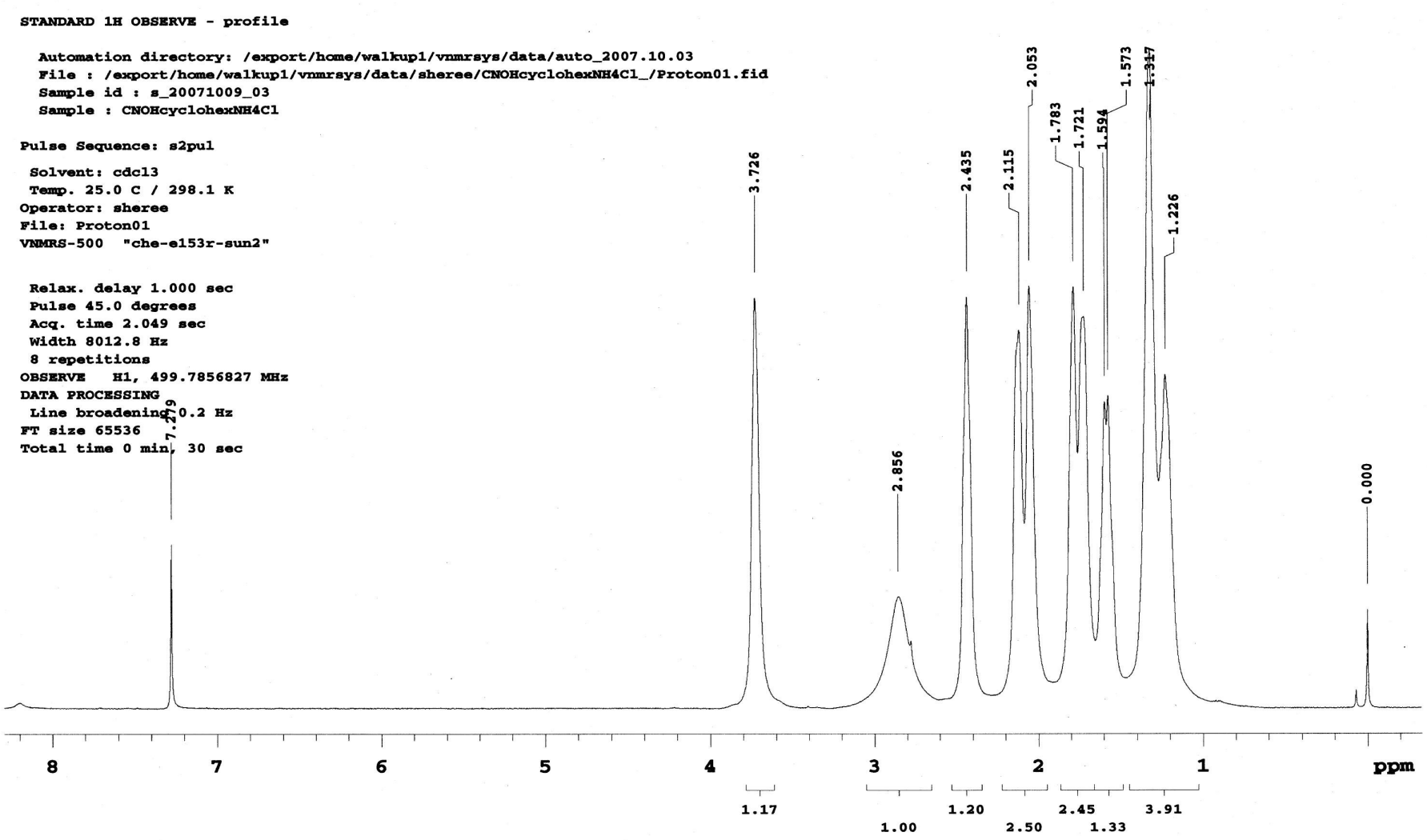
A.3.1. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2-*O*-tosyl)-β-CD (**12**).



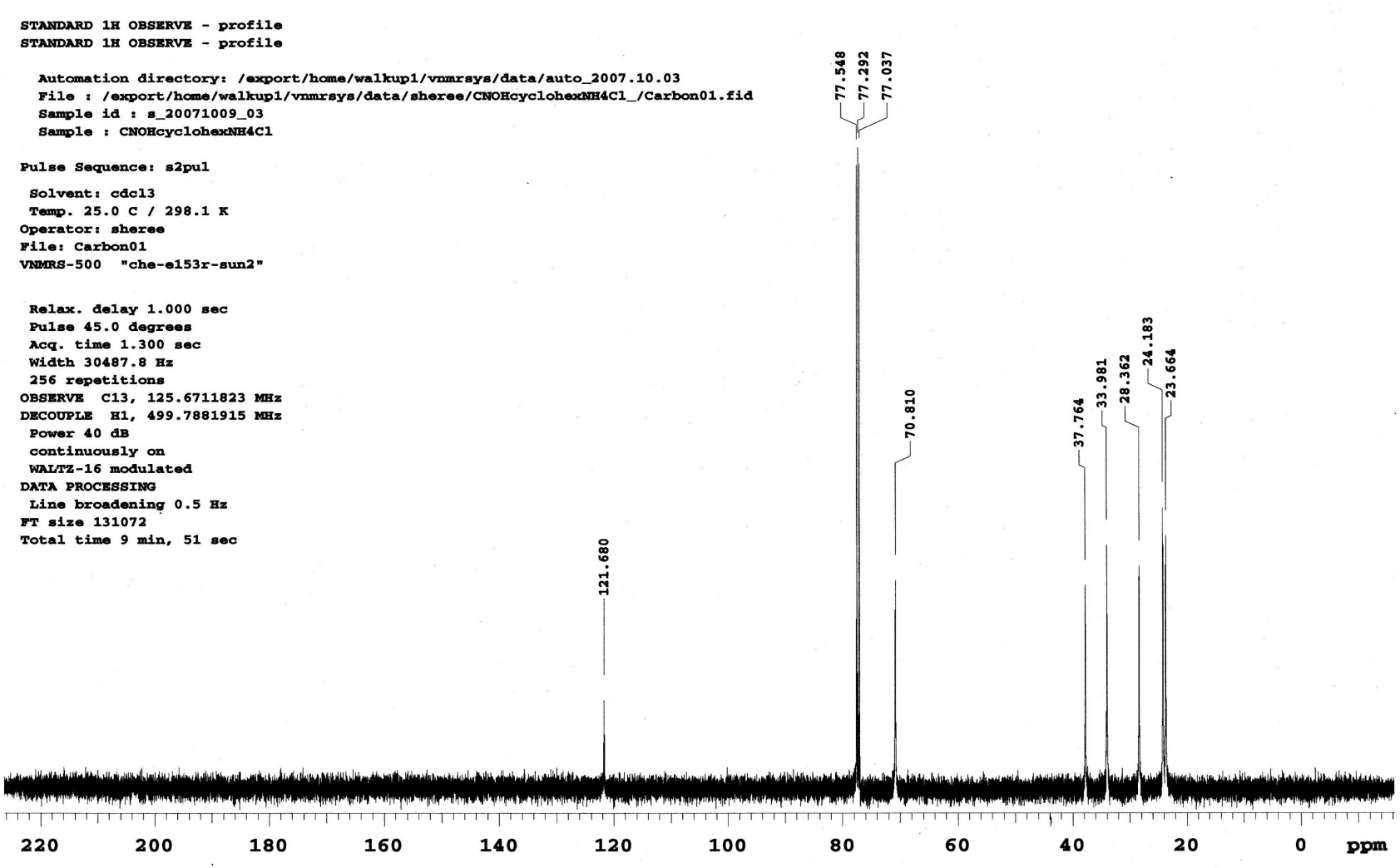
A.3.2. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-cyclomannoepoxide)-β-CD (**13**).



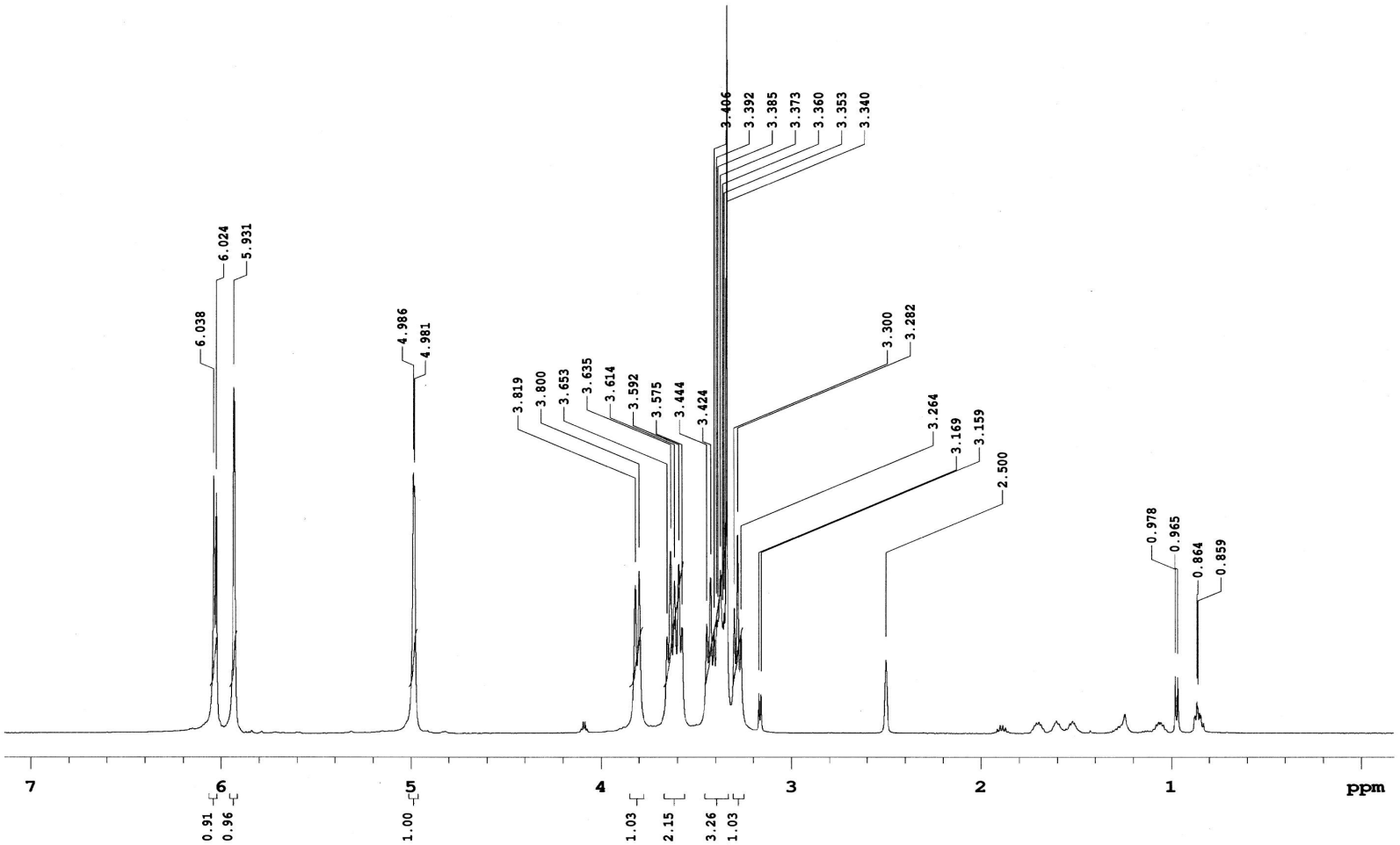
A.3.3. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-cyclomannoepoxide)-β-CD (**13**).



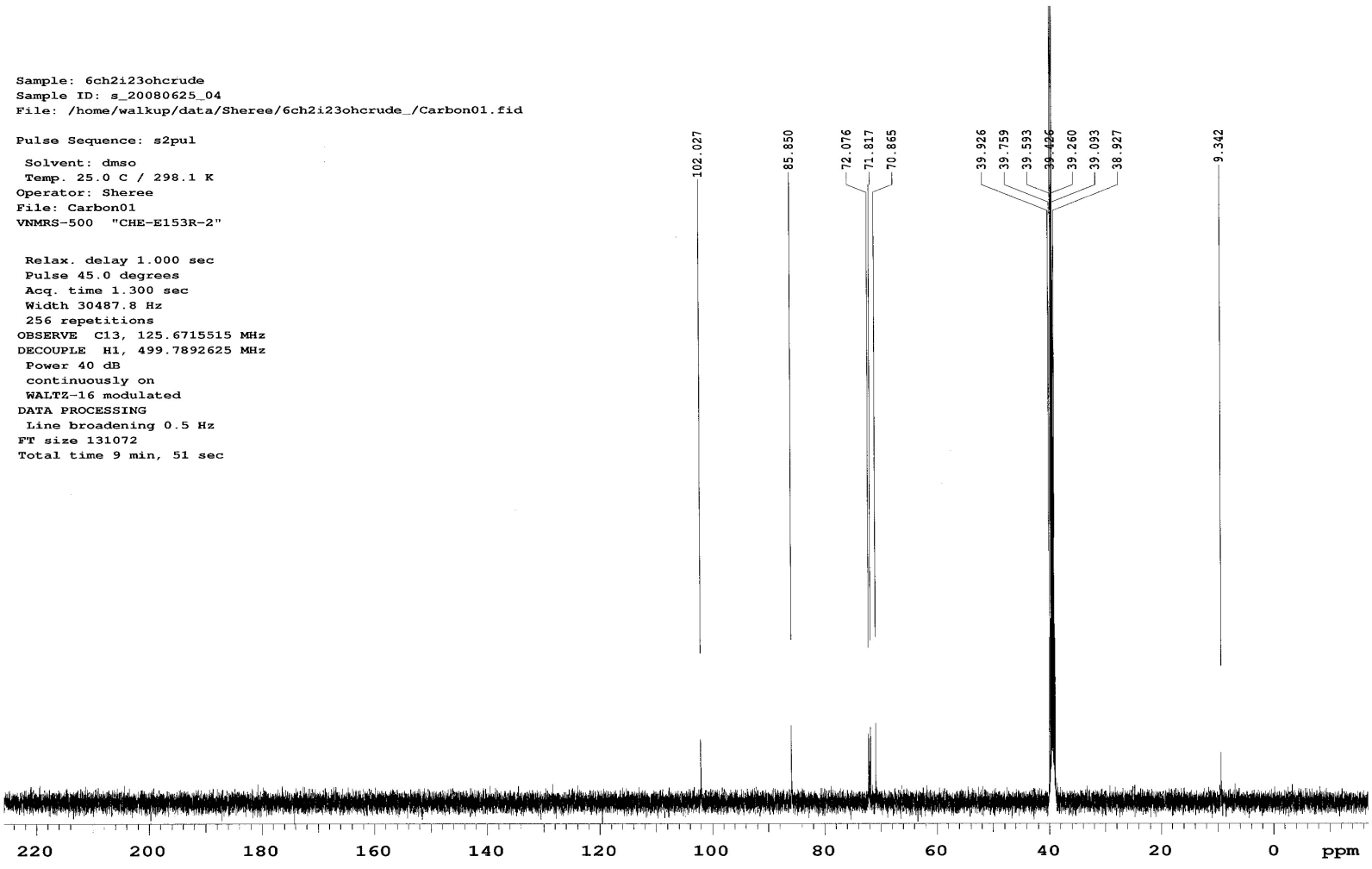
A.3.4. ^{1}H NMR ($CDCl_3$, 500 MHz) of *trans*-hydroxy-cyclohexanecarbonitrile.



