Supplementary Information (SI) for Organic & Biomolecular Chemistry.
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Supporting Information for:

A β-Hairpin Peptide Derived from Aβ Forms Different Oligomers in the Crystal State and in Aqueous Solution

Jason Zhu,[a] Adam G. Kreutzer,[a] Zhiwei Liu,[b] Xingyue Li,[a] Sabrina M. Richter,[a] Vojislava Pophristic,[b] James S. Nowick*[a,c,d]

[a]Department of Chemistry, University of California, Irvine, Irvine California 92697-2025, United States

[b]Department of Chemistry and Biochemistry, Rowan University, Glassboro, New Jersey 08028-1701, United States

[c]Department of Pharmaceutical Sciences, University of California, Irvine, Irvine California 92697-2025, United States

[d]Department of Molecular Biology and Biochemistry, University of California, Irvine, Irvine California 92697-2025, United States

*Corresponding author
James S. Nowick, 4126 Natural Sciences 1, University of California Irvine, Irvine, CA 92697

Email: jsnowick@uci.edu

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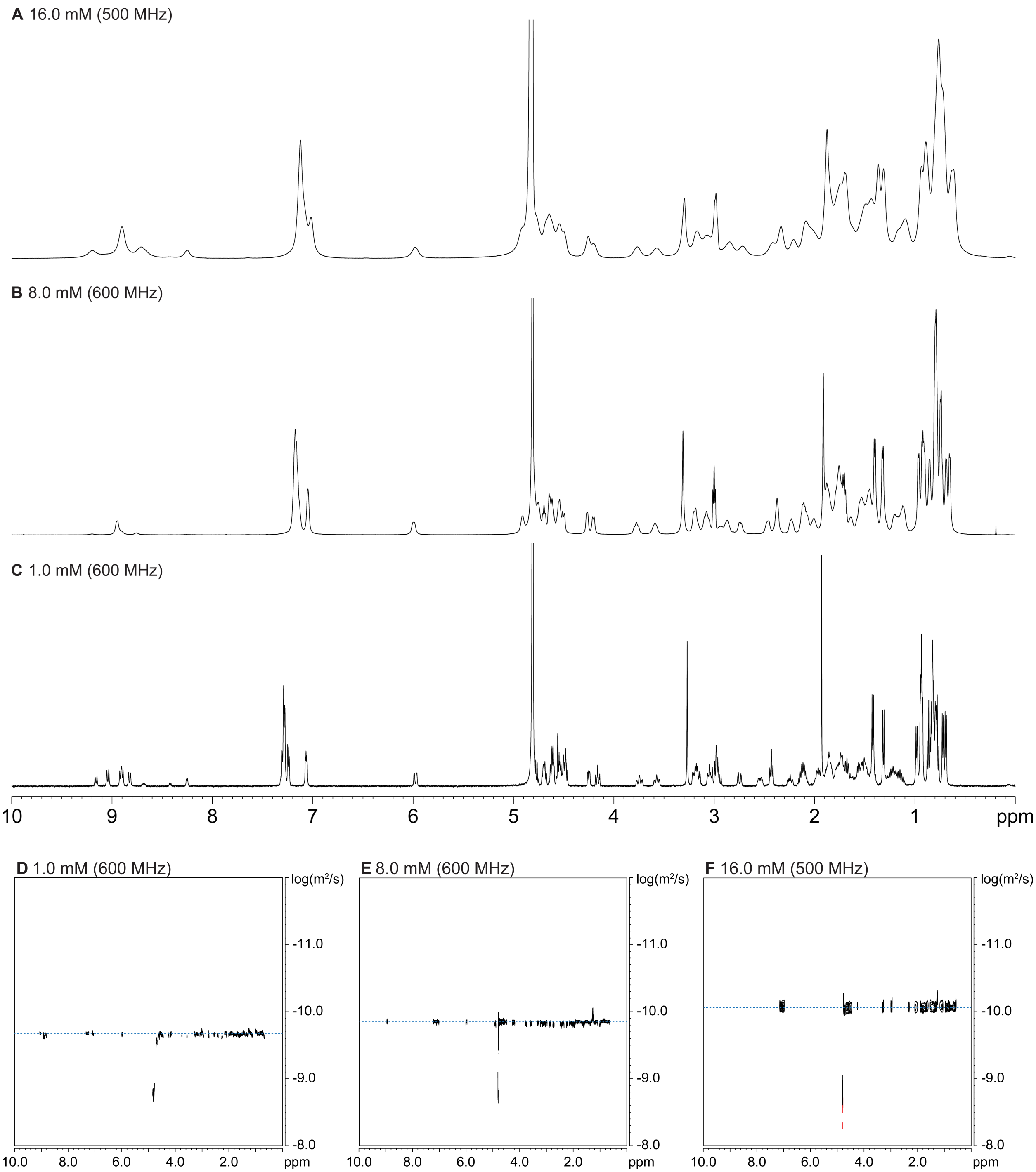
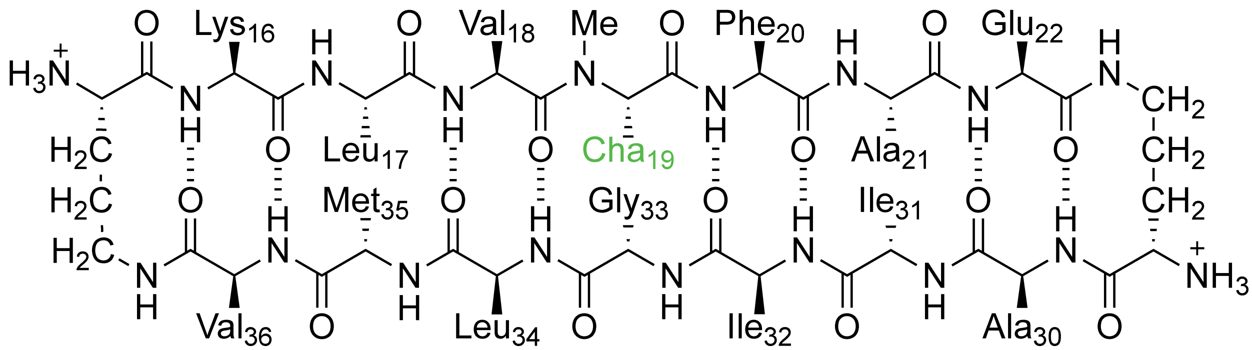


$10^{-9.6715} =$ $2.13 \pm 0.13 \times 10^{-10}\ m^2/s$

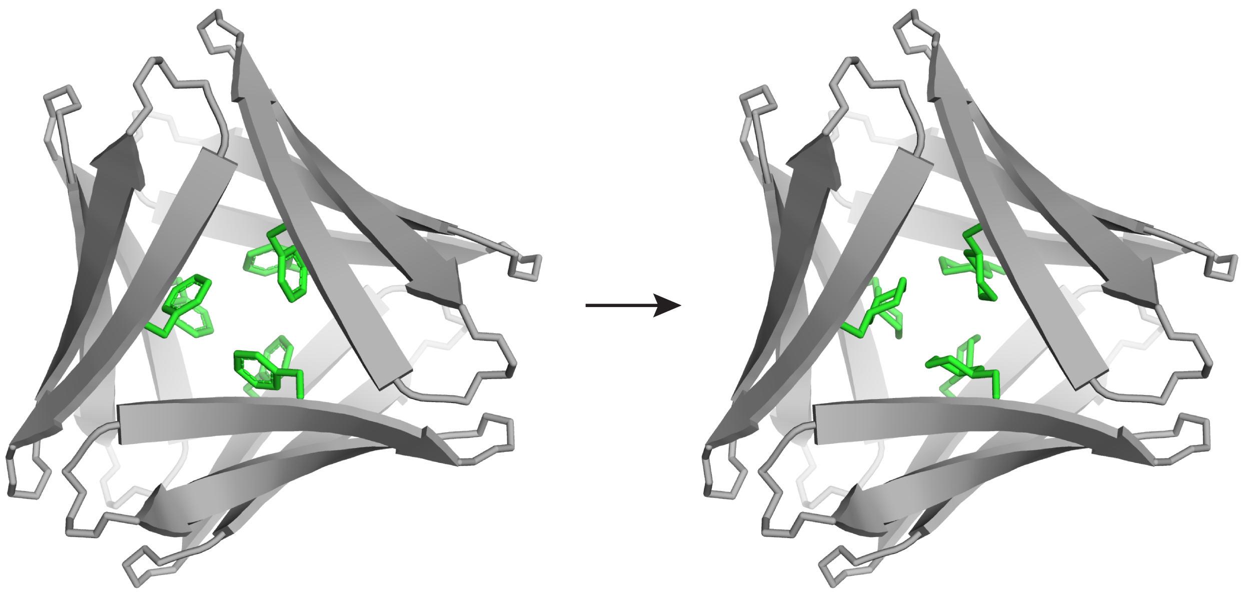
$10^{-9.8487} =$ $1.42 \pm 0.11 \times 10^{-10}\ m^2/s$

$10^{-10.063} =$ $0.86 \pm 0.13 \times 10^{-10}\ m^2/s$

Figure S1. ^1H NMR and DOSY spectra of peptide **1** at 1.0, 8.0, and 16.0 mM in D_2O at 298 K. The peaks between 7.5 – 10 ppm correspond to incomplete NH exchange with D_2O. The HOD peak is calibrated to $19.0 \times 10^{-10}\ m^2/s$.[1]



peptide **2**



Phe19 Cha19

Figure S2. Molecular models illustrating the Phe19 to Cha19 mutation shown in green. Models based on the peptide **1** crystallographic hexamer (PDB 5W4H).

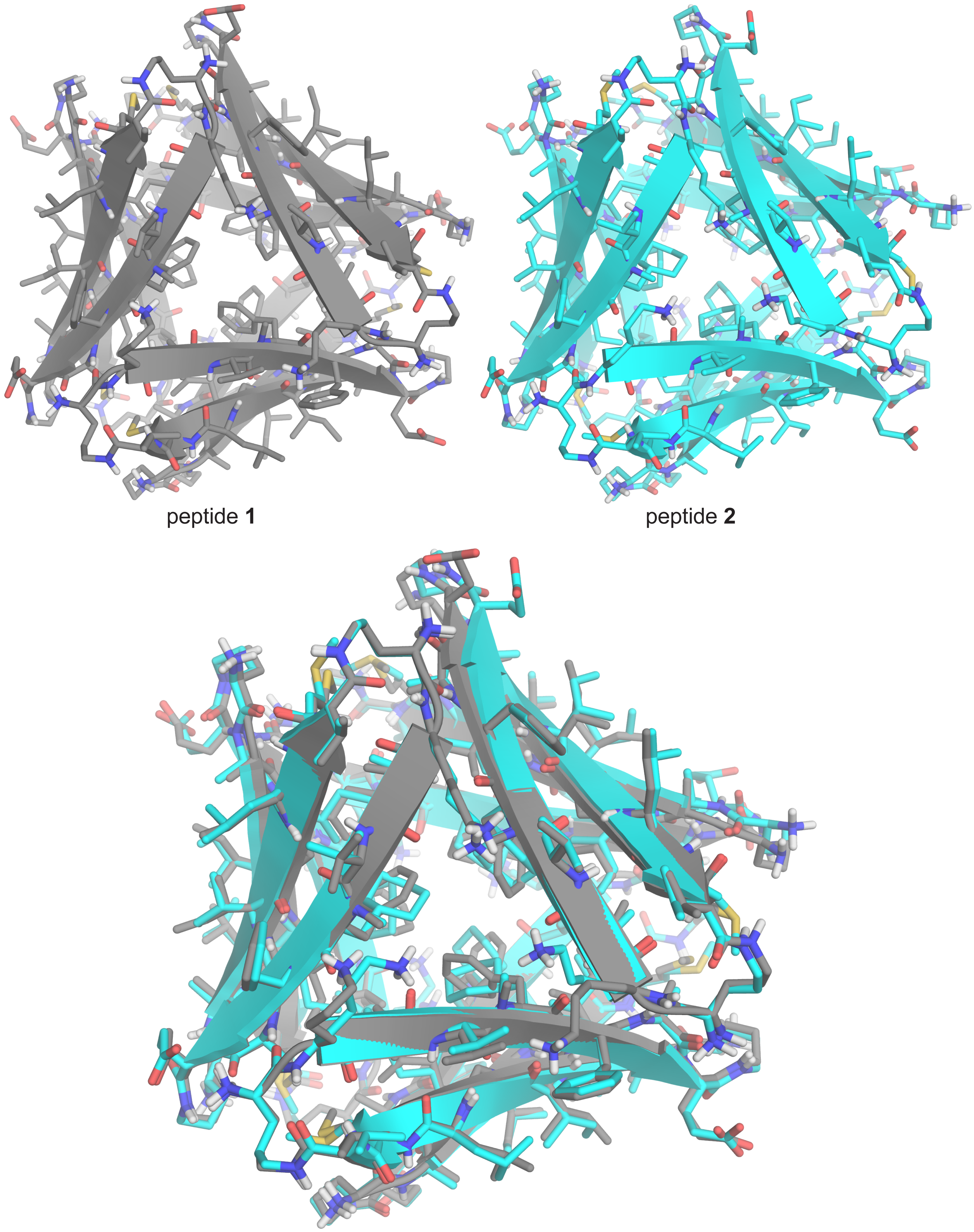


Figure S3. Peptide **1** and peptide **2** crystallographic hexamer side-by-side and overlay. Crystallographic coordinates taken from PDB 5W4H and 9EEC.

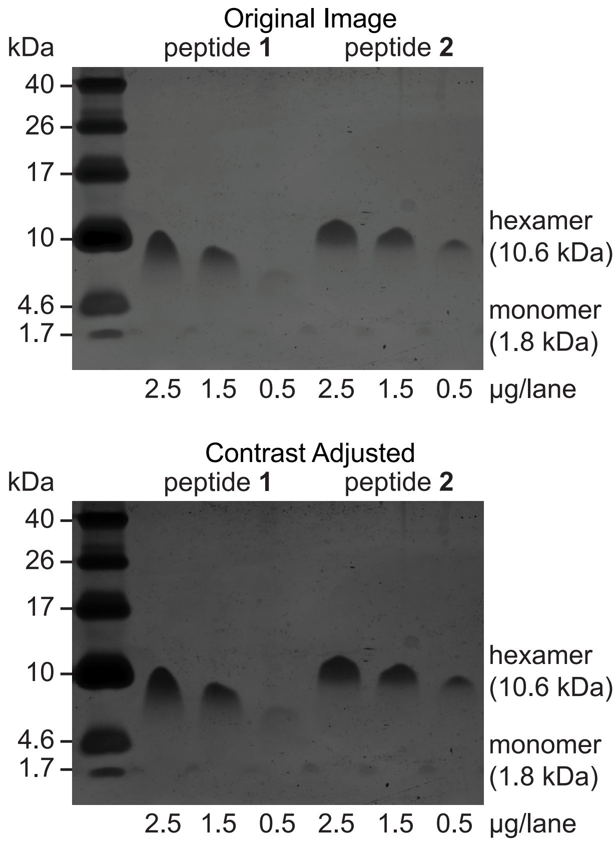


Figure S4. Two images of the silver-stained SDS-PAGE gel from Figure **3**. In the top image the contrast has not been adjusted; in the bottom image the contrast has been uniformly adjusted.

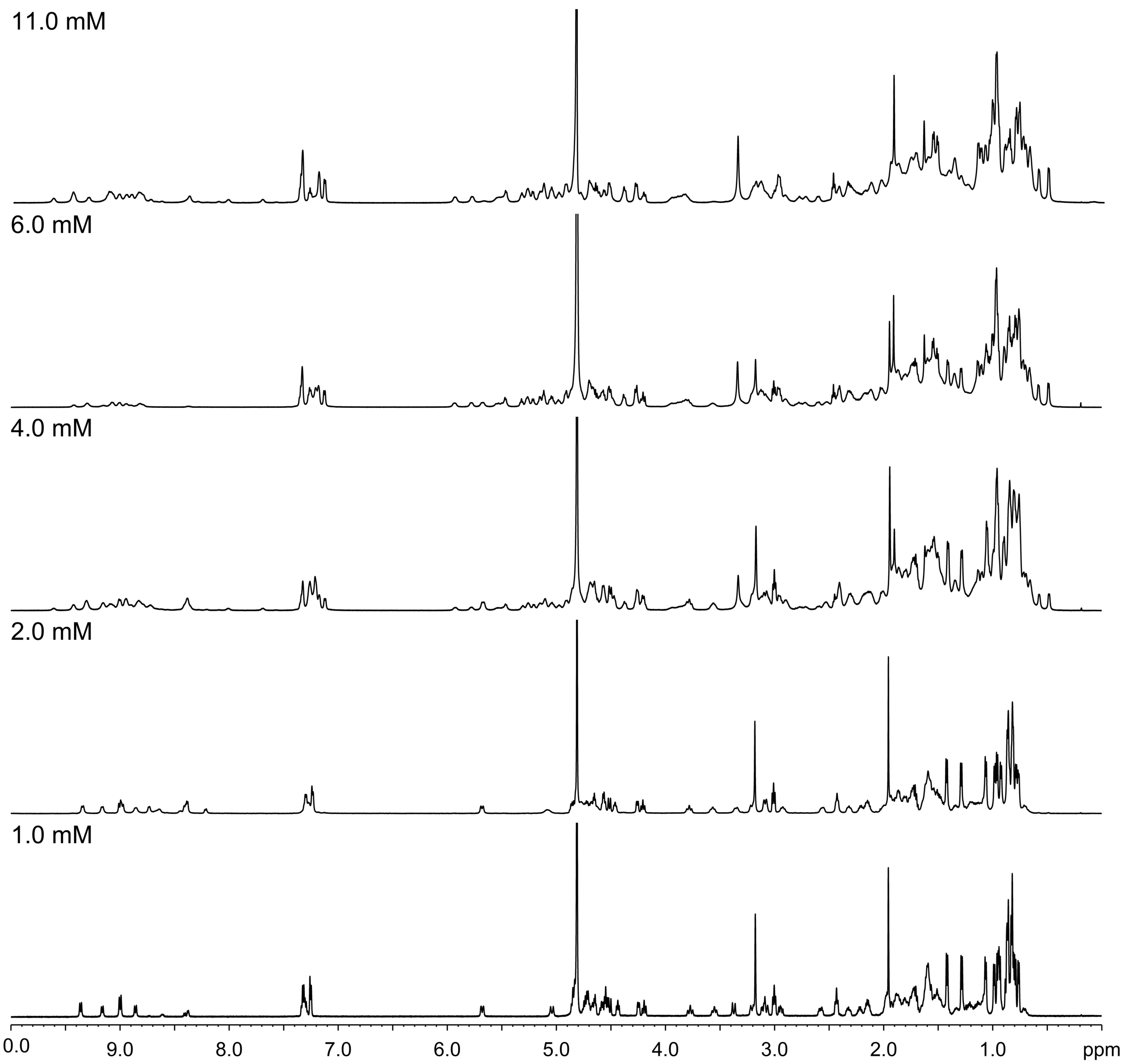


Figure S5. [1]H NMR stacked plot of peptide **2** between 1.0 and 11.0 mM in D_2O at 600 MHz and 298 K.

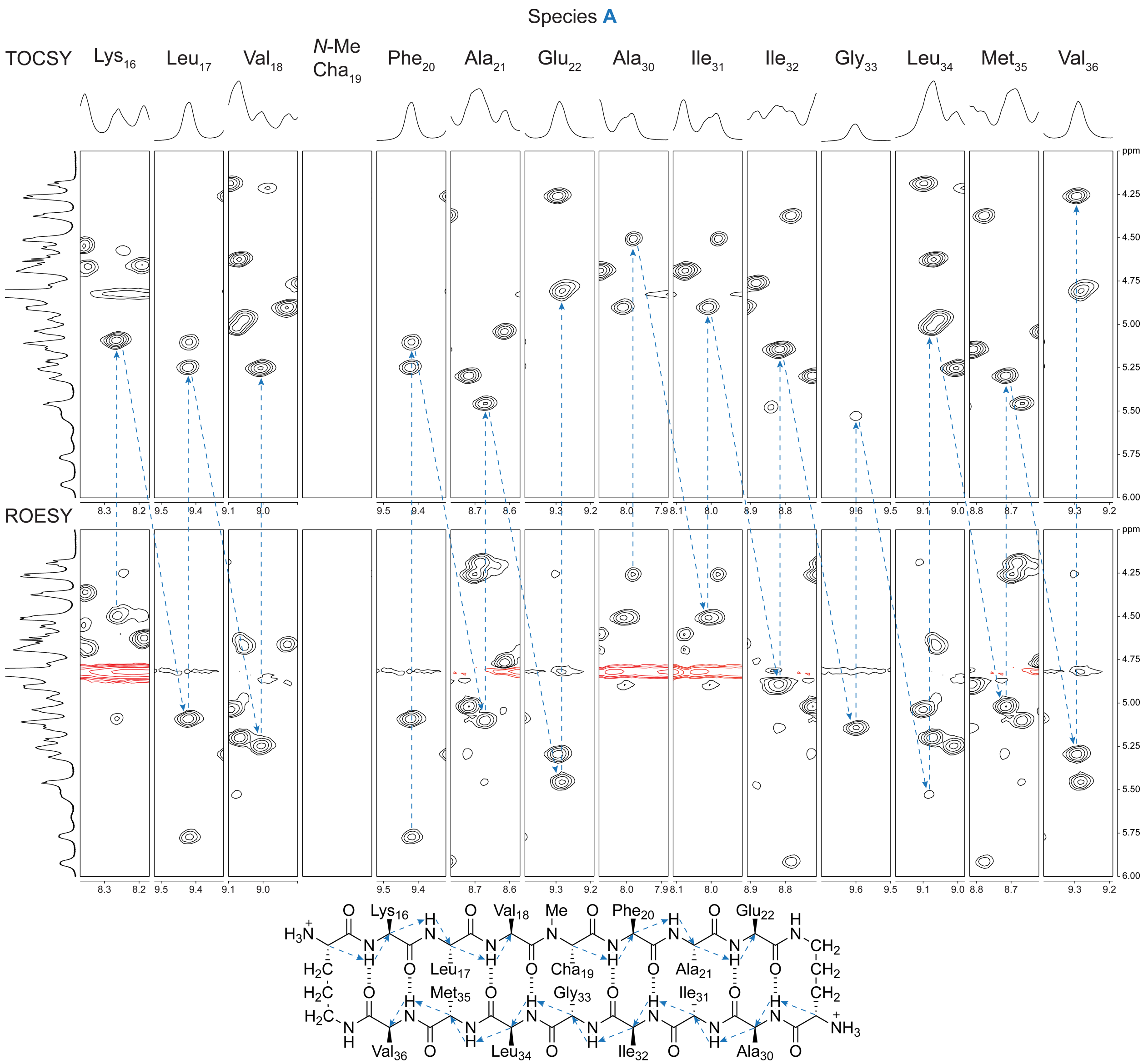


Figure S6. Strips from TOCSY and ROESY spectra showing peak assignments from species **A**. Spectra were recorded at 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K.

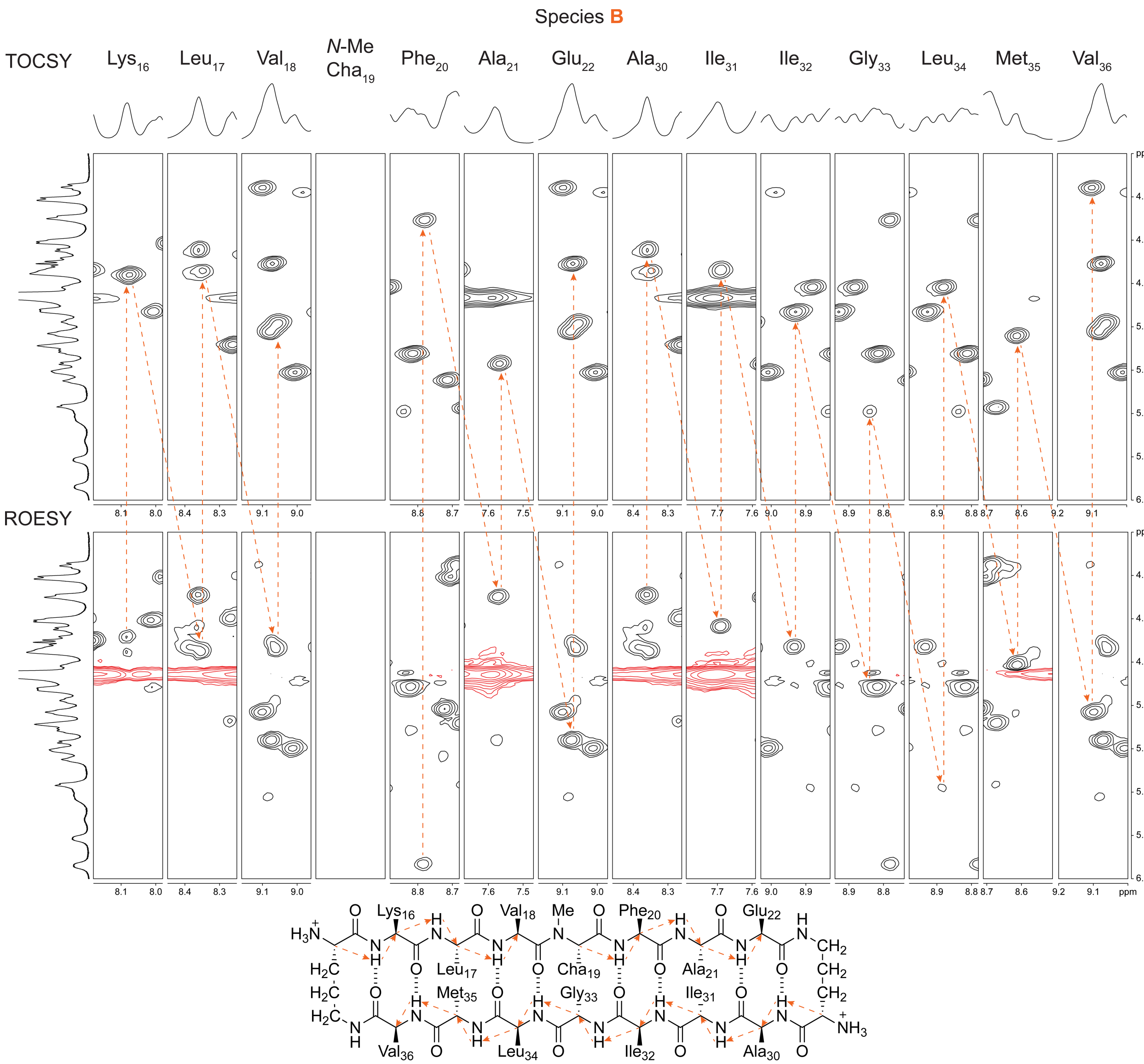


Figure S7. Strips from TOCSY and ROESY spectra showing peak assignments from species **B**. Spectra were recorded at 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K.

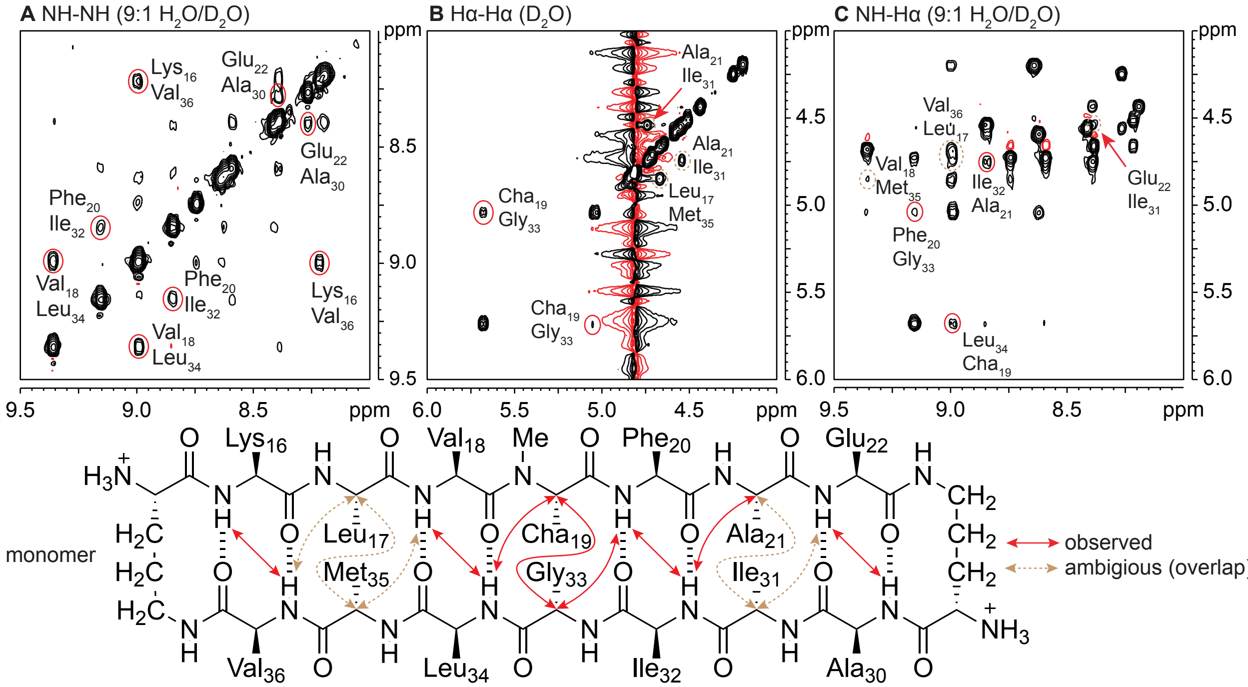


Figure S8. NOESY spectrum of peptide **2** showing key intramolecular NOEs associated with β-sheet folding for monomer. Spectra were recorded at 1.0 mM at 600 MHz and 298 K.

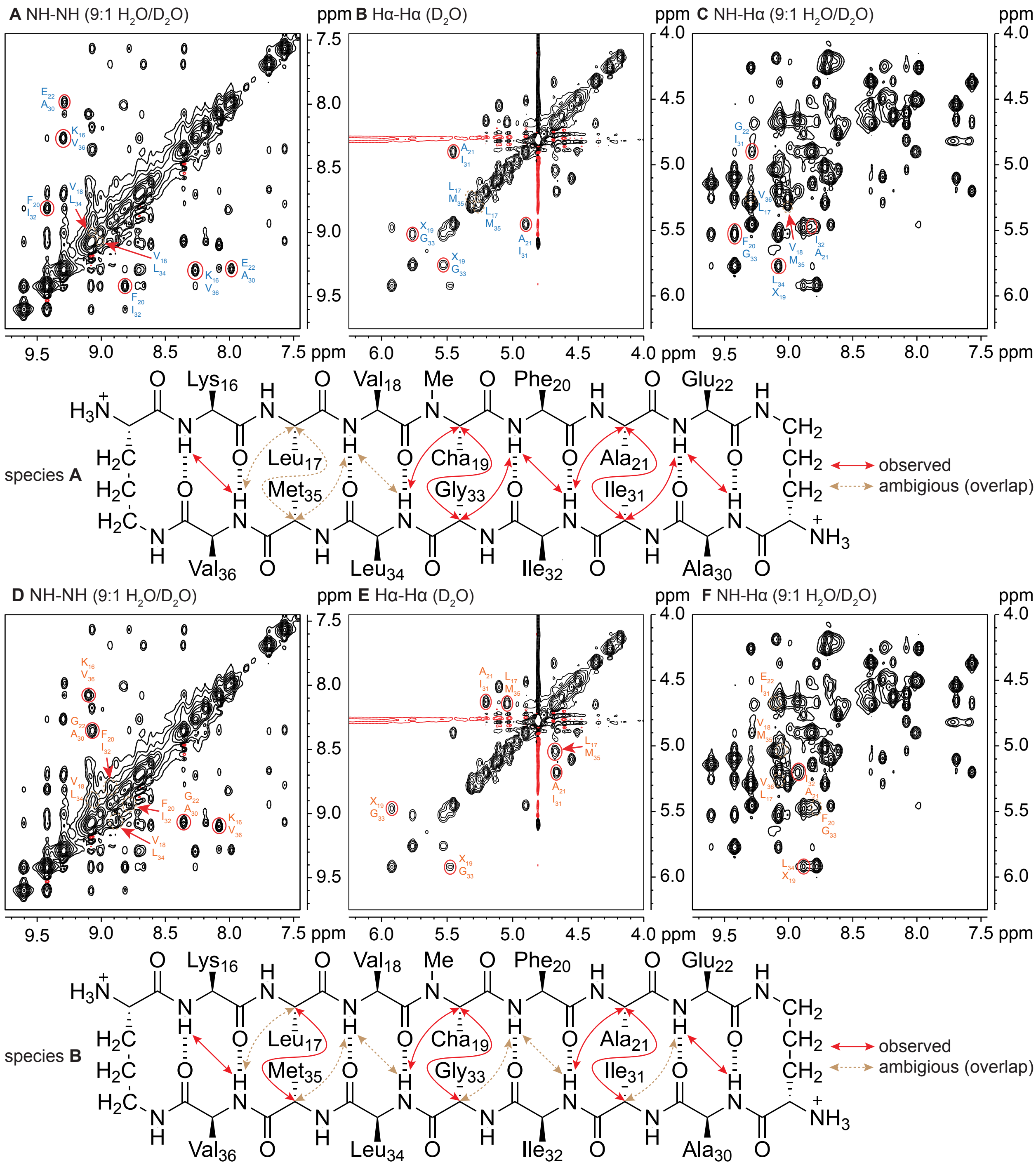


Figure S9. NOESY spectrum of peptide **2** showing key intramolecular NOEs associated with β-sheet folding for species **A** and **B**. Spectra were recorded at 11.0 mM at 600 MHz and 298 K. X_{19} refers to Cha_{19}.

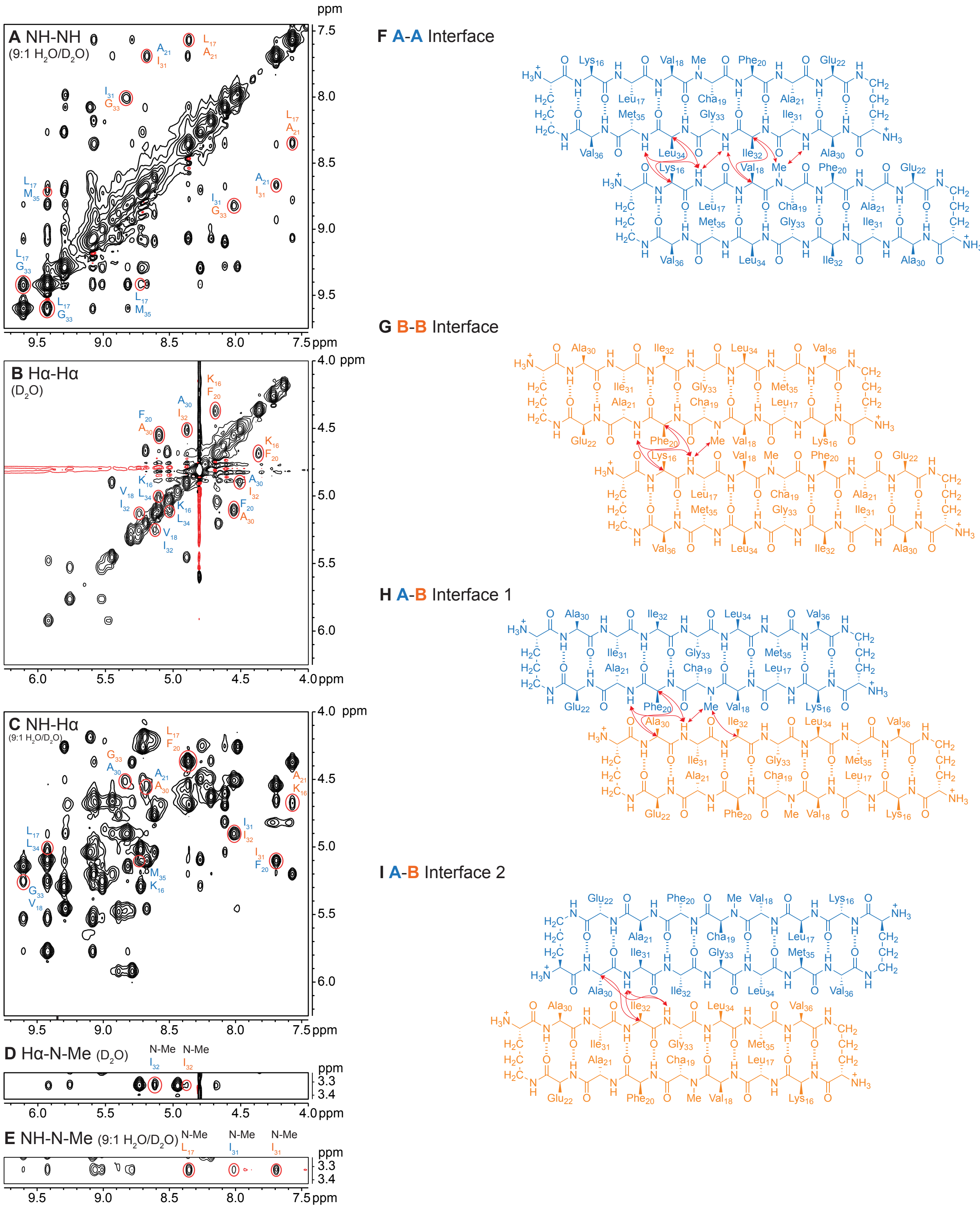


Figure S10. Summary of key peptide **2** intermolecular NOEs. NOESY spectra were recorded at 11.0 mM at 600 MHz and 298 K. Simplified ChemDraw illustrations of the intermolecular contacts are shown as dimers for visualization purposes only.

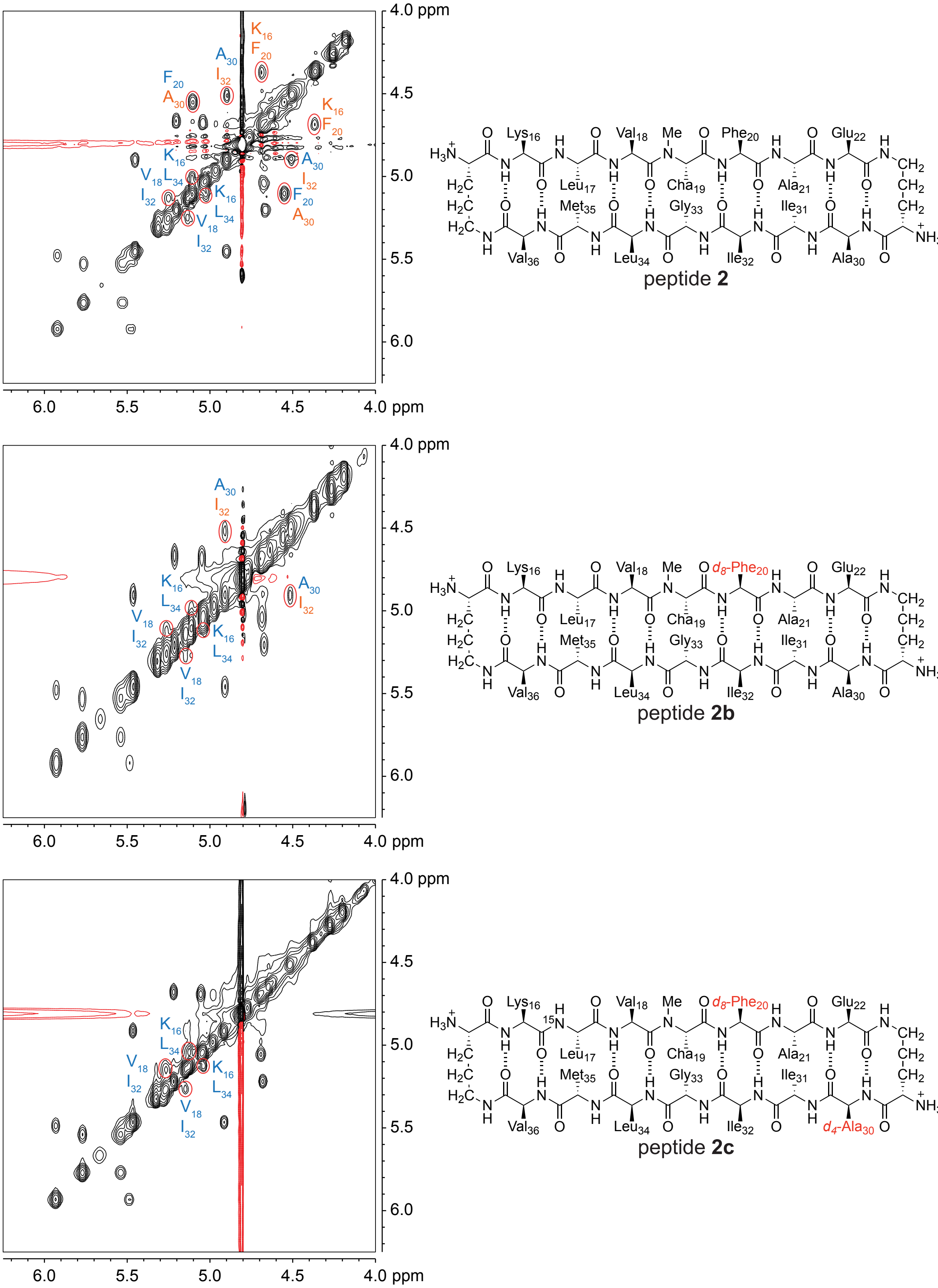


Figure S11. NOESY spectra of peptide **2**, **2b**, and **2c** showing key intermolecular Hα-to-Hα NOEs that disappear with deuteration. Peptide **2** was recorded at 600 MHz. Peptides **2b** and **2c** was recorded at 800 MHz. All spectra were recorded at 11.0 mM in D_2O at 298 K.

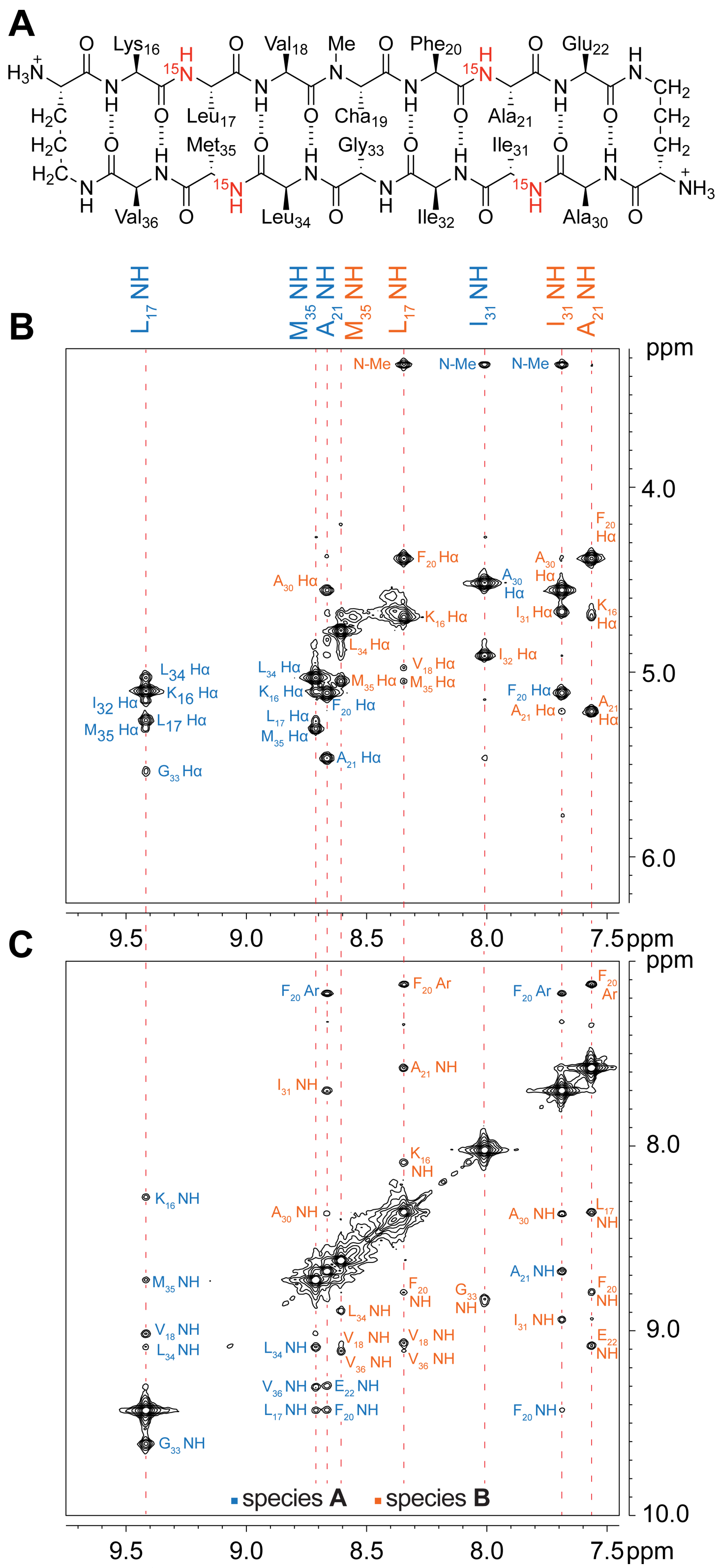
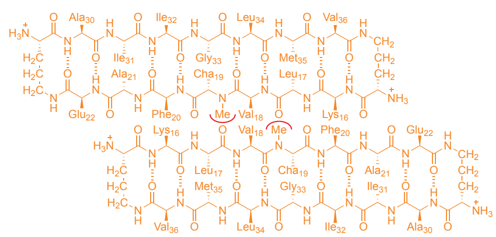


Figure S12. Fully annotated ^{15}N-edited NOESY HSQC spectrum of peptide **2d**. Chemical structure of peptide **2d**, highlighting the ^{15}N labels at Leu_{17}, Ala_{21}, Ile_{31}, and Met_{35} in red (A). ^{15}N-edited NOESY-HSQC spectrum of peptide **2d** showing all NOEs to either Hα or *N*-Me (B) and NH protons (C). The spectrum was recorded at 11.0 mM in 9:1 H_2O/D_2O at 800 MHz and 298 K.

In building the model of the asymmetric hexamer, we considered numerous other models that proved to be inconsistent with the NMR data. These oligomer models that are inconsistent with the data can be categorized both by the oligomer state and by key underlying assumptions made. We excluded all odd numbered oligomers because of the 1:1 integration of resonances associated with associated with species **A** and **B**. Additionally, the presence of multiple A-B intermolecular contacts indicates there is one oligomer species composed of species **A** and **B**.

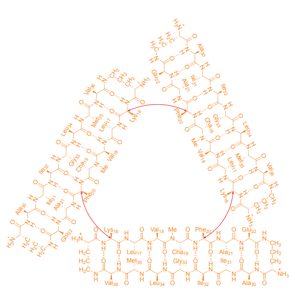
The quotient of diffusion coefficients of peptide **2** shown in Figure **S14** provides a rough estimate that peptide **2** is between a decamer and a tetradecamer. We considered all cases between a tetramer and hexadecamer to account for the possibility that the oligomer may be anisotropic (not perfectly spherical).

Tetramer: Species **B** forms a strong locus of intermolecular contacts between Lys_{16} Hα and Phe_{20} Hα. Assuming a tetramer, species **B** must make a dimer which is forbidden because of the two *N*-methyl groups on Cha_{19} would disrupt hydrogen bonding along the dimer interface.

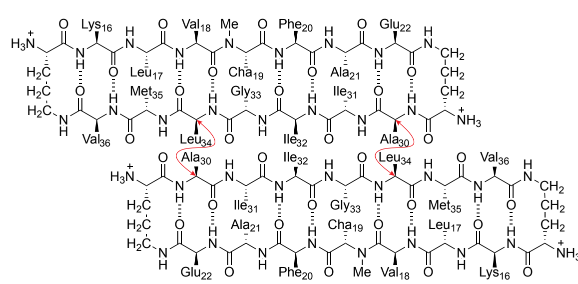


N-methyl groups disrupt dimer interface

Hexamer: Based on molecularity, there are three molecules of species **A** and **B**. The only way to satisfy the Lys_{16} Hα and Phe_{20} Hα for species **B** is to make triangular trimers analogous to that observed in the crystal structure (shown below on the left). We ruled out the crystallographic hexamer because we do not observe any NOE between Ala_{30} Hα and Leu_{34} Hα for either **A-A**, **A-B**, or **B-B** (shown below on the right) that would be expected based on the crystallographic hexamer. We could not conceptualize any other hexameric oligomer models outside of the proposed asymmetric hexamer.

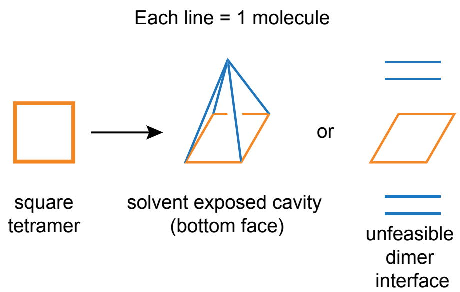


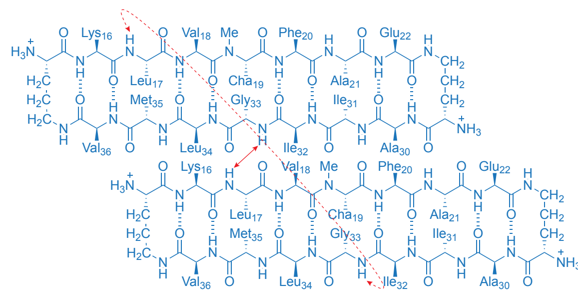
observed in solution-phase NMR



not observed in solution-phase NMR

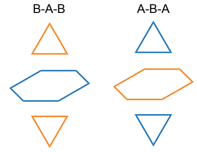
Octamer: Based on molecularity, there are four molecules of species **A** and four molecules of species **B**. We can exclude the possibility that species **B** makes two sets of dimers because the N-methyl groups on Cha_{19} would disrupt hydrogen bonding same as for the tetrameric models (shown above) of the oligomer. This leaves the possibility that species **B** makes a square tetramer where the Lys_{16} Hα and Phe_{20} Hα is located on the interior vertices of a square. There are two possible arrangements of the remaining four molecules of **A**: all four molecules of **A** above the plane of the square or two molecules of **A** below and two molecules of **B** above. In the first case, the model contains a solvent exposed cavity that is unlikely to assemble in aqueous solution. In the second case, there is no way to satisfy the observed **A-A** intermolecular contacts with an arrangement of dimers either above or below the plane of the square tetramer formed by species **B**.





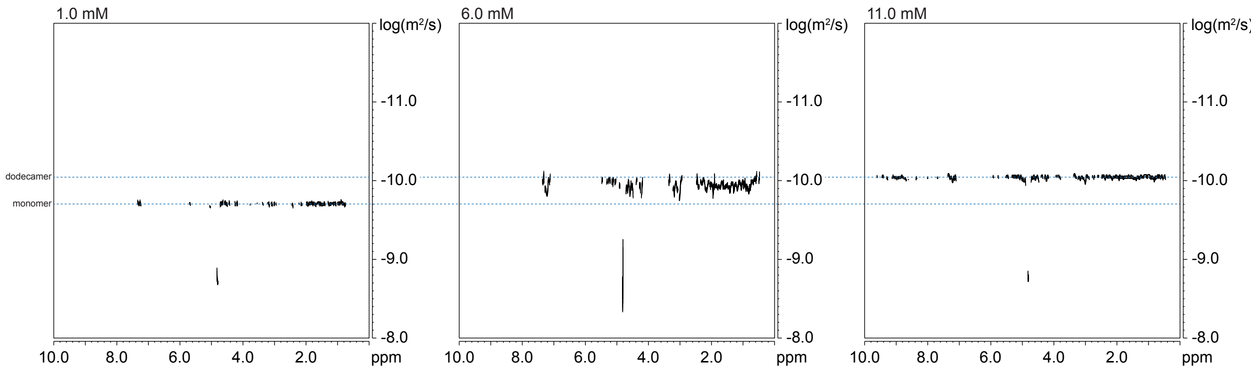
Decamer and Tetradecamer: By analogy described for the octamer oligomer model, we can make a pentamer or heptamer for species **B** using the Lys$_{16}$ Hα and Phe$_{20}$ Hα NOE. However, we run into the same issues encountered in the octamer oligomer model. The odd number of molecules of species **A** means that all five molecules of species **A** would have to pack on one face of the pentamer or heptamer. The resulting model runs into the same issues as the octamer with a solvent exposed cavity on one face.

Dodecamer: There are two main possibilities with a dodecamer model, species **B** can either make two triangular trimers as described in the asymmetric hexamer model or a hexamer based on the Lys$_{16}$ Hα and Phe$_{20}$ Hα NOEs. In the case where species **B** makes two triangular trimers, species A would have to hexameric structure that is sandwiched between the two species **B** triangular trimers (shown as B-A-B). We did not find a reasonable conformation for the species **A** monomer that simultaneously satisfied the **A-A** intermolecular NOEs (Leu$_{17}$ NH to Gly$_{33}$ NH) and two unique **A-B** intermolecular NOEs (Ala$_{30}$ Hα to Ile$_{32}$ Hα **B-A** and Ile$_{32}$ Hα to Phe$_{20}$ Hα **B-A**) within 5 Å. In the case where species **B** makes a hexamer, we considered placement of six molecules of species **A** on one face as well as a symmetrical placement of three molecules of **A** on both faces. The placement of six molecules of species **A** on the same face contains an exposed cavity. We did not find a reasonable arrangement of the three molecules of **A** on both faces of a hexamer formed by species **B** that simultaneously satisfied the **A-A** and two **A-B** intermolecular interfaces.



Hexadecamer: Given that species **B** cannot make dimers as described in the tetramer model, we considered whether species **B** can make two sets of square tetramers or one set of octamers. The hexadecamer models run into the same issues described in the tetramer and octamer model.

Figure S13. Consideration and elimination of alternative models of the asymmetric hexamer.



Calculations for monomer and dodecamer

For monomer at 1.0 mM, log $D_{monomer}$ = -9.703, $D_{monomer} = 10^{-9.703} = 1.98 \times 10^{-10}$ m^2/s.

For dodecamer at 11.0 mM, log $D_{dodecamer}$ = -10.044, $D_{dodecamer} = 10^{-10.044} = 0.904 \times 10^{-10}$ m^2/s.

To assess oligomer state,

Oligomer	Ratio
dimer	0.794
trimer	0.693
hexamer	0.550
decamer	0.464
undecamer	0.450
dodecamer	0.437
tridecamer	0.425
tetradecamer	0.415

Figure S14. DOSY NMR spectra of 1.0, 6.0, and 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K. The 11.0 mM spectrum shows one track with a diffusion coefficient of $0.904 \pm 0.08 \times 10^{-10}$ m^2/s. The 1.0 mM spectrum shows one track with a diffusion coefficient of $1.98 \pm 0.11 \times 10^{-10}$ m^2/s. The HOD peak is calibrated to 19.0×10^{-10} m^2/s.[1]

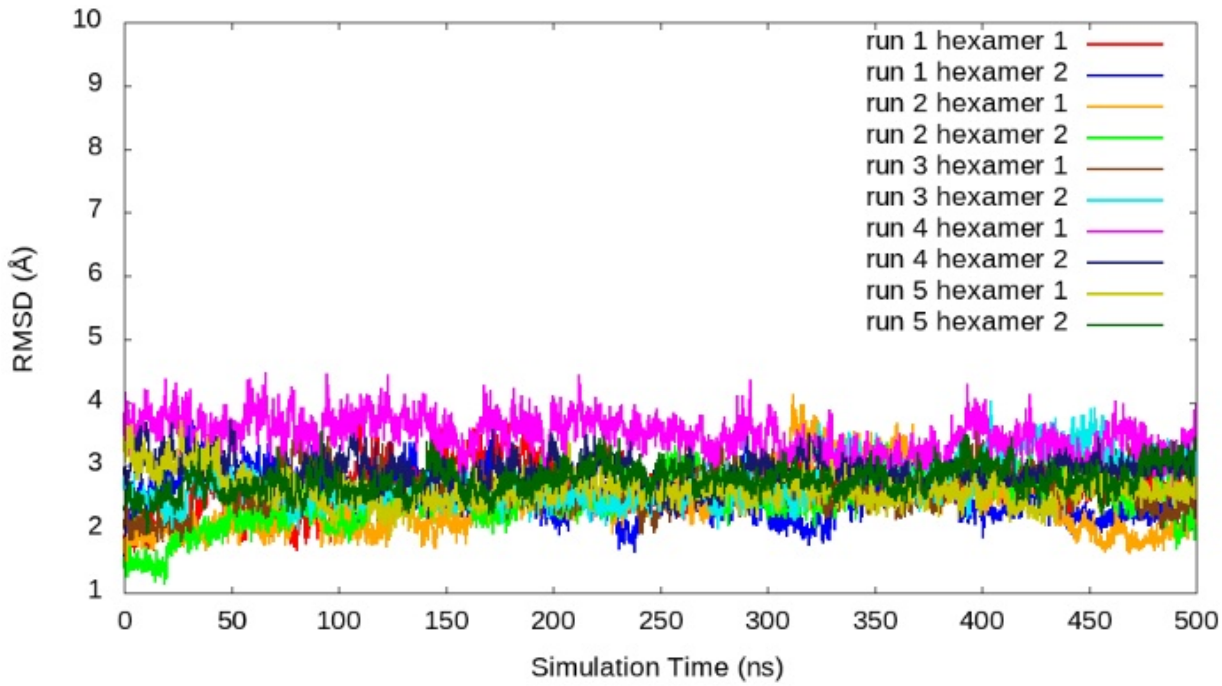


Figure S15. Root mean square displacement (RMSD) of hexamers backbone atoms with respect to the NMR-based molecular model.

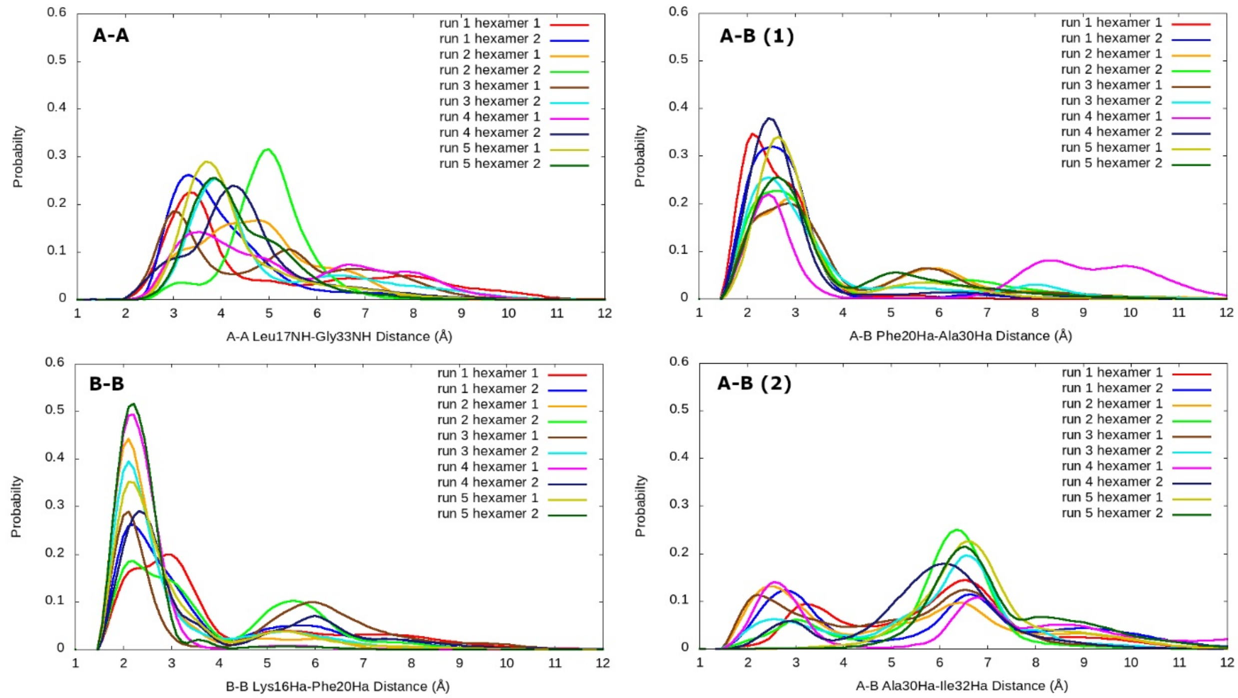


Figure S16. Probability distributions of key NOE distances observed during MD simulations. Each histogram is calculated from three equivalent pairs of proton-proton distances in one hexamer and among 50,000 frames (500 ns) per pair.

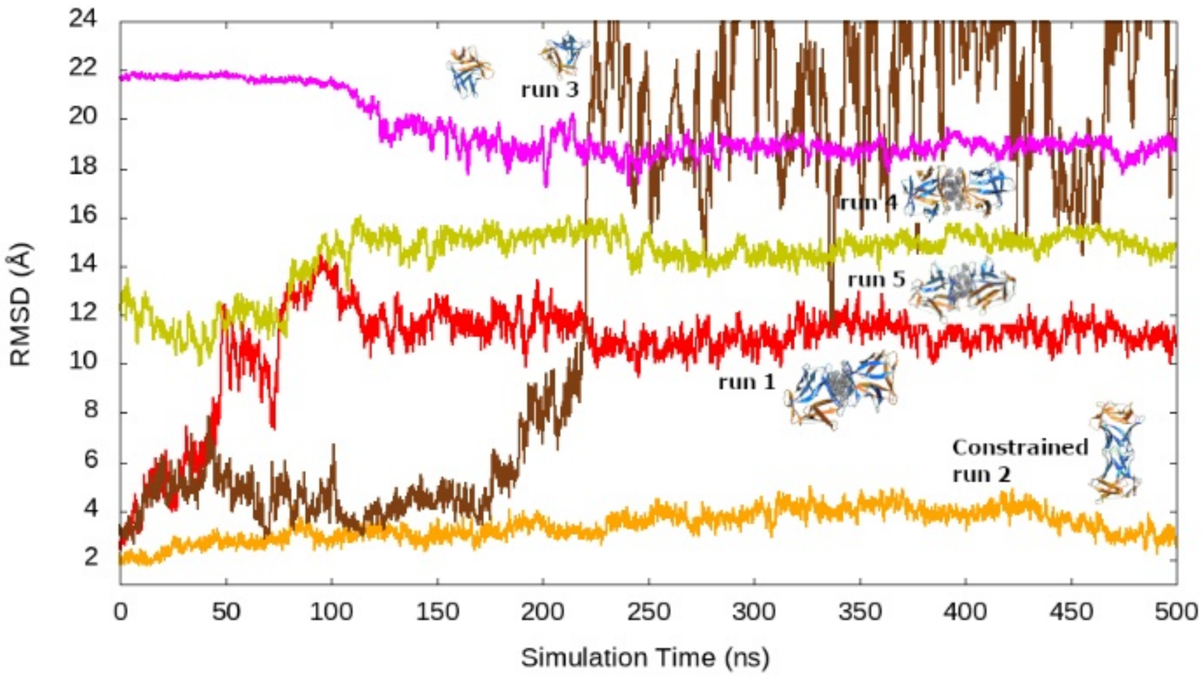
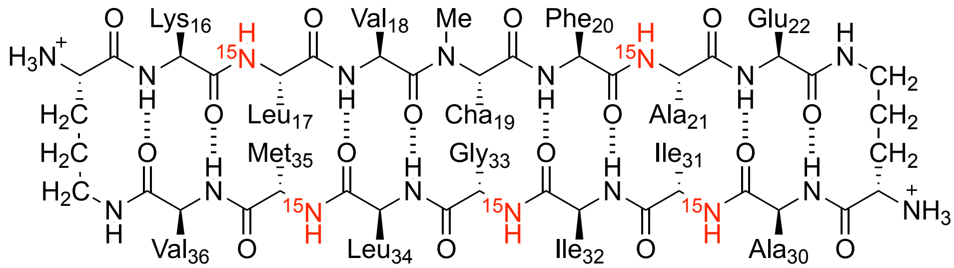


Figure S17. RMSD of dodecamer backbone atoms with respect to initial structure of run 1.

Table S1. Crystallographic properties, crystallization conditions, and data collection and model refinement statistics for peptide **2**.

PDB ID	9EEC
space group	*P*432
a, *b*, *c* (Å)	67.511 67.511 67.511
α, β, λ (°)	90, 90, 90
molecules per asymmetric unit	3
wavelength (Å)	1.0
resolution (Å)	38.98–1.58 (1.64–1.58)
total reflections	744201 (3145)
unique reflections	13661 (287)
multiplicity	62.6 (11.0)
completeness (%)	99.95 (99.09)
mean I/σ(I)	33.09 (0.06)
Wilson B factor	32.25
R_{merge}	0.05796 (15.74)
$R_{measure}$	0.05846 (16.44)
$CC_{1/2}$	1 (0.0818)
CC^*	1 (0.389)
R_{work}	0.2110 (0.3321)
R_{free}	0.2250 (0.3454)
number of non-hydrogen atoms	390
RMS_{bonds}	0.012
RMS_{angles}	1.41
Ramachandran favored (%)	100
Ramachandran allowed (%)	0
Ramachandran outliers (%)	0
rotamer outliers (%)	0
clashscore	13.29
average B-factor	40.47
number of TLS groups	3
ligands/ions	0
water molecules	15
crystallization conditions	0.1 M HEPES at pH 7.0, 0.25 M $MgCl_2$, 30% isopropanol

Table S2. Chemical shift tabulations for ^{15}N-labeled isotopologues **2a** and **2d**. Data taken from $^{1}H,^{15}N$ HSQC recorded at 1.0 mM or 11.0 mM peptide in 9:1 H_2O/D_2O at 800 MHz and 298 K.



Residue	^{15}N Chemical Shift (ppm)		
	monomer	species **A**	species **B**
Leu_{17}	123.3	125.2	123.3
Ala_{21}	127.0	121.8	125.8
Ile_{31}	121.8	121.7	120.5
Gly_{33}	111.4	109.9	104.7
Met_{35}	123.9	123.8	121.7

Table S3. Chemical shifts of NH and Hα protons of peptide **2** monomer, species **A**, and species **B** in D_2O or 9:1 H_2O/D_2O at 600 MHz and 298 K.

Residue	Chemical Shift (ppm)					
	monomer		species **A**		species **B**	
	NH	Hα	NH	Hα	NH	Hα
Lys_{16}	8.21	4.64	8.27	5.11	8.09	4.70
Leu_{17}	8.38	4.67	9.43	5.26	8.36	4.67
Val_{18}	9.35	4.83	9.01	5.26	9.06	4.97
N-Me Cha_{19}	-	5.68	-	5.77	-	5.92
Phe_{20}	9.15	4.71	9.42	5.11	8.79	4.37
Ala_{21}	8.59	4.74	8.67	5.46	7.57	5.21
Glu_{22}	8.39	4.44	9.29	4.82	9.08	4.63
Ala_{30}	8.26	4.55	7.99	4.51	8.37	4.55
Ile_{31}	8.41	4.54	8.02	4.90	7.70	4.66
Ile_{32}	8.84	4.59	8.82	5.14	8.94	4.90
Gly_{33}	8.62	5.04	9.61	5.53	8.85	5.48
Leu_{34}	8.98	4.72	9.08	5.04	8.89	4.77
Met_{35}	8.74	4.86	8.72	5.31	8.62	5.04
Val_{36}	8.99	4.19	9.30	4.26	9.10	4.18

Materials and Methods[2]

General information

Chemicals and Supplies All chemicals were used as received unless otherwise noted. Dry methylene chloride (CH_2Cl_2), *N,N*-dimethylformamide (DMF), and diethyl ether were obtained by passing through alumina under argon prior to use. The DMF was additionally passed through an amine scavenger resin column under argon. Amine-free *N,N*-dimethylformamide (DMF) and piperidine were purchased from Alfa Aesar. 2,4,6-collidine and diisopropylethylamine (DIPEA) was purchased from Thermo Scientific. 1,1,1,3,3,3-Hexafluoro-2-propanol (HFIP) and trifluoroacetic acid (TFA) were purchased from Oakwood Chemicals. Deionized water (18 MΩ) was obtained from a Barnstead NANOpure Diamond water purification system. HPLC grade acetonitrile and deionized water (18 MΩ), each spiked with 0.1% (v/v) trifluoroacetic acid (TFA), were used for analytical and preparative reverse-phase HPLC. Isotopically labeled alanine (^{15}N, 98% isotopic purity), glycine (^{15}N, 98% isotopic purity), isoleucine (^{15}N, 98% isotopic purity), leucine (^{15}N, 98% isotopic purity), methionine (^{15}N, 96-98% isotopic purity), alanine (d_4-Phe, 98% isotopic purity), and phenylalanine (d_8-Phe, 98% isotopic purity) were purchased from Cambridge *N*-(9-flurenylmethoxycarbonyloxy)succinimide (Fmoc-Osu) was purchased from GL Biochem. Isotope Laboratories. Deuterium oxide (D, 99.9%) was purchased from Sigma Aldrich.

Instrumentation All peptides were first purified on a Biotage® Isolera™ One system equipped with a Biotage® Sfr Bio C18 Duo 300 Å 20 µm column prior to purification on the Shimazu instrument equipped with an Agilent Zorbax 7 µm 300SB-C18 column (21.2 x 250 mm). Analytical reverse-phase HPLC was performed on an Agilent 1260 Infinity II instrument with a Phenomonex bioZen PEPTIDE 2.6 µm XB-C18 column. Yields refer to isolated purified compounds confirmed chromatographically. UV absorption was measured using the Thermo Scientific™ Varioskan™ LUX. Matrix-assisted desorption ionization time-of-flight (MALDI-TOF) mass spectrometry was performed on the AB SCIEX TOF/TOF 5800 system in linear reflector mode with α-cyano-4-hydroxycinnamic acid (CHCA) used as the matrix. All peptides were assumed to have one trifluoroacetate ion per ammonium group present in each peptide.

Synthesis and purification of peptides

Peptide synthesis procedure Macrocyclic peptides **1**, **2**, and subsequent homologues were synthesized by manual solid-phase peptide synthesis on 2-chlorotrityl resin, followed by HFIP cleavage, solution-phase cyclization, global deprotection, and reverse phase HPLC purification. A step-by-step procedure is detailed below.

Resin loading 2-chlorotrityl resin (300 mg, 1.07 mmol/g) and Boc-Orn(Fmoc)-OH (100 mg) was added to a 10-mL Bio-Rad Poly-Prep chromatography column. The resin was loaded overnight (8-12 h) in a solution of 6 mL DCM and 0.3 mL 2,4,6-collidine under gentle agitation. The loading solution was drained and the resin was washed three times with DCM. After washing, the resin was capped using a solution of DCM/MeOH/DIPEA (17:2:1, 8 mL) for 1 h under gentle agitation. The solution was drained and the resin was washed three times with DCM. The resin was dried under a flow of nitrogen. Once dry, loading was checked on the Varioskan through UV analysis (290 nm) of the Fmoc cleavage product. Nominal loading values range from 0.12 – 0.15 mmol.

Solid-phase peptide coupling The loaded was re-suspended in amine-free DMF, allowed to swell for 30 min, and transferred to a clean 25 mL coupling vessel. The linear peptide was synthesized from the *C*-terminus to the *N*-terminus using standard coupling cycles unless otherwise noted. A standard coupling cycle consists of the following: (1) Fmoc deprotection with 20% (v/v) piperidine in DMF (5 mL) for 10 min, (2) resin washing with DMF (5X), (3) coupling of the amino acid (4 equiv) with HCTU (4 equiv) in 20% (v/v) collidine in DMF for 30 min, and (4) resin washing with DMF (5X). For the amino acid after the *N*-methylated amino acid, the resin was double coupled for 1 h each using 4 equiv of HATU/HOAt. Once the linear synthesis was complete, the resin was transferred to a 10-mL Bio-Rad Poly-Prep chromatography column and washed with DCM (3X).

Cleavage of the linear peptide from resin The linear peptide was cleaved from using a cleavage solution of 20% (v/v) HFIP in DCM (7.5 mL) under gentle agitation. The filtrate was collected in a 1-L round-bottom flask. Another 7.5 mL of cleavage solution was added to the resin under gentle agitation for 30 min and combined with the original filtrate. The combined filtrates were concentrated under reduced pressure to afford a translucent film.

Cyclization of the linear peptide (125 mL) was added to dissolve the clear oil. HATU (6 equiv) and HOAt (6 equiv) were added to the solution. The solution was stirred before DIPEA (300 µL) was added dropwise. The flask was flushed three times with nitrogen and allowed to cyclize at room temperature for 2 d. The reaction was concentrated under reduced pressure (65 °C) to afford a beige solid. The round bottom was placed under vacuum to remove residual DMF.

Global deprotection of the cyclic peptide The protected peptide was dissolved in a mixture of TFA/TIPS/H_2O (90:5:5, 10 mL) with NH_4I (250 mg) and Me_2S (ca. 1 mL). The mixture was stirred for 1 h and transferred into two 50-mL conical tubes each containing 35 mL of chilled diethyl ether. Both conical tubes were centrifuged (600xg, 4 °C) to pellet the precipitate. The supernatant was decanted into a 125-mL Erlenmeyer flask. This process was repeated twice more by adding additional ether, shaking to break up the pellet, and centrifuging. The pellet was dried under reduced pressure.

Biotage Reverse-phase HPLC purification The dried peptide was dissolved in 20% (v/v) ACN in H_2O and purified on the Biotage® Isolera™ One system equipped with a Biotage® Sfr Bio C18 Duo 300 Å 20 µm column using an ACN/H_2O gradient (20 – 60%) over 8 column volumes. Fractions were analyzed by MALDI-TOF and analytical HPLC. Fractions containing predominantly desired peptide were combined and concentrated under reduced pressure to afford an off-white solid.

Shimadzu Reverse-phase HPLC purification The peptide was dissolved in 20% (v/v) ACN in H_2O and filtered through a 0.2 µm PVDF syringe filter to remove insoluble aggregates and purified by RP-HPLC using an ACN/H_2O gradient (25–45%, 15 mL/min) over 90 min. Fractions were analyzed by MALDI-TOF and analytical HPLC. Pure fractions were and lyophilized to afford peptide as white trifluoroacetate (TFA) salts.

Isotopologues **2a-d** were synthesized the same as peptides **1** and **2**. Fmoc-protected ^{15}N glycine replaced Gly_{33} in the preparation of **2a**. Fmoc-protected d_8 phenylalanine replaced Phe_{20} in preparation of **2b**. Fmoc-protected d_8 phenylalanine and d_4 alanine replaced Phe_{20} and Ala_{30} in preparation of **2c**. Fmoc-protected ^{15}N alanine, ^{15}N isoleucine, ^{15}N leucine, and ^{15}N methionine replaced Ala_{21}, Ile_{31}, Leu_{17}, and Met_{35} in preparation of **2d**.

X-ray crystallography of peptide 2

*Crystallization procedure of peptide **2*** Crystal screening trays for peptide **2** were prepared based on previously reported crystallization conditions for peptide 1 in a 4x6 matrix Hamptom VDX 24-well plate. Two optimization trays conditions were tested. In both trays, the HEPES buffer pH was varied in each row in increments of 0.5 pH units (6.5, 7.0, 7.5, and 8.0) and the isopropanol concentration in each column in increments of 2% (28%, 30%, 32%, 34%, 36%, 38%). The first well of the first tray was prepared by combining 100 µL of 1 M HEPES at pH 6.5, 250 µL of 1 M $MgCl_2$, 280 µL of isopropanol, and 370 µL of (18 MΩ water). The other wells were prepared in analogous fashion, by combining 100 µL of HEPES buffer of varying pH, isopropanol in varying amounts, $MgCl_2$, and 18 MΩ water for a total volume of 1 mL per well. The first well of the second tray was prepared by combining 100 µL of 1 M HEPES at pH 6.5, 200 µL of 1 M sodium citrate, 280 µL of isopropanol, and 420 µL of (18 MΩ water). The other wells for the second tray were prepared in analogous fashion.

Three hanging-drops were prepared per borosilicate glass slide by combining of peptide 2 (10 mg/mL in 18 MΩ water) and well solution in the following amounts: 1 µL: 1 µL, 2 µL: 1 µL, 1 µL: 2 µL of peptide solution to well solution. Slides were inverted and sealed by pressing firmly against the silicone grease surrounding each well.

Crystals of peptide **2** suitable for X-ray diffraction grew in ~3-5d. Crystals were harvested with a nylon loop attached to a copper or steel pin, cryoprotected using MPD and flash frozen in liquid nitrogen prior to data collection.

X-ray crystallographic data collection, data processing, and structure determination of peptide ***2***
Diffraction data for peptide **2** were collected on the Advanced Light Source Synchrotron at the Berkeley Center for Structural Biology on beamline 5.0.2. at 1.0 Å wavelength with 0.5° oscillation. Diffraction data were collected using b4. Diffraction data were integrated, scaled, and merged using XDS and pointless and aimless. The resolution was limited to the $CC_{1/2}$ resolution of 1.58 Å. The electron density map for peptide **2** was generated by molecular replacement with the coordinates from the X-ray crystallographic structure of peptide **1** (PDB 5W4H) as a search model using Phaser in the Phenix software suite 1.10.1. Molecular manipulations of the model were performed with Coot. Coordinates were refined in the *P*432 space group with phenix.refine. The optimal number and composition of the TLS groups were chosen automatically by phenix.refine.

Silver-stained SDS-PAGE

Pouring gels The reagents for Tricine SDS-PAGE were prepared according to recipes and procedures detailed in Schägger, N. *Nat. Protoc.* 2006, *1*, 16-22.[3] Fresh resolving separation gel (16%) were prepared by mixing the following: Acrylamide bis-acrylamide (AB-3) (5 mL), Gel Buffer (5 mL), H_2O, and 10% (w/v) ammonium persulfate solution (100 μL) and tetramethylethylenediamine TEMED (10 μL). The solution was poured into a cast and allowed to polymerize over 30 min. The resolving gel was topped with isopropanol (IPA) during the polymerization process. Once the resolving gel has set, the isopropanol was discarded and the resolving gel was rinsed with water. The stacking gel (4%) was prepared in an analogous fashion and poured on top of the resolving gel. The comb was inserted, and the gel was allowed to set for 15 min.

Loading and running Concentrated SDS-loading dye (6X) was mixed with peptide to give the working concentrations listed and loaded onto the gel lanes (12 µL per lane). The migration of peptides was compared with molecular weight protein ladder (Spectra^{TM} Multicolor Low Range Protein Ladder, ThermoFisher Scientific, catalog #: 26628). The gels were run at a constant 90 V at room temperature.

Silver-staining The reagents for silver staining were prepared according to procedures detailed in Simpson, R. J. *CHS Protoc*. **2007**.[4] [For best results, we prepared the sodium thiosulfate, silver nitrate, and developing solution fresh each time. The silver nitrate solution was chilled at 4 °C prior to use. Pure sodium carbonate (≥ 99.5 %) was used to prepare the developing solution.] The gel was rocked in 20 mL of fixing solution (MeOH/AcOH/H_2O, 50:5:45). Next, the fixing solution was discarded and 20 mL of fresh solution (MeOH/H_2O, 1:1) was added to the gel and rocked for 10 min before being discarded. After that, the gel was rocked in RO water for 10 min. The gel was submerged in 0.02 % (w/v) aqueous sodium thiosulfate solution and rinsed for 1 min with RO water three times. Freshly made chilled 0.1 % (w/v) aqueous silver nitrate solution (20 mL) was added to the gel and rocked for 20 min. The solution was discarded and the gel was rinsed again with RO water for 1 min three times. The gel was developed using 2% (w/v) sodium carbonate solution with 0.04% (w/v) formaldehyde. The gel then was submerged in 5% (v/v) aqueous acetic acid. The silver-stained gels were visualized with a ChemiDocTM MP Imaging System using the "Optimal Auto-exposure" exposure under the "Silver Stain Gel" application of the Image Lab Touch Software.

Circular dichroism (CD) spectroscopy

A 100 μM solution of each peptide was prepared by dissolving weighed portion of peptide in the appropriate volume of 10 mM sodium phosphate buffer at pH 7.4. Each solution was transferred to a 1 mM quartz cuvette for data acquisition. Circular dichroism spectra were acquired on a Jasco-810 circular dichroism spectropolarimeter at ambient temperature. Data was collected with a 0.5 nm pitch, 1 nm bandwidth, from 260 nm to 190 nm averaged over 5 accumulations with 11-point smoothing.

