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S U P P L E M E N T A R Y M A T E R I A L

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Sensitive and Regenerable Organochalcogen Probes for the Colorimetric Detection of Thiols

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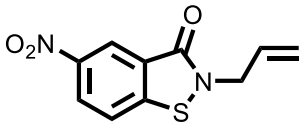
Contents

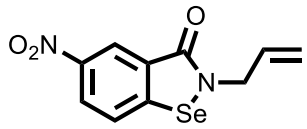
Experimental Section:

All NMR experiments were carried out on 400 MHz spectrometers in $CDCl_3$ or DMSO-d_6 and NMR chemical shifts are reported in ppm referenced to the solvent peaks of $CDCl_3$ (7.26 ppm for ^{1}H and 77.16 (± 0.06) ppm for ^{13}C, respectively) or DMSO-d_6 (2.50 ppm for ^{1}H and 39.50 ppm for ^{13}C, respectively). ^{77}Se NMR chemical shifts are reported relative to dimethyl selenide (0 ppm) in reference to diphenyl diselenide 461.0 ppm external standard. High resolution mass spectra (HRMS) and low resolution mass spectra (LRMS) are reported for ions of ^{80}Se. Mass analysis is performed on quadruple-time of flight (Q-TOF) mass spectrometer equipped with an ESI source (+ve). Ground (mortar and pestle) anhydrous K_2CO_3 powder was dried in an oven at 160 °C for 6 h and stored in a desiccator prior to use. Silica gel (60 mesh size) was used for column chromatography. TLC analysis of reaction mixtures was performed using silica gel plates. Reactions were concentrated at reduced pressure on a rotary evaporator. Amide precursors for the preparation of compounds **1-4** were prepared according to previously reported methods.[1-3]

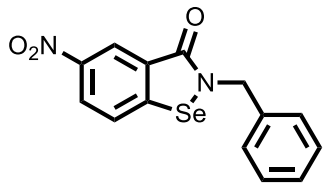


2-Allyl-5-nitrobenzo[d][1,2]selenazol-3(2H)-one (**1**) Copper iodide (79 mg, 0.4 mmol) and 1,10-phenanthroline (75 mg, 0.4 mmol) were dissolved in DMF (6 mL) and the resulting mixture was stirred for 15 min. To this brown colored solution, N-allyl-2-chloro-5-nitrobenzamide (0.4 g, 1.7 mmol), selenium powder (0.065 g, 2.0 mmol), and K_2CO_3 (0.35 g, 2.55 mmol) were added. The resulting reaction mixture was heated at 110^{o} C for 5 hours. The progress of reaction was monitored by TLC. The reaction mixture was then poured into brine solution (60 mL) and stirred for 3 h. The product was extracted with ethyl acetate (3 × 40 mL). The combined organic extracts were washed with distilled water (2 × 50 mL), dried over sodium sulphate and concentrated. The resulting yellow oil was purified by column

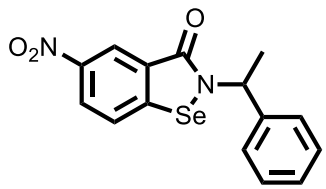
chromatography on SiO_2 eluted with dichloromethane. Yield 0.36 g (92%), mp 121-123 °C (123 °C).[1] ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.87 (d, *J* = 2.1 Hz, 1H), 8.44 (dd, *J* = 2.2, 8.8 Hz, 1H), 7.69 (d, *J* = 8.8 Hz, 1H), 6.0-5.86 (m, 1H), 5.43-5.32 (m, 2H), 4.52 (d, *J* = 6.3 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 163.7, 146.4, 146.1, 131.6, 126.2, 125.3, 122.7, 121.3, 120.4, 46.6. LRMS-ES$^+$*m/z*: 237.0217 (Calculated for $C_{10}H_8N_2O_3S + H^+$: 237.0328).



2-Allyl-5-nitrobenzo[d][1,2]selenazol-3(2H)-one (2) Allyl group containing Se-N heterocycle **2** was synthesized from N-allyl-2-chloro-5-nitrobenzamide[1] (0.5 g, 2.1 mmol) by using CuI (98 mg, 0.5 mmol), 1,10-phenanthroline (93 mg, 0.5 mmol), selenium powder (0.197 g, 2.5 mmol), and K_2CO_3 (0.43 g, 3.1 mmol) in DMF (8 mL). Reaction mixture was heated at 110°C for 24 hours. The progress of reaction was monitored by TLC. The reaction mixture was then poured into brine solution (80 mL) and stirred for 3 h. The product was extracted with ethyl acetate (3 × 50 mL). The combined organic extracts were washed with distilled water (2 × 50 mL) and dried over sodium sulphate. Ethyl acetate was evaporated under reduced pressure at 40 °C to obtain the crude product which was purified by column chromatography over silica gel by using hexane: ethyl acetate (8:2) as mobile phase. Yield 0.43 g (74%), mp 172-174 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.41 (d, *J* = 2.3 Hz, 1H), 8.36 (dd, *J* = 2.3, 8.8 Hz, 1H), 8.26 (d, *J* = 8.8 Hz, 1H), 5.98-5.85 (m, 1H), 5.28-5.16 (m, 2H), 4.34 (d, *J* = 5.6 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.2, 148.0, 146.5, 134.6, 129.3, 127.9, 125.7, 122.4, 118.4, 46.1. ^{77}Se NMR ($CDCl_3$): δ 866.1. HRMS-ES$^+$*m/z*: 306.9619 (Calculated for $C_{10}H_8N_2O_3{}^{80}Se$ + Na: 306.9593).

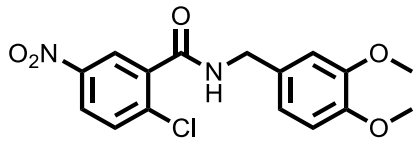


2-Benzyl-5-nitrobenzo[d][1,2]selenazol-3(2H)-one (3) Nitro group containing Se-N heterocycle **3** was synthesized from corresponding N-benzyl-2-chloro-5-nitrobenzamide[2] (0.4 g, 1.4 mmol), CuI (65 mg, 0.35 mmol), 1,10-phenanthroline (62 mg, 0.35 mmol), selenium powder (0.13 g, 1.7 mmol), and K_2CO_3 (0.29 g, 2.1 mmol) in DMF (7 mL) and refluxing for 16 h at 110^{o}C under nitrogen atmosphere. Progress of reaction was monitored by TLC. After this, reaction mixture was poured over brine solution (70 mL) and stirred for 3 h. Product was extracted by using ethyl acetate (50.0 mL x 3). Combined ethyl acetate layer was washed with distilled water (50 mL x 2), dried over sodium sulphate and ethyl acetate was distilled under reduced pressure at 40^o C. Resulted dark brown colored oil, which was purified by column chromatography using dichloromethane over silica gel gave yellow powder. Yield 0.37 g (82%). mp 206-208 °C (mp 206-208 ^{o}C).[2] ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.89 (d, *J* = 2.3 Hz, 1H), 8.39 (dd, *J* = 8.7, 2.3 Hz, 1H), 7.73 (d, *J* = 8.8 Hz, 1H), 7.37-7.34 (m, 5H), 5.02 (s, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.5, 147.0, 145.4, 136.5, 129.1, 128.8, 128.7, 128.6, 126.0, 124.9, 124.3, 49.1. ^{77}Se NMR ($CDCl_3$): δ 866.4 ppm. HRMS-ES$^+$ *m/z*: 334.9938 (Calculated for $C_{14}H_{10}N_2O_3{}^{80}Se + H^+$: 334.9930).



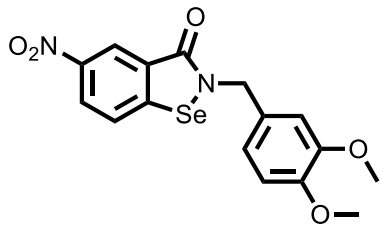
5-Nitro-2-(1-phenylethyl)benzo[d][1,2]selenazol-3(2H)-one (4) Nitro group containing Se-N heterocycle **4** was synthesized from 2-chloro-5-nitro-N-(1-phenylethyl)benzamide[3] (0.5 g, 1.6 mmol), CuI (78 mg, 0.4 mmol), 1,10-phenanthroline (73 mg, 0.4 mmol), selenium

powder (0.15 g, 2.0 mmol), and K_2CO_3 (0.33 g, 2.4 mmol) in DMF (8 mL). Brown colored reaction mixture was refluxed at 110^o C using refluxing condenser under nitrogen atmosphere. Progress of reaction was monitored by TLC. After this, reaction mixture poured over brine solution (80 mL) and stirred for 3 h. Product was extracted by using ethyl acetate (50.0mL x 3). Combined ethyl acetate layer was washed with distilled water (50 mL x 2), dried over sodium sulphate and ethyl acetate was distilled under reduced pressure at 40^o C. Resulted yellow colored oil, which was purified by column chromatography using dichloromethane over silica gel. Yield 0.46 g (81%). Mp 159-161 ^{o}C (163-165 °C).[3] ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.8 (d, *J* = 2.2 Hz, 1H), 8.34 (dd, *J* = 2.2, 8.7 Hz, 1H), 7.74 (d, *J* = 8.7 Hz, 1H), 7.42-7.34 (m, 5H), 5.87 (q, *J* = 6.7 Hz, 1H), 1.74 (d, *J* = 6.8 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.0, 146.8, 145.7, 140.6, 129.3, 128.97, 128.77, 127.3, 125.7, 125.0, 124.1, 53.6, 19.6. ^{77}Se NMR ($CDCl_3$): 853.6 ppm LRMS-ES$^+$ *m/z*: 349.5665 (Calculated for $C_{15}H_{12}N_2O_3$ ^{80}Se + H^+ : 349.0091).



2-Chloro-N-(3,4-dimethoxybenzyl)-5-nitrobenzamide (precursor of 5) 2-chloro-5-nitrobenzoyl chloride (0.38 g, 1.7 mmol) was dissolved in dry CH_2Cl_2 (25 mL) in a single neck flask and cooled to 0 ^{o}C. 3,4-dimethoxy benzyl amine (0.37 g, 2.2 mmol) and triethylamine (0.34 g, 3.4 mmol) in CH_2Cl_2 were slowly added to 2-chloro-5-nitrobenzoyl chloride solution by dropping funnel. Resulted reaction mixture stirred for 1 h at 0^{o}C and 12 h at room temperature. After this water (25 mL) was added to reaction flask and stirred for 30 min. CH_2Cl_2 (25 mL) was added to reaction mixture and water layer separated by separating funnel. CH_2Cl_2 layer was washed with 10% HCl (25 mL), and then with water (25 mL). Dichloromethane layer dried over Na_2SO_4, evaporated on rotary evaporator under vacuo.

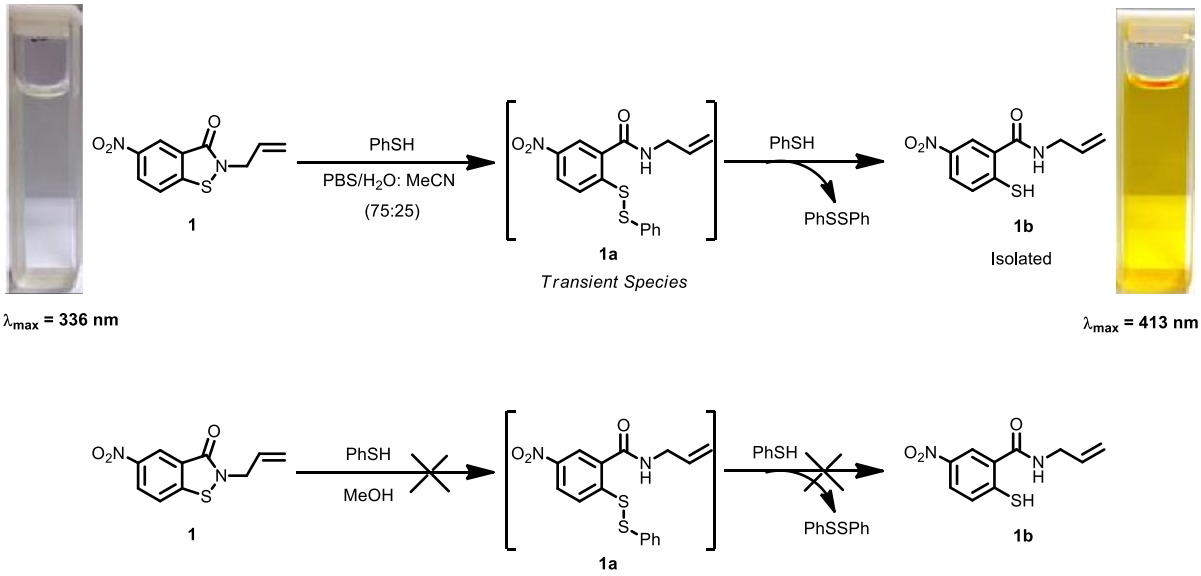
Resulted white solid was passed through silica gel using CH_2Cl_2 to obtain pure amide. Yield 0.56 g (92%), mp 124-126 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.3 (d, *J* = 2.6 Hz, 1H), 8.07 (dd, *J* = 2.7, 8.8 Hz, 1H), 7.49 (d, *J* = 8.8 Hz, 1H), 6.87 (t, *J* = 5.5 Hz, 1H), 6.82-6.79 (m, 2H), 6.76-6.71 (m, 1H),4.48 (d, *J* = 5.6 Hz, 2H), 3.78 (s, 3H), 3.77 (s, 3H) ^{13}C NMR (100 MHz, $CDCl_3$) δ 164.3, 149.1, 148.5, 146.3, 137.7, 136.5, 131.3, 129.8, 125.5, 124.7, 120.2, 111.2, 111.1, 55.88, 55.84, 44.1. HRMS-ES$^+$ *m/z*: 351.0764 (Calculated for $C_{16}H_{15}ClN_2O_5$ + H^+: 351.0742).



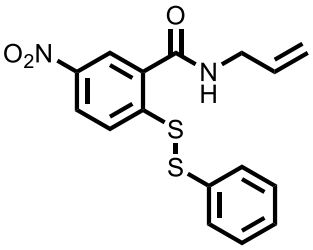
2-(3,4-Dimethoxybenzyl)-5-nitrobenzo[d][1,2]selenazol-3(2H)-one (5) Copper iodide (41 mg, 0.2 mmol) and 1,10-phenanthroline (38 mg, 0.2 mmol) were dissolved in DMF (5 mL) and the resulting mixture was stirred for 15 min. To this brown colored solution, 2-chloro-N-(3,4-dimethoxybenzyl)-5-nitrobenzamide (0.3 g, 0.85 mmol), selenium powder (0.080 g, 1.0 mmol), and K_2CO_3 (0.17 g, 1.3 mmol) were added. The resulting reaction mixture was heated at 110^o C for 20 hours. The progress of reaction was monitored by TLC. The reaction mixture was then poured into brine solution (50 mL) and stirred for 3 h. The product was extracted with ethyl acetate (3 × 40 mL). The combined organic extracts were washed with distilled water (2 × 50 mL) and dried over sodium sulphate. Ethyl acetate was evaporated under reduced pressure at 40 ^{o}C. Resulted crude product was purified by column chromatography over silica gel by using hexane: ethyl acetate (8:2) as mobile phase. Yield 0.21 g (62%), mp 190-192 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.45 (d, *J* = 2.2 Hz, 1H), 8.34 (dd, *J* = 2.2, 8.9 Hz, 1H), 8.25 (d, *J* = 8.8 Hz, 1H), 7.0 (s, 1H), 6.93-6.84 (m, 2H), 4.82 (s, 2H), 3.7 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.3, 149.6, 149.5, 146.9, 145.6, 128.95, 128.91, 125.9, 124.9,

124.2, 121.5, 111.9, 111.2, 56.0, 55.96, 49.2; ^{77}Se NMR ($CDCl_3$): 885.7 ppm. HRMS-ES$^+$ *m/z*: 395.0116 (Calculated for $C_{16}H_{14}N_2O_5{}^{80}Se + H^+$: 395.0141).

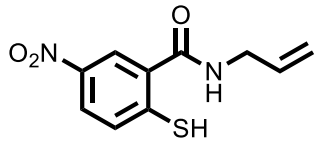
Characterization of intermediates 1a and 1b:



Reaction of probe **1** with PhSH in PBS (10 mM, pH 7.4): acetonitrile (75:25) mixture gives bright yellow colored thiol **1b** (λ_{max} =413 nm) *via* the formation of an unsymmetrical disulphide **1a** as transient species. The formation of **1b** with gradual addition of PhSH (0-2 equiv.) was monitored by UV spectrophotometer at λ 413 nm. However, absorbance at λ 413 nm remains constant beyond 2 equiv. of PhSH, (Figures 37 and 38), indicating 1:2 ratio for probe **1** and PhSH. The formation of thiol **1b** was confirmed by its isolation as yellow solid followed by its characterization by multinuclear NMR and HRMS techniques, while a transient species **1a** was confirmed by analysing reaction mixture by mass spectrometry. On the other hand, formation of thiol **1b** was suppressed in methanol and characteristic red shift was not observed upon addition of PhSH to probe **1** in methanol.

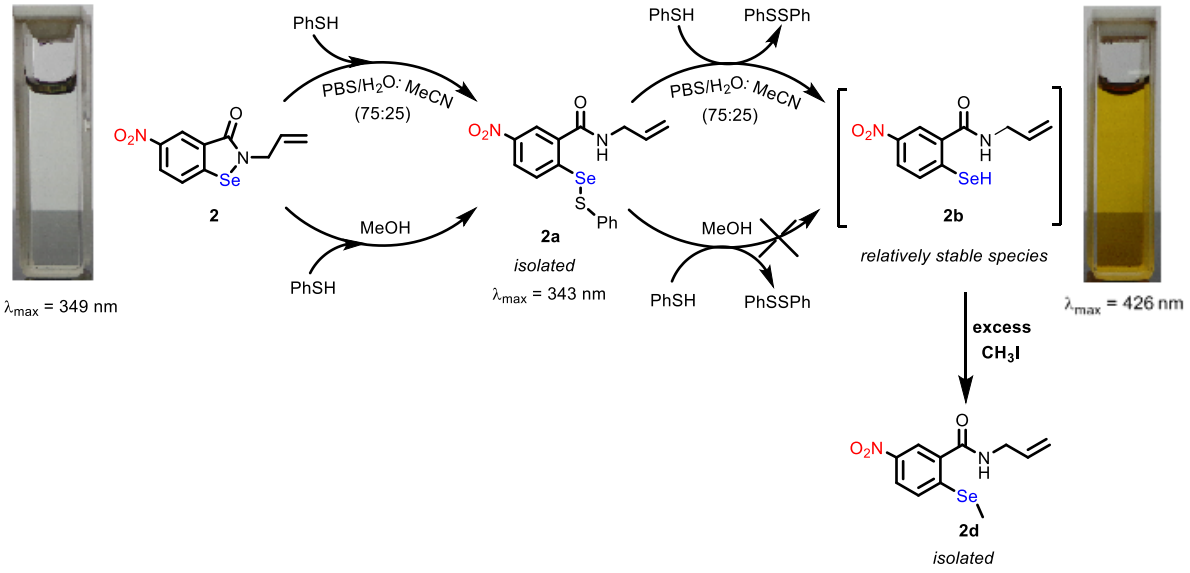


N-allyl-5-nitro-2-(phenyldisulfanyl)benzamide (1a): A freshly prepared (1:1) mixture of isothiazolone **1** and PhSH in distilled water: acetonitrile (75: 25) mixture was analysed by mass spectrometry and a peak was observed at *m*/*z*: 369.0474 (Calculated for $C_{16}H_{14}N_2O_3S_2$ + Na^+) attributed to unsymmetrical disulphide **1a** (See Figure S23).



N-allyl-2-mercapto-5-nitrobenzamide (1b): In a 10 mL capacity round bottom flask isothiazolone **1** (50 mg, 0.2 mmol) was added followed by addition of 5 mL PBS buffer (10mM, pH 7.4): acetonitrile (75: 25) mixture. To this, 4.0 mL PhSH (50 mg, 0.4 mmol) solution from the stock solution (300 mg PhSH in 25 mL acetonitrile) was added in a lot. Reaction mixture was stirred at room temperature for 1 h. Progress of reaction was monitored by TLC. Then, reaction mixture was extracted with ethyl acetate (10 mL x 2). Combined ethyl acetate layer was dried over Na_2SO_4 and solvent was evaporated under reduced pressure at 40 ^oC. Crude product was washed with hexane to obtain colorless solid. Yield 0.036 g (72%), mp 188-190 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 8.31 (s, 1H), 8.08 (dd, *J* =8.9, 2.2 Hz, 1H), 7.45 (d, *J* = 8.7 Hz, 2H), 6.27 (bs, 1H), 6.02-5.84 (m, 1H), 5.49 (s, 1H), 5.33-5.21 (m, 2H), 4.09 (t, *J* = 5.6 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 166.5, 144.72, 144.68, 133.1, 132.1, 125.0, 122.7, 117.7, 42.7; λ_{max} =413 nm; HRMS-ES$^+$*m*/*z*: 237.0325 (Calculated for $C_{10}H_{10}N_2O_3S$-H^+: 237.0328).

Characterization of intermediates 2a-2d and study on the solvent dependent formation of selenol

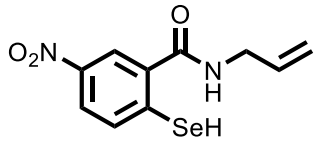


The reaction of probe **2** with PhSH in PBS (10 mM, pH 7.4): acetonitrile (75:25) mixture gave blue shift in UV-Vis spectroscopy with progressively increase in absorbance at λ_{max} = 343 nm upon addition of PhSH from 0 to 1 equivalent. The mechanistic study revealed that, blue shift observed is due to the formation of colorless selenenylsulfide **2a** which was isolated, characterized and found to show absorbance at λ_{max} = 343 nm under identical conditions (Figure 49). Selenenylsulfide **2a** upon further treatment with next equivalent of PhSH (1 to 2 equivalent) showed red shift with linear increase in absorbance at λ_{max} = 426 nm. The excess of PhSH does not lead to increase in absorbance at λ_{max} = 426 nm. The absorbance observed at λ_{max} = 426 nm was thought to be due to formation of corresponding selenol **2b**. Selenol **2b** was confirmed by mass spectral analysis of reaction mixture under identical conditions. It was also isolated and characterized in the form of methyl selenide **2d** by addition of excess of methyl iodide to the bright yellow colored reaction mixture of probe **2** and two equivalents of PhSH in PBS (10 mM, pH 7.4):acetonitrile (3:1) medium. The change in solvent from PBS (10 mM, pH 7.4): acetonitrile (75:25) to MeOH or MeCN could

offered only colorless **2a** and formation of selenol **2b** was not observed even with excess of PhSH (Figure 48). It confirms that, here generation of selenol **2b** is feasible only in the presence of aqueous medium. In order to evaluate the reversibility of probe **2**, we added an oxidising agent (*t*-BuOOH) to the selenol **2b** and waited for a minute. It resulted into colorless reaction mixture which upon recording UV spectrum turned out to be selenenyl sulphide **2a** (343 nm). To this, when PhSH (2 equivalent) solution was added, it again showed red shift to 426 nm with appearance of yellow color. This reversibility between the yellow colored species (λ = 426 nm) and colorless species (λ = 343 nm) is determined for more than ten cycles by successive addition of reductant (PhSH) and oxidant (*t*-BuOOH). Intermediate that forms by the reaction between selenol (**2b**) and *t*-BuOOH is characterized by mass spectrometry and turned out to be selenenic acid (**2c**).

N-allyl-5-nitro-2-(phenylseleninothioyl)benzamide (2a): In a 10 mL capacity round bottom flask isoselenazolone **1** (75 mg, 0.26 mmol) was charged followed by addition of 5 mL PBS buffer (10mM, pH 7.4): acetonitrile (75: 25) mixture. To this, 2.5 mL PhSH (30 mg, 0.26 mmol) solution from the stock solution (300 mg PhSH in 25 mL acetonitrile) was added in a lot. Reaction mixture was stirred at room temperature for 1 h. Progress of reaction was monitored by TLC. Then, reaction mixture was extracted with ethyl acetate (10 mL x 2). Combined ethyl acetate layer was dried over Na_2SO_4 and solvent was evaporated under reduced pressure at 40 ^oC. Crude product was given multiple washings with hexane to obtain pale yellow powder. Yield 0.086 g (84%), mp 112-114 °C. ^1H NMR (400 MHz, $CDCl_3$) δ 8.52-8.35 (m, 2H), 8.21 (dd, *J* = 2.2, 8.9 Hz, 1H), 7.45 (d, *J* = 7.8 Hz, 2H), 7.22 (t, *J* = 7.8 Hz, 1H), 7.19-7.13 (m, 1H), 6.75 (bs, 1H), 6.04-5.84 (m, 1H), 5.41-5.13 (m, 2H), 4.13 (t, *J* =

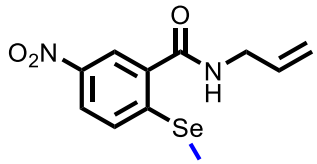
5.8 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 166.1, 147.8, 146.1, 135.6, 133.0, 131.1, 129.8, 129.3, 129.1, 127.1, 125.6, 121.3, 118.0, 43.0; ^{77}Se NMR ($CDCl_3$): 629.7 ppm. λ_{max} =343 nm; HRMS-ES$^+$ *m/z*: 416.9781 (calculated for $C_{16}H_{14}N_2O_3S^{80}Se$ + Na: 416.9783).



