ABSTRACT

Evaluation of Phenolic Protecting Groups Using Microwave Synthesis and Progress Towards a Benzosuberene Anti-Cancer Agent

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According to an epidemiology survey conducted by the American Cancer Society, about 1,596,670 new cancer cases were expected to be diagnosed in the year 2011. Cancer is a term used to categorize diseases in which abnormal cells divide uncontrollably and are able to invade normal tissue in the body. Most cancers can be classified into broad categories such as carcinoma, sarcoma, leukemia, lymphoma, and central nervous system cancers. Synthetic chemistry plays a key role in terms of the discovery of new anti-cancer agents. Robust protecting group strategies are paramount in successful target-directed synthetic campaigns. This study investigates the use of a microwave technique compared to conventional heating in the installation of selected phenolic protecting groups. In addition, this thesis covers the progress made towards the synthesis of a new functionalized benzosuberene anti-cancer agent.

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EVALUATION OF PHENOLIC PROTECTING GROUPS USING MICROWAVE SYNTHESIS AND PROGRESS TOWARDS A BENZOSUBERENE ANTI-CANCER AGENT

A Thesis Submitted to the Faculty of

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In Partial Fulfillment of the Requirements for the

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By

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ABBREVIATIONS

^{o} C	Degree Celsius
CA4	Combretastatin A-4
CA4P	Combretastatin A-4 phosphate
DMF	*N*,*N*-dimethylformamide
DNA	Deoxyribonucleic acid
h	Hour
Hz	Hertz
KO*t*Bu	Potassium *tert*-butoxide
MHz	Megahertz
min	Minutes
mM	Millimole
NMR	Nuclear magnetic resonance spectroscopy
THF	Tetrahydrofuran
VDA	Vascular disrupting agent

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This research project would not have been possible without the support of many people.

It is with immense gratitude that I acknowledge the support and help of

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Honors thesis by Mahmood Jamil Khan, dedicated

To,

My Parents

and

in Loving Memory of My Grandparents

مري أبــو أمي أور انكـــي والـــدين كـو مـيري محبـت قـا تحــف

CHAPTER ONE

Introduction

Cancer

In the United States, about 550,000 people die each year because of cancer. During the 1970's the cancer survival rate was about 50%, however; with the advent of new treatment options that figure has reached a 66% survival rate.[1]

Cancer is a commonly used term for cells that transform into abnormal cells within an organism and proliferate without control.[2] These cells are then able to invade other tissue types within the host and cause complications which eventually lead to death. Like other diseases, cancer is not confined to just one type of classification. Currently, there are more than one-hundred different types of cancer which are classified according to the organ or type of cell from which the cancer originates.

There are six primary categories that are used to classify cancer types.[2]

- Sarcoma: Cancer that arises from smooth muscle, fat, bone, and cartilage.
- Leukemia: Cancer that arises primarily from bone marrow.
- Carcinoma: Cancer that arises from skin and tissue that covers internal organs.
- Lymphoma: Cancer that arises in the immune system.
- Myeloma: Cancer that arises in plasma cells, a type of white blood cell that is responsible for the production of antibodies.
- CNS: Cancer that arises from the tissue located in the brain, brain stem, and spinal cord.

The uncontrollable growth of cells is the hallmark of cancer. When enough abnormally growing cells are produced, they form a mass of cells commonly called a tumor. The term tumor is not synonymous with cancer. Cancer by definition is malignant, meaning that the condition becomes progressively worse and can result in death. Tumors can be classified into three general categories: benign, pre-malignant, and malignant. The latter two tumor types are considered lethal.

A benign tumor is a tumor that lacks the ability to metastasize, meaning that it cannot spread from one organ to another. A pre-malignant tumor is the precursor to a malignant tumor in which there are prominent morphological alterations that, if left untreated, could lead to the development of a malignant tumor. Lastly, a malignant tumor is characterized by its ability to invade nearby parts of the body. This type of tumor can metastasize, or spread, to distant parts of the body through the blood system or the lymphatic system.

Typically, healthy cells are able to control their growth through a complex set of checkpoints that can even cause cell death if they become defective. Cancer occurs when problems arise in the genes that control the proliferation of the cell. These problems arise when there are damaged genes or when the faulty genes are inherited.[2]

Treatment of Cancer

There are three main different treatment options that are widely used today in treating and eradicating cancer from the body. The most common treatments are chemotherapy, radiation therapy, and surgery.

Chemotherapy

Chemotherapy is the treatment of cancer with chemical compounds or combinations of them into a standardized treatment regimen. This treatment acts on cancer by killing the cells that are in the cell division phase. Although chemotherapy targets cancer cells, it also inadvertently affects the replication of normal cells. In a general sense, most chemotherapeutic drugs work by inhibiting mitosis of the rapidly dividing cell. Since there are wide arrays of chemotherapeutic drugs, we will only discuss the broad scheme of its action on cancer cells. The main features of chemotherapy are that it arrests the multiplication of cells, assists the immune system for self-defense, and helps initiate the growth of essential enzymes in the body that help combat cancer. Following are the commonly used therapeutic drugs.[3]

- Alkylating agents: These agents impair cell function by forming covalent bonds between the phosphate, amino, carboxyl, and sulfhydryl groups in important molecules. They can also function by chemically modifying a cell's DNA. Ex: cisplatin, mechlorethamine.
- Anti-metabolites: These agents prevent purines and pyrimidines, the building blocks of DNA, from being incorporated into DNA during the S phase of the cell cycle. Ex: Sulfadiazine

- Alkaloids and terpenoids: Most of these agents are derived from plants and function by blocking the formation of microtubules during cell division. Ex: Vinblastine
- Topoisomerase inhibitors: These inhibitors function by blocking either type I or type II topoisomerases. Administration of this class of drugs interferes with transcription and replication of DNA by disrupting the proper super coiling of DNA.

  Ex: Topotecan

Radiation Therapy

This type of treatment is commonly applied to tumors that are considered cancerous. The utilization of ionizing radiation in this treatment works by damaging the DNA of tissue subjected to irradiation. The goal of radiation treatment is to irreversibly damage the DNA of the cancer cells so that they are unable to replicate effectively. In essence, radiation can alter the structure of DNA bases making them unfit to be incorporated into newly synthesized DNA. Another positive aspect of this treatment is that it takes advantage of a cancer cell's repair mechanism. When a particular cell undergoes transformation into a cancerous cell, it essentially turns off its DNA repair machinery. The lack of a repair mechanism, therefore; makes this radiation therapy more useful against cancer cells.[4]

Surgery

Surgery consists of an operation to physically remove or repair parts of the body to treat cancer. Surgery can be used to remove a cancerous tumor to relieve the symptoms caused by the cancer or to accurately diagnose the cancer type. This form of treatment is often supplemented with other common treatments such as chemotherapy and radiation therapy.

Vascular Disrupting Agents

Vascular disrupting agents (VDAs) are drugs that are designed to disrupt and damage the vasculature of well-established cancerous tumors. The "buckling" of the blood vessels after the administration of a VDA results in significant loss of blood flow to the tumor and usually leads to central necrosis. By definition, necrosis is the premature death of cells and or living tissue. VDAs can be classified as small-molecule and ligand based compounds. The focal point of this thesis, centers on small-molecule VDAs. Two common small-molecule VDAs are combretastatin A-4 and vadimezan.[5]

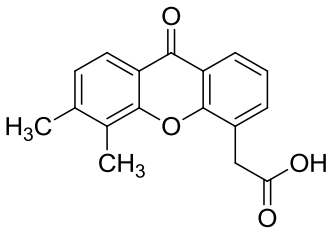


Figure 1.0: Structure of Vadimezan

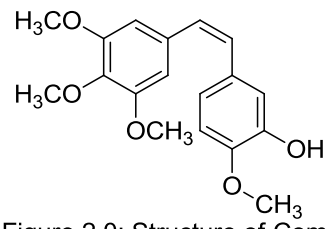


Figure 2.0: Structure of Combretastatin A-4

Combretastatin A-4 phosphate (CA4P) is a well-known disodium phosphate pro-drug that is currently in human clinical trials. It biologically functions as both a microtubule destabilizing drug and a vascular disrupting agent. The chemical structure of combretastatin A-4 phosphate is illustrated in figure 3.0. [10]

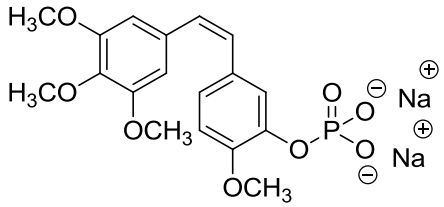


Figure 3.0: Chemical Structure of Combretastatin A-4 Phosphate

It is important to note that CA4P is rapidly converted by the body to CA4 through the action of non-specific phosphatase enzymes.[6] CA4 functions by inhibiting tubulin polymerization during cell division. CA4 binds to the colchicine site on the α, β-tubulin heterodimer and as a result disrupts the polymerization into microtubules during cellular division. CA4 also causes the endothelial cells within the tumor vasculature to buckle up causing decreased blood flow to the central tumor. Since tumors are known to form vasculature in a rapid fashion, the disruption of blood vessels is hallmark to the potency of CA4.[7]

Research

The research relevant to this study is related to the synthesis of benzosuberene and various compounds containing protecting groups. This two part study aimed to determine if subjecting protection reactions to microwave irradiation was more beneficial than a conventional heating approach commonly used in laboratories. Furthermore, this study will provide an overview of the installation of phenolic protecting groups and will describe the progress towards the synthesis of a functionalized benzosuberene-based anti-cancer agent.[9]

CHAPTER TWO

Materials and Methods

Materials

Chemicals and solvents for the reactions below were purchased from commercial sources such as Sigma-Aldrich, Sigma, and Alfa-Aesar.[8] ^{1}H NMR spectra were obtained in deuterated chloroform with 0.03% TMS using a Varian spectrometer at a frequency of 500 MHz. Peaks listed in this study are denoted as singlet (s), doublet (d), triplet (t), quartet (q) and coupling constants are reported in hertz (Hz). All solvents, glassware, and instruments used in our study were obtained by Baylor University.

Terminology

The compounds synthesized in this study are numbered in sequential order, 1-5. Furthermore, a letter designation accompanies each compound. Compound names that include "a" are hereto designated as being synthesized by microwave irradiation. Compound names that include the "b" are hereto designated as being synthesized by the conventional method. For example, compound **1a** was synthesized under microwave irradiation, while compound **1b** was synthesized under conventional conditions.

The compounds synthesized in the benzosuberene project are labeled with numbers and no letters.

Project Overview: Protecting Group Strategies and a Benzosuberene Anti-Cancer Agent

Two separate projects were carried out during the course of this thesis research. The first project focuses on tosylate protecting group strategies for various phenolic moieties under microwave conditions. Among the twenty reactions conducted, five reactions yielded significant results. The second project consisted of preparing an analogue of a functionalized benzosuberene-based anti-cancer agent. Details regarding strategies for each of these projects are described in the following sections.

Overall, each of the five reactions was performed twice, once under normal conditions at room temperature and once under microwave irradiation at 60 ^{o}C. Refer to figures 4.0 and 5.0 for a detailed reaction scheme.

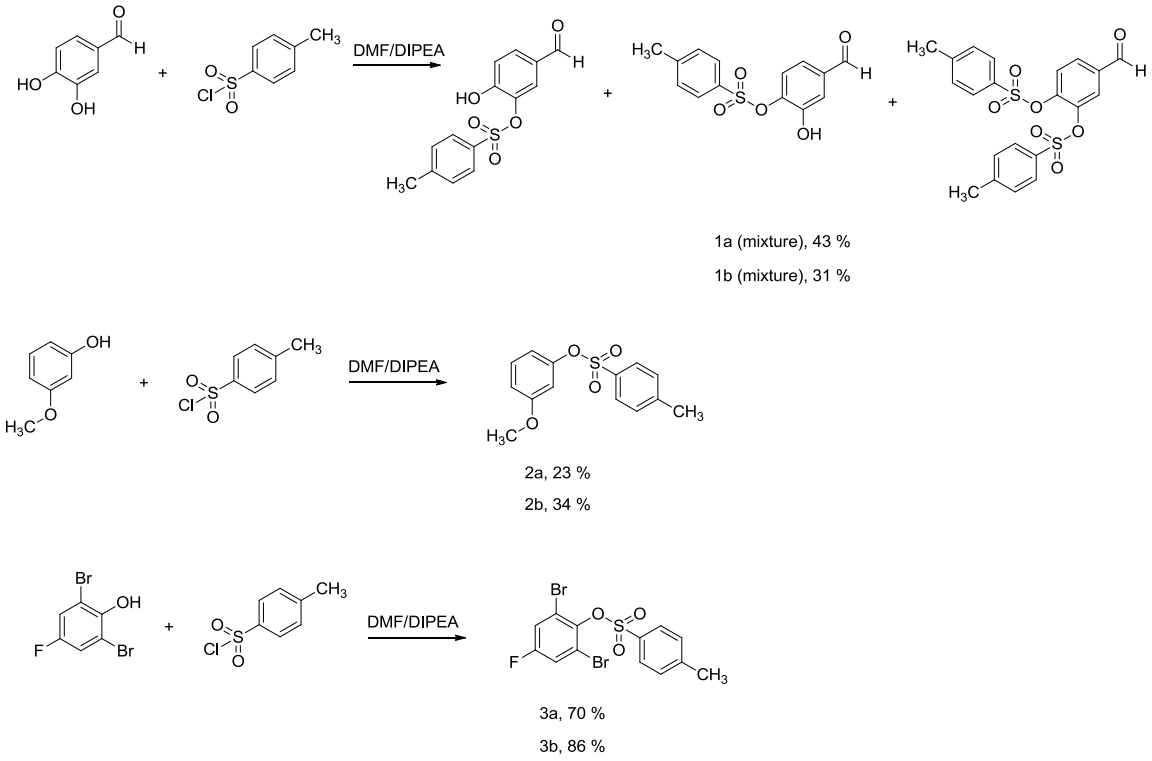


Figure 4.0: Reaction scheme for compounds 1a, 2a, 3a, 1b, 2b, 3b. Compounds 1a-3a were synthesized by microwave irradiation while compounds 1b-3b were synthesized by the conventional method.

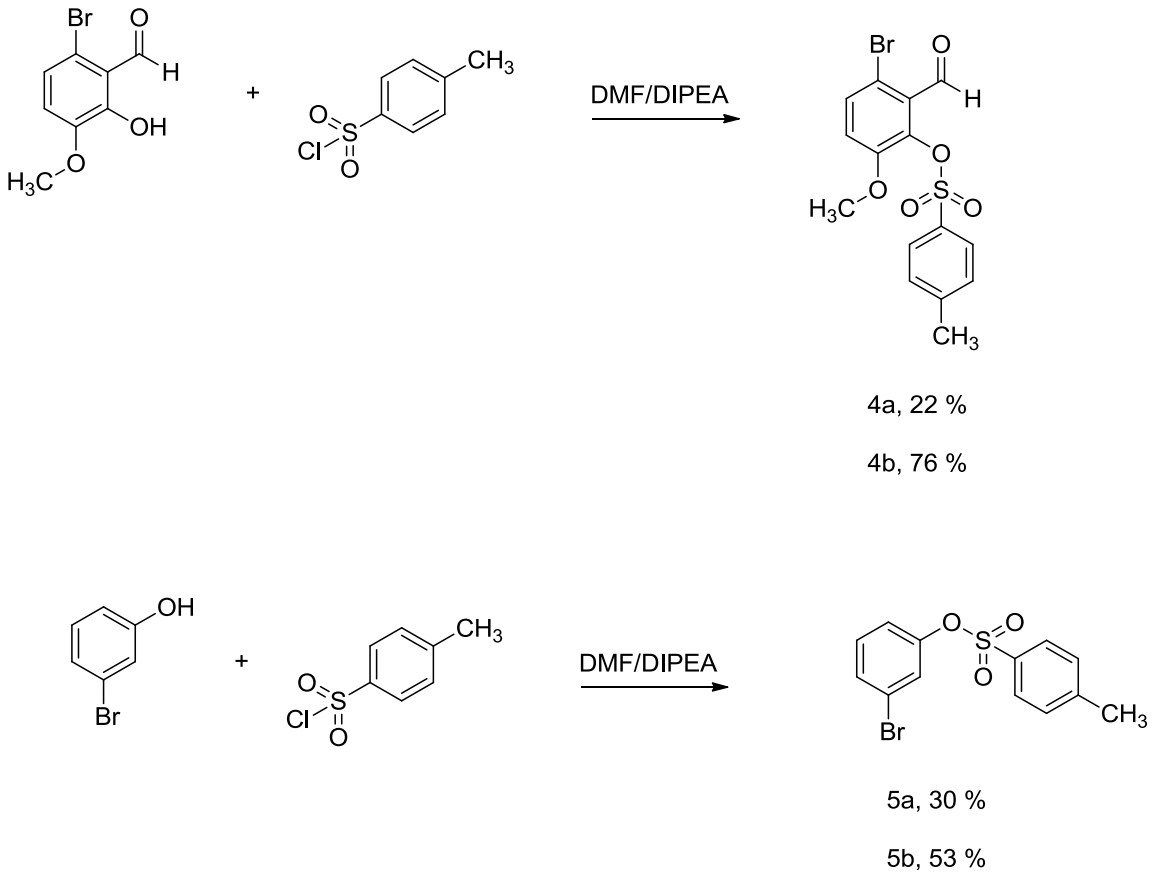


Figure 5.0: Reaction scheme for compounds 4a, 5a, 4b, 5b.
Compounds 4a-5a were synthesized by microwave irradiation while compounds 4b-5b were synthesized by the conventional method.

4-Formyl-1,2-phenylene *bis*(4-methylbenzenesulfonate) (1a)

The microwave reaction was conducted by combining 3,4-dihydroxybenzaldehyde (0.690 g, 4.99 mmol), *para*-toulenesulfonylchloride (1.920 g, 10.07 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reactor, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **1a** (0.909 g, 2.12 mmol, 43 %) as a brown solid. Since compound **1a** was a mixture of three products that were not separated, NMR shifts are not included.

3-Methoxyphenyl 4-methylbenzenesulfonate (2a)

The microwave reaction was conducted by combining of 3-methoxyphenol (0.620 g, 5.00 mmol), *para*-toulenesulfonylchloride (0.651 g, 3.41 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the

reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **2a** (0.304 g, 1.16 mmol, 23 %) as a solid.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.57 (s, 2H), 2.45 (s, 3H), 3.72 (s, 3H), 6.56 (dd, 2H, *J* = 12.0 Hz), 6.72 (d, 1H, *J* = 33 Hz), 7.16 (t, 1H, *J* = 8 Hz, 8 Hz), 7.28 (d, 2H, *J* = 13 Hz), 7.71 (d, 2H, *J* = 11 Hz)

2,6-Dibromo-4-fluorophenyl 4-methylbenzenesulfonate (3a)

The microwave reaction was conducted by combining 2,6-Dibromo-4-fluorophenol (1.86 g, 6.88 mmol), *para*-toulenesulfonylchloride (0.973 g, 5.10 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **3a** (1.95 g, 4.78 mmol, 69 %) as a solid.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.53 (s, 2H), 2.48 (s, 3H), 7.38 (dd, 2H, *J* = 8 Hz), 7.92 (d, 2H, *J* = 8.5 Hz)

3-Bromo-2-formyl-6-methoxyphenyl 4-methylbenzenesulfonate (4a)

The microwave reaction was conducted by combining 6-bromo-2-hydroxy-3-methylbenaldehyde (1.14 g, 5.30 mmol), *para*-toulenesulfonylchloride (1.44 g, 7.54 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^oC under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **4a** (0.412 g, 1.17 mmol, 22 %) as a viscous oil.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.48 (s, 3H), 3.62 (s, 3H), 6.97 (d, 1H, *J* = 8.5 Hz), 7.26 (s, 1H), 7.37 (d, 2H, J = 8Hz), 7.81 (d, 2H, *J* = 8.5 Hz), 10.07 (s, 1H)

3-Bromophenyl 4-methylbenzenesulfonate (5a)

The microwave reaction was conducted by combining 3-bromophenol (0.868 g, 5.02 mmol), *para*-toulenesulfonylchloride (1.44 g, 4.98 mmol), and dimethylformamide

(15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **5a** (0.346 g, 1.11 mmol, 30 %) as a brown solid.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.46 (s, 3H), 6.93 (ddd, 1H, *J* = 8.5 Hz, 10 Hz, 8.5 Hz), 7.25 (tt, 2H, *J* = 7Hz, 8 Hz, 8 Hz, 7 Hz), 7.34 (d, 2H, 8.5 Hz), 7.38 (ddd, 1H, *J* = 8.6 Hz, 10 Hz, 8.6 Hz), 7.75 (dd, 2H, *J* = 6.5 Hz, 7.0 Hz).