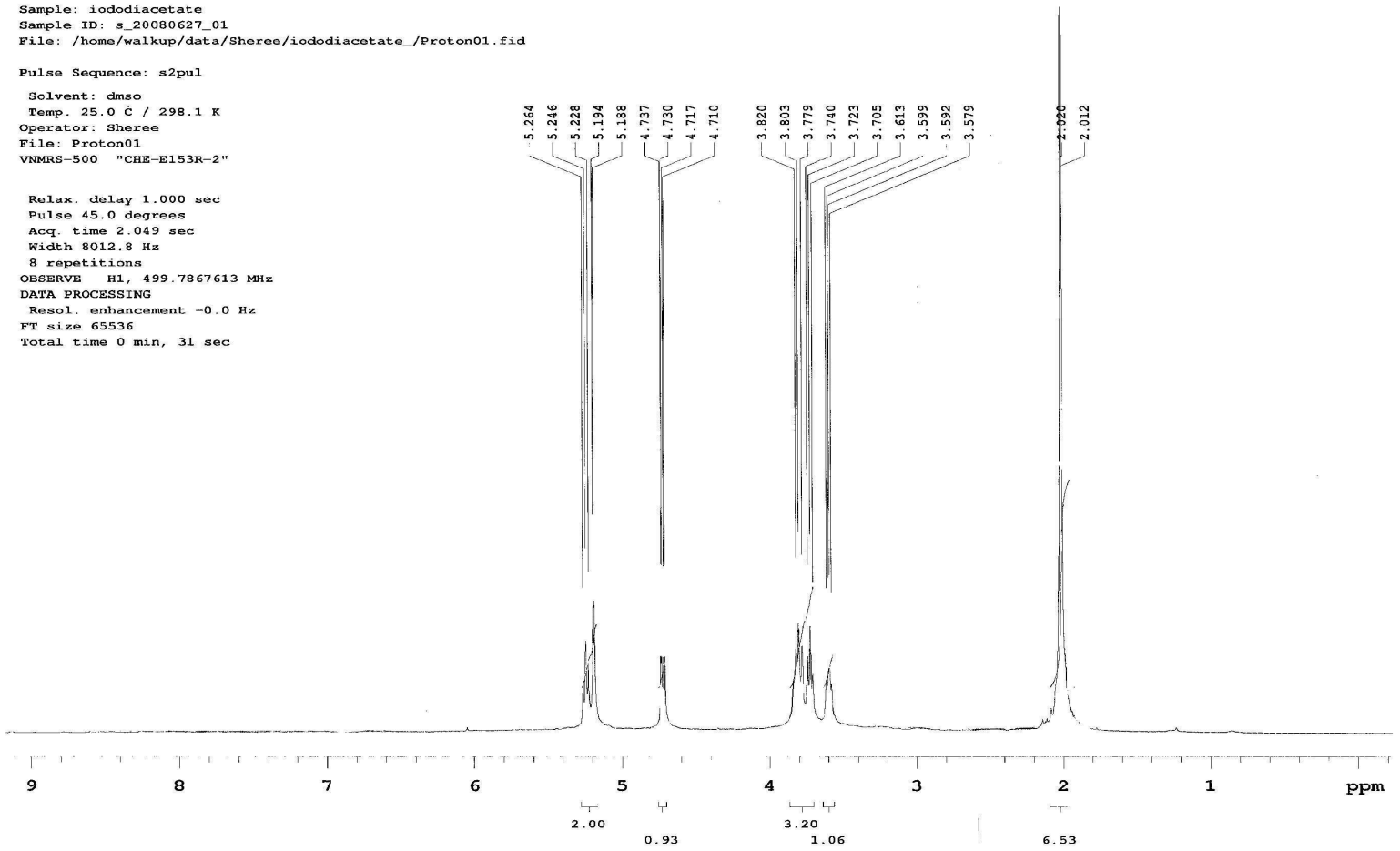
A.3.5. ^{13}C NMR ($CDCl_3$, 125 MHz) of *trans*-hydroxy-cyclohexanecarbonitrile.



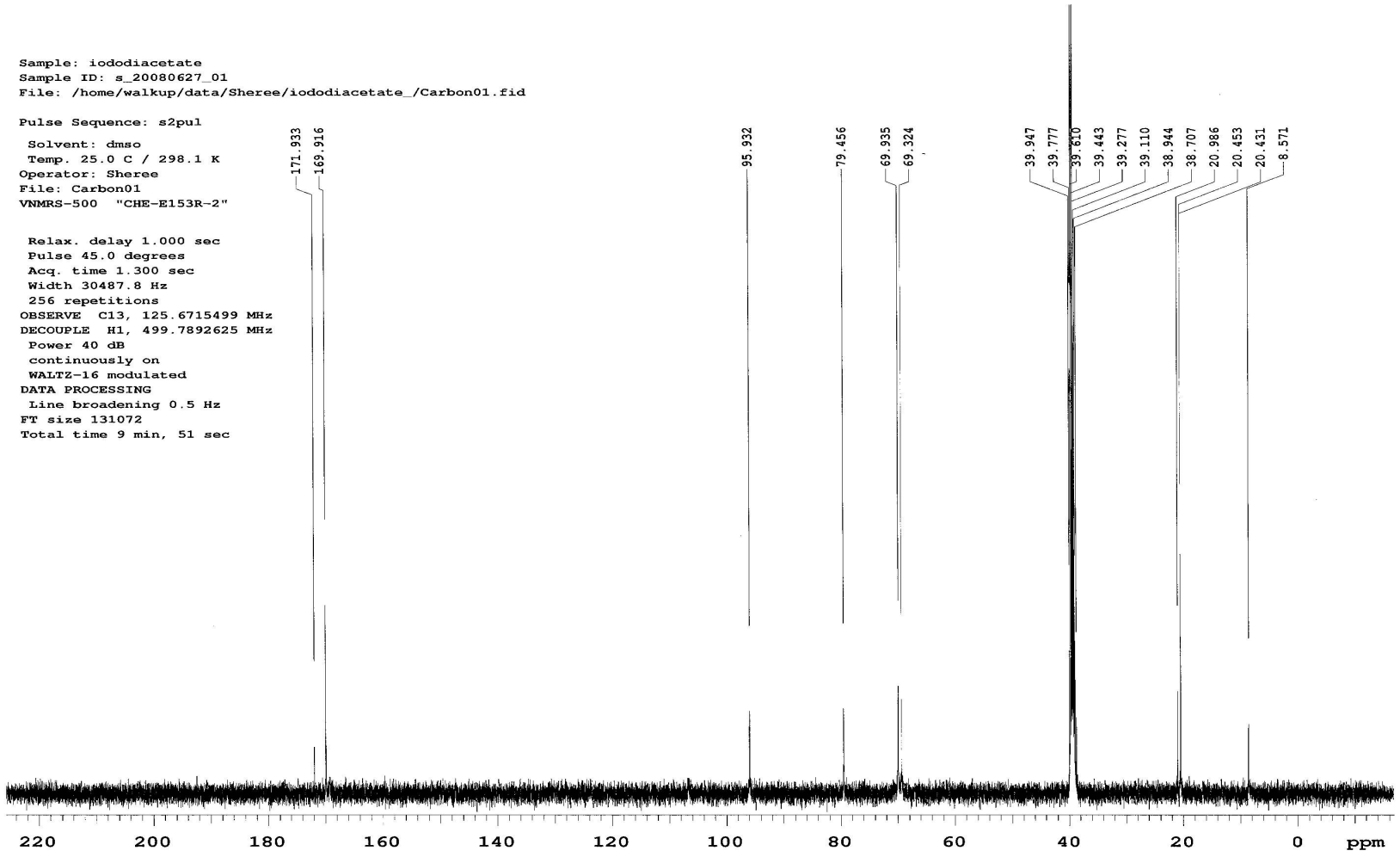
A.4.1. ^{1}H NMR (DMSO, 500 MHz) of *per*(6-*O*-*iodo*-2,3-dihydroxy)-β-CD (**15**).



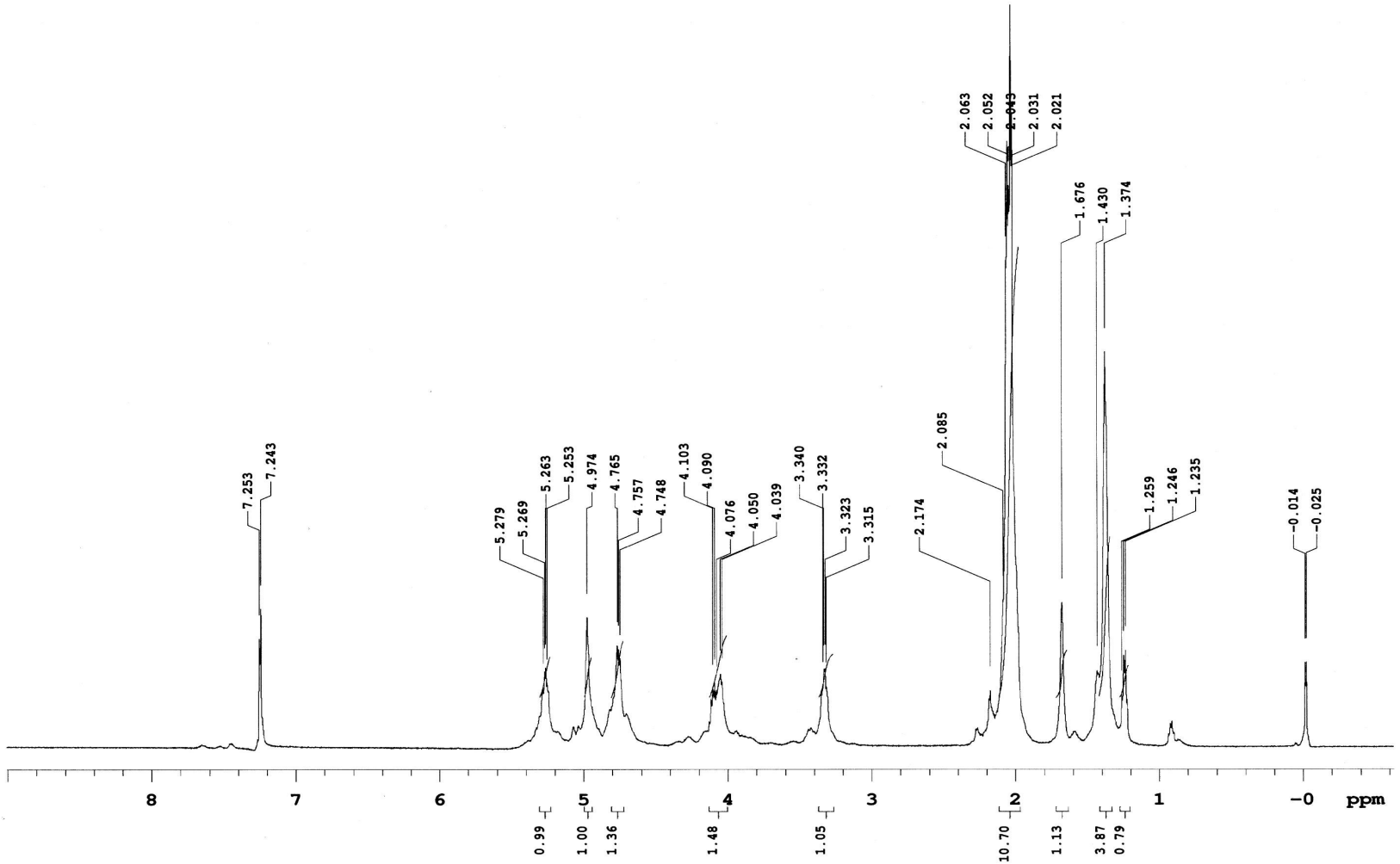
A.4.2. ^{13}C NMR (DMSO, 125 MHz) of *per*(6-*O*-*iodo*-2,3-dihydroxy)-β-CD (**15**).



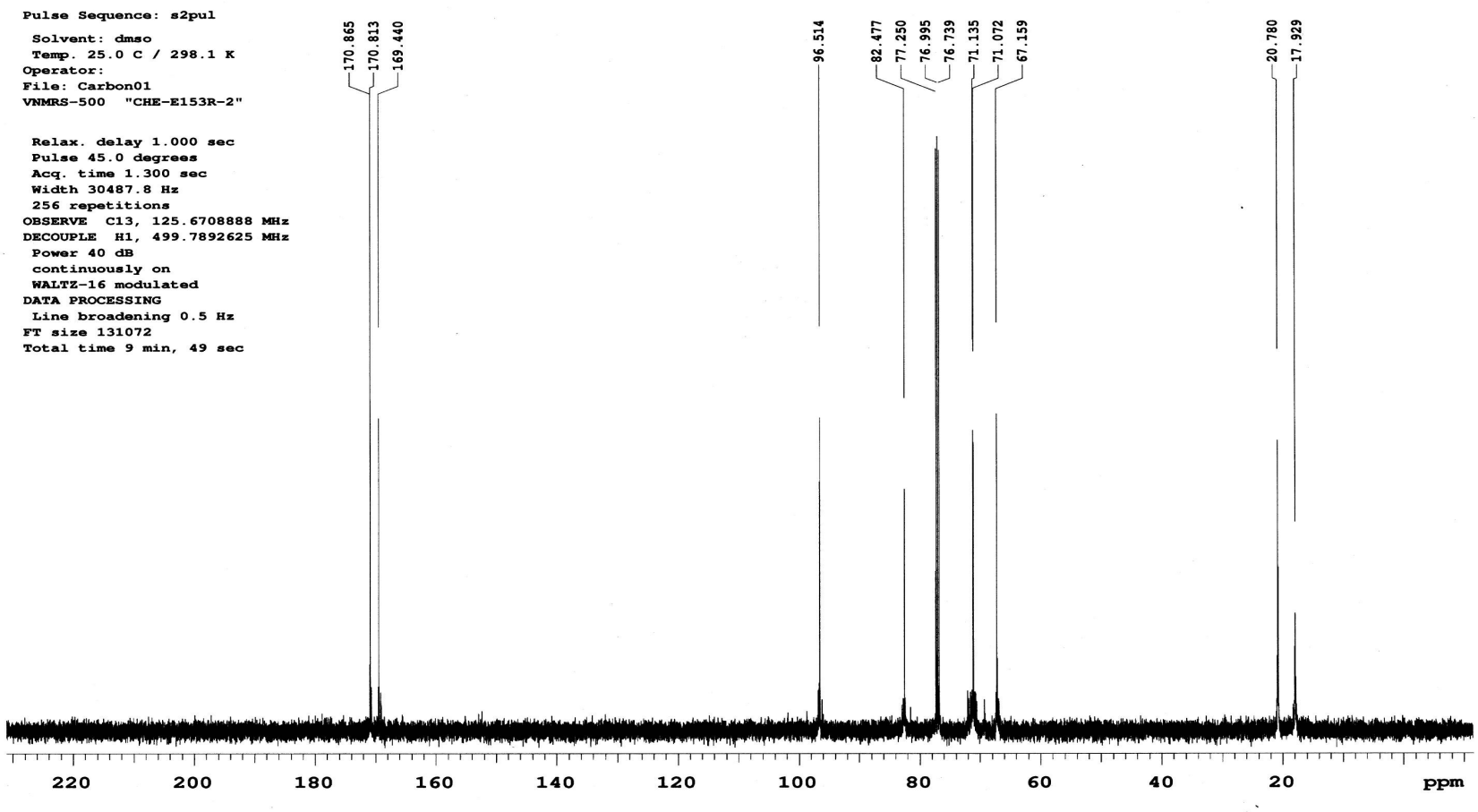
A.4.3. ^{1}H NMR (DMSO, 500 MHz) of *per*(6-*O*-*iodo*-2,3-diacetoxy)-β-CD (**16**).



