SYNTHESIS OF β-LACTAM BELACTOSIN A ANALOGS AS POTENTIAL 20S PROTEASOME INHIBITORS

by

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ABSTRACT

Belactosin A is a naturally occurring proteasome inhibitor with potent anti-tumor activity, however it does not possess the properties necessary to be used clinically. Like other proteasome inhibitors, such as carfilzomib and bortezomib, which are currently used to treat myelomas, belactosin A exhibits a peptidomimetic backbone with a serine trap at the C-terminal end. Key structural features of the natural product include a cyclopropane ring and a terminal β-lactone as the serine trap.

While several syntheses of belactosin A have been reported, only a few analogs have been prepared, all of which maintained the β-lactone as the serine trap. Though β-lactams are classic serine traps, only two have been reported as proteasome inhibitors. The synthesis of novel β-lactam analogs of belactosin A is reported here, with the key step being coupling of cyclopropyl peptidomimetics to β-lactams. The use of phenylalanine, leucine, and valine as starting materials leads to benzyl, isobutyl, and isopropyl analogs of the natural product. Attempts were made to optimize functionalization of the cyclopropyl backbones for coupling to the β-lactams, which lead to the proposal of a modified synthetic route to achieve the proposed analogs.

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CHAPTER I: INTRODUCTION

20S Proteasome as an antitumor target

Proteasome structure and function

The 26S proteasome is a multi-subunit complex that plays a central role in the ubiquitin-proteasome pathway for regulated protein degradation, and its activity affects a multitude of cellular processes. The 26S proteasome is comprised of a 20S proteolytic core and two 19S regulatory "caps" (Figure 1a). In the ubiquitin-proteasome pathway, proteins are marked for degradation by ubiquitination. The 19S regulatory cap binds to the ubiquitin chain and allows the protein to enter the barrel-like proteolytic core where it is proteolyzed and released (Figure 1b). Because its inhibition induces apoptosis (cell death) in proliferating cells and prevents angiogenesis (development of new blood vessels), the 20S proteasome is an attractive anti-tumor target. Proteasome inhibition also blocks NF-κB activation necessary to produce interleukin growth factors required by cancerous cells [1]. Many anti-tumor chemotherapies currently in clinical use, such as doxorubicin and radiotherapy, stop tumor growth by damaging DNA. Proteasome inhibition has been found to also work synergistically with these therapies by preventing the activation of DNA repair enzymes [6].

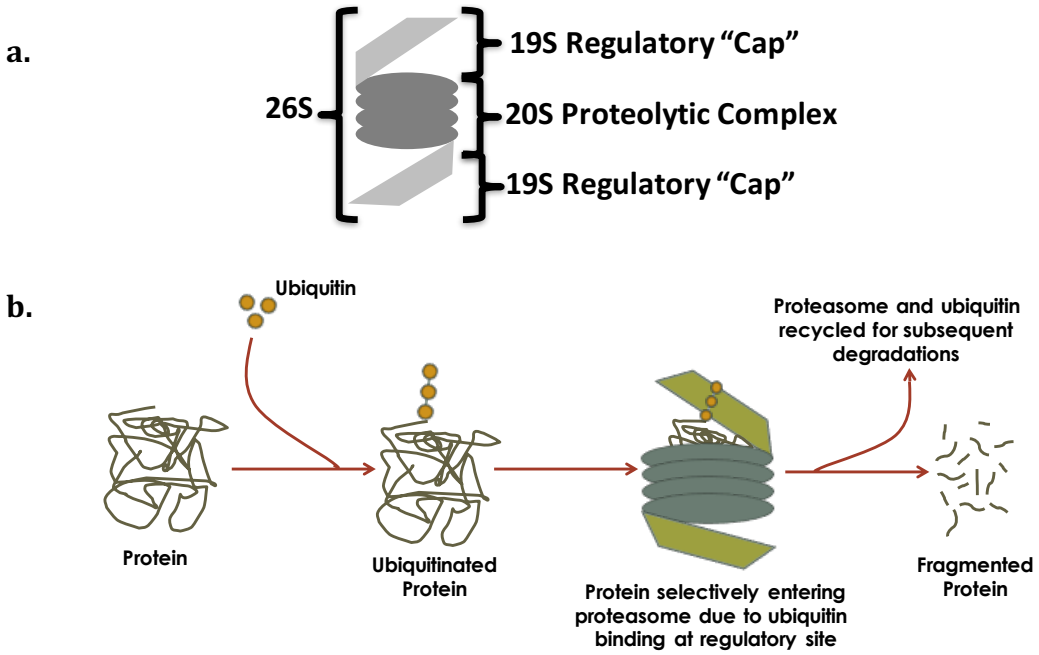


Figure 1: Proteasome structure and function: (a) Simplified graphic representing the regulatory and catalytic components of the 26S proteasome (b) Ubiquitin-proteasome pathway for regulated protein degradation.

The structure of eukaryotic 20S proteasomes can be further broken down into two β-rings that each consist of three β-subunits for a total of two chymotrypsin-like (CT-L), two trypsin-like (T-L), and two caspase-like (C-L) active sites that specifically cleave after hydrophobic/aromatic, basic, and acidic residues respectively [1]. The majority of proteasome inhibitors target a combination of these active sites, but the CT-L subunits are the most frequently targeted. While CT-L activity has historically been considered the "rate-limiting step" of protein degradation, recent studies have shown that all six subunits contribute equally and that relative activities are dependent on the specific substrates and the cell-type.

Some cancer cell lines have been shown to be more dependent on C-L and T-L activities and are less susceptible to drugs that primarily target the CT-L subunits [2].

Proteasome mechanism of action

In general, the active site of a protease contains a nucleophilic catalytic residue that is activated through some form of catalytic cascade. The most common mode of activation is through what is termed the catalytic triad, which consists of an acidic residue (Asp or Glu), a basic histidine (His) residue, and the nucleophilic residue. Proteases are classified primarily by the identity of this nucleophilic residue and by the mechanisms through which this residue acts upon the peptide bond. The most common proteases are metallo (34%), serine (30%), and cysteine (26%) proteases, but the aspartic (4%) proteases have also been well characterized [3]. However, the β-subunits of the proteasome are unique in that they utilize an N-terminal threonine (T1) activated by a lysine residue (K33) as their catalytic mechanism rather than an acid-activated histidine. Eva Huber and her colleagues suggest that the N-terminal amine plays a significant role in stabilizing the incoming protein substrate as well as donating a proton to the C-terminal cleavage product and subsequently activating a water molecule for cleavage of the N-terminal product [4]. This mechanism is depicted in Figure 2.

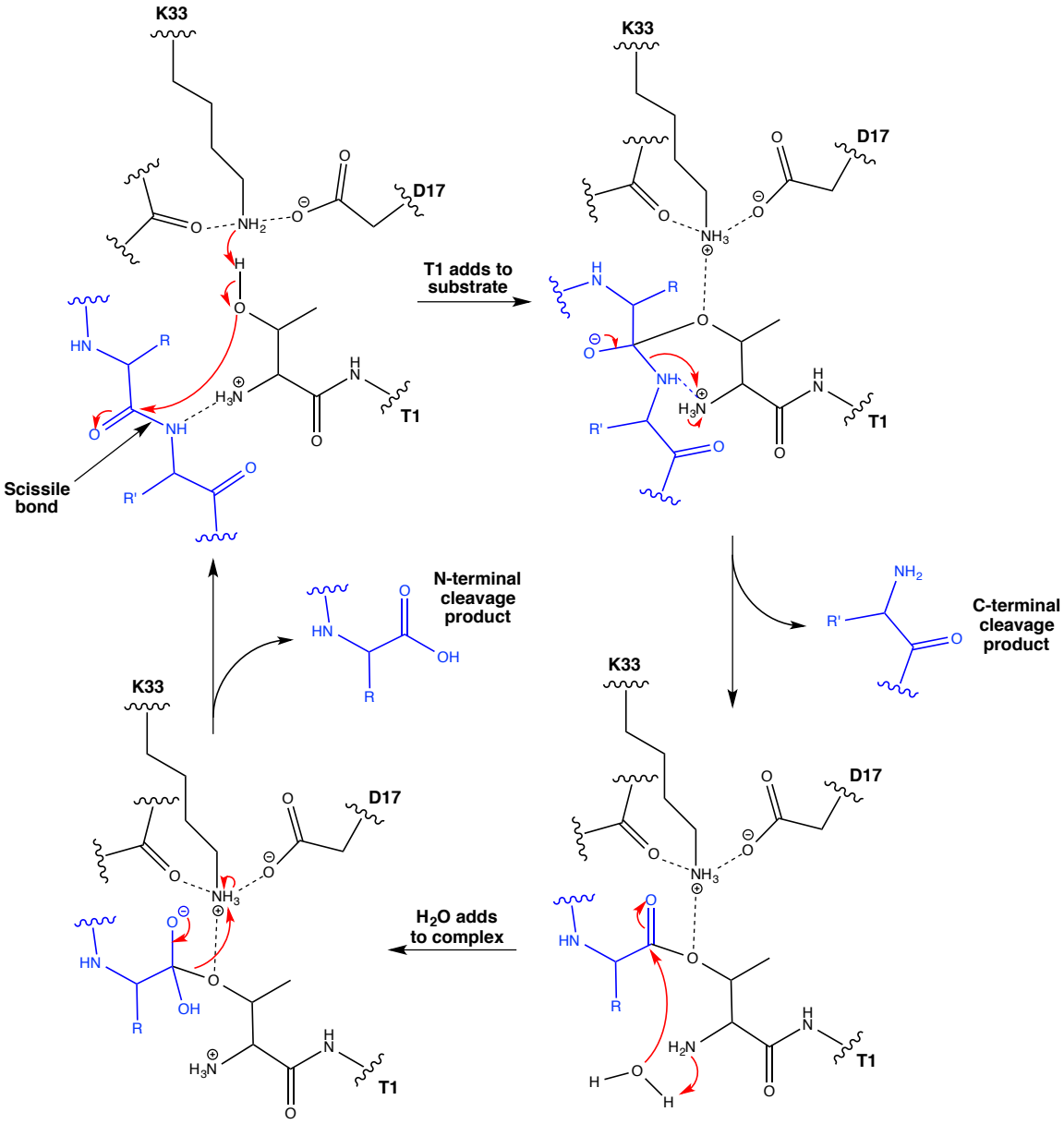


Figure 2: Proteasome catalytic mechanism for protein cleavage. Substrate highlighted in blue.

Serine traps as structural elements for proteasome inhibition

In order to occupy the active site and trap the nucleophilic threonine residue, most proteasome inhibitors take advantage of traditionally electrophilic structural moieties that have been termed "serine traps" as warheads. Most drugs containing serine traps bind covalently and irreversibly to their target, but there are some drugs, particularly borates, that have been found to be reversible inhibitors. Figure 3 displays the most common serine traps that will be discussed.

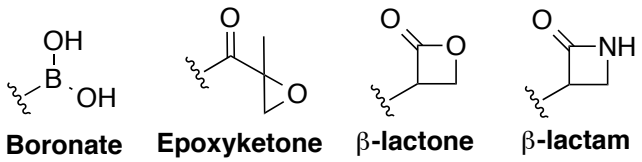


Figure 3: Common serine traps that target nucleophilic catalysts.

Boronate inhibitors

Boronates are slow binding, but highly selective proteasome inhibitors. Though boronates are considered to be reversible inhibitors, they maintain a considerably slow dissociation rate that renders them essentially irreversible within the timescale of a typical cell culture experiment. The electrophilic boron atom captures the nucleophile in a covalent bond that is only semi-reversible (Figure 4).

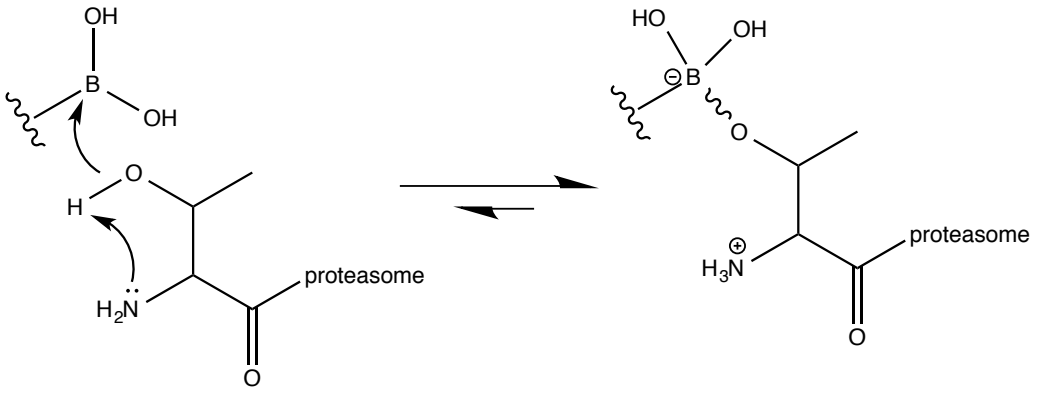


Figure 4: General mechanism of boronates as reversible 20S-proteasome inhibitors [1].

Boronates have been found to be highly selective for proteasome inhibition over other proteases. Boron and sulfur have a very weak interaction, which makes boronates poor inhibitors of cysteine proteases. Boronates have also been found to be 1000-fold weaker serine protease inhibitors than they are proteasome inhibitors. This is due to the increase in the rate of hydrolysis of the boronate-serine adduct [1]. In the early stages of its development, researchers searched extensively to identify any other targets of the boronate proteasome inhibitor and cancer drug bortezomib (Figure 5), but they failed [5].

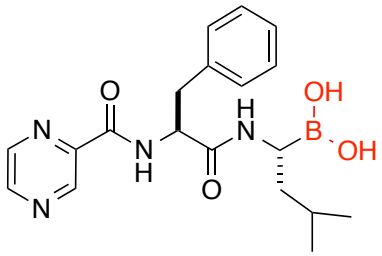


Figure 5: Structure of bortezomib. Boronate serine trap is highlighted in red.

Bortezomib was the first proteasome inhibitor to be used clinically to treat cancer. It was approved by the FDA in 2003 for the treatment of multiple myeloma and gained later approval in 2006 for the treatment of mantle cell lymphoma. The drug is currently involved in over 200 active clinical trials for various combination therapies with other chemotherapeutic agents. The limitations of toxicity in dosing, however, have rendered bortezomib unsatisfactory for the treatment of solid tumors [6]. The structurally related drug, CEP-18770 (Figure 6), has been found to exhibit similar activity to bortezomib, but it has a much more favorable cytotoxicity profile towards healthy cells [7].

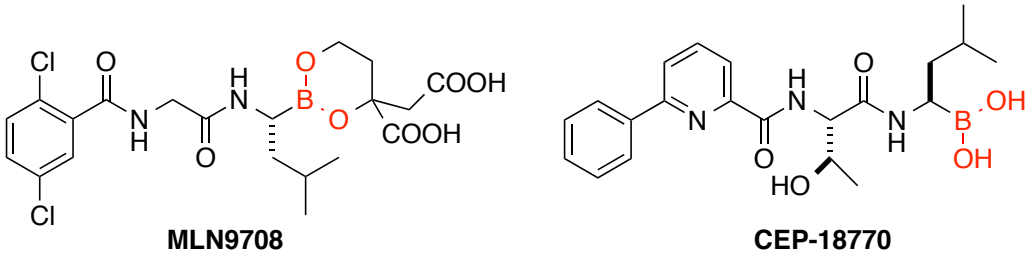


Figure 6: Structure of cancer drug candidates, MLN9708 and CEP-18770, currently in Phase I clinical trials. The boronate of MLN9708 is hydrolyzed in vivo to its active form. Boronate serine trap is highlighted in red for both compounds.

CEP-18770 is currently in Phase I trials for the treatment of solid tumors and non-Hodgkin's lymphoma [7]. Another boronate currently in clinical trials is MLN9708 (Figure 6). The boronate of MLN9708 is masked in the ring, but hydrolyzes to the

active form immediately upon entering the plasma. MLN9708 is currently in Phase I clinical trials for the treatment of lymphoma and non-hematological malignancies and in Phase II trials for treatment of multiple myeloma. All three of the boronate drugs mentioned selectively inhibit the CT-L subunit of the 20S proteasome [6].

Epoxyketone inhibitors

Epoxyketones react through a unique mechanism that incorporates both the hydroxyl and free amine of the N-terminal threonine to covalently bind the drug to the active site through formation of a six-membered morpholino ring (Figure 7)[8].

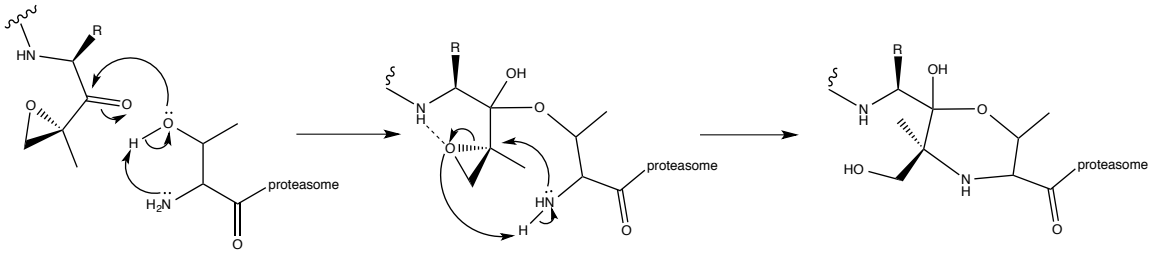


Figure 7: General mechanism of peptide epoxyketones as irreversible 20S proteasome inhibitors.

The dual nature of the epoxyketone mechanism to target both the hydroxyl and terminal amine provides epoxyketone drugs with a high selectivity towards the unique N-terminal catalytic residue of the proteasome and renders the drugs as entirely irreversible inhibitors [1].

Epoxomicin (Figure 8) is a natural product isolated from an unidentified actinomycete bacterial strain based on its anti-tumor activity in mice [9]. It was the

first epoxyketone identified as a proteasome inhibitor, but it doesn't have the necessary pharmaceutical properties to be a clinical drug.

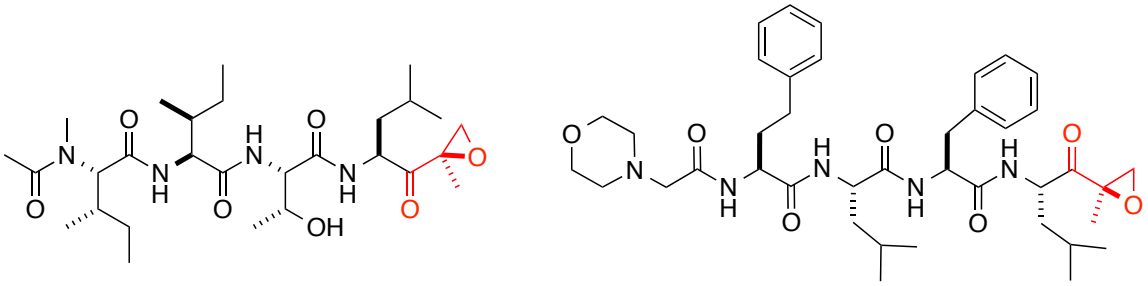


Figure 8: Structure of epoxomicin and carfilzomib. Epoxyketone serine trap is highlighted in red.

Carfilzomib (Figure 8) was discovered as an analog of epoxomicin with improved properties. In Phase I clinical trials, carfilzomib exhibited equal potency, greater selectivity, and less peripheral neuropathy than bortezomib [6]. In 2012, carfilzomib was approved by the FDA for the treatment of multiple myeloma [10]. Both carfilzomib and bortezomib can only be administered intravenously, but proteasome inhibitor therapy requires twice weekly dosing that would make an orally available inhibitor advantageous. Oprozomib (Figure 9) was developed as an analog of carfilzomib to address this need [6].

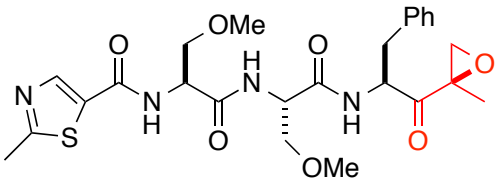


Figure 9: Structure of oprozomib. Epoxyketone serine trap is highlighted in red.

Oprozomib has demonstrated similar activity to carfilzomib as an orally bioavailable drug and is currently in Phase I clinical trials for treatment of hematologic malignancies and solid tumors [11]. The oral bioavailability of oprozomib compared to carfilzomib is attributed to the truncated size and incorporation of the methylserine residues. Di- and tripeptides are more likely to cross intestinal epithelial barriers and be absorbed into the blood stream compared to tetrapeptides like carfilzomib. An SAR study led by Han-Jie Zhou of Proteolix identified that the tripeptide form retained the greatest activity against the proteasome. He also identified the methylserine residues provided better metabolic stability and solubility for oral bioavailability than the bulky residues of carfilzomib and bortezomib. Once the side chains were optimized, the N-cap was optimized for potency revealing that 2-Me-5-thiazole provided the best results [12].

β-Lactone and β-lactam inhibitors

β-Lactones are the least stable of the proteasome inhibitors. Lactone esters are readily hydrolyzed by water at neutral pH and exist in equilibrium with the lactathione form that results from reacting with free glutathione in mammalian

cells. Though most β-lactone proteasome inhibitors are considered irreversible, the adduct formed with the proteasome is slowly ($t_{1/2} \approx 20$ h) hydrolyzed by water allowing for recovery of proteolytic activity. The general mechanism of an "irreversible" β-lactone inhibitor is depicted in Figure 10.

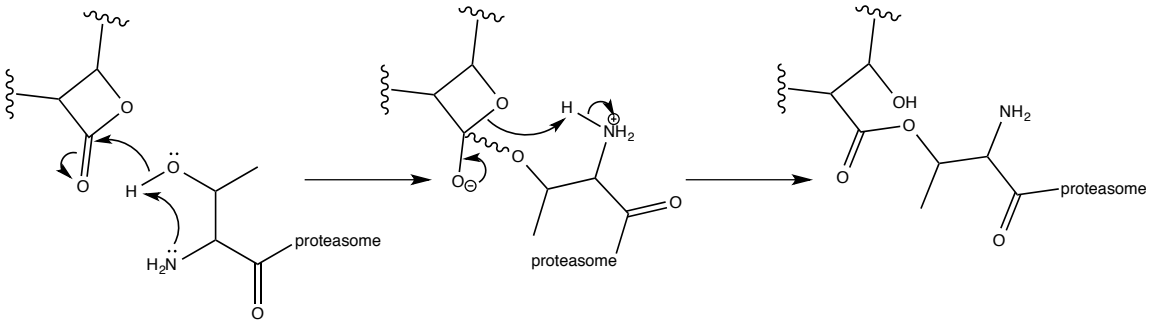


Figure 10: General mechanism of β-lactones as irreversible 20S proteasome inhibitors.

Salinosporamide A (also known as marizomib) is the most notable β-lactone proteasome inhibitor that is in clinical development (Figure 11).

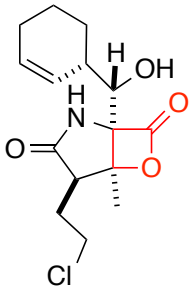


Figure 11: Structure of salinosporamide A. β-lactone serine trap is highlighted in red.

Salinosporamide A was isolated from the marine bacterium *Salinospora tropica* and has been found to bind irreversibly to all three types of β-subunits in the proteasome, whereas all of the other drugs discussed thus far primarily only target the CT-L subunit [6]. The irreversibility of salinosporamide A has been suggested to be due to the involvement of the chloroethyl substituent after opening of the β-lactone ring. E. J. Corey and colleagues have suggested that the chloroethyl undergoes an intramolecular substitution with the hydroxyl to form a tetrahydrofuran derivative that reduces the rate of dissociation (Figure 12) [13].

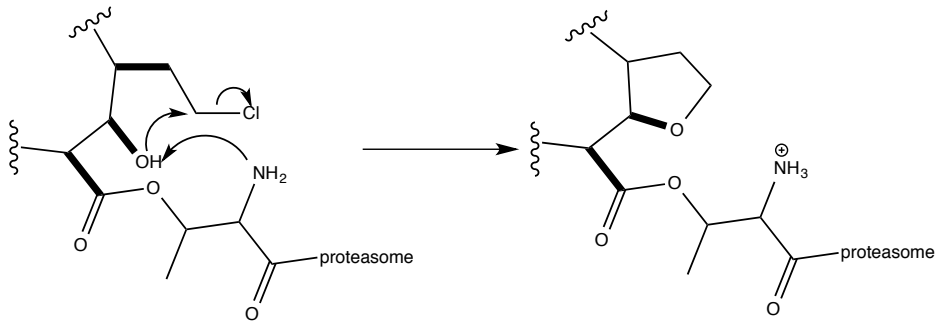
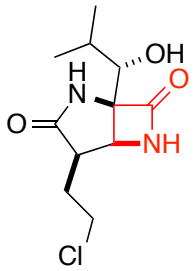


Figure 12: Extension of β-lactone mechanism against 20S proteasome with tetrahydrofuran ring closure of the chloroethyl of salinosporamide A.

Corey and colleagues' suggestion is based on the identification of the tetrahydrofuran derivative as a product when they treated salinosporin derivatives with benzylamine, which they used as a chemical model for the nucleophilic N-terminal threonine of the proteasome [13].

Corey and colleagues have also worked to develop a β-lactam analog of salinosporamide A (Figure 13).



1

Figure 13: Corey β-lactam analog of salinosporamide A. β-lactam highlighted in red.

Though **1** is suspected to undergo the same mechanism as salinosporamide A, the rate of proteasome inhibition of **1** is significantly slower. However, Corey states that the slow rate is "more than compensated for" by the increased aqueous stability of **1** under physiological conditions compared to salinosporamide A [14].

The only other β-lactam proteasome inhibitors that have been investigated are those that have been synthesized by Patricia Imbach at Novartis Institutes for BioMedical Research in Switzerland. Novartis had previously reported a non-covalent 20S proteasome inhibitor that selectively targeted the CT-L β-subunits. Imbach applied computational modeling of this scaffold to guide the optimum placement of a β-lactam moiety and synthesized several analogs fitting this model (Figure 14).

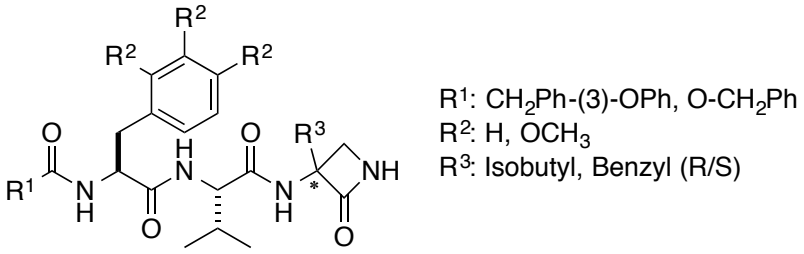


Figure 14: Scaffold of Novartis β-lactam proteasome inhibitors.

Imbach found that the R configuration at the β-lactam was necessary to properly orient the bulky substituent into the binding pocket and that the methoxy substituents at R^2 provided the most potent activity. Changes at R^1 and R^3 had little effect on the activity. This observation is characteristic of covalent inhibitors, which do not depend as greatly on optimization of intermolecular interactions for potency. Covalent inhibition was confirmed by mass spectrometry implying that these β-lactam compounds are irreversible inhibitors. The antiproliferative activity of the β-lactam derivatives against human breast carcinoma cell line MDA-MB-435 was also competitive, reaching an IC_{50} as low as 32 nM [15].

Belactosin A and associated analogs as proteasome inhibitors

Belactosin A (Figure 15) was first isolated in 2000 as a novel *Streptomyces* metabolite following observation of both antitumor and antibiotic activities [16].

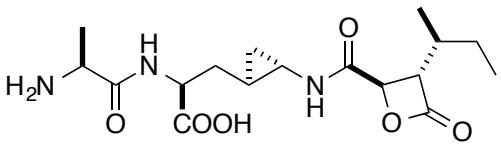


Figure 15: Structure of belactosin A.

In 2004, the antitumor activity was identified to be the result of irreversible 20S proteasome inhibition at the CT-L β-subunit [17]. There have been three reports of total syntheses of belactosin A [18, 19, 20], but Satoshi Shuto and colleagues are the only group to have developed analogs of the compound as well.

Shuto's first structure-activity relationship (SAR) study of belactosin A was to synthesize all of the possible stereoisomers to identify the most potent form, but the most important observation of this study came forth from their serendipitous decision to also test the benzyl carbamate (Cbz) protected vinyl synthetic intermediates for proteasome inhibition (Figure 16).

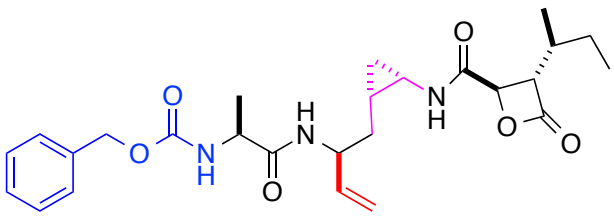
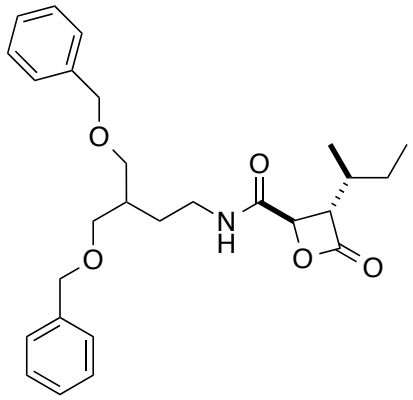


Figure 16: Structure of lead compound in Shuto's SAR study of belactosin A analogs exhibiting an unnatural *cis*-cyclopropyl (pink). The vinyl (red) and Cbz (blue) substituents were determined to increase potency 20-fold.

The vinyl and Cbz substituents were found to significantly enhance the potency of the compound. The most potent *cis*/L-*anti* analog (Figure 16) was found to be 20 times more potent than belactosin A [21]. This compound was then used as the lead for a subsequent SAR study that looked at different bulky hydrophobic substituents in place of the Cbz-alanine and the vinyl substituents [22]. Both of these studies showed that the addition of bulky hydrophobic aromatic substituents greatly enhanced potency and selectivity towards the CT-L subunit. Based on these analogs, Shuto developed a nonpeptidic derivative of belactosin (Figure 17) and coupled it to peptide boronate and epoxyketone fragments.



CT-L activity: IC_{50} = 29 nM
HCT116 cell growth: IC_{50} > 10 μM

Figure 17: Shuto's nonpeptidic belactosin derivative [23].

The configuration of the resulting compounds (Figure 18) was designed to maximize all possible binding pocket interactions based on observations of the protein crystal structure of both fragments bound to the active site [24, 25]. Note

the incredible increase in potency of the boronate derivative with an IC_{50} of 32 nM against cancer cell lines. This activity is competitive with the leading clinical drug bortezomib (IC_{50} = 10 nM) [22].

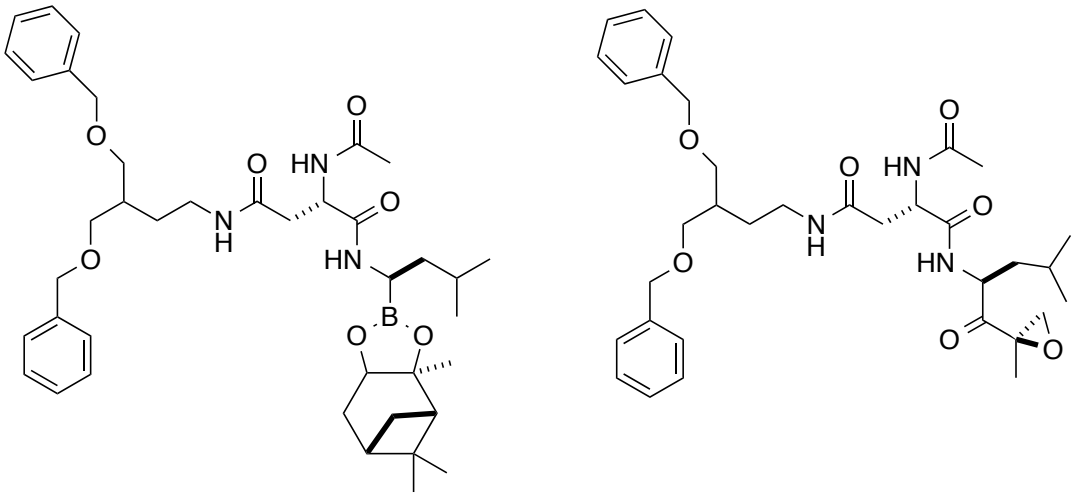


Figure 18: Shuto's nonpeptidic belactosin derivatives with alternative peptide boronate and epoxyketone warheads. The addition of the peptide warhead maximizes binding pocket interactions with the CT-L active site [24, 25].

Goals of project

To date, Shuto's lab is the only group to synthesize and investigate analogs of belactosin A. No analogs have been investigated that maintain the cyclopropyl peptidomimetic core with an alternative serine trap warhead to the natural β-

lactone. Based on the success of Imbach and Corey's β-lactam proteasome inhibitors and the limited number of β-lactam inhibitors in existence, nine β-lactam belactosin A analogs are proposed herein for synthesis and investigation of activity against the 20S proteasome (Figure 19).

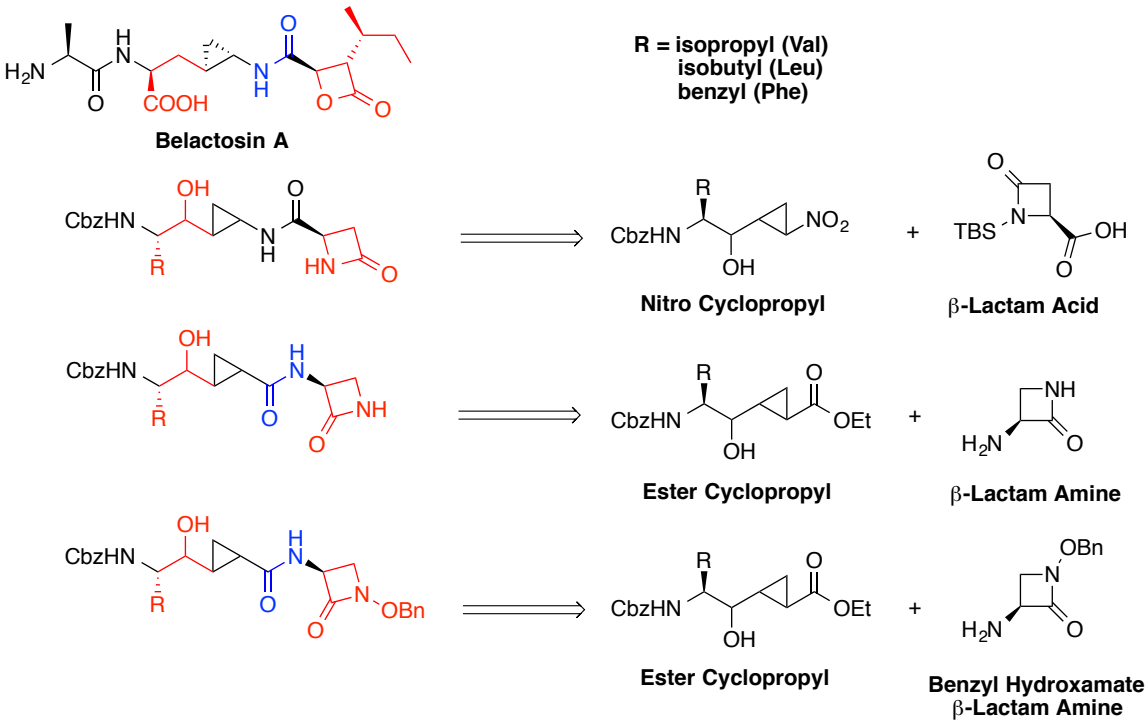


Figure 19: Projected library of nine analogs and retrosynthetic analysis. Each analog will have four possible stereoisomers.

The nitro and ester cyclopropyl peptidomimetic core elements were previously developed in Norma Dunlap's lab [26, 27]. The key steps in synthesizing the proposed analogs are the syntheses of each of the three proposed β-lactam fragments and optimizing the procedure for coupling the β-lactams to the

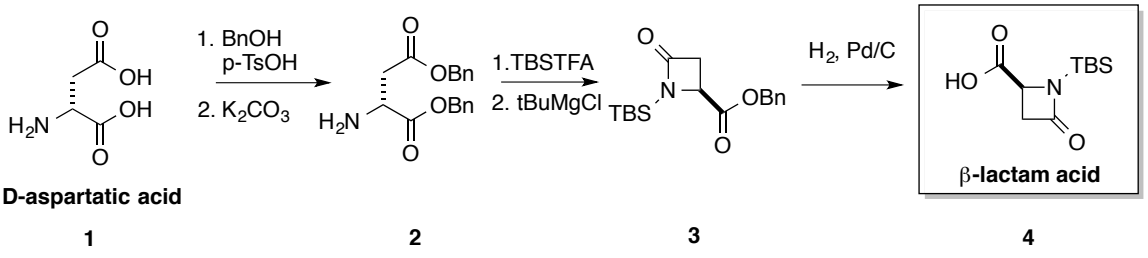
cyclopropyl core fragment. Once the synthesis of the analogs is completed, biological assays will be conducted to evaluate their inhibitory effects on the 20S proteasome. The goal of this project is to complete the β-lactam amine analog series and, time permitting, further explore optimization of the amine reduction for the β-lactam acid analog series.

Synthesis of β-lactams

β-Lactam acids

In developing a synthesis for the natural product dealanylalahopcin, Jack Baldwin engineered a method to synthesize β-lactam acid **4** from the readily available D-aspartic acid. This process proceeds through a base (*t*BuMgCl) catalyzed cyclization of a dibenzyl ester, N-silyl intermediate. Hydrogenolysis of the benzyl ester provides the final β-lactam acid product (Scheme 1) [28].

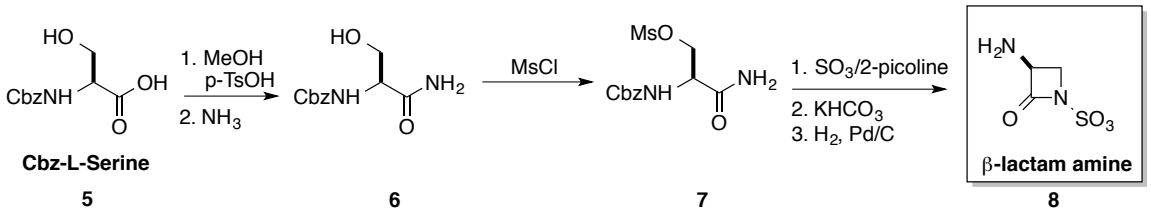
Scheme 1: Synthesis of β-lactam acid following Baldwin's procedure.



β-Lactam amines

In 1988, a synthesis was developed at Roche involving an intramolecular cyclization of a sulfate-activated amide by displacing a mesylate leaving group to afford a β-lactam amine. These β-lactam amines synthesized by Roche generally exhibited additional substitutions at the β-position of the unnatural amino acids, whereas the desired β-lactam **8** has no additional substitutions on the β-lactam ring. Applying this method to Cbz-L-serine as a cheap starting material would theoretically afford the sulfonated β-lactam amine **8** (Scheme 2) [29].

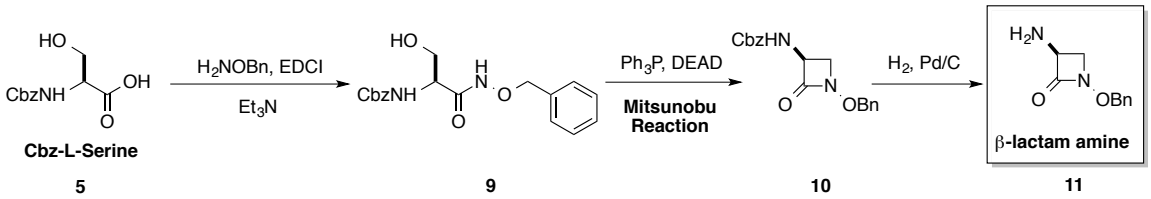
Scheme 2: Synthesis of β-lactam via sulfonation following Roche procedure.



Cbz-L-Serine **5** **6** **7** **8**

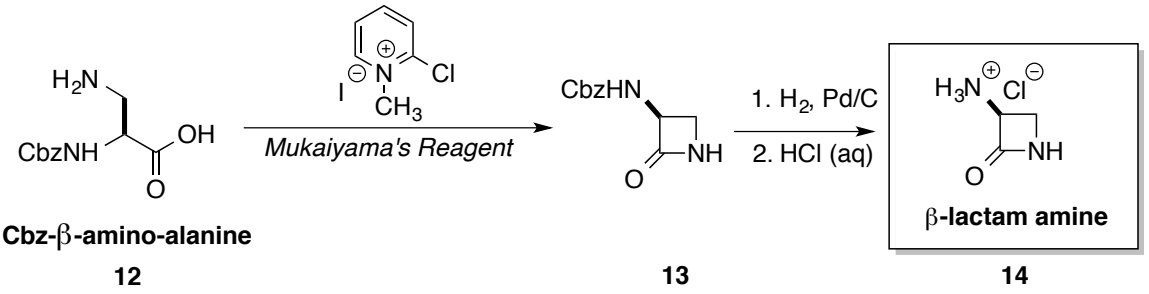
Another method for synthesizing β-lactam amines was developed by Marvin Miller in 1980. This method would also utilize Cbz-L-serine as a starting material, but it goes through a benzyl hydroxamate rather than a sulfonate in order to activate that amide for cyclization to afford β-lactam **11**. This method also utilizes a Mitsunobu reaction to transform the serine hydroxyl into a good leaving group rather than a mesylate (Scheme 3) [30].

Scheme 3: Synthesis of β-lactam via *O*-benzyl hydroxamate following Miller procedure



A third method for synthesizing β-lactam amines is a one-step reaction developed by Shital Chattopadhyay in 2015. This method does not require going through any intermediates in order to activate an amide for cyclization. Instead Cbz-β-amino-alanine, an unnatural amino acid, is used as the starting material and 2-chloro-1-methyl pyridinium iodide (Mukaiyama's reagent) is used to activate the acid, resulting in cyclization (Scheme 4) [31].

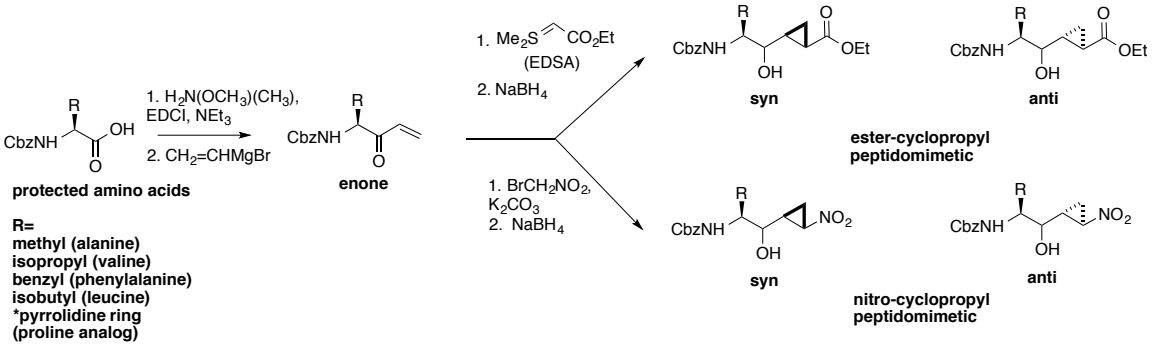
Scheme 4: Synthesis of β-lactam via Mukaiyama's reagent following Saha's procedure.



Synthesis of cyclopropyl backbone

The cyclopropyl backbones are a signature of Dunlap's group. The synthesis utilizes protected amino acids as the starting material, providing an easy route to R-group variability. The amino acids are converted to Weinreb amides and treated with vinylmagnesium bromide in order to form the enone intermediate. From this intermediate, the synthesis diverges to form either the ester or the nitro cyclopropyl product via treatment with ethyl (dimethylsulfuranylidene)acetate (EDSA) and bromonitromethane, respectively. After cyclopropyl formation, the resulting ketone is reduced by sodium borohydride to yield the final product (Scheme 5) [26, 27].

Scheme 5: Synthesis of cyclopropyl backbone core.

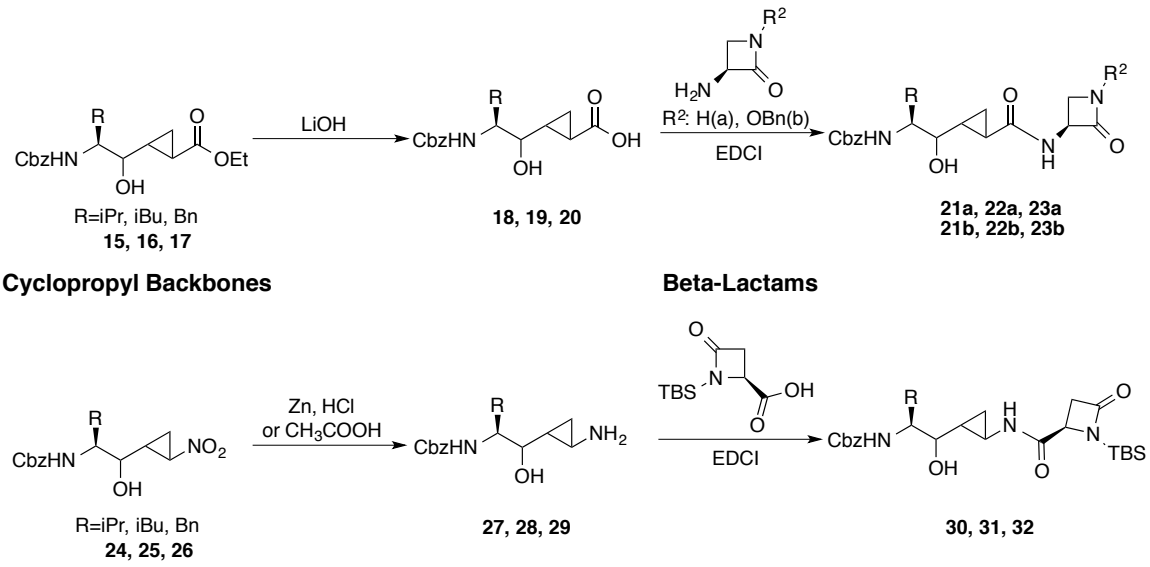


β-Lactam coupling

Finally, in order for the β-lactams to be coupled to the cyclopropyl backbones, the ester series must be hydrolyzed to an acid and the nitro series must

be reduced to an amine. Once this is completed, the two pieces can be coupled through EDCI catalyzed amide bond formation in order to form the initial precursor to the proposed analogs (Scheme 6).

Scheme 6: Coupling of β-lactams to amino acid-derived cyclopropyl backbones.



These precursors are lacking the Cbz-alanine unit at the end opposing the β-lactam. In Shuto's SAR analysis, he was able to show that the analogs were able to maintain activity without the alanine unit as long as a benzyl substituent was in place at the terminal end [22]. Based on these observations, these precursors will be evaluated for cancer cell cytotoxicity prior to addition of the Cbz-alanine terminal unit.

CHAPTER II: MATERIALS AND METHODS

Instruments, materials, and reagents

NMR data are obtained using a 500MHz FT-NMR model ECA-500 JEOL (Peabody, MA) purchased with funding provided by the National Science Foundation through the NSF-MRI program (#0321211). Chemical shifts are reported in parts per million (ppm) in reference to tetramethylsilane (TMS). Splitting patterns are represented by the following abbreviations: s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), dd (doublet of doublets), ddd (doublet of doublet of doublets), dt (doublet of triplets) and br (broad signal). Coupling constants (J values) are recorded in Hz. High-resolution electrospray ionization-mass spectrometry (ESI-MS) was performed at Notre Dame University, Notre Dame, Indiana. All other MS data was collected on Waters Synapt HDMS QToF instrument at Middle Tennessee State University.

Thin-layer chromatography (TLC) was performed on glass plates coated with silica gel with UV active backing purchased from Fisher Scientific, Pittsburgh, PA. TLC plates were analyzed utilizing UV light (254 nm) absorbance and subsequent staining with either phosphomolybdic acid (PMA), ninhydrin, or anisaldehyde (reagent grade, Aldrich, Milwaukee, WI) stain solutions or utilizing an iodine chamber. Flash column chromatography was performed with silica gel, 32-63 micron ASTM (reagent grade, Fisher Scientific, Pittsburgh, PA), and, where

indicated, flash column chromatography was also performed on an ISCO CombiFlash R_f 200 Teledyne ISCO, (Lincoln, NE) using a Teledyne ISCO cartridge preloaded with 5g of normal phase silica and a Teledyne ISCO preloaded 12g flash column.

Dichloromethane (DCM), methanol (MeOH), acetone, ethyl acetate (EA), ethanol (EtOH), and hexanes (Hex) were purchased from Fisher Scientific, Pittsburgh, PA. Chloroform was purchased reagent grade from Acros Organic, New Jersey, USA. Anhydrous tetrahydrofuran (THF) was obtained using a Pure Solv solvent purification system (Model PS-MD-3, Innovative Technology, Amesbury, MA). Deutero-chloroform ($CDCl_3$), Deutero-methanol (Methanol-d_4), and Deutero-acetone (Acetone-d_6) were purchased from Aldrich, Milwaukee, WI. Solvent extractions were performed using EA or DCM where indicated and washed with either distilled water, 1M hydrochloric acid (HCl), saturated sodium bicarbonate ($NaHCO_3$), and/or brine (reagent grade, Fisher Scientific, Pittsburgh, PA). The organic layer was dried with magnesium sulfate ($MgSO_4$) (Fisher Scientific, Pittsburgh, PA) and filtered. Evaporation of solvents was achieved using a Heidolph rotary evaporator (Model G3, Schwabach, Germany).

Triethylamine (Et_3N) and zinc metal (powdered) were obtained from Fisher Scientific, Pittsburgh, PA. (S)-3-Amino-2-(benzyloxycarbonylamino)-propanoic acid (95%) was purchased from Matrix Scientific. All other reagents were purchased from Sigma Aldrich (Milwaukee, WI). Where indicated, catalytic hydrogenation was performed on a Parr hydrogenation apparatus (Mod# A16CA, Moline, IL) using a GE Motor (Mod# 5KH35LNB1645X, RPM 1725).

Synthetic methods

β-Lactam acid

(L)-Dibenzyl aspartate (2): To a solution of D-aspartic acid (2.0 g, 15.0 mmol) in 6 mL of benzene, benzyl alcohol (12.5 mL, 120.0 mmol) and p-toluenesulfonic acid (5.7 g, 30.0 mmol) were added and the reaction was refluxed with a Dean-Stark trap for 3 hrs. The reaction solution was cooled to room temperature and transferred to an Erlenmeyer flask with an additional 15 mL of benzene and 25 mL of diethyl ether where the product was allowed to slowly crystallize out of solution over 4 weeks under refrigeration. The crystals were collected to yield 4.24 g (58%) of dibenzyl ester tosylate salt. To a 25 mL aqueous solution containing 1.6 g K_2CO_3, 1.2 g of the dibenzyl ester tosylate salt was added. The aqueous solution was extracted three times with EA. The combined organic layers were dried over $MgSO_4$ and evaporated to give 660 mg (85%) of the dibenzyl ester amine **2**.

(4*S*)-Benzyl-*N*-(*t*-butyldimethylsilyl)azetidin-2-one-4-carboxylate (3): To a solution of amine **2** (660 mg, 2.10 mmol) in 9 mL of dry acetonitrile (MeCN), *tert*-butyldimethylsilyl chloride (TBSCl) (64 mg, 0.42 mmol) and *N*-*tert*-butyldimethylsilyl-*N*-methyltrifluoroacetamide (MTBSTFA) (0.9 mL, 4.2 mmol) were added and the reaction mixture was allowed to stir at room temperature. After 1.5 hrs, the solvent was evaporated and placed under high vacuum conditions for 26 hrs to remove remaining MTBSTFA, providing 983.5 mg (110%) of crude silylated

product. The crude N-silylated dibenzyl ester was then suspended in 10 mL of anhydrous diethyl ether under argon and cooled to 0˚C. Once cooled, 1.1 mL of 2.0 M *tert*-butyl magnesium chloride (*t*BuMgCl) (2.3 mmol) in diethyl ether was added dropwise over 45 min resulting in the formation of a pale yellow precipitate. After the reaction mixture stirred for 17 hrs at room temperature, 15 mL of aqueous ammonium chloride (NH_4Cl) was added and allowed to stir for an additional 20 min. The reaction mixture was extracted twice with EA and the combined organic layers were then washed with brine. The organic layer was then dried over $MgSO_4$, filtered, and evaporated. The crude product was chromatographed on a 20 x 150 mm silica gel column eluting sequentially with 1:10, 1:7, and 1:5 EA/Hex to give 477 mg (71%) of β-lactam **3**. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.41 – 7.33 (m, 5H, aryl), 5.19 (s, 2H, benzyl C$\underline{H}_2$), 4.07 (dd, J = 6.2, 2.8 Hz, 1H, β-lactam C$\underline{H}$), 3.33 (dd, J = 15.1, 6.0 Hz, 1H, β-lactam C$\underline{H}_2$), 3.07 (dd, J = 15.0, 2.8 Hz, 1H, β-lactam C$\underline{H}_2$), 0.93 (s, 9H, 3 TBS *t*Bu C$\underline{H}_3$), 0.25 (s, 3H, TBS C$\underline{H}_3$), 0.06 (s, 3H, TBS C$\underline{H}_3$).

(4*S*)-*N*-(t-Butyldimethylsilyl)azetidin-2-one-4-carboxylic acid (4): To a solution of β-lactam **3** (477 mg, 1.49 mmol) in 40 mL anhydrous THF, 500 mg of 10% Pd/C was added. The reaction vessel was flushed twice with an argon balloon and twice with a H_2 balloon and stirred under H_2 (1 atm). After 20 hrs, the reaction mixture was filtered through celite, washing twice with EA, and the solvent was evaporated to yield 302 mg (88%) of β-lactam acid **4** as a white solid. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 4.05 (dd, J = 6.1, 2.9 Hz, 1H, β-lactam C$\underline{H}$), 3.37 (dd, J = 15.2, 6.1 Hz, 1H, β-lactam C$\underline{H}_2$), 3.10 (dd, J = 15.2, 2.8 Hz, 1H, β-lactam C$\underline{H}_2$), 0.90 (s, 9H, 3

TBS *t*Bu C$\underline{H}_3$), 0.09 (s, 6H, 2 TBS C$\underline{H}_3$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 175.76 (β-lactam $\underline{C}$=O), 171.49 ($\underline{C}$OOH), 48.69 (β-lactam $\underline{C}$H), 43.94 (β-lactam $\underline{C}H_2$), 25.69 (3 TBS *t*Bu $\underline{C}H_3$), 18.60 (TBS *t*Bu 4˚$\underline{C}$), -3.58 (2 TBS $\underline{C}H_3$).

β-Lactam amine

***N*-Benzyloxycarbonyl-L-serine amide (6)**: Cbz-L-Serine **5** (1.05 g, 4.39 mmol) was dissolved in 10 mL of MeOH and one drop of concentrated HCl was added. The solution was allowed to stir at room temperature for 24.5 hr. The reaction mixture was then extracted with EA and water and the organic layer was dried over $MgSO_4$, filtered, and the solvent evaporated to yield 1.03 g Cbz-L-serine methyl ester (92.4%).

Cbz-L-serine methyl ester (891.3 mg, 3.52 mmol) was dissolved in 50 mL of MeOH and ammonia gas (NH_3) was bubbled into the solution for 7 min at 0°C. The excess gas was allowed to vent into water. The reaction mixture was then allowed to stir at room temperature overnight. After 25.5 hr the reaction mixture was poured into 30 mL of 1M HCl, extracted with EA twice, and washed with brine. The organic layer was dried over $MgSO_4$, filtered, and the solvent evaporated to yield 382 mg crude product (45.6%). The amide **6** was then purified by recrystallization from EA. The crude product was suspended in 4.5 mL EA and heated in a 60°C water bath until the product was dissolved. The mixture was then allowed to cool to room temperature before being placed in an ice bath. The resulting white rod-shaped crystals were filtered through a Buchner funnel and dried yielding 127.2 mg (33.3%

recovery) of product. m. p. = 119.2-120°C. ^{1}H-NMR (500 MHz, Acetone-d_6): δ7.40 – 7.26 (m, 5H, aryl C<u>H</u>), 7.07 (br s, 1H, N<u>H</u>), 6.58 (br s, 1H, N<u>H</u>), 6.37 (br s, 1H, N<u>H</u>), 5.06 (s, 2H, Cbz C<u>H</u>$_2$), 4.22 (m, 1H, α-C<u>H</u>), 3.78 (m, 2H, β-C<u>H</u>$_2$), ^{13}C-NMR (125 MHz, Acetone-d_6): δ172.4(amide <u>C</u>=O), 155.5 (Cbz <u>C</u>=O), 137.3 (4° aryl <u>C</u>), 128.4 – 127.8 (5 aryl <u>C</u>), 66.0 (β-<u>C</u>H$_2$), 62.5 (Cbz <u>C</u>H$_2$), 56.6 (α-<u>C</u>H).

***N*-Benzyloxycarbonyl-L-serine(*O*-mesyl) amide (7):** Cbz-L-serine amide **6** (313.7 mg, 1.317 mmol) was dissolved in 4 mL of 1,2-dimethoxyethane under argon, cooled to -10°C, and treated with 550 μL (3.950 mmol) of Et_3N. An excess (255 μL, 3.292 mmol) of mesyl chloride (MsCl) was added dropwise. It is important to note that the appearance of yellow precipitate during addition of MsCl is an indicator that the reagent is being added too fast or the cooling bath is not cold enough which will reduce the product yield. The mixture stirred at -10°C for one hour before being slowly diluted with 6 mL of saturate brine and extracted with an equal volume of EA three times. The organic layer was dried over $MgSO_4$, filtered, and evaporated to yield a crude yellow oil. The oil was purified by trituration out of hexane and vacuum-dried to yield a white solid **7** (340.2 mg, 81.6%): ^{1}H-NMR (500 MHz, Acetone-d_6): δ7.49 – 7.18 (m, 5H, aryl C<u>H</u>), 6.89 (br s, 1H, N<u>H</u>), 5.10 (m, 2H, Cbz C<u>H</u>$_2$), 4.59 (m, 1H, α-C<u>H</u>), 4.50 (m, 2H, β-C<u>H</u>$_2$), 3.06 (s, 3H, sulfonyl-C<u>H</u>$_3$); 13C-NMR (125 MHz, Acetone-d_6): δ170.4 (amide <u>C</u>=O), 156.2 (Cbz <u>C</u>=O), 137.1 (4° aryl <u>C</u>), 128.5 – 127.9 (5 aryl <u>C</u>'s), 69.4 (β-<u>C</u>H$_2$), 66.4 (Cbz <u>C</u>H$_2$), 54.03 (α-<u>C</u>H), 36.4 (sulfonyl-<u>C</u>H$_3$).

***O*-Benzyl-α-*N*-Cbz-L-serine hydroxamate (9):** Cbz-L-serine **5** (1.0236 g, 4.28 mmol) was dissolved in 20 mL DCM and stirred at -10°C (ice/acetone) for 10

minutes. Once cooled, 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDCI) (822 mg, 4.28 mmol), Et_3N (242 μL, 4.28 mmol), and *O*-benzylhydroxylamine hydrochloride (683 mg, 4.28 mmol) were added in succession. The reaction mixture was stirred for 21.5 hrs, slowly coming to room temperature. The reaction mixture was diluted with 25 mL DCM and poured into 50 mL of 1M HCl. The aqueous layer was back-extracted with two 25 mL portions of DCM. The organic layers were combined, dried with $MgSO_4$, filtered, and evaporated. The crude product was purified by Combiflash using a 40 g prefilled column (1:1 to 4:1 EA/Hex for 3 min, hold 4:1 EA/Hex for 7 min, 4:1 EA/Hex to 100% EA for 4 min, and hold 100% EA for 3 min) to yield 154.5 mg (9.5%) of product **9** and 870.7 mg (69%) of an undesired dimer of product **9** (See Discussion). ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 9.65 (br s, 1H, hydroxamate N<u>H</u>), 7.30 (m, 10H, aryl), 5.96 (br d, J = 8.0 Hz, 1H, Cbz N<u>H</u>), 5.02 (m, 2H, Cbz C<u>H</u>$_2$), 4.83 (s, 2H, hydroxamate C<u>H</u>$_2$), 4.11 (dt, J = 8.9, 4.5 Hz, 1H, α-C<u>H</u>), 3.87 (dd, J = 11.2, 4.0 Hz, 1H, β-C<u>H</u>$_2$), 3.56 (dd, J = 11.5, 5.8 Hz, 1H, β-C<u>H</u>$_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 168.46 (hydroxamate <u>C</u>=O), 156.52 (Cbz <u>C</u>=O), 135.88 – 134.89 (2 4˚ aryl <u>C</u>), 129.31 – 128.01 (10 aryl <u>C</u>), 78.33 (hydroxamate <u>C</u>H$_2$), 67.35 (Cbz <u>C</u>H$_2$), 62.33 (β-<u>C</u>H$_2$), 53.56 (α-<u>C</u>H$_2$). Mass spectrum (ESI-MS) *m/z* ($C_{18}H_{21}N_2O_5$) calculated for (M+1) 345.1445, found 345.1446.

3-[(Cbz)amino]-2-azetidinone (13): To a solution of 2-chloro-1-methyl pyridinium iodide (Mukaiyama's reagent) (1.113 g, 4.36 mmol) in 150 mL dry MeCN, 575 μL (3.27 mmol) of diisopropylethylamine (DIPEA) was added and stirred over low heat. In 50 mL of dry MeCN, 519.0 mg (2.18 mmol) of **12** was sonicated to

achieve even suspension. This suspension was added dropwise over the course of 10 min. The reaction mixture was refluxed at 70˚C for 1 hr, allowing for full dissolution of reagents, and slowly cooled to stir at room temperature for an additional 27 hrs. The reaction mixture was poured into 50 mL of brine and extracted with two equal portions of EA. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated. The crude product was chromatographed on 20 x 150 mm silica gel eluting sequentially with 1:2, 1:1, 2:1, and 4:1 EA/Hex to afford 200.3 mg (42%) of the Cbz-protected β-lactam amine **13**. ^{1}H-NMR (500 MHz, Acetone-d_6) δ 7.47 – 7.06 (m, 5H, aryl), 6.93 (br s, 1H, N<u>H</u>), 4.96 (s, 2H, Cbz C<u>H</u>$_2$), 4.73 (m, 1H, β-lactam C<u>H</u>), 3.42 (t, J = 5.4 Hz, 1H, β-lactam C<u>H</u>$_2$), 3.13 (dd, J = 4.8, 2.7 Hz, 1H, β-lactam C<u>H</u>$_2$). ^{13}C-NMR (125 MHz, Acetone-d$_6$) δ 167.66 (β-lactam <u>C</u>=O), 155.62 (Cbz <u>C</u>=O), 137.21(4˚ aryl <u>C</u>), 128.40 – 127.91 (5 aryl <u>C</u>), 66.10 (Cbz-<u>C</u>H$_2$), 59.31 (β-lactam <u>C</u>H), 43.39 (β-lactam <u>C</u>H$_2$). Mass spectrum (ESI-MS) *m/z* ($C_{11}H_{12}N_2NaO_3$) calculated for (M+23) 243.0740, found 243.0732.

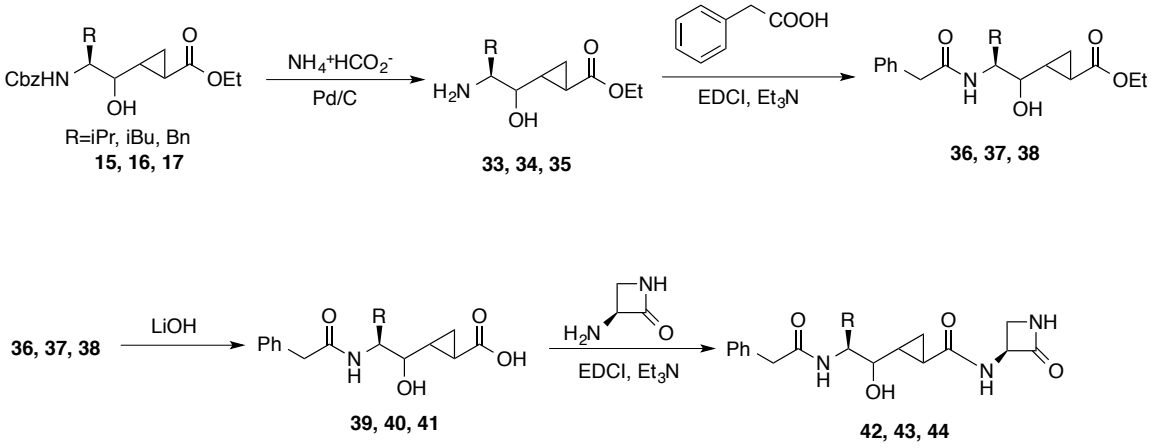
3-[Amino]-2-azetidinone hydrochloride (14): β-lactam **13** (96.4 mg, 0.438 mmol) was dissolved in EA and transferred to glass Parr bottle and diluted to approximately 50 mL. The 10% Pd/C (225 mg) was added and the reaction mixture was kept under H_2 (35 psi) on Parr shaker for 21.5 hrs. The reaction mixture was then filtered through celite and evaporated to give 54.5 mg of crude product. The crude product was resuspended in 25 mL EA and extracted three times with an equal volume of 1M HCl. The aqueous layers were combined and lyophilized to yield 50.6 mg (94%) of the hydrochloride salt **14**. ^{1}H-NMR (500 MHz, Methanol-d_4) δ 3.60

(m, 1H, β-lactam C<u>H</u>), 2.69 (dd, J = 25.0, 6.6 Hz, 1H, β-lactam C<u>H</u>$_2$), 2.69 (dd, J = 52.1, 6.7 Hz, 1H, β-lactam C<u>H</u>$_2$), 2.47 (d, J = 2.5 Hz, 1H, N<u>H</u>). ^{13}C-NMR (125 MHz, Methanol-d_4) δ 167.46 (<u>C</u>=O), 49.74 (β-lactam <u>C</u>H), 38.43 (β-lactam <u>C</u>H$_2$).