4-Formyl-1,2-phenylene bis(4-methylbenzenesulfonate) (1b)

The reaction was conducted by combining 3,4-dihydroxybenzaldehyde (0.712 g, 5.15 mmol), *para*-toulenesulfonylchloride (1.992 g, 10.44 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the

reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **1b** (0.613 g, 1.43 mmol, 31 %) as a solid. Since compound **1b** was a mixture of three products that were not separated, NMR shifts are not included.

3-Methoxyphenyl 4-methylbenzenesulfonate (2b)

The reaction was conducted by combining 3-methoxyphenol (0.620 g, 4.99 mmol), *para*-toulenesulfonylchloride (0.651 g, 3.42 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **2b** (0.449 g, 1.71 mmol, 34 %) as a white crystal.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.572 (s, 2H), 2.44 (s, 3H), 3.72 (s, 3H), 6.56 (dd, 2H, *J* = 12.0 Hz), 6.72 (d, 1H, *J* = 33 Hz), 7.16 (t, 1H, *J* = 8 Hz, 8 Hz), 7.28 (d, 2H, *J* = 13 Hz), 7.71 (d, 2H, *J* = 11 Hz)

2,6-Dibromo-4-fluorophenyl 4-methylbenzenesulfonate (3b)

The reaction was conducted by combining 2,6-Dibromo-4-fluorophenol (1.30 g, 4.83 mmol), *para*-toulenesulfonylchloride (0.986 g, 5.17 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **3b** (1.69 g, 4.14 mmol, 86 %) as a brown solid.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 1.52 (s, 2H), 2.48 (s, 3H), 7.38 (dd, 2H, *J* = 8 Hz), 7.91 (d, 2H, *J* = 8.5 Hz)

3-Bromo-2-formyl-6-methoxyphenyl 4-methylbenzenesulfonate (4b)

The reaction was conducted by combining 6-bromo-2-hydroxy-3-methylbenaldehyde (1.16 g, 5.38 mmol), *para*-toulenesulfonylchloride (1.427 g, 7.48 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **4b** (1.43 g, 4.06 mmol, 76 %) as a viscous oil.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.47 (s, 3H), 3.62 (s, 3H), 6.96 (d, 1H, *J* = 8.5 Hz), 7.26 (s, 1H), 7.37 (d, 2H, *J* = 8 Hz), 7.80 (d, 2H, *J* = 8.5 Hz), 10.07 (s, 1H)

3-Bromophenyl 4-methylbenzenesulfonate (5b)

The reaction was conducted by combining 3-bromophenol (0.866 g, 5.05 mmol), *para*-toulenesulfonylchloride (0.647g, 3.39 mmol), and dimethylformamide (15 mL) into a 20 mL microwave safe vial. After one minute of stirring, *N*,*N*-diisopropylethylamine (0.647 g, 5.00 mmol) was added. The reaction mixture was allowed to stir for one minute to ensure homogeneity. The reaction solution was then placed into the microwave

reaction apparatus, the Biotage Initiator, and was stirred for 30 min at 60 ^{o}C under constant microwave irradiation. The contents of the reaction vessel were transferred to a separatory funnel and 20 mL of deionized water was added. The products were isolated by extraction with EtOAc (3 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford the tosylate protected analogue **5b** (0.824 g, 2.65 mmol, 53 %) as a solid.

^{1}H-NMR (500 MHz, $CDCl_3$) δ 2.46 (s, 3H), 6.93 (ddd, 1H, *J* = 8.5 Hz, 10 Hz, 8.5 Hz), 7.25 (tt, 2H, *J* = 7 Hz, 8 Hz, 8 Hz, 7 Hz), 7.34 (d, 2H, 8.5 Hz), 7.38 (ddd, 1H, *J* = 8.6 Hz, 10 Hz, 8.6 Hz), 7.725 (dd, 2H, *J* = 6.5 Hz, 7.0 Hz).

Benzosuberene Synthesis

5-(2-Methoxyphenyl)pent-4-enoic acid (6)

To a solution of 4-(bromotriphenylphosphoranyl)butanoic acid (32.1 g, 74.9 mmol) in THF (200 mL) in a flask, KO*t*Bu (16.7 g, 148 mmol) was added and the reaction mixture was allowed to stir for 15 mintues. 2-Methoxybenzaldehyde (6.88 g, 50.5 mmol) dissolved in in THF (25 mL) was added drop wise to the reaction mixture over 5 minutes. The reaction was stirred continuously for 5 hours. On completion the reaction was quenched with H_2O (50 mL) and extracted with EtOAc (4 x 15 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford 5-(2-methoxyphenyl)pent-4-enoic acid**, 6** (8.34 g, 40.4 mmol, 80 %).

5-(2-Methoxyphenyl)pentanoic acid (7)

The reaction, compound **7**, was conducted by combining 5-(2-methoxyphenyl)pent-4-enoic acid (8.34 g, 40.4 mmol) to EtOH (100 mL) and was allowed to stir for 1 minute in a 250 mL roundbottom flask. Next 10% Pd-C (0.40 g) was added to the reaction vessel. Following the addition of the catalyst, 3 liters of H_2 (0.27 g, 269 mmol) were introduced into the reaction vessel via rubber balloons. The reaction was subjected to constant stirring for 24 hours. The crude product was isolated by filtration and the excess EtOH was removed under reduced pressure (rotary evaporation) to afford 5-(2-methoxyphenyl)pentanoic acid, **7** (5.16 g, 24.8 mmol, 61 %).

1-Methoxy-6,7,8,9-tetrahydro-5H-benzo[7]annulen-5-one (8)

Compound **8**, was synthesized by combining 5-(2-methoxyphenyl) pentanoic acid (1.00 g, 4.81 mmol) to Eaton's reagent (60 mL) , 1-methoxy-6,7,8,9-tetrahydro-5H-benzo[7]annulen-5-one (17.8 g, 74.7 mmol) in a round bottom flask. The reaction was subjected to constant stirring for 48 hours. The product was quenched with H_2O (100 mL) and extracted with EtOAc (4 x 50 mL). The combined EtOAc layers were dried over Na_2SO_4 (anhydrous). After filtration, the solvent was removed under reduced pressure (rotary evaporation) to afford 1-methoxy-6,7,8,9-tetrahydro-5H-benzo[7]annulen-5-one, **8** (0.512 g, 2.69 mmol, 56 %)

CHAPTER Three

Results and Discussion

Over the course of this study, close to thirty different compounds were modified with phenolic protecting groups. Many of the compounds were either synthesized in low yields or were contaminated with impurities. Of the thirty compounds synthesized, five yielded promising results. Figures 4.0 and 5.0 summarize the protection of five compounds studied in this thesis.

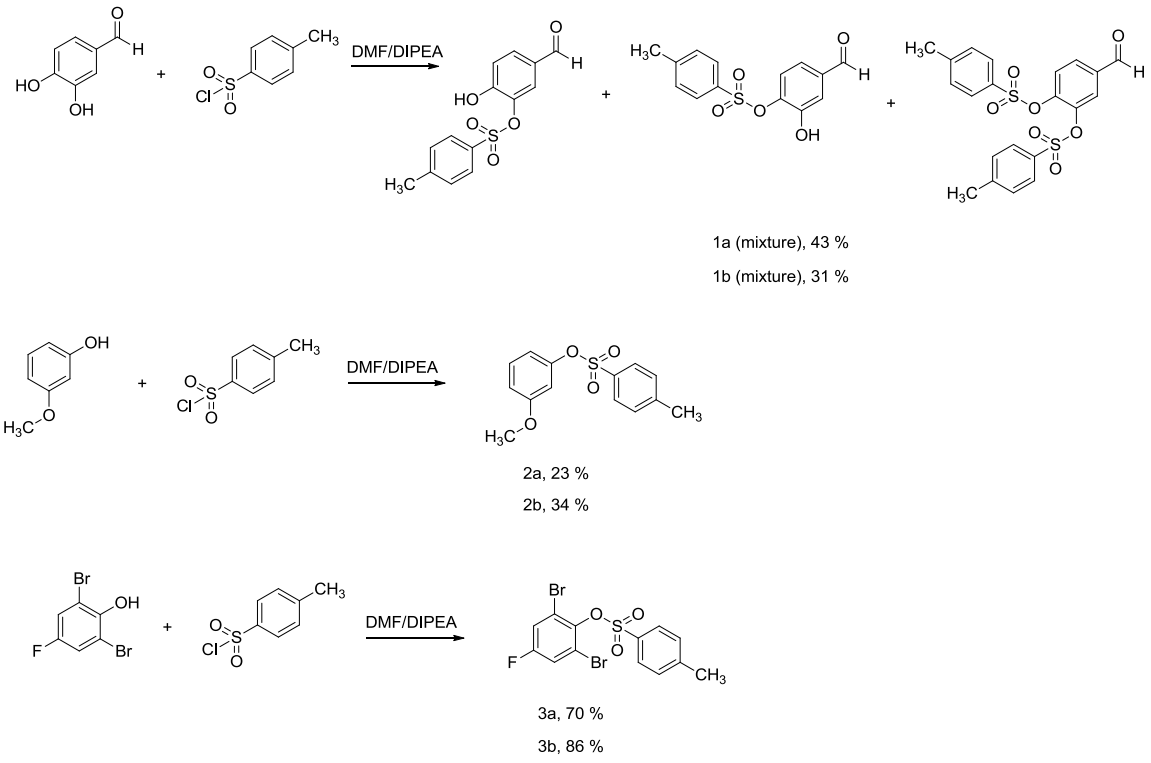


Figure 4.0: Reaction scheme for compounds 1a, 2a, 3a, 1b, 2b, 3b. Compounds 1a-3a were synthesized by microwave irradiation while compounds 1b-3b were synthesized by the conventional method.

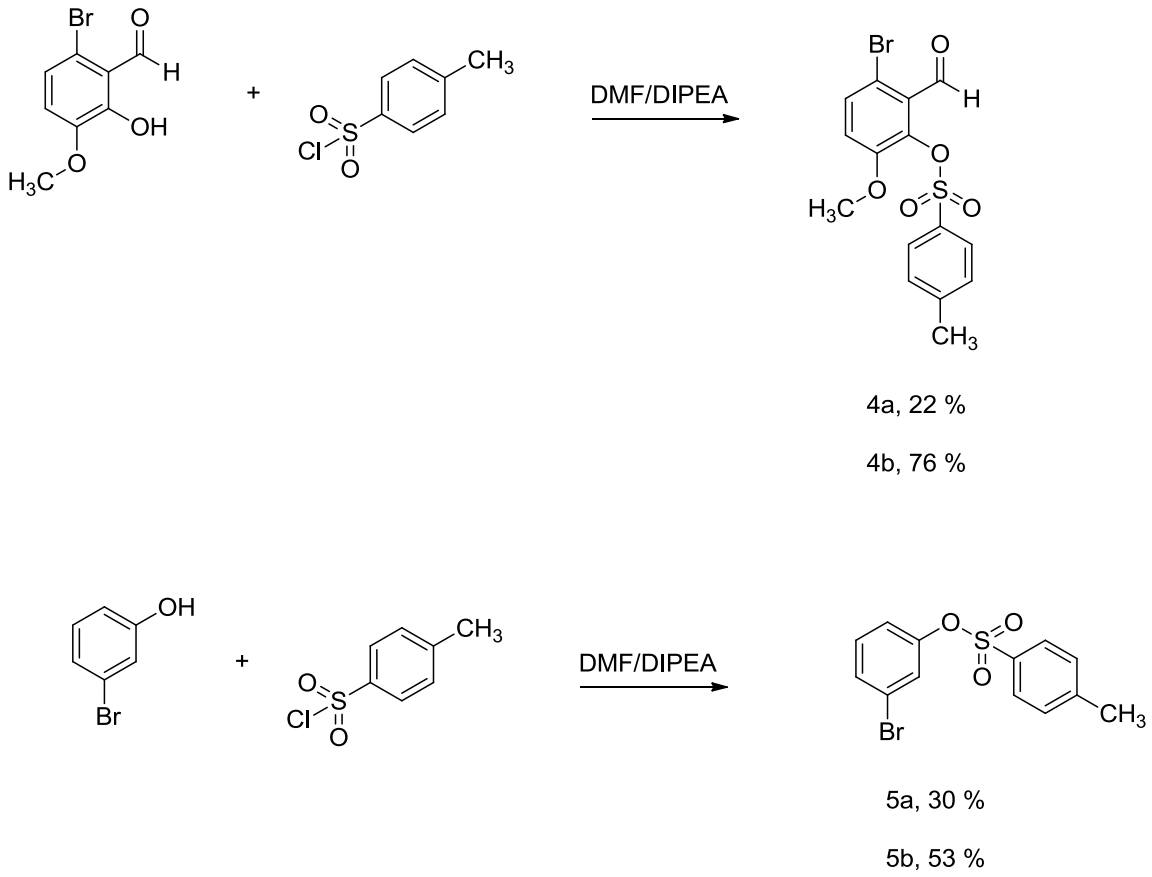


Figure 5.0: Reaction scheme for compounds 4a, 5a, 4b, 5b.
Compounds 4a-5a were synthesized by microwave irradiation while compounds 4b-5b were synthesized by the conventional method.

Figure 4.0 and 5.0 outline the steps taken to prepare the protected compounds, **1a-5a, 1b-5b**. It quickly became evident that the microwave reactions, **1a-5a**, resulted in lower yields. After successfully completing the protection reactions summarized in figure 4.0 and 5.0, NMR analyses were conducted to ensure that the compounds isolated were indeed the ones that were desired. Although thirty reactions were performed, only five of them were significant. This study proposes that there were either thermodynamic and or structural barriers that did not allow these compounds to be synthesized. Nonetheless, each reaction was completed and purified using column chromatography. In general, NMR analyses were conducted to properly characterize the reaction products. It is worthy to note that the NMR spectra for compound **1a** and **1b** are not included. Due to the presence of two hydroxyl groups, there are three possible outcomes of the protection reaction. Since the product mixture of compounds was not separated into individual compounds, their NMR spectra are not included. The yields for the protected compounds can be found in figure 4.0 and 5.0

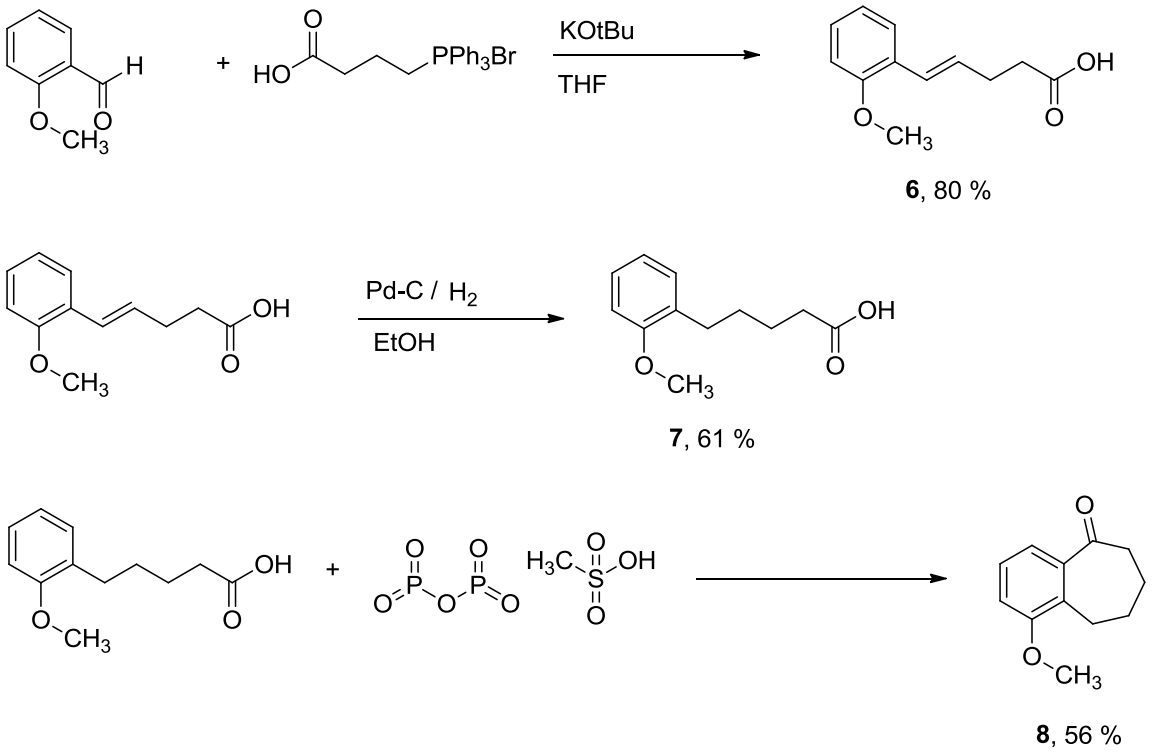


Figure 6.0: Reaction scheme for compounds 6-8

Figure 6.0 outlines the synthesis of the final product, compound **8**. Compound **6**, synthesized through a Wittig reaction, was reduced to afford compound **7.** Cyclization proceeded through an Eaton's reagent mediated Friedel-Crafts acylation reaction resulting in compound **8**. The presence of a methoxy group on the benzene ring became a problem during the cyclization of compound **7**. Because the methoxy group is an ortho-para director, it is hypothesized that compound **8** was being synthesized as an eight membered ring rather than a seven membered ring. However, it is important to also note that the occurrence of an intermolecular reaction between compound **7** cannot be ruled out. Despite multiple attempts to synthesize the desired compound, at this point it seems

that the cyclization of compound **7** to compound **8** is not feasible due to the presence of the ortho-para directing methoxy group.

CHAPTER FOUR

Conclusions

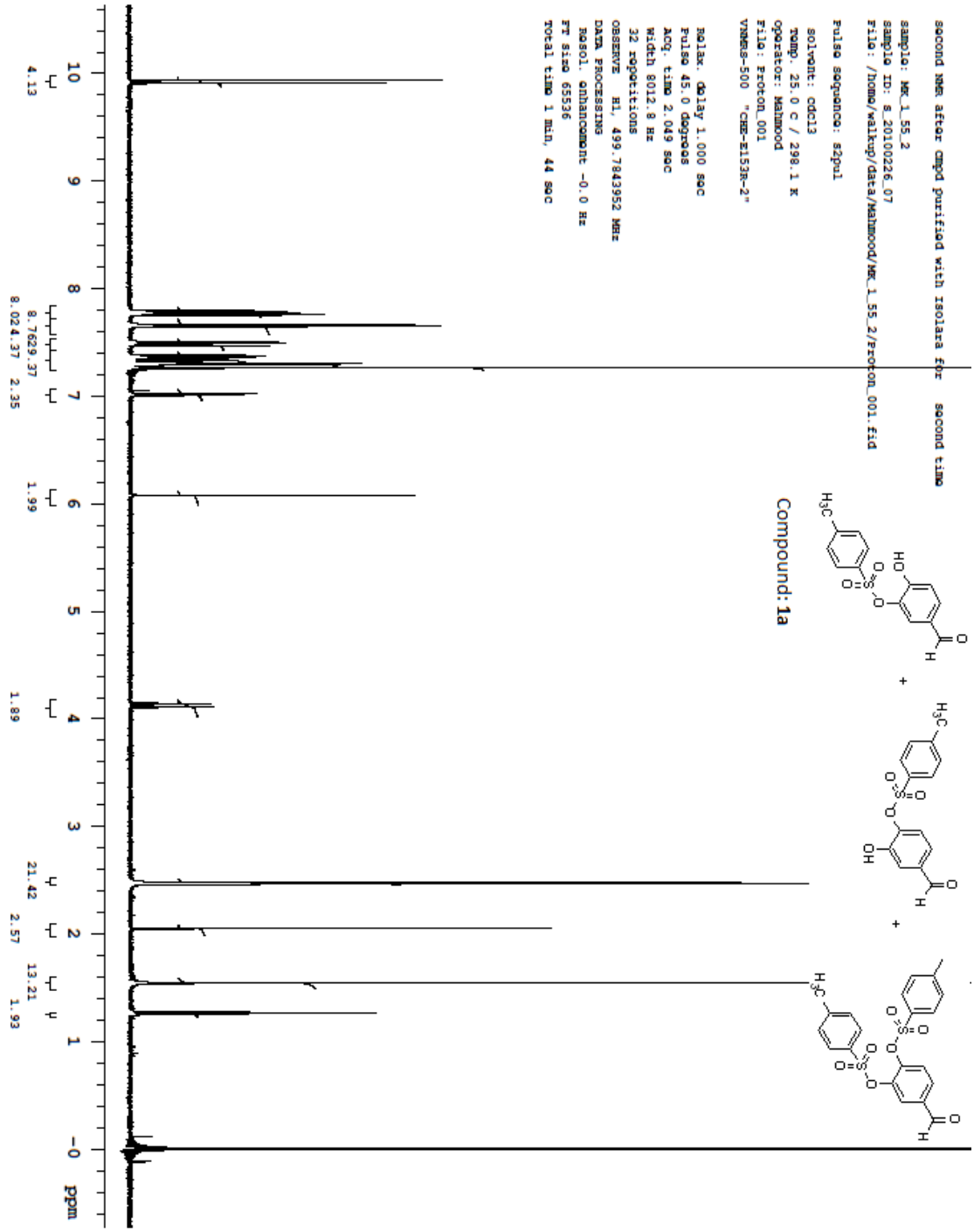
The application of efficient protecting group strategies is paramount for the successful preparation of a functional target molecule through multi-step synthetic routes. This study evaluates the installation of tosyl protecting groups on phenolic moieties utilizing both microwave and conventional strategies. For a subset of the compounds studied, conventional heating was more effective than microwave irradiation. Future studies will survey a larger sample of tosylate reactions under microwave conditions. In addition, it is believed that applying this concept to bio-reductive triggers could be very useful.