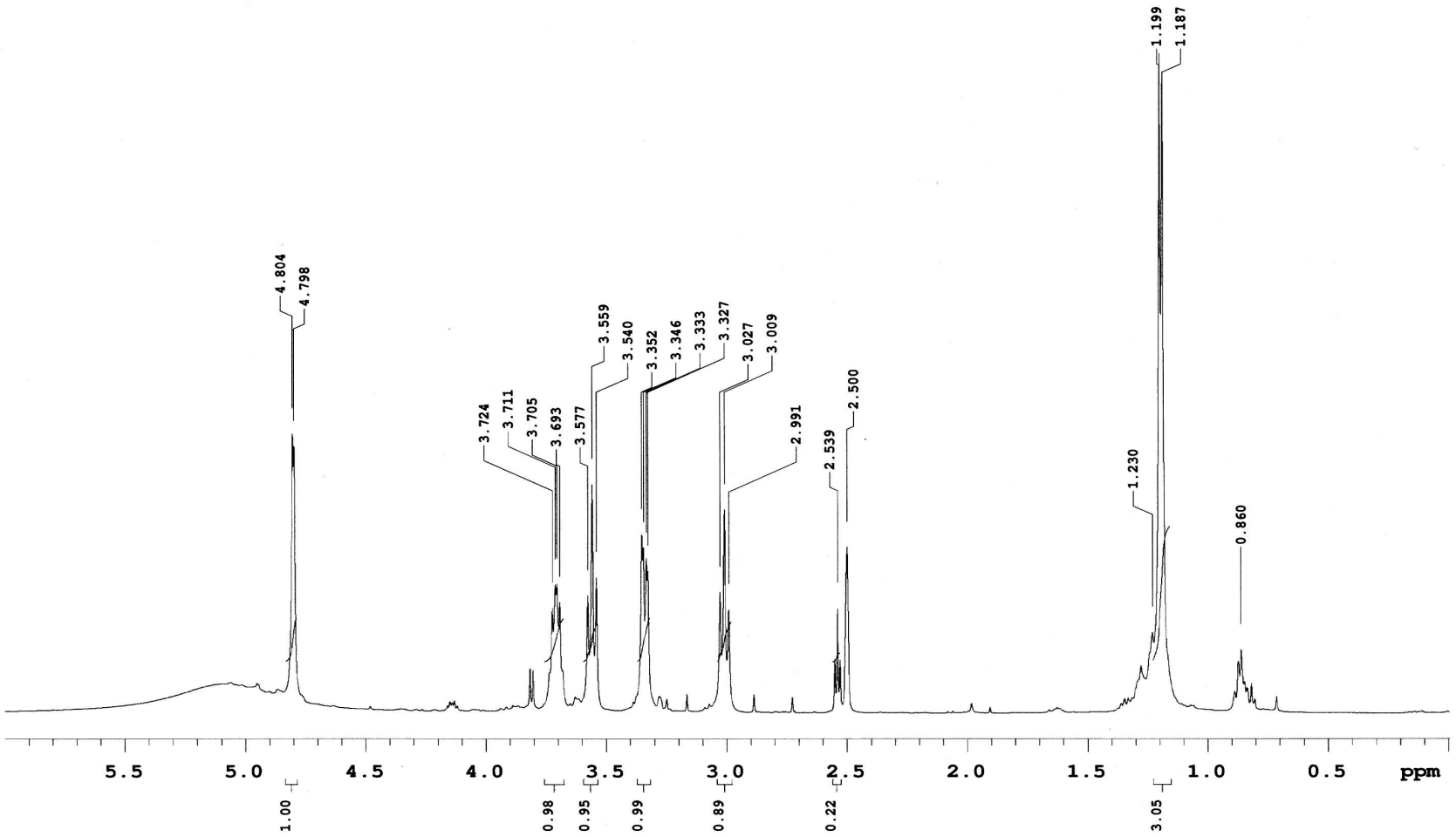
A.4.4 ^{13}C NMR (DMSO, 125 MHz) of *per*(6-*O*-*iodo*-2,3-diacetoxy)-β-CD (**16**).



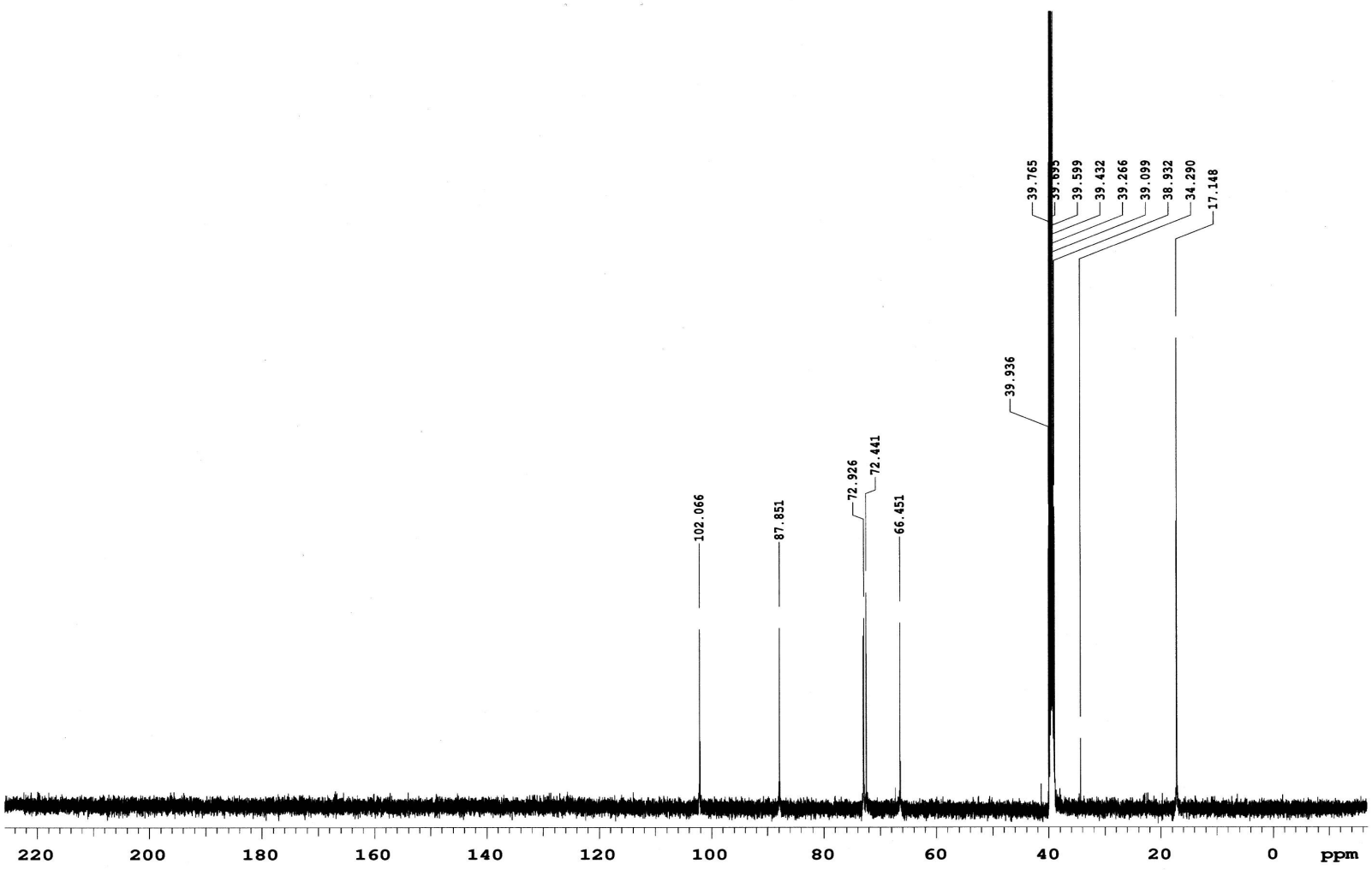
A.4.5. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-deoxy-2,3-diacetoxy)-β-CD (**17**).



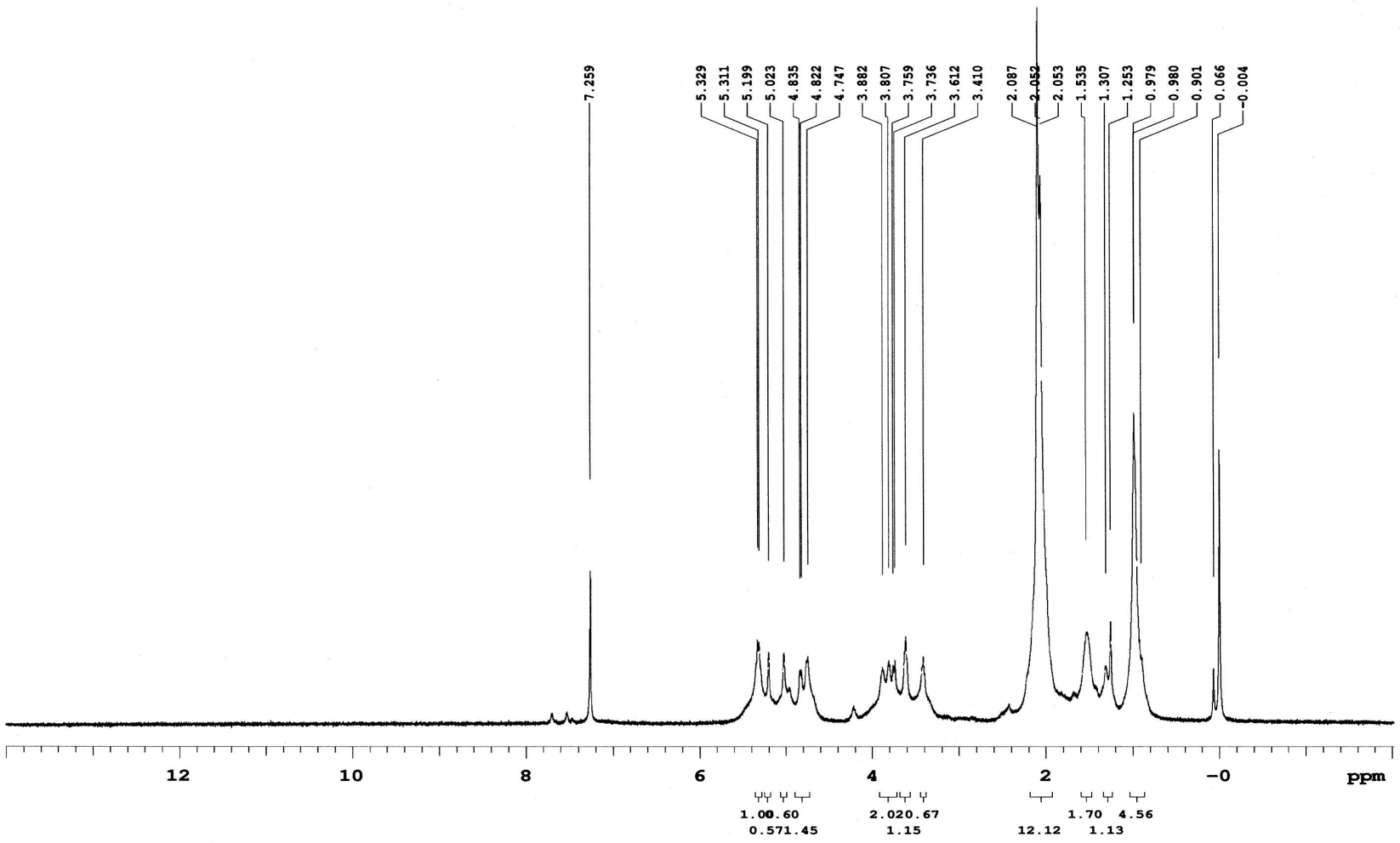
A.4.6. ^{13}C NMR (DMSO, 125 MHz) of *per*(6-deoxy-2,3-diacetoxy)-β-CD (**17**).



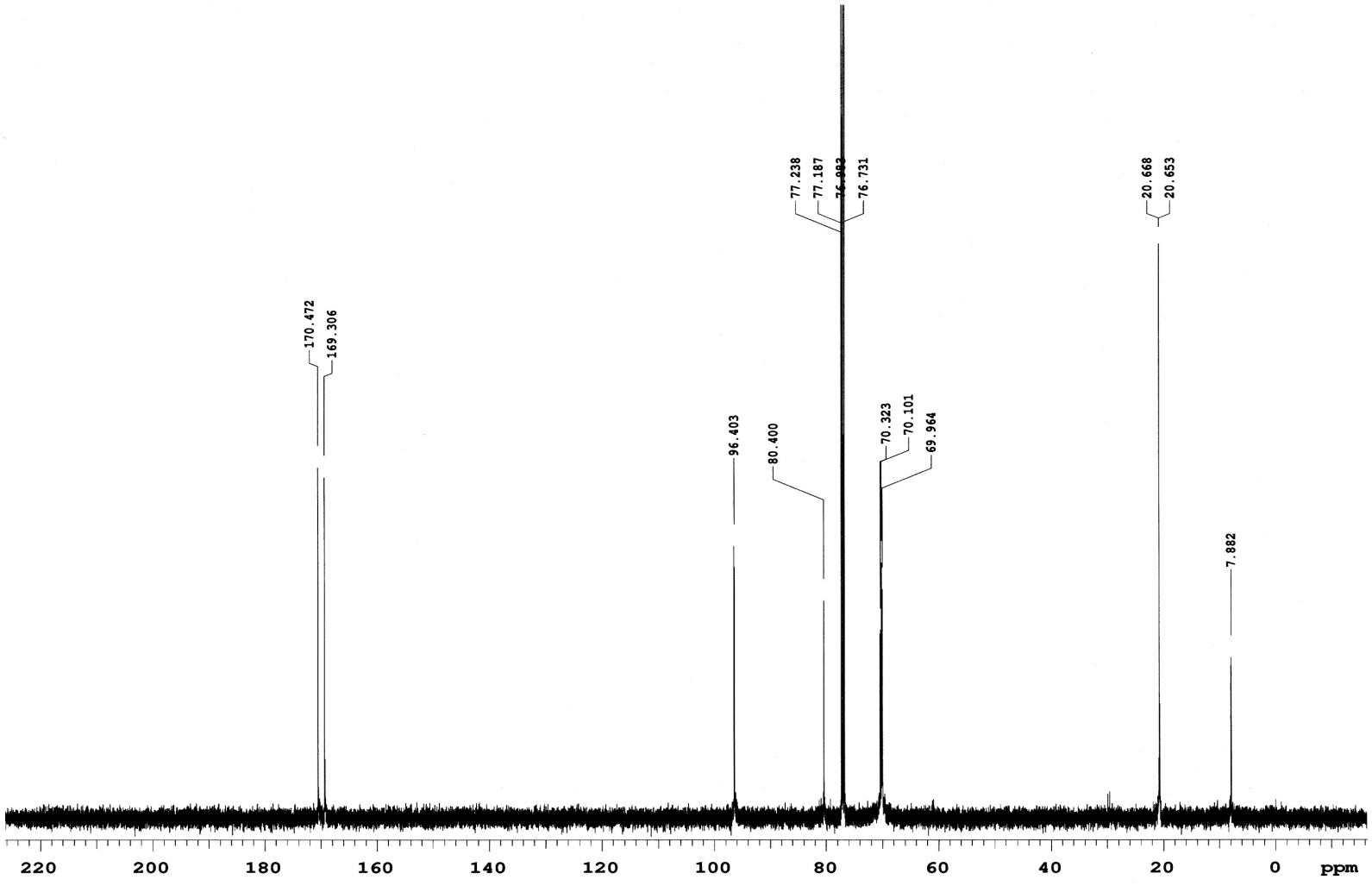
A.4.7. ^{1}H NMR (DMSO, 500 MHz) of *per*(6-deoxy)-β-CD (**14**).