N-allyl-2-hydroseleno-5-nitrobenzamide (2b): A freshly prepared (1:2) mixture of isoselenazolone **2** and PhSH in distilled water: acetonitrile (75: 25) mixture was analysed mass spectrometrically and a peak was observed at *m/z*: 284.9679 (calculated for $C_{10}H_{10}N_2O_3Se$ - H^+) attributed to selenol **2b** (See Figure S31).

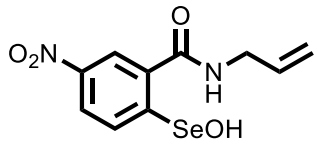
Existence of selenol **2b** in aqueous medium was further validated by a separate experiment involving its in situ trapping by methyl iodide.

^{1}H, ^{13}C, ^{77}Se NMR and mass spectrometric study of methyl selenide (2d) obtained by reaction of 2, two equivalent of PhSH and excess of methyl iodide:



N-allyl-2-(methylselanyl)-5-nitrobenzamide (2d): Isoselenazolone **2** (50 mg, 0.2 mmol) and solution of PhSH (3.3 mL, 0.4 mmol) were mixed thoroughly in PBS buffer (10mM, pH 7.4): acetonitrile (75: 25) mixture to get bright orange colored solution. To this, excess of methyl iodide (85 mg, 0.6 mmol) was added in a lot. Resulted pale yellow reaction mixture was stirred at room temperature for 30 minutes. Then, reaction mixture was extracted with ethyl acetate (10 mL x 2). Combined ethyl acetate was dried over Na_2SO_4 followed by evaporation of solvent under reduced pressure at 40 ^{o}C. Obtained crude product was washed

with hexane to obtain yellow solid. Yield 0.038 g (68%), mp 176-178 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.3 (d, *J* = 2.3 Hz, 2H), 8.16 (dd, *J* = 8.8, 2.3 Hz, 1H), 7.5 (d, *J* = 8.8 Hz, 1H), 6.23 (bs, 1H), 6.0-5.88 (m, 1H), 5.34-5.20 (m, 2H), 4.1 (t, *J* = 5.8 Hz, 2H), 2.33 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 166.3, 146.5, 144.9, 134.4, 133.3, 128.6, 125.0, 121.8, 117.6, 42.7, 7.4; ^{77}Se NMR ($CDCl_3$): 279.2 ppm. HRMS-ES^{+}*m*/*z*: 301.0085 (Calculated for $C_{11}H_{12}N_2O_3{}^{80}Se + H^+$: 301.0086).



N-allyl-2-(hydroxyselanyl)-5-nitrobenzamide (2c): A freshly prepared (1:2:1) mixture of **2**, PhSH and *tert*-butyl hydroperoxides (TBHP) in distilled water: acetonitrile (75: 25) mixture was analysed mass spectrometrically and peaks were observed at *m*/*z*: 300.9788 (Calculated for $C_{10}H_{10}N_2O_4Se- H^+$) attributed to selenenic acid **2c**. (See Figure S36)

Figure S1 ^{1}H NMR of **1**

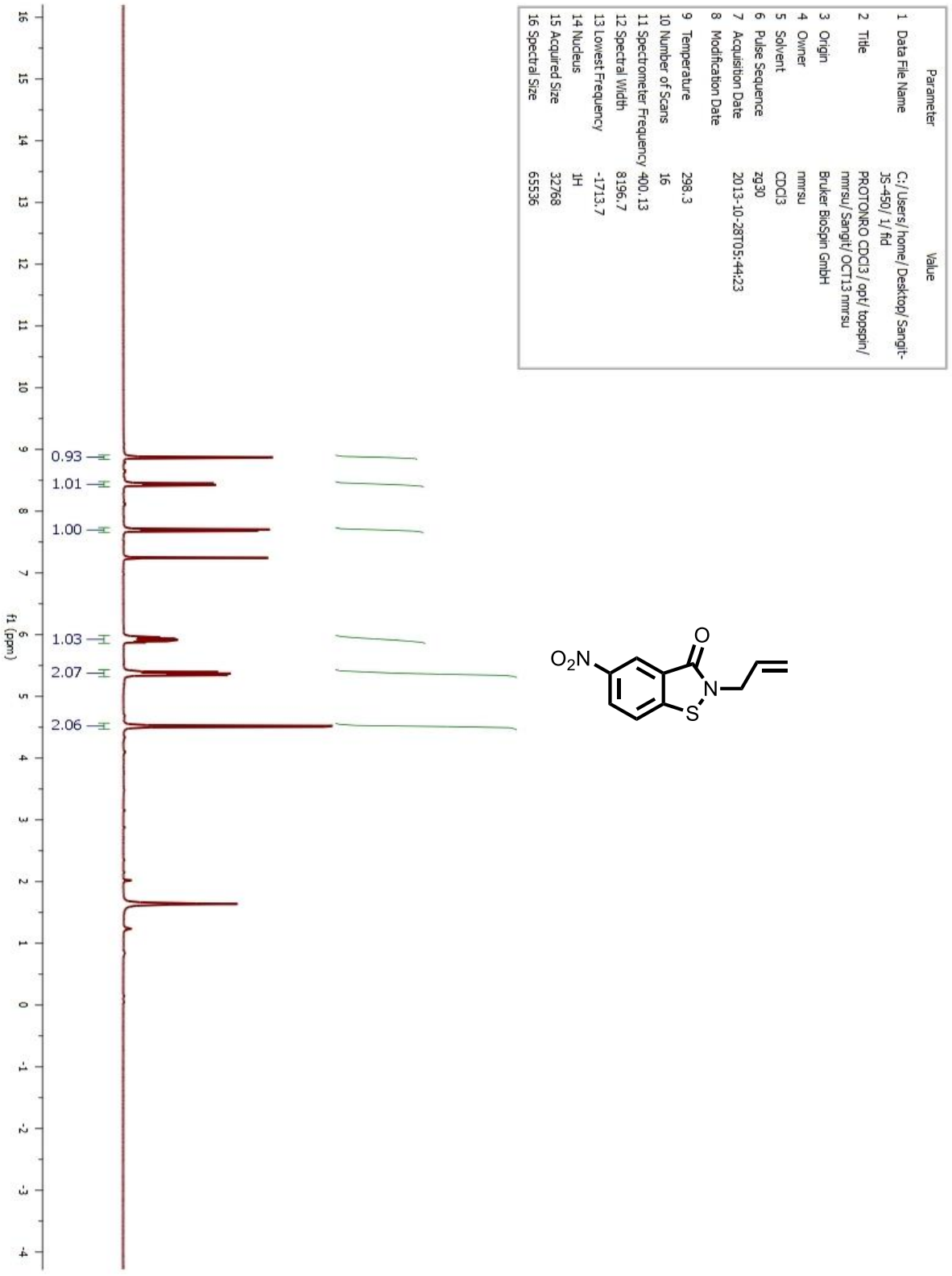


Figure S2 ^{13}C NMR of **1**

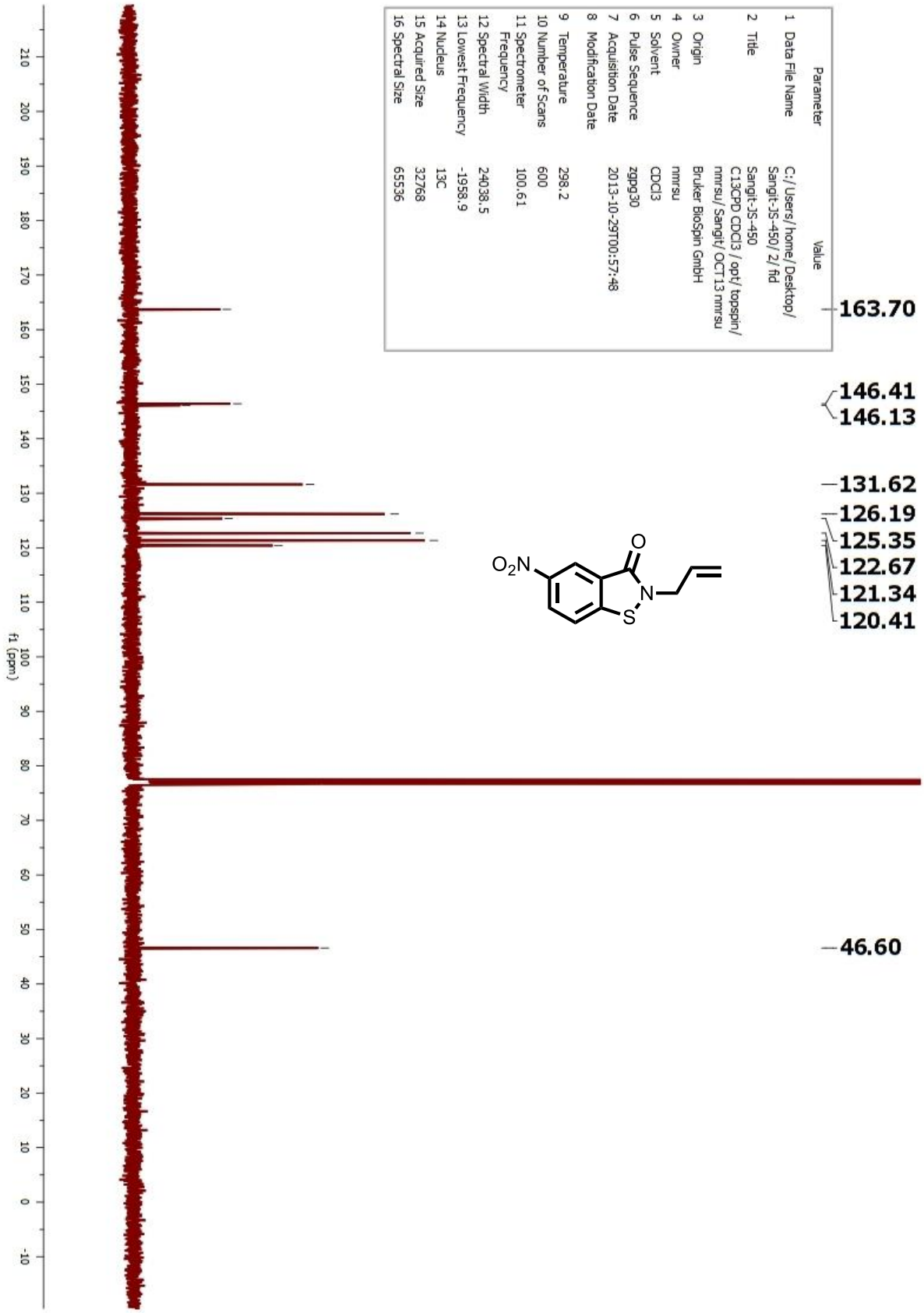


Figure S3 Mass spectra of **1**

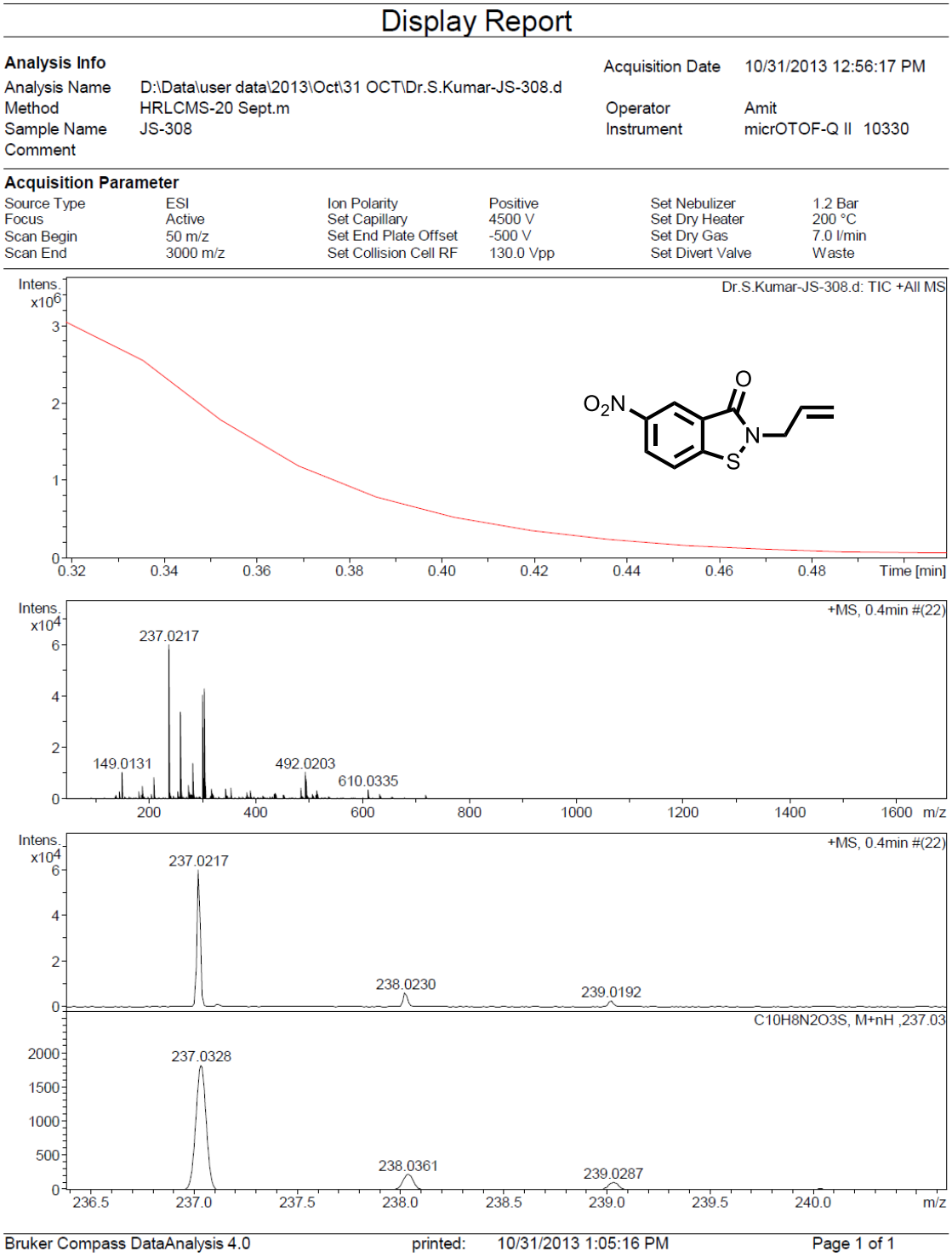


Figure S4 ^{1}H NMR of **2**

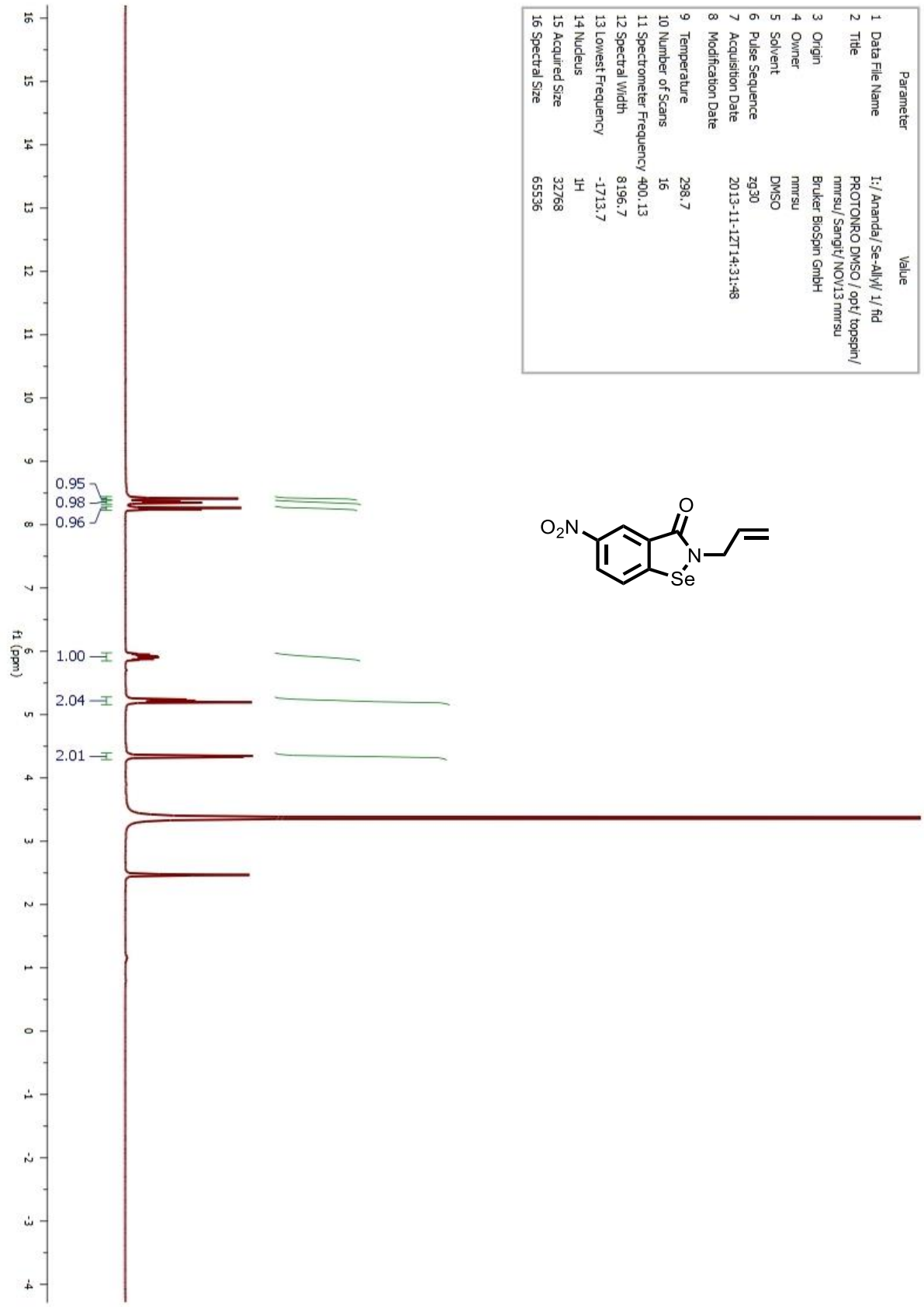


Figure S5 ^{13}C NMR of **2**

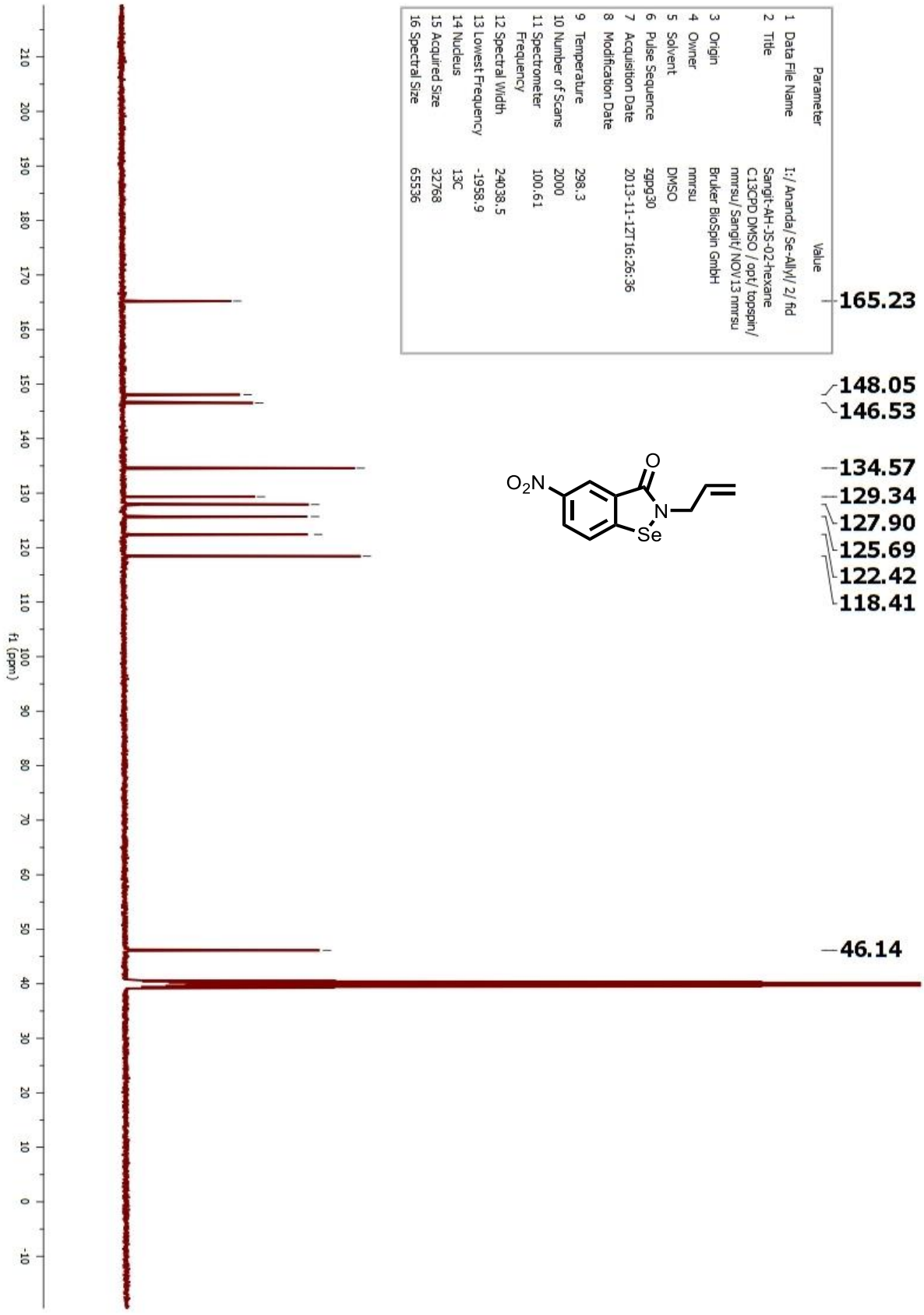
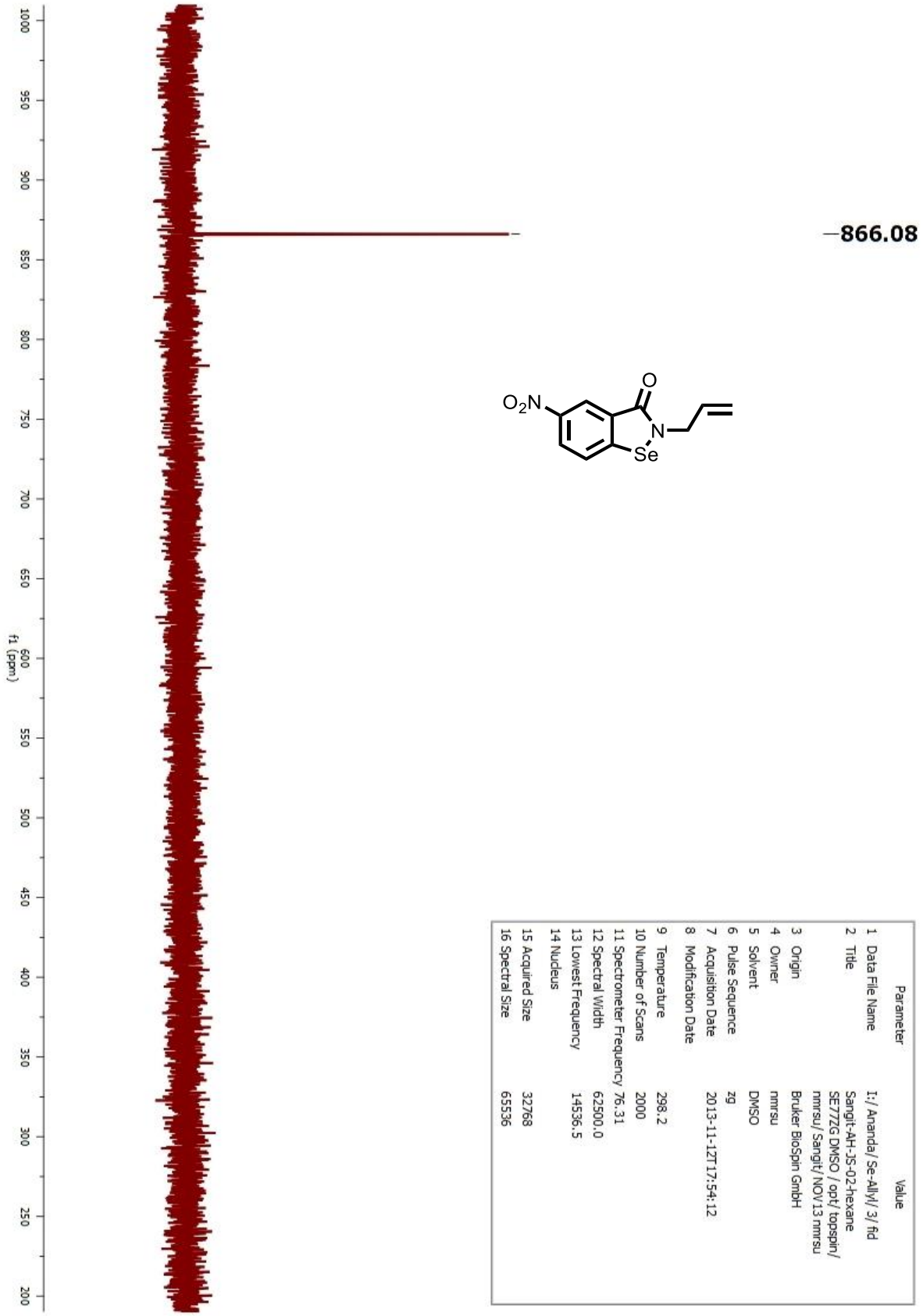