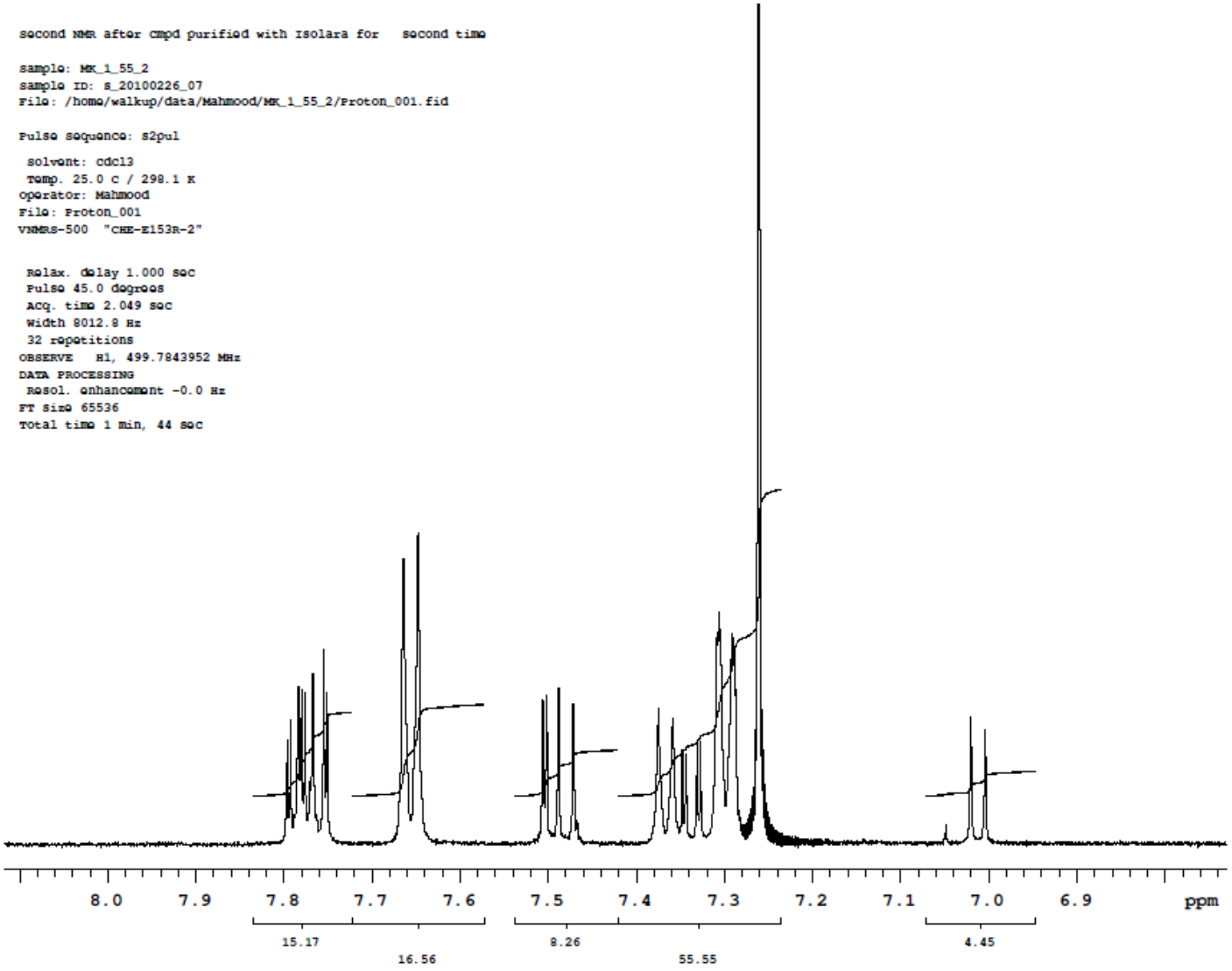
Moreover, the goal to synthesize benzosuberene was not successful. In the future, the hope is to apply the knowledge gained from this study in the installation of protecting groups to help combat the incorrect cyclization of compound **8**. Nonetheless, the synthetic campaign towards a benzosuberene analogue yielded valuable practical knowledge in the realm of anti-cancer compound synthesis.

The synthesis documented in this outline a method for installing protecting groups on phenolic moieties and a route for synthesizing a benzosuberene, an anti-cancer agent. Despite some hurdles encountered in this project, it is reported that a conventional heating method for installing protecting groups is most efficient and that the synthesis of a benzosuberene analogue was not successful due to unforeseen circumstances.

APPENDIX

Appendix A: NMR Spectra

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solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
8.0
7.9
7.8
7.7
7.6
7.5
7.4
7.3
7.2
7.1
7.0
6.9
ppm
15.17
16.56
8.26
55.55
4.45

```
second NMR after cmpd purified with Isolara for   second time

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Sample ID: S_20100226_07
File: /home/walkup/data/Mahmood/MK_1_55_2/Proton_001.fid

Pulse Sequence: s2pul

 Solvent: cdcl3
 Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500  "CHE-E153R-2"


 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 Acq. time 2.049 sec
 Width 8012.8 Hz
 32 repetitions
OBSERVE   H1, 499.7843952 MHz
DATA PROCESSING
 Resol. enhancement -0.0 Hz
FT Size 65536
Total time 1 min, 44 sec
```

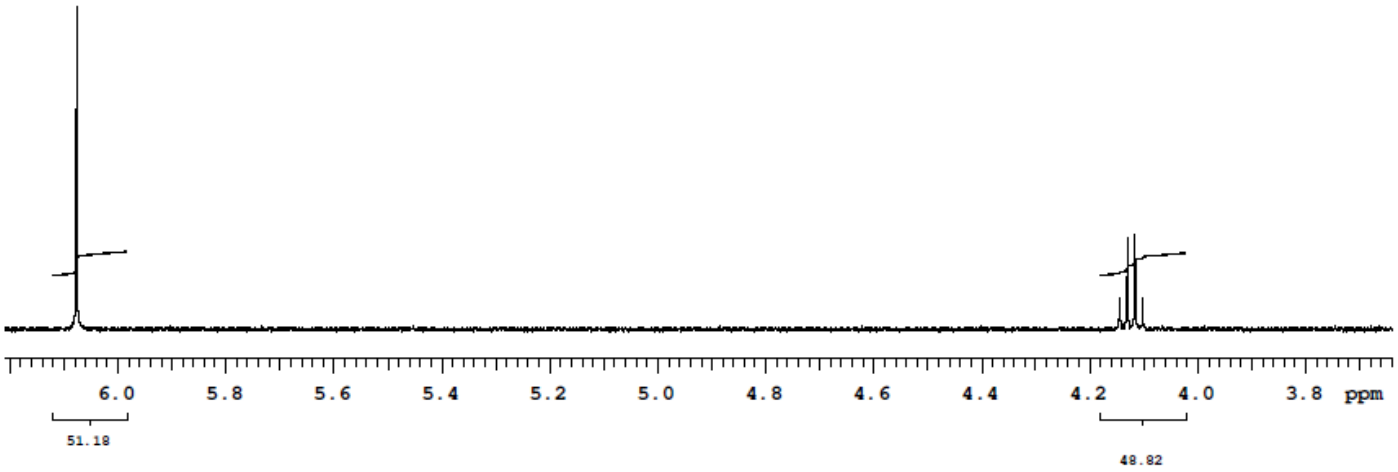

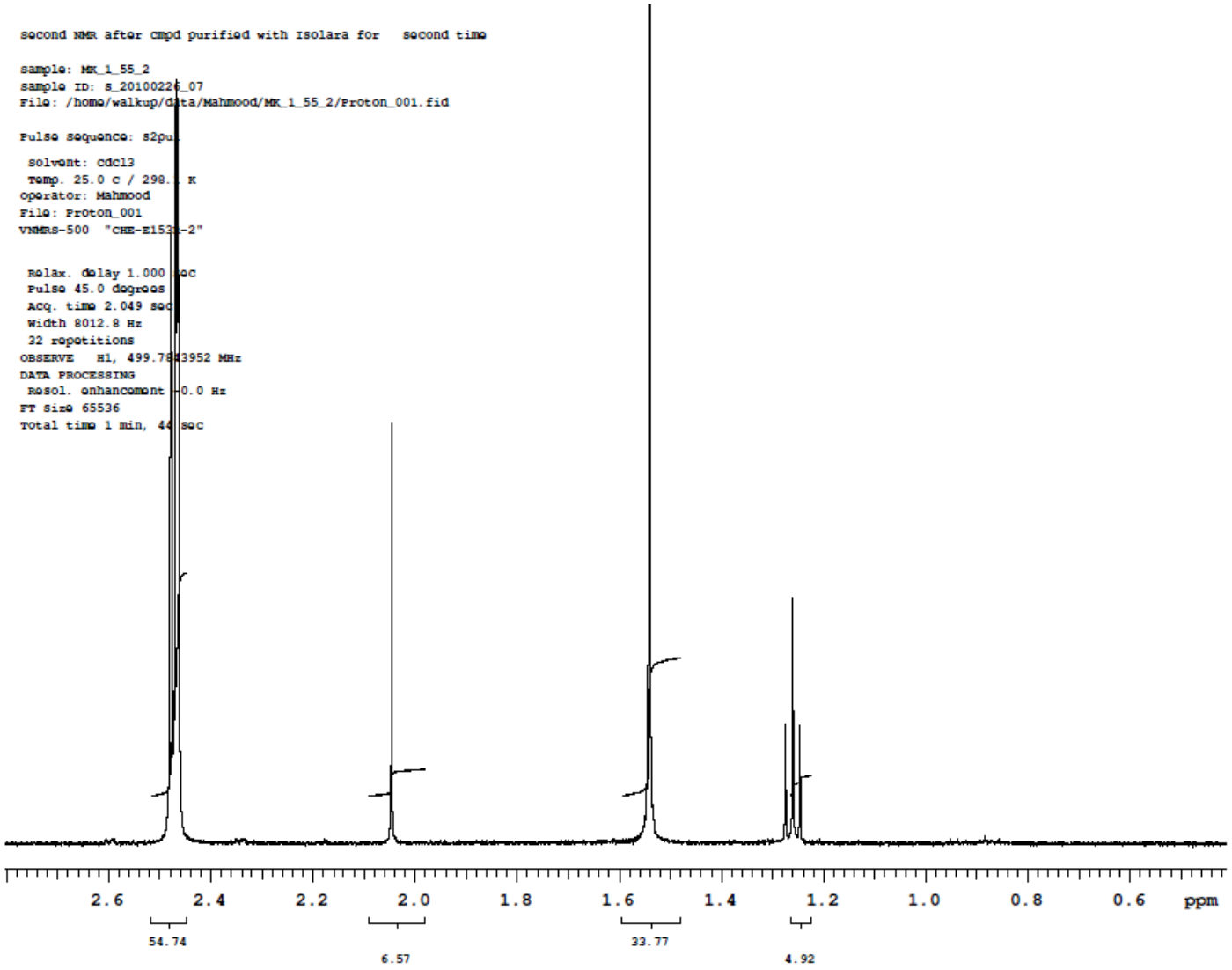
second NMR after cmpd purified with Isolara for second time
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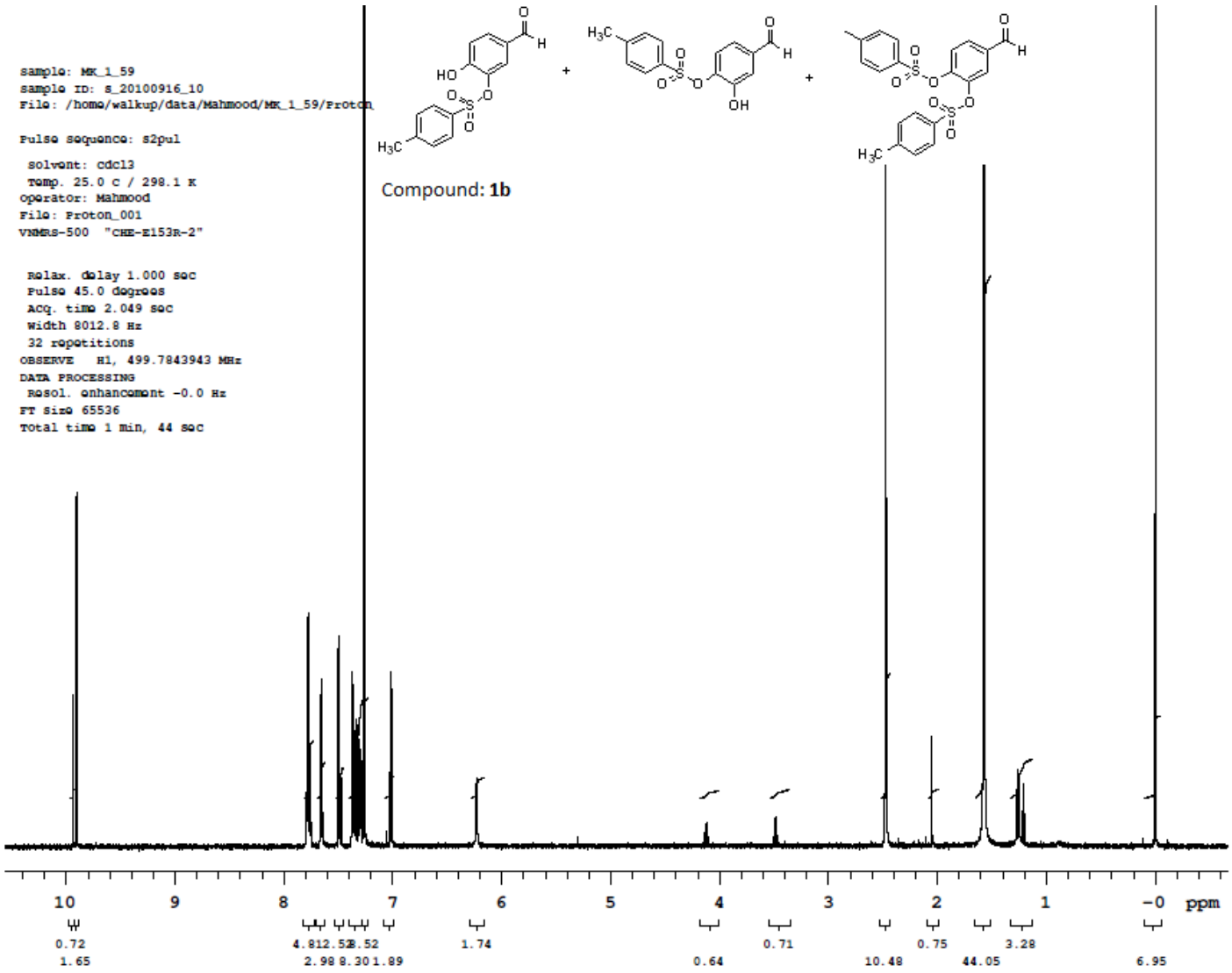
Temp. 25.0 c / 298. K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 4
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
ppm
54.74
6.57
33.77
4.92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Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
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Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
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DATA PROCESSING
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FT size 65536
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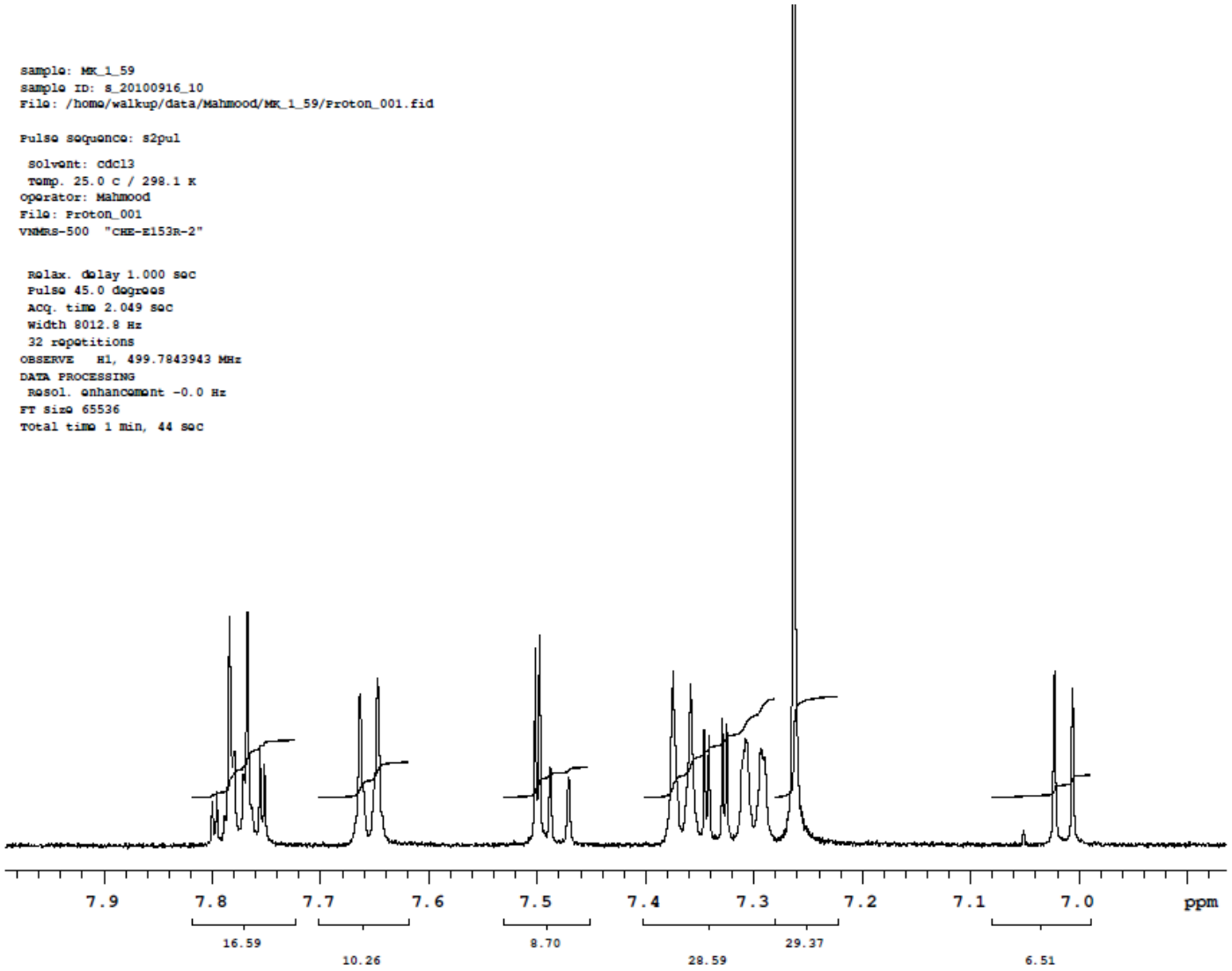
Compound: 1b
10
9
8
7
6
5
4
3
2
1
-0
ppm
0.72
1.65
4.81
2.98
2.52
8.30
1.89
1.74
0.64
0.71
10.48
0.75
44.05
3.28
6.95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Operator: Mahmood
File: Proton_001
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Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
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Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
7.9
7.8
7.7
7.6
7.5
7.4
7.3
7.2
7.1
7.0
ppm
16.59
10.26
8.70
28.59
29.37
6.51

```
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 Solvent: cdcl3
 Temp. 25.0 c / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500  "CHE-E153R-2"


 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 Acq. time 2.049 sec
 Width 8012.8 Hz
 32 repetitions
OBSERVE   H1, 499.7843943 MHz
DATA PROCESSING
 Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
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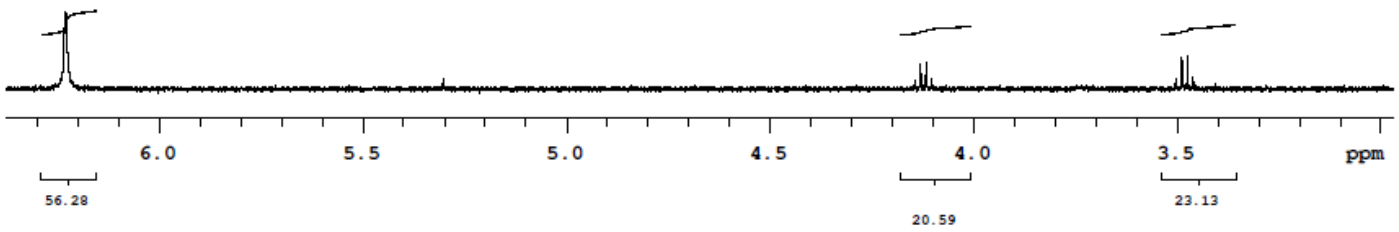

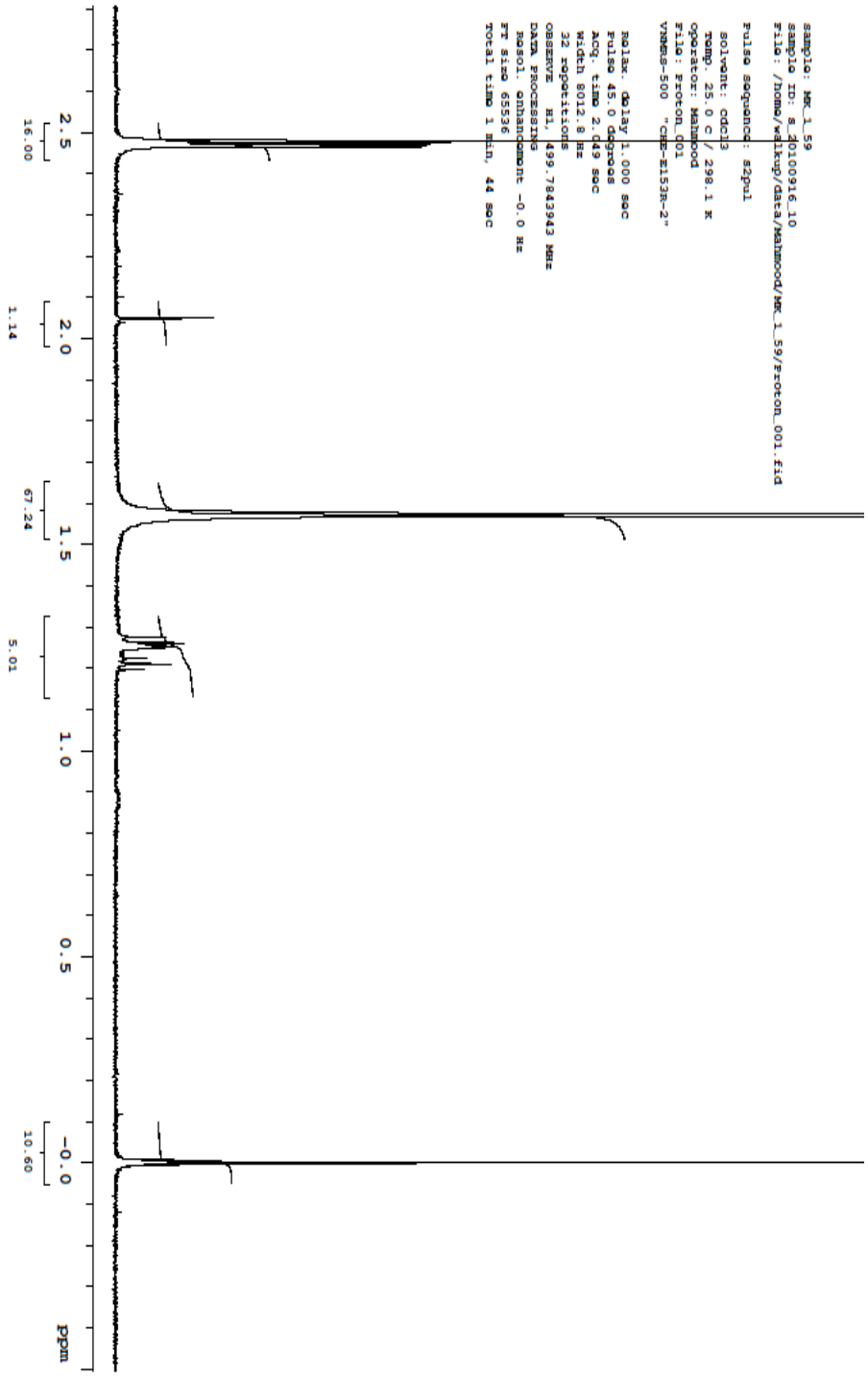
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operator: Mahmood
file: Proton_001
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Pulse 45.0 degrees
Acq. time 2.049 sec
width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
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1.5
1.0
0.5
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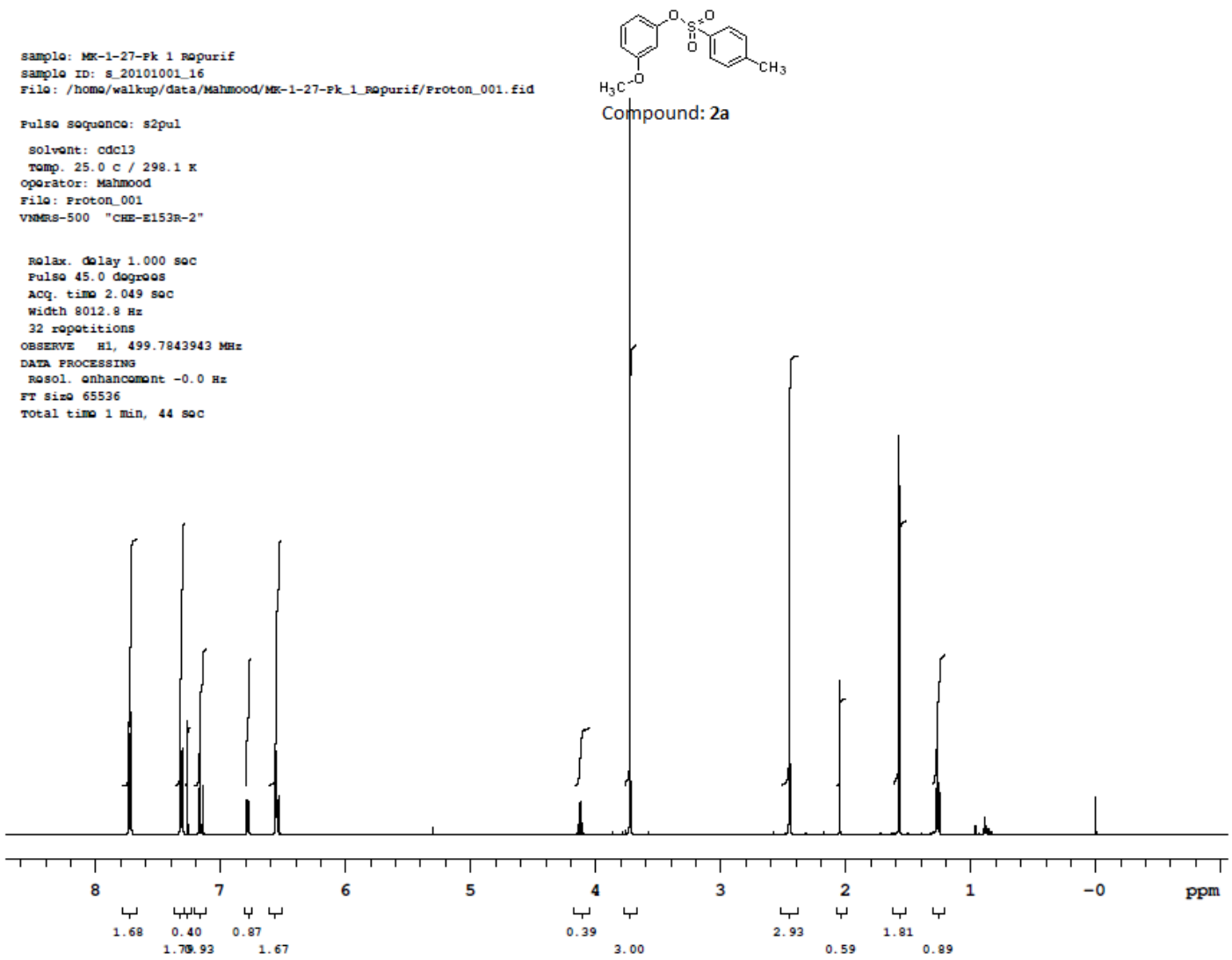
ppm
16.00
1.14
67.24
5.01
10.60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Solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
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Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
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8
7
6
5
4
3
2
1
-0
ppm
1.68
0.40
0.87
1.70
0.93
1.67
0.39
3.00
2.93
0.59
1.81
0.89