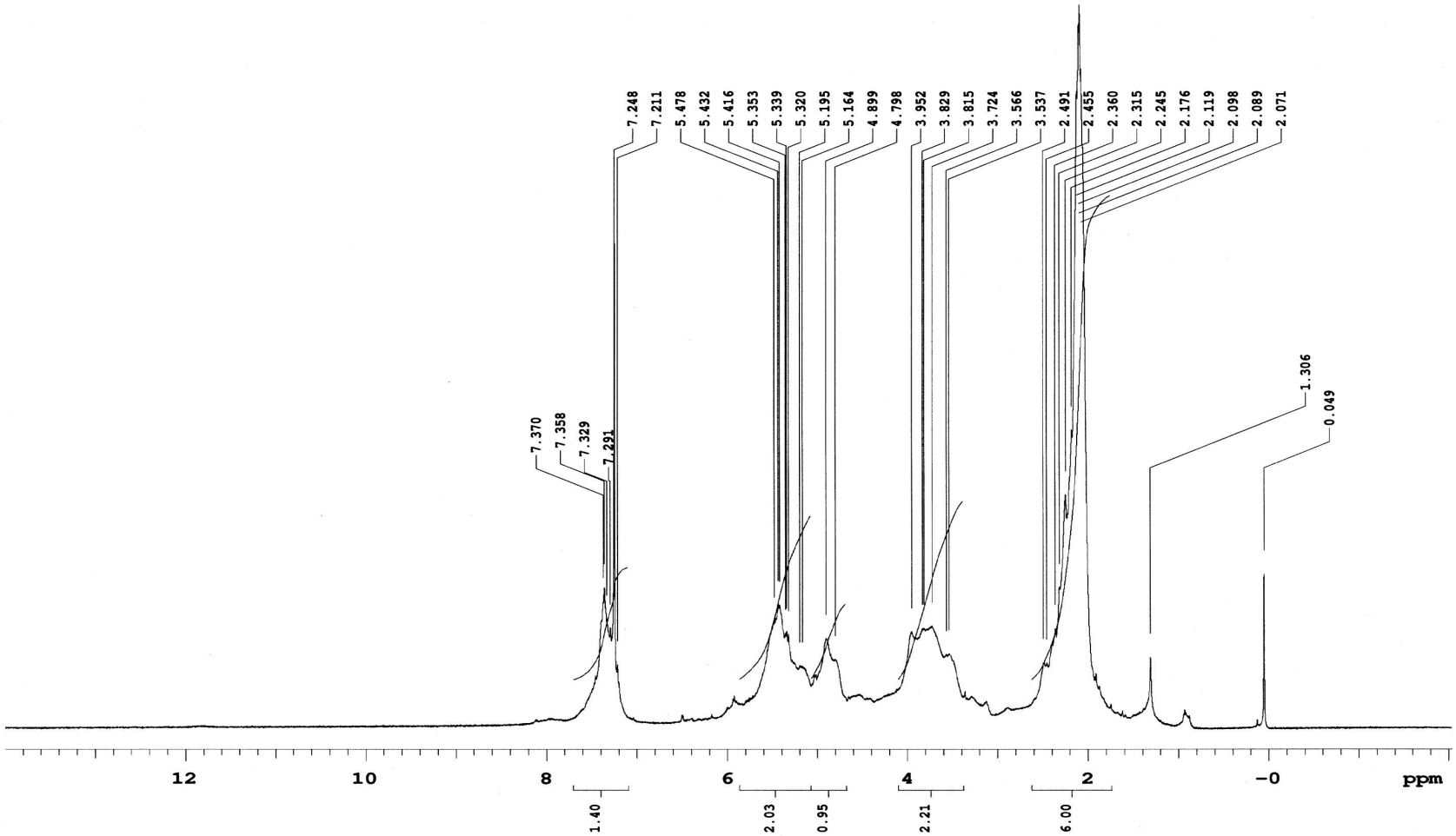
A.4.8. ^{13}C NMR (DMSO, 125 MHz) of *per*(6-deoxy)-β-CD (**14**).



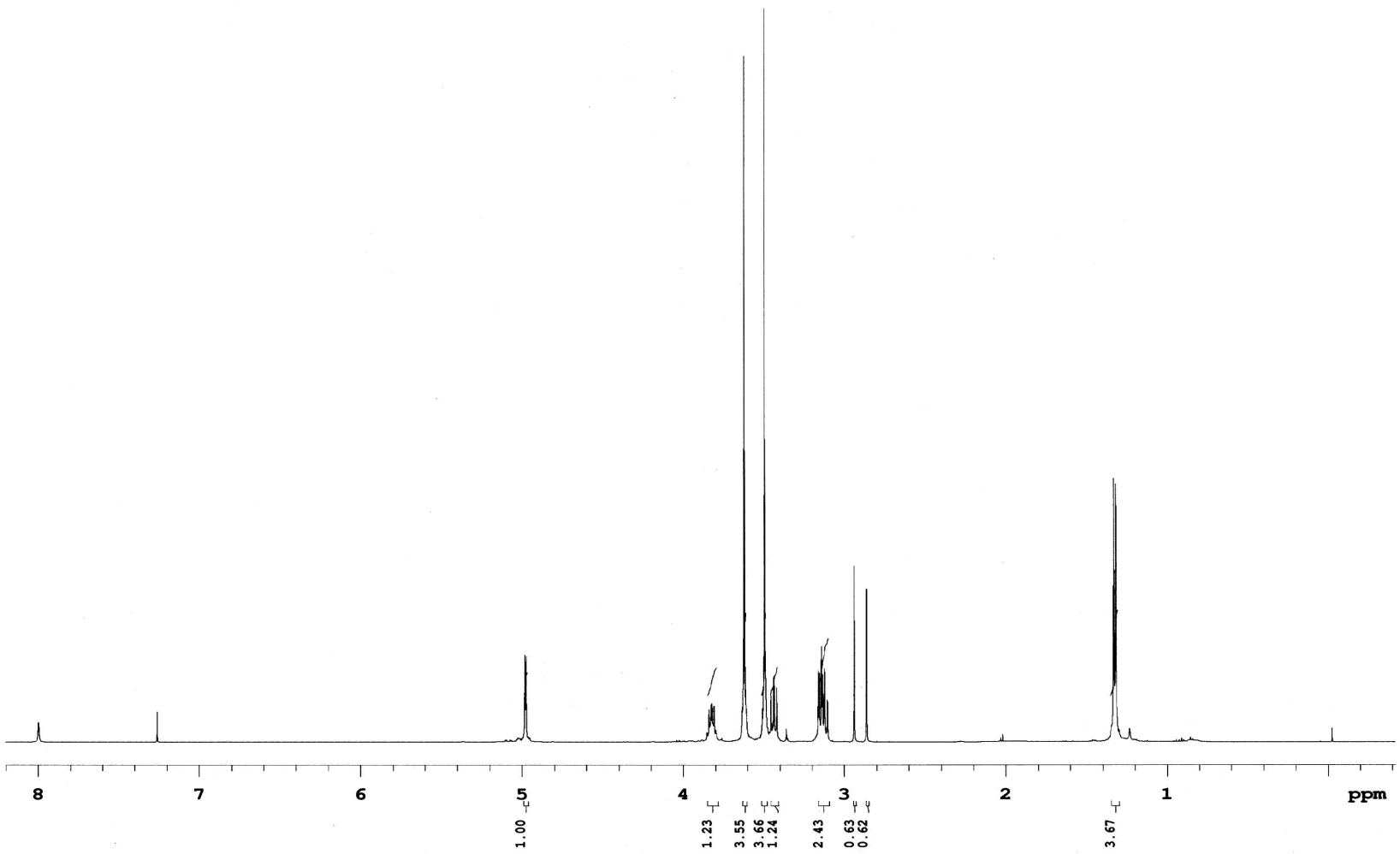
A.4.9. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-methyl-2,3-diacetoxy)-β-CD (**18**).



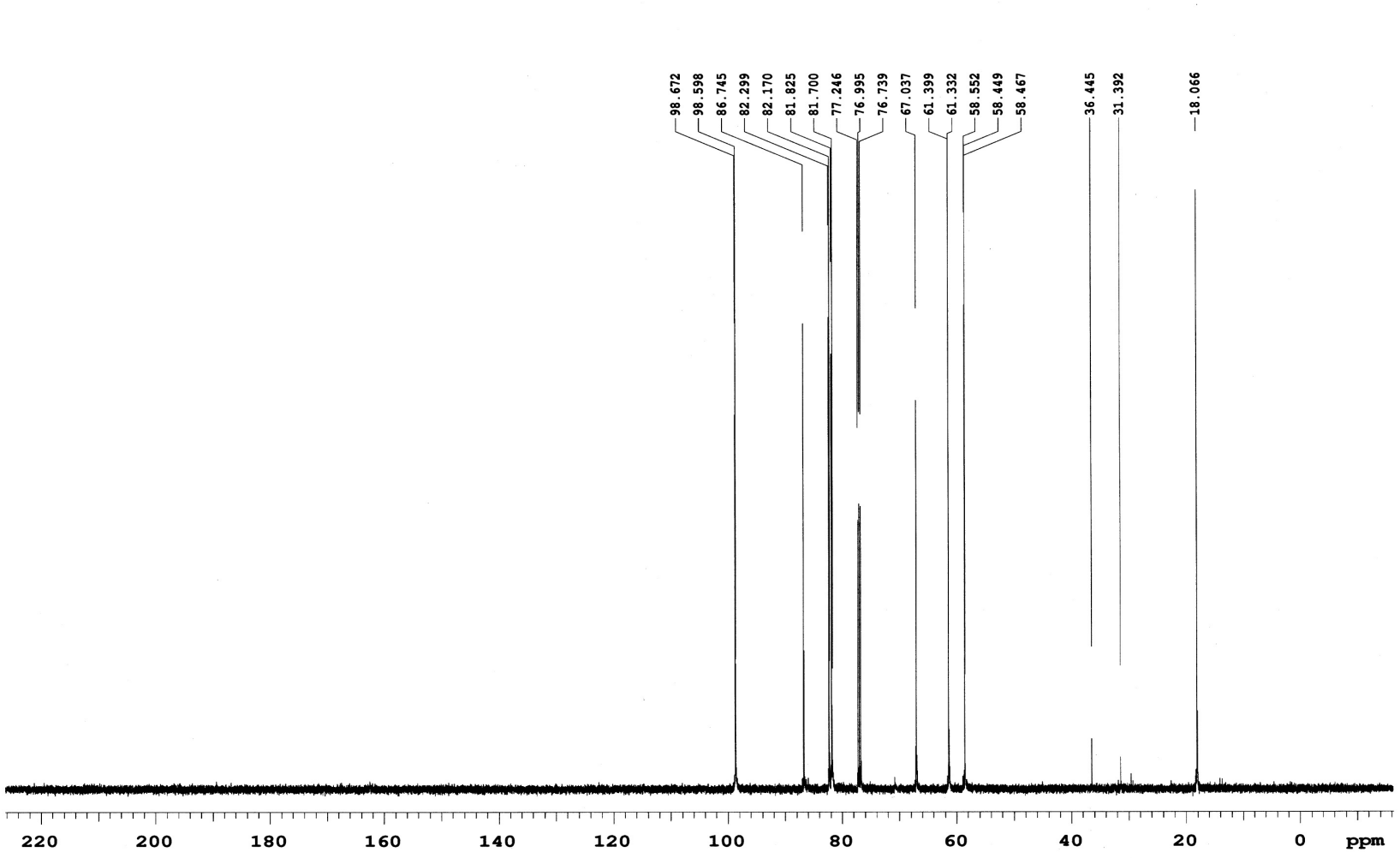
A.4.10. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-methyl-2,3-diacetoxy)-β-CD (**18**).



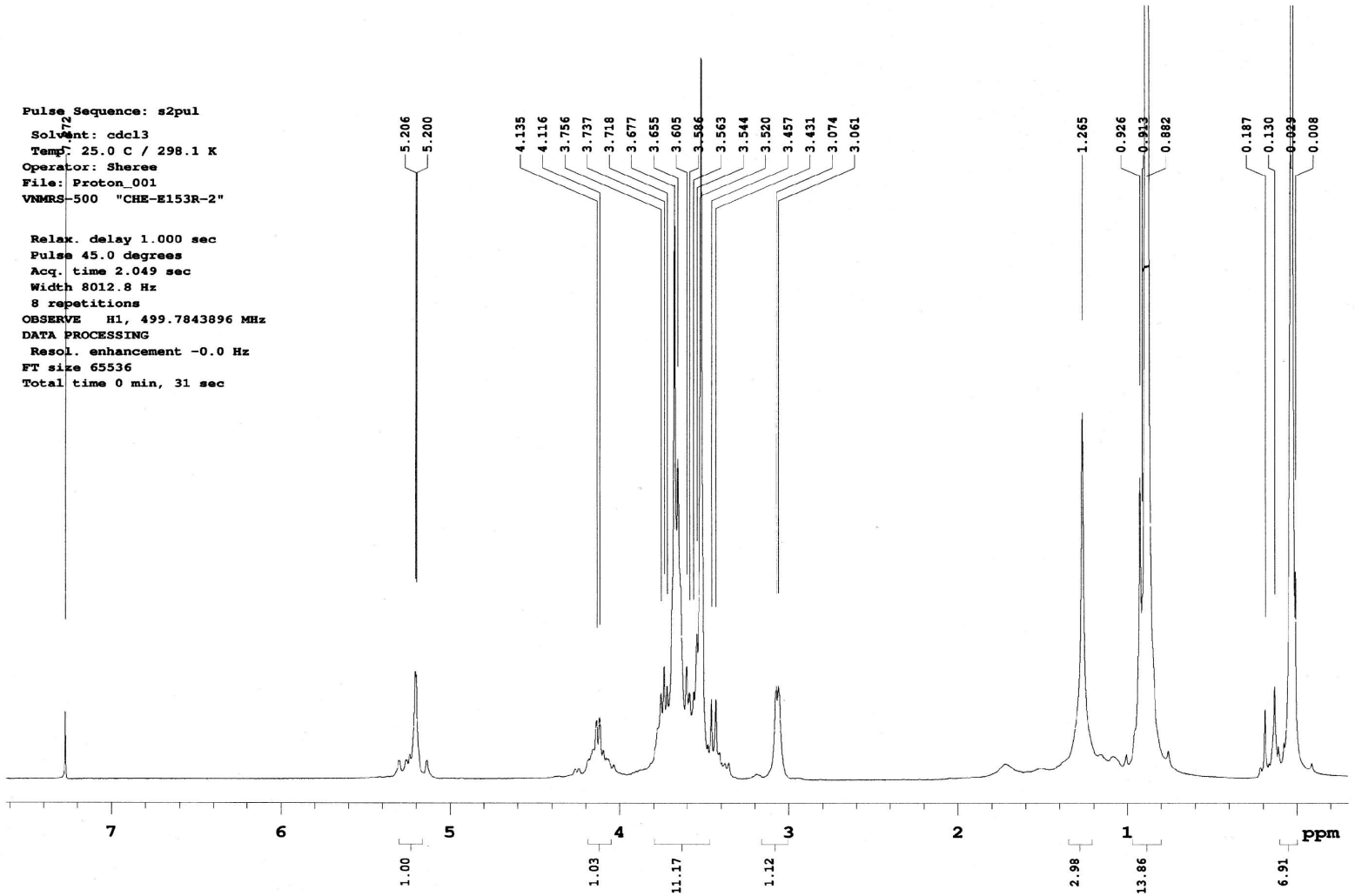
A.4.11. ^{1}H NMR (DMF-d_6, 500 MHz) of *per*(6-phenyl-2,3-diacetoxy)-β-CD (**19**).