Fmoc-Protection of Isotopically Labeled Amino Acids

Fmoc-protected d_4-Ala-OH and Fmoc-protected d_8-Phe-OH were prepared as previously reported.[2]

Fmoc-Protection of ^{15}N-Labeled Amino Acids

Fmoc [^{15}N] Ala-OH: A 100-mL one-neck round-bottom flask equipped with a magnetic stirring bar was charged with ^{15}N-labeled alanine (0.5 g, 5.6 mmol), and Fmoc-OSu (1.80 g, 5.3 mmol). A solution of 1:1 CH_3CN/H_2O (25 mL) and Et_3N (782 μL, 5.6 mmol)) were added then the reaction mixture was stirred for 15 min. Additional Et_3N was added until a clear solution formed (pH 7.2). The solution was stirred for an additional 1 h. The solution was poured into a solution of 1.0 M HCl (125 mL) in a 250 mL beaker while stirring vigorously. The Fmoc-[^{15}N]Ala-OH precipitated from the solution and the solid was isolated by filtering the mixture through a 7 cm Büchner funnel equipped with a piece of filter paper. The solid was dried by aspirating air through the funnel, suspended in EtOAc (ca. 100 mL) to form a clear solution, dried over Na_2SO_4, filtered, and then concentrated by rotary evaporation. The isolated solid was dried overnight under vacuum and ground up to give ca. 1.64 g (94%) of white powder.

^{1}H NMR (600 MHz, DMSO-d$_6$): δ 12.53 (br s, 1H), 7.89 (d, J = 7.5 Hz, 2H), 7.72 (d, J = 6.7 Hz, 2H), 7.66 (dd, $^1J_{HN}$ = 93.1, 7.2 Hz, 1H), 7.42 (t, J = 7.4 Hz, 2H), 7.33 (appar t, J = 7.4 Hz, 2H), 4.32 – 4.26 (m, 2H), 4.22 (t, J = 7.0 Hz, 1H), 4.00 (quint, J = 7.2 Hz, 1H), 1.28 (dd, J = 7.4, 1.5 Hz, 3H) ppm.

^{13}C NMR (150.9 MHz, DMSO-d$_6$): δ 174.4, 155.8 (d, $^1J_{CN}$ = 26.2 Hz), 143.86, 143.82, 140.73, 140.72, 127.6, 127.1, 125.3, 120.12, 120.11, 65.6, 49.2 (d, $^1J_{CN}$ = 12.4 Hz), 46.6, 17.1 ppm. [The following pairs of peaks correspond to the diastereotopic carbons of the Fmoc protecting group: 143.86 and 143.82; 140.73 and 140.72; 120.12 and 120.11. There are 2 coincident carbons at 127.6, 127.1, and 125.3 ppm that could not be resolved.]

Fmoc-[^{15}N] Ile-OH: A 50-mL one-neck round-bottom flask equipped with a magnetic stirring bar was charged with ^{15}N-labeled isoleucine (0.25 g, 1.9 mmol), and Fmoc-OSu (0.61 g, 1.8 mmol). A solution of 1:1 CH_3CN/H_2O (12.5 mL) and Et_3N (264 L, 1.9 mmol) were added then the reaction mixture was stirred for 15 min. Additional Et_3N was added until a clear solution formed (pH 7.5). The solution was stirred for an additional 1 h. The solution was poured into a solution of 1.0 M HCl (62.5 mL) in a 100 mL beaker while stirring vigorously. The Fmoc-[^{15}N]Ile-OH precipitated from the solution and the solid was isolated by filtering the mixture through a 7 cm Büchner funnel equipped with a piece of filter paper. The solid was dried by aspirating air through the funnel, suspended in EtOAc (ca. 50 mL) to form a homogenous solution, dried over Na_2SO_4, filtered, concentrated by rotary evaporation, and then stripped twice with DCM. The isolated solid was dried overnight under vacuum and ground up to give ca. 0.64 g (95%) of white powder.

^{1}H NMR (600 MHz, DMSO-d$_6$): δ 12.58 (br s, 1H), 7.89 (d, J = 7.9 Hz, 2H), 7.75 (dd, J = 7.4, 2.6 Hz, 2H), 7.61 (dd, $^1J_{HN}$ = 92.9, 8.6 Hz, 1H), 7.42 (t, J = 7.4 Hz, 2H), 7.36 – 7.28 (m, 2H), 4.36 –

4.16 (m, 3H), 3.92 (ddd, J = 8.3, 6.6, 1.5 Hz, 1H), 1.87 – 1.75 (m, 1H), 1.49 – 1.38 (m, 1H), 1.29 – 1.19 (m, 1H), 0.88 (d, J = 6.9 Hz, 3H), 0.85 (t, J = 7.4 Hz, 3H) ppm.

^{13}C NMR (150.9 MHz, DMSO-d$_6$): 173.3, 156.3 (d, $^{1}J_{CN}$ = 26.5 Hz), 143.9, 143.8, 140.71, 140.69, 127.65, 127.64, 127.07, 127.05, 125.38, 125.37, 120.11, 120.10, 65.7, 58.5 (d, $^{1}J_{CN}$ = 12.3 Hz), 46.6, 36.0, 24.7, 15.6, 11.2 ppm. [The following pairs of peaks correspond to the diastereotopic carbons of the Fmoc protecting group: 143.9 and 143.8; 140.71 and 140.69; 127.65 and 127.64; 127.07 and 127.05; 125.38 and 125.37; 120.11 and 120.10.]

Fmoc-[^{15}N] Leu-OH: A 100-mL one-neck round-bottom flask equipped with a magnetic stirring bar was charged with ^{15}N-labeled leucine (1.0 g, 7.6 mmol), and Fmoc-OSu (2.44 g, 7.2 mmol). A solution of 1:1 CH_3CN/H_2O (50 mL) and Et_3N (1.062 mL, 7.6 mmol)) were added then the reaction mixture was stirred for 15 min. Additional Et_3N was added until a clear solution formed (pH 7.3). The solution was stirred for an additional 1 h. The solution was poured into a solution of 1.0 M HCl (250 mL) in a 400 mL beaker while stirring vigorously. The Fmoc-[^{15}N]Leu-OH precipitated from the solution and the solid was isolated by filtering the mixture through a 7 cm Büchner funnel equipped with a piece of filter paper. The solid was dried by aspirating air through the funnel, suspended in EtOAc (ca. 200 mL) to form a slightly turbid solution, dried over Na_2SO_4, filtered, concentrated by rotary evaporation, and then stripped twice with DCM. The isolated solid was dried overnight under vacuum and ground up to give ca. 2.54 g (94%) of white powder.

^{1}H NMR (600 MHz, DMSO-d$_6$): δ 12.53 (br s, 1H), 7.89 (d, J = 7.6 Hz, 2H), 7.72 (d, J = 7.5 Hz, 2H), 7.63 (dd, $^{1}J_{HN}$ = 92.7, 8.3 Hz, 1H), 7.42 (t, J = 7.4 Hz, 2H), 7.32 (ddd, J = 7.4, 3.5, 1.0 Hz, 2H), 4.37 – 4.25 (m, 2H), 4.22 (t, J = 7.1 Hz, 1H), 4.02 – 3.94 (m, 1H), 1.72 – 1.61 (m, 1H), 1.61 – 1.52 (m, 1H), 1.52 – 1.42 (m, 1H), 0.90 (d, J = 6.6 Hz, 3H), 0.85 (d, J = 6.5 Hz, 3H) ppm.

^{13}C NMR (150.9 MHz, DMSO-d$_6$): δ 174.4, 156.1 (d, $^1J_{CN}$ = 26.4 Hz), 143.9, 143.8, 140.73, 140.71, 127.64, 127.62, 127.06, 127.04, 125.29, 125.26, 120.12, 120.10, 65.5, 52.1 (d, $^1J_{CN}$ = 12.3 Hz), 46.7, 24.3, 22.9, 21.1 ppm. [The following pairs of peaks correspond to the diastereotopic carbons of the Fmoc protecting group: 143.9 and 143.8; 140.73 and 140.71; 127.64 and 127.62; 127.06 and 127.04; 125.29 and 125.26; 120.12 and 120.10.]

Fmoc-[^{15}N] Met-OH: A 100-mL one-neck round-bottom flask equipped with a magnetic stirring bar was charged with ^{15}N-labeled methionine (0.5 g, 3.4 mmol), and Fmoc-OSu (1.07 g, 3.2 mmol). A solution of 1:1 CH_3CN/H_2O (25 mL) and Et_3N (466 L, 3.4 mmol) were added then the reaction mixture was stirred for 15 min. Additional Et_3N was added until a clear solution formed (pH 7.4). The solution was stirred for an additional 1 h. The solution was poured into a solution of 1.0 M HCl (125 mL) in a 250 mL beaker while stirring vigorously. The Fmoc-[^{15}N]Met-OH precipitated from the solution and the solid was isolated by filtering the mixture through a 7 cm Büchner funnel equipped with a piece of filter paper. The solid was dried by aspirating air through the funnel, suspended in EtOAc (ca. 100 mL) to form a clear solution, dried over Na_2SO_4, filtered, concentrated by rotary evaporation, and then stripped twice with DCM. The isolated solid was dried overnight under vacuum and ground up to give ca. 1.18 g (95%) of white powder.

^{1}H NMR (600 MHz, DMSO-d$_6$): δ 12.66 (br s, 1H), 7.89 (d, J = 7.6 Hz, 2H), 7.72 (dd, J = 7.6, 2.7 Hz, 2H), 7.68 (dd, $^1J_{HN}$ = 92.6, 8.2 Hz, 1H), 7.42 (t, J = 7.4 Hz, 2H), 7.33 (t, J = 7.1 Hz, 2H), 4.37 – 4.26 (m, 2H), 4.23 (t, J = 7.1 Hz, 1H), 4.10 (appar ddd, J = 8.6, 8.5 4.2 Hz, 1H), 2.57 – 2.45 (m, 2H), 2.04 (s, 3H), 2.00 – 1.92 (m, 1H), 1.92 – 1.83 (m, 1H) ppm.

^{13}C NMR (150.9 MHz, DMSO-d$_6$): δ 173.7, 156.2 (d, $^1J_{CN}$ = 26.3 Hz), 143.84, 143.77, 140.73, 140.72, 127.6, 127.1, 125.3, 125.2, 120.13, 120.12, 65.6, 52.6 (d, $^1J_{CN}$ = 12.4 Hz), 46.7, 30.3, 29.8, 14.5 ppm. [The following pairs of peaks correspond to the diastereotopic carbons of the Fmoc protecting group: 143.84 and 143.77; 140.73 and 140.72; 125.3 and 125.2; 120.13 and 120.12. There are 2 coincident carbons at 127.6 and 127.1 ppm that could not be resolved.]

Sample Preparation NMR spectroscopic studies of peptides were performed in D_2O or 9:1 H_2O/D_2O. NANOpure water was used in all 9:1 H_2O/D_2O samples. The solutions were prepared gravimetrically by dissolving a weighed portion of peptide in the appropriate volume of solvent. The peptides were dissolved by vortexing and sonicating the sample the sample until the sample appeared homogenous. The molecular weight of peptides was calculated as TFA salts with all amino groups assumed to be protonated. [For peptide **2**, the samples needed to be sonicated and vortexed for at least 30 minutes before the peptide fully dissolved. Furthermore, peptide **2** is prone to precipitation out upon exposure to dissolved ions (e.g. Na^+, Cl^-, etc.) particularly at the peptide higher concentrations (> 6 mM).]

NMR Spectroscopy of Unlabeled Peptides

Data acquisition NMR spectra were of unlabeled peptides were generally recorded on a Bruker 600 MHz spectrometer with a Bruker CBBFO helium-cooled cryoprobe or TBI probe. Presaturation water suppression was applied as needed. TOCSY spectra were recorded with 2048 points in the f_2 dimension and 256 increments in the f_1 dimension with NS = 8 or 16 and a spinlock mixing time between 60 and 200 ms. NOESY spectra were recorded with 2048 points in the f_2 and 256 increments in the f_1 with NS = 8 or 16 and a mixing time of 1 s. A 200-ms mixing time was used for samples containing higher peptide concentrations to limit spin diffusion. ROESY (EXSY) spectra were recorded with 2048 points in the f_2 and 256 increments in the f_1 with NS = 8 and a mixing time of 150-ms.

Data processing NMR spectra were processed with the Bruker TopSpin software. Manual baseline correction was applied to both dimensions after manually phasing the spectra. For TOCSY, NOESY, and ROESY spectra run in D_2O, the spectra were referenced to the solvent residual (δ = 4.81). Solvent suppressed NMR spectra were referenced to diagnostic peaks in the D_2O spectra.

Diffusion-Ordered Spectroscopy (DOSY) Experiments DOSY experiments[5] (@dosy1D.c with the ledbpgp2s1d pulse program) were performed on a Bruker 600 MHz spectrometer equipped with a Bruker CBBFO helium-cooled cryoprobe, with a diffusion delay (Δ) of 75-ms. Variable diffusion gradient lengths (δ) were selected to achieve the appropriate signal attenuation (ca. 20 x) between the 5% – 95% gradient strength. Sixteen sets of FIDs were Fourier transformed in Bruker's Topspin software to give a pseudo-2D spectra. After phasing and baseline correction, each pseudo-2D spectra was calibrated using the residual HOD peak (1.9 x 10^{-9} m^2/s at 298 K). The diffusion coefficients were read as logarithmic values and converted to linear values.

NMR Spectroscopy of ^{15}N-Labeled Peptides

^{1}H,^{15}N HSQC and ^{1}H,^{15}N NOESY-HSQC (^{15}N-edited NOESY) Data Acquisition NMR spectra for isotopically labeled peptides were recorded on the Avance 800 MHz NMR spectrometer equipped with a TXI probe. BioTop was used to optimize parameters for all isotopically labeled experiments run on the 800 MHz spectrometer. ^{1}H,^{15}N HSQC spectra were generally recorded with 2048 points in the f_2 dimension and 282 increments in the f_1 dimension NS = 16. ^{1}H,^{15}N NOESY-HSQC were

generally recorded with 2048 points in the f_3 dimension (^{1}H), 1 increment in the f_2 dimension (^{15}N), and 1024 increments in the f_1 dimension (^{1}H) with NS = 16 and a 120-ms mixing time.

^{1}H,^{15}N HSQC and ^{1}H,^{15}N NOESY-HSQC (^{15}N-edited NOESY) Data Processing NMR spectra were Fourier transformed in Bruker TopSpin software with forward linear prediction and a Qsine weighting function. Manual baseline correction was applied to both dimensions after manually phasing the spectra. The ^{1}H,^{15}N HSQC spectra were processed to a final matrix size of 2048 x 256 real points, using the Qsine weighting function and forward linear prediction. The ^{1}H,^{15}N NOESY-HSQC spectra were processed to a final matrix size of 2048 x 1024 real points, using the Qsine weighting function and forward linear prediction. The ^{1}H,^{15}N-HSQC and ^{15}N-edited NOESY-HSQC spectra were referenced using the xiref command in TopSpin.

Peak Assignments Residue specific peak assignments were determined by performing an NH to Hα walk. Within each residue, the NH proton gives a strong cross peak to the Hα proton in the TOCSY spectra. Between two adjacent residues the Hα (*i*) gives a strong cross peak to the NH proton (*i+1*) on the adjacent residue in the ROESY spectra. The TOCSY and ROESY enables sequential peak assignments of each strand in macrocyclic peptide for both monomer and species **A** and **B**. For the top-strand bearing the *N*-methyl amino acid, the ROESY spectra provides a strong crosspeak between the *N*-methyl group and Hα of Val_{18}. Intramolecular NOEs in the NOESY spectra were used to corroborate the peak assignments of the top and bottom strands. In cases of heavy overlap in either the NH or Hα region (particularly for the peptide **2** oligomer), all possible cases were considered and invalid cases that were inconsistent with the NMR data were ruled out. The peak assignments for species **A** and **B** were cross-checked with the ^{1}H,^{15}N HSQC spectra of

^{15}N labeled analogues (peptides **2a** and **2d**) and with the ^{1}H NMR spectra of the deuterated analogs (peptides **2b** and **2c**).

Molecular Modeling of the Asymmetric Hexamer

We used PyMOL in conjugation with MacroModel to build energy minimized models (local minima) with constraints that are consistent with the observed intermolecular contacts and symmetry requirements. We started with one copy of the monomer from the crystallographic hexamer for species **B**. Then, we used the sculpt function in PyMOL to alter the conformation of the monomer to better match the observed **A**-**B** intermolecular contacts on the exterior of the triangular trimer while also preserving the network of interstrand intramolecular hydrogen bonds (ca. 1.8 – 2.2 Å). Three copies of the species **B** monomer were initially aligned to the crystallographic triangular trimers. Then, we manually adjusted the position of the three molecules to better accommodate the observed **A**-**B** intermolecular contacts while preserving approximate C_3 symmetry. From there, we used the crystallographic coordinates of a more-twisted hairpin (PDB 1HS0) to model our initial conformation for species **A**. We grafted on our sequence onto the 1HS0 structure and locally minimized the conformation using the clean function in PyMOL.[6] The additional twisting of the hairpin for species **A** better accommodated the top-to-bottom strand **A**-**A** intermolecular contacts. Three molecules of **A** were positioned manually in PyMOL onto the triangular trimers while simultaneously satisfying the **A**-**A** intermolecular contacts, **A**-**B** intermolecular contacts, and approximate C_3 symmetry.

We minimized (local minima) our rough model of the hexamer in MacroModel using the MMFFs force field with GB/SA water solvation and two to three distance constraints along each of the observed **A-A**, **A-B** (1), **A-B** (2), and **B-B** interfaces. Force constants were 100 kJ/mol-Å^2 for all interatomic distance with a ± 1 Å tolerance. We used a 2 Å distance for strong NOEs, 3 Å distance for moderate NOEs, and 4 Å distance for weaker NOEs. The PRCG method was used with a maximum of 9000 iterations and a 0.05 kJ/Å-mol convergence threshold on the gradient. The coordinates were exported in .pdb file format and imported into PyMOL. The initial coordinates of the solution phase dodecamer were generated by duplicating the hexamer structure and manually positioning the relative orientation of the two hexamers.

Molecular Dynamic Simulations on a pair of hexamers

Molecular dynamics simulation was performed using the Amber22 software package,[7] with the FF19SB force field for protein.[8] The non-standard residues CHA and ORN are generated utilizing the same RESP charge fitting protocol[9] as the amino acid residues in Amber22. The system was solvated with explicit OPC water molecules[10] in a periodic box of ~100Å in each dimension, and just the necessary number of counterions (Na^+ or Cl^-) to neutralize the overall charge of the system. Equilibration of the system followed a consistent protocol which began with short minimization runs to fix possible bad contacts in initial structure, proceeded to gradually heating the system to 300 K, then constant pressure (NPT) simulation at 1 atm and 300 K to equilibrate the simulation box size to be consistent with experimental density of an aqueous solution. In the case of run 4 and 5, a 50 ns run with distance constraints to optimize the hydrophobic contacts between the desired surfaces is carried out for each system. Finally, a production run (no constraints except for run 2) was performed in the NVT ensemble at 300 K for 500 ns, with a timestep of 1 fs. The trajectories were then analyzed, focusing on the structural features and conformational changes, with the cpptraj program in the Amber22 package.

We initially hypothesized that the exposed hydrophobic ends of the cylindrin-like trimer might form a contact. We thus started with a pair of simulations in which two hexamers are arranged with the ends of the cylindrin-like trimer facing each other and stabilized by a network of intermolecular hydrogen bonds between the two cylindrins' Orn_1 and Met_{35} residues. Simulation 1 was run without distance constraints, while simulation 2 was run with additional distance constraints along the hypothesized interface. The constrained dimer of hexamers was stable for the duration of simulation 2. However, in simulation 1 the hypothesized interface broke apart, and the two hexamers rearranged to come together on the sides of two cylindrin-like trimer, stabilized by hydrophobic contacts involving the side chains of residues Val_{18}, Phe_{20}, Ala_{30}, Ile_{32}, Leu_{34}, and Val_{36}. Simulation 3, starting with a frame from simulation 2 and then removing the constraints along the interface, also resulted in the interface between the two hexamers breaking apart. Collectively, simulations 1–3 show that the hypothesized configuration of the dodecamer is not stable, but that there are other possible stable arrangements of two hexamers.

To further investigate possible stable arrangements of two hexamers, we ran two additional simulations that varied the relative orientation of the two hexamers. Simulation 4 started with the solvent exposed ends of the triangular trimer subunits of each hexamer stacked on top of each other while simulation 5 started with the cylindrin-like trimer subunits of each hexamer arranged in a side-to-side manner. In both simulations 4 and 5, similar to that of simulation 1, the two hexamers stayed together with stabilizing hydrophobic contacts between the side chains of residues Val_{18}, Phe_{20}, Ala_{30}, Ile_{32}, Leu_{34}, and Val_{36} in between the two asymmetric hexamers.

References and Notes

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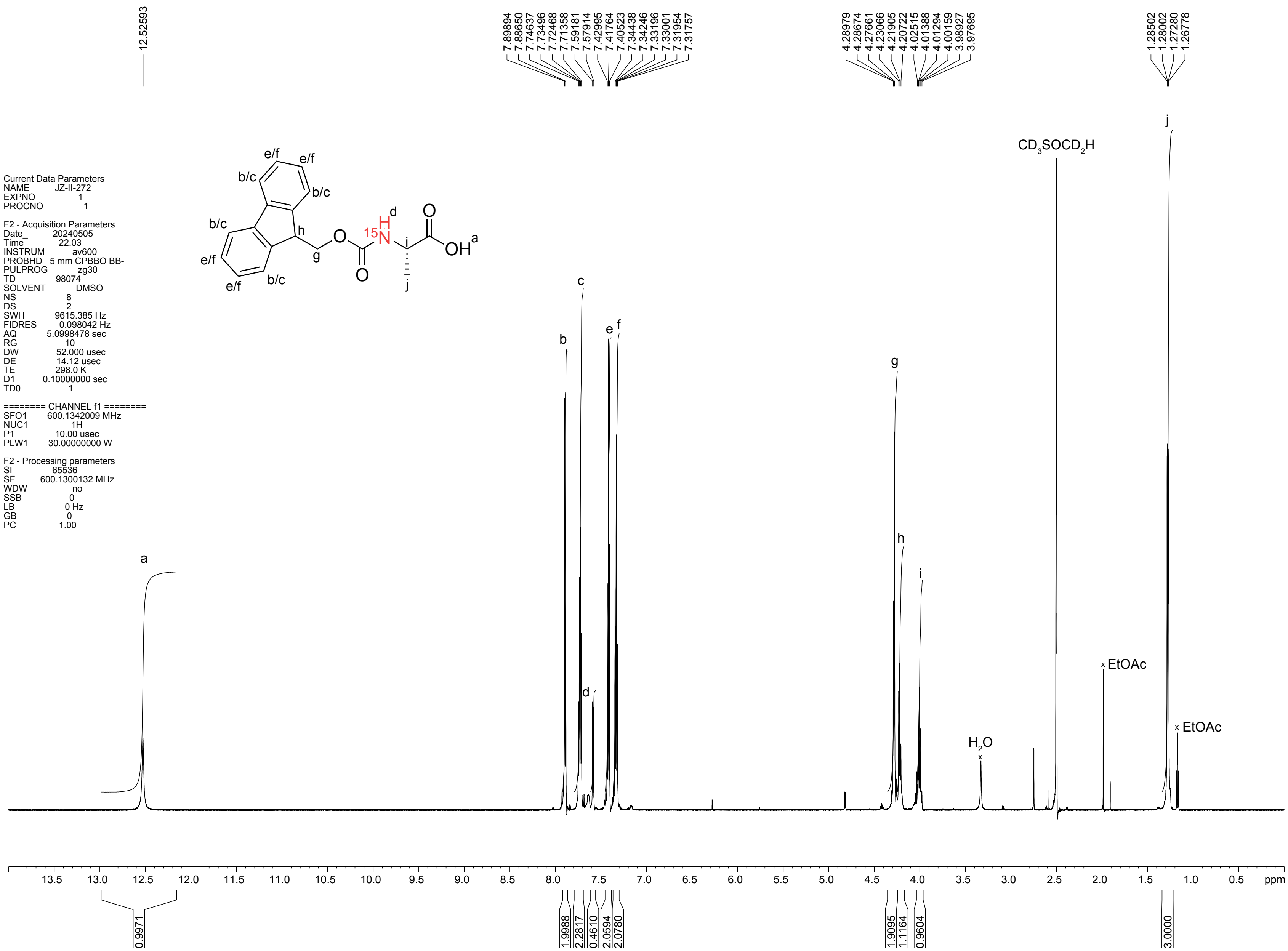
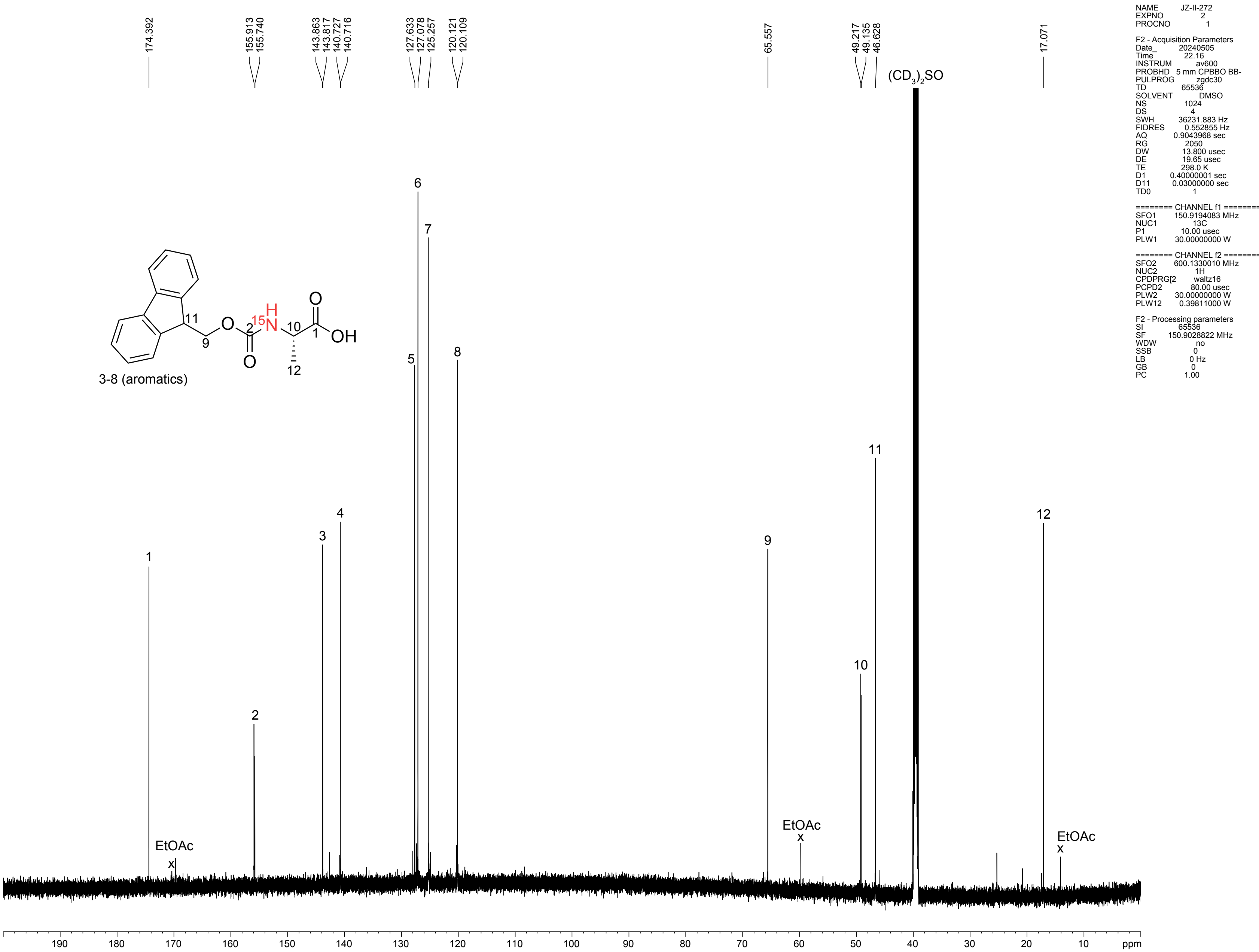
NMR Characterization Data

^{1}H NMR, and ^{13}C NMR spectra of Fmoc-^{15}N-Ala-OH in CD_3SOCD_3 at 600 MHz and 298 K.





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49.135
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======== CHANNEL f2 ========
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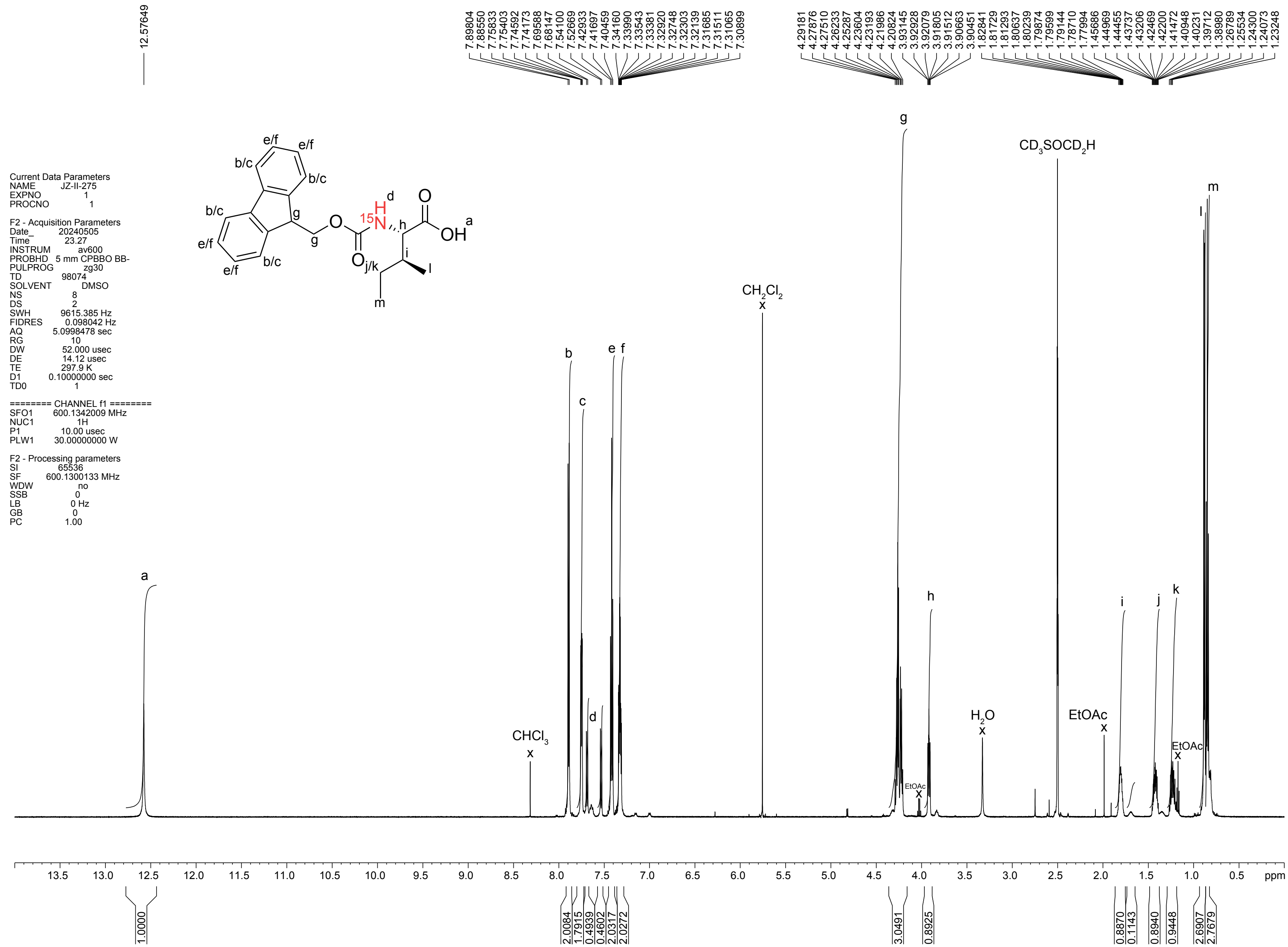
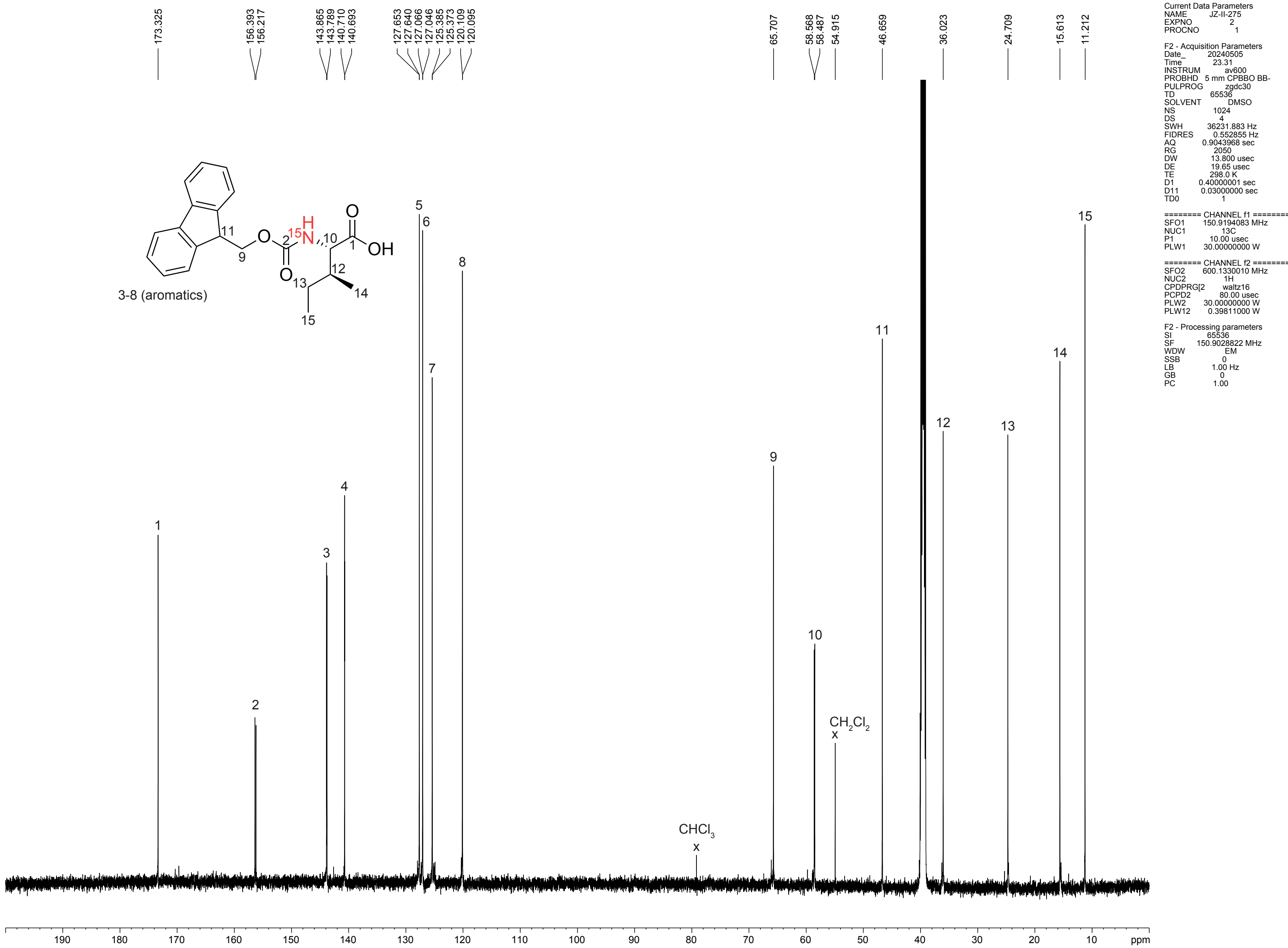
^{1}H NMR, and ^{13}C NMR spectra of Fmoc-^{15}N-Ile-OH in CD_3SOCD_3 at 600 MHz and 298 K.





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120.109
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65.707
58.568
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24.709
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3-8 (aromatics)
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CH2Cl2
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RG 2050
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DE 19.65 usec
TE 298.0 K
D1 0.40000001 sec
D11 0.03000000 sec
TD0 1
======== CHANNEL f1 ========
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NUC1 13C
P1 10.00 usec
PLW1 30.00000000 W
======== CHANNEL f2 ========
SFO2 600.1330010 MHz
NUC2 1H
CPDPRG[2 waltz16
PCPD2 80.00 usec
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F2 - Processing parameters
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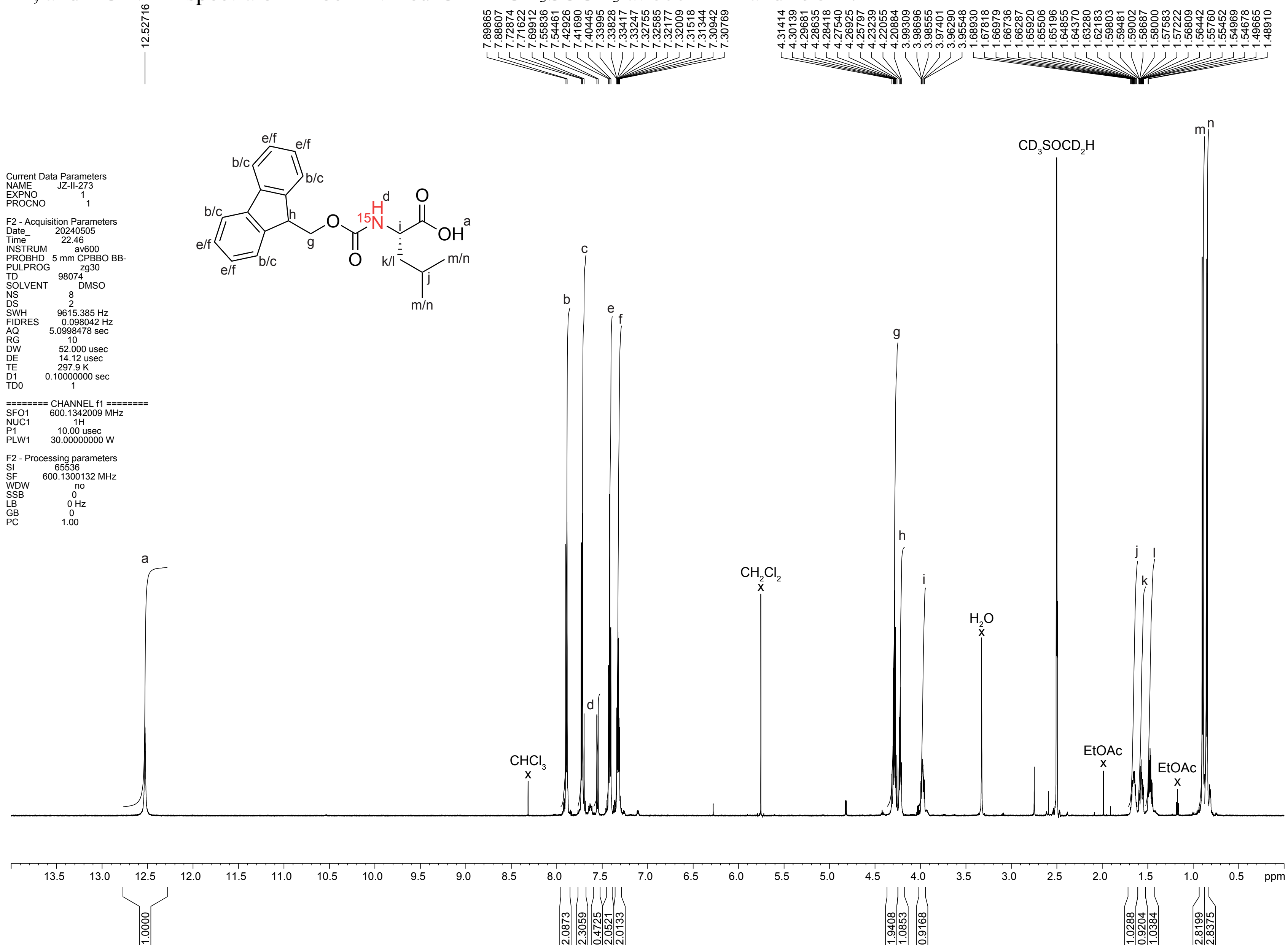
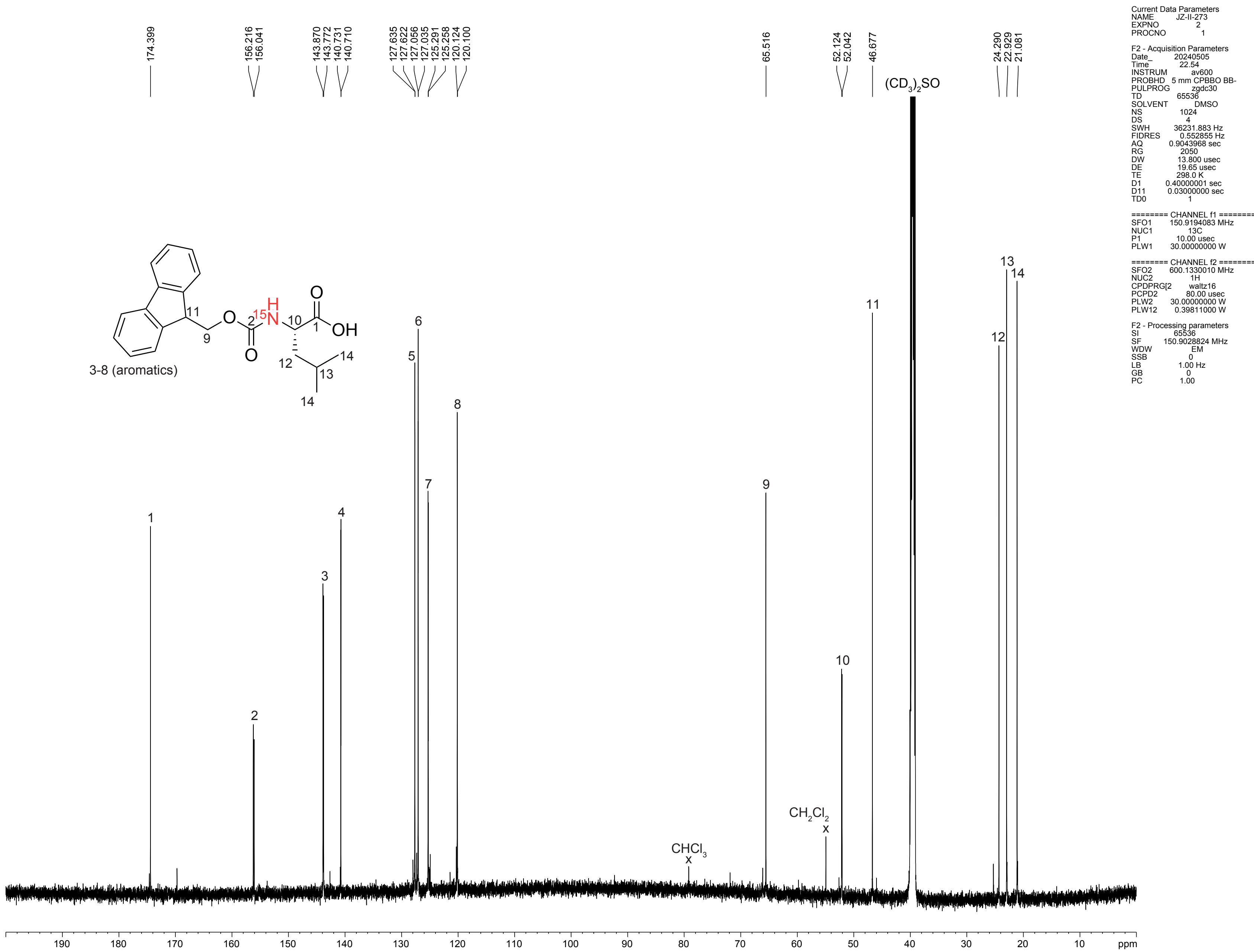
^{1}H NMR, and ^{13}C NMR spectra of Fmoc-^{15}N-Leu-OH in CD_3SOCD_3 at 600 MHz and 298 K.

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CH_2Cl_2
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PROCNO 1
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FIDRES 0.552855 Hz
AQ 0.9043968 sec
RG 2050
DW 13.800 usec
DE 19.65 usec
TE 298.0 K
D1 0.40000001 sec
D11 0.03000000 sec
TD0 1
======== CHANNEL f1 ========
SFO1 150.9194083 MHz
NUC1 13C
P1 10.00 usec
PLW1 30.00000000 W
======== CHANNEL f2 ========
SFO2 600.1330010 MHz
NUC2 1H
CPDPRG[2 waltz16
PCPD2 80.00 usec
PLW2 30.00000000 W
PLW12 0.39811000 W
F2 - Processing parameters
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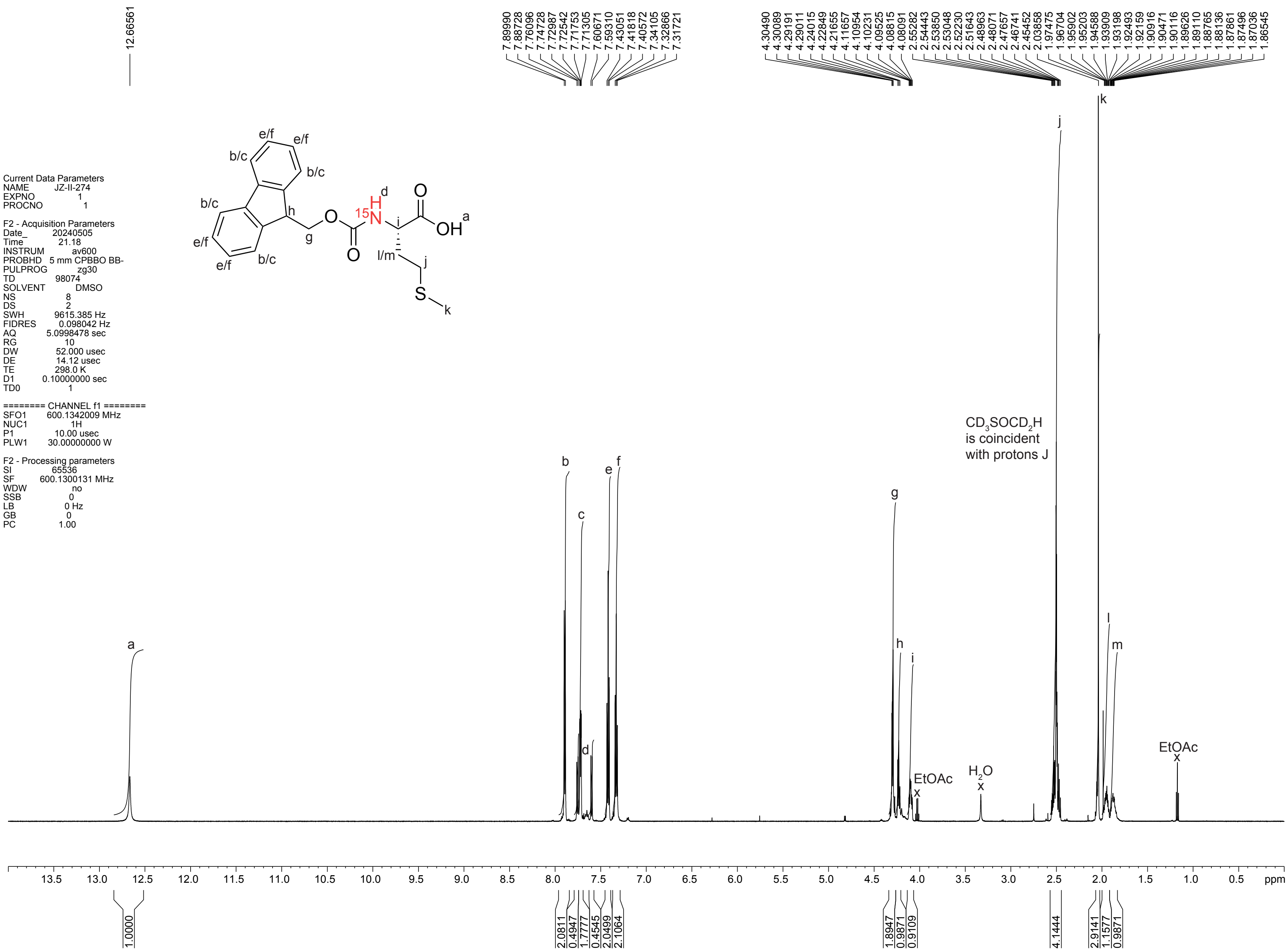
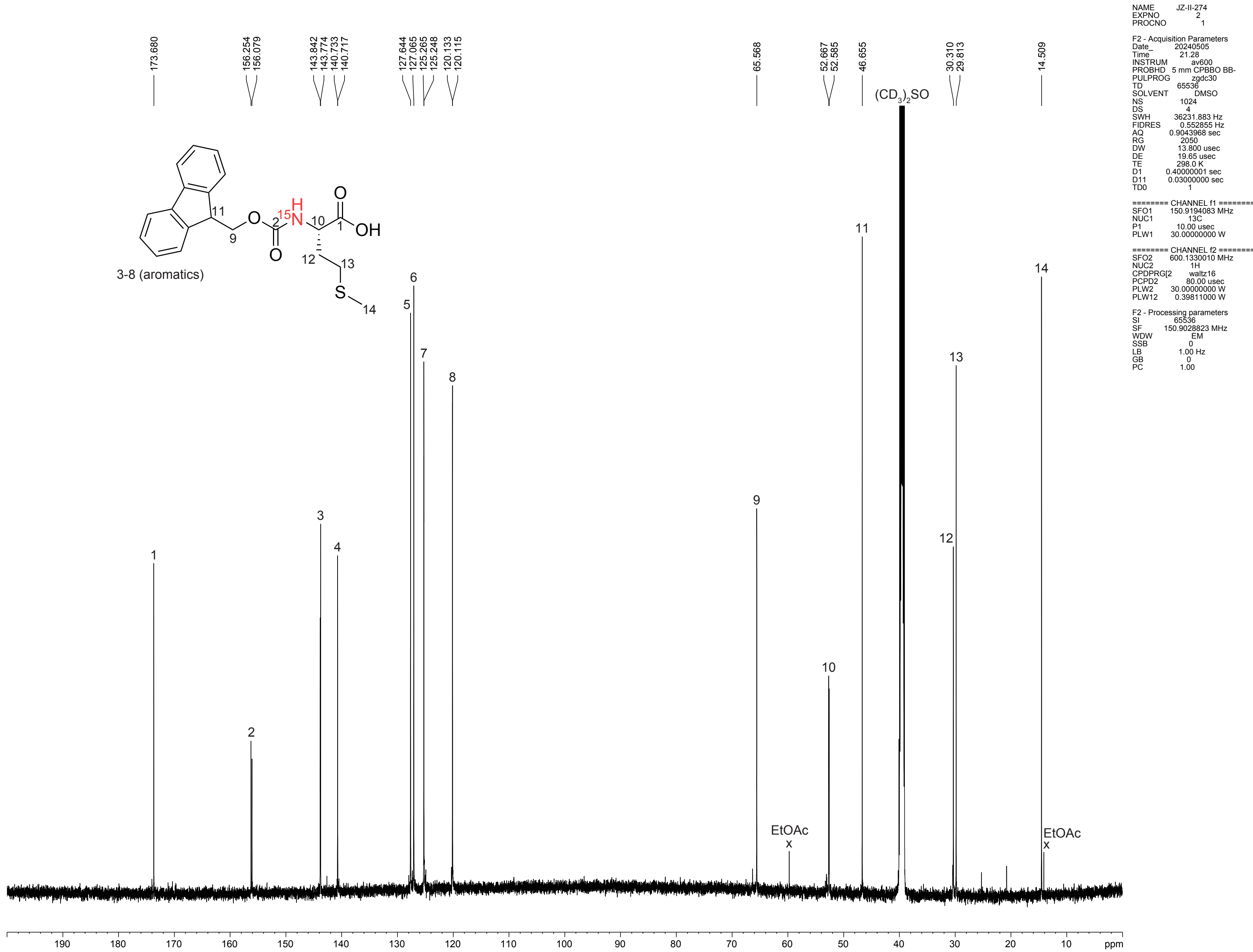
SF 150.9028824 MHz
WDW EM
SSB 0
LB 1.00 Hz
GB 0
PC 1.00

[1]H NMR, and [13]C NMR spectra of Fmoc-[15]N-Met-OH in CD_3SOCD_3 at 600 MHz and 298 K.

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EtOAc
x
ppm
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PROCNO 1
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FIDRES 0.552855 Hz
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RG 2050
DW 13.800 usec
DE 19.65 usec
TE 298.0 K
D1 0.40000001 sec
D11 0.03000000 sec
TD0 1
======== CHANNEL f1 ========
SFO1 150.9194083 MHz
NUC1 13C
P1 10.00 usec
PLW1 30.00000000 W
======== CHANNEL f2 ========
SFO2 600.1330010 MHz
NUC2 1H
CPDPRG[2 waltz16
PCPD2 80.00 usec
PLW2 30.00000000 W
PLW12 0.39811000 W
F2 - Processing parameters
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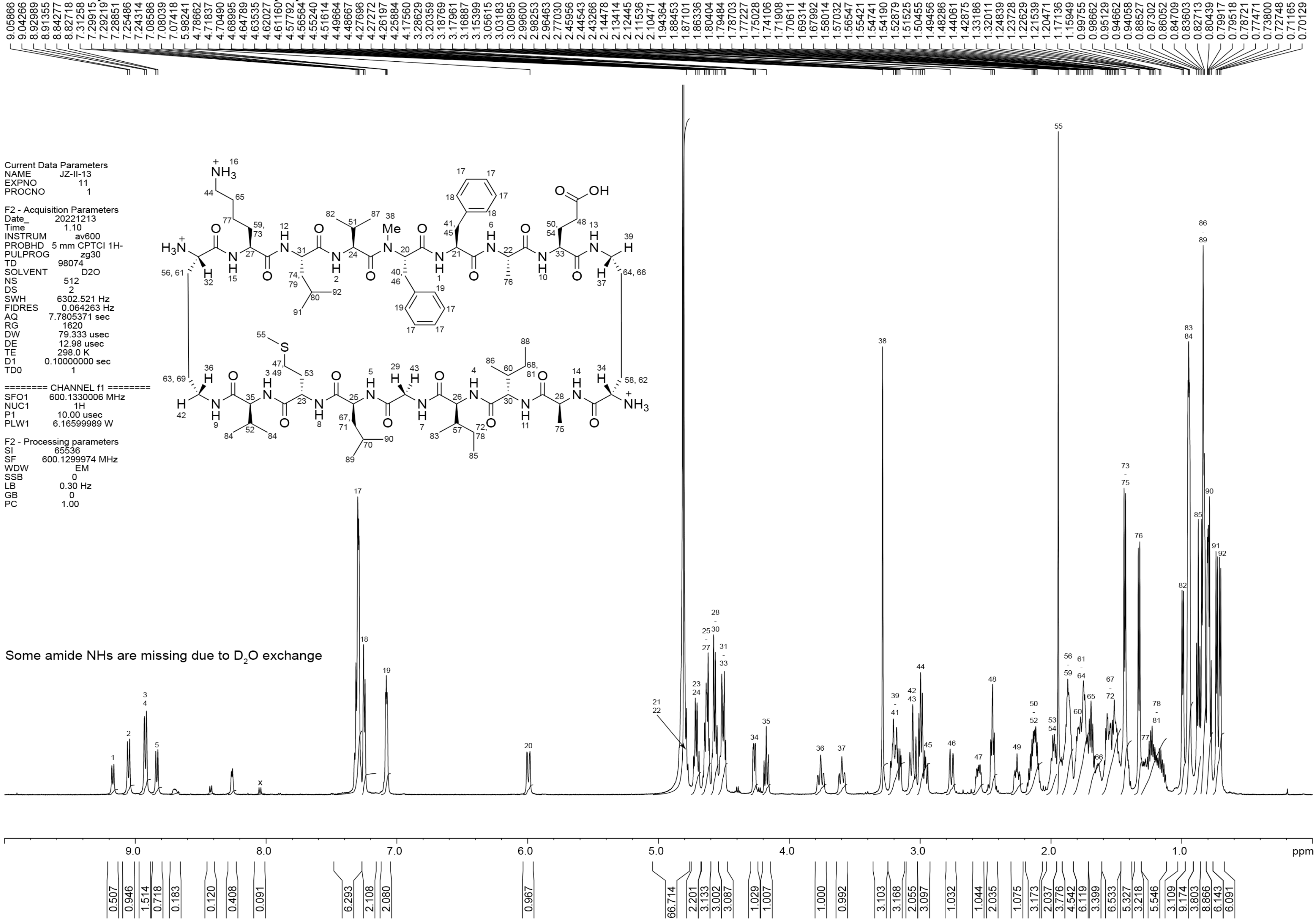
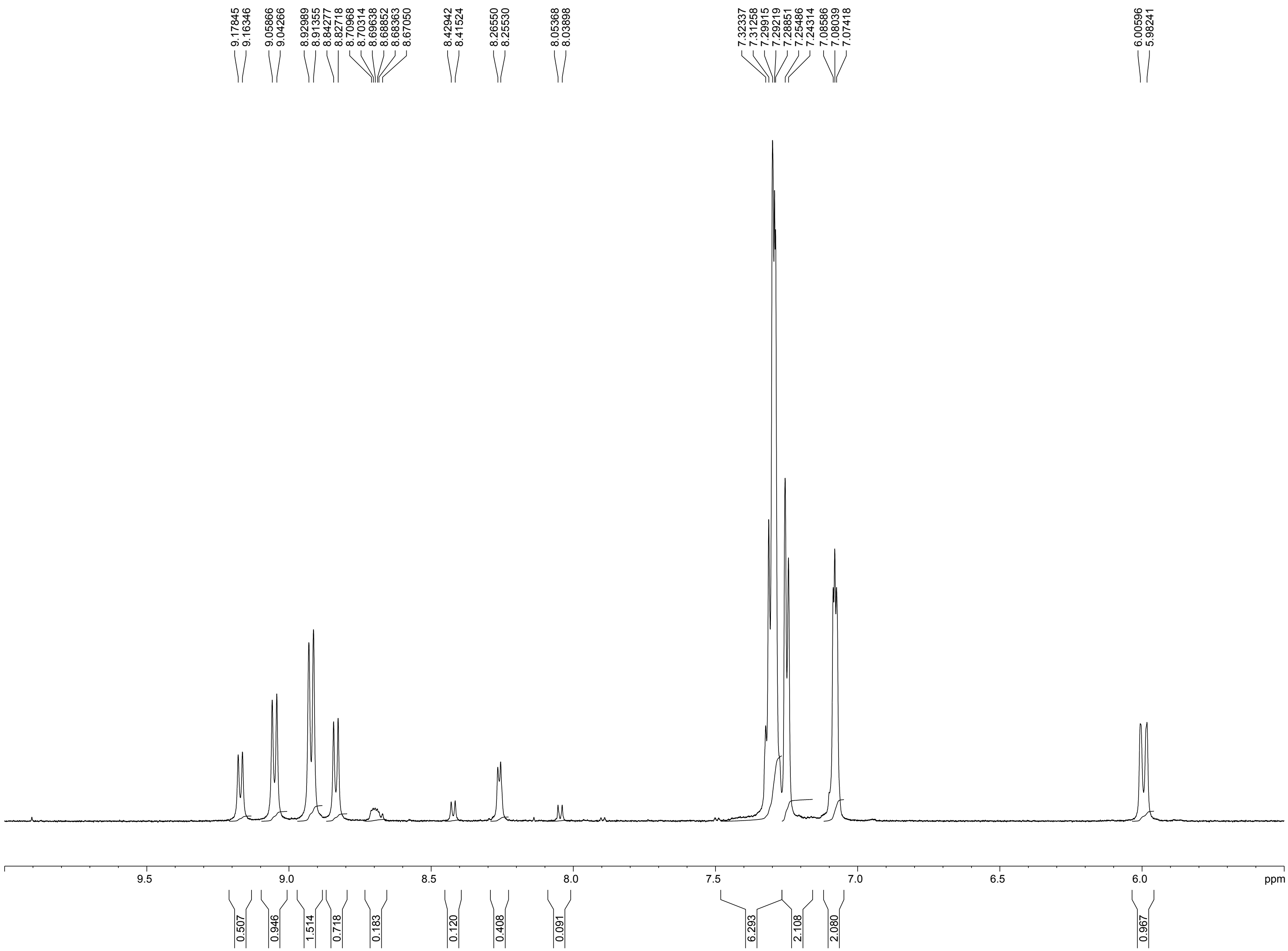
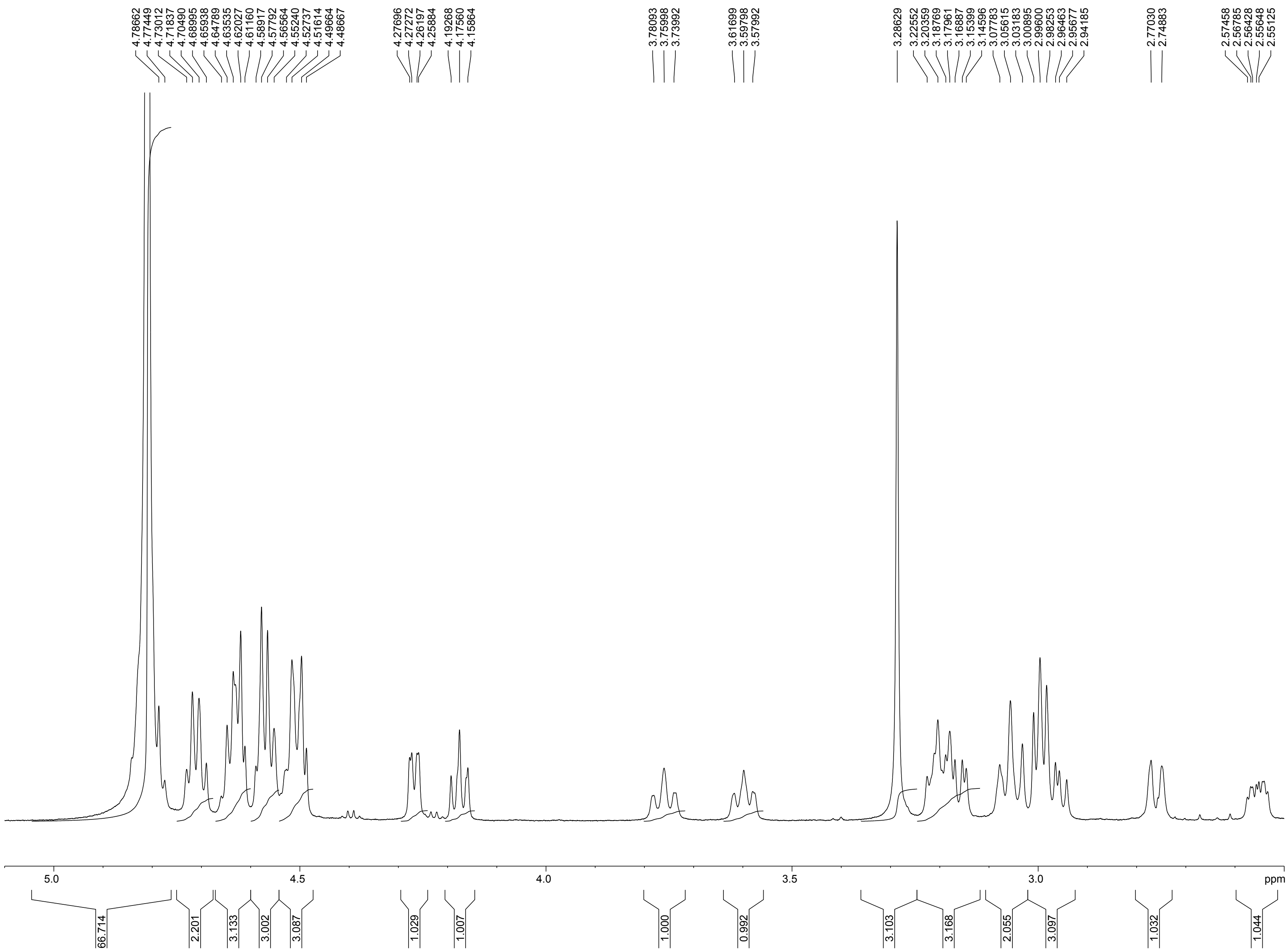
^{1}H NMR spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K



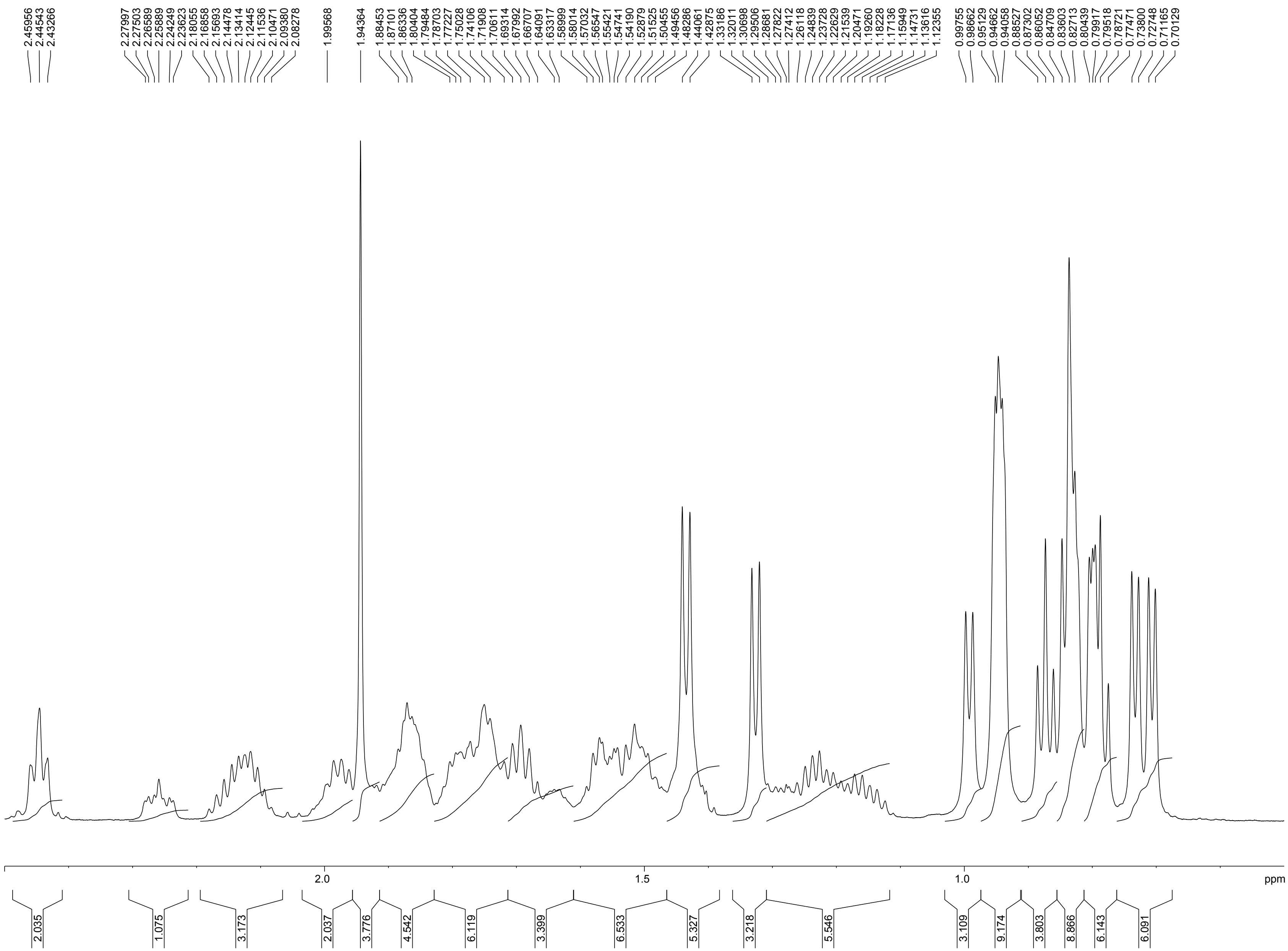
^{1}H NMR spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K



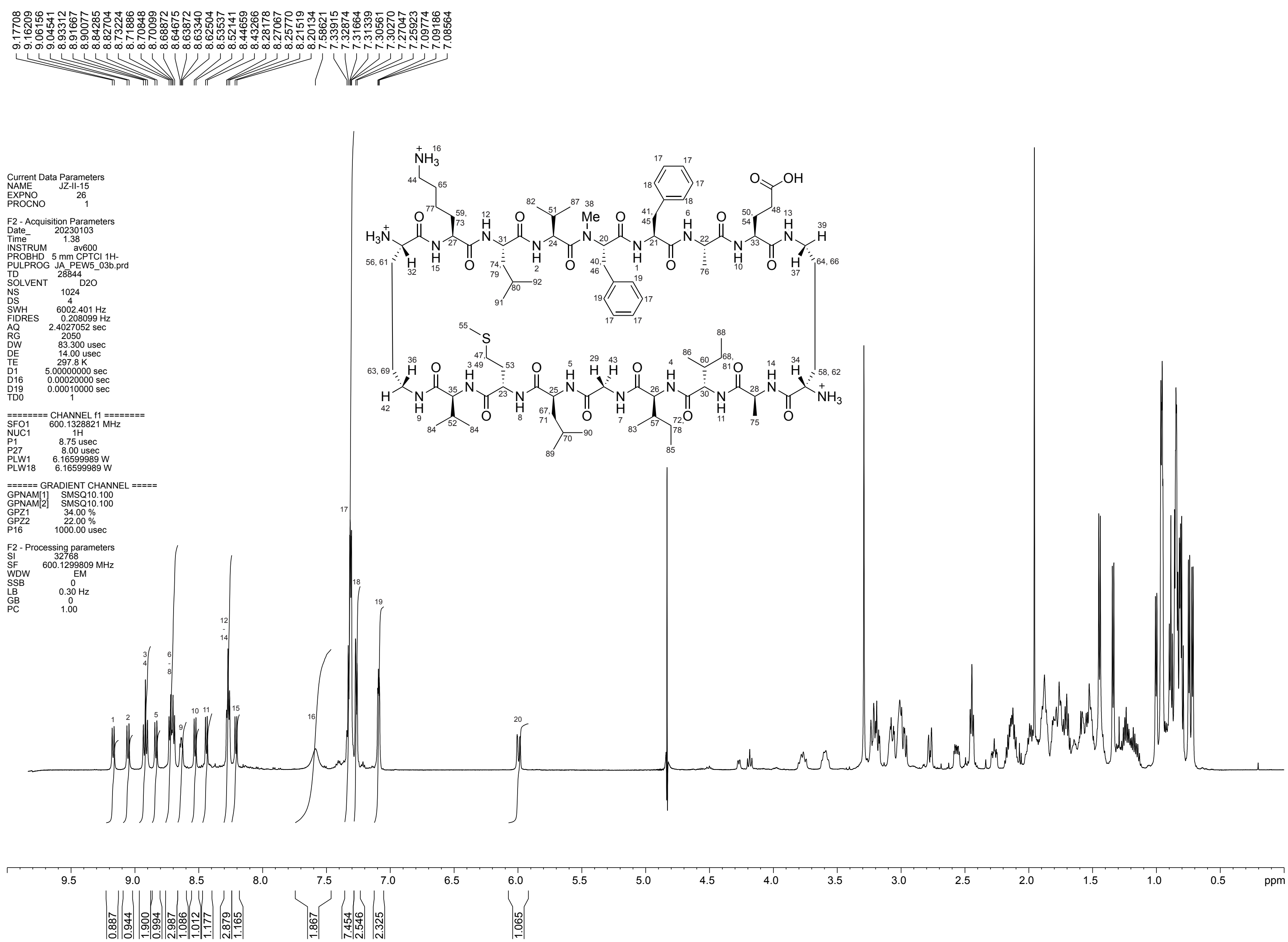
[1]H NMR spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K



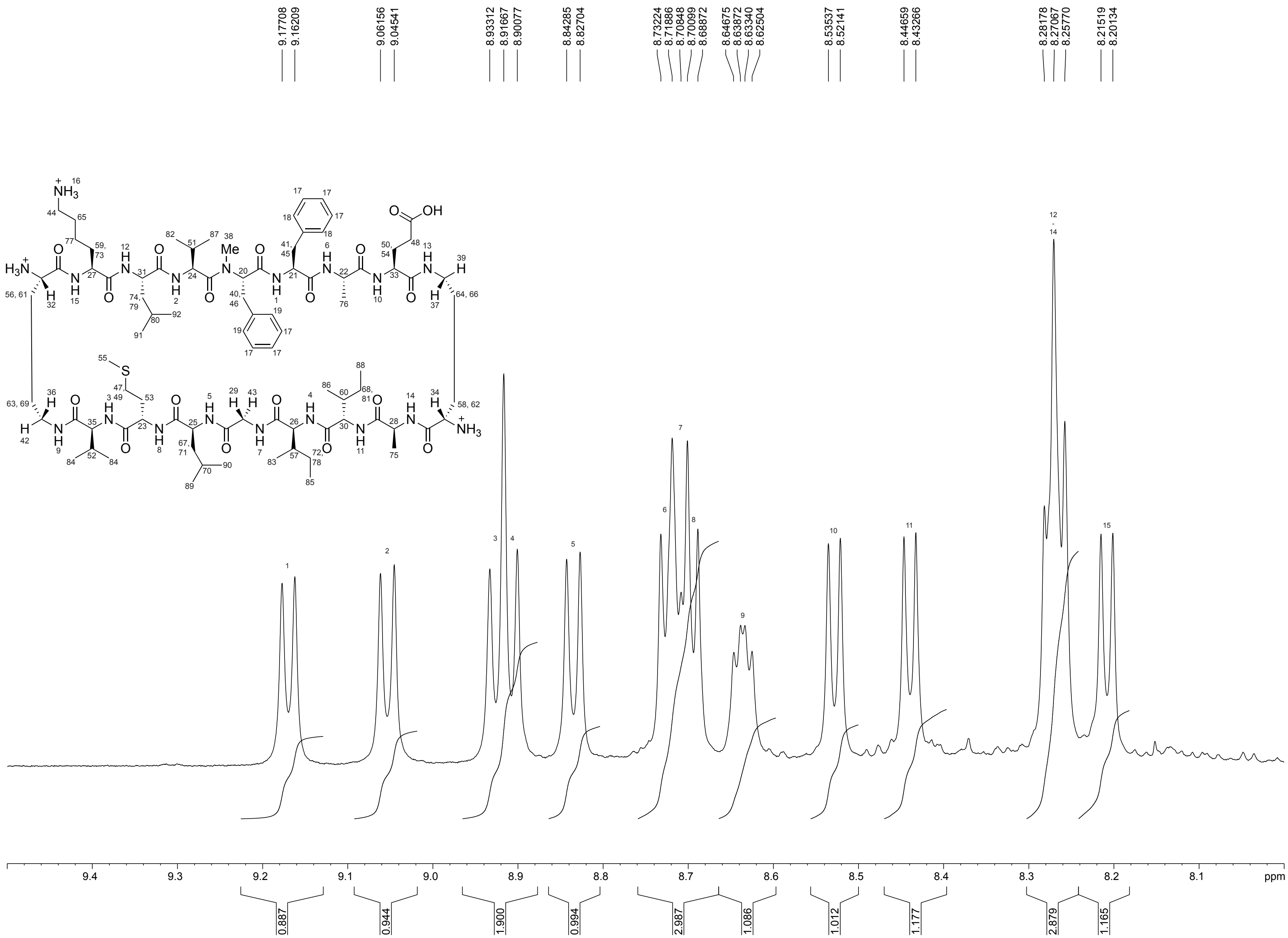
^{1}H NMR spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K



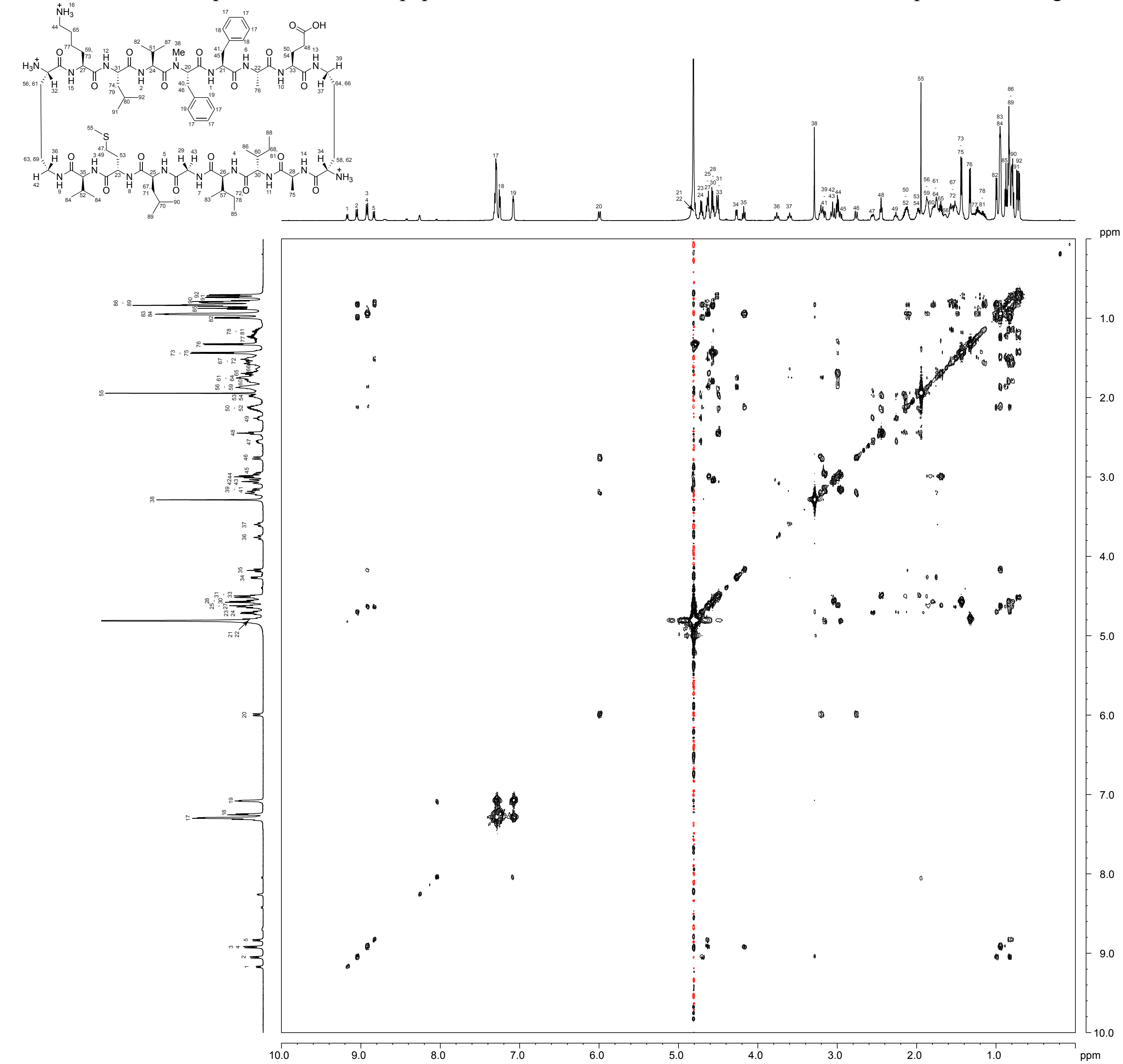
^{1}H NMR spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak.