```
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Sample ID: s_20101001_16
File: /home/walkup/data/Mahmood/MK-1-27-Pk_1_Repurif/Proton_001.fid

Pulse sequence: s2pul

 Solvent: cdcl3
 Temp. 25.0 c / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500  "CHE-E153R-2"


 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 Acq. time 2.049 sec
 Width 8012.8 Hz
 32 repetitions
OBSERVE   H1, 499.7843943 MHz
DATA PROCESSING
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FT size 65536
Total time 1 min, 44 sec
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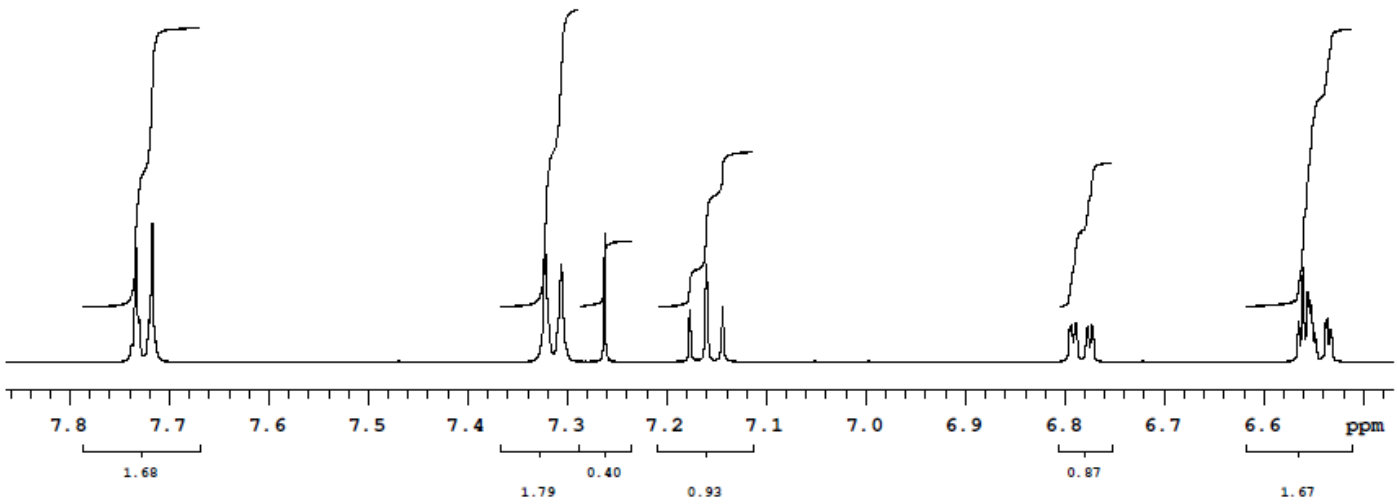