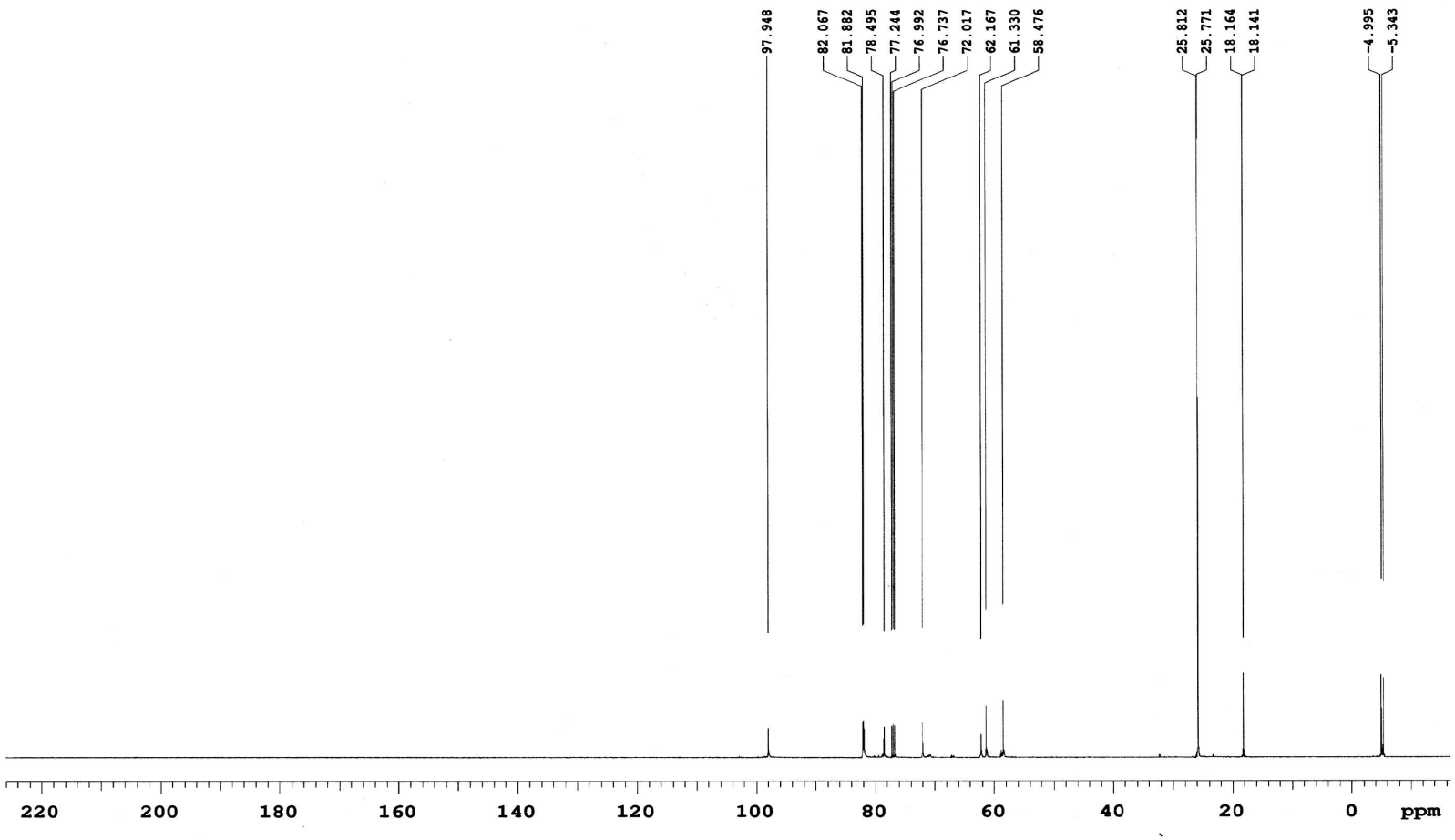
A.4.12. ^{1}H NMR (DMSO-d_6, 500 MHz) of *per*(6-deoxy-2,3-dimethoxy)-β-CD (**20**).



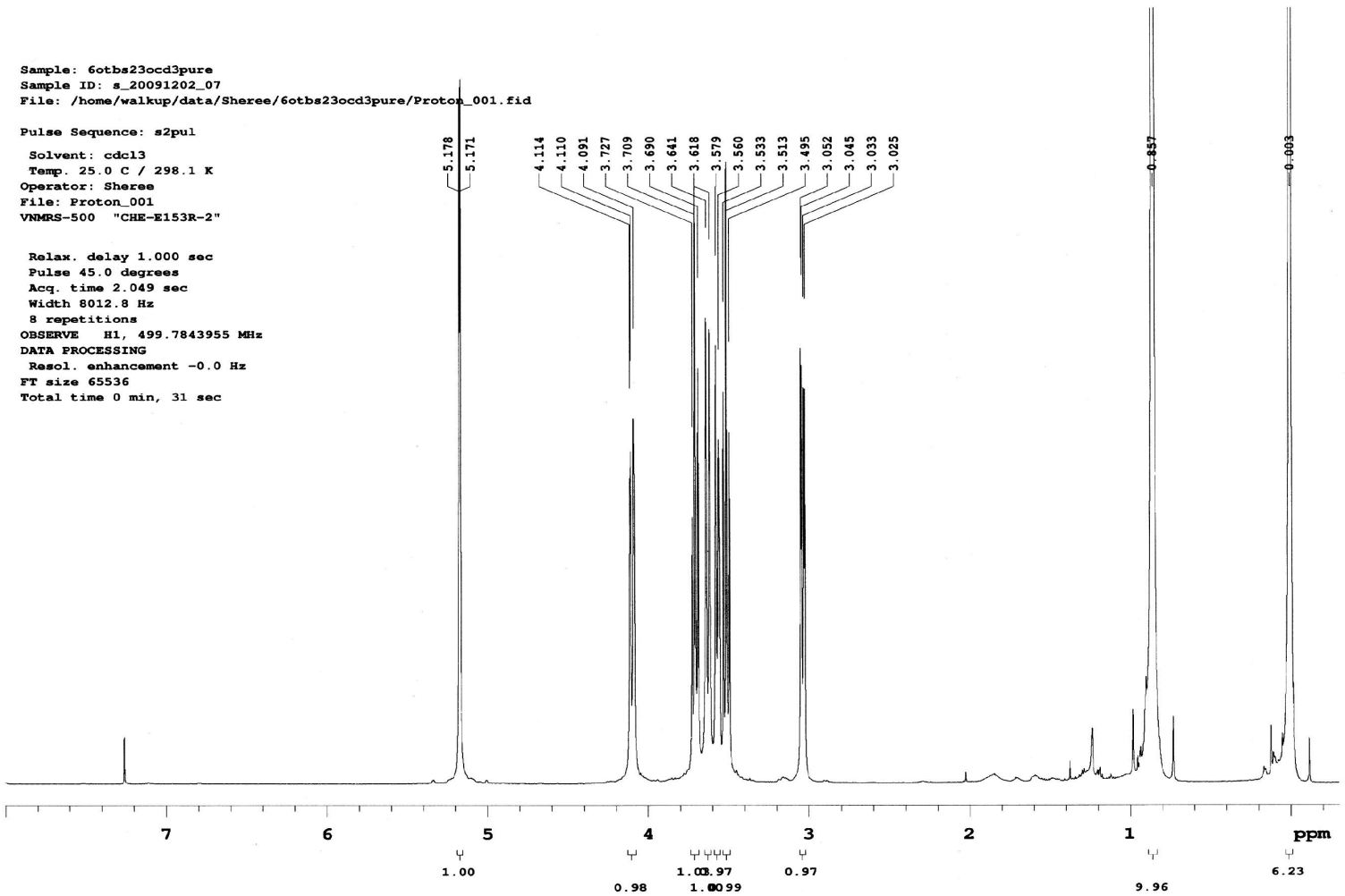
A.4.13. ^{13}C NMR (DMSO-d_6, 125 MHz) of *per*(6-deoxy-2,3-dimethoxy)-β-CD (**20**).



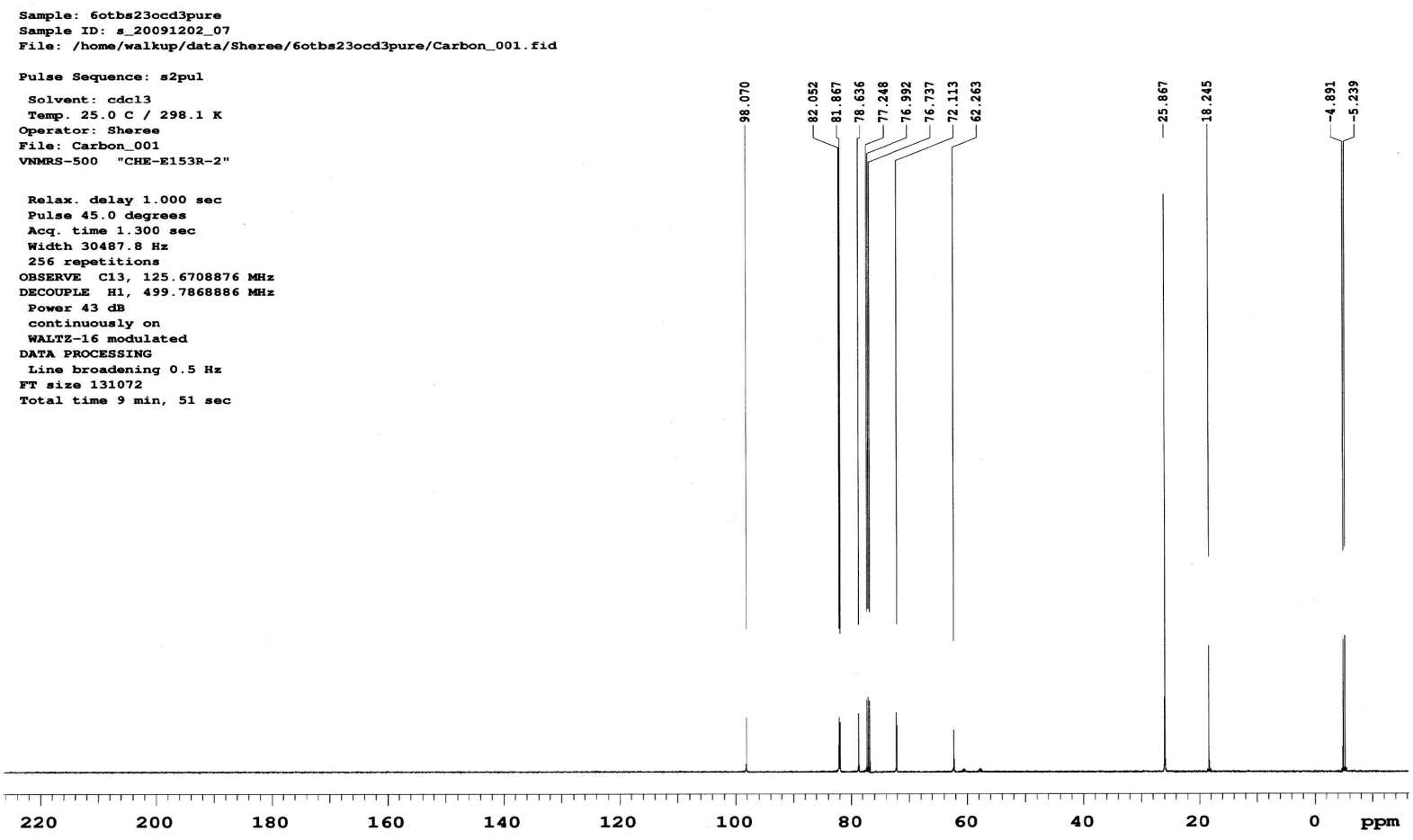
A.4.14. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-dimethoxy)-β-CD (**21**).



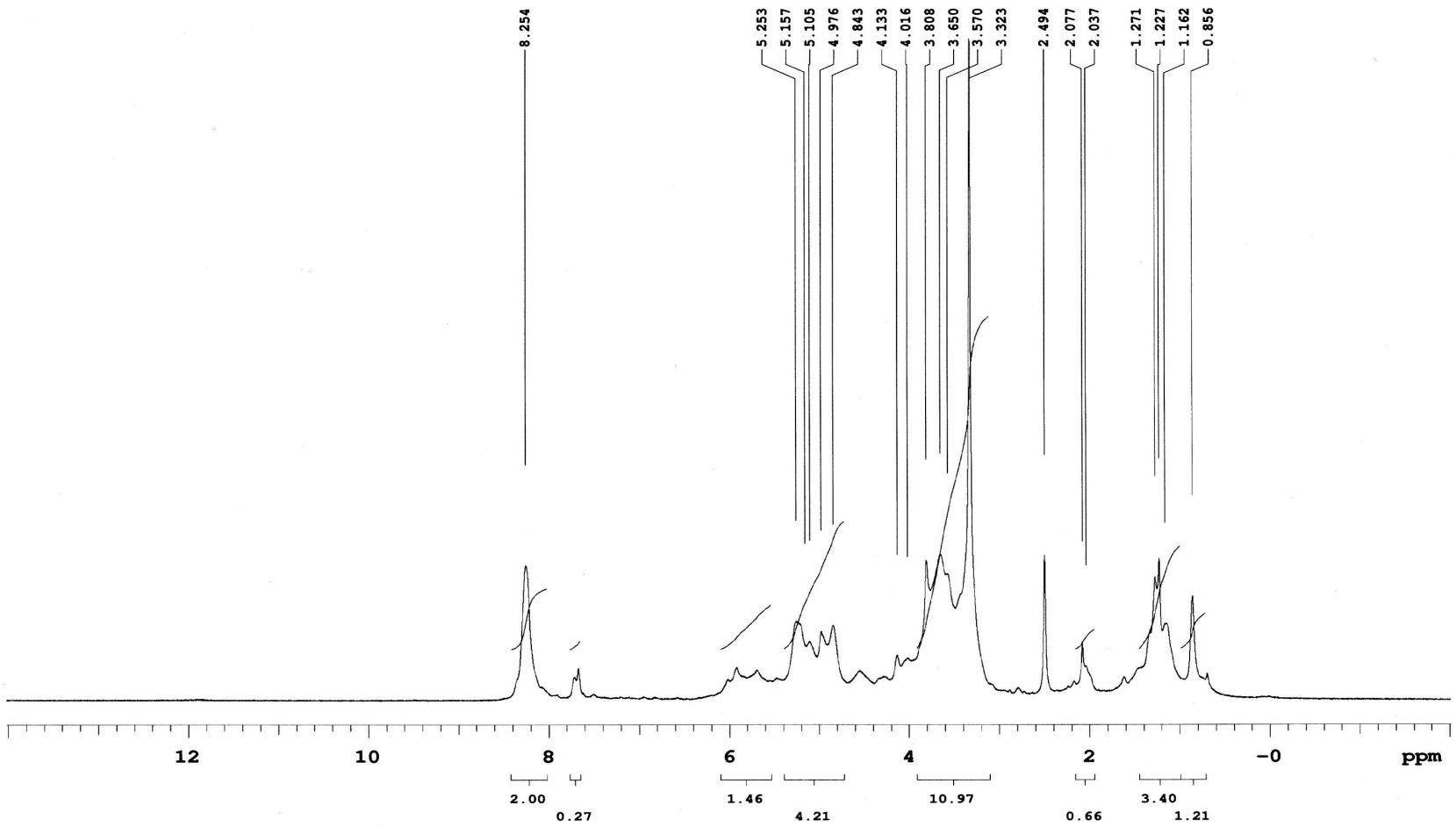
A.4.15. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-dimethoxy)-β-CD (**21**).