^{1}H NMR spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak



^{1}H NMR 2D TOCSY spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K with 200-ms spin-lock mixing time



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FIDRES 3.082584 Hz
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RG 1620
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DE 10.00 usec
TE 298.0 K
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D1 2.00000000 sec
D9 0.20000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
IN0 0.00015840 sec
L1 126

======== CHANNEL f1 ========
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P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
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PLW10 0.63095999 W

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GPNAM[2] SMSQ10.100
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GPZ2 10.00 %
P16 1000.00 usec

F1 - Acquisition parameters
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FIDRES 49.321339 Hz
SW 10.520 ppm
FnMODE QF

F2 - Processing parameters
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SF 600.1299988 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 QF
SF 600.1299987 MHz
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SSB 0
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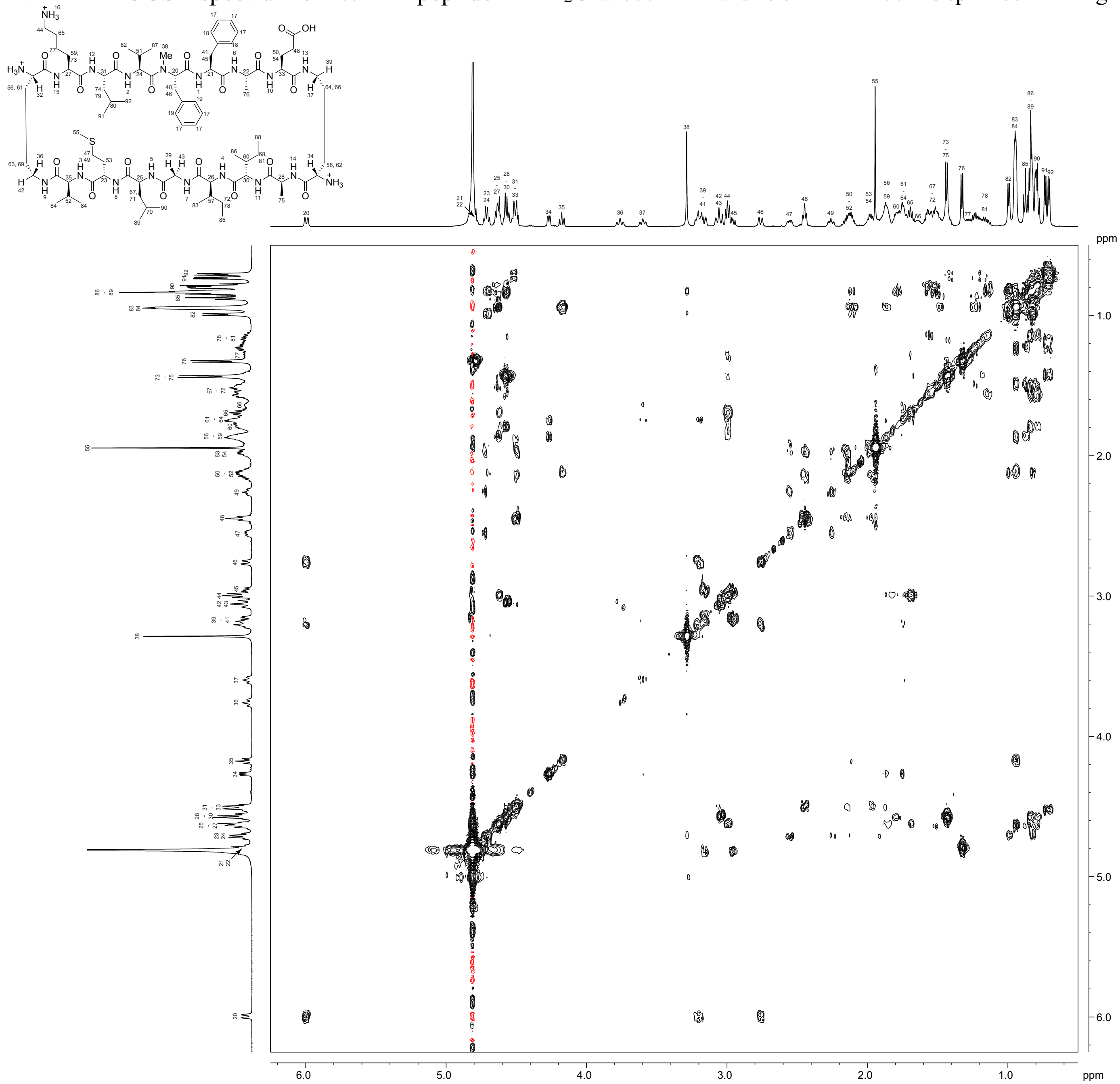
GB 0

[1]H NMR 2D TOCSY spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K with 200-ms spin-lock mixing time



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PROCNO               1

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SOLVENT            D2O
NS                   8
DS                  16
SWH         6313.131 Hz
FIDRES      3.082584 Hz
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RG                1620
DW          79.200 usec
DE           10.00 usec
TE             298.0 K
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D1       2.00000000 sec
D9       0.20000000 sec
D12      0.00002000 sec
D16      0.00020000 sec
IN0      0.00015840 sec
L1                 126

======== CHANNEL f1 ========
SFO1      600.1330006 MHz
NUC1                 1H
P1              10.00 usec
P5              16.67 usec
P6              25.00 usec
P7              50.00 usec
P17           2500.00 usec
PLW1          6.16599989 W
PLW10         0.63095999 W

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GPNAM[2]     SMSQ10.100
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GPZ2            10.00 %
P16          1000.00 usec

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SFO1        600.133 MHz
FIDRES     49.321339 Hz
SW           10.520 ppm
FnMODE               QF

F2 - Processing parameters
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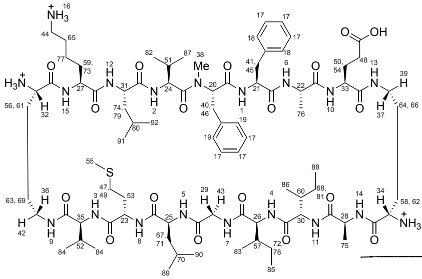
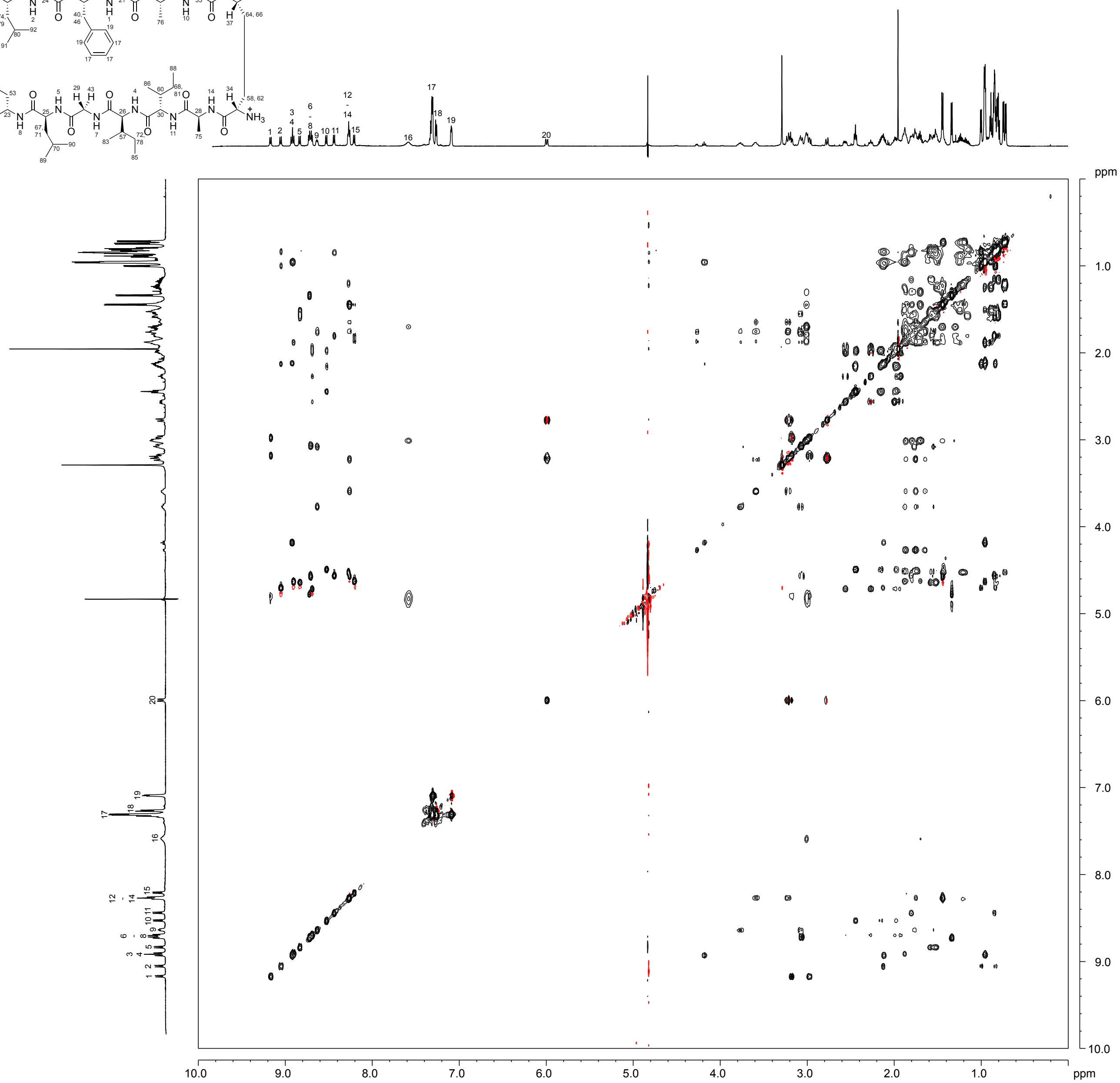
WDW                SINE
SSB                   0
LB                 0 Hz
GB                    0
PC                 1.40

F1 - Processing parameters
SI                 1024
MC2                  QF
SF       600.1299987 MHz
WDW                SINE
SSB                   0
LB                 0 Hz
GB                    0
```

[1]H NMR 2D TOCSY spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 60-ms spin-lock mixing time



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D1       2.00000000 sec
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D12      0.00002000 sec
D16      0.00020000 sec
D19      0.00010000 sec
IN0      0.00015140 sec
L1                  36
P2          16.00 usec

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NUC1                1H
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P6          25.00 usec
P7          50.00 usec
P17       2500.00 usec
P27          8.00 usec
PLW1      6.16599989 W
PLW10     0.63095999 W
PLW18     6.16599989 W

====== GRADIENT CHANNEL =====
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GPZ2           22.00 %
P16       1000.00 usec

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SW          11.006 ppm
FnMODE     States-TPPI

F2 - Processing parameters
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SF     600.1299799 MHz
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SSB                  2
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GB                   0
PC                1.00

F1 - Processing parameters
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MC2        States-TPPI
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WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
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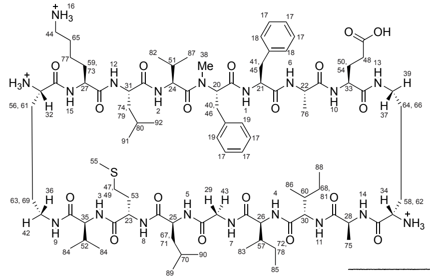
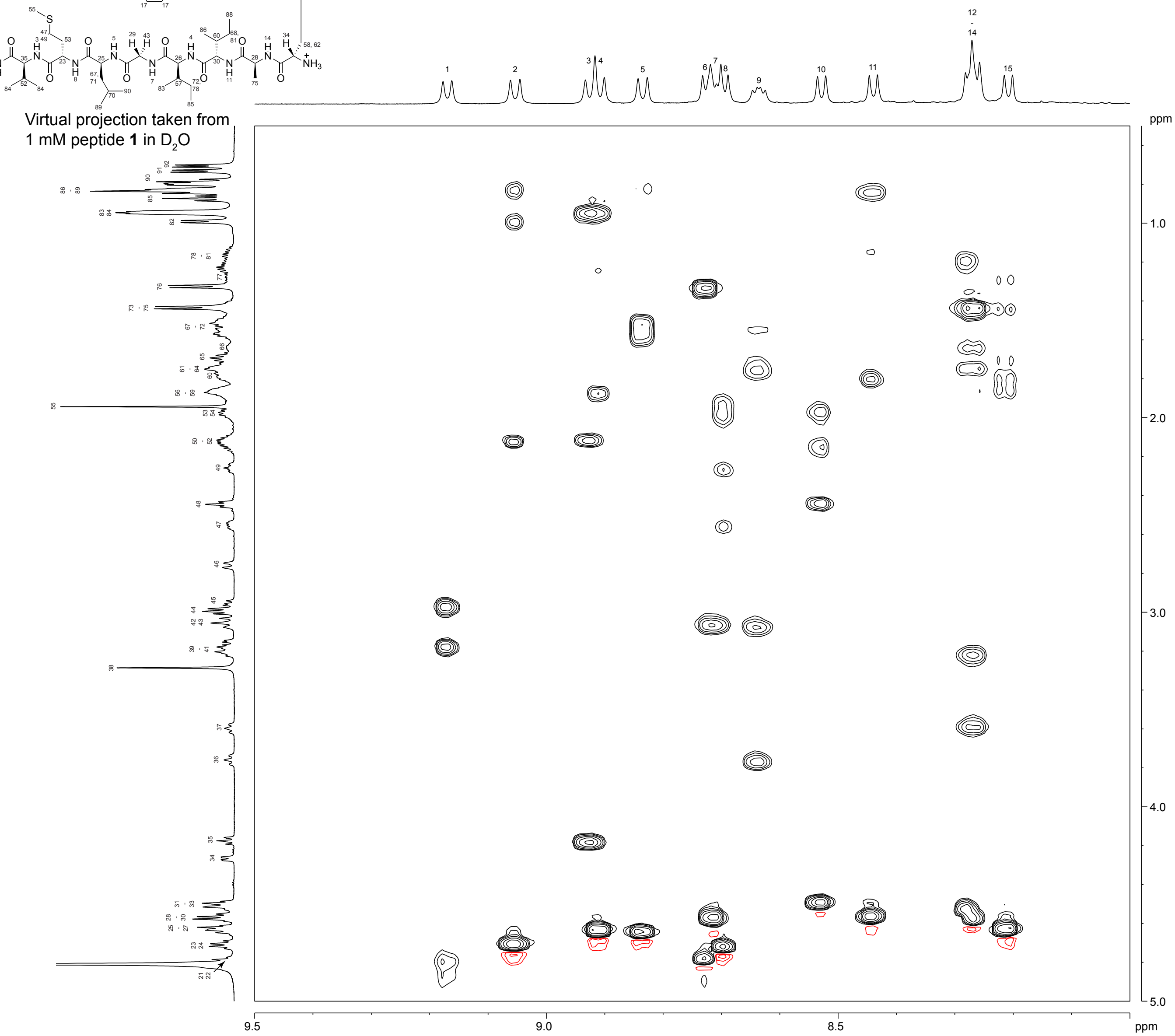
[1]H NMR 2D TOCSY spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 60-ms spin-lock mixing time





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SOLVENT D2O
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DS 16
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FIDRES 3.223688 Hz
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DE 10.00 usec
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D16 0.00020000 sec
D19 0.00010000 sec
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======== CHANNEL f1 ========
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P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
P27 8.00 usec
PLW1 6.16599989 W
PLW10 0.63095999 W
PLW18 6.16599989 W

====== GRADIENT CHANNEL =====
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GPNAM[2] SMSQ10.100
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GPZ2 22.00 %
P16 1000.00 usec

F1 - Acquisition parameters
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FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE States-TPPI

F2 - Processing parameters
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SF 600.1299799 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299805 MHz
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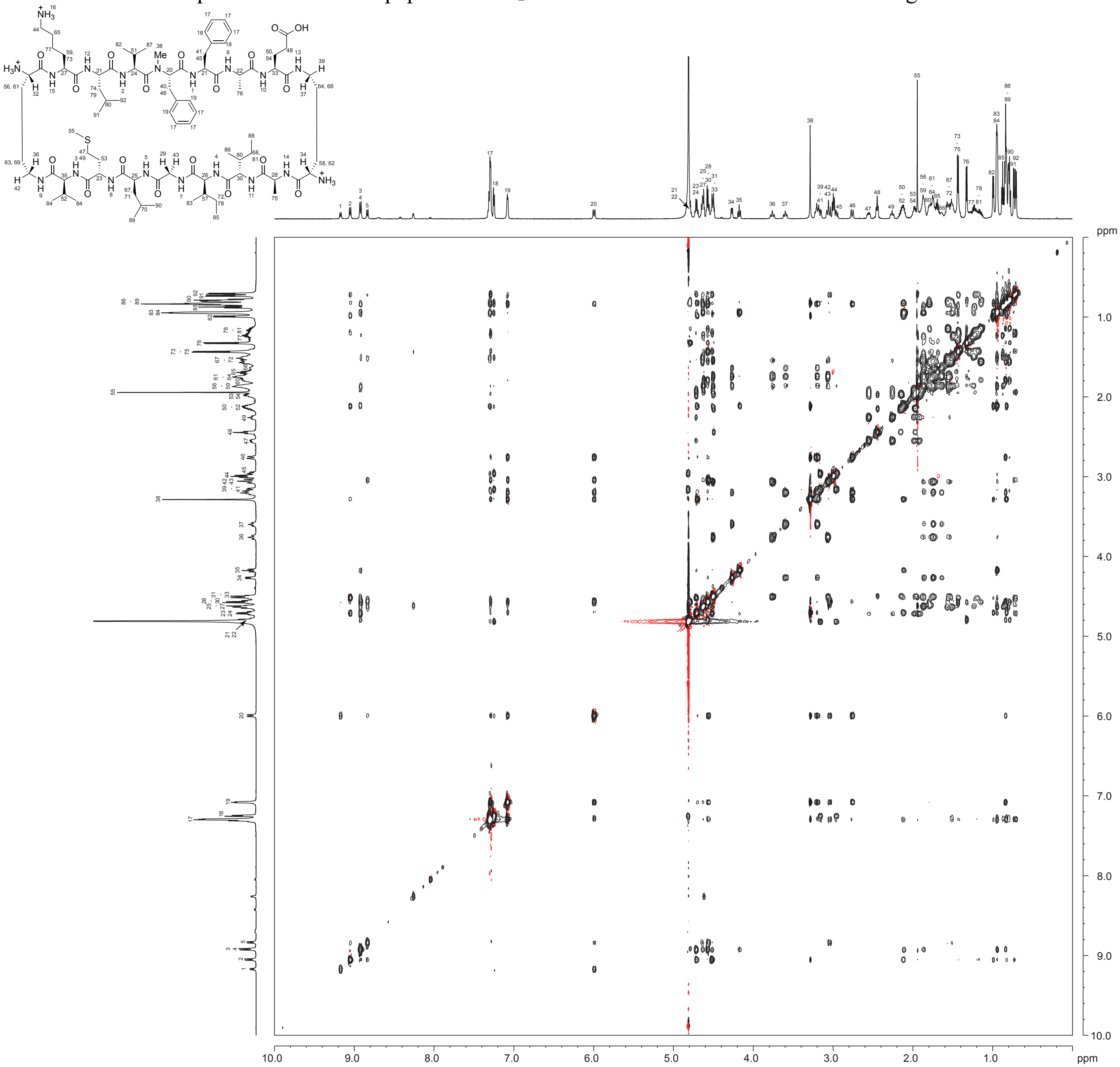
^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K with 1-s mixing time



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DS 16
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FIDRES 3.077403 Hz
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RG 2050
DW 79.333 usec
DE 18.63 usec
TE 297.9 K
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D1 2.00000000 sec
D8 1.00000000 sec
D16 0.00020000 sec
IN0 0.00015860 sec

======== CHANNEL f1 ========
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NUC1 1H
P1 10.00 usec
P2 20.00 usec
PLW1 6.16599989 W

====== GRADIENT CHANNEL =====
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GPZ1 40.00 %
P16 1000.00 usec

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FIDRES 49.259144 Hz
SW 10.506 ppm
FnMODE States-TPPI

F2 - Processing parameters
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SF 600.1299945 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299942 MHz
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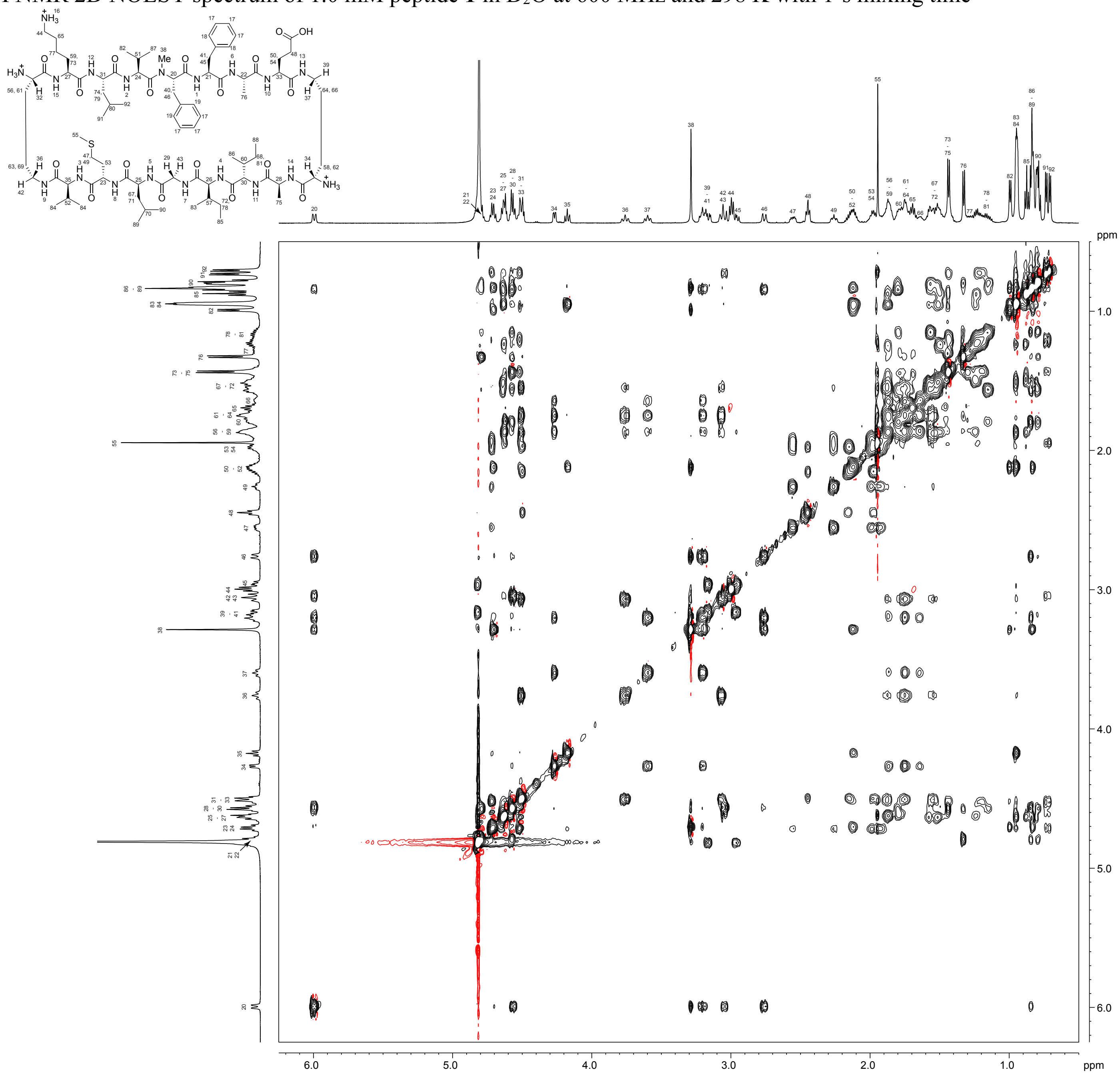
SSB 2
LB 0 Hz
GB 0

[1]H NMR 2D NOESY spectrum of 1.0 mM peptide **1** in D_2O at 600 MHz and 298 K with 1-s mixing time



Current Data Parameters
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EXPNO 13
PROCNO 1

F2 - Acquisition Parameters
Date_ 20221213
Time 2.34
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG noesygpph
TD 2048
SOLVENT D2O
NS 16
DS 16
SWH 6302.521 Hz
FIDRES 3.077403 Hz
AQ 0.1624747 sec
RG 2050
DW 79.333 usec
DE 18.63 usec
TE 297.9 K
D0 0.00006657 sec
D1 2.00000000 sec
D8 1.00000000 sec
D16 0.00020000 sec
IN0 0.00015860 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
P2 20.00 usec
PLW1 6.16599989 W

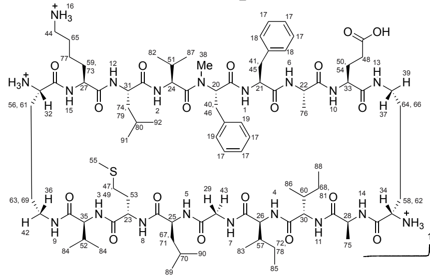
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPZ1 40.00 %
P16 1000.00 usec

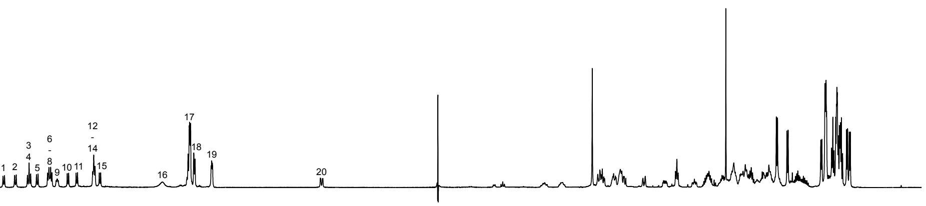
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 49.259144 Hz
SW 10.506 ppm
FnMODE States-TPPI

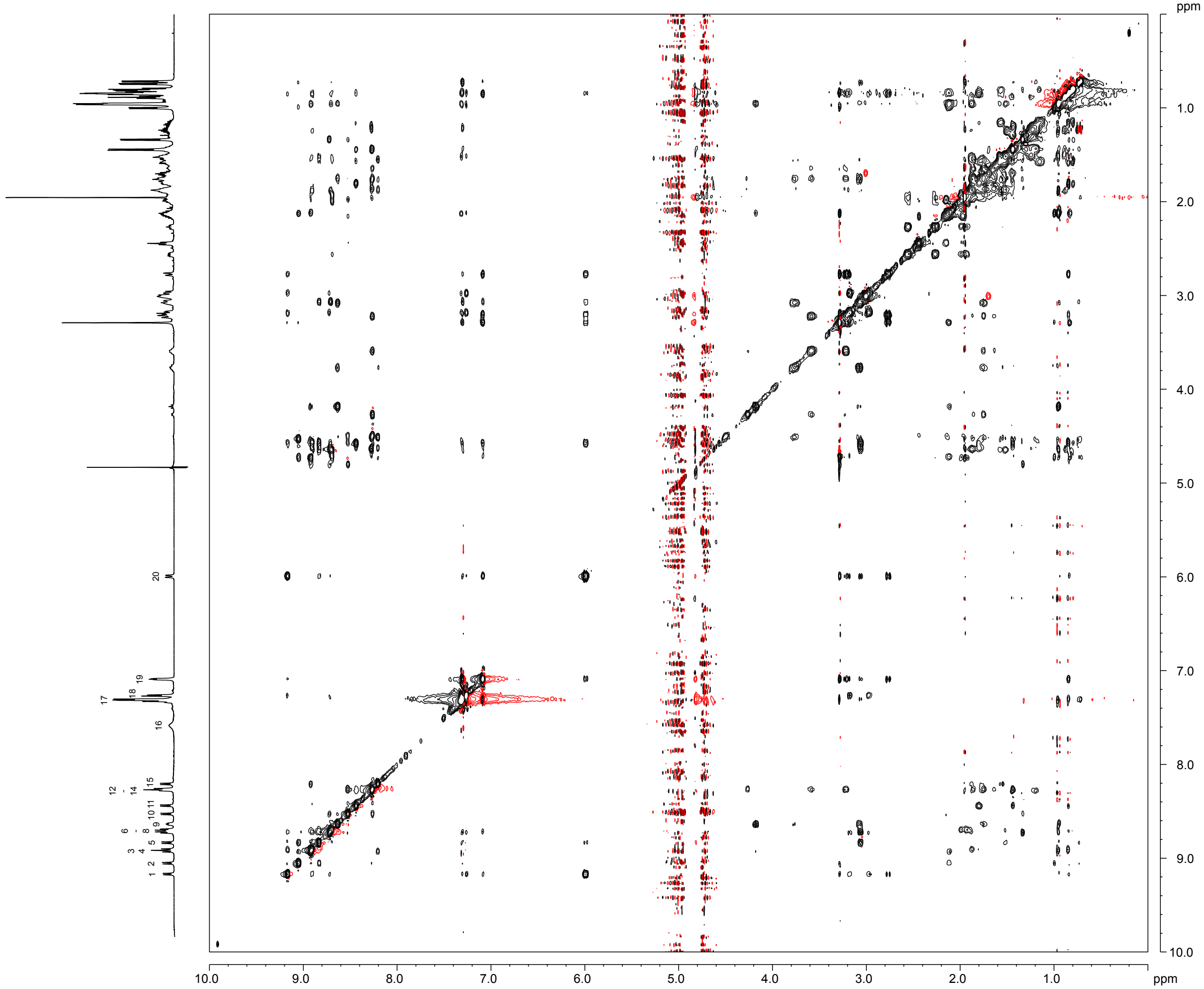
F2 - Processing parameters
SI 1024
SF 600.1299945 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299942 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time

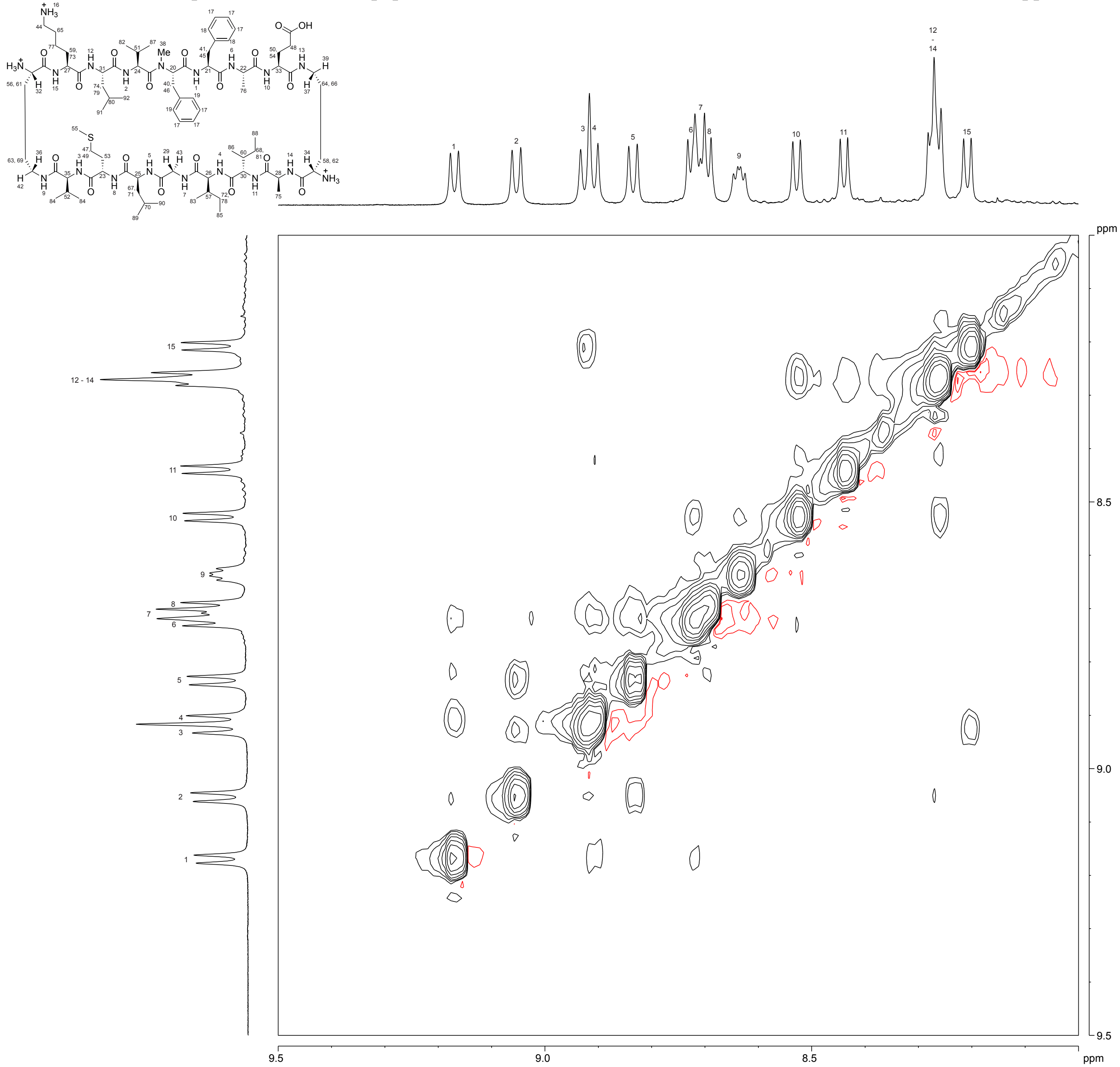






Current Data Parameters	
NAME	JZ-II-15
EXPNO	28
PROCNO	1
F2 - Acquisition Parameters	
Date_	20230103
Time	2.57
INSTRUM	av600
PROBHD	5 mm CPTCI 1H-
PULPROG	noesygpphw5
TD	2048
SOLVENT	D2O
NS	16
DS	16
SWH	6602.113 Hz
FIDRES	3.223688 Hz
AQ	0.1551019 sec
RG	2050
DW	75.733 usec
DE	10.00 usec
TE	297.7 K
D0	0.00006551 sec
D1	2.00000000 sec
D8	1.00000000 sec
D11	0.03000000 sec
D12	0.00002000 sec
D13	0.00000400 sec
D16	0.00020000 sec
D19	0.00010000 sec
IN0	0.00015140 sec
======== CHANNEL f1 ========	
SFO1	600.1328968 MHz
NUC1	1H
P1	8.00 usec
P27	8.00 usec
PLW1	6.16599989 W
PLW18	6.16599989 W
PLW32	0.00000063 W
====== GRADIENT CHANNEL =====	
GPNAM[1]	SMSQ10.100
GPNAM[2]	SMSQ10.100
GPZ1	34.00 %
GPZ2	22.00 %
P16	1000.00 usec
F1 - Acquisition parameters	
TD	256
SFO1	600.1329 MHz
FIDRES	51.601719 Hz
SW	11.006 ppm
FnMODE	States-TPPI
F2 - Processing parameters	
SI	1024
SF	600.1299831 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.00
F1 - Processing parameters	
SI	1024
MC2	States-TPPI
SF	600.1299804 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time

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Current Data Parameters
NAME          JZ-II-15
EXPNO               28
PROCNO               1

F2 - Acquisition Parameters
Date_         20230103
Time              2.57
INSTRUM          av600
PROBHD   5 mm CPTCI 1H-
PULPROG    noesygpphw5
TD                2048
SOLVENT            D2O
NS                  16
DS                  16
SWH        6602.113 Hz
FIDRES     3.223688 Hz
AQ       0.1551019 sec
RG                2050
DW          75.733 usec
DE           10.00 usec
TE             297.7 K
D0      0.00006551 sec
D1      2.00000000 sec
D8      1.00000000 sec
D11     0.03000000 sec
D12     0.00002000 sec
D13     0.00000400 sec
D16     0.00020000 sec
D19     0.00010000 sec
IN0     0.00015140 sec

======== CHANNEL f1 ========
SFO1     600.1328968 MHz
NUC1                  1H
P1              8.00 usec
P27             8.00 usec
PLW1        6.16599989 W
PLW18       6.16599989 W
PLW32       0.00000063 W

====== GRADIENT CHANNEL =====
GPNAM[1]      SMSQ10.100
GPNAM[2]      SMSQ10.100
GPZ1             34.00 %
GPZ2             22.00 %
P16         1000.00 usec

F1 - Acquisition parameters
TD                   256
SFO1        600.1329 MHz
FIDRES      51.601719 Hz
SW            11.006 ppm
FnMODE       States-TPPI

F2 - Processing parameters
SI                  1024
SF       600.1299831 MHz
WDW                QSINE
SSB                    2
LB                  0 Hz
GB                     0
PC                  1.00

F1 - Processing parameters
SI                  1024
MC2          States-TPPI
SF       600.1299804 MHz
WDW                QSINE
SSB                    2
LB                  0 Hz
GB                     0
```
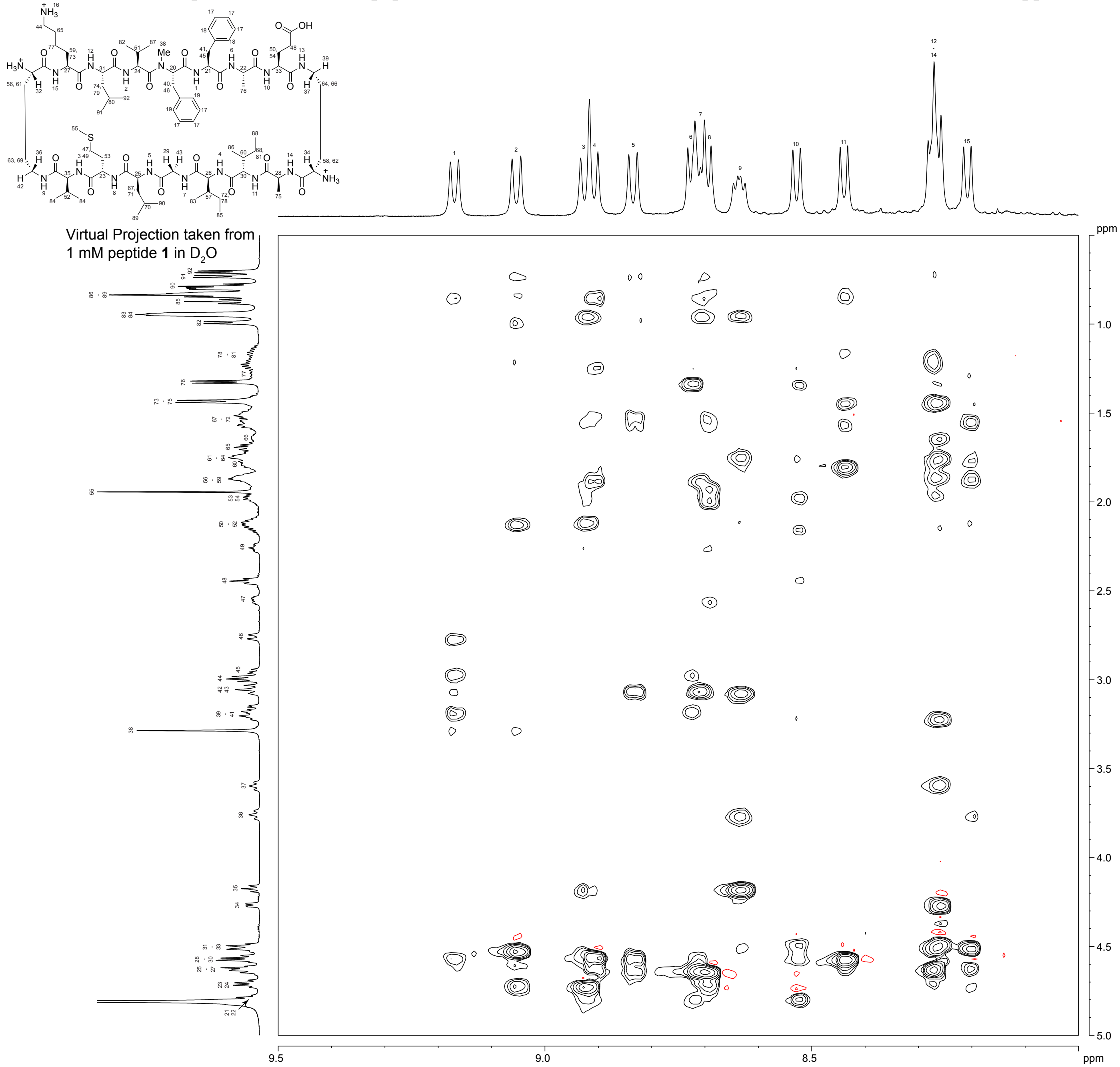

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **1** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time

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Current Data Parameters
NAME          JZ-II-15
EXPNO               28
PROCNO               1

F2 - Acquisition Parameters
Date_         20230103
Time              2.57
INSTRUM          av600
PROBHD   5 mm CPTCI 1H-
PULPROG    noesygpphw5
TD                2048
SOLVENT            D2O
NS                  16
DS                  16
SWH        6602.113 Hz
FIDRES     3.223688 Hz
AQ       0.1551019 sec
RG                2050
DW         75.733 usec
DE          10.00 usec
TE             297.7 K
D0      0.00006551 sec
D1      2.00000000 sec
D8      1.00000000 sec
D11     0.03000000 sec
D12     0.00002000 sec
D13     0.00000400 sec
D16     0.00020000 sec
D19     0.00010000 sec
IN0     0.00015140 sec

======== CHANNEL f1 ========
SFO1    600.1328968 MHz
NUC1                 1H
P1               8.00 usec
P27              8.00 usec
PLW1       6.16599989 W
PLW18      6.16599989 W
PLW32      0.00000063 W

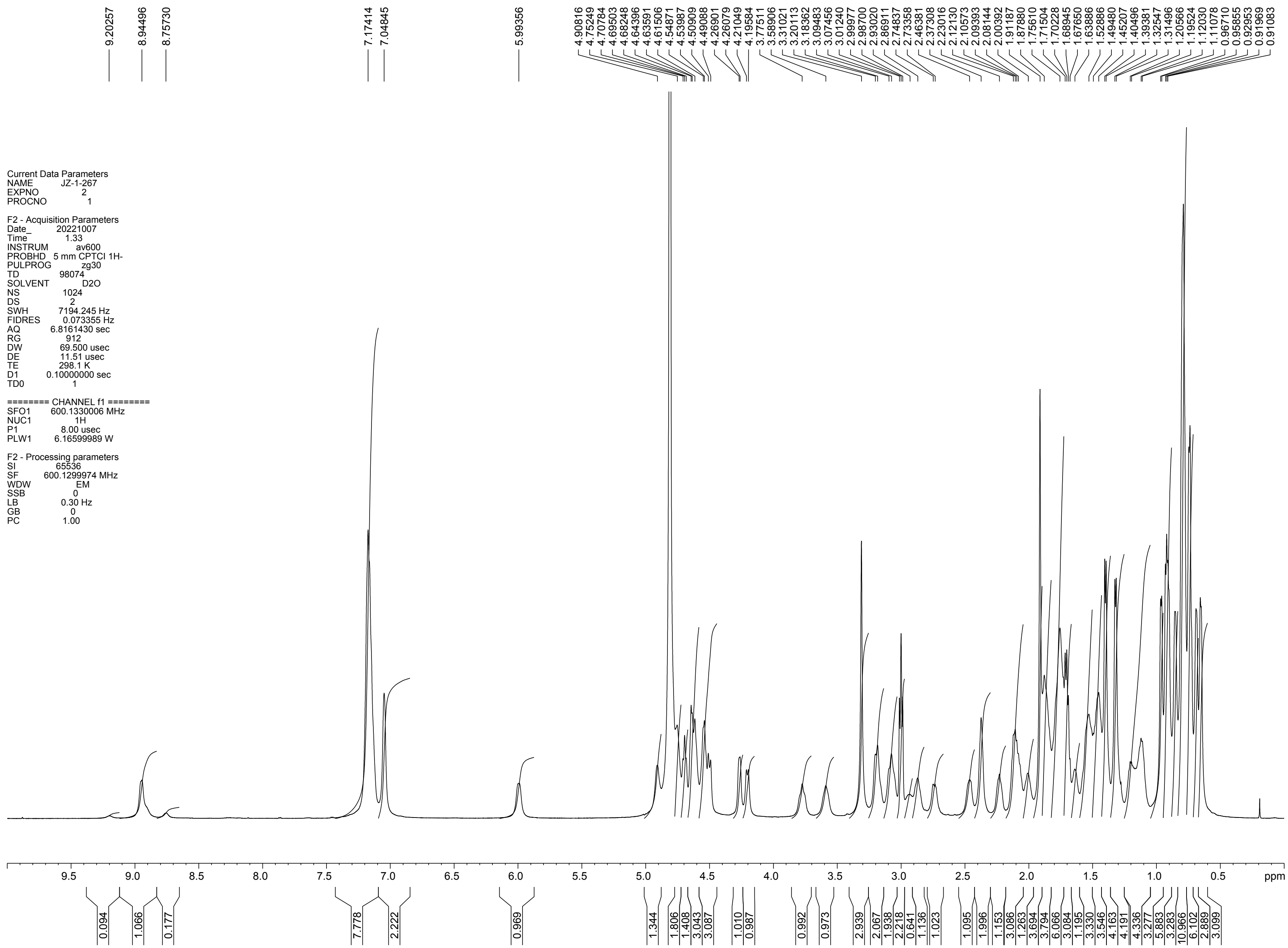
====== GRADIENT CHANNEL =====
GPNAM[1]     SMSQ10.100
GPNAM[2]     SMSQ10.100
GPZ1            34.00 %
GPZ2            22.00 %
P16        1000.00 usec

F1 - Acquisition parameters
TD                  256
SFO1       600.1329 MHz
FIDRES    51.601719 Hz
SW           11.006 ppm
FnMODE      States-TPPI

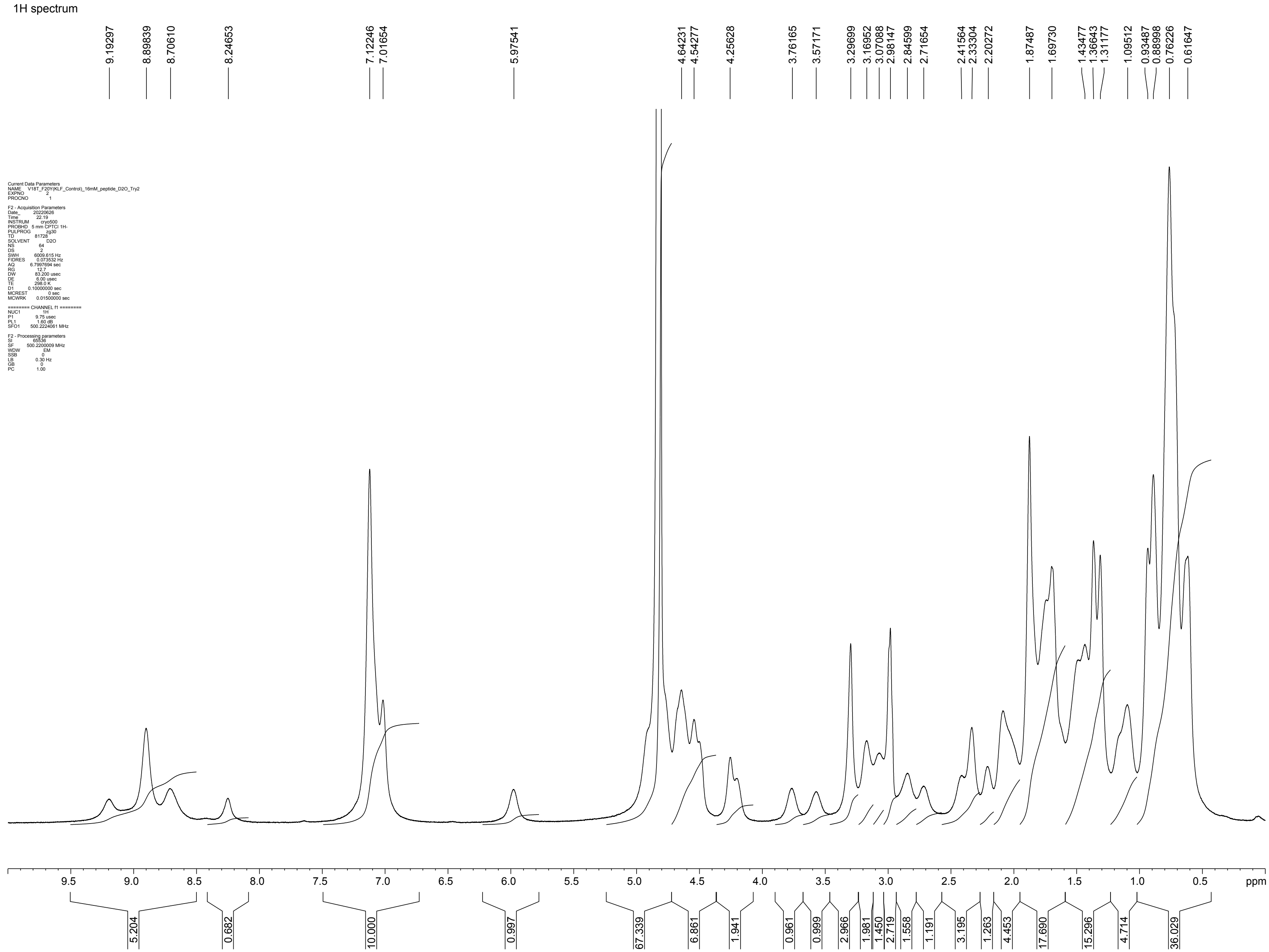
F2 - Processing parameters
SI                 1024
SF      600.1299831 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
PC                 1.00

F1 - Processing parameters
SI                 1024
MC2         States-TPPI
SF      600.1299804 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
```

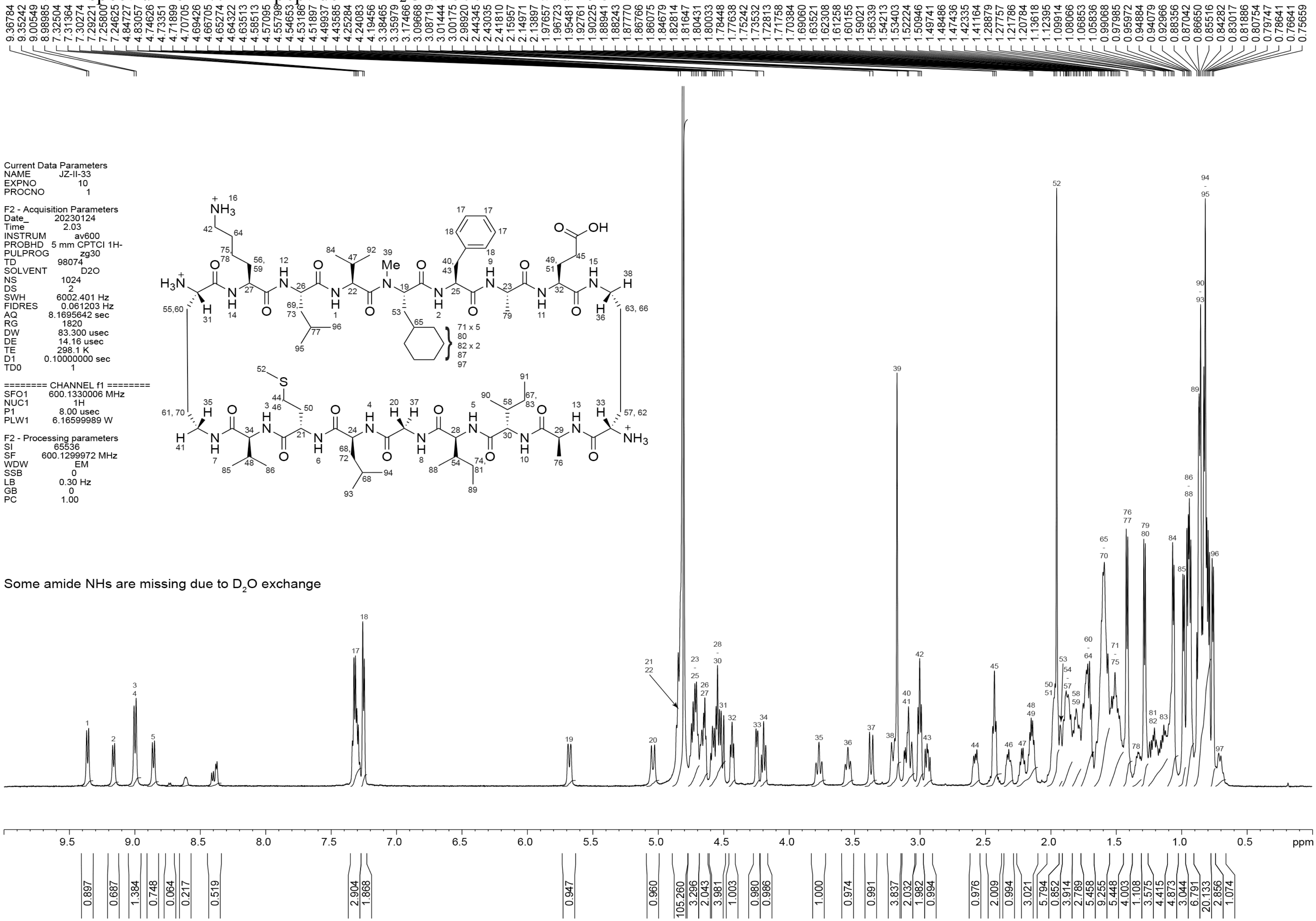
^1H NMR spectrum of 8.0 mM peptide **1** in D_2O at 600 MHz and 298 K



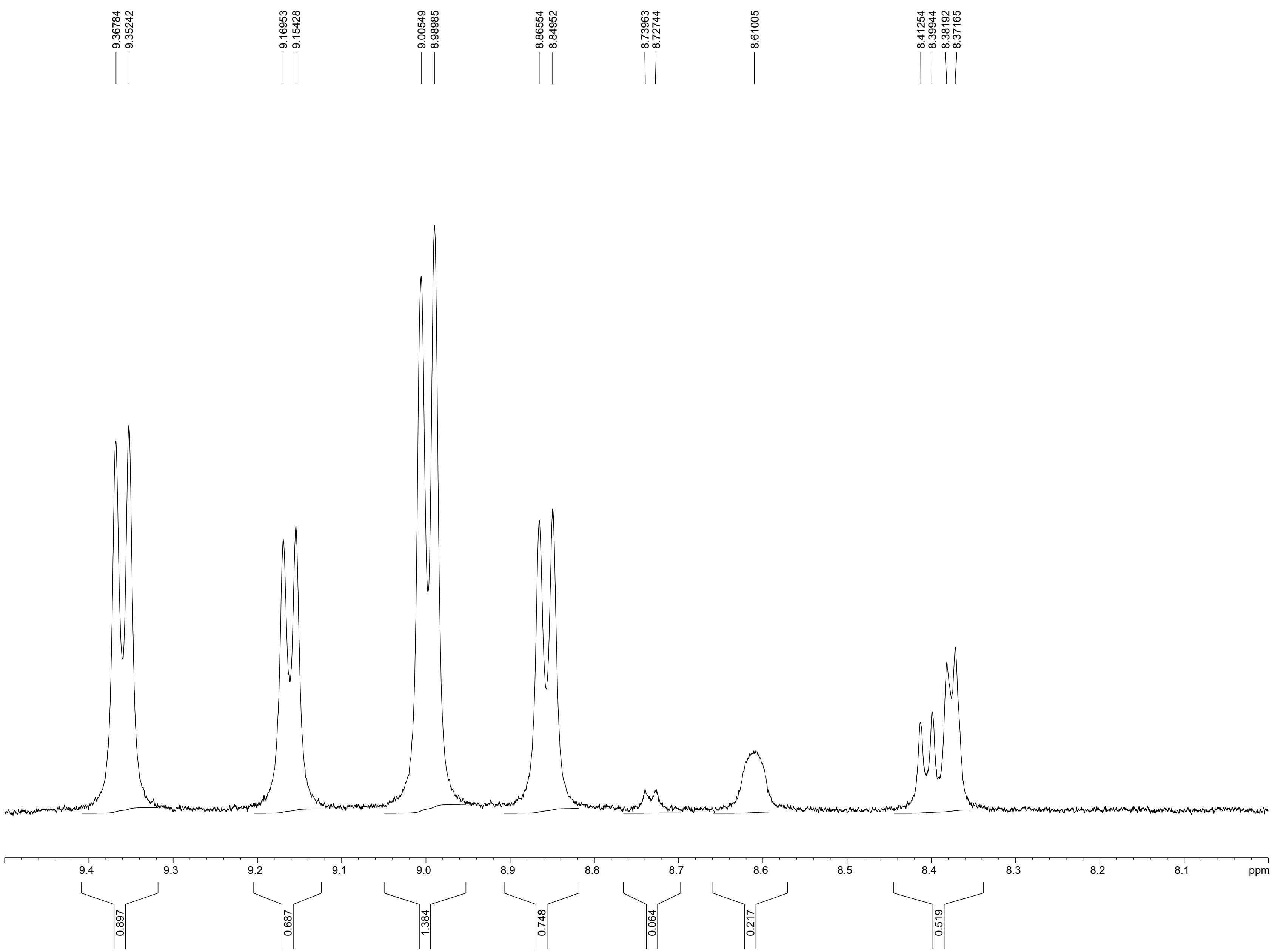
^{1}H NMR spectrum of 16.0 mM peptide **1** in D_2O at 500 MHz and 298 K



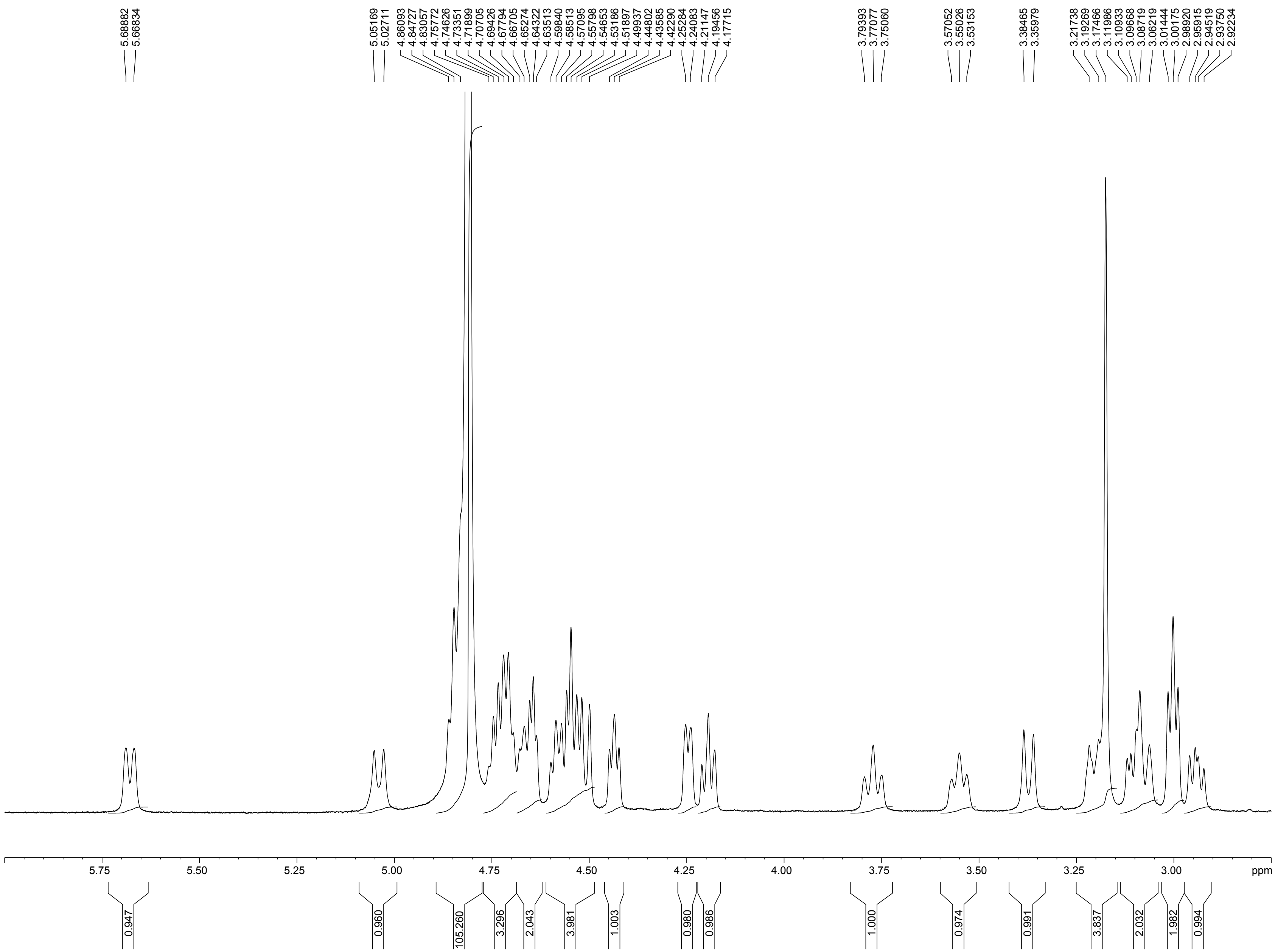
^{1}H NMR spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K



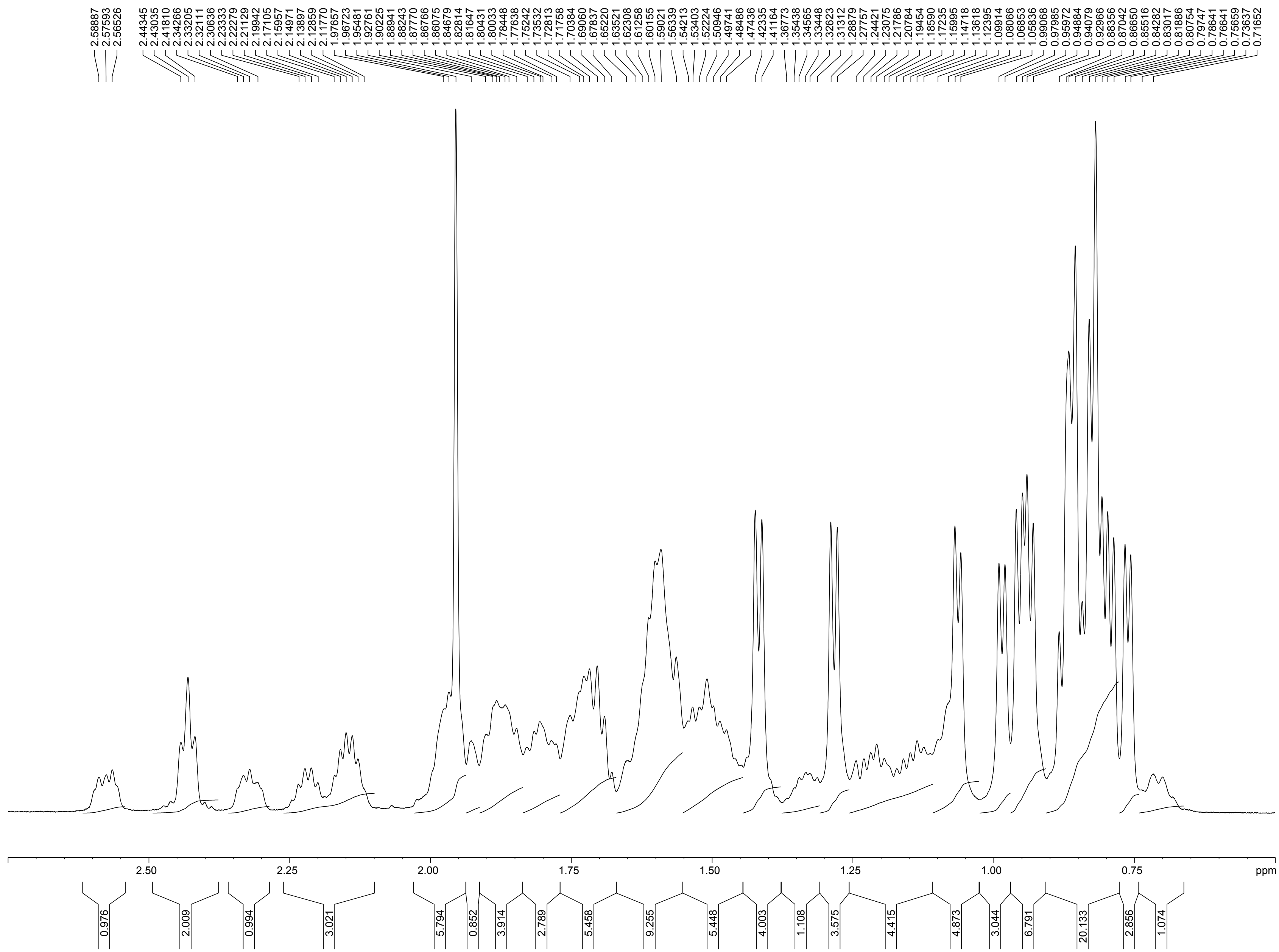
^{1}H NMR spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K



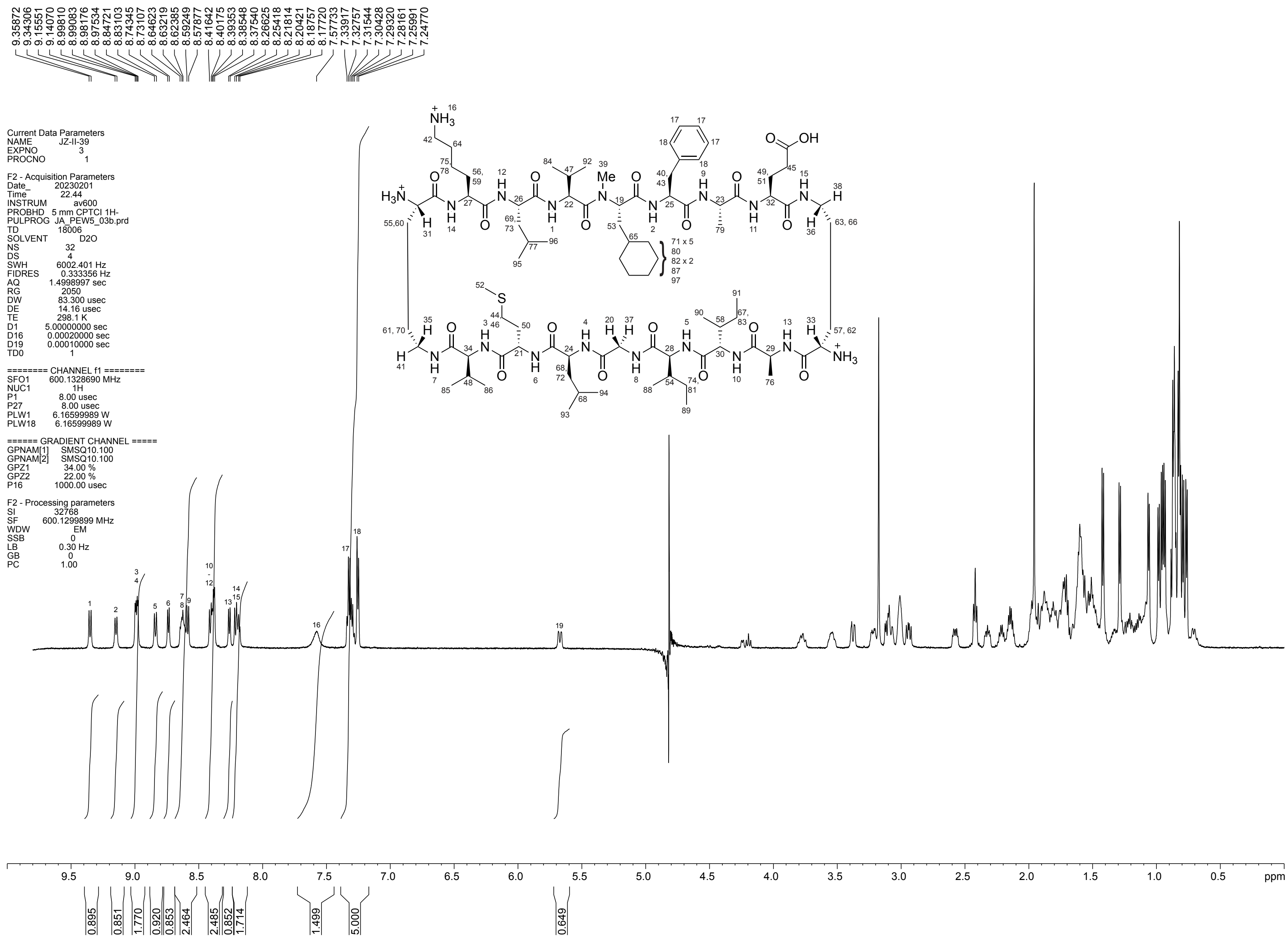
[1]H NMR spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K



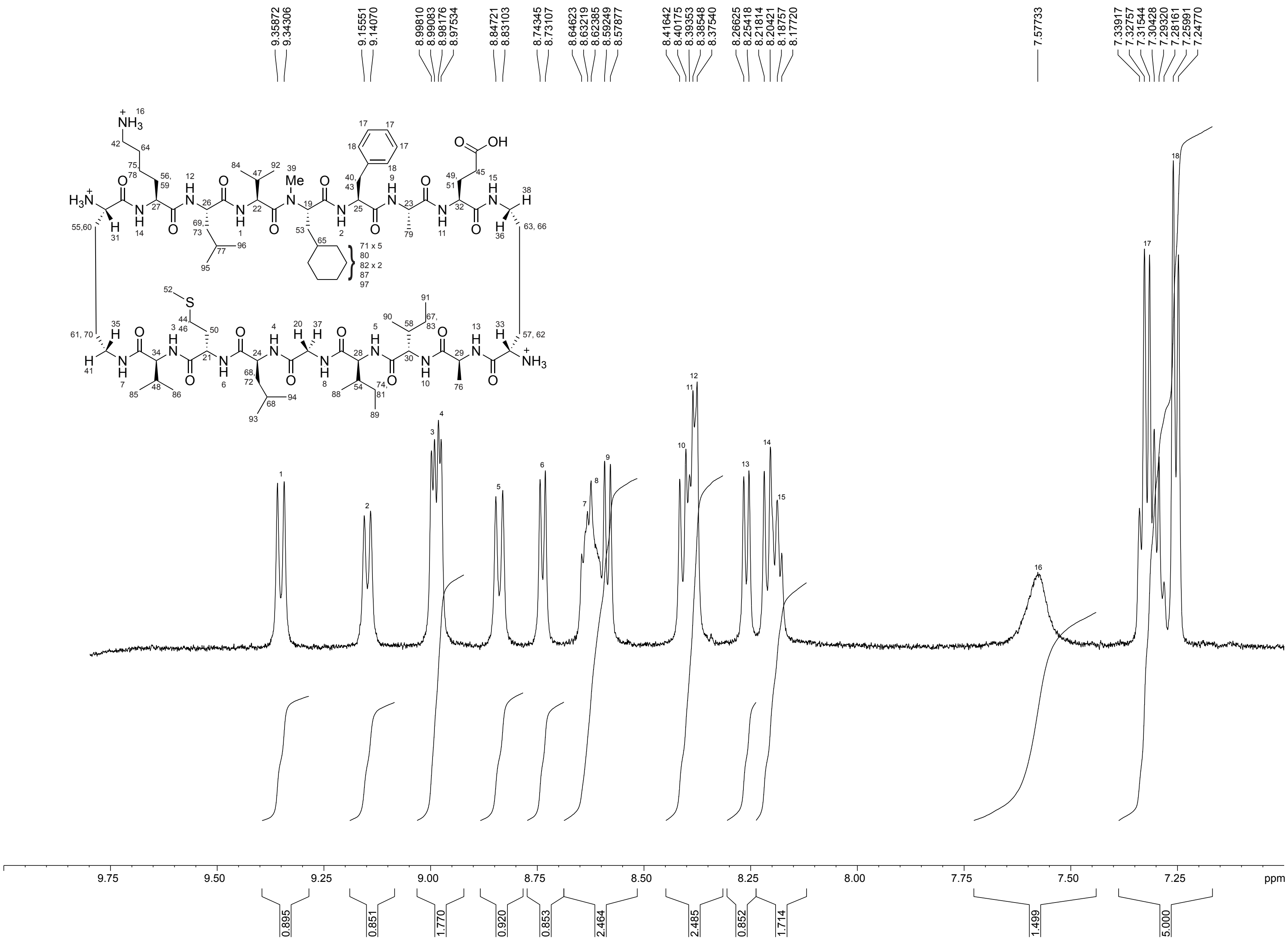
^{1}H NMR spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K



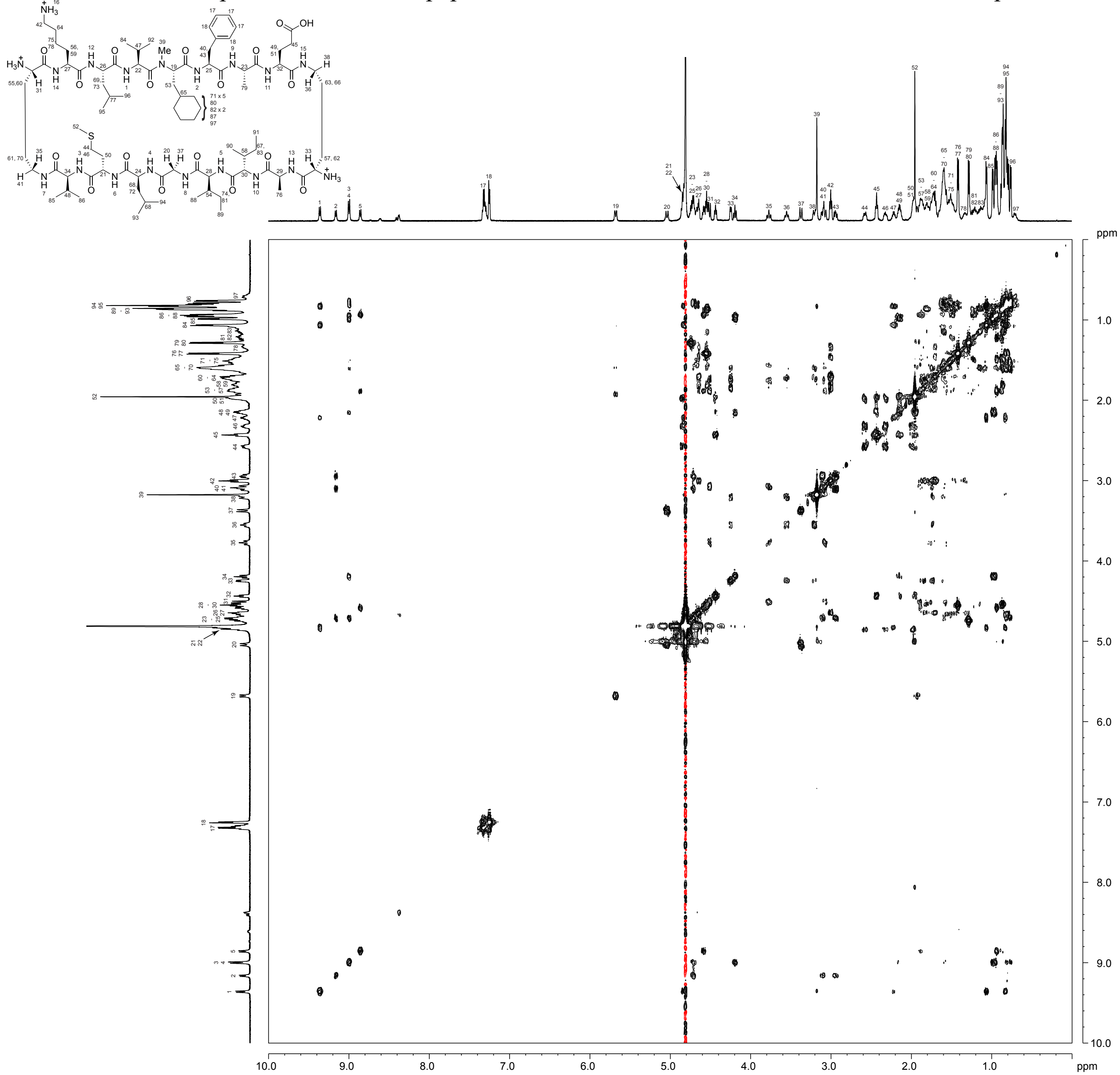
^1H NMR spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak



^{1}H NMR spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak



^{1}H NMR 2D TOCSY spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 100-ms spin-lock mixing time



Current Data Parameters
NAME JZ-II-33
EXPNO 11
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230124
Time 2.04
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG mlevgp_mo.prd
TD 2048
SOLVENT D2O
NS 16
DS 16
SWH 6009.615 Hz
FIDRES 2.934382 Hz
AQ 0.1703936 sec
RG 1820
DW 83.200 usec
DE 10.00 usec
TE 298.1 K
D0 0.00000300 sec
D1 2.00000000 sec
D9 0.10000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
IN0 0.00016640 sec
L1 60

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
PLW1 6.16599989 W
PLW10 0.63095999 W

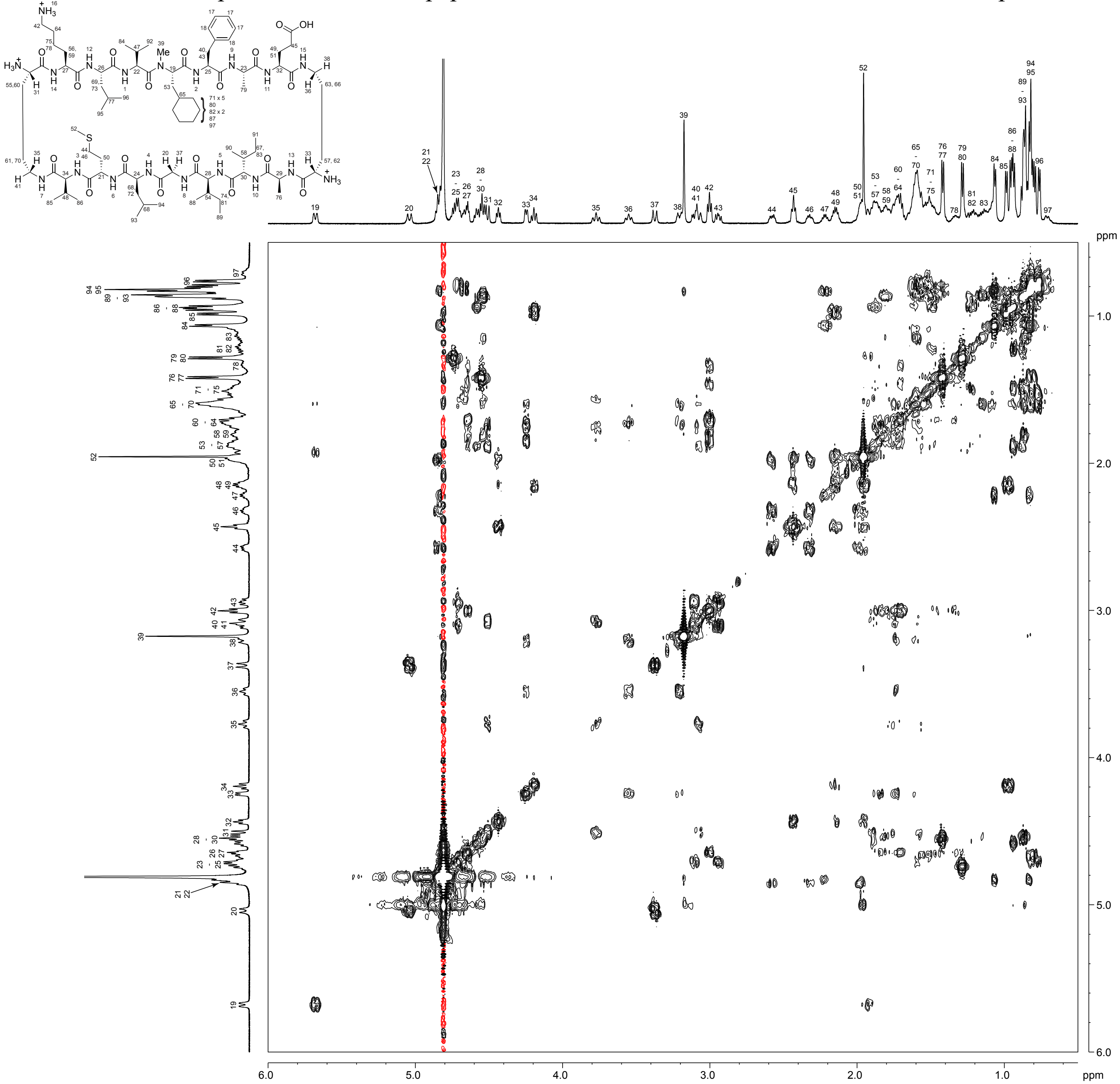
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 10.00 %
GPZ2 10.00 %
P16 1000.00 usec

F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 46.950119 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 1024
SF 600.1299967 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 QF
SF 600.1299968 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0

[1]H NMR 2D TOCSY spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 100-ms spin-lock mixing time



Current Data Parameters
NAME JZ-II-33
EXPNO 11
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230124
Time 2.04
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG mlevgp_mo.prd
TD 2048
SOLVENT D2O
NS 16
DS 16
SWH 6009.615 Hz
FIDRES 2.934382 Hz
AQ 0.1703936 sec
RG 1820
DW 83.200 usec
DE 10.00 usec
TE 298.1 K
D0 0.00000300 sec
D1 2.00000000 sec
D9 0.10000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
IN0 0.00016640 sec
L1 60

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
PLW1 6.16599989 W
PLW10 0.63095999 W

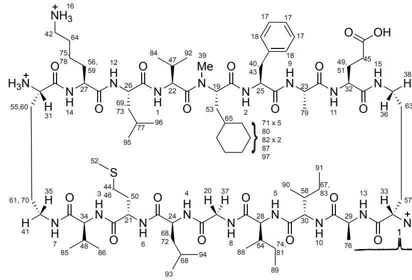
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 10.00 %
GPZ2 10.00 %
P16 1000.00 usec

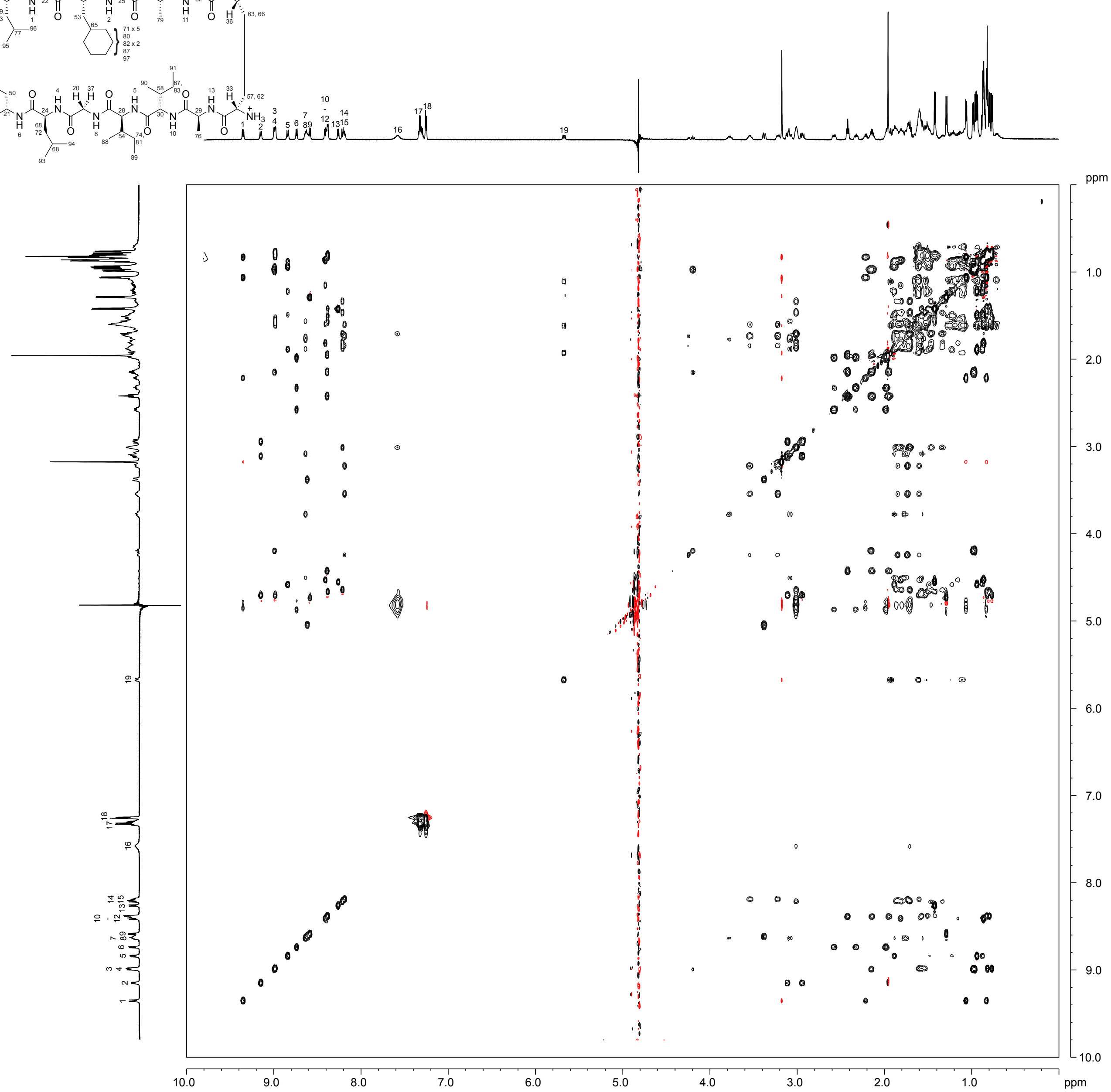
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 46.950119 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 1024
SF 600.1299967 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 QF
SF 600.1299968 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0

^1H NMR 2D TOCSY spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-ms spin-lock mixing time



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Current Data Parameters
NAME          JZ-II-39
EXPNO                5
PROCNO               1

F2 - Acquisition Parameters
Date_         20230202
Time              0.47
INSTRUM          av600
PROBHD   5 mm CPTCI 1H-
PULPROG     mlevgpphw5
TD                2048
SOLVENT            D2O
NS                  16
DS                  16
SWH        6009.615 Hz
FIDRES     2.934382 Hz
AQ       0.1703936 sec
RG                2050
DW         83.200 usec
DE          10.00 usec
TE             298.1 K
D0      0.00007411 sec
D1      2.00000000 sec
D9      0.20000000 sec
D12     0.00002000 sec
D16     0.00020000 sec
D19     0.00010000 sec
IN0     0.00016640 sec
L1                 120
P2          16.00 usec

======== CHANNEL f1 ========
SFO1    600.1328692 MHz
NUC1                1H
P1           8.00 usec
P5          16.67 usec
P6          25.00 usec
P7          50.00 usec
P17       2500.00 usec
P27          8.00 usec
PLW1      6.16599989 W
PLW10     0.63095999 W
PLW18     6.16599989 W

====== GRADIENT CHANNEL =====
GPNAM[1]    SMSQ10.100
GPNAM[2]    SMSQ10.100
GPZ1           34.00 %
GPZ2           22.00 %
P16       1000.00 usec

F1 - Acquisition parameters
TD                 256
SFO1      600.1329 MHz
FIDRES    46.950119 Hz
SW          10.014 ppm
FnMODE     States-TPPI

F2 - Processing parameters
SI                1024
SF     600.1299901 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
PC                1.00

F1 - Processing parameters
SI                1024
MC2        States-TPPI
SF     600.1299893 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
```
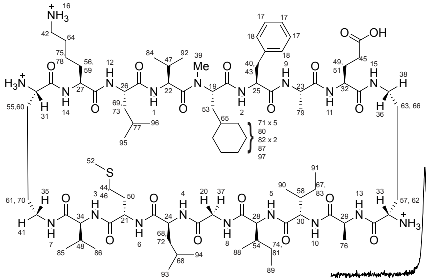
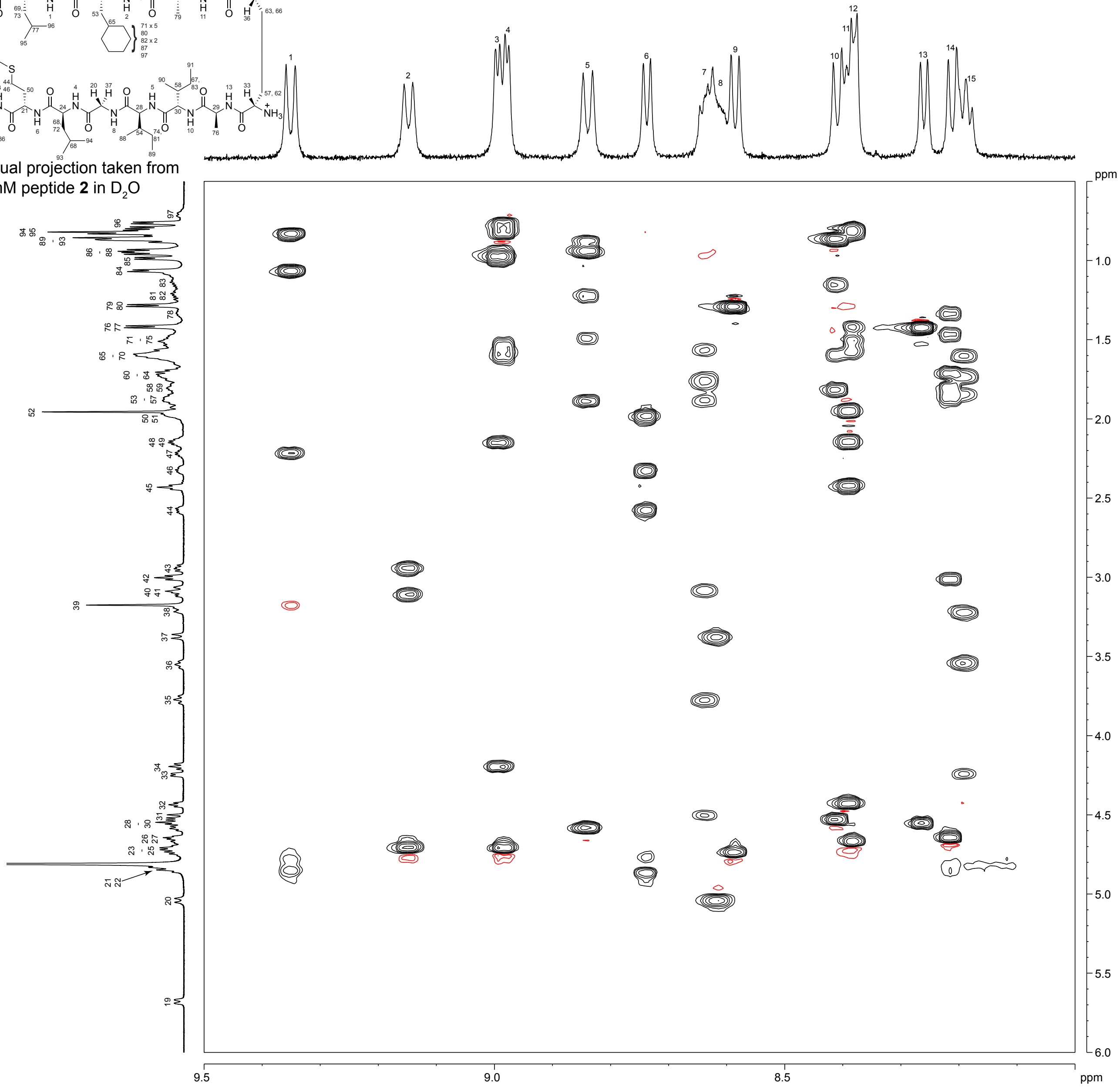