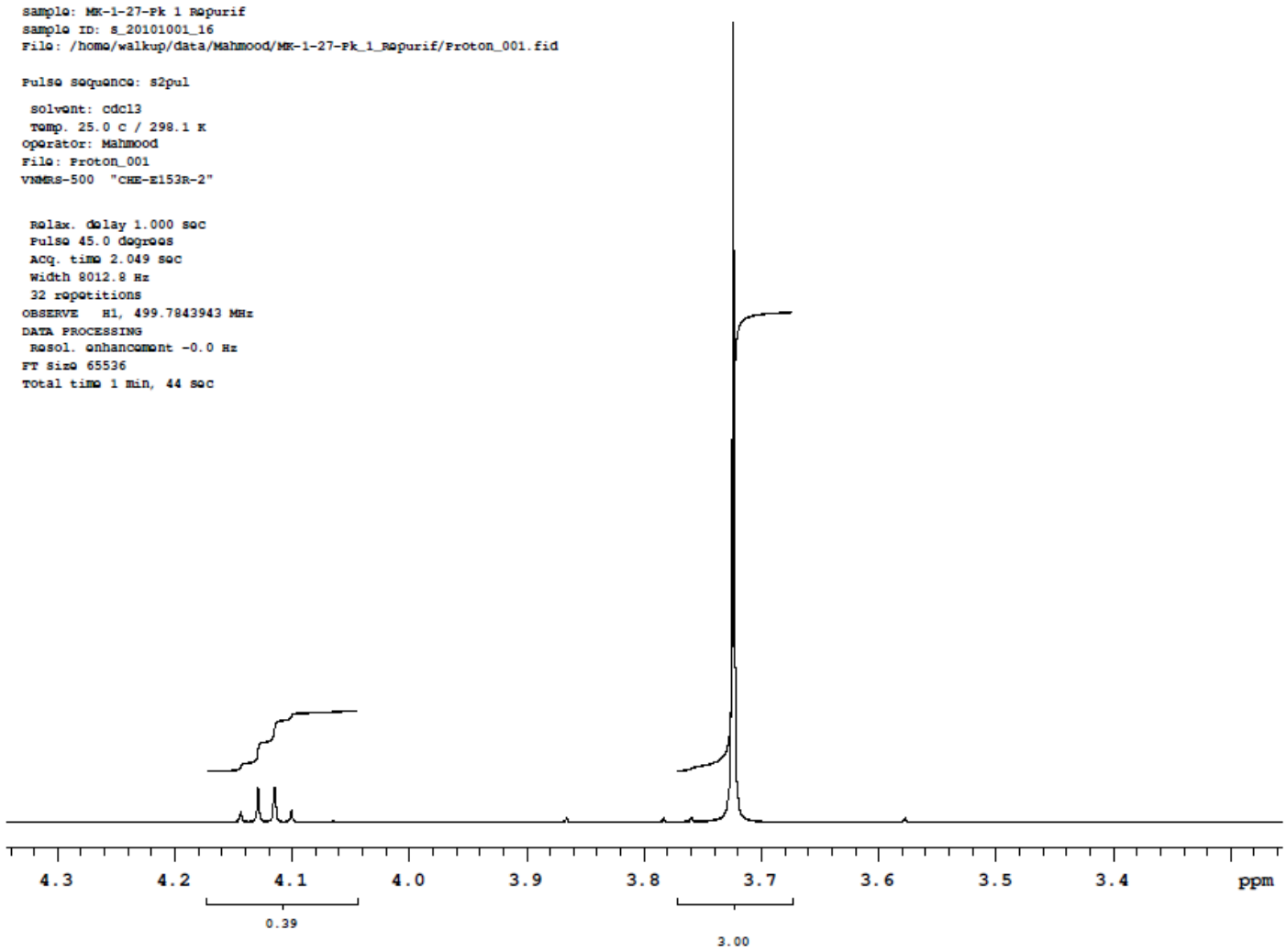
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ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
DATA PROCESSING
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FT Size 65536
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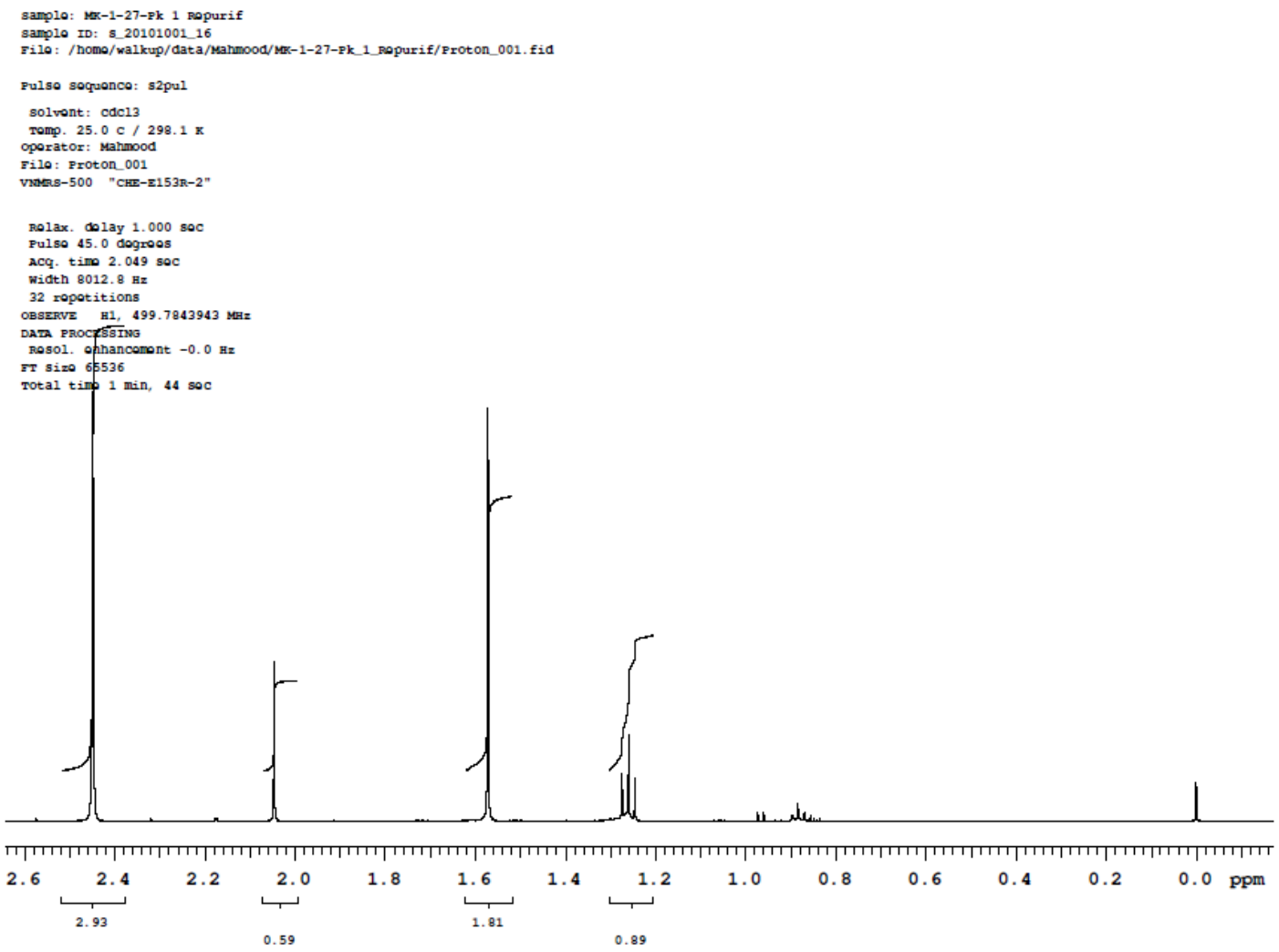
4.2
4.1
4.0
3.9
3.8
3.7
3.6
3.5
3.4
ppm
0.39
3.00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Operator: Mahmood
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ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
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DATA PROCESSING
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FT size 65536
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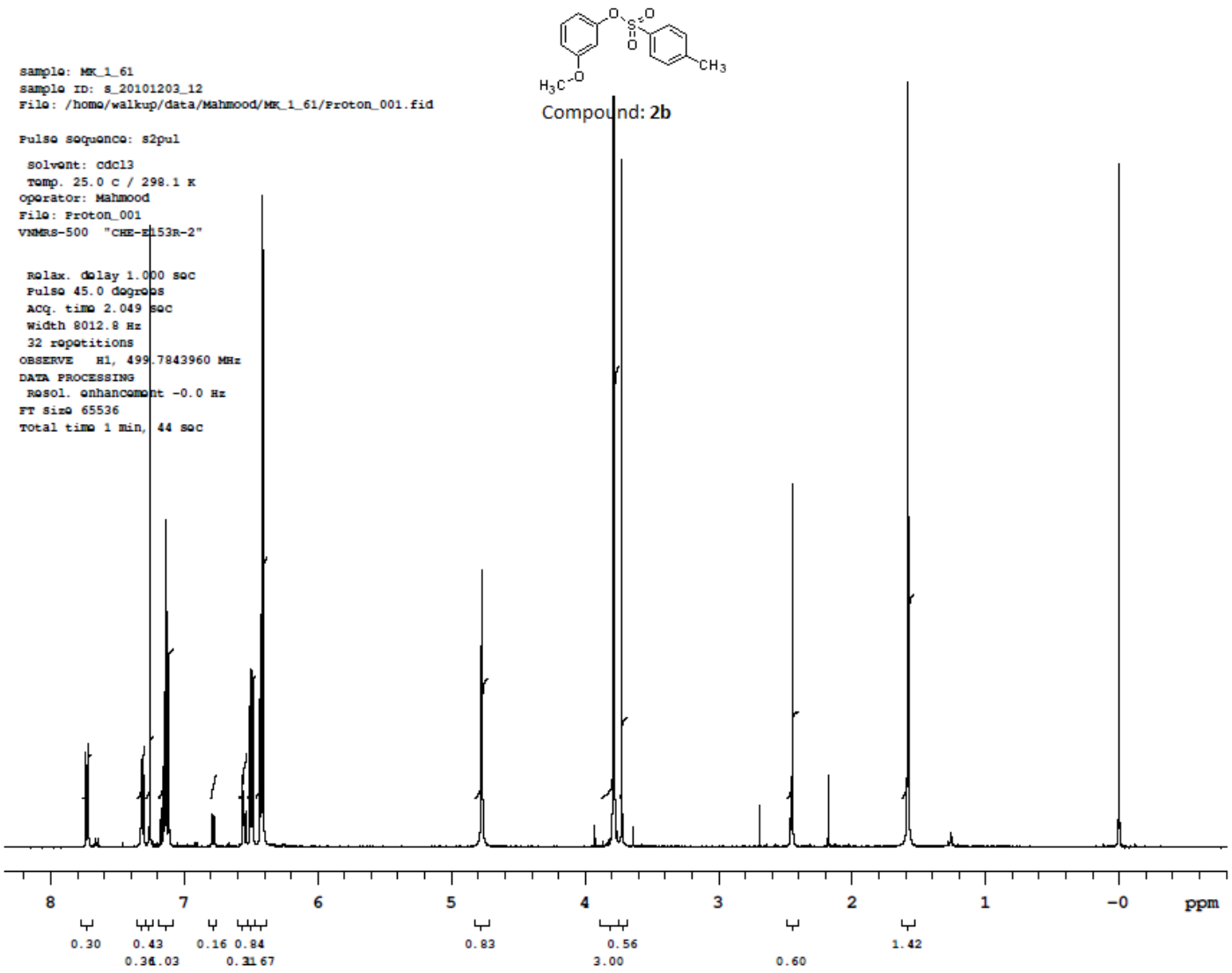
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
ppm
2.93
0.59
1.81
0.89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Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843960 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Compound: 2b
8
7
6
5
4
3
2
1
-0
ppm
0.30
0.43
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0.84
0.83
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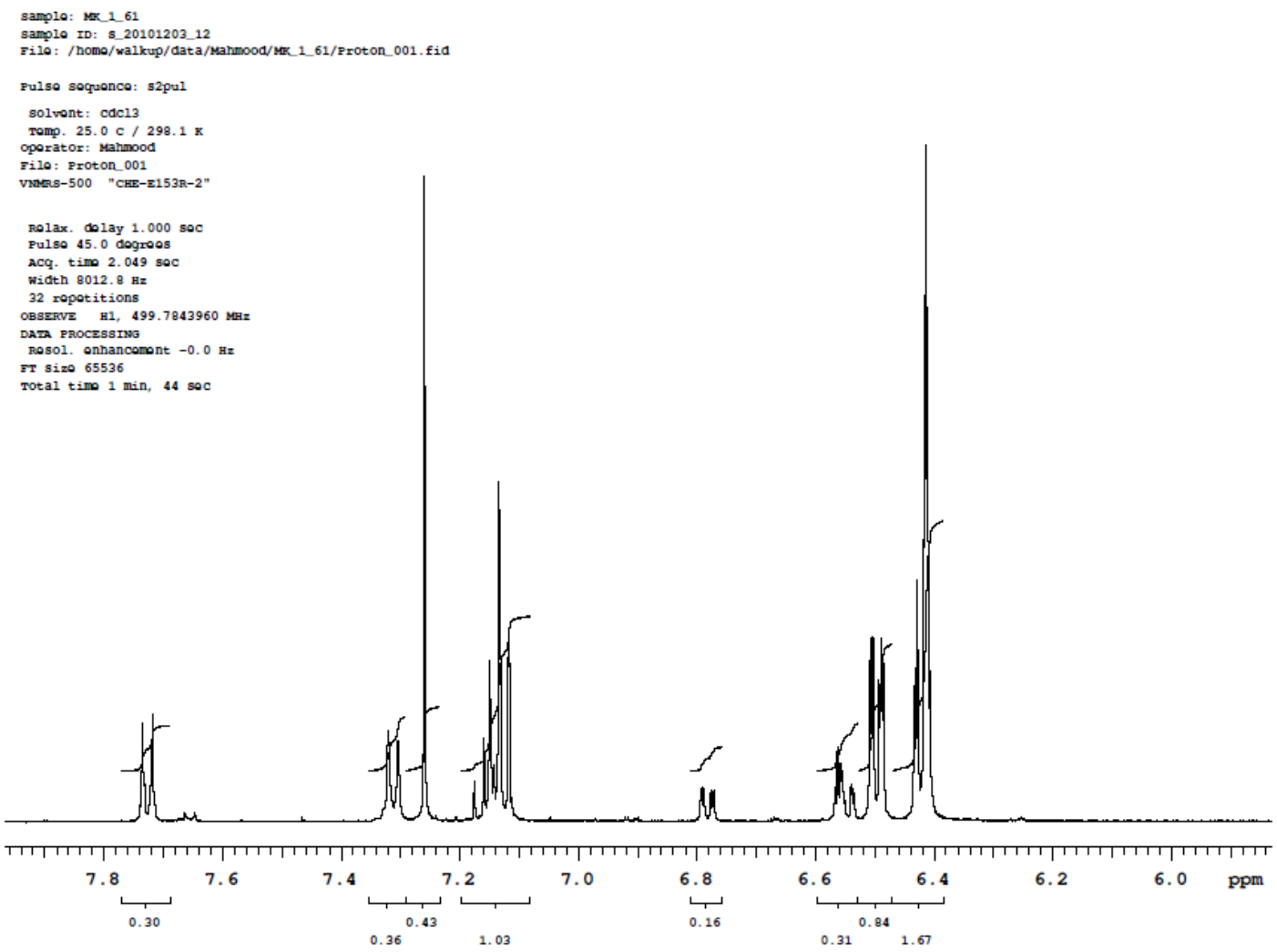
1.67
3.00
0.60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Operator: Mahmood
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Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843960 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
7.8
7.6
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
ppm
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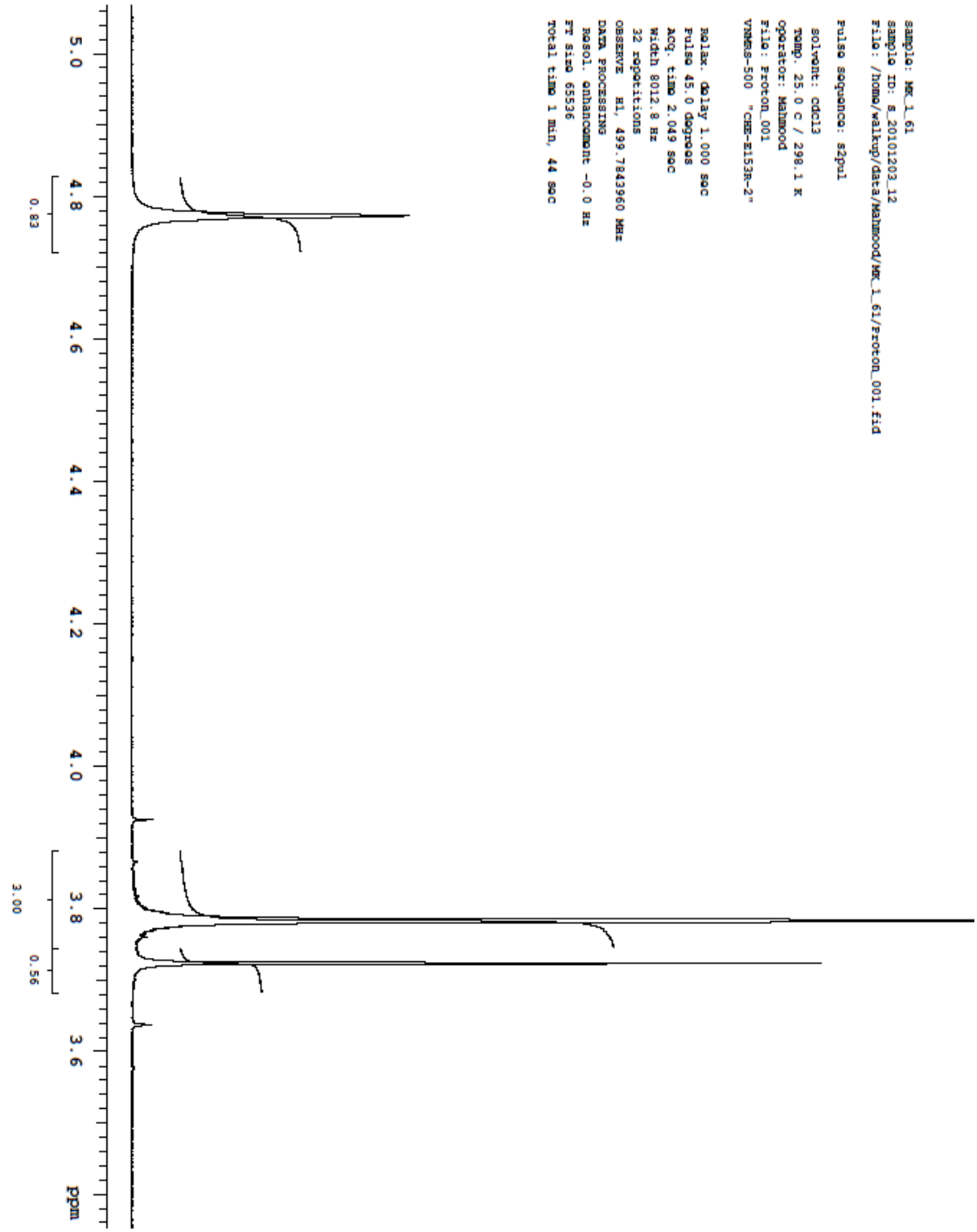
0.36
0.43
1.03
0.16
0.31
0.84
1.67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solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
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Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843960 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
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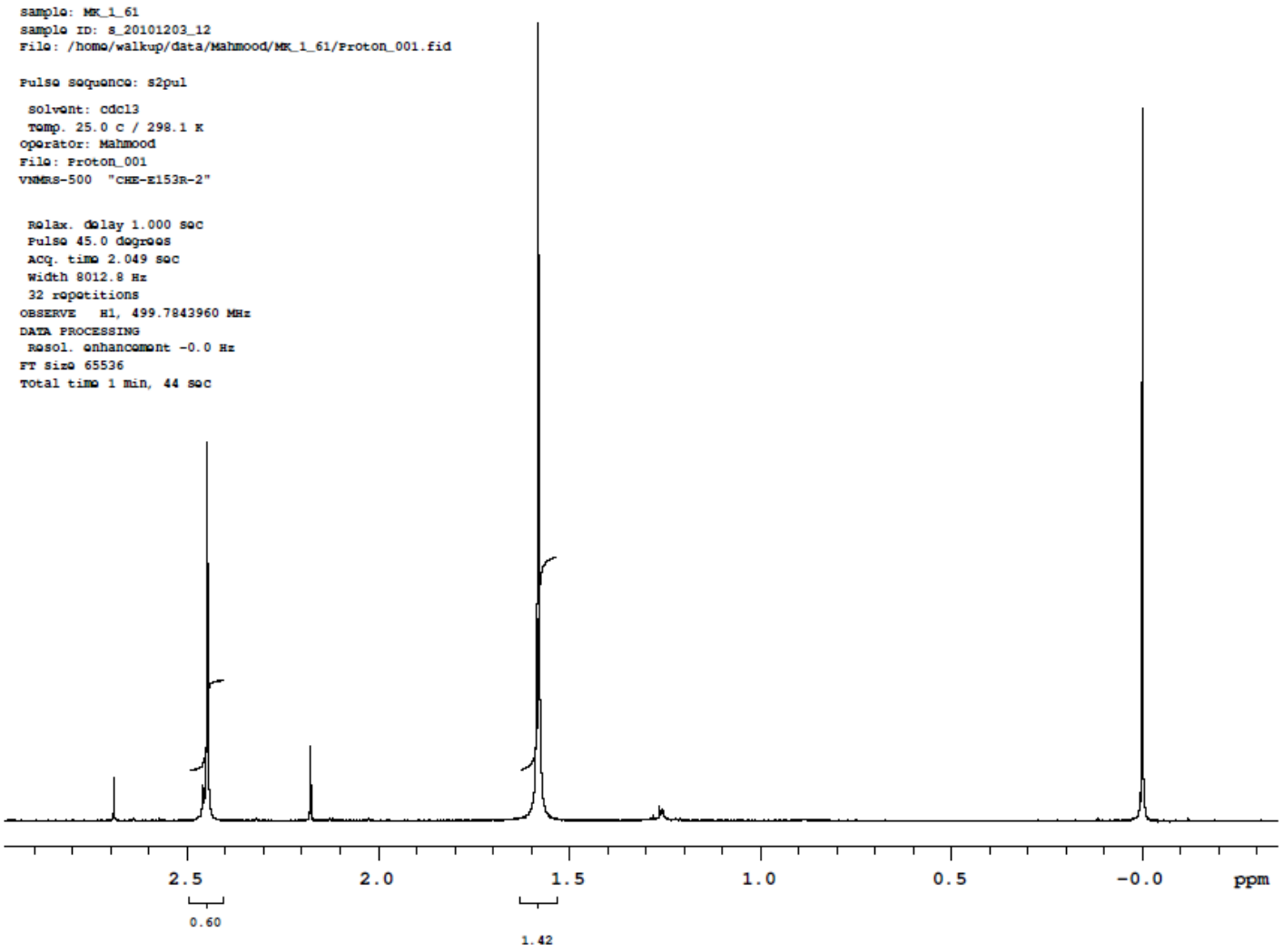
4.6
4.4
4.2
4.0
3.8
3.6
ppm
0.83
3.00
0.56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Operator: Mahmood
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Pulse 45.0 degrees
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Width 8012.8 Hz
32 repetitions
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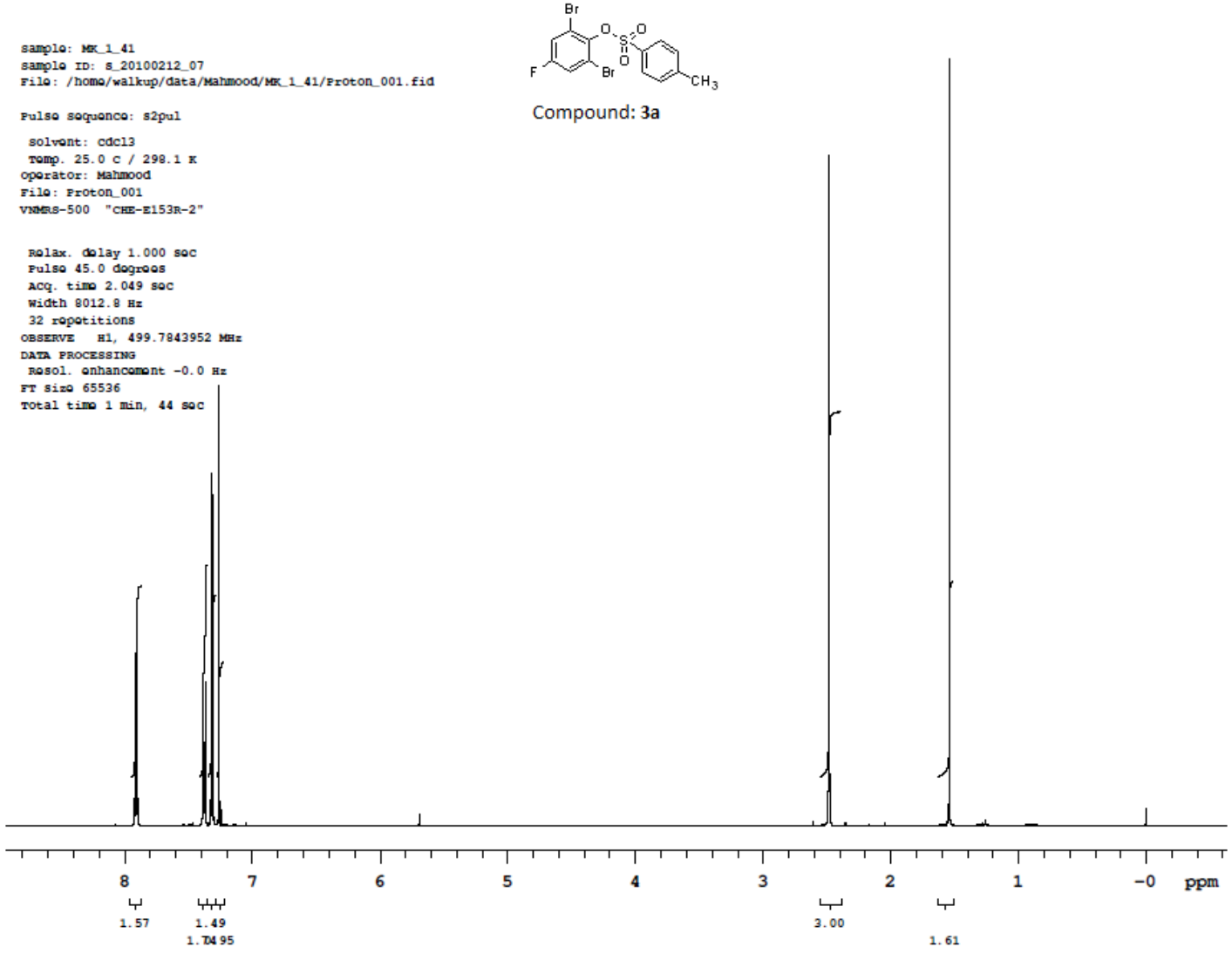
1.5
1.0
0.5
-0.0
ppm
0.60
1.42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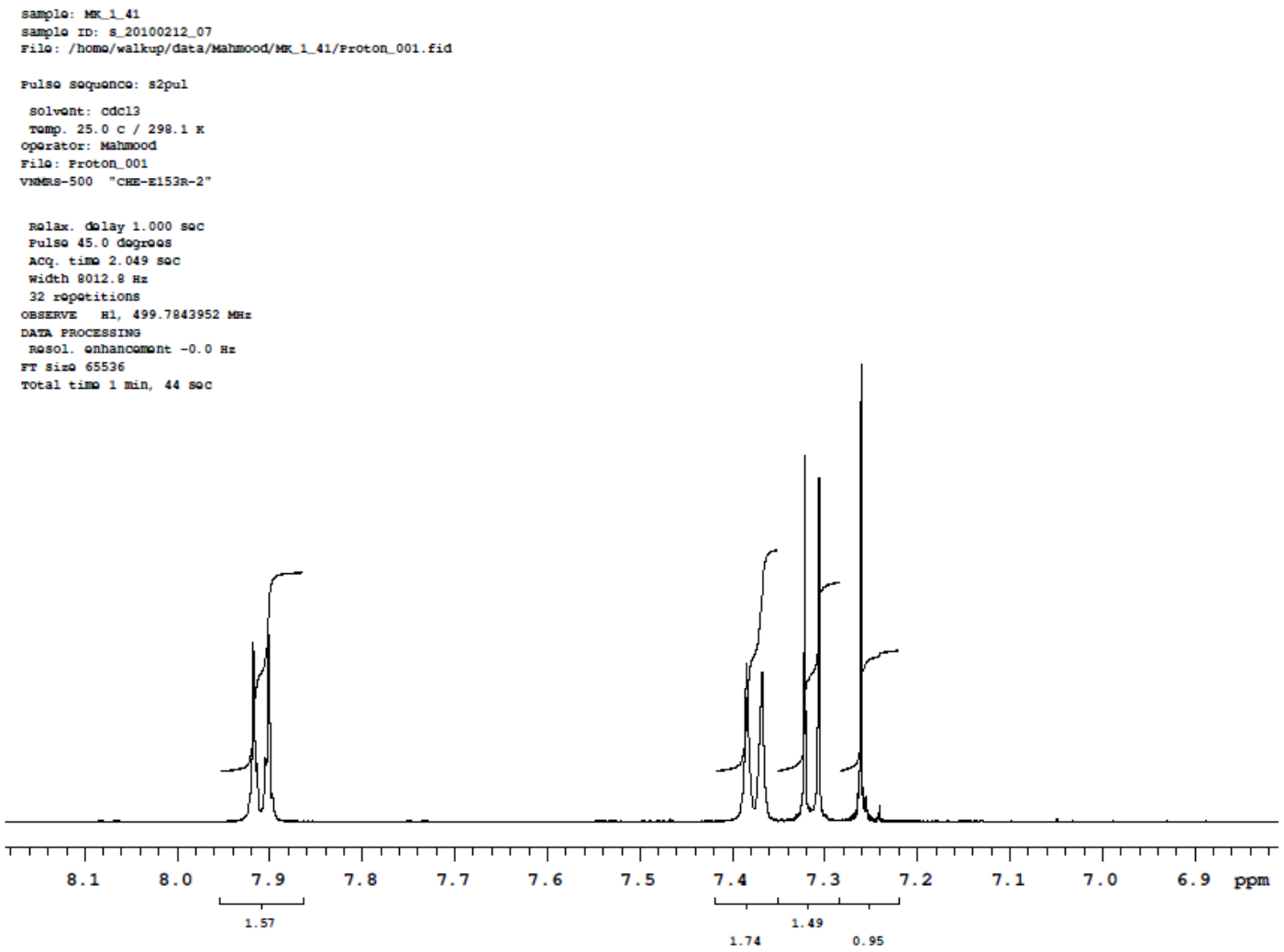
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Br
O
S
O
O
F
Br
CH3
Compound: 3a
8
7
6
5
4
3
2
1
-0
ppm
1.57
1.49
1.7495
3.00
1.61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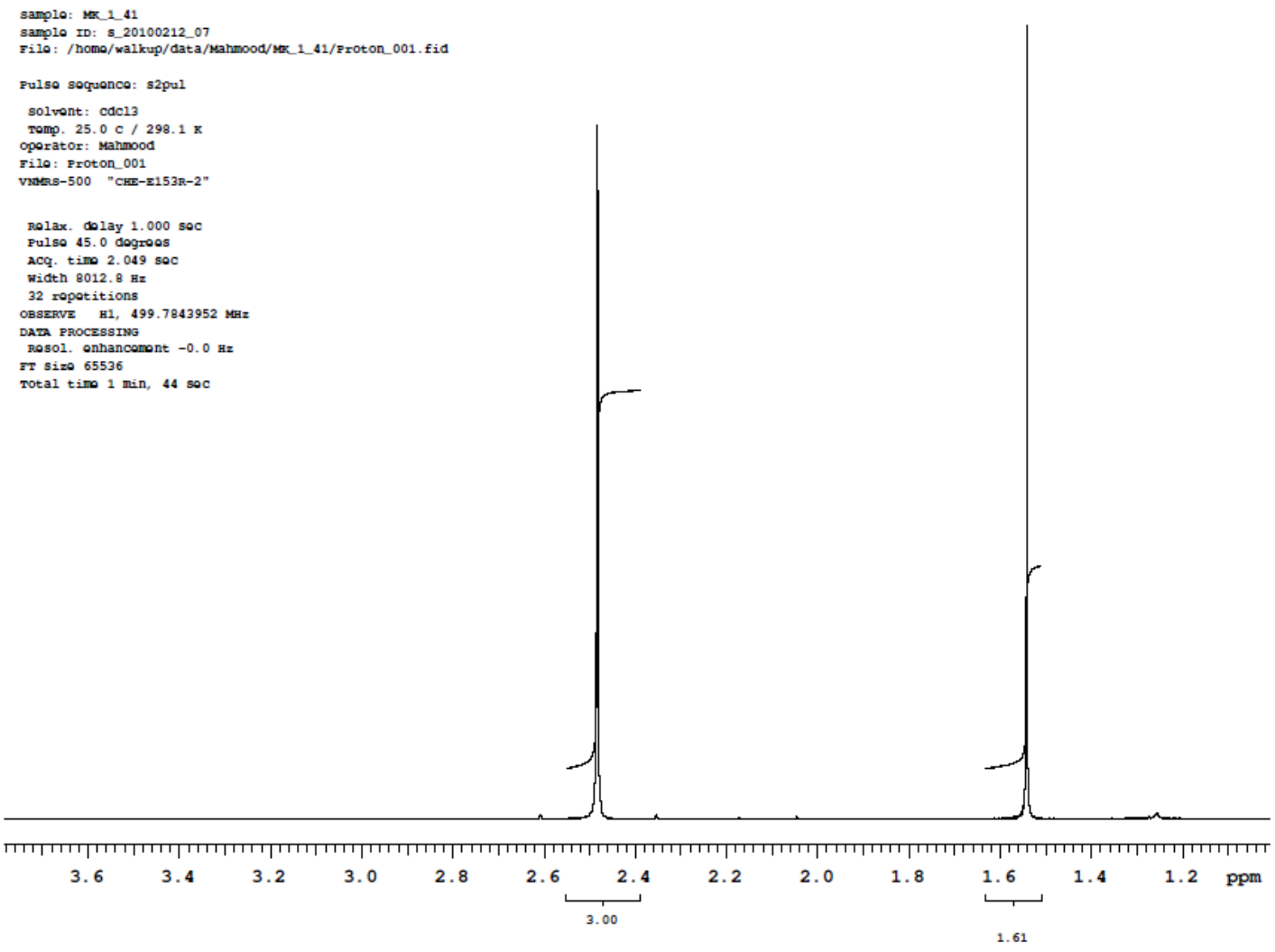
sample ID: s_20100212_07
File: /home/walkup/data/Mahmood/MK_1_41/Proton_001.fid
Pulse sequence: s2pul
Solvent: cdcl3
Temp. 25.0 c / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
8.1
8.0
7.9
7.8
7.7
7.6
7.5
7.4
7.3
7.2
7.1
7.0
6.9
ppm
1.57
1.74
1.49
0.95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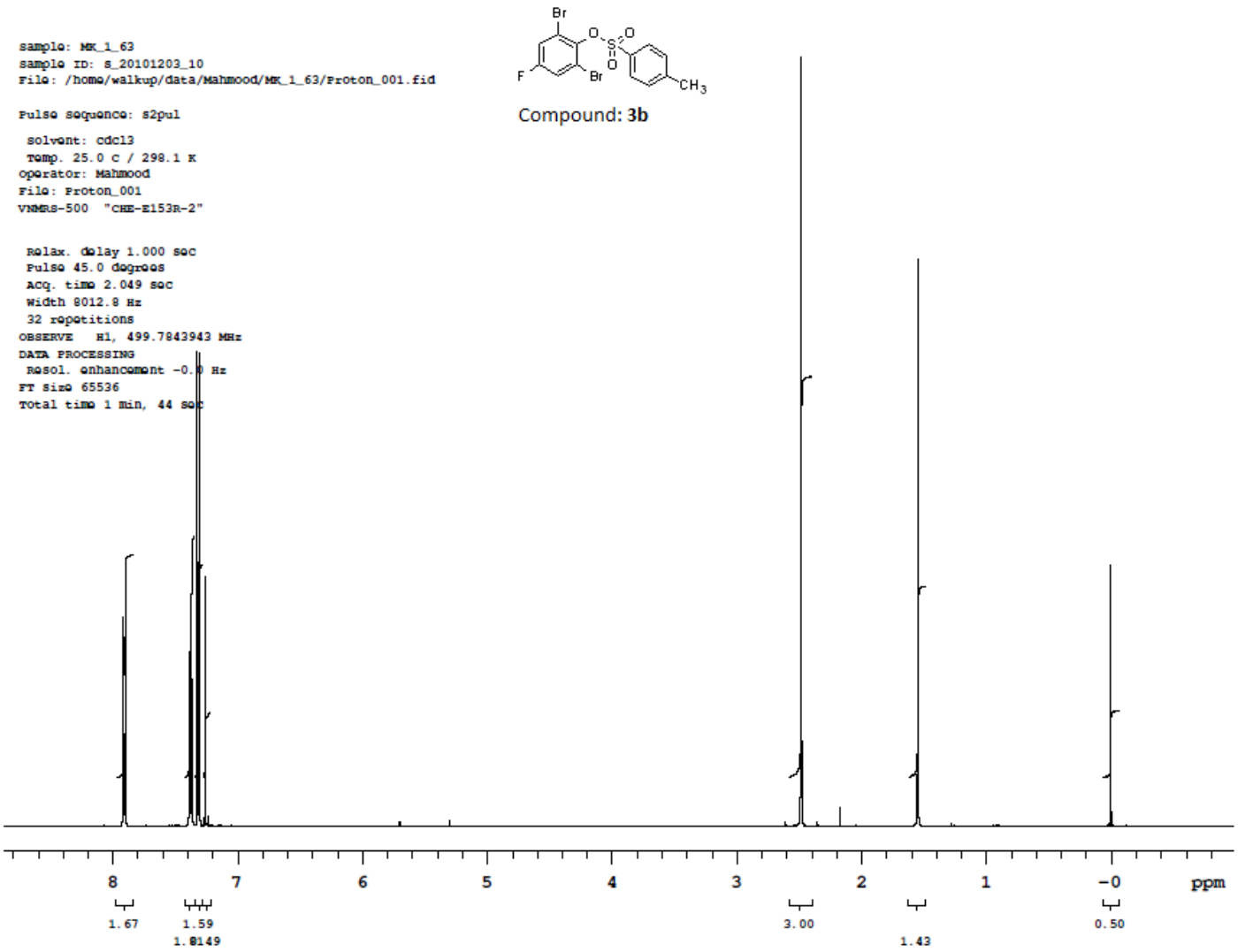
sample ID: s_20100212_07
File: /home/walkup/data/Mahmood/MK_1_41/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT Size 65536
Total time 1 min, 44 sec
3.6
3.4
3.2
3.0
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
ppm
3.00
1.61