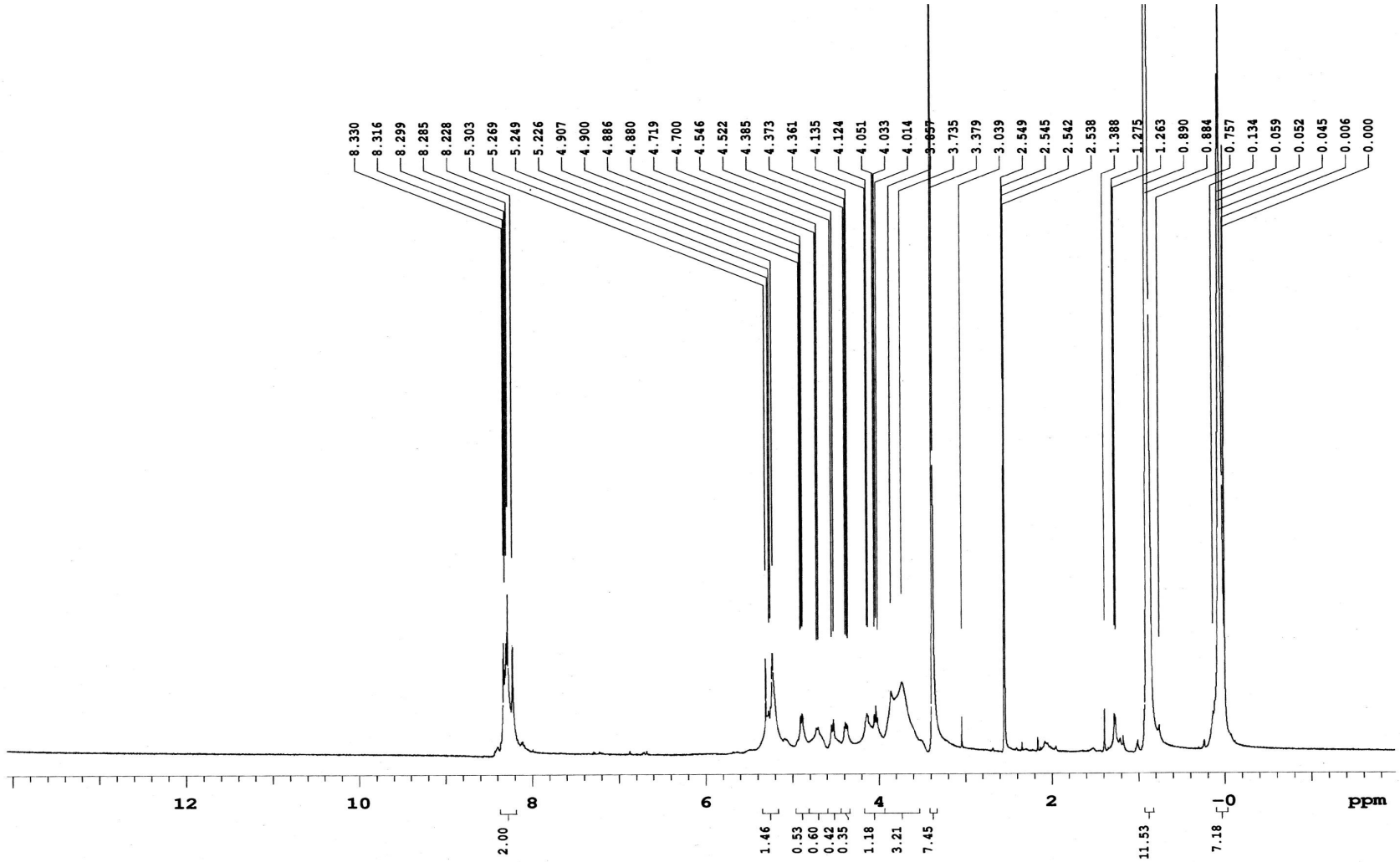
A.4.16. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-bis-($^{2}H_3$)methoxy)-β-CD (**22**).



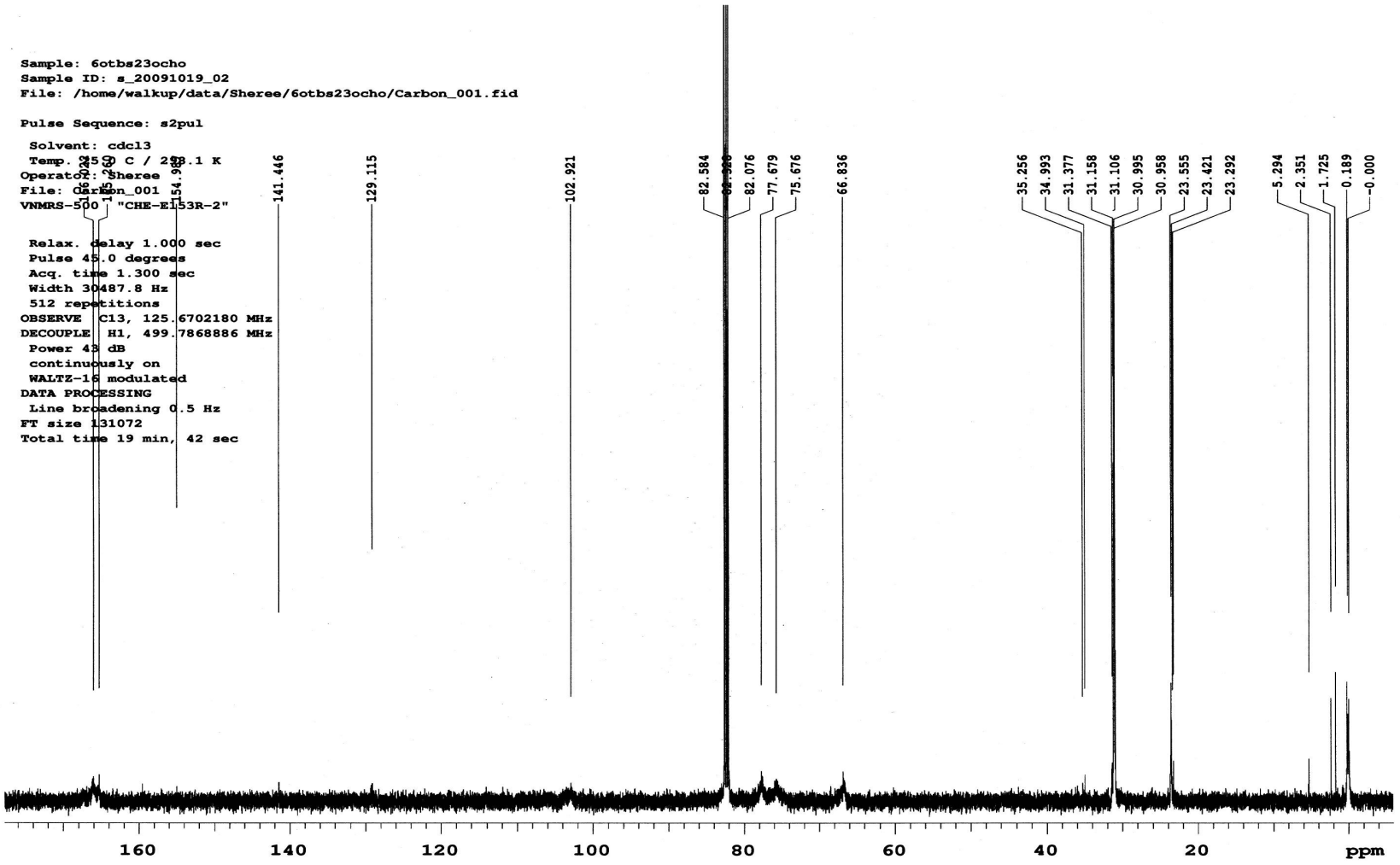
A.4.17. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-bis-($^2H_3$)methoxy)-β-CD (**22**).



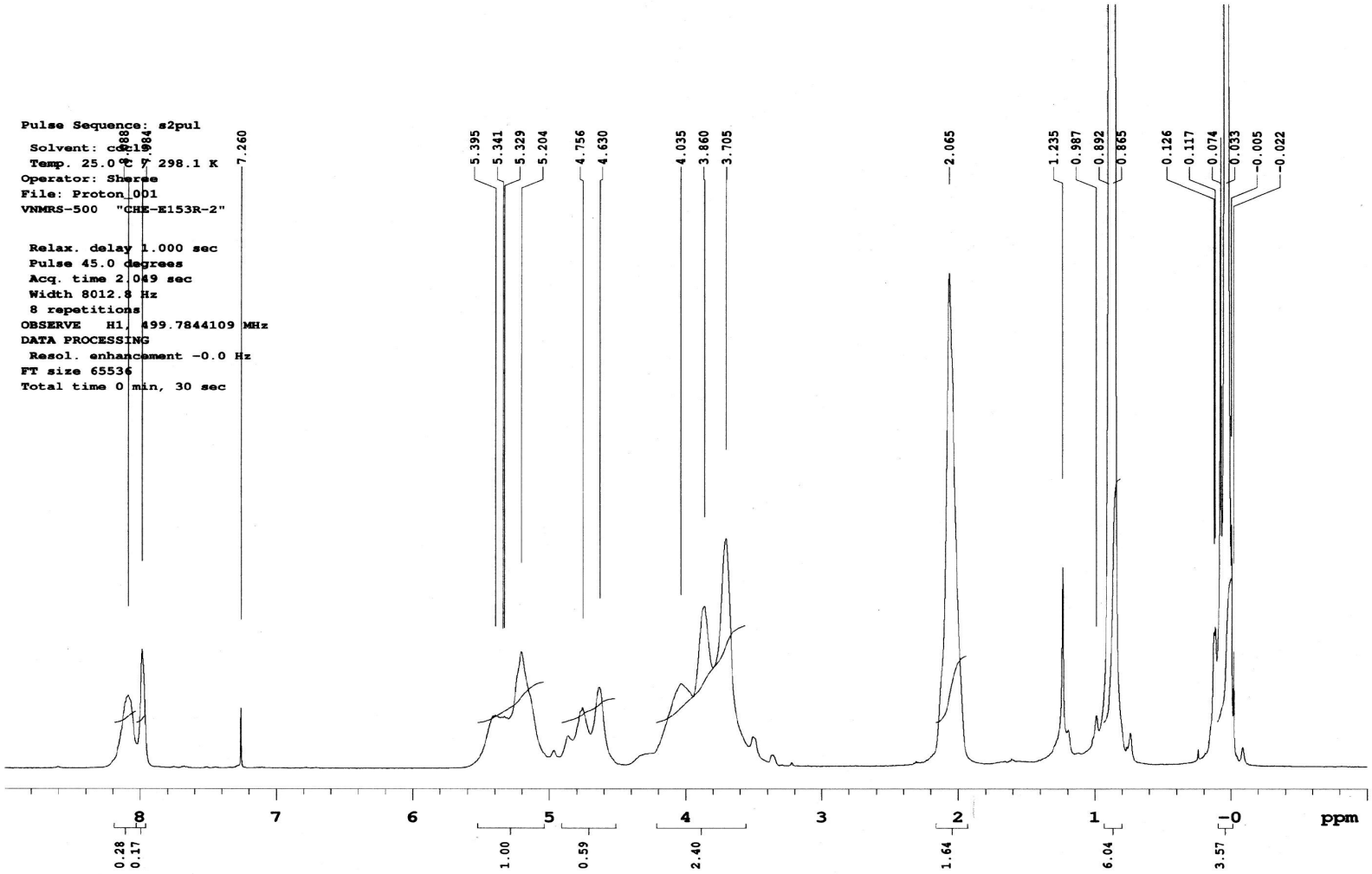
A.4.18. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-deoxy-2,3-*O*-diformyl)-β-CD (**24**).



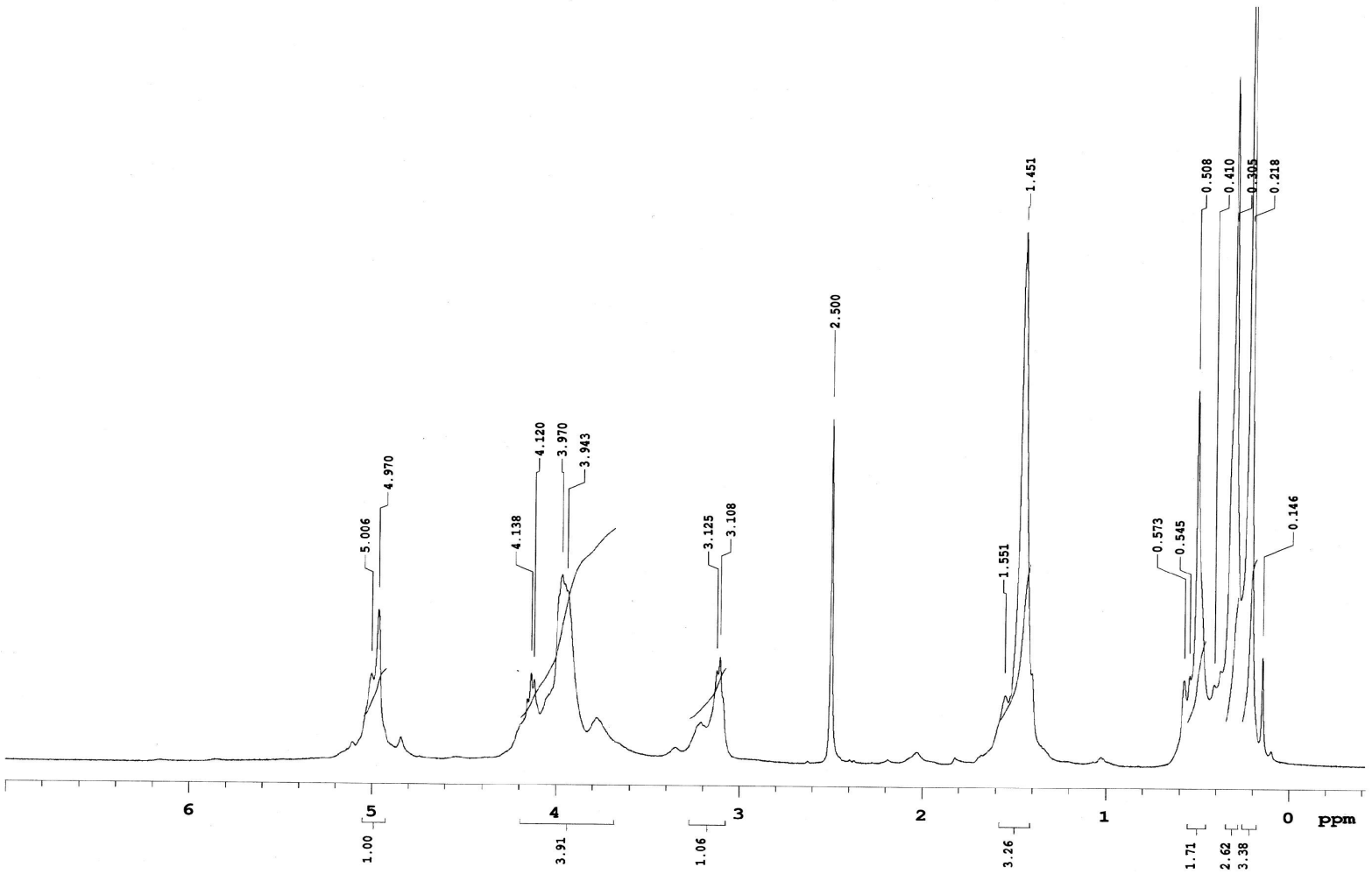
A.4.19. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-*O*-diformyl)-β-CD (**25**).



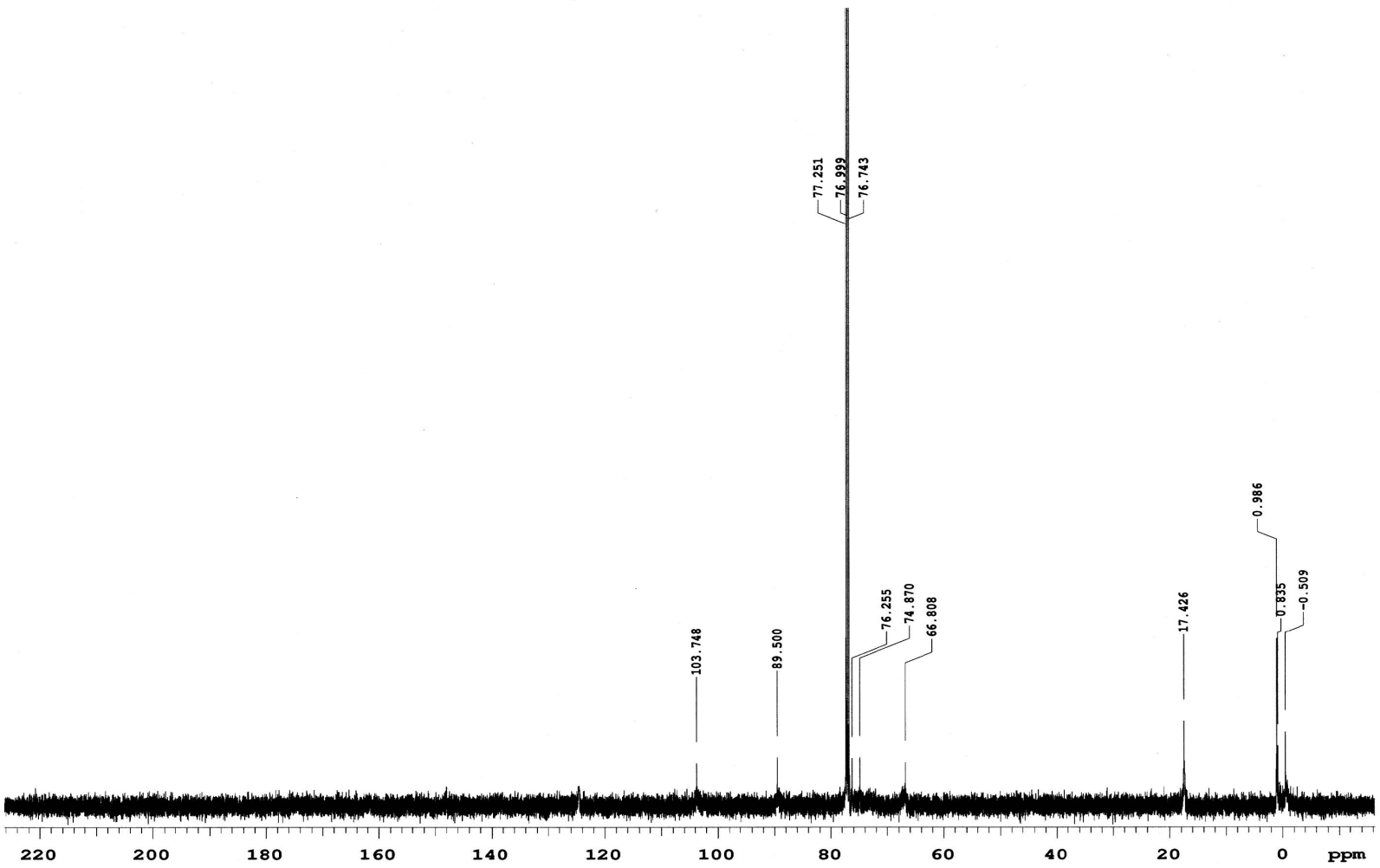
A.4.20. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-*O*-diformyl)-β-CD (**25**).