[1]H NMR 2D TOCSY spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-ms spin-lock mixing time





Current Data Parameters
NAME JZ-II-39
EXPNO 5
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230202
Time 0.47
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG mlevgpphw5
TD 2048
SOLVENT D2O
NS 16
DS 16
SWH 6009.615 Hz
FIDRES 2.934382 Hz
AQ 0.1703936 sec
RG 2050
DW 83.200 usec
DE 10.00 usec
TE 298.1 K
D0 0.00007411 sec
D1 2.00000000 sec
D9 0.20000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
D19 0.00010000 sec
IN0 0.00016640 sec
L1 120
P2 16.00 usec

======== CHANNEL f1 ========
SFO1 600.1328692 MHz
NUC1 1H
P1 8.00 usec
P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
P27 8.00 usec
PLW1 6.16599989 W
PLW10 0.63095999 W
PLW18 6.16599989 W

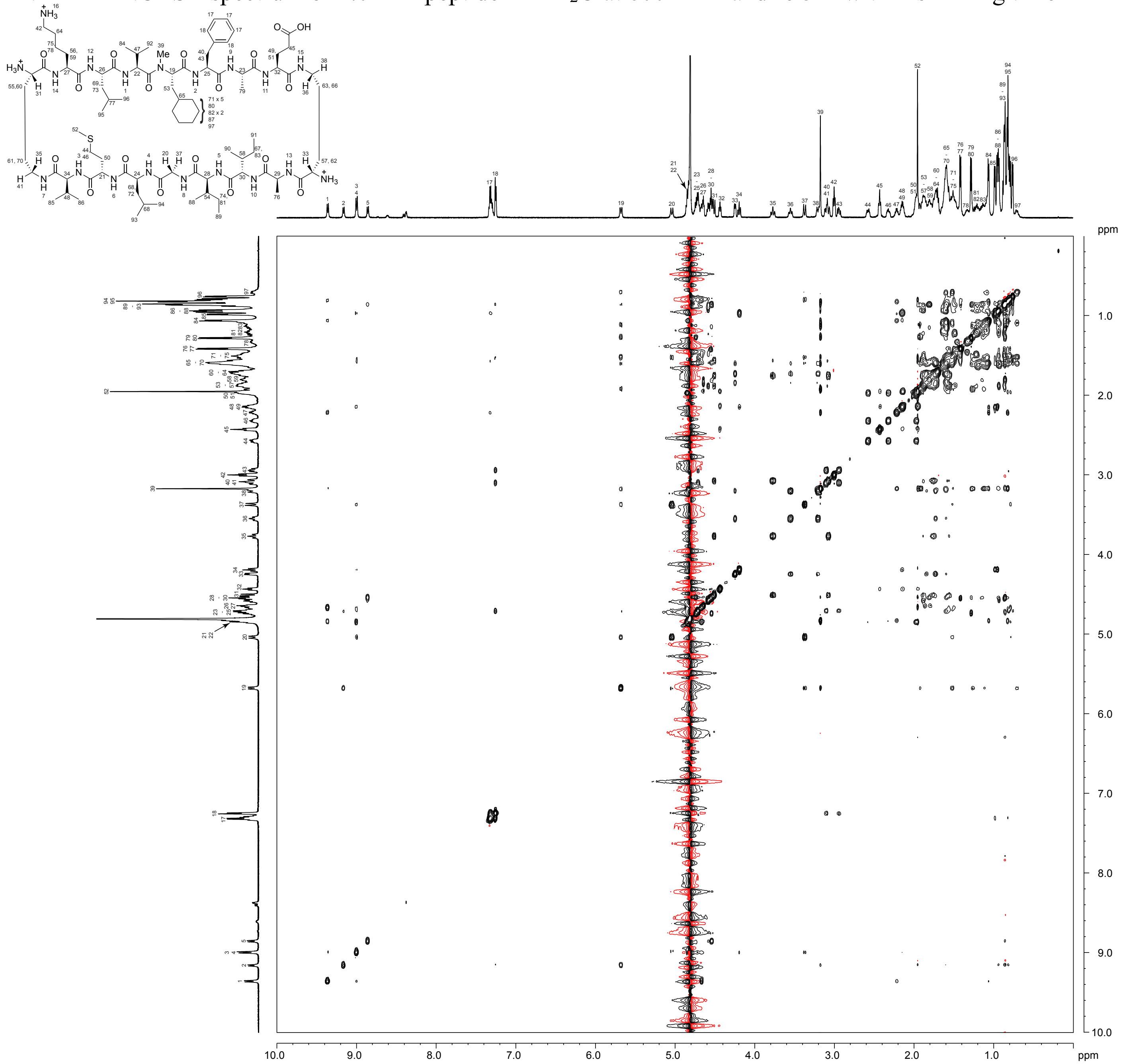
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 34.00 %
GPZ2 22.00 %
P16 1000.00 usec

F1 - Acquisition parameters
TD 256
SFO1 600.1329 MHz
FIDRES 46.950119 Hz
SW 10.014 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299901 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299893 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 1-s mixing time



Current Data Parameters
NAME JZ-II-33
EXPNO 12
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230124
Time 4.43
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG noesyphrv
TD 2048
SOLVENT D2O
NS 16
DS 16
SWH 6002.401 Hz
FIDRES 2.930860 Hz
AQ 0.1705984 sec
RG 812
DW 83.300 usec
DE 10.74 usec
TE 298.1 K
D0 0.00007311 sec
D1 2.00000000 sec
D8 1.00000000 sec
IN0 0.00016660 sec

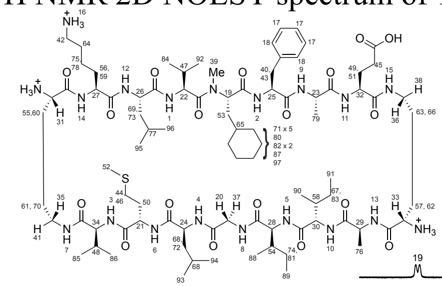
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
PLW1 6.16599989 W

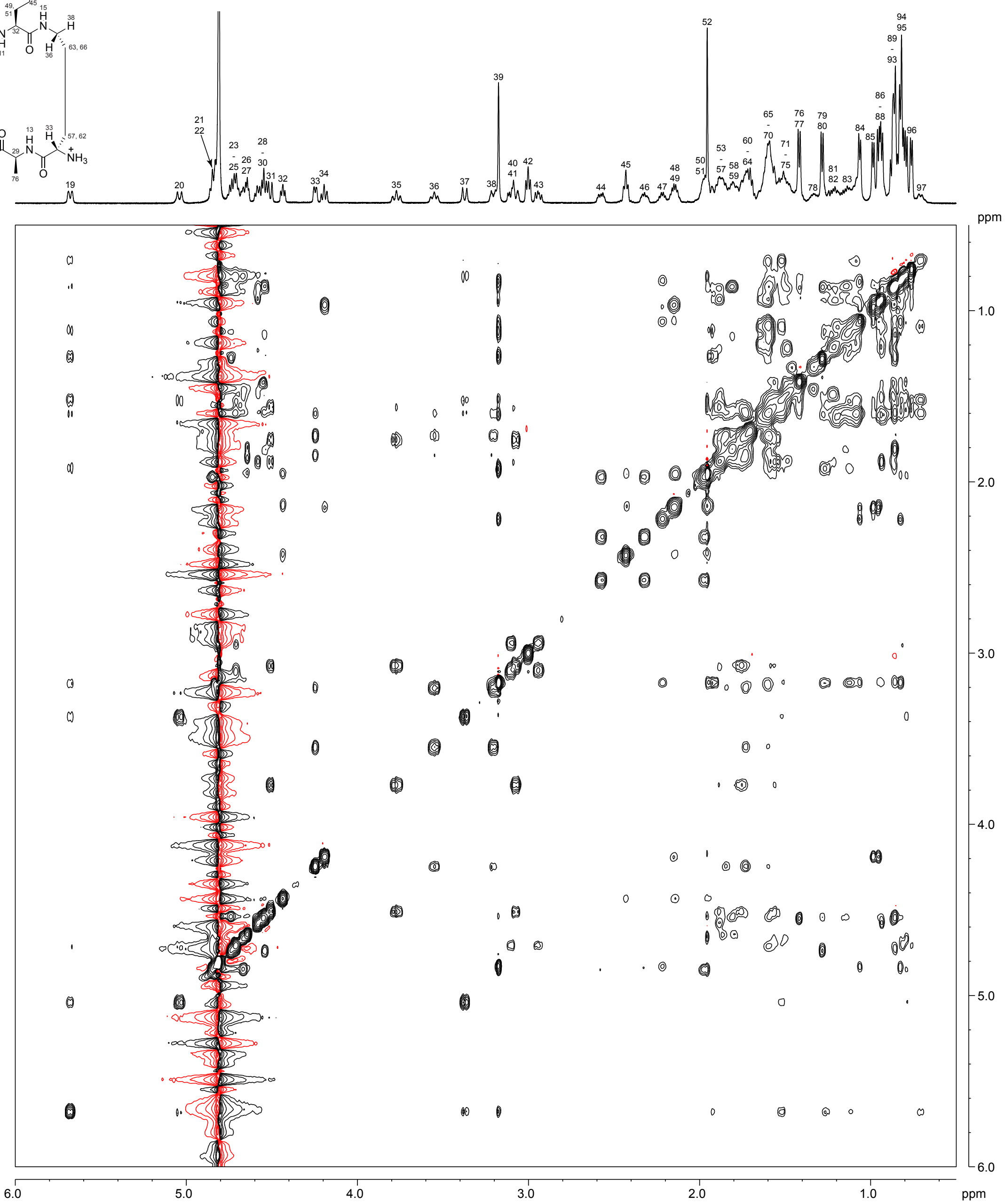
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 46.893757 Hz
SW 10.002 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299968 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299977 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

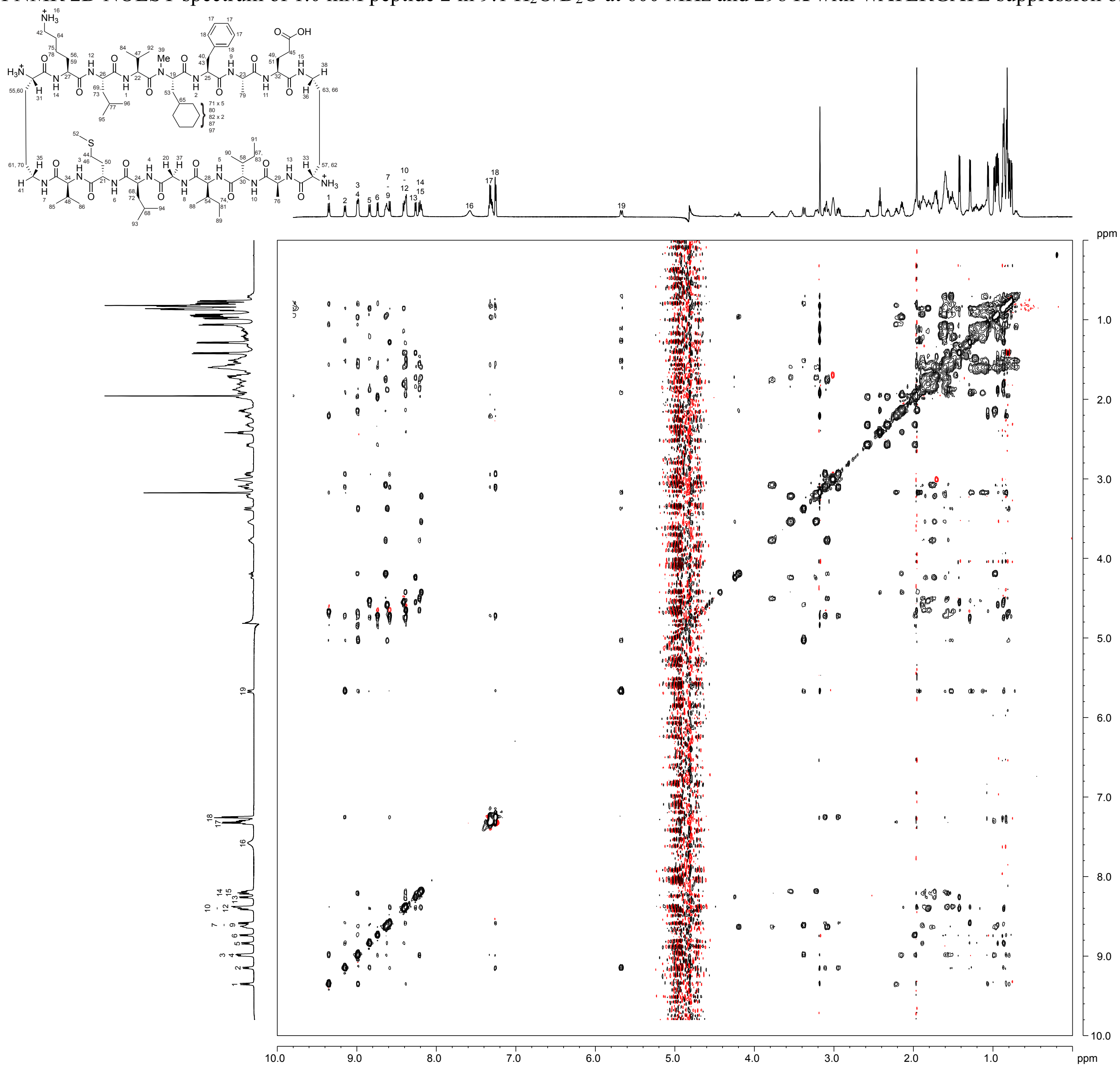
^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 1-s mixing time





Current Data Parameters	
NAME	JZ-II-33
EXPNO	12
PROCNO	1
F2 - Acquisition Parameters	
Date_	20230124
Time	4.43
INSTRUM	av600
PROBHD	5 mm CPTCI 1H-
PULPROG	noesyphrv
TD	2048
SOLVENT	D2O
NS	16
DS	16
SWH	6002.401 Hz
FIDRES	2.930860 Hz
AQ	0.1705984 sec
RG	812
DW	83.300 usec
DE	10.74 usec
TE	298.1 K
D0	0.00007311 sec
D1	2.00000000 sec
D8	1.00000000 sec
IN0	0.00016660 sec
======== CHANNEL f1 ========	
SFO1	600.1330006 MHz
NUC1	1H
P1	8.00 usec
PLW1	6.16599989 W
F1 - Acquisition parameters	
TD	256
SFO1	600.133 MHz
FIDRES	46.893757 Hz
SW	10.002 ppm
FnMODE	States-TPPI
F2 - Processing parameters	
SI	1024
SF	600.1299968 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.40
F1 - Processing parameters	
SI	1024
MC2	States-TPPI
SF	600.1299977 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time

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Current Data Parameters
NAME          JZ-II-39
EXPNO                6
PROCNO               1

F2 - Acquisition Parameters
Date_         20230202
Time              3.31
INSTRUM          av600
PROBHD   5 mm CPTCI 1H-
PULPROG     noesygpphw5
TD                2048
SOLVENT            D2O
NS                  16
DS                  16
SWH         6009.615 Hz
FIDRES      2.934382 Hz
AQ         0.1703936 sec
RG                2050
DW          83.200 usec
DE           10.00 usec
TE             298.1 K
D0        0.00007301 sec
D1        2.00000000 sec
D8        1.00000000 sec
D11       0.03000000 sec
D12       0.00002000 sec
D13       0.00000400 sec
D16       0.00020000 sec
D19       0.00010000 sec
IN0       0.00016640 sec

======== CHANNEL f1 ========
SFO1      600.1328692 MHz
NUC1                1H
P1             8.00 usec
P27            8.00 usec
PLW1        6.16599989 W
PLW18       6.16599989 W
PLW32       0.00000063 W

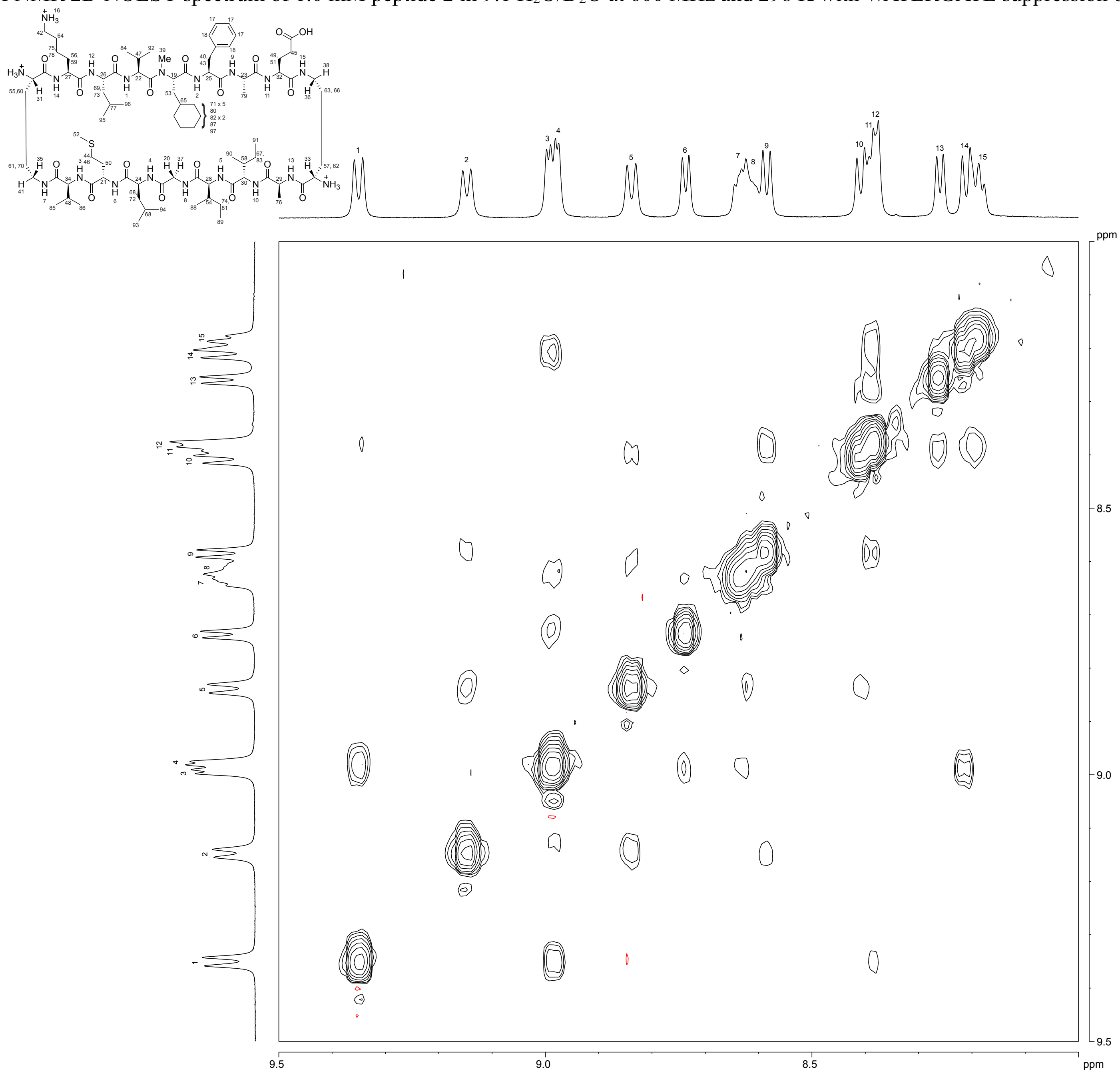
====== GRADIENT CHANNEL =====
GPNAM[1]     SMSQ10.100
GPNAM[2]     SMSQ10.100
GPZ1            34.00 %
GPZ2            22.00 %
P16         1000.00 usec

F1 - Acquisition parameters
TD                 256
SFO1       600.1329 MHz
FIDRES     46.950119 Hz
SW          10.014 ppm
FnMODE      States-TPPI

F2 - Processing parameters
SI                1024
SF      600.1299892 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
PC                1.00

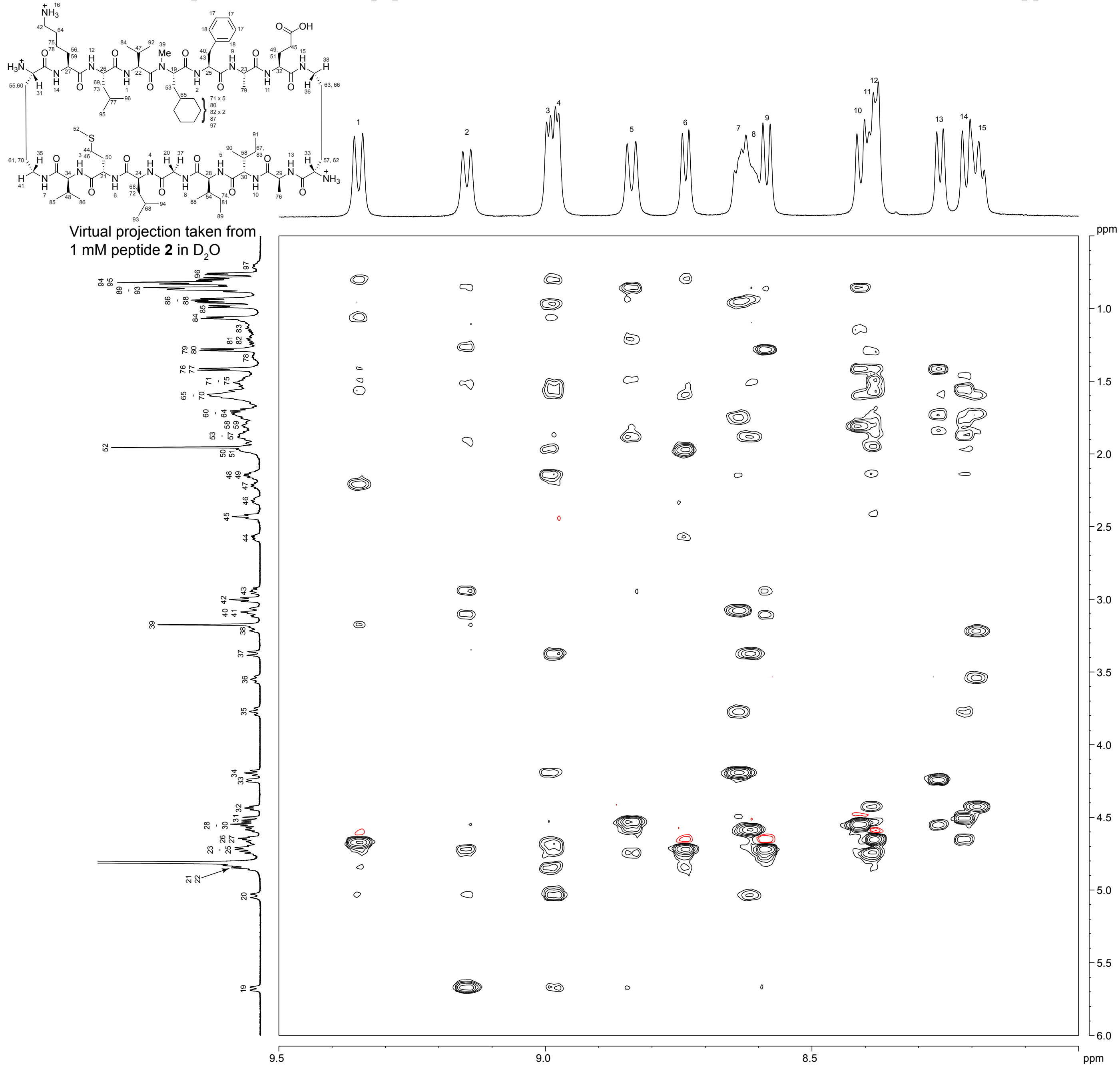
F1 - Processing parameters
SI                1024
MC2         States-TPPI
SF      600.1299908 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
```

^{1}H NMR 2D NOESY spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time



Current Data Parameters	
NAME	JZ-II-39
EXPNO	6
PROCNO	1
F2 - Acquisition Parameters	
Date_	20230202
Time	3.31
INSTRUM	av600
PROBHD	5 mm CPTCI 1H-
PULPROG	noesygpphw5
TD	2048
SOLVENT	D2O
NS	16
DS	16
SWH	6009.615 Hz
FIDRES	2.934382 Hz
AQ	0.1703936 sec
RG	2050
DW	83.200 usec
DE	10.00 usec
TE	298.1 K
D0	0.00007301 sec
D1	2.00000000 sec
D8	1.00000000 sec
D11	0.03000000 sec
D12	0.00002000 sec
D13	0.00000400 sec
D16	0.00020000 sec
D19	0.00010000 sec
IN0	0.00016640 sec
======== CHANNEL f1 ========	
SFO1	600.1328692 MHz
NUC1	1H
P1	8.00 usec
P27	8.00 usec
PLW1	6.16599989 W
PLW18	6.16599989 W
PLW32	0.00000063 W
====== GRADIENT CHANNEL =====	
GPNAM[1]	SMSQ10.100
GPNAM[2]	SMSQ10.100
GPZ1	34.00 %
GPZ2	22.00 %
P16	1000.00 usec
F1 - Acquisition parameters	
TD	256
SFO1	600.1329 MHz
FIDRES	46.950119 Hz
SW	10.014 ppm
FnMODE	States-TPPI
F2 - Processing parameters	
SI	1024
SF	600.1299892 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.00
F1 - Processing parameters	
SI	1024
MC2	States-TPPI
SF	600.1299908 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

[1]H NMR 2D NOESY spectrum of 1.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 1-s mixing time



Current Data Parameters

NAME	JZ-II-39
EXPNO	6
PROCNO	1

F2 - Acquisition Parameters

Date_	20230202
Time	3.31
INSTRUM	av600
PROBHD	5 mm CPTCI 1H-
PULPROG	noesygpphw5
TD	2048
SOLVENT	D2O
NS	16
DS	16
SWH	6009.615 Hz
FIDRES	2.934382 Hz
AQ	0.1703936 sec
RG	2050
DW	83.200 usec
DE	10.00 usec
TE	298.1 K
D0	0.00007301 sec
D1	2.00000000 sec
D8	1.00000000 sec
D11	0.03000000 sec
D12	0.00002000 sec
D13	0.00000400 sec
D16	0.00020000 sec
D19	0.00010000 sec
IN0	0.00016640 sec

======== CHANNEL f1 ========

SFO1	600.1328692 MHz
NUC1	1H
P1	8.00 usec
P27	8.00 usec
PLW1	6.16599989 W
PLW18	6.16599989 W
PLW32	0.00000063 W

====== GRADIENT CHANNEL =====

GPNAM[1]	SMSQ10.100
GPNAM[2]	SMSQ10.100
GPZ1	34.00 %
GPZ2	22.00 %
P16	1000.00 usec

F1 - Acquisition parameters

TD	256
SFO1	600.1329 MHz
FIDRES	46.950119 Hz
SW	10.014 ppm
FnMODE	States-TPPI

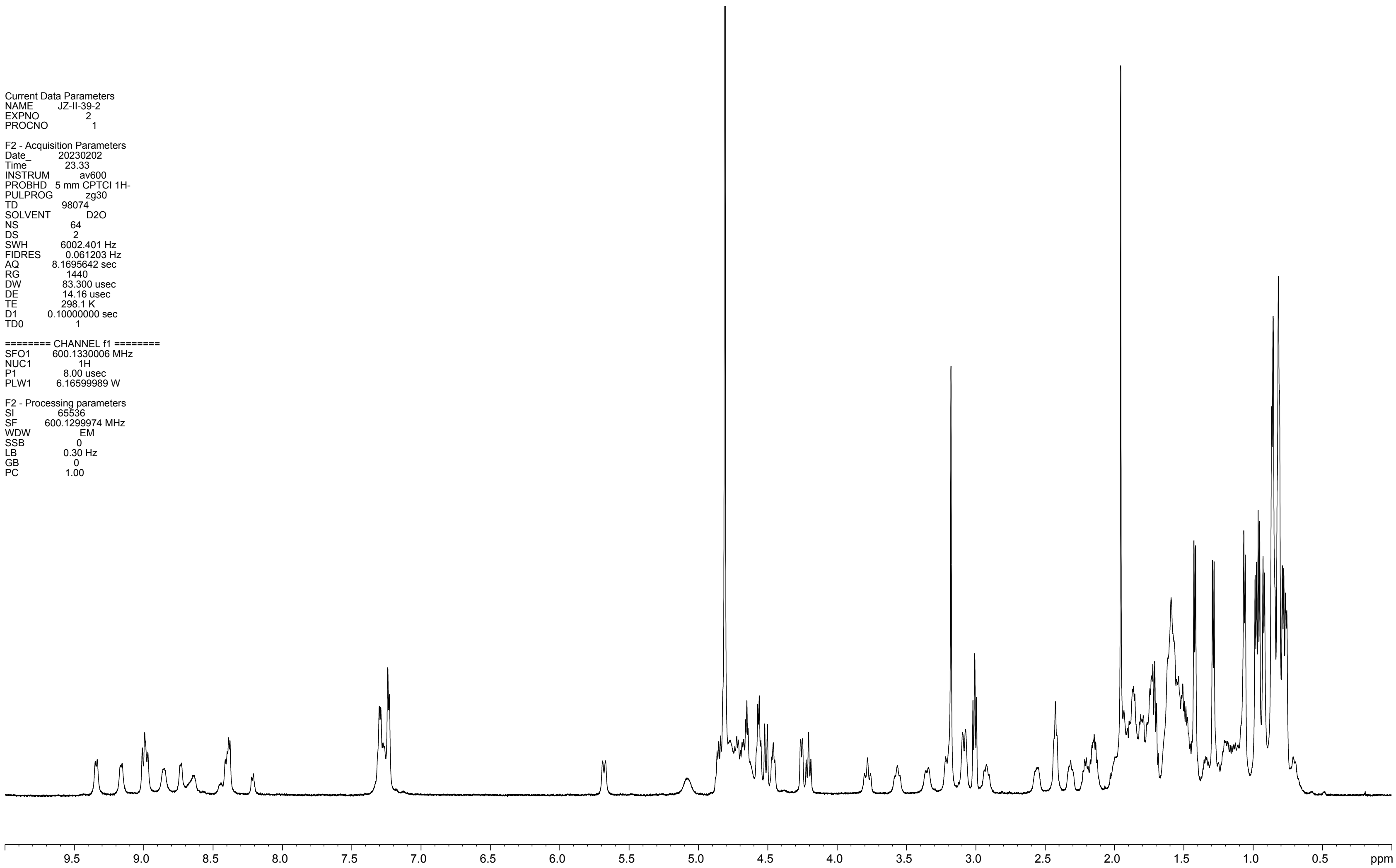
F2 - Processing parameters

SI	1024
SF	600.1299892 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.00

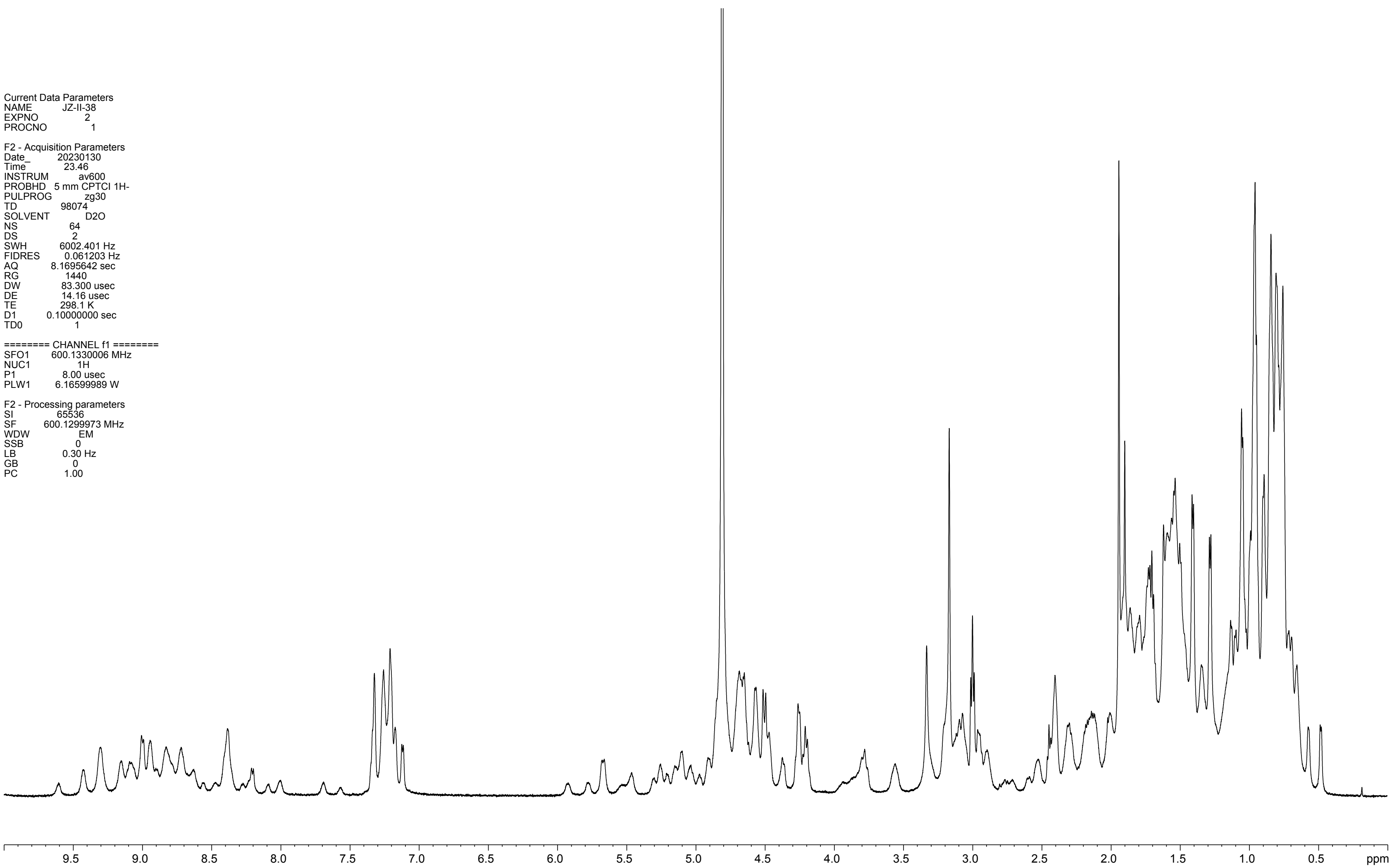
F1 - Processing parameters

SI	1024
MC2	States-TPPI
SF	600.1299908 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

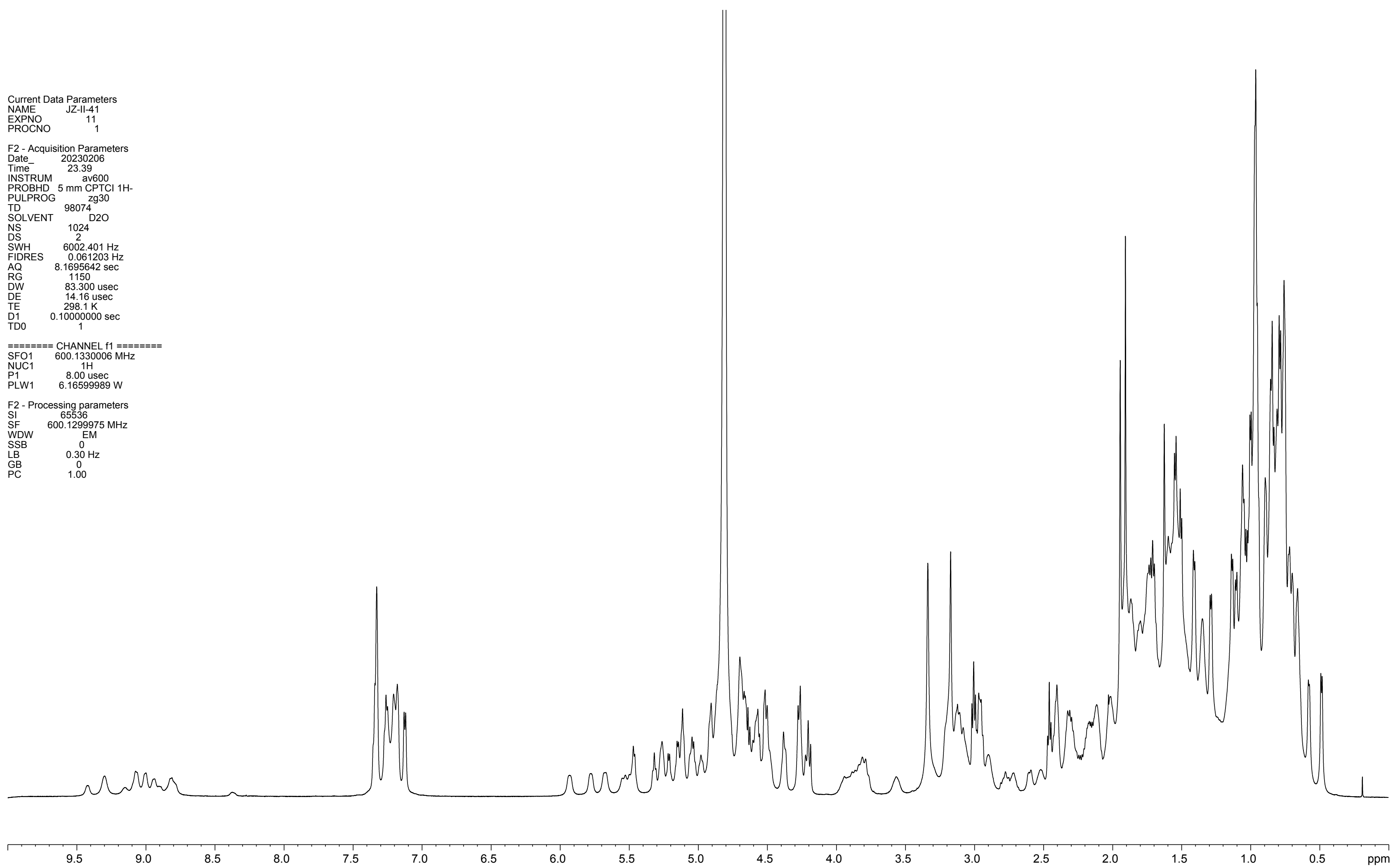
^{1}H NMR spectrum of 2.0 mM peptide **2** in D_2O at 600 MHz and 298 K



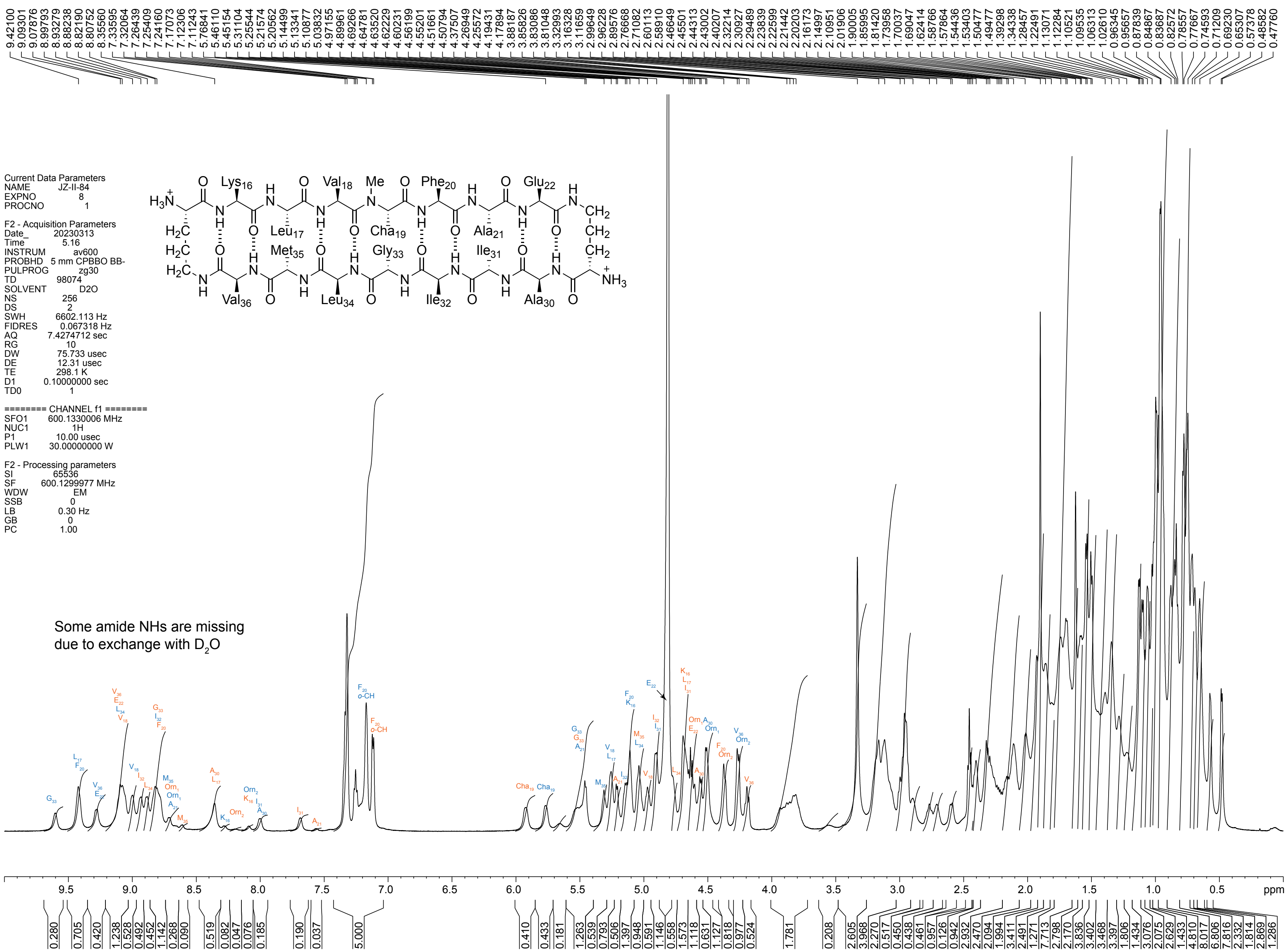
[1]H NMR spectrum of 4.0 mM peptide **2** in D_2O at 600 MHz and 298 K



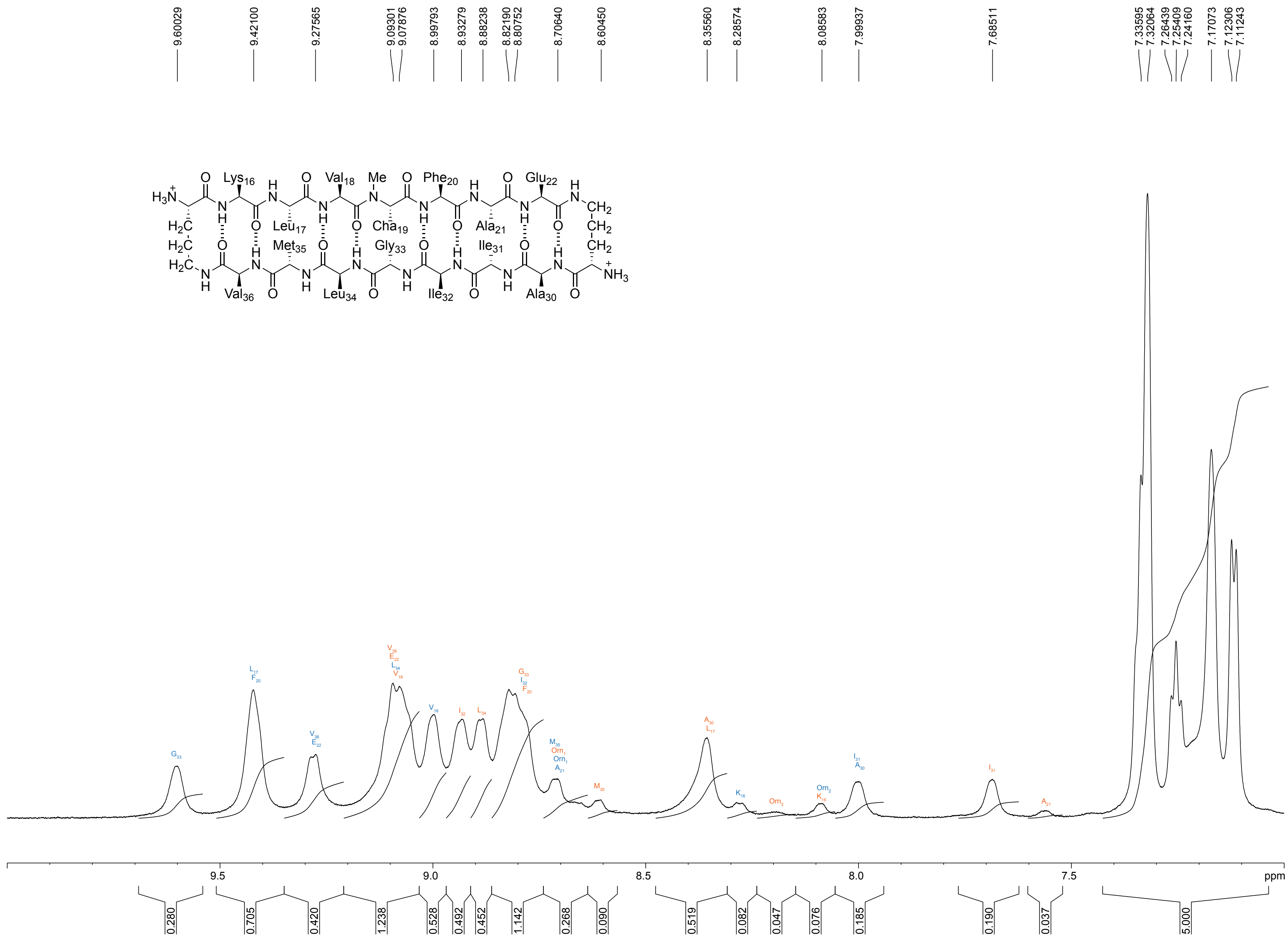
^{1}H NMR spectrum of 6.0 mM peptide **2** in D_2O at 600 MHz and 298 K



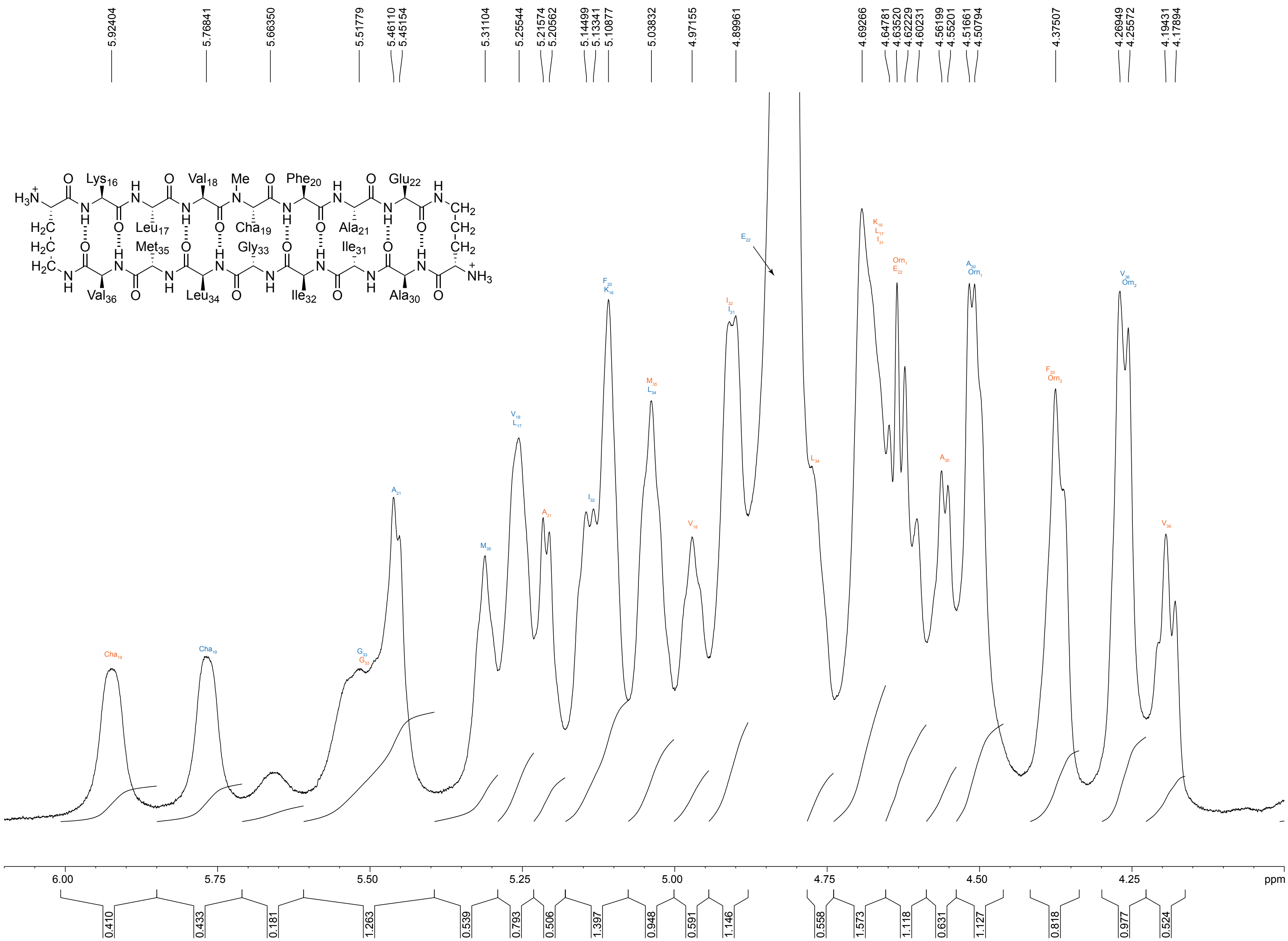
^{1}H NMR spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K



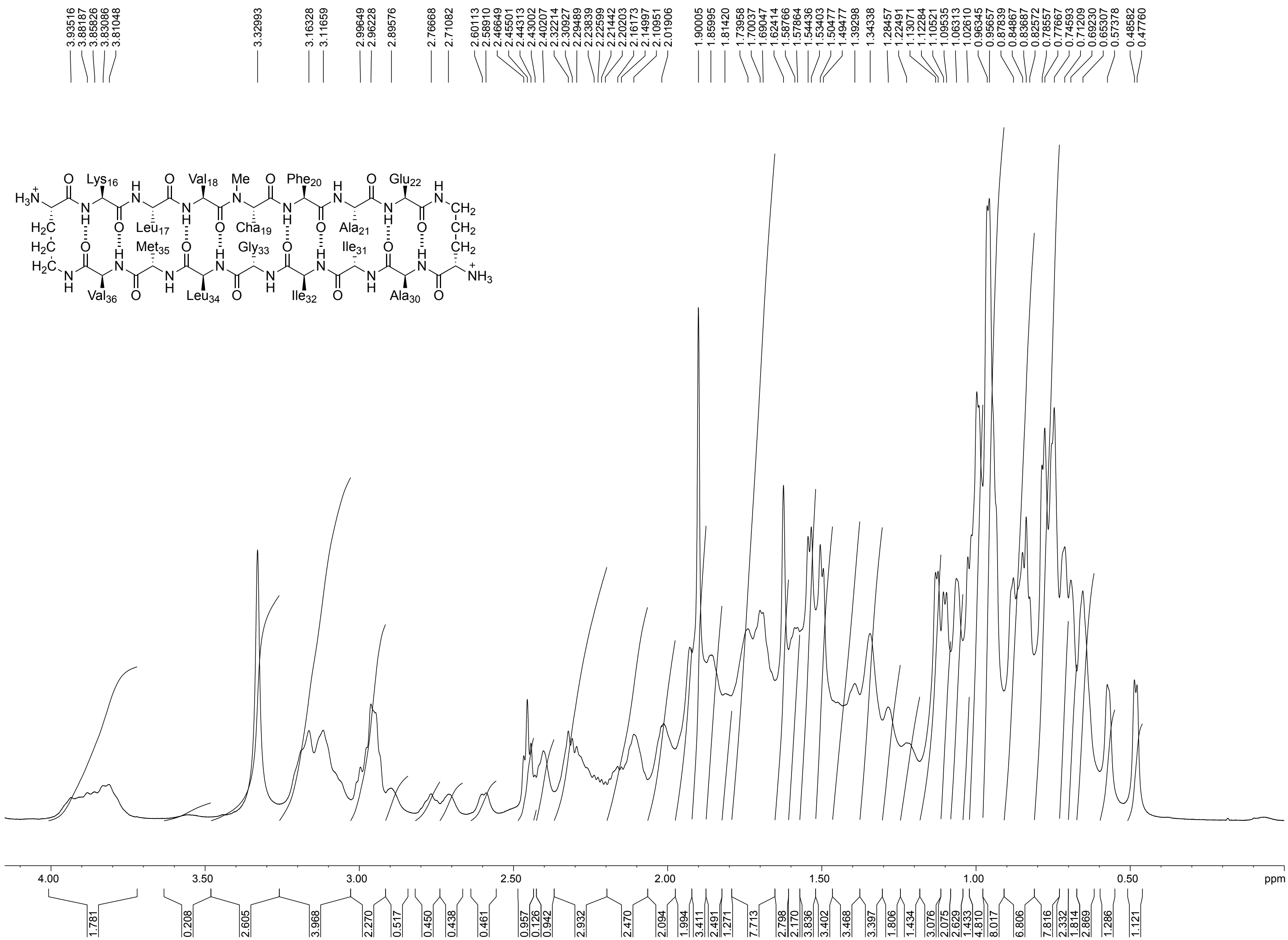
^{1}H NMR spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K



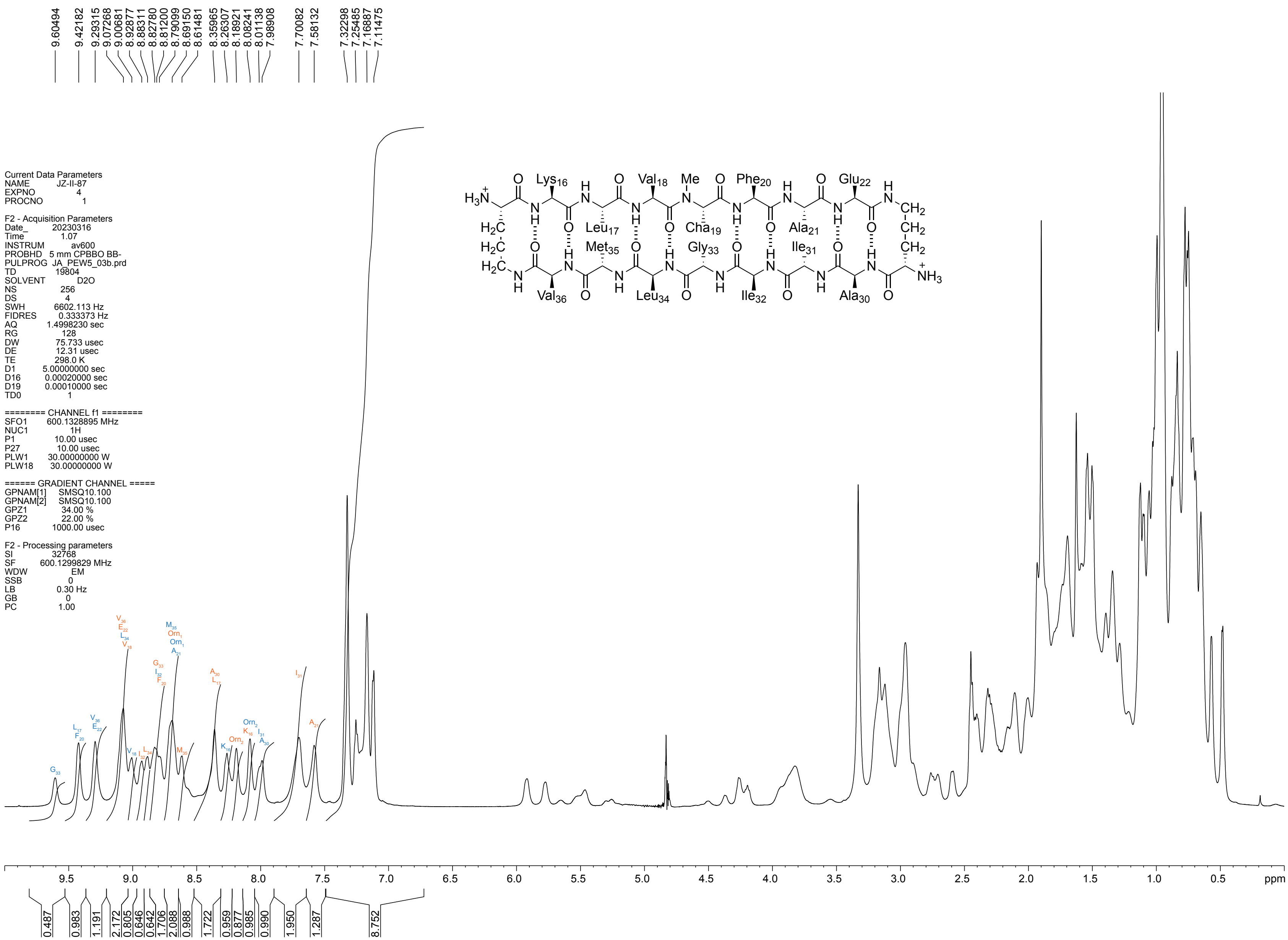
^1H NMR spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K



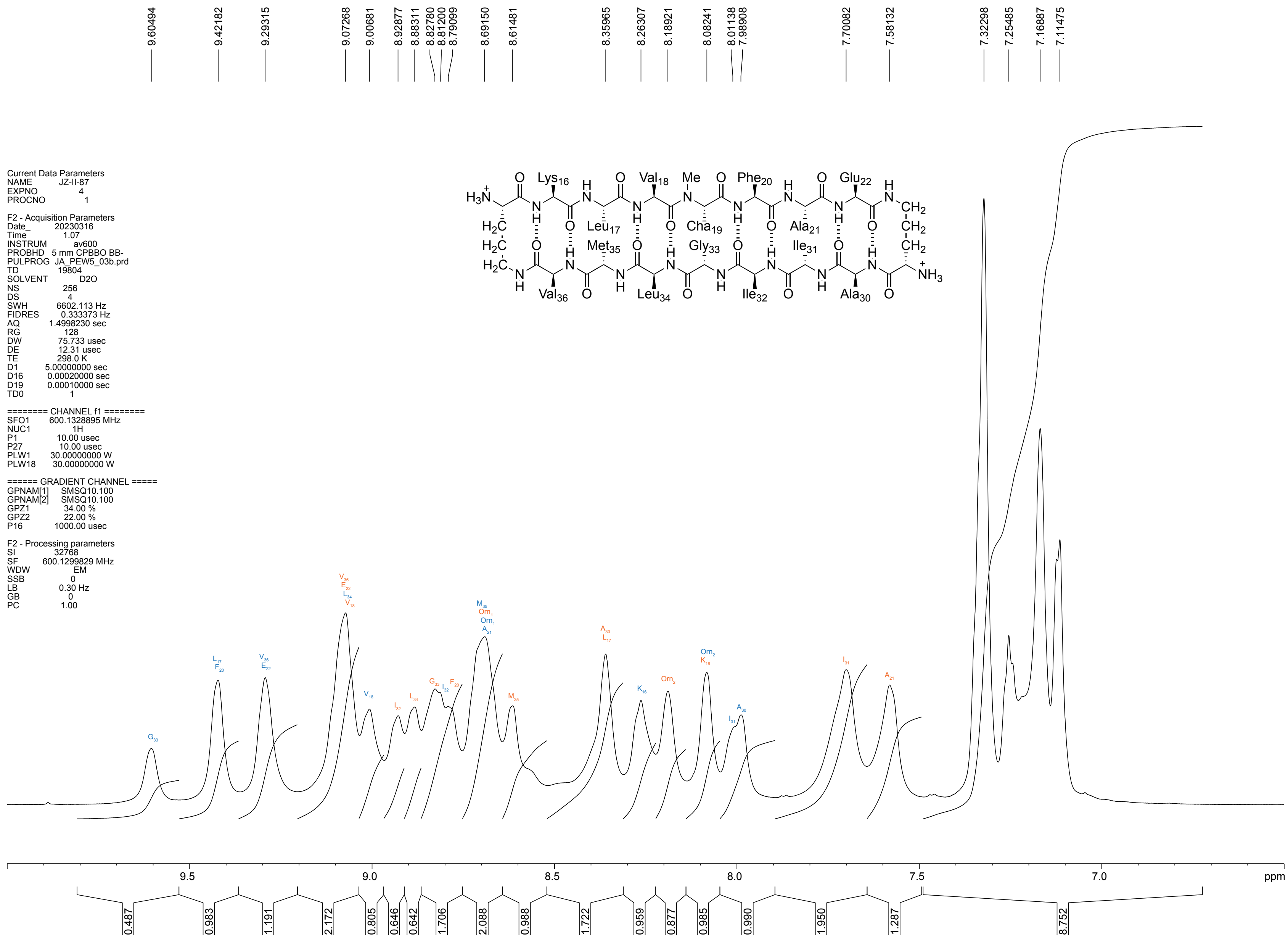
[1]H NMR spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K



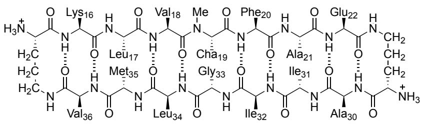
[1]H NMR spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak

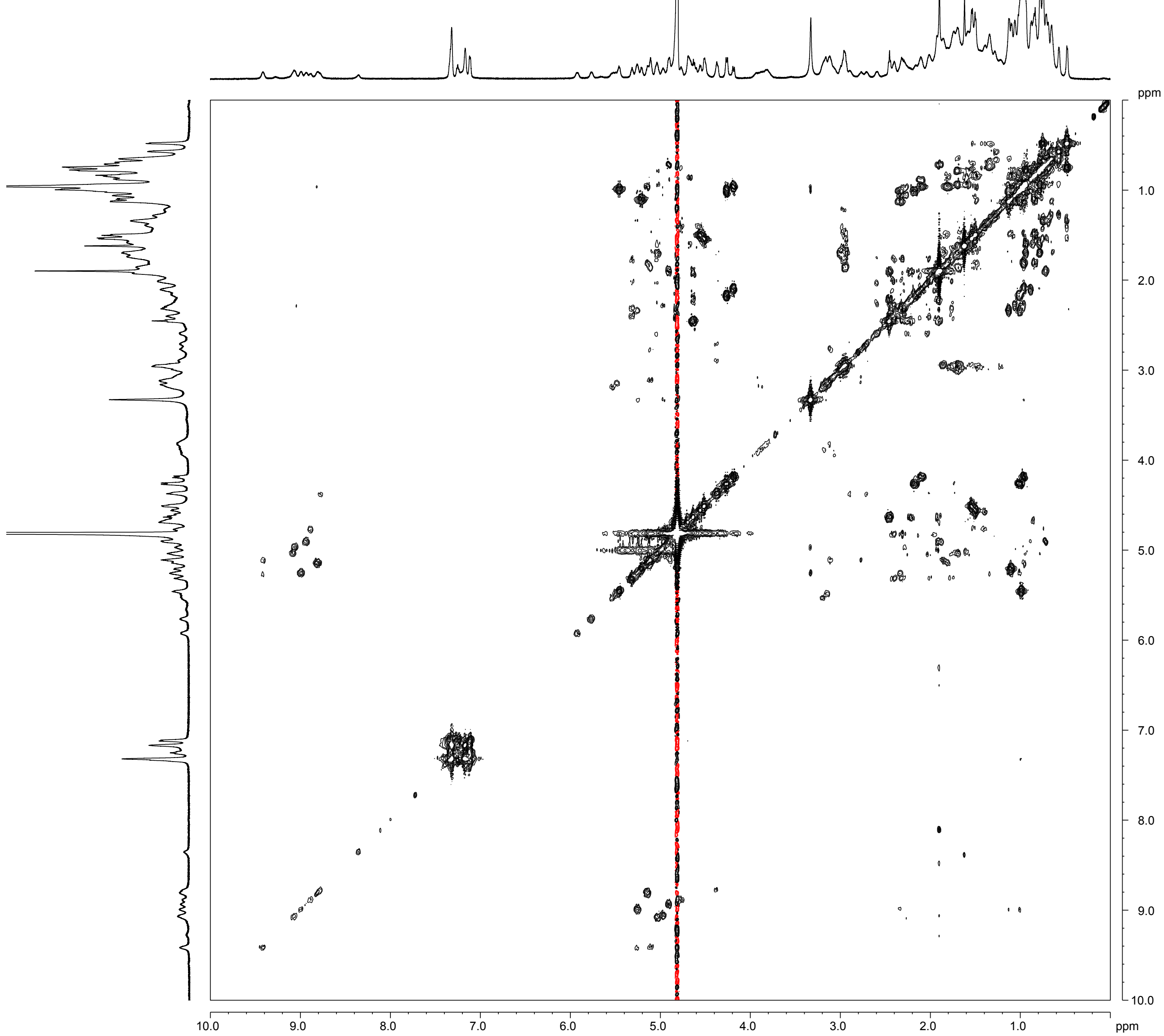


[1]H NMR spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak



^{1}H NMR 2D TOCSY spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 50-ms spin-lock mixing time





Current Data Parameters
NAME JZ-II-84
EXPNO 15
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230321
Time 3.02
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG mlevgp_mo.prd
TD 2048
SOLVENT D2O
NS 8
DS 16
SWH 6602.113 Hz
FIDRES 3.223688 Hz
AQ 0.1551019 sec
RG 10
DW 75.733 usec
DE 10.00 usec
TE 298.0 K
D0 0.00000300 sec
D1 2.00000000 sec
D9 0.05000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
IN0 0.00015140 sec
L1 30

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
PLW1 30.00000000 W
PLW10 5.15000010 W

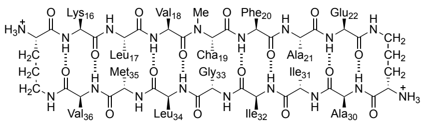
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 10.00 %
GPZ2 10.00 %
P16 1000.00 usec

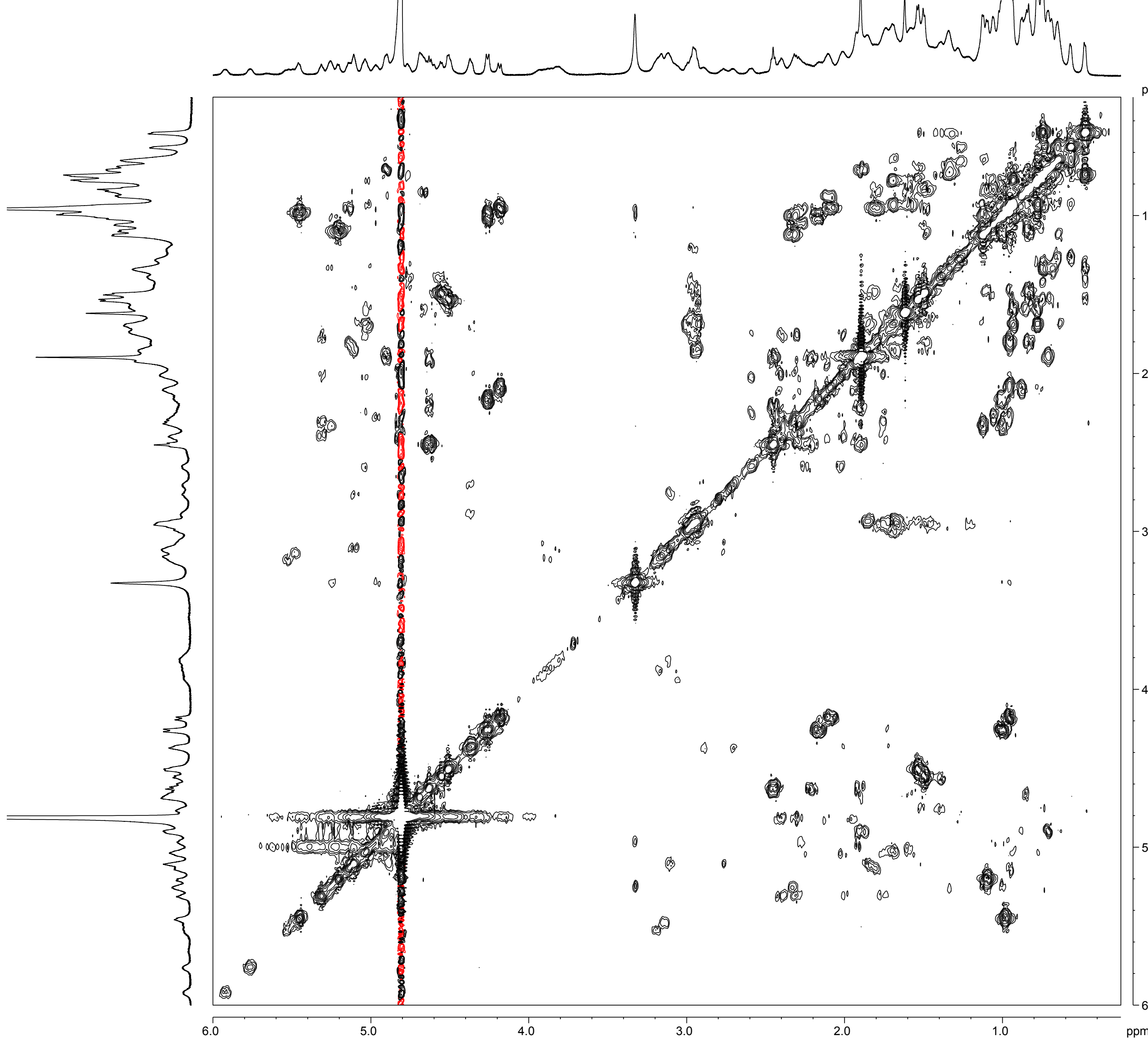
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE QF

F2 - Processing parameters
SI 1024
SF 600.1299980 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 QF
SF 600.1299978 MHz
WDW SINE
SSB 0
LB 0 Hz
GB 0

^{1}H NMR 2D TOCSY spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 50-ms spin-lock mixing time



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Current Data Parameters
NAME            JZ-II-84
EXPNO                 15
PROCNO                 1

F2 - Acquisition Parameters
Date_           20230321
Time                3.02
INSTRUM            av600
PROBHD   5 mm CPBBO BB-
PULPROG    mlevgp_mo.prd
TD                  2048
SOLVENT              D2O
NS                     8
DS                    16
SWH           6602.113 Hz
FIDRES        3.223688 Hz
AQ          0.1551019 sec
RG                    10
DW            75.733 usec
DE             10.00 usec
TE               298.0 K
D0         0.00000300 sec
D1         2.00000000 sec
D9         0.05000000 sec
D12        0.00002000 sec
D16        0.00020000 sec
IN0        0.00015140 sec
L1                    30

======== CHANNEL f1 ========
SFO1       600.1330006 MHz
NUC1                  1H
P1             10.00 usec
P5             16.67 usec
P6             25.00 usec
P7             50.00 usec
P17          2500.00 usec
PLW1       30.00000000 W
PLW10       5.15000010 W

====== GRADIENT CHANNEL =====
GPNAM[1]      SMSQ10.100
GPNAM[2]      SMSQ10.100
GPZ1             10.00 %
GPZ2             10.00 %
P16          1000.00 usec

F1 - Acquisition parameters
TD                   256
SFO1         600.133 MHz
FIDRES      51.601719 Hz
SW            11.006 ppm
FnMODE                QF

F2 - Processing parameters
SI                  1024
SF        600.1299980 MHz
WDW                 SINE
SSB                    0
LB                  0 Hz
GB                     0
PC                  1.40

F1 - Processing parameters
SI                  1024
MC2                   QF
SF        600.1299978 MHz
WDW                 SINE
SSB                    0
LB                  0 Hz
GB                     0
```
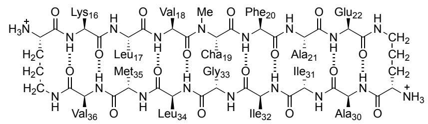
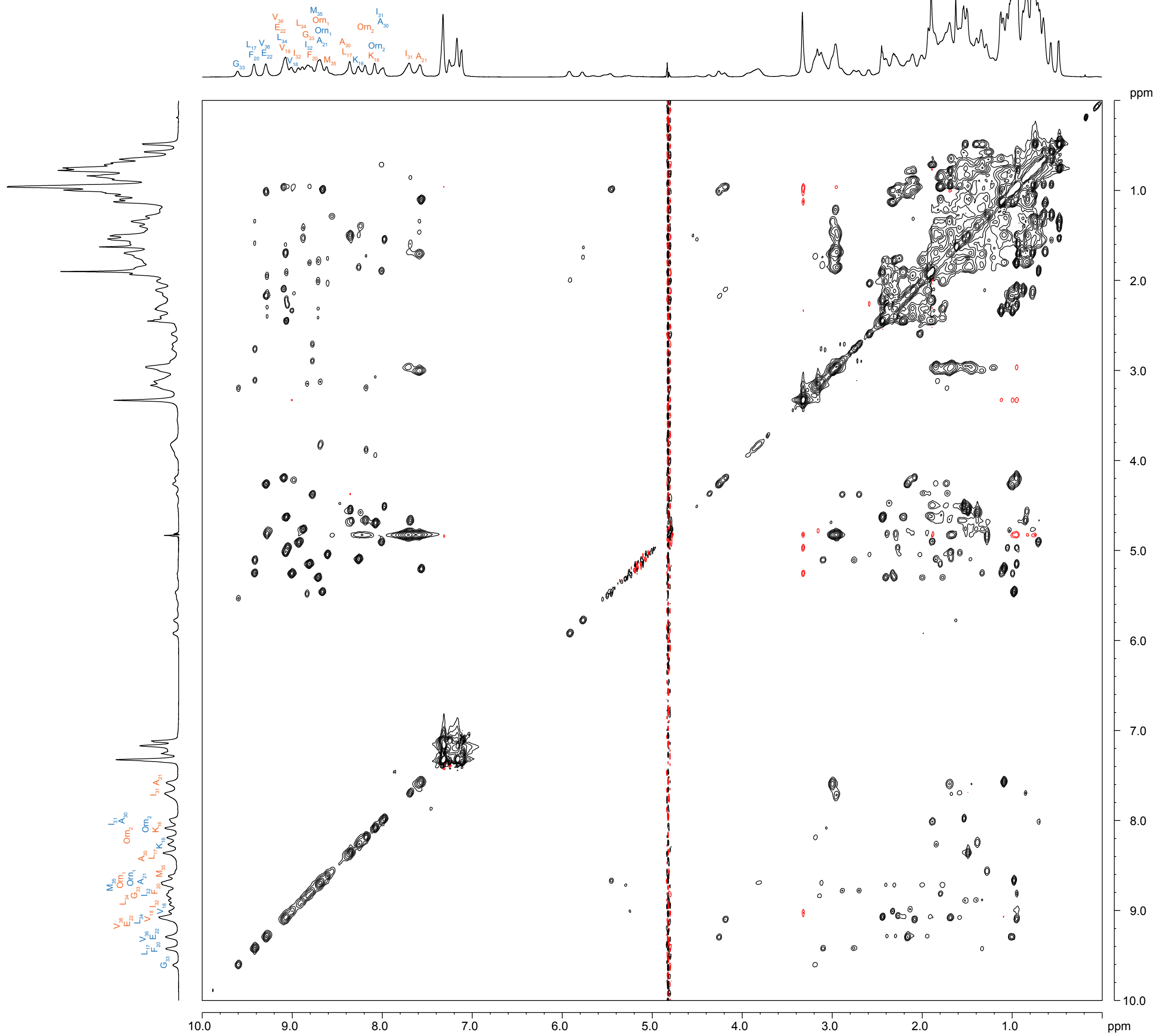