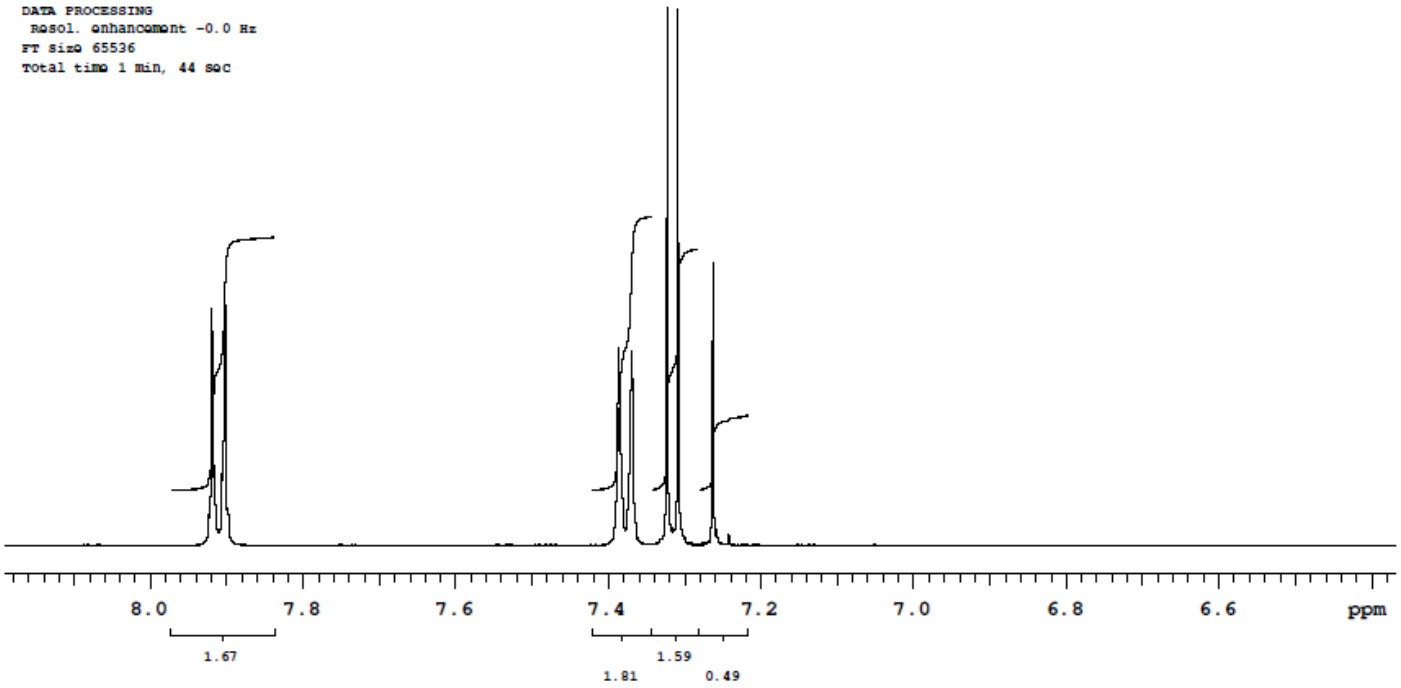

sample: MK_1_63
sample ID: s_20101203_10
File: /home/walkup/data/Mahmood/MK_1_63/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Br
O
S
O
O
F
Br
CH3
Compound: 3b
8
7
6
5
4
3
2
1
−0
ppm
1.67
1.59
1.8149
3.00
1.43
0.50

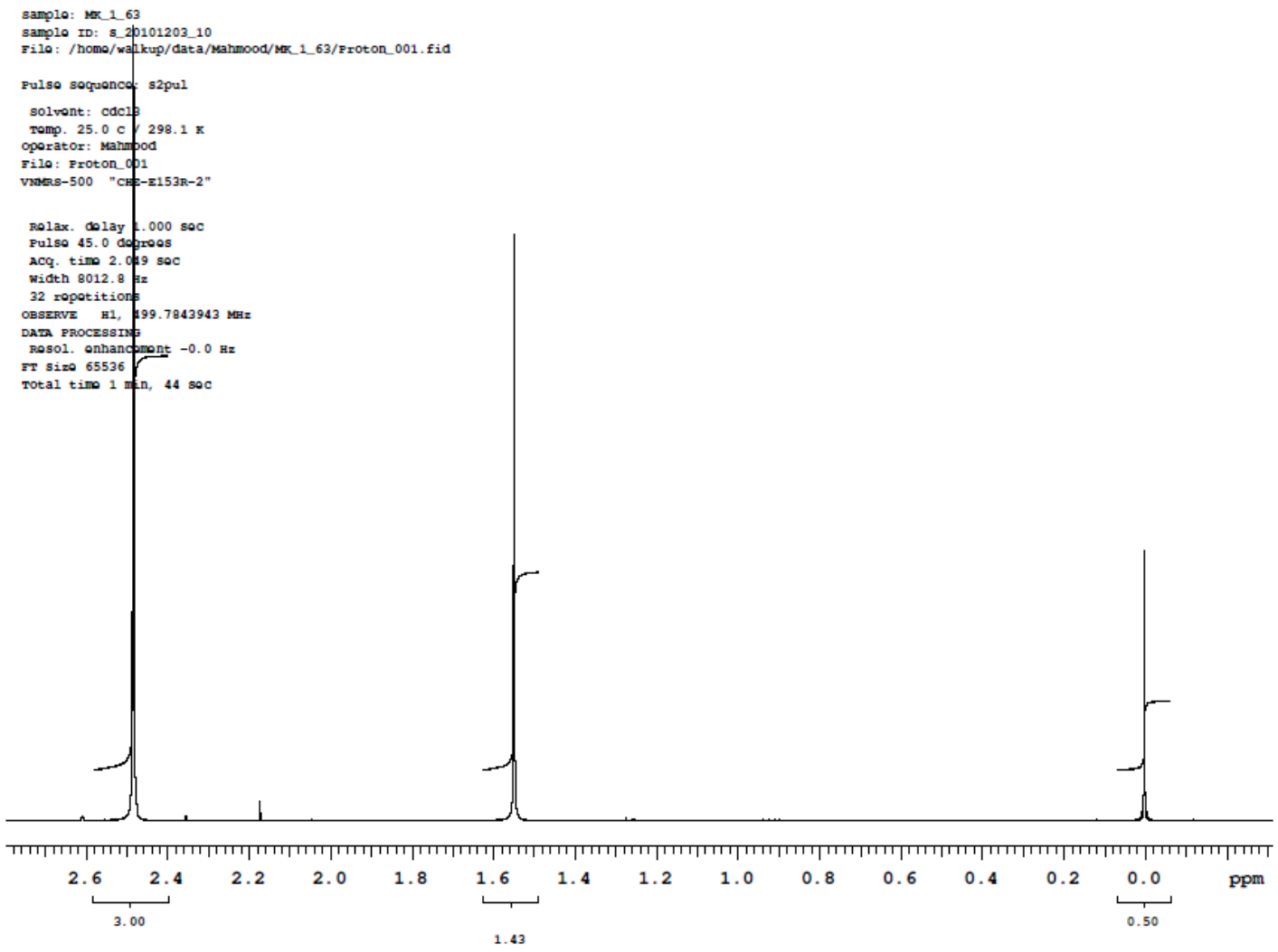
sample: MK_1_63
sample ID: s_20101203_10
File: /home/walkup/data/Mahmood/MK_1_63/Proton_001.fid