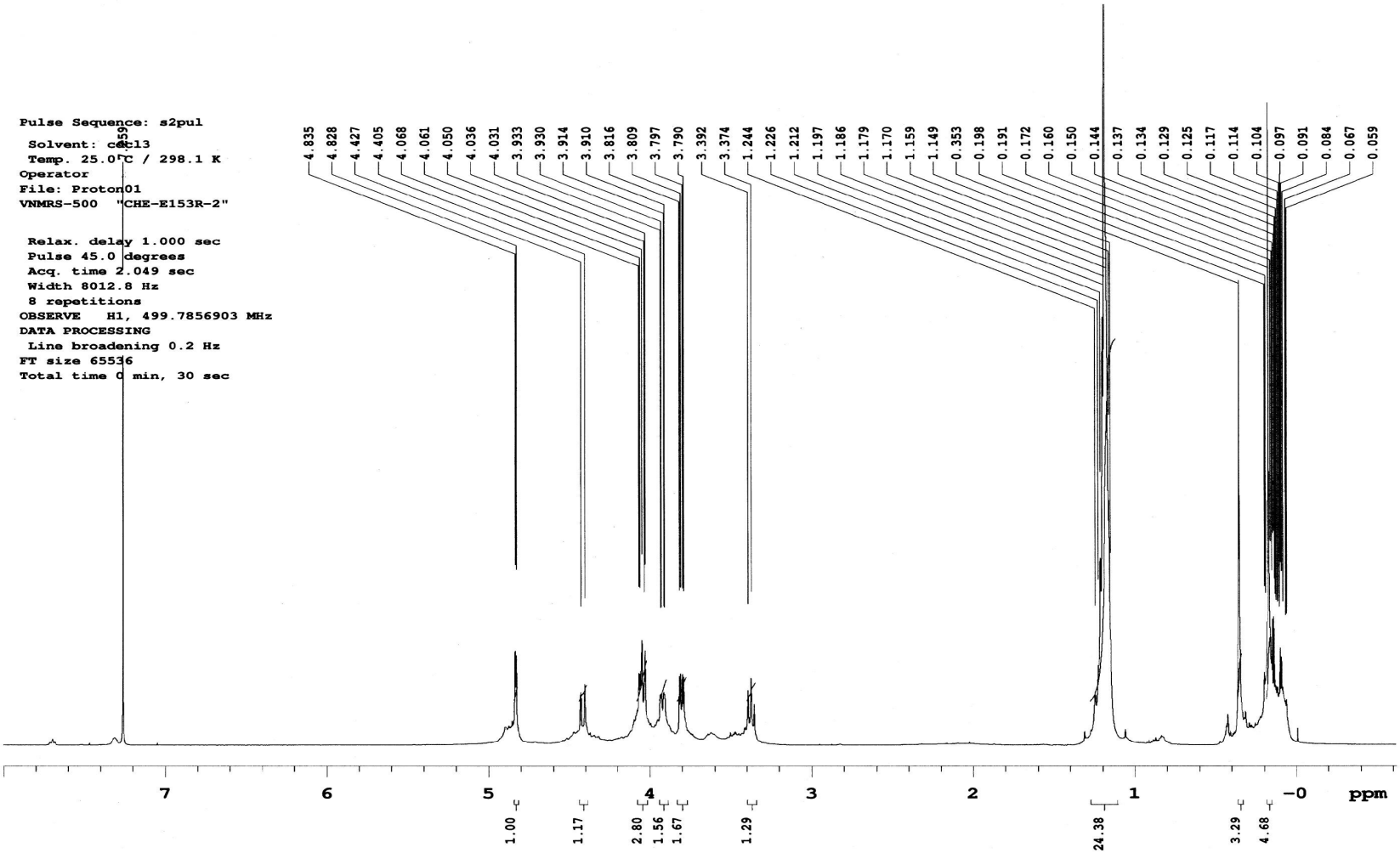
A.4.21. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-di-O-(45% formyl/55% acetyl)-β-CD (**BU 7**).



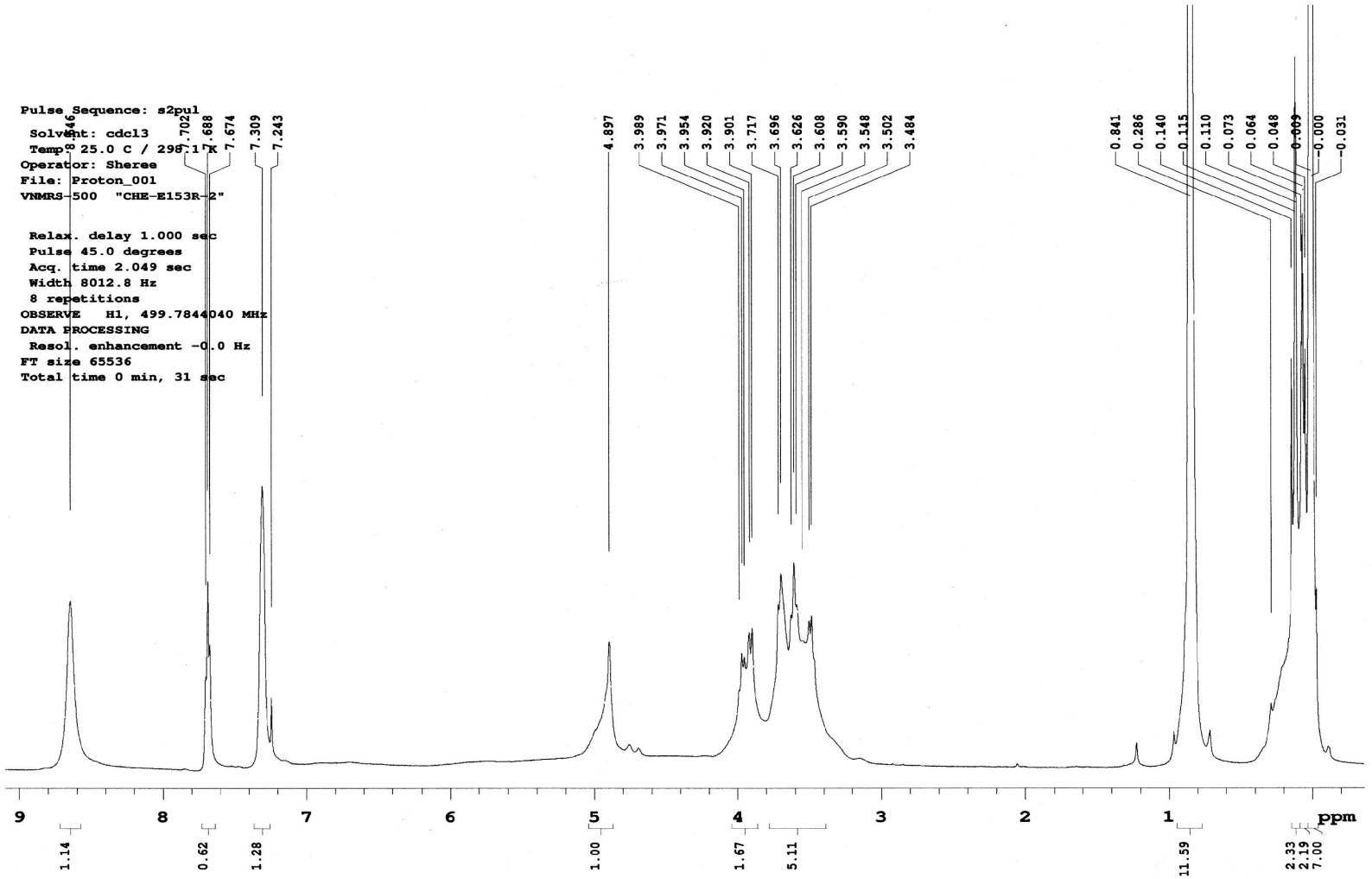
A.4.22. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-deoxy-2,3-di-*O*-cyclodimethylsilyl)-β-CD (**42**).



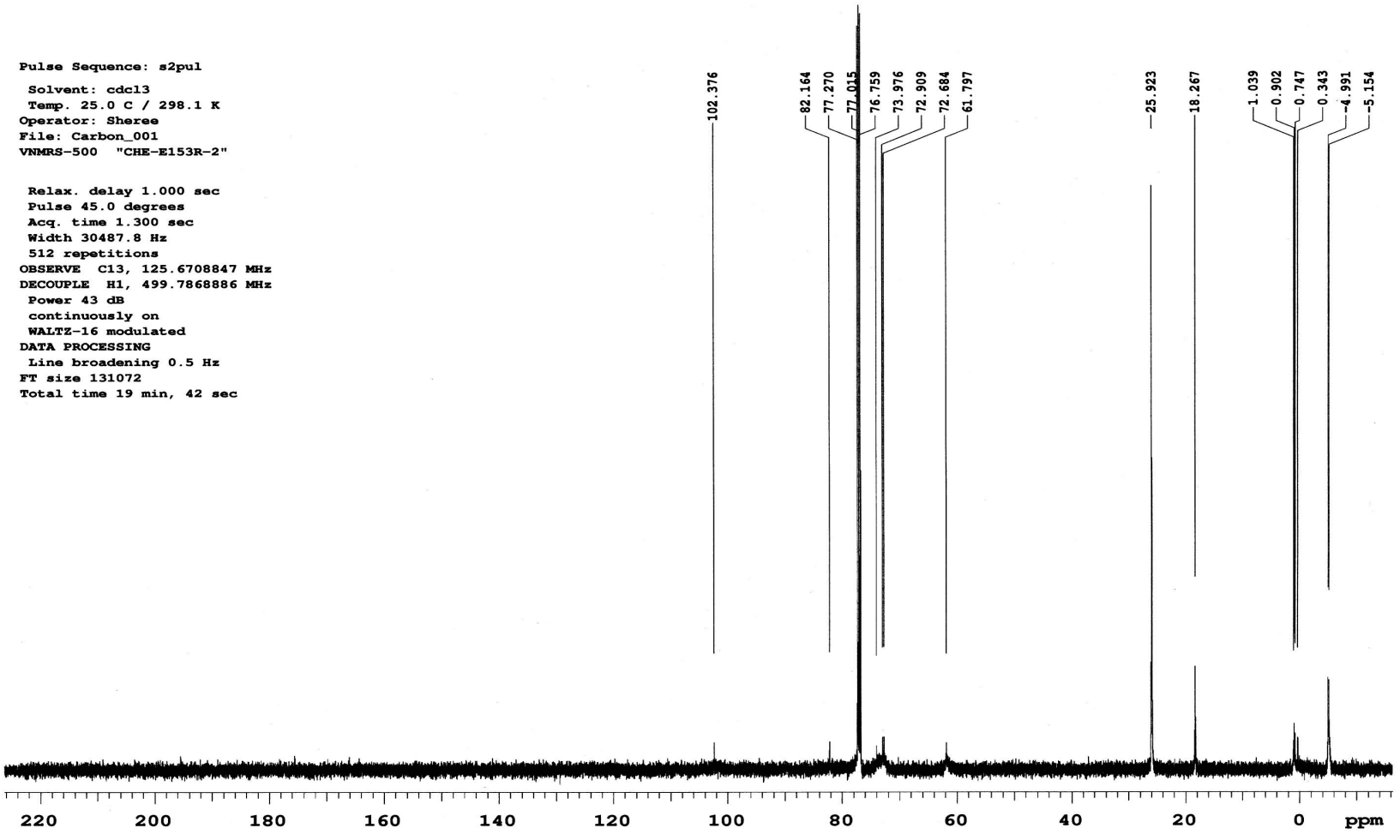
A.4.23. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-deoxy-2,3-di-*O*-cyclodimethylsilyl)-β-CD (**42**).



A.4.24. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-di-*O*-cyclodimethylsilyl)-β-CD (**43**).



A.4.25. ^{1}H NMR ($CDCl_3$, 500 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-di-*O*-cyclodimethylsilyl)-γ-CD (**BU 4**).



A.4.26. ^{13}C NMR ($CDCl_3$, 125 MHz) of *per*(6-*O*-*tert*-butyldimethylsilyl-2,3-di-*O*-cyclodimethylsilyl)-γ-CD (**BU 4**).

APPENDIX B

Tables

Table B.5.1. Column Parameters of CycloSil B

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
(2,3-dimethoxy,6-O-*tert*-butyldimethylsilyl)	200	220	>100:1	Hydrogen	11 psi	370,000

Table B.5.2. Column Parameters of Chiraldex A-PH

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
(*S*)-2-hydroxypropyl methyl ether-3,6-dimethyl	200	220	>100:1	Hydrogen	12 psi	102,653

Table B.5.3. Column Parameters of Chiraldex B-PH

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
(*S*)-2-hydroxypropyl methyl ether-3,6-dimethyl	200	220	>100:1	Hydrogen	12 psi	70,000

Table B.5.4. Column Parameters of Restek Rt-βDEXsa

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-diacetoxy-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	17 psi	105,000

Table B.5.5. Column Parameters of **BU 1**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-*O*-dimethoxy-6-deoxy	230	250	>100:1	Hydrogen	23 psi	300,000

Table B.5.6. Column Parameters of **BU 2**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-cyclodimethyl-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	11.3 psi	100,000

Table B.5.7 Column Parameters of **BU 3**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-cyclodimethyl-6-deoxy	230	250	>100:1	Hydrogen	10.2 psi	1,100

Table B.5.8. Column Parameters of **BU 4**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-O-cyclodimethylsilyl-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	10 psi	475,000

Table B.5.9. Column Parameters of **BU 5**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-O-diformyl-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	10 psi	29,000

Table B.5.10. Column Parameters of **BU 6**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-bis($^2H_3$)methoxy-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	6.1 psi	253,000

Table B.5.11. Column Parameters of **BU 7**

CYCLODEXTRIN	T°C MAX		SPLIT FLOW	CARRIER GAS	HEAD PRESSURE	*N*
	Isothermal	Programmed				(dodecane)
2,3-dimixed-6-*O*-*tert*-butyldimethylsilyl	230	250	>100:1	Hydrogen	10.2 psi	56,000

Table B.5.12. Enantioseparation Data of CSPs.