[1]H NMR 2D TOCSY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 60-ms spin-lock mixing time



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Current Data Parameters
NAME          JZ-II-87
EXPNO                5
PROCNO               1

F2 - Acquisition Parameters
Date_         20230316
Time              1.08
INSTRUM          av600
PROBHD   5 mm CPBBO BB-
PULPROG     mlevgpphw5
TD                2048
SOLVENT            D2O
NS                   8
DS                  16
SWH        6602.113 Hz
FIDRES     3.223688 Hz
AQ       0.1551019 sec
RG                 128
DW         75.733 usec
DE          10.00 usec
TE             298.0 K
D0      0.00006533 sec
D1      2.00000000 sec
D9      0.06000000 sec
D12     0.00002000 sec
D16     0.00020000 sec
D19     0.00010000 sec
IN0     0.00015140 sec
L1                  36
P2          20.00 usec

======== CHANNEL f1 ========
SFO1    600.1328896 MHz
NUC1                1H
P1          10.00 usec
P5          16.67 usec
P6          25.00 usec
P7          50.00 usec
P17       2500.00 usec
P27         10.00 usec
PLW1     30.00000000 W
PLW10     5.15000010 W
PLW18    30.00000000 W

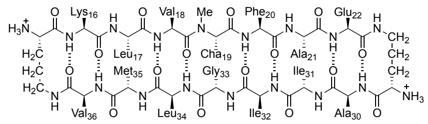
====== GRADIENT CHANNEL =====
GPNAM[1]    SMSQ10.100
GPNAM[2]    SMSQ10.100
GPZ1           34.00 %
GPZ2           22.00 %
P16       1000.00 usec

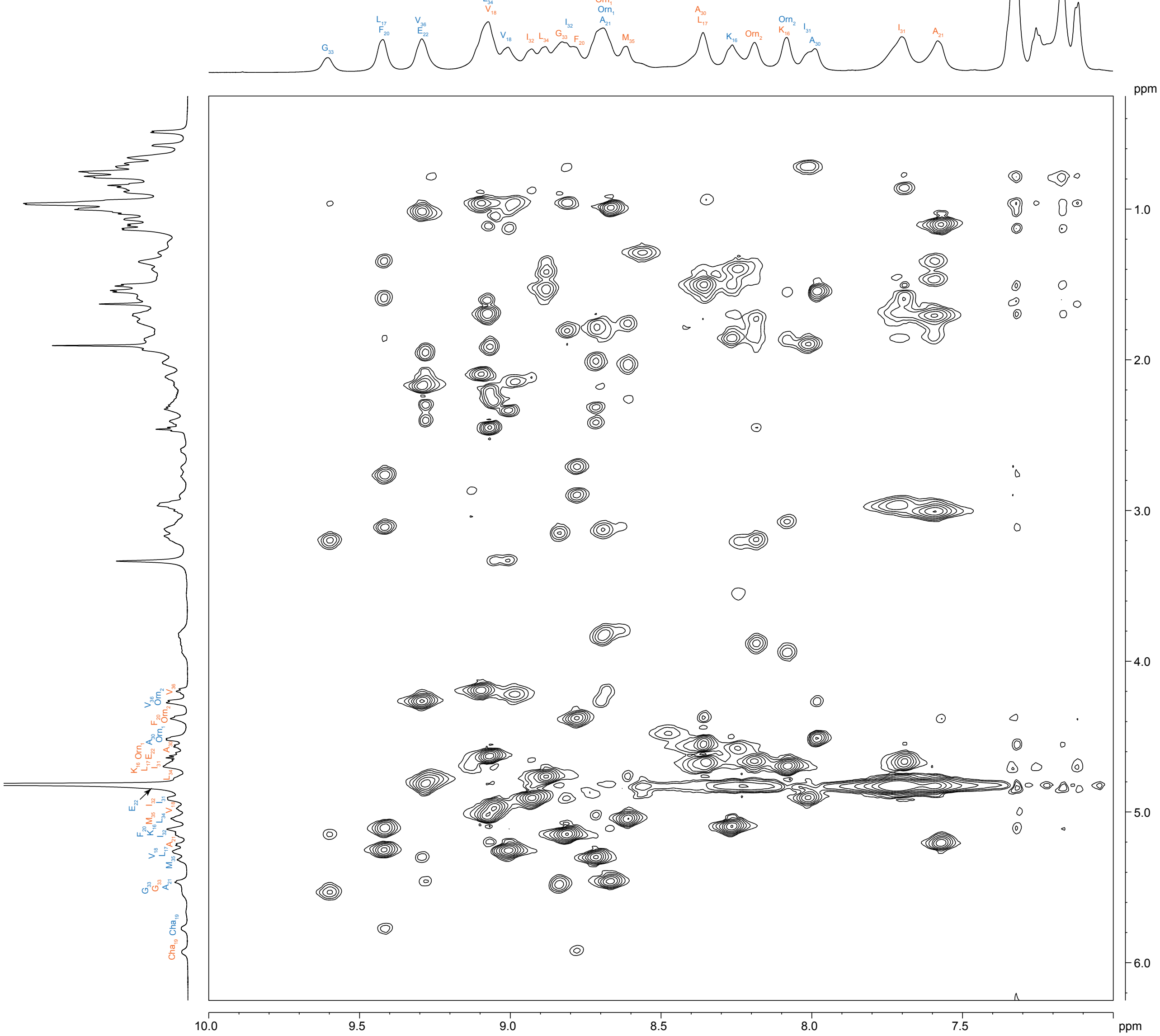
F1 - Acquisition parameters
TD                 256
SFO1      600.1329 MHz
FIDRES    51.601719 Hz
SW          11.006 ppm
FnMODE     States-TPPI

F2 - Processing parameters
SI                1024
SF     600.1299867 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
PC                1.40

F1 - Processing parameters
SI                1024
MC2        States-TPPI
SF     600.1299818 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
```

^1H NMR 2D TOCSY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 60-ms spin-lock mixing time





Current Data Parameters
NAME JZ-II-87
EXPNO 5
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230316
Time 1.08
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG mlevgpphw5
TD 2048
SOLVENT D2O
NS 8
DS 16
SWH 6602.113 Hz
FIDRES 3.223688 Hz
AQ 0.1551019 sec
RG 128
DW 75.733 usec
DE 10.00 usec
TE 298.0 K
D0 0.00006533 sec
D1 2.00000000 sec
D9 0.06000000 sec
D12 0.00002000 sec
D16 0.00020000 sec
D19 0.00010000 sec
IN0 0.00015140 sec
L1 36
P2 20.00 usec

======== CHANNEL f1 ========
SFO1 600.1328896 MHz
NUC1 1H
P1 10.00 usec
P5 16.67 usec
P6 25.00 usec
P7 50.00 usec
P17 2500.00 usec
P27 10.00 usec
PLW1 30.00000000 W
PLW10 5.15000010 W
PLW18 30.00000000 W

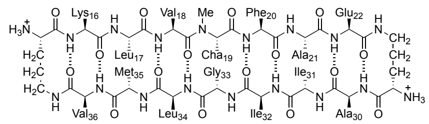
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 34.00 %
GPZ2 22.00 %
P16 1000.00 usec

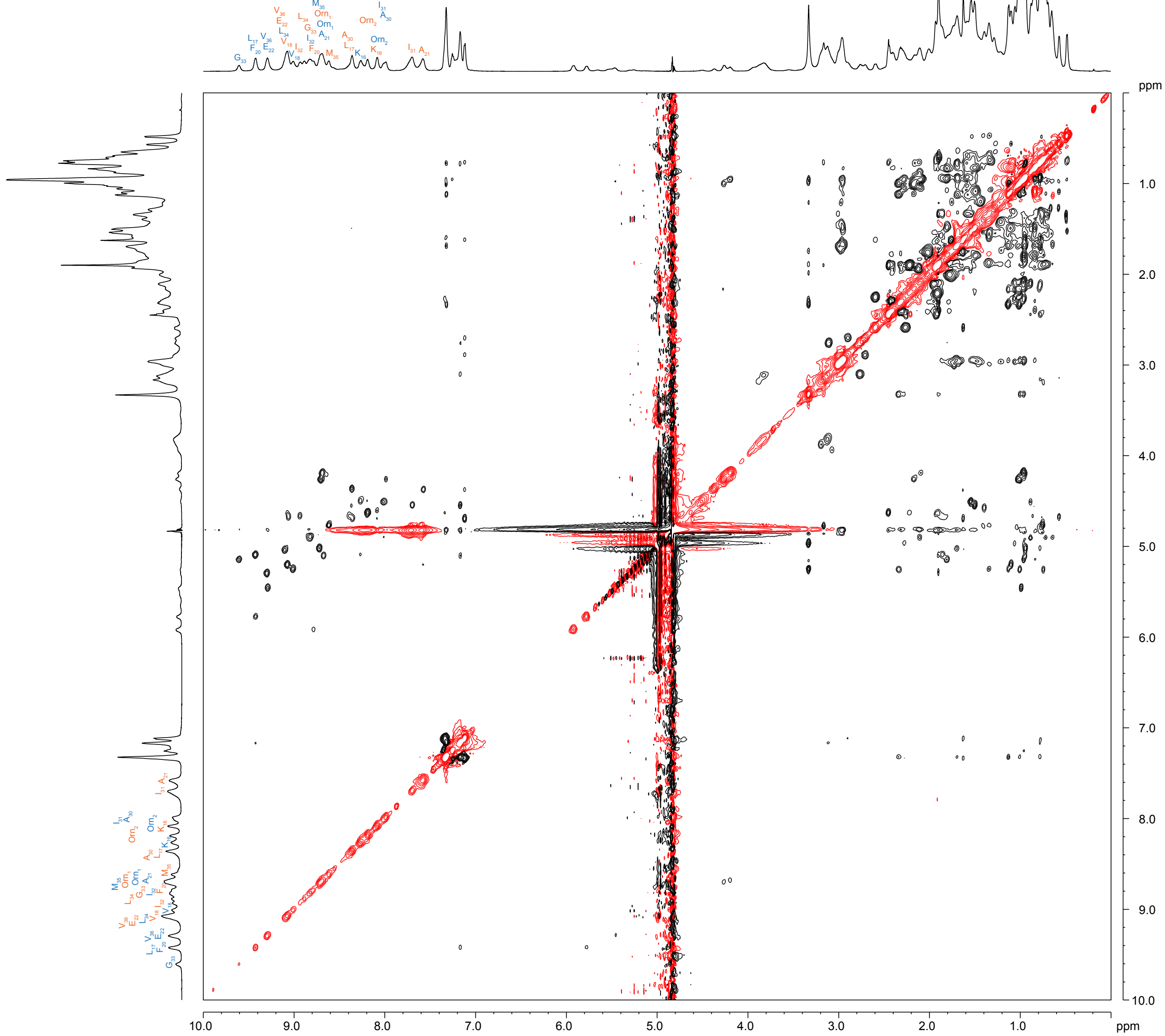
F1 - Acquisition parameters
TD 256
SFO1 600.1329 MHz
FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299867 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299818 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

[1]H NMR 2D ROESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 150-ms spin-lock mixing time



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Current Data Parameters
NAME          JZ-II-87
EXPNO                8
PROCNO               1

F2 - Acquisition Parameters
Date_         20230316
Time              5.38
INSTRUM          av600
PROBHD   5 mm CPBBO BB-
PULPROG  roesygpph19.2.prd
TD                2048
SOLVENT            D2O
NS                   8
DS                  16
SWH        6602.113 Hz
FIDRES     3.223688 Hz
AQ        0.1551019 sec
RG                 645
DW          75.733 usec
DE           10.00 usec
TE             298.0 K
D0       0.00006533 sec
D1       2.00000000 sec
D8       0.15000001 sec
D12      0.00002000 sec
D16      0.00020000 sec
D19      0.00010000 sec
IN0      0.00015140 sec
L4                 417
P15      150000.00 usec

======== CHANNEL f1 ========
SFO1     600.1328896 MHz
NUC1                1H
P0           10.00 usec
P1           10.00 usec
P25         180.00 usec
P27          10.00 usec
PLW1      30.00000000 W
PLW18     30.00000000 W
PLW27      0.31623000 W

====== GRADIENT CHANNEL =====
GPNAM[1]     SMSQ10.100
GPNAM[2]     SMSQ10.100
GPZ1            30.00 %
GPZ2            30.00 %
P16        1000.00 usec

F1 - Acquisition parameters
TD                 256
SFO1      600.1329 MHz
FIDRES    51.601719 Hz
SW          11.006 ppm
FnMODE     States-TPPI

F2 - Processing parameters
SI                1024
SF     600.1299845 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
PC                1.40

F1 - Processing parameters
SI                1024
MC2        States-TPPI
SF     600.1299844 MHz
WDW              QSINE
SSB                  2
LB                0 Hz
GB                   0
```
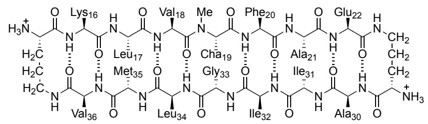
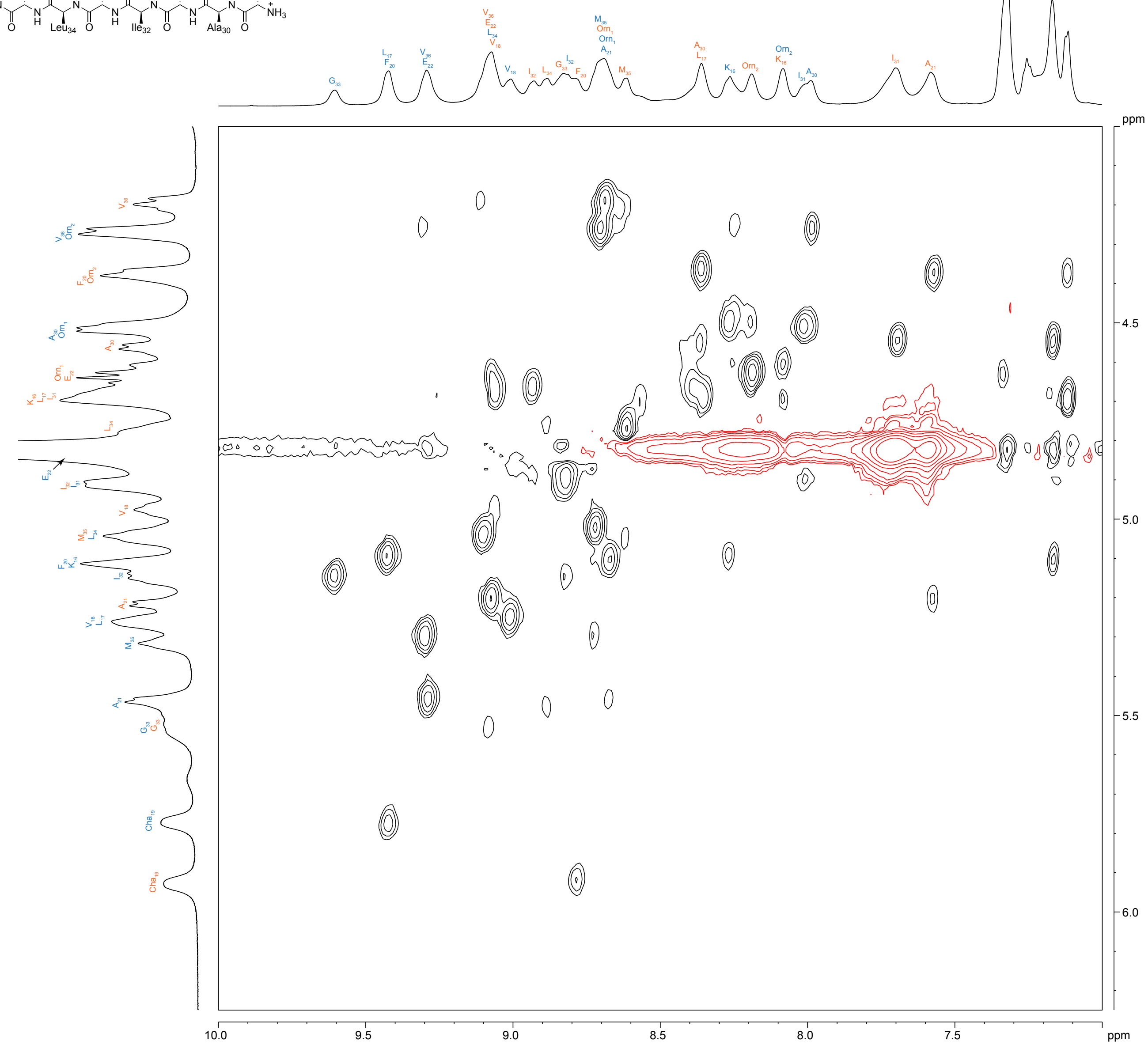
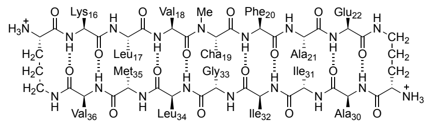

^{1}H NMR 2D ROESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 150-ms spin-lock mixing time

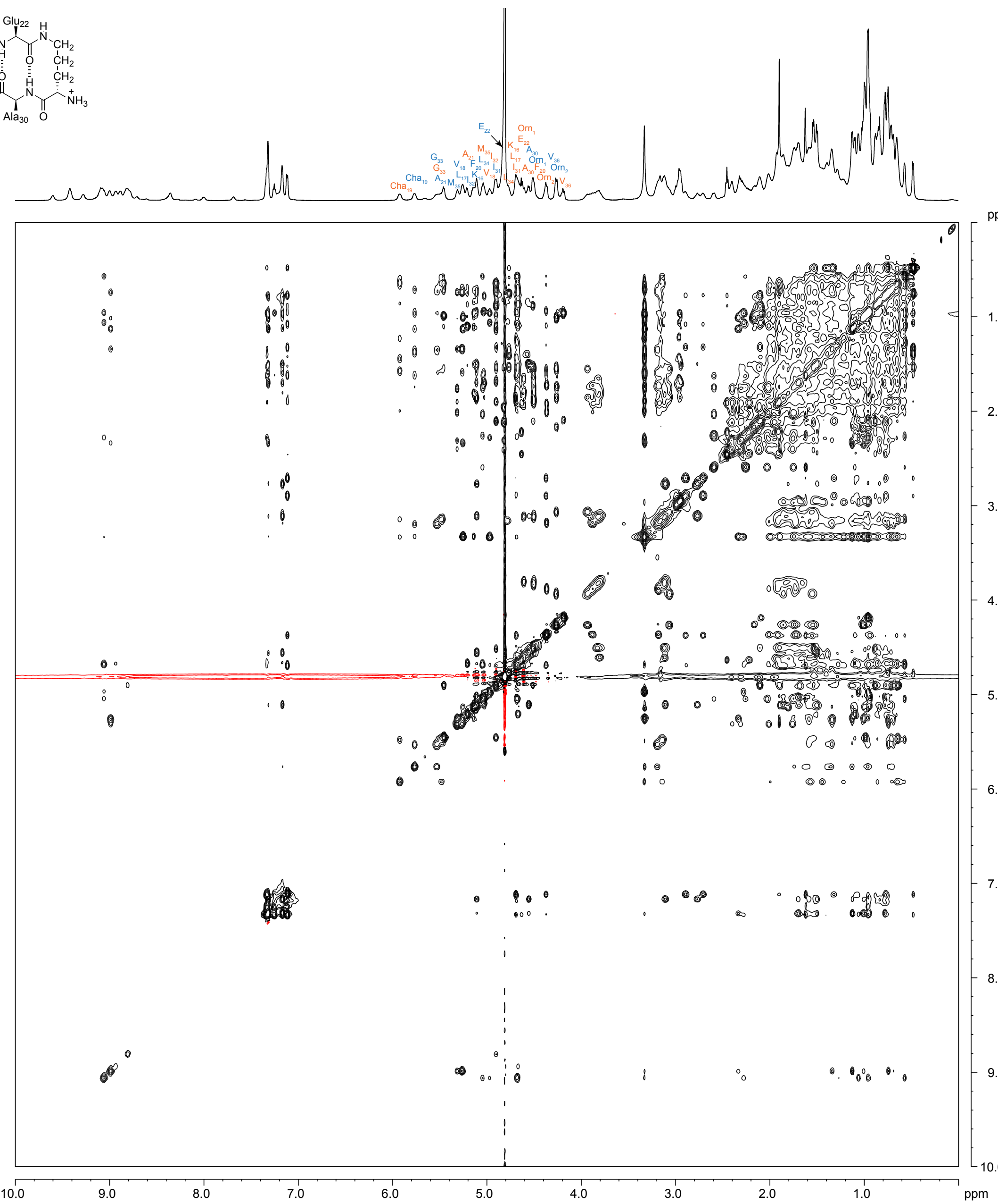




Current Data Parameters	
NAME	JZ-II-87
EXPNO	8
PROCNO	1
F2 - Acquisition Parameters	
Date_	20230316
Time	5.38
INSTRUM	av600
PROBHD	5 mm CPBBO BB-
PULPROG	roesygpph19.2.prd
TD	2048
SOLVENT	D2O
NS	8
DS	16
SWH	6602.113 Hz
FIDRES	3.223688 Hz
AQ	0.1551019 sec
RG	645
DW	75.733 usec
DE	10.00 usec
TE	298.0 K
D0	0.00006533 sec
D1	2.00000000 sec
D8	0.15000001 sec
D12	0.00002000 sec
D16	0.00020000 sec
D19	0.00010000 sec
IN0	0.00015140 sec
L4	417
P15	150000.00 usec
======== CHANNEL f1 ========	
SFO1	600.1328896 MHz
NUC1	1H
P0	10.00 usec
P1	10.00 usec
P25	180.00 usec
P27	10.00 usec
PLW1	30.00000000 W
PLW18	30.00000000 W
PLW27	0.31623000 W
====== GRADIENT CHANNEL =====	
GPNAM[1]	SMSQ10.100
GPNAM[2]	SMSQ10.100
GPZ1	30.00 %
GPZ2	30.00 %
P16	1000.00 usec
F1 - Acquisition parameters	
TD	256
SFO1	600.1329 MHz
FIDRES	51.601719 Hz
SW	11.006 ppm
FnMODE	States-TPPI
F2 - Processing parameters	
SI	1024
SF	600.1299845 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.40
F1 - Processing parameters	
SI	1024
MC2	States-TPPI
SF	600.1299844 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

[1]H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 200-msec mixing time





Current Data Parameters
NAME JZ-II-101
EXPNO 2
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230505
Time 19.00
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG noesygpph
TD 2048
SOLVENT D2O
NS 8
DS 16
SWH 6602.113 Hz
FIDRES 3.223688 Hz
AQ 0.1551019 sec
RG 36
DW 75.733 usec
DE 17.51 usec
TE 297.9 K
D0 0.00006297 sec
D1 2.00000000 sec
D8 0.20000000 sec
D16 0.00020000 sec
IN0 0.00015140 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
P2 20.00 usec
PLW1 30.00000000 W

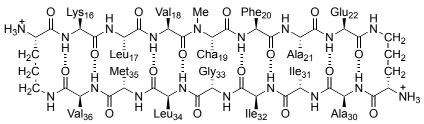
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPZ1 40.00 %
P16 1000.00 usec

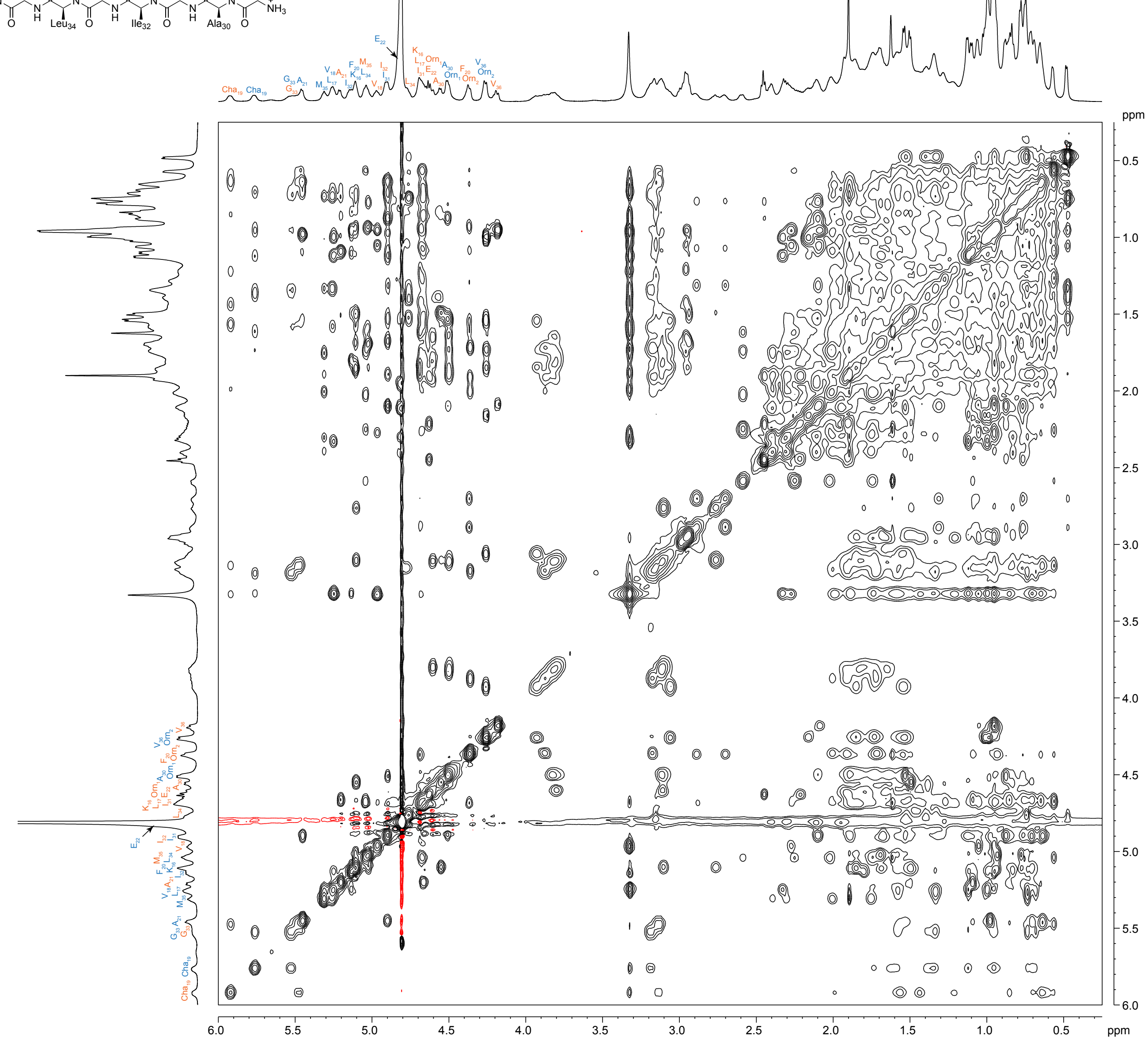
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299980 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299979 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

^{1}H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in D_2O at 600 MHz and 298 K with 200-msec mixing time





Current Data Parameters
NAME JZ-II-101
EXPNO 2
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230505
Time 19.00
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG noesygpph
TD 2048
SOLVENT D2O
NS 8
DS 16
SWH 6602.113 Hz
FIDRES 3.223688 Hz
AQ 0.1551019 sec
RG 36
DW 75.733 usec
DE 17.51 usec
TE 297.9 K
D0 0.00006297 sec
D1 2.00000000 sec
D8 0.20000000 sec
D16 0.00020000 sec
IN0 0.00015140 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
P2 20.00 usec
PLW1 30.00000000 W

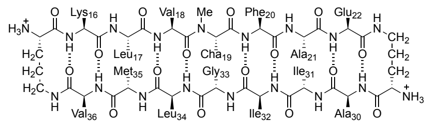
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPZ1 40.00 %
P16 1000.00 usec

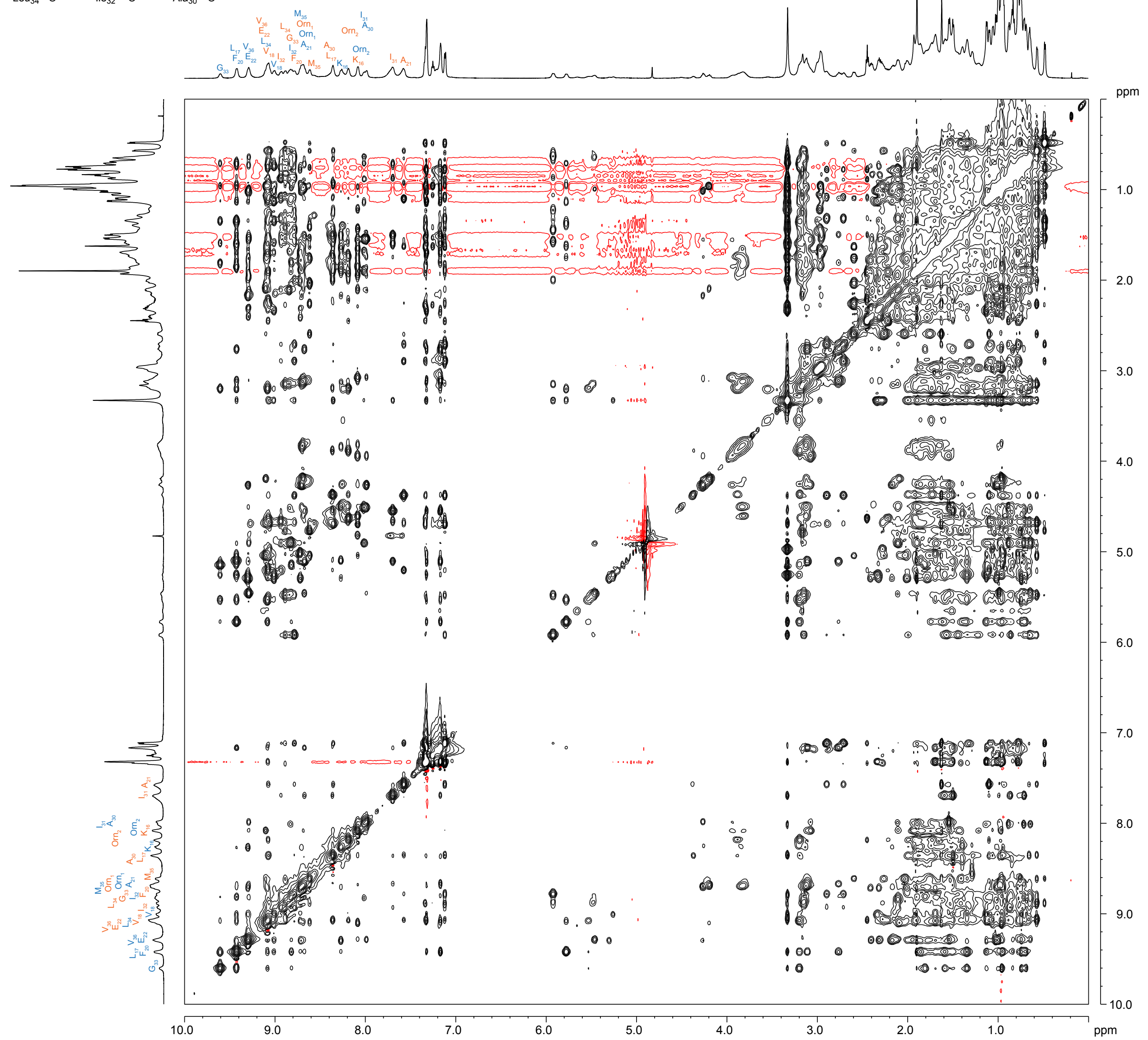
F1 - Acquisition parameters
TD 256
SFO1 600.133 MHz
FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299980 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299979 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

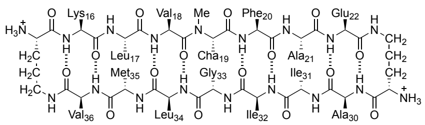
[1]H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-msec mixing time

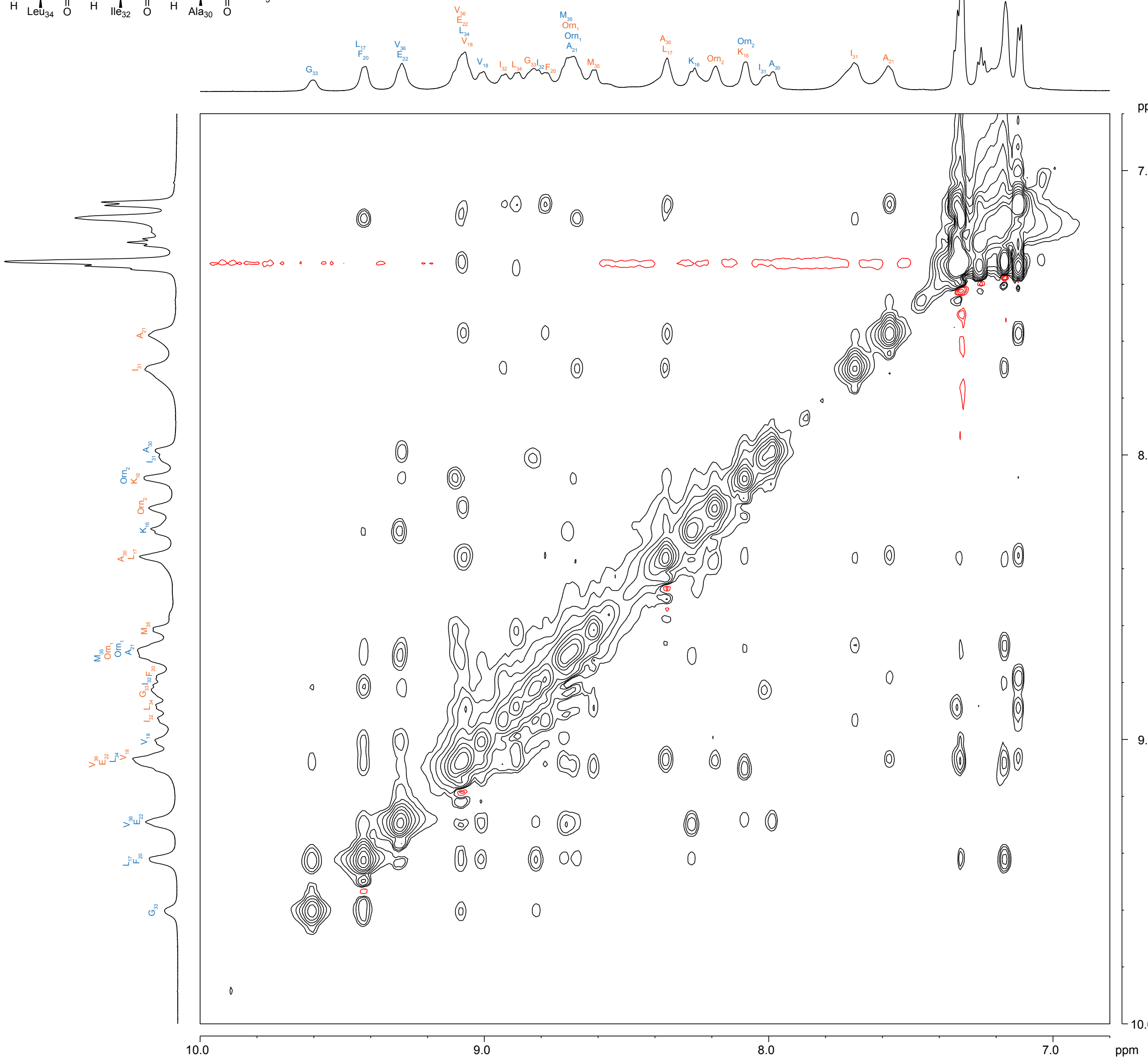




Current Data Parameters	
NAME	JZ-II-101
EXPNO	12
PROCNO	1
F2 - Acquisition Parameters	
Date_	20230514
Time	11.52
INSTRUM	av600
PROBHD	5 mm CPBBO BB-
PULPROG	noesygpphw5
TD	2048
SOLVENT	D2O
NS	8
DS	16
SWH	6602.113 Hz
FIDRES	3.223688 Hz
AQ	0.1551019 sec
RG	456
DW	75.733 usec
DE	10.00 usec
TE	298.0 K
D0	0.00006297 sec
D1	2.00000000 sec
D8	0.20000000 sec
D11	0.03000000 sec
D12	0.00002000 sec
D13	0.00000400 sec
D16	0.00020000 sec
D19	0.00010000 sec
IN0	0.00015140 sec
======== CHANNEL f1 ========	
SFO1	600.1328842 MHz
NUC1	1H
P1	10.00 usec
P27	10.00 usec
PLW1	30.00000000 W
PLW18	30.00000000 W
PLW32	0.00000461 W
====== GRADIENT CHANNEL =====	
GPNAM[1]	SMSQ10.100
GPNAM[2]	SMSQ10.100
GPZ1	34.00 %
GPZ2	22.00 %
P16	1000.00 usec
F1 - Acquisition parameters	
TD	256
SFO1	600.1329 MHz
FIDRES	51.601719 Hz
SW	11.006 ppm
FnMODE	States-TPPI
F2 - Processing parameters	
SI	1024
SF	600.1299848 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0
PC	1.40
F1 - Processing parameters	
SI	1024
MC2	States-TPPI
SF	600.1299846 MHz
WDW	QSINE
SSB	2
LB	0 Hz
GB	0

[1]H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-msec mixing time





Current Data Parameters
NAME JZ-II-101
EXPNO 12
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230514
Time 11.52
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG noesygpphw5
TD 2048
SOLVENT D2O
NS 8
DS 16
SWH 6602.113 Hz
FIDRES 3.223688 Hz
AQ 0.1551019 sec
RG 456
DW 75.733 usec
DE 10.00 usec
TE 298.0 K
D0 0.00006297 sec
D1 2.00000000 sec
D8 0.20000000 sec
D11 0.03000000 sec
D12 0.00002000 sec
D13 0.00000400 sec
D16 0.00020000 sec
D19 0.00010000 sec
IN0 0.00015140 sec

======== CHANNEL f1 ========
SFO1 600.1328842 MHz
NUC1 1H
P1 10.00 usec
P27 10.00 usec
PLW1 30.00000000 W
PLW18 30.00000000 W
PLW32 0.00000461 W

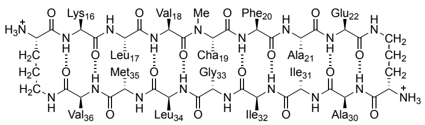
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPNAM[2] SMSQ10.100
GPZ1 34.00 %
GPZ2 22.00 %
P16 1000.00 usec

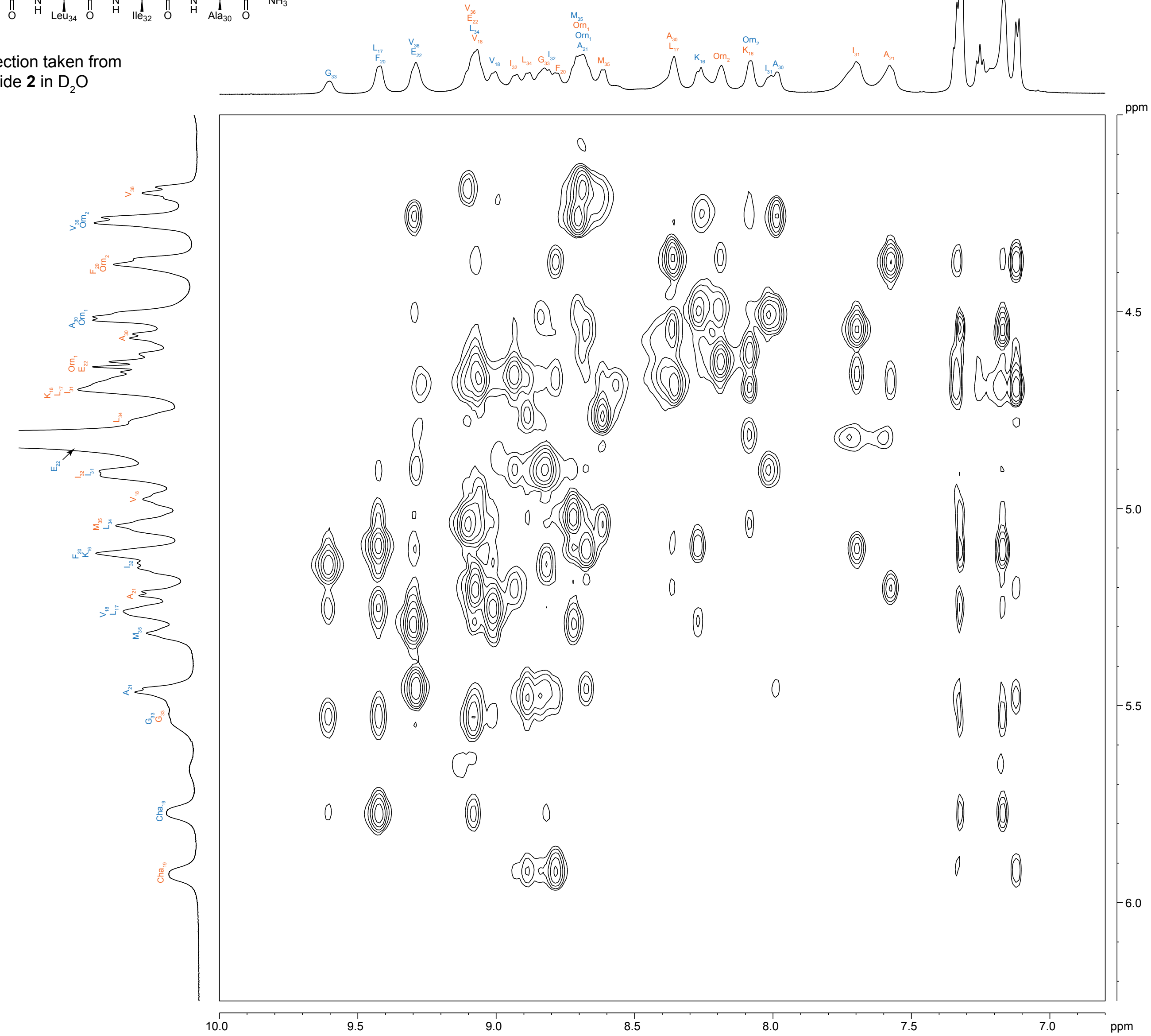
F1 - Acquisition parameters
TD 256
SFO1 600.1329 MHz
FIDRES 51.601719 Hz
SW 11.006 ppm
FnMODE States-TPPI

F2 - Processing parameters
SI 1024
SF 600.1299848 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 1.40

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 600.1299846 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

[1]H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-msec mixing time



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Current Data Parameters
NAME          JZ-II-101
EXPNO               12
PROCNO               1

F2 - Acquisition Parameters
Date_         20230514
Time             11.52
INSTRUM          av600
PROBHD   5 mm CPBBO BB-
PULPROG    noesygpphw5
TD                2048
SOLVENT            D2O
NS                   8
DS                  16
SWH        6602.113 Hz
FIDRES     3.223688 Hz
AQ       0.1551019 sec
RG                 456
DW         75.733 usec
DE          10.00 usec
TE             298.0 K
D0      0.00006297 sec
D1      2.00000000 sec
D8      0.20000000 sec
D11     0.03000000 sec
D12     0.00002000 sec
D13     0.00000400 sec
D16     0.00020000 sec
D19     0.00010000 sec
IN0     0.00015140 sec

======== CHANNEL f1 ========
SFO1    600.1328842 MHz
NUC1                 1H
P1           10.00 usec
P27          10.00 usec
PLW1     30.00000000 W
PLW18    30.00000000 W
PLW32     0.00000461 W

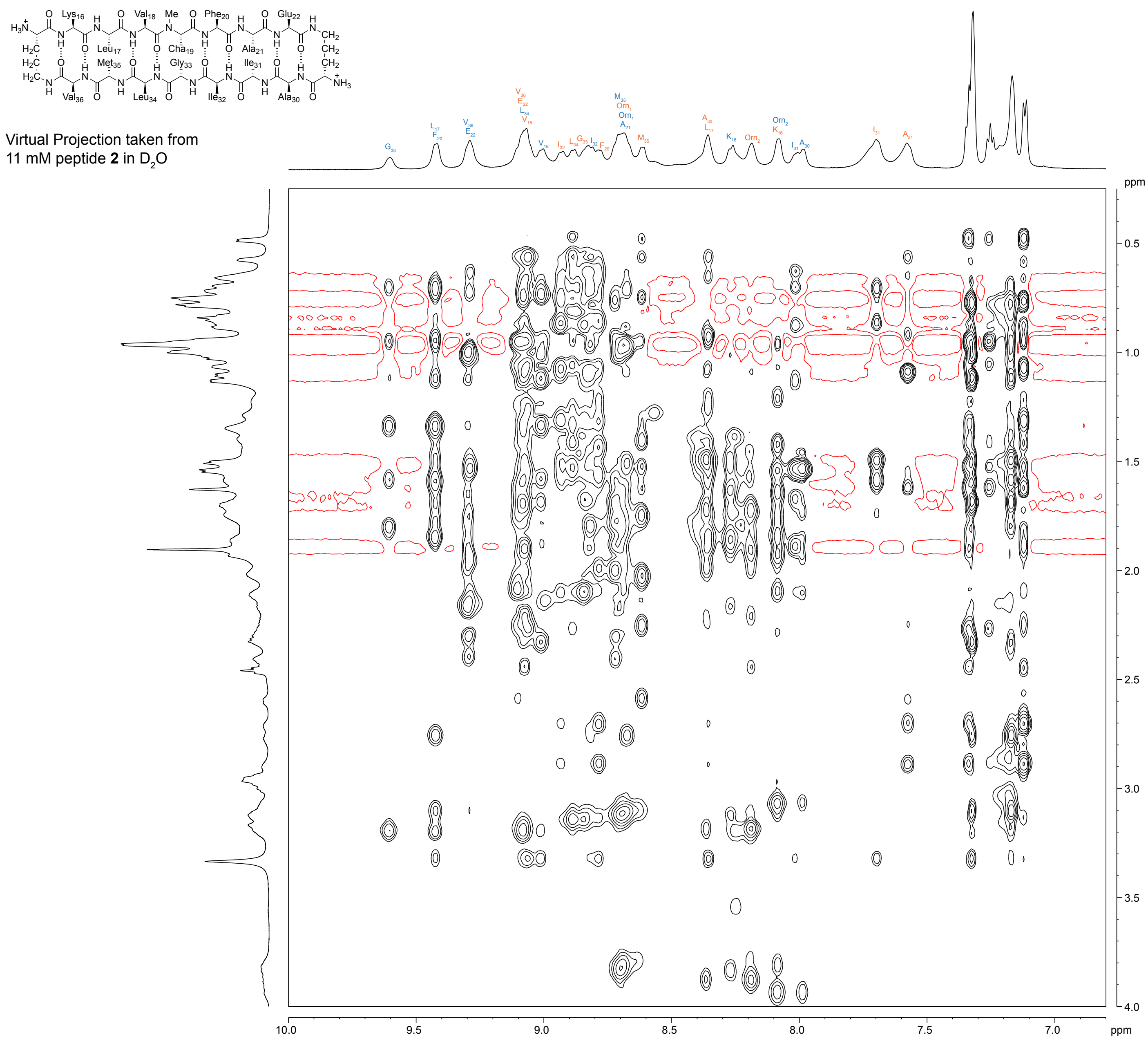
====== GRADIENT CHANNEL =====
GPNAM[1]     SMSQ10.100
GPNAM[2]     SMSQ10.100
GPZ1            34.00 %
GPZ2            22.00 %
P16        1000.00 usec

F1 - Acquisition parameters
TD                  256
SFO1       600.1329 MHz
FIDRES     51.601719 Hz
SW          11.006 ppm
FnMODE      States-TPPI

F2 - Processing parameters
SI                 1024
SF      600.1299848 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
PC                 1.40

F1 - Processing parameters
SI                 1024
MC2         States-TPPI
SF      600.1299846 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
```

[1]H NMR 2D NOESY spectrum of 11.0 mM peptide **2** in 9:1 H_2O/D_2O at 600 MHz and 298 K with WATERGATE suppression of the HOD peak and 200-ms mixing time




```
Current Data Parameters
NAME          JZ-II-101
EXPNO               12
PROCNO               1

F2 - Acquisition Parameters
Date_          20230514
Time              11.52
INSTRUM           av600
PROBHD   5 mm CPBBO BB-
PULPROG     noesygpphw5
TD                 2048
SOLVENT             D2O
NS                    8
DS                   16
SWH          6602.113 Hz
FIDRES       3.223688 Hz
AQ          0.1551019 sec
RG                  456
DW           75.733 usec
DE            10.00 usec
TE              298.0 K
D0         0.00006297 sec
D1         2.00000000 sec
D8         0.20000000 sec
D11        0.03000000 sec
D12        0.00002000 sec
D13        0.00000400 sec
D16        0.00020000 sec
D19        0.00010000 sec
IN0        0.00015140 sec

======== CHANNEL f1 ========
SFO1       600.1328842 MHz
NUC1                 1H
P1             10.00 usec
P27            10.00 usec
PLW1       30.00000000 W
PLW18      30.00000000 W
PLW32       0.00000461 W

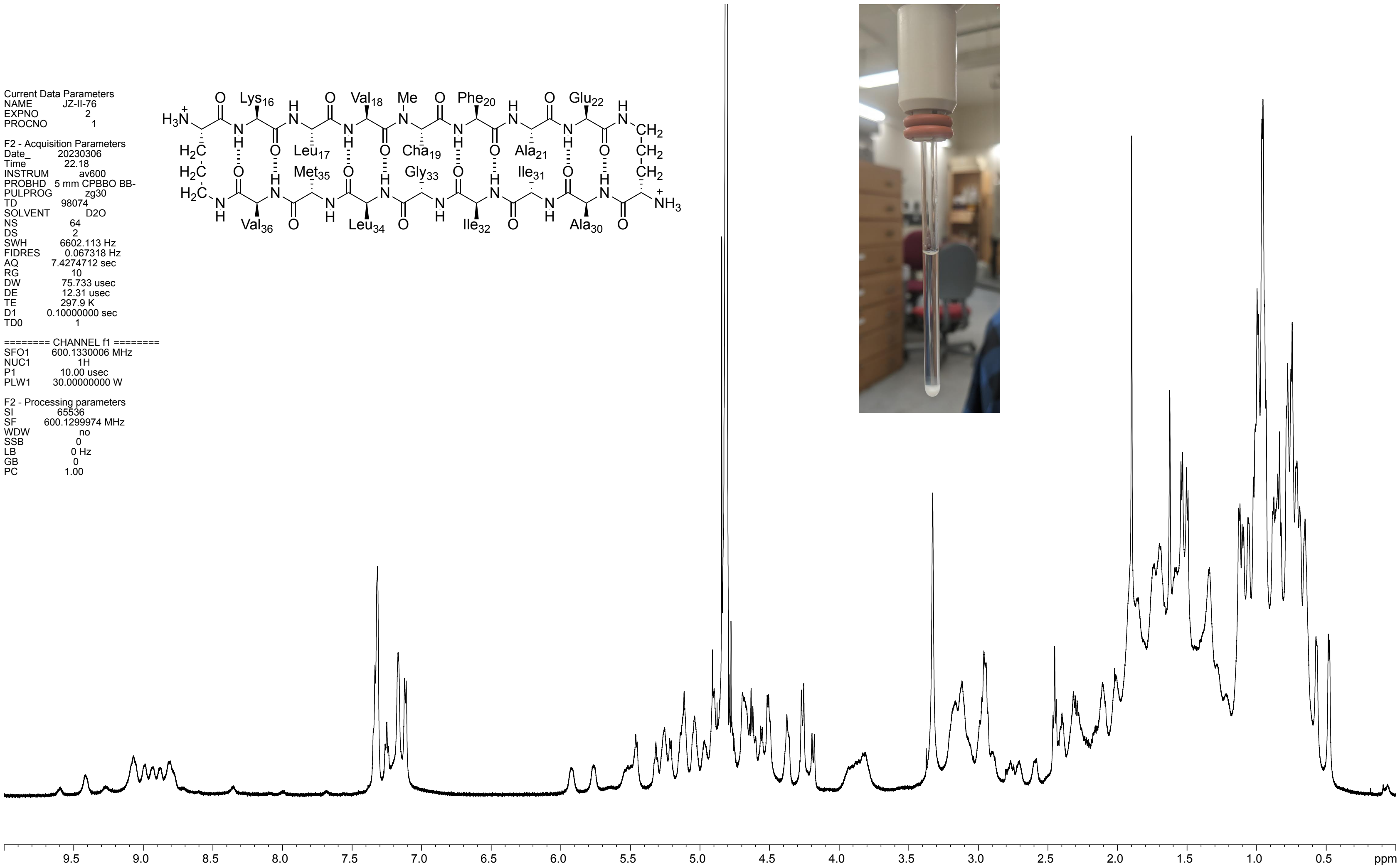
====== GRADIENT CHANNEL =====
GPNAM[1]     SMSQ10.100
GPNAM[2]     SMSQ10.100
GPZ1            34.00 %
GPZ2            22.00 %
P16          1000.00 usec

F1 - Acquisition parameters
TD                  256
SFO1         600.1329 MHz
FIDRES      51.601719 Hz
SW           11.006 ppm
FnMODE      States-TPPI

F2 - Processing parameters
SI                 1024
SF         600.1299848 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
PC                 1.40