Cyclopropyl acids

*NOTE: Due to the carboxybenzyl (Cbz) group not being stable under the basic conditions required for the ester hydrolysis an alternative analog series was proposed (Scheme 7). Refer to discussion for further elaboration.

Scheme 7: Alternative synthesis of cyclopropyl acids for coupling to β-lactam amine.



Ethyl 2-((S)-2-amino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylate (33): To a solution of **15** (171.1 mg, 0.487 mmol) in 25 mL of MeOH, ammonium formate (NH_4HCO_2) (828.4 mg, 13.1 mmol) and 10% Pd/C (300 mg) were added. After 50 min stirring at room temperature, the reaction

mixture was filtered through celite and the solvent evaporated. The resulting slurry was then resuspended in saturated $NaHCO_3$ and extracted three times with EA. The aqueous layer was brought to a pH of 8 with 1M NaOH and extracted two additional times with EA. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to yield 54.9 mg (52%) of amine **33** as a clear oil. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 4.13 (m, 2H, OC$\underline{H}_2$CH$_3$), 3.66 – 3.42 (m, 1H, OC$\underline{H}$), 2.48 (m, 1H, α-C$\underline{H}$), 1.75 – 1.51 (m, 2H, 2 cyclopropyl C$\underline{H}$), 1.46 (m, 1H, β-C$\underline{H}$), 1.26 (m, 4H, cyclopropyl C$\underline{H}_2$, OCH$_2$C$\underline{H}_3$), 1.13 – 1.00 (m, 2H, cyclopropyl C$\underline{H}_2$), 0.97-0.85 (m, 7H, 2 C$\underline{H}_3$, cyclopropyl C$\underline{H}_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 174.41 (C$\underline{C}$=O), 71.47 – 69.41 (O-C$\underline{H}$), 62.13 – 61.95 (α-CH), 60.62 (O$\underline{C}$H$_2$CH$_3$), 23.39 – 22.32 (β-C$\underline{H}$), 20.20 – 19.23 (2 C$\underline{H}_3$), 16.78 – 16.51 (cyclopropyl C$\underline{H}$), 14.34 (OCH$_2$C$\underline{H}_3$), 12.13 – 10.80 (cyclopropyl C$\underline{H}_2$).

Ethyl 2-((S)-2-amino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylate (35): To a solution of **17** (303.3 mg, 0.764 mmol) in 50 mL of MeOH, NH_4HCO_2 (1.3 g, 20.6 mmol) and 10% Pd/C (603.3 mg) were added. After 40 min stirring at room temperature, the reaction mixture was filtered through celite and the solvent evaporated. The resulting slurry was then resuspended in saturated $NaHCO_3$ and extracted three times with EA. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to yield 169.2 mg (84%) of amine **35** as a yellow oil. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.42 – 6.98 (m, 5H, aryl), 4.10 (m, 3H, α-C$\underline{H}$, OC$\underline{H}_2$CH$_3$), 3.13 (m, 1H, OC$\underline{H}$), 3.04 – 2.36 (m, 2H, β-C$\underline{H}_2$), 1.78 – 1.48 (m, 2H, 2 cyclopropyl C$\underline{H}$), 1.36 – 1.11 (m, 3H, OCH$_2$C$\underline{H}_3$), 1.11 –

0.79 (m, 2H, cyclopropyl C$\underline{H}_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 174.22 ($\underline{C}$=O), 139.22 (4˚ aryl $\underline{C}$), 129.32 – 126.51(5 aryl $\underline{C}$), 75.26 – 72.90 (O-$\underline{C}$H), 60.69 (O$\underline{C}H_2CH_3$), 57.21 (α-$\underline{C}$H), 40.79 – 38.87 (β-$\underline{C}H_2$), 26.23 – 23.67 (cyclopropyl $\underline{C}$H), 17.79 – 17.28 (cyclopropyl $\underline{C}$H), 14.35 ($OCH_2\underline{C}H_3$), 12.58 – 11.43 (cyclopropyl $\underline{C}H_2$).

Ethyl 2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylate (36): To a solution of phenylacetic acid (34.4 mg, 0.253 mmol) in 1 mL of DCM, (1-Cyano-2-ethoxy-2-oxoethylidenaminooxy)dimethylamino-morpholino-carbenium hexafluorophosphate (COMU) (108 mg, 0.253 mmol) was added and allowed to stir at 0˚C for 15 min to activate the acid. After 15 min, amine **33** (54.9 mg, 0.253 mmol) was suspended in 1 mL of DCM and added to the reaction mixture followed by DIPEA (89 μL, 0.506 mmol). After stirring at 0˚C for 1 hr, the reaction mixture was allowed to come to room temperature and stir overnight. After 26 hrs, the reaction mixture was poured into 25 mL of EA and extracted sequentially with two 5 mL portions of 1M HCl, two 5 mL portions of $NaHCO_3$, and two 5 mL portions of brine. The organic layer was dried over $MgSO_4$ and evaporated to yield 116.7 mg of crude product. The product was purified on a 15 x 100 mm silica column eluting with 1:10 EA/Hex, 1:5 EA/Hex, 1:1 EA/Hex, 4:1 EA/Hex, 100% EA, and 100% MeOH. The product was identified to elute off the column in 1:5 EA/Hex, affording 37.1 mg (44%) of amide **36** with some impurities. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.49 – 6.96 (m, 5H, aryl), 4.24 – 3.98 (m, 2H, O$\underline{C}H_2CH_3$), 3.83 – 3.47 (m, 3H, benzyl amide $\underline{C}H_2$, $\underline{C}$H), 3.41 – 3.09 (m, 1H, $\underline{C}$H), 2.16 (m, 1H, β-$\underline{C}$H), 1.80 – 0.71 (m, 13H, 3 $\underline{C}H_3$, 2

cyclopropyl C<u>H</u>, cyclopropyl C<u>H</u>$_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 173.29 – 173.14 (<u>C</u>=O), 168.90 (<u>C</u>=O), 134.98 (4˚ aryl <u>C</u>), 129.36 – 126.99 (5 aryl <u>C</u>), 80.21 (O-<u>C</u>H), 65.05 ((α-<u>C</u>H), 60.91 (O-<u>C</u>H$_2$CH$_3$), 43.66 (benzyl amide <u>C</u>H$_2$), 28.97 (β-<u>C</u>H), 27.68 – 26.78 (cyclopropyl <u>C</u>H), 21.71 (CH$_3$), 20.45 (CH$_3$), 14.31 (O-<u>C</u>H$_2$CH$_3$), 12.36 (cyclopropyl <u>C</u>H$_2$). Mass spectrum (ESI-MS) *m/z* ($C_{19}H_{28}NO_4$) calculated for (M+1) 334.2013, found 334.2011.

Ethyl 2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylate (38): To a solution of phenylacetic acid (87 mg, 0.643 mmol) in 8 mL of DCM, EDCI (123 mg, 0.643 mmol) was added and allowed to stir for 15 min to activate the acid. After 15 min, amine **35** (169.2 mg, 0.643 mmol) was suspended in 8 mL of DCM and added to the reaction mixture followed by Et_3N (638 μL, 4.83 mmol). After stirring at room temperature for 21 hrs, the reaction mixture was poured into 25 mL of 1M HCl and extracted two times with an equal volume of EA. The organic layers were combined and washed with saturated $NaHCO_3$. The organic layer was dried over $MgSO_4$, filtered, and evaporated to give 141.6 mg of crude product. The product was further purified by triturating with hexane to afford 56.8 mg (23%) of amide **38**. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.49 – 6.76 (m, 10H, aryl), 5.57 (dd, J = 16.8, 7.3 Hz, 1H, N<u>H</u>), 4.32 – 4.16 (m, 1H, α-C<u>H</u>), 4.16 – 4.00 (m, 2H, OC<u>H</u>$_2$CH$_3$), 3.69 – 3.20 (m, 3H, benzyl amide C<u>H</u>$_2$, O-C<u>H</u>), 3.03 – 2.58 (m, 2H, β-C<u>H</u>$_2$), 1.76 – 1.39 (m, 2H, 2 cyclopropyl C<u>H</u>), 1.32 – 1.17 (m, 3H, OCH$_2$C<u>H</u>$_3$), 1.18 – 0.83 (m, 2H, cyclopropyl C<u>H</u>$_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 174.24 (<u>C</u>=O), 172.60 (<u>C</u>=O), 137.39 (4˚ aryl <u>C</u>), 134.27 (4˚ aryl <u>C</u>), 129.53 – 126.75 (10 aryl <u>C</u>),

74.80 – 73.26 (O-CH), 60.79 (O-CH_2CH_3), 57.10 – 56.94 (α-CH), 43.59 – 43.55 (benzyl amide CH_2), 35.81 – 35.41 (β-CH_2), 24.36 – 16.79 (2 cyclopropyl CH), 14.38 (O-CH_2CH_3), 11.92 – 11.36 (cyclopropyl CH_2). Mass spectrum (ESI-MS) *m/z* ($C_{23}H_{28}NO_4$) calculated for (M+1) 382.2013, found 382.2001.

2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylic acid (39): To a solution of ester **36** (37.1 mg, 0.111 mmol) in 6 mL THF/ 1 mL H_2O, lithium hydroxide monohydrate (40 mg, 0.952 mmol) was added and the reaction was stirred at room temperature. After 4 hrs, the reaction mixture was poured into 5 mL EA/ 5 mL H_2O and the organic layer was collected for starting material. The aqueous layer was acidified to pH=2 and extracted three times with DCM. The DCM organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to give 12.8 mg of crude product. The EA layer was also found to contain product, which afforded an additional 12.9 mg for a combined mass of 25.7 mg (75%) of crude product with some starting material still present. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.55 – 6.97 (m, 5H, aryl), 3.67 (s, 2H, benzyl amide CH_2), 3.58 (dd, J = 8.4, 4.5 Hz, 1H, α-CH), 3.21 (m, 1H, O-CH), 2.29 – 2.08 (m, 1H, β-CH), 1.74 – 1.66 (m, 1H, cyclopropyl CH), 1.61 (m, 1H, cyclopropyl CH), 1.23 – 1.18 (m, 1H, cyclopropyl CH_2), 1.17 (d, J = 6.6 Hz, 3H, CH_3), 1.12 (d, J = 7.0 Hz, 3H, CH_3), 0.91 – 0.78 (m, 1H, cyclopropyl CH_2). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 177.48 (C=O), 168.95 (C=O), 134.74 (4˚ aryl C), 129.87 – 126.01 (5 aryl C), 82.76 – 77.64 (O-CH), 70.23 – 62.82 (α-CH), 43.48 (benzyl amide CH_2), 28.81 (β-CH), 27.69 –

27.17 (cyclopropyl CH), 21.02 (CH_3), 20.30 (CH_3), 19.98 (cyclopropyl CH), 12.71 (cyclopropyl CH_2).

2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylic acid (41): To a solution of ester **38** (56.8 mg, 0.149 mmol) in 8 mL THF/ 2 mL H_2O, lithium hydroxide monohydrate (25 mg, 0.596 mmol) was added and the reaction was stirred at room temperature. After 5 hrs, the reaction mixture was poured into 5 mL EA/ 5 mL H_2O and the organic layer was collected for starting material. The aqueous layer was acidified to pH=2 and extracted three times with DCM. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to afford 19.9 mg (38%) of crude product. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.46 – 6.81 (m, 10H, aryl), 5.57 (m, 1H, NH), 4.23 (m, 1H, α-CH), 3.65 (s, 2H, benzyl amide CH_2), 3.62 – 3.40 (m, 1H, O-CH), 2.93 (m, 1H, β-CH_2), 2.65 (m, 1H, β-CH_2), 1.71 (m, 1H, cyclopropyl CH), 1.65 – 1.45 (m, 1H, cyclopropyl CH), 1.17 – 0.77 (m, 2H, cyclopropyl CH_2). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 176.75 (C=O), 173.25 (C=O), 137.07 (4˚ aryl C), 134.06 (4˚ aryl C), 129.55 – 126.83 (10 aryl C), 56.97 (α-CH), 43.39 (O-CH), 41.17 (benzyl amide CH_2), 35.58 (β-CH_2), 24.90 (cyclopropyl CH), 17.03 (cyclopropyl CH), 12.18 (cyclopropyl CH_2).

Cyclopropyl amines

2-((S)-2-(benzylcarbonylamino)-1-hydroxy-4-methylpentyl) cyclopropaneamine (28): To a solution of nitrocyclopropyl **25** (36 mg, 0.11 mmol)

in 2 mL isopropyl alcohol and 1 mL 1M HCl, 30 mesh granular Zn (130 mg, 2.0 mmol) was added and the solution was stirred at room temperature. After 35 min, the reaction mixture was gravity filtered, washing with ethyl acetate, and poured into saturated $NaHCO_3$ (aq). The aqueous solution was extracted two times with EA and the organic layers were combined, dried over $MgSO_4$, and evaporated to yield 49 mg (100%) of crude cyclopropylamine.

2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropaneamine (29): To a solution of nitrocyclopropyl **26** (60 mg, 0.1g mmol) in 3 mL isopropyl alcohol and 1.5 mL 1M HCl, Zn dust (190 mg, 2.9 mmol) was added and the solution was stirred at room temperature. After 40 min, the reaction mixture was gravity filtered into a separatory funnel containing 20 mL of water and 20 mL of EA. The aqueous layer was extracted twice with EA and the organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to afford 44 mg (81%) of crude cyclopropylamine. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.74 – 6.84 (m, 10H, aryl), 5.59 – 5.14 (m, 1H, α-C<u>H</u>), 5.14 – 4.86 (m, 2H, Cbz C<u>H</u>$_2$), 4.20 – 3.74 (m, 2H, O-C<u>H</u>$_2$), 3.24 – 2.70 (m, 2H, β-C<u>H</u>$_2$, cyclopropyl C<u>H</u>), 2.35 – 1.89 (m, 1H, cyclopropyl C<u>H</u>), 1.08 – 0.41 (m, 1H, cyclopropyl C<u>H</u>$_2$). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 156.74 (<u>C</u>=O), 138.30 – 136.55 (2 4˚ aryl <u>C</u>), 129.28 – 126.51 (10 aryl <u>C</u>), 75.57 – 74.27 (<u>C</u>H), 66.78 (Cbz <u>C</u>H$_2$), 57.22 (<u>C</u>H), 38.44 (β-<u>C</u>H$_2$), 29.80 – 25.51 (<u>C</u>H), 12.70 (cyclopropyl <u>C</u>H$_2$).

Belactosin analogs

Benzyl *N*-[(2S)-1-{2-[1-(*tert*-butyldimethylsilyl)-4-oxoazetidine-2-amido]cyclopropyl}-1-hydroxy-4-methylpentan-2-yl]carbamate (31): To a solution of β-lactam **4** (27 mg, 0.11 mmol) in 1.0 mL DCM, EDCI (21 mg, 0.11 mmol) was added and the reaction mixture was stirred at room temperature for 10 min to activate the acid. Once the acid was activated, amine **28** (33 mg, 0.11 mmol) was added in 1 mL DCM followed by 0.2 mL Et_3N. After stirring at room temperature for 18 hrs, the reaction mixture was poured into 1M HCl and extracted two times with EA. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to give 226 mg of crude product. The crude product was chromatagraphed on 25 x 100 mm silica gel eluting sequentially with 1:10, 1:5, 1:1, and 4:1 EA/Hex to afford 15.2 mg (27%) of belactosin analog **31**. The ^{1}H-NMR was complex, but key peaks were recognized. Mass spectrum (ESI-MS) *m/z* ($C_{27}H_{44}N_3O_5Si$) calculated for (M+1) 518.3020, found 518.3045.

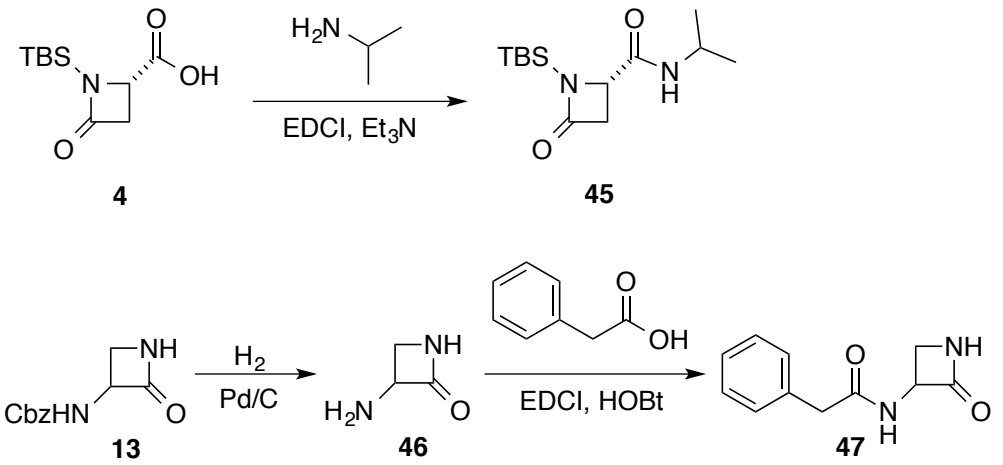
Benzyl *N*-[(2S)-1-{2-[1-(*tert*-butyldimethylsilyl)-4-oxoazetidine-2-amido]cyclopropyl}-1-hydroxy-4-phenylpropan-2-yl]carbamate (32): To a solution of β-lactam **4** (30 mg, 0.129 mmol) in 1.0 mL DCM, EDCI (25 mg, 0.129 mmol) was added and the reaction mixture was stirred at room temperature for 15 min to activate the acid. Once the acid was activated, amine **29** (44 mg, 0.129 mmol) was added in 1 mL DCM followed by 0.2 mL Et_3N. After stirring at room temperature for 18 hrs, the reaction mixture was poured into 1M HCl and extracted two times with EA. The organic layers were combined, dried over $MgSO_4$, filtered, and

evaporated to give 89 mg of crude product. The crude product was chromatagraphed on 15 x 100 mm silica gel eluting sequentially with 1:10, 1:6, 1:3, 1:1, and 3:1 EA/Hex to afford 19 mg (27%) of belactosin analog **32**. The ^{1}H-NMR was complex, but key peaks were recognized. Mass spectrum (ESI-MS) *m/z* ($C_{30}H_{42}N_3O_5Si$) calculated for (M+1) 552.2888, found 552.2878.

Model coupling for proof of concept

Due to limited materials and time, a model coupling was performed to prove that a method had been developed to successfully couple the β-lactams to a respective amine and carboxylic acid (Scheme 8).

Scheme 8: Model coupling of β-lactams.



1-(*tert*-Butyldimethylsilyl)-4-oxo-*N*-(propan-2-yl)azetidine-2-carboxamide (45): To a solution of acid **4** (20.0 mg, 0.087 mmol) in 2 mL of DCM, EDCI (16.7 mg, 0.087 mmol) was added and allowed to stir for 15 min to activate the

acid. After 15 min, 2-aminopropane (7.5 μL, 0.087 mmol) was added followed by an excess of Et_3N (0.2 mL) and the reaction was stirred at room temperature for 22.5 hrs. Once complete, the reaction mixture was poured into 1M HCl and extracted two times with EA. The organic layers were combined, dried over $MgSO_4$, filtered, and evaporated to afford 14.9 mg (63%) of **45**. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 5.89 (d, J = 8.1 Hz, 1H, NH), 4.11 (dt, J = 13.8, 6.9, 6.9 Hz, 1H, CH), 3.93 (dd, J = 6.2, 2.8 Hz, 1H, β-lactam CH), 3.38 (dd, J = 15.7, 6.6 Hz, 1H, β-lactam CH_2), 2.97 (dd, J = 15.5, 2.9 Hz, 1H, β-lactam CH_2), 1.17 (t, J = 6.8 Hz, 6H, 2 CH_3), 0.97 (s, 9H, 3 TBS tBu CH_3), 0.30 (s, 3H, TBS CH_3), 0.15 (s, 3H, TBS CH_3). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 172.44 (C=O), 170.74 (C=O), 50.61 (β-lactam CH), 48.57 (CH), 44.93 (β-lactam CH_2), 26.31 (3 TBS tBu CH_3), 22.64 (CH_3), 22.57 (CH_3), 18.67 (TBS tBu 4° C), -5.42 (TBS CH_3), -6.02 (TBS CH_3).

***N*-(2-oxoazetidin-3-yl)-2-phenylacetamide (52)**: To a solution of β-lactam **13** (73.9 mg, 0.336 mmol) in 50 mL of ethyl acetate, 184.3 mg of 10% Pd/C was added and placed under H_2 at 25 psi to react on a Parr shaker for 2 hrs. After 2 hrs, the reaction suspension was filtered through celite and evaporated to provide **46** in an assumed 100% yield of 41 mg. A solution of phenylacetic acid (45.7 mg, 0.336 mmol) and 1-hydroxybenzotriazole (HOBt) (45.4 mg, 0.336 mmol) in 2 mL of DCM was cooled to 0°C and EDCI (64.5 mg, 0.336 mmol) was added. The reaction was stirred at 0°C for 30 min and at room temperature for an additional hour. Amine **46** was suspended in 2 mL of DCM and added to the activated acid followed by Et_3N (89 μL, 0.672 mmol) to stir overnight at room temperature. After 21 hrs, the reaction

mixture was poured into 1M HCl and extracted three times with EA. The organic layers were combined and washed with $NaHCO_3$. The organic layer was dried over $MgSO_4$, filtered, and evaporated to provide 15.4 mg of crude product. The product was purified by trituration with 2:3 EA/Hex to afford 3.8 mg (5.5%) of **47** with minor impurities. ^{1}H-NMR (500 MHz, Chloroform-*d*) δ 7.59 – 6.98 (m, 5H, aryl), 6.13 (d, J = 7.2 Hz, 1H, NH), 4.93 (ddd, J = 7.5, 5.2, 2.5 Hz, 1H, β-lactam CH), 3.68 – 3.54 (m, 3H, β-lactam CH_2, Ph-CH_2), 3.34 – 3.19 (m, 1H, β-lactam CH_2), 1.85 – 1.75 (m, 1H, β-lactam NH). ^{13}C-NMR (125 MHz, Chloroform-*d*) δ 174.66 (C=O), 171.50 (C=O), 134.10 (4˚ aryl C), 129.57 – 127.37 (5 aryl C), 58.20 (CH), 44.67 (CH_2), 43.46 (CH_2). Mass spectrum (ESI-MS) *m/z* ($C_{11}H_{13}N_2O_2$) calculated for (M+1) 205.0972, found 205.0948.

CHAPTER III: RESULTS AND DISCUSSION

Synthesis of the β-lactams

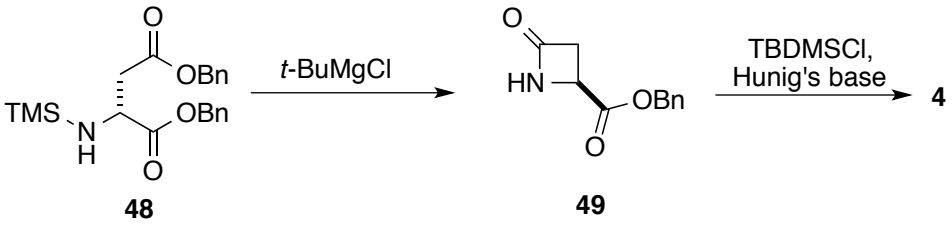
Although several β-lactam syntheses have been reported, the β-lactam cyclization can be difficult to achieve. Intramolecular cyclizations require a great deal of dilution and controlled rates of reagent addition to prevent any intermolecular coupling. Many of these reported syntheses also require that the nitrogen of the amide bond have a certain nucleophilicity and/or pK_a of its associated hydrogen. Obtaining the specified conditions of the previously reported methods proved to be quite challenging. A discussion of the issues encountered with each method is described herein.

β-Lactam acid

For the β-lactam acid, there was no difficulty in obtaining the silylated dibenzylester, but several attempts were made to close the ring using *t*BuMgCl. In the experimental write-up, Baldwin instructs to add the *t*BuMgCl dropwise in the cyclization step, taking the dibenzyl ester directly to β-lactam **3** [28]. However, when adding at a rate of 3 drops every 10 sec, the product was obtained at only an 8.2% yield and a 17% recovery of desilyl dibenzyl ester **2**. A subsequent attempt utilized the same drop rate but with an increased the amount of *t*BuMgCl from 1.2 to 1.6 equivalents only provided the product at a 28% yield and an increase to 2.0

equivalents yielded no product. Bringing the amount of *t*BuMgCl back to 1.2 equivalents and slowing the rate to only 2 drops every 5 sec, completing the addition in about 15 min, also yielded no cyclized product. Finally, upon further investigation in the literature, it was discovered that in the procedure that cyclized the closely related N-TMS dibenzyl ester **48** to β-lactam **43** (Scheme 9) Baldwin listed an addition rate of 23.0 mL of 2M *t*-BuMgCl over 45 min [28].

Scheme 9: Alternative synthesis of β-lactam acid used by Baldwin et al.



When this same rate was used for the cyclization of the N-TBS dibenzyl ester, the cyclization to β-lactam **3** was achieved in a 71% yield (Scheme 1).

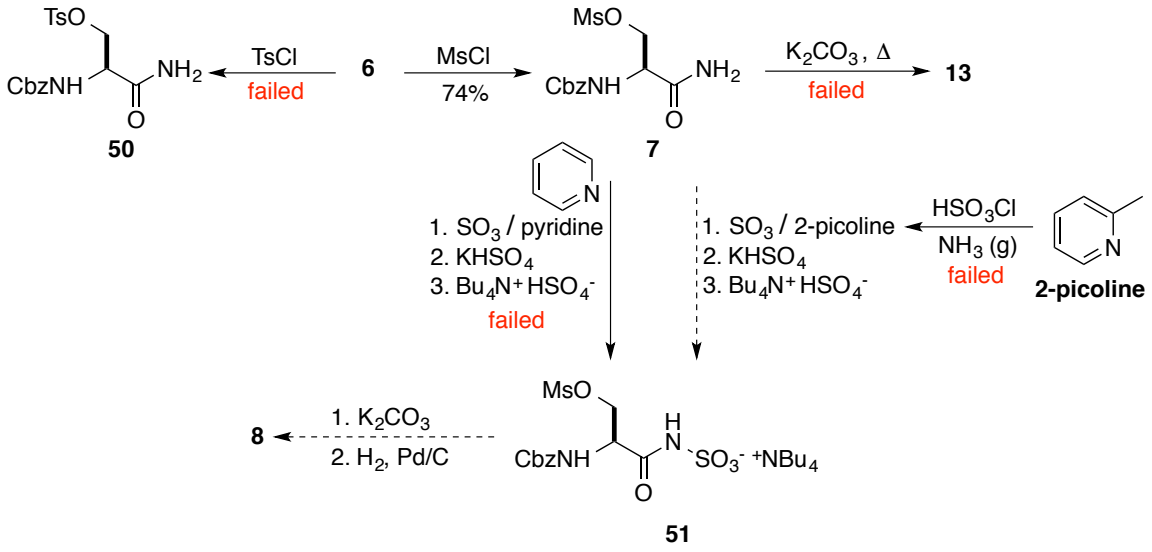
β-Lactam amine

Formation of the β-lactam amine proved to be especially challenging. The primary challenge was in the formation of either a C-N bond or an amide bond in order to close the ring. Three different procedures were attempted to accomplish this synthesis with the Saha procedure being the most successful.

In following the Roche procedure as outlined in Scheme 2, several attempts and modifications were made due to availability of materials. Serine amide **6** was

readily accessible and since tosyl chloride (TsCl) was already in stock, an attempt was made to synthesize tosylate **50** rather than mesylate **7**, but the reaction inexplicably and consistently failed (Scheme 10).

Scheme 10: Summation of attempted syntheses of β-lactam amine following Roche procedure. Dashed reaction arrows denote reactions that remain unattempted.



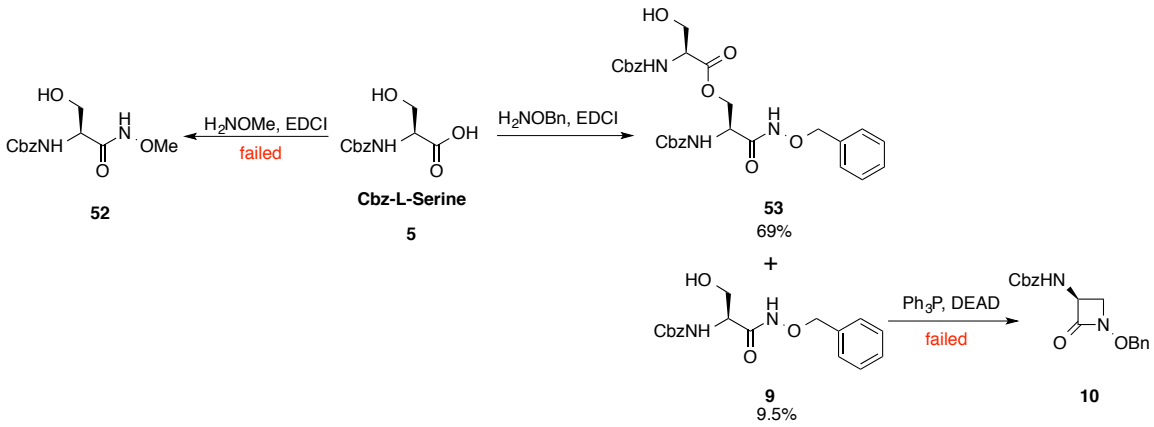
Mesyl chloride (MsCl) was purchased from Sigma Aldrich, and the mesylation resulted in a 74% yield of **7**. An attempt was made to directly cyclize **7** to β-lactam **13** using potassium carbonate and heat, but the reaction failed (Scheme 10). Further research revealed that an electron withdrawing group (EWG) must be attached to the amide nitrogen in order for cyclization to occur. This is because the EWG significantly lowers the pK_a of the amide, resulting in selective deprotonation and inhibition of undesired proton-transfer processes. Without the selectivity provided

by an EWG, direct cyclizations of primary amides fail [32]. This knowledge revealed the necessity of the sulfate intermediate in the Roche procedure. Sulfonation of **7** was first attempted using a pyridine-sulfur trioxide complex, but these attempts repeatedly failed. Further research revealed that an extensive study had previously been conducted that showed that the 2-picoline-sulfur trioxide complex was the only complex capable of providing the desired product **51**. All other complexes, including pyridine-sulfur trioxide, result in incomplete conversion to the sulfonated product [33]. An attempt was made to synthesize the 2-picoline-sulfur trioxide complex, but when that failed, other routes to the β-lactam amine that did not require sulfonation were investigated.

The Miller procedure provided an alternative route to the β-lactam amine. Rather than using a sulfonate as an EWG, Miller utilized N-alkoxy groups, specifically an N-benzyloxy, to lower the pK_a of the amide. Initially, an attempt was made to couple methoxyamine to the serine acid to yield product **52** (Scheme 11). The rationale behind this decision was to keep the substituents on the β-lactam as small and unobtrusive as possible. However, after several failed attempts to synthesize **52** utilizing both DMF/H_2O and methylene chloride as reaction solvents and running the reaction at both room temperature and -10˚C, efforts switched to Miller's N-benzyloxy instead. Compound **9** was synthesized in a 9.5% yield along with an undesired dimer **53** appearing in a 69% yield. Initially, it was thought that dimer **53** was the desired product due to the ^{1}H-NMR signals being closer in

similarity to those reported in the literature than those presented by product **9** in $CDCl_3$.

Scheme 11: Summation of attempted syntheses of β-lactam amine following Miller procedure.



The literature had reported the data in 1:1 $CDCl_3/CD_3COCD_3$, which prevented an exact comparison. However, HR-MS of **53** did not show the calculated peak of the desired product (*m/z*: 345.1445). Rather, the primary peak was a *m/z* of 588.1883, which matches the calculated value of the dimer at 588.1958. The verification of the dimer structure of **53** was then verified upon looking closer at the integrations of the ^{1}H-NMR. Cyclization of **9** repeatedly failed to give the desired β-lactam **10**. After β-lactam **13** was successfully synthesized via Saha's procedure, it was decided to forgo this route as well.

The cyclization of amino acid **12** utilizing Mukaiyama's reagent as a catalyst worked splendidly to provide β-lactam **13** in one step with yields as high as 60%.

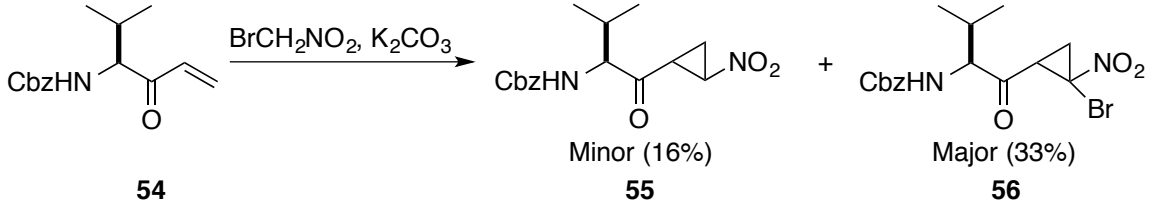
Transfer hydrogenation using ammonium formate as a hydrogen donor was the first route attempted to remove the Cbz, and, while it may have been successful, the product was too small and polar and could not be extracted out of the aqueous layer. Catalytic hydrogenation utilizing a Parr shaker was attempted next, and provided the β-lactam as a primary amine with a 100% yield. However, the primary amine was not stable. To solve this issue, the primary amine was extracted from ethyl acetate with 1M HCl following catalytic hydrogenation and the water was lyophilized to provide a 94% yield of the more stable ammonium salt **14**.

Synthesis of the cyclopropyl amines and esters

Nitrocyclopropanation

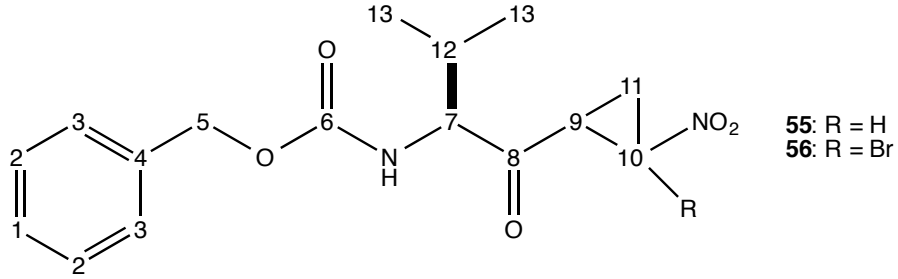
In order to pursue each series, more nitrocyclopropyls and cyclopropyl esters needed to be synthesized as starting material following the procedure previously developed by Dunlap's lab [26, 27]. However, issues were encountered in trying to repeat the nitrocyclopropanation of enones. Rather than forming the valine nitrocyclopropyl **55** as expected, the major product isolated was bromonitrocyclopropyl **56** (Scheme 12). The identity of **56** was confirmed through both 1D and 2D NMR spectroscopy and mass spectrometry. Table 1 shows a comparison of ^{1}H and ^{13}C shifts for product **55** and **56**.

Scheme 12: Synthesis of undesired bromonitrocyclopropyl products.



54 **55** **56**

Table 1: Comparison of ^{1}H and ^{13}C NMR shifts for products **55** and **56**.

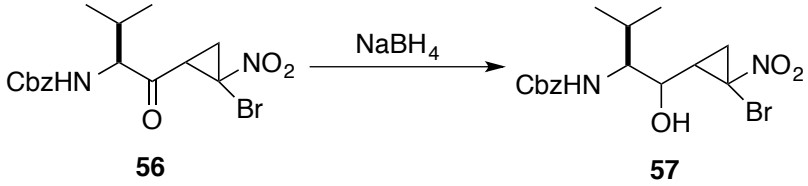


Position	δ ^{1}H-NMR (mult, *J* Hz)		δ ^{13}C-NMR (type)	
	55	**56**	**55**	**56**
1	7.32 (m)	7.31 (m)	128.43 (CH)	128.44 (CH)
2			128.29 (CH)	128.22 (CH)
3			128.70 (CH)	128.70 (CH)
4	---	---	136.09 (q)	136.02 (q)
5	5.10 (d, 3.11)	5.07 (m)	67.39 (CH_2)	67.43 (CH_2)
6	---	---	156.44 (q)	156.53 (q)
7	4.56 (m)	4.45 (dd, 8.1, 5.4)	60.74 (CH)	62.39 (CH)
8	---	---	203.58 (q)	204.87 (q)
9	3.10 (ddt, 9.6, 7.1, 2.3)	5.19 (dd, 9.4, 6.6)	29.43 (CH)	76.29 (CH)
10	4.56 (m)	---	65.74 (CH)	93.48 (q)
11	2.08 (dq, 9.8, 4.8)	3.53 (m)	18.73 (CH_2)	37.17 (CH_2)
	1.70 (m)			
12	2.28 (m)	2.18 (m)	29.88 (CH)	29.23
13	1.05 (dd, 6.7, 2.2)	1.01 (d, 6.9)	19.79 (CH_3)	19.87 (CH_3)
	0.82 (dd, 6.9, 4.2)	0.86 (d, 6.8)	17.05 (CH_3)	17.34 (CH_3)
NH	5.37 (d, 6.86)	5.43 (br d, 8.0)	---	---

The predicted *m/z* and isotopic pattern of **56** was 398 (100%), 399 (17%), 400 (100%), 401 (17%). Using Waters TOF MS ES+, masses of 421, 422, 423, and 424 were identified and correspond to the mass of **56** with a sodium adduct (+23).

To further verify whether the bromine was attached to C10 and not C9, **56** was reduced to the alcohol using $NaBH_4$ following the procedure cited previously by Dunlap's lab [26, 27] to give **57** (Scheme 13).

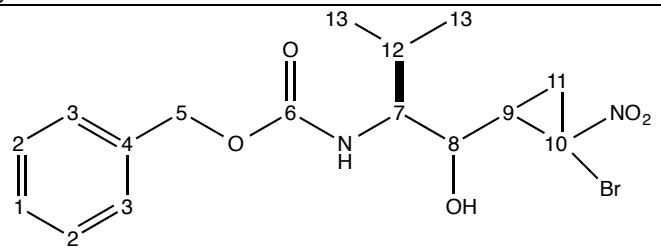
Scheme 13: $NaBH_4$ reduction of valine bromonitrocyclopropyl.



56 **57**

Using both 1D and 2D NMR spectroscopy, it was verified that H8 and H9 are on adjacent carbons and, thus, that the bromine and nitro groups were attached to the same carbon. COSY showed a clear coorelation between H8 and H9, as well as a coorelation between H9 and H11 and between H8 and H7. It is also interesting that one stereoisomer also shows a coorelation between H8 and H11. Table 2 summarizes the ^{1}H-NMR shifts of **57** along with the COSY interactions that confirmed these assignments.

Table 2: Summary of ^{1}H-NMR and COSY data of **57.**



**Note: The correlation between H8 and H11 only occurs for one stereoisomer. This data corresponds to a mixture of four stereoisomers.*

Position	δ ^{1}H-NMR (mult, *J* Hz)	COSY Coorelations
1	7.33 (m)	---
2		
3		
4	---	---
5	5.07 (m)	---
6	---	---
7	3.76 (ddd, 10.0, 7.7, 5.4)	H8, H12, NH
8	3.59 (td, 8.4, 2.3)	H7, H9, *H11
9	4.83 (ddd, 9.9, 7.8, 2.3)	H11
10	---	---
11	3.45 (dd, 16.8, 7.8)	*H8, H9
	3.20 (dd, 16.8, 9.9)	
12	2.07 (m)	H7, H13
13	0.93 (d, 6.8)	H12
	0.88 (d, 6.7)	
NH	4.99 (d, 10.2)	H7
OH	1.76 (s)	---

A multitude of different reaction conditions were attempted in order to prevent the formation of **56**, but ultimately it was determined to be the consequence of poor purification of enone **54** by the CombiFlash. Prior to these attempts, the enone had always been purified by manual column chromatography. Once the enone was repurified by manual column chromatography, the reaction proceeded with little to no formation of undesired product **56**. Due to the extra mass of undetected impurities from the CombiFlash purification, an excess of

bromonitromethane was used. It is suspected that this excess produced enough Br_2 to form dibromonitromethane *in situ* that could react through a carbene cyclopropanation mechanism rather than the nucleophilic cyclopropanation of Michael-acceptors (enones) (Figure 20).

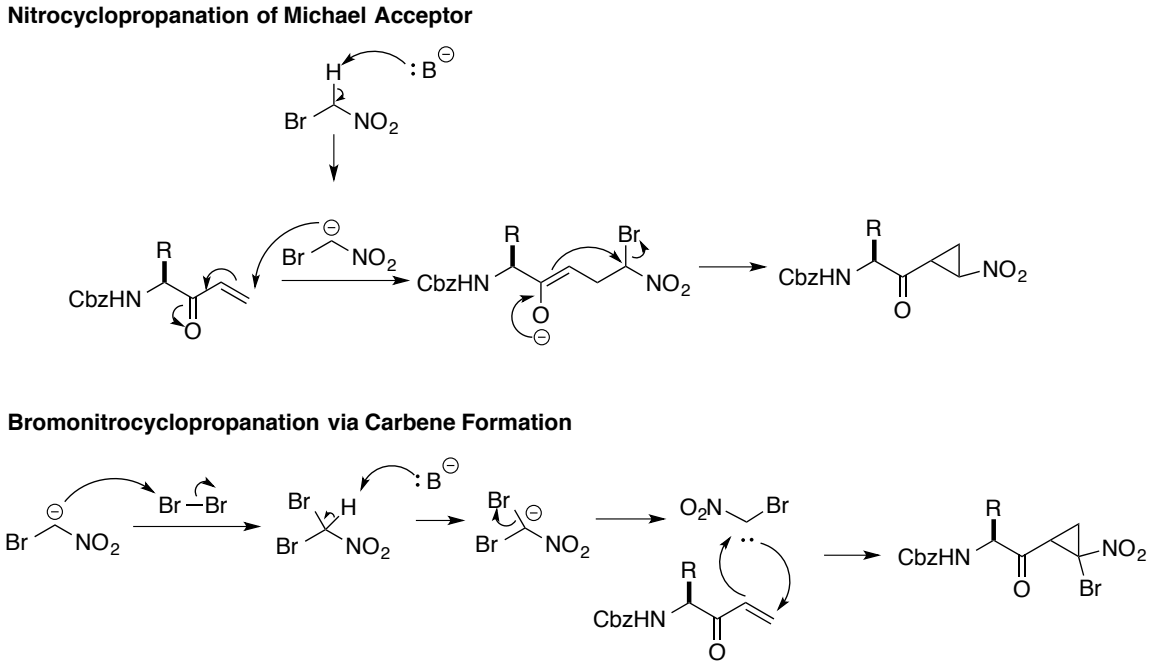


Figure 20: Mechanism of nitro- versus bromonitrocyclopropanation.

In the original publication of this reaction, Ballini noted that the bromonitromethane had to be added in several fractions, otherwise the yields would be "decreased substantially." Ballini proposed that this may be due to the bromonitromethane undergoing partial decomposition under basic conditions [34]. However, the reduced yield that Ballini observed may have actually been due to the side formation of the carbene when bromonitromethane is in excess.

Cyclopropyl amines

Functionalization of the cyclopropyl backbones for coupling proved to be one of the greater challenges of this project. To reduce the nitro to an amine, a reagent had to be chosen that could selectively reduce the nitro without opening the cyclopropyl ring or removing the Cbz-protecting group. Previous work in Dunlap's lab determined that transfer hydrogenation, while easily reducing the nitro, quickly removed the Cbz and was not an optimal reduction method. The only reduction method that seemed to work was Zn and HCl (Scheme 6). However, the amine product from this reaction could not be purified, making product verification quite challenging. The only verification came from comparing crude ^{1}H-NMR spectra of the product in question to that of the amine products that had successfully coupled to the β-lactam acid. The lack of consistent crude spectra and many failed or low yielding coupling attempts proved this reduction method to exhibit limited reproducibility and questionable yields. Further work is needed to optimize another reduction method as well as developing a purification method for the final amine product.

Cyclopropyl acids

Functionalization of the cyclopropyl ester to an acid via a base catalyzed hydrolysis (Scheme 6) presented some unanticipated complications. From the crude ^{1}H-NMR data of the products of the hydrolysis of the esters **15-17**, it was

determined that a side reaction was occurring that was eliminating a benzyl alcohol from the Cbz. It is suspected that the basic reaction conditions deprotonated the alcohol creating a strong nucleophile that could then attack the carbonyl of the Cbz to form a favored five-membered oxazolidinone ring (Figure 21).

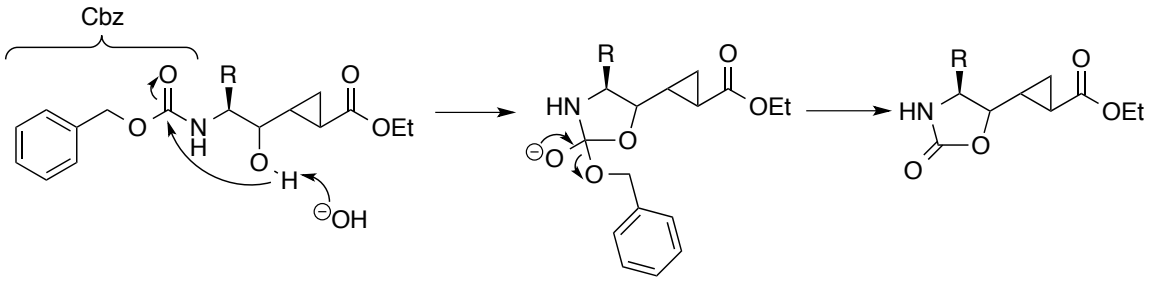


Figure 21: Proposed mechanism for undesired intramolecular cyclization.

To solve this issue, an alternative synthesis was developed to replace the Cbz carbamate with a benzyl amide that would not be prone to the same nucleophilic attack and intramolecular cyclization (Scheme 7). While this synthesis did not exhibit the best yields, particularly regarding the coupling of phenylacetic acid and ester hydrolysis, acids **39** and **41** were successfully obtained. However, the removal of the Cbz on leucine ester **16** by transfer hydrogenation was unsuccessful at both 45 min and 4 hr reaction times. The explanation is uncertain as to why this method removed the Cbz from **15** and **17**, but failed to remove it from **16**. Catalytic hydrogenation under high pressure on the Parr shaker will be used in future attempts to obtain amine **34** to complete the series.

Coupling of β-lactams to cyclopropyl backbones

Due to the many issues encountered in functionalizing the cyclopropyl backbones, only analogs **31** and **32** were successfully synthesized, but with a yield of only 27% for the two steps. Because of the inability to purify the amines **27-29** it is still unclear whether the low yield was due to the reduction or the coupling step. After performing the model coupling of β-lactam acid **4** and 2-aminopropane (Scheme 8), it was found that product **45** was afforded in a 63% yield. While this does not necessarily prove that the reduction is the primary step affecting the yield, it does prove that it is possible to couple β-lactam **4** to amines in relatively high yields. If the reduction is not the issue, then the yield of the coupling could also be hindered by the alcohol of **27-29** competing with the amine as a nucleophile capable of attacking the activated acid.

Regrettably, the coupling of acid **41** with β-lactam **14** failed and acid **41** was unable to be recovered. Due to limited time and resources, a model system was also developed to determine if β-lactam **14** could be successfully coupled to phenylacetic acid. When this reaction also failed to afford **47**, it was determined that the ammonium salt, though theoretically more stable, was not reacting as it should. Upon further investigation into the literature, a procedure for coupling β-lactam **46** to an acid activated with *N*-hydroxysuccinimide and *N,N'*-dicyclohexylcarbodiimide (DCC) was uncovered. In this procedure, the filtrate containing β-lactam **46**

following catalytic hydrogenation was directly added to a solution containing the activated acid and reacted on a rotary evaporator where the reaction mixture was allowed to slowly concentrate to the appropriate volume. Once the reaction mixture was reduced to a third of its original volume, it was removed from vacuum and stirred at 35˚C for 18 hours [35]. The key aspects of this procedure reveal a likely instability of the β-lactam amine requiring that it be reacted immediately after formation. It also revealed that an additional boost in acid activation may be required, which was the function of *N*-hydroxysuccinimide in the procedure.

Based on these findings, an alternative synthesis of the model-coupling product **47** was attempted. This procedure utilized HOBt to aid EDCI in acid activation, which operates through the same mechanism as *N*-hydroxysuccinimide, and amine **46** was added immediately following the work-up of the catalytic hydrogenation. Though product **47** was successfully attained by this method providing proof of concept, the yield was only 5.5%. Modifications to the reaction conditions will need to be explored to increase this yield before another attempt is made to couple the β-lactam amine to one of the cyclopropyl acids.

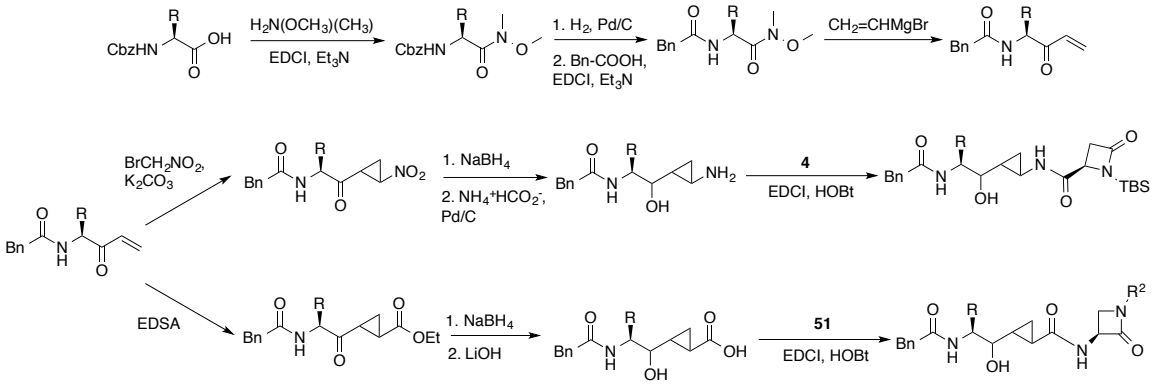
Conclusions

Belactosin A is a β-lactone natural product that has been shown to halt tumor growth via proteasome inhibition. Several analogs of its structure have previously been explored in attempts to improve efficacy and reduce toxicity, but none of these

analogs utilized a β-lactam as a serine trap. The goal of this project was to synthesize novel β-lactam analogs of belactosin A based on the cyclopropyl backbones previously developed by Dunlap.

While only two analogs were successfully obtained, the many issues encountered in this synthesis have been illuminating. Due to the necessity of switching the Cbz-protecting group for a benzyl amide in order to achieve acids **39-41**, a new avenue for the total synthetic design of this project can now be explored. Due to the low yields observed with coupling phenylacetic acid to amines that also contain alcohols in close proximity, it is worth trying to couple the phenylacetic acid prior to the reduction of the ketone to the alcohol. To accomplish this, the Cbz would need to be removed from the Weinreb amide. This is because the Weinreb amide is much less susceptible to the nucleophilic attack of free amines compared to the ketone. Once the benzyl amide cyclopropyl alcohols are obtained, they can be functionalized to the respective amine and acid. With the benzyl amide in place, it will be worth retrying transfer hydrogenation to reduce the nitro to an amine. When this was attempted on the Cbz-protected nitrocyclopropyls before, the nitro was reduced, but the Cbz was also removed. With the benzyl amides, the transfer hydrogenation should only reduce the nitro, eliminating the previous issue. Hopefully, this switch will allow for higher yields to be obtained. The proposal for this new synthetic strategy is shown in Scheme 14.

Scheme 14: Proposed alternative synthetic strategy for belactosin A analogs.



The incorporation of the benzyl amide also creates the opportunity to explore other bulky hydrophobic groups at that position. Two enones that would be especially interesting and useful to try to bring to cyclopropyl amines and acids are shown in Figure 22. The phenylethylamide provides an additional carbon of length, making it more comparable to the Cbz. The benzyl amide alanine would mimic the alanine residue seen in belactosin-A, making it even more comparable to Shuto's analogs.

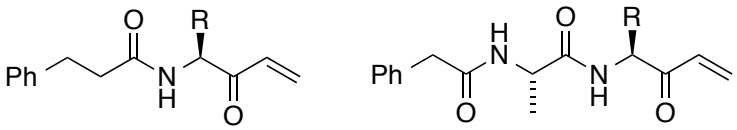


Figure 22: Additional enones that could be explored to expand on the belactosin A analogs.

As shown in Scheme 14 and Figure 22, there are many areas where the synthesis of β-lactam belactosin A analogs can be expanded. Once the model coupling procedures are fully optimized, they can be applied to the cyclopropyl amines and acids, which by this new synthetic strategy, may be able to be afforded in higher yields. When more analogs are obtained, they can be assayed for proteasome inhibition to identify those exhibiting the greatest activity. The most promising analogs can then be further studied to identify which stereoisomers are the most active.

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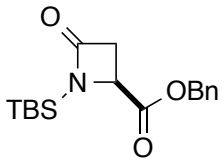
APPENDICES

APPENDIX A: NMR DATA

(4*S*)-Benzyl *N*-(*t*-butyldimethylsilyl)azetidin-2-one-4-carboxylate (3)

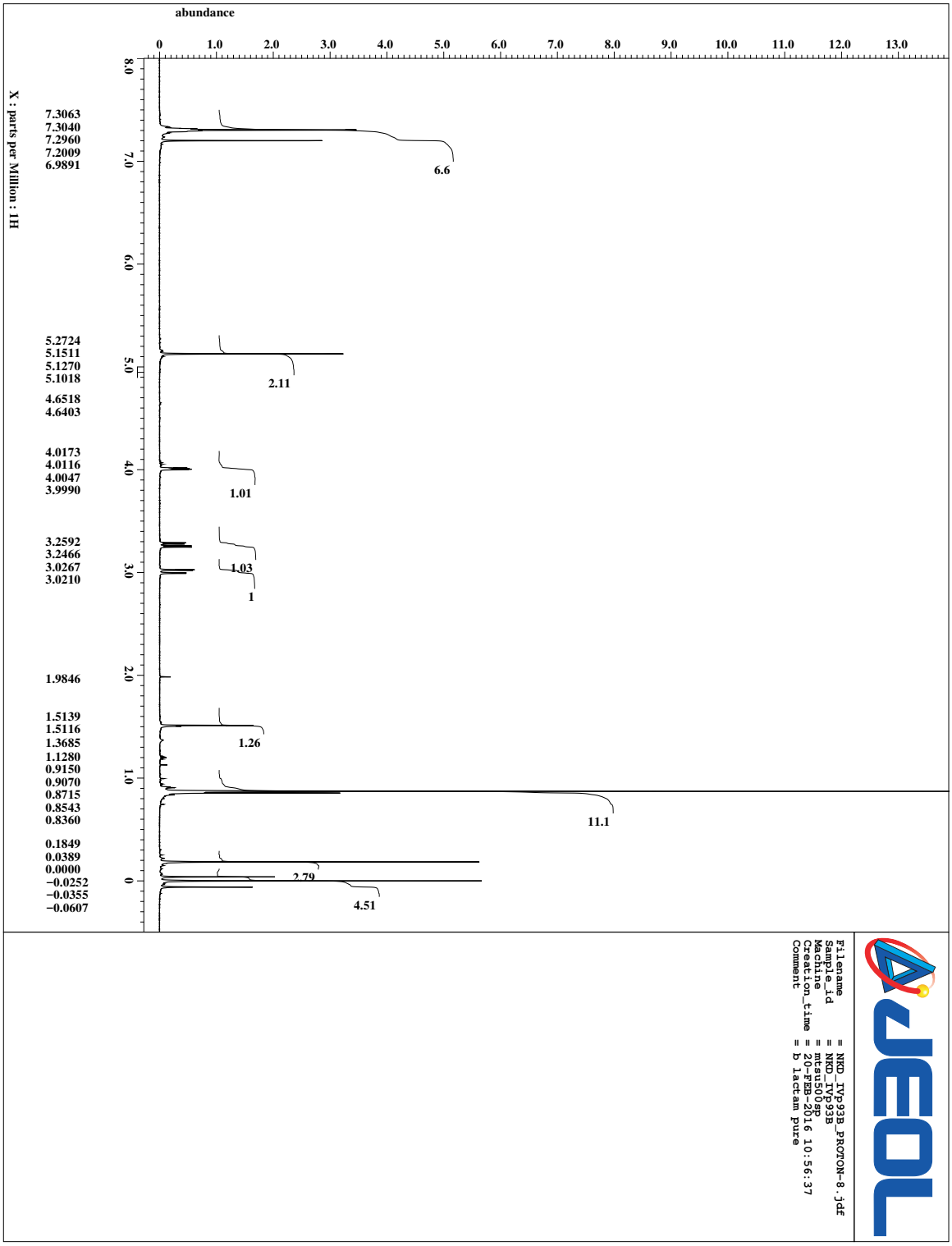


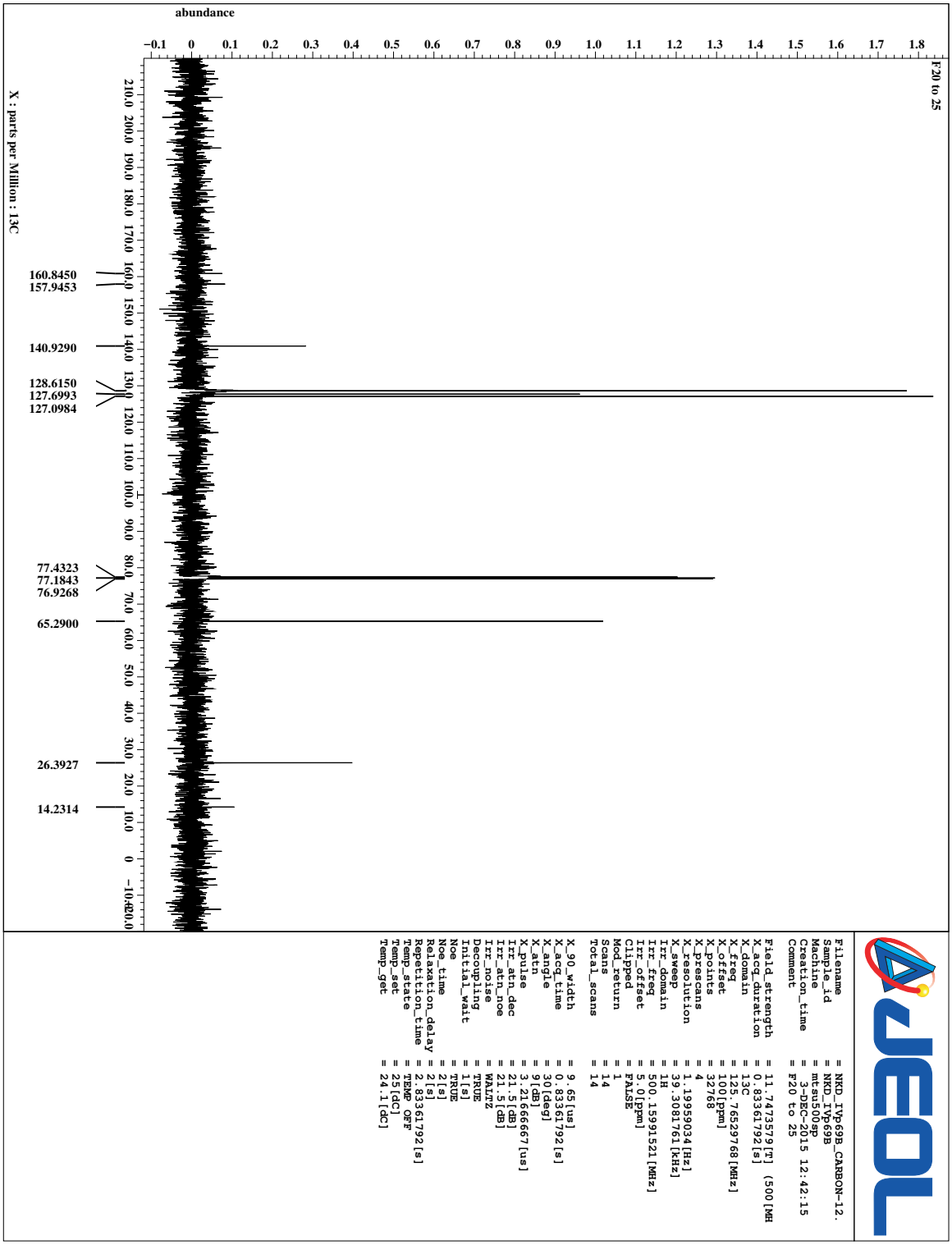
NMR

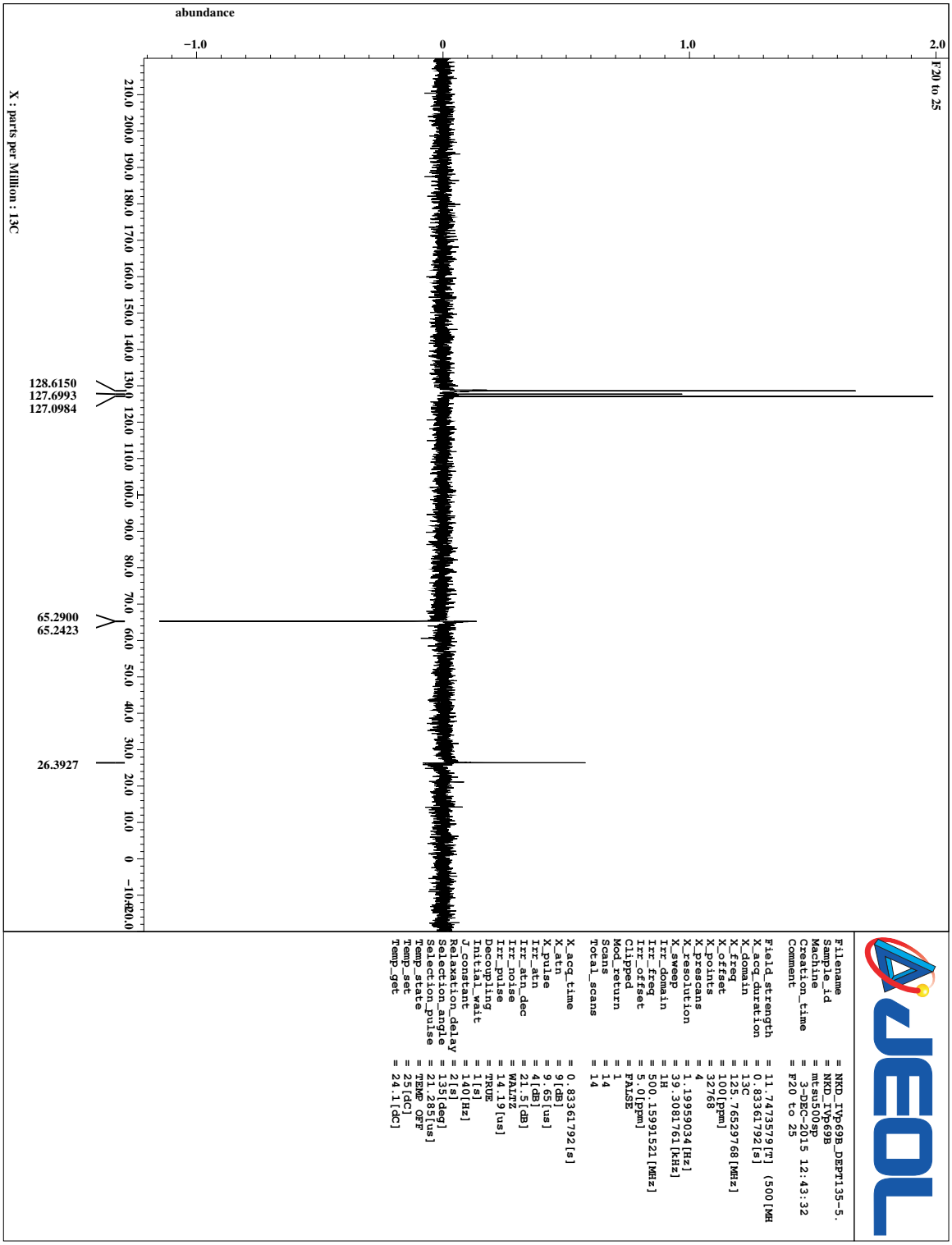
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

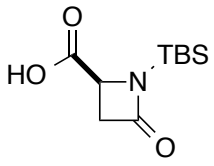
$DEPT_{135}$ ($CDCl_3$)


abundance
X : parts per Million : 1H
7.3063
7.3040
7.2960
7.2009
6.9891
5.2724
5.1511
5.1270
5.1018
4.6518
4.6403
4.0173
4.0116
4.0047
3.9990
3.2592
3.2466
3.0267
3.0210
1.9846
1.5139
1.5116
1.3685
1.1280
0.9150
0.9070
0.8715
0.8543
0.8360
0.1849
0.0389
0.0000
−0.0252
−0.0355
−0.0607
6.6
2.11
1.01
1.03
1
1.26
11.1
2.79
4.51
Filename = NKD_IVp93B_PROTON-8.jdf
Sample_id = NKD_IVp93B
Machine = mtsu500sp
Creation_time = 20-FEB-2016 10:56:37
Comment = b lactam pure
JEOL


abundance
F20 to 25
X : parts per Million : 13C
160.8450
157.9453
140.9290
128.6150
127.6993
127.0984
77.4323
77.1843
76.9268
65.2900
26.3927
14.2314
Filename = NMD_IVp69B_CARBON-12.
Sample_id = NMD_IVp69B
Machine = mt-gus500mp
Creation_time = 3-DEC-2015 12:42:15
Comment = F20 to 25
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.3633792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_90_width = 9.65[us]
X_acq_time = 0.8336792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.21666667[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.8336792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 24.1[dC]


abundance
−1.0
0
1.0
2.0
F20 to 25
X : parts per Million : 13C
128.6150
127.6993
127.0984
65.2900
65.2423
26.3927
Filename = NKD_IVp69B_DEPT135-5.
Sample_id = NKD_IVp69B
Machine = mt-su500ap
Creation_time = 3-DEC-2015 12:43:32
Comment = F20 to 25
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.63361792[s]
X_domain = 1H
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 9.65[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 24.1[dC]