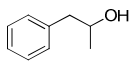
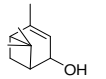
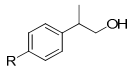
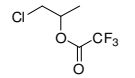
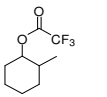
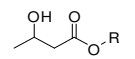
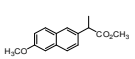
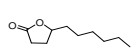
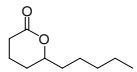
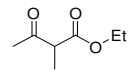
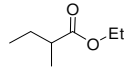
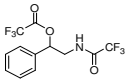
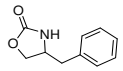
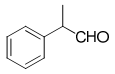
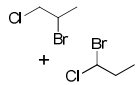
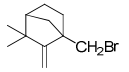
Structure	R=	T°C	J&W CycloSil B			Chiraldex A-PH			Chiraldex B-PH			Restek βDEXsa			BU 1			BU 2			BU 3			BU 4			BU 5			BU 6			BU 7		
			k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s
		80→160 @ 5°C/min	9.55	1.014	2.413	8.97	1.003	0.465	-	-	-	10.40	1.008	2.063	7.87	1.007	1.140	-	-	-	-	-	-	-	-	-	-	-	-	7.74	1.010	1.436	-	-	-
		80→120 @ 2°C/min	-	-	-	-	-	-	7.55	1.032	4.219	-	-	-	13.53	1.007	1.463	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.92	0.970	1.092
	-H	120→200 @ 5°C/min	5.09	1.016	1.756	-	-	-	-	-	-	5.38	1.028	4.089	-	-	-	-	-	-	-	-	-	-	-	-	-	-	3.824	1.011	0.913	-	-	-
	-CH_3		6.40	1.012	1.595	-	-	-	-	-	-	6.83	1.018	3.283	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5.034	1.008	0.842	-	-	-
	-OCH_3		9.31	1.007	0.902	-	-	-	-	-	-	8.37	1.025	3.985	7.97	-	1.181	-	-	-	-	-	-	-	-	-	-	-	7.680	1.006	0.830	6.68	0.92	0.86
	-Cl		9.41	1.011	1.514	-	-	-	-	-	-	8.67	1.043	6.806	-	-	-	-	-	-	-	-	-	-	-	-	-	-	7.902	1.004	0.546	-	-	-
		80→120 @ 2°C/min	-	-	-	-	-	-	-	-	-	5.90	1.021	1.760	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	cis/trans	80→120 @ 2°C/min	6.42	1.046	3.652	-	-	-	-	-	-	3.71	1.013	0.715	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.896	1.024	1.267	-	-	-
			6.74	1.036	2.853							4.43	1.028	1.552															2.975	1.018	1.033	4.381	0.992	0.925
	-CH_3	60→120 @ 2°C/min	8.74	1.024	2.025	-	-	-	-	-	-	25.11	1.028	4.38	6.24	1.011	1.217	-	-	-	-	-	-	-	-	-	-	-	5.897	1.024	1.669	-	-	-
	-CH_2CH_3		11.42	1.027	3.168	10.49	1.005	0.680	-	-	-	25.95	1.045	6.67	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8.711	1.019	1.838	-	-	-
	-$CH(CH_3)_2$		12.61	1.046	5.751	10.88	1.008	0.982	-	-	-	26.71	1.011	1.80	10.12	1.014	1.957	-	-	-	-	-	-	-	-	-	-	-	9.872	1.034	3.573	-	-	-
		150→200x 10 @ 5°C/min	18.20	1.01	0.83	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		100→195 @ 5°C/min	12.77	1.009	1.974	-	-	-	11.82	1.002	0.759	12.55	1.029	4.561	10.12	1.014	1.957	-	-	-	-	-	-	-	-	-	-	-	8.82	1.005	0.646	-	-	-
		120→195 @ 5°C/min	10.04	1.002	0.334	9.17	1.003	0.861	-	-	-	9.64	1.033	4.526	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		90→120 @ 2°C/min	-	-	-	-	-	-	-	-	-	9.26	1.029	3.572	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		60→100 @ 2°C/min	3.58	1.050	3.025	-	-	-	-	-	-	3.87	1.020	1.062	2.97	1.013	1.077	-	-	-	-	-	-	-	-	-	-	-	2.89	1.027	1.526	-	-	-

Table B.5.13. Enantioseparation Data of CSPs.

Structure	R=	T°C	J&W CycloSil B			Chiraldex A-PH			Chiraldex B-PH			Restek βDEXsa			BU 1			BU 2			BU 3			BU 4			BU 5			BU 6			BU 7		
			k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s	k'	α	R_s
	-C_8		-	-	-	4.61	1.010	1.134	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-C_{10}	100→195	-	-	-	7.73	1.006	1.407	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-C_{12}	@ 5°C/min	-	-	-	11.04	1.003	0.989	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	-$CH_2OC_6H_5$		-	-	-	3.85	1.008	1.042	-	-	-	10.23	1.030	4.223	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		80→180 @ 5°C/min	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		60→120 @ 5°C/min	4.75	1.02	1.74	-	-	-	2.02	1.017	0.528	-	-	-	3.67	1.019	1.257	2.12	1.014	0.672	-	-	-	-	-	-	-	-	-	3.55	1.017	1.279	-	-	-
		120→200x 5 @ 5°C/min	13.61	1.03	4.72	8.25	1.006	1.886	-	-	-	29.66	1.011	2.676	9.79	1.012	3.289	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8.11	0.896	1.240
		150→220x 10 @ 5°C/min	16.91	1.008	0.852	-	-	-	9.07	1.024	2.983	8.35	1.047	2.021	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	14.93	0.852	1.103
		80→190 @ 5°C/min	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		80→190 @ 5°C/min	7.86	1.005	0.607	5.78	1.008	1.170	-	-	-	11.51	1.014	3.015	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	7.17	1.023	2.801	5.51	0.981	1.997
		60→100 @ 2°C/min	-	-	-	-	-	-	-	-	-	10.54	1.045	3.75	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
			3.79	1.017	0.953							13.00	1.037	3.61																					
		100→190 @ 5°C/min	-	-	-	2.40	1.019	1.514	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
		80→190 @ 5°C/min	17.46	1.010	2.376	16.35	1.006	2.220	16.19	1.010	4.271	10.82	1.010	1.852	14.39	1.007	2.369	-	-	-	-	-	-	-	-	-	-	-	-	13.56	1.004	0.687	-	-	-

REFERENCES

1. Lough, W. J.; Wainer, I. W. *Chirality in Natural and Applied Science*. Blackwell Science, Ltd.: Oxford, 2002.

2. Friedman, L.; Miller, J. G. *Science* **1971**, *172*, 1044.

3. Allenmark, Stig G. *Chromatographic Enantioseparations: Methods and Application.* Ellis Horwood Ltd.: England, 1988.

4. Albert, A.. *Selective Toxicity, 5th Ed*. Chapman & Hall: London, 1973.

5. Blaschke, G.; Kraft, P.; Fickenstscher, K.; Kohler, F.; *Arzneim. Forsch*. **1979**, *29*, 1690.

6. Smith, Michael B.; March, Jerry. *Advanced Organic Chemistry: Reaction, Mechanism, Structure, 5th Edition.* John Wiley & Sons: New York, 2001.

7. Zief, Morris; Crane, Laura. *Chromatographic Chiral Separations*. Marcel Dekker, Inc.: New York, 1988.

8. Subramanian, G. *Chiral Separation Techniques: A Practical Approach.* Wiley-VCH: New York, 2001.

9. Consiglio, P.; Pino, L.; Flowers I.; Pittman, C. U. Jr, *J. Chem. Soc. Chem. Commun.* **1983**, 612.

10. Viswanathan. T; Toland, A. *J. Chem. Ed.* 1995, 72, 945-946.

11. Miller, James M. *Chromatography: Concepts & Contrasts*. John Wiley & Sons, Inc.: New York, 1988.

12. Snyder, L. R.; Kirkland, J. J. *Introduction to Modern Liquid Chromatography 2nd Edition*. John Wiley & Sons: New York, 1979.

13. James, A. T.; Martin, A J. P., *Biochem. J*. **1952**, *50*, 679-690.

14. Martin, A. J. P; Synge, R. L. M. *Biochem J. (Lond.)* **1941**, *35*, 1358.

15. Grob, Robert L.; Barry, Eugene F. *Modern Practice of Gas Chromatography*, John Wiley & Sons: New Jersey, 2004.

16. Schurig V.; *Angew. Che., Int. Ed. Engl*. **1977**, *16*, 110.

17. Schurig, V. *Journal of Chromatography A* **1994**, *66*, 111-129.

18. Koscielski, T.; Sybilska, D; Jurczak, J. *J. Chromatog.* **1983**, *280*, 131.

19. Dodziuk, Helena. *Cyclodextrins and Their Complexes.* Wiley-VCH: Poland, 2006.

20. Koshland, D. E. *Angew. Chem. Int. Ed. Engl*. **1994**, *33*, 2475.

21. Cserhati, Tibor; Forgacs, Esther. *Cyclodextrins in Chromatography*. RSC: United Kingdom, 2003.

22. Easton, Christopher J.; Lincoln, Stephen F. *Modified Cyclodextrins: Scaffolds and Templates for Supramolecular Chemistry.* Imperial College Press: London, 1999.

23. Shpigun, O. A.; Ananieva, I. A.; Budanova, N. Yu Shapovalova, E. N. *Russian Chemical Reviews* **2003**, *72*, 1035-1054.

24. Koscielski, T.; Sybilska, D.; Jurczak, J. *J. Chromatogr*. **1983**, *261*, 357-362.

25. Schurig, V.; Novotny, H. P.; *J. Chromatogr*. **1988**, *441*, 155-163.

26. Schurig, V. *J. Chromatog. A* **2001**, *906*, 275-299.

27. Konig, W, *Enantioselective Gas Chromatography with Modified Cyclodextins*, Huthig Heidelerg, 1992.

28. Armstrong, D. W, Li, W. Y, Pitha, J. *Anal. Chem*. **1990**, *62*, 214-217.

29. Fugedi, Peter. *Carbohydrate Research* **1989**, *192*, 366-369.

30. Crich, David; Vinod, A. U.; Picione, John. *J. Org. Chem*. **2003**, *68*, 8453-8458.

31. Baer, Hans H.; Defaye, Jacques, Gonzales, Santoyo F. *Carbohydrate Research* **1992**, *228*, 307-314.

32. Kim, Byoung; Lee, Su; Park, Keung; Lee, Kwang; Park, Jung. J. *High. Resol. Chromatogr.* **1997**, *20*, 208-212.

33. Melton, L. D.; Slessor, K. N. *Carbohydr. Res.* **1971**, *18*, 29.

34. VanEtten, R. L., Clowes, G. A.; Sebastian, M. L.; Bender, J. *J. Am. Chem. Soc.* **1967**, *89*, 3253.

35. Barret, A. Barton, D.; Bielski, R.; McCombie, S. *J. Chem. Soc. Chem. Commun*. **1977**, 866.

36. Bargiotti, A. Hanessian, S.; LaRue, M. *Tetrahedron Lett*. **1978**, 737.

37. Mukaiyama, T.; Sato, T.; Hanna, J. *Chem. Lett*. **1973**, 1041.

38. Tipson, R. S.; Cohen, A. *Carbohydrate Research* **1965**, *1*, 338-340.

39. Fukudome, Makoto; Shiratani, Tomonori; Immel, Stefan; Nogami, Yasuyoshi; Yuan, De-Qi; Fujita, Kahee. *Angew. Chem. Int. Ed*. **2005**, *44*, 4201-4204.

40. Corey, E. J.; Winter. R. A. E. *J. Am. Chem. Soc*. **1963**, *85*, 2677-2676.

41. Tyrlik, S.; Wolochowicz, I. *Bull. Soc. Chim. Fr*. **1973**, 2147.

42. McMurry, J. E.; Fleming, M. P. *J. Am. Chem. Soc*. **1974**, *96*, 4708.

43. Rieke, R. D., Hudnall, P. M. *J. Am. Chem. Soc*. **1972**, *94*, 7178.

44. McMurry, John E.; Fleming, Michael, P.; Kees, Kenneth, Krepski, Larry R. J. *Org. Chem.* **1978**, *43*, 3255-3266.

45. Garegg, Per J.; Samuelsson, Bertil. *Synthesis* **1979**, 469.; Garegg, Per J. *Pure & Appl. Chem*. **1984**, *56*, 845-858.

46. Dess, Daniel, B.; Martin, J. C. *J. Am. Chem. Soc*. **1991**, *113*, 7277-7287.

47. Mancuso, A. J.;Swern, D.; *Synthesis* **1981**, 165.

48. Frigerio et al., U.S. Patent 5,510,538, 1996.

49. Frigerio, Marco; Santagostino, Marco; Sputore, Simona. *J. Org. Chem*. **1999**, *64*, 4537-4538.

50. Khan, Abdul R.; Barton, Lawrence; D'Souza, Valerian. *J. Chem. Comm. Commun.* **1992**; Khan, Abdul, Barton, Lawrence; D'Souza Valerian. *J. Org. Chem*. **1996**, *61*, 8301-8303.

51. Zhang, Ping; Coleman, Andrew. W. *Supramolecular Chemistry* **1993**, *2*, 255-263.

52. Ashton, Peter, R.; Koniger, Rainer; Stoddart, Fraser, J. *J. Org. Chem*. **1996**, *61*, 903-908.

53. Yan, Jiaming; Watanabe, Ryosuke; Yamaguchi, Masatoshi; Yuan, De-Qi; Fujita, Kahee. *Tetrahedron Letters* **1999**, *40*, 1513-1514.

54. Yuan, De-Qi, Tahara, Tsutom; Chen, Wen-Hua; Okabe, Yuji; Yang, Cheng; Yagi, Youchi; Nogami, Yasuyshi; Fukudome, Makoto; Fujita, Kahee. *J. Org. Chem*. **2003**, *68*, 9456-9466.

55. VanEtten, R. L.; Sebastian, J. F.; Clowes, G. A.; Bender, M. L.. *J. Am. Chem. Soc*. **1967**, *89*, 3242.

56. Buschmann, H.; Mutihac, L.; Schollmeyer, E. *Supramolecular Chemistry* **2005**, *17*, 447-451.

57. (a)Chini, Marco, Crotti, Paolo; Favero, Lucilla; Macchia, Franco. *Tetrahedron Letters* **1991**, *32*, 4775-4778. (b) Caron, M.; Sharpless, K. B. *J. Org. Chem*. **1985**, *50*, 1557-1560. (c) Yang, et al. *Tetrahedron* **1988**, *44*, 6305-6314.

58. Shitangkoon, Aroonsiri; Vigh, Gyula. *Journal of Chromatography A* **1996**, *738*, 31-42.

59. Kobor, F.; Angermund, K. and Schomburg, G., *J. High Resolut. Chromatogr*. **1993**, *16*, 299.

60. Gadelle, A.; Defaye, J. *Angew. Chem*. **1991**, *103*, 94-95.

61. (a) Gong, H.; Andrews, R.S.; Zuccarello, J. L.; Lee, S. J.; Gagne, M. R. *Org. Lett*. **2009**, *11*, 879.; (b) Spiegel, S. A.; Wiberg, K. B.; Schacherer, L. N.; Medeiros, M. R.; Wood, J. L. *J. Am. Chem. Soc*. **2005**, *127*, 12513.

62. Narayanam, Jagan, J. R.; Rucker, Joesph, W.; Stephenson, Corey R. J. *J. Am. Chem. Soc*. **2009**, *131*, 8756-8757.

63. Taylor, Richard, J. K., *Organocopper Reagents: A Practical Approach*. Oxford University Press: New York, 1994.

64. (a) Krimen, Lewis I. *Organic Syntheses* **1988**, *6*, 8.; (b) *Med. Res. Chem*. **2007**, *15*, 452-462.

65. Buc, Saul R. *Organic Syntheses* **1963**, *4*, 101.

66. Ishikawa, Tsutomu. *Superbases for Organic Synthesis*. John Wiley & Sons, Ltd.: United Kingdom, 2009.

67. DiMagno, Stephen G.; Sun, Haoran, US Patent 7,592,486 B2 2009.

68. Advanced Separation Technologies (Astec), *Chiraldex GC Columns: a guide to using cyclodextrin bonded phases for separations by capillary gas chromatography*, Whippany, New Jersey, 2002.