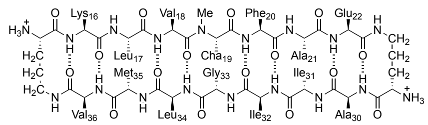
F1 - Processing parameters
SI                 1024
MC2         States-TPPI
SF         600.1299846 MHz
WDW               QSINE
SSB                   2
LB                 0 Hz
GB                    0
```

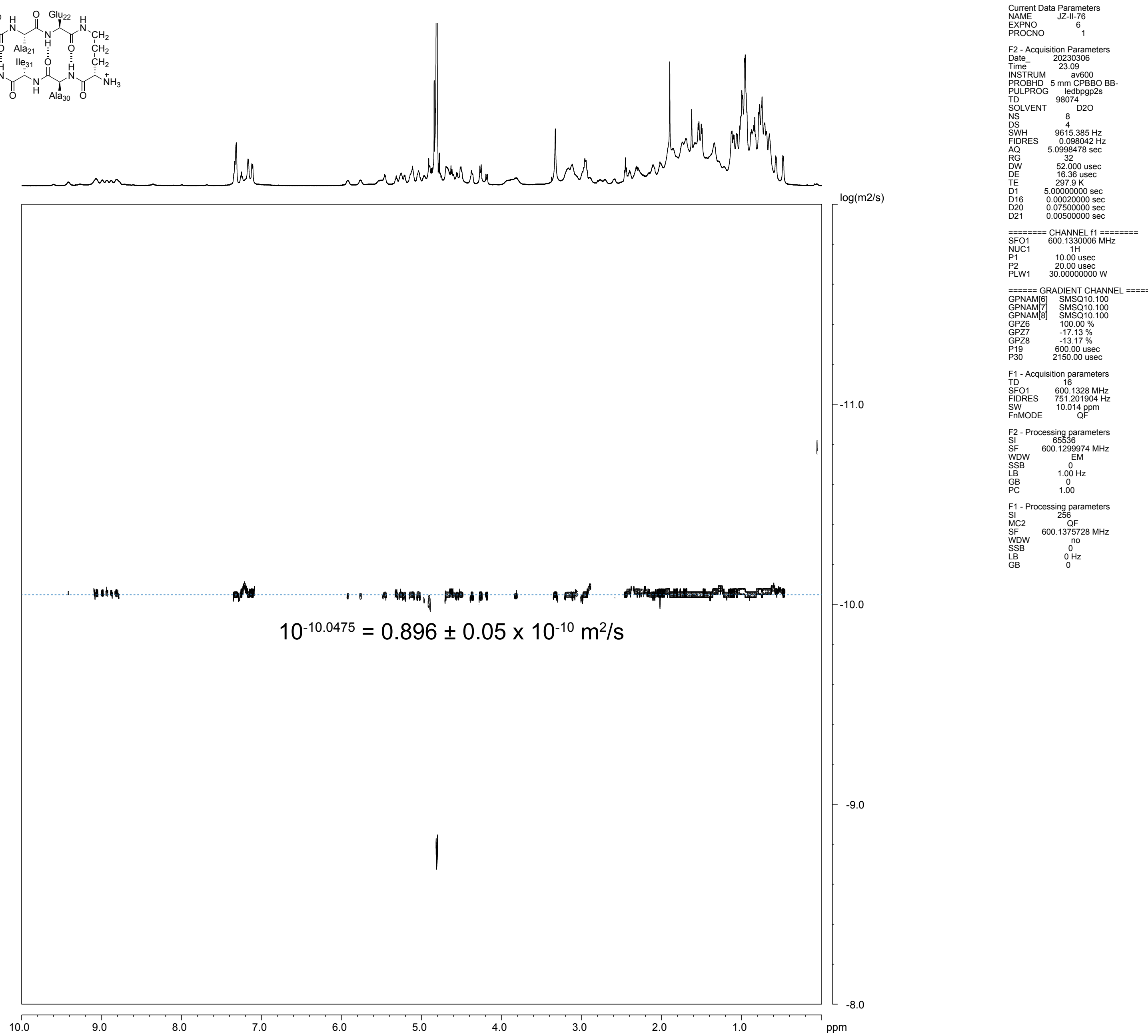
[1]H NMR spectrum of nominal 16.0 mM peptide **2** in D_2O at 600 MHz and 298 K



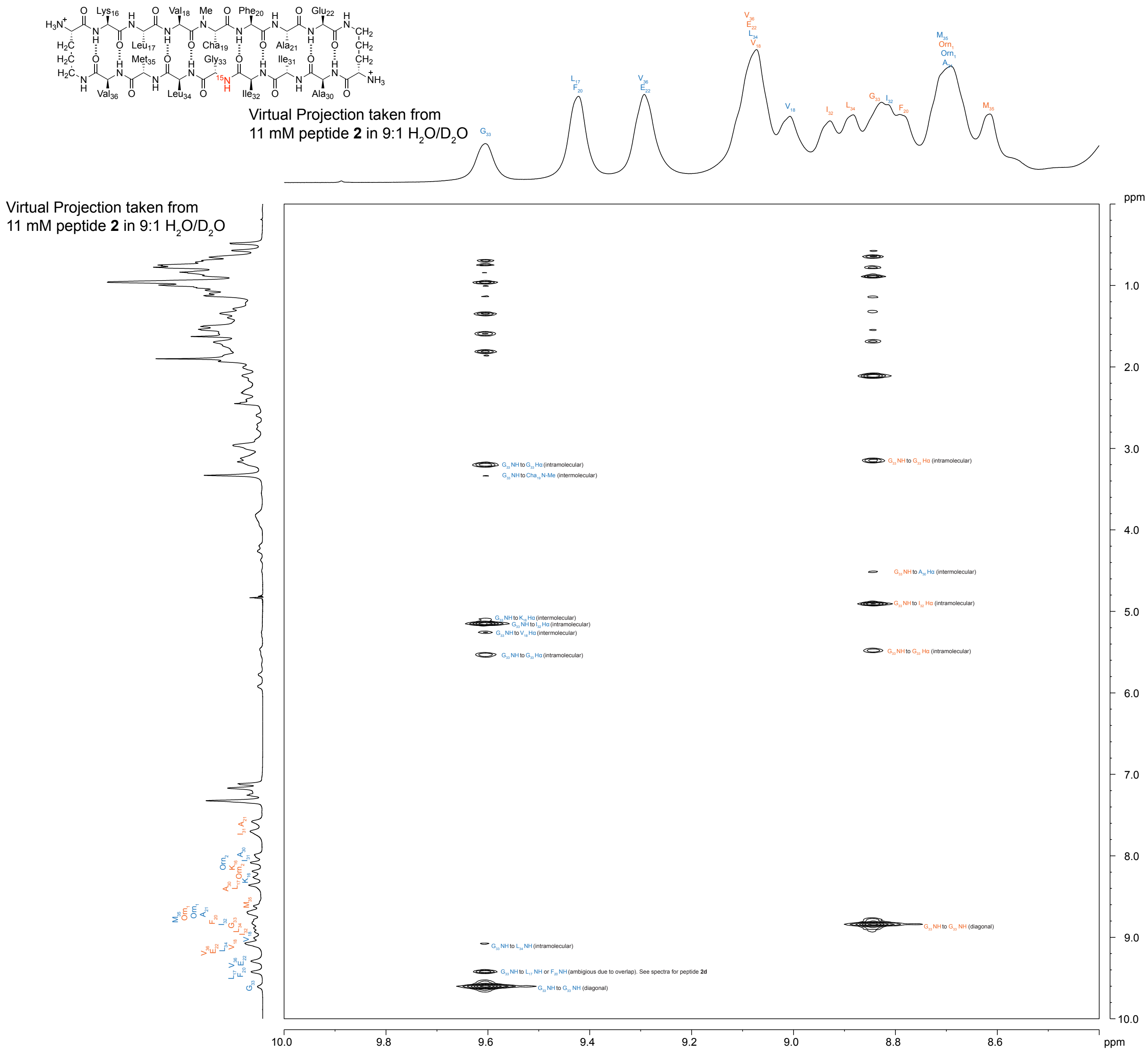
Note: The concentration is nominal because peptide **2** is not fully soluble at 16.0 mM.

^{1}H NMR DOSY spectrum of nominal 16.0 mM peptide **2** in D_2O at 600 MHz and 298 K





^{15}N-edited NOESY spectrum of 11.0 mM peptide **2a** in 9:1 H_2O/D_2O at 800 MHz and 298 K with WATERGATE suppression of the HOD peak and 120-msec mixing time



Current Data Parameters
NAME JZ-II-299
EXPNO 14
PROCNO 15

F2 - Acquisition Parameters
Date_ 20240704
Time 7.54 h
INSTRUM Avance
PROBHD Z8435_0053 (PH
PULPROG noesyhsqcf3gpwg3d
TD 2048
SOLVENT H2O+D2O
NS 16
DS 32
SWH 9615.385 Hz
FIDRES 9.390024 Hz
AQ 0.1064960 sec
RG 101
DW 52.000 usec
DE 6.50 usec
TE 298.0 K
CNST4 90.0000000
D0 0.00000300 sec
D1 1.00000000 sec
D8 0.12000000 sec
D10 0.00000300 sec
D11 0.03000000 sec
D16 0.00020000 sec
D26 0.00277778 sec
IN0 0.00005210 sec
IN10 0.00017626 sec
TDav 1
ZGOPTNS
SFO1 799.7537563 MHz
NUC1 1H
P1 8.38 usec
P2 16.76 usec
P11 1000.00 usec
P29 4000.00 usec
PLW1 27.04899025 W
SPNAM[1] Sinc1.1000
SPOAL1 0.500
SPOFFS1 0 Hz
SPW1 0.00548277 W
SPNAM[11 Sinc1.1000
SPOAL11 0.500
SPOFFS11 0 Hz
SPW11 0.00034232 W
SFO3 81.0474452 MHz
NUC3 15N
CPDPRG[3 garp
P21 32.79 usec
P22 65.58 usec
PCPD3 220.00 usec
PLW3 430.00000000 W
PLW16 9.55510044 W
GPNAM[3] SMSQ10.100
GPX3 0 %
GPY3 0 %
GPZ3 -50.00 %
GPNAM[4] SMSQ10.100
GPX4 0 %
GPY4 0 %
GPZ4 40.00 %
GPNAM[5] SMSQ10.100
GPX5 0 %
GPY5 0 %
GPZ5 50.00 %
GPNAM[6] SMSQ10.100
GPX6 0 %
GPY6 0 %
GPZ6 30.00 %
GPNAM[7] SMSQ10.100
GPX7 0 %
GPY7 0 %
GPZ7 31.00 %
P16 1000.00 usec
P30 3000.00 usec
P31 5000.00 usec

======== F2 INDIRECT DIMENSION ========
td2 1
sw_F2 35.000000

======== F1 INDIRECT DIMENSION ========
td1 1024
sw_F1 12.000000

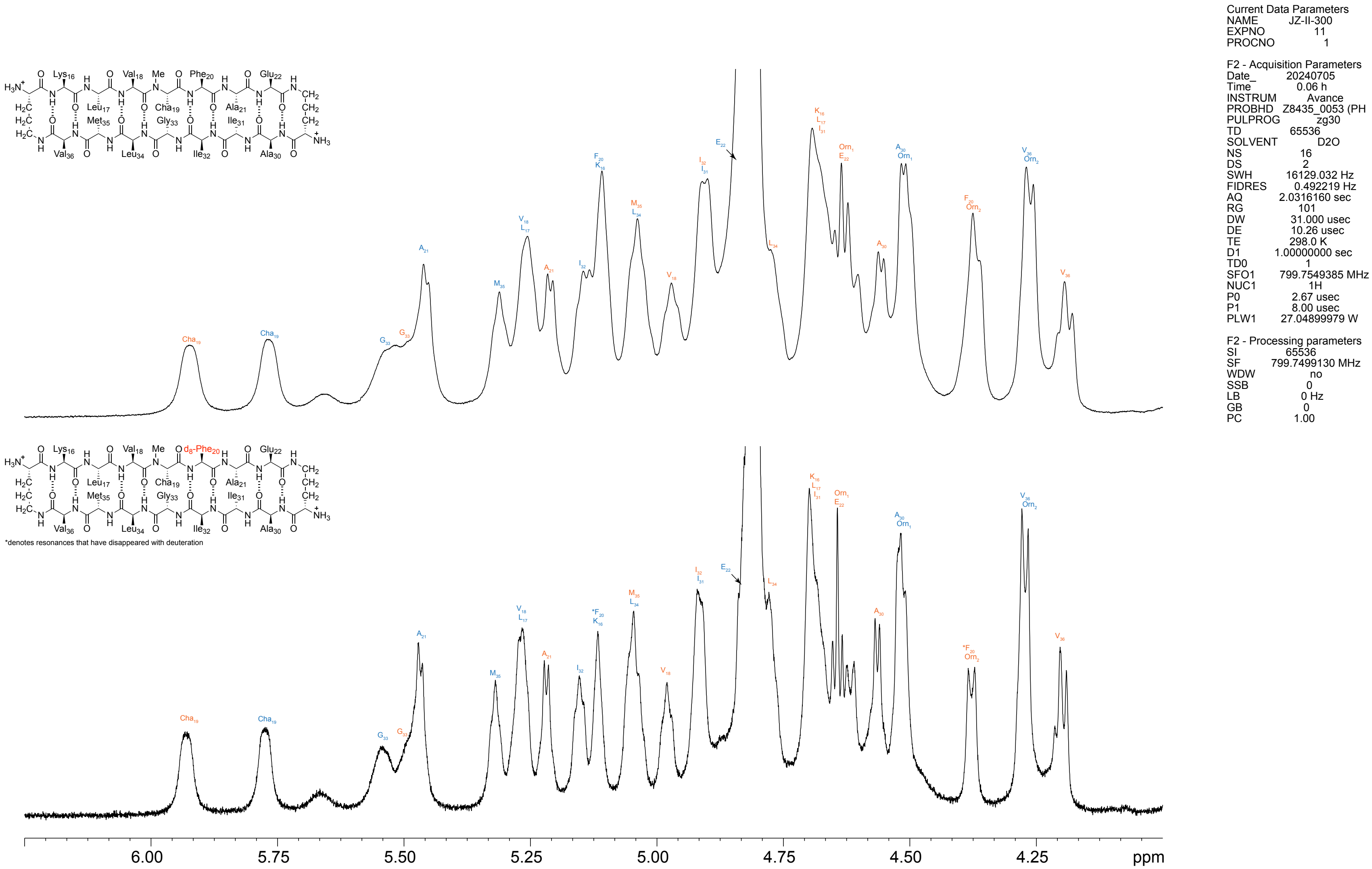
F1 - Acquisition parameters
TD 1
SFO1 81.04745 MHz
FIDRES 5673.321289 Hz
SW 35.000 ppm
FnMODE States-TPPI

F3 - Acquisition parameters
SW 12.000 ppm
SFO1 799.7538 MHz
HZpPT 1.171514 Hz
TD 1024

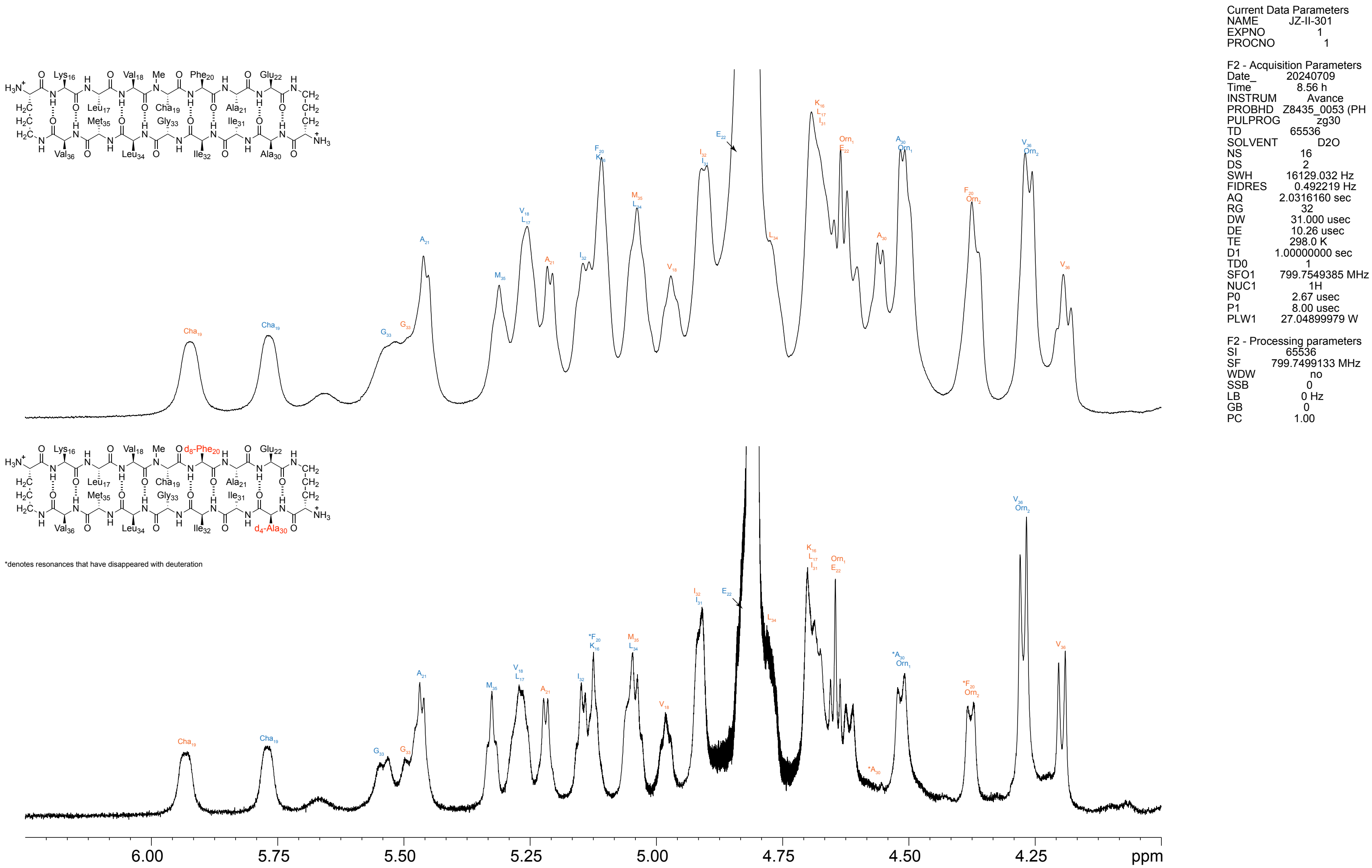
F2 - Processing parameters
SI 2048
SF 799.7498992 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0
PC 4.00

F1 - Processing parameters
SI 1024
MC2 States-TPPI
SF 799.7498984 MHz
WDW QSINE
SSB 2
LB 0 Hz
GB 0

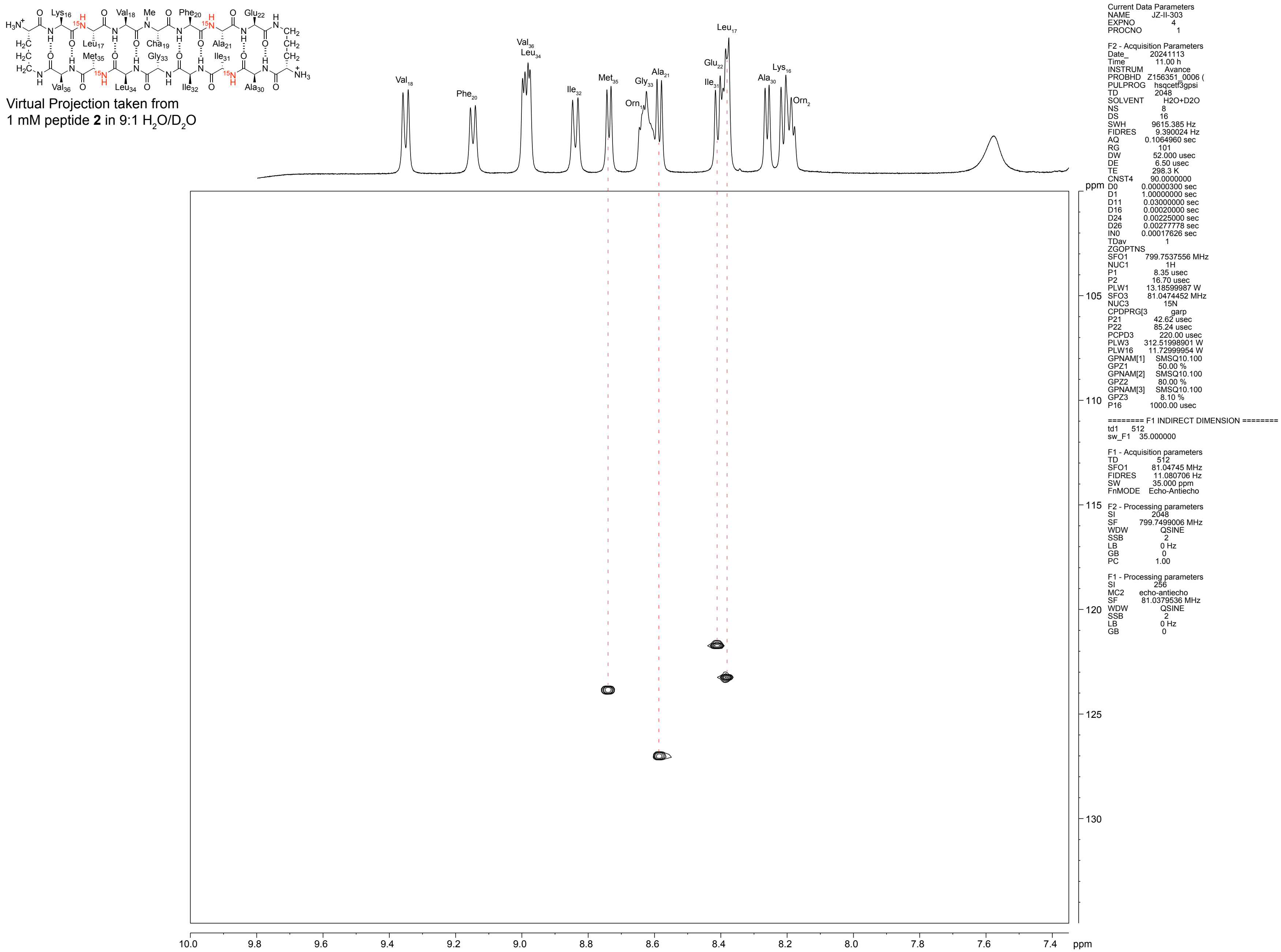
[1]H NMR stacked plot of 11.0 mM peptide **2** and **2b** showing key Hα resonances that disappear with deuteration in D_2O at 298 K. Peptide **2** taken at 600 MHz and peptide **2b** taken at 800 MHz.



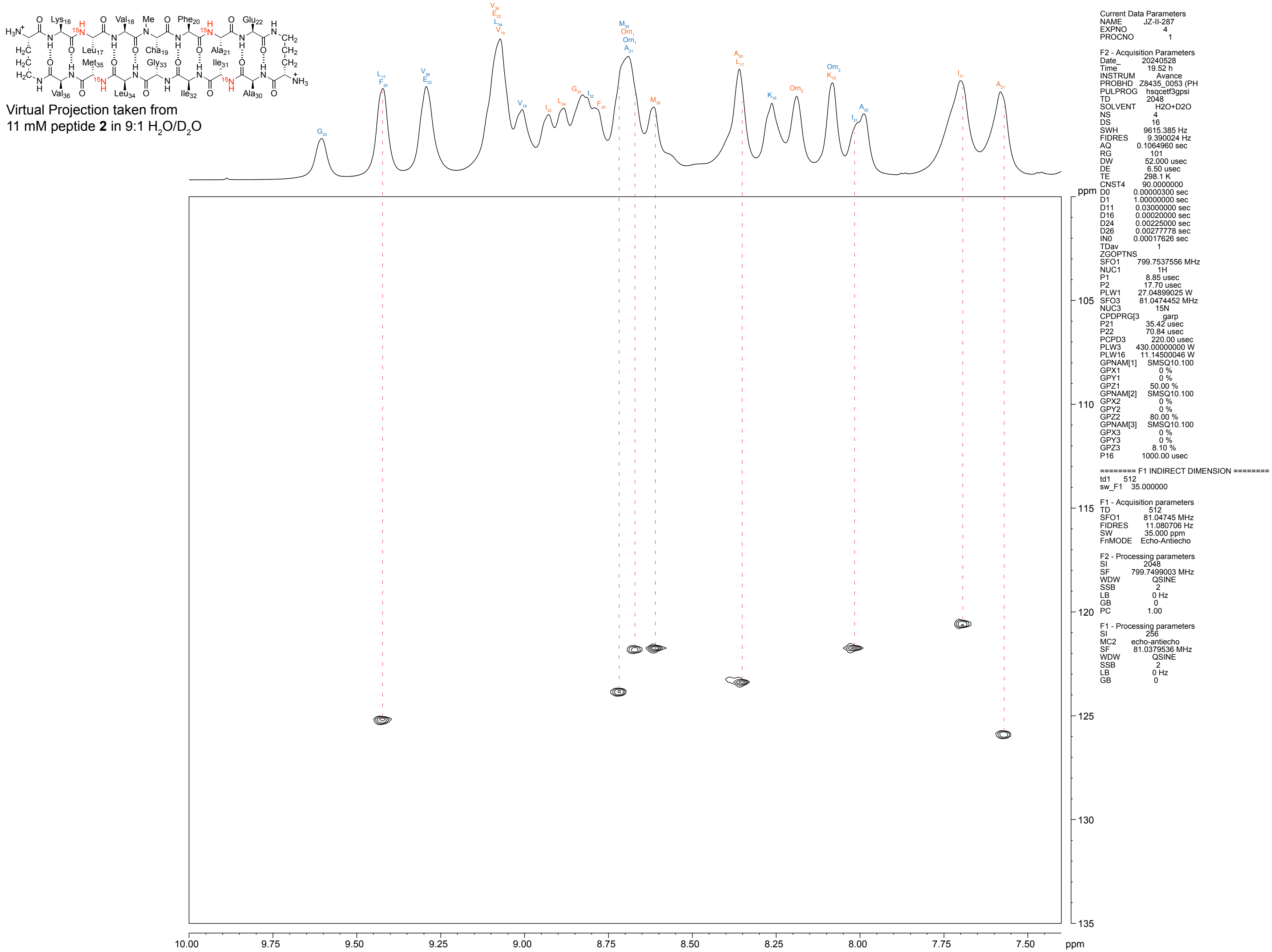
^{1}H NMR stacked plot of 11.0 mM peptide **2** and **2c** showing key Hα resonances that disappear with deuteration in D_2O at 298 K. Peptide **2** taken at 600 MHz and peptide **2c** taken at 800 MHz



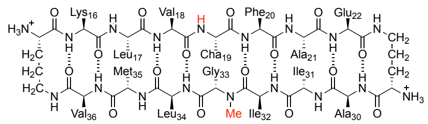
^{1}H,^{15}N HSQC spectrum of 1.0 mM peptide **2d** in 9:1 H_2O/D_2O at 800 MHz and 298 K with WATERGATE suppression of the HOD peak



$^1H,^{15}N$ HSQC spectrum of 11.0 mM peptide **2d** in 9:1 H_2O/D_2O at 800 MHz and 298 K with WATERGATE suppression of the HOD peak



^{1}H NMR spectrum of 1.0 mM peptide **3a** in D_2O at 600 MHz and 298 K

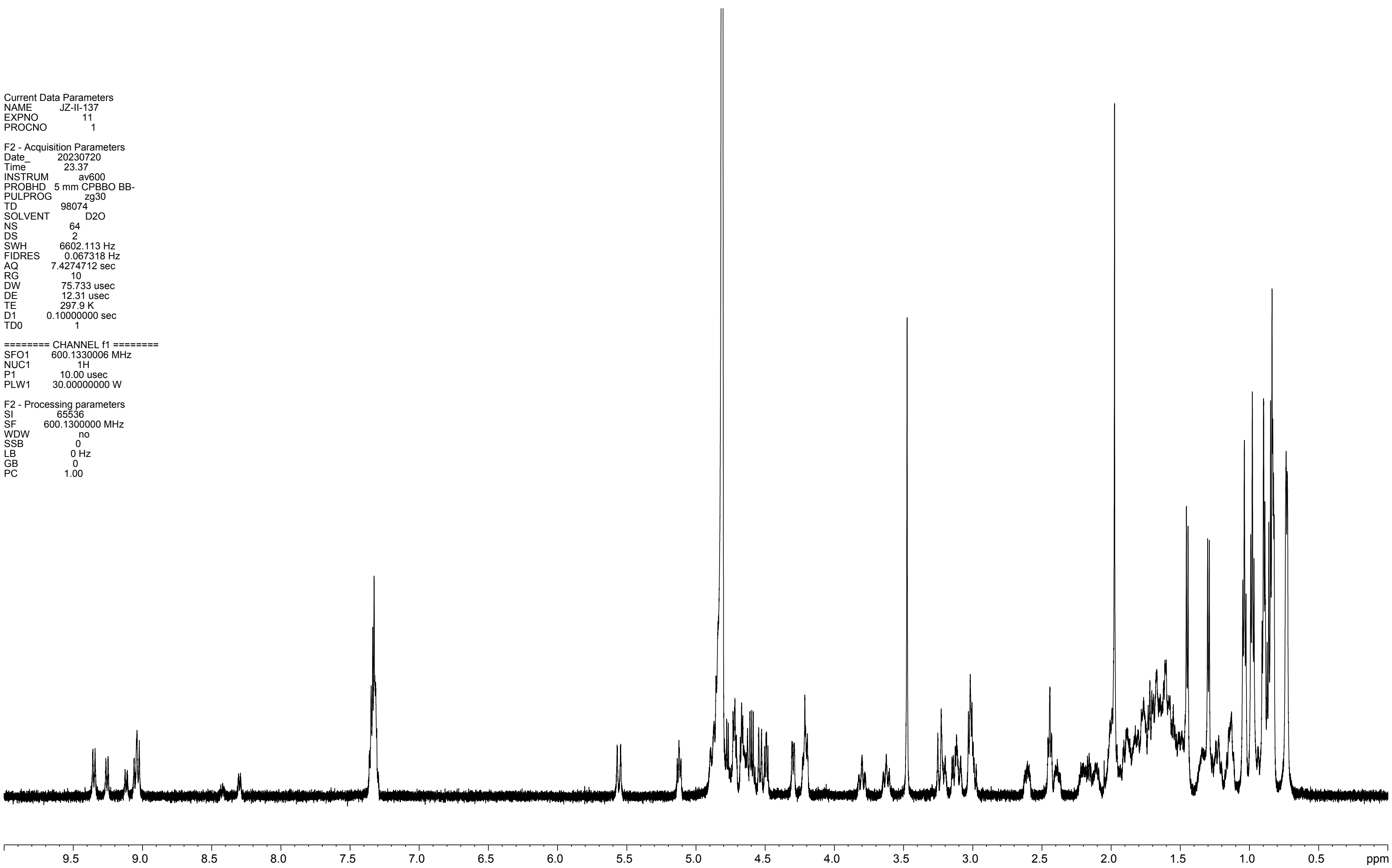


Current Data Parameters
NAME JZ-II-137
EXPNO 11
PROCNO 1

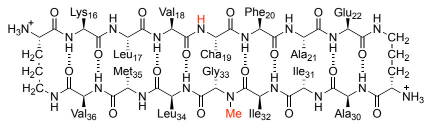
F2 - Acquisition Parameters
Date_ 20230720
Time 23.37
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 64
DS 2
SWH 6602.113 Hz
FIDRES 0.067318 Hz
AQ 7.4274712 sec
RG 10
DW 75.733 usec
DE 12.31 usec
TE 297.9 K
D1 0.10000000 sec
TD0 1

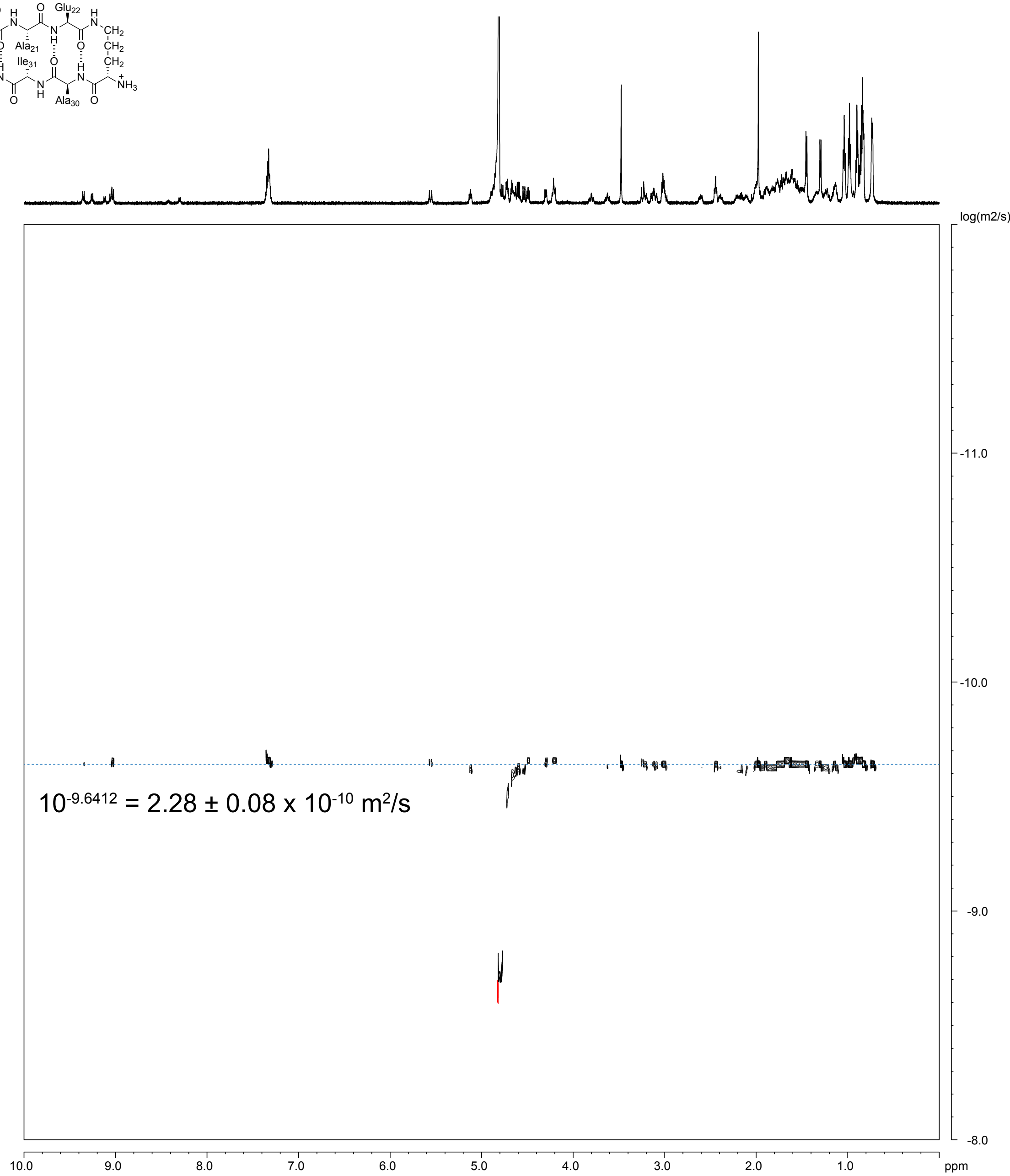
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1300000 MHz
WDW no
SSB 0
LB 0 Hz
GB 0
PC 1.00



^{1}H NMR DOSY spectrum of 1.0 mM peptide **3a** in D_2O at 600 MHz and 298 K





Current Data Parameters

NAME	JZ-II-137
EXPNO	17
PROCNO	1

F2 - Acquisition Parameters

Date_	20230721
Time	0.09
INSTRUM	av600
PROBHD	5 mm CPBBO BB-
PULPROG	ledbpgp2s
TD	98074
SOLVENT	D2O
NS	8
DS	4
SWH	6602.113 Hz
FIDRES	0.067318 Hz
AQ	7.4274712 sec
RG	57
DW	75.733 usec
DE	17.67 usec
TE	298.0 K
D1	5.00000000 sec
D16	0.00020000 sec
D20	0.07500000 sec
D21	0.00500000 sec

======== CHANNEL f1 ========

SFO1	600.1330006 MHz
NUC1	1H
P1	9.75 usec
P2	19.50 usec
PLW1	30.00000000 W

====== GRADIENT CHANNEL =====

GPNAM[6]	SMSQ10.100
GPNAM[7]	SMSQ10.100
GPNAM[8]	SMSQ10.100
GPZ6	100.00 %
GPZ7	-17.13 %
GPZ8	-13.17 %
P19	600.00 usec
P30	1400.00 usec

F1 - Acquisition parameters

TD	16
SFO1	600.1328 MHz
FIDRES	751.201904 Hz
SW	10.014 ppm
FnMODE	QF

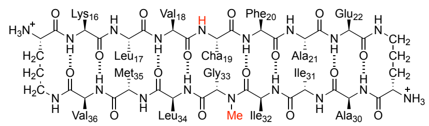
F2 - Processing parameters

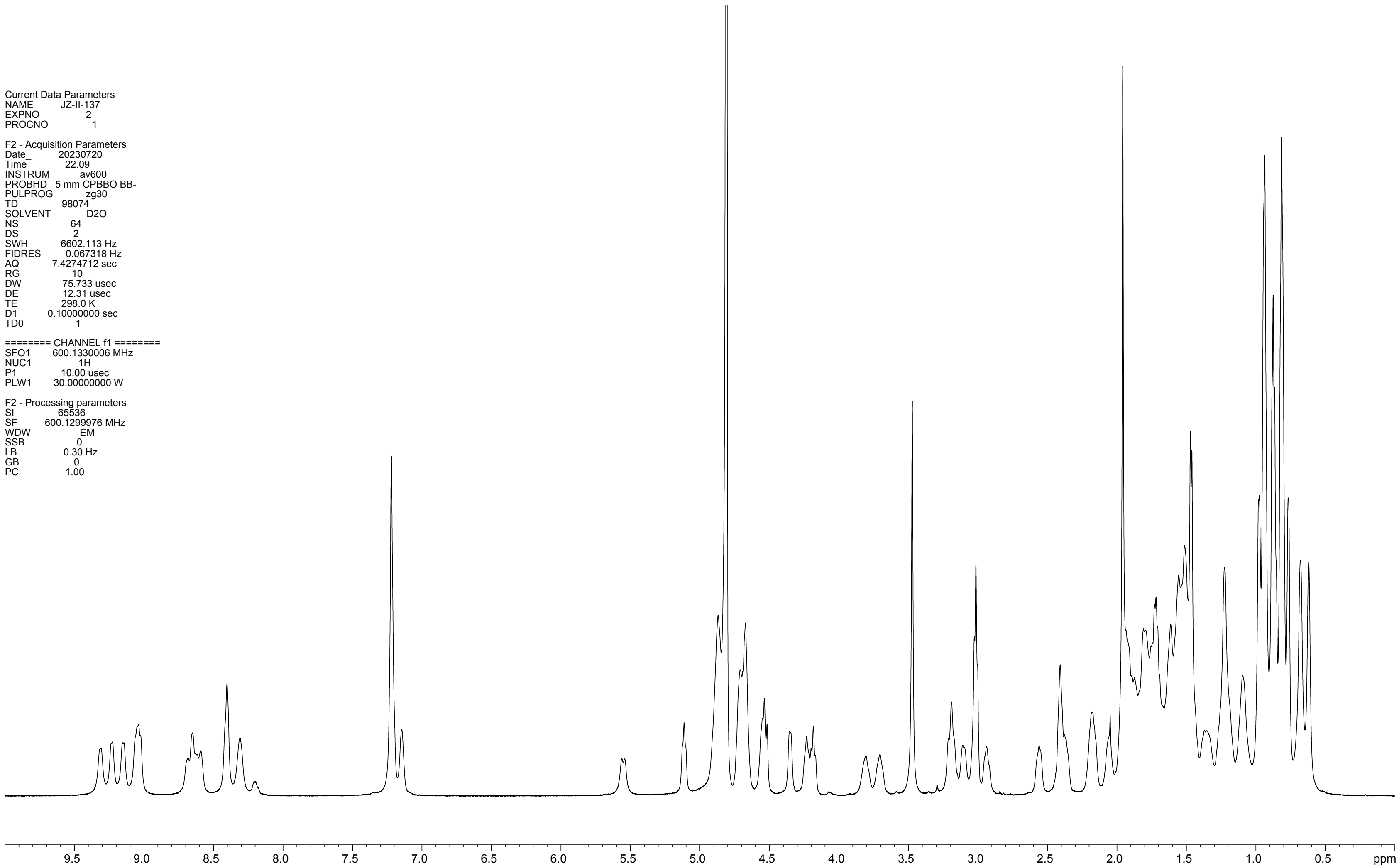
SI	65536
SF	600.1300054 MHz
WDW	EM
SSB	0
LB	5.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

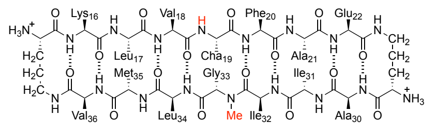
SI	256
MC2	QF
SF	600.1376011 MHz
WDW	no
SSB	0
LB	0 Hz
GB	0

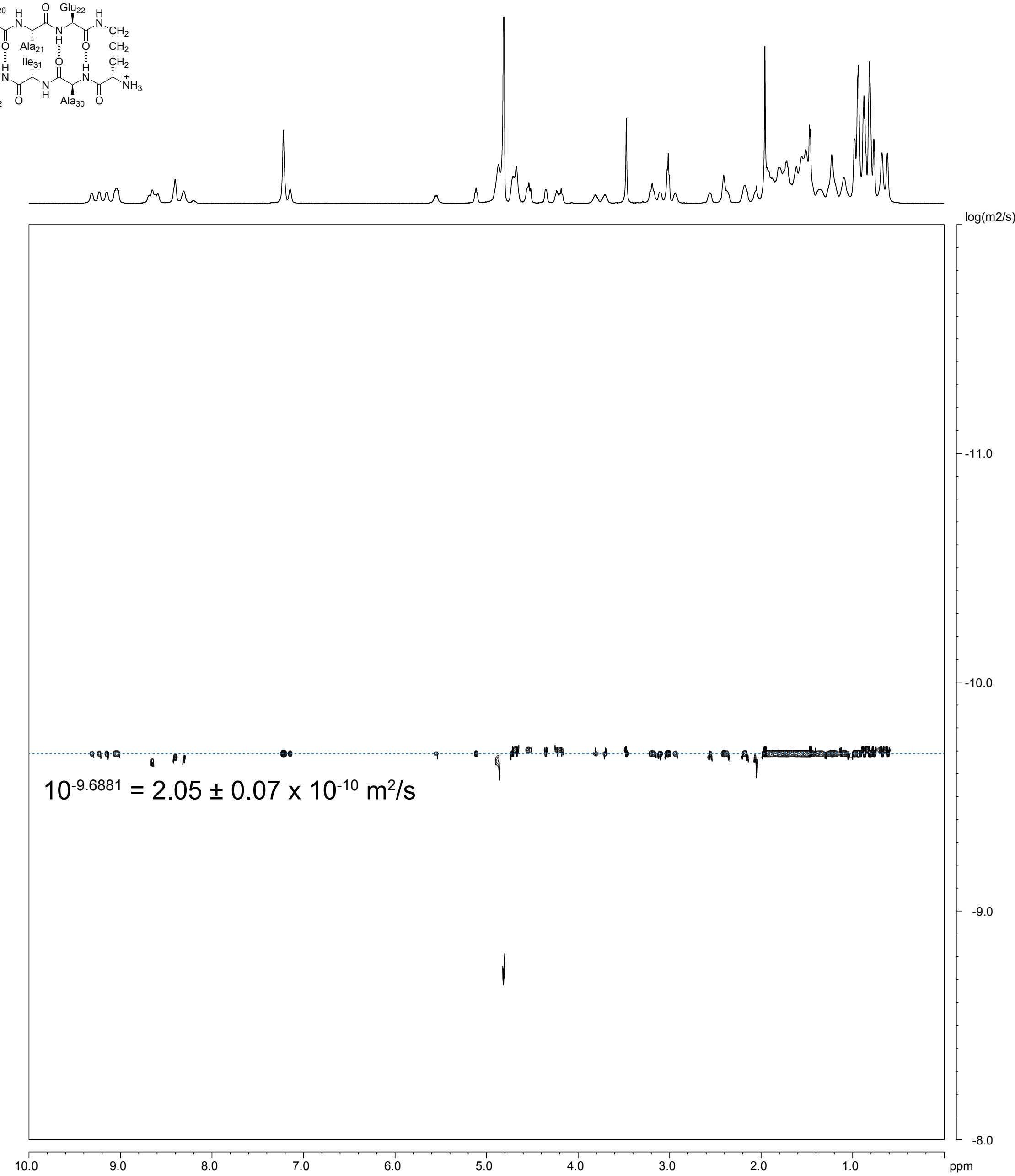
^{1}H NMR spectrum of 8.0 mM peptide **3a** in D_2O at 600 MHz and 298 K





^{1}H NMR DOSY spectrum of 8.0 mM peptide **3a** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME JZ-II-137
EXPNO 7
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230720
Time 22.53
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 6602.113 Hz
FIDRES 0.067318 Hz
AQ 7.4274712 sec
RG 32
DW 75.733 usec
DE 16.87 usec
TE 298.0 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 11.00 usec
P2 22.00 usec
PLW1 30.00000000 W

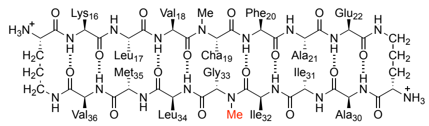
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 1400.00 usec

F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299971 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 256
MC2 QF
SF 600.1376103 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

^{1}H NMR spectrum of 1.0 mM peptide **3b** in D_2O at 600 MHz and 298 K

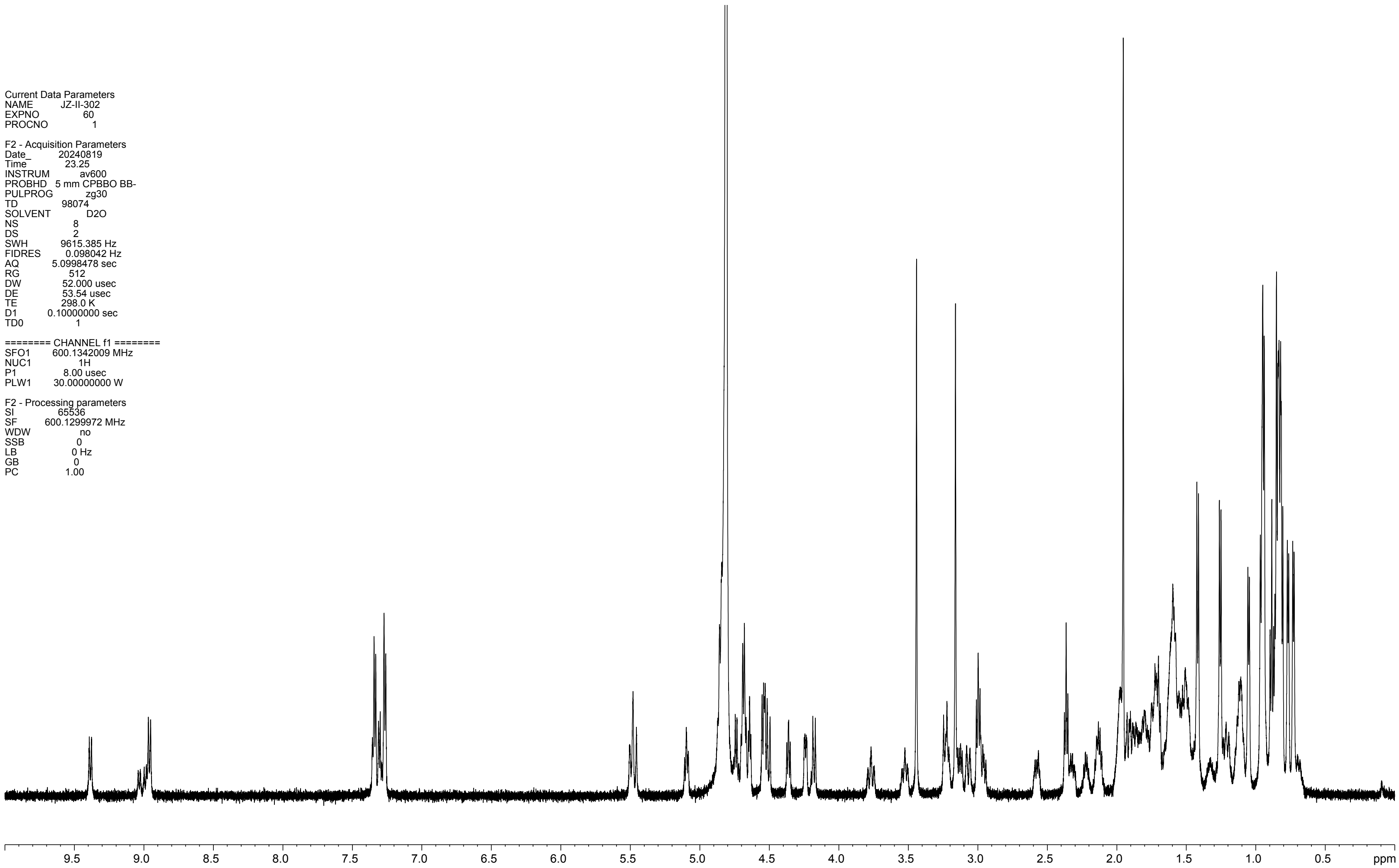


Current Data Parameters
NAME JZ-II-302
EXPNO 60
PROCNO 1

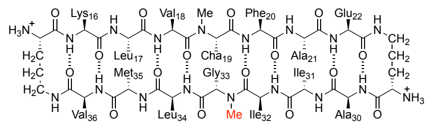
F2 - Acquisition Parameters
Date_ 20240819
Time 23.25
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 8
DS 2
SWH 9615.385 Hz
FIDRES 0.098042 Hz
AQ 5.0998478 sec
RG 512
DW 52.000 usec
DE 53.54 usec
TE 298.0 K
D1 0.10000000 sec
TD0 1

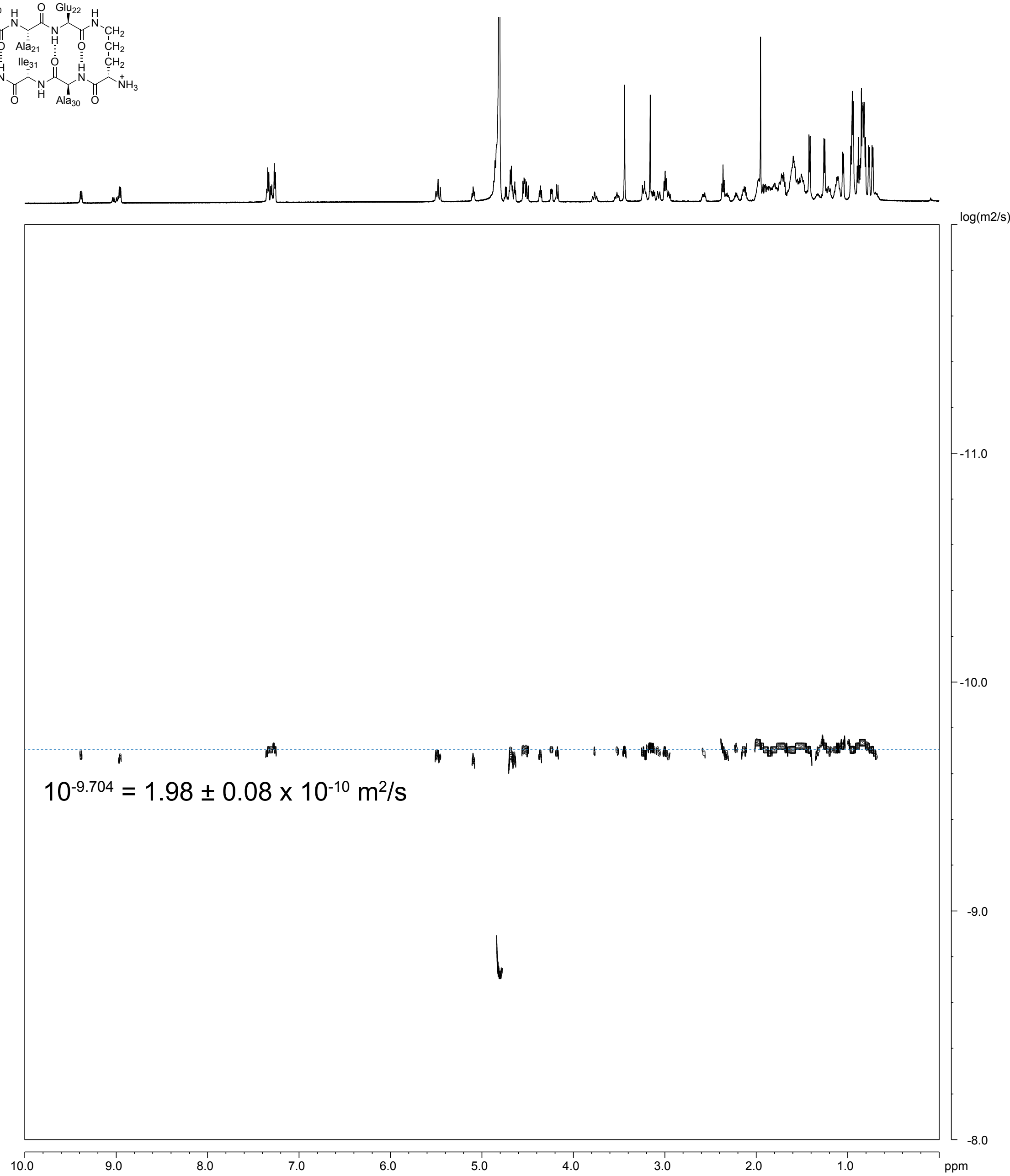
======== CHANNEL f1 ========
SFO1 600.1342009 MHz
NUC1 1H
P1 8.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1299972 MHz
WDW no
SSB 0
LB 0 Hz
GB 0
PC 1.00



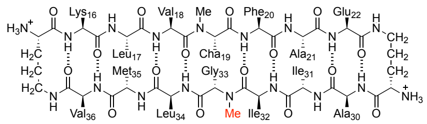
^{1}H NMR DOSY spectrum of 1.0 mM peptide **3b** in D_2O at 600 MHz and 298 K

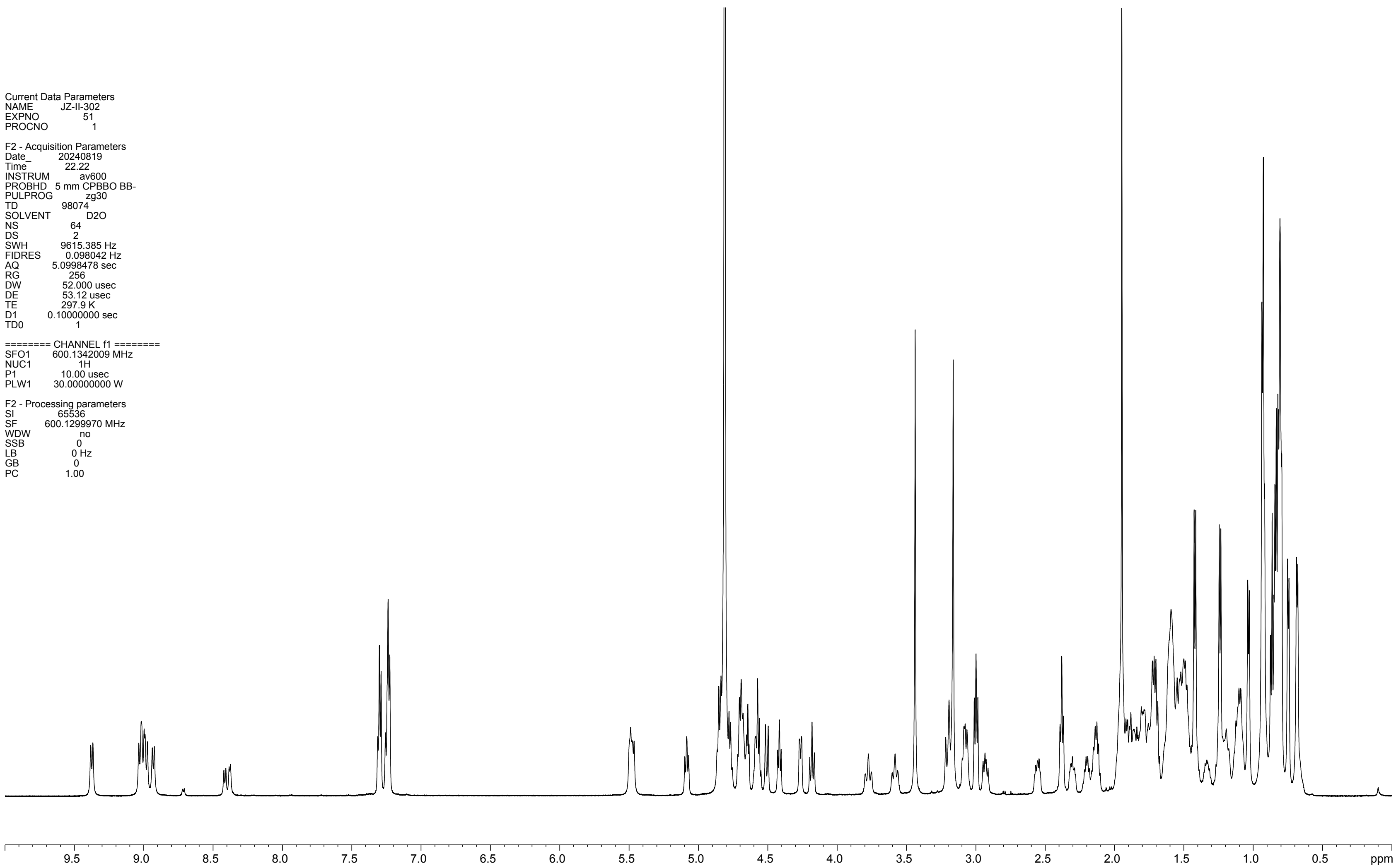




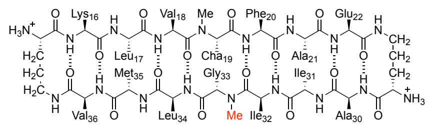
Current Data Parameters	
NAME	JZ-II-302
EXPNO	65
PROCNO	1
F2 - Acquisition Parameters	
Date_	20240819
Time	23.54
INSTRUM	av600
PROBHD	5 mm CPBBO BB-
PULPROG	ledbpgp2s
TD	98074
SOLVENT	D2O
NS	8
DS	4
SWH	9615.385 Hz
FIDRES	0.098042 Hz
AQ	5.0998478 sec
RG	28.5
DW	52.000 usec
DE	17.63 usec
TE	297.9 K
D1	5.00000000 sec
D16	0.00020000 sec
D20	0.07500000 sec
D21	0.00500000 sec
======== CHANNEL f1 ========	
SFO1	600.1342009 MHz
NUC1	1H
P1	8.00 usec
P2	16.00 usec
PLW1	30.00000000 W
====== GRADIENT CHANNEL =====	
GPNAM[6]	SMSQ10.100
GPNAM[7]	SMSQ10.100
GPNAM[8]	SMSQ10.100
GPZ6	100.00 %
GPZ7	-17.13 %
GPZ8	-13.17 %
P19	600.00 usec
P30	1600.00 usec
F1 - Acquisition parameters	
TD	16
SFO1	600.1328 MHz
FIDRES	751.201904 Hz
SW	10.014 ppm
FnMODE	QF
F2 - Processing parameters	
SI	65536
SF	600.1299973 MHz
WDW	EM
SSB	0
LB	5.00 Hz
GB	0
PC	1.00
F1 - Processing parameters	
SI	256
MC2	QF
SF	600.1375822 MHz
WDW	no
SSB	0
LB	0 Hz
GB	0

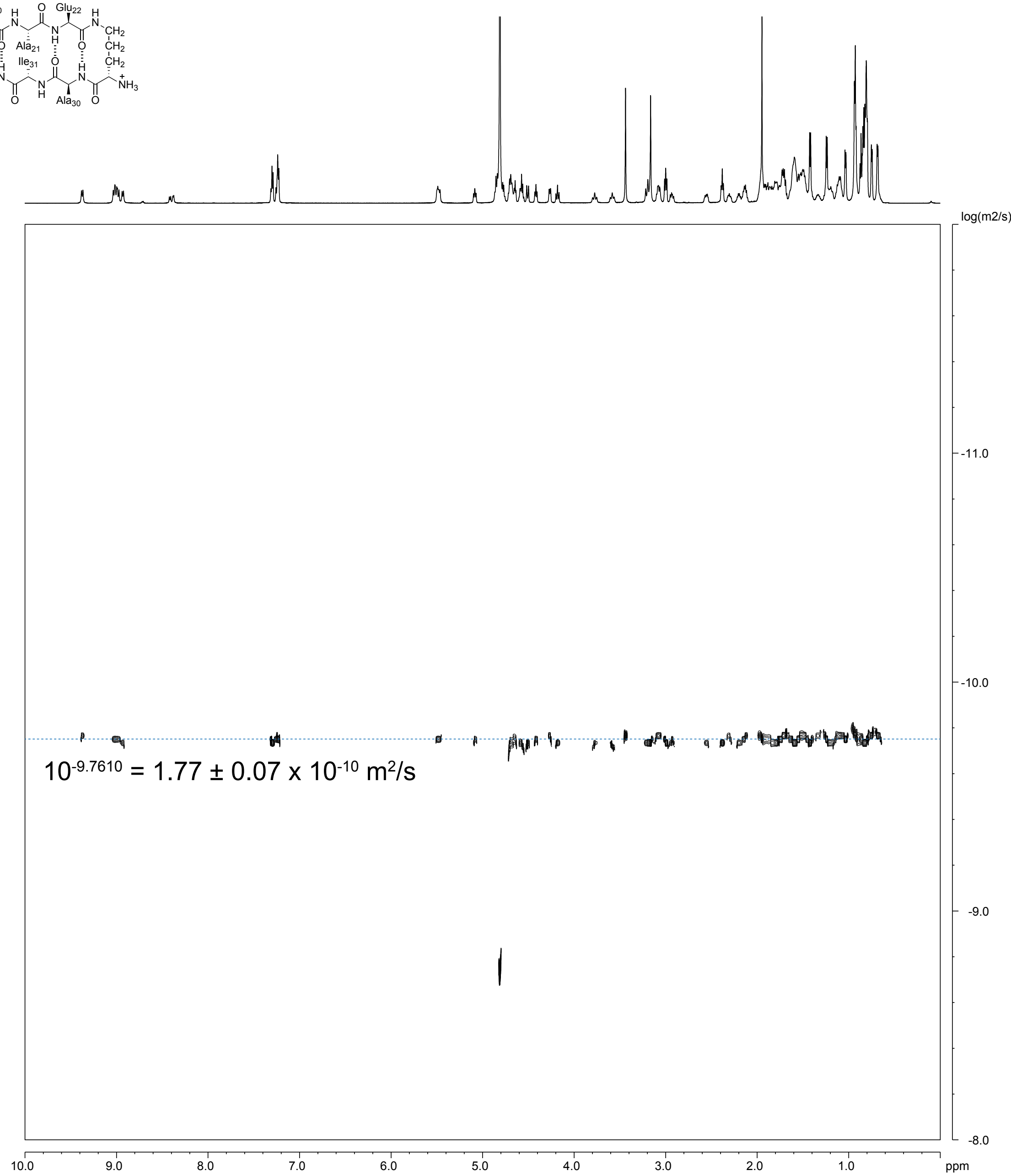
^1H NMR spectrum of 4.0 mM peptide **3b** in D_2O at 600 MHz and 298 K





^{1}H NMR DOSY spectrum of 4.0 mM peptide **3b** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME JZ-II-302
EXPNO 55
PROCNO 1

F2 - Acquisition Parameters
Date_ 20240819
Time 22.45
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 9615.385 Hz
FIDRES 0.098042 Hz
AQ 5.0998478 sec
RG 32
DW 52.000 usec
DE 17.63 usec
TE 298.0 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1342009 MHz
NUC1 1H
P1 8.00 usec
P2 16.00 usec
PLW1 30.00000000 W

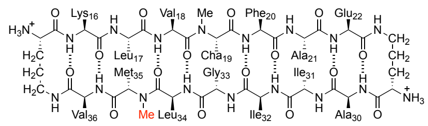
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 1900.00 usec

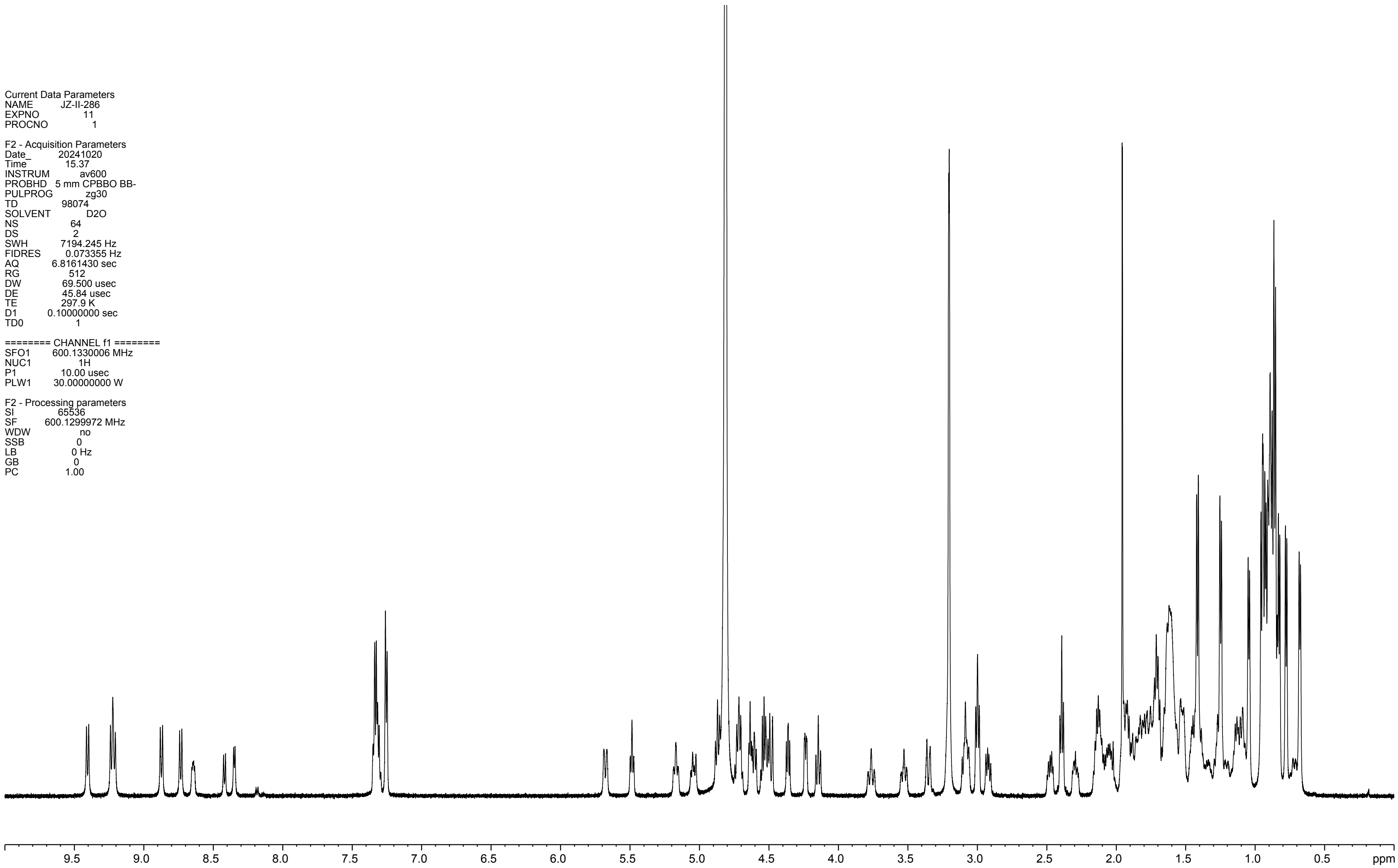
F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299972 MHz
WDW EM
SSB 0
LB 5.00 Hz
GB 0
PC 1.00

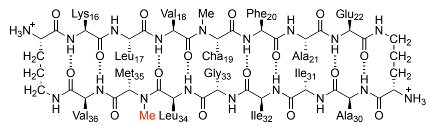
F1 - Processing parameters
SI 256
MC2 QF
SF 600.1375634 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

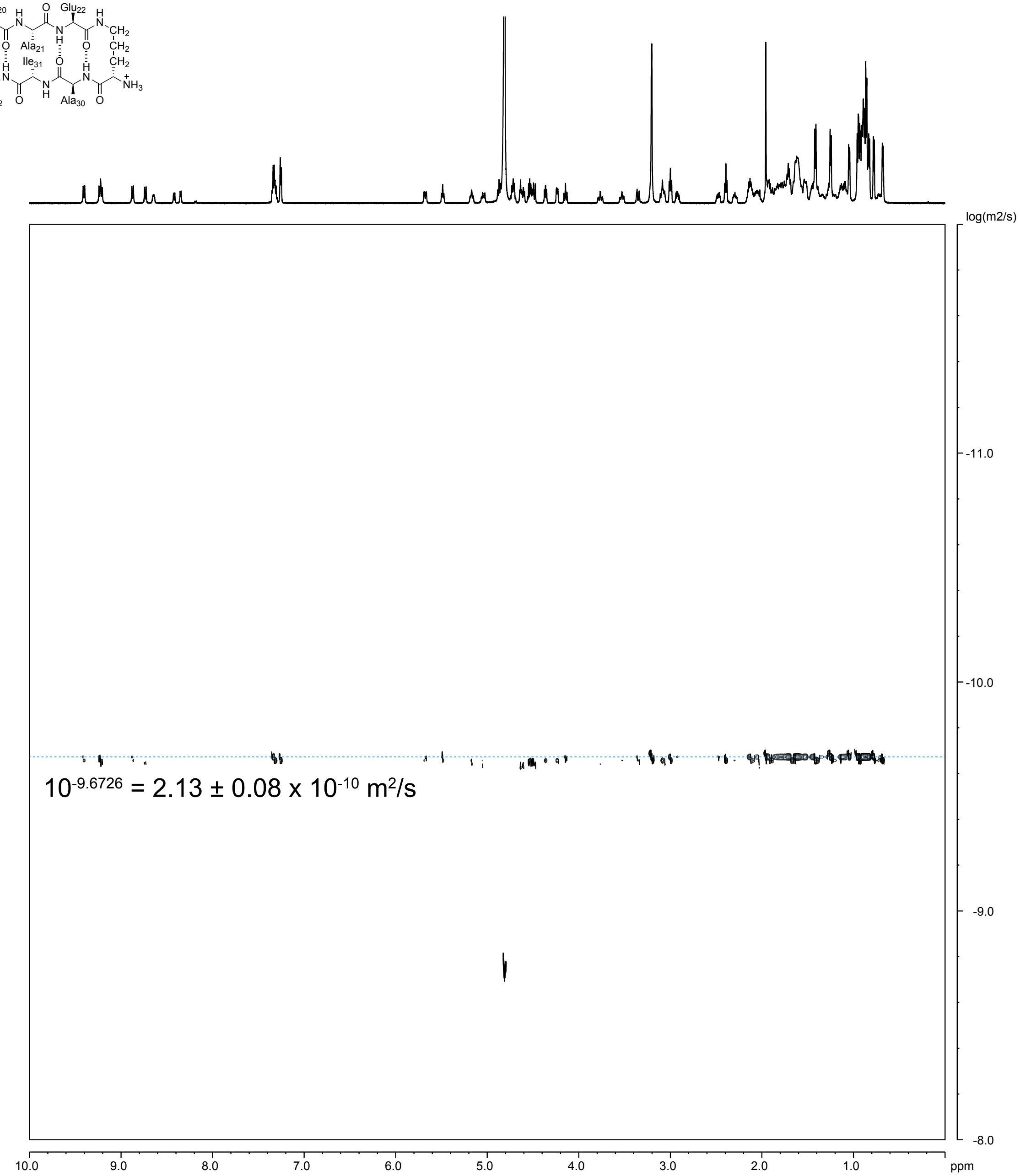
[1]H NMR spectrum of 1.0 mM peptide **3c** in D_2O at 600 MHz and 298 K





^{1}H NMR DOSY spectrum of 1.0 mM peptide **3c** in D_2O at 600 MHz and 298 K



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Current Data Parameters
NAME          JZ-II-286
EXPNO               15
PROCNO               1

F2 - Acquisition Parameters
Date_          20241020
Time              16.12
INSTRUM           av600
PROBHD   5 mm CPBBO BB-
PULPROG       ledbpgp2s
TD                98074
SOLVENT             D2O
NS                    8
DS                    4
SWH         7194.245 Hz
FIDRES      0.073355 Hz
AQ         6.8161430 sec
RG                 50.8
DW           69.500 usec
DE            16.15 usec
TE              298.0 K
D1       5.00000000 sec
D16      0.00020000 sec
D20      0.07500000 sec
D21      0.00500000 sec

======== CHANNEL f1 ========
SFO1     600.1328866 MHz
NUC1                 1H
P1             9.00 usec
P2            18.00 usec
PLW1      30.00000000 W

====== GRADIENT CHANNEL =====
GPNAM[6]     SMSQ10.100
GPNAM[7]     SMSQ10.100
GPNAM[8]     SMSQ10.100
GPZ6           100.00 %
GPZ7           -17.13 %
GPZ8           -13.17 %
P19          600.00 usec
P30         1550.00 usec

F1 - Acquisition parameters
TD                   16
SFO1       600.1328 MHz
FIDRES    751.201904 Hz
SW           10.014 ppm
FnMODE               QF

F2 - Processing parameters
SI                65536
SF       600.1299974 MHz
WDW                  EM
SSB                   0
LB              5.00 Hz
GB                    0
PC                 1.00

F1 - Processing parameters
SI                  256
MC2                  QF
SF       600.1375821 MHz
WDW                  no
SSB                   0
LB                 0 Hz
GB                    0
```
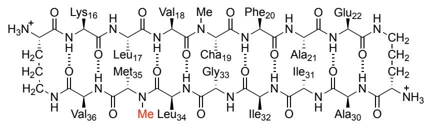
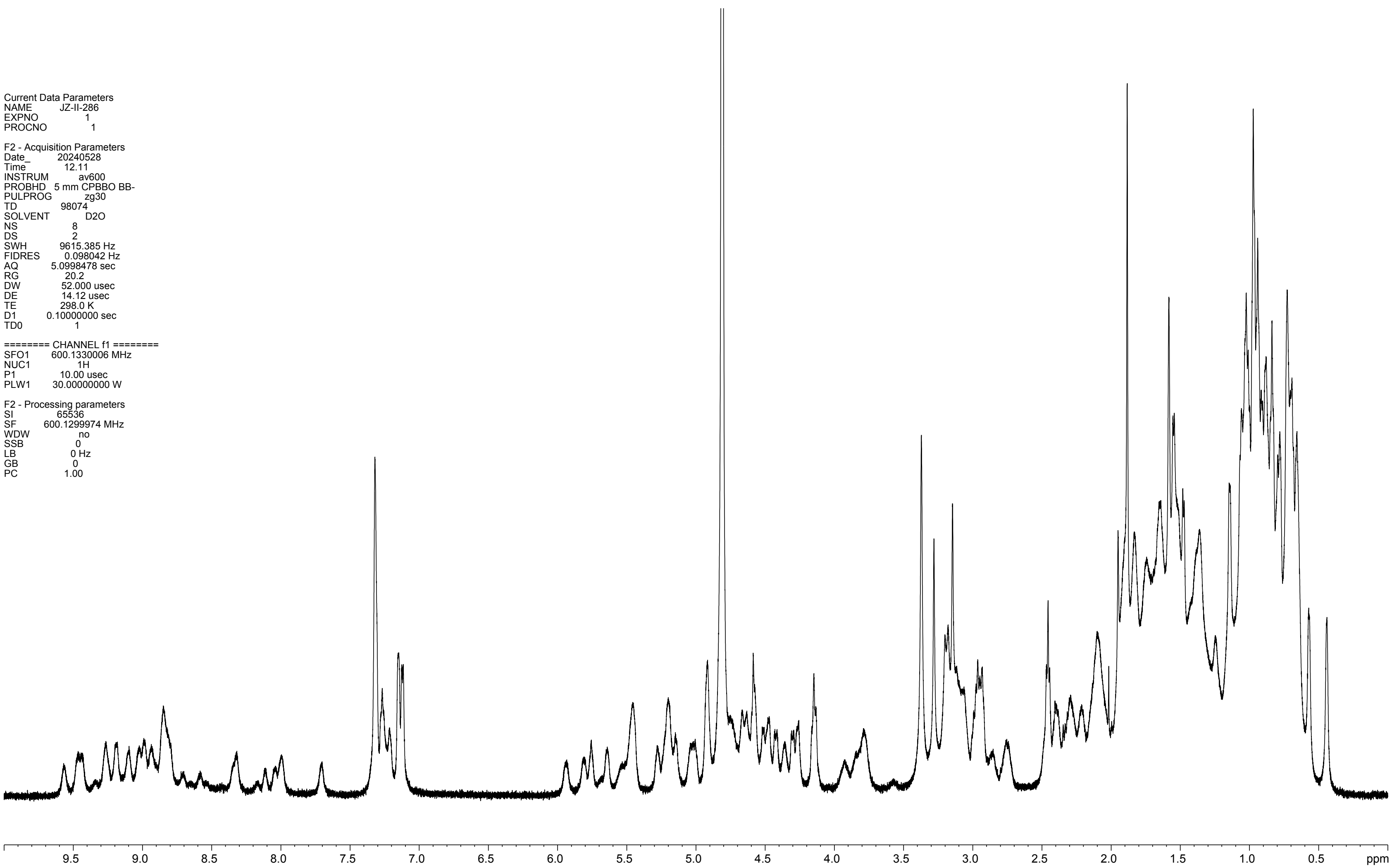
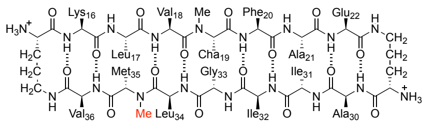
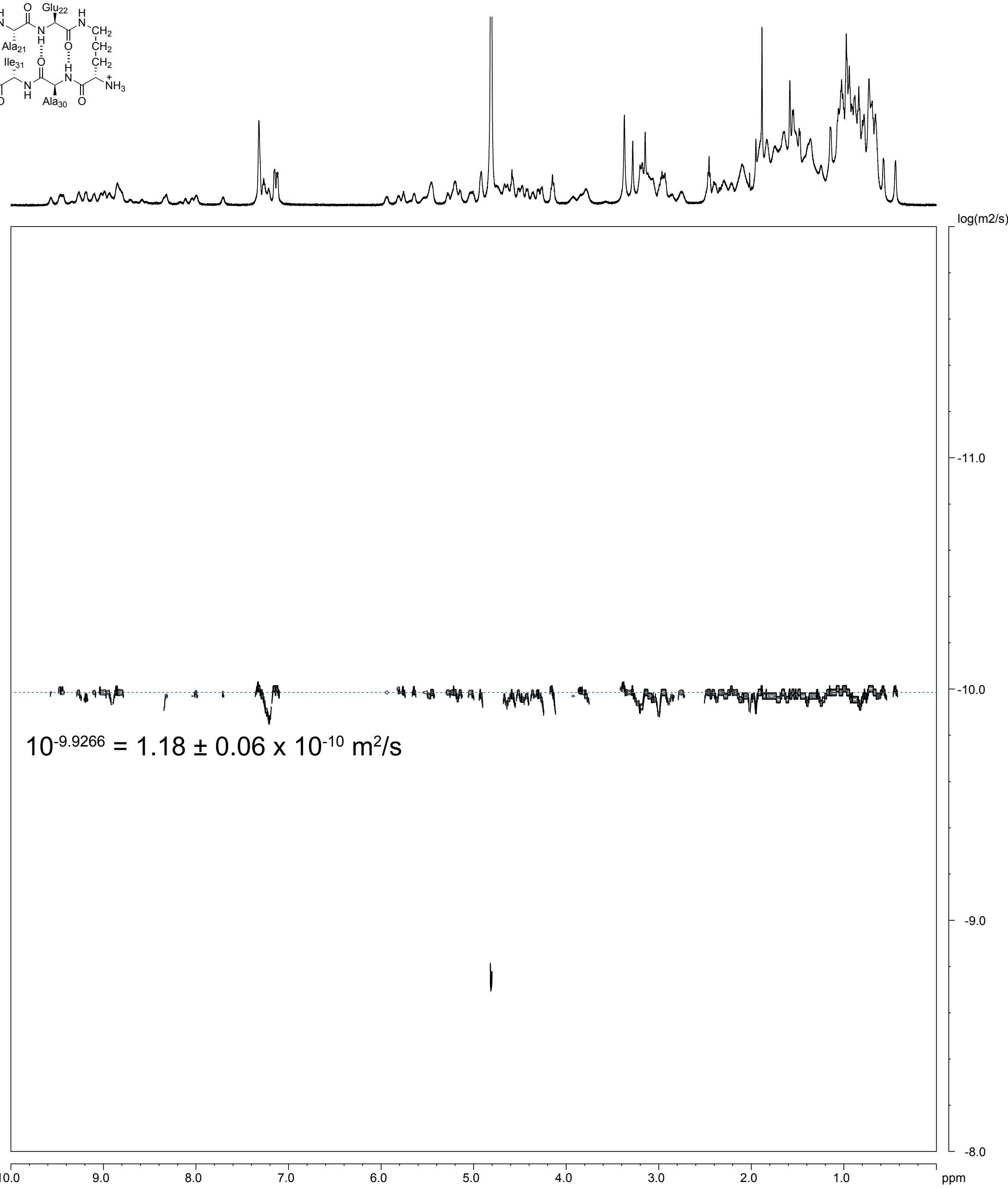

^1H NMR spectrum of 11.0 mM peptide **3c** in D_2O at 600 MHz and 298 K





^{1}H NMR DOSY spectrum of 11.0 mM peptide **3c** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME JZ-II-286
EXPNO 5
PROCNO 1

F2 - Acquisition Parameters
Date_ 20240528
Time 12.31
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 9615.385 Hz
FIDRES 0.098042 Hz
AQ 5.0998478 sec
RG 50.8
DW 52.000 usec
DE 16.36 usec
TE 297.9 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
P2 20.00 usec
PLW1 30.00000000 W

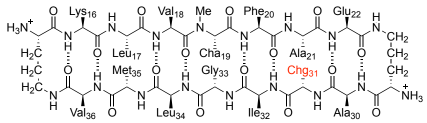
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 2300.00 usec

F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299976 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 256
MC2 QF
SF 600.1375728 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

[1]H NMR spectrum of 1.0 mM peptide **4a** in D_2O at 600 MHz and 298 K

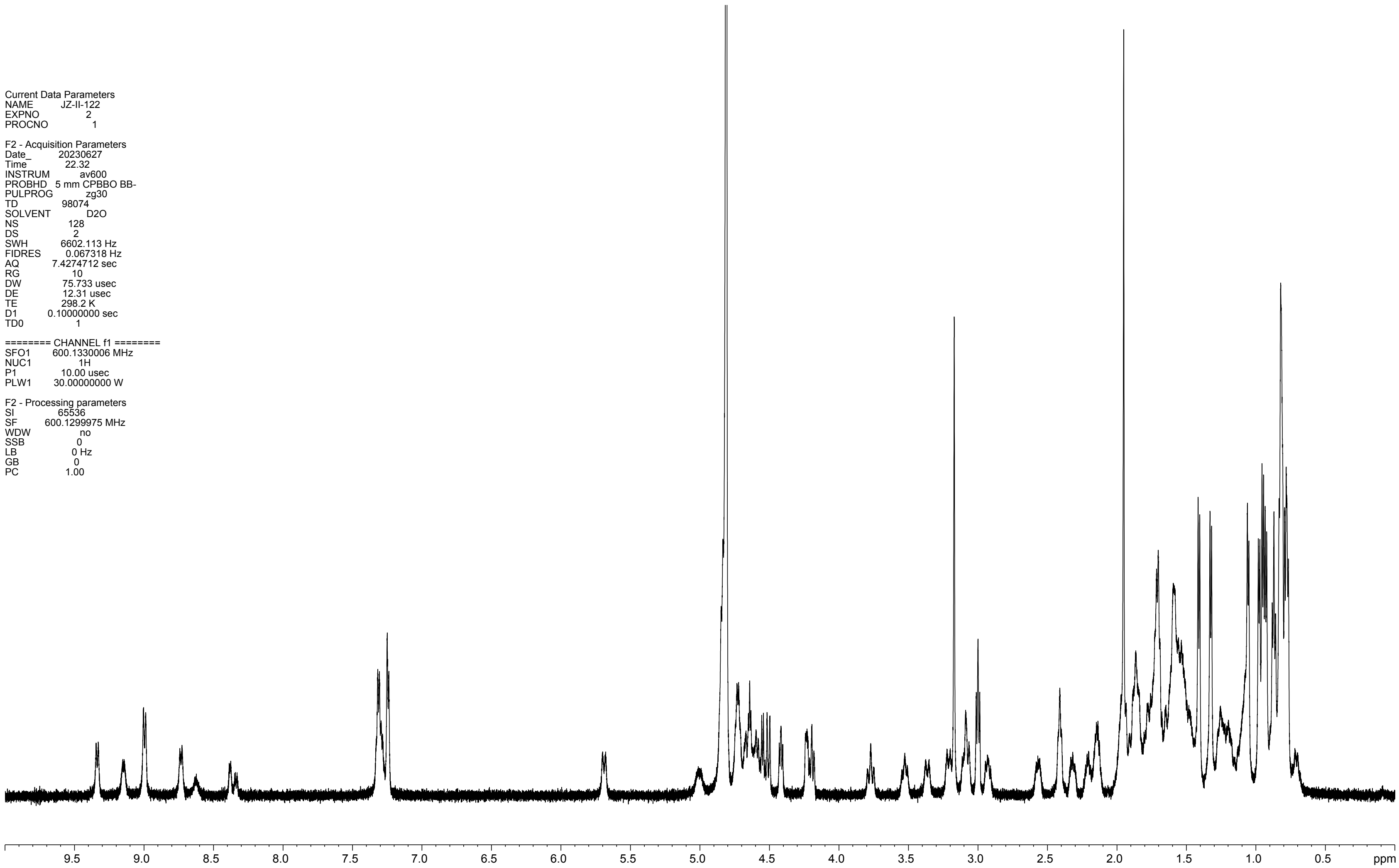


Current Data Parameters
NAME JZ-II-122
EXPNO 2
PROCNO 1

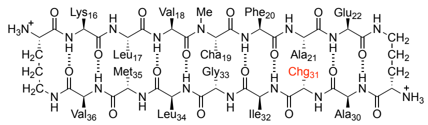
F2 - Acquisition Parameters
Date_ 20230627
Time 22.32
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 128
DS 2
SWH 6602.113 Hz
FIDRES 0.067318 Hz
AQ 7.4274712 sec
RG 10
DW 75.733 usec
DE 12.31 usec
TE 298.2 K
D1 0.10000000 sec
TD0 1

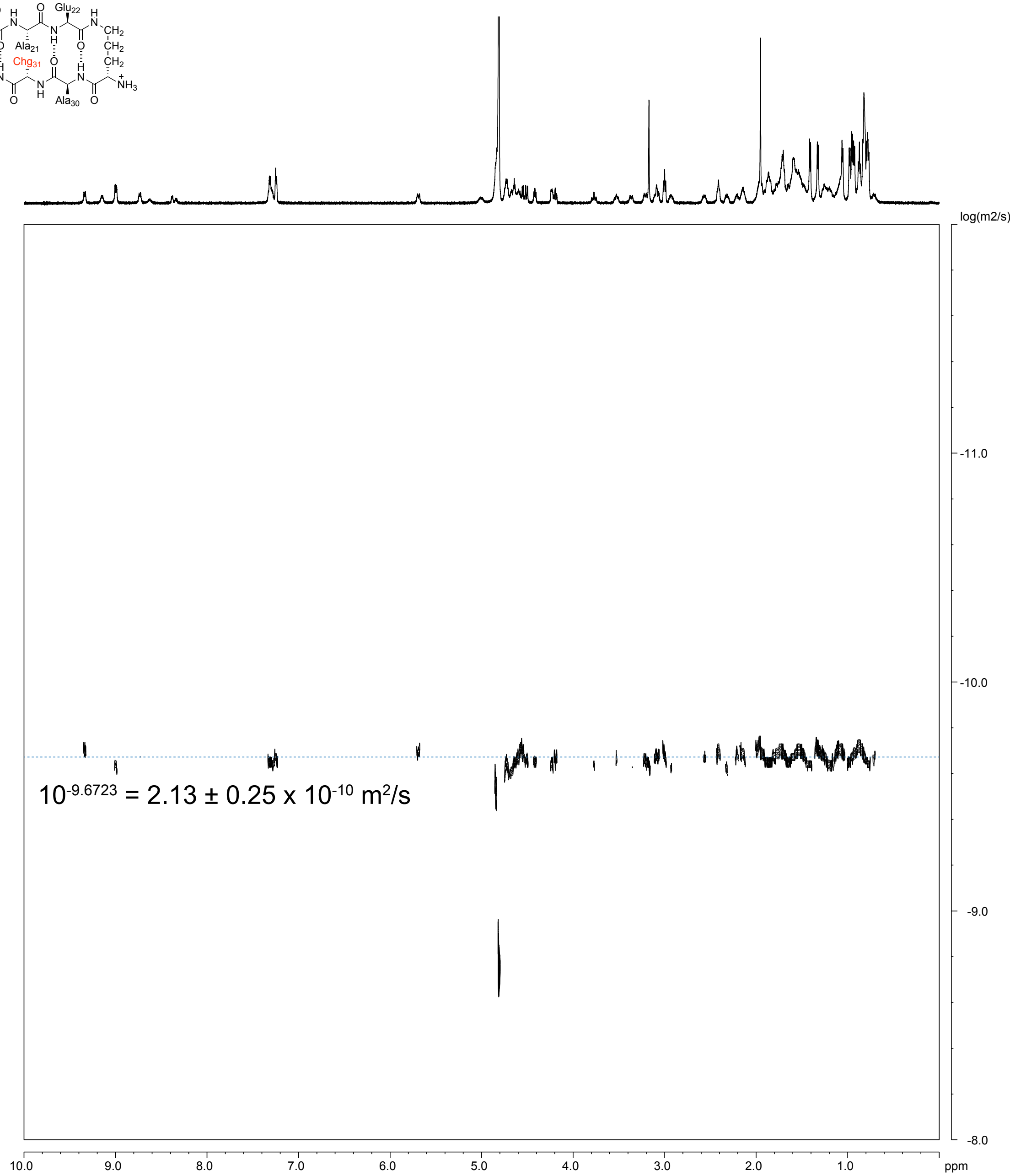
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1299975 MHz
WDW no
SSB 0
LB 0 Hz
GB 0
PC 1.00



^{1}H NMR DOSY spectrum of 1.0 mM peptide **4a** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME JZ-II-122
EXPNO 8
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230627
Time 23.54
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 6602.113 Hz
FIDRES 0.067318 Hz
AQ 7.4274712 sec
RG 71.8
DW 75.733 usec
DE 18.78 usec
TE 298.2 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
P2 16.00 usec
PLW1 30.00000000 W

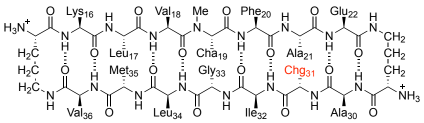
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 1700.00 usec

F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299977 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 256
MC2 QF
SF 600.1375450 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

^{1}H NMR spectrum of 6.0 mM peptide **4a** in D_2O at 600 MHz and 298 K

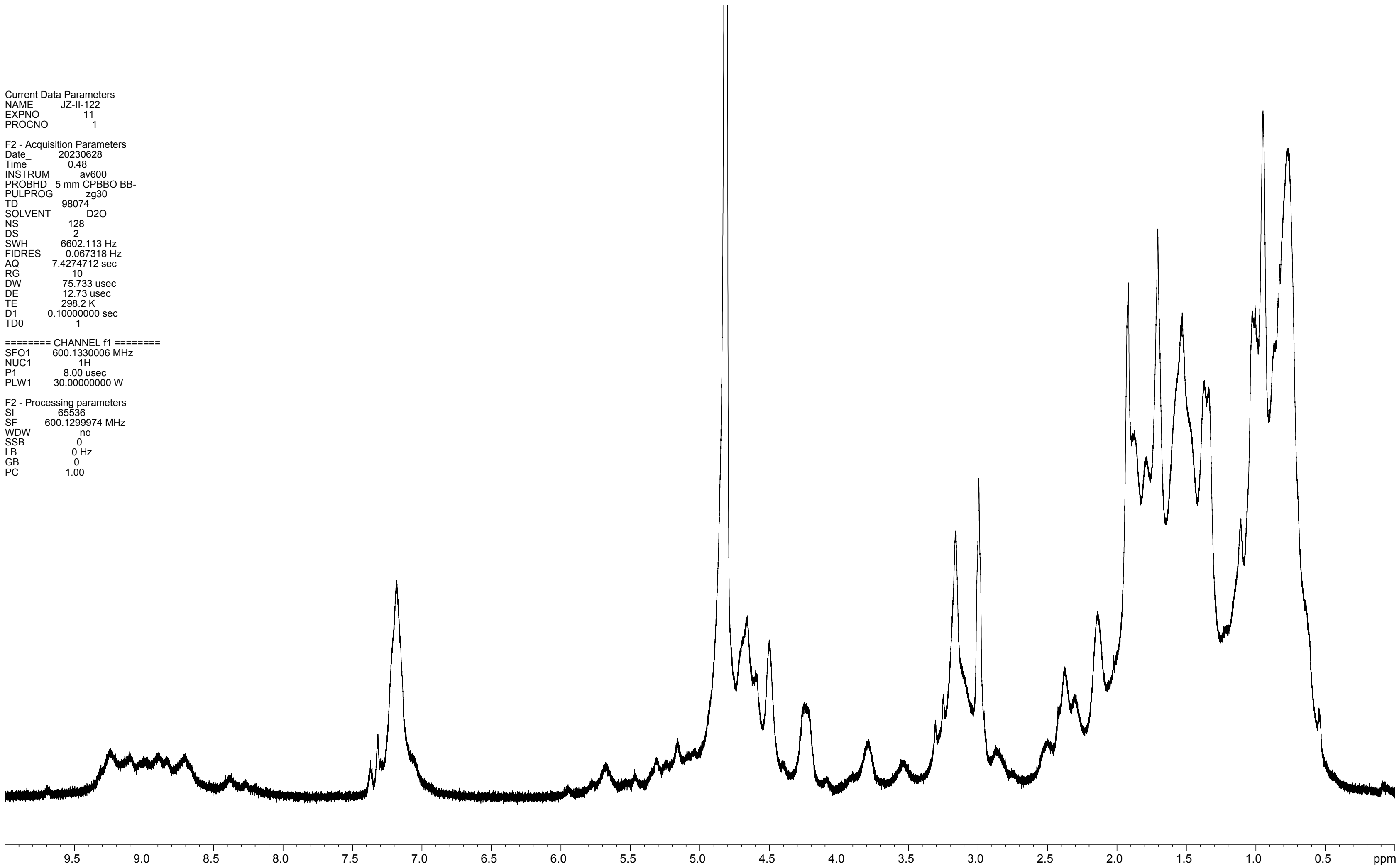


Current Data Parameters
NAME JZ-II-122
EXPNO 11
PROCNO 1

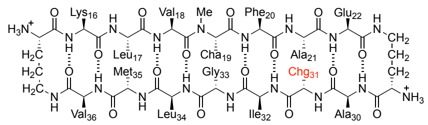
F2 - Acquisition Parameters
Date_ 20230628
Time 0.48
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 128
DS 2
SWH 6602.113 Hz
FIDRES 0.067318 Hz
AQ 7.4274712 sec
RG 10
DW 75.733 usec
DE 12.73 usec
TE 298.2 K
D1 0.10000000 sec
TD0 1

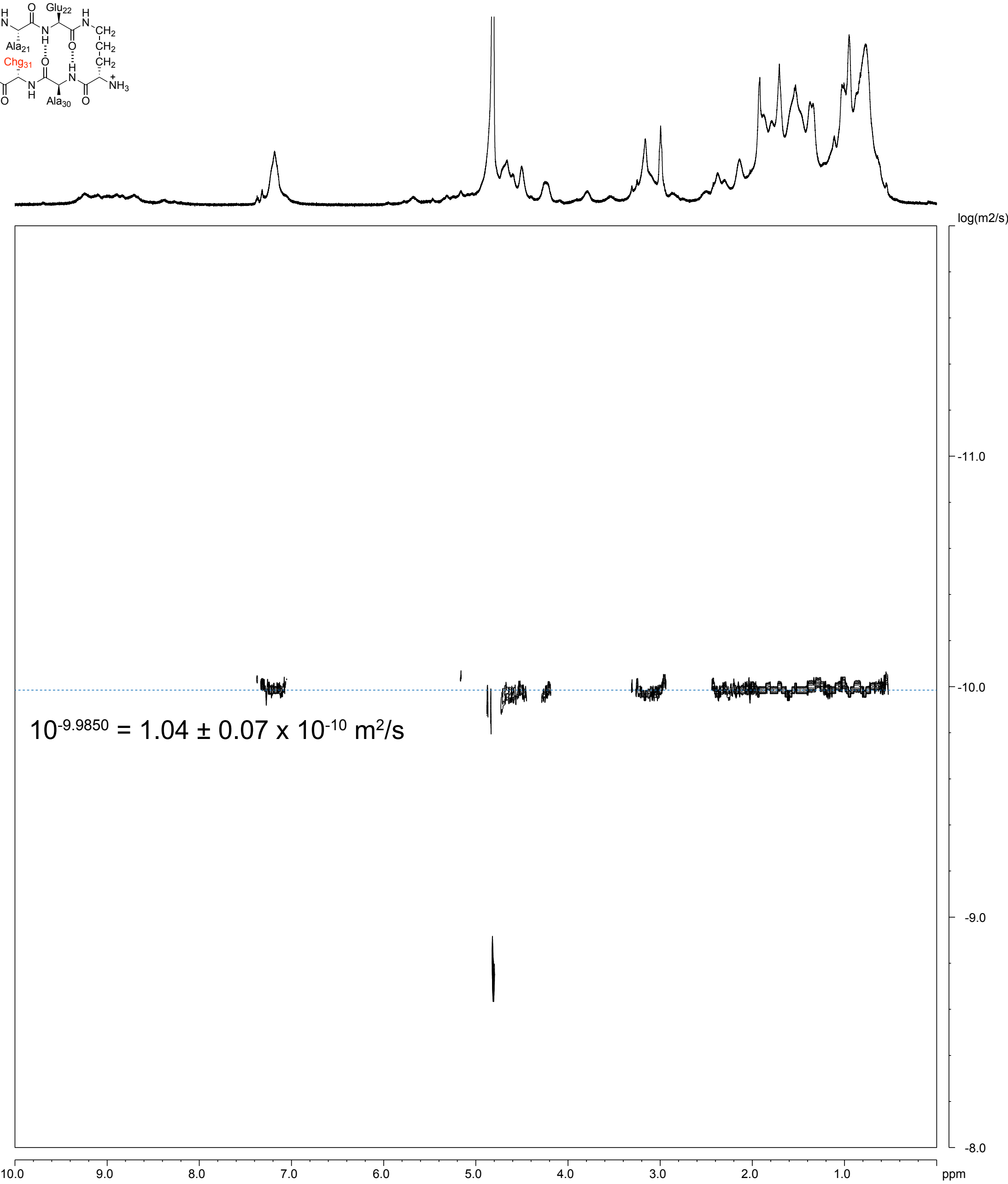
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1299974 MHz
WDW no
SSB 0
LB 0 Hz
GB 0
PC 1.00



^{1}H NMR DOSY spectrum of 6.0 mM peptide **4a** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME JZ-II-122
EXPNO 14
PROCNO 1

F2 - Acquisition Parameters
Date_ 20230628
Time 1.18
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 9615.385 Hz
FIDRES 0.098042 Hz
AQ 5.0998478 sec
RG 57
DW 52.000 usec
DE 17.63 usec
TE 298.2 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 8.00 usec
P2 16.00 usec
PLW1 30.00000000 W

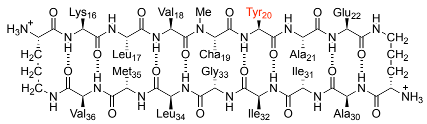
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 2400.00 usec

F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299974 MHz
WDW EM
SSB 0
LB 2.00 Hz
GB 0
PC 1.00

F1 - Processing parameters
SI 256
MC2 QF
SF 600.1375542 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

^1H NMR spectrum of 1.0 mM peptide **4b** in D_2O at 600 MHz and 298 K

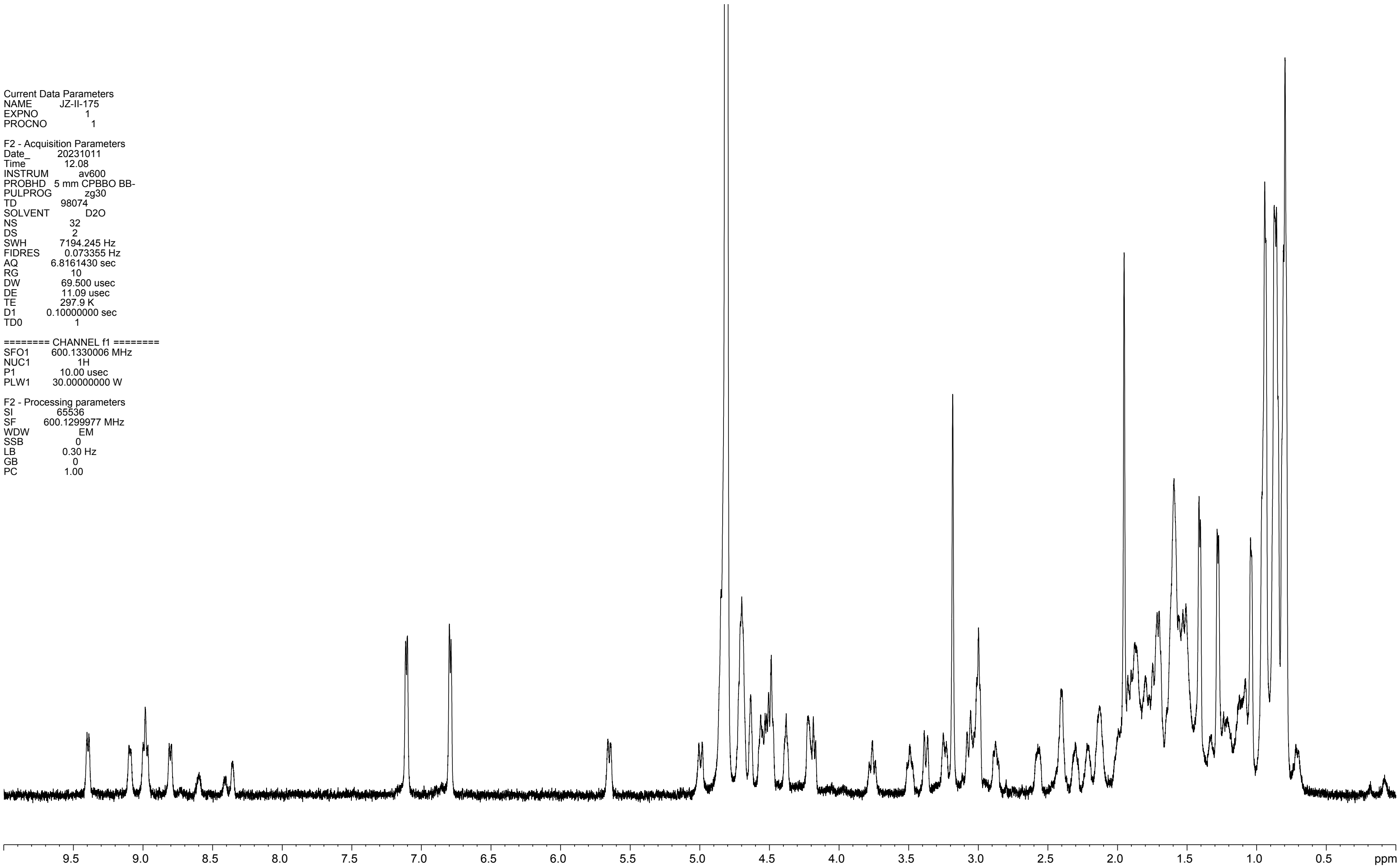


Current Data Parameters
NAME JZ-II-175
EXPNO 1
PROCNO 1

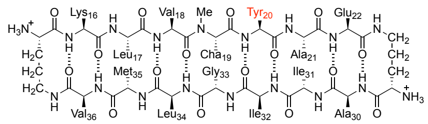
F2 - Acquisition Parameters
Date_ 20231011
Time 12.08
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 32
DS 2
SWH 7194.245 Hz
FIDRES 0.073355 Hz
AQ 6.8161430 sec
RG 10
DW 69.500 usec
DE 11.09 usec
TE 297.9 K
D1 0.10000000 sec
TD0 1