Figure S6 ^{77}Se NMR of **2**



	Parameter	Value
1	Data File Name	I:/ Ananda/ Se-Allyl/ 3/ fid
2	Title	Sangit-AH-JS-02-hexane SE77ZG DMSO / opt/ topspin/ nmrsu/ Sangit/ NOV13 nmrsu
3	Origin	Bruker BioSpin GmbH
4	Owner	nmrsu
5	Solvent	DMSO
6	Pulse Sequence	zg
7	Acquisition Date	2013-11-12T17:54:12
8	Modification Date	
9	Temperature	298.2
10	Number of Scans	2000
11	Spectrometer Frequency	76.31
12	Spectral Width	62500.0
13	Lowest Frequency	14536.5
14	Nucleus	
15	Acquired Size	32768
16	Spectral Size	65536

Figure S7 Mass spectra of **2**

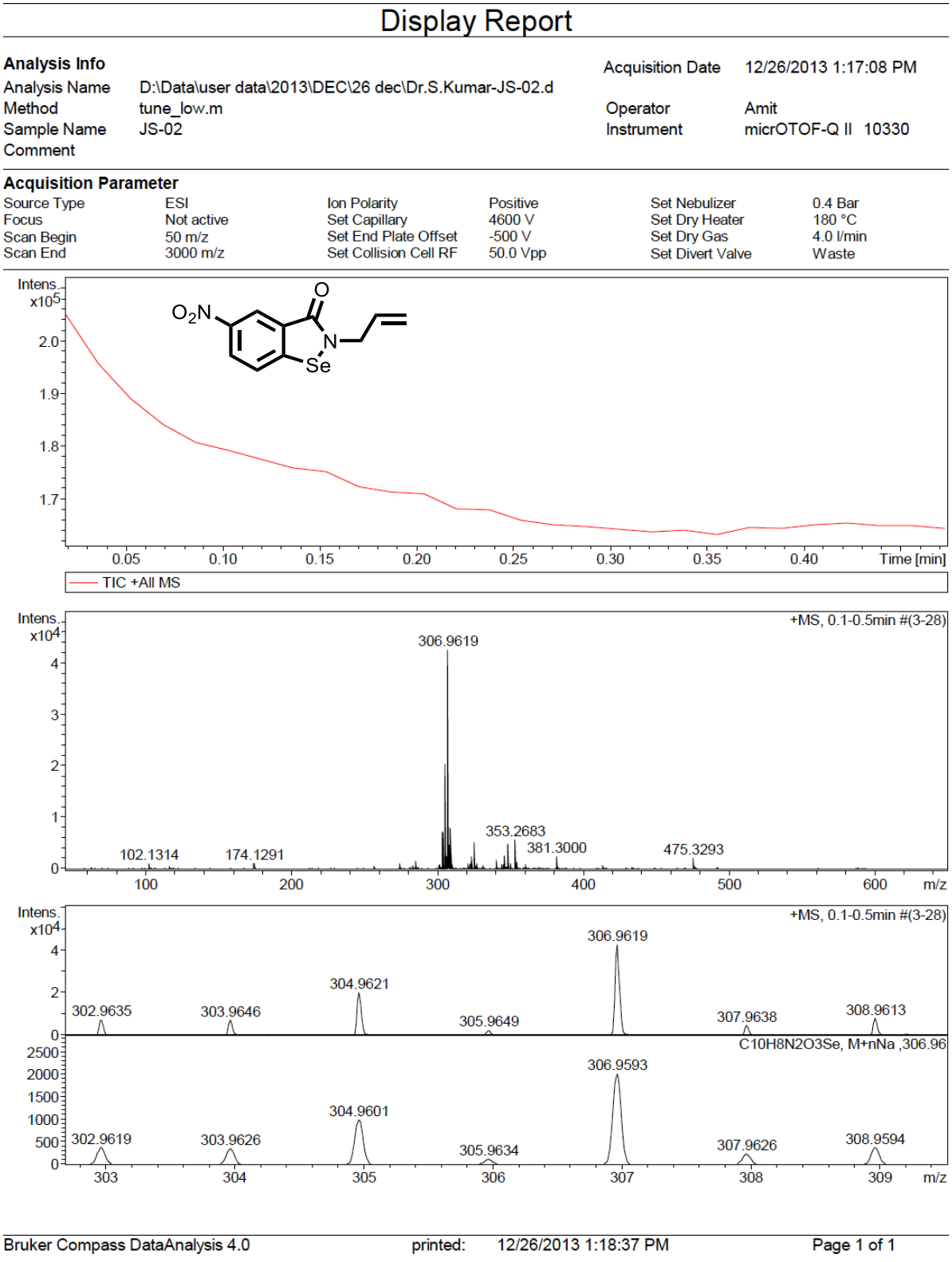


Figure S8 ^{1}H NMR of **3**

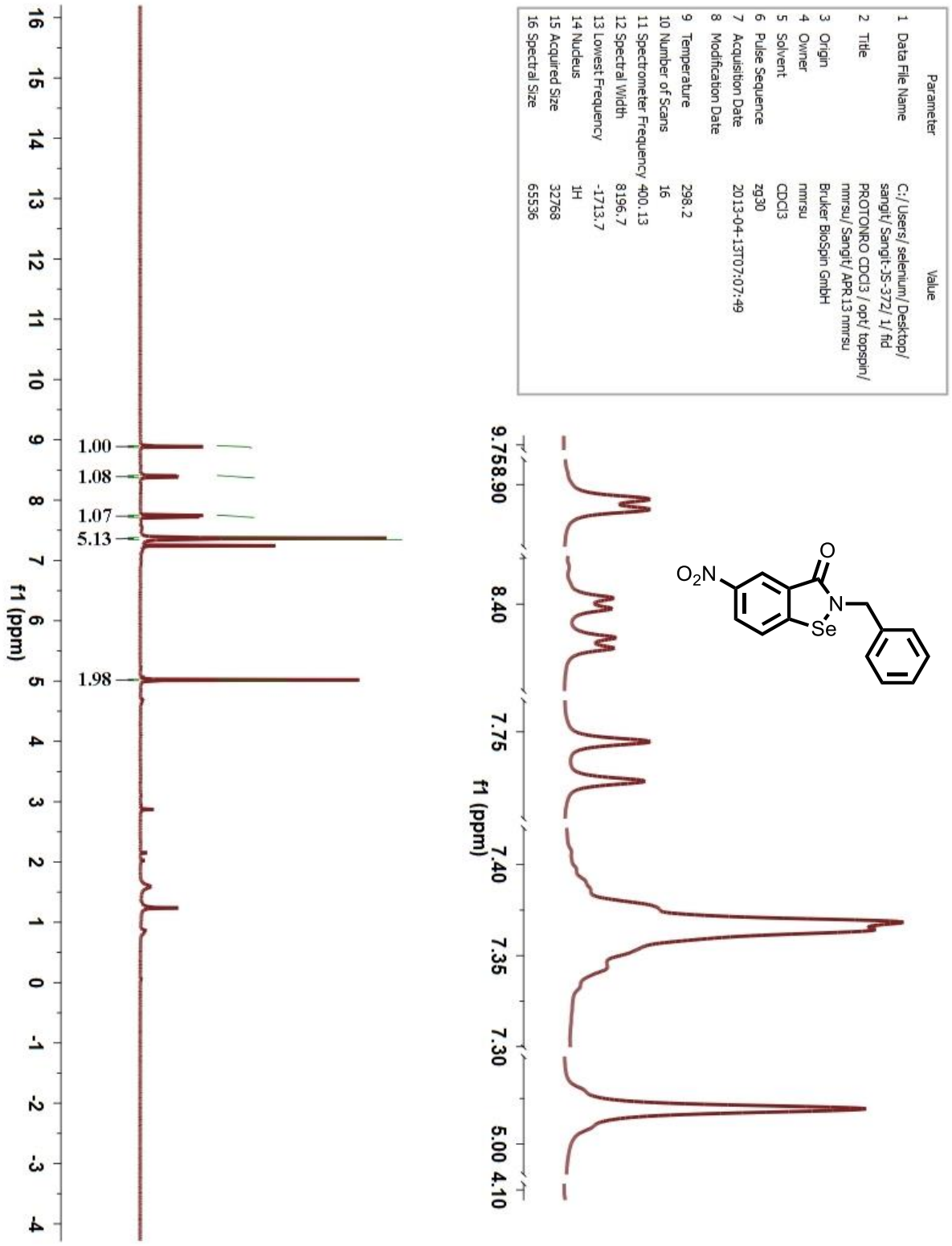


Figure S9 ^{13}C NMR of **3**

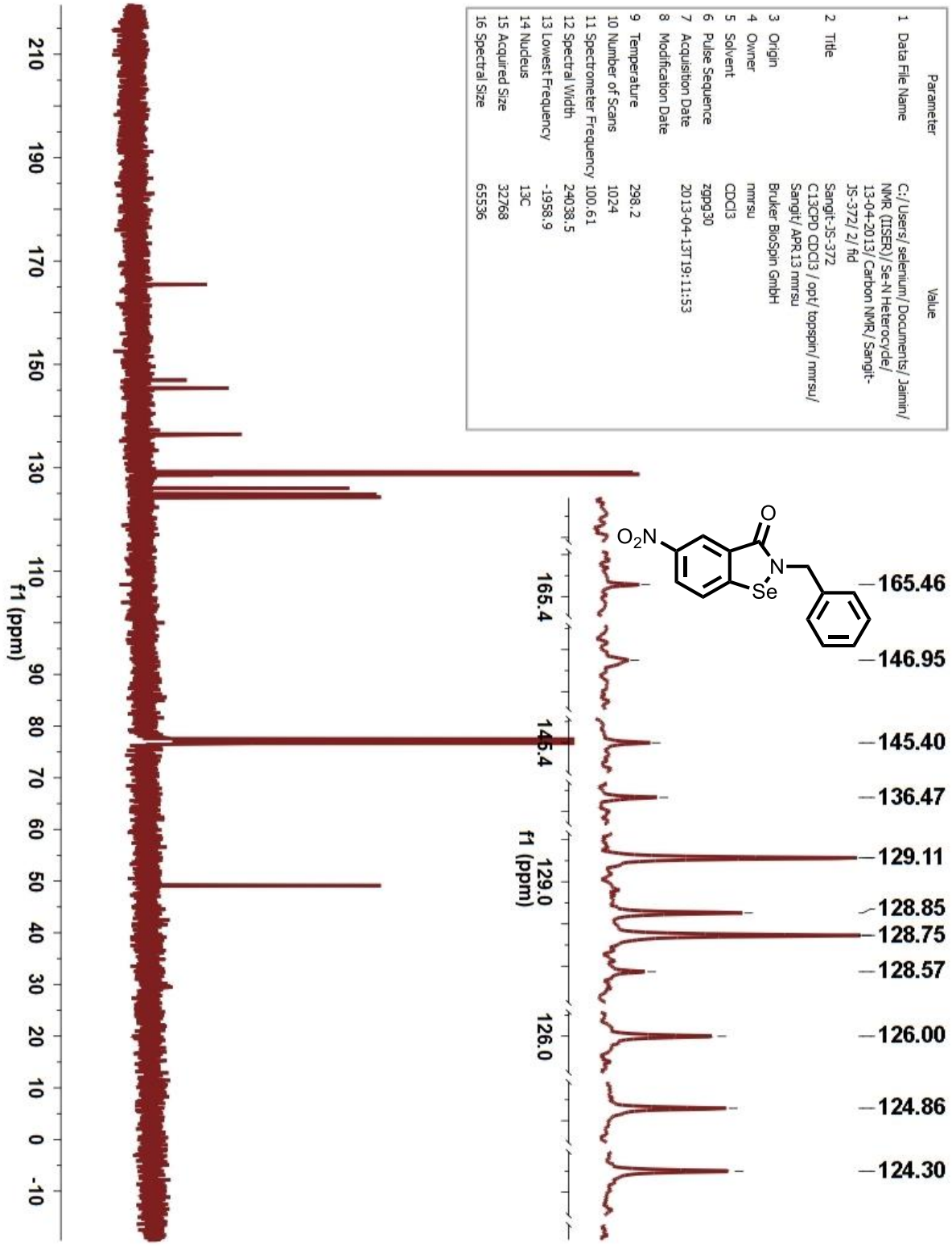


Figure S10 ^{77}Se NMR of **3**

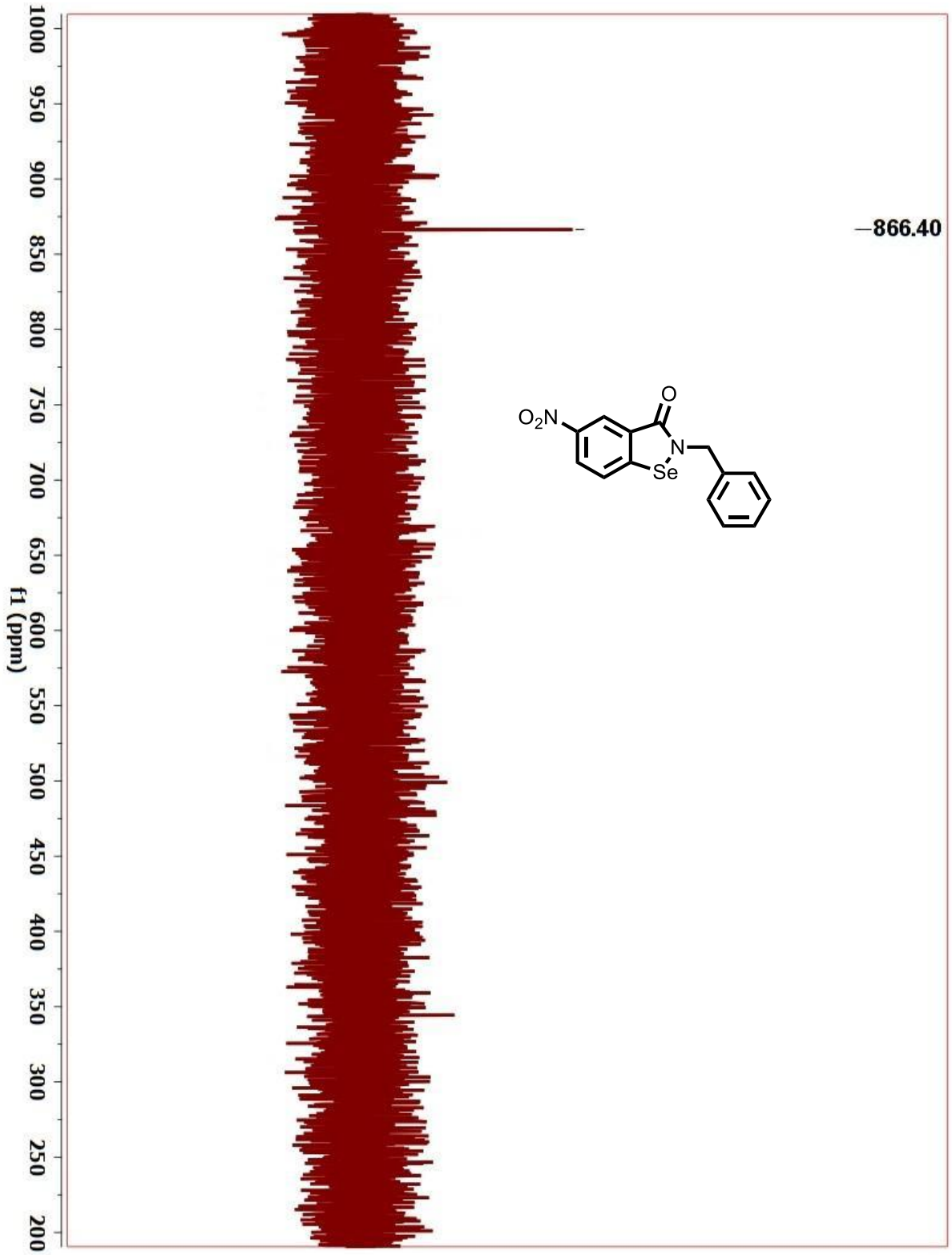


Figure S11 HRMS of **3**

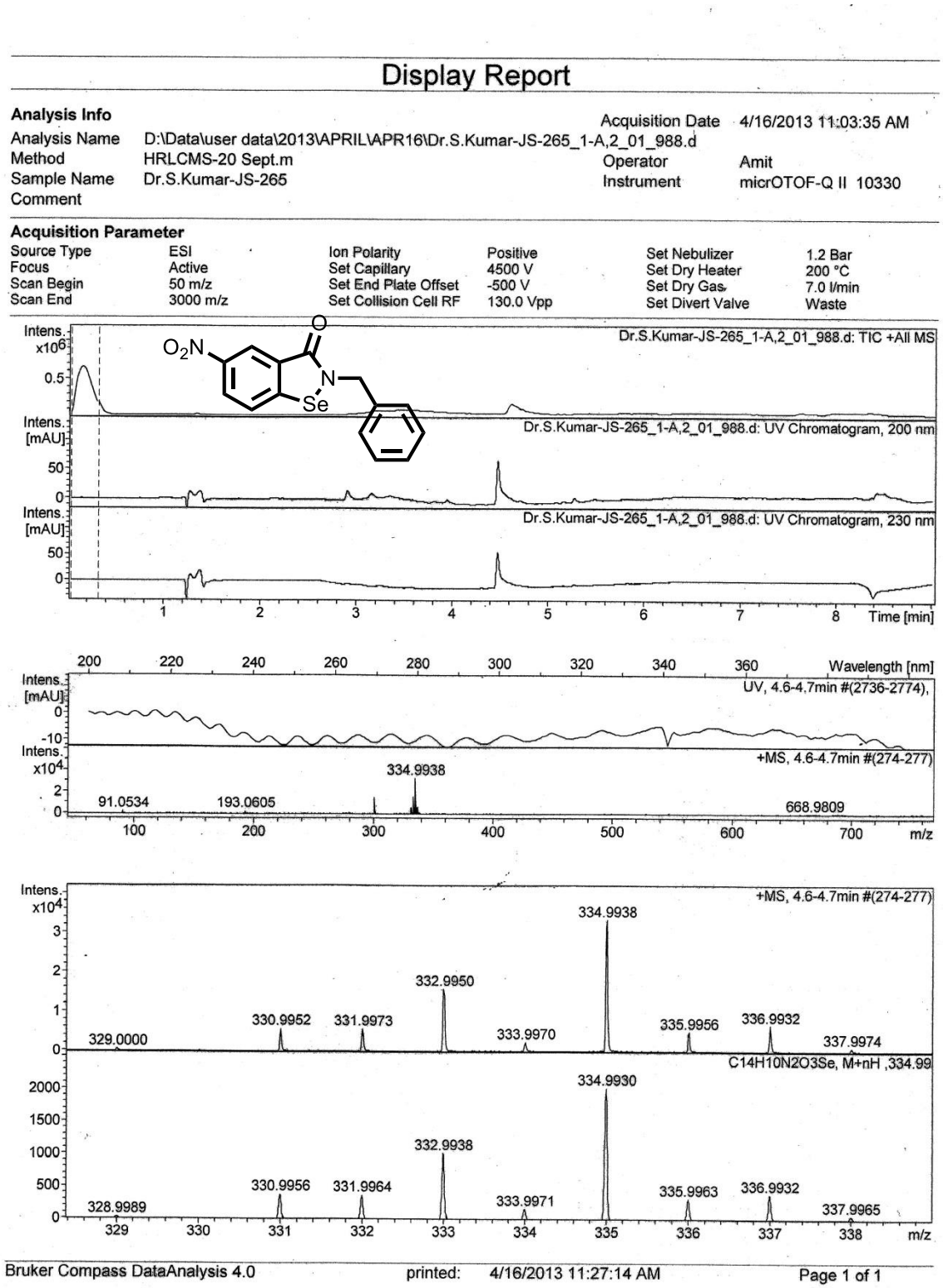


Figure S12 ^{1}H NMR of **4**

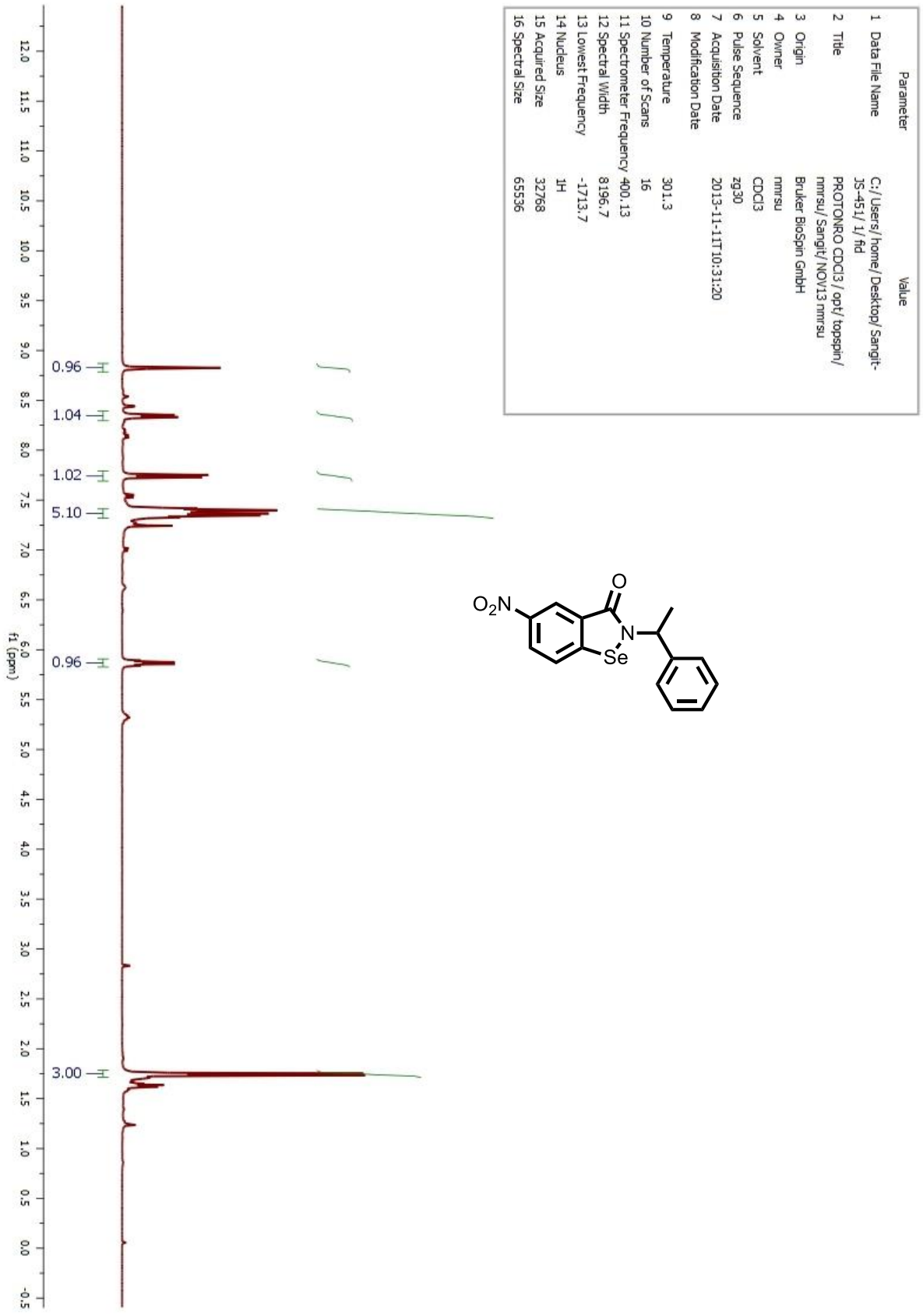


Figure S13 ^{13}C NMR of **4**

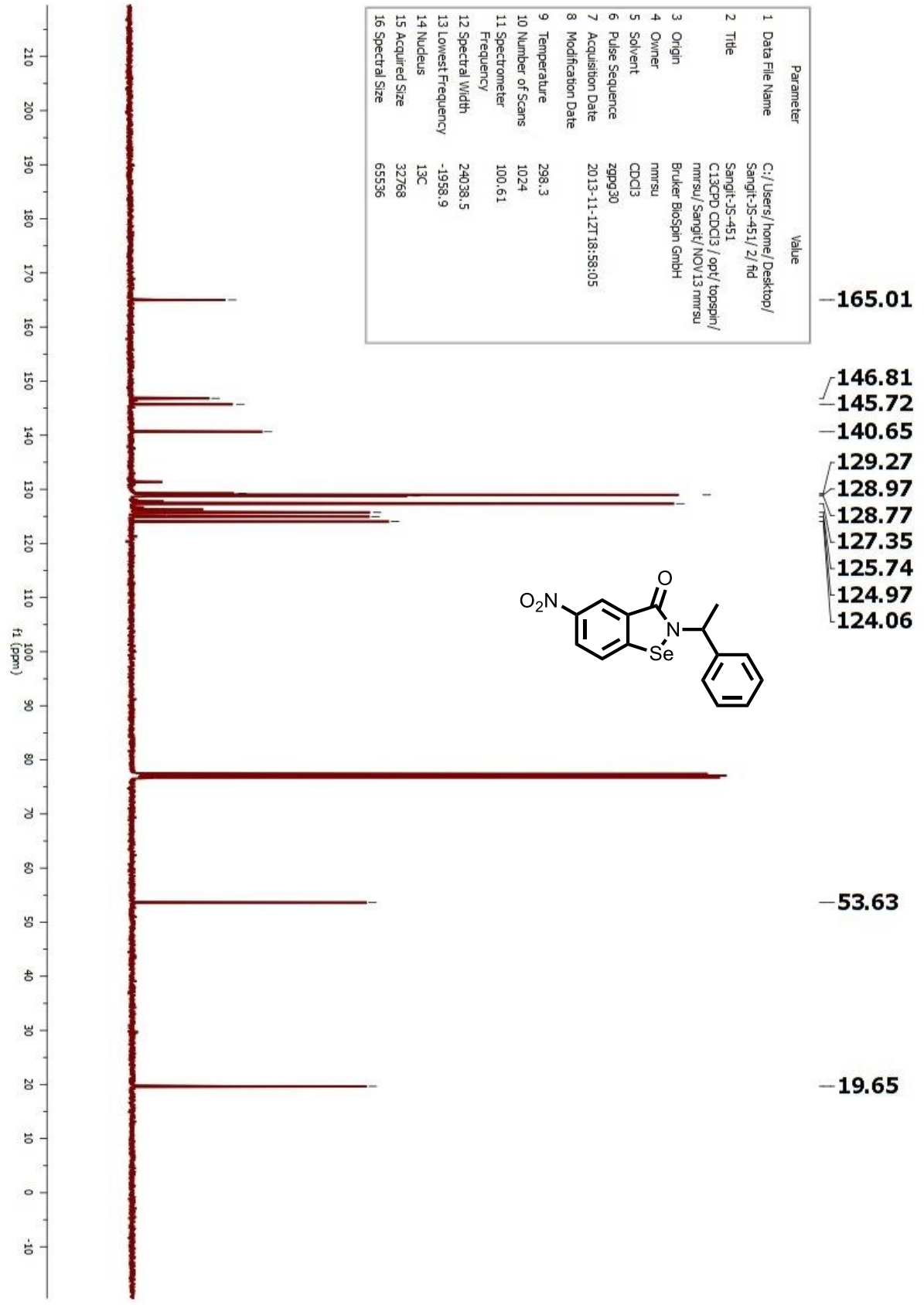


Figure S14 ^{77}Se NMR of **4**