(4*S*)-*N*-(*t*-Butyldimethylsilyl)azetidin-2-one-4-carboxylic acid (4)

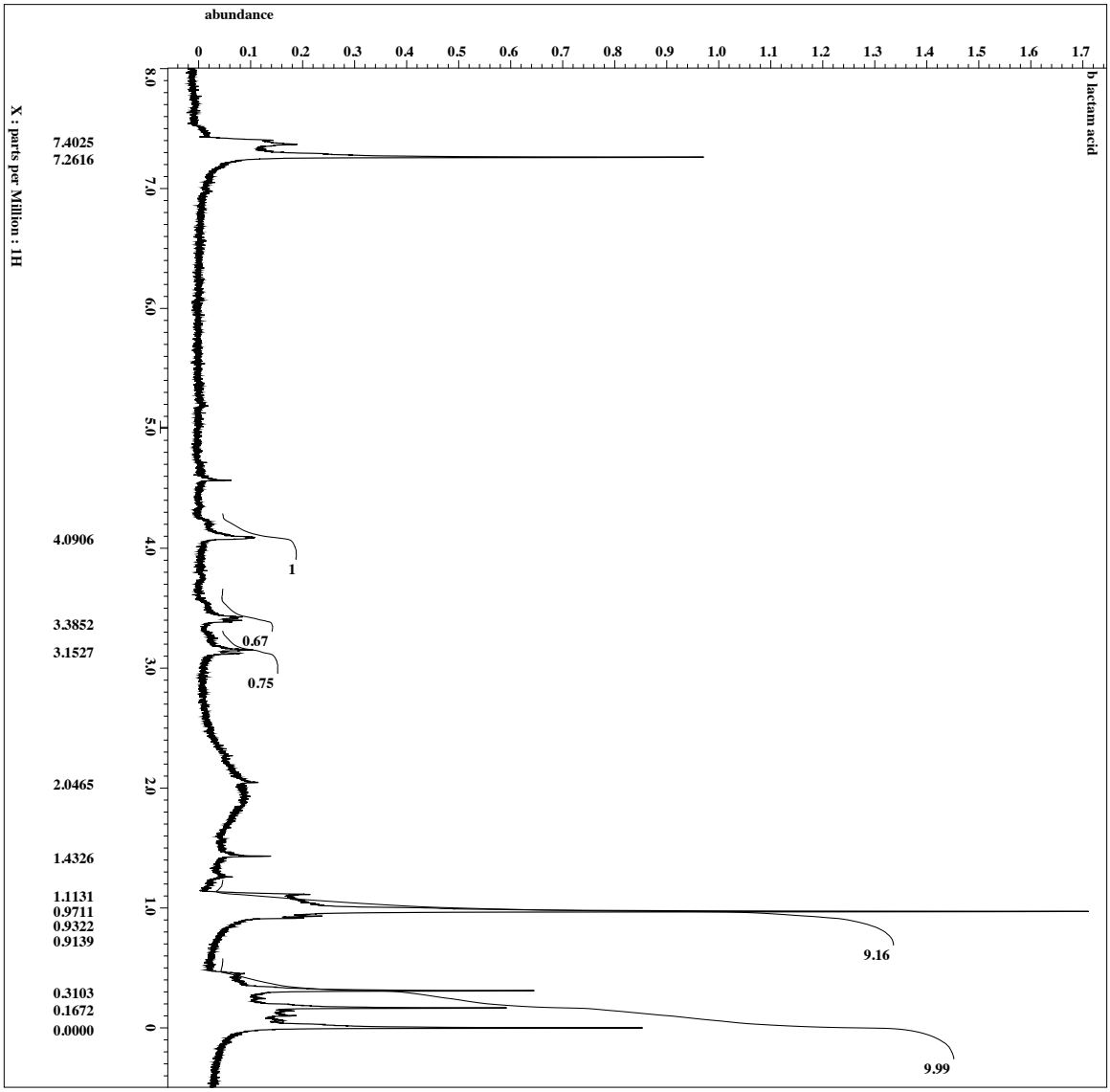


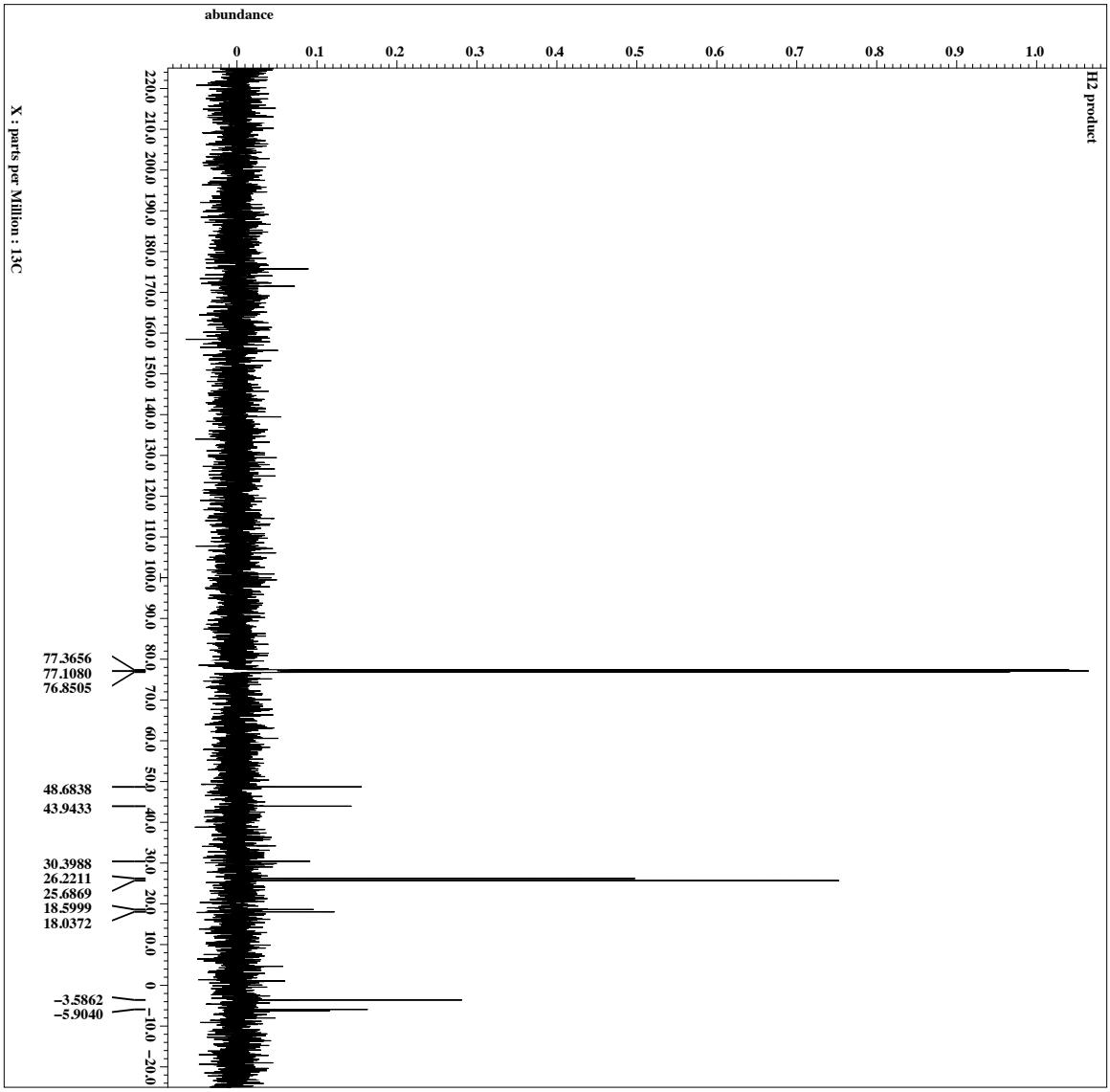
NMR

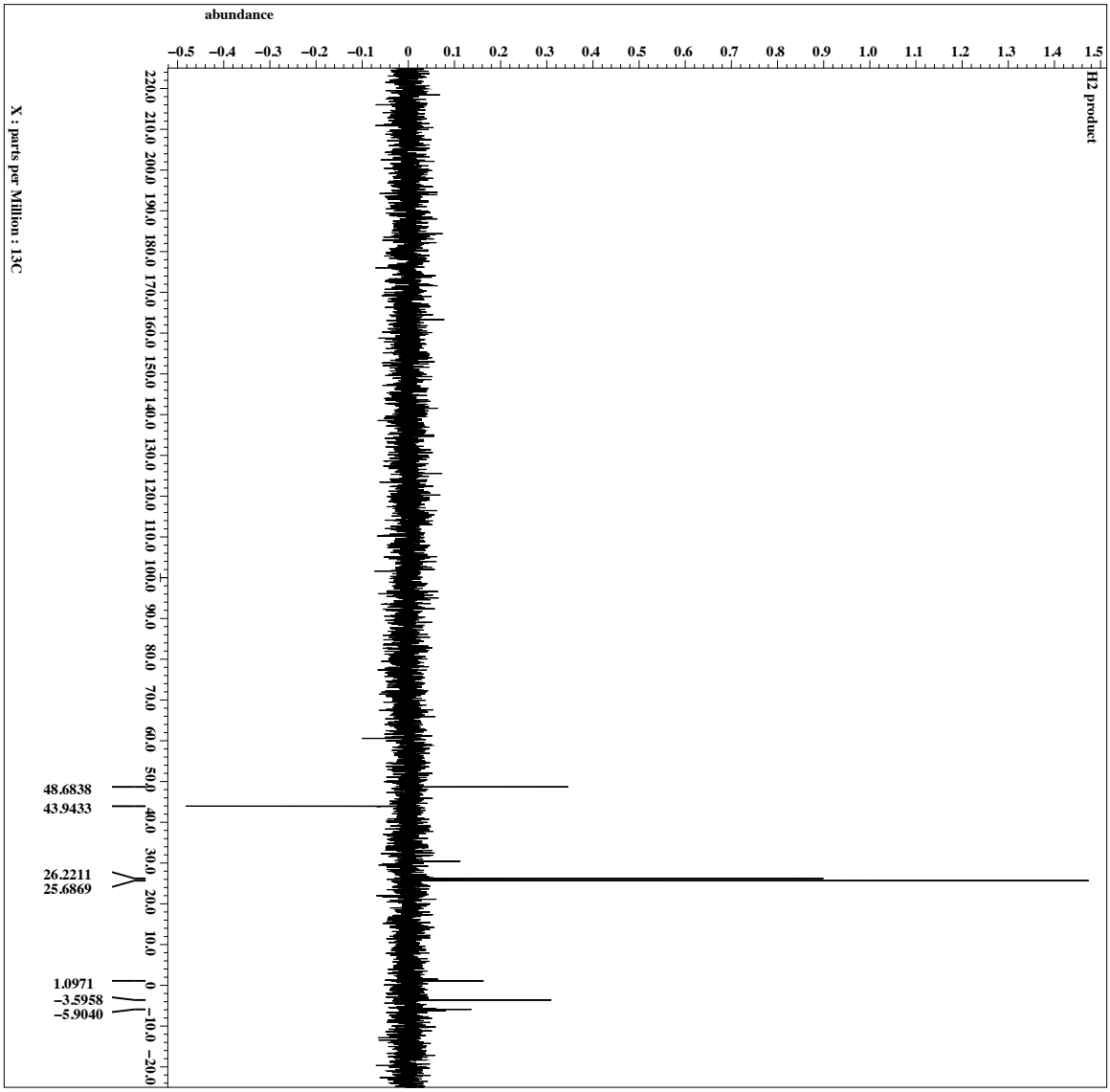
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

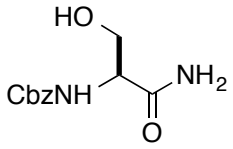
$DEPT_{135}$ ($CDCl_3$)


b lactam acid
abundance
X : parts per Million : 1H
7.4025
7.2616
4.0906
3.3852
3.1527
2.0465
1.4326
1.1131
0.9711
0.9322
0.9139
0.3103
0.1672
0.0000
1
0.67
0.75
9.16
9.99
Filename = beta_lactam acid-3.jd
Sample_id = NKD_typ89
Machine = mt-eu500pp
Creation_time = 23-FEB-2016 11:00:27
Comment = b lactam acid
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dc]
Temp_get = 23[dc]


H2 product
abundance
X : parts per Million : 13C
77.3656
77.1080
76.8505
48.6838
43.9433
30.3988
26.2211
25.6869
18.5999
18.0372
−3.5862
−5.9040
Filename = NKD_ivp73cr_CARBON-7.
Sample_id = NKD_ivp73cr
Machine = mt-su500#2
Creation_time = 9-DEC-2015 09:31:30
Comment = H2 product
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_90_width = 9.65[us]
X_acq_time = 0.83361792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.21666667[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.83361792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 24.1[dC]


H2 product
abundance
X : parts per Million : 13C
48.6838
43.9433
26.2211
25.6869
1.0971
−3.5958
−5.9040
Filename = NKD_IVp73cr_DEPT135-5
Sample_id = NKD_IVp73cr
Machine = mt-eu500#2
Creation_time = 9-DEC-2015 09:32:49
Comment = H2 product
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 9.65[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 24.1[dC]

_N_-benzyloxycarbonyl-L-serine amide (6)

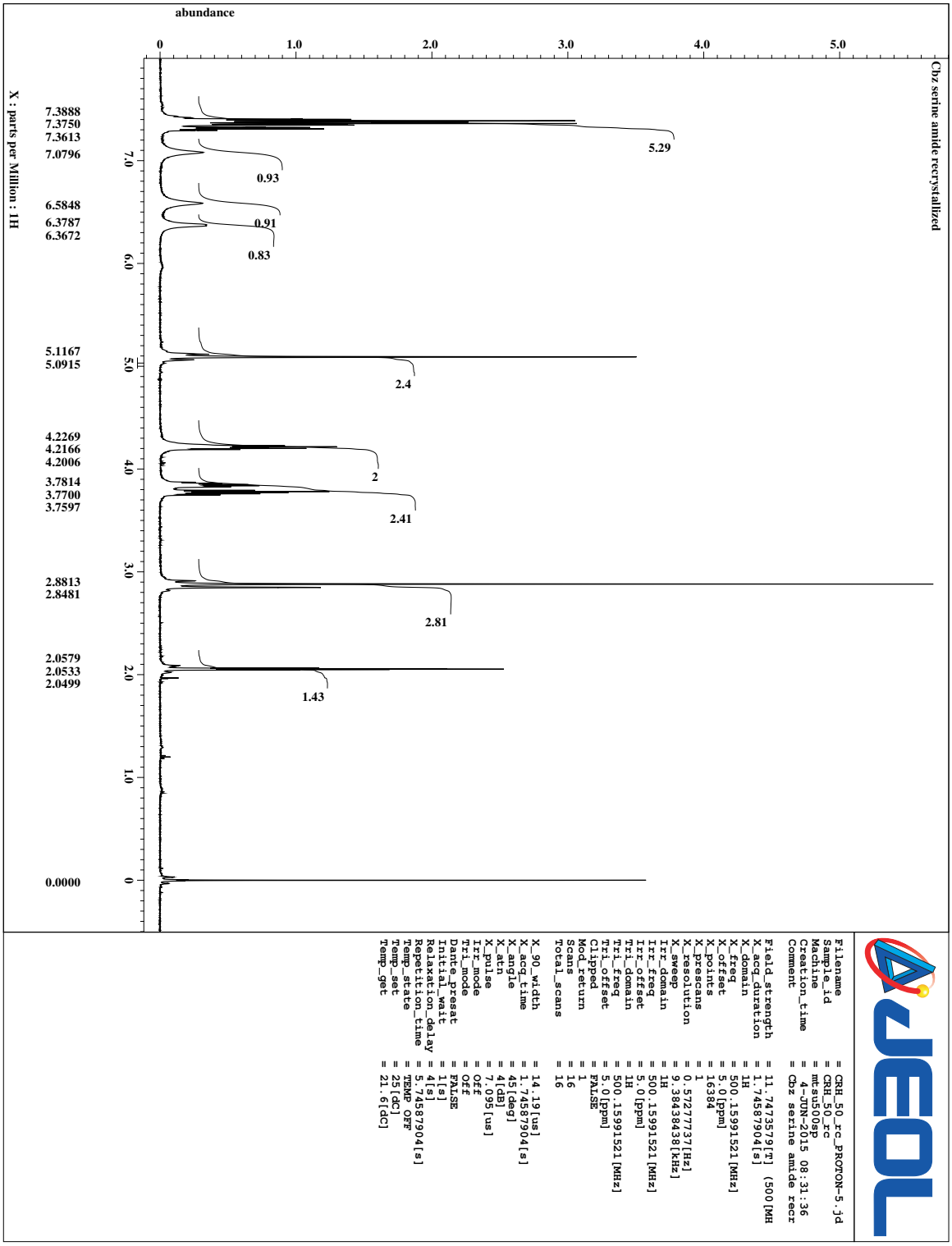


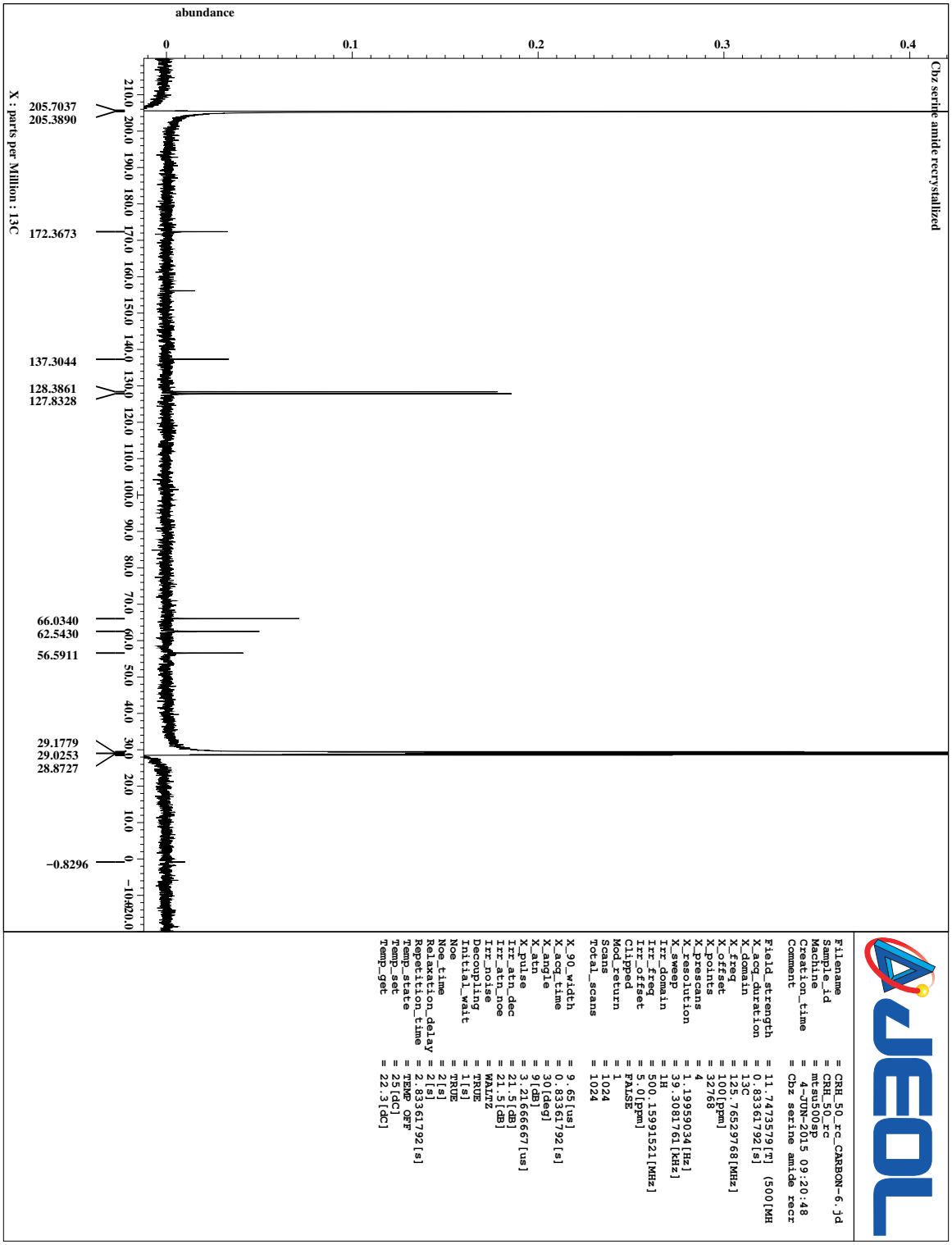
NMR

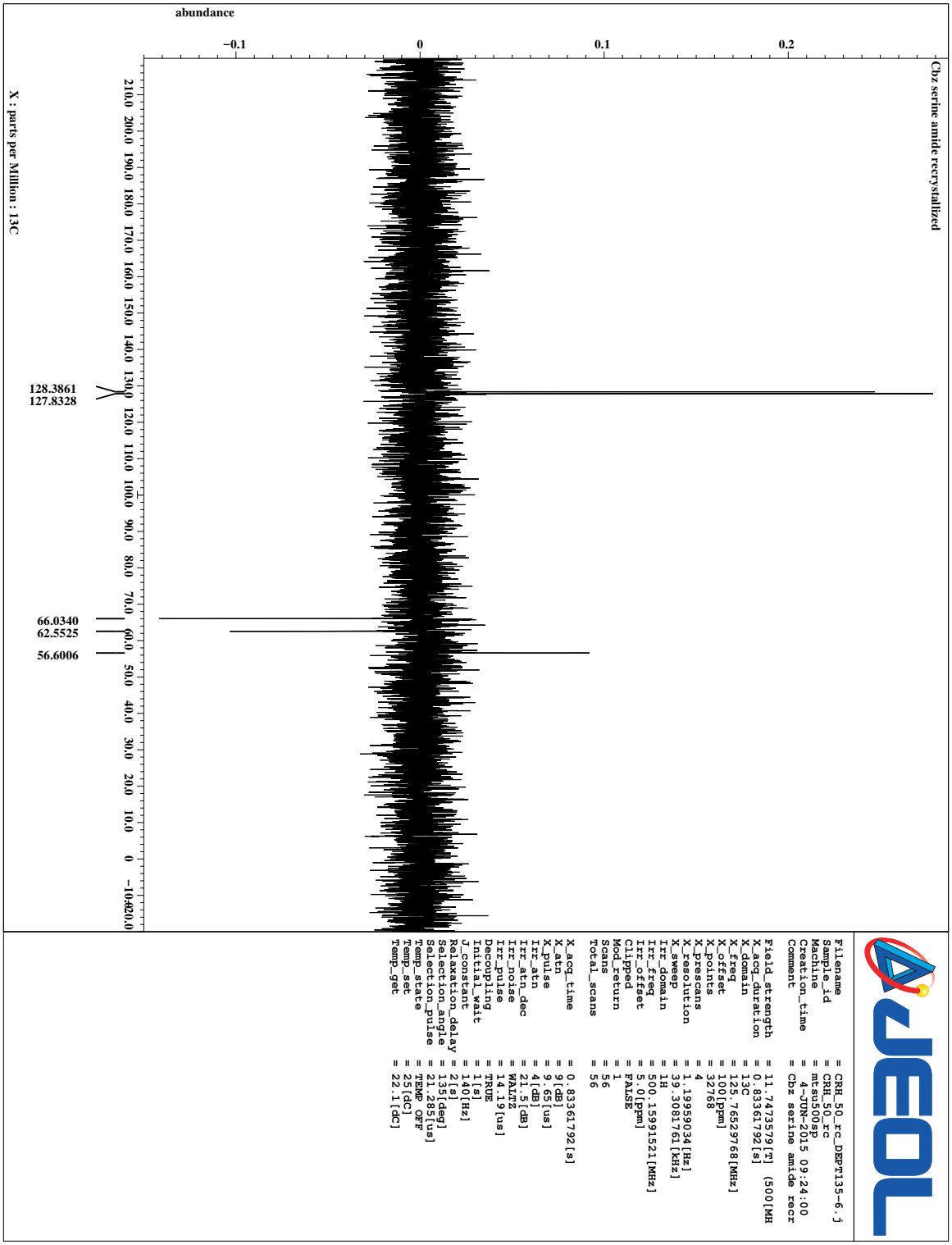
^{1}H-NMR (Acetone-d_6)

^{13}C-NMR (Acetone-d_6)

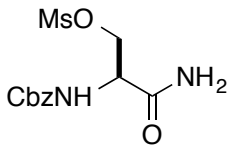
DEPT$_{135}$ (Acetone-d_6)


Cbz serine amide recrystallized
abundance
X : parts per Million : 1H
7.3888
7.3750
7.3613
7.0796
6.5848
6.3787
6.3672
5.1167
5.0915
4.2269
4.2166
4.2006
3.7814
3.7700
3.7597
2.8813
2.8481
2.0579
2.0533
2.0499
0.0000
5.29
0.93
0.91
0.83
2.4
2
2.41
2.81
1.43
Filename = CRH_50_rc_PROTON-5.jd
Sample_id = CRH_50_rc
Machine = mr-us500pp
Creation_time = 4-JUN-2015 08:31:36
Comment = Cbz serine amide recr
Field_strength = 11.747579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.3843843[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.6[dC]


Cbz serine amide recrystallized
abundance
0
0.1
0.2
0.3
0.4
X : parts per Million : 13C
205.7037
205.3890
172.3673
137.3044
128.3861
127.8328
66.0340
62.5430
56.5911
29.1779
29.0253
28.8727
−0.8296
Filename = CRH_50_rc_CARBON-6.jd
Sample_id = CRH_50_rc
Machine = mt-su500ap
Creation_time = 4-JUN-2015 09:20:48
Comment = Cbz serine amide recr
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.8336179[s]
X_domain = 13C
X_freq = 125.7652976[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.1599152[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 1024
Total_scans = 1024
X_90_width = 9.65[us]
X_acq_time = 0.8336179[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.216666667[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.833617792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.3[dC]


Cbz serine amide recrystallized
abundance
−0.1
0
0.1
0.2
128.3861
127.8328
66.0340
62.5525
56.6006
X : parts per Million : 13C
210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0 −10.0 −20.0
JEOL
Filename = CRH_50_rc_DEPT135-6.j
Sample_id = CRH_50_rc
Machine = mt-su500ap
Creation_time = 4-JUN-2015 09:24:00
Comment = Cbz serine amide recr
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 56
Total_scans = 56
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 9.65[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.1[dC]

_N_-benzyloxycarbonyl-L-serine(O-mesyl) Amide (7)



NMR

^{1}H-NMR (Acetone-d_6)

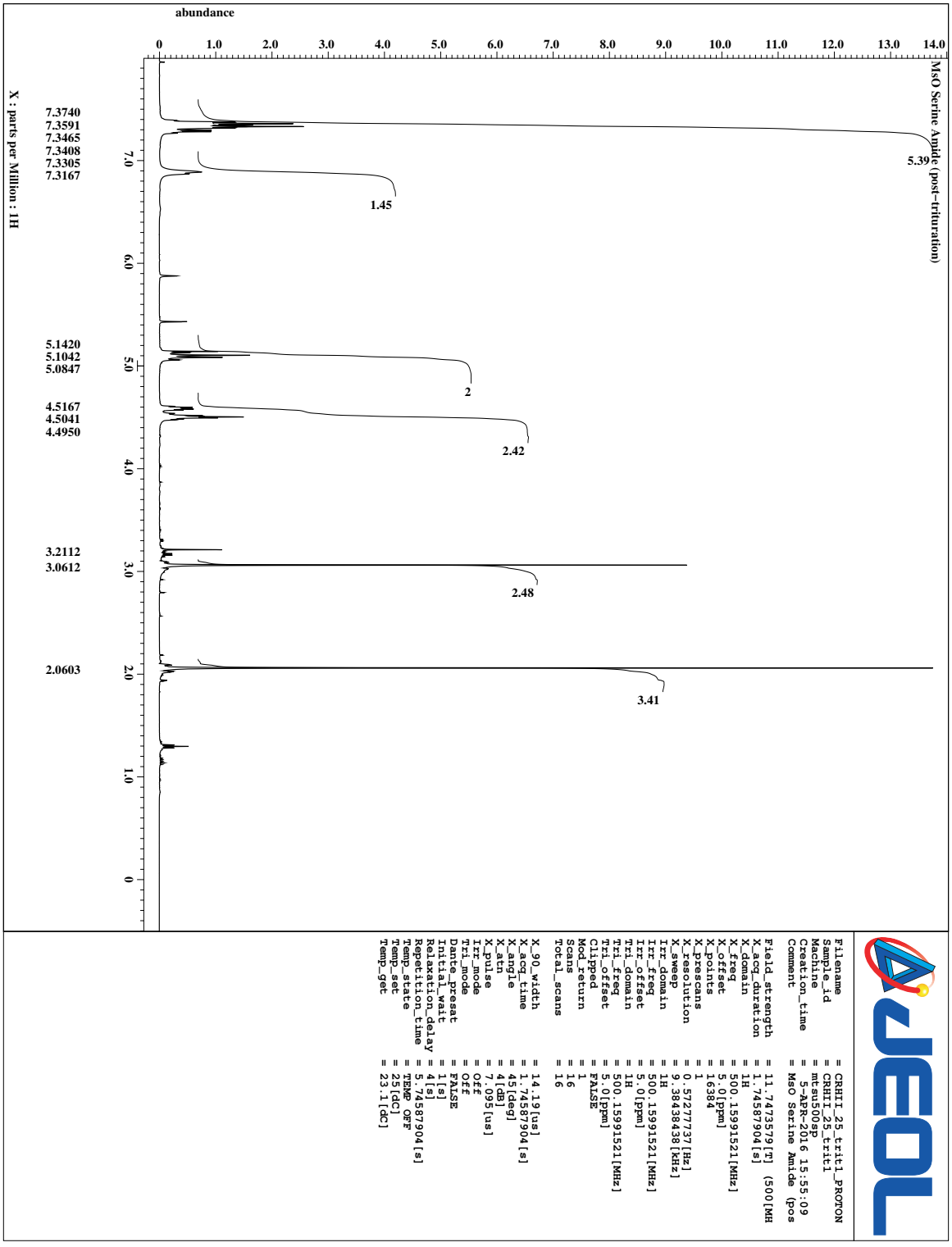
^{13}C-NMR (Acetone-d_6)

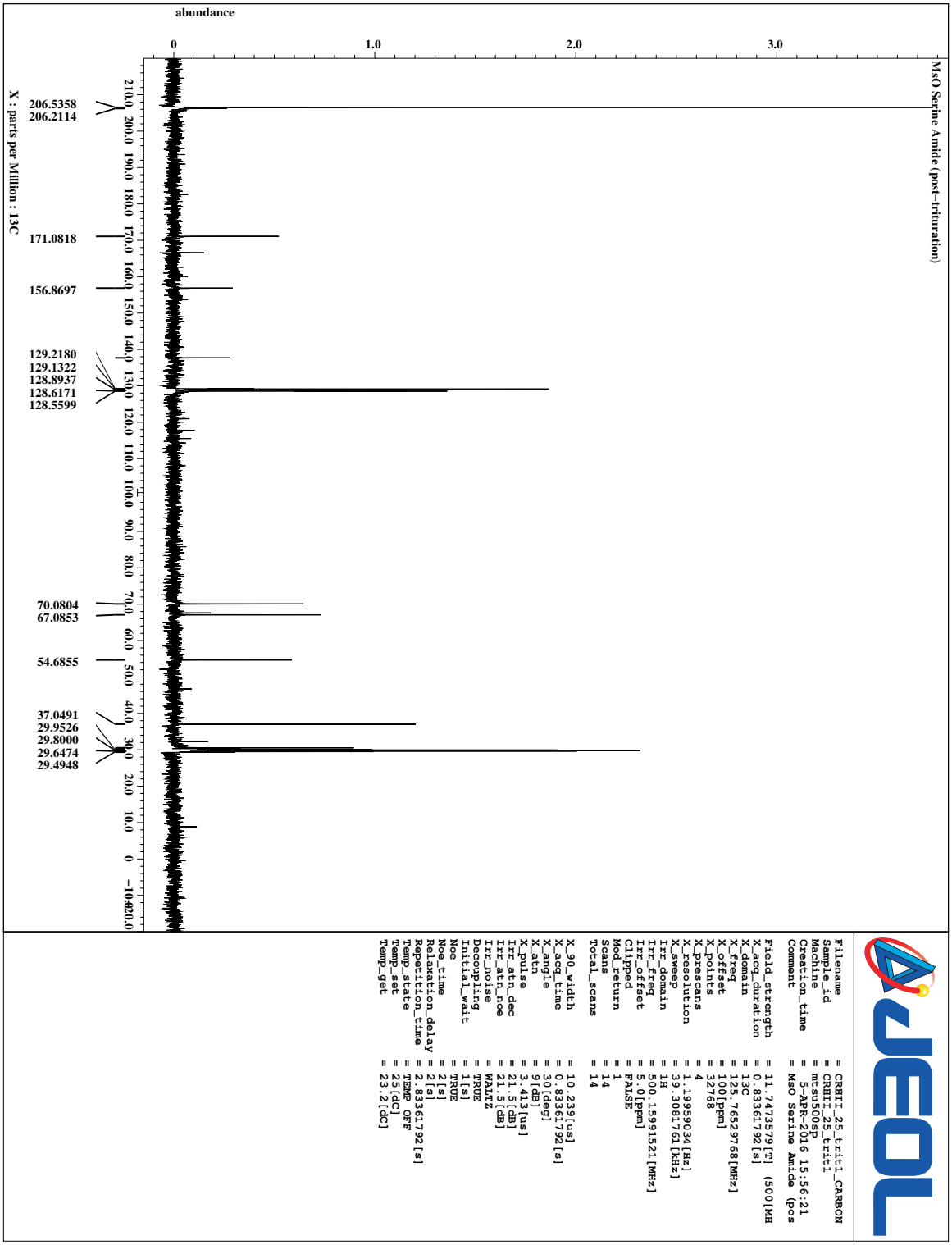
DEPT$_{135}$ (Acetone-d_6)

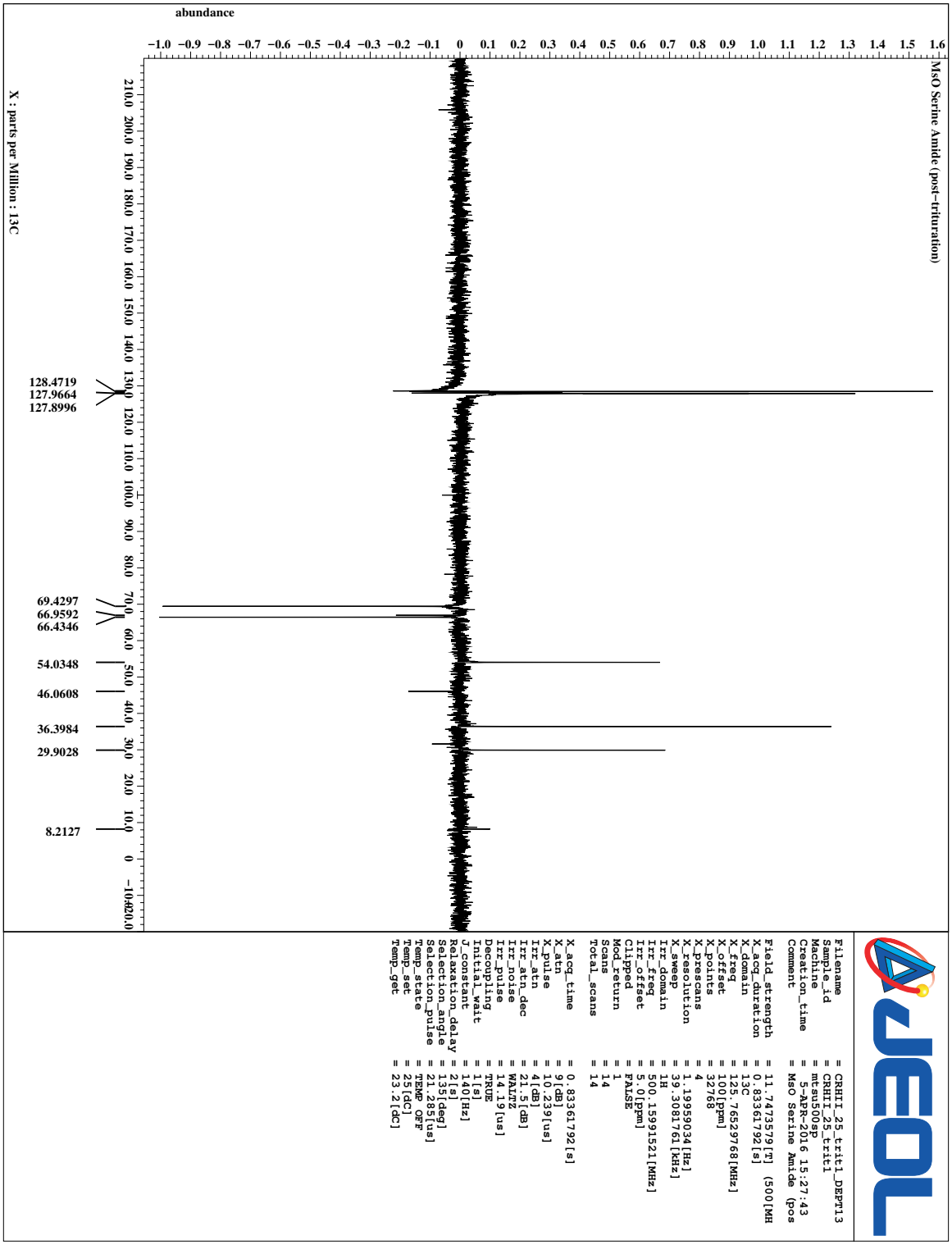
COSY (Acetone-d_6)

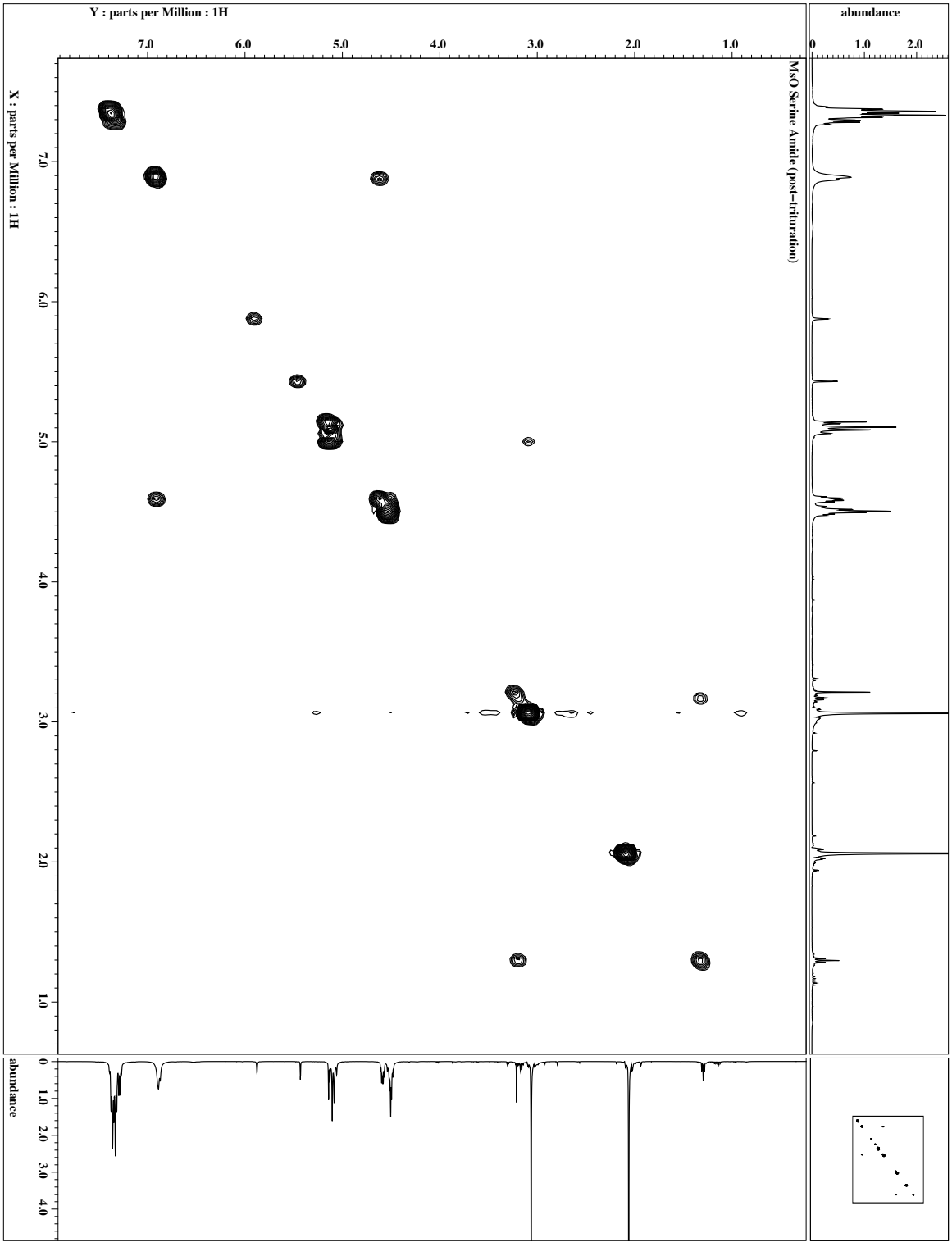
HMQC (Acetone-d_6)

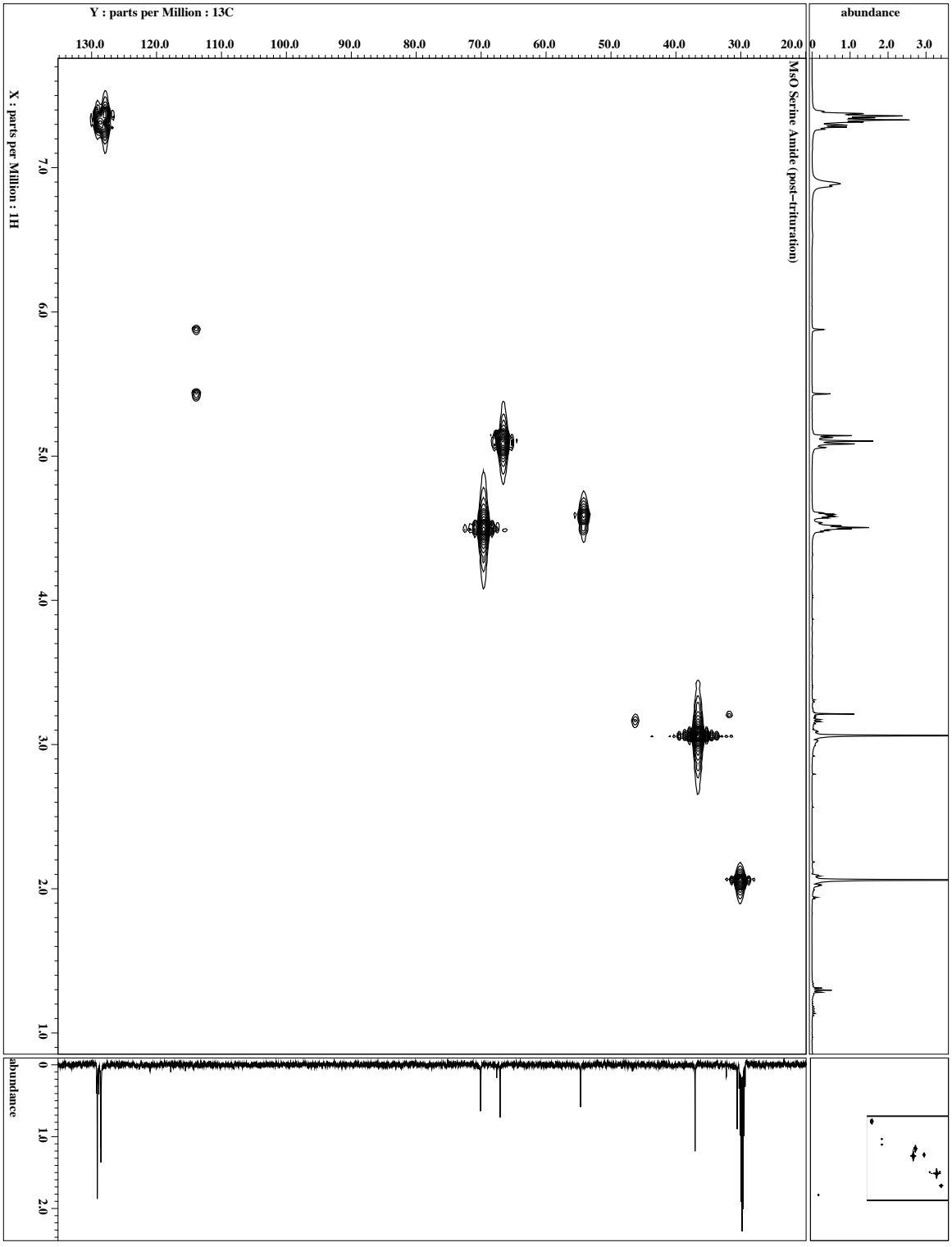
HMBC (Acetone-d_6)

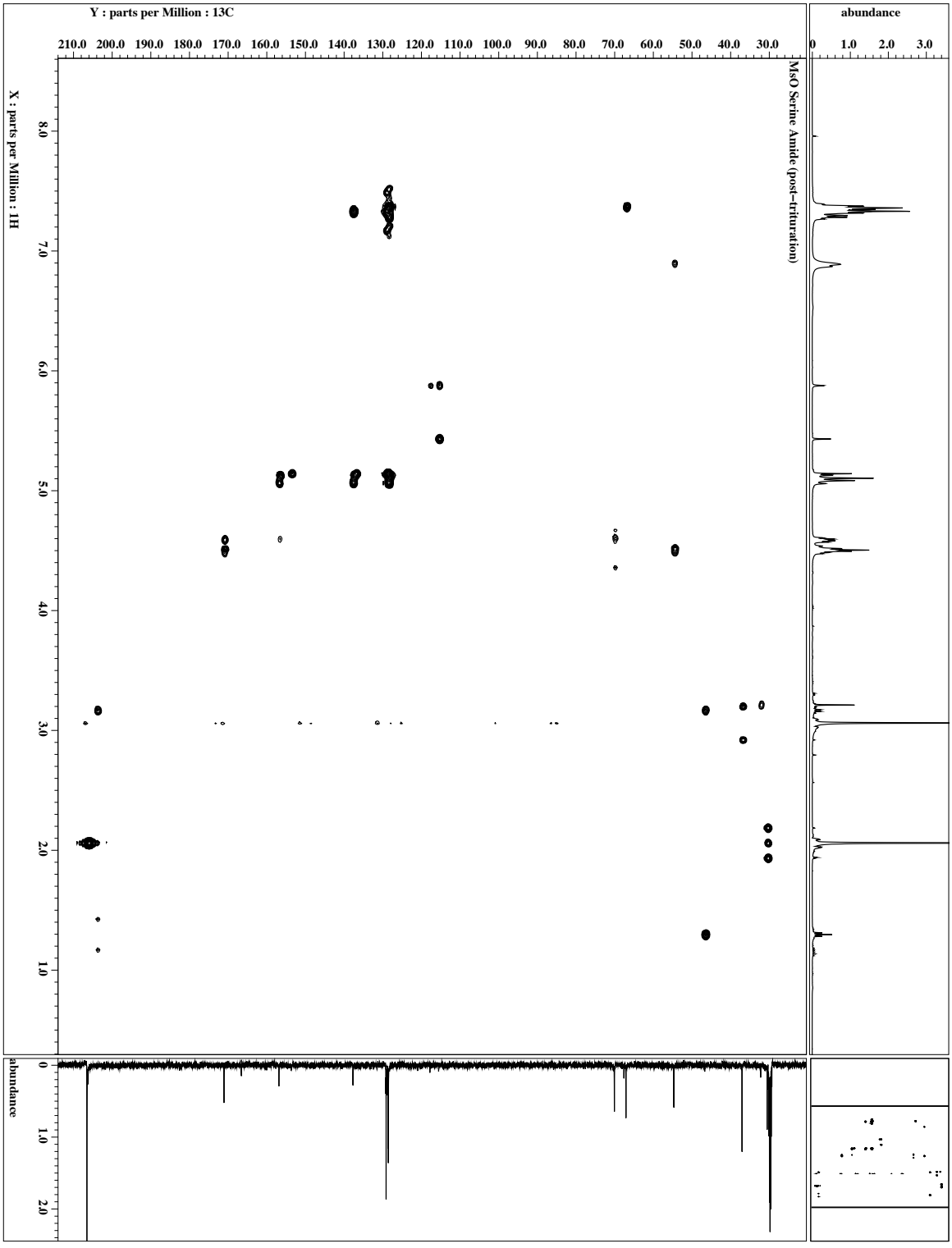

abundance
MsO Serine Azide (post-trituration)
X : parts per Million : 1H
7.3740
7.3591
7.3465
7.3408
7.3305
7.3167
5.1420
5.1042
5.0847
4.5167
4.5041
4.4950
3.2112
3.0612
2.0603
5.39
1.45
2
2.42
2.48
3.41
Filename = CRHI1_25_trit1_PROTON
Sample_id = CRHI1_25_trit1
Machine = mt-su500asp
Creation_time = 5-APR-2016 15:55:09
Comment = MsO Serine Amide (pos
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.1[dC]


MsO Serine Amide (post-trituration)
abundance
0
1.0
2.0
3.0
X : parts per Million : 13C
206.5358
206.2114
171.0818
156.8697
129.2180
129.1322
128.8937
128.6171
128.5599
70.0804
67.0853
54.6855
37.0491
29.9526
29.8000
29.6474
29.4948
JEOL
Filename = CRHI1_25_trit1_CARBON
Sample_id = CRHI1_25_trit1
Machine = mt-su500asp
Creation_time = 5-APR-2016 15:56:21
Comment = MsO Serine Amide (pos
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.8336179[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_90_width = 10.239[us]
X_acq_time = 0.8336179[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.8336179[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.2[dC]

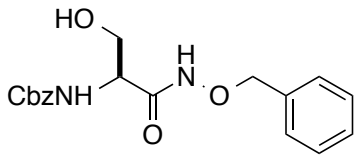

MsO Serine Amide (post-trituration)
abundance
X : parts per Million : 13C
128.4719
127.9664
127.8996
69.4297
66.9592
66.4346
54.0348
46.0608
36.3984
29.9028
8.2127
Filename = CRHI1_25_trit1_DEPT13
Sample_id = CRHI1_25_trit1
Machine = mt-su500sp
Creation_time = 5-APR-2016 15:27:43
Comment = MsO Serine Amide (pos
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.2[dC]


Y : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
abundance
0
1.0
2.0
MsO Serine Amide (post-trituration)
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
abundance
0
1.0
2.0
3.0
4.0


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
MsO Serine Amide (post-trituration)
abundance
0
1.0
2.0
3.0
abundance
0
1.0
2.0


Y : parts per Million : 13C
X : parts per Million : 1H
MsO Serine Amide (post-trituration)
abundance
abundance

_O_-Benzyl-α-_N_-Cbz-L-serine hydroxamate (9)



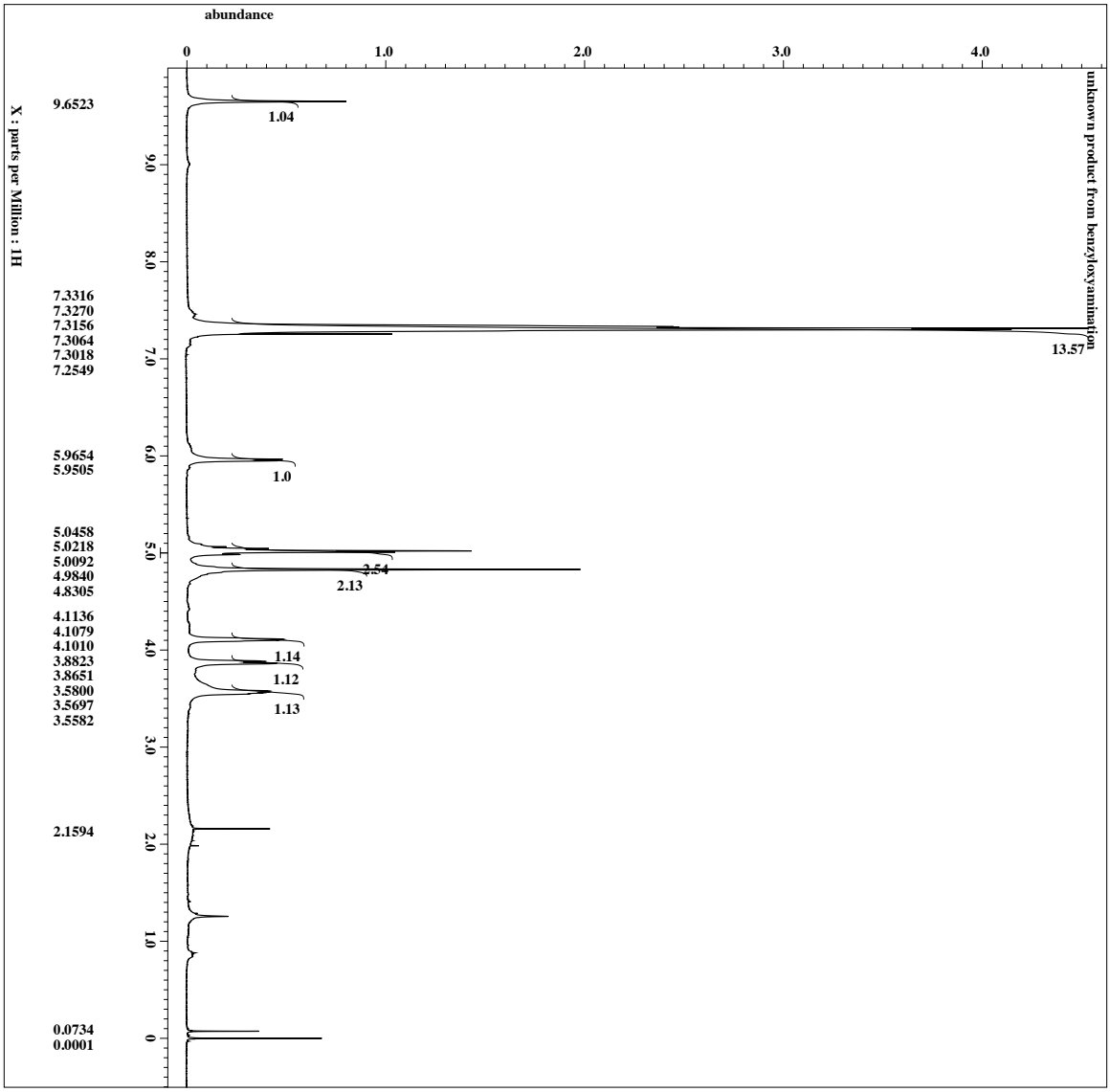
NMR

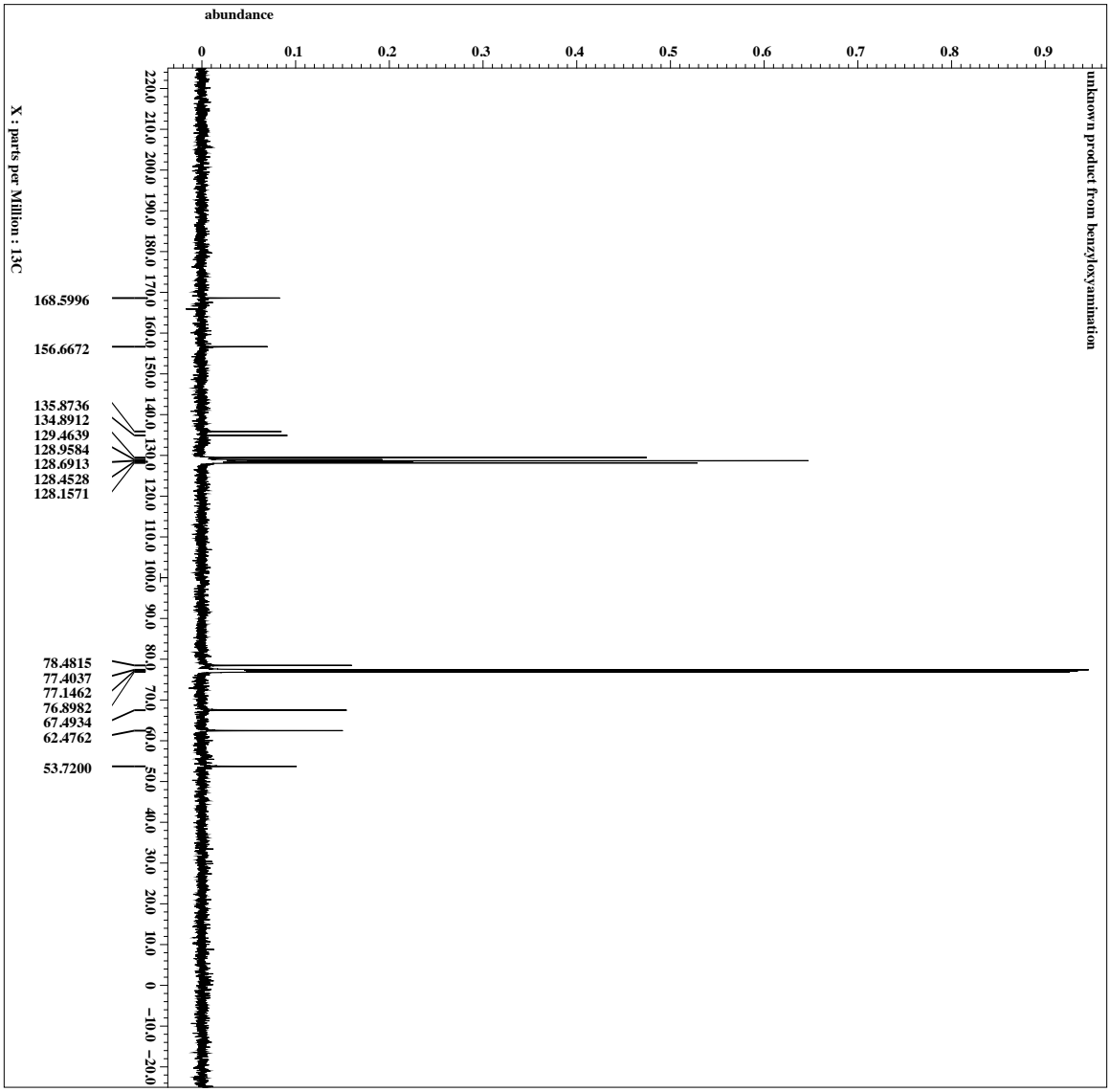
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

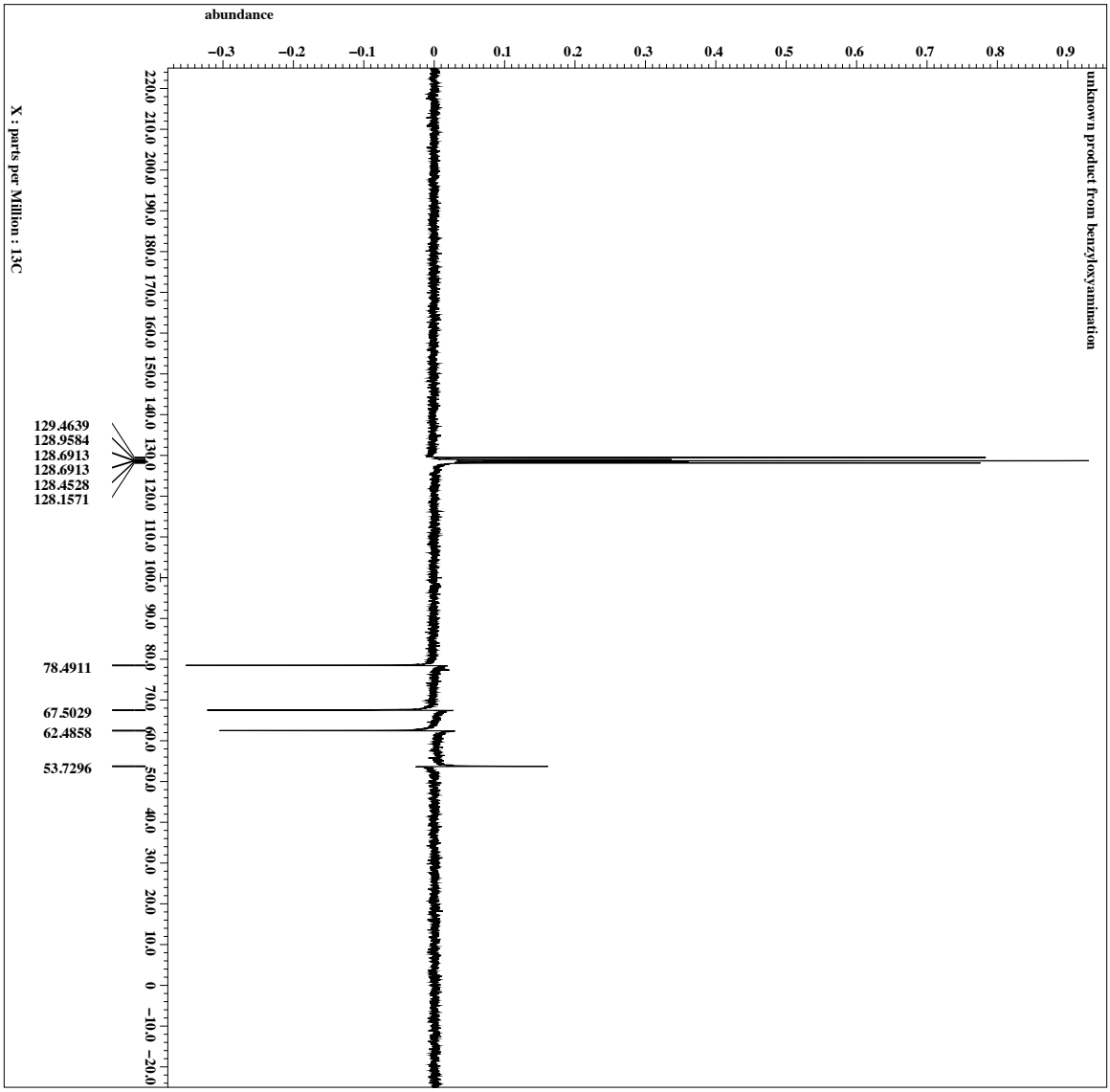
$DEPT_{135}$ ($CDCl_3$)

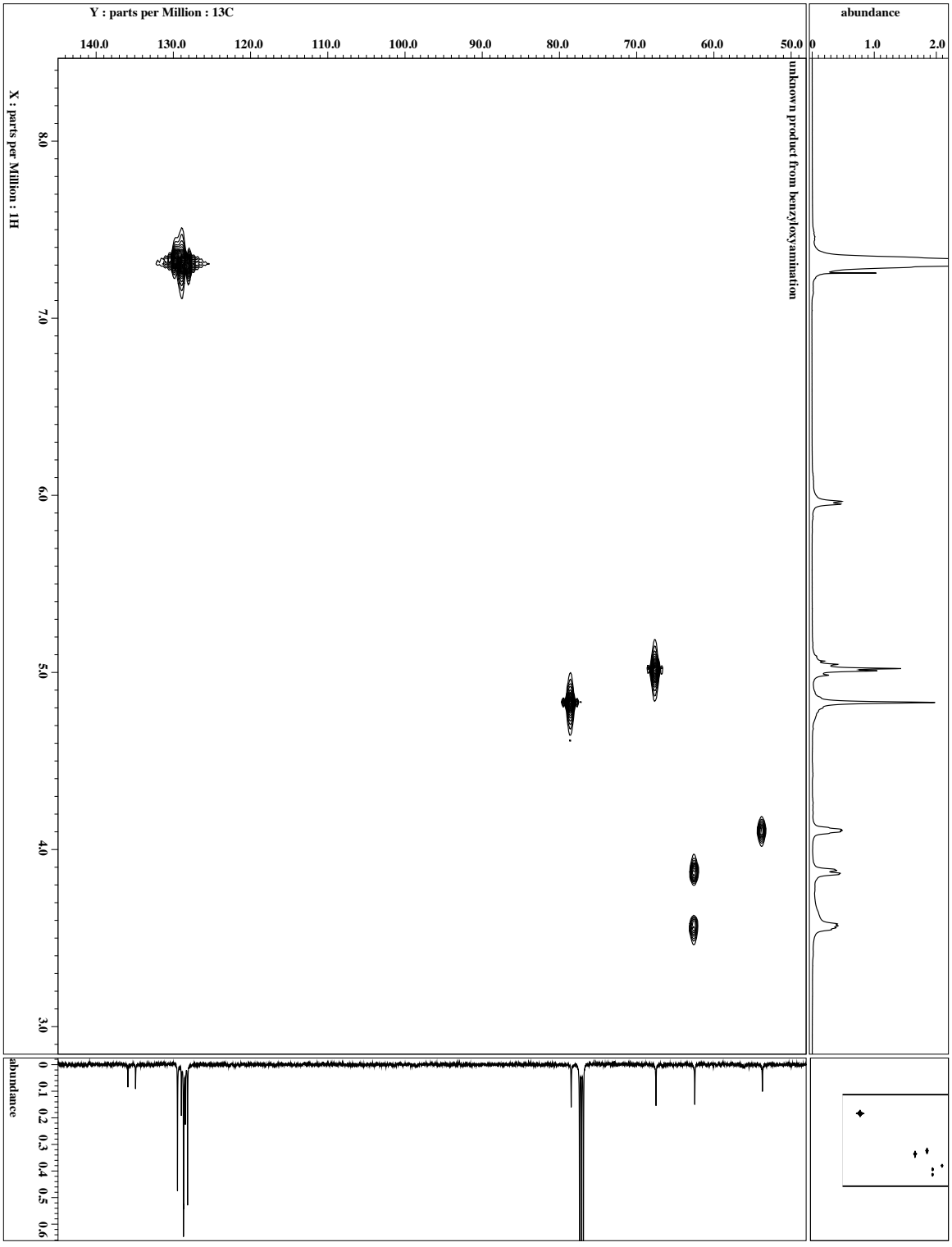
HMQC ($CDCl_3$)


abundance
unknown product from benzyloxyamination
X : parts per Million : 1H
9.6523
7.3316
7.3270
7.3156
7.3064
7.3018
7.2549
5.9654
5.9505
5.0458
5.0218
5.0092
4.9840
4.8305
4.1136
4.1079
4.1010
3.8823
3.8651
3.5800
3.5697
3.5582
2.1594
0.0734
0.0001
1.04
13.57
1.0
2.54
2.13
1.14
1.12
1.13
Filename = CRHIII_117b_PROTON-7.
Sample_id = CRHIII_117b
Machine = mt-su500ap
Creation_time = 28-FEB-2017 15:26:20
Comment = unknown product from
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.3[dC]