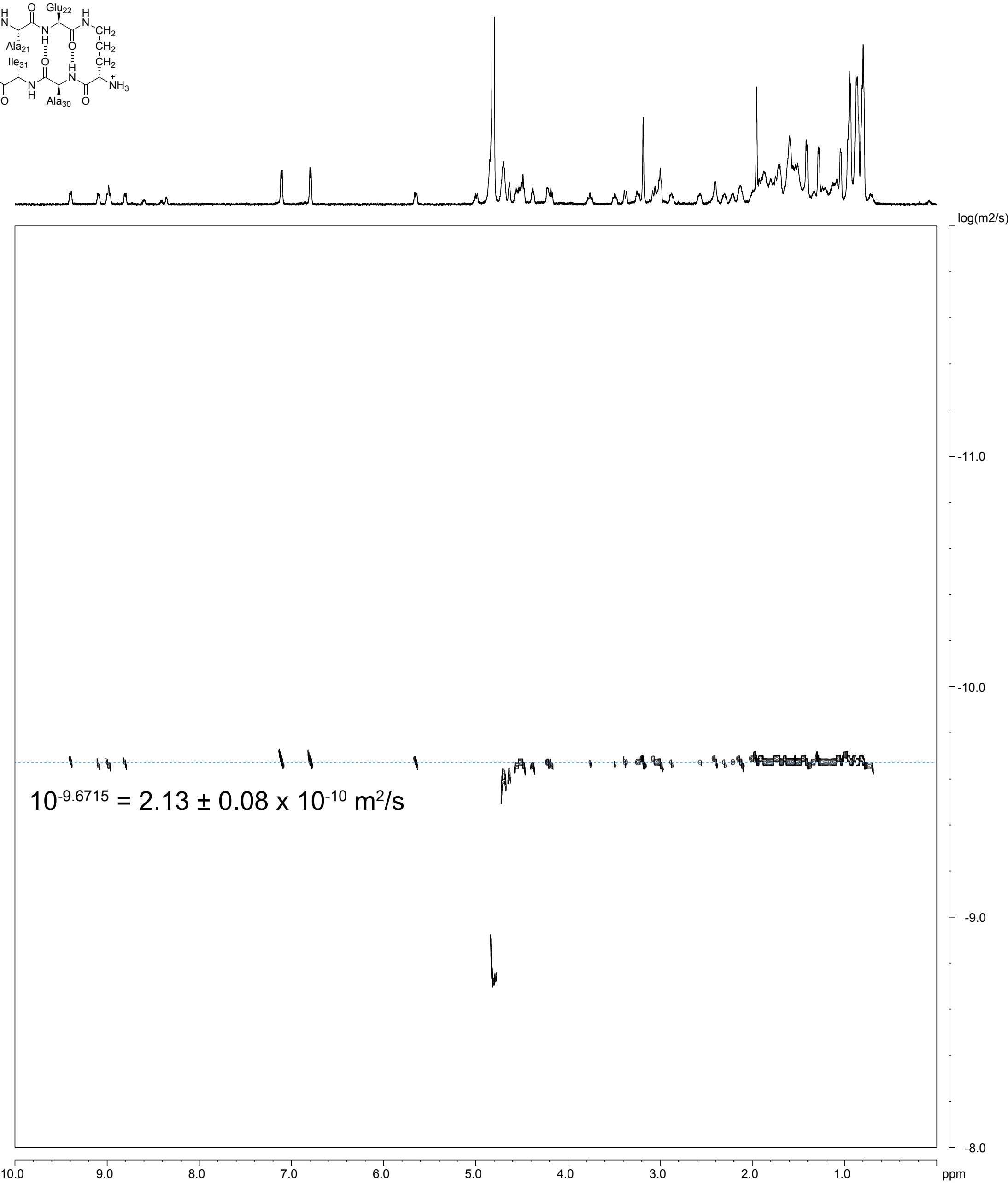
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1299977 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00



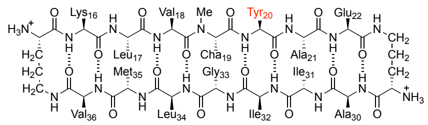
^{1}H NMR DOSY spectrum of 1.0 mM peptide **4b** in D_2O at 600 MHz and 298 K





Current Data Parameters	
NAME	JZ-II-175
EXPNO	4
PROCNO	1
F2 - Acquisition Parameters	
Date_	20231011
Time	12.34
INSTRUM	av600
PROBHD	5 mm CPBBO BB-
PULPROG	ledbpgp2s
TD	98074
SOLVENT	D2O
NS	8
DS	4
SWH	9615.385 Hz
FIDRES	0.098042 Hz
AQ	5.0998478 sec
RG	50.8
DW	52.000 usec
DE	16.36 usec
TE	297.9 K
D1	5.00000000 sec
D16	0.00020000 sec
D20	0.07500000 sec
D21	0.00500000 sec
======== CHANNEL f1 ========	
SFO1	600.1330006 MHz
NUC1	1H
P1	10.00 usec
P2	20.00 usec
PLW1	30.00000000 W
====== GRADIENT CHANNEL =====	
GPNAM[6]	SMSQ10.100
GPNAM[7]	SMSQ10.100
GPNAM[8]	SMSQ10.100
GPZ6	100.00 %
GPZ7	-17.13 %
GPZ8	-13.17 %
P19	600.00 usec
P30	1550.00 usec
F1 - Acquisition parameters	
TD	16
SFO1	600.1328 MHz
FIDRES	751.201904 Hz
SW	10.014 ppm
FnMODE	QF
F2 - Processing parameters	
SI	65536
SF	600.1299978 MHz
WDW	EM
SSB	0
LB	10.00 Hz
GB	0
PC	1.00
F1 - Processing parameters	
SI	256
MC2	QF
SF	600.1375820 MHz
WDW	no
SSB	0
LB	0 Hz
GB	0

^{1}H NMR spectrum of 11.0 mM peptide **4b** in D_2O at 600 MHz and 298 K

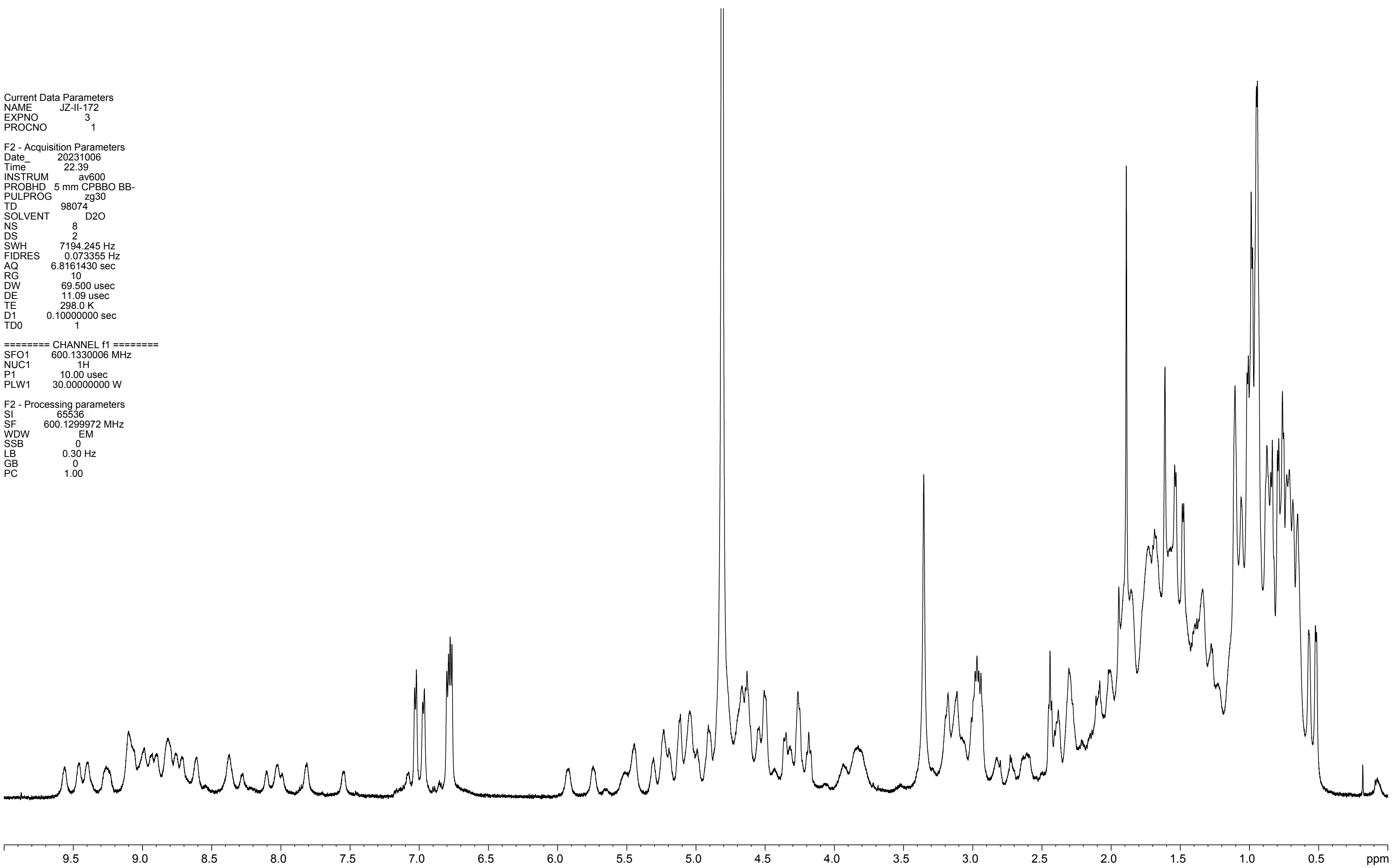


Current Data Parameters
NAME JZ-II-172
EXPNO 3
PROCNO 1

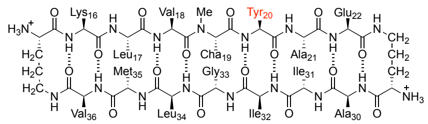
F2 - Acquisition Parameters
Date_ 20231006
Time 22.39
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 8
DS 2
SWH 7194.245 Hz
FIDRES 0.073355 Hz
AQ 6.8161430 sec
RG 10
DW 69.500 usec
DE 11.09 usec
TE 298.0 K
D1 0.10000000 sec
TD0 1

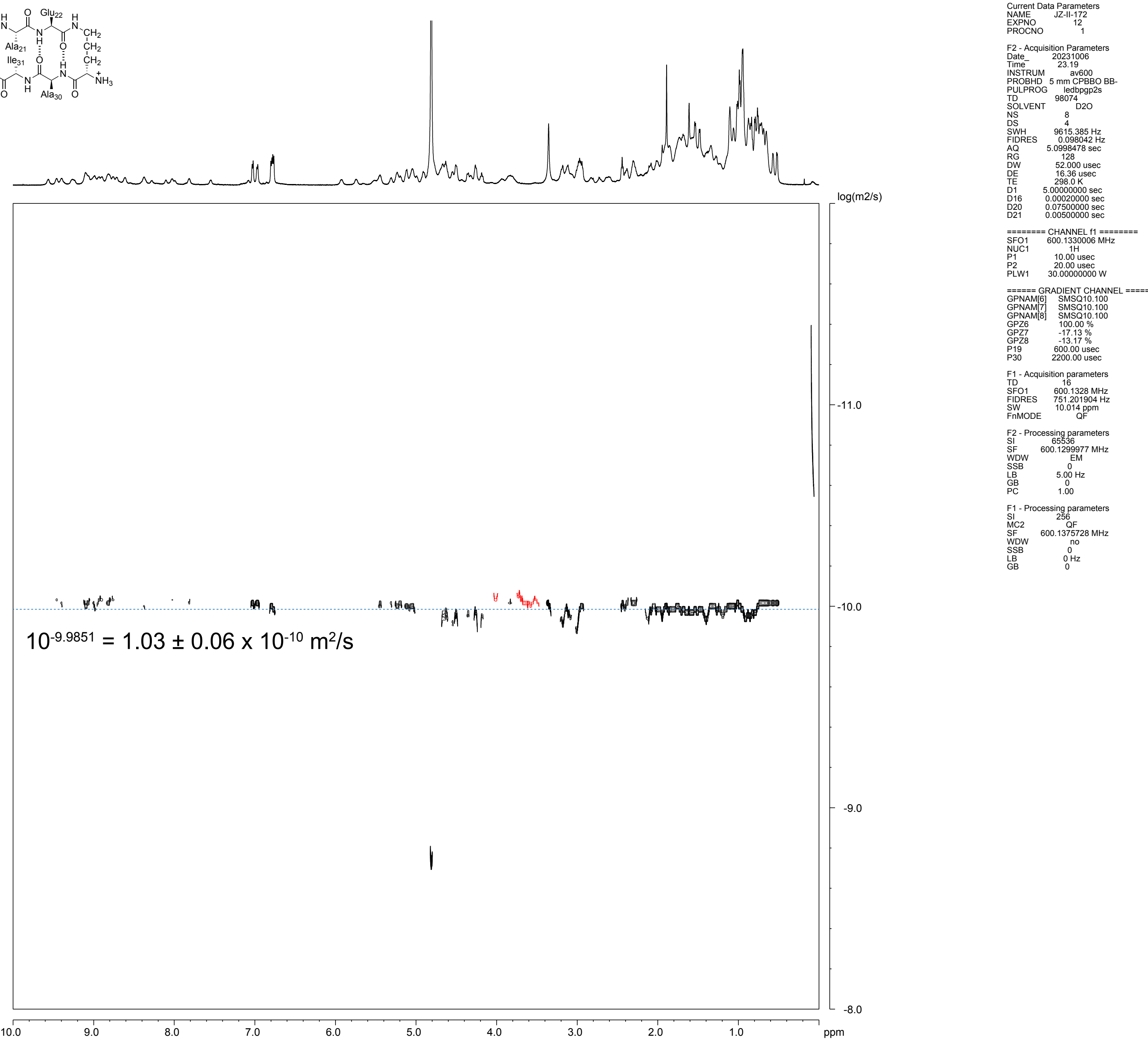
======== CHANNEL f1 ========
SFO1 600.1330006 MHz
NUC1 1H
P1 10.00 usec
PLW1 30.00000000 W

F2 - Processing parameters
SI 65536
SF 600.1299972 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

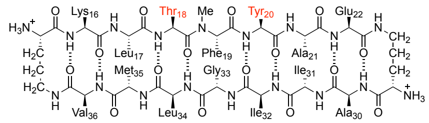


^{1}H NMR DOSY spectrum of 11.0 mM peptide **4b** in D_2O at 600 MHz and 298 K





^{1}H NMR spectrum of 1.0 mM peptide **5** in D_2O at 600 MHz and 298 K

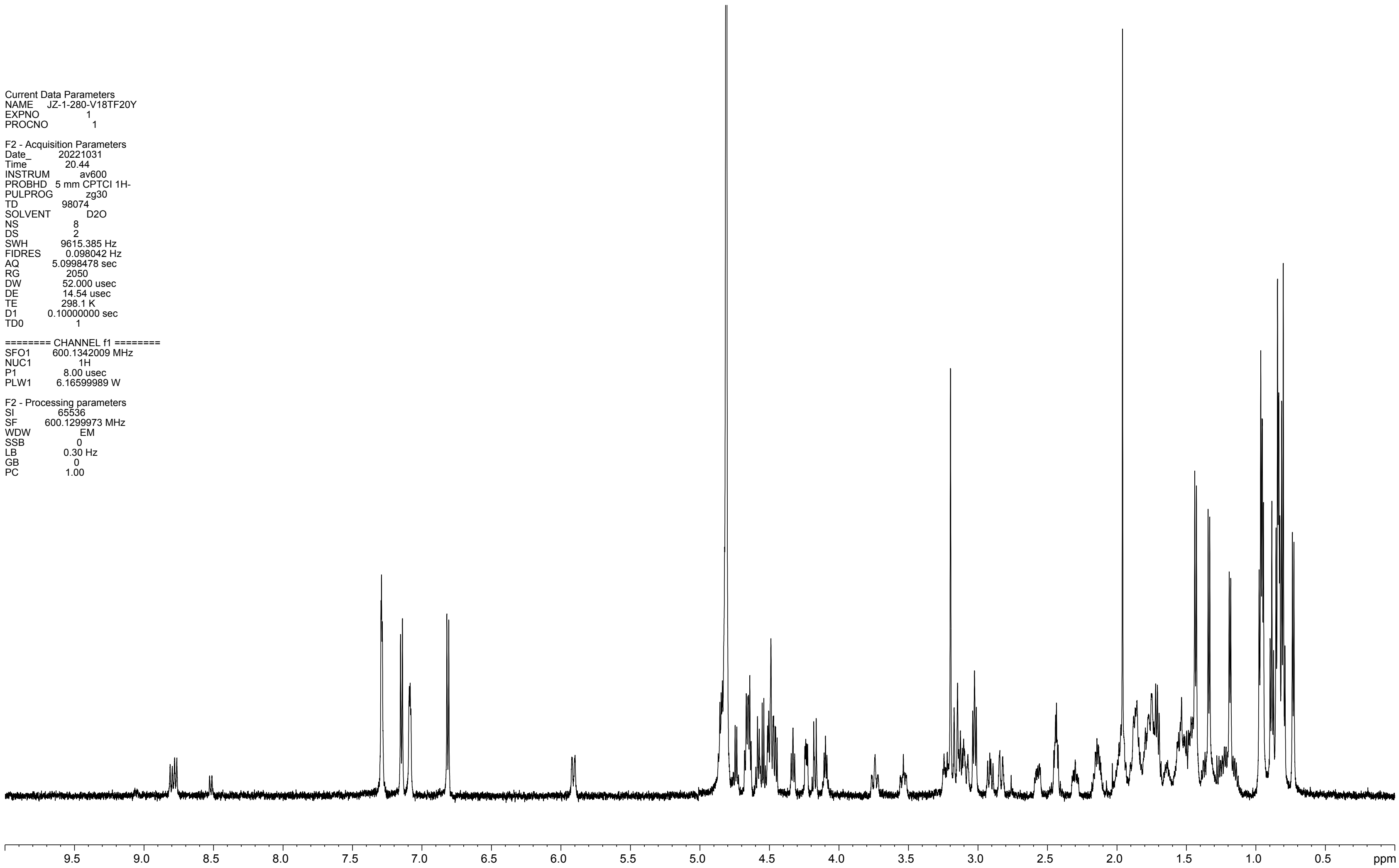


Current Data Parameters
NAME JZ-1-280-V18TF20Y
EXPNO 1
PROCNO 1

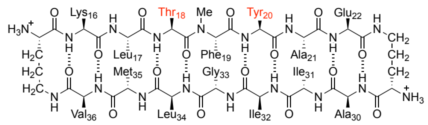
F2 - Acquisition Parameters
Date_ 20221031
Time 20.44
INSTRUM av600
PROBHD 5 mm CPTCI 1H-
PULPROG zg30
TD 98074
SOLVENT D2O
NS 8
DS 2
SWH 9615.385 Hz
FIDRES 0.098042 Hz
AQ 5.0998478 sec
RG 2050
DW 52.000 usec
DE 14.54 usec
TE 298.1 K
D1 0.10000000 sec
TD0 1

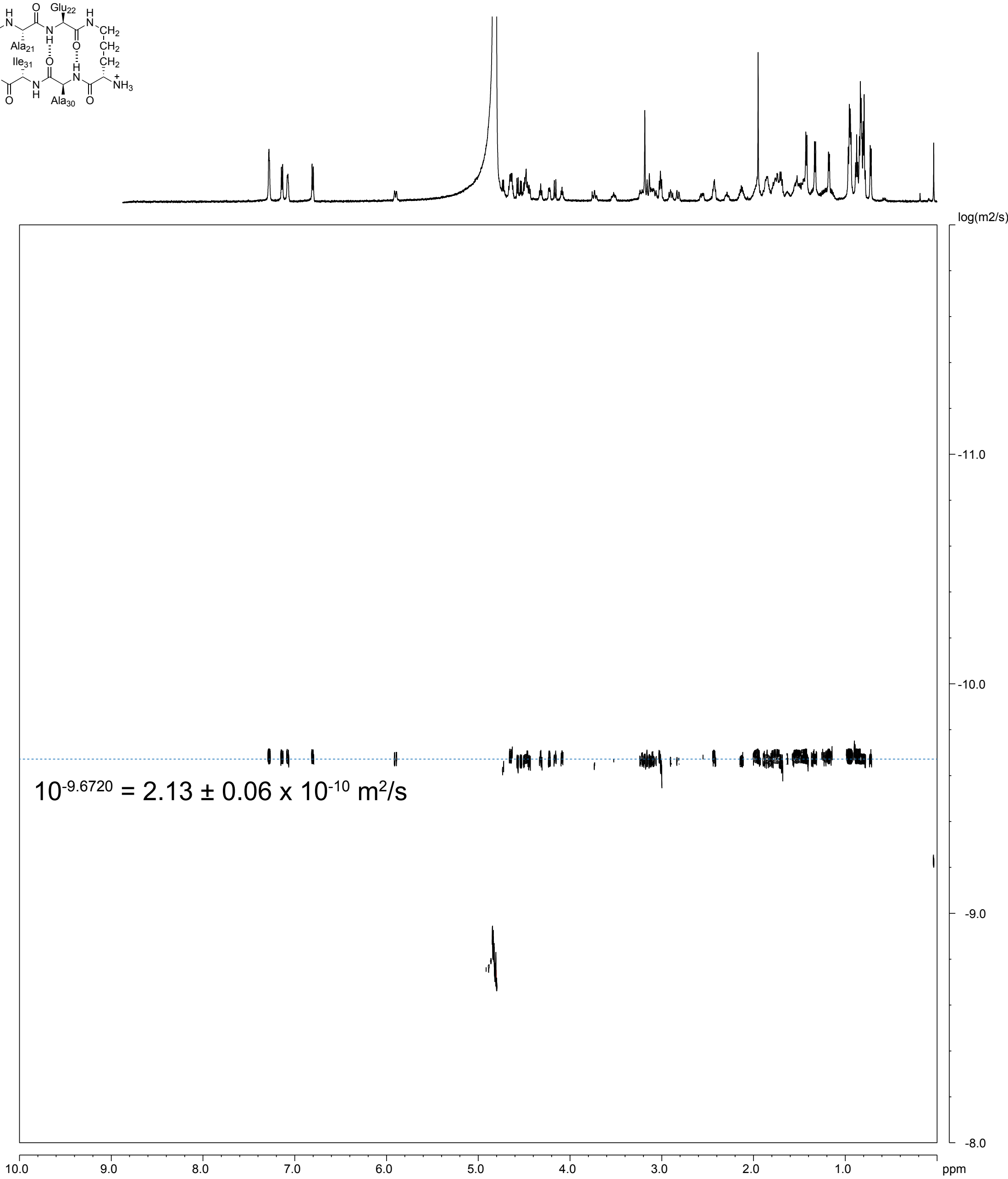
======== CHANNEL f1 ========
SFO1 600.1342009 MHz
NUC1 1H
P1 8.00 usec
PLW1 6.16599989 W

F2 - Processing parameters
SI 65536
SF 600.1299973 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00



^{1}H NMR DOSY spectrum of 1.0 mM peptide **5** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME V18T_F20Y(KLF)_1.0mM_peptide_D2O
EXPNO 5
PROCNO 1

F2 - Acquisition Parameters
Date_ 20220412
Time 22.50
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 5482.456 Hz
FIDRES 0.055901 Hz
AQ 8.9443483 sec
RG 181
DW 91.200 usec
DE 21.95 usec
TE 298.0 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1324563 MHz
NUC1 1H
P1 10.25 usec
P2 20.50 usec
PLW1 30.00000000 W

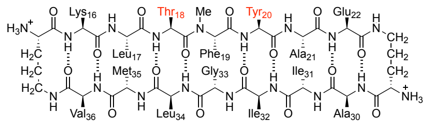
====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 1350.00 usec

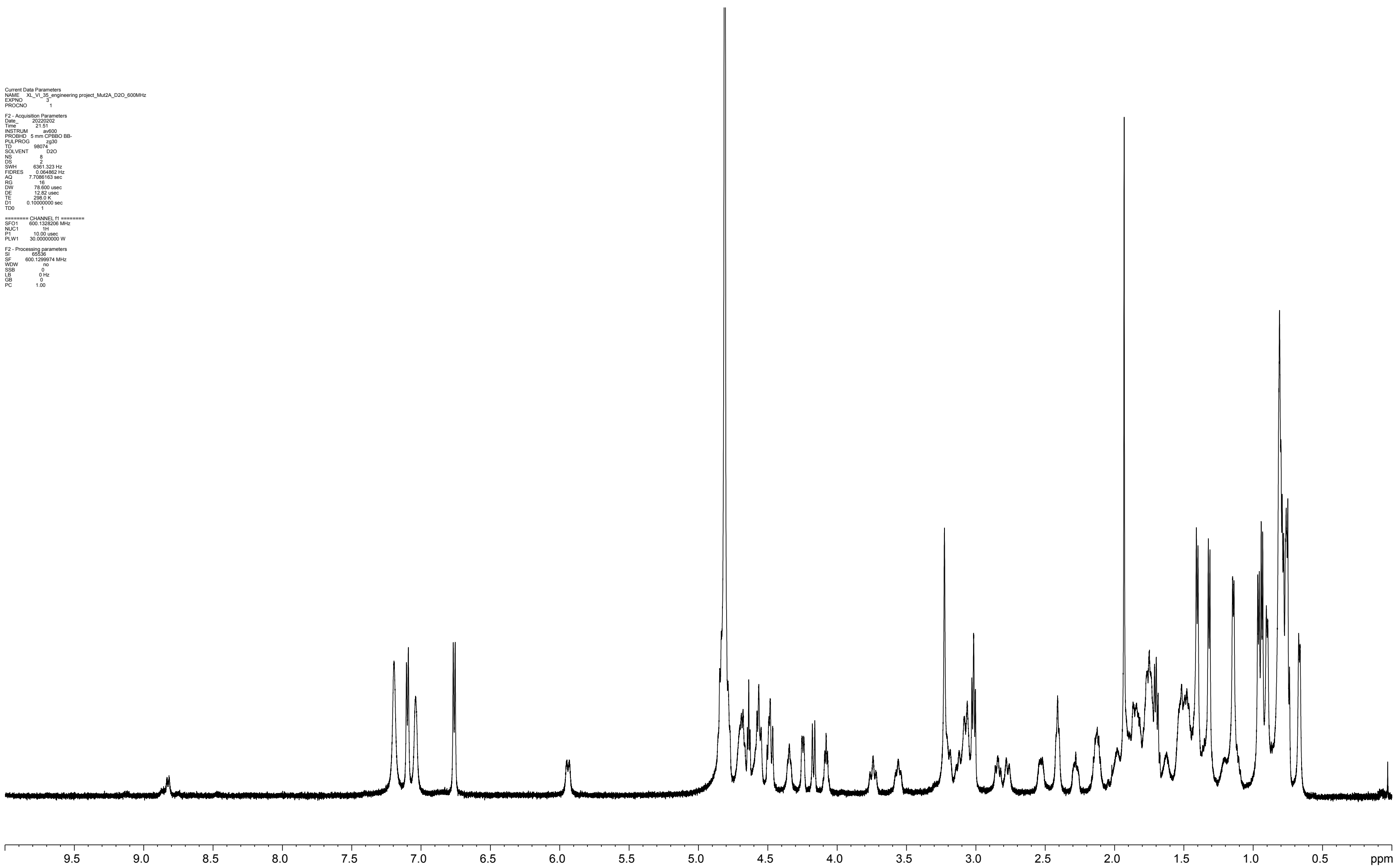
F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299973 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

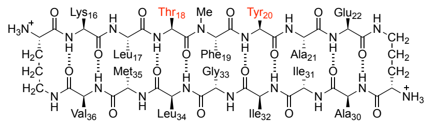
F1 - Processing parameters
SI 256
MC2 QF
SF 600.1372381 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

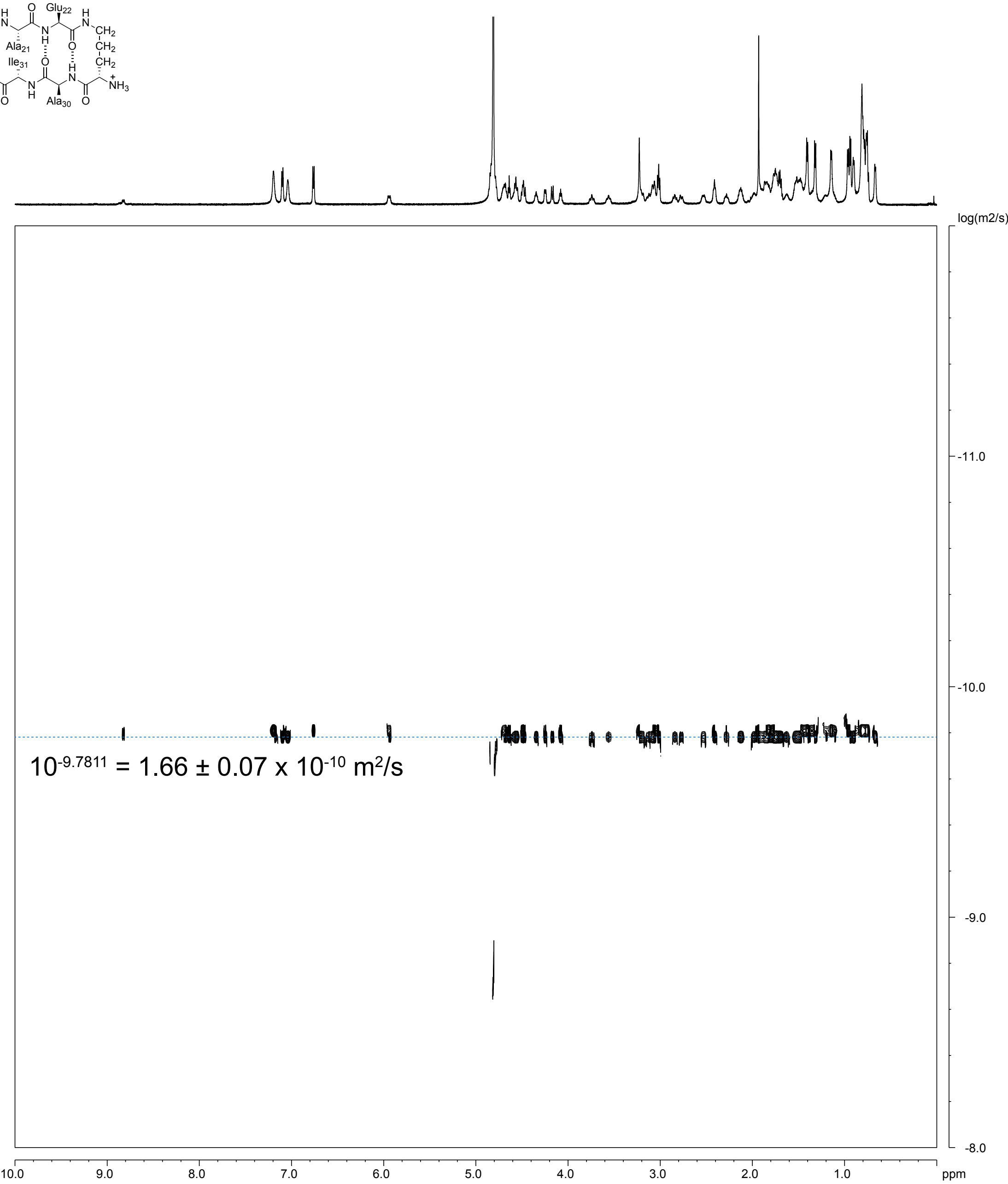
[1]H NMR spectrum of 8.0 mM peptide **5** in D_2O at 600 MHz and 298 K





^{1}H NMR DOSY spectrum of 8.0 mM peptide **5** in D_2O at 600 MHz and 298 K





Current Data Parameters
NAME XL_VI_35_engineering project_Mut2A_D2
EXPNO 7
PROCNO 1

F2 - Acquisition Parameters
Date_ 20220202
Time 22.23
INSTRUM av600
PROBHD 5 mm CPBBO BB-
PULPROG ledbpgp2s
TD 98074
SOLVENT D2O
NS 8
DS 4
SWH 6361.323 Hz
FIDRES 0.064862 Hz
AQ 7.7086163 sec
RG 114
DW 78.600 usec
DE 18.70 usec
TE 298.0 K
D1 5.00000000 sec
D16 0.00020000 sec
D20 0.07500000 sec
D21 0.00500000 sec

======== CHANNEL f1 ========
SFO1 600.1328206 MHz
NUC1 1H
P1 9.50 usec
P2 19.00 usec
PLW1 30.00000000 W

====== GRADIENT CHANNEL =====
GPNAM[6] SMSQ10.100
GPNAM[7] SMSQ10.100
GPNAM[8] SMSQ10.100
GPZ6 100.00 %
GPZ7 -17.13 %
GPZ8 -13.17 %
P19 600.00 usec
P30 1800.00 usec

F1 - Acquisition parameters
TD 16
SFO1 600.1328 MHz
FIDRES 751.201904 Hz
SW 10.014 ppm
FnMODE QF

F2 - Processing parameters
SI 65536
SF 600.1299977 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

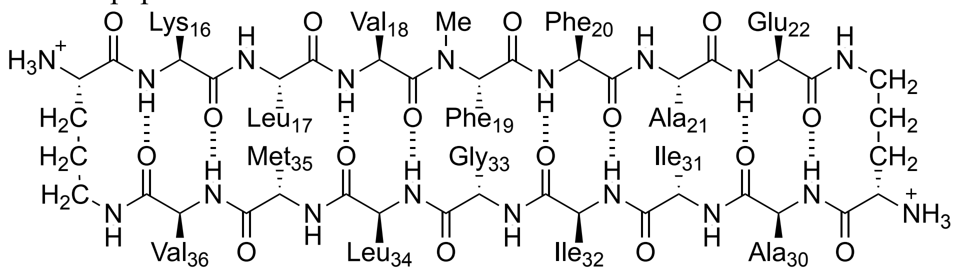
F1 - Processing parameters
SI 256
MC2 QF
SF 600.1371767 MHz
WDW no
SSB 0
LB 0 Hz
GB 0

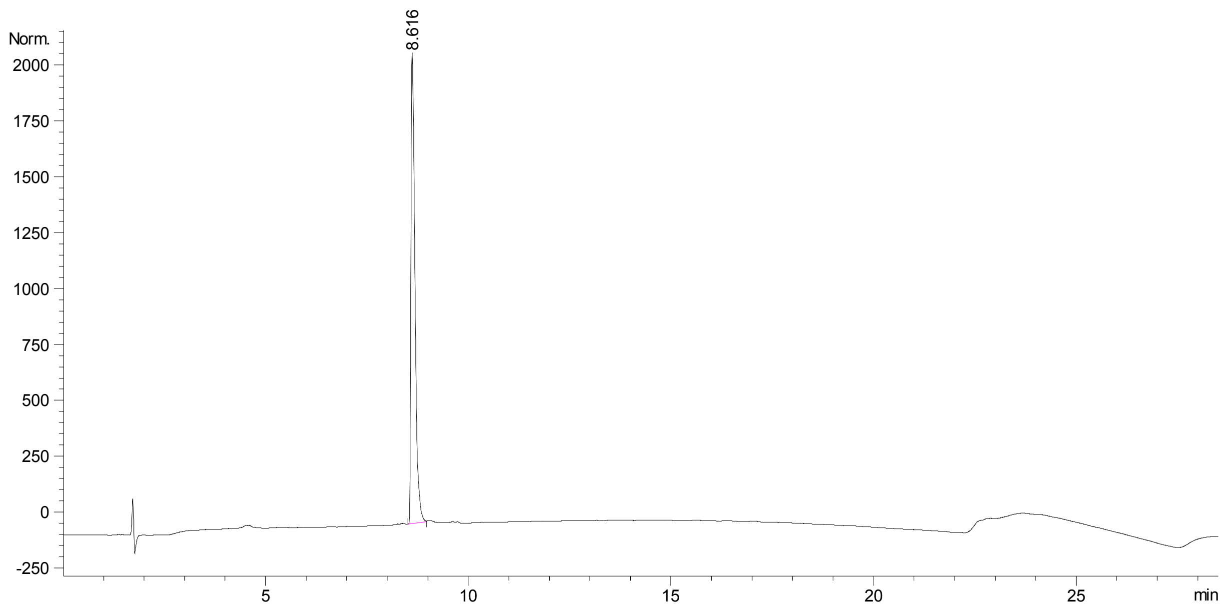
Peptide Characterization Data

Summary of purified peptides yields. Percent yields were calculated based on the resin loading.

Peptide	Yield (mg)	% Yield	Calculated MW as a TFA salt
1	42	11	2117.3 (•3 TFA)
2	37	16	2123.4 (•3 TFA)
2a	59	13	2124.4 (•3 TFA)
2b	55	21	2131.4 (•3 TFA)
2c	61	20	2135.5 (•3 TFA)
2d	41	16	2127.3 (•3 TFA)
3a	73	26	2123.4 (•3 TFA)
3b	7	8	2137.4 (•3 TFA)
3c	45	16	2137.4 (•3 TFA)
4a	73	24	2149.4 (•3 TFA)
4b	29	10	2149.4 (•3 TFA)
5	38	12	2149.4 (•3 TFA)

Characterization of peptide **1**

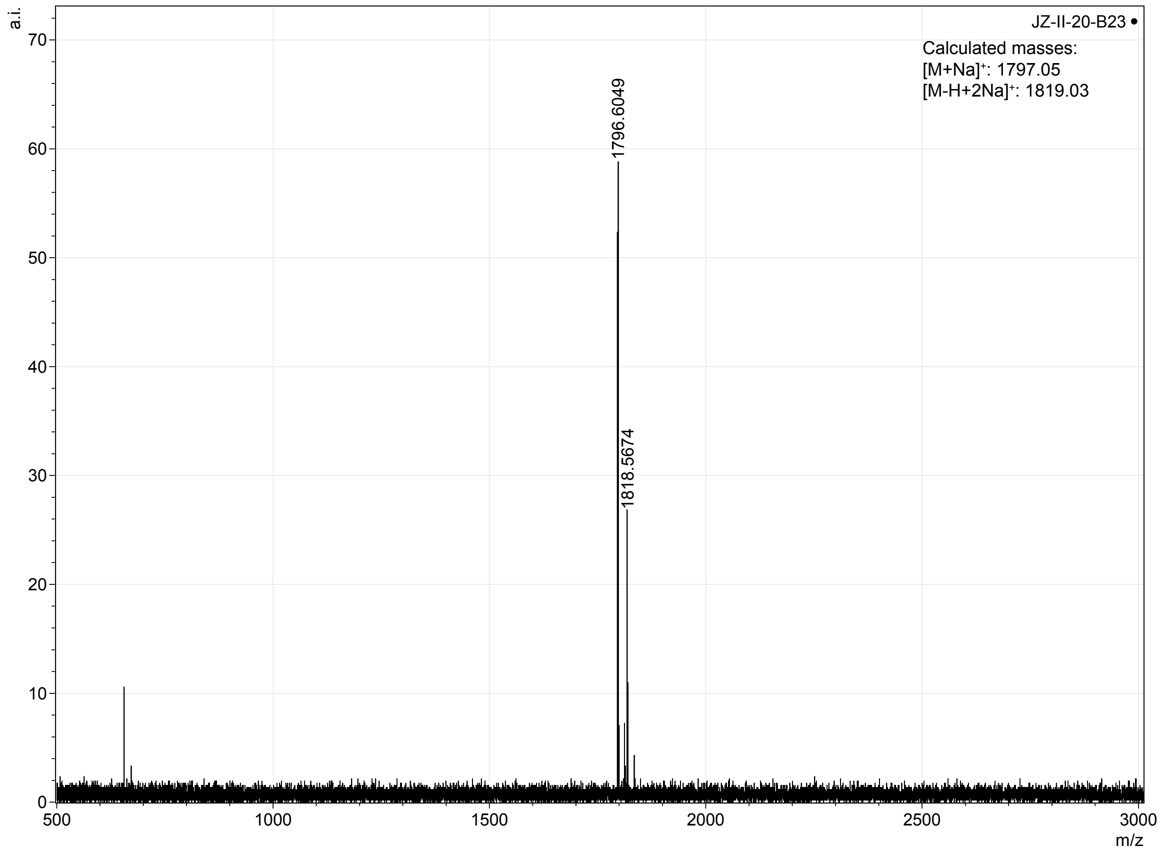


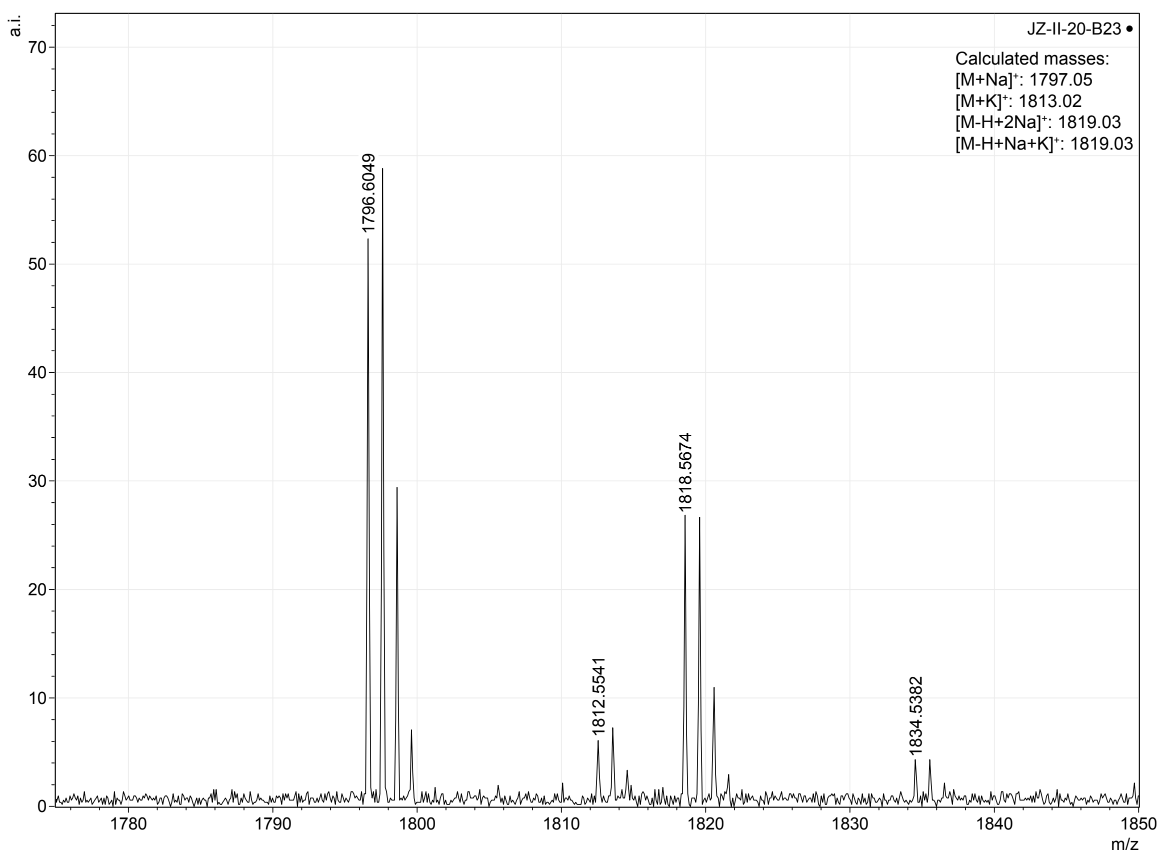


Signal 1: MWD1 A, Sig=214,4 Ref=off

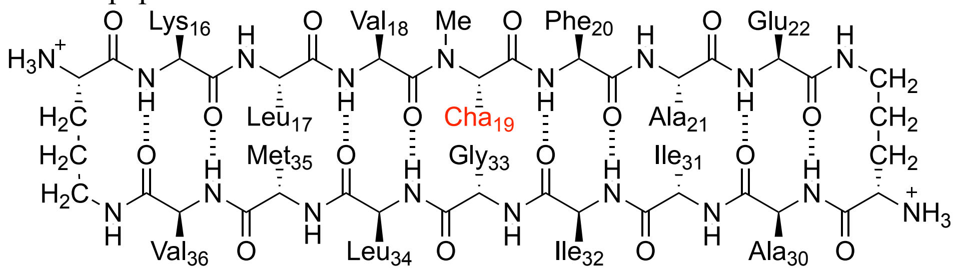
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.616	MM	0.1152	1.31734e4	1905.11182	100.0000

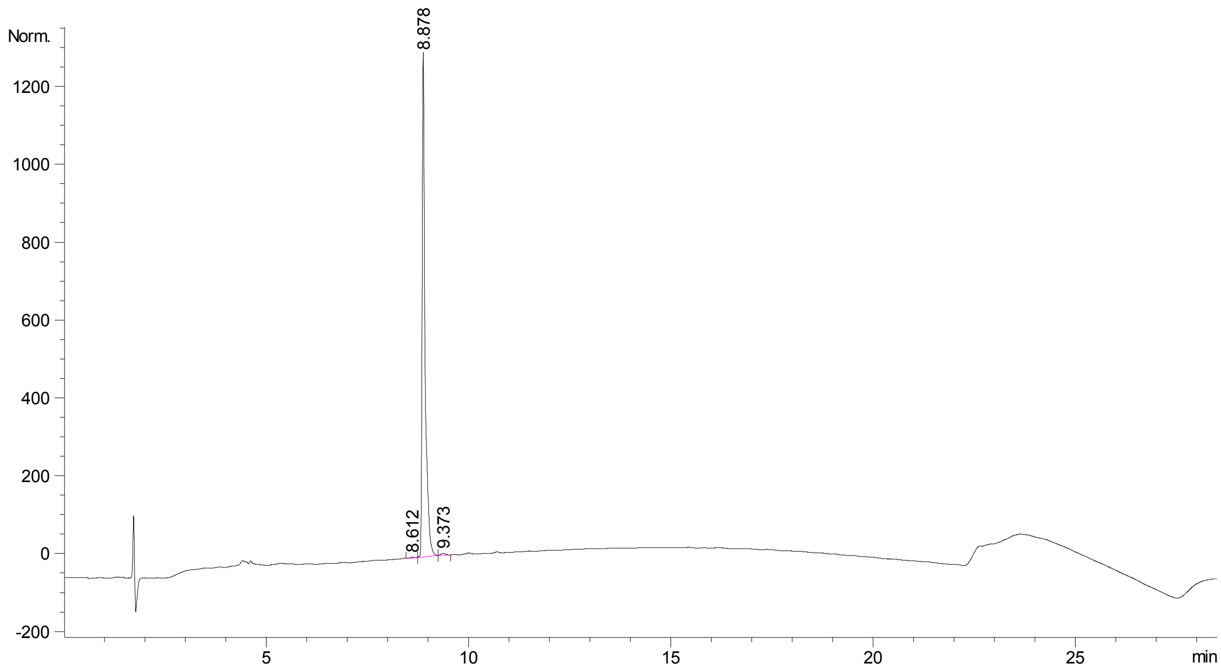
Totals : 1.31734e4 1905.11182


JZ-II-20-B23
Calculated masses:
[M+Na]+: 1797.05
[M-H+2Na]+: 1819.03
1796.6049
1818.5674
a.i.
m/z


JZ-II-20-B23
Calculated masses:
[M+Na]+: 1797.05
[M+K]+: 1813.02
[M-H+2Na]+: 1819.03
[M-H+Na+K]+: 1819.03
1796.6049
1812.5541
1818.5674
1834.5382
a.i.
m/z

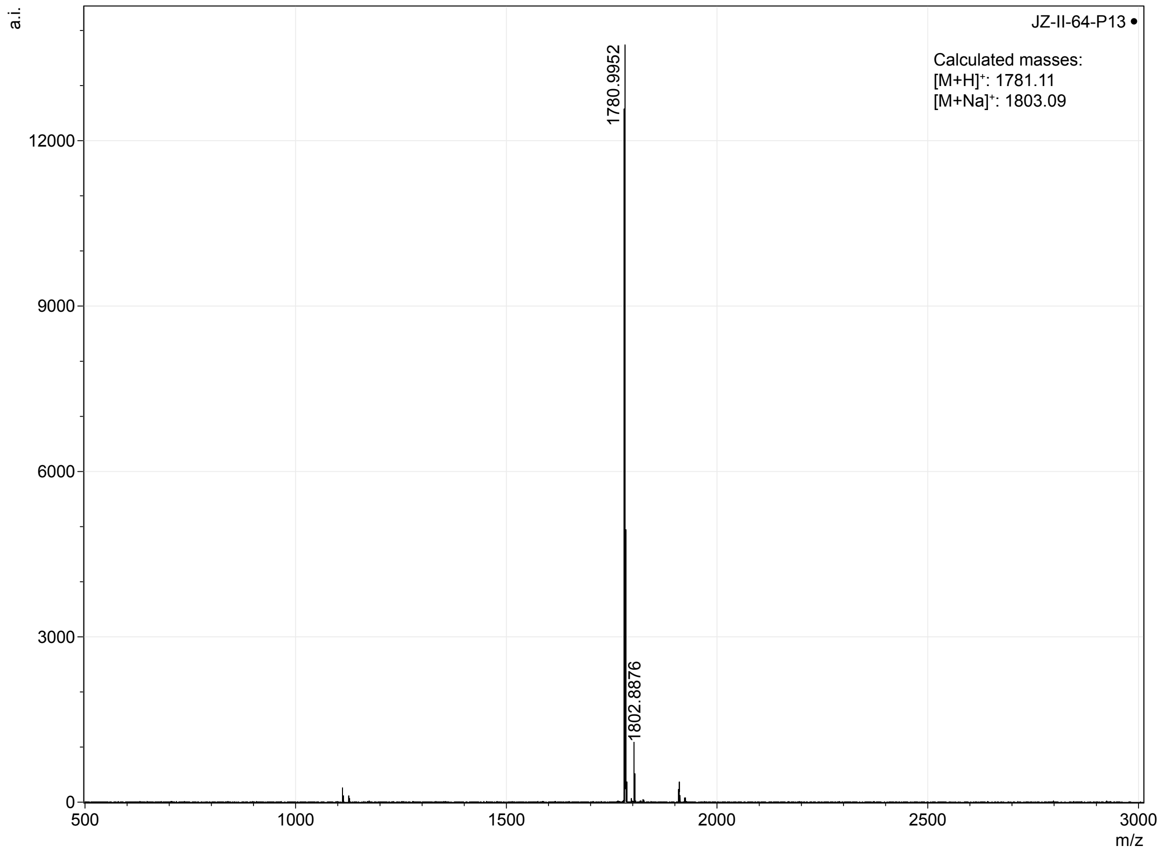
Characterization of peptide **2**

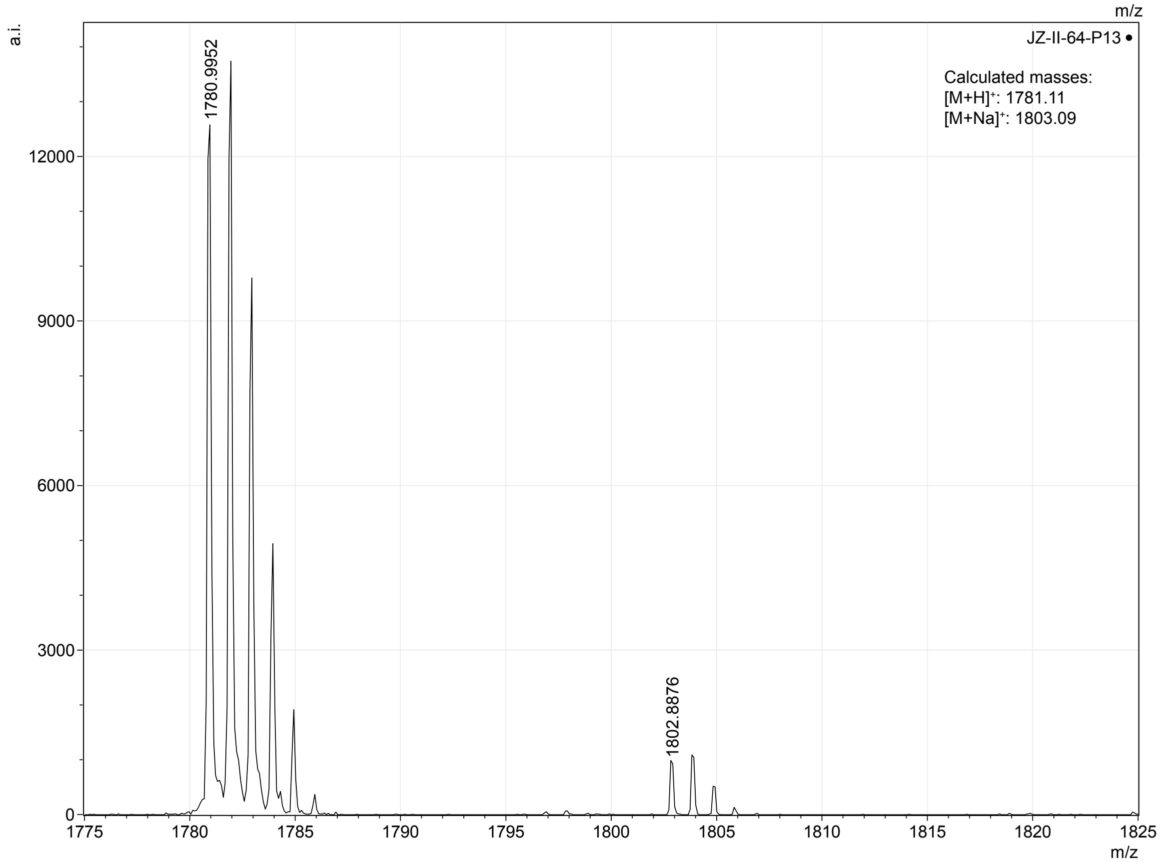




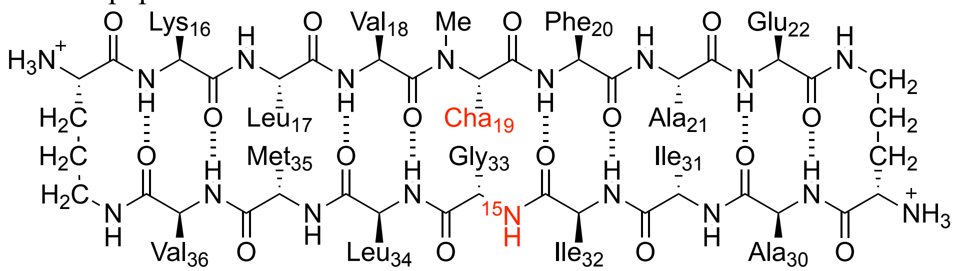
Signal 1: MWD1 A, Sig=214,4 Ref=off

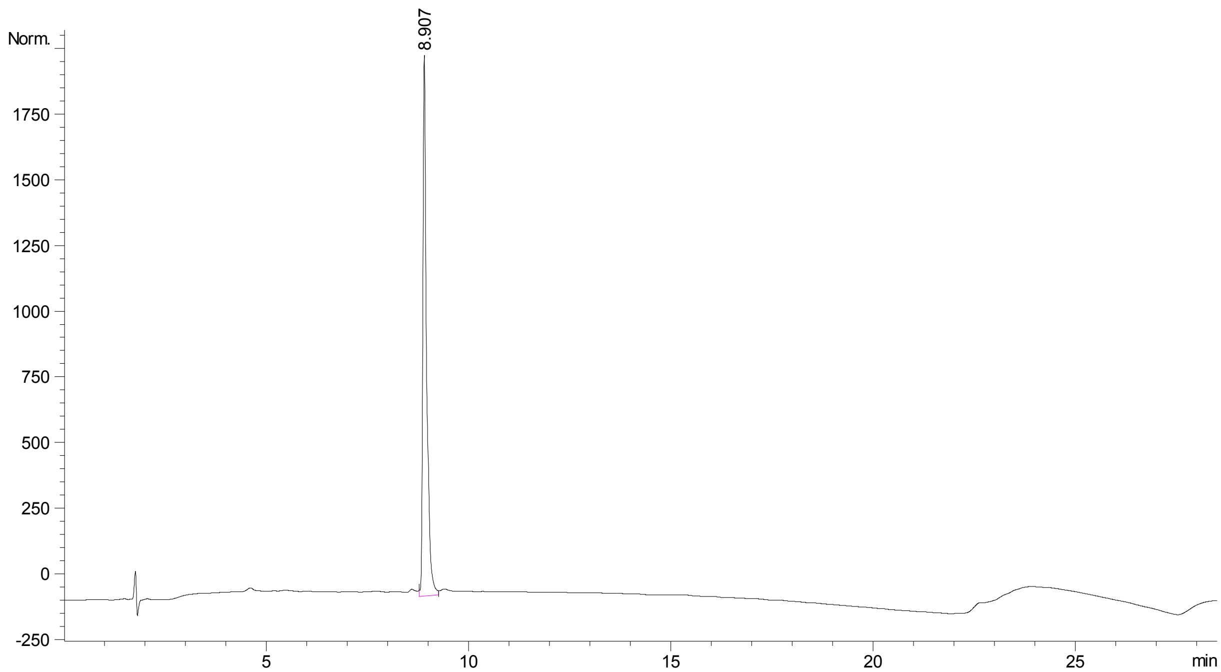
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.612	MM	0.1604	19.43673	2.01966	0.3197
2	8.878	MM	0.0855	6016.14404	1172.66187	98.9668
3	9.373	MM	0.1565	43.37235	4.61924	0.7135
Totals :				6078.95312	1179.30076	


JZ-II-64-P13
Calculated masses:
[M+H]+: 1781.11
[M+Na]+: 1803.09
1780.9952
1802.8876
a.i.
m/z

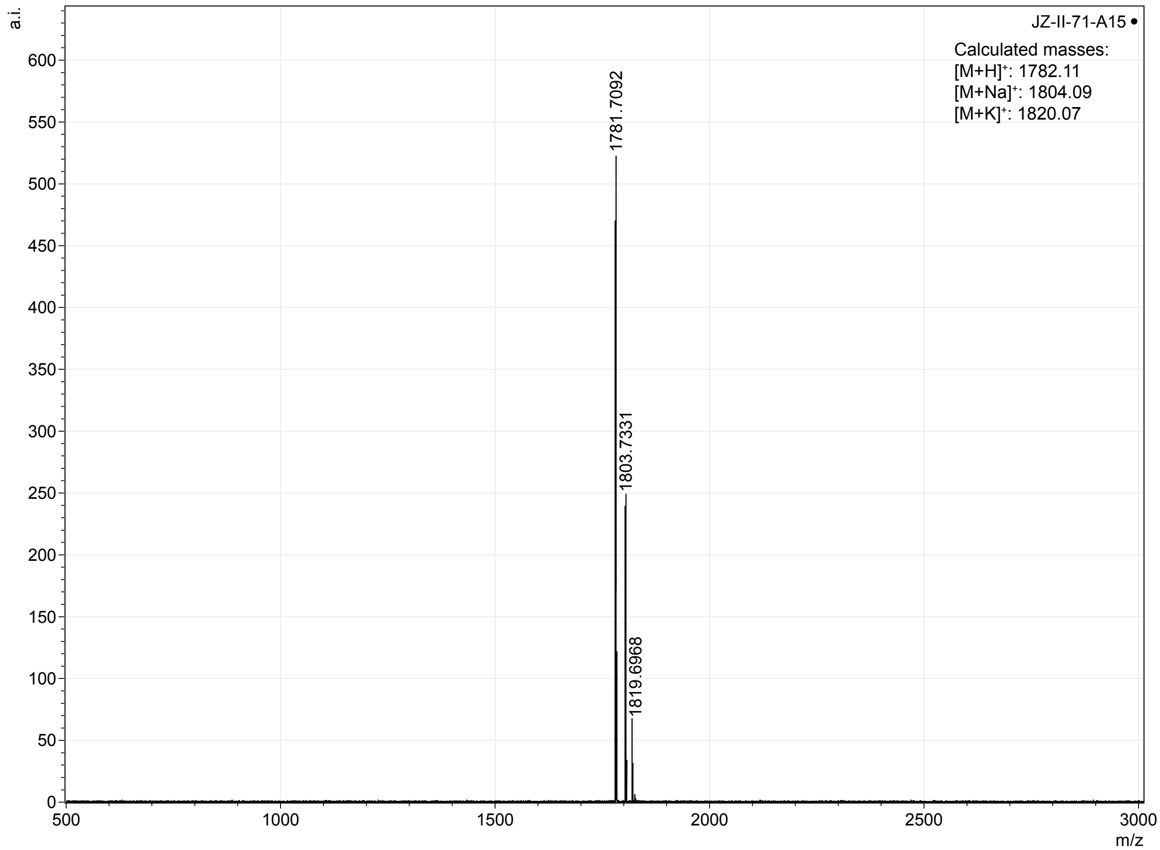

JZ-II-64-P13
Calculated masses:
[M+H]+: 1781.11
[M+Na]+: 1803.09
1780.9952
1802.8876
a.i.
m/z

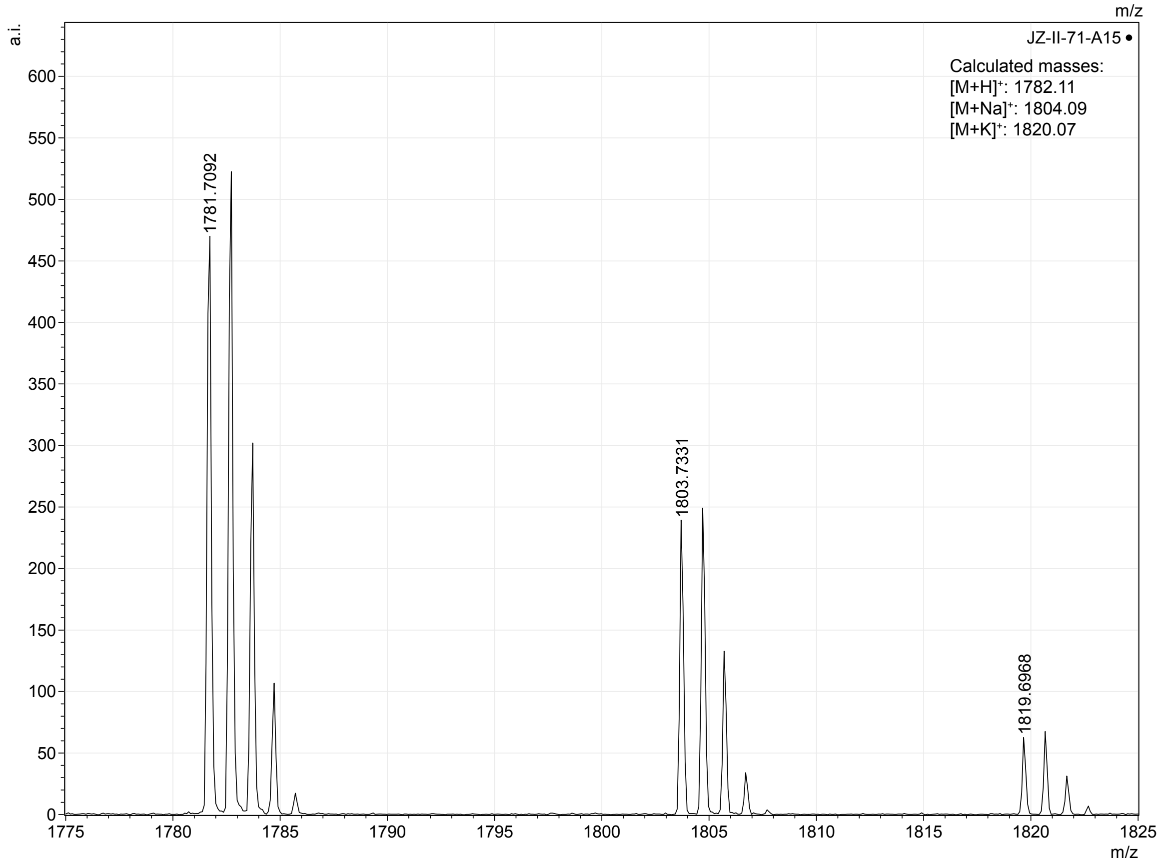
Characterization of peptide **2a**



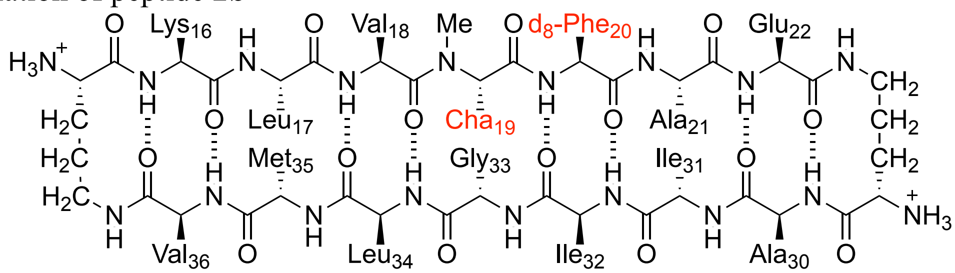


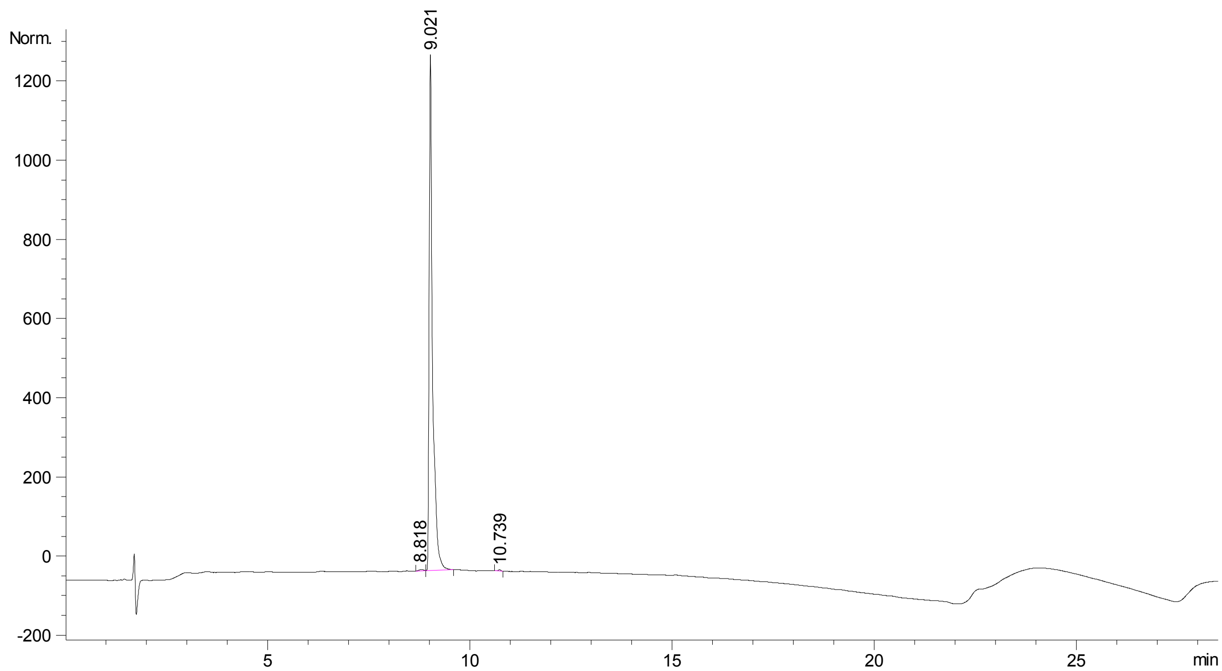
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.907	MM	0.1084	1.20888e4	1859.29480	100.0000
Totals :				1.20888e4	1859.29480	


JZ-II-71-A15
Calculated masses:
[M+H]+: 1782.11
[M+Na]+: 1804.09
[M+K]+: 1820.07
1781.7092
1803.7331
1819.6968
a.i.
m/z


JZ-II-71-A15
Calculated masses:
[M+H]+: 1782.11
[M+Na]+: 1804.09
[M+K]+: 1820.07
1781.7092
1803.7331
1819.6968
a.i.
m/z

Characterization of peptide **2b**

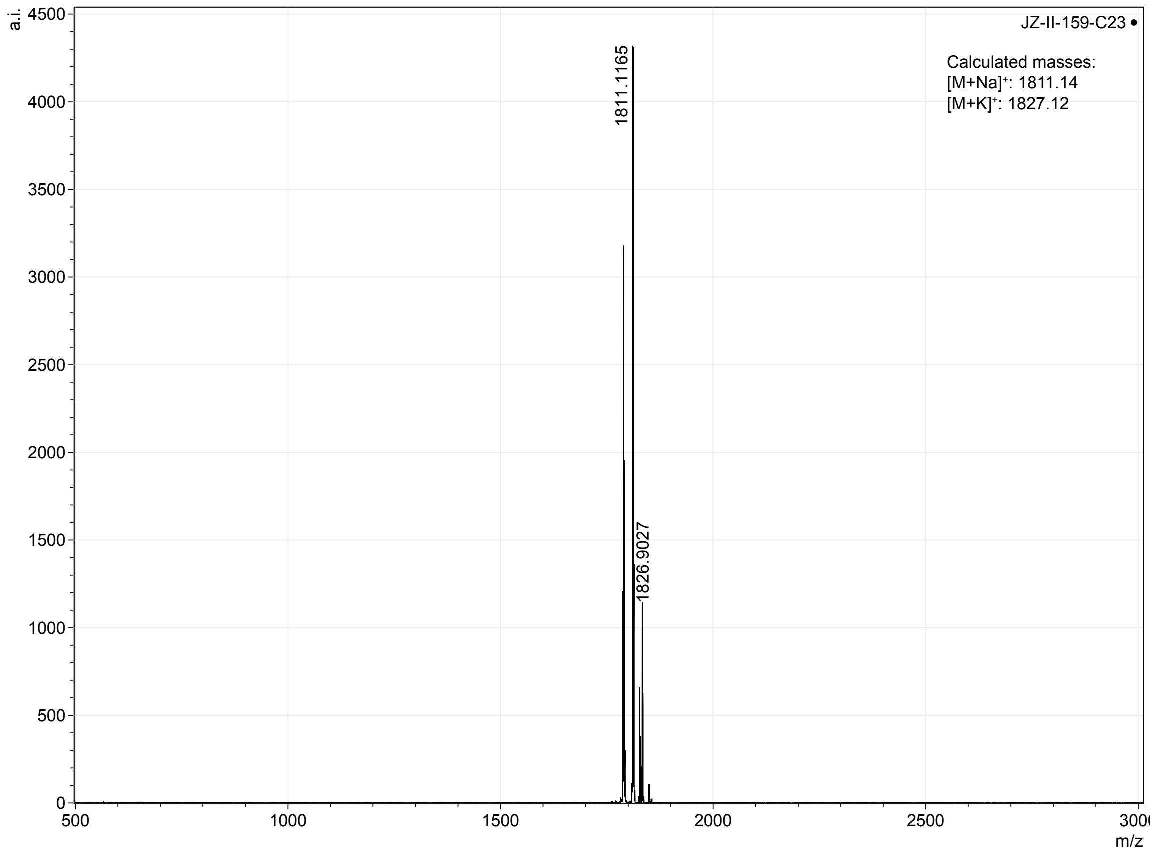


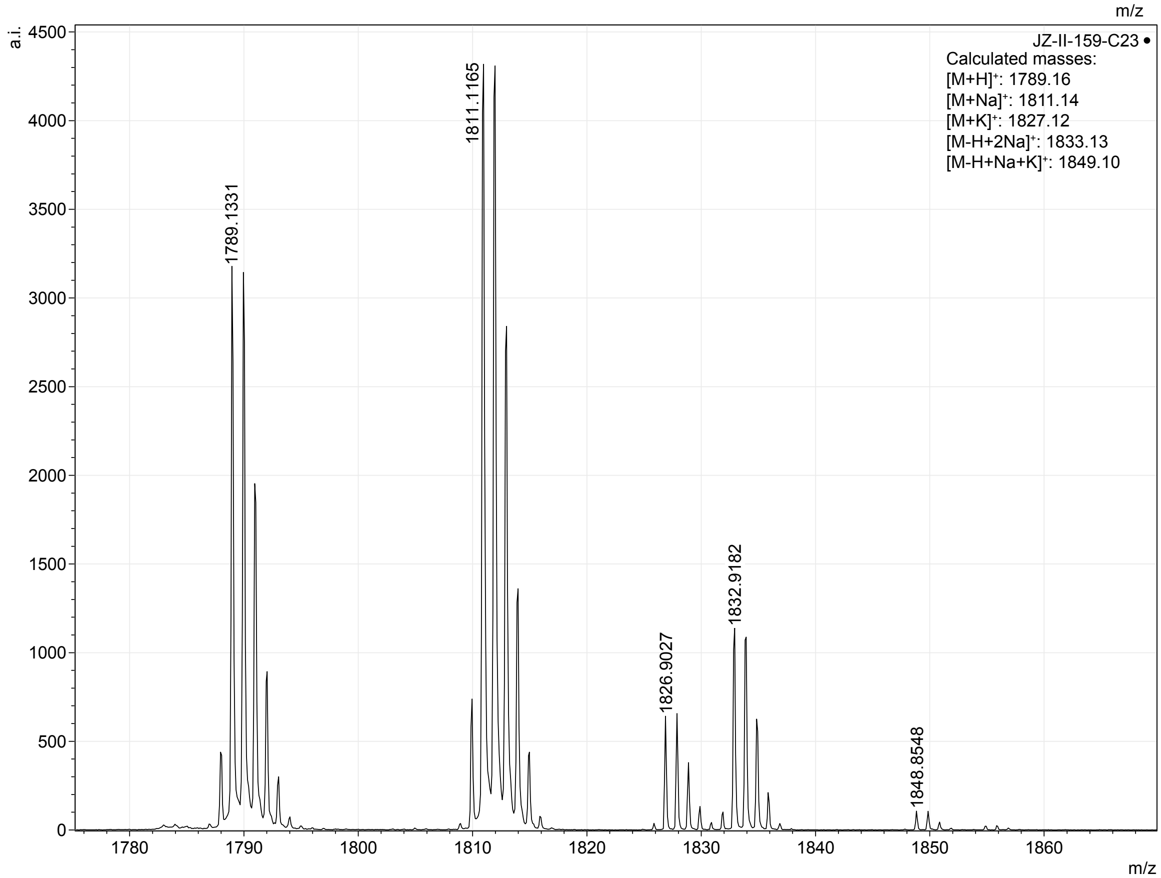


Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.818	MM	0.1128	23.98382	2.85144	0.3505
2	9.021	MM	0.0958	6802.32568	1184.01550	99.4053
3	10.739	MM	0.0890	16.70873	3.12815	0.2442

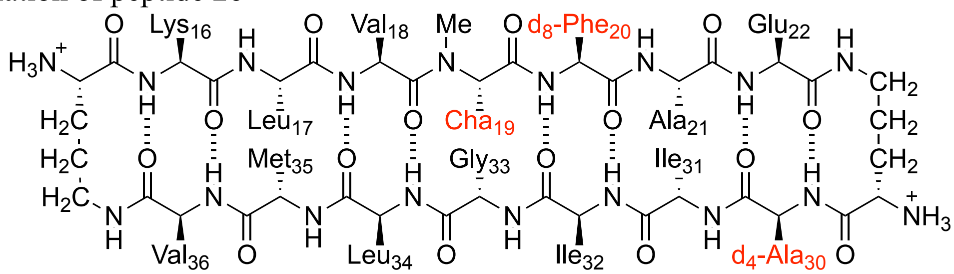
Totals : 6843.01823 1189.99509

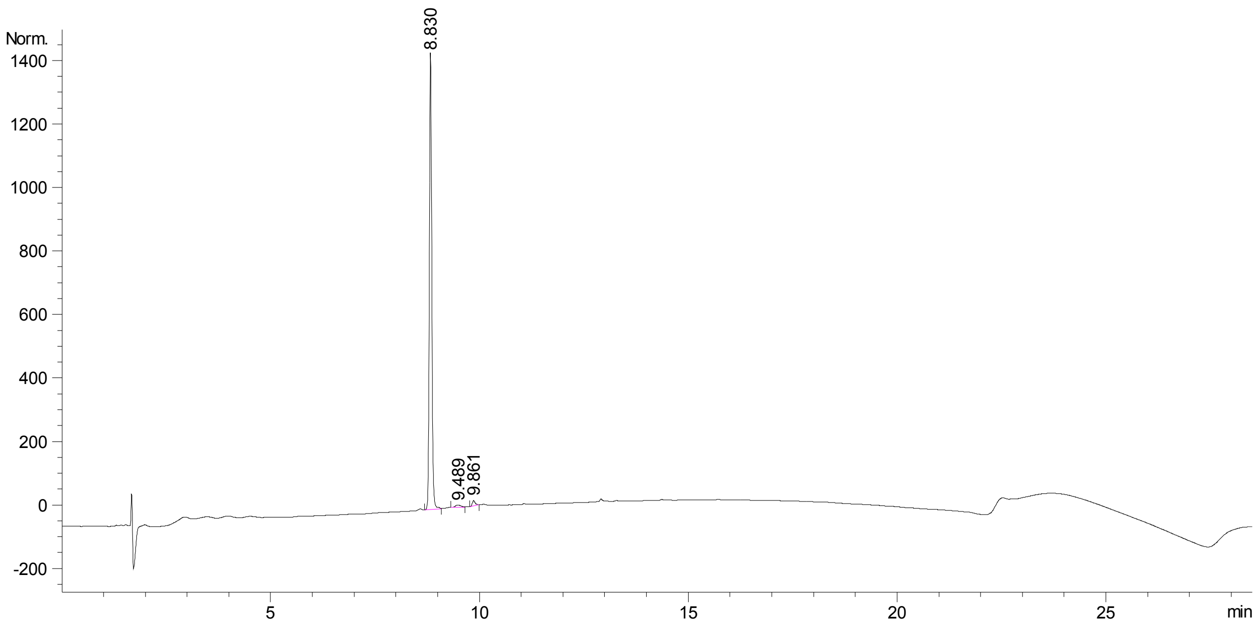
Note: The commercially available d_8-Phe$_{20}$ (98%) contains incomplete deuteration at Hα as reported by Li, X.; *et. al*.[2] This is reflected in the MALDI-TOF characterization data.