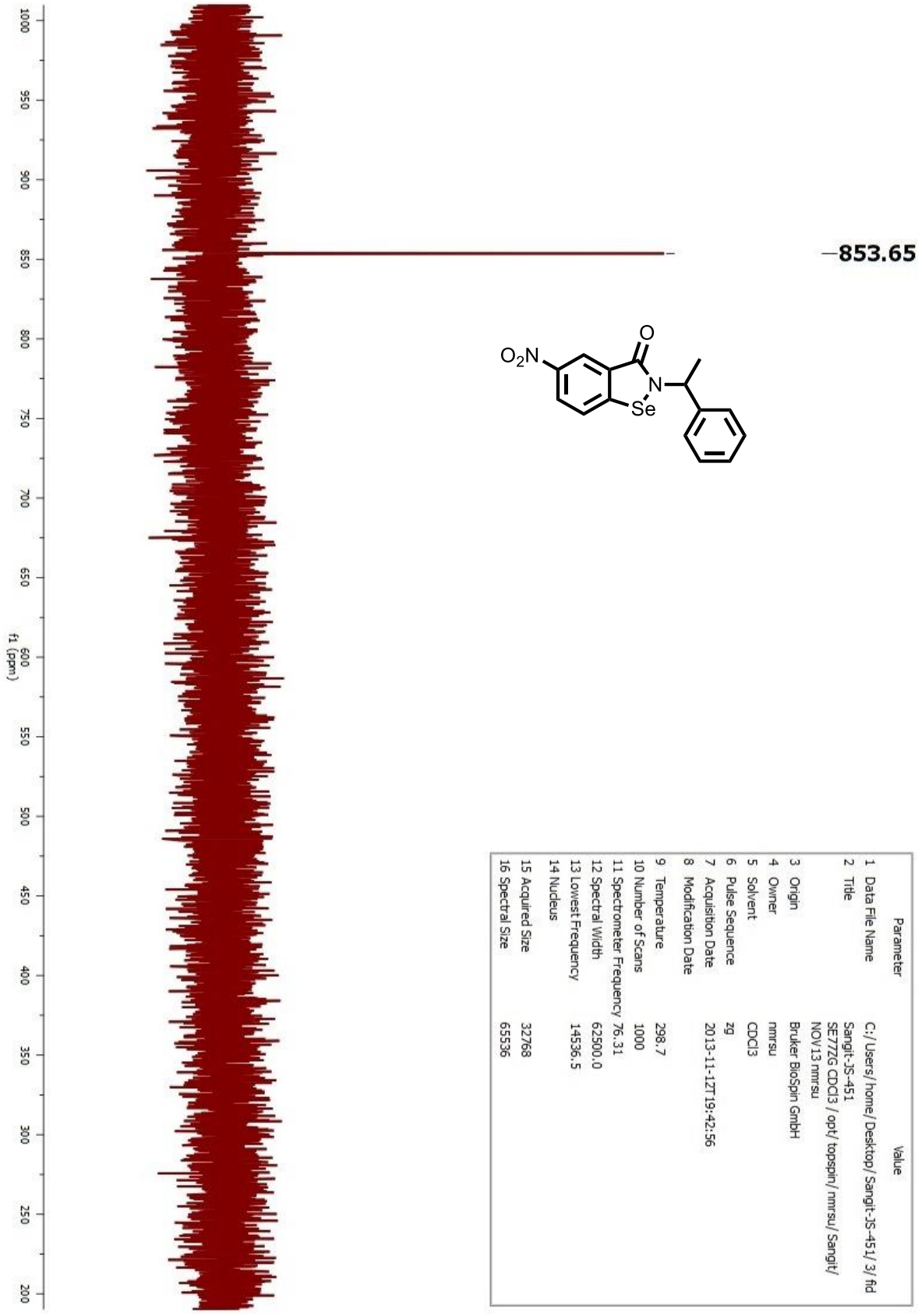


Figure S15 Mass spectra for **4**

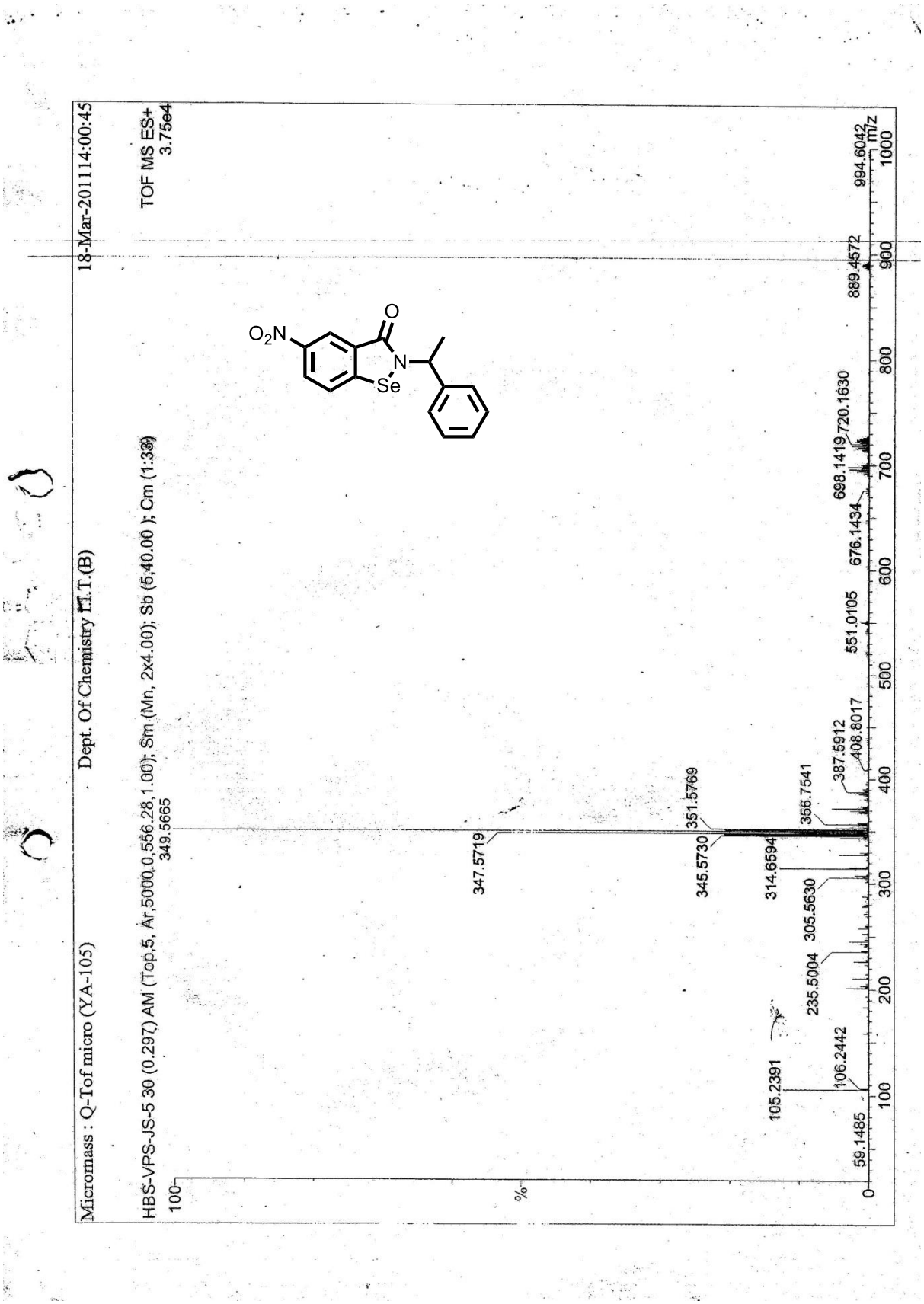


Figure S16 ^{1}H NMR of precursor for **5**

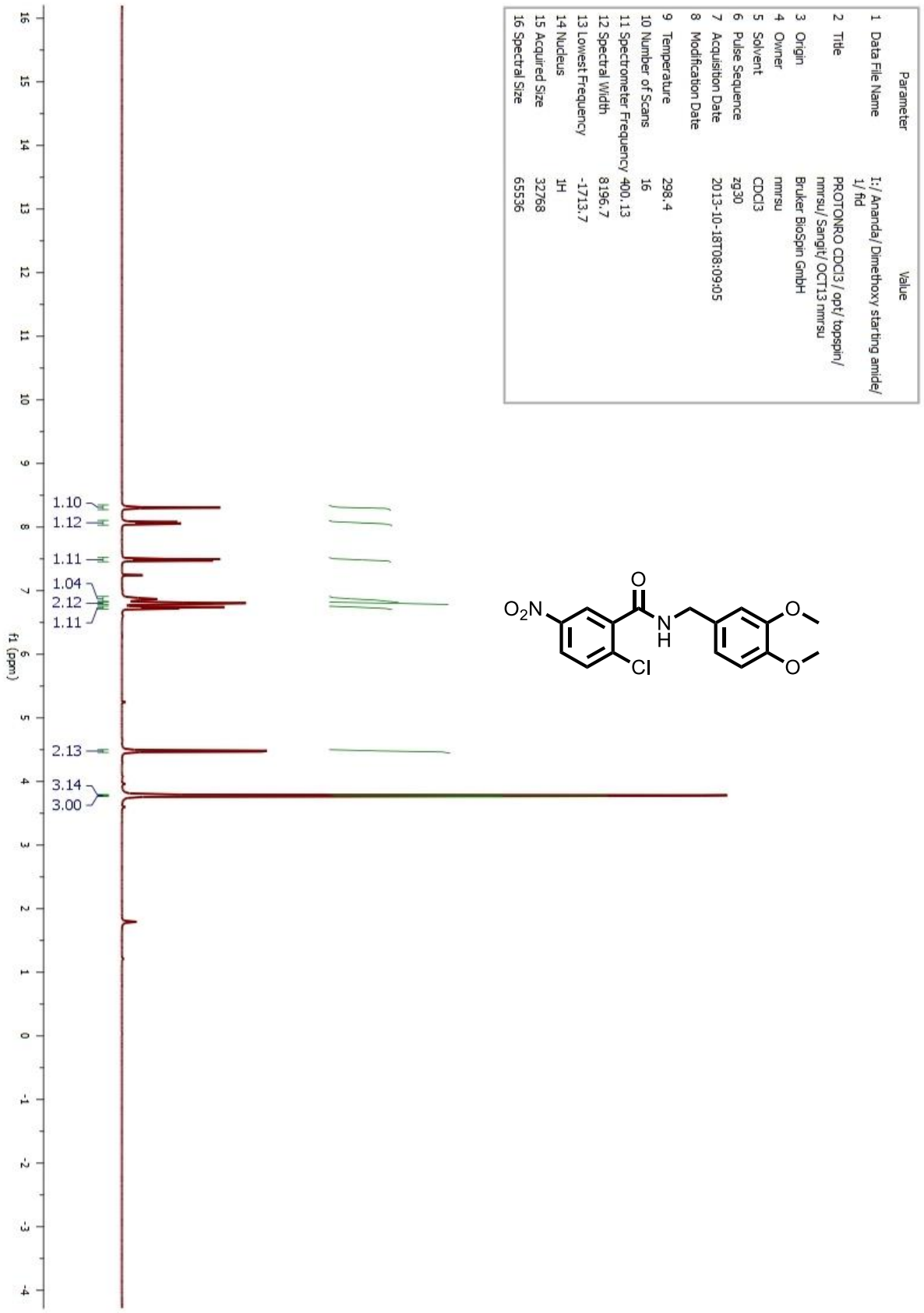


Figure S17 ^{13}C NMR of precursor for **5**

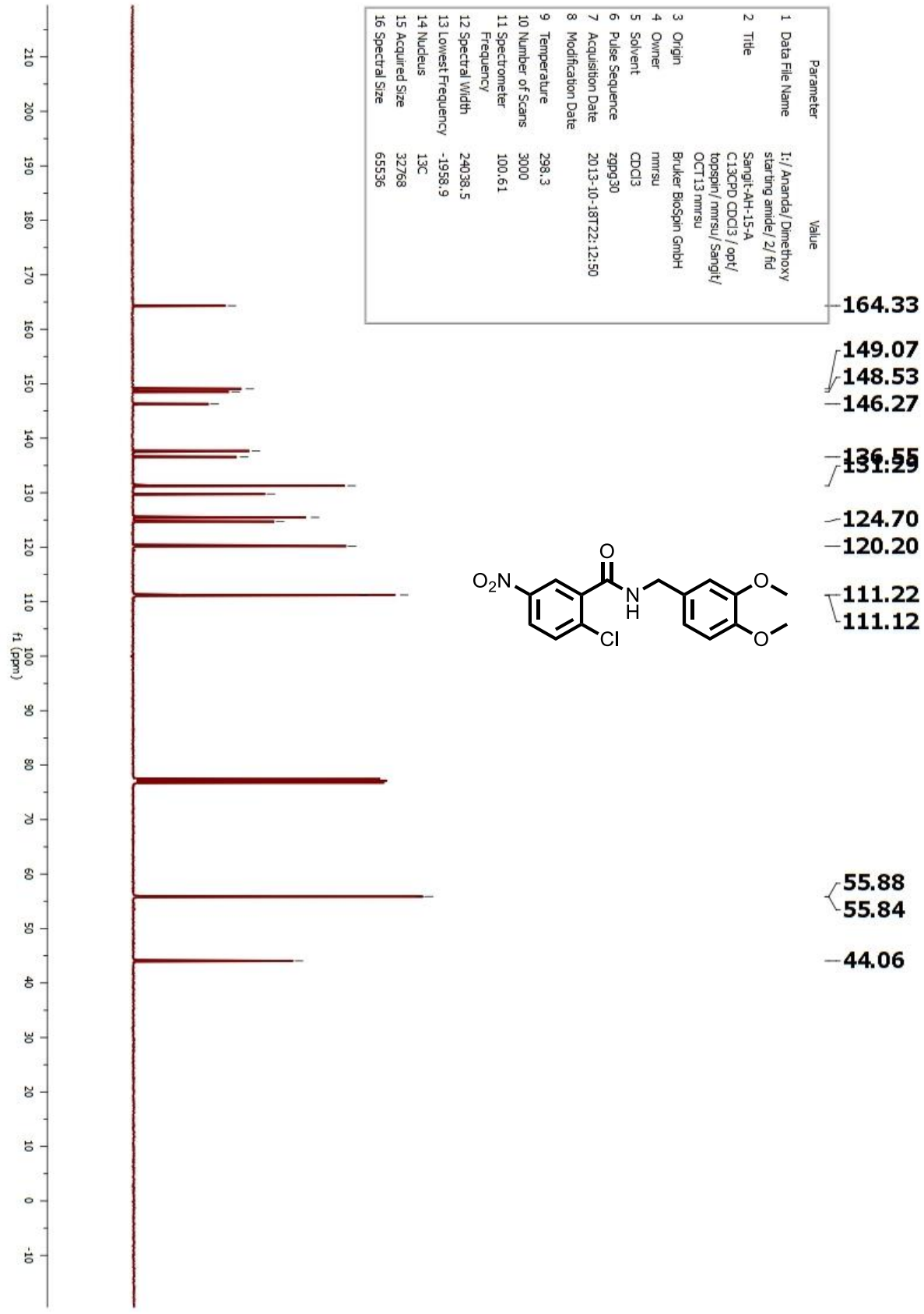


Figure S18 HRMS of precursor for **5**

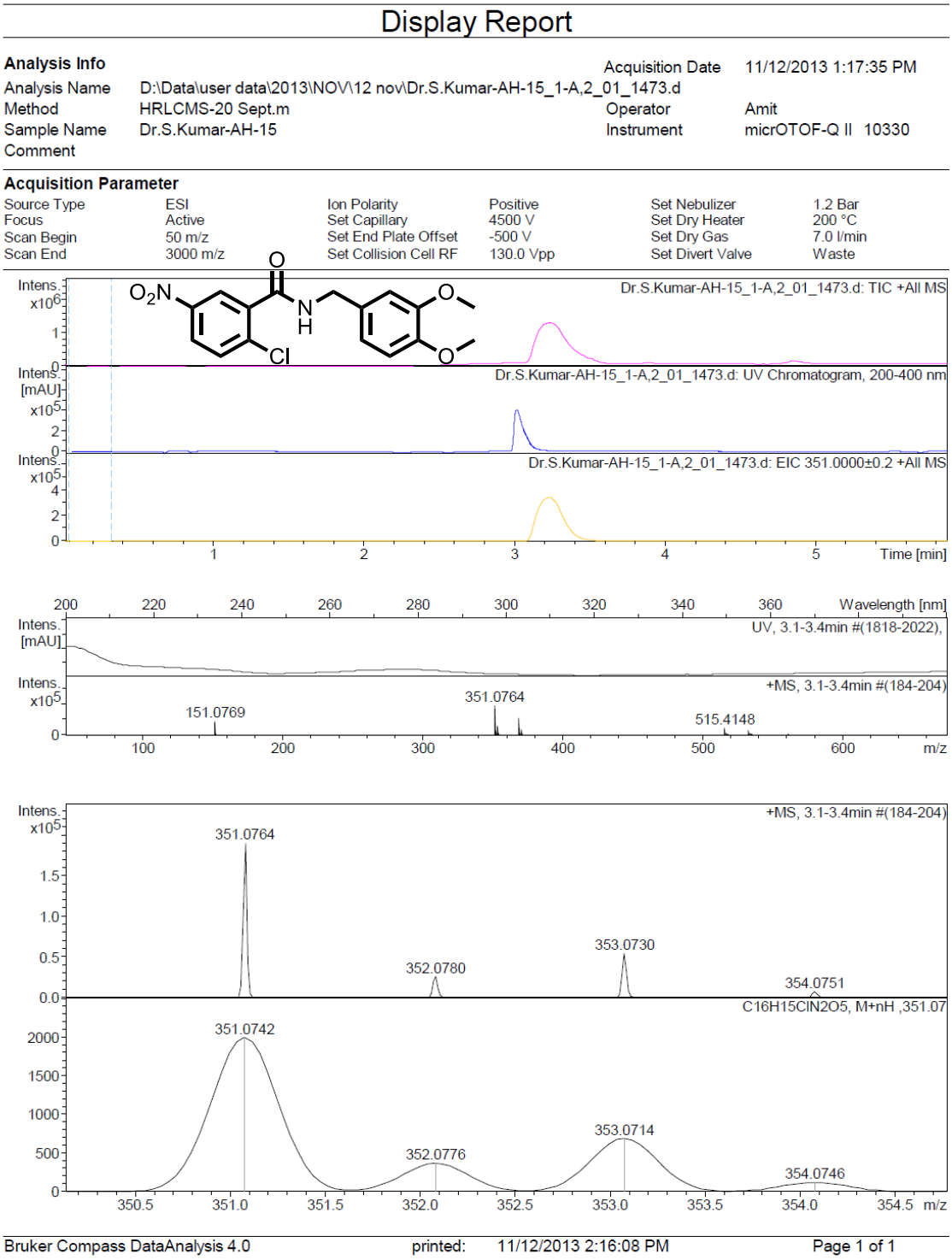


Figure S19 ^{1}H NMR of **5**

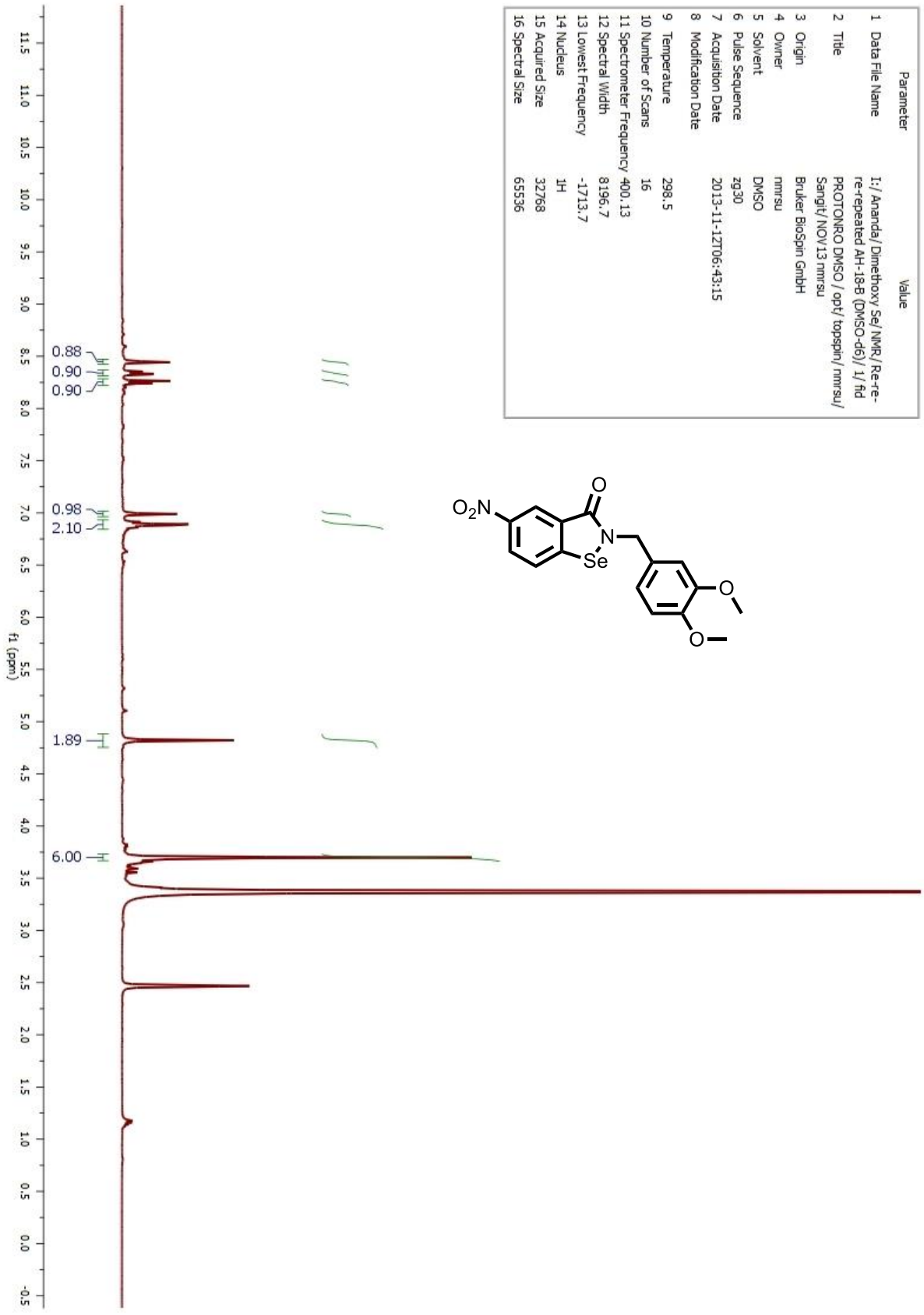


Figure S20 ^{13}C NMR of **5**

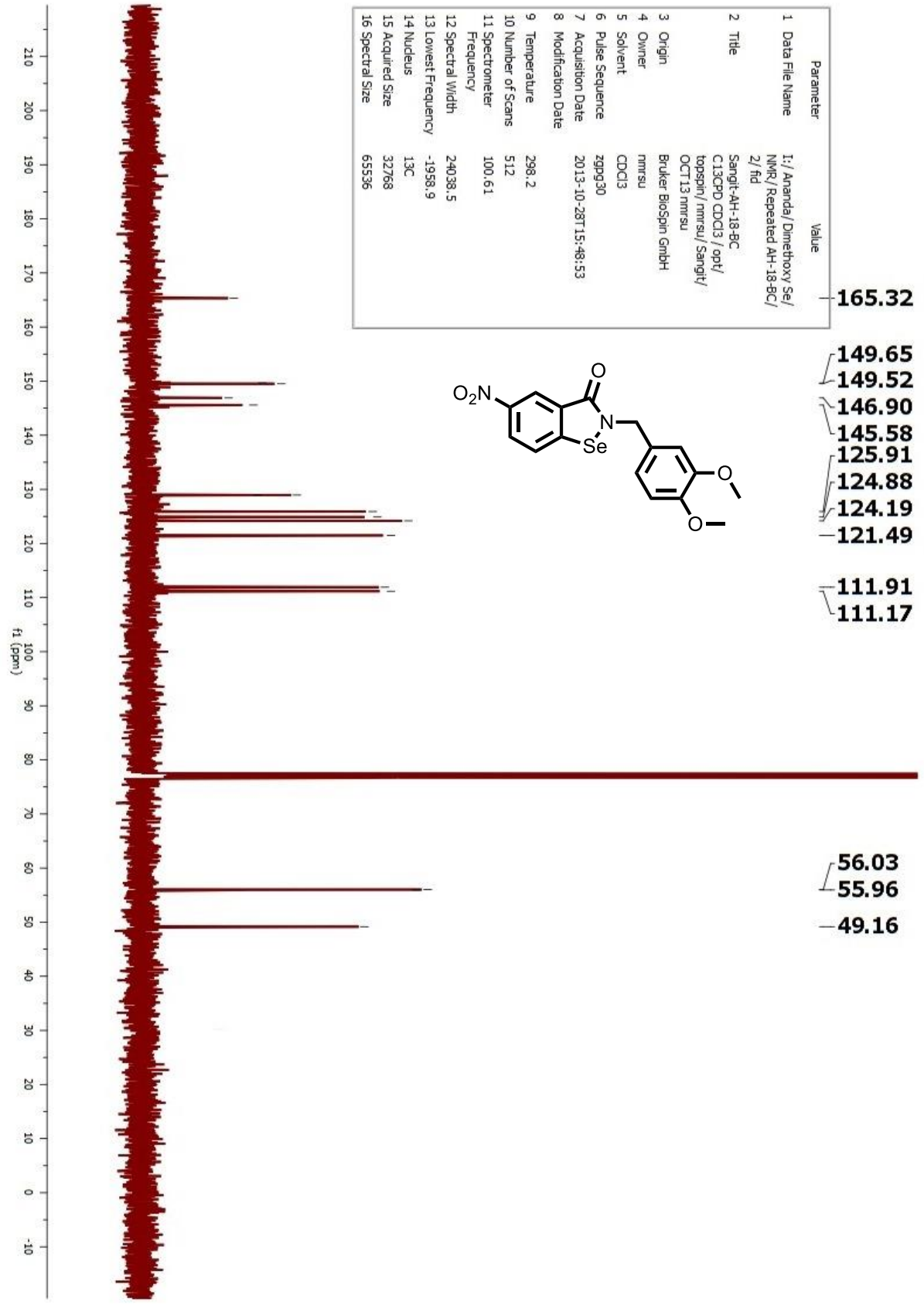
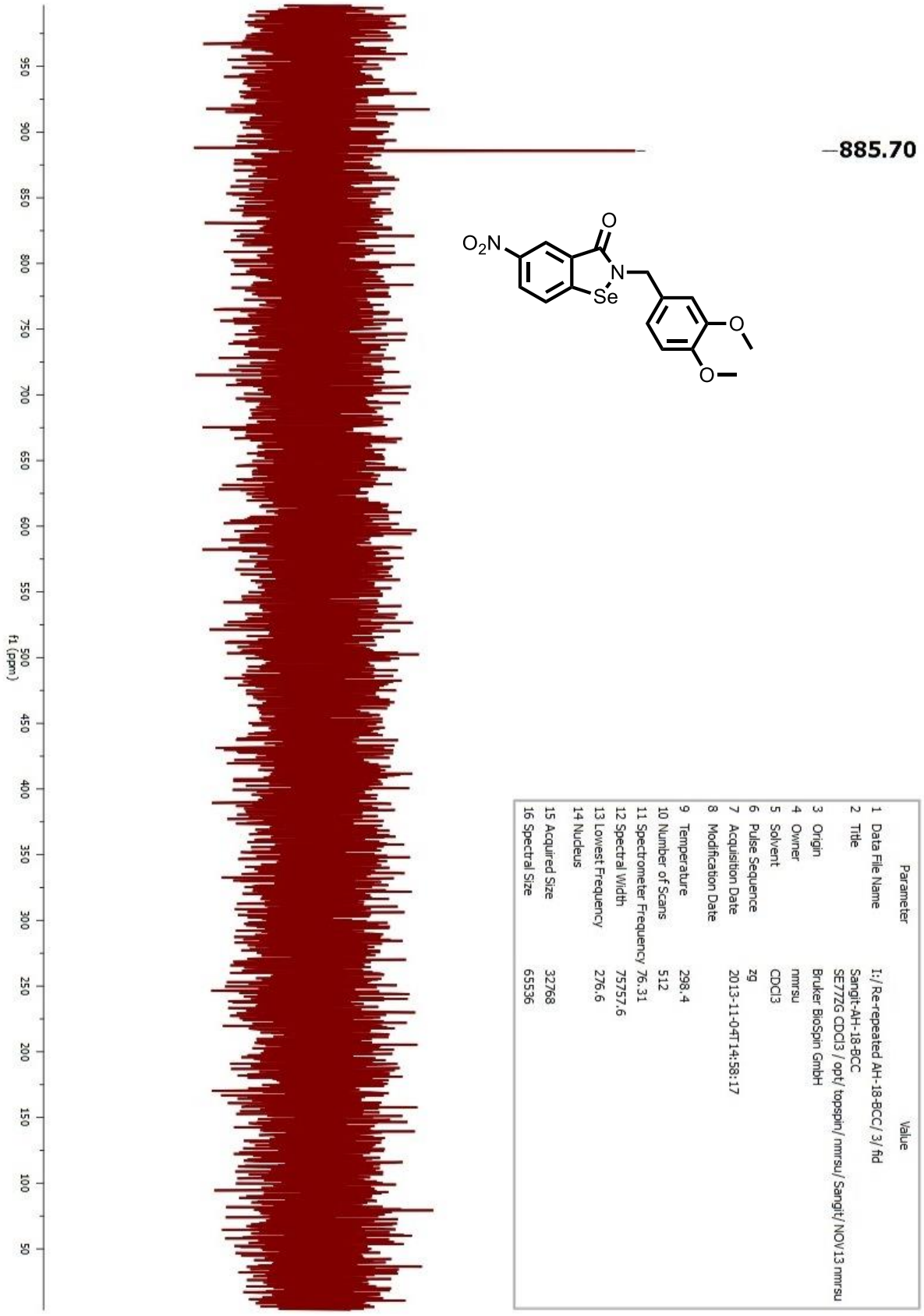


Figure S21 ^{77}Se NMR of **5**



Parameter	Value
1 Data File Name	I:/ Re-repeated AH-18-BCC/ 3/ fid
2 Title	Sangit-AH-18-BCC SE77ZG CDCl3 / opt/ topspin/ nmrsu/ Sangit/ NOV13 nmrsu
3 Origin	Bruker BioSpin GmbH
4 Owner	nmrsu
5 Solvent	CDCl3
6 Pulse Sequence	zg
7 Acquisition Date	2013-11-04T14:58:17
8 Modification Date	
9 Temperature	298.4
10 Number of Scans	512
11 Spectrometer Frequency	76.31
12 Spectral Width	75757.6
13 Lowest Frequency	276.6
14 Nucleus	
15 Acquired Size	32768
16 Spectral Size	65536