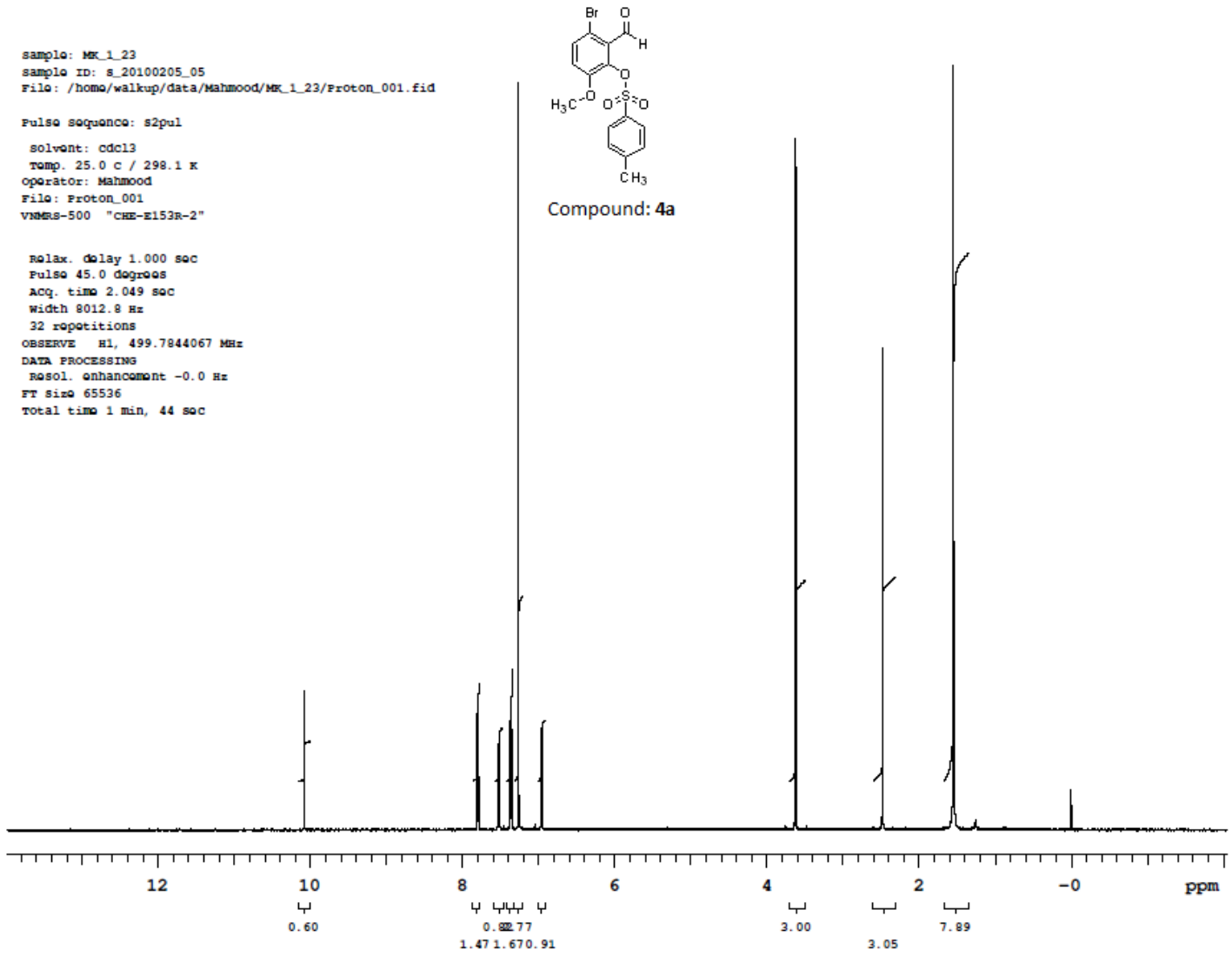
Pulse sequence: s2pul

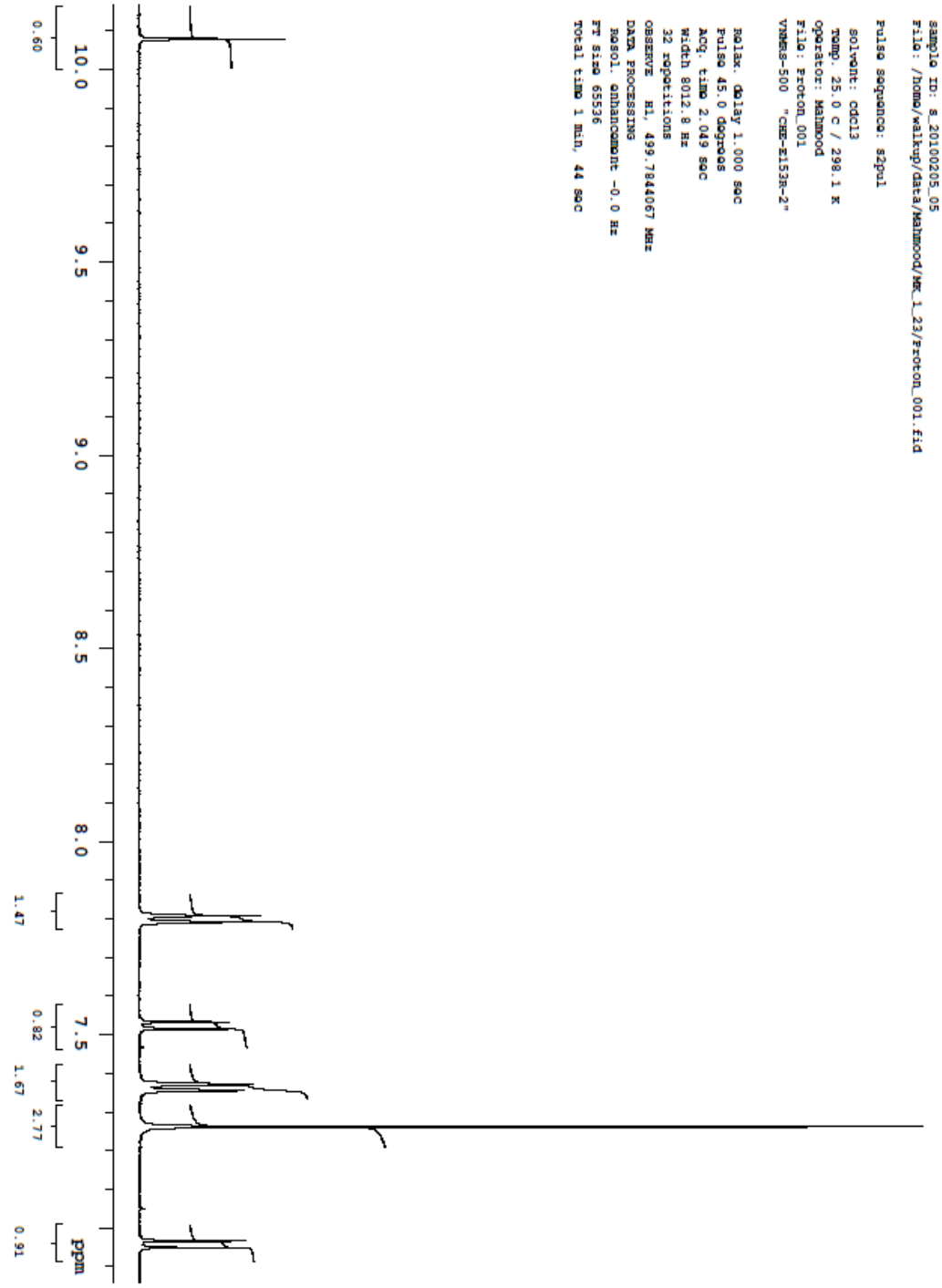
 Solvent: cdcl3
 Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"

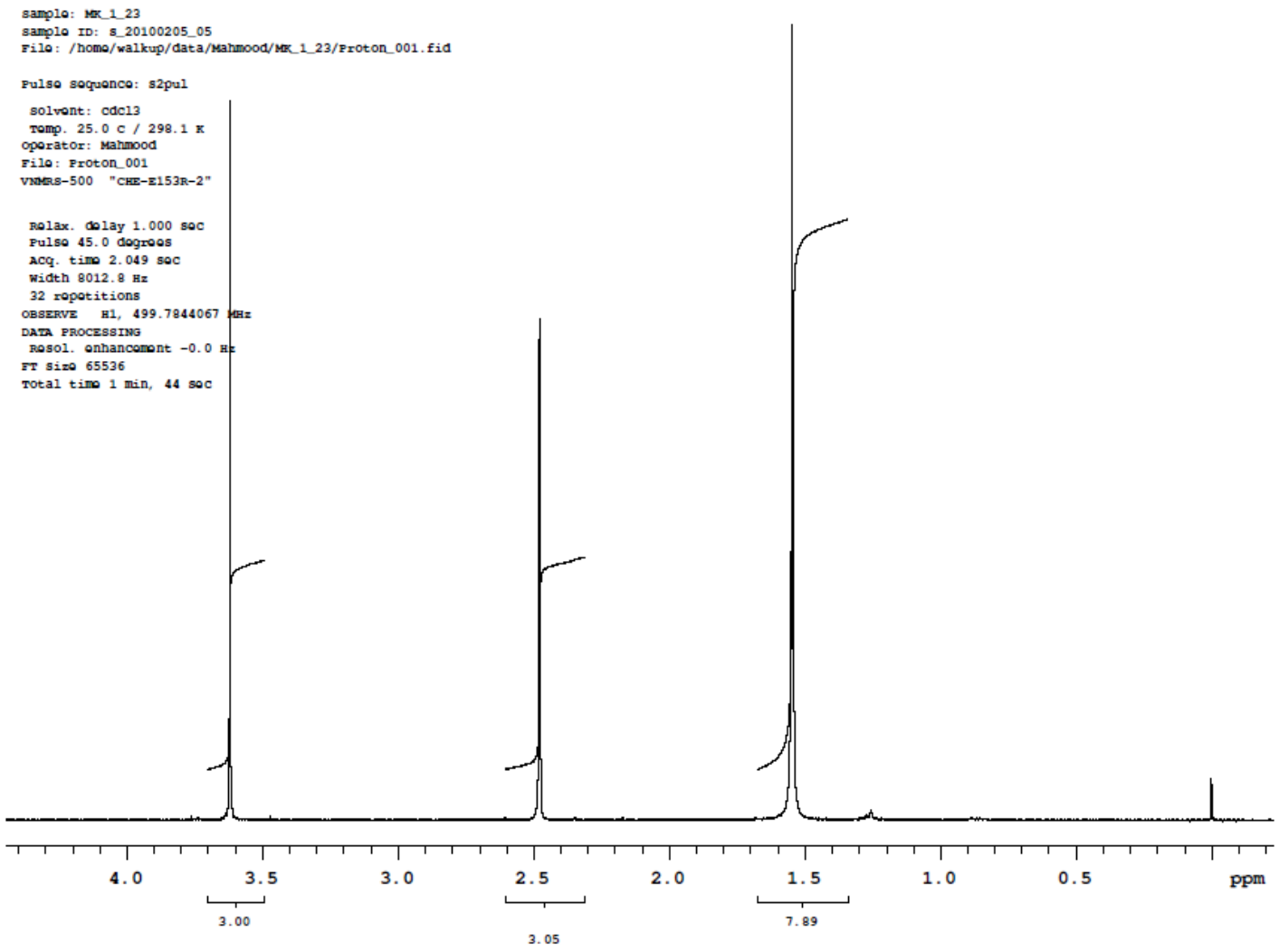
 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 Acq. time 2.049 sec
 Width 8012.8 Hz
 32 repetitions
OBSERVE   H1, 499.7843943 MHz
DATA PROCESSING
 Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec

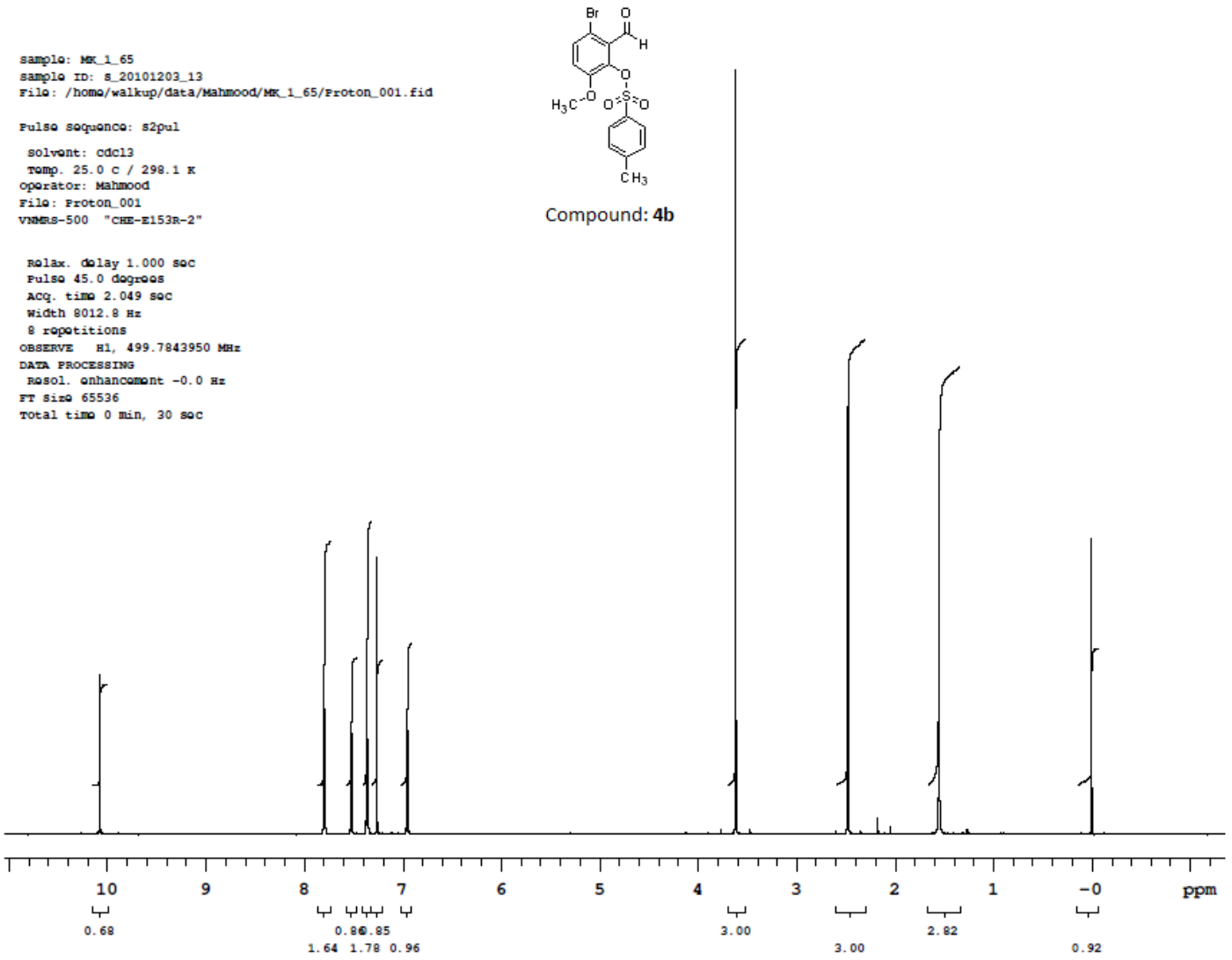



sample: MK_1_63
sample ID: s_20101203_10
File: /home/walkup/data/Mahmood/MK_1_63/Proton_001.fid
Pulse sequence: s2pul
Solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843943 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
ppm
3.00
1.43
0.50


sample: MK_1_23
sample ID: s_20100205_05
File: /home/walkup/data/Mahmood/MK_1_23/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 c / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7844067 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Br
O
H
O
H3C
O
S
O
O
CH3
Compound: 4a
12
10
8
6
4
2
-0
ppm
0.60
1.47
1.67
0.91
3.00
3.05
7.89


sample: MK_1_23
sample ID: s_20100205_05
file: /home/walkup/data/Mahmood/MK_1_23/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153H-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7844067 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
10.0
9.5
9.0
8.5
8.0
7.5
ppm
0.60
1.47
0.82
1.67
2.77
0.91


Sample: MK_1_23
Sample ID: s_20100205_05
File: /home/walkup/data/Mahmood/MK_1_23/Proton_001.fid
Pulse Sequence: s2pul
Solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7844067 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT Size 65536
Total time 1 min, 44 sec
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
ppm
3.00
3.05
7.89

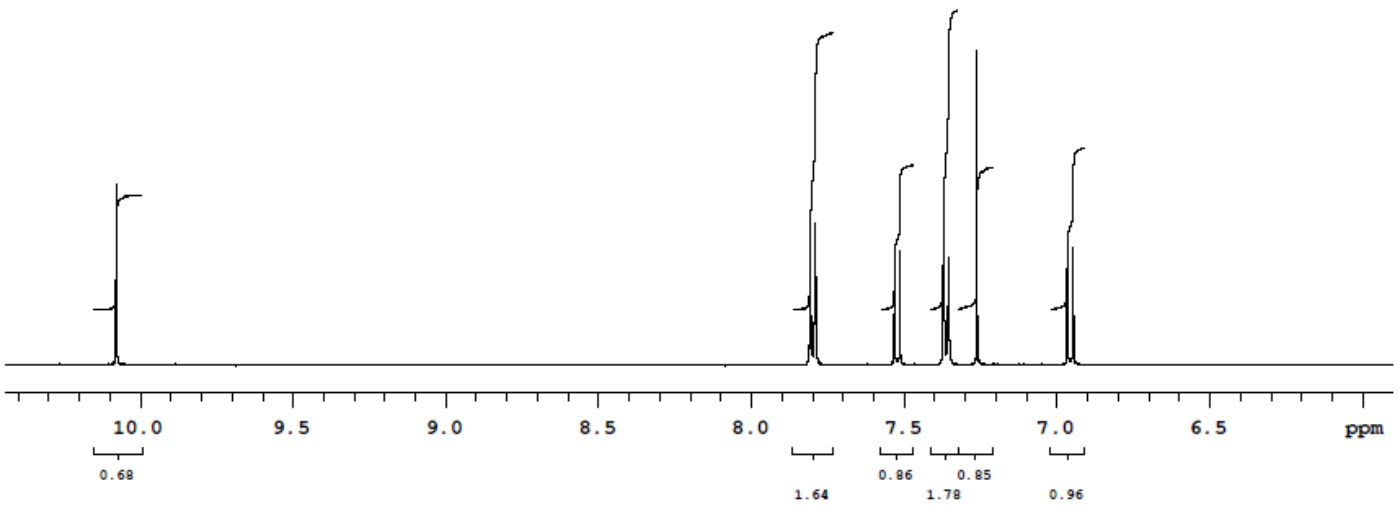

sample: MK_1_65
sample ID: s_20101203_13
File: /home/walkup/data/Mahmood/MK_1_65/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
8 repetitions
OBSERVE H1, 499.7843950 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 0 min, 30 sec
Br
O
H
O
H3C
O
S
O
O
CH3
Compound: 4b
10
9
8
7
6
5
4
3
2
1
−0
ppm
0.68
1.64
0.86
1.78
0.85
0.96
3.00
3.00
2.82
0.92

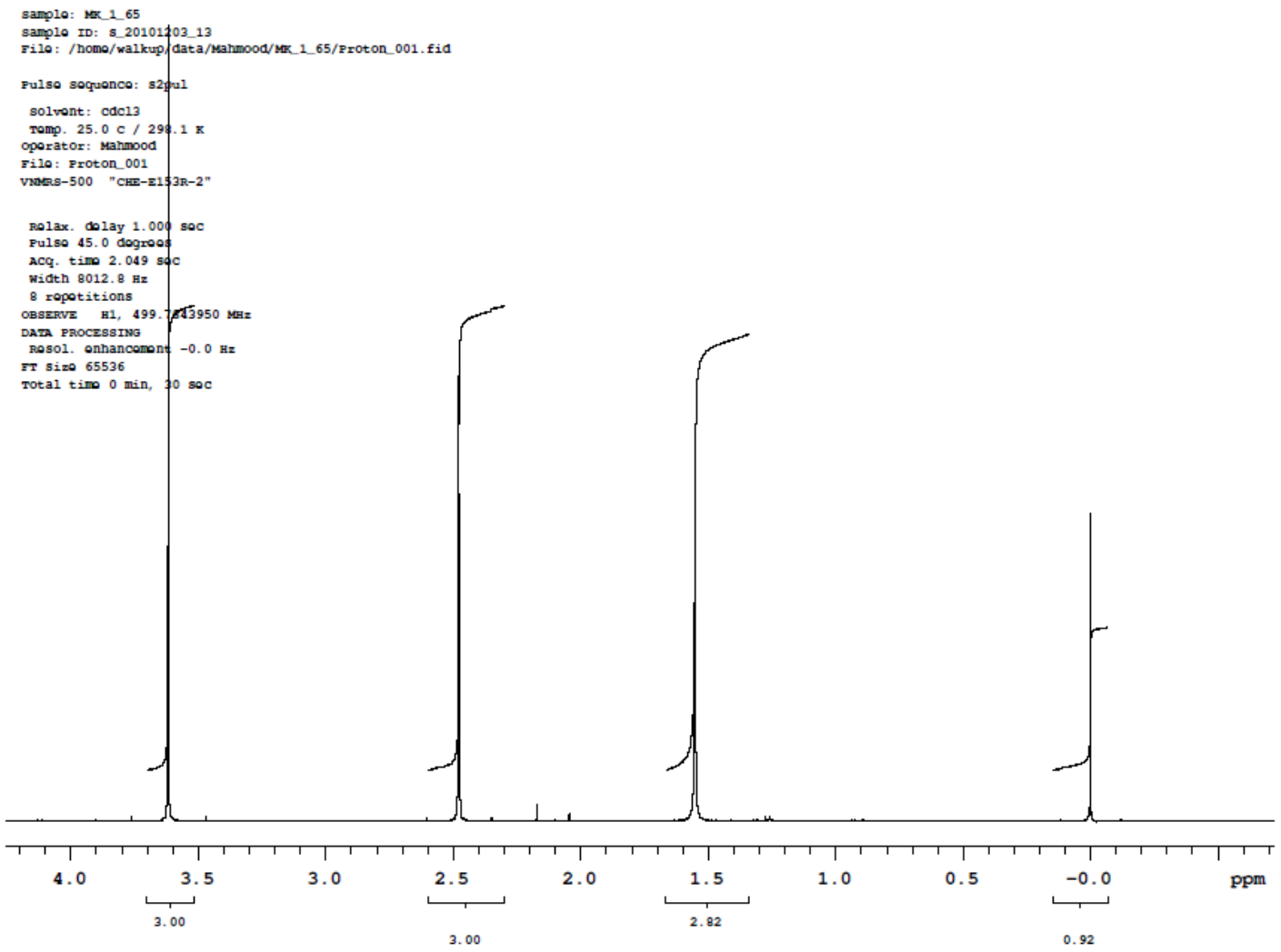
sample: MK_1_65
sample ID: s_20101203_13
File: /home/walkup/data/Mahmood/MK_1_65/Proton_001.fid