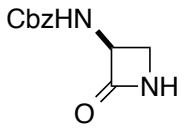



```
Filename                = CRHITI_117b_CARBON-4.
Sample_id               = CRHITI_117b
Machine                 = mt-su500ap
Creation_time           = 28-FEB-2017 14:59:29
Comment                 = unknown product from

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 0.83361792[s]
X_domain                = 13C
X_freq                  = 125.76529768[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 1.19959034[Hz]
X_sweep                 = 39.3081761[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 500
Total_scans             = 500

X_90_width              = 10.239[us]
X_acq_time              = 0.83361792[s]
X_angle                 = 30[deg]
X_atn                   = 9[dB]
X_pulse                 = 3.413[us]
Irr_atn_dec             = 21.5[dB]
Irr_atn_noe             = 21.5[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 2.83361792[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 21.6[dC]
```


unknown product from benzyloxyamination
abundance
X : parts per Million : 13C
129.4639
128.9584
128.6913
128.6913
128.4528
128.1571
78.4911
67.5029
62.4858
53.7296
Filename = CRHIII_117b_DEPT135-4
Sample_id = CRHIII_117b
Machine = mt-su500ap
Creation_time = 28-FEB-2017 15:23:45
Comment = unknown product from
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 500
Total_scans = 500
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 13C
140.0
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
X : parts per Million : 1H
8.0
7.0
6.0
5.0
4.0
3.0
unknown product from benzyloxyamination
abundance
0
1.0
2.0
abundance
0
0.1
0.2
0.3
0.4
0.5
0.6

3-[(Cbz)amino]-2-azetidinone (13)



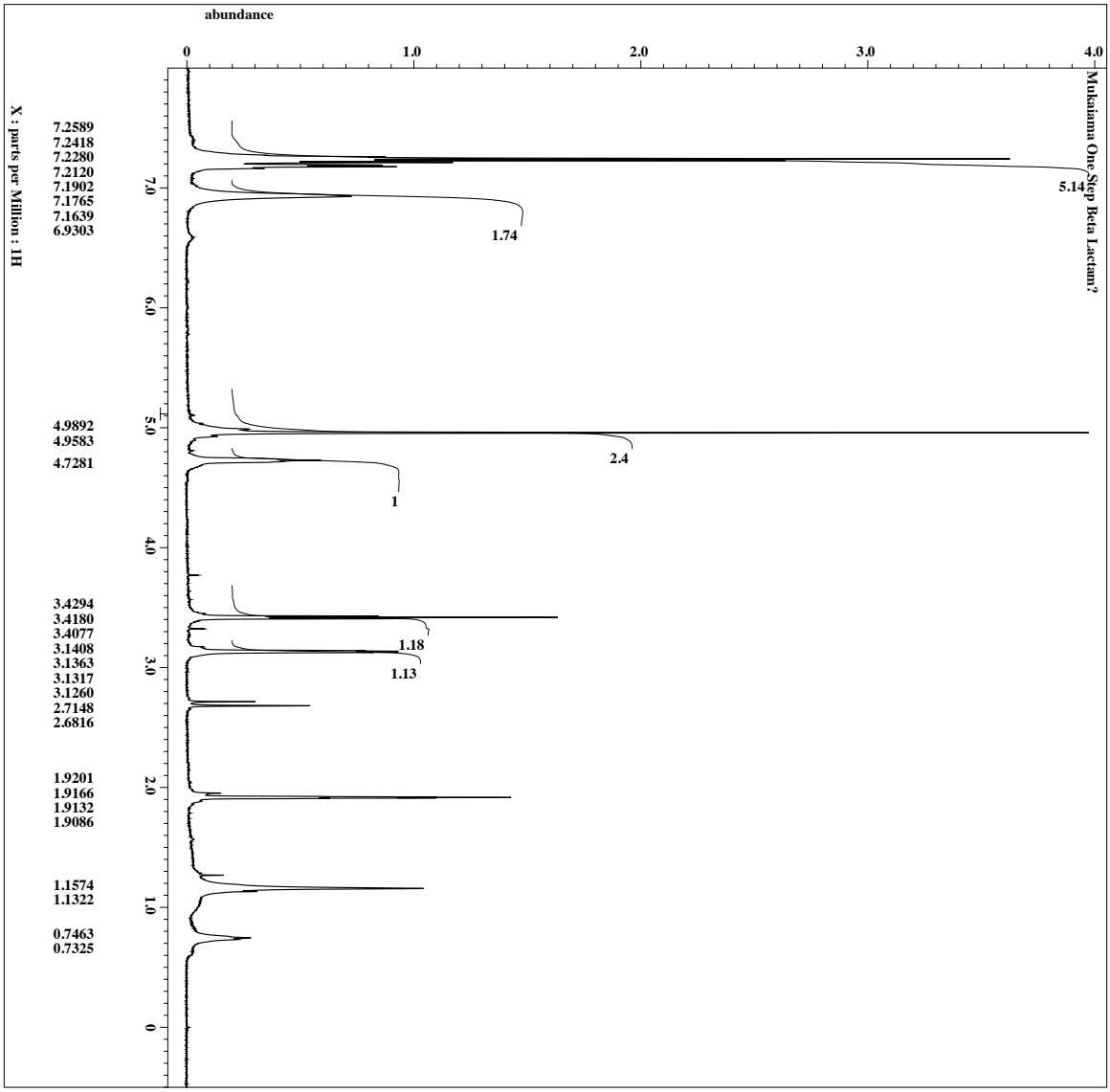
NMR

^{1}H-NMR (Acetone-d_6)

^{13}C-NMR (Acetone-d_6)

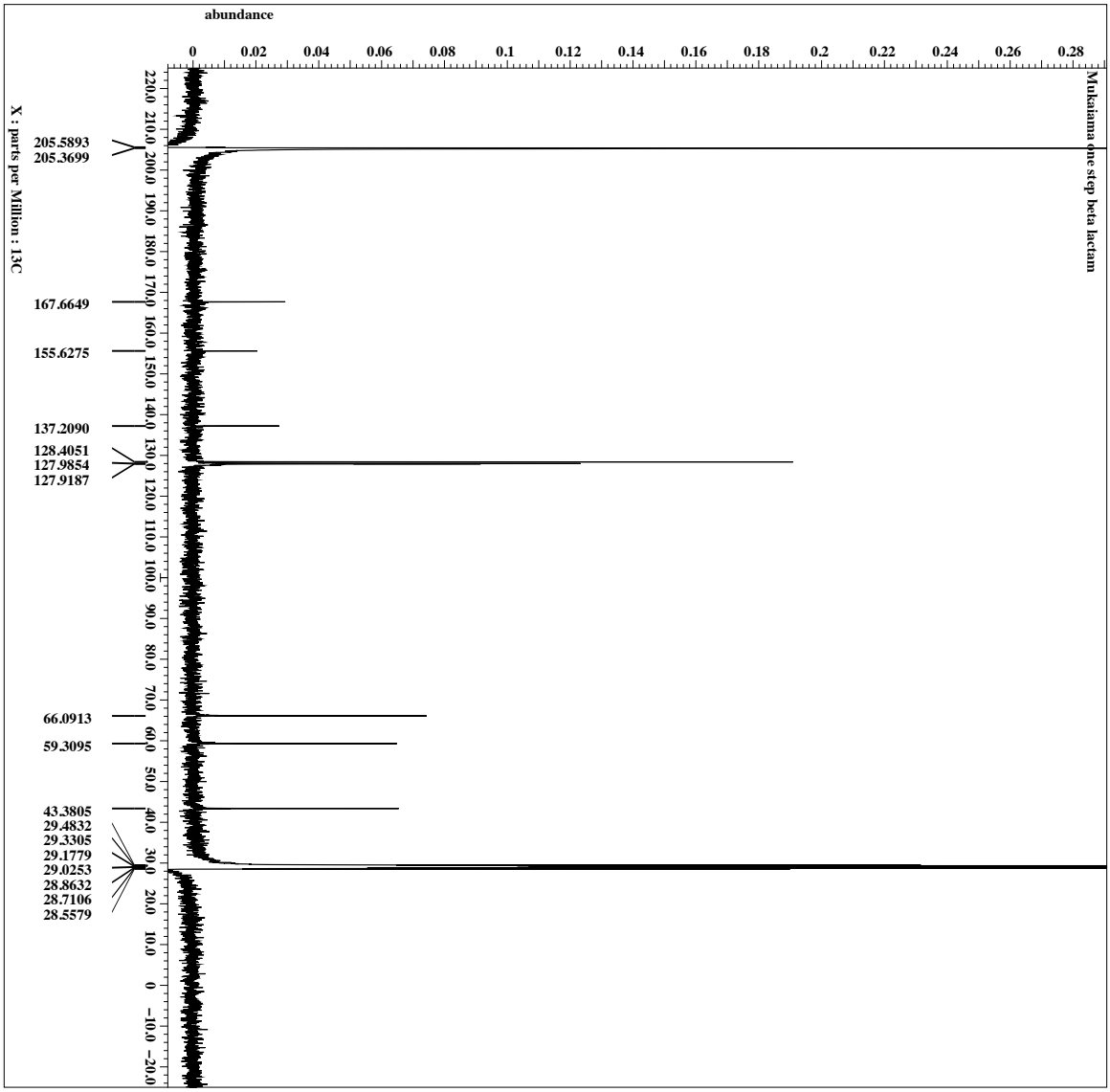
DEPT$_{135}$ (Acetone-d_6)

COSY (Acetone-d_6)



JEOL

```
Filename                = CRHIT_111C_PROTON-8.j
Sample_id               = CRHIT_111C
Machine                 = mt-su500ap
Creation_time           = 21-OCT-2016 18:21:44
Comment                 = Mukaiama One Step Bet

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 1.74587904[s]
X_domain                = 1H
X_freq                  = 500.15991521[MHz]
X_offset                = 5.0[ppm]
X_points                = 16384
X_prescans              = 1
X_resolution            = 0.57277737[Hz]
X_sweep                 = 9.38384438[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Tri_domain              = 1H
Tri_freq                = 500.15991521[MHz]
Tri_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 16
Total_scans             = 16

X_90_width              = 14.19[us]
X_acq_time              = 1.74587904[s]
X_angle                 = 45[deg]
X_atn                   = 4[dB]
X_pulse                 = 7.095[us]
Irr_mode                = Off
Tri_mode                = Off
Dante_presat            = OFF
Initial_wait            = 1[s]
Relaxation_delay        = 4[s]
Repetition_time         = 5.74587904[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 22.8[dC]
```

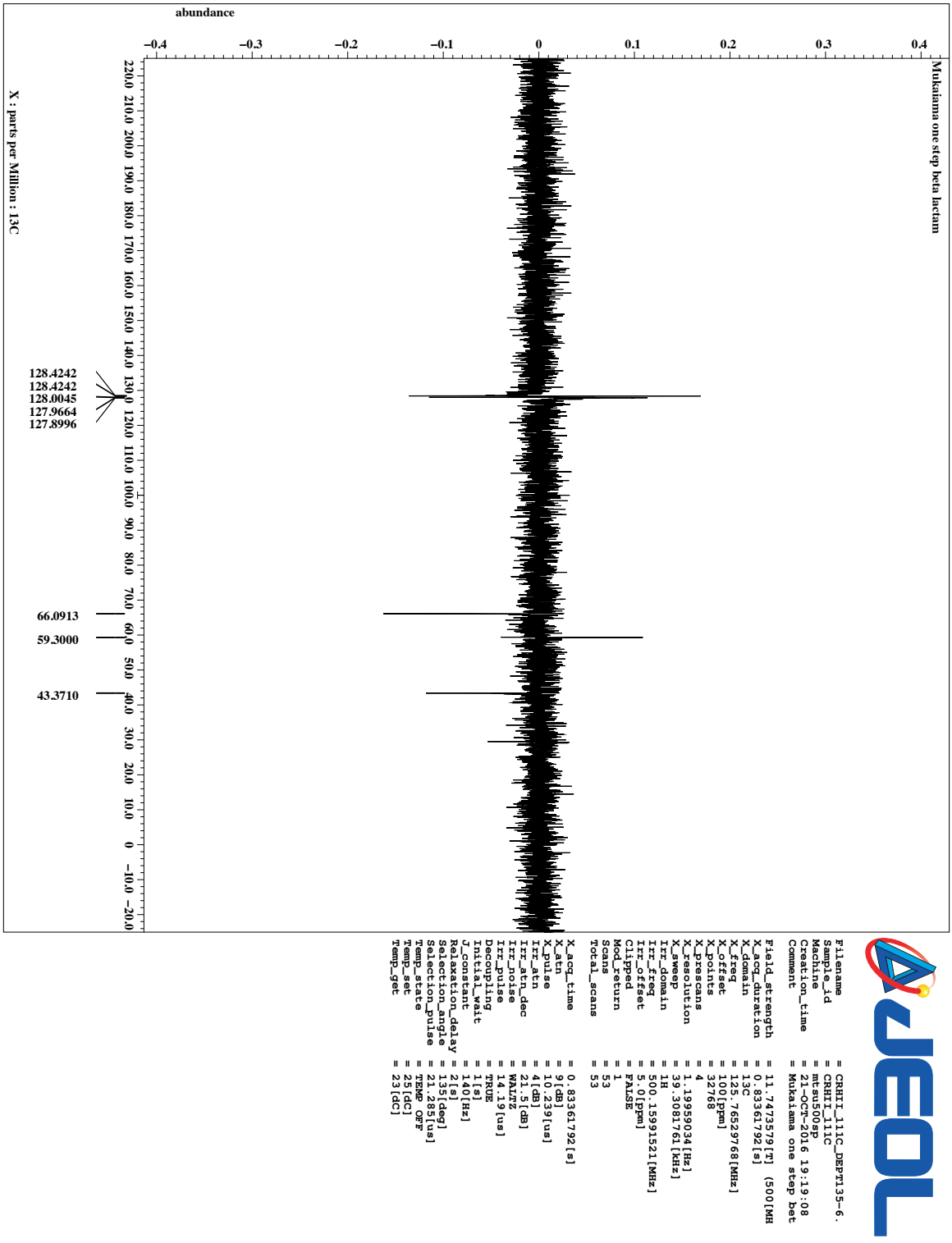


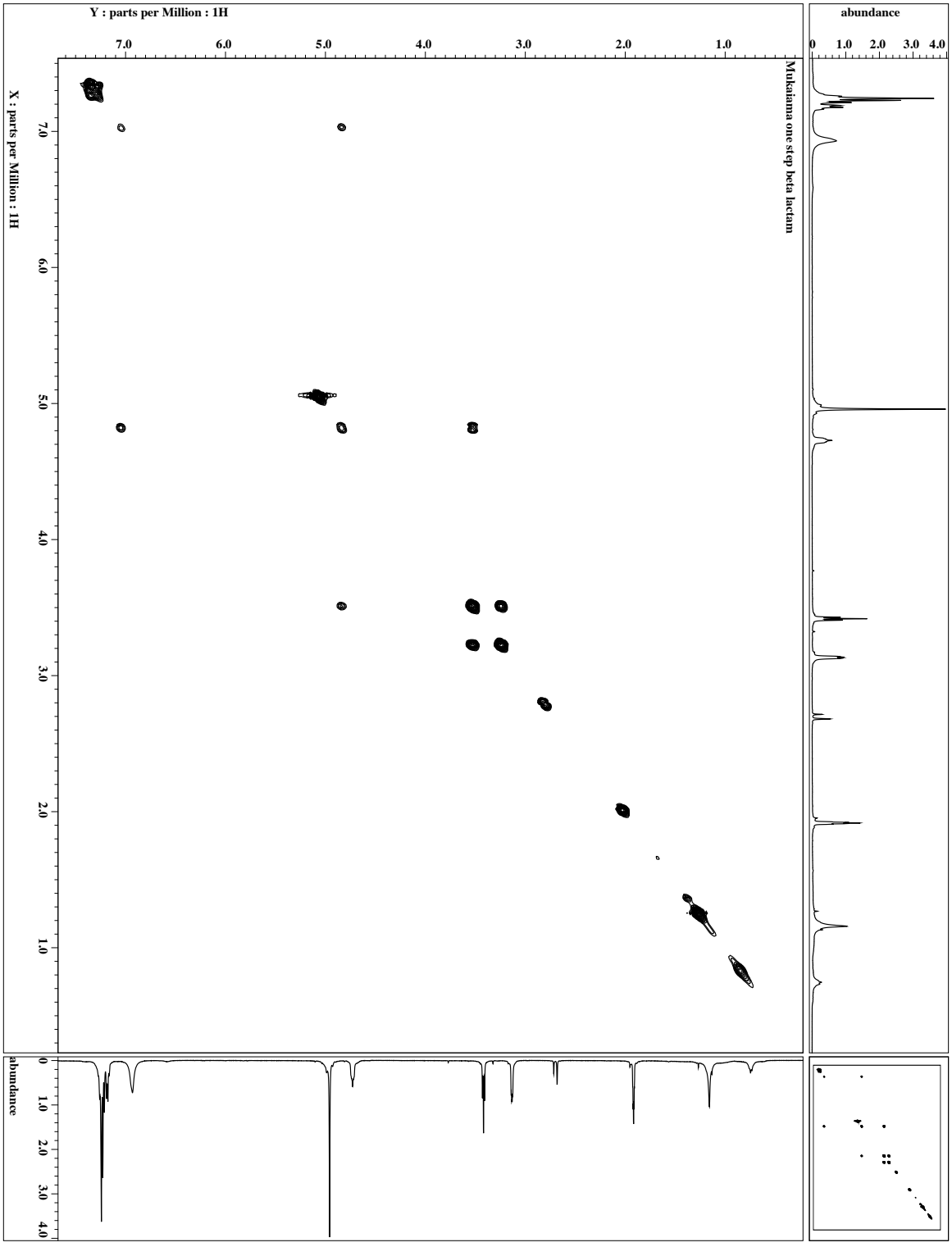
JEOL

Filename = CRHI_111C_CARBON-5.j
Sample_id = CRHI_111C
Machine = mt-su500ap
Creation_time = 21-OCT-2016 19:16:05
Comment = Mukaiama one step bet

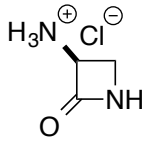
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 1024
Total_scans = 1024

X_90_width = 10.239[us]
X_acq_time = 0.83361792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.83361792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.2[dC]


Mukaiama one step beta lactam
abundance
X : parts per Million : 13C
128.4242
128.4242
128.0045
127.9664
127.8996
66.0913
59.3000
43.3710
Filename = CRHII_111C_DEPT135-6.
Sample_id = CRHII_111C
Machine = mt-su500ap
Creation_time = 21-OCT-2016 19:19:08
Comment = Mukaiama one step bet
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 53
Total_scans = 53
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
abundance
Mukaiama one step beta lactam

3-[amino]-2-azetidinone hydrochloride (14)



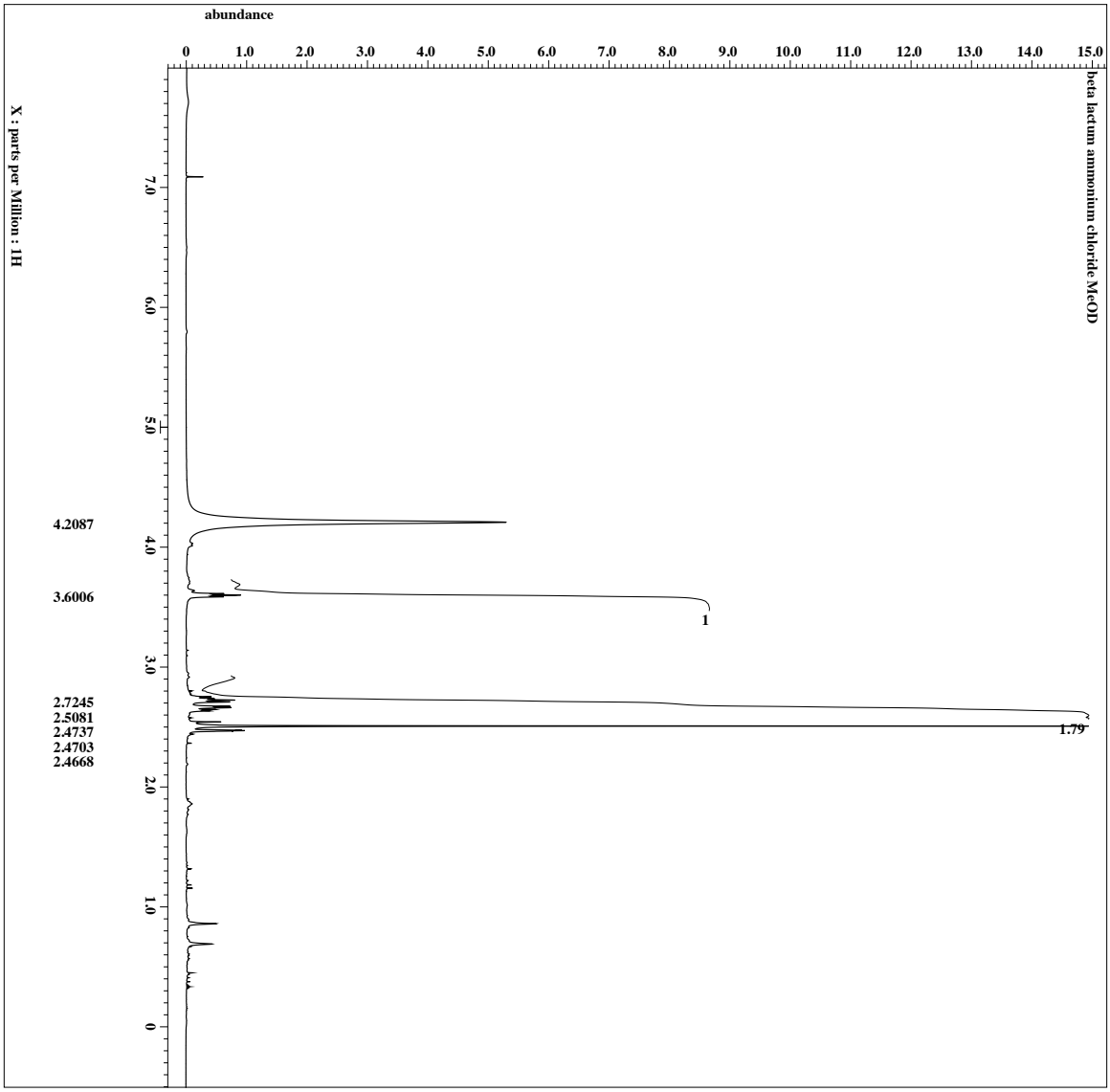
NMR

^{1}H-NMR (Methanol-d_4)

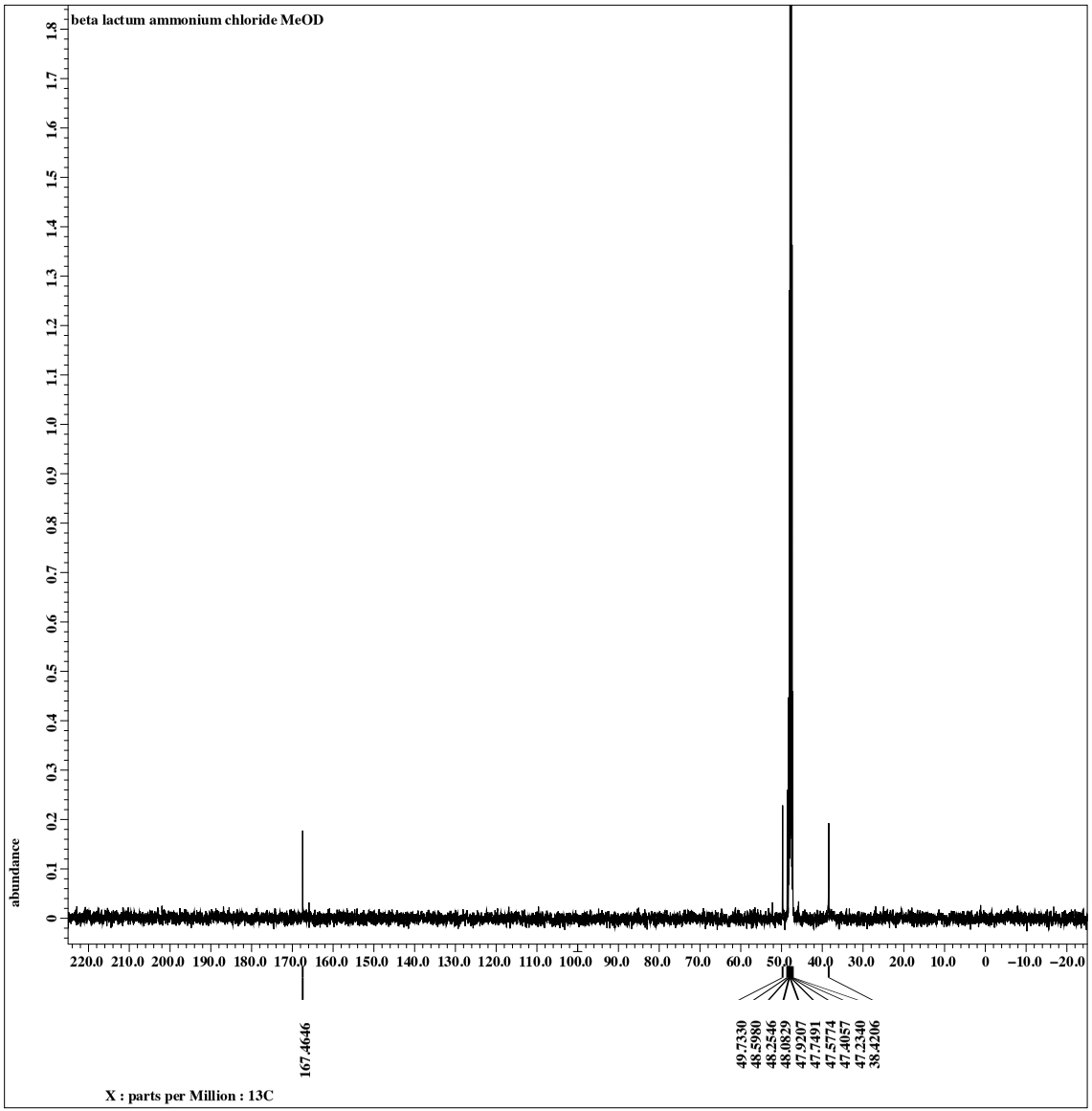
^{13}C-NMR (Methanol-d_4)

DEPT$_{135}$ (Methanol-d_4)

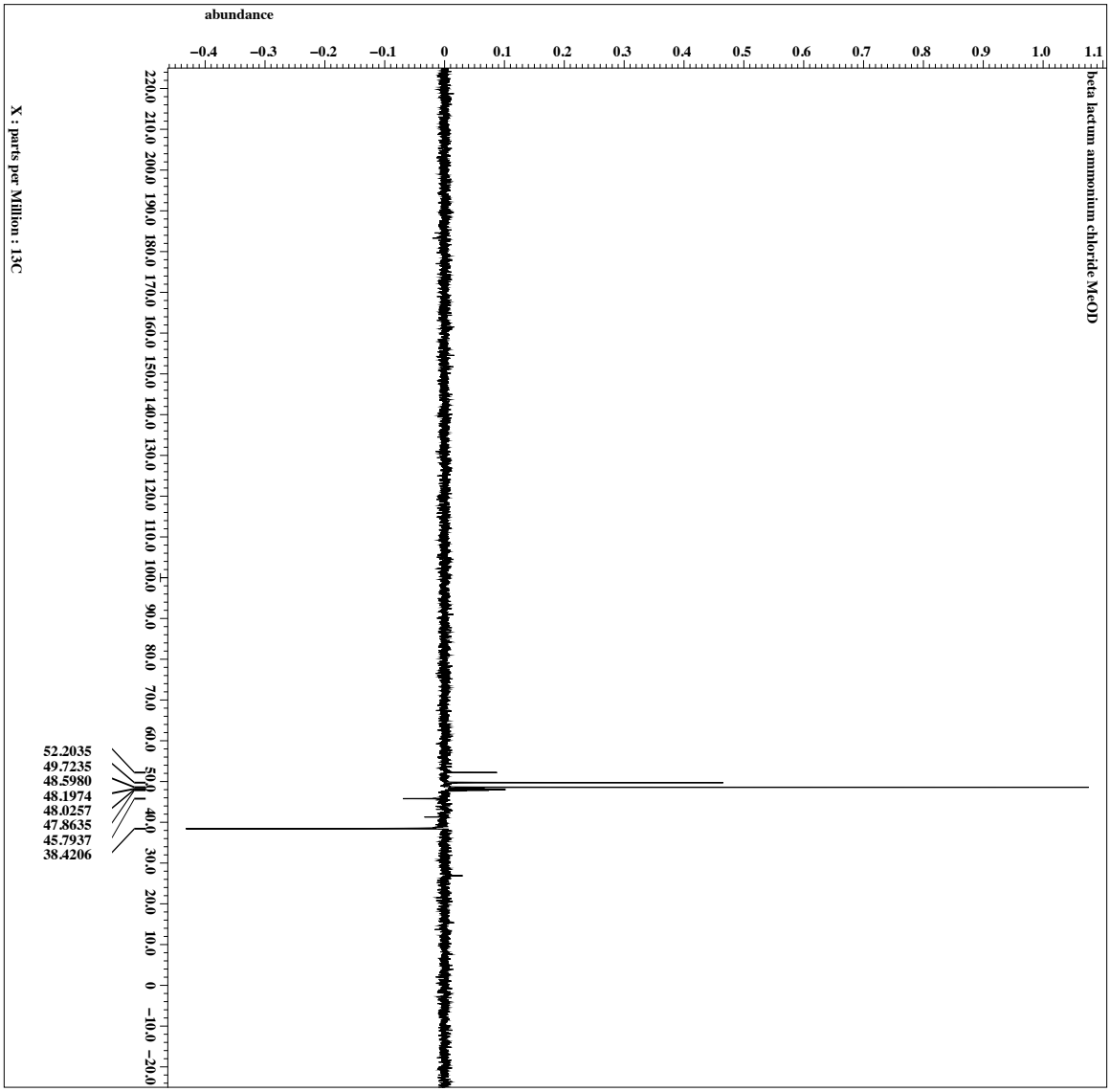
HMQC (Methanol-d_4)

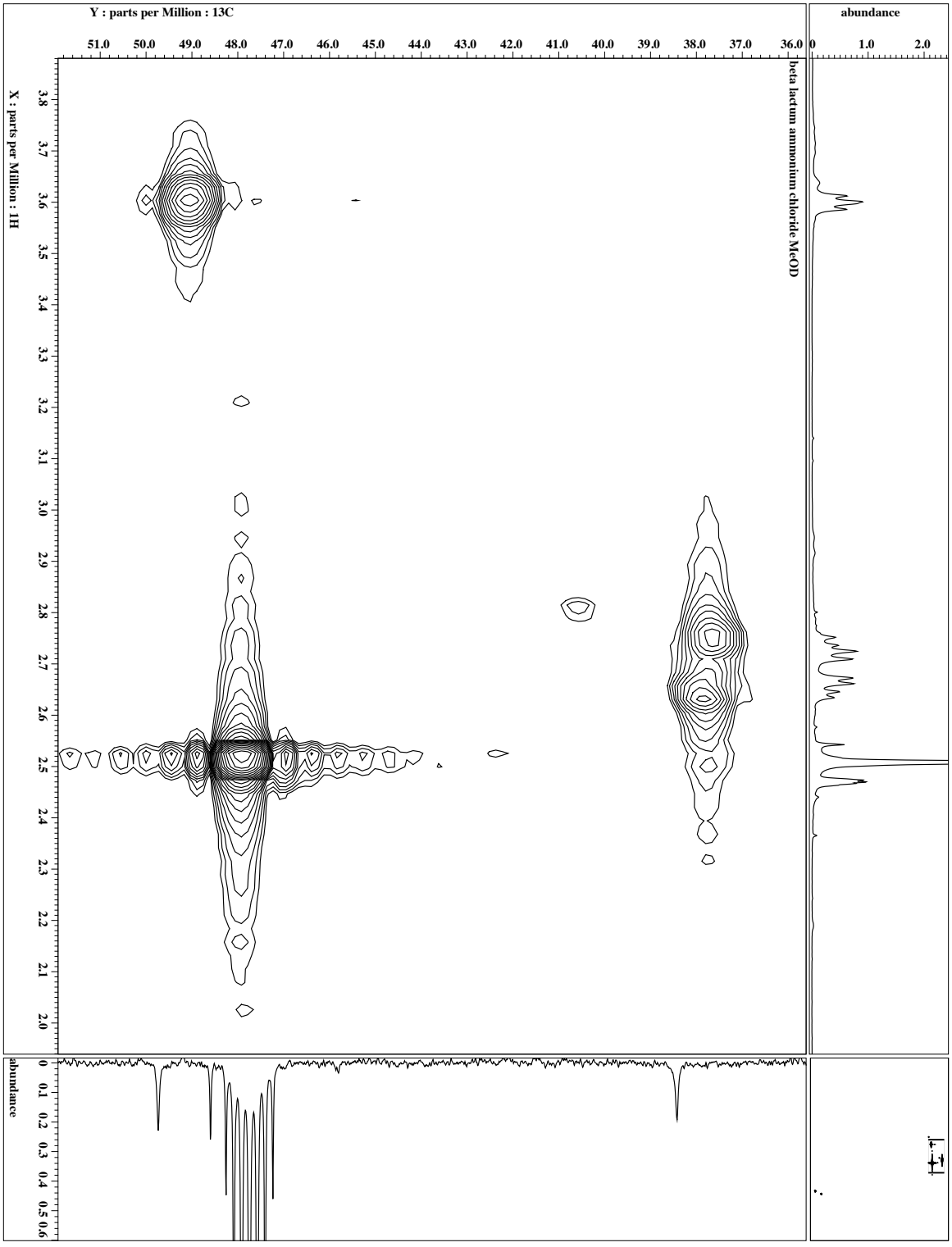



```
Filename          = CRHIII_11a_PROTON-11.
Sample_id         = CRHIII_11a
Machine           = mt-su500ap
Creation_time     = 26-JAN-2017 08:52:16
Comment           = beta lactum ammonium

Field_strength    = 11.7473579[T] (500[MH
X_acq_duration    = 1.74587904[s]
X_domain          = 1H
X_freq            = 500.15991521[MHz]
X_offset          = 5.0[ppm]
X_points          = 16384
X_prescans        = 1
X_resolution      = 0.57277737[Hz]
X_sweep           = 9.38438438[kHz]
Irr_domain        = 1H
Irr_freq          = 500.15991521[MHz]
Irr_offset        = 5.0[ppm]
Tri_domain        = 1H
Tri_freq          = 500.15991521[MHz]
Tri_offset        = 5.0[ppm]
Clipped           = FALSE
Mod_return        = 1
Scans             = 16
Total_scans       = 16

X_90_width        = 14.19[us]
X_acq_time        = 1.74587904[s]
X_angle           = 45[deg]
X_atn             = 4[dB]
X_pulse           = 7.095[us]
Irr_mode          = Off
Tri_mode          = Off
Dante_presat      = FALSE
Initial_wait      = 1[s]
Relaxation_delay  = 4[s]
Repetition_time   = 5.74587904[s]
Temp_state        = TEMP OFF
Temp_set          = 25[dC]
Temp_get          = 21.3[dC]
```

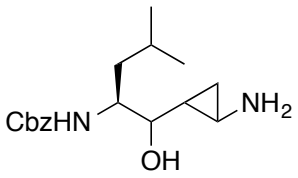



```
Filename          = CRHIII_11a_CARBON-11.
Sample_id         = CRHIII_11a
Machine           = mtsu500sp
Creation_time     = 26-JAN-2017 10:00:21
Comment           = beta lactum ammonium

Field_strength    = 11.7473579[T] (500[MH
X_acq_duration    = 0.83361792[s]
X_domain          = 13C
X_freq            = 125.76529768[MHz]
X_offset          = 100[ppm]
X_points          = 32768
X_prescans        = 4
X_resolution      = 1.19959034[Hz]
X_sweep           = 39.3081761[kHz]
Irr_domain        = 1H
Irr_freq          = 500.15991521[MHz]
Irr_offset        = 5.0[ppm]
Clipped           = FALSE
Mod_return        = 1
Scans             = 100
Total_scans       = 100

X_90_width        = 10.239[us]
X_acq_time        = 0.83361792[s]
X_angle           = 30[deg]
X_atn             = 9[dB]
X_pulse           = 3.413[us]
Irr_atn_dec       = 21.5[dB]
Irr_atn_noe       = 21.5[dB]
Irr_noise         = WALTZ
Decoupling        = TRUE
Initial_wait      = 1[s]
Noe               = TRUE
Noe_time          = 2[s]
Relaxation_delay  = 2[s]
Repetition_time   = 2.83361792[s]
Temp_state        = TEMP OFF
Temp_set          = 25[dC]
Temp_get          = 21.7[dC]
```


beta lactum ammonium chloride MeOD
abundance
X : parts per Million : 13C
52.2035
49.7235
48.5980
48.1974
48.0257
47.8635
45.7937
38.4206
Filename = CRHIII_11a_DEPT135-4.
Sample_id = CRHIII_11a
Machine = mt-su500ap
Creation_time = 26-JAN-2017 10:11:48
Comment = beta lactum ammonium
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_sweep = 1H
Irr_domain = 500.15991521[MHz]
Irr_freq = 5.0[ppm]
Irr_offset = TRUE
Clipped = 1
Mod_return = 230.0
Scans = 230.0
Total_scans
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 13C
51.0 50.0 49.0 48.0 47.0 46.0 45.0 44.0 43.0 42.0 41.0 40.0 39.0 38.0 37.0 36.0
X : parts per Million : 1H
3.8 3.7 3.6 3.5 3.4 3.3 3.2 3.1 3.0 2.9 2.8 2.7 2.6 2.5 2.4 2.3 2.2 2.1 2.0
beta lactum ammonium chloride MeOD
abundance
0 1.0 2.0
abundance
0 0.1 0.2 0.3 0.4 0.5 0.6

2-((*S*)-2-(benzylcarbonylamino)-1-hydroxy-4-methylpentyl) cyclopropaneamine (28)

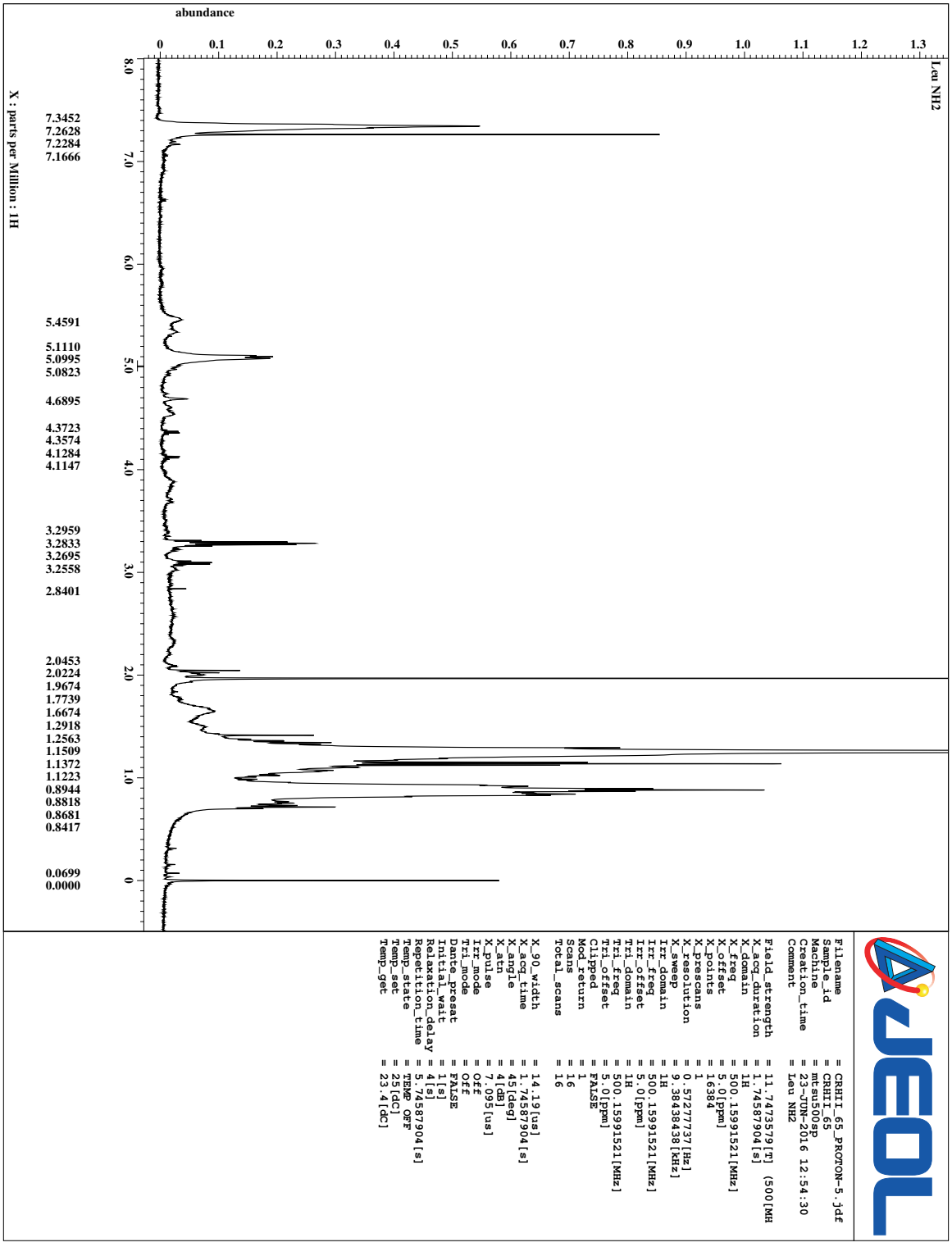


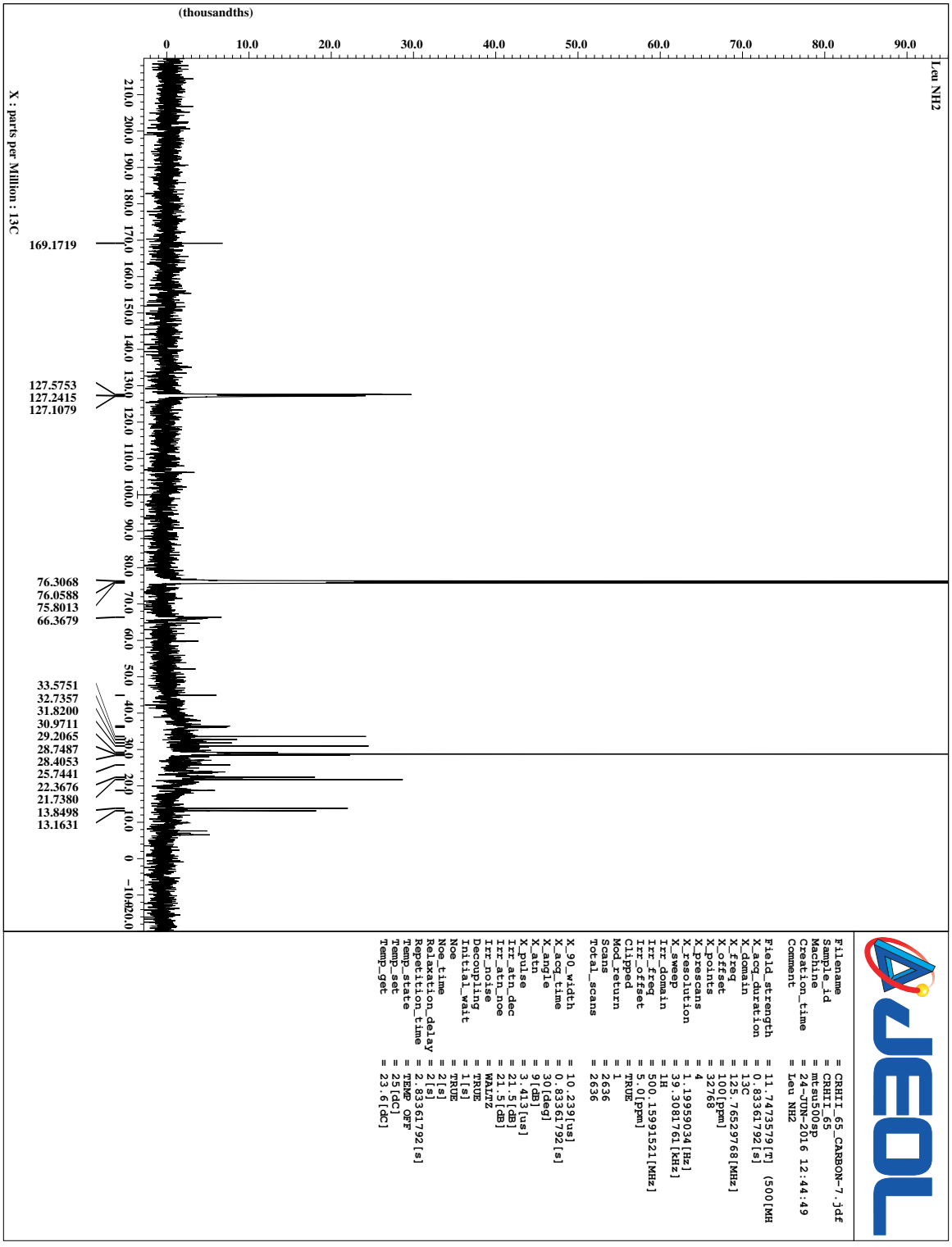
NMR

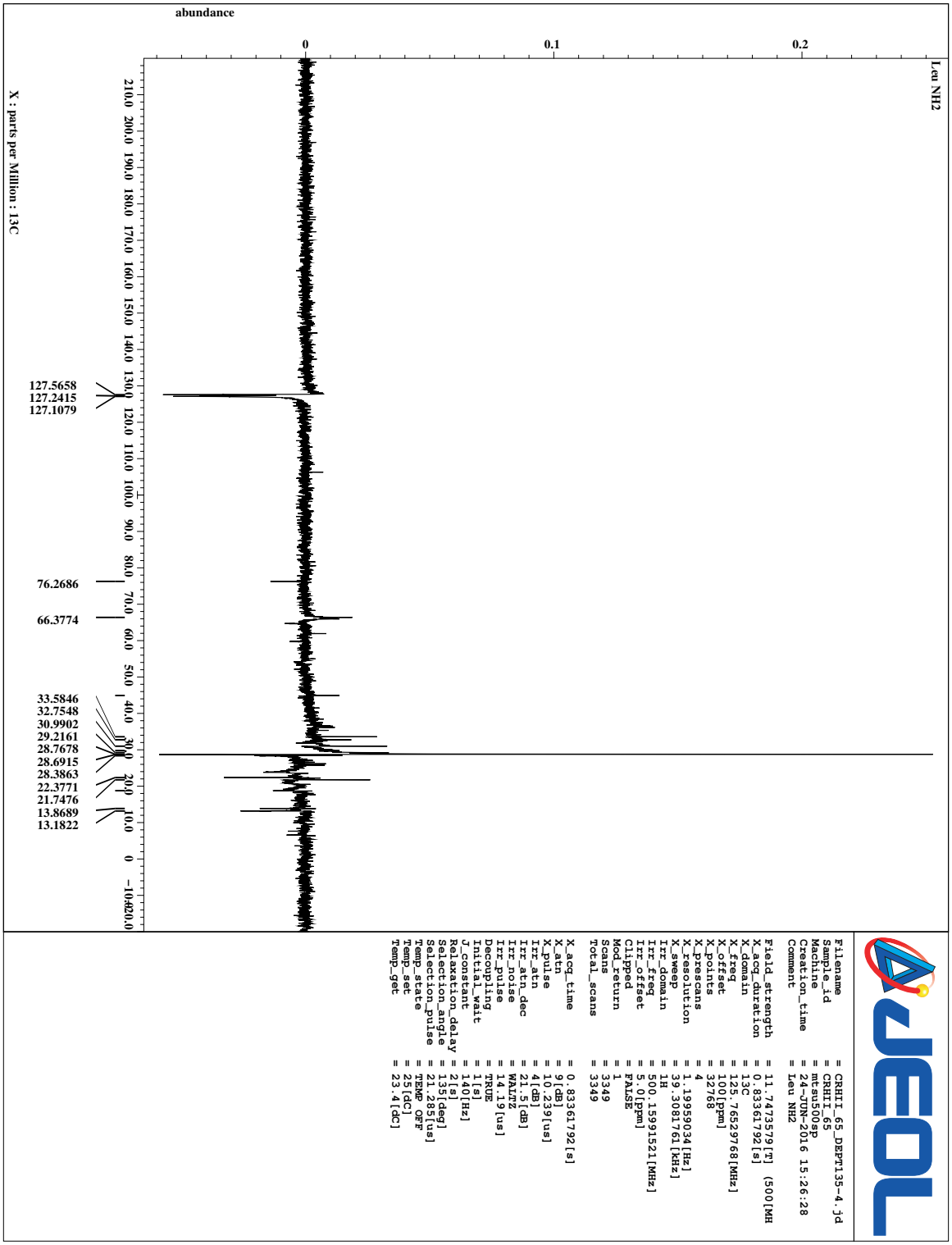
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

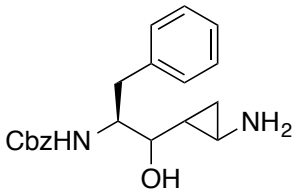
$DEPT_{135}$ ($CDCl_3$)


Leu NH2
abundance
X : parts per Million : 1H
7.3452
7.2628
7.2284
7.1666
5.4591
5.1110
5.0995
5.0823
4.6895
4.3723
4.3574
4.1284
4.1147
3.2959
3.2833
3.2695
3.2558
2.8401
2.0453
2.0224
1.9674
1.7739
1.6674
1.2918
1.2563
1.1509
1.1372
1.1223
0.8944
0.8818
0.8681
0.8417
0.0699
0.0000
Filename = CRHI1_65_PROTON-5.jdf
Sample_id = CRHI1_65_
Machine = mt-su500sp
Creation_time = 23-JUN-2016 12:54:30
Comment = Leu NH2
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.4[dC]


(thousandths)
0
10.0
20.0
30.0
40.0
50.0
60.0
70.0
80.0
90.0
Leu NH2
X : parts per Million : 13C
210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0 −10 −20.0
169.1719
127.5753
127.2415
127.1079
76.3068
76.0588
75.8013
66.3679
33.5751
32.7357
31.8200
30.9711
29.2065
28.7487
28.4053
25.7441
22.3676
21.7380
13.8498
13.1631
Filename = CRHI_65_CARBON-7.jdf
Sample_id = CRHI_65_
Machine = mt-su500ap
Creation_time = 24-JUN-2016 12:44:49
Comment = Leu NH2
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.8336179[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = TRUE
Mod_return = 1
Scans = 2636
Total_scans = 2636
X_90_width = 10.239[us]
X_acq_time = 0.8336179[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.8336179[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.6[dC]


Leu NH2
abundance
X : parts per Million : 13C
127.5658
127.2415
127.1079
76.2686
66.3774
33.5846
32.7548
30.9902
29.2161
28.7678
28.6915
28.3863
22.3771
21.7476
13.8689
13.1822
Filename = CRHI1_65_DEPT135-4.jd
Sample_id = CRHI1_65_
Machine = mt-su500ap
Creation_time = 24-JUN-2016 15:26:28
Comment = Leu NH2
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 3349
Total_scans = 3349
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.4[dC]

2-((*S*)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropaneamine (29)



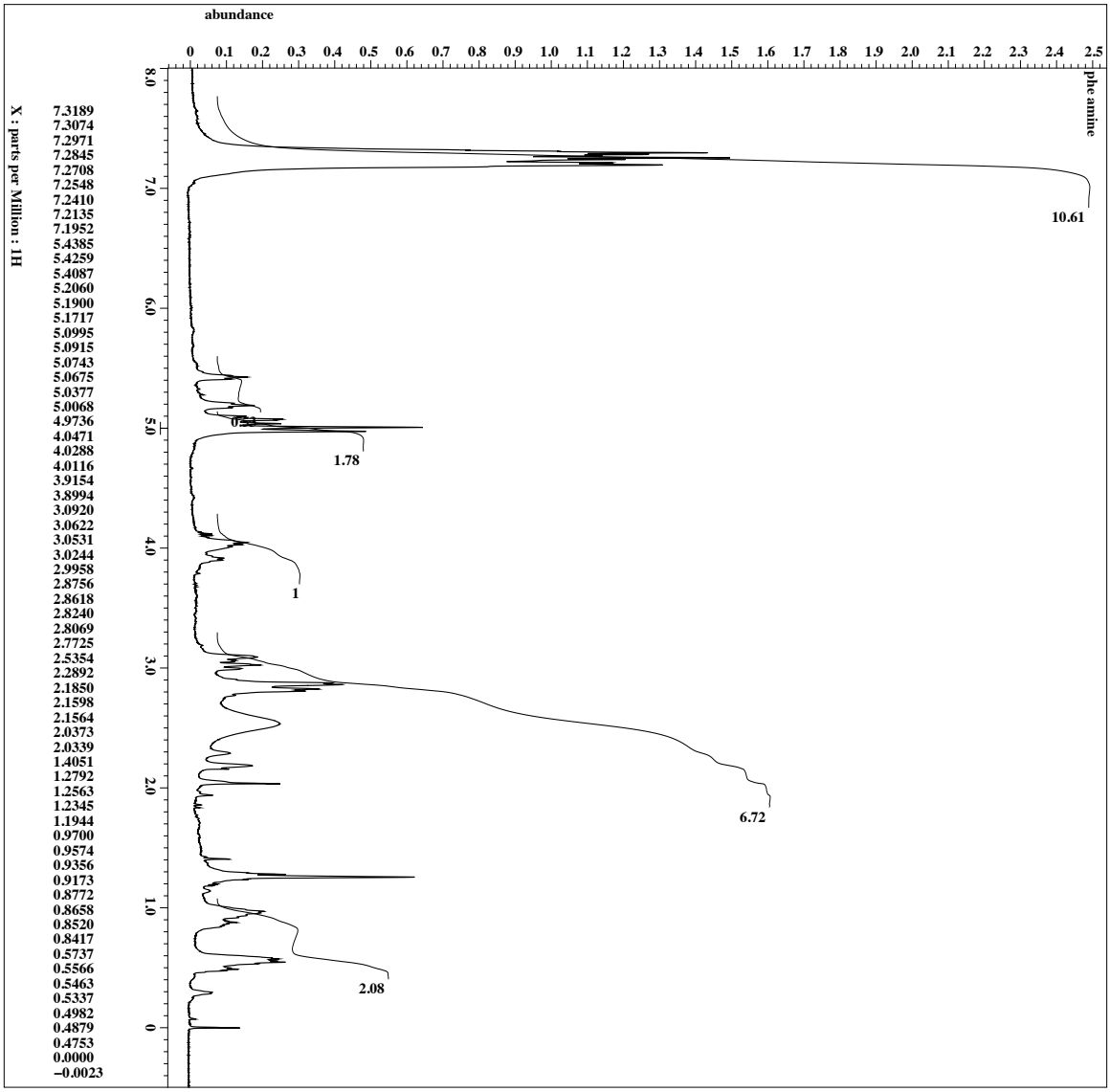
NMR

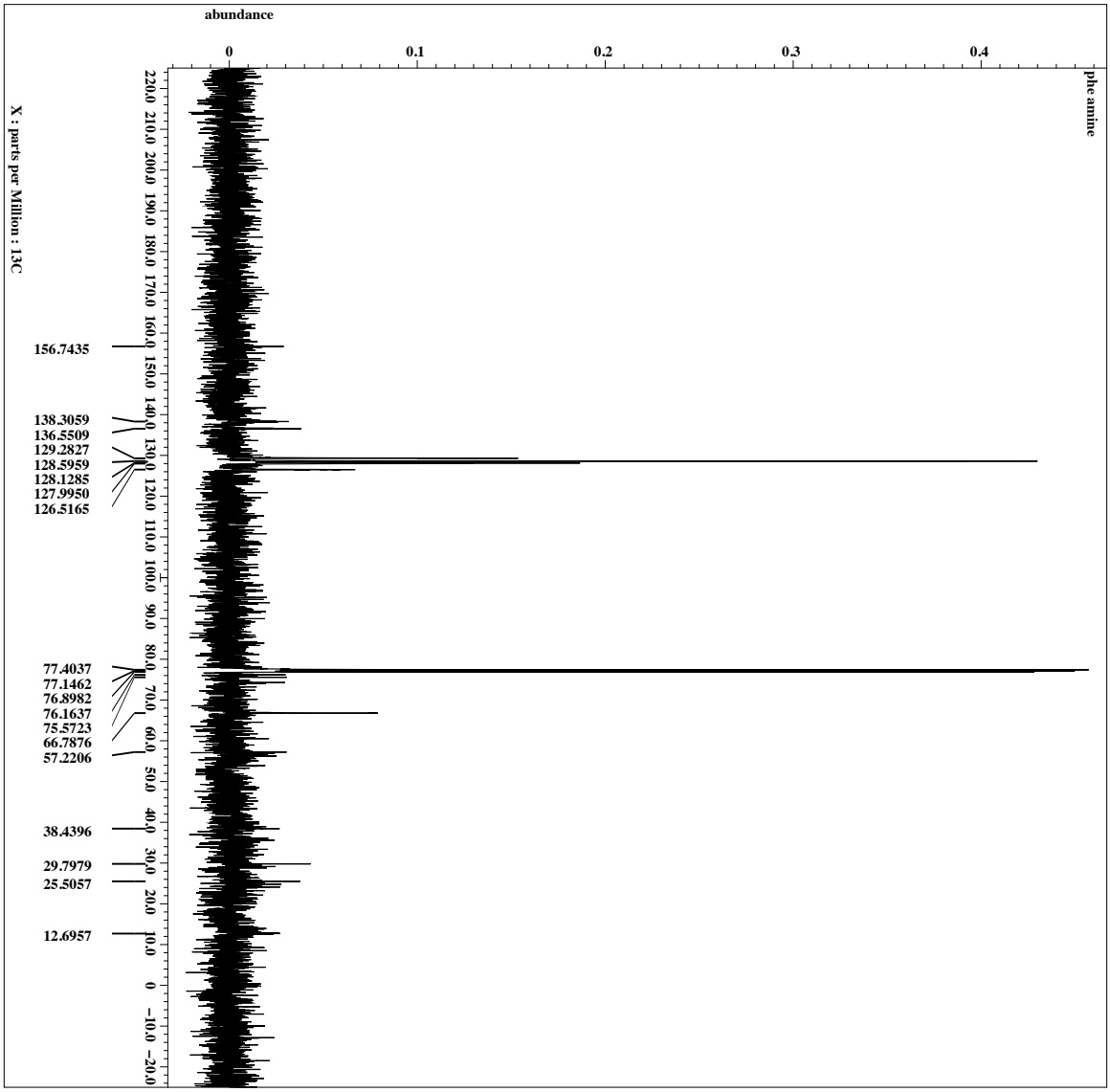
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

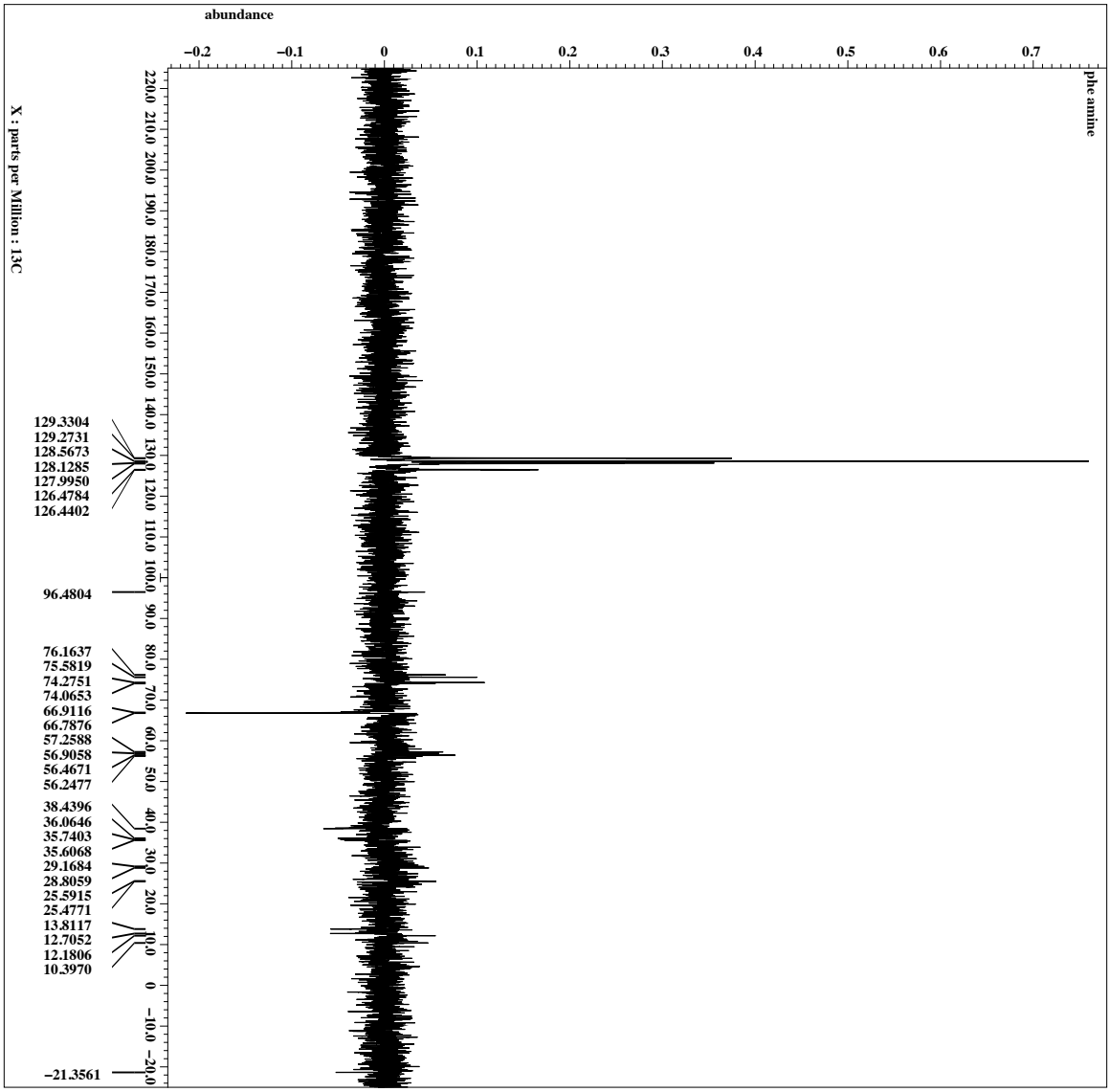
$DEPT_{135}$ ($CDCl_3$)