Figure S22 HRMS of **5**

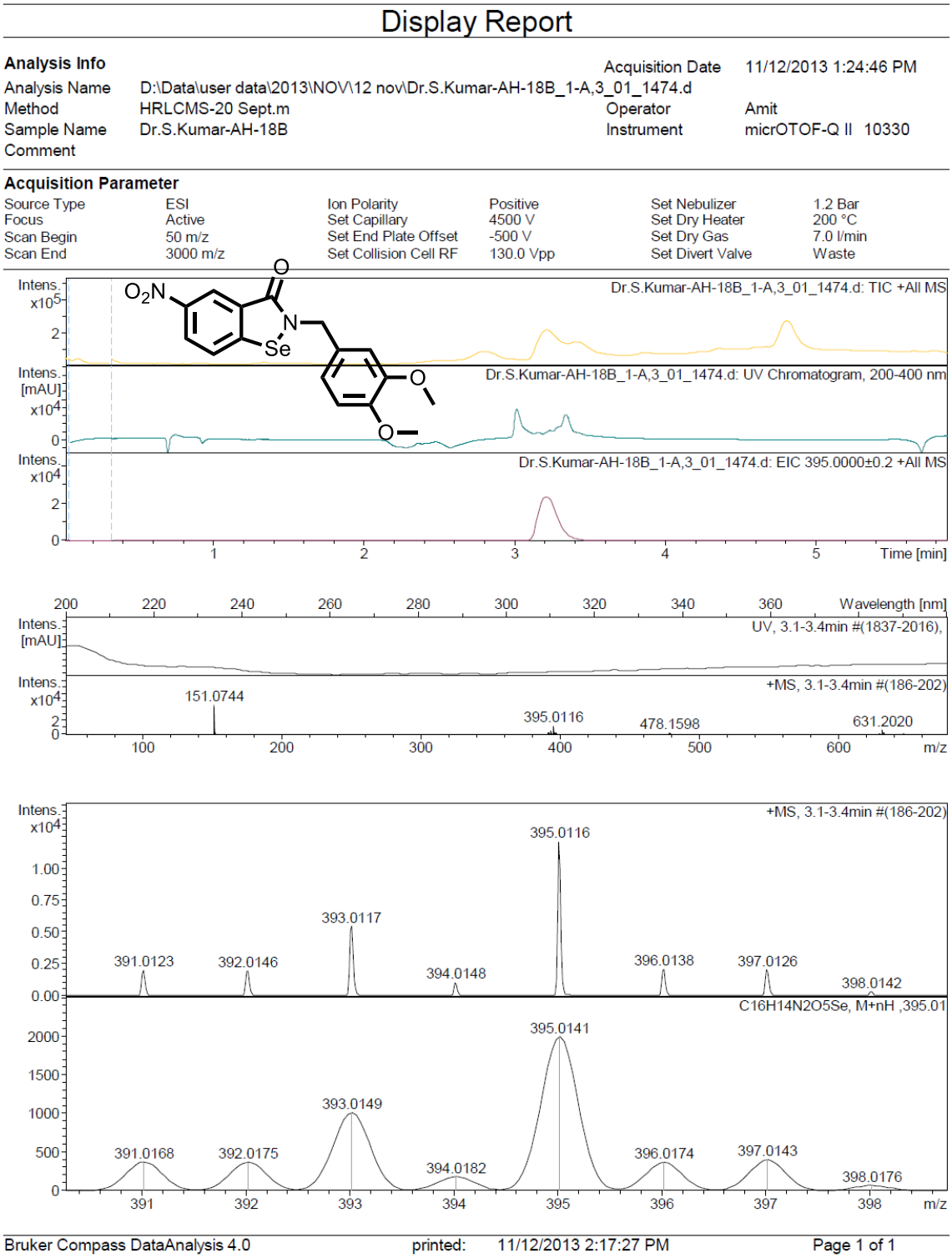


Figure S23 Mass spectra of **1a**

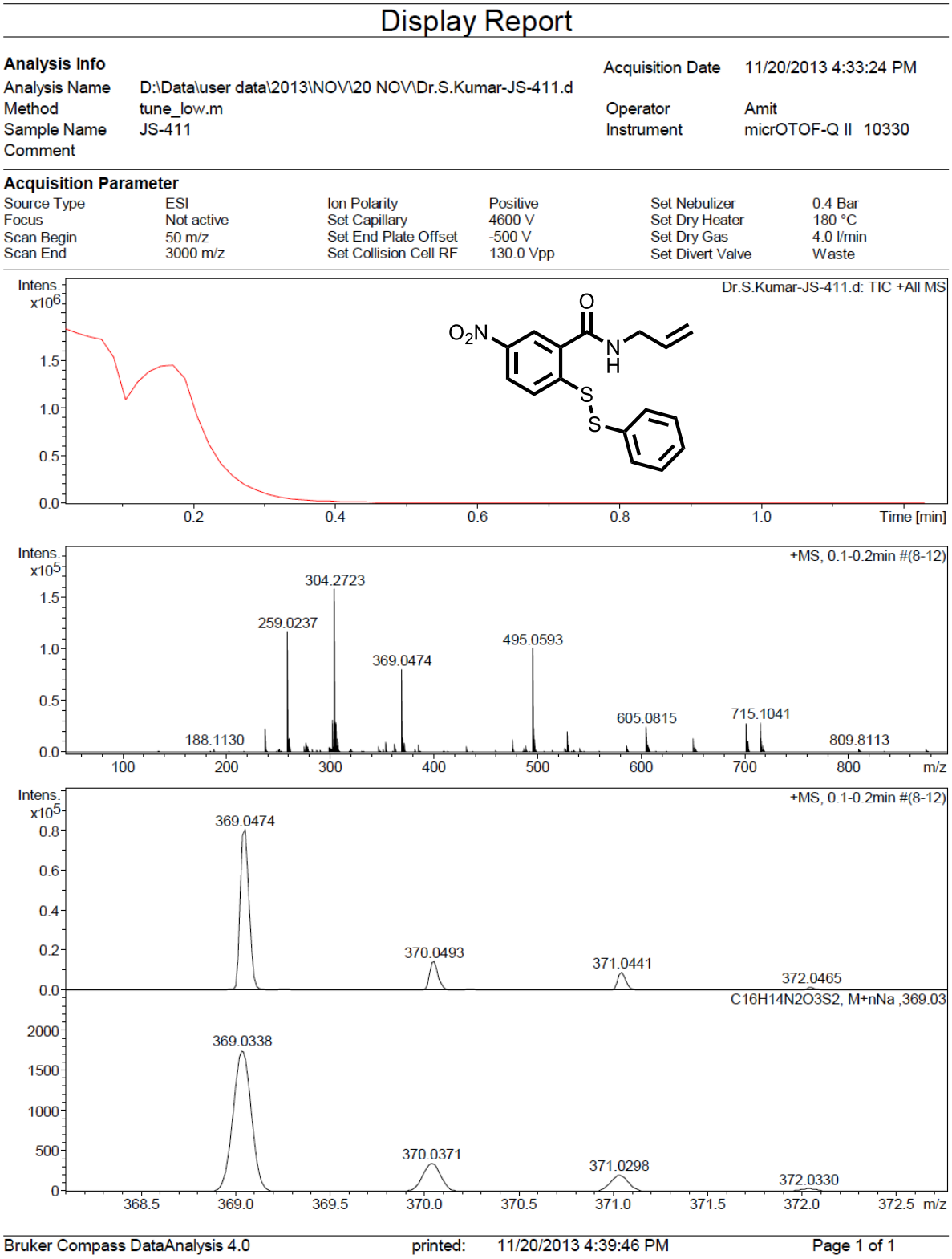


Figure S24 ^{1}H NMR of **1b**

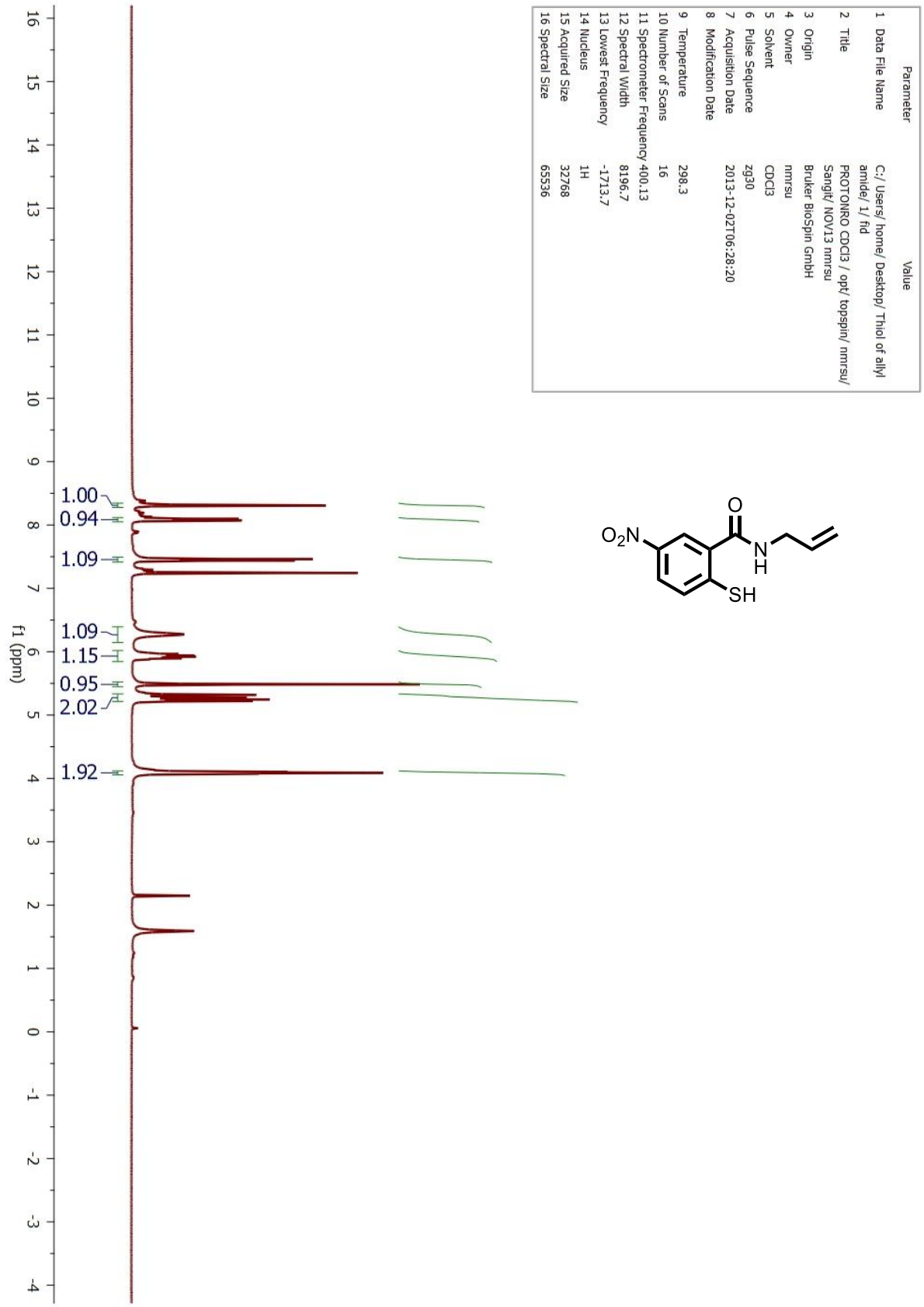


Figure S25 ^{13}C NMR of **1b**

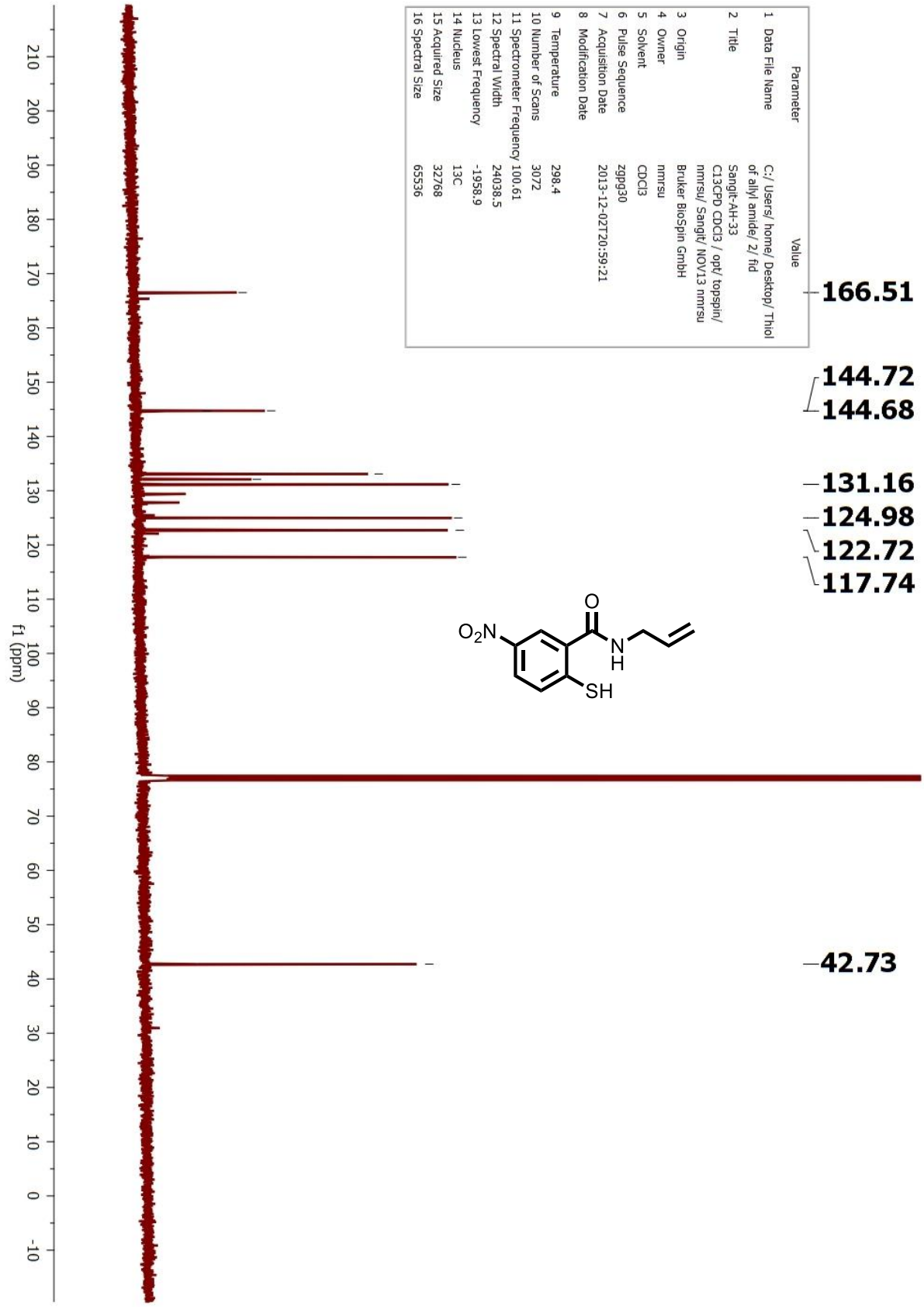


Figure S26 Mass spectra of **1b**

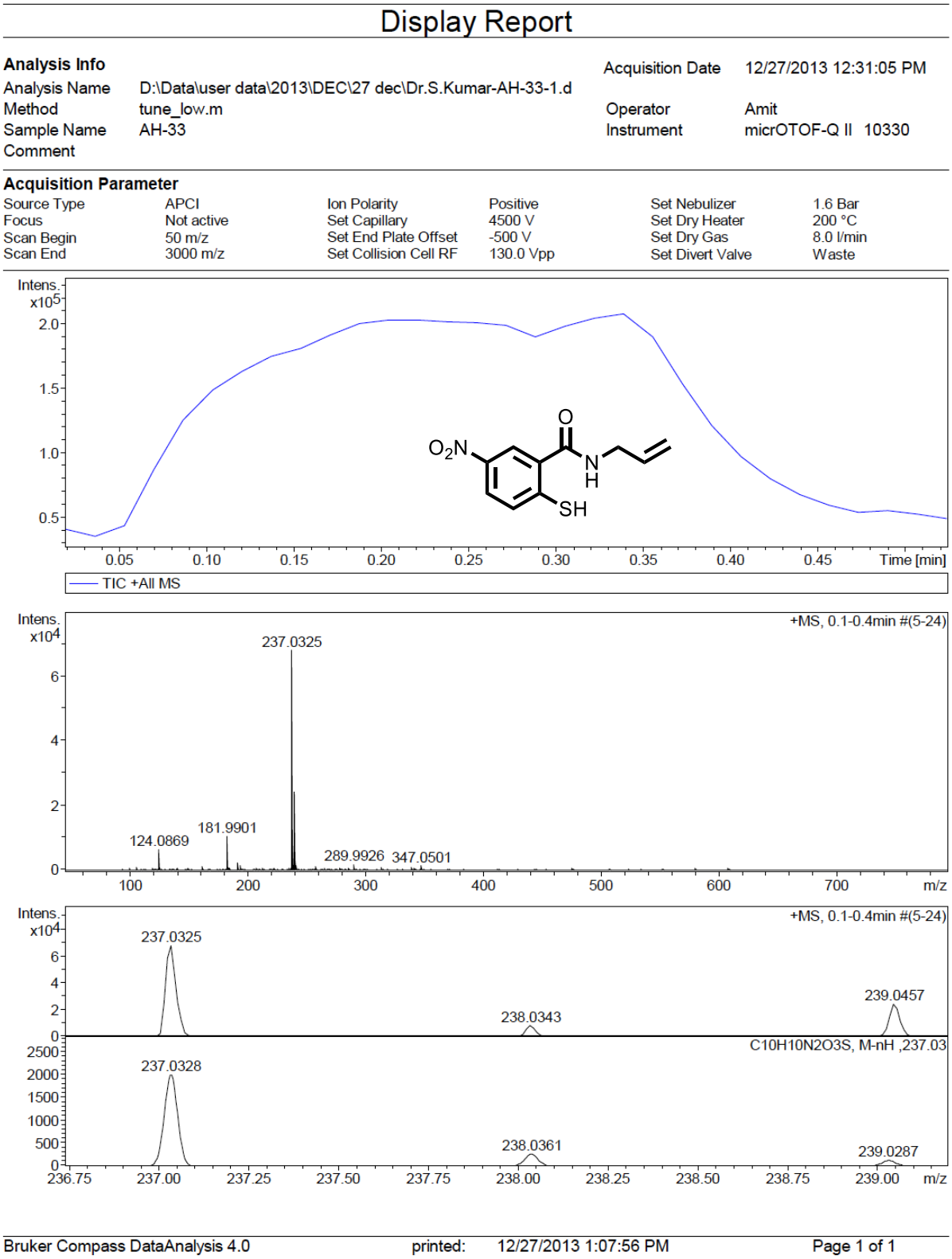


Figure S27 ^{1}H NMR of intermediate **2a**

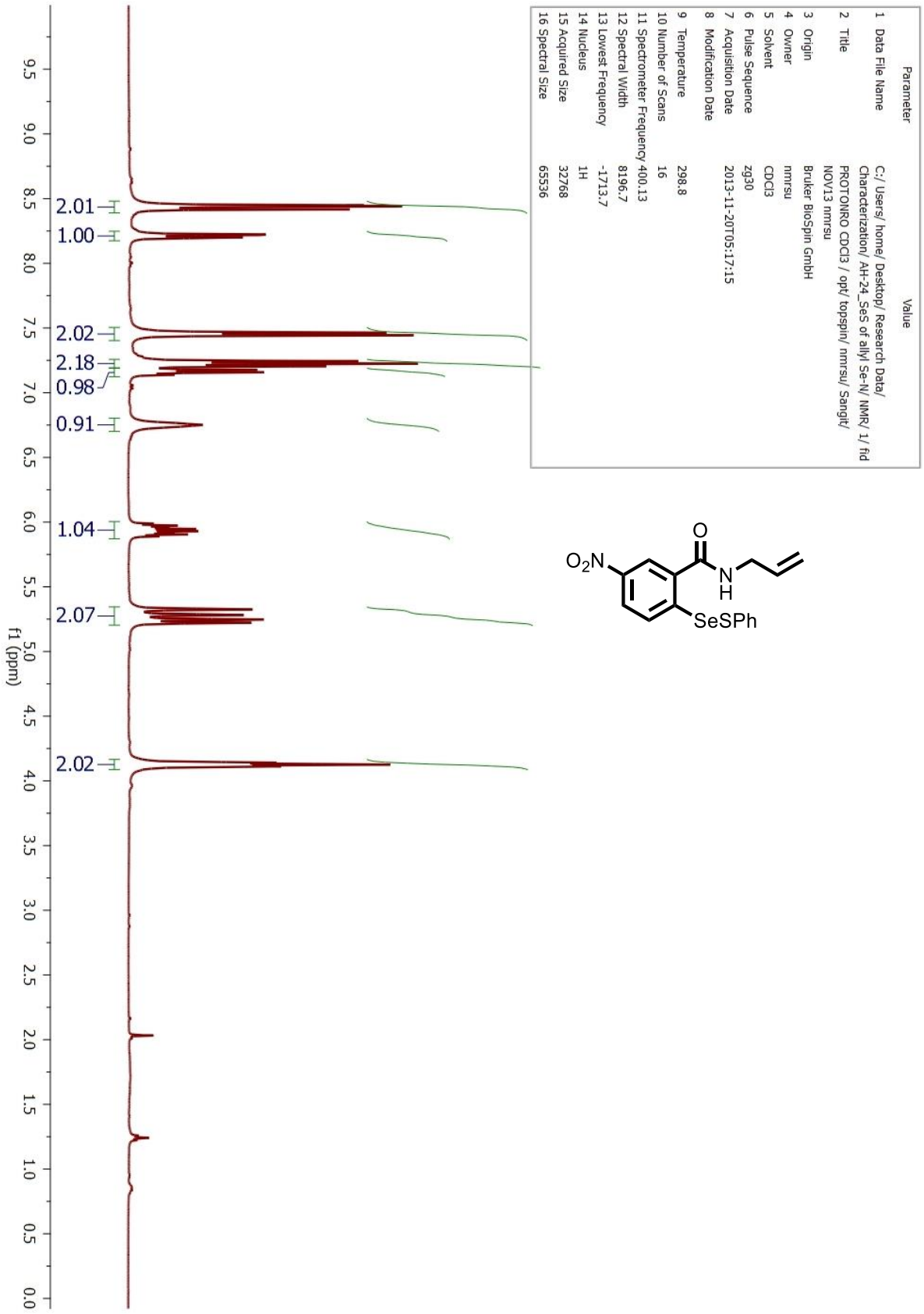


Figure S28 ^{13}C NMR of intermediate **2a**

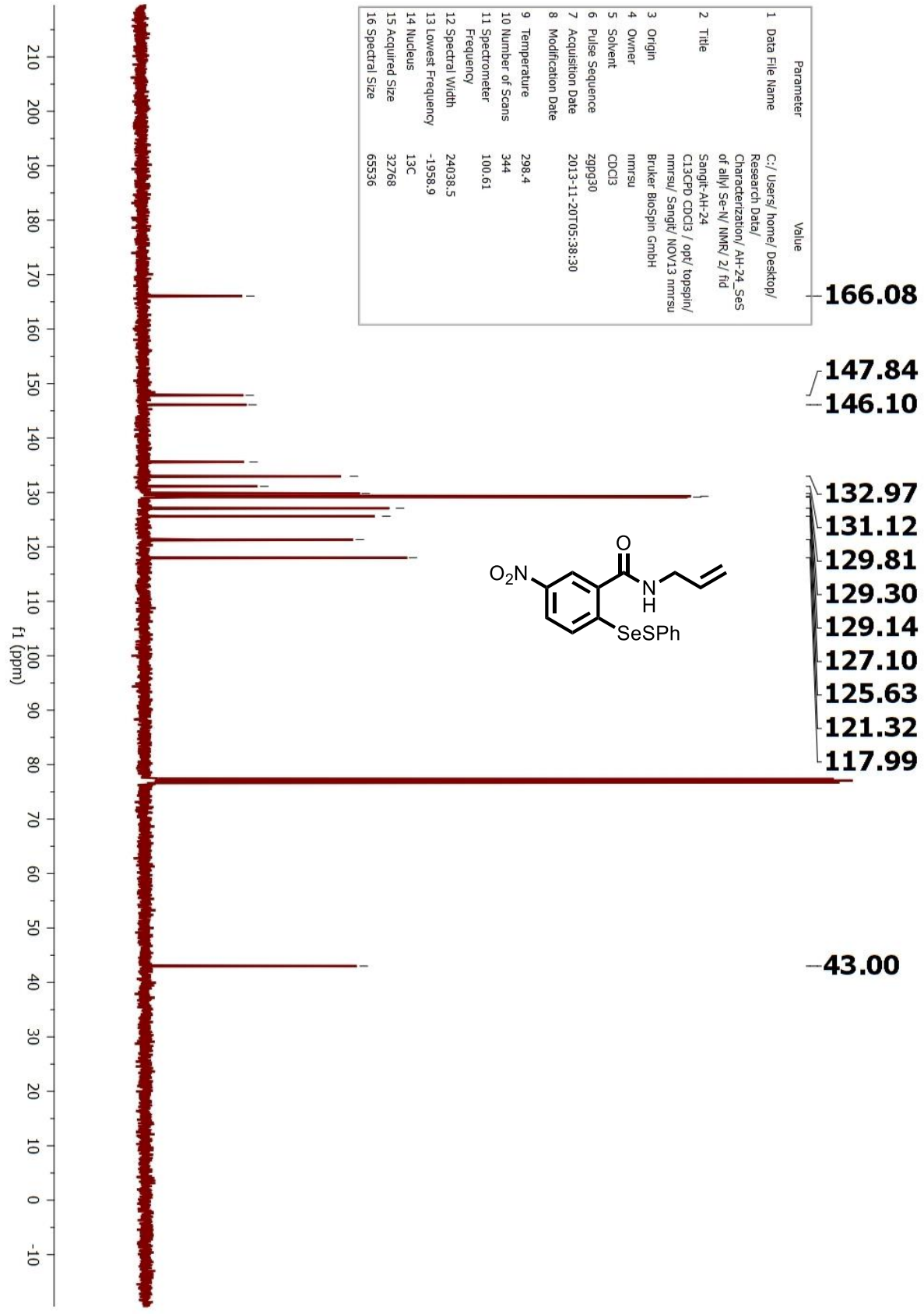


Figure S29 ^{77}Se NMR of intermediate **2a**

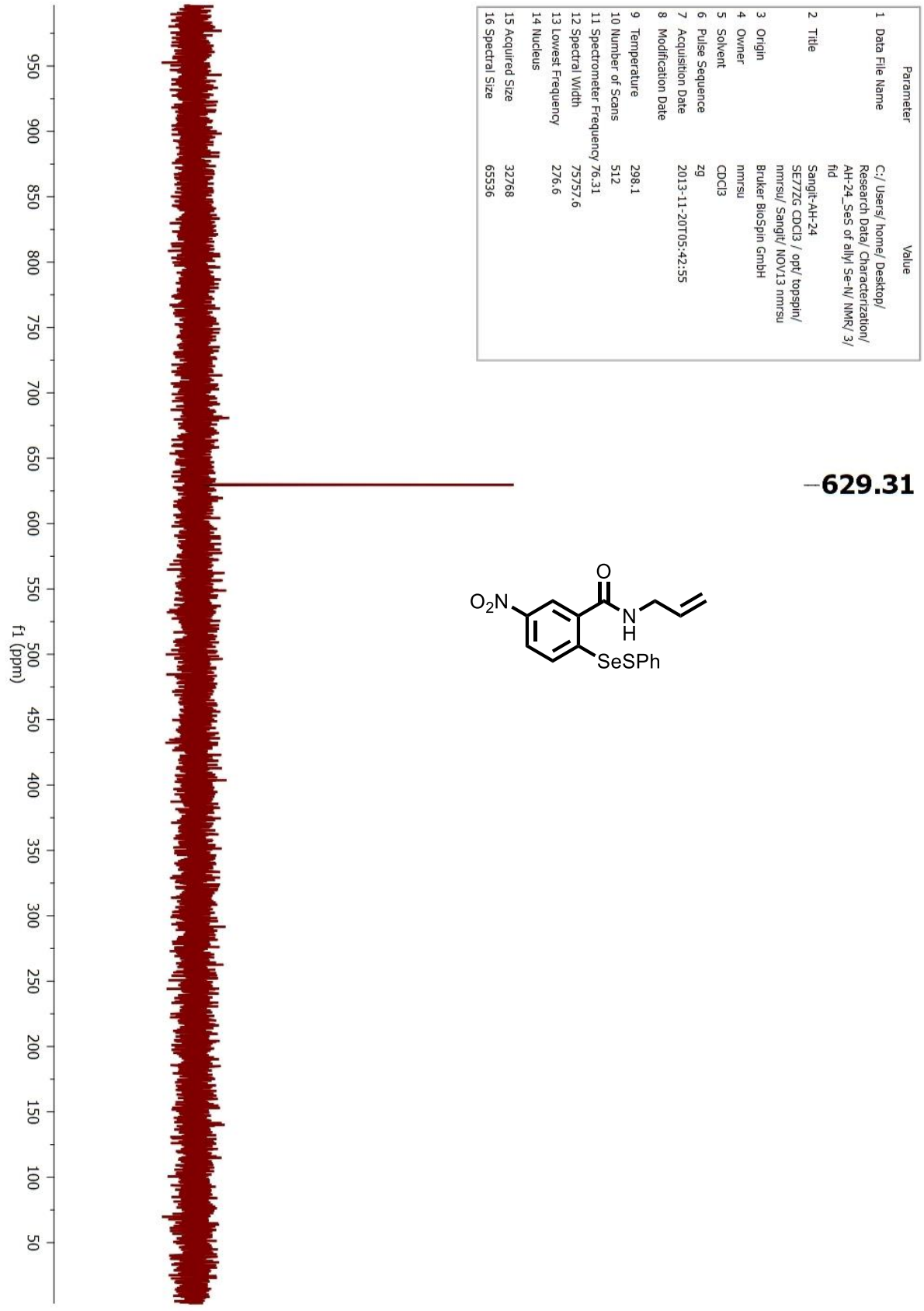