JZ-II-159-C23
Calculated masses:
[M+Na]+: 1811.14
[M+K]+: 1827.12
1811.1165
1826.9027
a.i.
m/z


JZ-II-159-C23
Calculated masses:
[M+H]+: 1789.16
[M+Na]+: 1811.14
[M+K]+: 1827.12
[M-H+2Na]+: 1833.13
[M-H+Na+K]+: 1849.10
1789.1331
1811.1165
1826.9027
1832.9182
1848.8548
a.i.
m/z

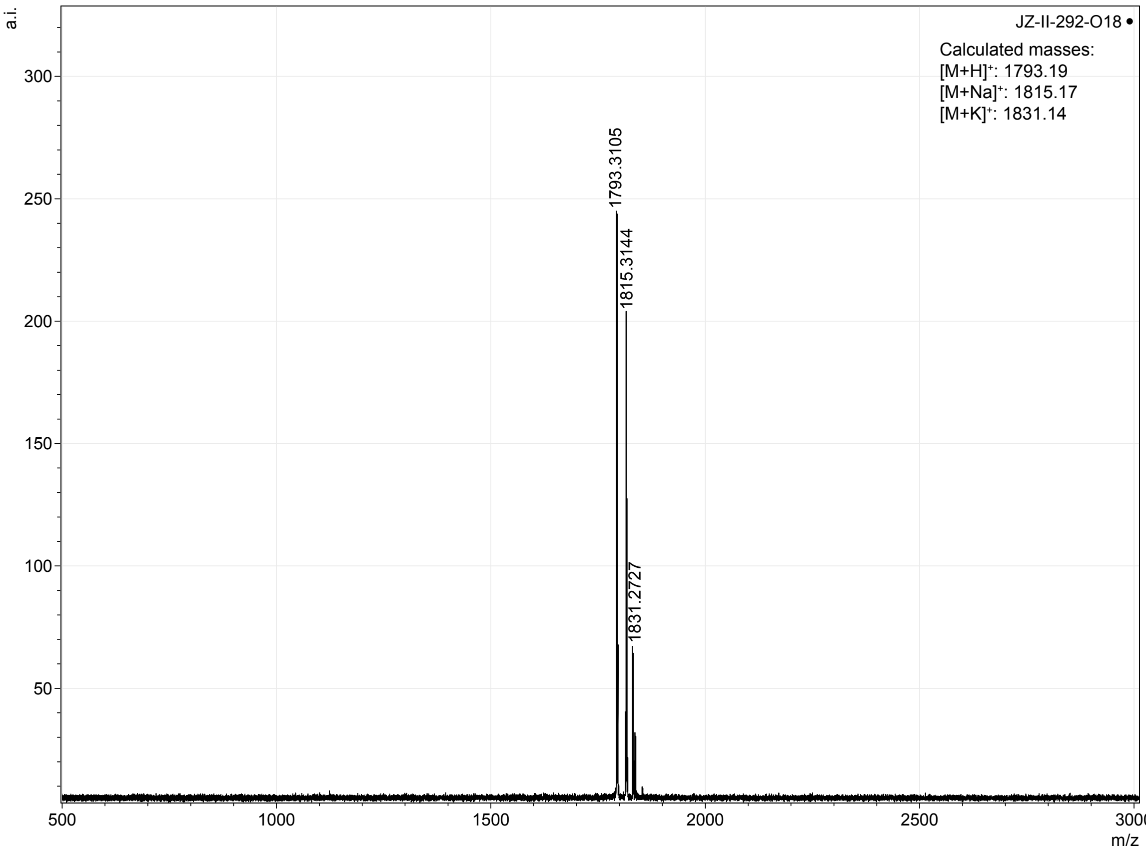
Characterization of peptide **2c**

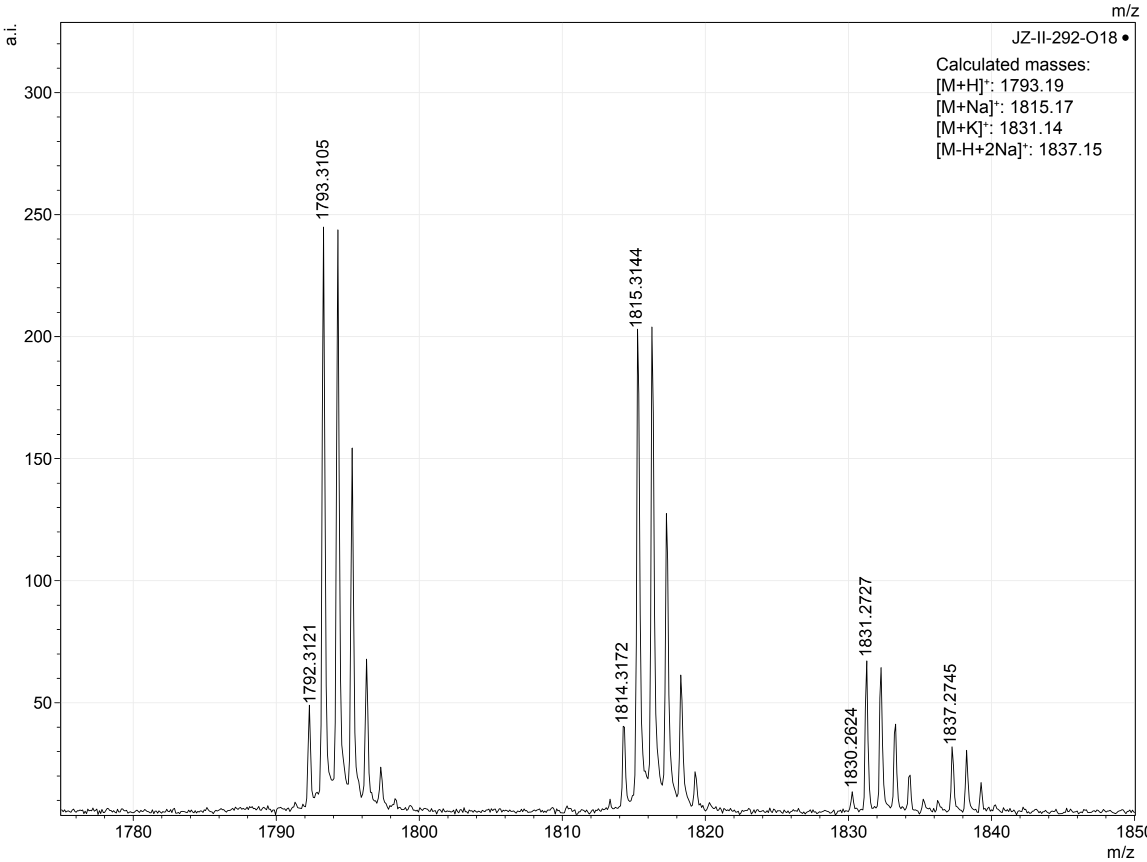




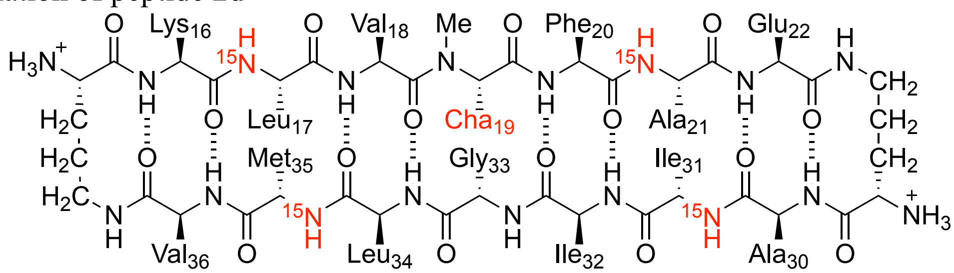
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.830	MM	0.0661	5163.70215	1301.25464	97.4876
2	9.489	MM	0.1440	56.28535	6.51516	1.0626
3	9.861	MM	0.0826	76.78871	15.48572	1.4497
Totals :				5296.77621	1323.25551	

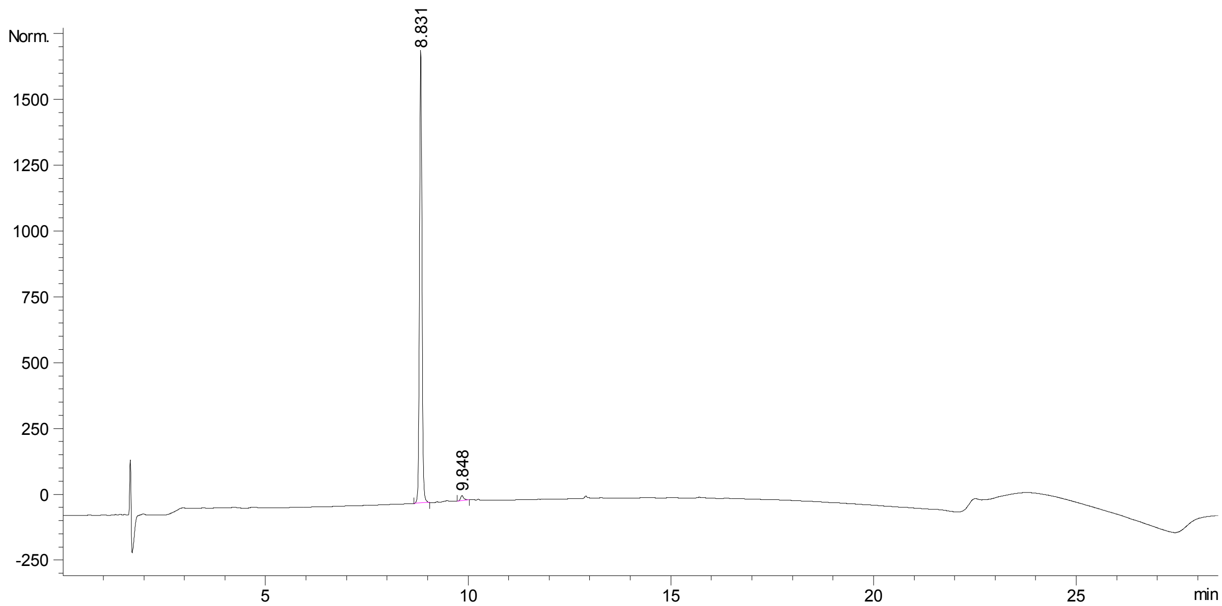
Note: The commercially available d_8-Phe$_{20}$ (98%) contains incomplete deuteration at Hα as reported by Li, X.; *et. al.*[2] This is reflected in the MALDI-TOF characterization data.


a.i.
JZ-II-292-O18
Calculated masses:
[M+H]+: 1793.19
[M+Na]+: 1815.17
[M+K]+: 1831.14
1793.3105
1815.3144
1831.2727
m/z


a.i.
JZ-II-292-O18
Calculated masses:
[M+H]+: 1793.19
[M+Na]+: 1815.17
[M+K]+: 1831.14
[M-H+2Na]+: 1837.15
1793.3105
1815.3144
1792.3121
1814.3172
1831.2727
1837.2745
1830.2624
m/z

Characterization of peptide **2d**

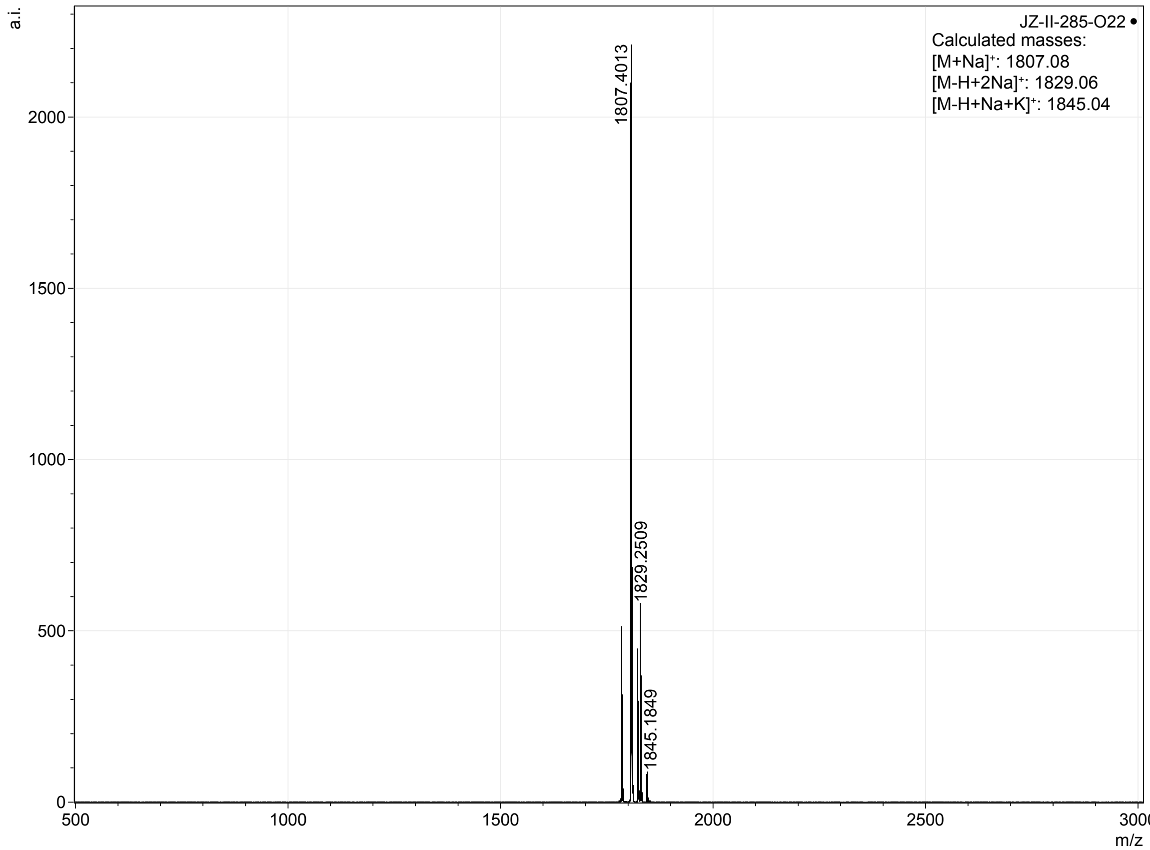


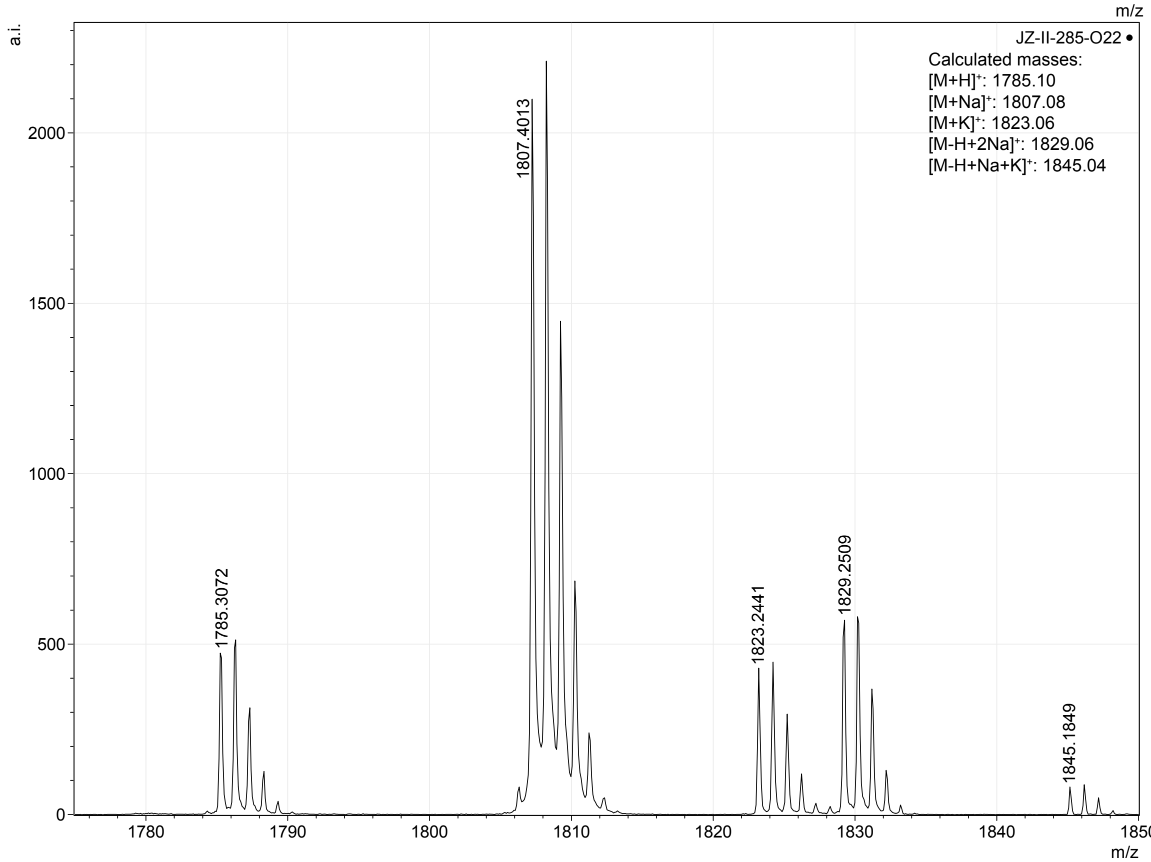


Signal 1: MWD1 A, Sig=214,4 Ref=off

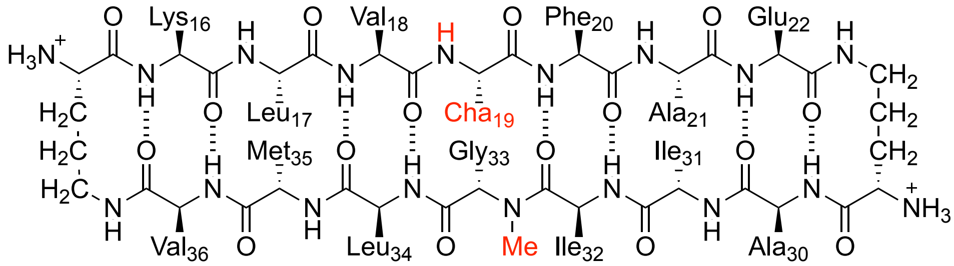
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.831	MM	0.0694	6482.80225	1557.91272	98.5461
2	9.848	MM	0.0910	95.64141	17.52157	1.4539

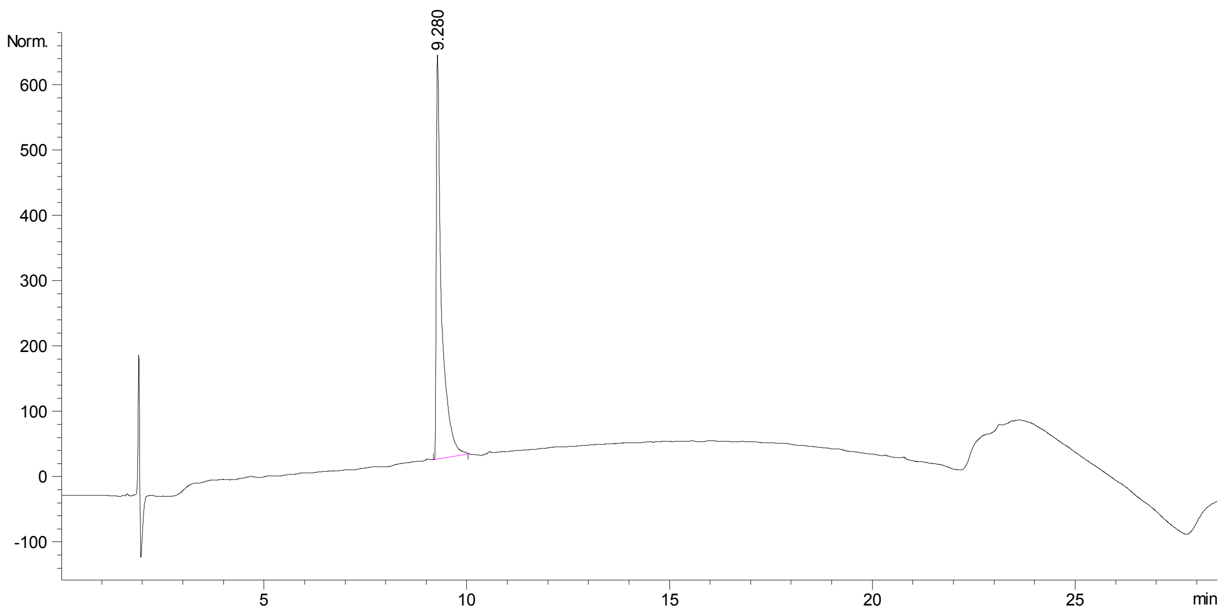
Totals : 6578.44366 1575.43429


a.i.
JZ-II-285-O22
Calculated masses:
[M+Na]+: 1807.08
[M-H+2Na]+: 1829.06
[M-H+Na+K]+: 1845.04
1807.4013
1829.2509
1845.1849
m/z

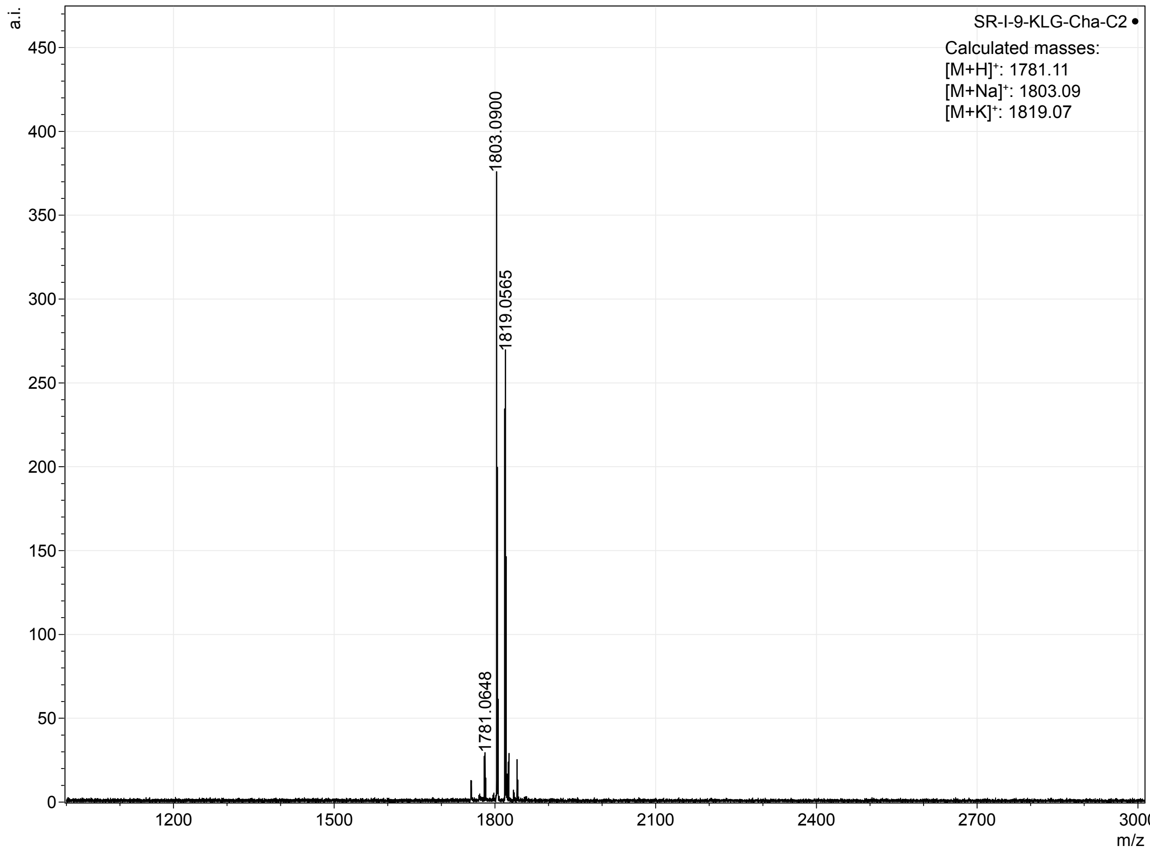

a.i.
JZ-II-285-O22
Calculated masses:
[M+H]+: 1785.10
[M+Na]+: 1807.08
[M+K]+: 1823.06
[M-H+2Na]+: 1829.06
[M-H+Na+K]+: 1845.04
1785.3072
1807.4013
1823.2441
1829.2509
1845.1849
m/z

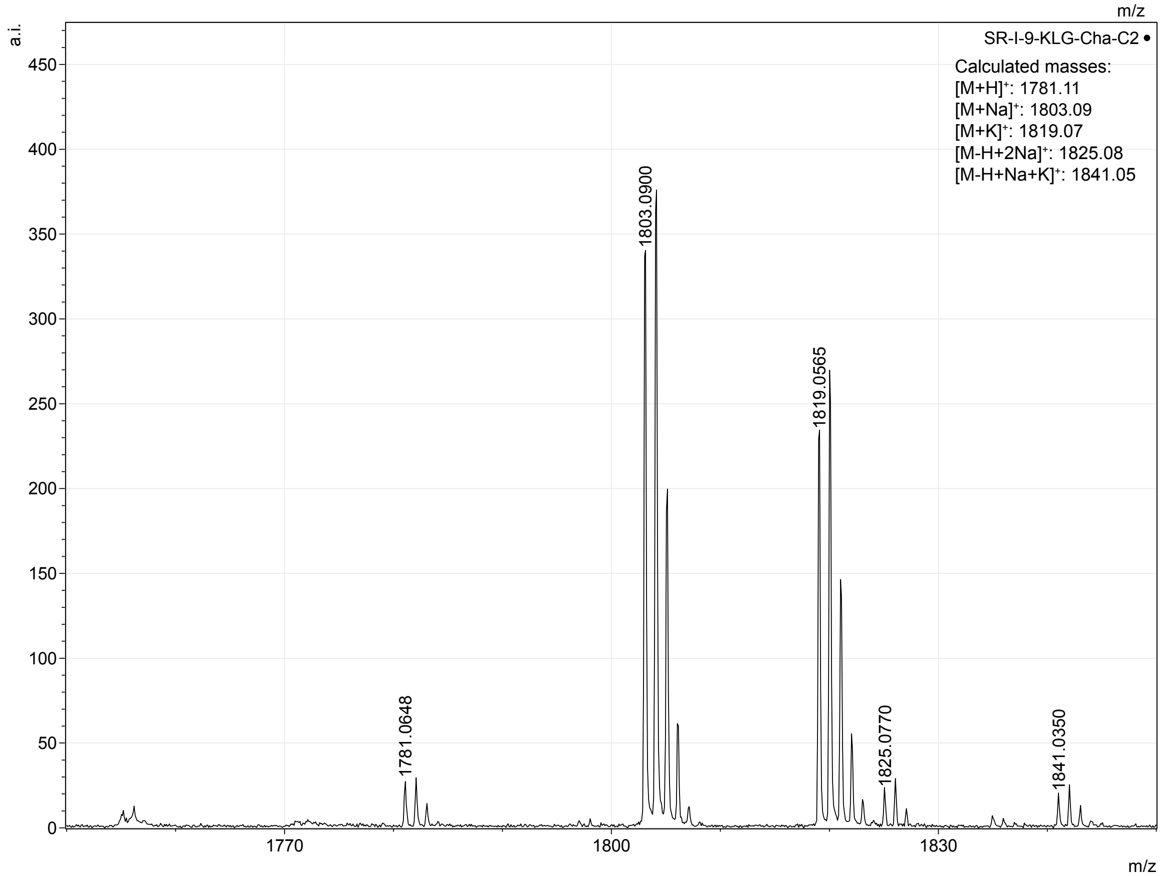
Characterization of peptide **3a**



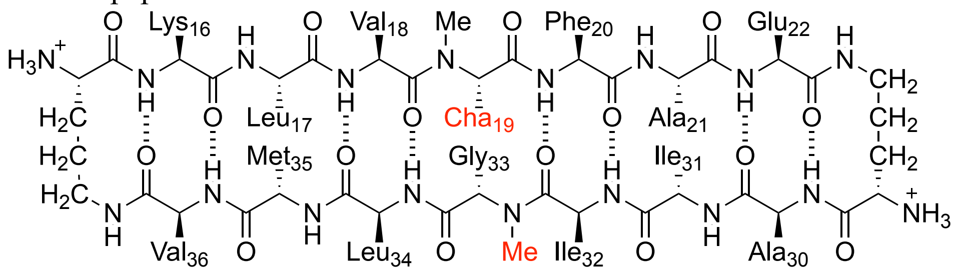


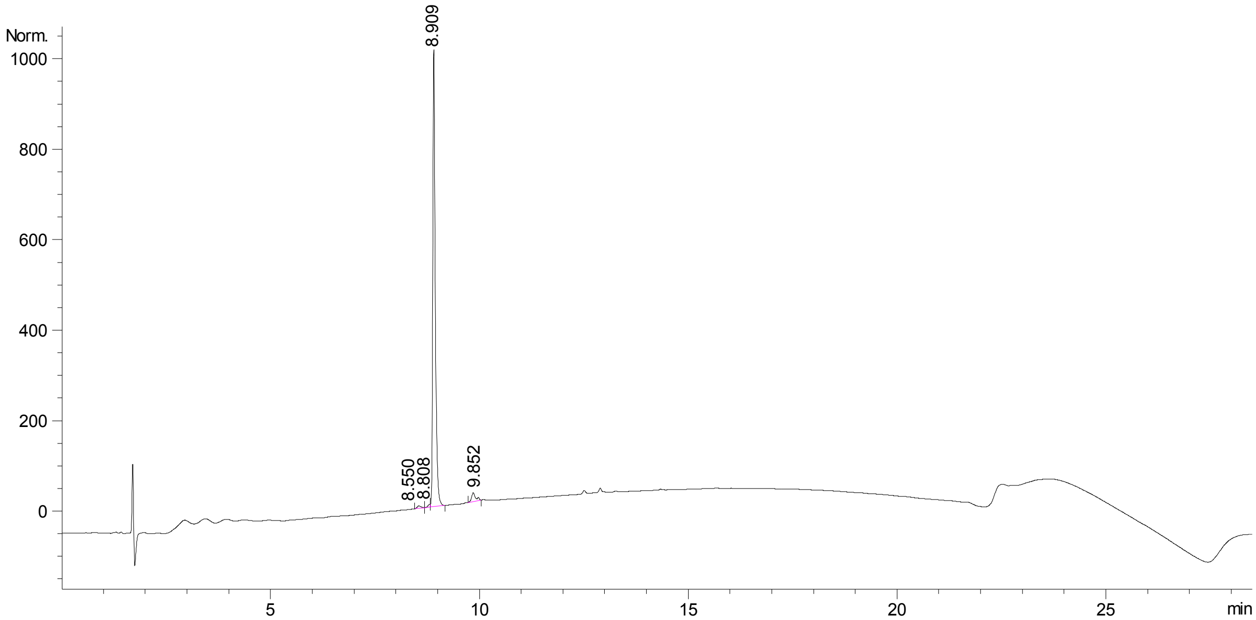
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	9.280	MM	0.1487	4993.32324	559.54315	100.0000
Totals :				4993.32324	559.54315	


SR-I-9-KLG-Cha-C2
Calculated masses:
[M+H]+: 1781.11
[M+Na]+: 1803.09
[M+K]+: 1819.07
1803.0900
1819.0565
1781.0648
a.i.
m/z

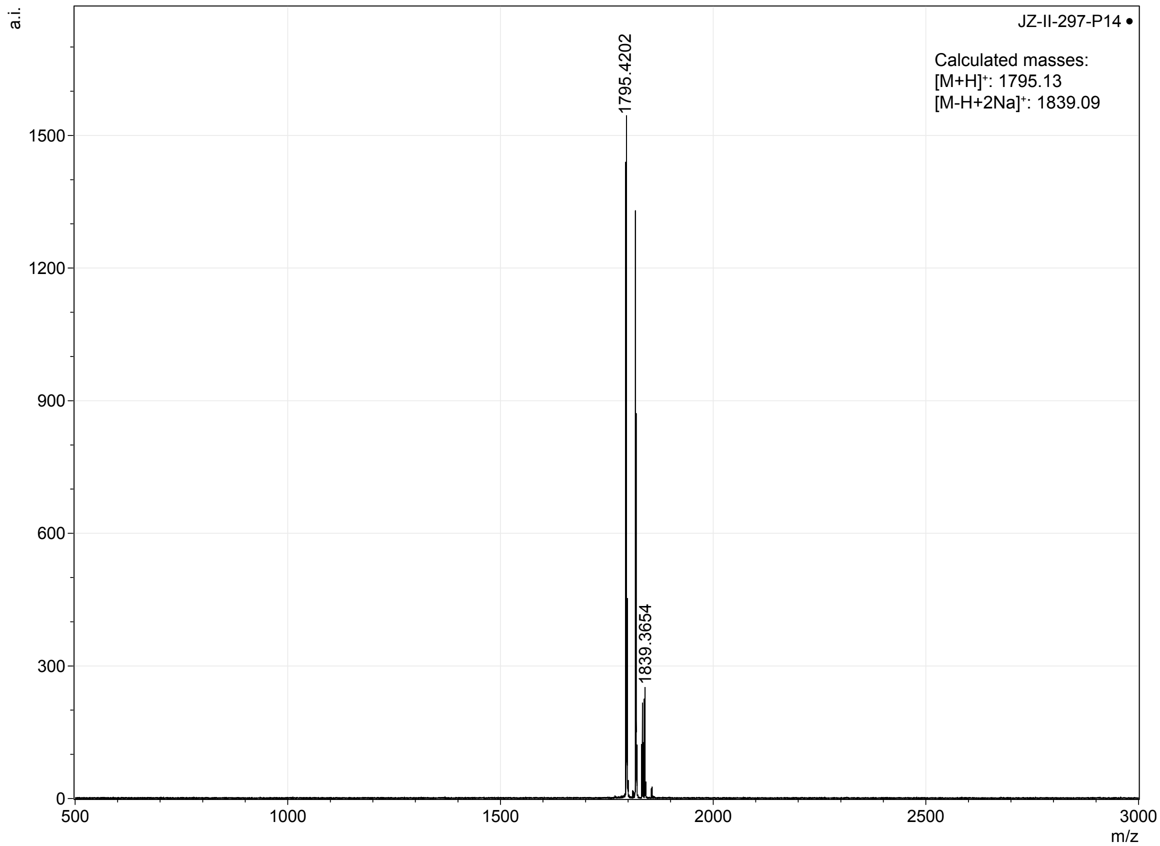

SR-I-9-KLG-Cha-C2
Calculated masses:
[M+H]+: 1781.11
[M+Na]+: 1803.09
[M+K]+: 1819.07
[M-H+2Na]+: 1825.08
[M-H+Na+K]+: 1841.05
1803.0900
1819.0565
1781.0648
1825.0770
1841.0350
a.i.
m/z

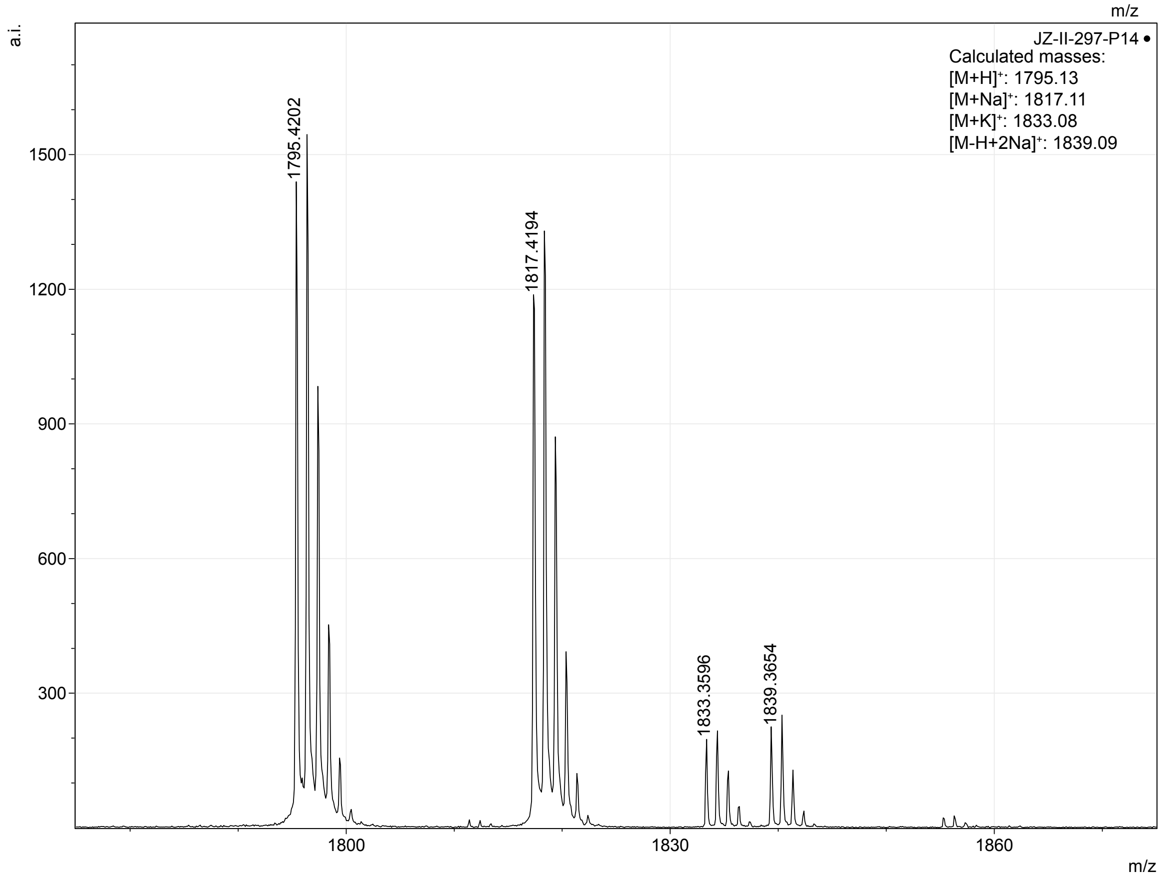
Characterization of peptide **3b**



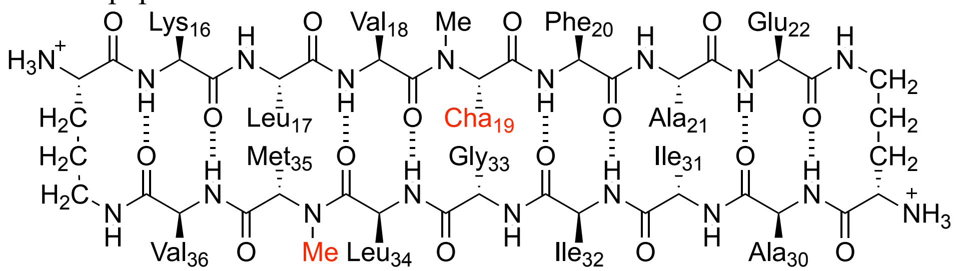


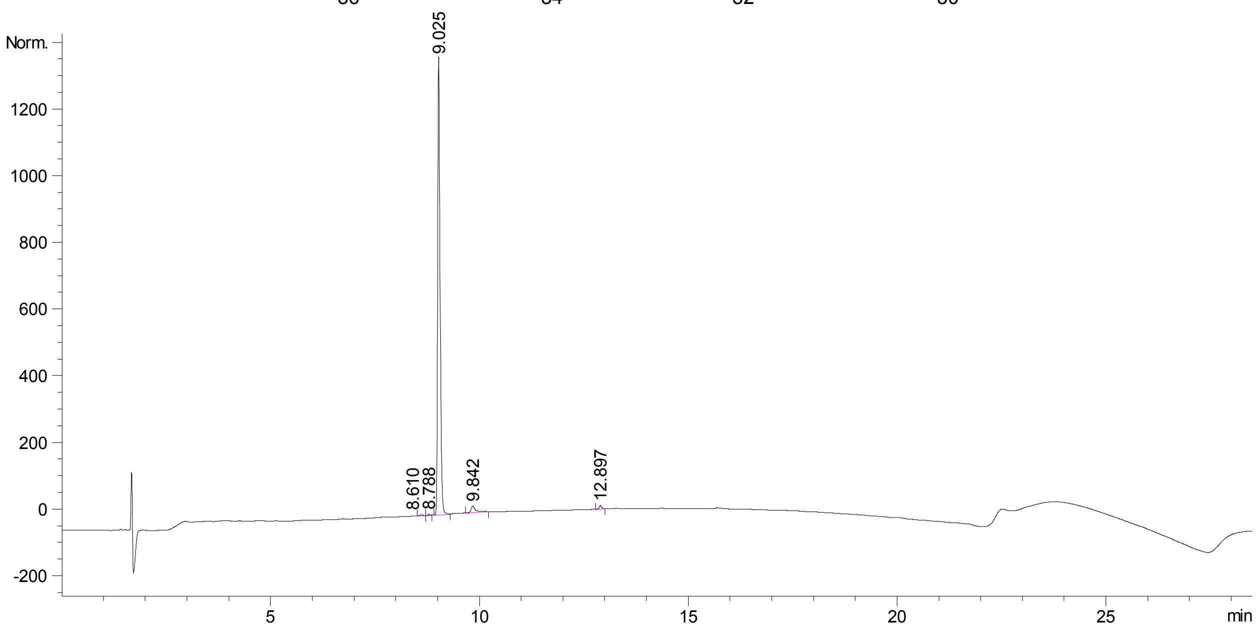
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.550	MM	0.1083	31.36832	4.82610	0.7690
2	8.808	MF	0.0674	24.03691	5.94412	0.5893
3	8.909	FM	0.0711	3902.27173	915.22583	95.6695
4	9.852	MM	0.1110	121.23396	18.20858	2.9722
Totals :				4078.91091	944.20463	


a.i.
JZ-II-297-P14
Calculated masses:
[M+H]+: 1795.13
[M-H+2Na]+: 1839.09
1795.4202
1839.3654
m/z

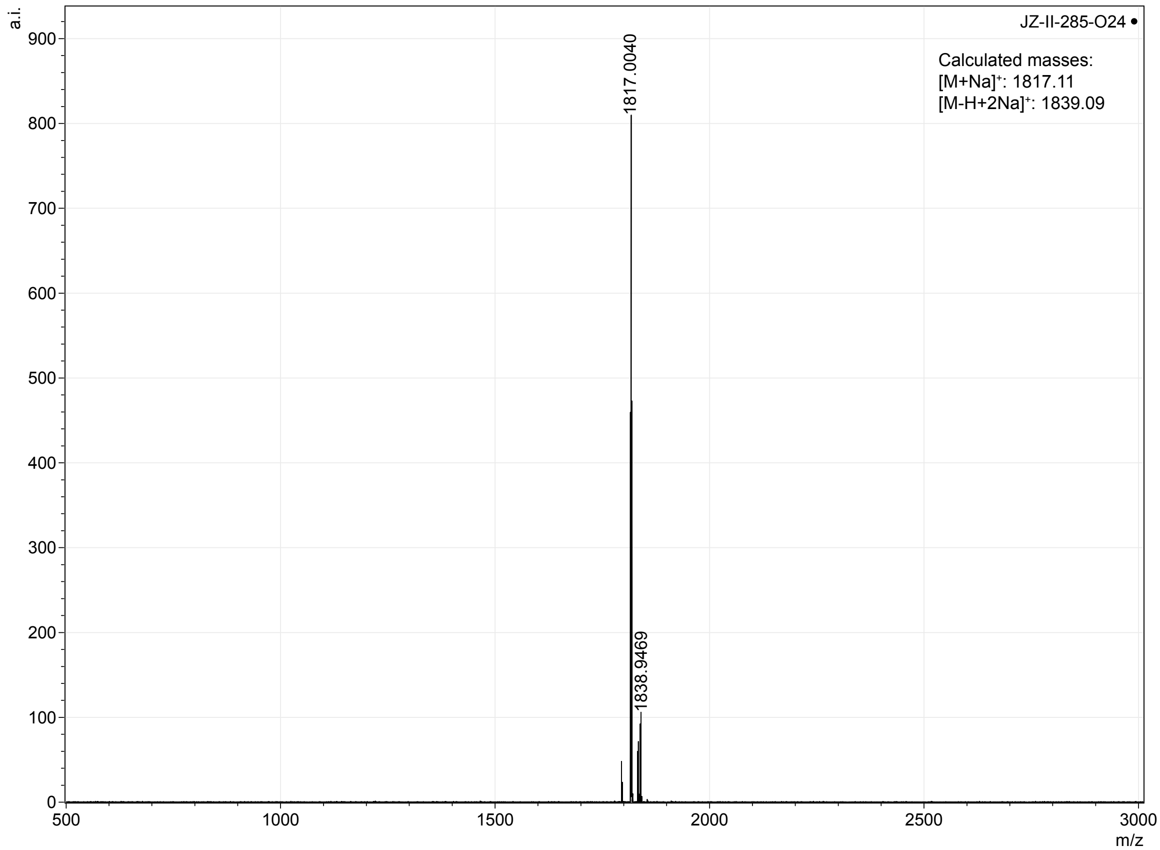

a.i.
JZ-II-297-P14
Calculated masses:
[M+H]+: 1795.13
[M+Na]+: 1817.11
[M+K]+: 1833.08
[M-H+2Na]+: 1839.09
1795.4202
1817.4194
1833.3596
1839.3654
m/z

Characterization of peptide **3c**

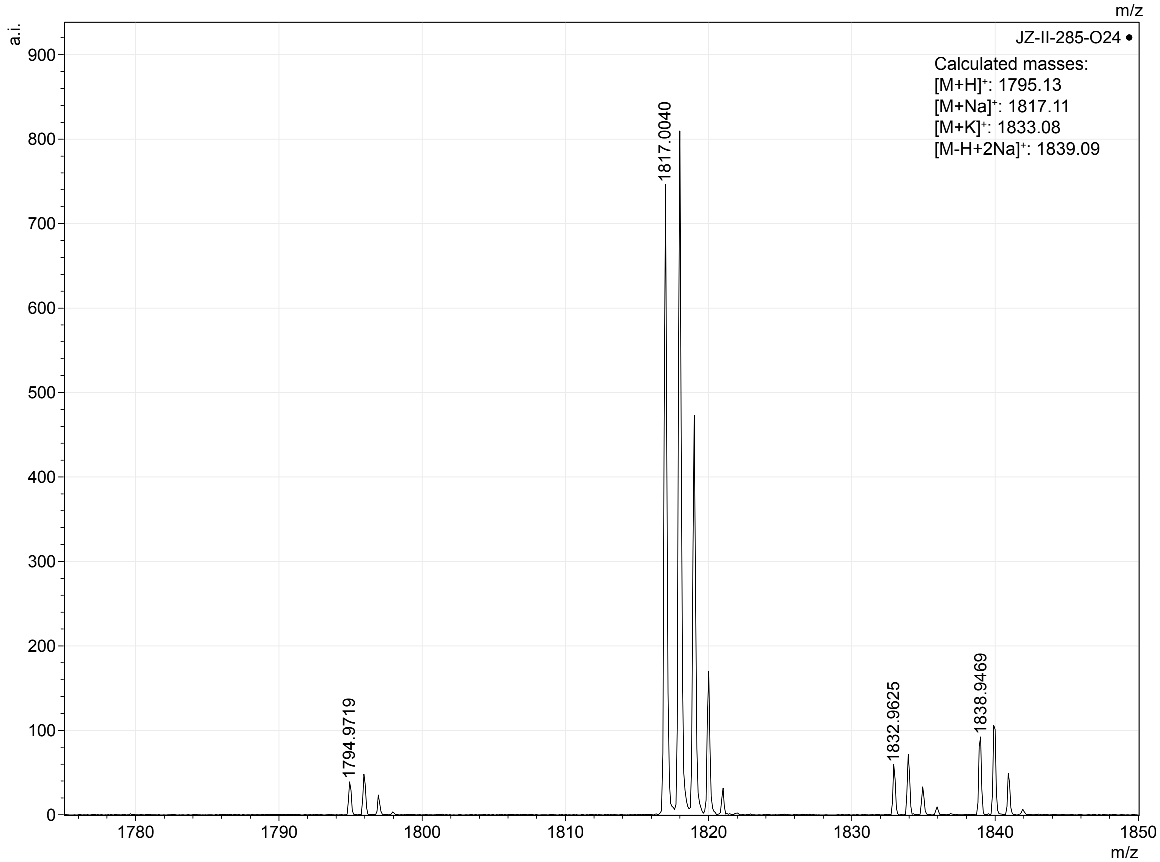




Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.610	MM	0.1104	19.06510	2.87894	0.3598
2	8.788	MM	0.0902	16.20865	2.99636	0.3059
3	9.025	MM	0.0675	5045.52930	1245.53979	95.2275
4	9.842	MM	0.1477	170.85822	19.28419	3.2247
5	12.897	MM	0.0808	46.73169	9.63660	0.8820

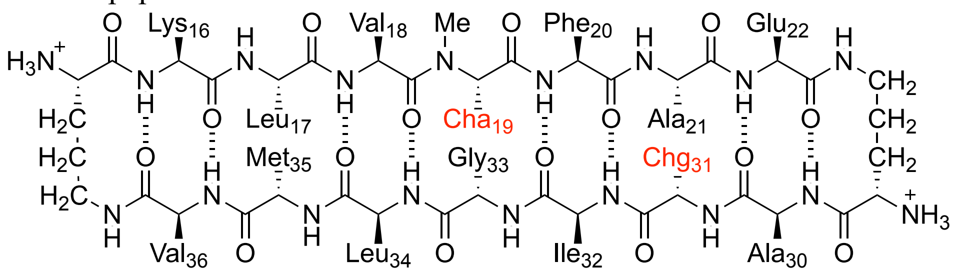


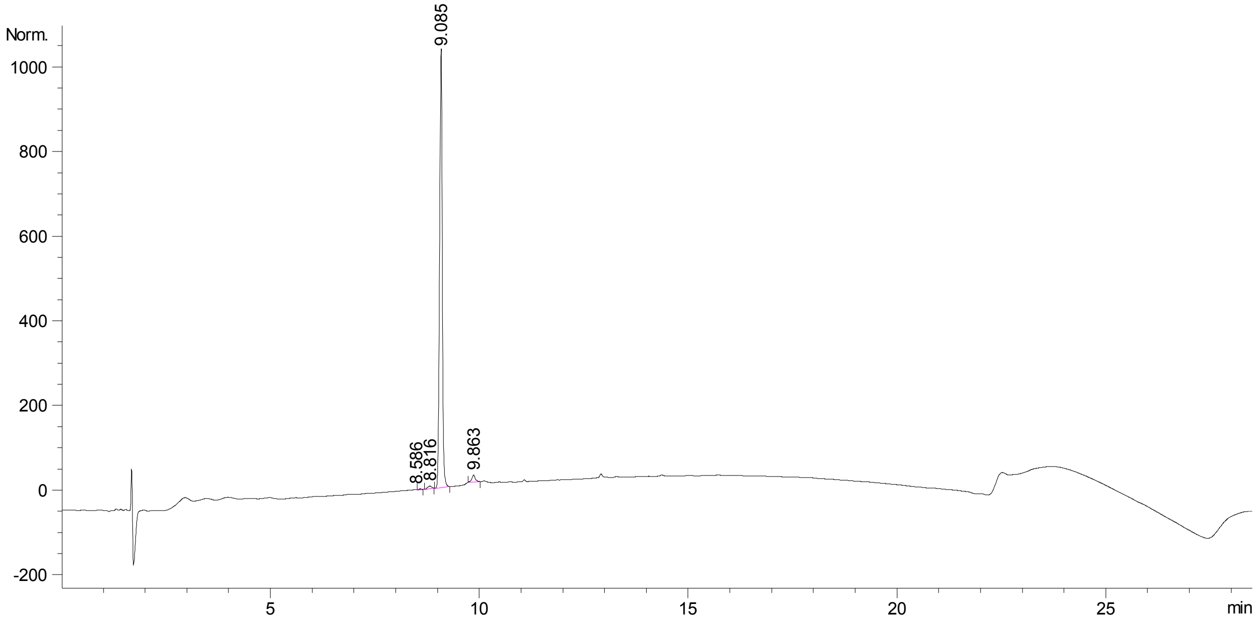
a.i.
JZ-II-285-O24
Calculated masses:
[M+Na]+: 1817.11
[M-H+2Na]+: 1839.09
1817.0040
1838.9469
m/z



a.i.
JZ-II-285-O24
Calculated masses:
[M+H]+: 1795.13
[M+Na]+: 1817.11
[M+K]+: 1833.08
[M-H+2Na]+: 1839.09
1794.9719
1817.0040
1832.9625
1838.9469
m/z

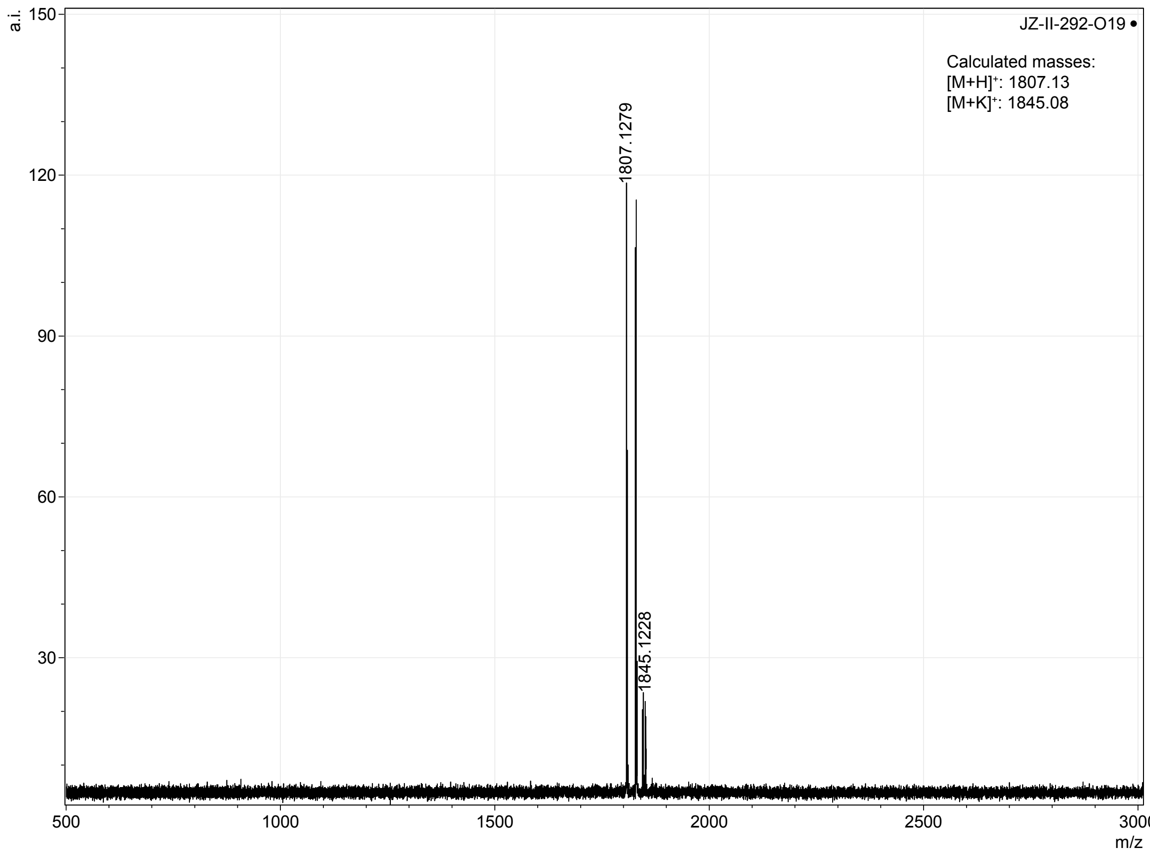
Characterization of peptide **4a**

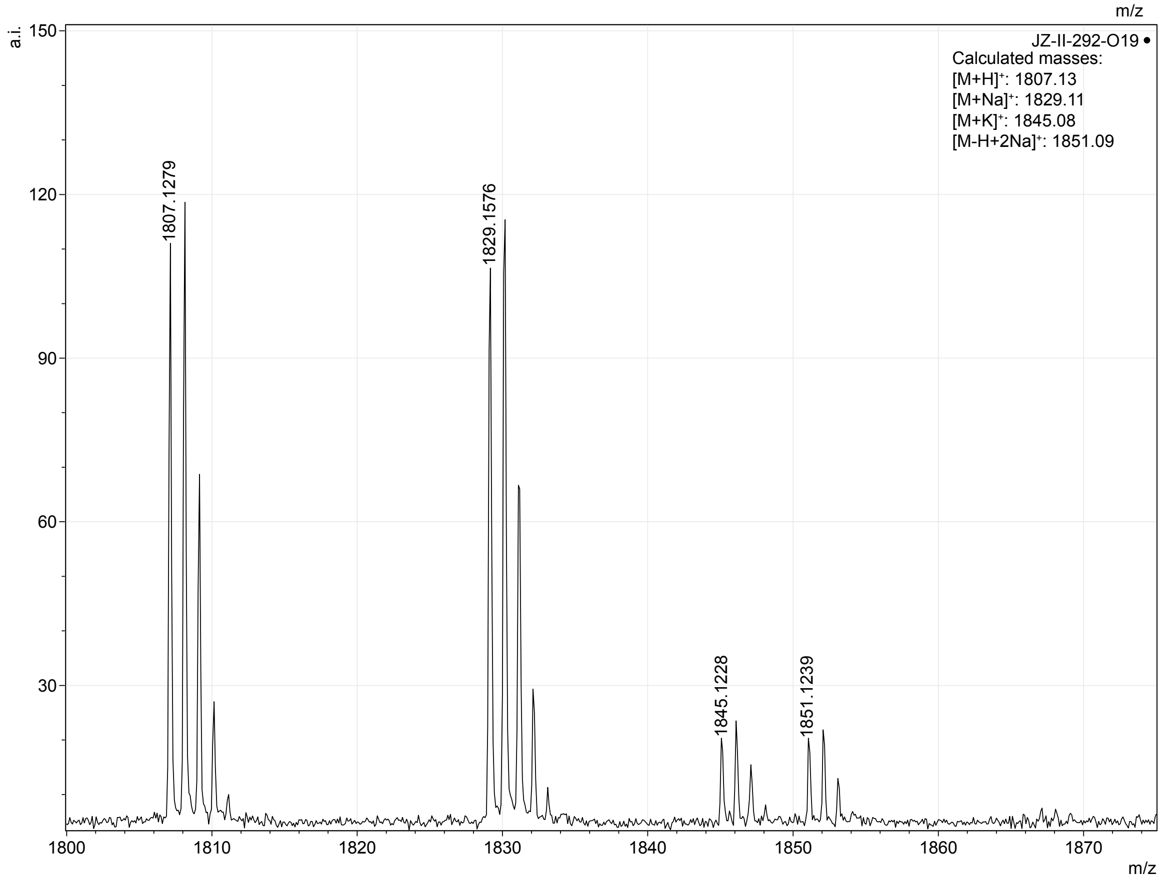




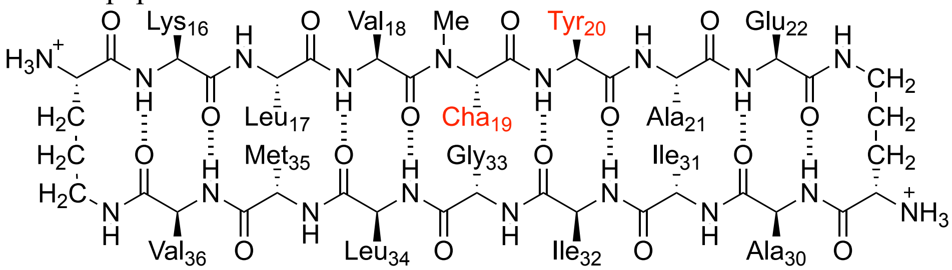
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.586	MM	0.0781	9.93242	2.11904	0.2322
2	8.816	MM	0.1096	37.04632	5.63426	0.8662
3	9.085	MM	0.0735	4142.23828	938.64838	96.8498
4	9.863	MM	0.0959	87.75318	15.24404	2.0518

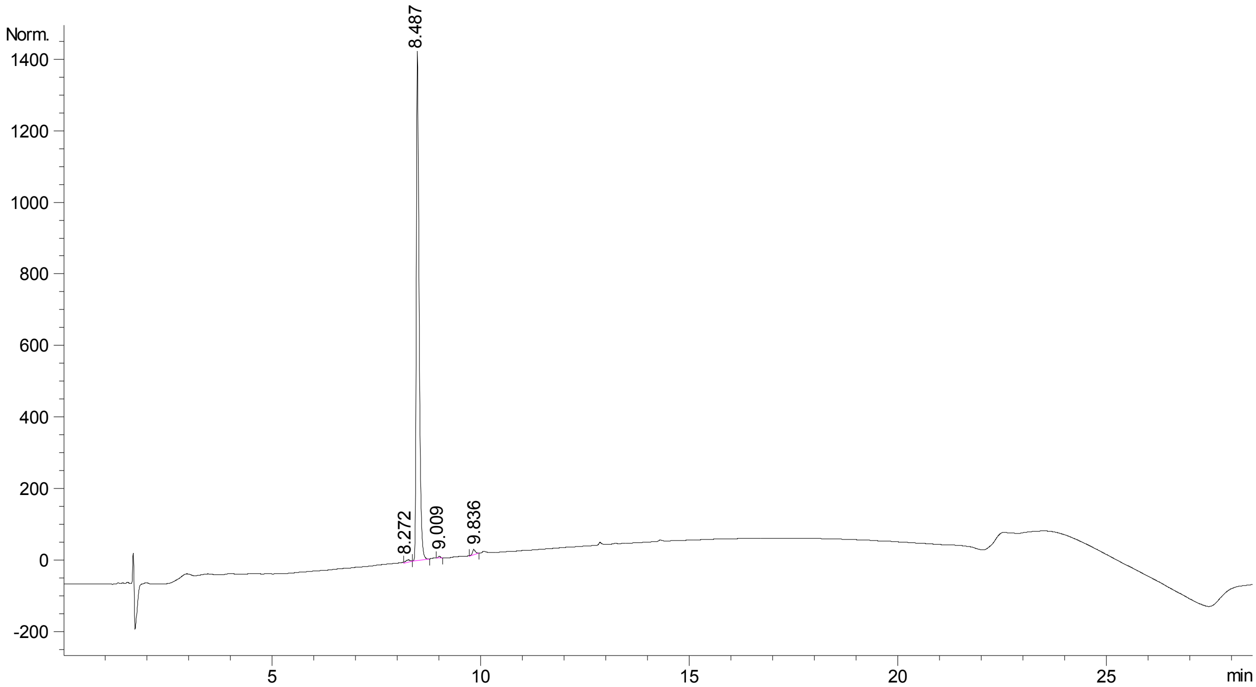
Totals : 4276.97020 961.64571


JZ-II-292-O19
Calculated masses:
[M+H]+: 1807.13
[M+K]+: 1845.08
1807.1279
1845.1228
a.i.
m/z


JZ-II-292-O19
Calculated masses:
[M+H]+: 1807.13
[M+Na]+: 1829.11
[M+K]+: 1845.08
[M-H+2Na]+: 1851.09
1807.1279
1829.1576
1845.1228
1851.1239
a.i.
m/z

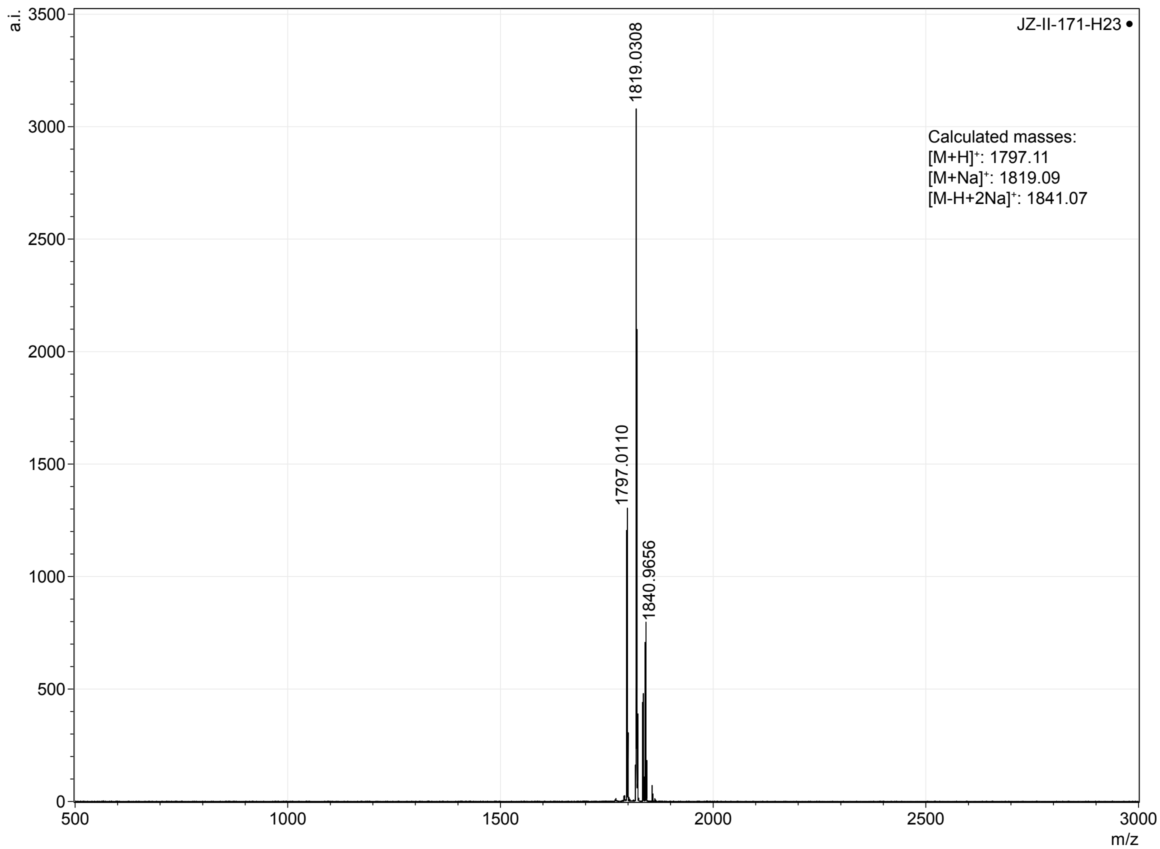
Characterization of peptide **4b**



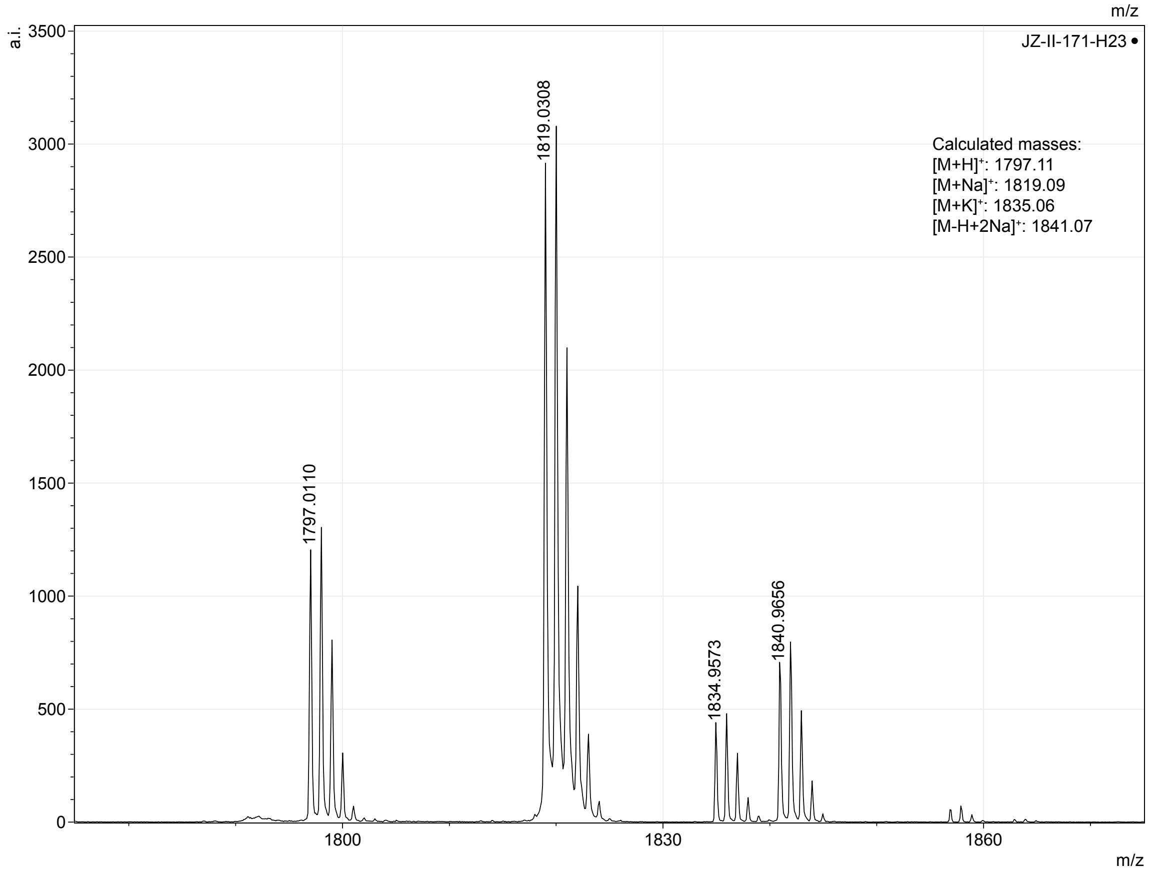


Signal 1: MWD1 A, Sig=214,4 Ref=off

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.272	MM	0.1254	43.77868	5.81827	0.7408
2	8.487	MM	0.0748	5778.48975	1287.12500	97.7799
3	9.009	MM	0.0588	16.15449	4.57509	0.2734
4	9.836	MM	0.0870	71.26708	13.64624	1.2059
Totals :				5909.69000	1311.16459	

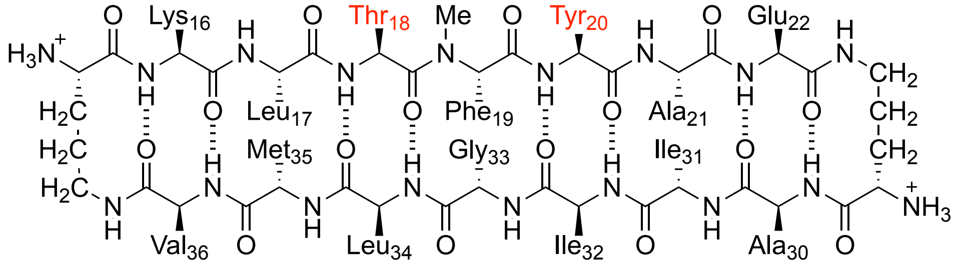


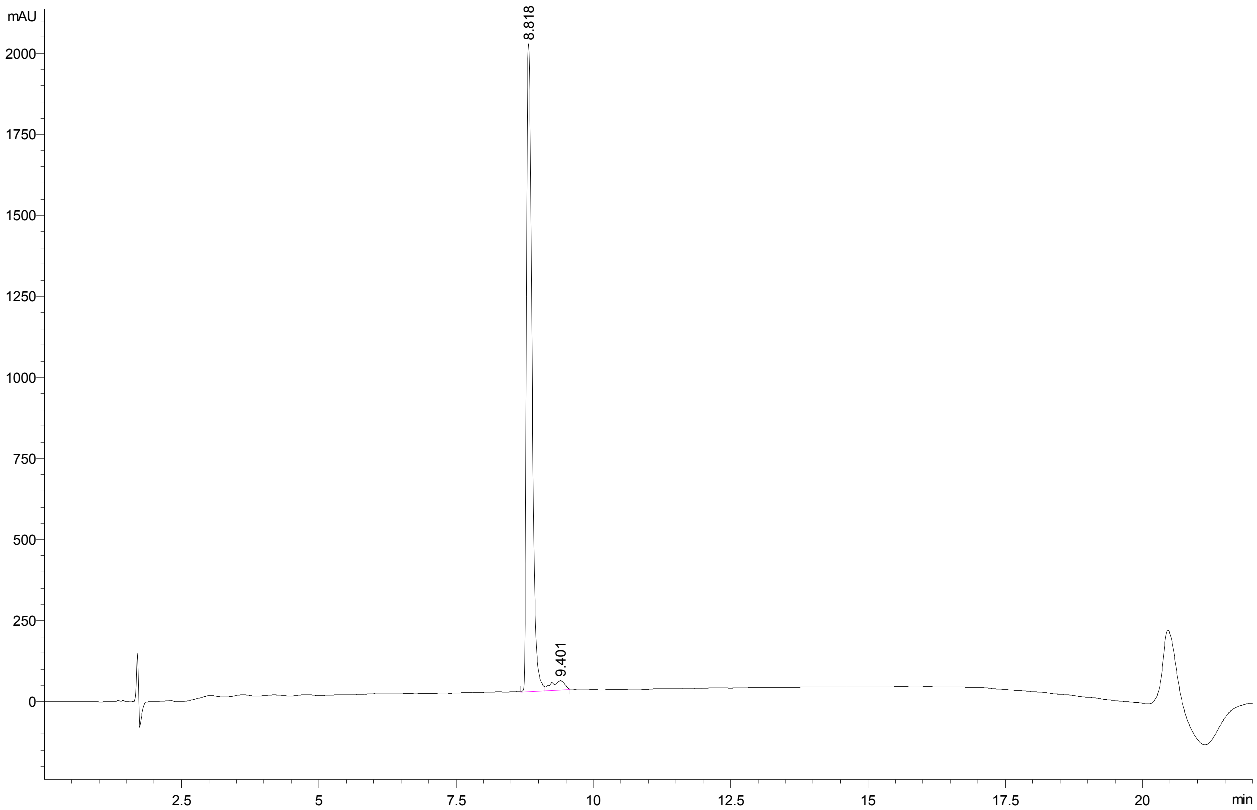
a.i.
JZ-II-171-H23
1819.0308
1797.0110
1840.9656
Calculated masses:
[M+H]+: 1797.11
[M+Na]+: 1819.09
[M-H+2Na]+: 1841.07
m/z



a.i.
JZ-II-171-H23
1819.0308
1797.0110
1834.9573
1840.9656
Calculated masses:
[M+H]+: 1797.11
[M+Na]+: 1819.09
[M+K]+: 1835.06
[M-H+2Na]+: 1841.07
m/z

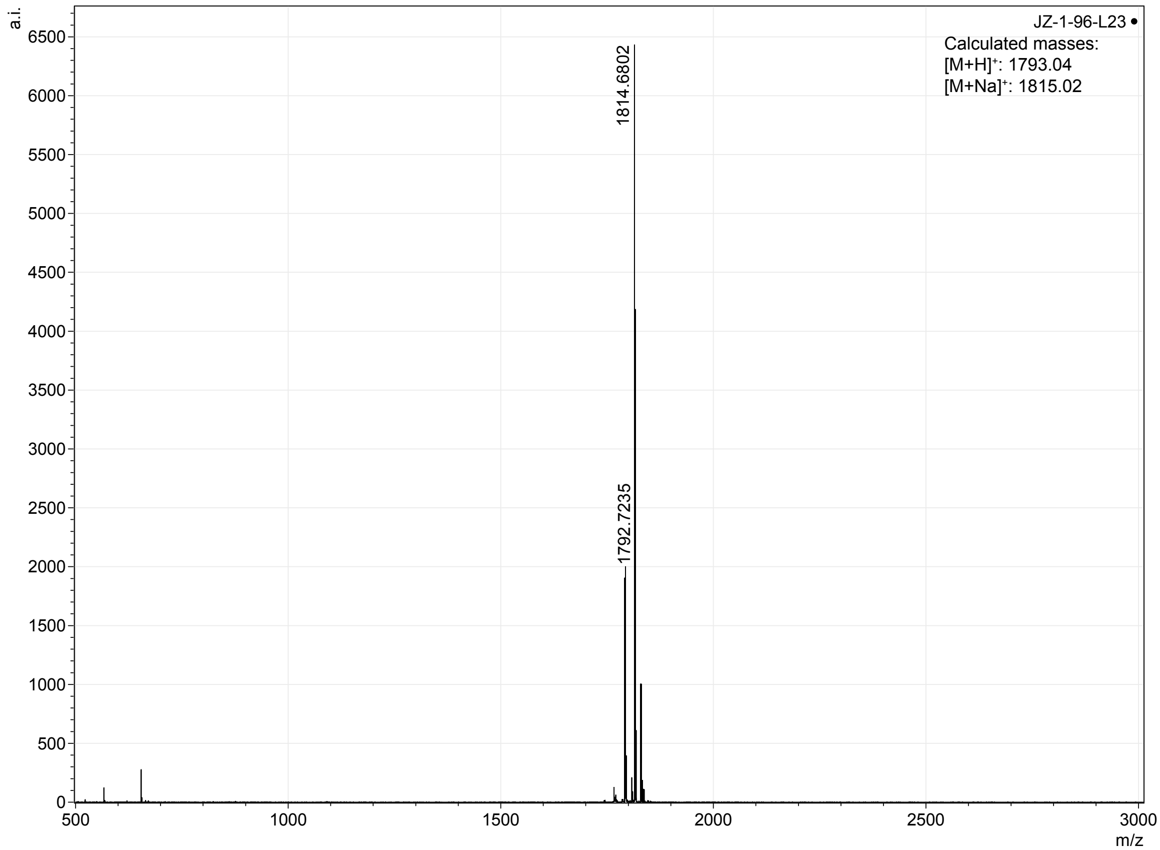
Characterization of peptide **5**

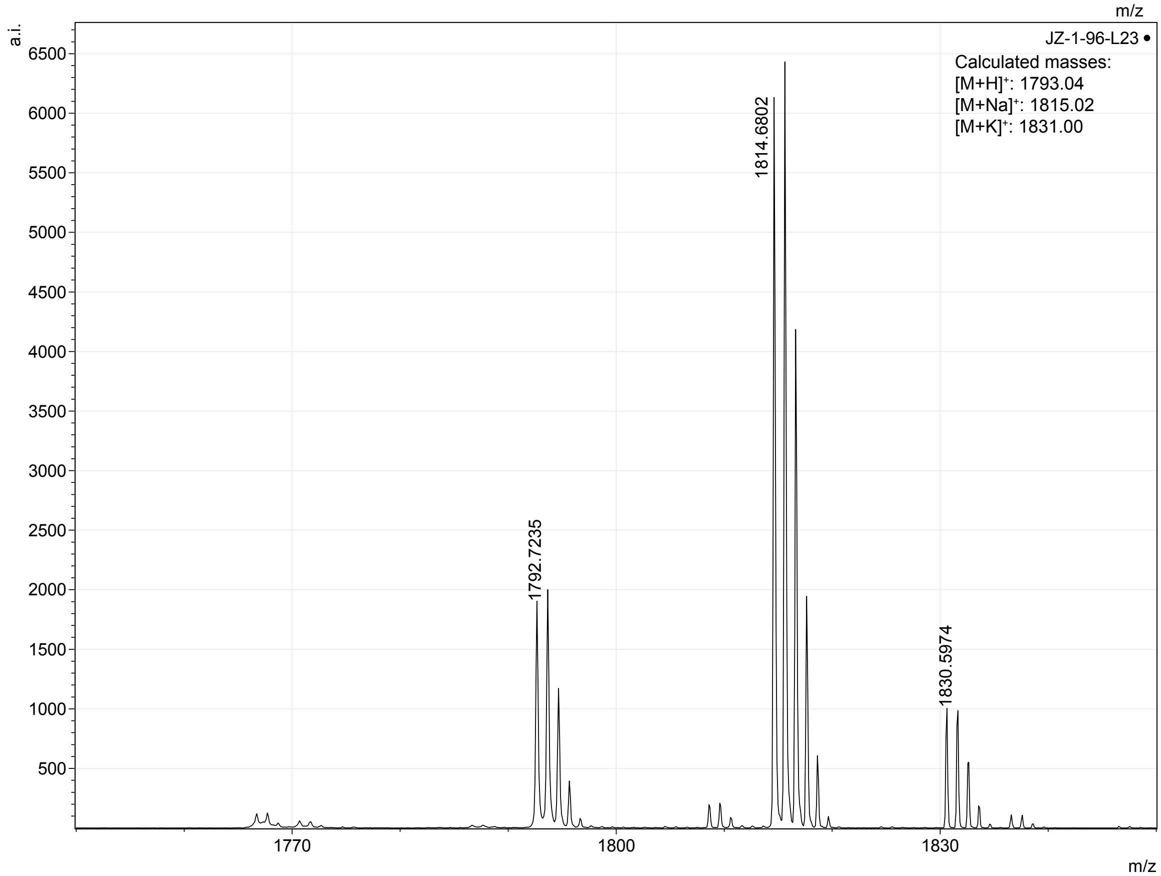




Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	8.818	MF	0.1172	1.40598e4	2000.00208	96.5700
2	9.401	FM	0.2852	499.37866	29.17830	3.4300

Totals : 1.45591e4 2029.18037


JZ-1-96-L23
Calculated masses:
[M+H]+: 1793.04
[M+Na]+: 1815.02
1814.6802
1792.7235
a.i.
m/z


JZ-1-96-L23
Calculated masses:
[M+H]+: 1793.04
[M+Na]+: 1815.02
[M+K]+: 1831.00
1814.6802
1792.7235
1830.5974
a.i.
m/z