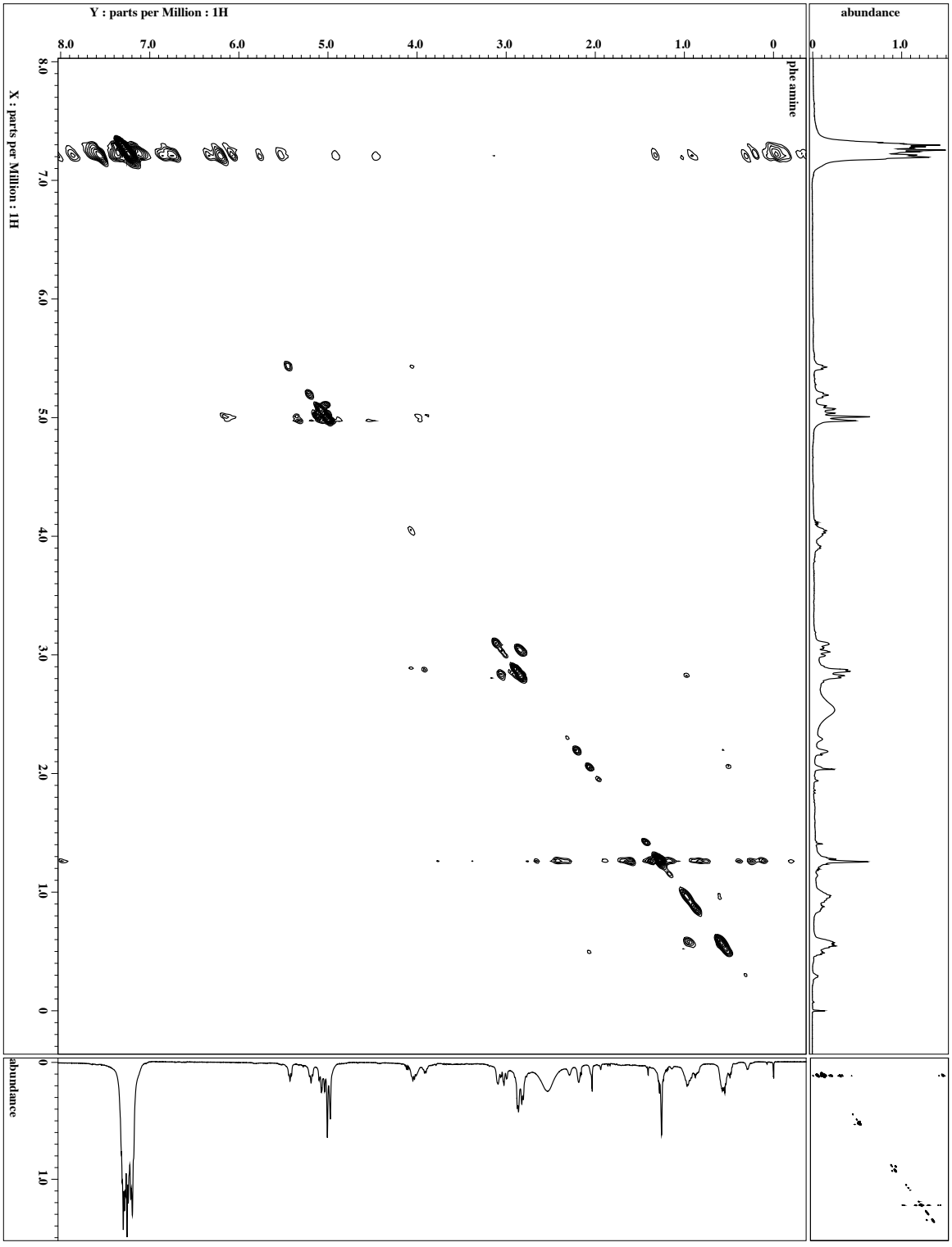
COSY ($CDCl_3$)


abundance
phe amine
10.61
1.78
1
6.72
2.08
X : parts per Million : 1H
7.3189
7.3074
7.2971
7.2845
7.2708
7.2548
7.2410
7.2135
7.1952
5.4385
5.4259
5.4087
5.2060
5.1900
5.1717
5.0995
5.0915
5.0743
5.0675
5.0377
5.0068
4.9736
4.0471
4.0288
4.0116
3.9154
3.8994
3.0920
3.0622
3.0531
3.0244
2.9958
2.8756
2.8618
2.8240
2.8069
2.7725
2.5354
2.2892
2.1850
2.1598
2.1564
2.0373
2.0339
1.4051
1.2792
1.2563
1.2345
1.1944
0.9700
0.9574
0.9356
0.9173
0.8772
0.8658
0.8520
0.8417
0.5737
0.5566
0.5463
0.5337
0.4982
0.4879
0.4753
0.0000
−0.0023
Filename = NKD_ivp99_PROTON-7.jd
Sample_id = NKD_ivp99
Machine = mt-eu500pp
Creation_time = 23-FEB-2016 11:07:51
Comment = phe amine
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23[dC]

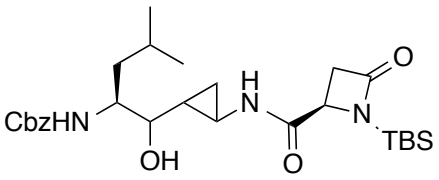



```
Filename                = NKD_1vp99_CARBON-6.jd
Sample_id               = NKD_1vp99
Machine                 = mt-eu500pp
Creation_time           = 23-FEB-2016 11:22:13
Comment                 = phe amine

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 0.83361792[s]
X_domain                = 13C
X_freq                  = 125.76529768[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 1.19959034[Hz]
X_sweep                 = 39.3081761[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 69
Total_scans             = 69

X_90_width              = 9.65[us]
X_acq_time              = 0.83361792[s]
X_angle                 = 30[deg]
X_atn                   = 9[dB]
X_pulse                 = 3.21666667[us]
Irr_atn_dec             = 21.5[dB]
Irr_atn_noe             = 21.5[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 2.83361792[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 23.2[dC]
```


phe amine
abundance
X : parts per Million : 13C
129.3304
129.2731
128.5673
128.1285
127.9950
126.4784
126.4402
96.4804
76.1637
75.5819
74.2751
74.0653
66.9116
66.7876
57.2588
56.9058
56.4671
56.2477
38.4396
36.0646
35.7403
35.6068
29.1684
28.8059
25.5915
25.4771
13.8117
12.7052
12.1806
10.3970
−21.3561
Filename = NKD_ivp99_DEPT135-6.j
Sample_id = NKD_ivp99
Machine = mt-eu500pp
Creation_time = 23-FEB-2016 11:24:24
Comment = phe amine
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 34
Total_scans = 34
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 9.65[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.1[dC]

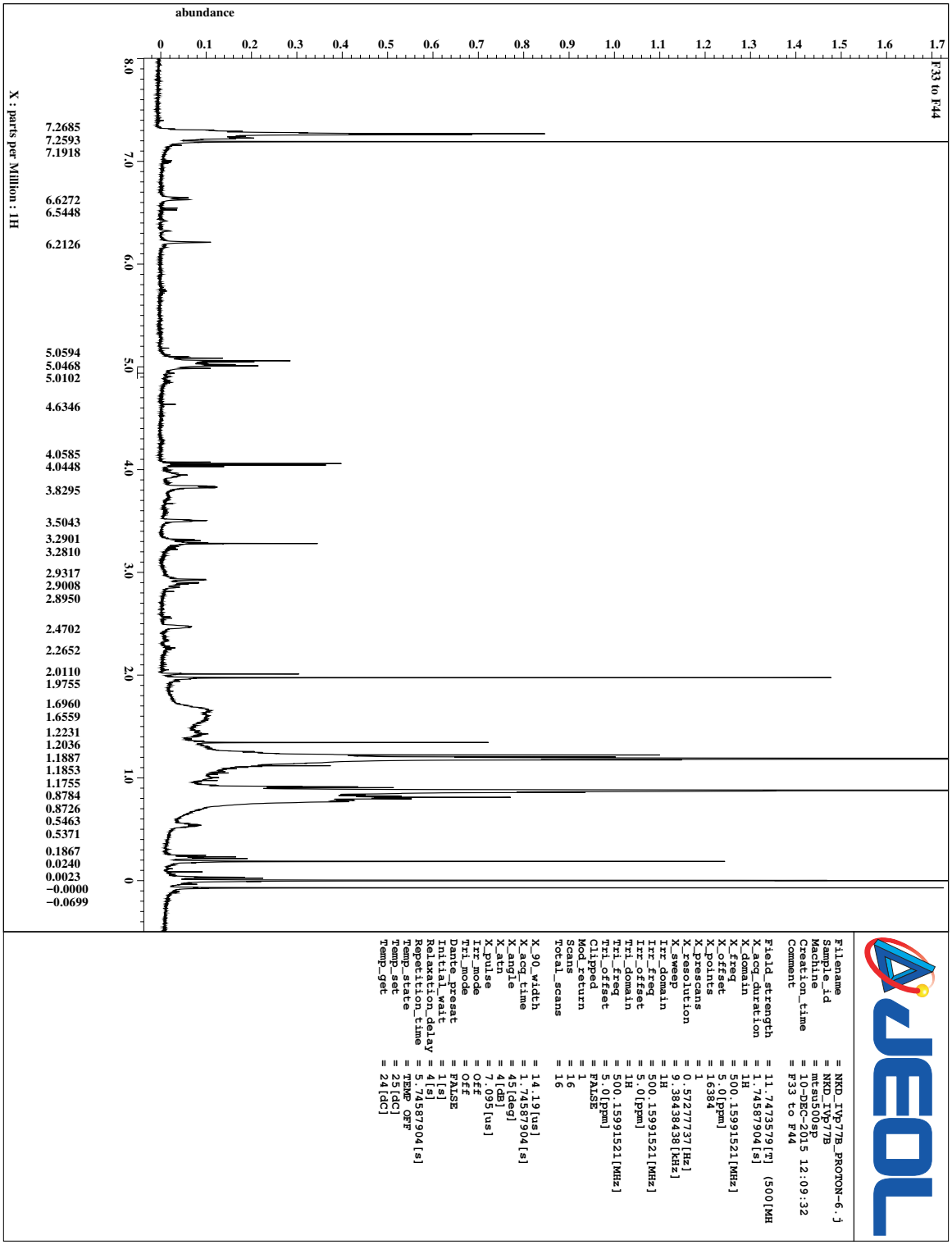

Y : parts per Million : 1H
abundance
X : parts per Million : 1H
phe amine
abundance

Benzyl N-[(2*S*)-1-{2-[1-(tert-butyldimethylsilyl)-4-oxoazetidine-2-amido]cyclopropyl}-1-hydroxy-4-methylpentan-2-yl]carbamate (31)

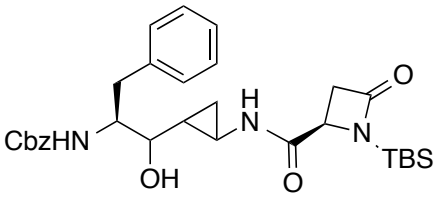


NMR

^{1}H-NMR ($CDCl_3$)


abundance
F33 to F44
X : parts per Million : 1H
7.2685
7.2593
7.1918
6.6272
6.5448
6.2126
5.0594
5.0468
5.0102
4.6346
4.0585
4.0448
3.8295
3.5043
3.2901
3.2810
2.9317
2.9008
2.8950
2.4702
2.2652
2.0110
1.9755
1.6960
1.6559
1.2231
1.2036
1.1887
1.1853
1.1755
0.8784
0.8726
0.5463
0.5371
0.1867
0.0240
0.0023
−0.0000
−0.0699
Filename = NKD_IVb77B_PROTON-6.j
Sample_id = NKD_IVb77B
Machine = mt-su500-sp
Creation_time = 10-DEC-2015 12:09:32
Comment = F33 to F44
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = FALSE
Relaxation_delay = 1[s]
Repetition_time = 4[s]
Temp_state = 5.74587904[s]
Temp_set = TEMP OFF
Temp_get = 25[dC]
= 24[dC]

Benzyl N-[(2*S*)-1-{2-[1-(tert-butyldimethylsilyl)-4-oxoazetidine-2-amido]cyclopropyl}-1-hydroxy-4-phenylpropan-2-yl]carbamate (32)



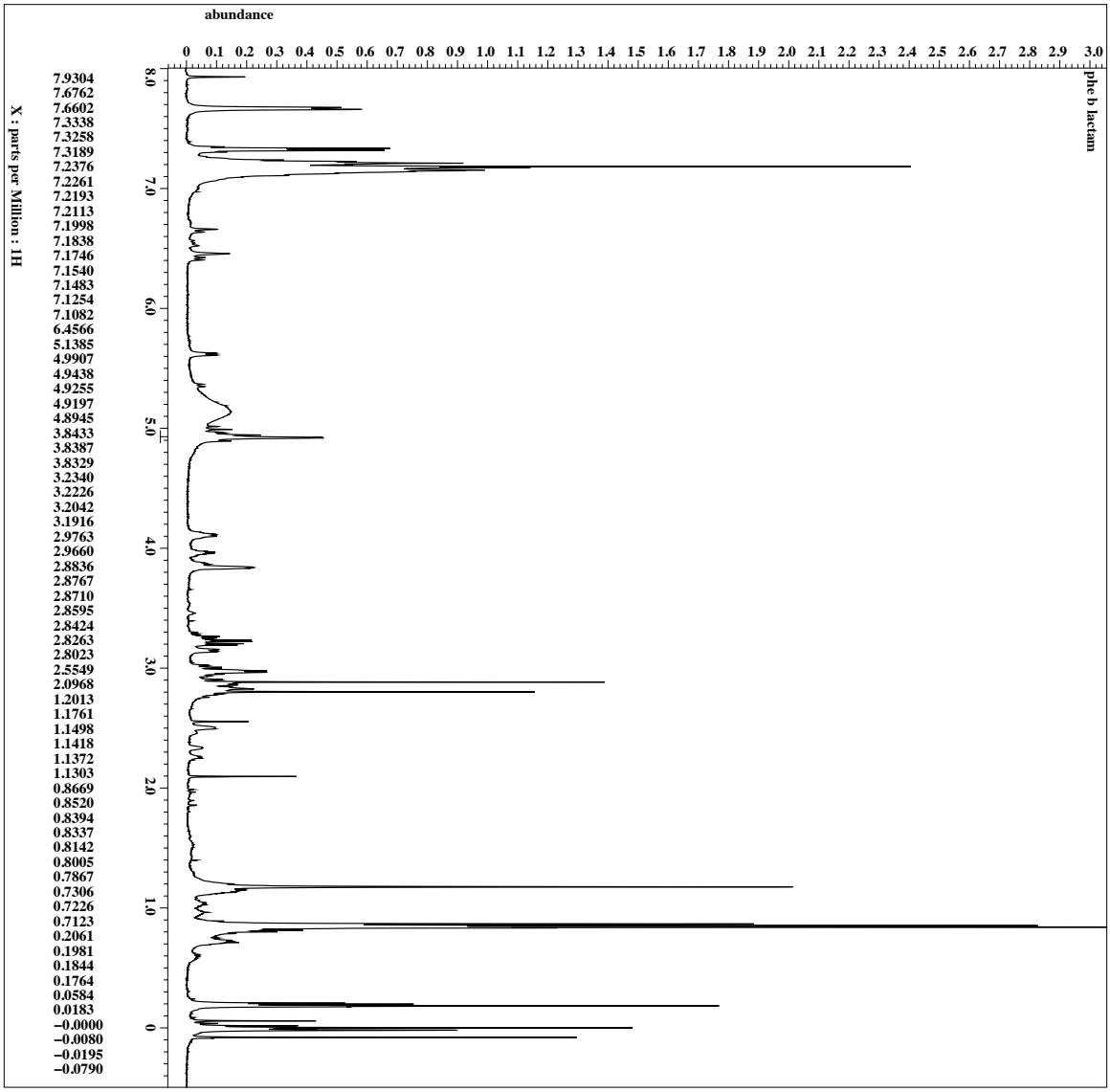
NMR

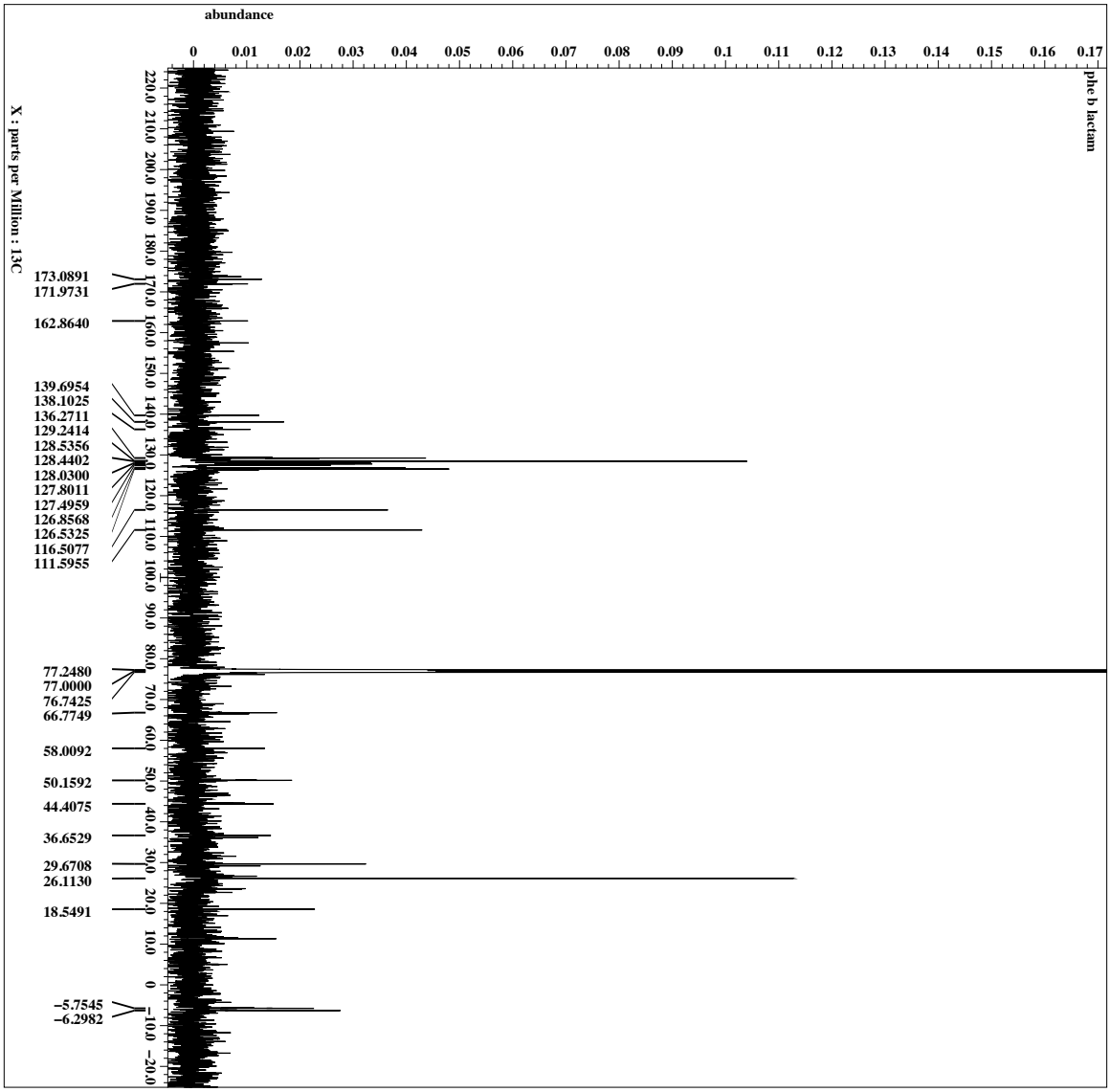
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

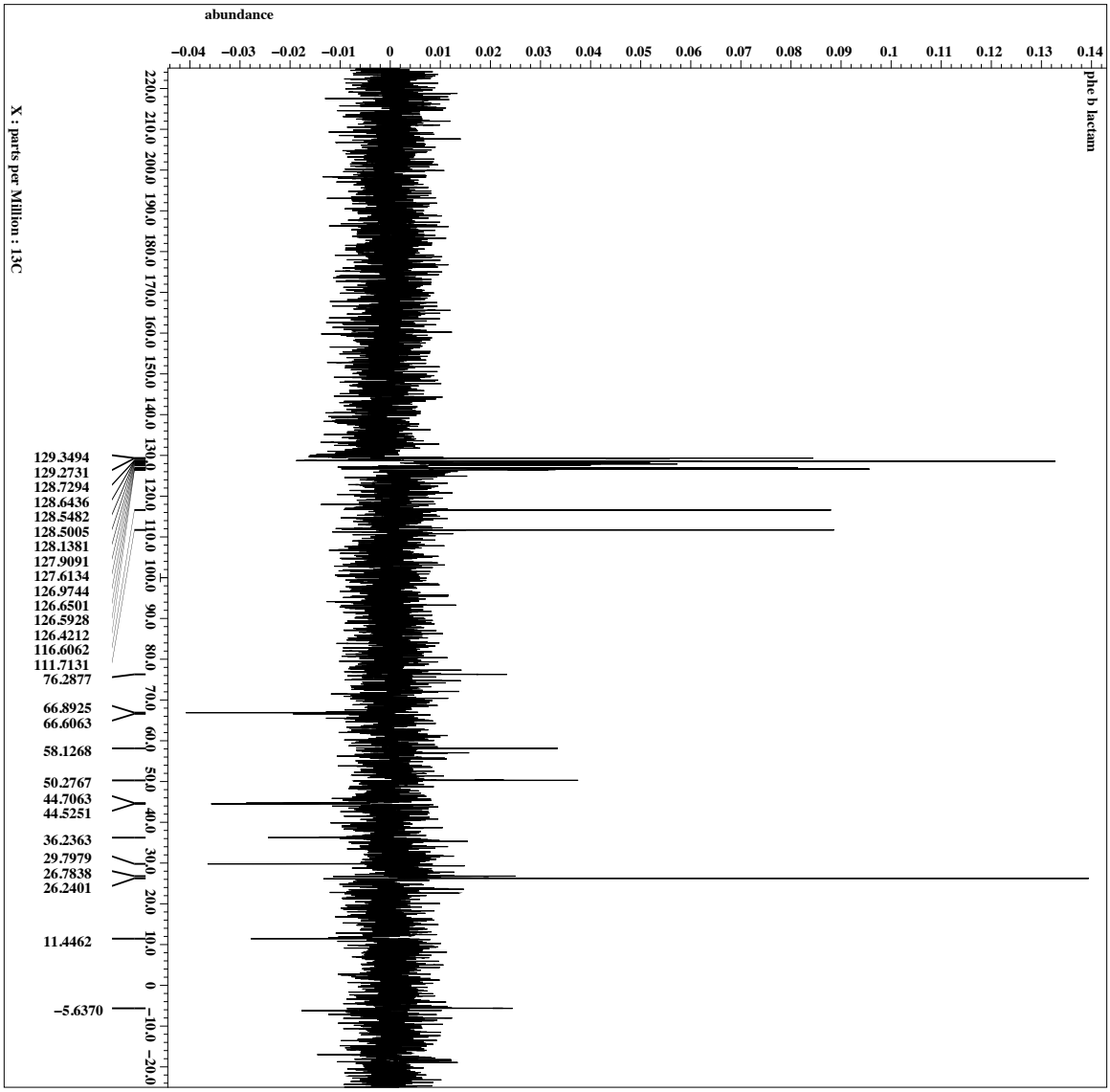
HMQC ($CDCl_3$)

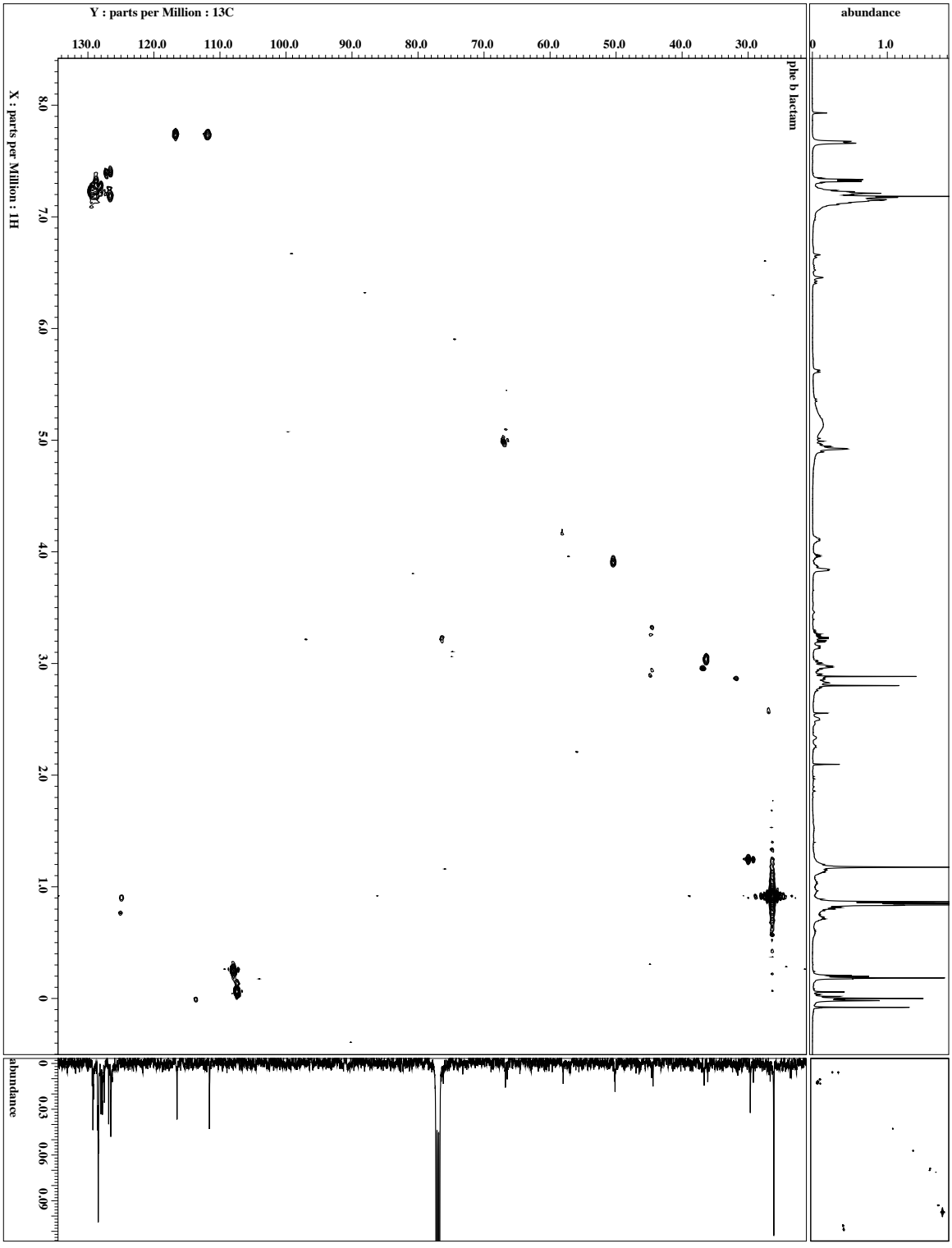

abundance
phe b lactam
X : parts per Million : 1H
7.9304
7.6762
7.6602
7.3338
7.3258
7.3189
7.2376
7.2261
7.2193
7.2113
7.1998
7.1838
7.1746
7.1540
7.1483
7.1254
7.1082
6.4566
5.1385
4.9907
4.9438
4.9255
4.9197
4.8945
3.8433
3.8387
3.8329
3.2340
3.2226
3.2042
3.1916
2.9763
2.9660
2.8836
2.8767
2.8710
2.8595
2.8424
2.8263
2.8023
2.5549
2.0968
1.2013
1.1761
1.1498
1.1418
1.1372
1.1303
0.8669
0.8520
0.8394
0.8337
0.8142
0.8005
0.7867
0.7306
0.7226
0.7123
0.2061
0.1981
0.1844
0.1764
0.0584
0.0183
−0.0000
−0.0080
−0.0195
−0.0790
Filename = NKD_ivp97B_PROTON-6.j
Sample_id = NKD_ivp97B
Machine = mt-eu500ssp
Creation_time = 2-MAR-2016 16:13:26
Comment = phe b lactam
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 64
Total_scans = 64
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.4[dC]



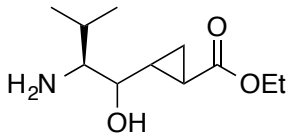
JEOL

Filename	= NKD_ivp97B_CARBON-7.j
Sample_id	= NKD_ivp97B
Machine	= mt-eu500pp
Creation_time	= 2-MAR-2016 17:02:19
Comment	= phe b lactam
Field_strength	= 11.7473579[T] (500[MH
X_acq_duration	= 0.83361792[s]
X_domain	= 13C
X_freq	= 125.76529768[MHz]
X_offset	= 100[ppm]
X_points	= 32768
X_prescans	= 4
X_resolution	= 1.19959034[Hz]
X_sweep	= 39.3081761[kHz]
Irr_domain	= 1H
Irr_freq	= 500.15991521[MHz]
Irr_offset	= 5.0[ppm]
Clipped	= TRUE
Mod_return	= 1
Scans	= 1024
Total_scans	= 1024
X_90_width	= 9.65[us]
X_acq_time	= 0.83361792[s]
X_angle	= 30[deg]
X_atn	= 9[dB]
X_pulse	= 3.21666667[us]
Irr_atn_dec	= 21.5[dB]
Irr_atn_noe	= 21.5[dB]
Irr_noise	= WALTZ
Decoupling	= TRUE
Initial_wait	= 1[s]
Noe	= TRUE
Noe_time	= 2[s]
Relaxation_delay	= 2[s]
Repetition_time	= 2.83361792[s]
Temp_state	= TEMP OFF
Temp_set	= 25[dC]
Temp_get	= 23[dC]


abundance
phe b lactam
X : parts per Million : 13C
129.3494
129.2731
128.7294
128.6436
128.5482
128.5005
128.1381
127.9091
127.6134
126.9744
126.6501
126.5928
126.4212
116.6062
111.7131
76.2877
66.8925
66.6063
58.1268
50.2767
44.7063
44.5251
36.2363
29.7979
26.7838
26.2401
11.4462
−5.6370
Filename = NKD_ivp97B_DEPT135-6.
Sample_id = NKD_ivp97B
Machine = mt-su500ap
Creation_time = 3-MAR-2016 00:34:07
Comment = phe b lactam
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 328
Total_scans = 328
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 9.65[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.7[dC]


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
X : parts per Million : 1H
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
phe b lactam
abundance
0
1.0
abundance
0
0.03
0.06
0.09

Ethyl 2-((S)-2-amino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylate (33)



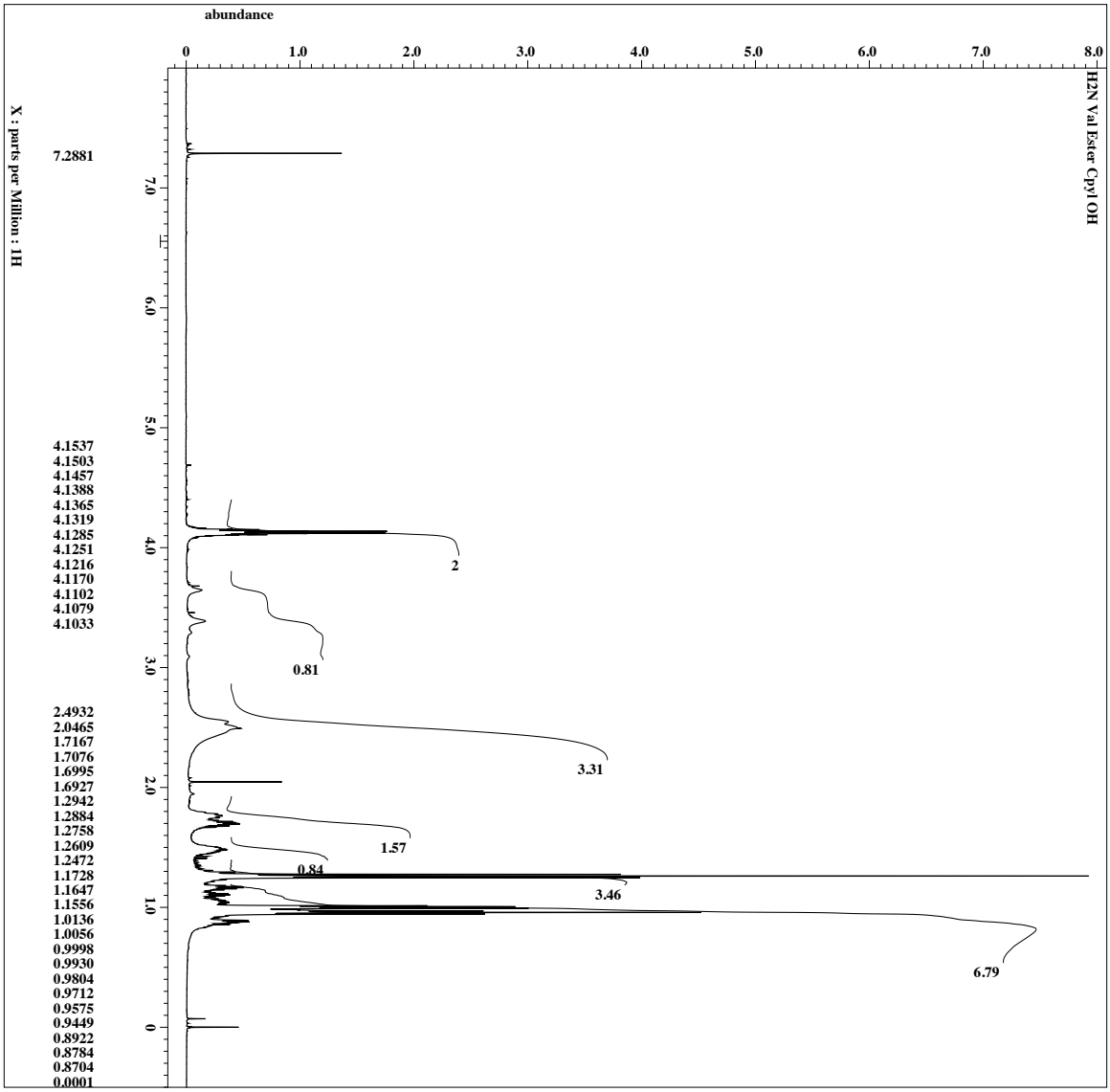
NMR

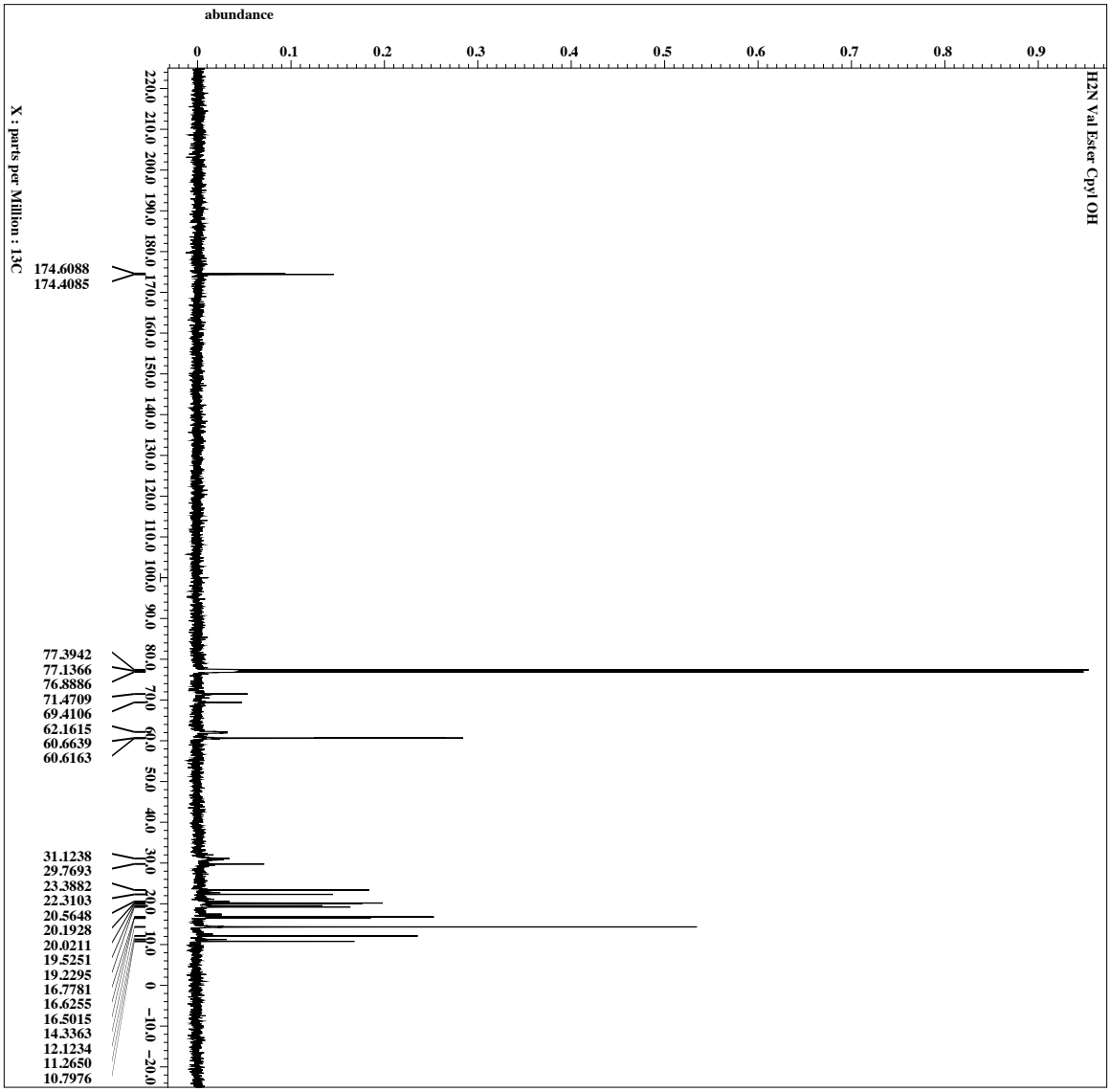
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

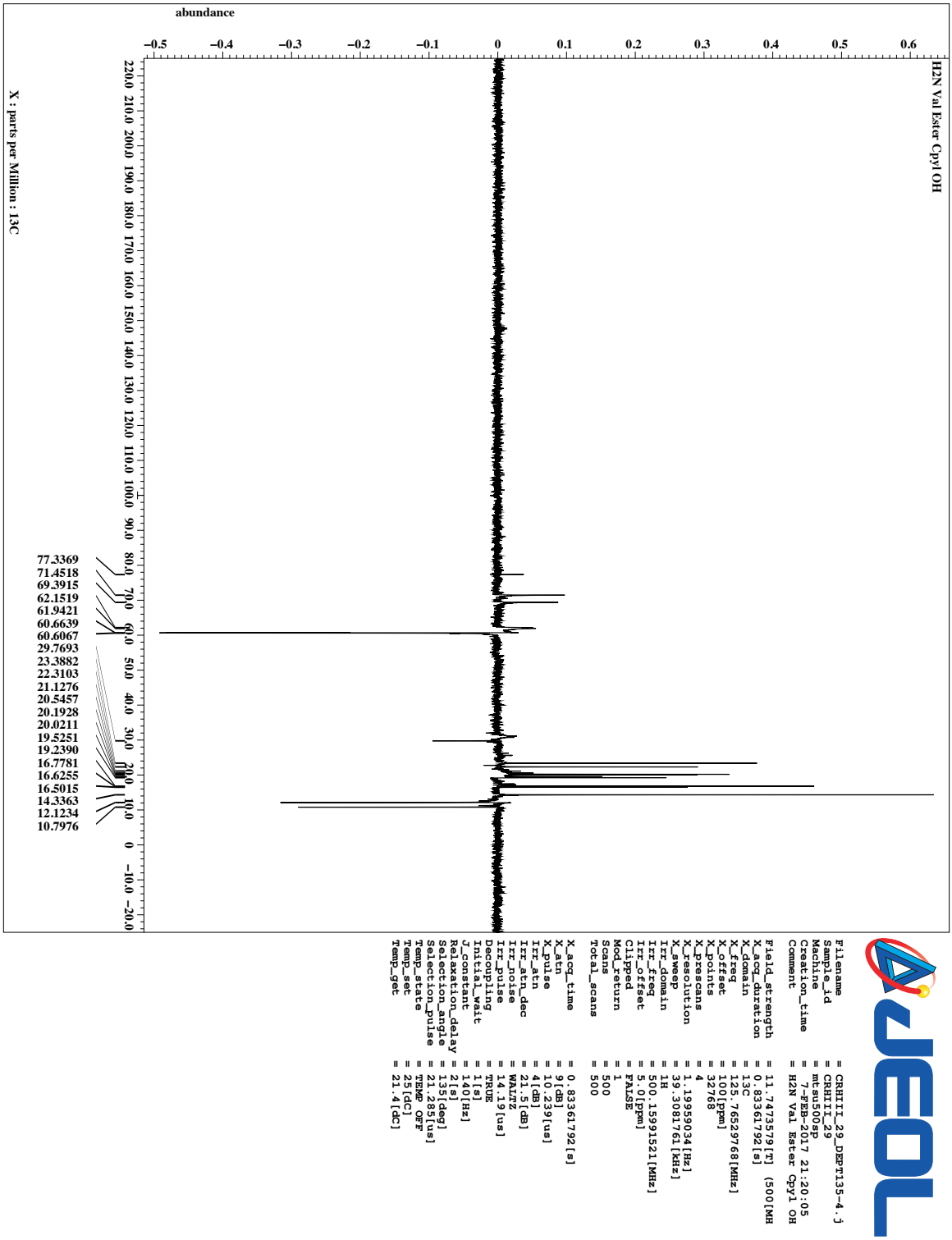
$DEPT_{135}$ ($CDCl_3$)

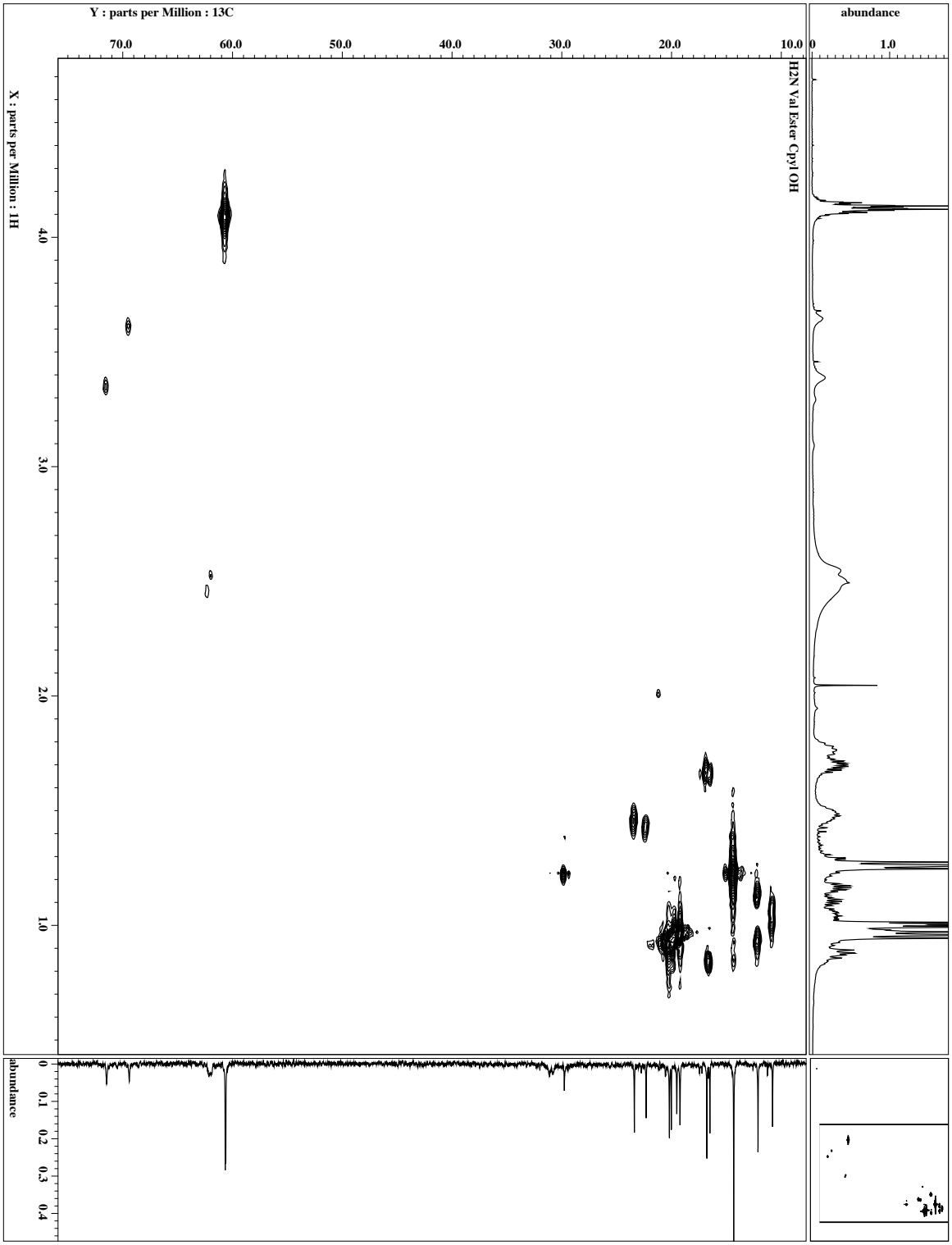
HMQC ($CDCl_3$)


abundance
H2N Val Ester Cpyl OH
X : parts per Million : 1H
7.2881
4.1537
4.1503
4.1457
4.1388
4.1365
4.1319
4.1285
4.1251
4.1216
4.1170
4.1102
4.1079
4.1033
2.4932
2.0465
1.7167
1.7076
1.6995
1.6927
1.2942
1.2884
1.2758
1.2609
1.2472
1.1728
1.1647
1.1556
1.0136
1.0056
0.9998
0.9930
0.9804
0.9712
0.9575
0.9449
0.8922
0.8784
0.8704
0.0001
2
0.81
3.31
1.57
0.84
3.46
6.79
Filename = CRHIII_29_PROTON-9.jd
Sample_id = CRHIII_29
Machine = mt-su500dp
Creation_time = 7-FEB-2017 20:27:44
Comment = H2N Val Ester Cpyl OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.1599152[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.1599152[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.1599152[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.3[dC]

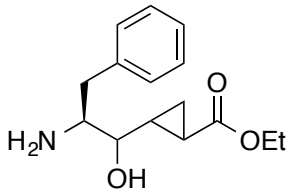



```
Filename                = CRHIII_29_CARBON-5.jd
Sample_id               = CRHIII_29
Machine                 = mt-su500-pp
Creation_time           = 7-FEB-2017 20:55:49
Comment                 = H2N Val Ester Cpyl OH

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 0.83361792[s]
X_domain                = 13C
X_freq                  = 125.76529768[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 1.19959034[Hz]
X_sweep                 = 39.3081761[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 500
Total_scans             = 500

X_90_width              = 10.239[us]
X_acq_time              = 0.83361792[s]
X_angle                 = 30[deg]
X_atn                   = 9[dB]
X_pulse                 = 3.413[us]
Irr_atn_dec             = 21.5[dB]
Irr_atn_noe             = 21.5[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 2.83361792[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 21.7[dC]
```


H2N Val Ester Cpyl OH
abundance
X : parts per Million : 13C
77.3369
71.4518
69.3915
62.1519
61.9421
60.6639
60.6067
29.7693
23.3882
22.3103
21.1276
20.5457
20.1928
20.0211
19.5251
19.2390
16.7781
16.6255
16.5015
14.3363
12.1234
10.7976
Filename = CRHIII_29_DEPT135-4.j
Sample_id = CRHIII_29
Machine = mt-su500dp
Creation_time = 7-FEB-2017 21:20:05
Comment = H2N Val Ester Cpyl OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 500
Total_scans = 500
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 13C
70.0
60.0
50.0
40.0
30.0
20.0
10.0
abundance
0
1.0
X : parts per Million : 1H
4.0
3.0
2.0
1.0
H2N Val Ester Cpyl OH
abundance
0
0.1
0.2
0.3
0.4

Ethyl 2-((S)-2-amino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylate (35)

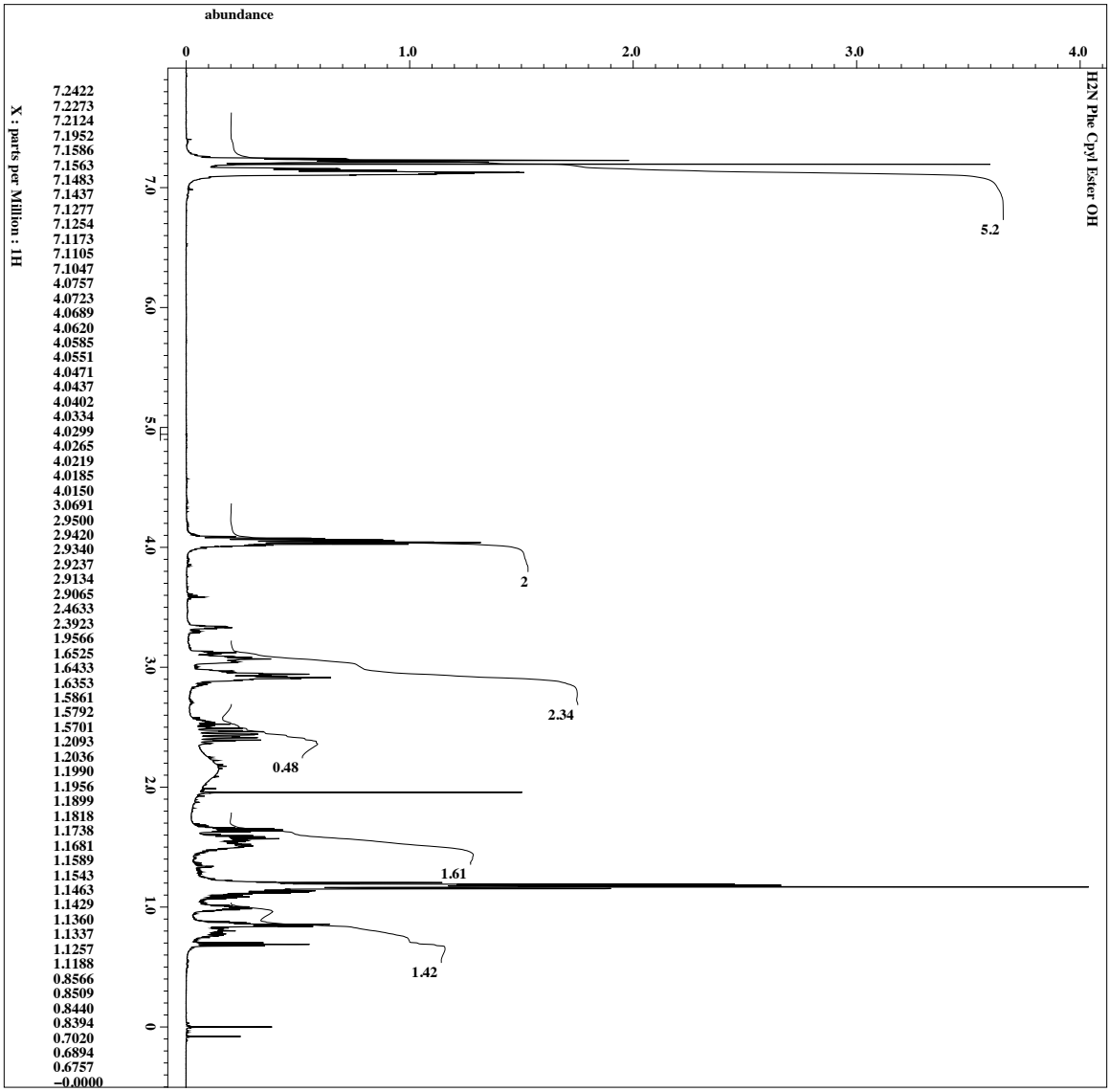


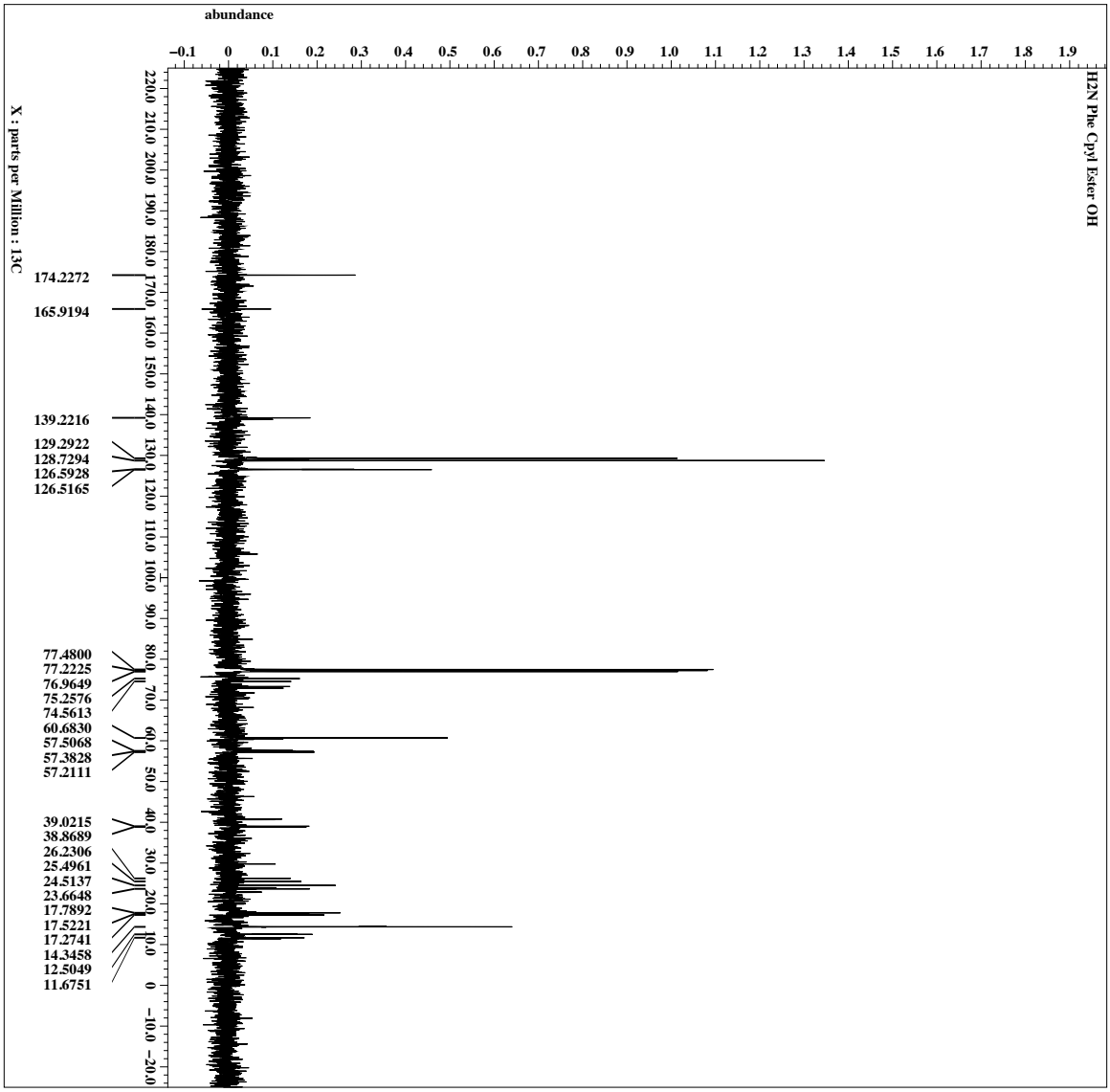
NMR

^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

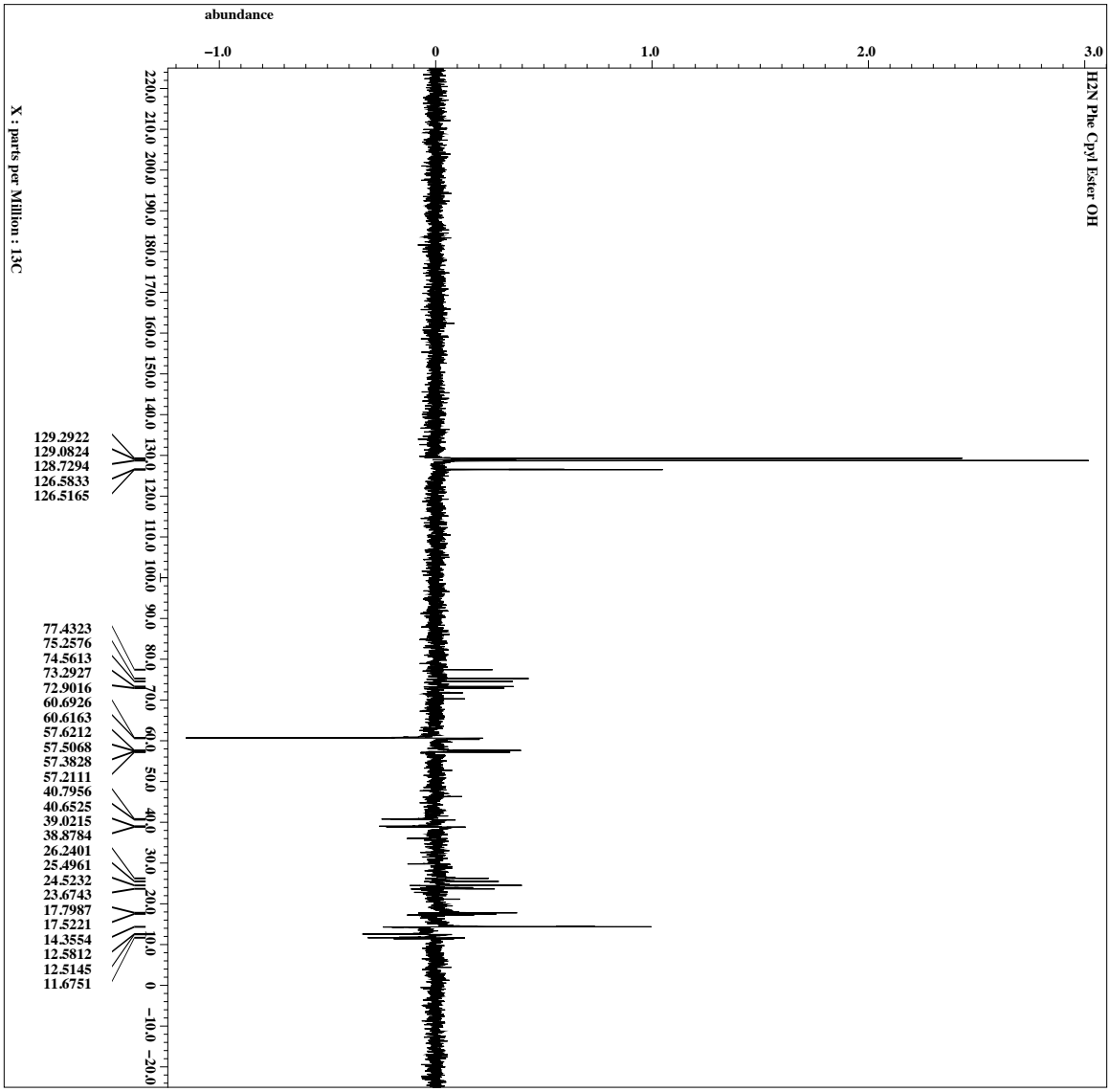
$DEPT_{135}$ ($CDCl_3$)


H2N Phe Cpyl Ester OH
abundance
0
1.0
2.0
3.0
4.0
5.2
2
2.34
0.48
1.61
1.42
X : parts per Million : 1H
7.2422
7.2273
7.2124
7.1952
7.1586
7.1563
7.1483
7.1437
7.1277
7.1254
7.1173
7.1105
7.1047
4.0757
4.0723
4.0689
4.0620
4.0585
4.0551
4.0471
4.0437
4.0402
4.0334
4.0299
4.0265
4.0219
4.0185
4.0150
3.0691
2.9500
2.9420
2.9340
2.9237
2.9134
2.9065
2.4633
2.3923
1.9566
1.6525
1.6433
1.6353
1.5861
1.5792
1.5701
1.2093
1.2036
1.1990
1.1956
1.1899
1.1818
1.1738
1.1681
1.1589
1.1543
1.1463
1.1429
1.1360
1.1337
1.1257
1.1188
0.8566
0.8509
0.8440
0.8394
0.7020
0.6894
0.6757
-0.0000
Filename = CRHIII_15_PROTON-5.jd
Sample_id = CRHIII_15
Machine = mt-su500ap
Creation_time = 26-JAN-2017 12:12:24
Comment = H2N Phe Cpyl Ester OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.2[dC]

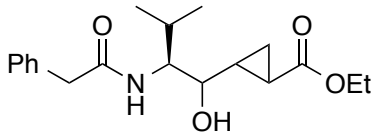


JEOL

Filename	= CRHIII_15_CARBON-4.jd
Sample_id	= CRHIII_15
Machine	= mt-su500ap
Creation_time	= 26-JAN-2017 12:13:49
Comment	= H2N Phe Cpyl Ester OH
Field_strength	= 11.7473579[T] (500[MH
X_acq_duration	= 0.83361792[s]
X_domain	= 13C
X_freq	= 125.76529768[MHz]
X_offset	= 100[ppm]
X_points	= 32768
X_prescans	= 4
X_resolution	= 1.19959034[Hz]
X_sweep	= 39.3081761[kHz]
Irr_domain	= 1H
Irr_freq	= 500.15991521[MHz]
Irr_offset	= 5.0[ppm]
Clipped	= FALSE
Mod_return	= 1
Scans	= 14
Total_scans	= 14
X_90_width	= 10.239[us]
X_acq_time	= 0.83361792[s]
X_angle	= 30[deg]
X_atn	= 9[dB]
X_pulse	= 3.413[us]
Irr_atn_dec	= 21.5[dB]
Irr_atn_noe	= 21.5[dB]
Irr_noise	= WALTZ
Decoupling	= TRUE
Initial_wait	= 1[s]
Noe	= TRUE
Noe_time	= 2[s]
Relaxation_delay	= 2[s]
Repetition_time	= 2.83361792[s]
Temp_state	= TEMP OFF
Temp_set	= 25[dC]
Temp_get	= 21.4[dC]


H2N Phe Cpyl Ester OH
abundance
X : parts per Million : 13C
129.2922
129.0824
128.7294
126.5833
126.5165
77.4323
75.2576
74.5613
73.2927
72.9016
60.6926
60.6163
57.6212
57.5068
57.3828
57.2111
40.7956
40.6525
39.0215
38.8784
26.2401
25.4961
24.5232
23.6743
17.7987
17.5221
14.3554
12.5812
12.5145
11.6751
Filename = CRHIII_15_DEPT135-5.j
Sample_id = CRHIII_15
Machine = mt-su500ap
Creation_time = 26-JAN-2017 12:15:06
Comment = H2N Phe Cpyl Ester OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.3[dC]

Ethyl 2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylate (36)



NMR

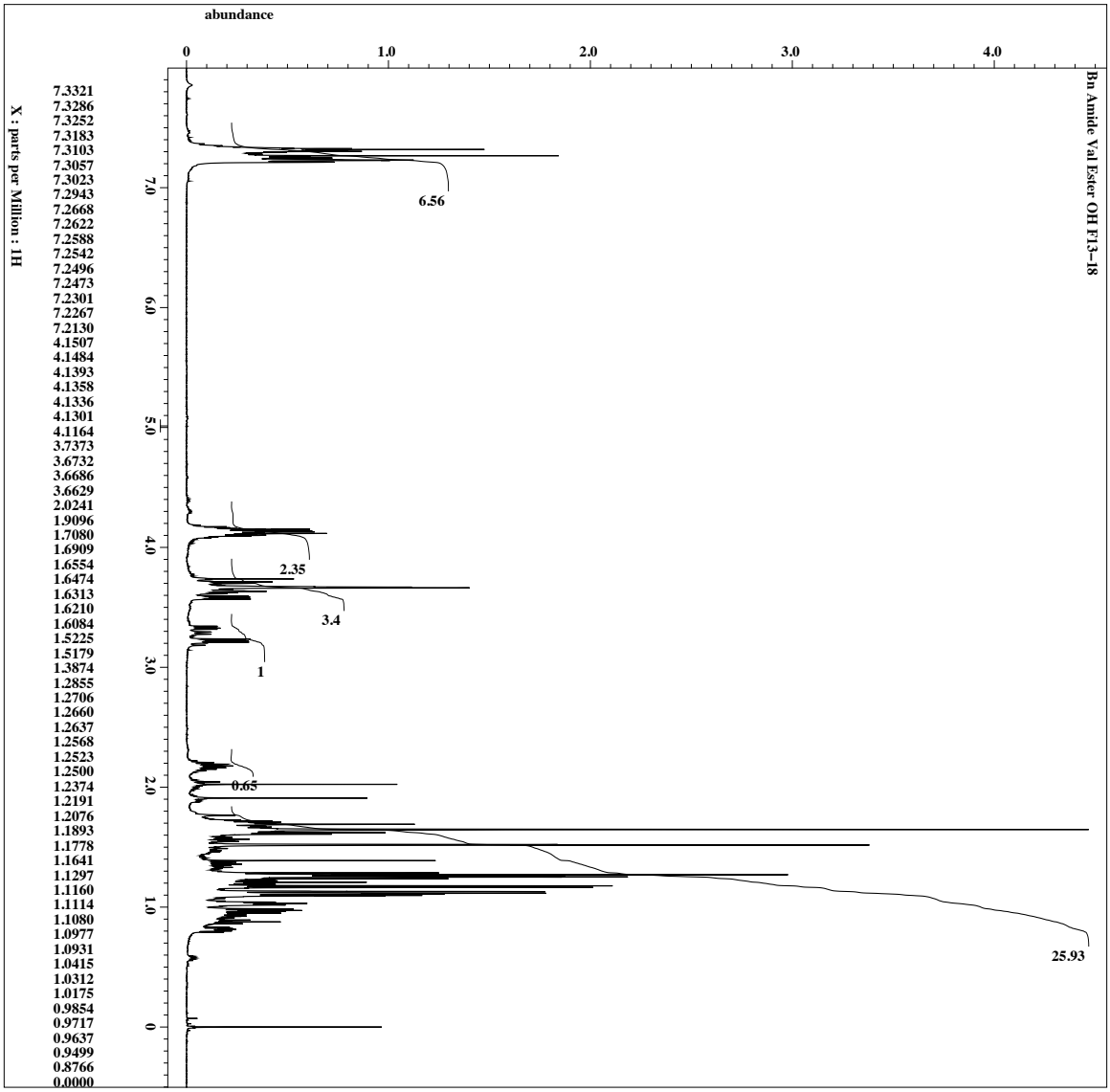
^{1}H-NMR ($CDCl_3$)