Figure S30 HRMS of intermediate **2a**

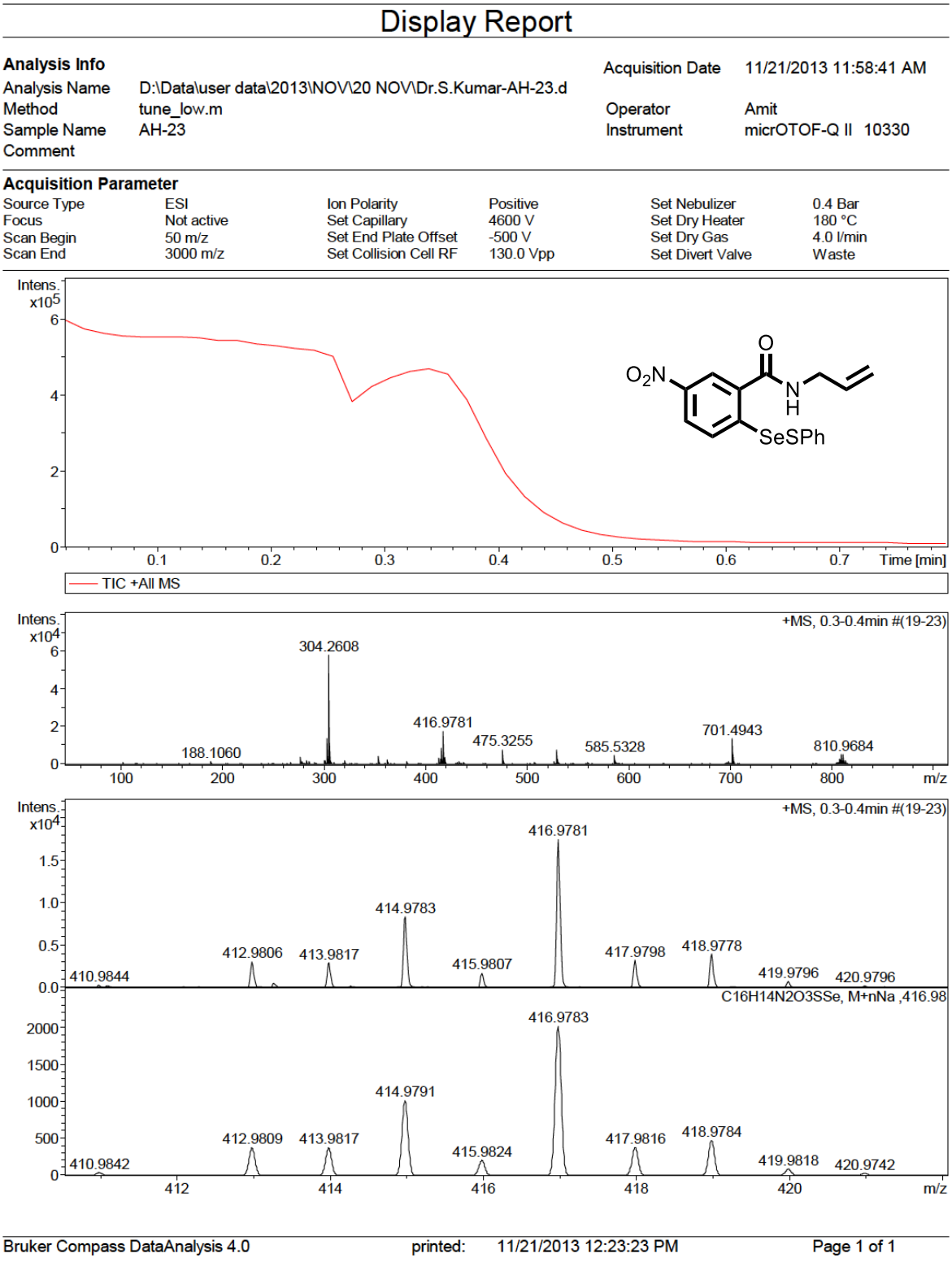


Figure S31 Mass spectra of intermediate **2b**

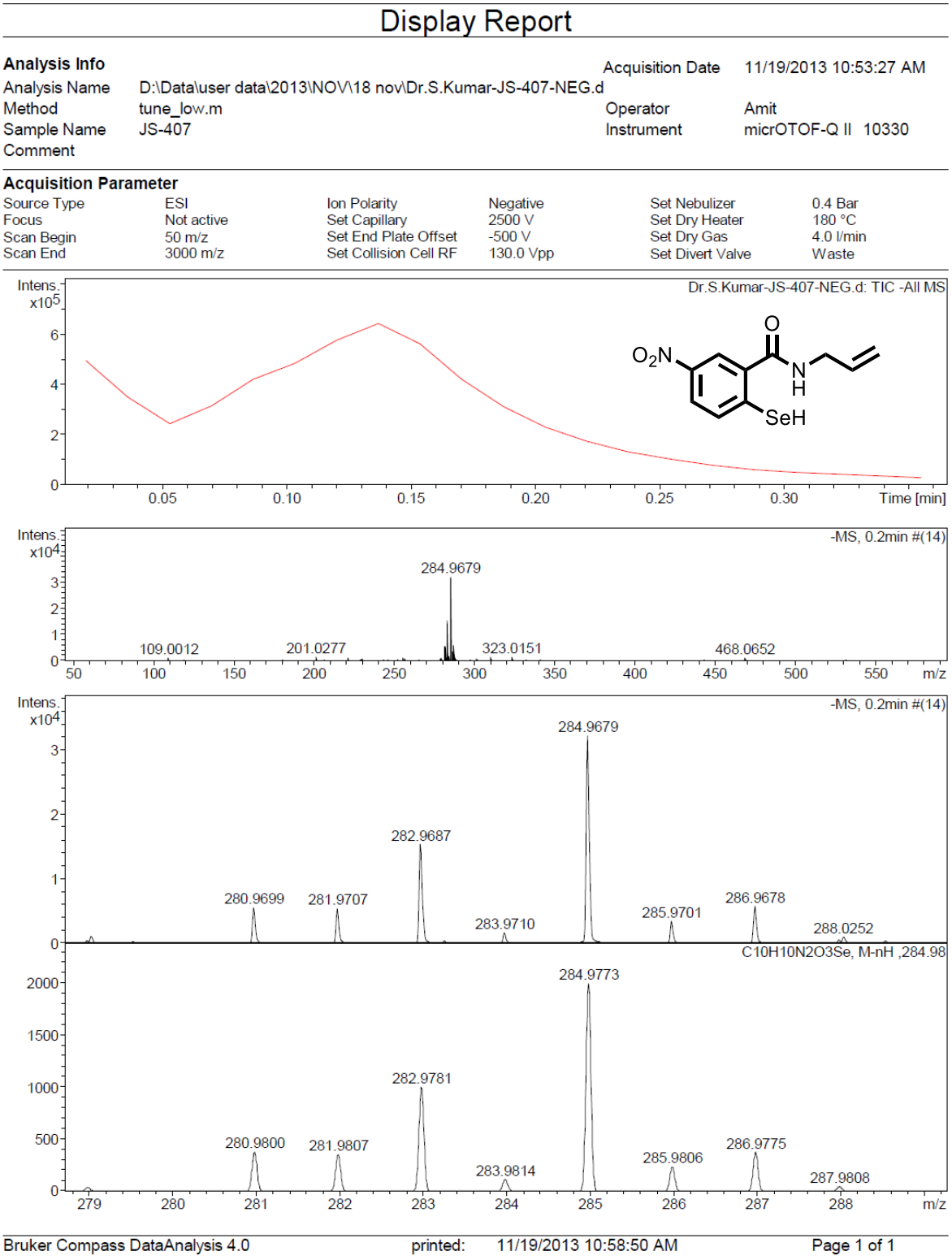


Figure S32 ^{1}H NMR of intermediate **2d**

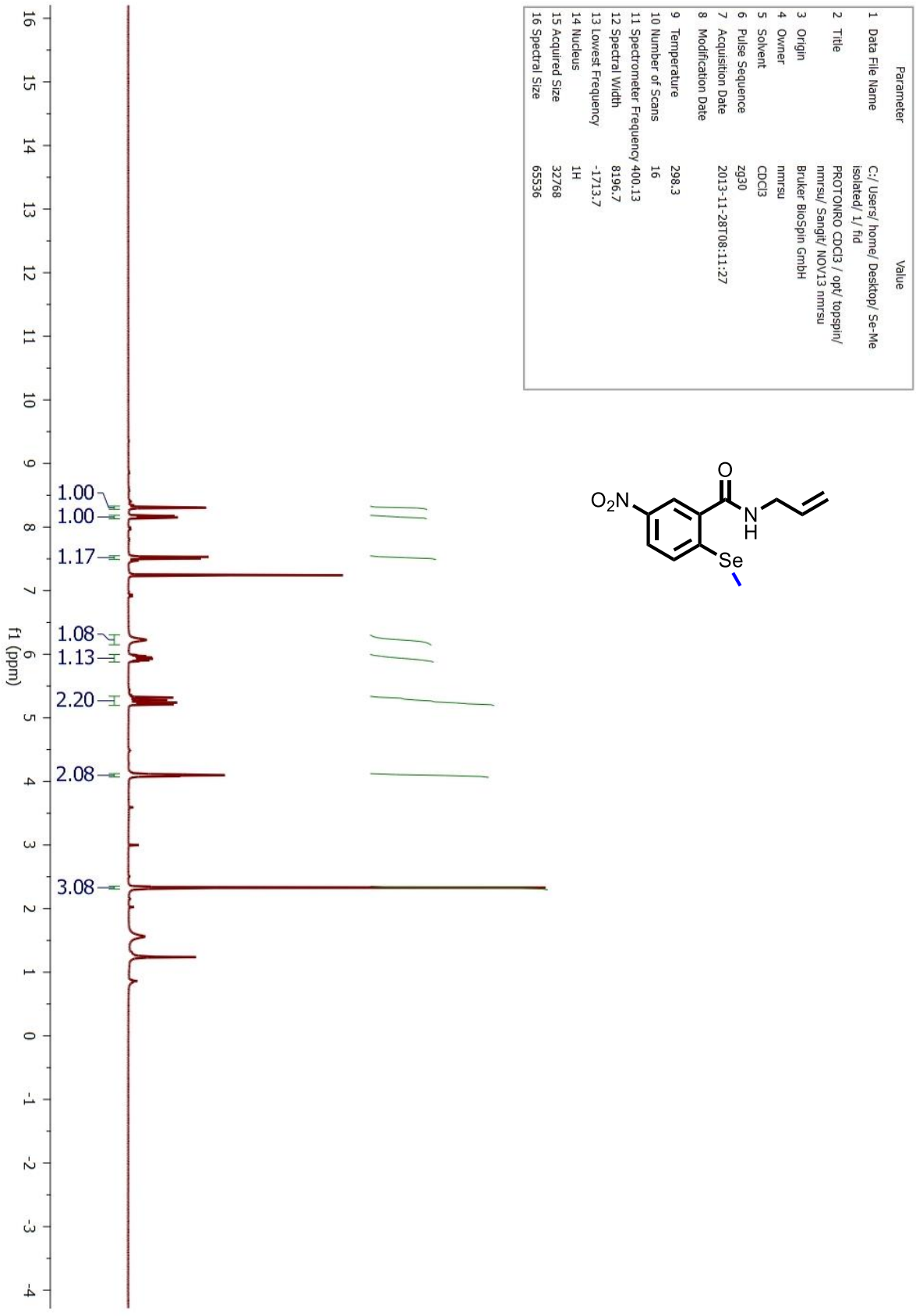


Figure S33 ^{13}C NMR of intermediate **2d**

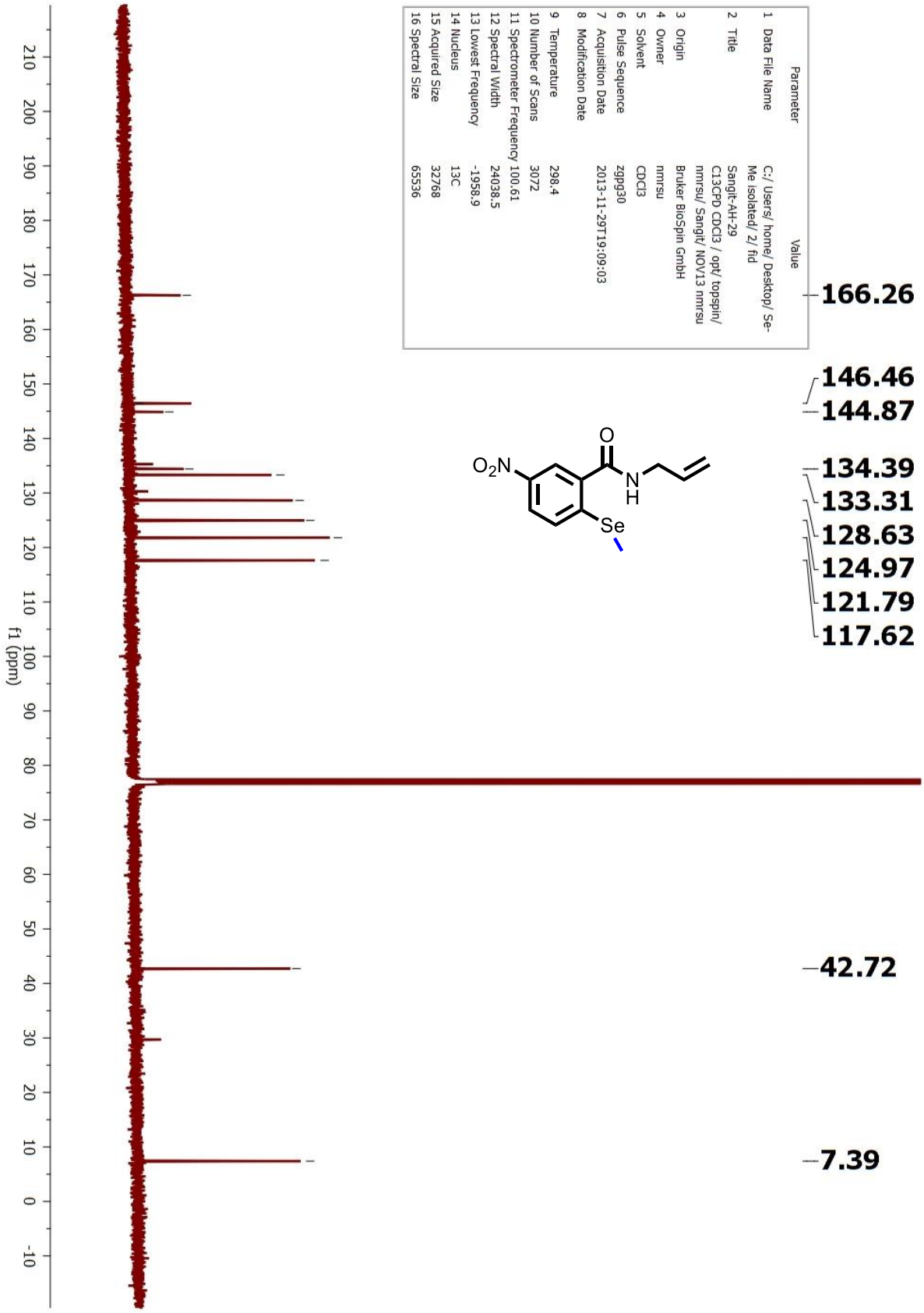


Figure S34 ^{77}Se NMR of intermediate **2d**

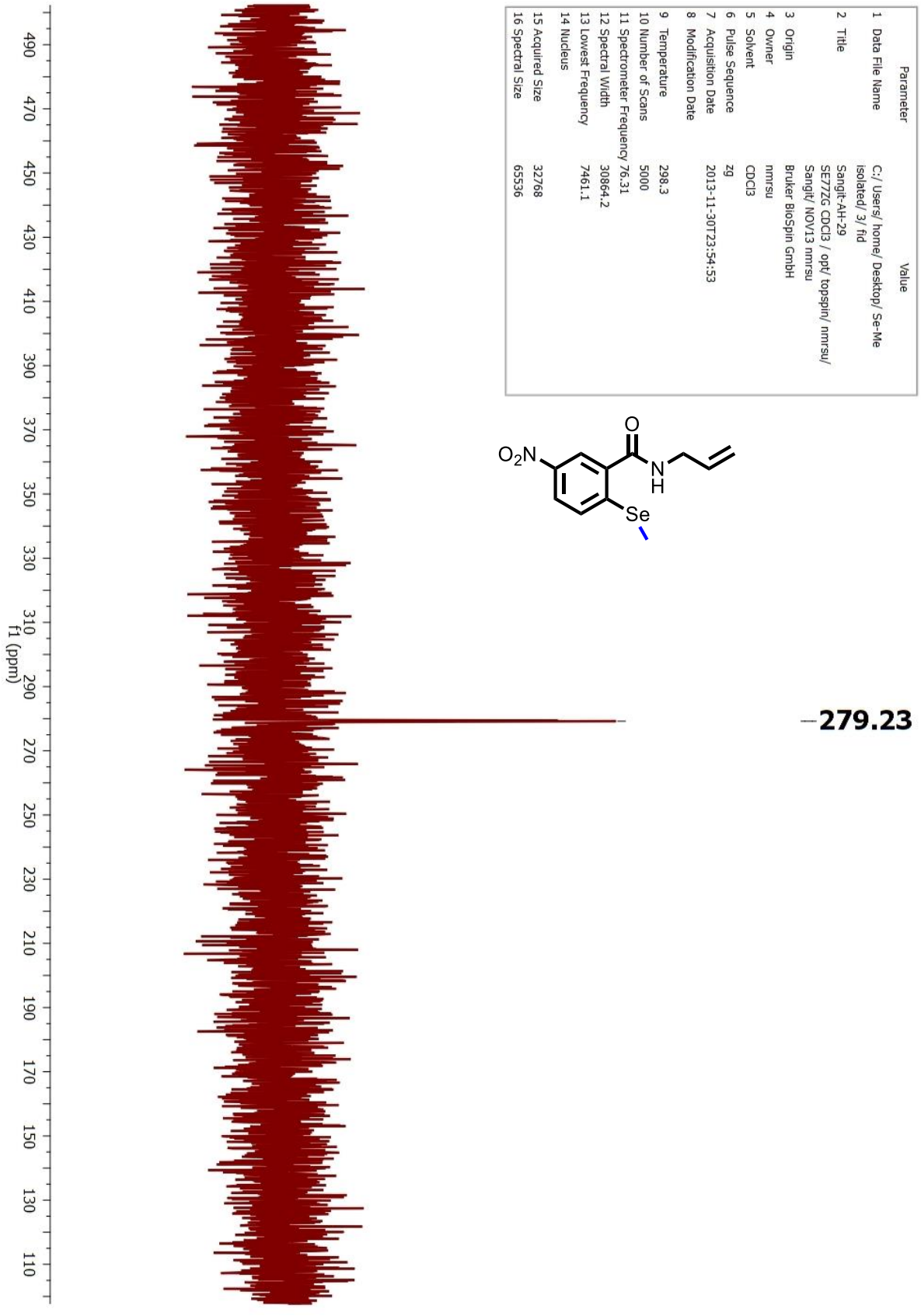


Figure S35 HRMS of intermediate **2d**

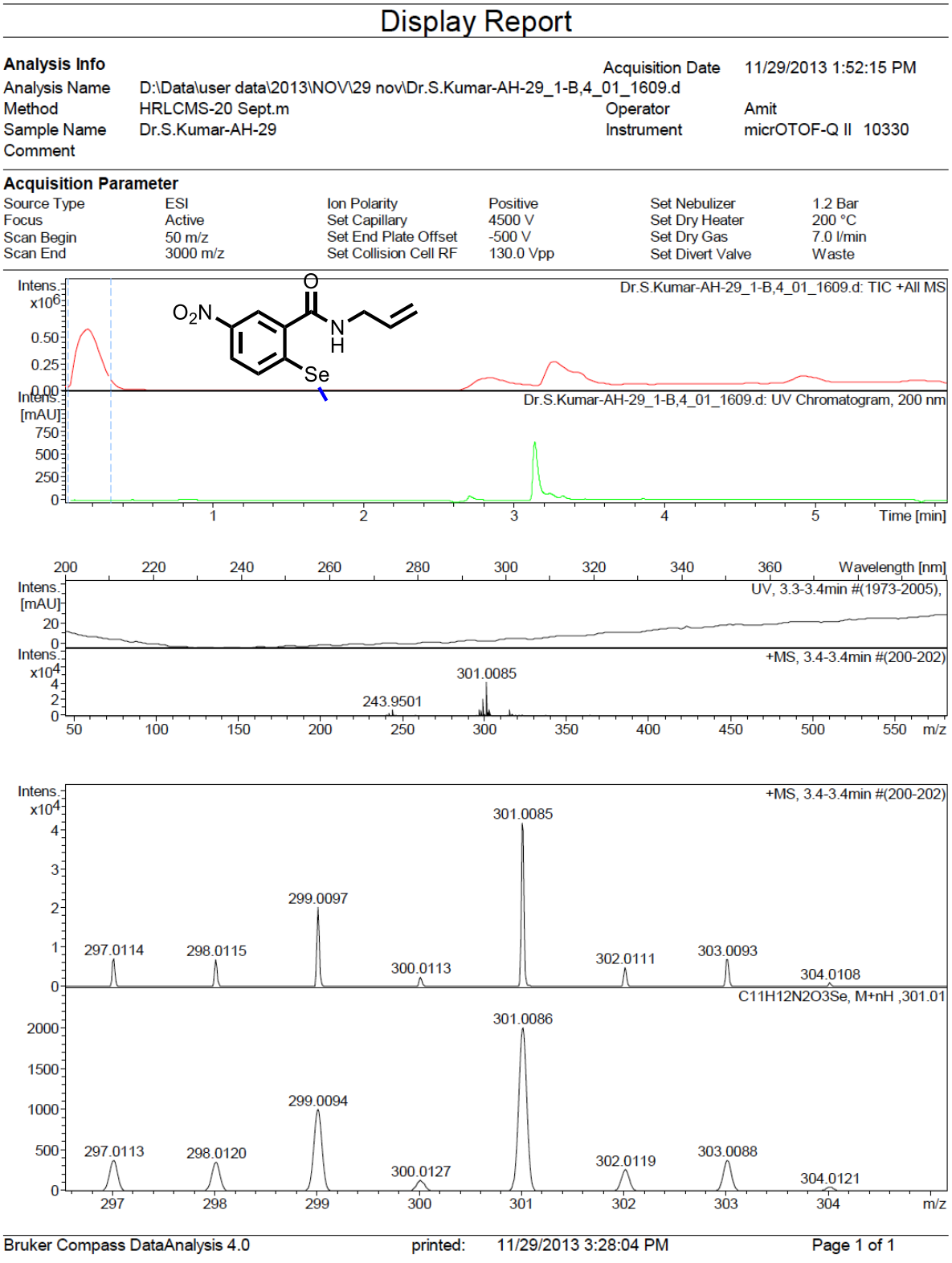
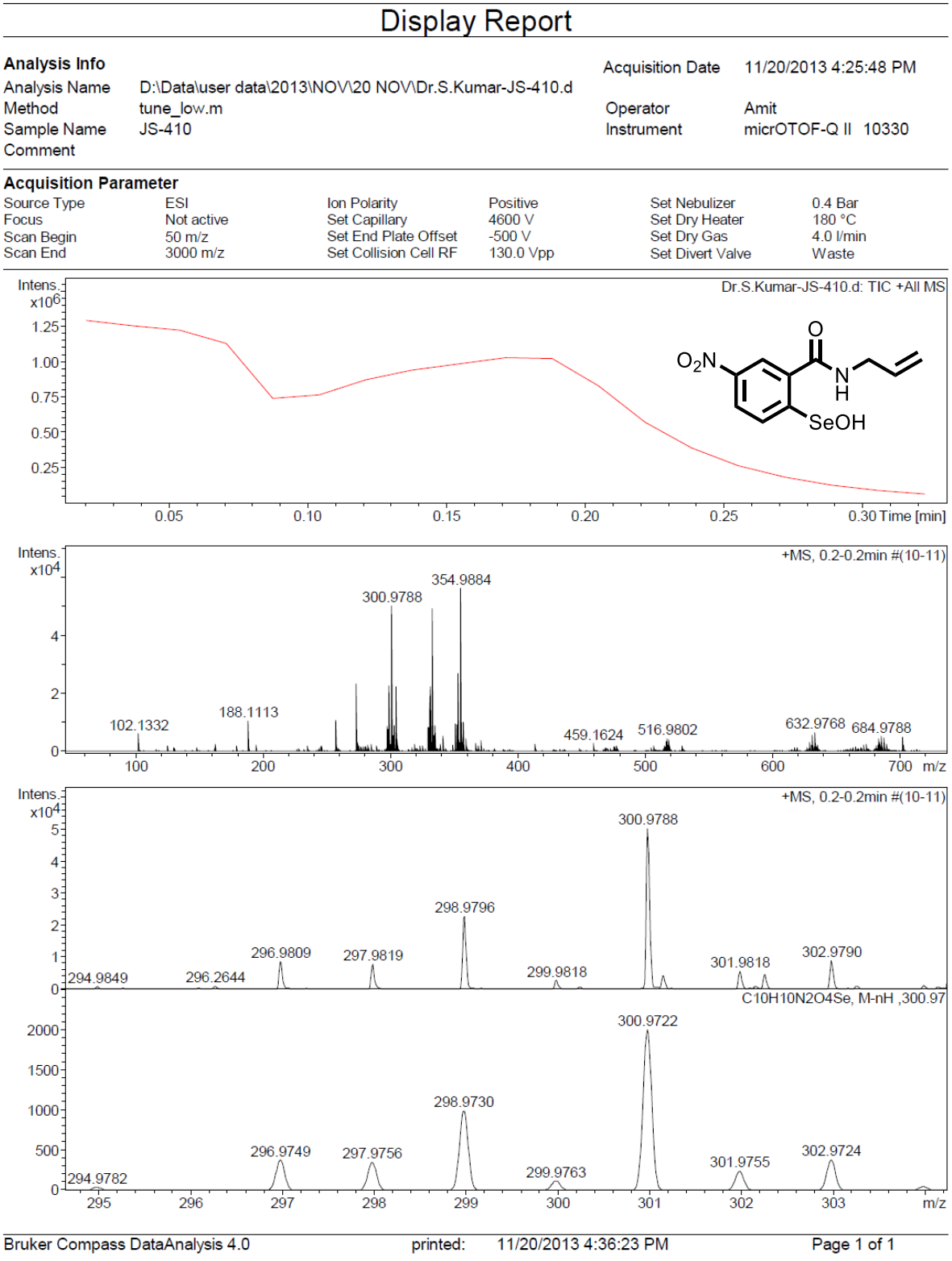


Figure S36 Mass spectra of intermediate **2c**



UV-Vis spectroscopic data for compound 1