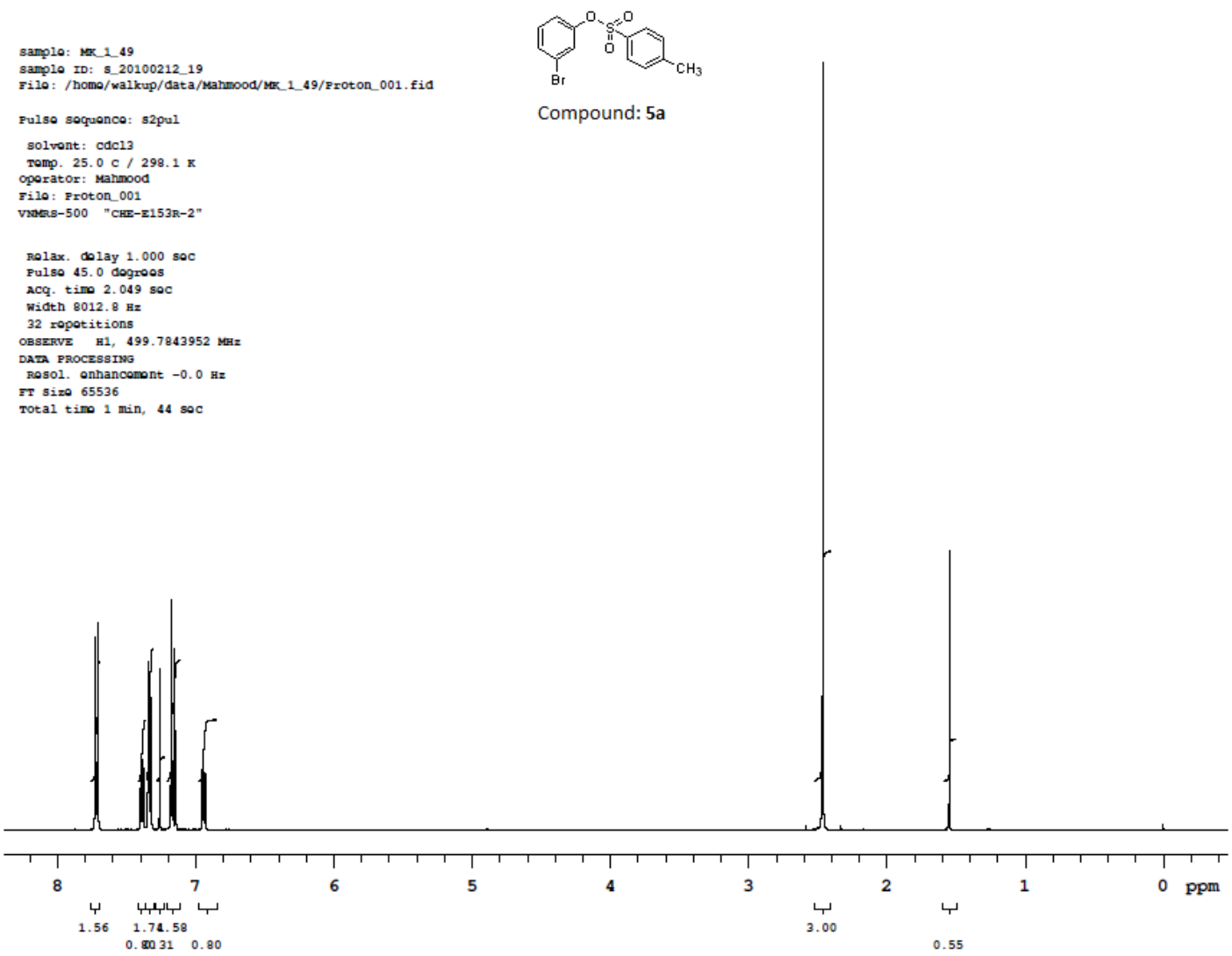
Pulse sequence: s2pul

 solvent: cdcl3
 Temp. 25.0 c / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"

 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 Acq. time 2.049 sec
 Width 8012.8 Hz
 8 repetitions
OBSERVE   H1, 499.7843950 MHz
DATA PROCESSING
 Resol. enhancement -0.0 Hz
FT size 65536
Total time 0 min, 30 sec




sample: MK_1_65
sample ID: s_20101203_13
File: /home/walkup/data/Mahmood/MK_1_65/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
8 repetitions
OBSERVE H1, 499.7743950 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 0 min, 30 sec
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
-0.0
ppm
3.00
3.00
2.82
0.92

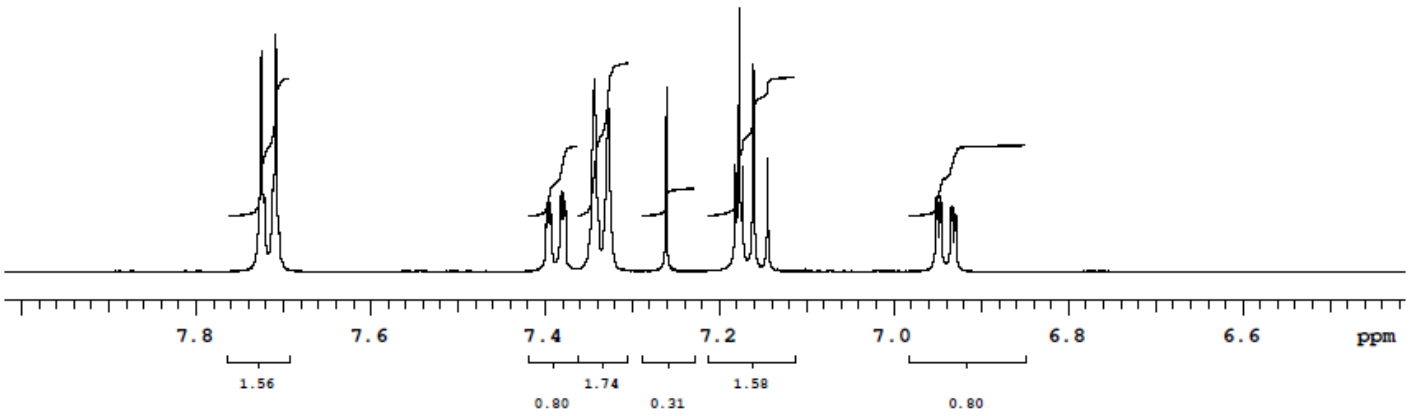

sample: MK_1_49
sample ID: s_20100212_19
File: /home/walkup/data/Mahmood/MK_1_49/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Br
CH3
Compound: 5a
8
7
6
5
4
3
2
1
0
ppm
1.56
1.74
4.58
0.80
3.00
0.55

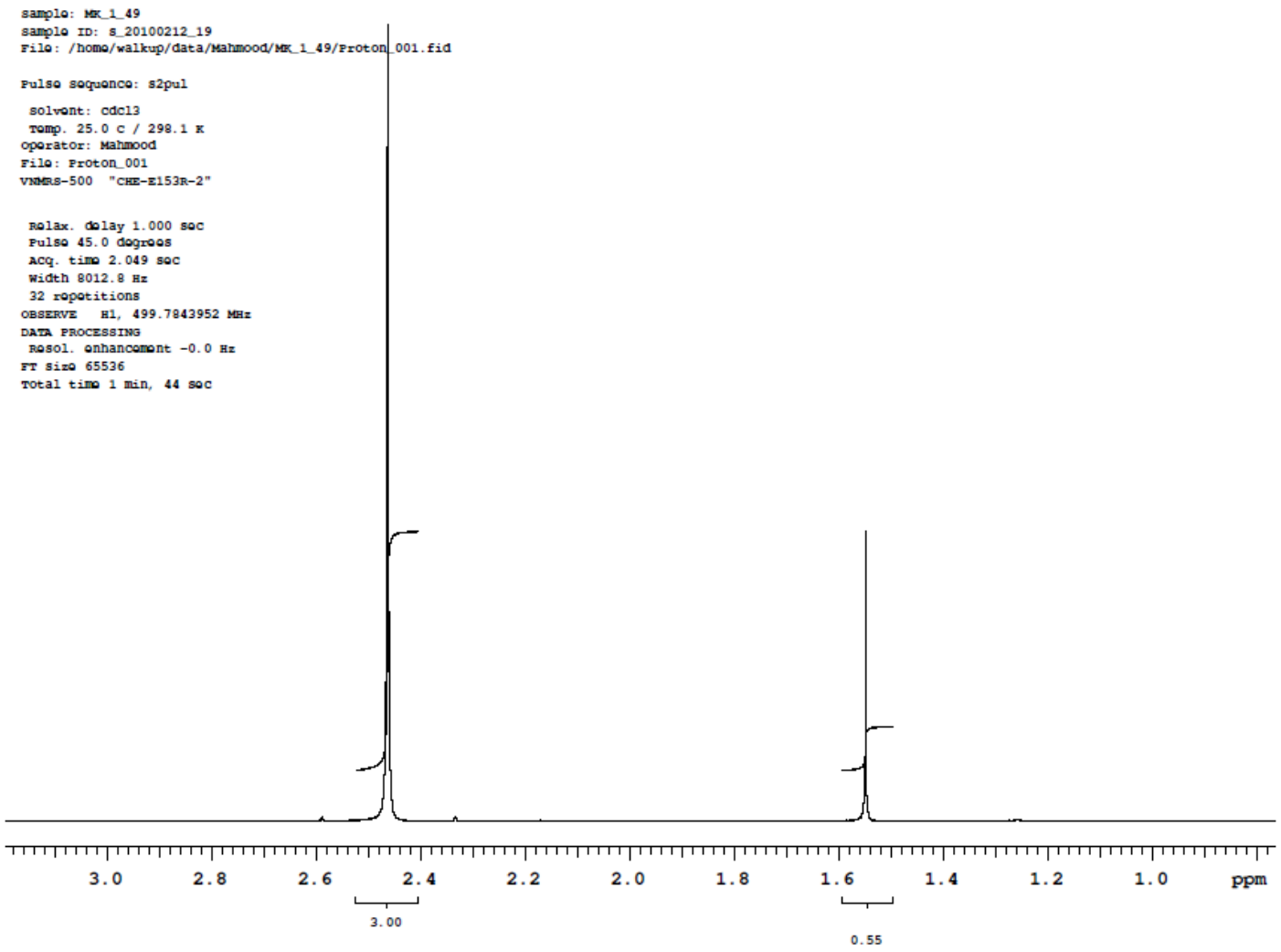
sample: MK_1_49
sample ID: s_20100212_19
File: /home/walkup/data/Mahmood/MK_1_49/Proton_001.fid

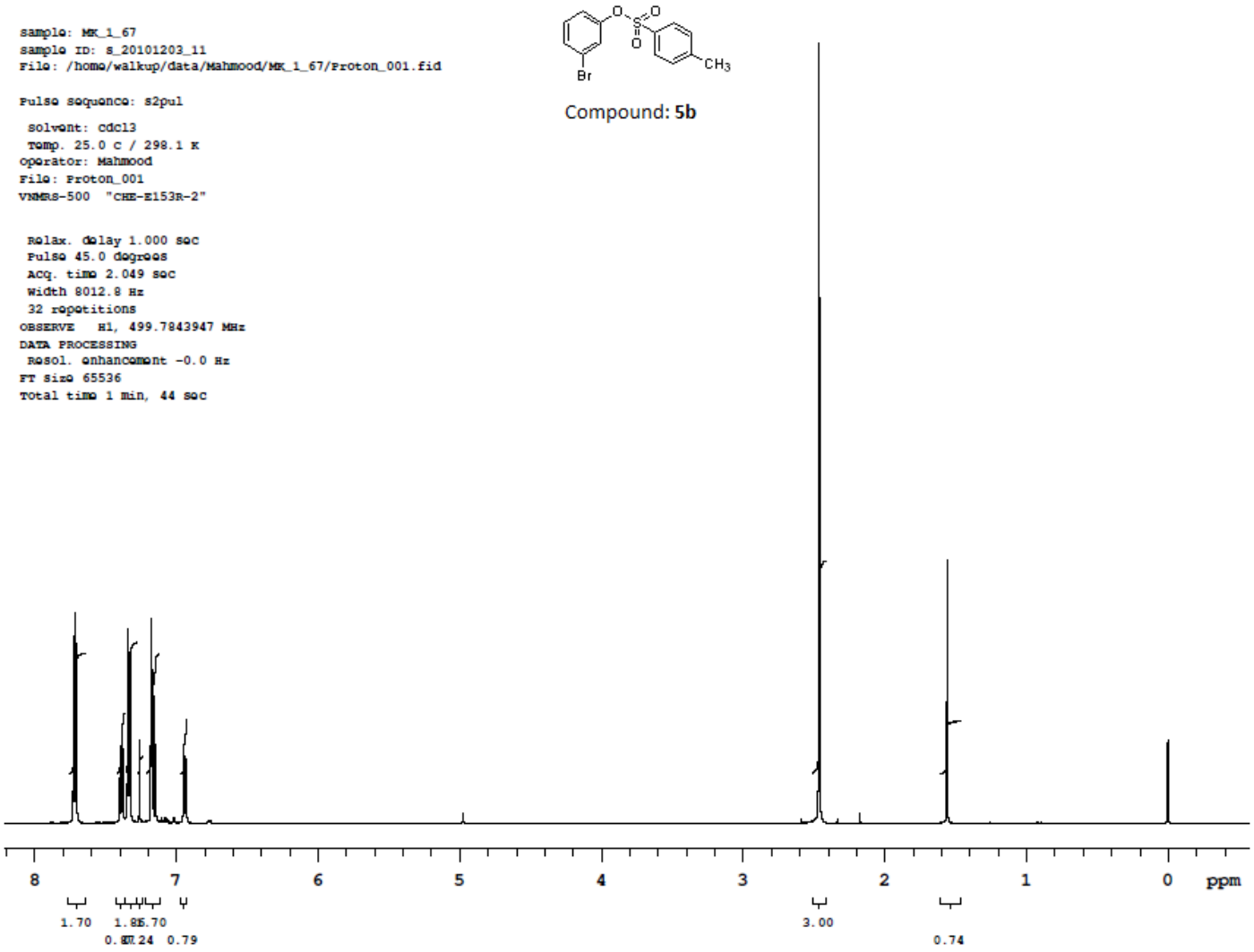
Pulse sequence: s2pul

solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT Size 65536
Total time 1 min, 44 sec




sample: MK_1_49
sample ID: s_20100212_19
File: /home/walkup/data/Mahmood/MK_1_49/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843952 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
3.0
2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
ppm
3.00
0.55

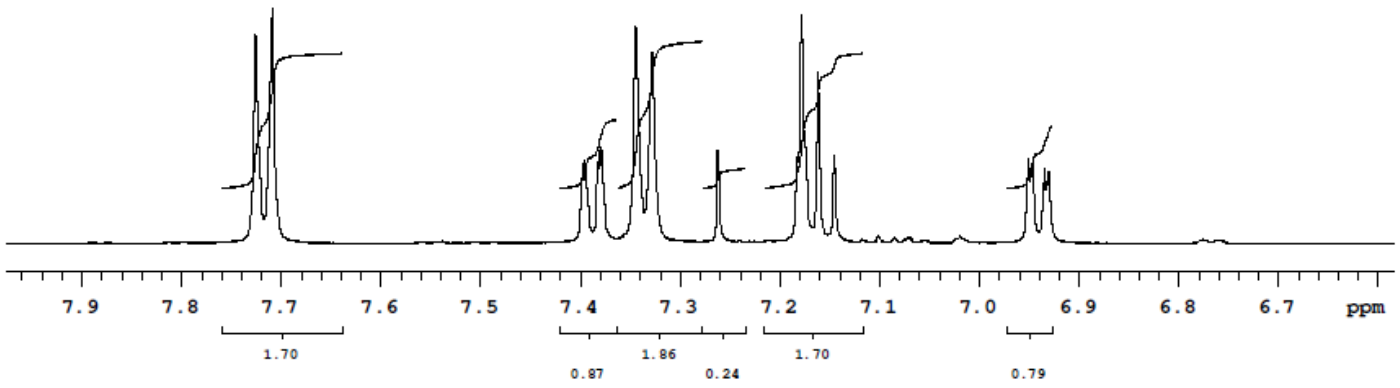

sample: MK_1_67
sample ID: s_20101203_11
File: /home/walkup/data/Mahmood/MK_1_67/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
ACQ. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843947 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
Br
CH3
Compound: 5b
8
7
6
5
4
3
2
1
0
ppm
1.70
1.85
5.70
0.79
3.00
0.74

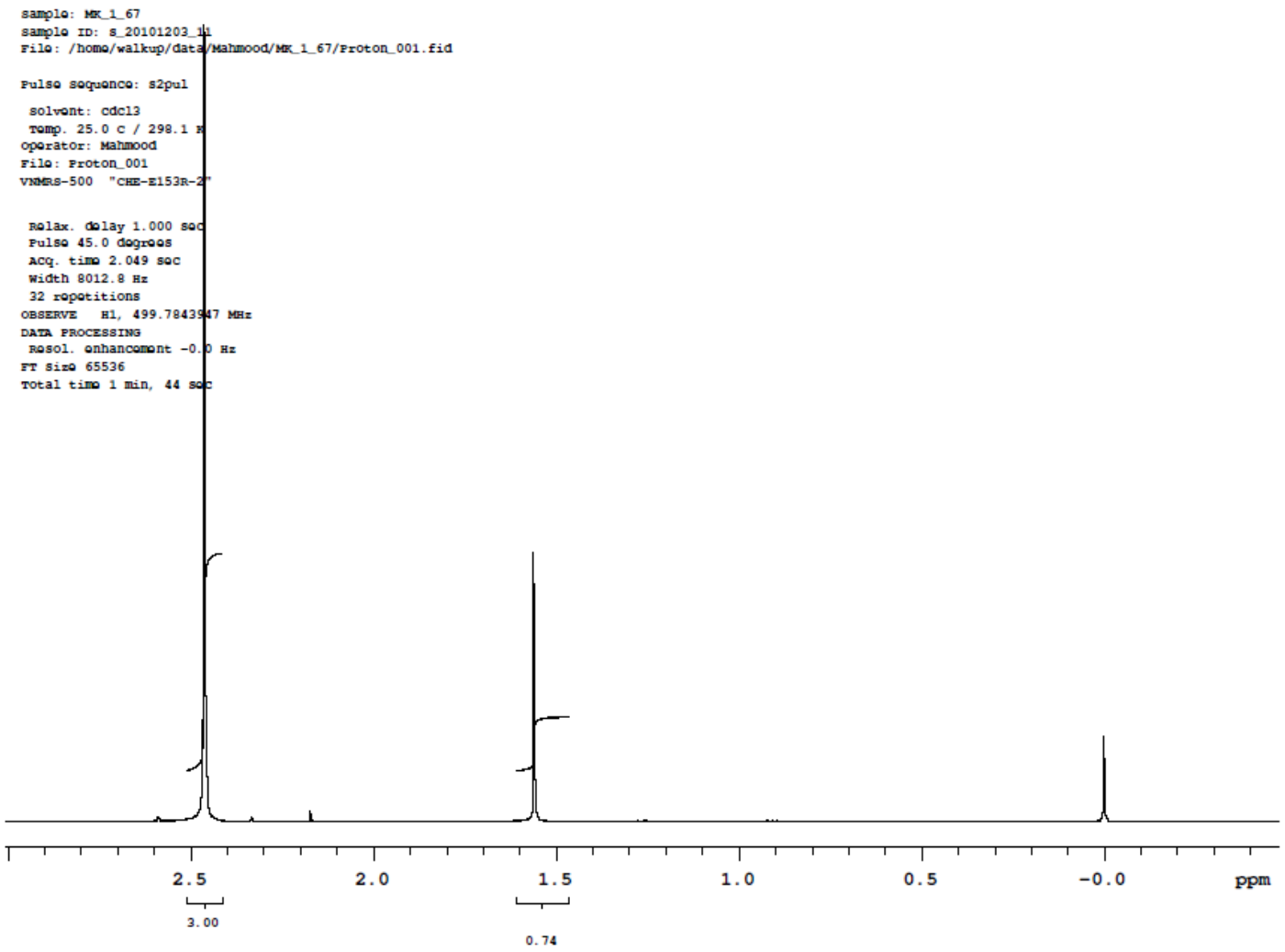
```
sample: MK_1_67
sample ID: s_20101203_11
File: /home/walkup/data/Mahmood/MK_1_67/Proton_001.fid

Pulse sequence: s2pul

 solvent: cdcl3
 Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500  "CHE-E153R-2"


 Relax. delay 1.000 sec
 Pulse 45.0 degrees
 ACQ. time 2.049 sec
 Width 8012.8 Hz
 32 repetitions
OBSERVE   H1, 499.7843947 MHz
DATA PROCESSING
 Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
```




sample: MK_1_67
sample ID: s_20101203_11
File: /home/walkup/data/Mahmood/MK_1_67/Proton_001.fid
Pulse sequence: s2pul
solvent: cdcl3
Temp. 25.0 C / 298.1 K
Operator: Mahmood
File: Proton_001
VNMRS-500 "CHE-E153R-2"
Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.049 sec
Width 8012.8 Hz
32 repetitions
OBSERVE H1, 499.7843947 MHz
DATA PROCESSING
Resol. enhancement -0.0 Hz
FT size 65536
Total time 1 min, 44 sec
2.5
2.0
1.5
1.0
0.5
-0.0
ppm
3.00
0.74

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