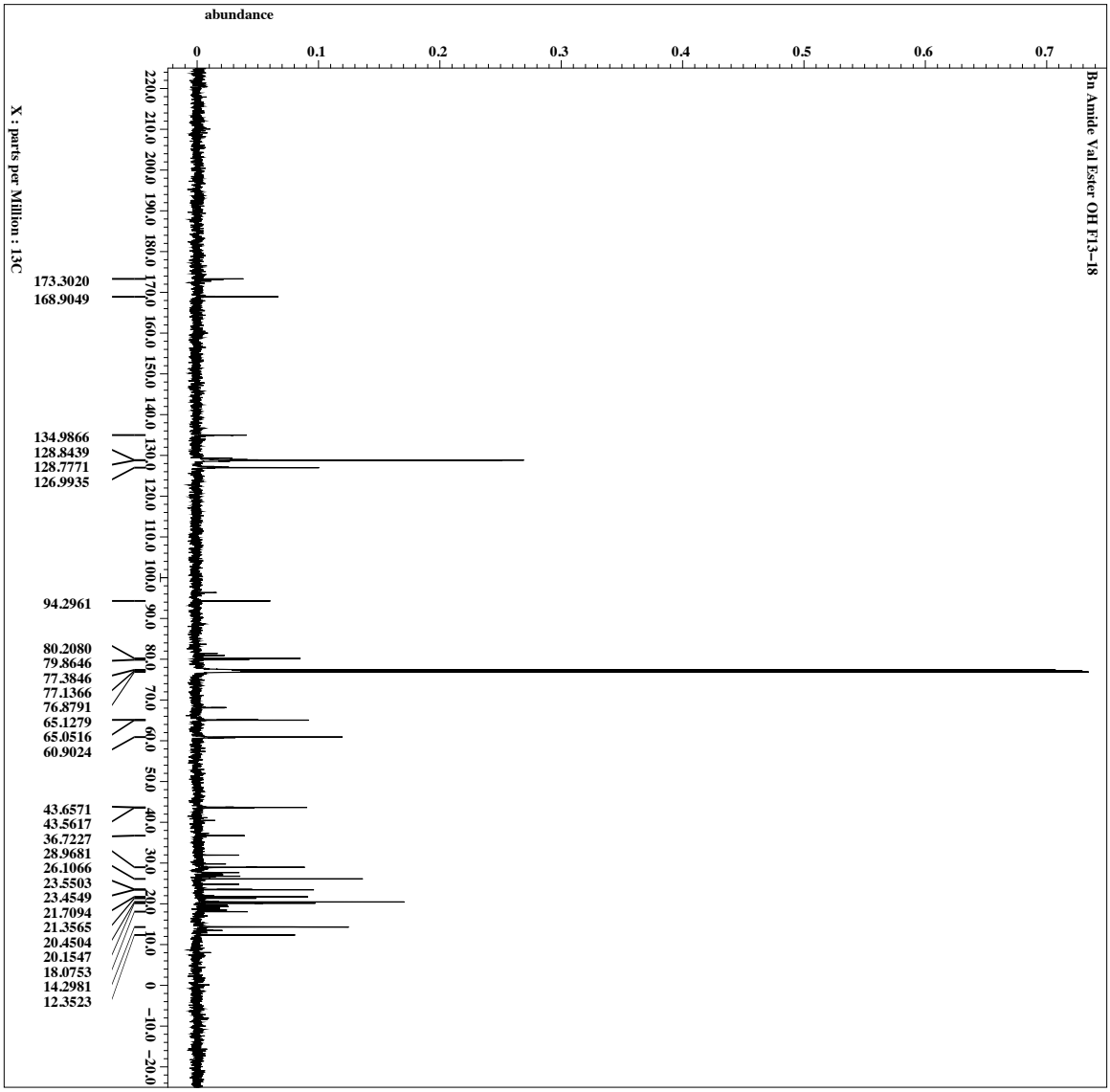
^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

COSY ($CDCl_3$)

HMQC ($CDCl_3$)

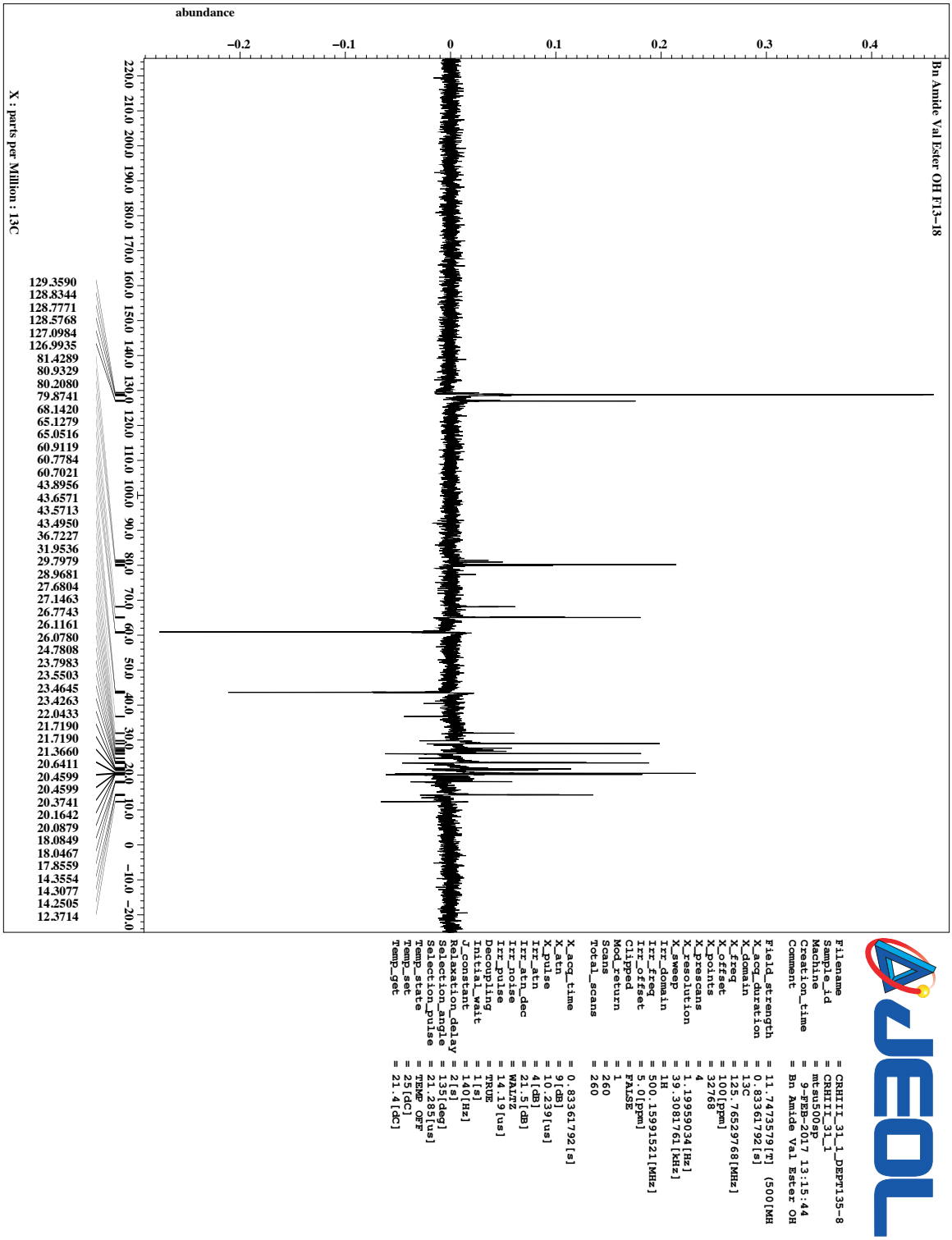

Bn Amide Val Ester OH F13-18
abundance
0
1.0
2.0
3.0
4.0
6.56
2.35
3.4
1
0.65
25.93
X : parts per Million : 1H
7.3321
7.3286
7.3252
7.3183
7.3103
7.3057
7.3023
7.2943
7.2668
7.2622
7.2588
7.2542
7.2496
7.2473
7.2301
7.2267
7.2130
4.1507
4.1484
4.1393
4.1358
4.1336
4.1301
4.1164
3.7373
3.6732
3.6686
3.6629
2.0241
1.9096
1.7080
1.6909
1.6554
1.6474
1.6313
1.6210
1.6084
1.5225
1.5179
1.3874
1.2855
1.2706
1.2660
1.2637
1.2568
1.2523
1.2500
1.2374
1.2191
1.2076
1.1893
1.1778
1.1641
1.1297
1.1160
1.1114
1.1080
1.0977
1.0931
1.0415
1.0312
1.0175
0.9854
0.9717
0.9637
0.9499
0.8766
0.0000
Filename = CRHIII_31_1_PROTON-5.
Sample_id = CRHIII_31_1
Machine = mt-su500pp
Creation_time = 9-FEB-2017 12:38:30
Comment = Bn Amide Val Ester OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38384438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.3[dC]

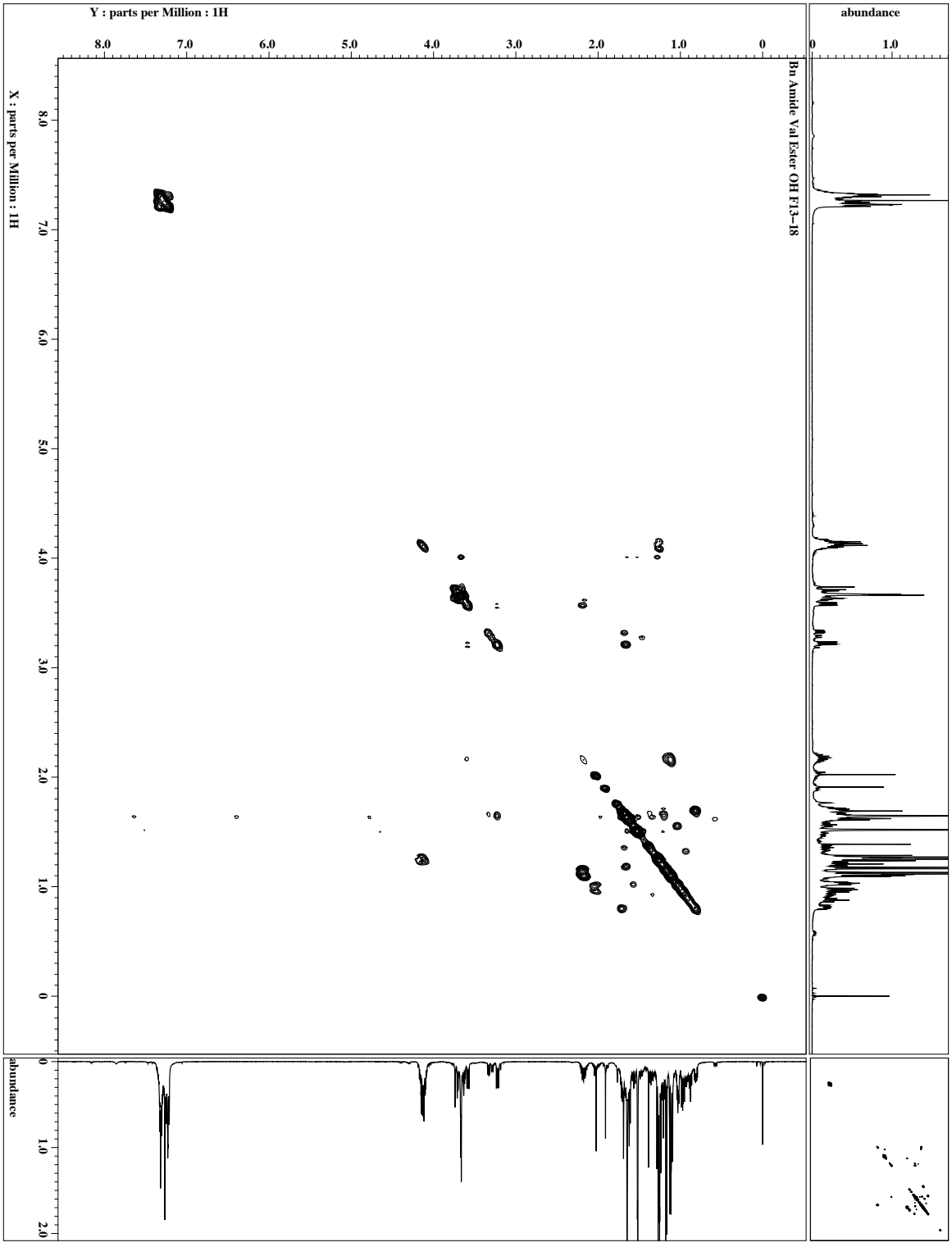


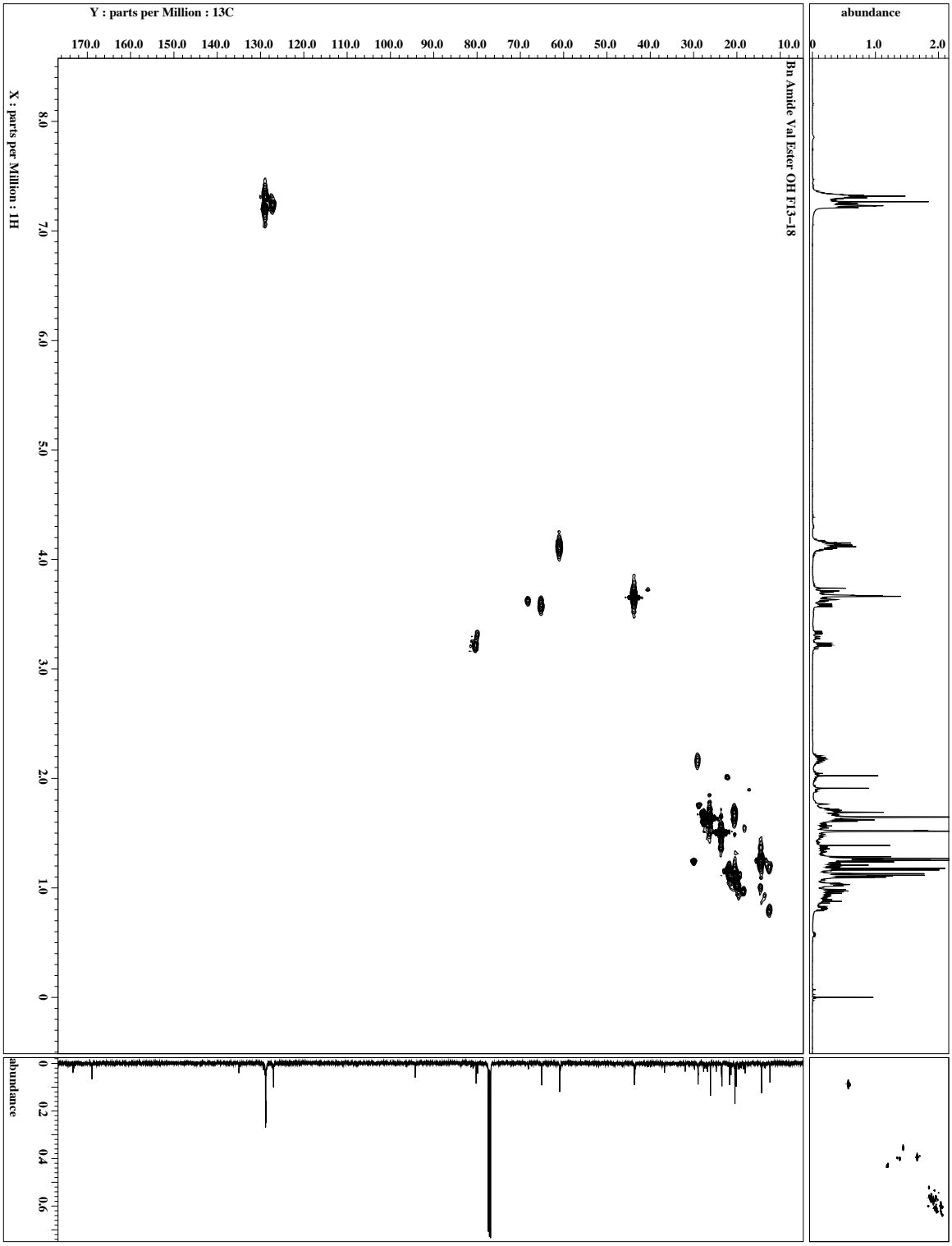
Filename = CRHIII_31_1_CARBON-18
Sample_id = CRHIII_31_1
Machine = mt-su500ap
Creation_time = 9-FEB-2017 13:02:51
Comment = Bn Amide Val Ester OH

Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = TRUE
Mod_return = 1
Scans = 500
Total_scans = 500

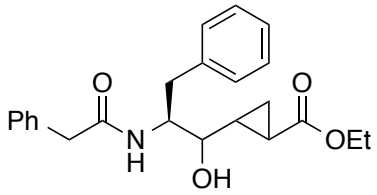
X_90_width = 10.239[us]
X_acq_time = 0.83361792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.83361792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.6[dC]


Bn Amide Val Ester OH F13-18
abundance
X : parts per Million : 13C
129.3590
128.8344
128.7771
128.5768
127.0984
126.9935
81.4289
80.9329
80.2080
79.8741
68.1420
65.1279
65.0516
60.9119
60.7784
60.7021
43.8956
43.6571
43.5713
43.4950
36.7227
31.9536
29.7979
28.9681
27.6804
27.1463
26.7743
26.1161
26.0780
24.7808
23.7983
23.5503
23.4645
23.4263
22.0433
21.7190
21.7190
21.3660
20.6411
20.4599
20.4599
20.3741
20.1642
20.0879
18.0849
18.0467
17.8559
14.3554
14.3077
14.2505
12.3714
Filename = CRHIII_31_1_DEPT135-8
Sample_id = CRHIII_31_1
Machine = mt-su500ap
Creation_time = 9-FEB-2017 13:15:44
Comment = Bn Amide Val Ester OH
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 260
Total_scans = 260
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 1H
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
abundance
Bn Amide Val Ester OH F13-18
X : parts per Million : 1H
abundance


Y : parts per Million : 13C
170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0
X : parts per Million : 1H
8.0 7.0 6.0 5.0 4.0 3.0 2.0 1.0 0
Bn Amide Val Ester OH F13–18
abundance
0 1.0 2.0
abundance
0 0.2 0.4 0.6

Ethyl 2-((S)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylate (38)



NMR

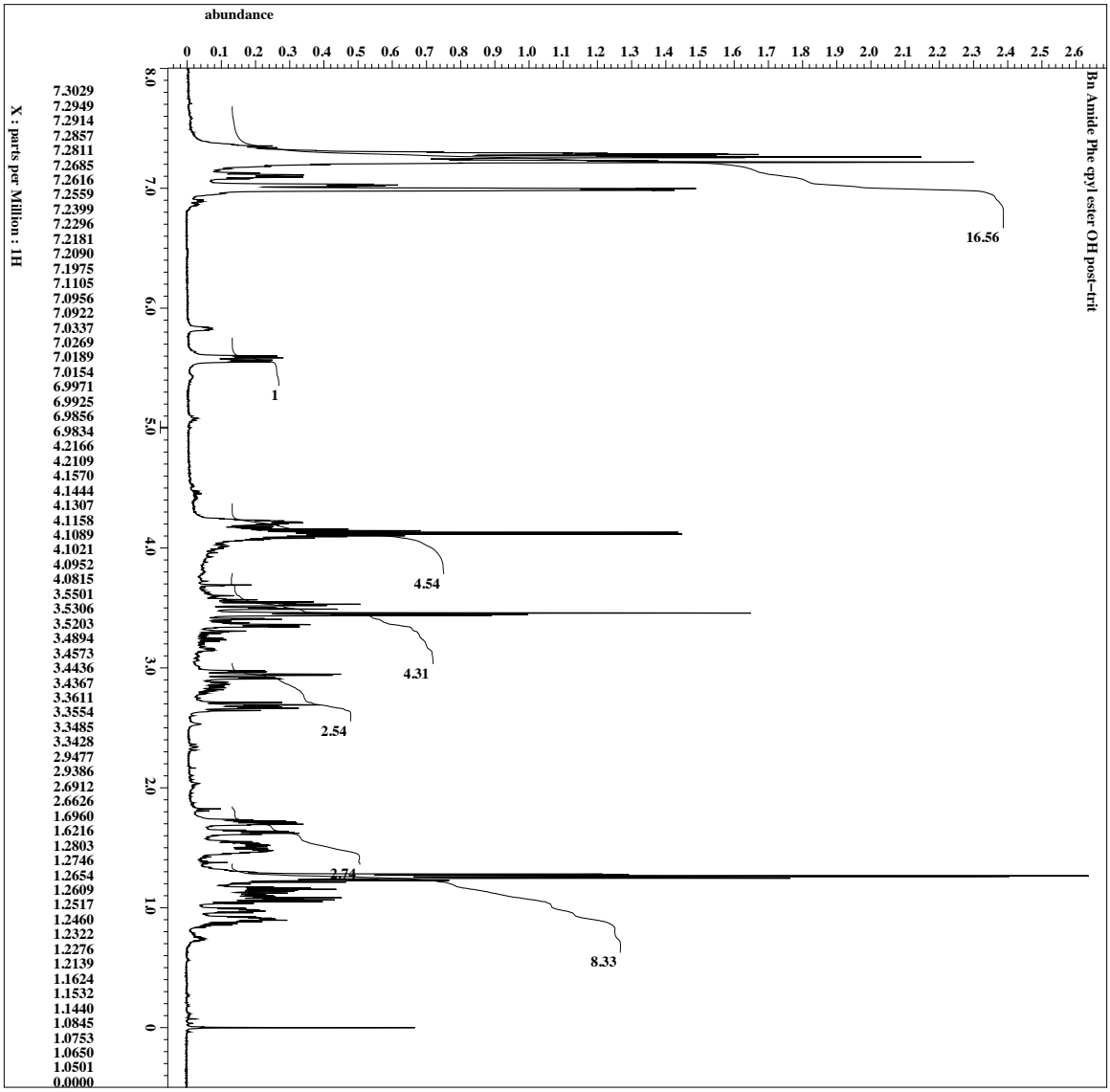
^{1}H-NMR ($CDCl_3$)

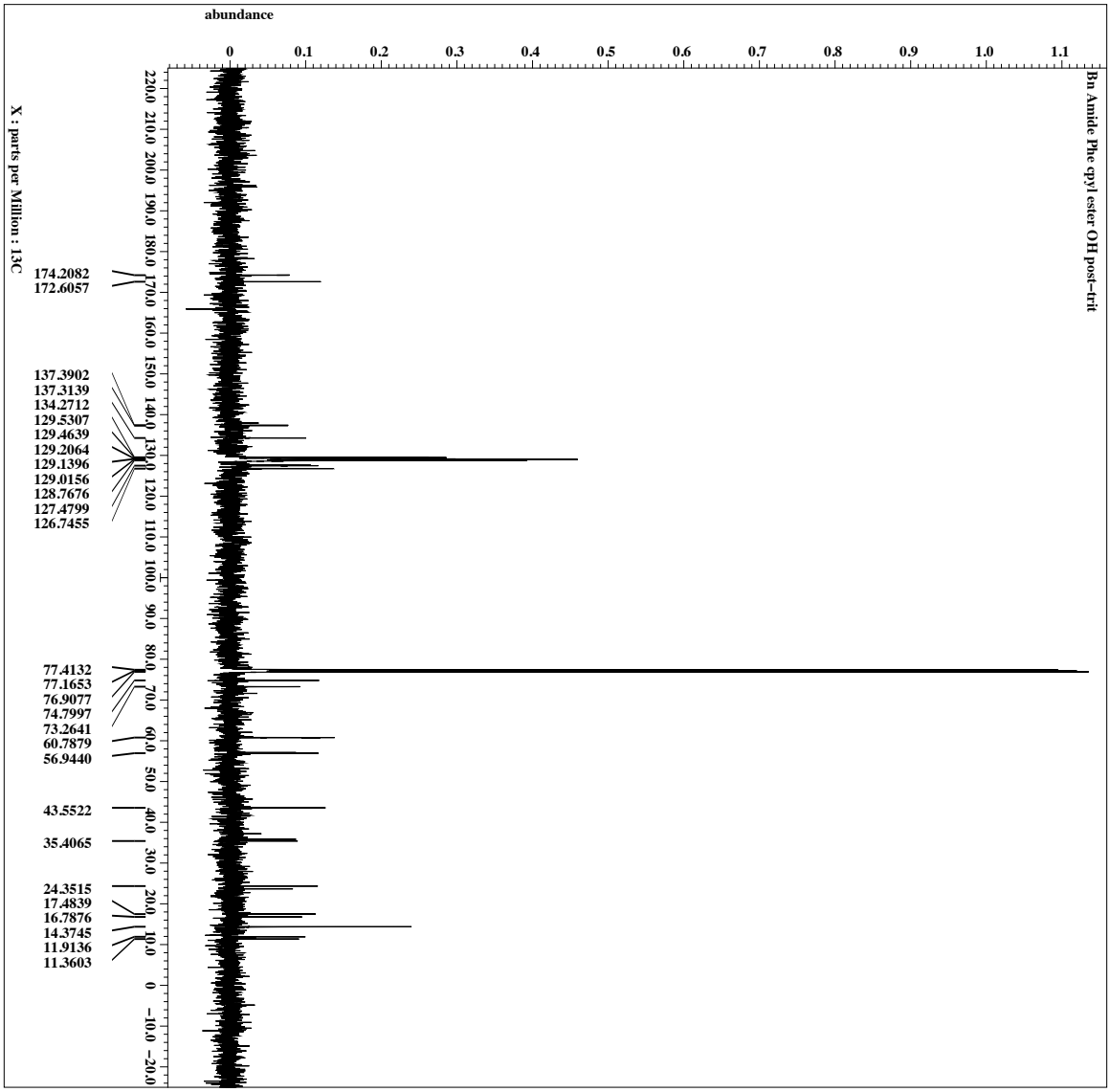
^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

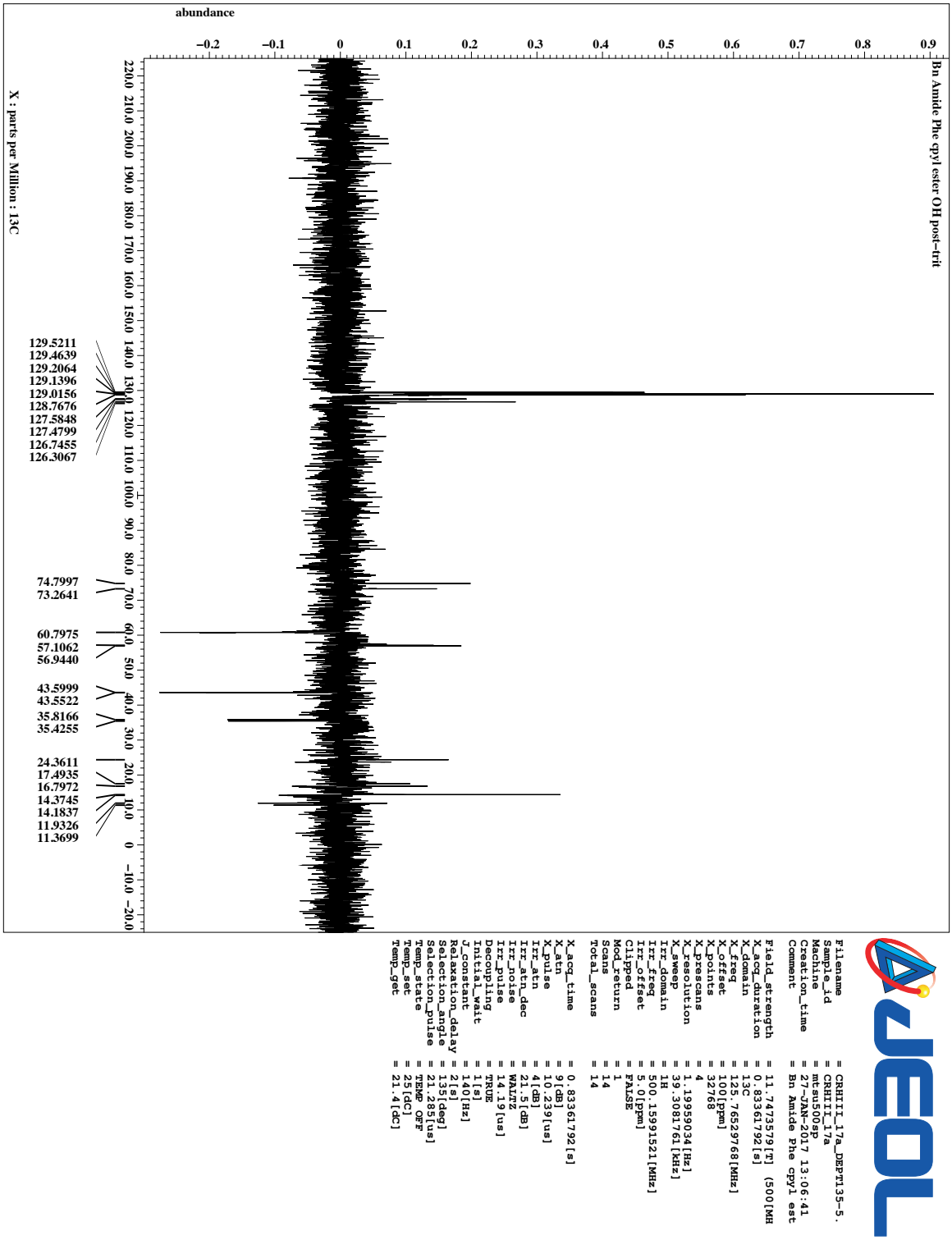
COSY ($CDCl_3$)

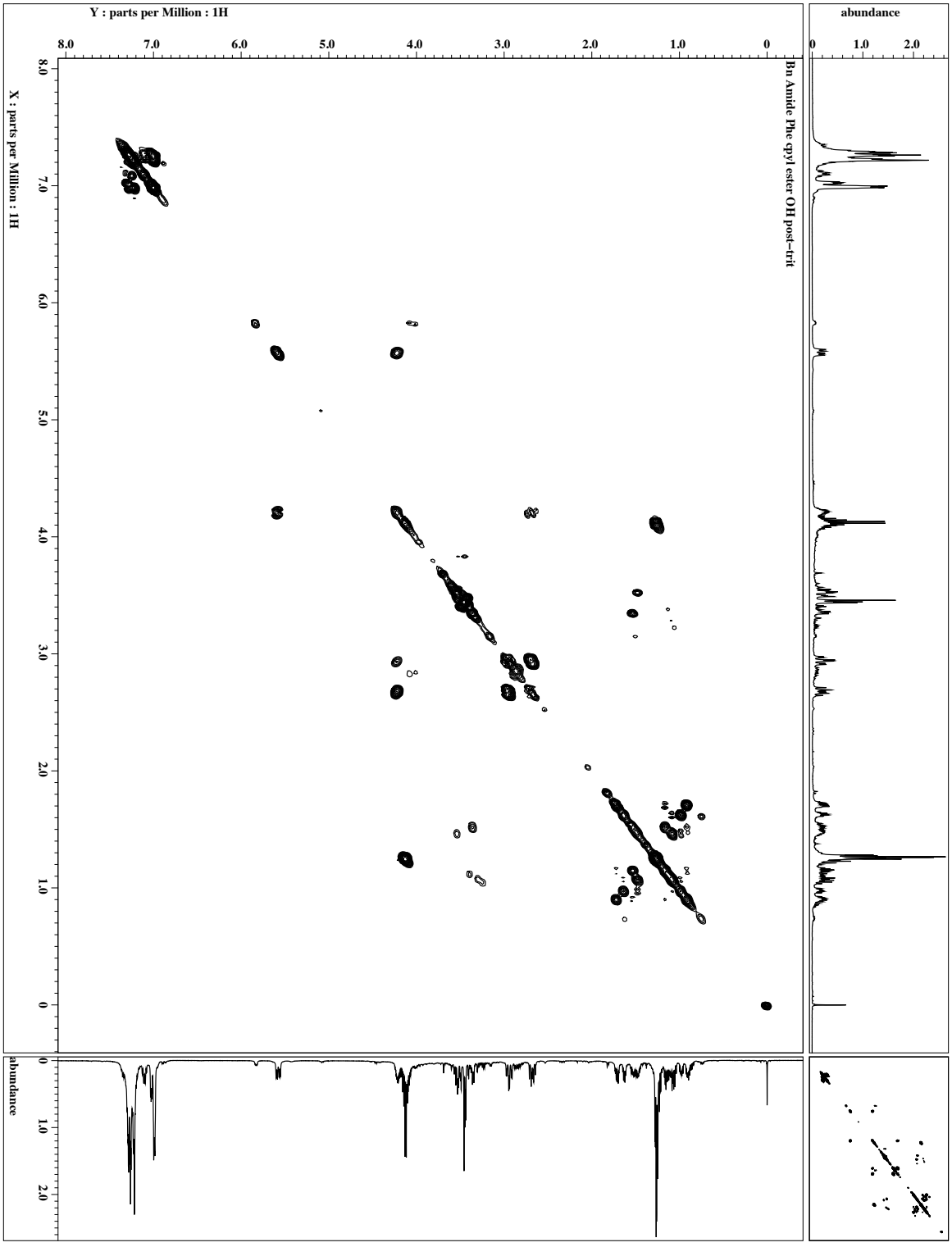
HMQC ($CDCl_3$)

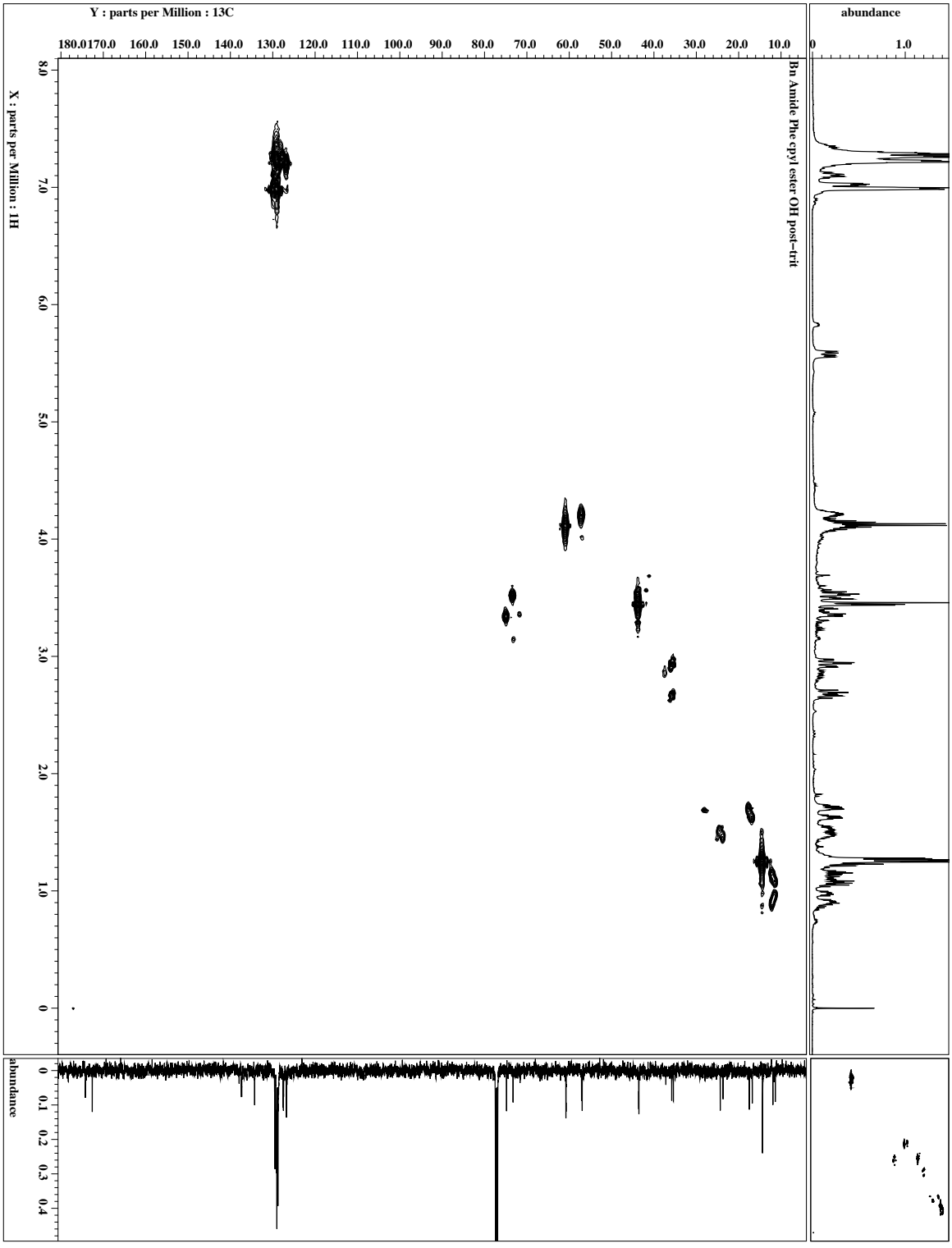

Bn Amide Phe cpyl ester OH post-trit
abundance
16.56
1
4.54
4.31
2.54
2.74
8.33
X : parts per Million : 1H
7.3029
7.2949
7.2914
7.2857
7.2811
7.2685
7.2616
7.2559
7.2399
7.2296
7.2181
7.2090
7.1975
7.1105
7.0956
7.0922
7.0337
7.0269
7.0189
7.0154
6.9971
6.9925
6.9856
6.9834
4.2166
4.2109
4.1570
4.1444
4.1307
4.1158
4.1089
4.1021
4.0952
4.0815
3.5501
3.5306
3.5203
3.4894
3.4573
3.4436
3.4367
3.3611
3.3554
3.3485
3.3428
2.9477
2.9386
2.6912
2.6626
1.6960
1.6216
1.2803
1.2746
1.2654
1.2609
1.2517
1.2460
1.2322
1.2276
1.2139
1.1624
1.1532
1.1440
1.0845
1.0753
1.0650
1.0501
0.0000
Filename = CRHIII_17a_PROTON-5.j
Sample_id = CRHIII_17a
Machine = mt-su500ap
Creation_time = 27-JAN-2017 13:00:45
Comment = Bn Amide Phe cpyl est
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.1[dC]



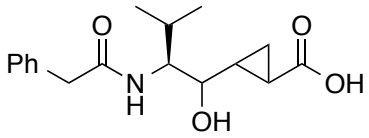
Filename	= CRHIII_17a_CARBON-5.j
Sample_id	= CRHIII_17a
Machine	= mt-su500ap
Creation_time	= 27-JAN-2017 13:05:28
Comment	= Bn Amide Phe cpyl est
Field_strength	= 11.7473579[T] (500[MH
x_acq_duration	= 0.83361792[s]
x_domain	= 13C
x_freq	= 125.76529768[MHz]
x_offset	= 100[ppm]
x_points	= 32768
x_prescans	= 4
x_resolution	= 1.19959034[Hz]
x_sweep	= 39.3081761[kHz]
Irr_domain	= 1H
Irr_freq	= 500.15991521[MHz]
Irr_offset	= 5.0[ppm]
Clipped	= FALSE
Mod_return	= 1
Scans	= 58
Total_scans	= 58
X_90_width	= 10.239[us]
X_acq_time	= 0.83361792[s]
X_angle	= 30[deg]
X_atn	= 9[dB]
X_pulse	= 3.413[us]
Irr_atn_dec	= 21.5[dB]
Irr_atn_noe	= 21.5[dB]
Irr_noise	= WALTZ
Decoupling	= TRUE
Initial_wait	= 1[s]
Noe	= TRUE
Noe_time	= 2[s]
Relaxation_delay	= 2[s]
Repetition_time	= 2.83361792[s]
Temp_state	= TEMP OFF
Temp_set	= 25[dC]
Temp_get	= 21.5[dC]


Bn Amide Phe cpyl ester OH post-trit
abundance
X : parts per Million : 13C
129.5211
129.4639
129.2064
129.1396
129.0156
128.7676
127.5848
127.4799
126.7455
126.3067
74.7997
73.2641
60.7975
57.1062
56.9440
43.5999
43.5522
35.8166
35.4255
24.3611
17.4935
16.7972
14.3745
14.1837
11.9326
11.3699
Filename = CRHIII_17a_DEPT135-5.
Sample_id = CRHIII_17a
Machine = mt-su500ap
Creation_time = 27-JAN-2017 13:06:41
Comment = Bn Amide Phe cpyl est
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
Bn Amide Phe epyl ester OH post-trit
abundance
abundance


Y : parts per Million : 13C
180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0
X : parts per Million : 1H
8.0 7.0 6.0 5.0 4.0 3.0 2.0 1.0 0
Bn Amide Phe cpyl ester OH post-trit
abundance
0 1.0
abundance
0 0.1 0.2 0.3 0.4

2-((*S*)-2-(benzylcarbonylamino)-1-hydroxy-3-methylbutyl) cyclopropanecarboxylic acid (39)



NMR

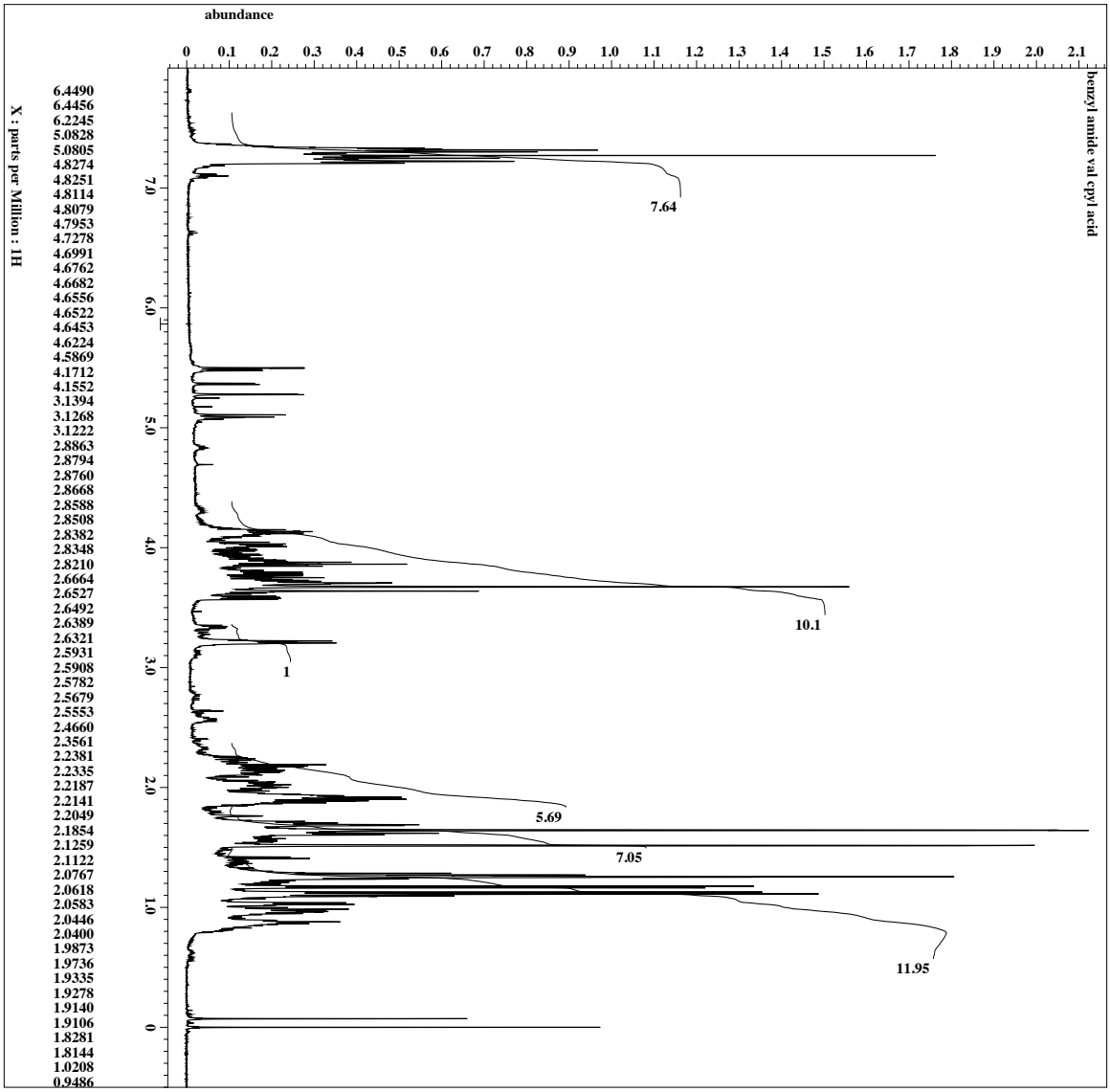
^{1}H-NMR ($CDCl_3$)

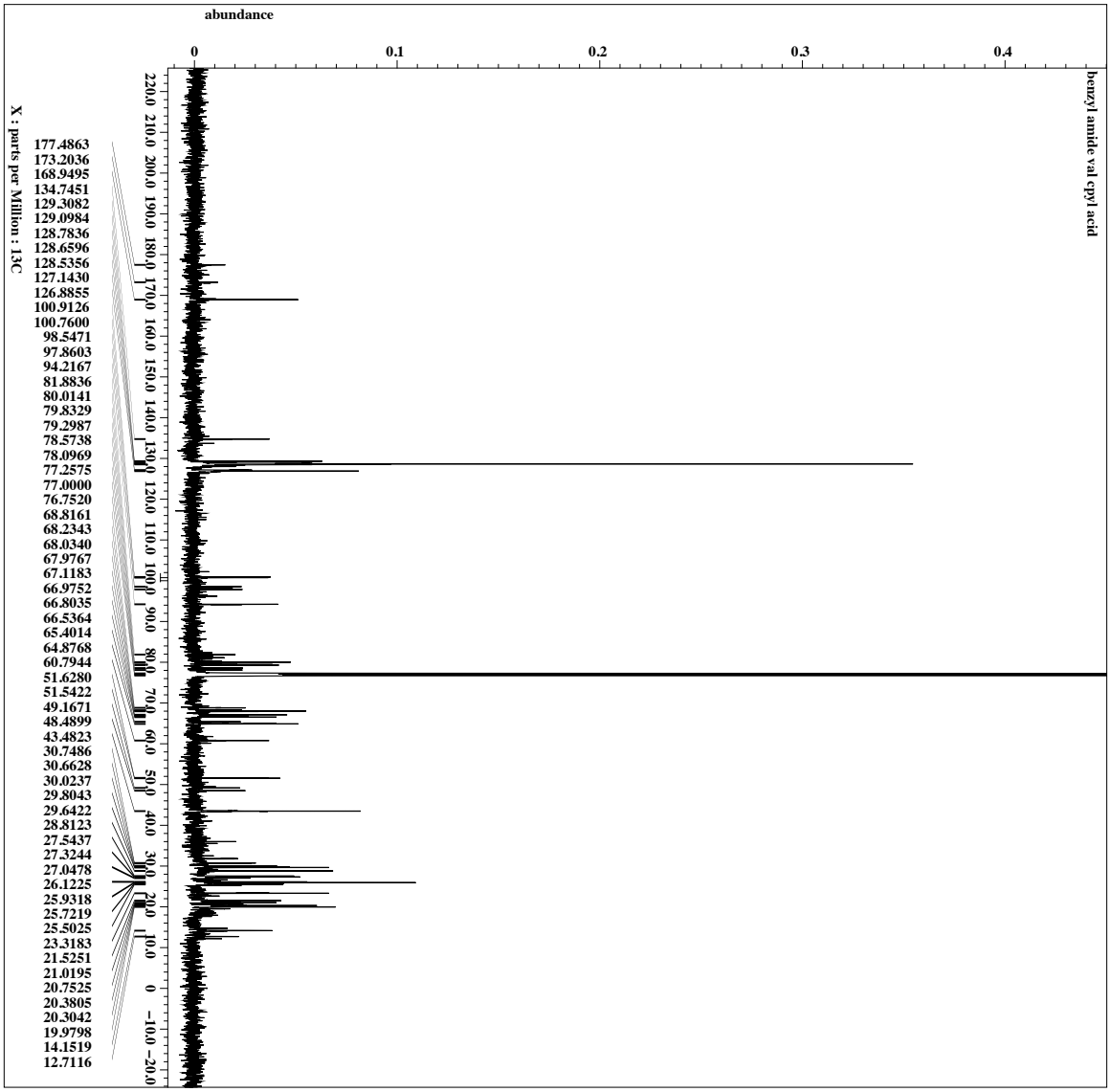
^{13}C-NMR ($CDCl_3$)

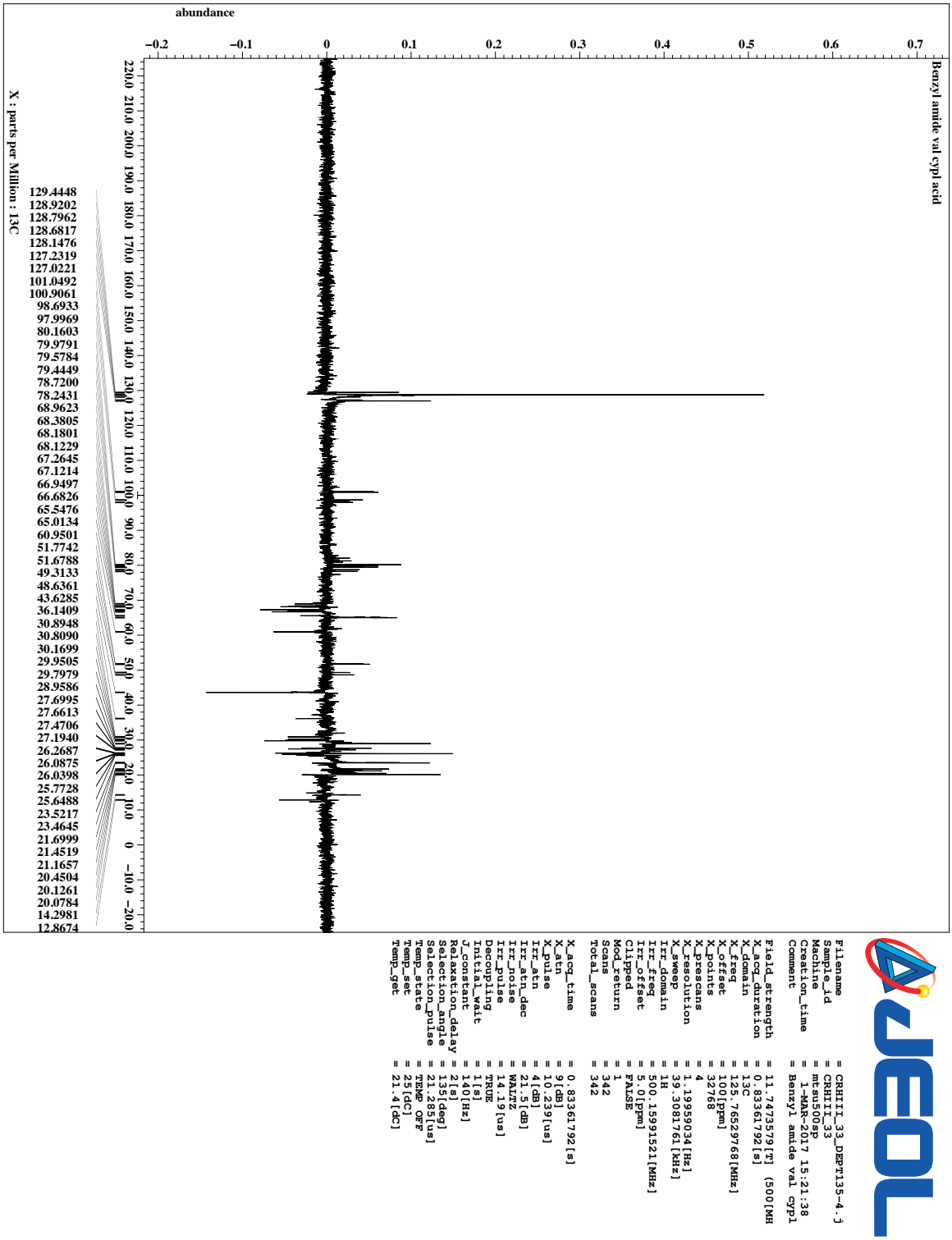
$DEPT_{135}$ ($CDCl_3$)

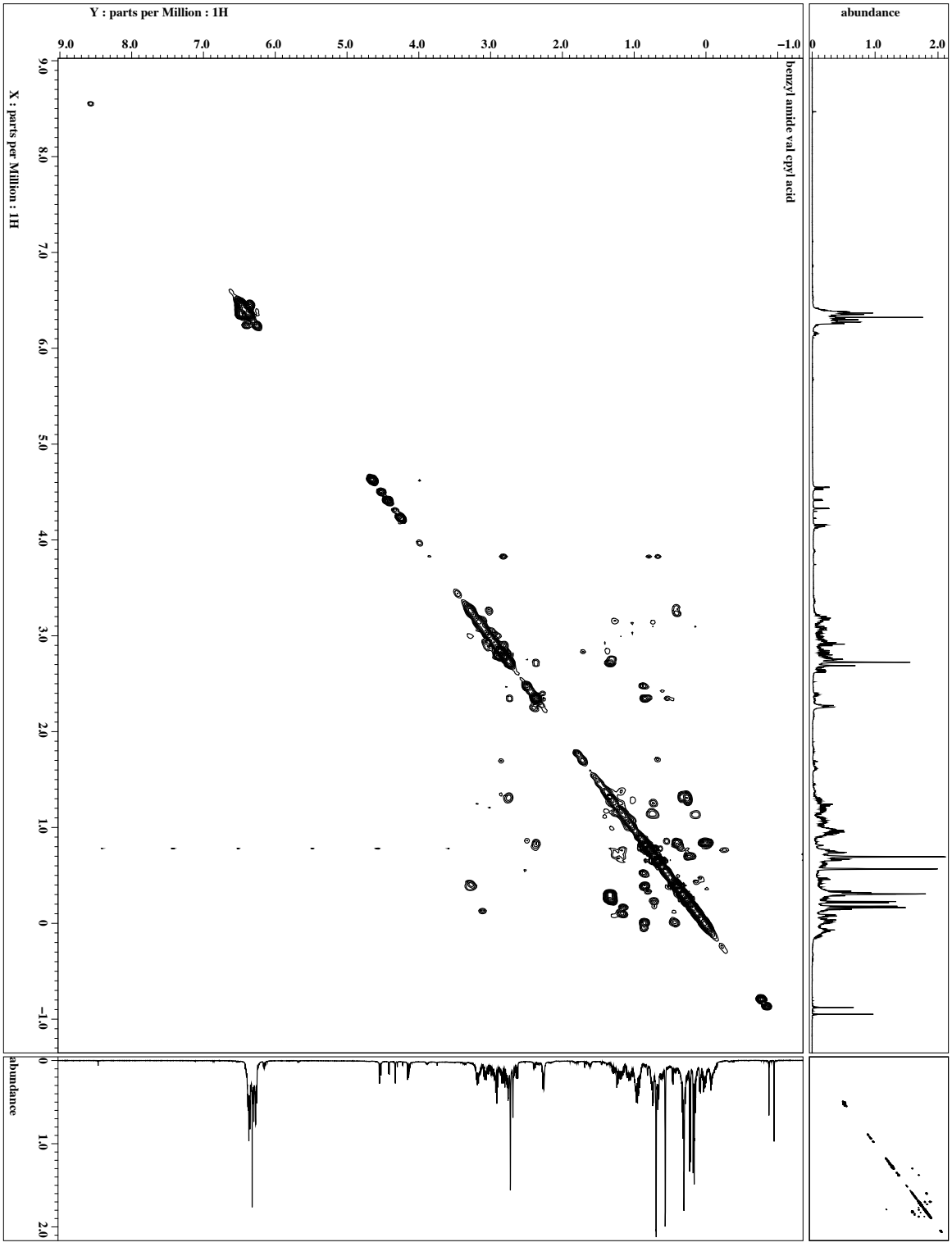
COSY ($CDCl_3$)

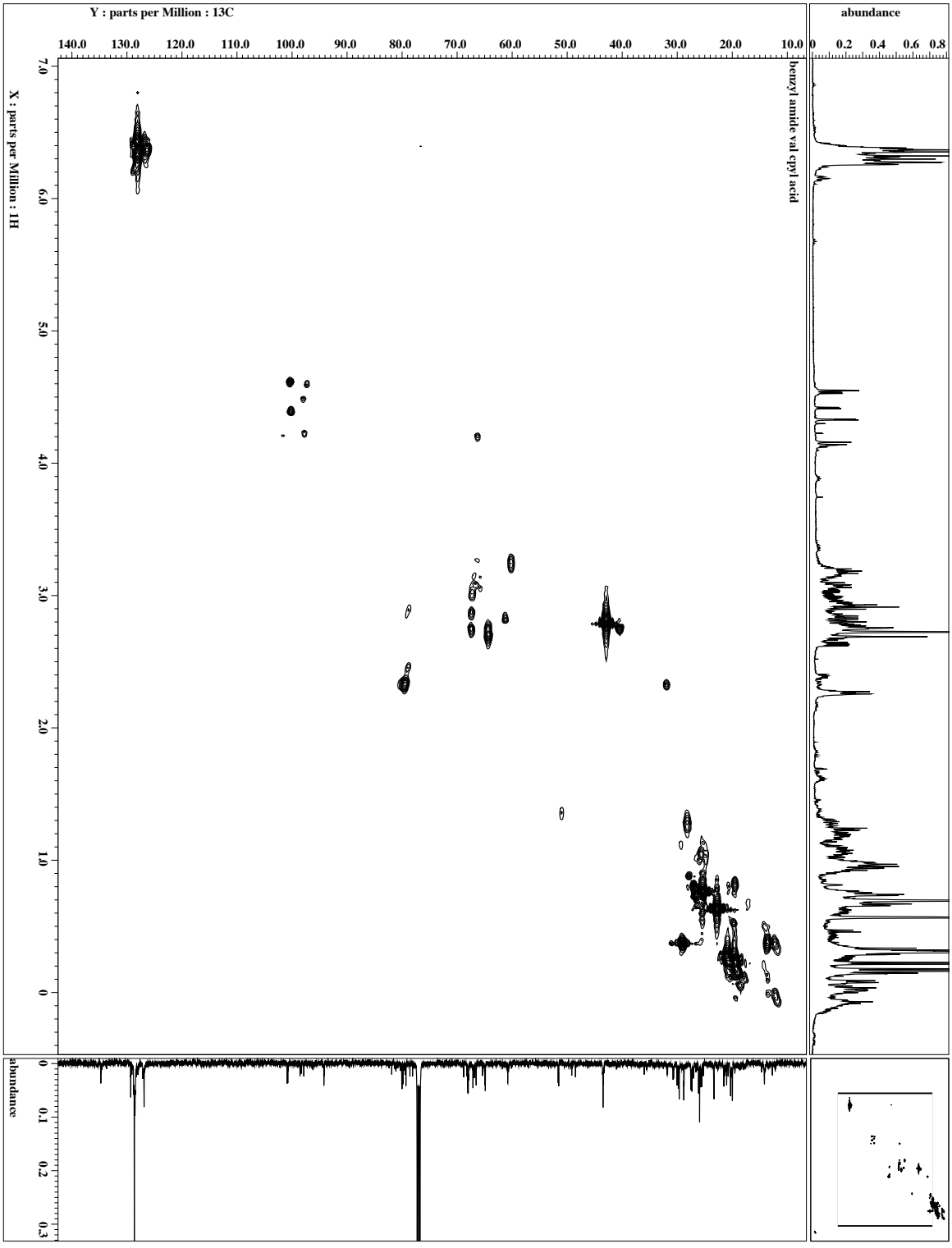
HMQC ($CDCl_3$)


abundance
benzyl amide val cpyl acid
7.64
10.1
1
5.69
7.05
11.95
X : parts per Million : 1H
6.4490
6.4456
6.2245
5.0828
5.0805
4.8274
4.8251
4.8114
4.8079
4.7953
4.7278
4.6991
4.6762
4.6682
4.6556
4.6522
4.6453
4.6224
4.5869
4.1712
4.1552
3.1394
3.1268
3.1222
2.8863
2.8794
2.8760
2.8668
2.8588
2.8508
2.8382
2.8348
2.8210
2.6664
2.6527
2.6492
2.6389
2.6321
2.5931
2.5908
2.5782
2.5679
2.5553
2.4660
2.3561
2.2381
2.2335
2.2187
2.2141
2.2049
2.1854
2.1259
2.1122
2.0767
2.0618
2.0583
2.0446
2.0400
1.9873
1.9736
1.9335
1.9278
1.9140
1.9106
1.8281
1.8144
1.0208
0.9486
Filename = CRHIII_33_PROTON-10.j
Sample_id = CRHIII_33
Machine = mt-su500pp
Creation_time = 1-MAR-2017 15:38:56
Comment = benzyl amide val cpyl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38384438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.2[dC]

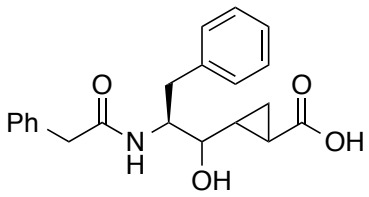

benzyl amide val cpyl acid
abundance
0
0.1
0.2
0.3
0.4
220.0 210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0 0 −10.0 −20.0
X : parts per Million : 13C
177.4863
173.2036
168.9495
134.7451
129.3082
129.0984
128.7836
128.6596
128.5356
127.1430
126.8855
100.9126
100.7600
98.5471
97.8603
94.2167
81.8836
80.0141
79.8329
79.2987
78.5738
78.0969
77.2575
77.0000
76.7520
68.8161
68.2343
68.0340
67.9767
67.1183
66.9752
66.8035
66.5364
65.4014
64.8768
60.7944
51.6280
51.5422
49.1671
48.4899
43.4823
30.7486
30.6628
30.0237
29.8043
29.6422
28.8123
27.5437
27.3244
27.0478
26.1225
25.9318
25.7219
25.5025
23.3183
21.5251
21.0195
20.7525
20.3805
20.3042
19.9798
14.1519
12.7116
JEOL
Filename = CRHIII_33_CARBON-9.jd
Sample_id = CRHIII_33
Machine = mt-nm500pp
Creation_time = 1-MAR-2017 16:31:08
Comment = benzyl amide val cpyl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 1024
Total_scans = 1024
X_90_width = 10.239[us]
X_acq_time = 0.83361792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.83361792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.6[dC]


Benzyl amide val cypl acid
abundance
X : parts per Million : 13C
129.4448
128.9202
128.7962
128.6817
128.1476
127.2319
127.0221
101.0492
100.9061
98.6933
97.9969
80.1603
79.9791
79.5784
79.4449
78.7200
78.2431
68.9623
68.3805
68.1801
68.1229
67.2645
67.1214
66.9497
66.6826
65.5476
65.0134
60.9501
51.7742
51.6788
49.3133
48.6361
43.6285
36.1409
30.8948
30.8090
30.1699
29.9505
29.7979
28.9586
27.6995
27.6613
27.4706
27.1940
26.2687
26.0875
26.0398
25.7728
25.6488
23.5217
23.4645
21.6999
21.4519
21.1657
20.4504
20.1261
20.0784
14.2981
12.8674
Filename = CRHIII_33_DEPT135-4.j
Sample_id = CRHIII_33
Machine = mt-su500pp
Creation_time = 1-MAR-2017 15:21:38
Comment = Benzyl amide val cypl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 342
Total_scans = 342
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.4[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
abundance
abundance
benzyl amide val cpyl acid


Y : parts per Million : 13C
X : parts per Million : 1H
abundance
benzyl amide val cpyl acid

2-((*S*)-2-(benzylcarbonylamino)-1-hydroxy-3-phenylpropyl) cyclopropanecarboxylic acid (41)



NMR

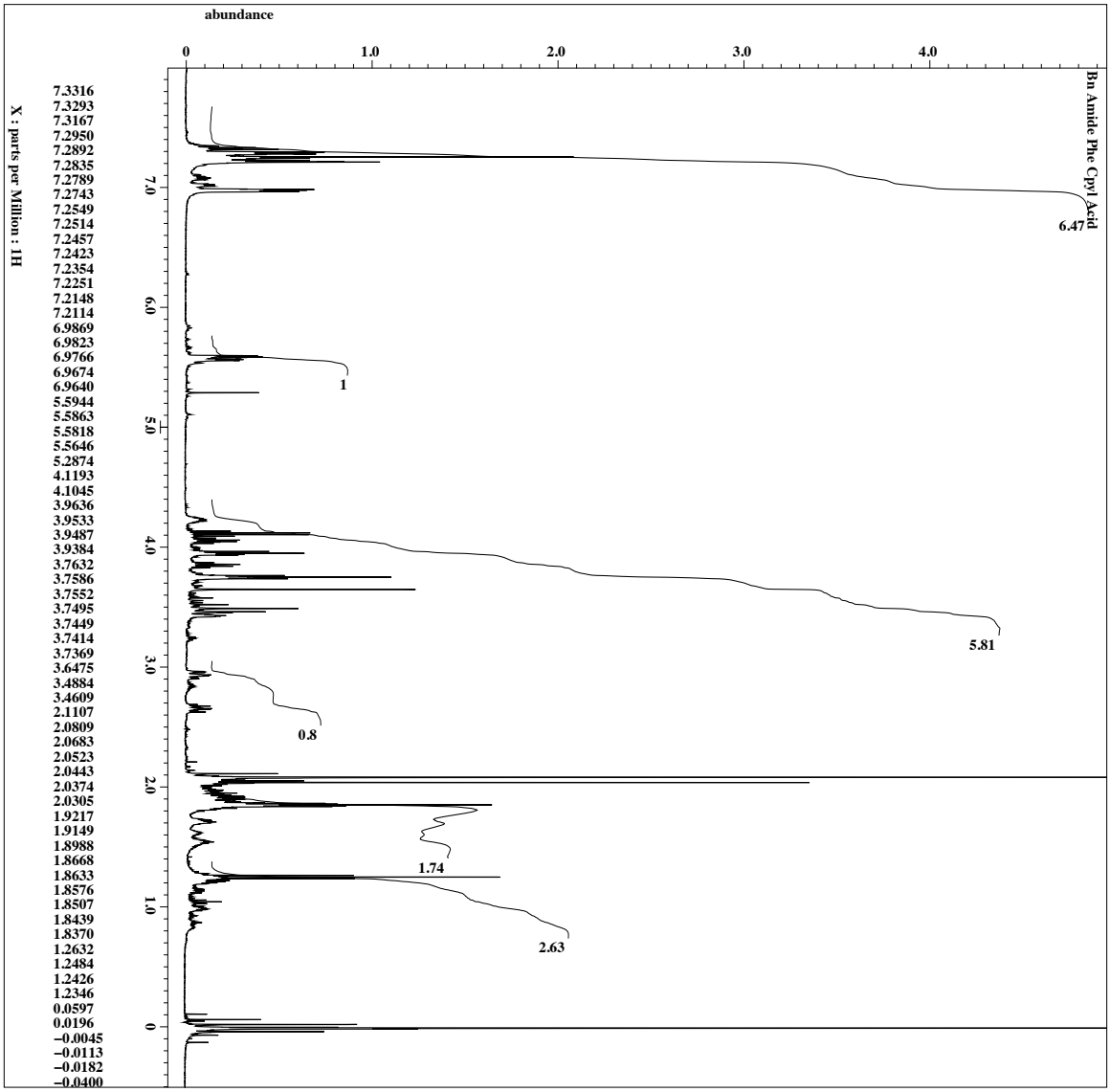
^{1}H-NMR ($CDCl_3$)