Figure 37. Thiol detection limit for compound **1**.

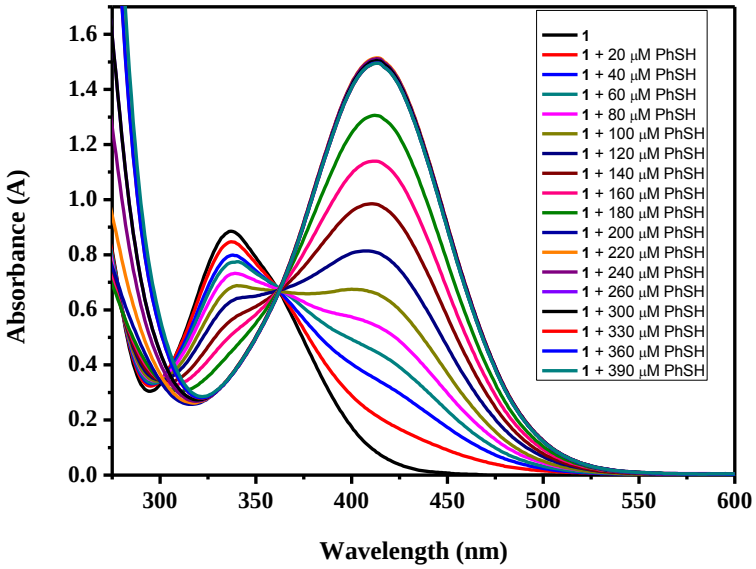


Figure 38. Linear change in absorbance at λ = 413 nm with change in conc.of PhSH

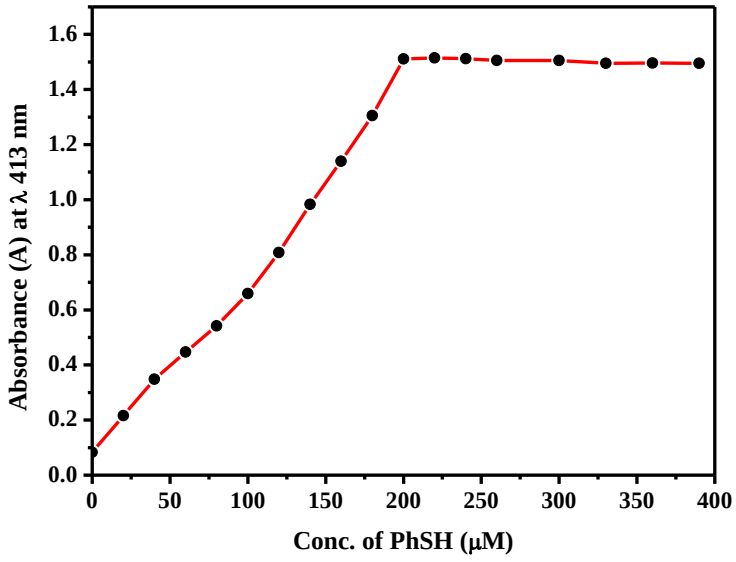


Figure 39. Selectivity of compound **1** towards different thiols and organic compounds.

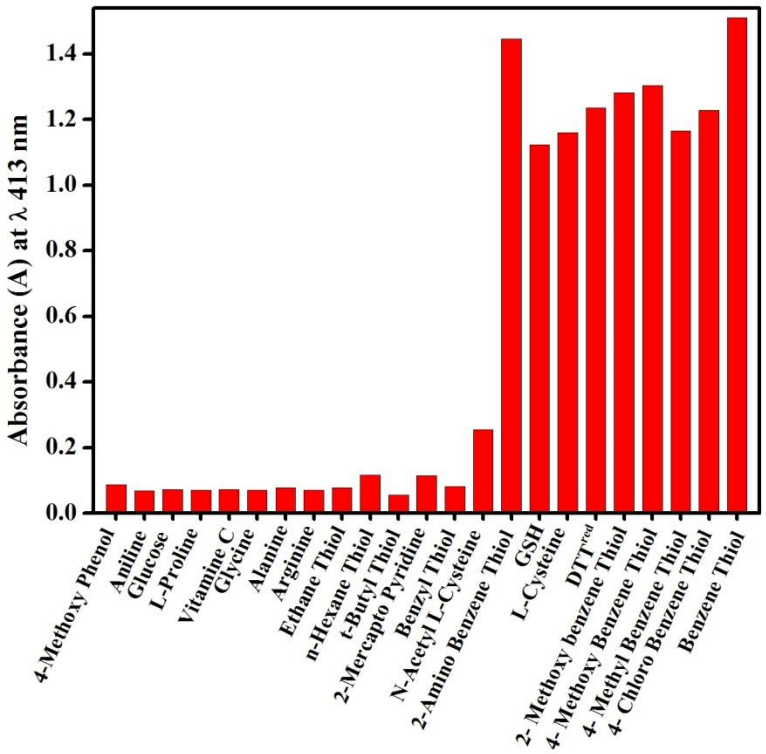
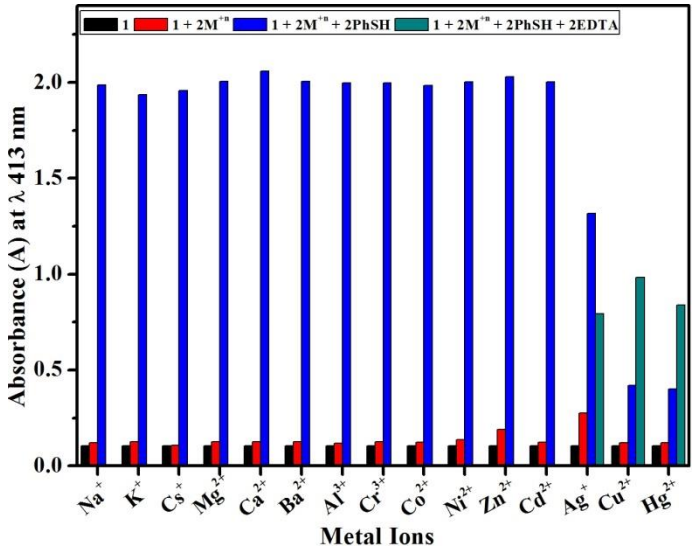
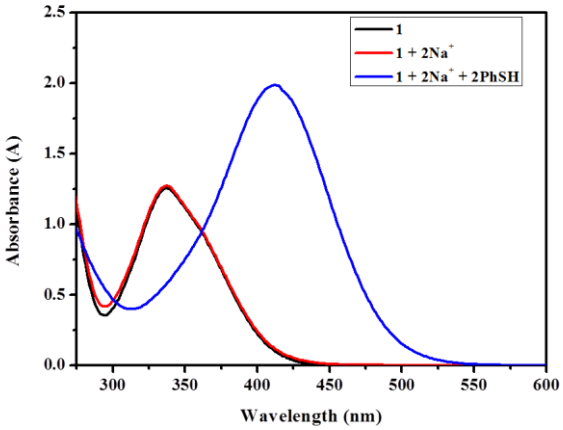
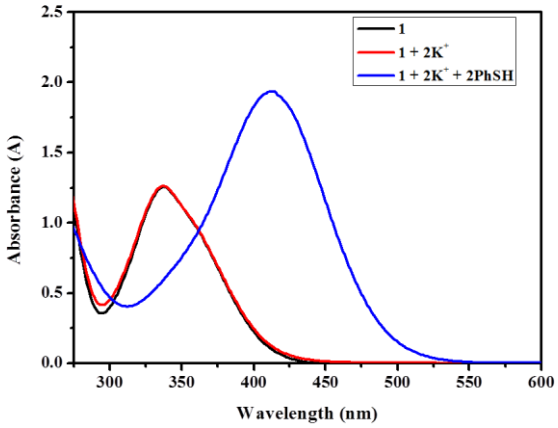
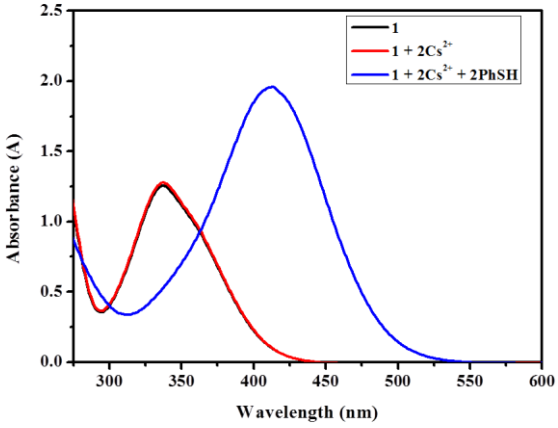


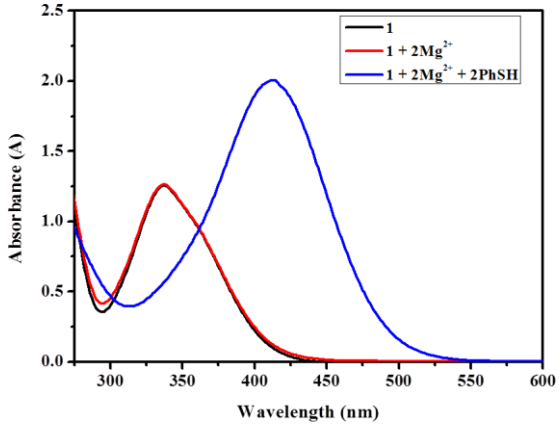
Figure 40. Metal ion inhibition effects for compound **1**.