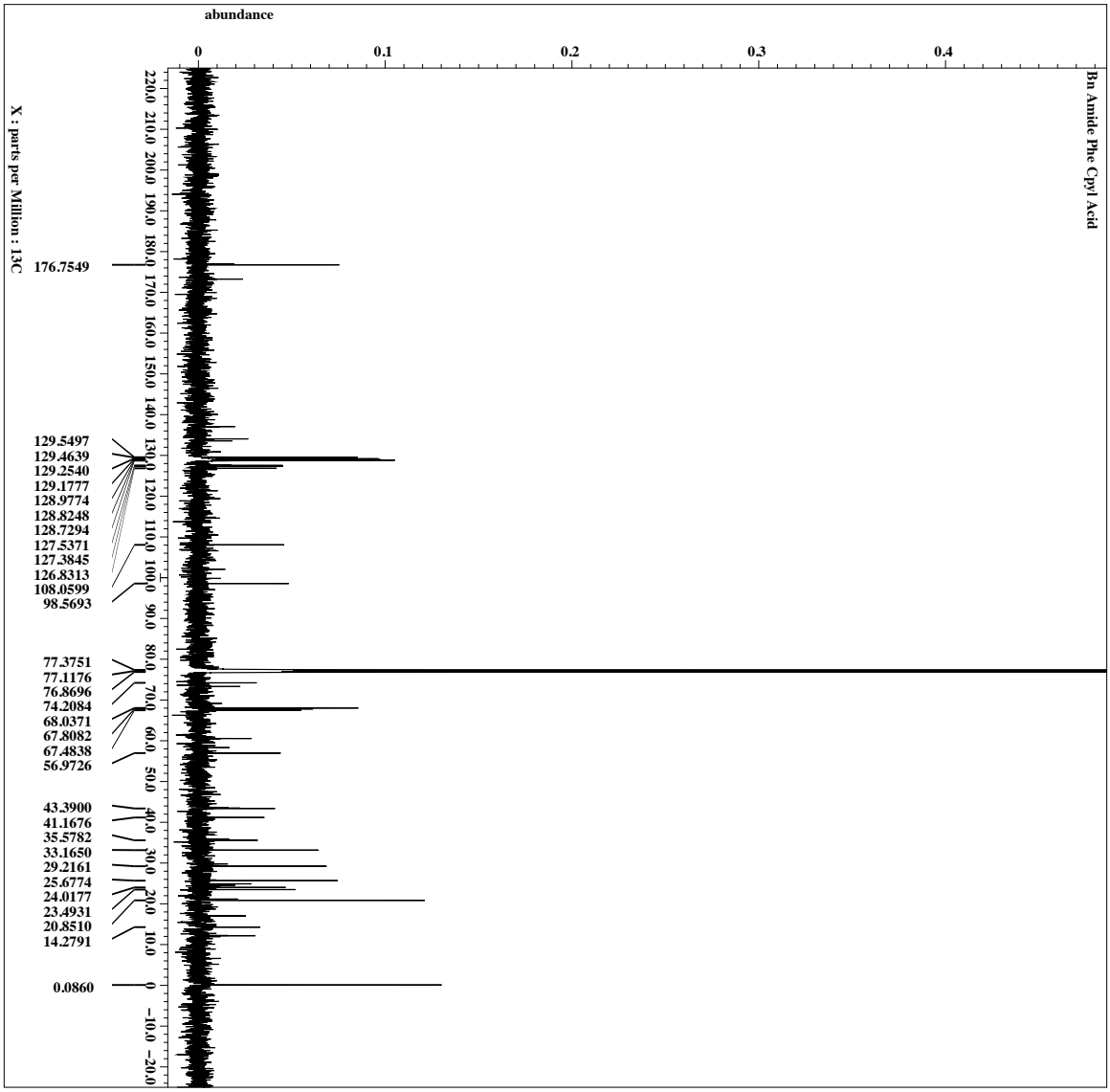
^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

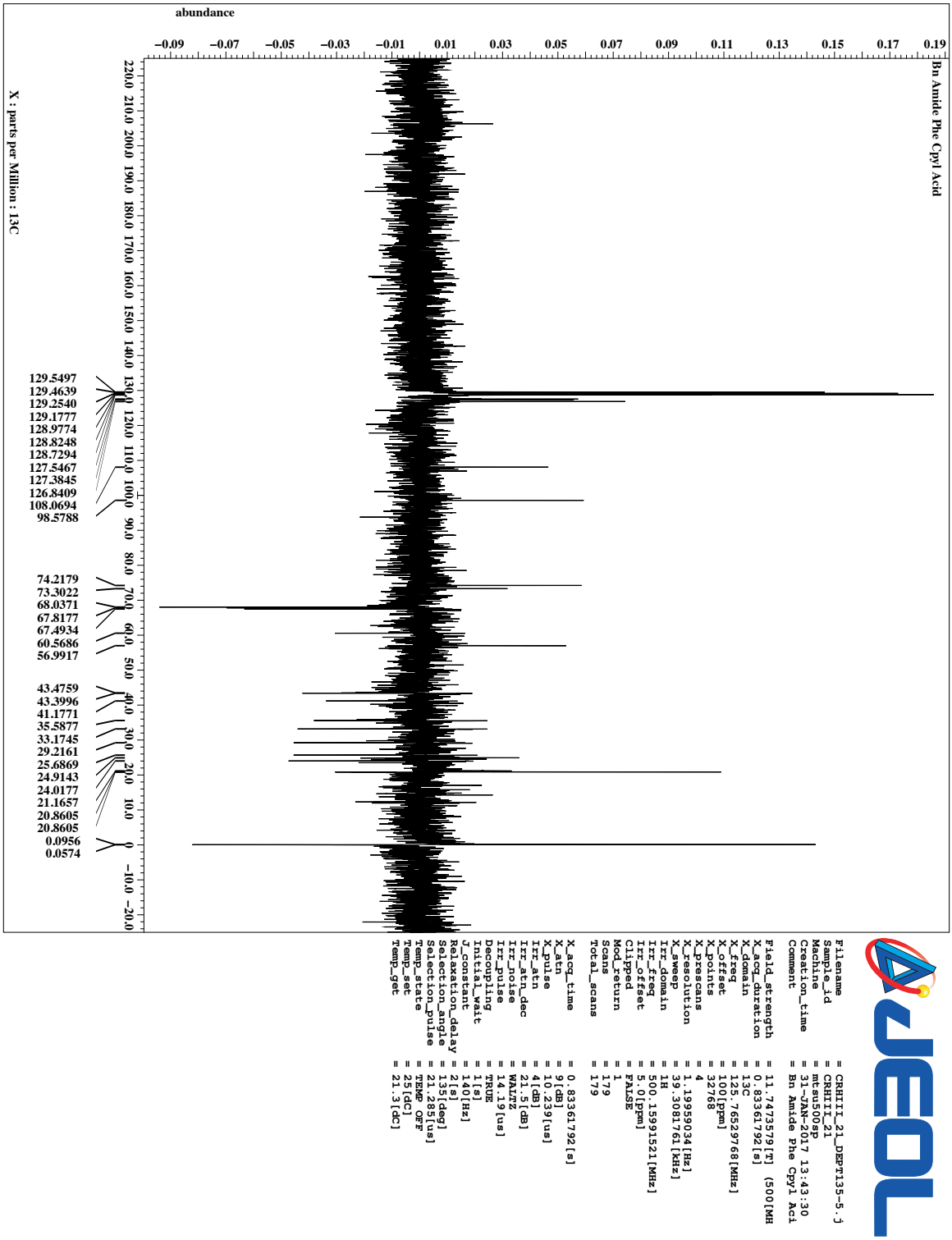
COSY ($CDCl_3$)

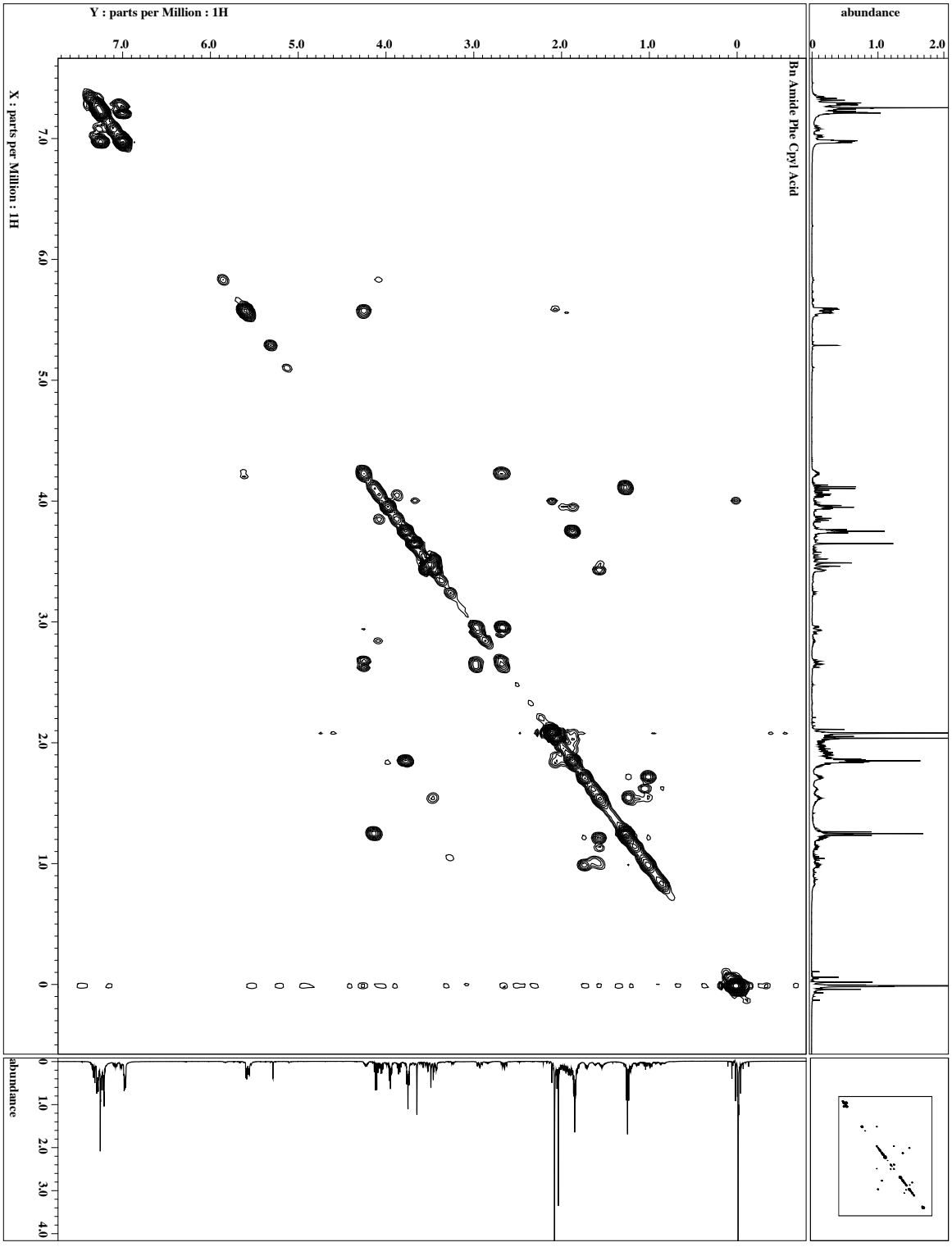
HMQC ($CDCl_3$)

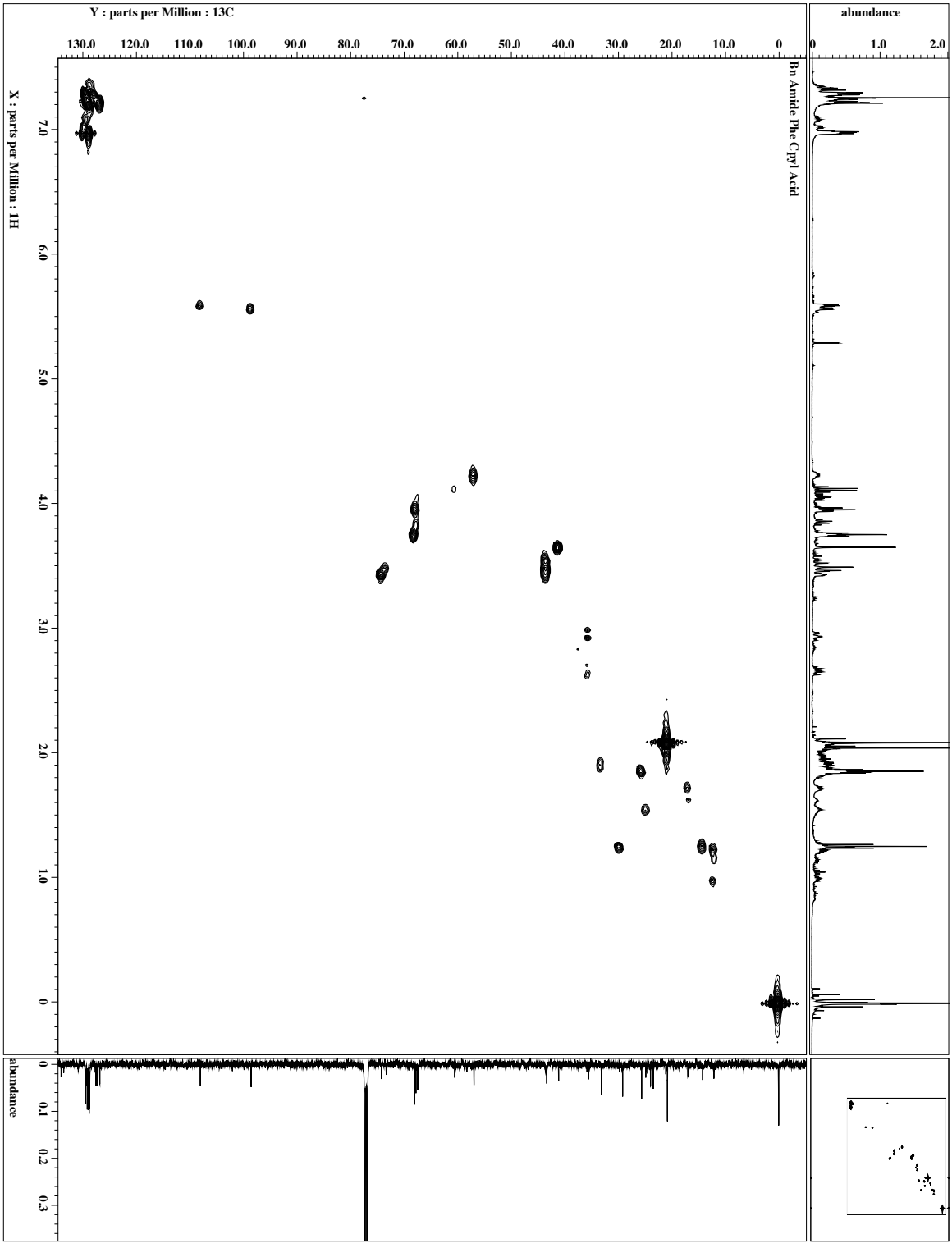

Bn Amide Phe Cpyl Acid
abundance
X : parts per Million : 1H
6.47
1
5.81
0.8
1.74
2.63
Filename = CRHIII_21_PROTON-14.j
Sample_id = CRHIII_21
Machine = mt-su500ap
Creation_time = 31-JAN-2017 13:57:55
Comment = Bn Amide Phe Cpyl Aci
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.2[dC]



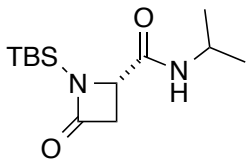

```
Filename                = CRHITI_21_CARBON-11.j
Sample_id               = CRHITI_21
Machine                 = mt-su500pp
Creation_time           = 31-JAN-2017 14:17:21
Comment                 = Bn Amide Phe Cpyl Aci

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 0.83361792[s]
X_domain                = 13C
X_freq                  = 125.76529768[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 1.19959034[Hz]
X_sweep                 = 39.3081761[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 400
Total_scans             = 400

X_90_width              = 10.239[us]
X_acq_time              = 0.83361792[s]
X_angle                 = 30[deg]
X_atn                   = 9[dB]
X_pulse                 = 3.413[us]
Irr_atn_dec             = 21.5[dB]
Irr_atn_noe             = 21.5[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 2.83361792[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 21.6[dC]
```


Bn Amide Phe Cpyl Acid
abundance
X : parts per Million : 13C
129.5497
129.4639
129.2540
129.1777
128.9774
128.8248
128.7294
127.5467
127.3845
126.8409
108.0694
98.5788
74.2179
73.3022
68.0371
67.8177
67.4934
60.5686
56.9917
43.4759
43.3996
41.1771
35.5877
33.1745
29.2161
25.6869
24.9143
24.0177
21.1657
20.8605
20.8605
0.0956
0.0574
Filename = CRHIII_21_DEPT135-5.j
Sample_id = CRHIII_21
Machine = mt-su500pp
Creation_time = 31-JAN-2017 13:43:30
Comment = Bn Amide Phe Cpyl Aci
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 179
Total_scans = 179
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.3[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
abundance
Bn Amide Phe Cpyl Acid


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
Bn Amide Phe Cpyl Acid
abundance
0
1.0
2.0
abundance
0
0.1
0.2
0.3

1-(tert-butyldimethylsilyl)-4-oxo-N-(propan-2-yl)azetidine-2-carboxamide (45)



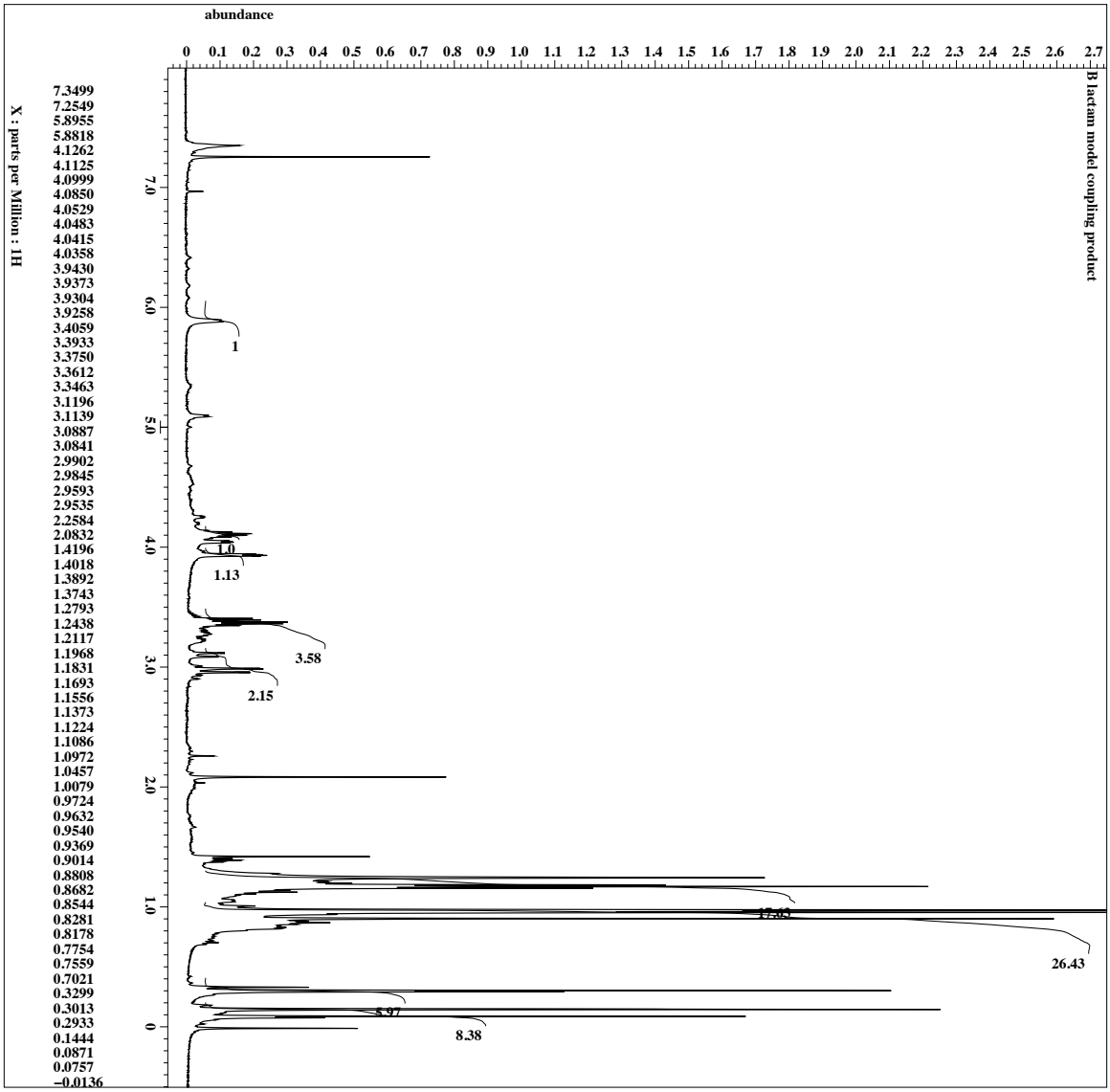
NMR

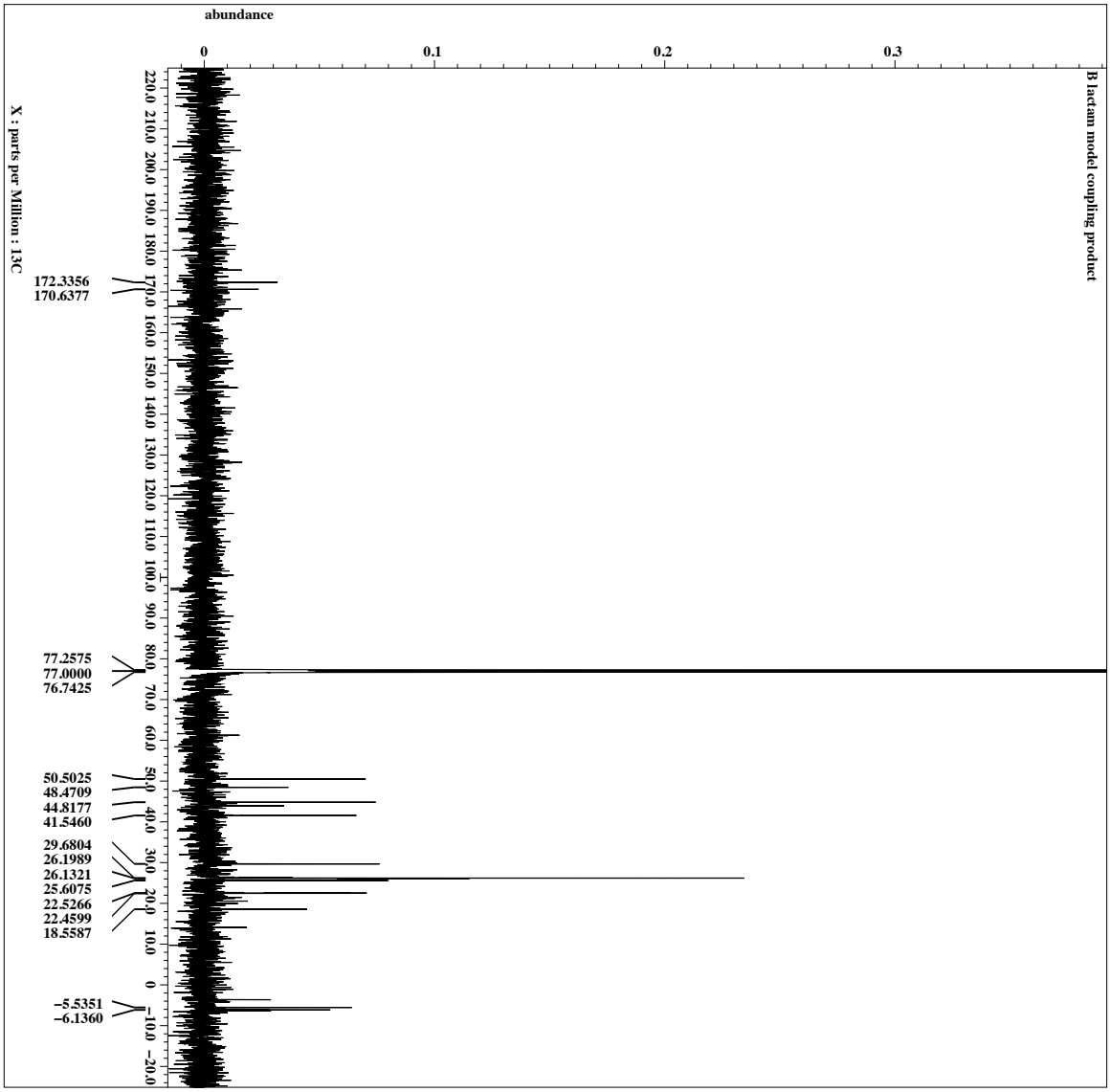
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

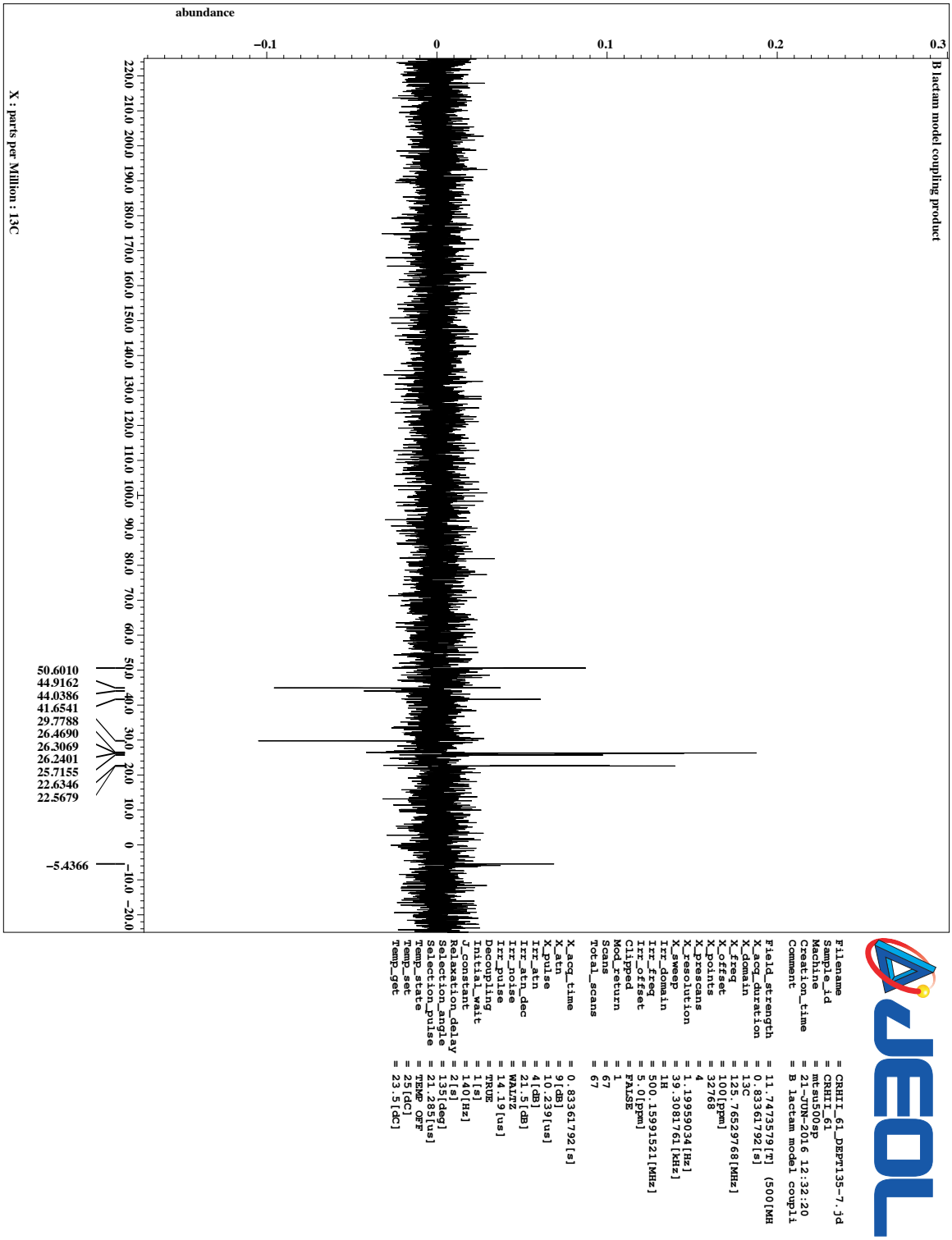
HMQC ($CDCl_3$)

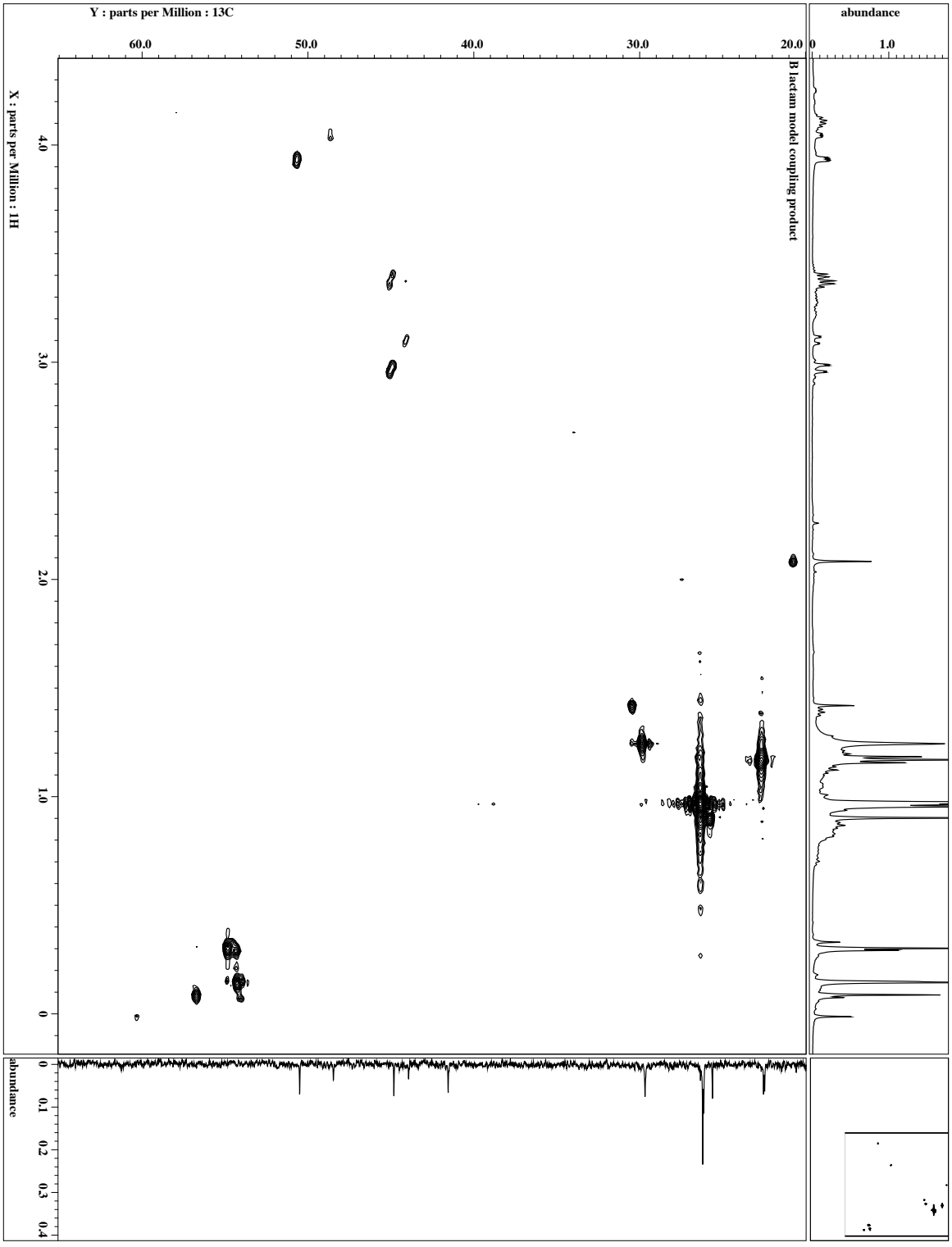

B lactam model coupling product
abundance
X : parts per Million : 1H
1
1.13
3.58
2.15
17.45
26.43
5.97
8.38
Filename = CRHII_61_PROTON-17.jd
Sample_id = CRHII_61
Machine = mt-su500ap
Creation_time = 21-JUN-2016 13:34:03
Comment = B lactam model coupli
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.4[dC]



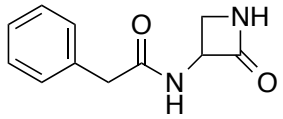
JEOL

Filename	= CRHII_61_CARBON-14.jd
Sample_id	= CRHII_61
Machine	= mt-su500ap
Creation_time	= 21-JUN-2016 12:47:31
Comment	= B lactam model coupli
Field_strength	= 11.7473579[T] (500[MH
X_acq_duration	= 0.83361792[s]
X_domain	= 13C
X_freq	= 125.76529768[MHz]
X_offset	= 100[ppm]
X_points	= 32768
X_prescans	= 4
X_resolution	= 1.19959034[Hz]
X_sweep	= 39.3081761[kHz]
Irr_domain	= 1H
Irr_freq	= 500.15991521[MHz]
Irr_offset	= 5.0[ppm]
Clipped	= FALSE
Mod_return	= 1
Scans	= 259
Total_scans	= 259
X_90_width	= 10.239[us]
X_acq_time	= 0.83361792[s]
X_angle	= 30[deg]
X_atn	= 9[dB]
X_pulse	= 3.413[us]
Irr_atn_dec	= 21.5[dB]
Irr_atn_noe	= 21.5[dB]
Irr_noise	= WALTZ
Decoupling	= TRUE
Initial_wait	= 1[s]
Noe	= TRUE
Noe_time	= 2[s]
Relaxation_delay	= 2[s]
Repetition_time	= 2.83361792[s]
Temp_state	= TEMP OFF
Temp_set	= 25[dC]
Temp_get	= 23.7[dC]


B lactam model coupling product
abundance
X : parts per Million : 13C
50.6010
44.9162
44.0386
41.6541
29.7788
26.4690
26.3069
26.2401
25.7155
22.6346
22.5679
−5.4366
Filename = CRHII_61_DEPT135-7.jd
Sample_id = CRHII_61
Machine = mt-su500ap
Creation_time = 21-JUN-2016 12:32:20
Comment = B lactam model coupli
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 67
Total_scans = 67
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.5[dC]


Y : parts per Million : 13C
60.0
50.0
40.0
30.0
20.0
X : parts per Million : 1H
4.0
3.0
2.0
1.0
0
B lactam model coupling product
abundance
0
1.0
abundance
0
0.1
0.2
0.3
0.4

N-(2-oxoazetidin-3-yl)-2-phenylacetamide (47)



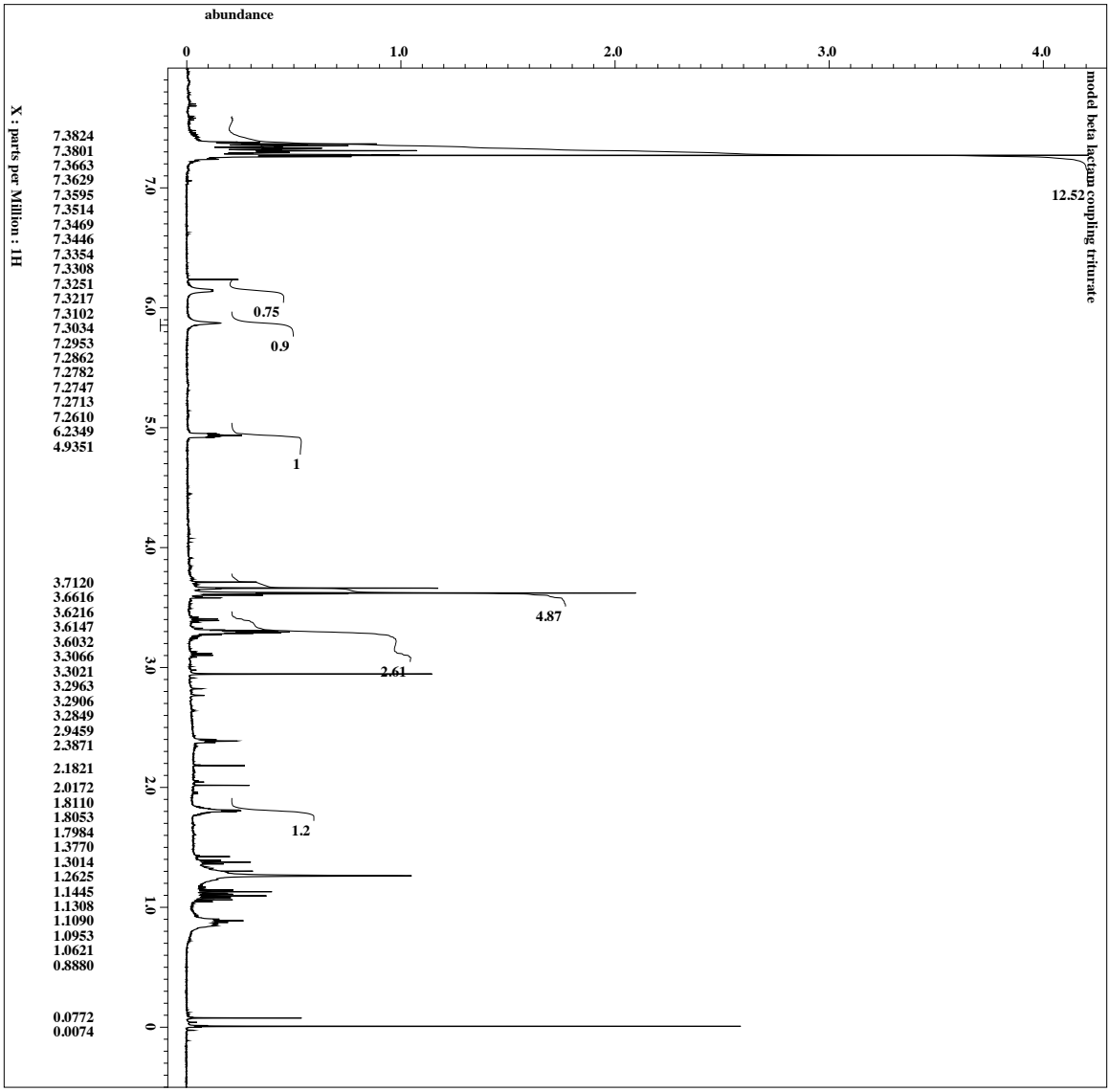
NMR

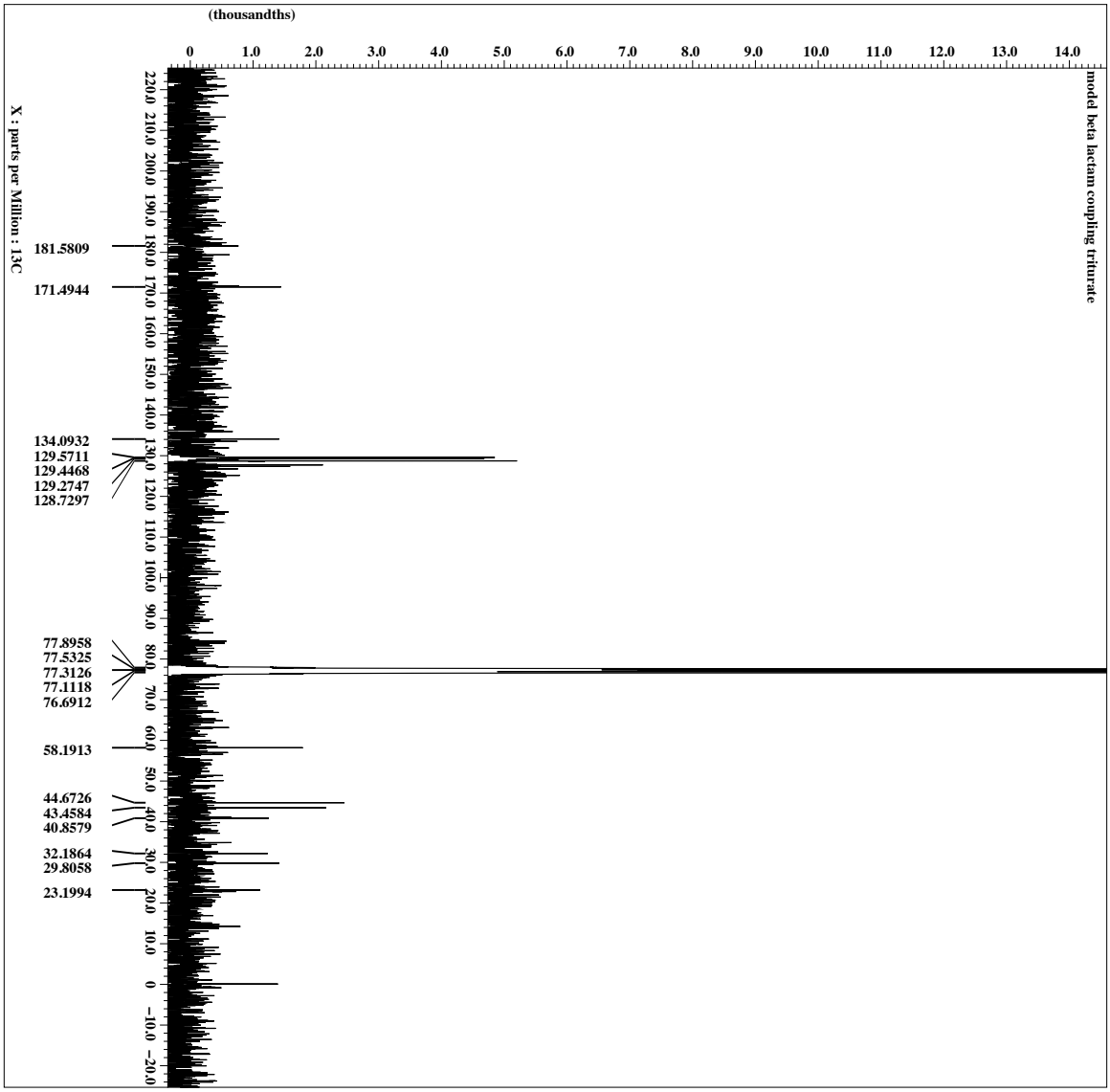
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

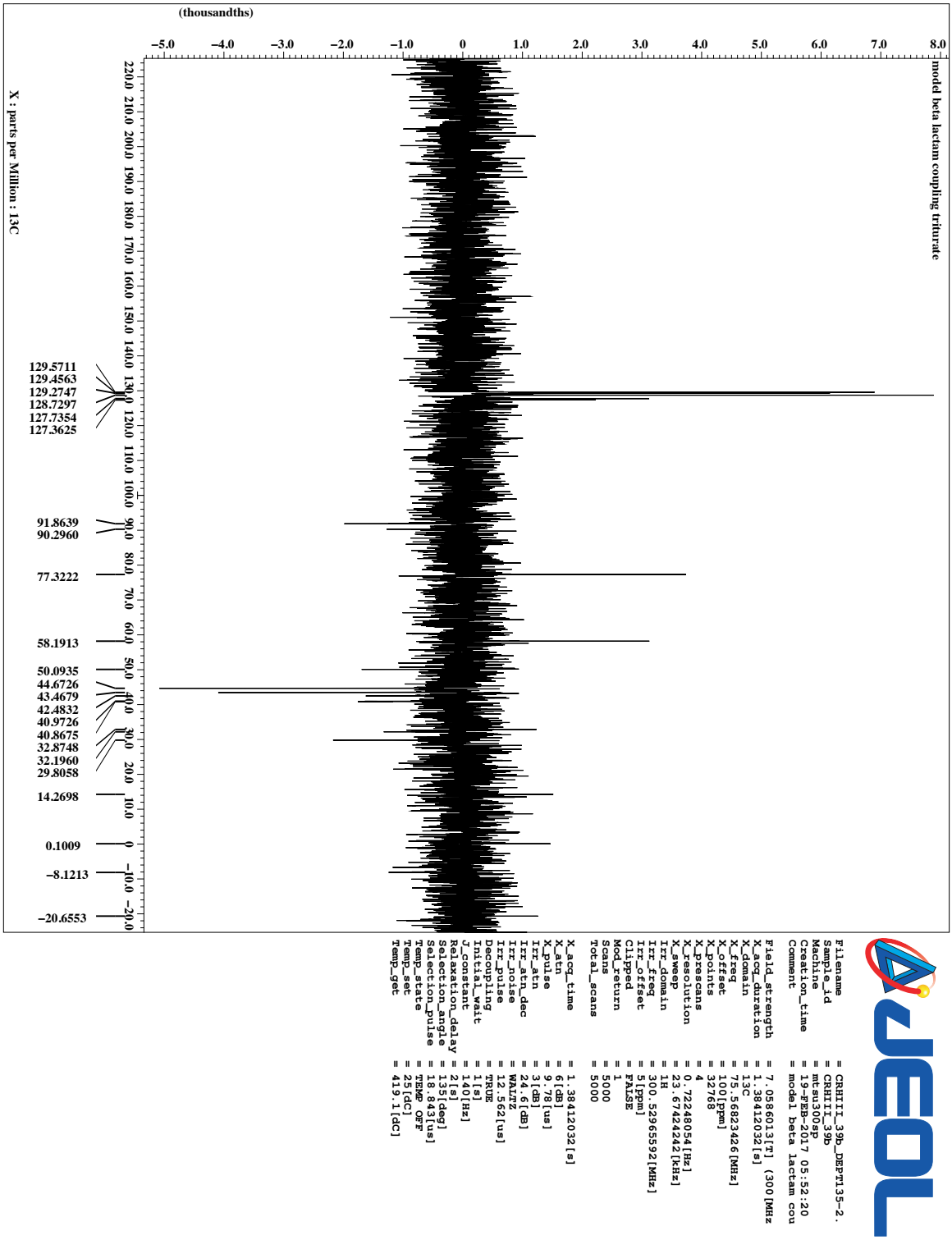
COSY ($CDCl_3$)

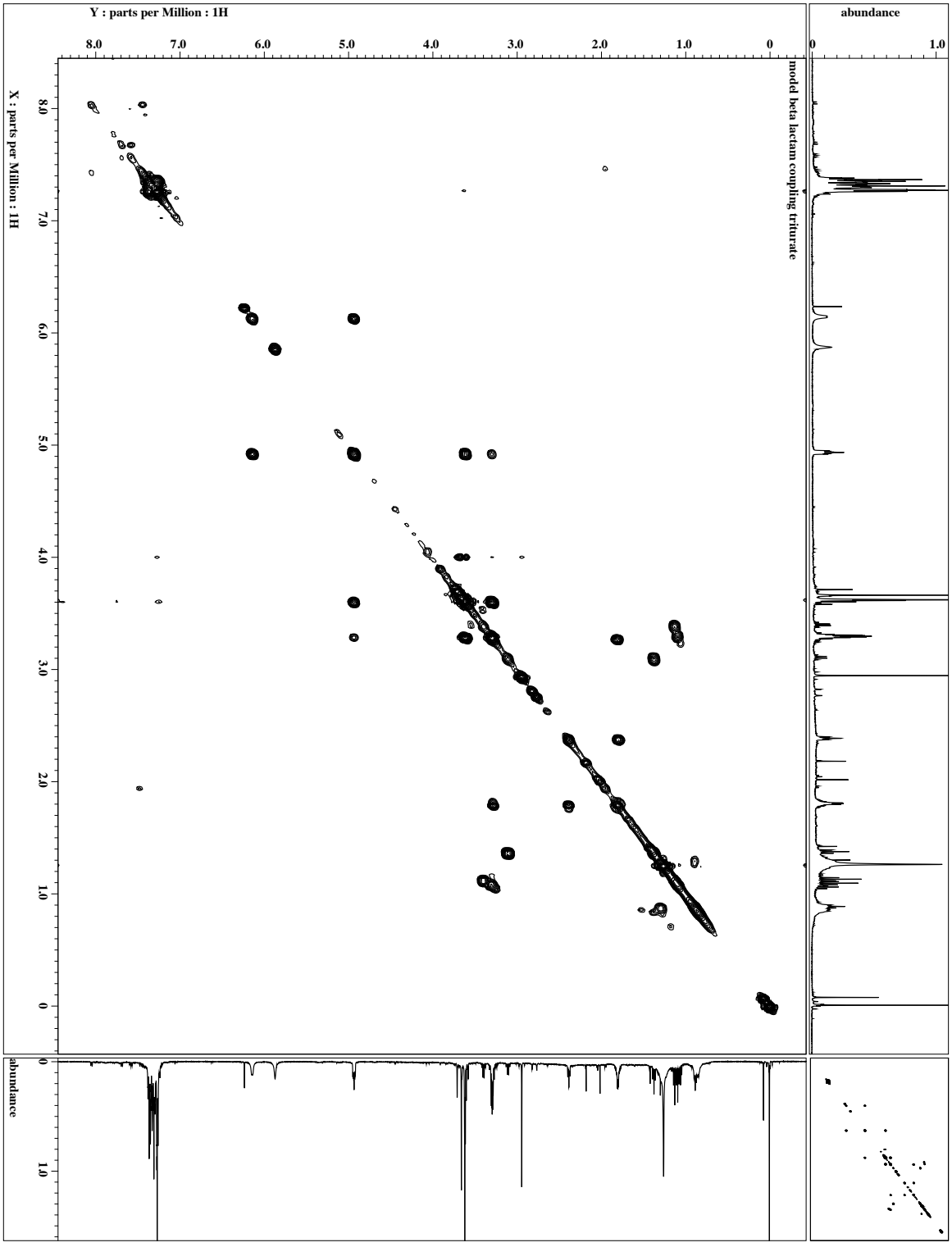

abundance
model beta lactam coupling triturate
12.52
0.75
0.9
1
4.87
2.61
1.2
X : parts per Million : 1H
7.3824
7.3801
7.3663
7.3629
7.3595
7.3514
7.3469
7.3446
7.3354
7.3308
7.3251
7.3217
7.3102
7.3034
7.2953
7.2862
7.2782
7.2747
7.2713
7.2610
6.2349
4.9351
3.7120
3.6616
3.6216
3.6147
3.6032
3.3066
3.3021
3.2963
3.2906
3.2849
2.9459
2.3871
2.1821
2.0172
1.8110
1.8053
1.7984
1.3770
1.3014
1.2625
1.1445
1.1308
1.1090
1.0953
1.0621
0.8880
0.0772
0.0074
Filename = CRHIII_39b_PROTON-8.j
Sample_id = CRHIII_39b
Machine = mt-su500ap
Creation_time = 18-FEB-2017 12:37:01
Comment = model beta lactam cou
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 74
Total_scans = 74
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 21.2[dC]



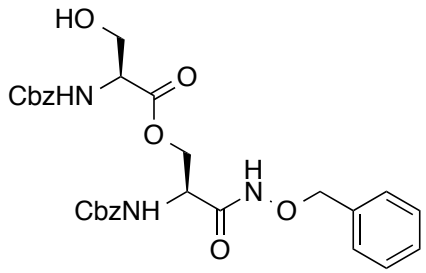
JEOL

```
Filename                = CRHIII_39b_CARBON-4.j
Sample_id               = CRHIII_39b
Machine                 = mt-eu300pp
Creation_time           = 19-FEB-2017 01:09:06
Comment                 = model beta lactam cou

Field_strength          = 7.0586013[T] (300[MHz
X_acq_duration          = 1.3841203[s]
X_domain                = 13C
X_freq                  = 75.5682342[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 0.72248054[Hz]
X_sweep                 = 23.674242[kHz]
Irr_domain              = 1H
Irr_freq                = 300.52965592[MHz]
Irr_offset              = 5[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 10000
Total_scans             = 10000

X_90_width              = 9.78[us]
X_acq_time              = 1.3841203[s]
X_angle                 = 30[deg]
X_atn                   = 6[dB]
X_pulse                 = 3.26[us]
Irr_atn_dec             = 24.6[dB]
Irr_atn_noe             = 24.6[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 3.3841203[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 293.4[dC]
```


model beta lactam coupling triurate
(thousandths)
X : parts per Million : 13C
129.5711
129.4563
129.2747
128.7297
127.7354
127.3625
91.8639
90.2960
77.3222
58.1913
50.0935
44.6726
43.4679
42.4832
40.9726
40.8675
32.8748
32.1960
29.8058
14.2698
0.1009
−8.1213
−20.6553
Filename = CRHIII_39b_DEPT135-2.
Sample_id = CRHIII_39b
Machine = mt-su300spp
Creation_time = 19-FEB-2017 05:52:20
Comment = model beta lactam cou
Field_strength = 7.0586013[T] (300[MHz
X_acq_duration = 1.38412032[s]
X_domain = 13C
X_freq = 75.56823426[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 0.72248054[Hz]
X_sweep = 23.67424242[kHz]
Irr_domain = 1H
Irr_freq = 300.52965592[MHz]
Irr_offset = 5[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 5000
Total_scans = 5000
X_acq_time = 1.38412032[s]
X_atn = 6[dB]
X_pulse = 9.78[us]
Irr_atn = 31[dB]
Irr_atn_dec = 24.6[dB]
Irr_noise = WALTZ
Irr_pulse = 12.562[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 18.843[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 419.1[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
abundance
model beta lactam coupling triurate

(2*S*)-2-[(Benzyloxy)carbamoyl]-2-{[(benzyloxy)carbonyl]amino}ethyl (2*S*)-2-{[(benzyloxy)carbonyl]amino}-3-hydroxypropanoate (53)



NMR

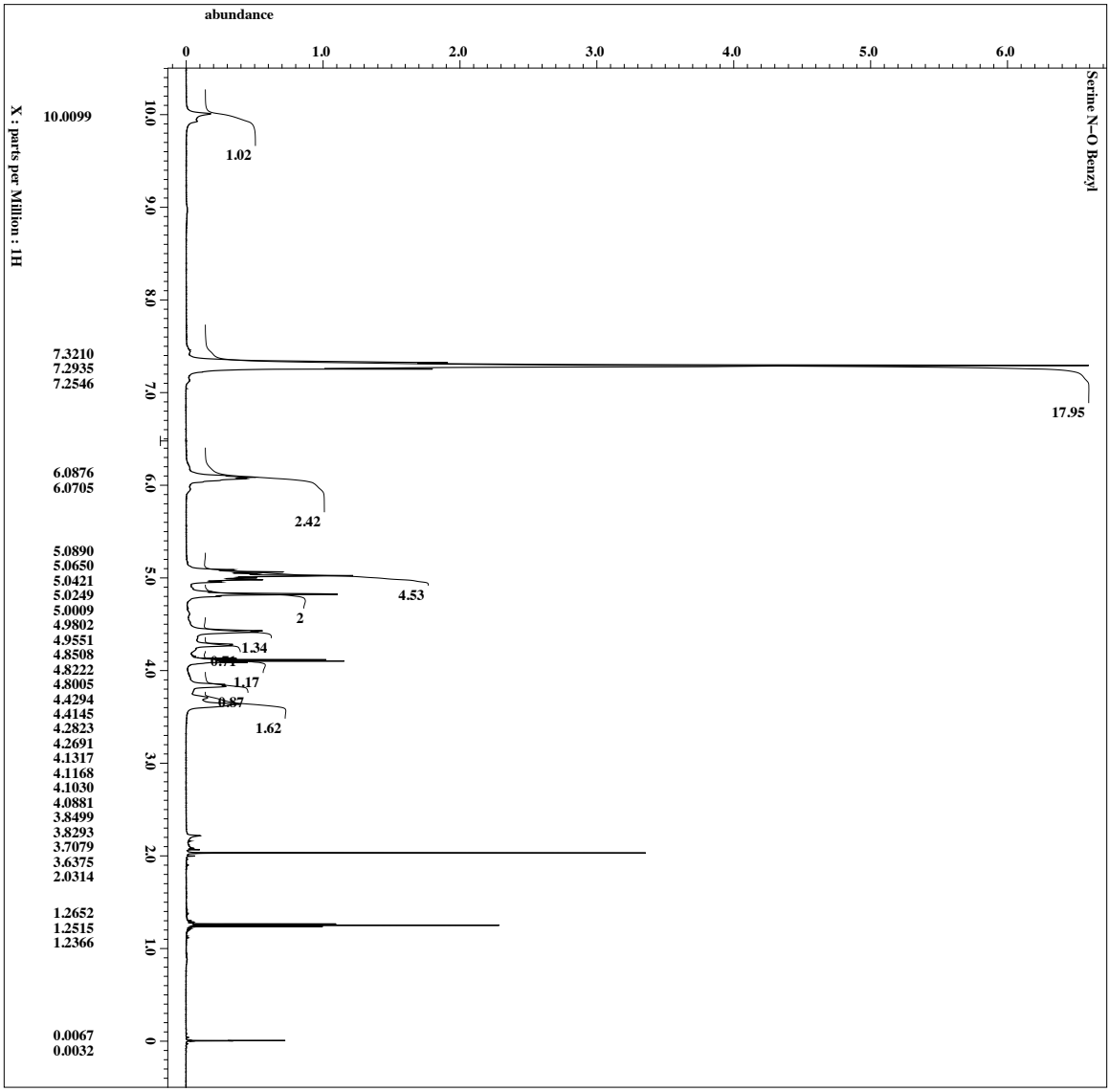
^{1}H-NMR ($CDCl_3$)

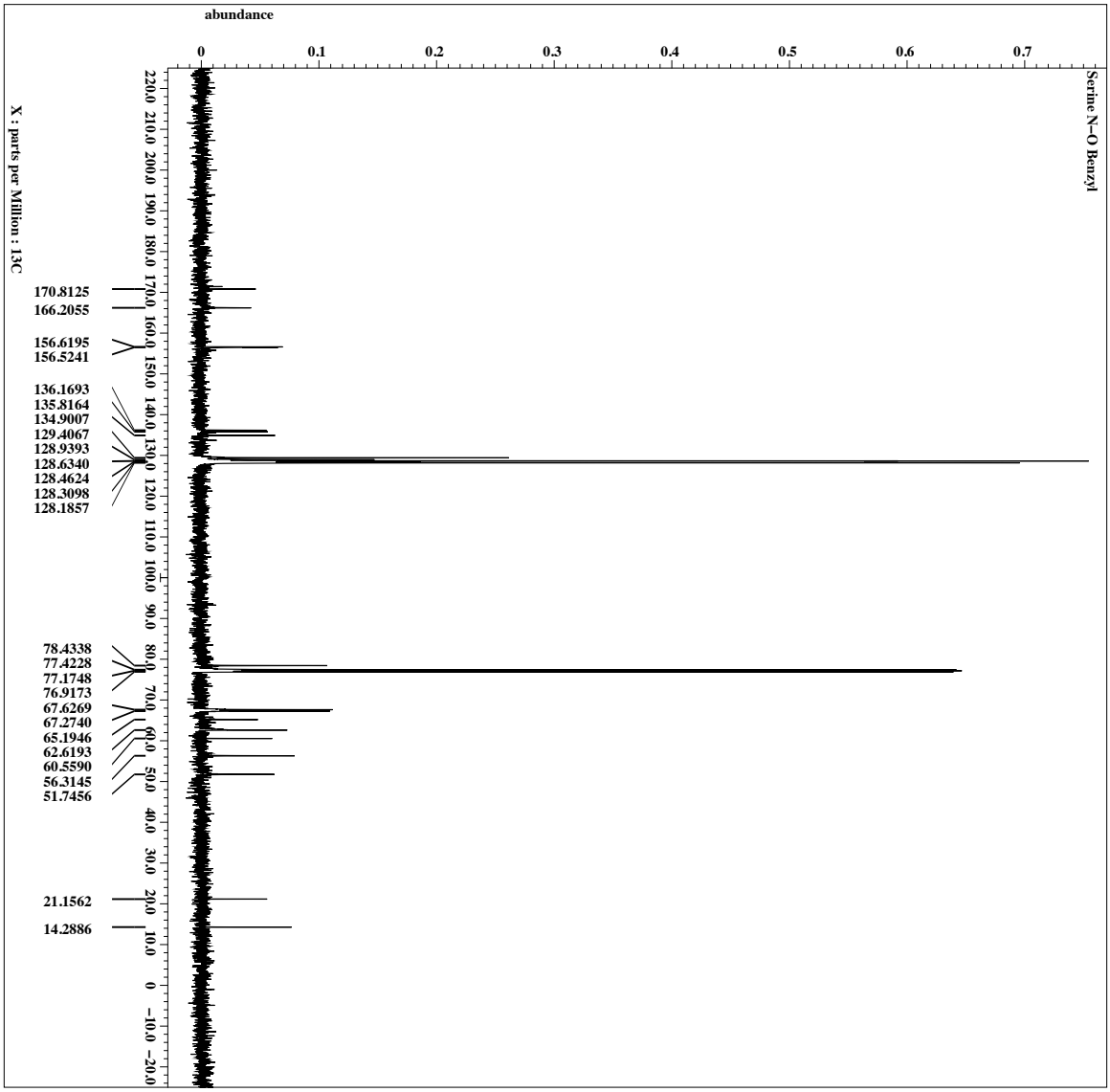
^{13}C-NMR ($CDCl_3$)

$DEPT_{135}$ ($CDCl_3$)

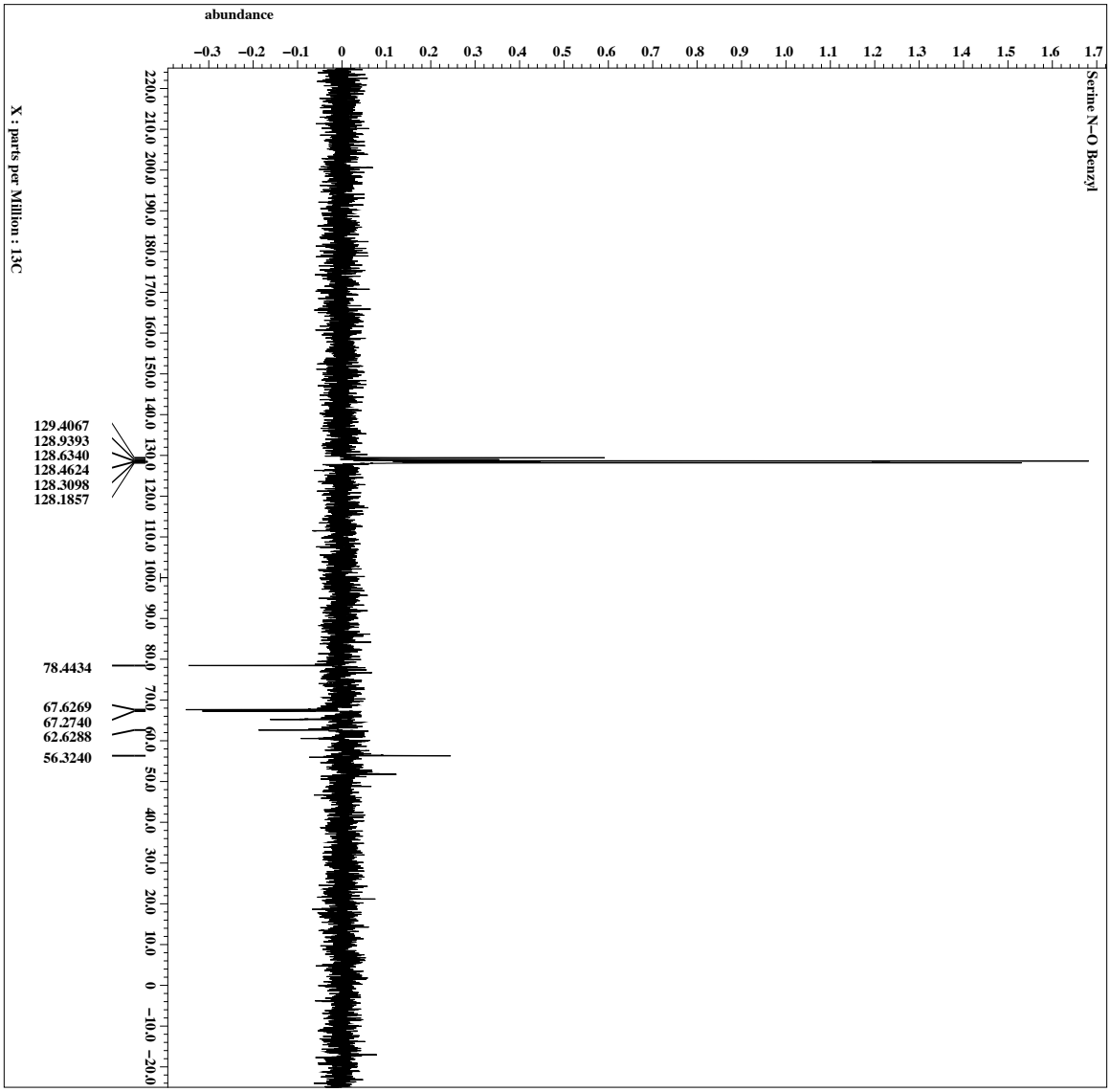
COSY ($CDCl_3$)

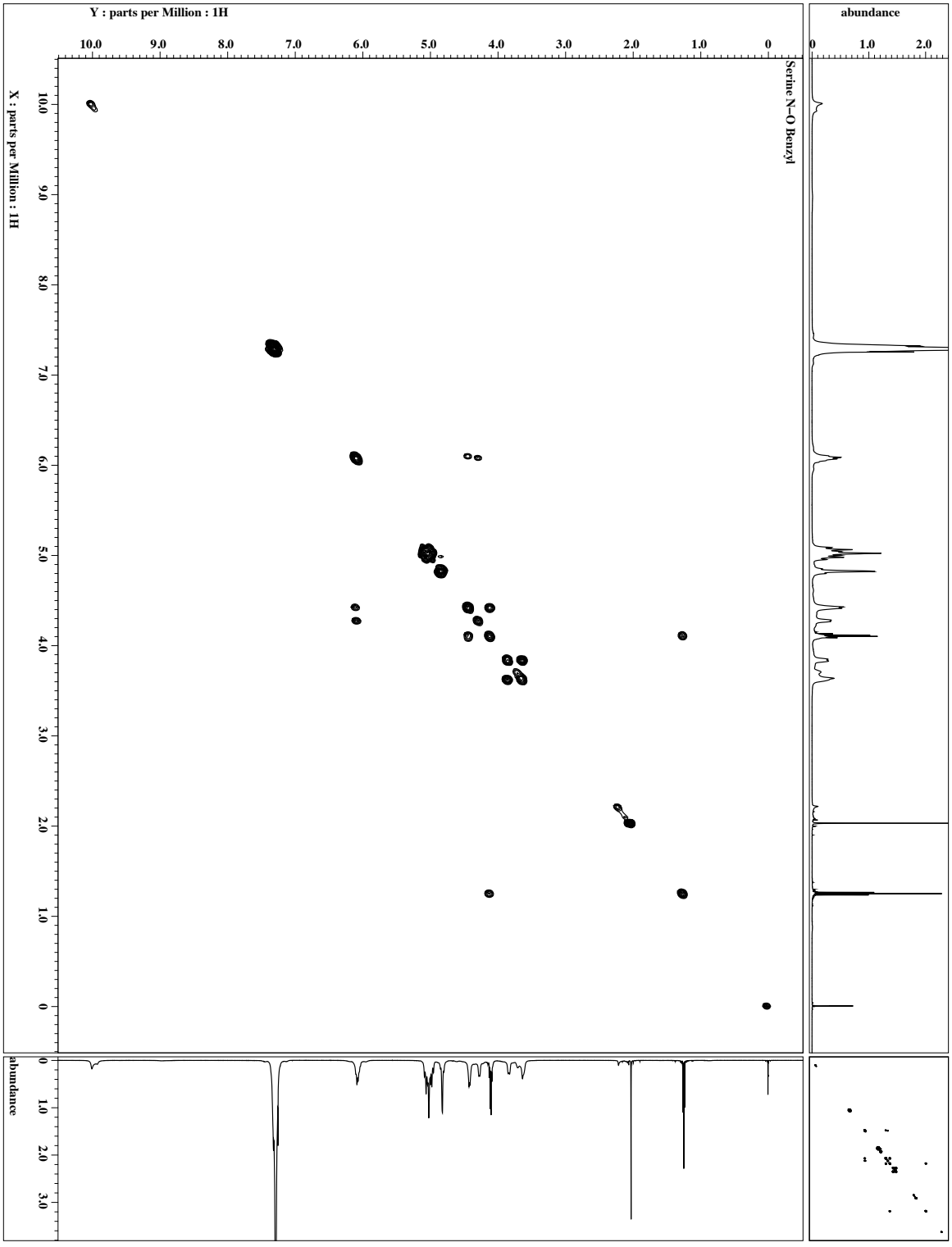
HMQC ($CDCl_3$)

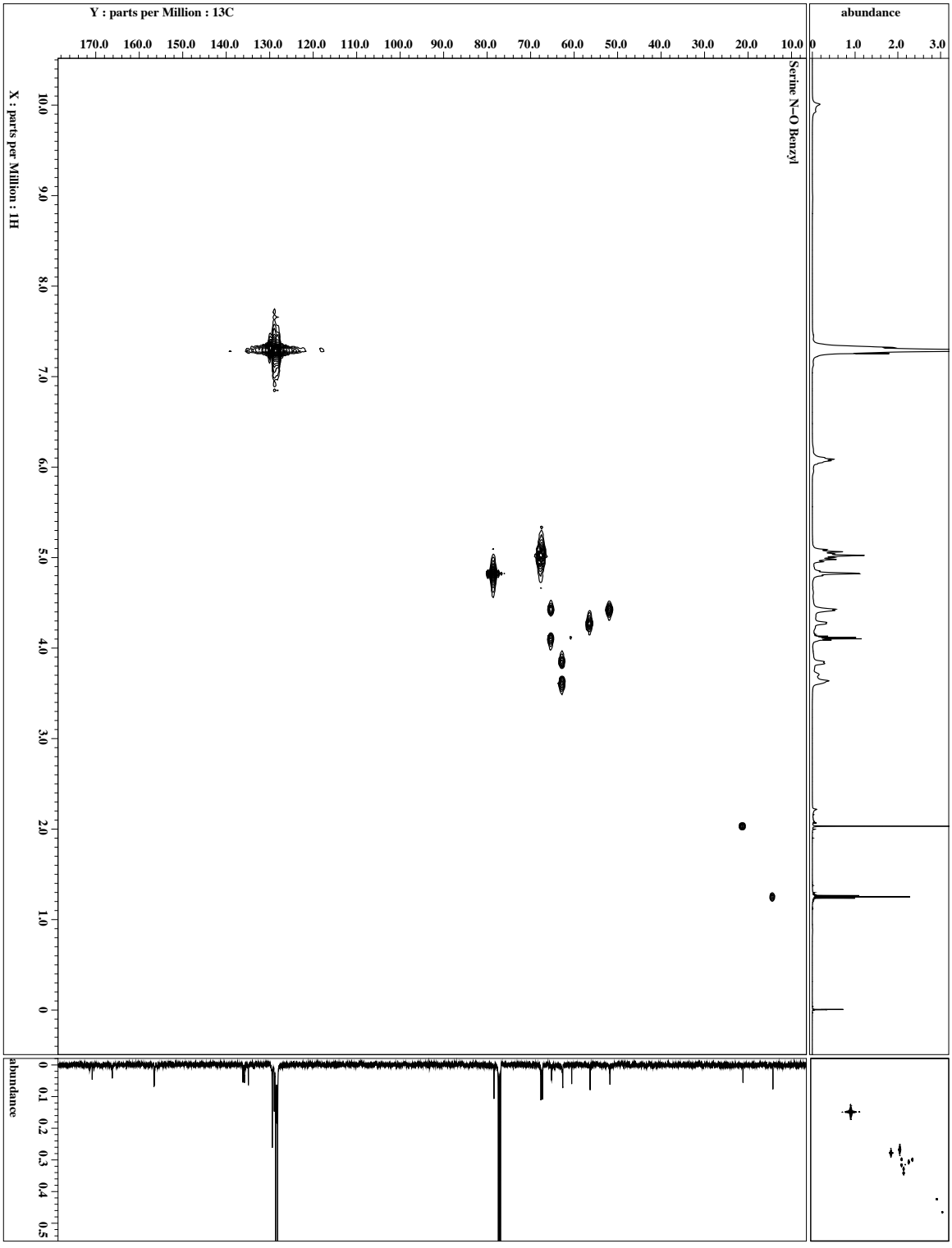

abundance
Serine N-O Benzyl
X : parts per Million : 1H
10.0099
7.3210
7.2935
7.2546
6.0876
6.0705
5.0890
5.0650
5.0421
5.0249
5.0009
4.9802
4.9551
4.8508
4.8222
4.8005
4.4294
4.4145
4.2823
4.2691
4.1317
4.1168
4.1030
4.0881
3.8499
3.8293
3.7079
3.6375
2.0314
1.2652
1.2515
1.2366
0.0067
0.0032
1.02
17.95
2.42
4.53
2
1.34
0.74
1.17
0.87
1.62
Filename = CRHIT_117A_PROTON-8.j
Sample_id = CRHIT_117A
Machine = mt-su500ap
Creation_time = 28-NOV-2016 13:35:13
Comment = Serine N-O Benzyl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = FALSE
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.8[dC]



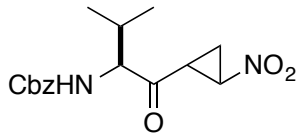

```
Filename                = CRHIT_117A_CARBON-8.j
Sample_id               = CRHIT_117A
Machine                 = mt-su500ap
Creation_time           = 28-NOV-2016 14:07:38
Comment                 = Serine N-O Benzyl

Field_strength          = 11.7473579[T] (500[MH
X_acq_duration          = 0.83361792[s]
X_domain                = 13C
X_freq                  = 125.76529768[MHz]
X_offset                = 100[ppm]
X_points                = 32768
X_prescans              = 4
X_resolution            = 1.19959034[Hz]
X_sweep                 = 39.3081761[kHz]
Irr_domain              = 1H
Irr_freq                = 500.15991521[MHz]
Irr_offset              = 5.0[ppm]
Clipped                 = FALSE
Mod_return              = 1
Scans                   = 270
Total_scans             = 270

X_90_width              = 10.239[us]
X_acq_time              = 0.83361792[s]
X_angle                 = 30[deg]
X_atn                   = 9[dB]
X_pulse                 = 3.413[us]
Irr_atn_dec             = 21.5[dB]
Irr_atn_noe             = 21.5[dB]
Irr_noise               = WALTZ
Decoupling              = TRUE
Initial_wait            = 1[s]
Noe                     = TRUE
Noe_time                = 2[s]
Relaxation_delay        = 2[s]
Repetition_time         = 2.83361792[s]
Temp_state              = TEMP OFF
Temp_set                = 25[dC]
Temp_get                = 23.1[dC]
```


Serine N-O Benzyl
abundance
X : parts per Million : 13C
129.4067
128.9393
128.6340
128.4624
128.3098
128.1857
78.4434
67.6269
67.2740
62.6288
56.3240
Filename = CRHIT_117A_DEPT135-4.
Sample_id = CRHIT_117A
Machine = mt-su500ap
Creation_time = 28-NOV-2016 13:41:45
Comment = Serine N-O Benzyl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.8336792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 14
Total_scans = 14
X_acq_time = 0.8336792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23[dC]


Y : parts per Million : 1H
X : parts per Million : 1H
abundance
Serine N-O Benzyl


Y : parts per Million : 13C
X : parts per Million : 1H
abundance
Serine N-O Benzyl

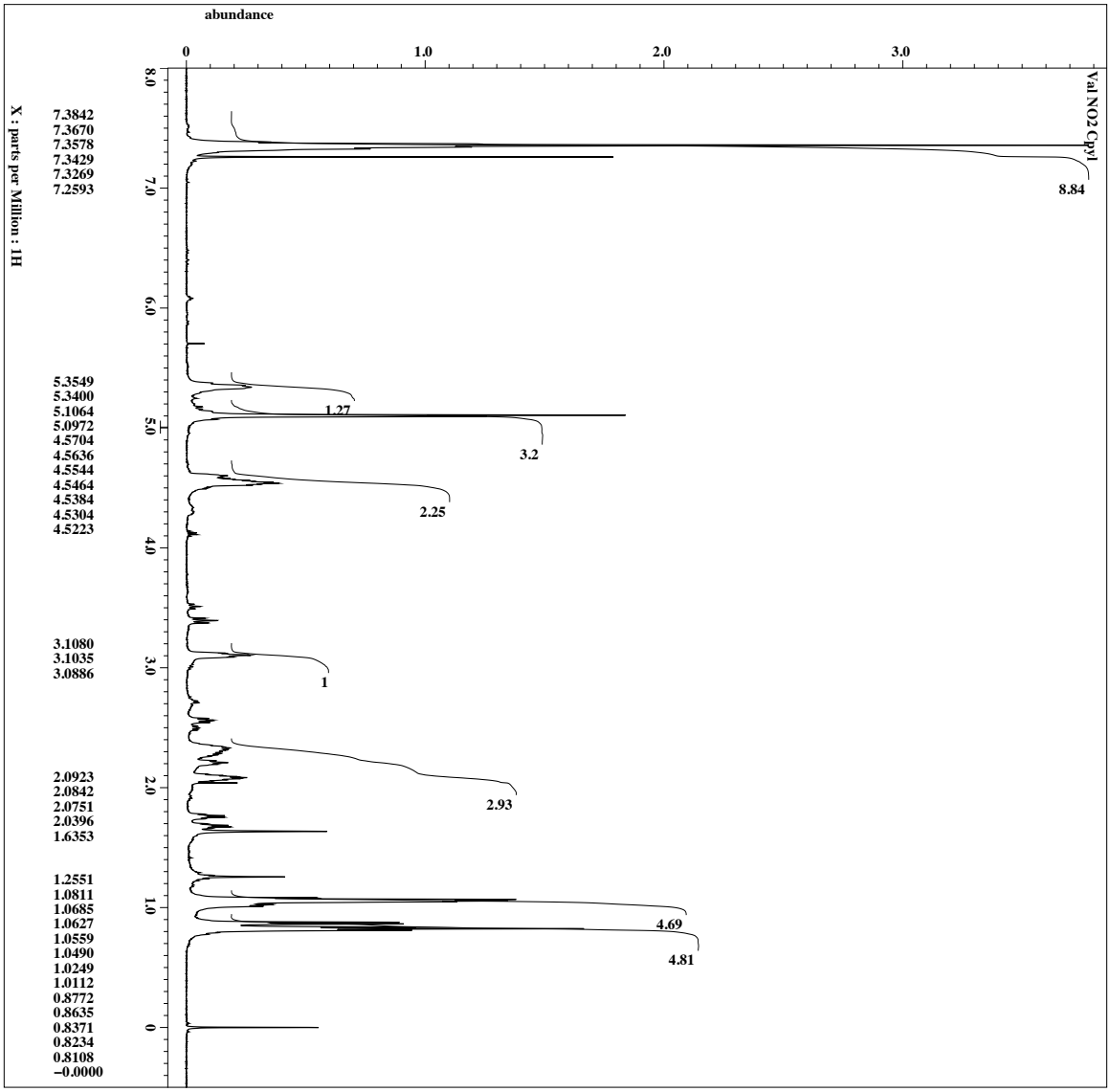
Benzyl N-[3-methyl-1-(2-nitrocyclopropyl)-1-oxobutan-2-yl]carbamate (55)

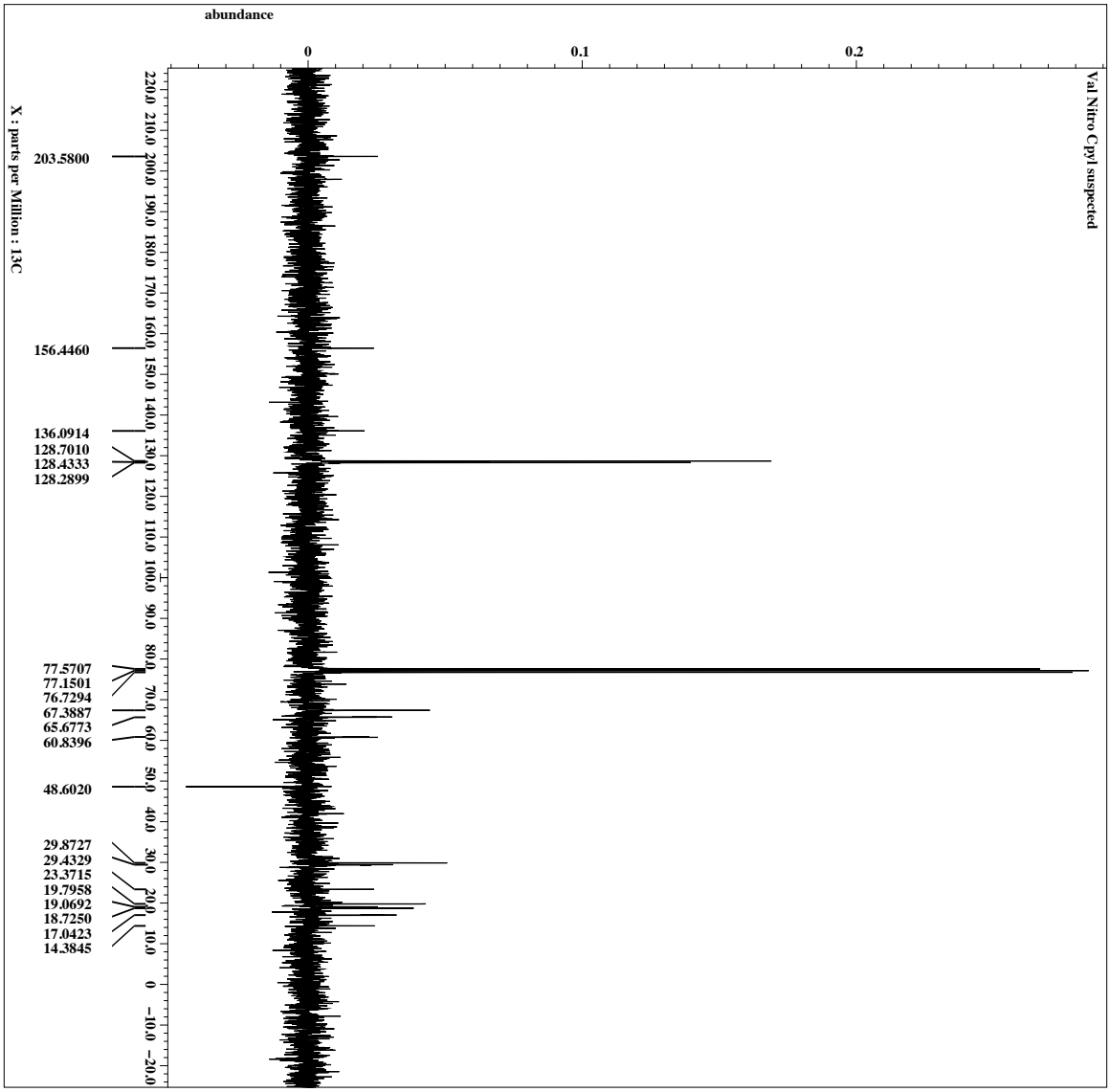


NMR

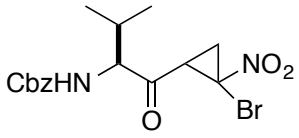
^{1}H-NMR ($CDCl_3$)

^{13}C-NMR ($CDCl_3$)


abundance
0
1.0
2.0
3.0
Val NO2 Cyl
8.84
4.81
4.69
3.2
2.93
2.25
1.27
1
X : parts per Million : 1H
7.3842
7.3670
7.3578
7.3429
7.3269
7.2593
5.3549
5.3400
5.1064
5.0972
4.5704
4.5636
4.5544
4.5464
4.5384
4.5304
4.5223
3.1080
3.1035
3.0886
2.0923
2.0842
2.0751
2.0396
1.6353
1.2551
1.0811
1.0685
1.0627
1.0559
1.0490
1.0249
1.0112
0.8772
0.8635
0.8371
0.8234
0.8108
−0.0000
Filename = CRHII_51_PROTON-5.jdf
Sample_id = CRHII_51
Machine = mt-su500ap
Creation_time = 31-MAY-2016 17:38:46
Comment = Val NO2 Cyl
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 24[dC]


Val Nitro Cpyl suspected
abundance
0
0.1
0.2
X : parts per Million : 13C
203.5800
156.4460
136.0914
128.7010
128.4333
128.2899
77.5707
77.1501
76.7294
67.3887
65.6773
60.8396
48.6020
29.8727
29.4329
23.3715
19.7958
19.0692
18.7250
17.0423
14.3845
Filename = CRHII_39_3_CARBON-7.j
Sample_id = CRHII_39_3
Machine = mt-eu300sp
Creation_time = 20-MAY-2016 11:52:56
Comment = Val Nitro Cpyl suspec
Field_strength = 7.0586013[T] (300[MHz
X_acq_duration = 1.3841203[s]
X_domain = 13C
X_freq = 75.5682342[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 0.7224805[Hz]
X_sweep = 23.6742424[kHz]
Irr_domain = 1H
Irr_freq = 300.5296559[MHz]
Irr_offset = 5[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 48
Total_scans = 48
X_90_width = 9.78[us]
X_acq_time = 1.3841203[s]
X_angle = 30[deg]
X_atn = 6[dB]
X_pulse = 3.26[us]
Irr_atn_dec = 24.6[dB]
Irr_atn_noe = 24.6[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 3.3841203[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.7[dC]

Benzyl N-[1-(2-bromo-2-nitrocyclopropyl)-3-methyl-1-oxobutan-2-yl]carbamate (56)



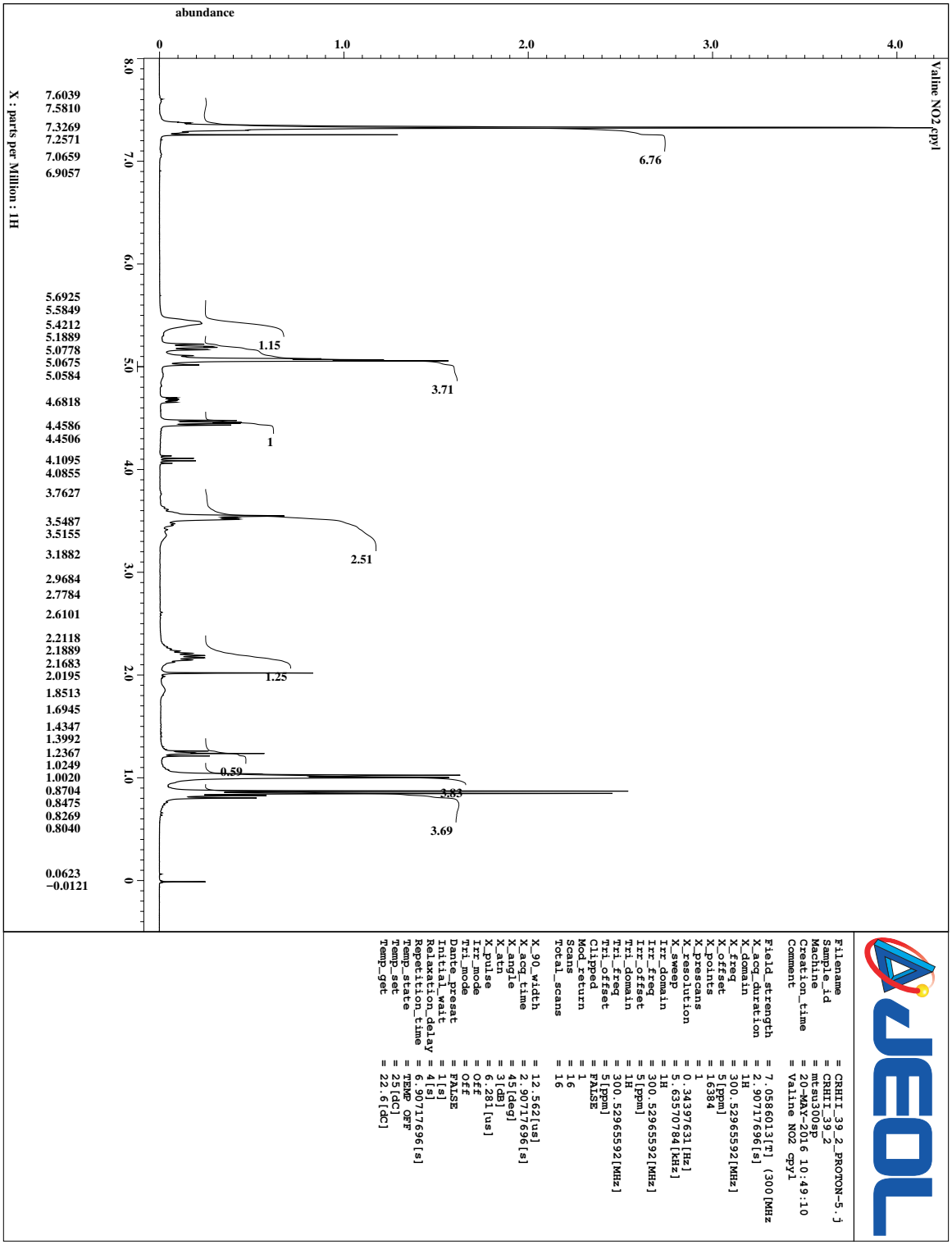
NMR

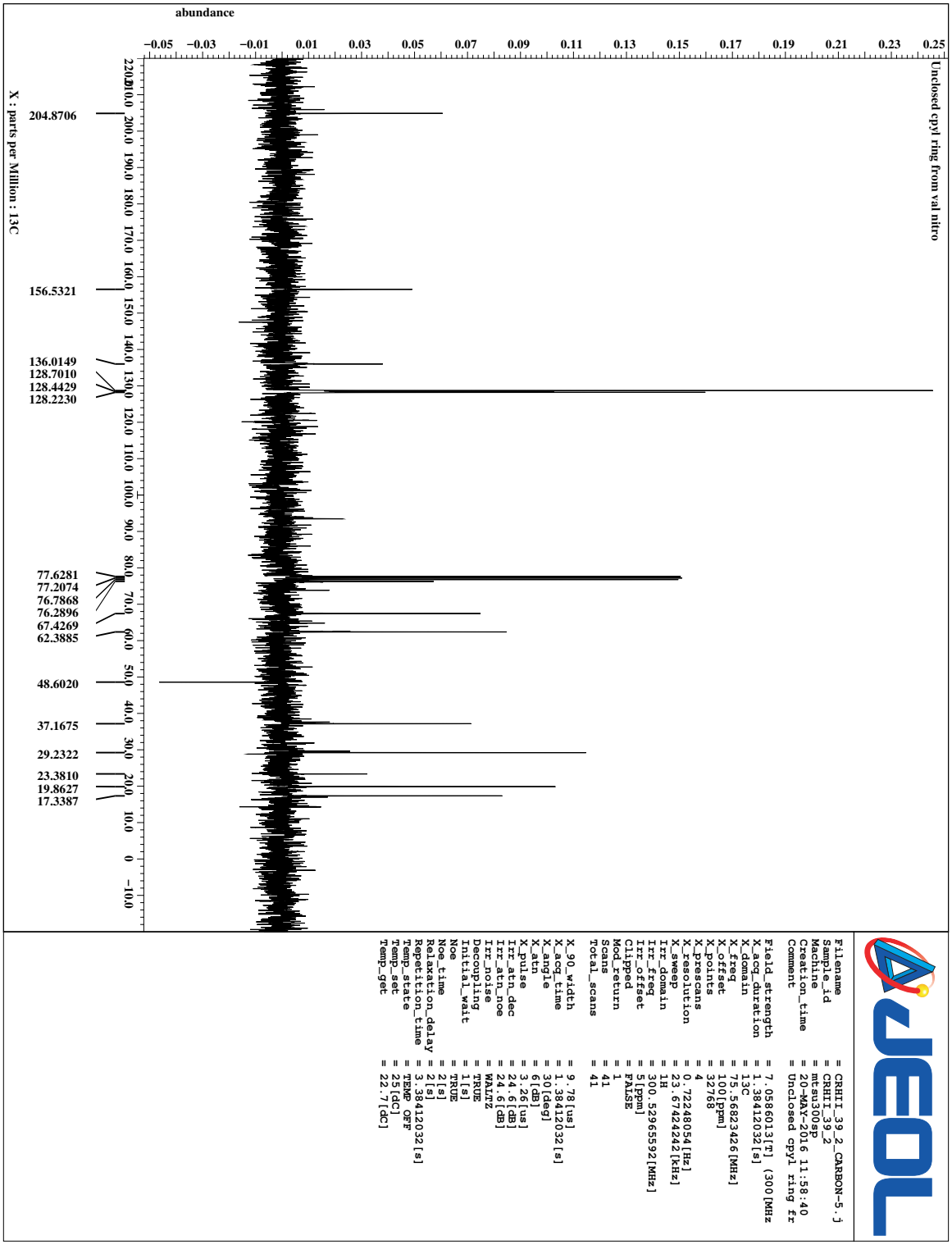
^{1}H-NMR ($CDCl_3$)

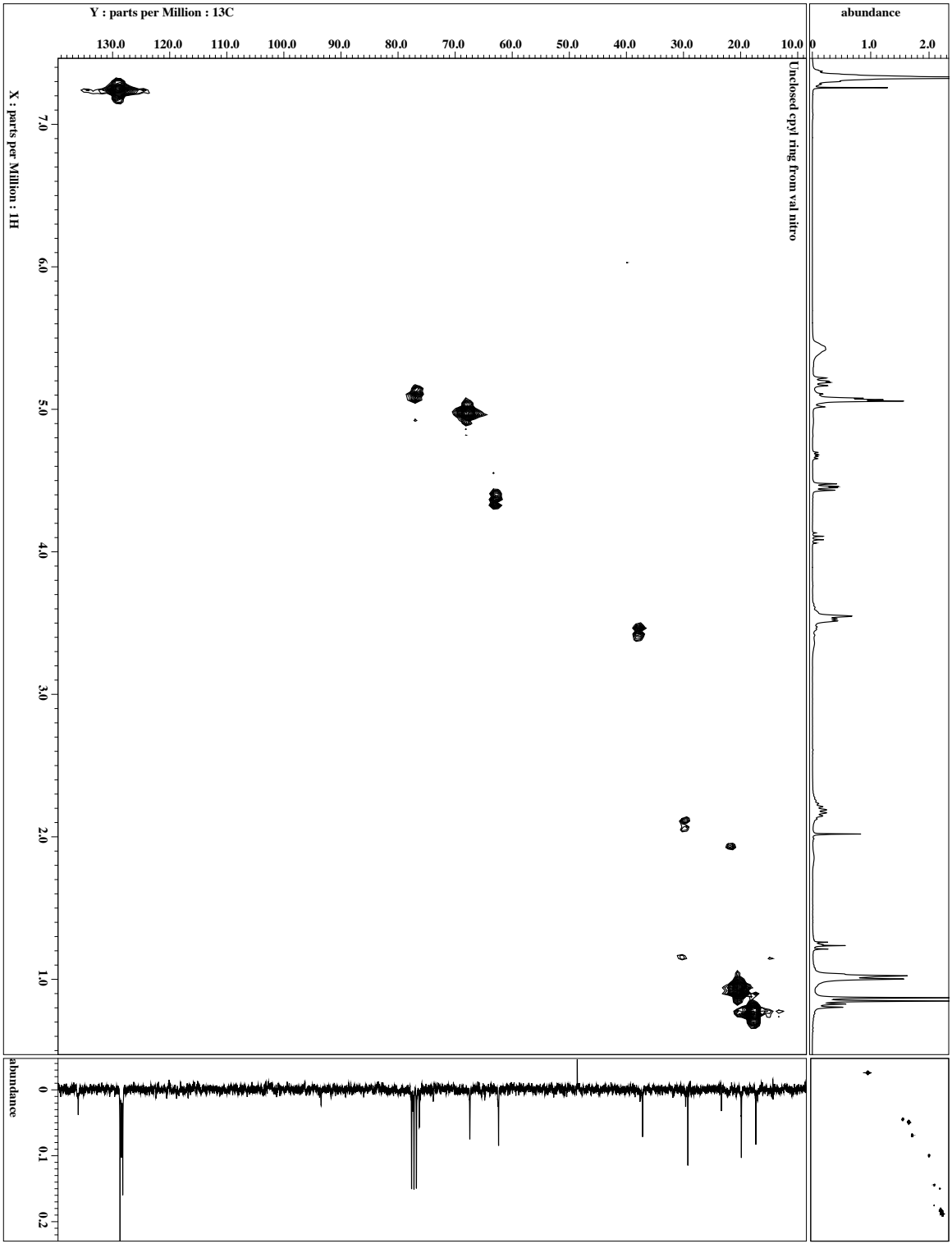
^{13}C-NMR ($CDCl_3$)

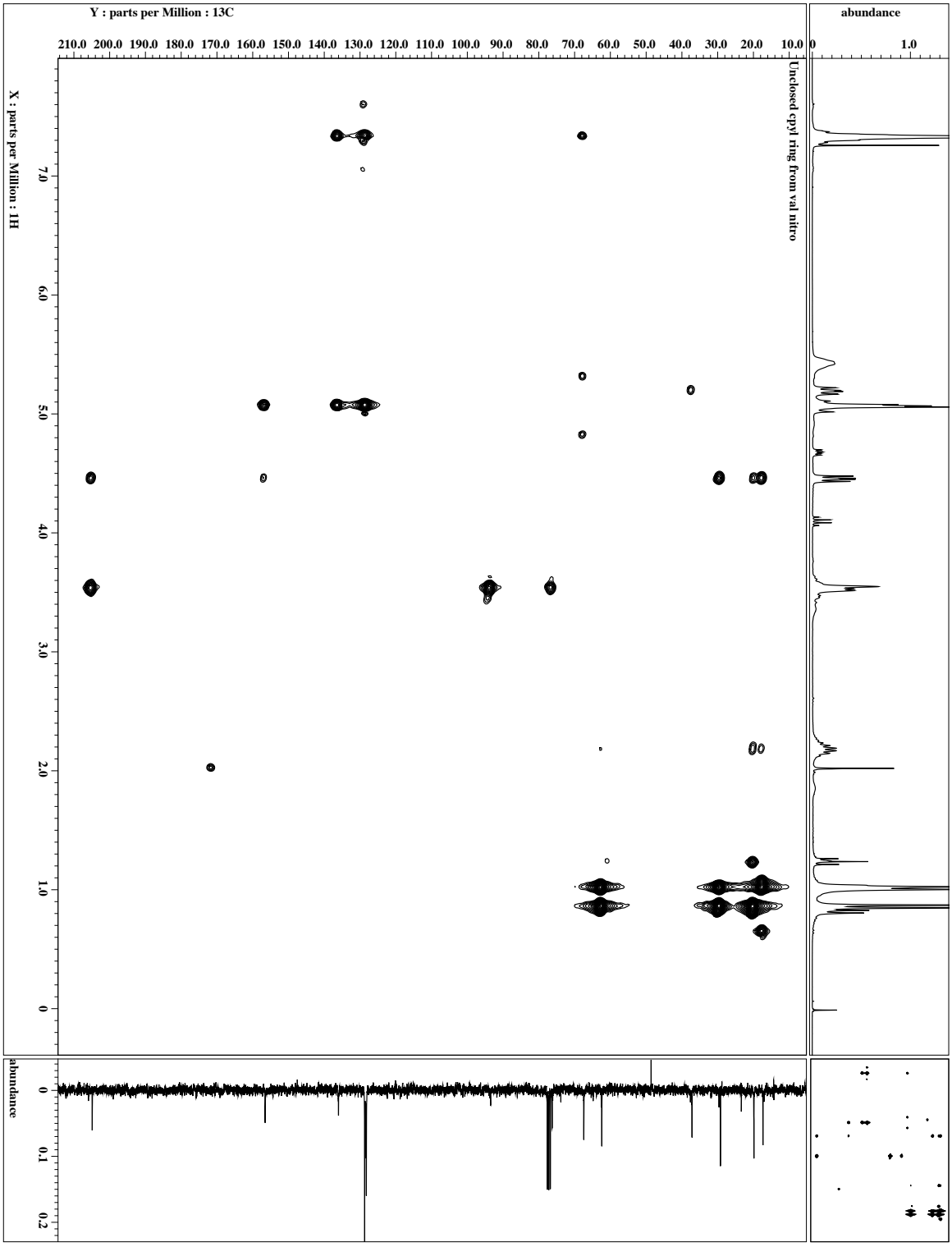
HMQC ($CDCl_3$)

HMBC ($CDCl_3$)

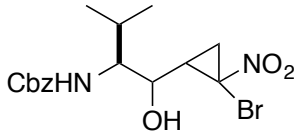

abundance
Valine NO2 cpyl
X : parts per Million : 1H
7.6039
7.5810
7.3269
7.2571
7.0659
6.9057
5.6925
5.5849
5.4212
5.1889
5.0778
5.0675
5.0584
4.6818
4.4586
4.4506
4.1095
4.0855
3.7627
3.5487
3.5155
3.1882
2.9684
2.7784
2.6101
2.2118
2.1889
2.1683
2.0195
1.8513
1.6945
1.4347
1.3992
1.2367
1.0249
1.0020
0.8704
0.8475
0.8269
0.8040
0.0623
−0.0121
6.76
1.15
3.71
1
2.51
1.25
0.59
3.83
3.69
Filename = CRHI_39_2_PROTON-5.j
Sample_id = CRHI_39_2
Machine = mt-su300sp
Creation_time = 20-MAY-2016 10:49:10
Comment = Valine NO2 cpyl
Field_strength = 7.0586013[T] (300[MHz
X_acq_duration = 2.90717696[s]
X_domain = 1H
X_freq = 300.52965592[MHz]
X_offset = 5[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.34397631[Hz]
X_sweep = 5.63570784[kHz]
Irr_domain = 1H
Irr_freq = 300.52965592[MHz]
Irr_offset = 5[ppm]
Tri_domain = 1H
Tri_freq = 300.52965592[MHz]
Tri_offset = 5[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 12.562[us]
X_acq_time = 2.90717696[s]
X_angle = 45[deg]
X_atn = 3[dB]
X_pulse = 6.281[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 6.90717696[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.6[dC]


Unclosed cpyl ring from val nitro
abundance
X : parts per Million : 13C
204.8706
156.5321
136.0149
128.7010
128.4429
128.2230
77.6281
77.2074
76.7868
76.2896
67.4269
62.3885
48.6020
37.1675
29.2322
23.3810
19.8627
17.3387
Filename = CRHI1_39_2_CARBON-5.j
Sample_id = CRHI1_39_2
Machine = mt-su300ap
Creation_time = 20-MAY-2016 11:58:40
Comment = Unclosed cpyl ring fr
Field_strength = 7.0586013[T] (300[MHz
X_acq_duration = 1.3841203[s]
X_domain = 13C
X_freq = 75.5682426[MHz]
X_offset = 100[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 0.72248054[Hz]
X_sweep = 23.674242[kHz]
X_domain = 1H
Irr_freq = 300.52965592[MHz]
Irr_offset = 5[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 41
Total_scans = 41
X_90_width = 9.78[us]
X_acq_time = 1.3841203[s]
X_angle = 30[deg]
X_atn = 6[dB]
X_pulse = 3.26[us]
Irr_atn_dec = 24.6[dB]
Irr_atn_noe = 24.6[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 3.3841203[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 22.7[dC]


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
Unclosed cpyl ring from val nitro
abundance
0
1.0
2.0
abundance
0
0.1
0.2


Y : parts per Million : 13C
210.0 200.0 190.0 180.0 170.0 160.0 150.0 140.0 130.0 120.0 110.0 100.0 90.0 80.0 70.0 60.0 50.0 40.0 30.0 20.0 10.0
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
Unclosed cpyl ring from val nitro
abundance
0
1.0
abundance
0
0.1
0.2

Benzyl N-[1-(2-bromo-2-nitrocyclopropyl)-1-hydroxy-3-methylbutan-2-yl]carbamate (57)



NMR

^{1}H-NMR ($CDCl_3$)

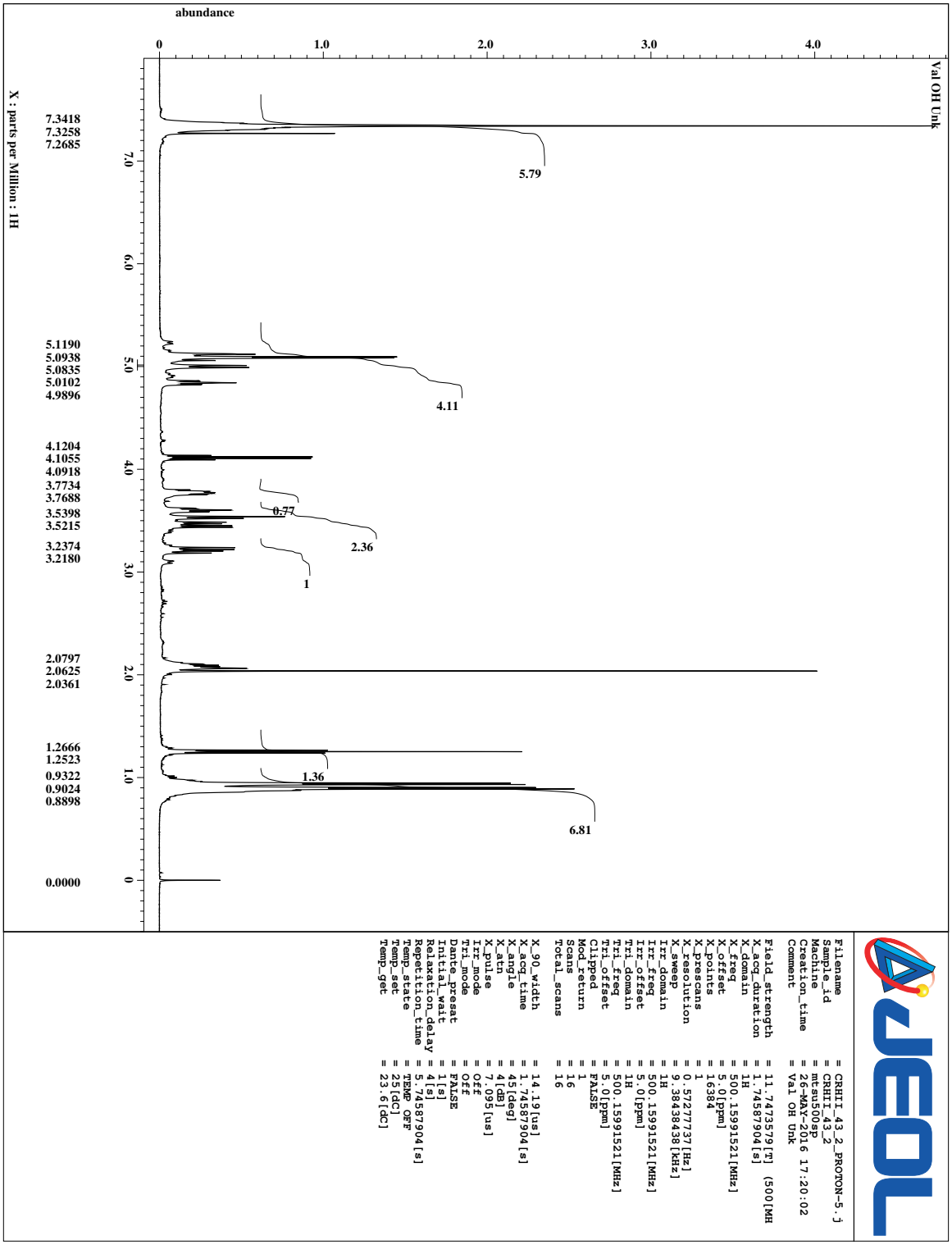
^{13}C-NMR ($CDCl_3$)

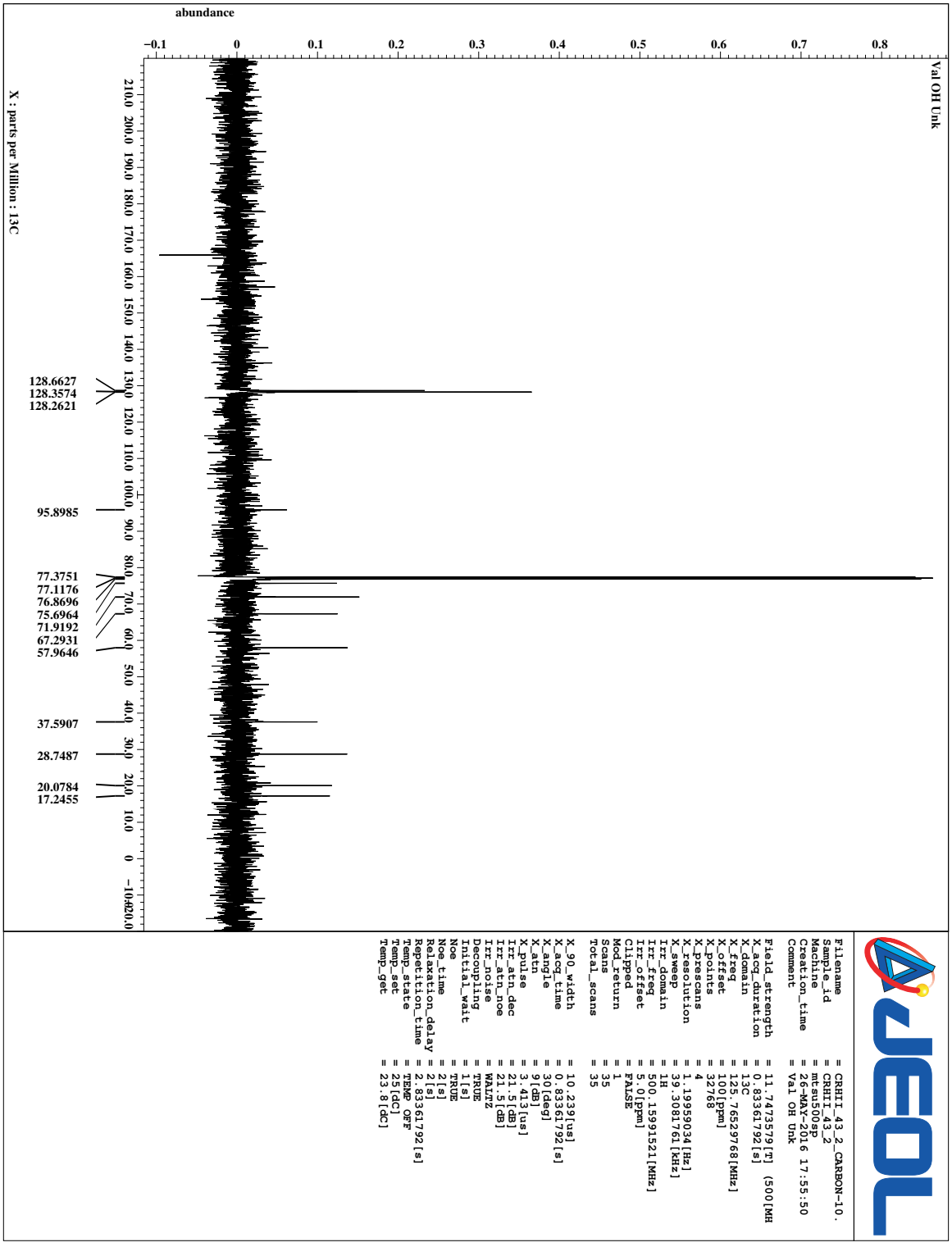
$DEPT_{135}$ ($CDCl_3$)

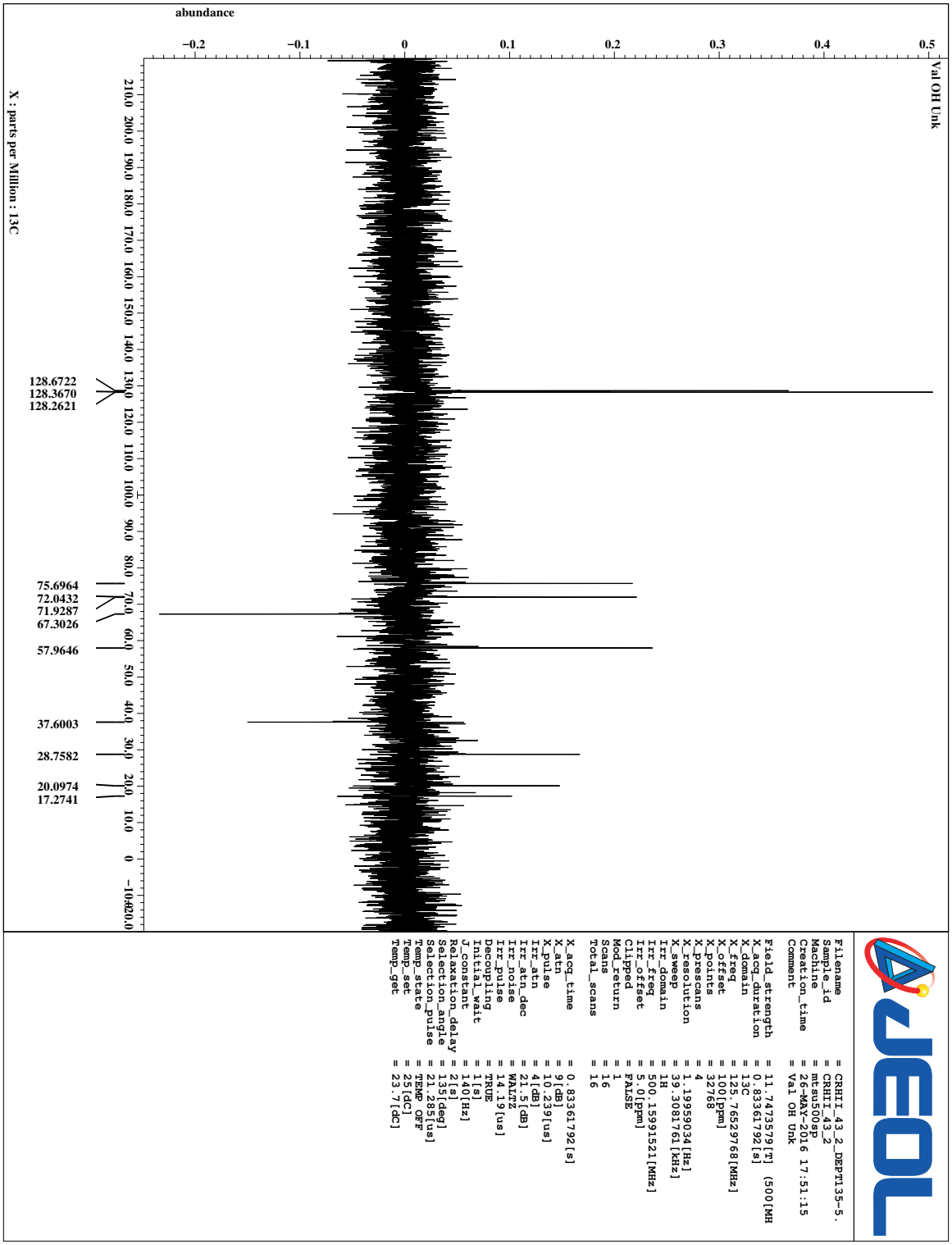
COSY ($CDCl_3$)

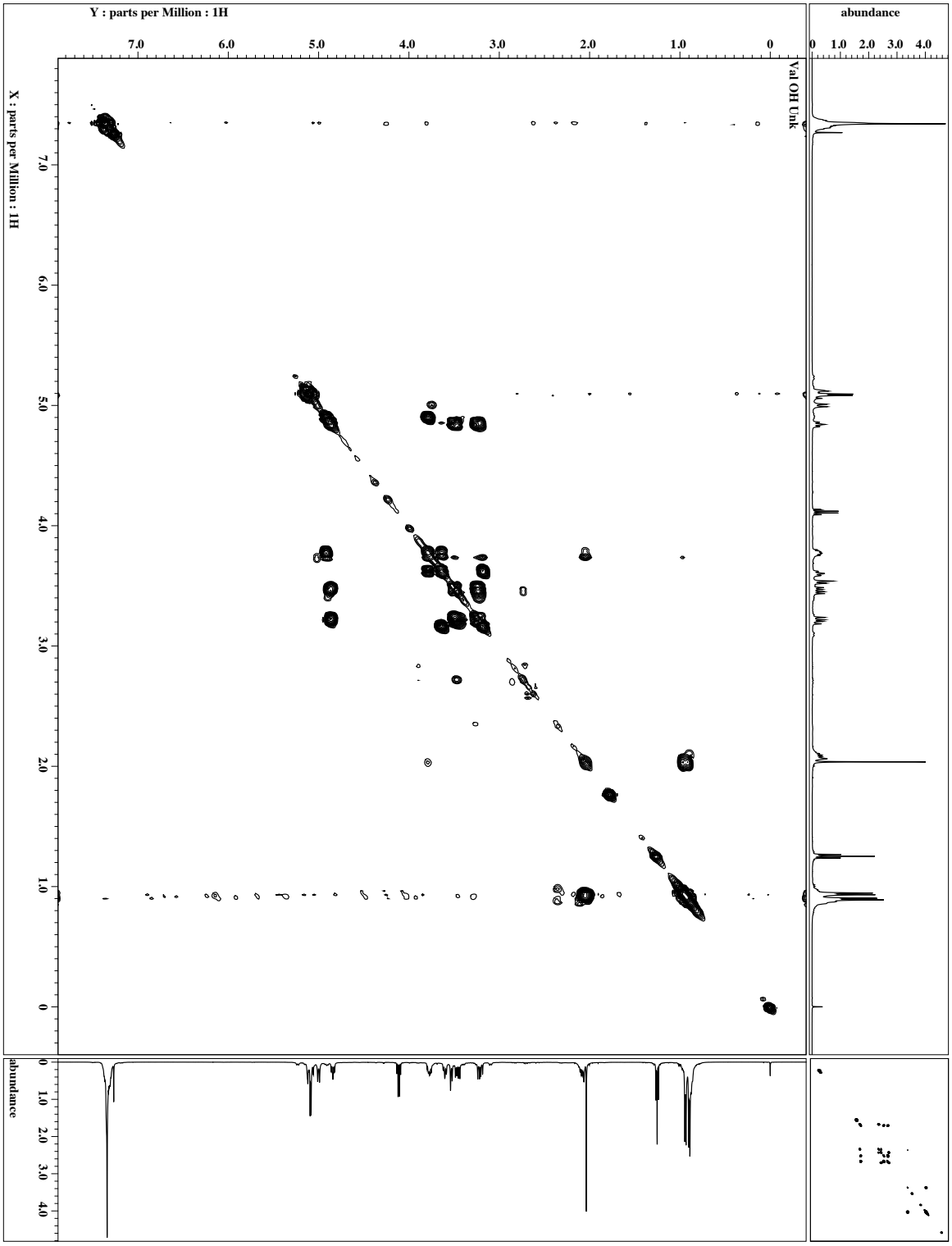
HMQC ($CDCl_3$)

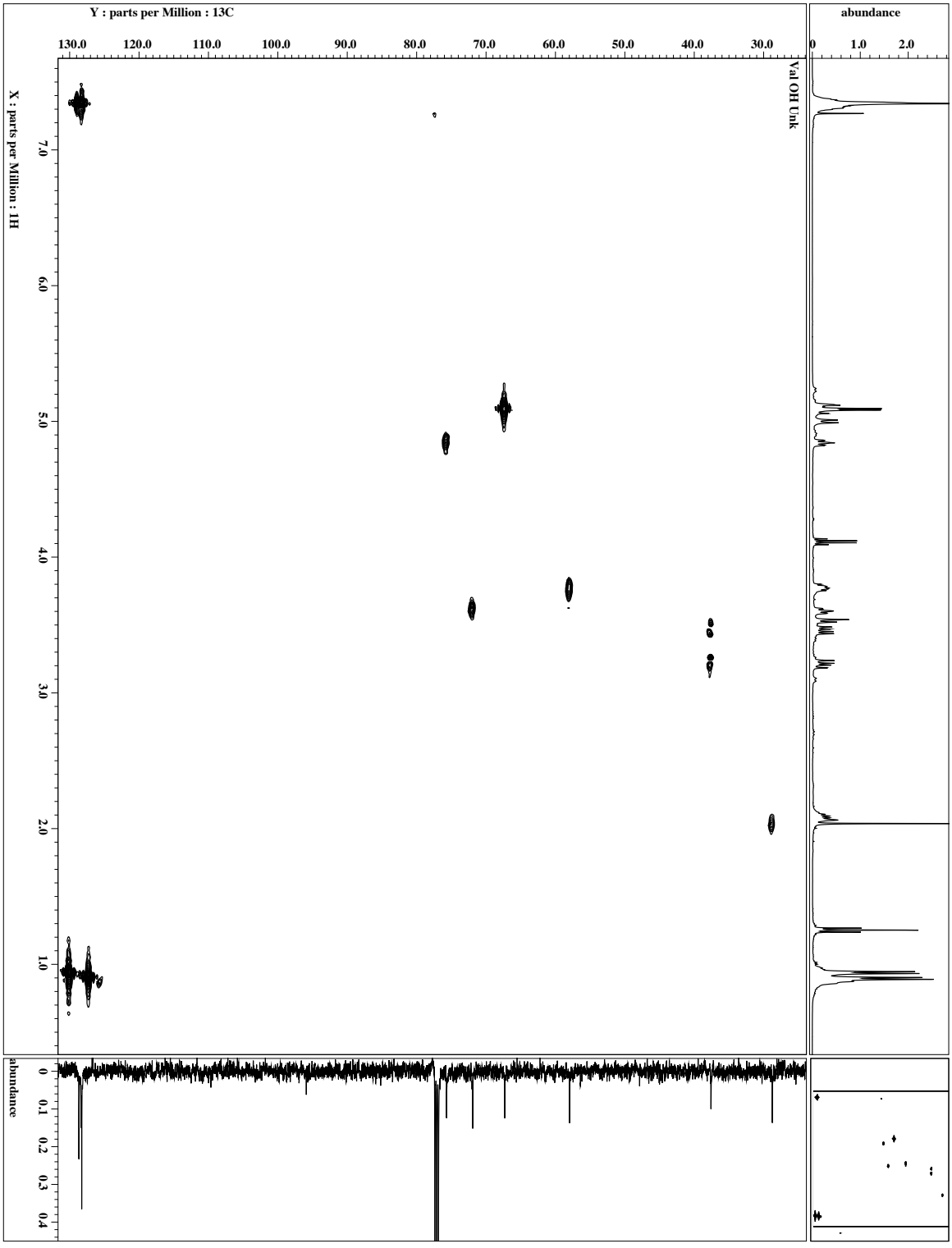
HMBC ($CDCl_3$)

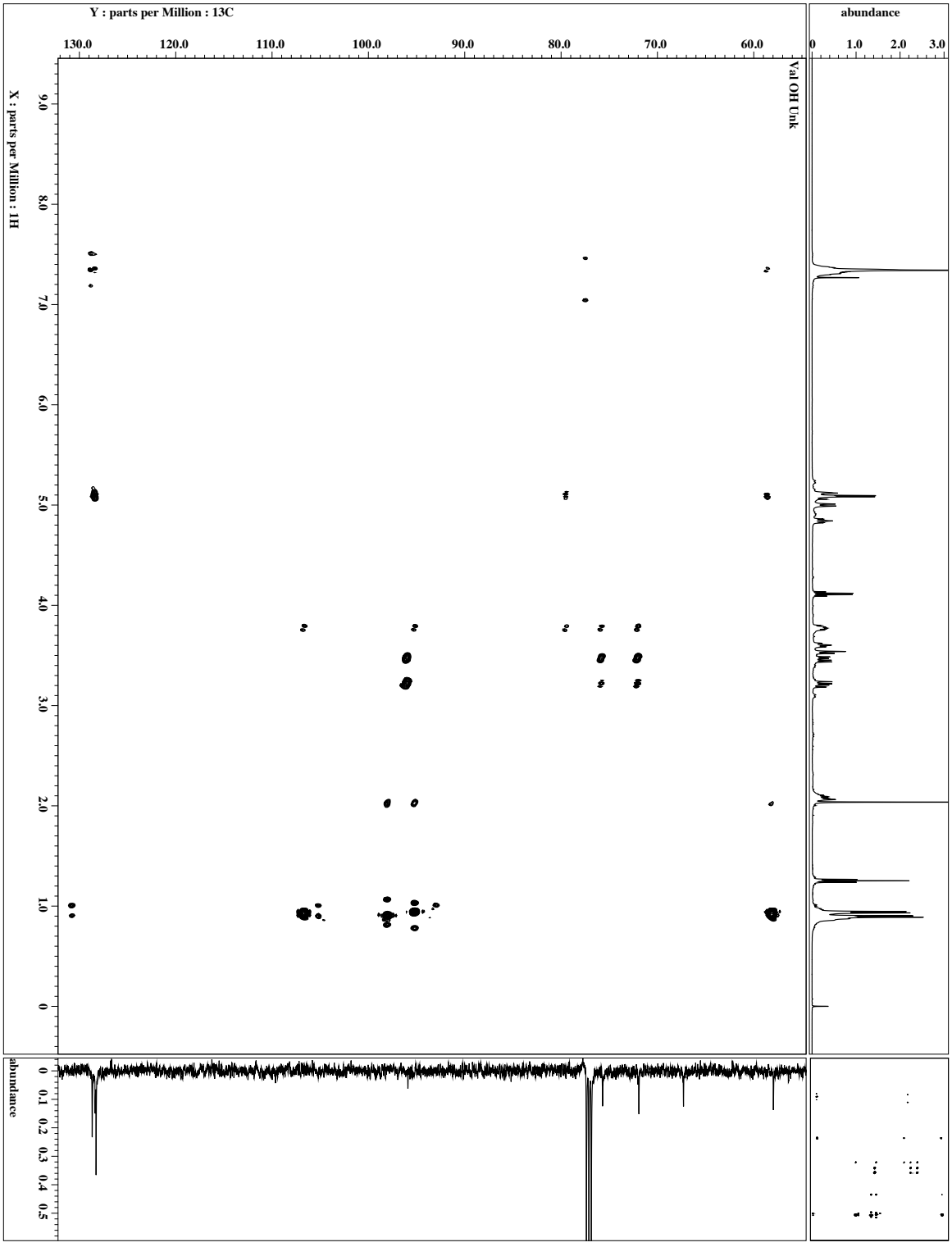

abundance
Val OH Unk
X : parts per Million : 1H
7.3418
7.3258
7.2685
5.1190
5.0938
5.0835
5.0102
4.9896
4.1204
4.1055
4.0918
3.7734
3.7688
3.5398
3.5215
3.2374
3.2180
2.0797
2.0625
2.0361
1.2666
1.2523
0.9322
0.9024
0.8898
0.0000
5.79
4.11
0.77
2.36
1
1.36
6.81
Filename = CRHII_43_2_PROTON-5.j
Sample_id = CRHII_43_2
Machine = mt-su500sp
Creation_time = 26-MAY-2016 17:20:02
Comment = Val OH Unk
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 1.74587904[s]
X_domain = 1H
X_freq = 500.15991521[MHz]
X_offset = 5.0[ppm]
X_points = 16384
X_prescans = 1
X_resolution = 0.57277737[Hz]
X_sweep = 9.38438438[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Tri_domain = 1H
Tri_freq = 500.15991521[MHz]
Tri_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_90_width = 14.19[us]
X_acq_time = 1.74587904[s]
X_angle = 45[deg]
X_atn = 4[dB]
X_pulse = 7.095[us]
Irr_mode = Off
Tri_mode = Off
Dante_presat = OFF
Initial_wait = 1[s]
Relaxation_delay = 4[s]
Repetition_time = 5.74587904[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.6[dC]


Val OH Unk
abundance
X : parts per Million : 13C
128.6627
128.3574
128.2621
95.8985
77.3751
77.1176
76.8696
75.6964
71.9192
67.2931
57.9646
37.5907
28.7487
20.0784
17.2455
Filename = CRHII_43_2_CARBON-10.
Sample_id = CRHII_43_2
Machine = mt-su500ap
Creation_time = 26-MAY-2016 17:55:50
Comment = Val OH Unk
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
X_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 35
Total_scans = 35
X_90_width = 10.239[us]
X_acq_time = 0.83361792[s]
X_angle = 30[deg]
X_atn = 9[dB]
X_pulse = 3.413[us]
Irr_atn_dec = 21.5[dB]
Irr_atn_noe = 21.5[dB]
Irr_noise = WALTZ
Decoupling = TRUE
Initial_wait = 1[s]
Noe = TRUE
Noe_time = 2[s]
Relaxation_delay = 2[s]
Repetition_time = 2.83361792[s]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.8[dC]


Val OH Unk
abundance
X : parts per Million : 13C
128.6722
128.3670
128.2621
75.6964
72.0432
71.9287
67.3026
57.9646
37.6003
28.7582
20.0974
17.2741
Filename = CRHII_43_2_DEPT135-5.
Sample_id = CRHII_43_2
Machine = mt-su500ap
Creation_time = 26-MAY-2016 17:51:15
Comment = Val OH Unk
Field_strength = 11.7473579[T] (500[MH
X_acq_duration = 0.83361792[s]
X_domain = 13C
X_freq = 125.76529768[MHz]
X_offset = 100.[ppm]
X_points = 32768
X_prescans = 4
X_resolution = 1.19959034[Hz]
X_sweep = 39.3081761[kHz]
Irr_domain = 1H
Irr_freq = 500.15991521[MHz]
Irr_offset = 5.0[ppm]
Clipped = FALSE
Mod_return = 1
Scans = 16
Total_scans = 16
X_acq_time = 0.83361792[s]
X_atn = 9[dB]
X_pulse = 10.239[us]
Irr_atn = 4[dB]
Irr_atn_dec = 21.5[dB]
Irr_noise = WALTZ
Irr_pulse = 14.19[us]
Decoupling = TRUE
Initial_wait = 1[s]
J_constant = 140[Hz]
Relaxation_delay = 2[s]
Selection_angle = 135[deg]
Selection_pulse = 21.285[us]
Temp_state = TEMP OFF
Temp_set = 25[dC]
Temp_get = 23.7[dC]


Y : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
abundance
0
1.0
2.0
3.0
4.0
Val OH Unk
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
abundance
0
1.0
2.0
3.0
4.0


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
50.0
40.0
30.0
X : parts per Million : 1H
7.0
6.0
5.0
4.0
3.0
2.0
1.0
Val OH Unk
abundance
0
1.0
2.0
abundance
0
0.1
0.2
0.3
0.4


Y : parts per Million : 13C
130.0
120.0
110.0
100.0
90.0
80.0
70.0
60.0
X : parts per Million : 1H
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0
Val OH Unk
abundance
0
1.0
2.0
3.0
abundance
0
0.1
0.2
0.3
0.4
0.5