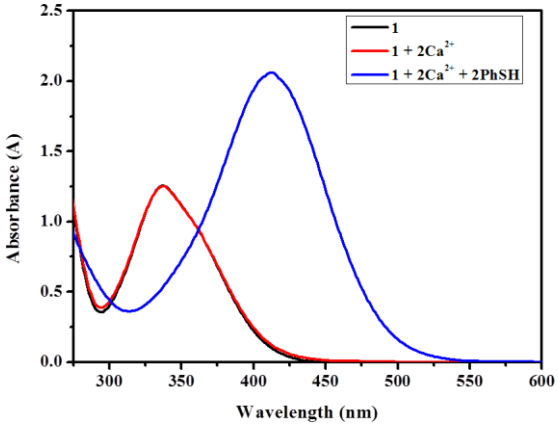


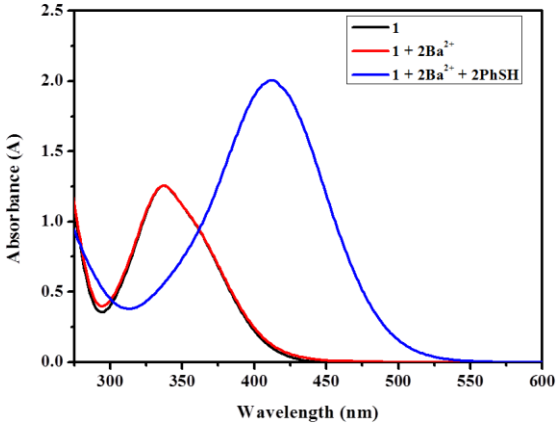

1
1 + 2Na+
1 + 2Na+ + 2PhSH
Absorbance (A)
Wavelength (nm)

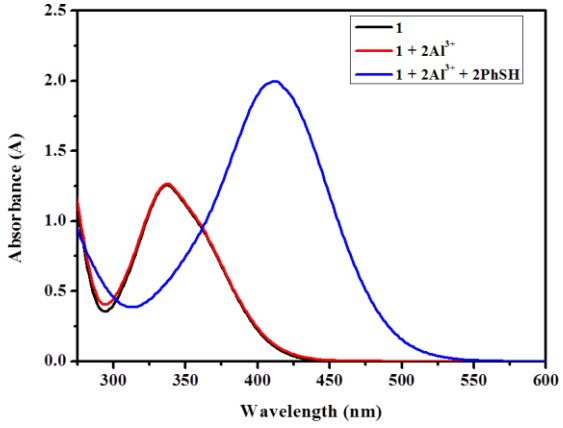

1
1 + 2K+
1 + 2K+ + 2PhSH
Absorbance (A)
Wavelength (nm)

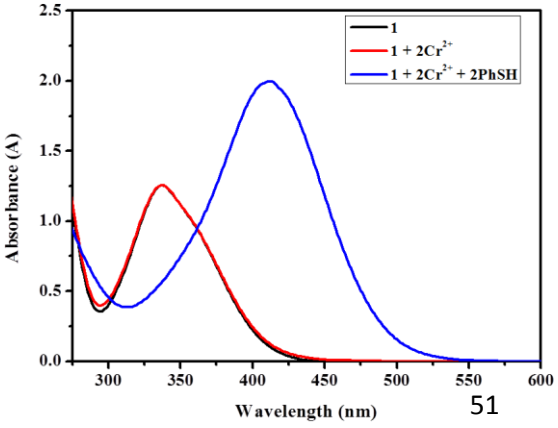

1
1 + 2Cs2+
1 + 2Cs2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

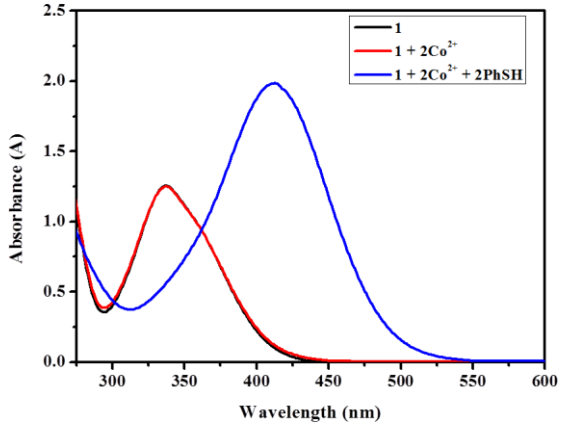

1
1 + 2Mg2+
1 + 2Mg2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

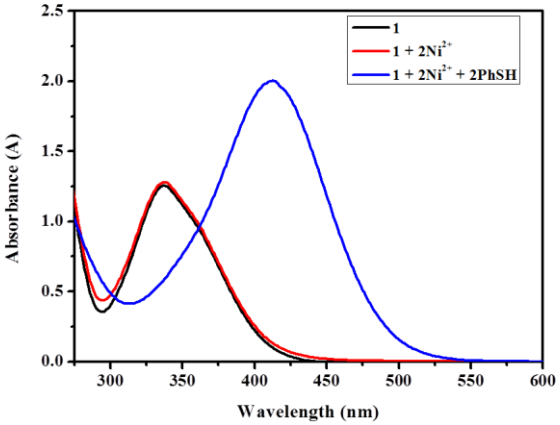

1
1 + 2Ca2+
1 + 2Ca2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

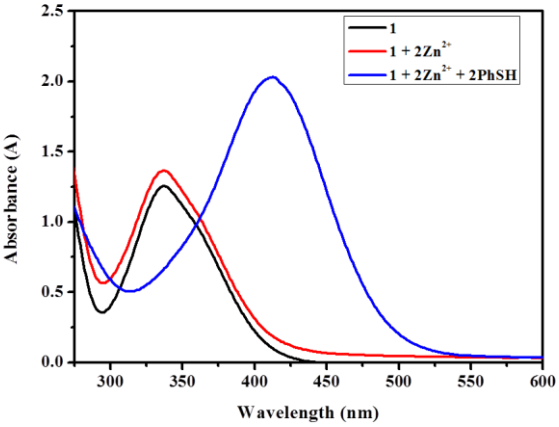

1
1 + 2Ba2+
1 + 2Ba2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

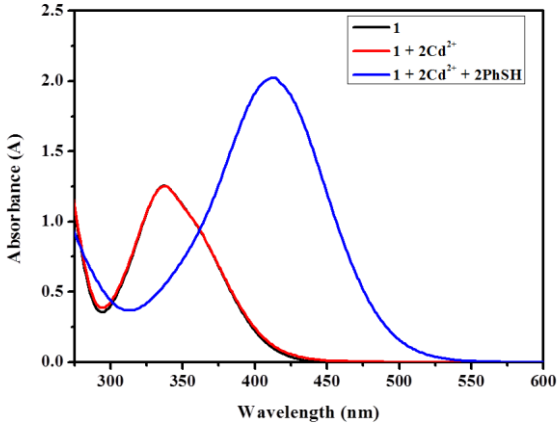

1
1 + 2Al3+
1 + 2Al3+ + 2PhSH
Absorbance (A)
Wavelength (nm)

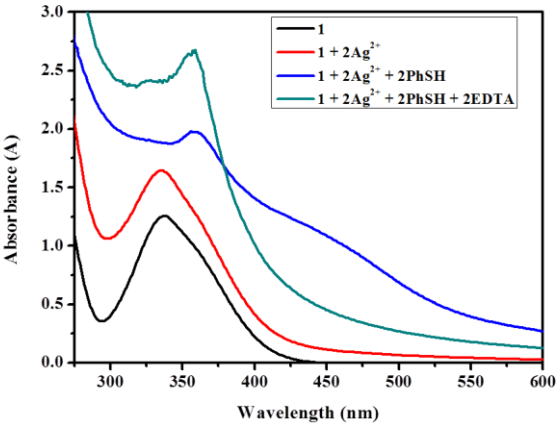

1
1 + 2Cr2+
1 + 2Cr2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

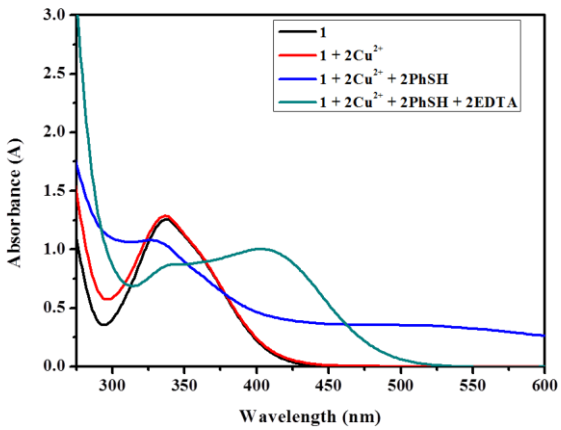

1
1 + 2Co2+
1 + 2Co2+ + 2PhSH
Absorbance (A)
Wavelength (nm)

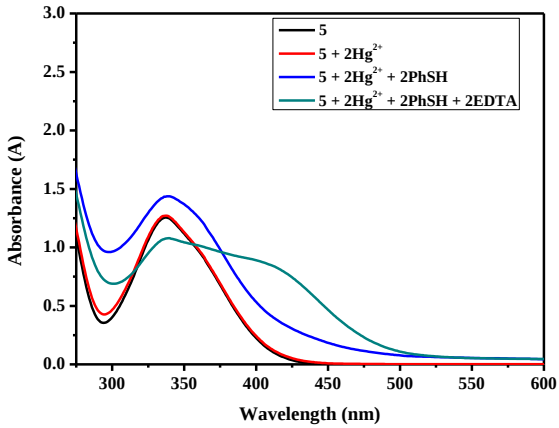

1
1 + 2Ni2+
1 + 2Ni2+ + 2PhSH
Absorbance (A)
Wavelength (nm)


1
1 + 2Zn2+
1 + 2Zn2+ + 2PhSH
Absorbance (A)
Wavelength (nm)


1
1 + 2Cd2+
1 + 2Cd2+ + 2PhSH
Absorbance (A)
Wavelength (nm)


1
1 + 2Ag2+
1 + 2Ag2+ + 2PhSH
1 + 2Ag2+ + 2PhSH + 2EDTA
Absorbance (A)
Wavelength (nm)


1
1 + 2Cu2+
1 + 2Cu2+ + 2PhSH
1 + 2Cu2+ + 2PhSH + 2EDTA
Absorbance (A)
Wavelength (nm)


5
5 + 2Hg2+
5 + 2Hg2+ + 2PhSH
5 + 2Hg2+ + 2PhSH + 2EDTA
Absorbance (A)
Wavelength (nm)

UV-Vis spectroscopic data for compound 2

Figure 41. Thiol detection limit for compound **2**.

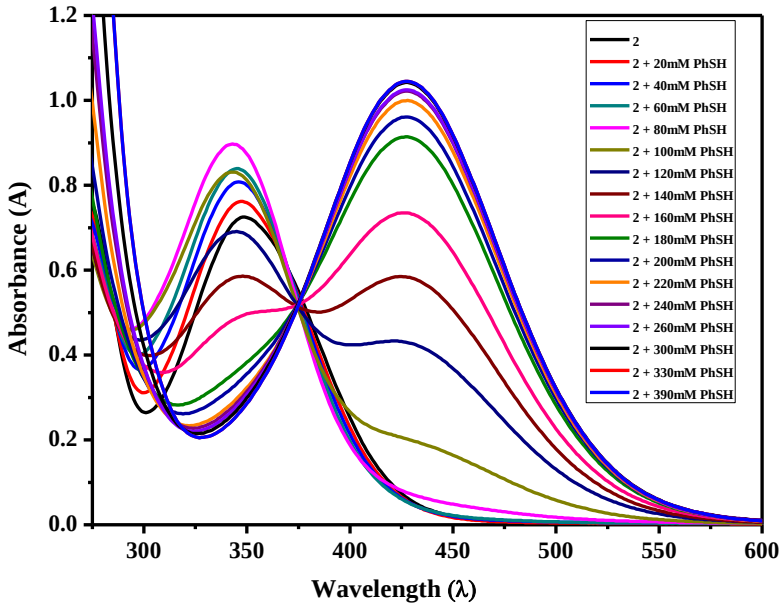


Figure 42. Change in absorbance at λ = 426 nm with change in conc. of PhSH

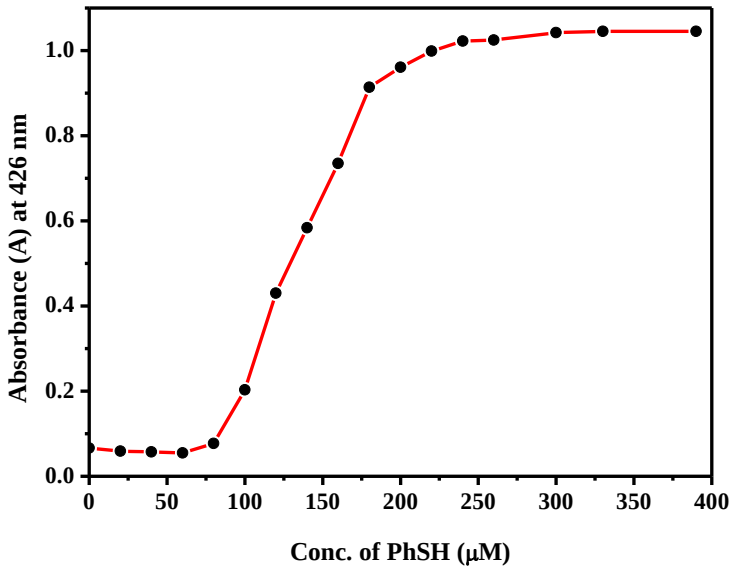


Figure 43. Change in absorbance at λ = 343 nm with change in conc. of PhSH

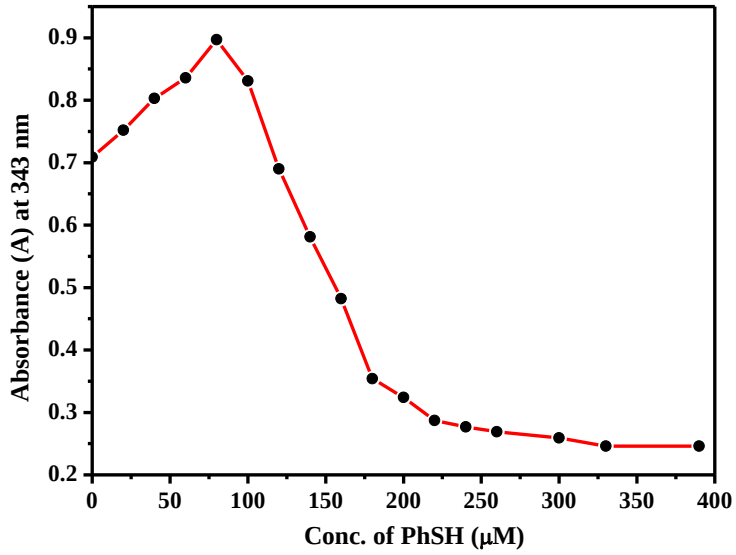


Figure 44. Selectivity of probe **2** against various organic compounds and thiols.

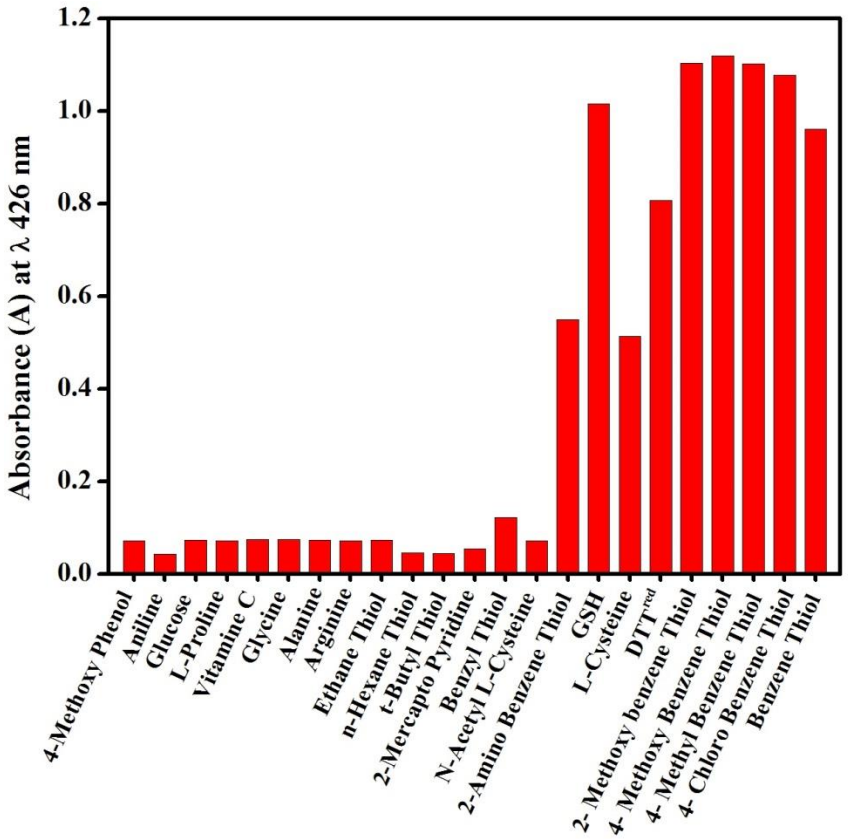
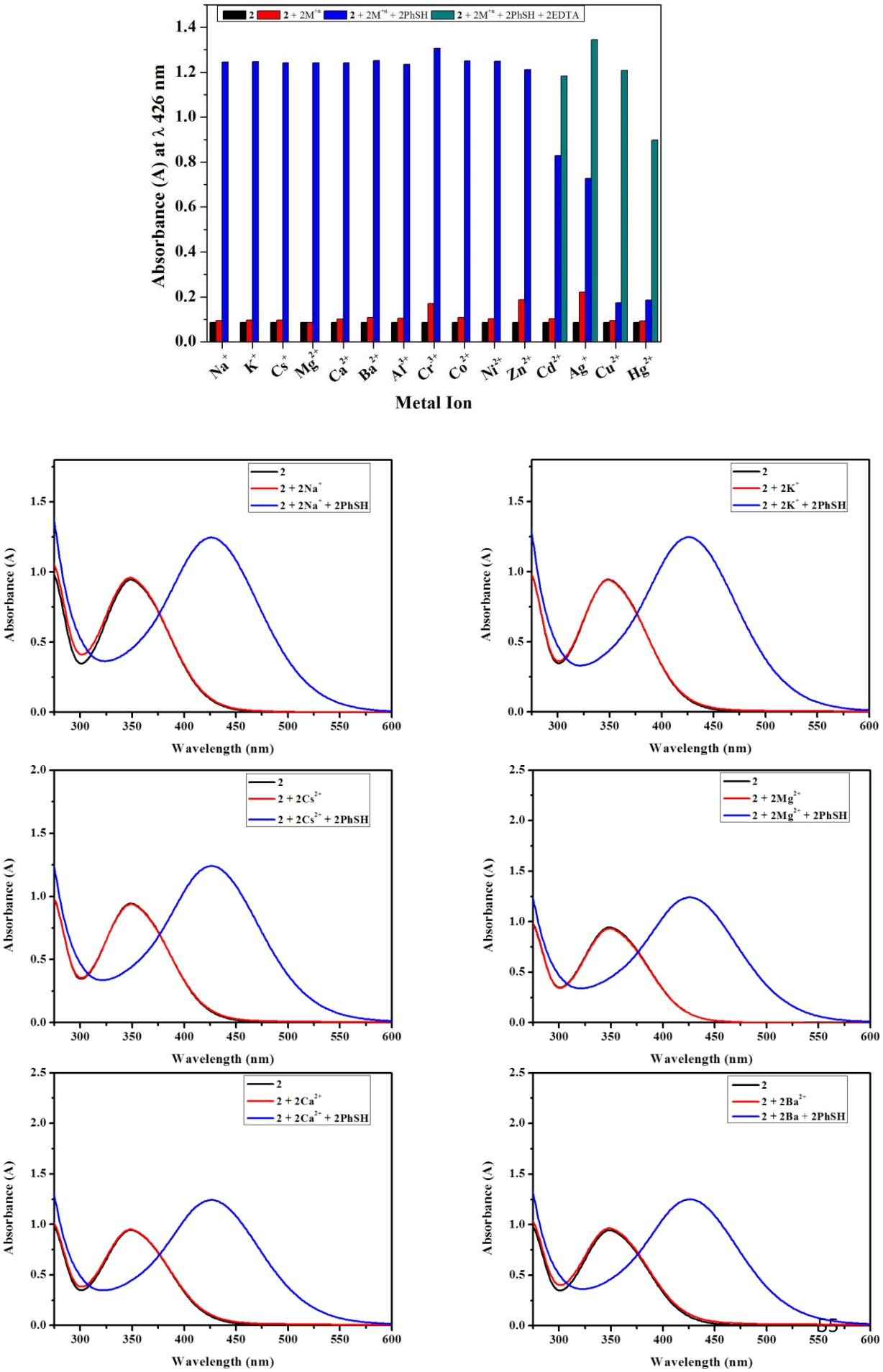
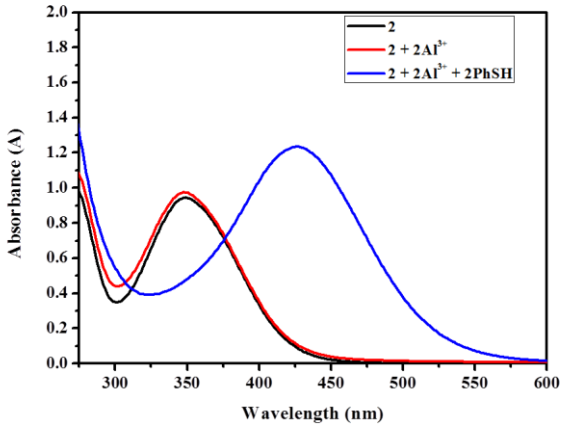
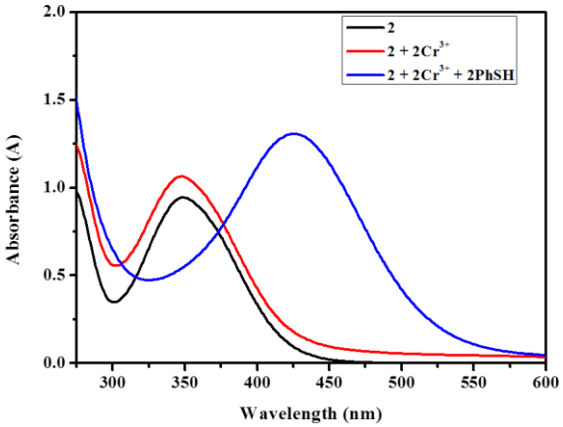
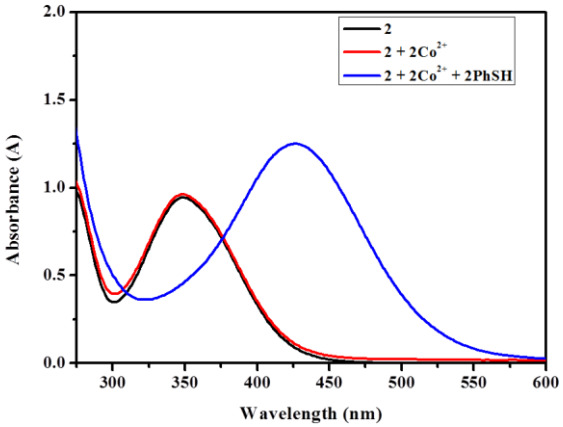


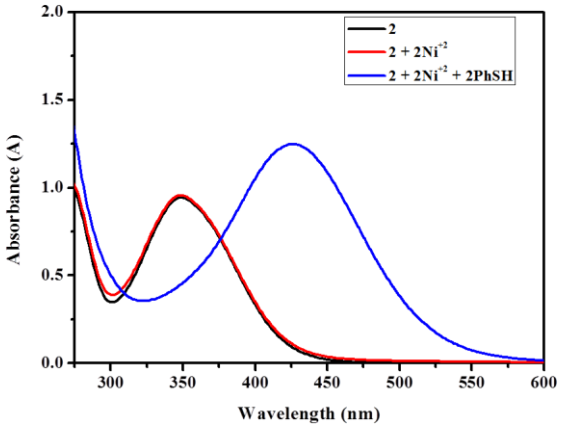
Figure 45. Metal ion inhibition effects for probe **2**.

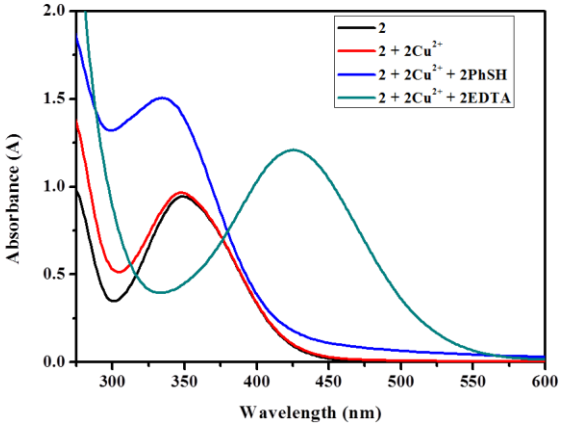


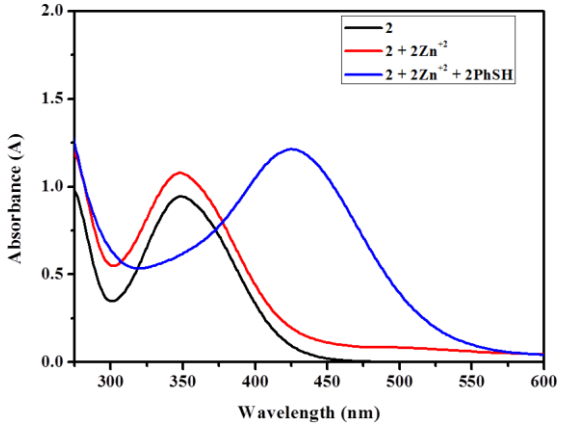

2
2 + 2Al3+
2 + 2Al3+ + 2PhSH
Absorbance (A)
Wavelength (nm)

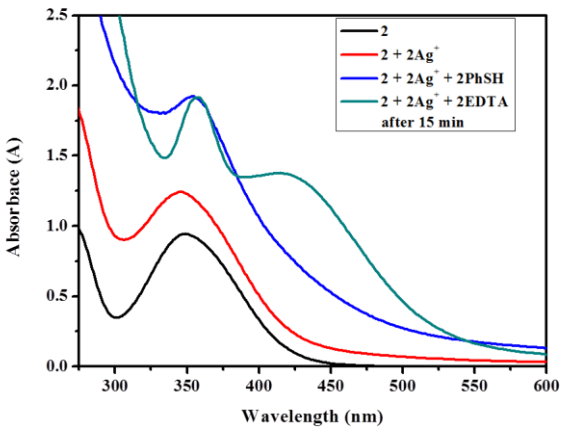

2
2 + 2Cr3+
2 + 2Cr3+ + 2PhSH
Absorbance (A)
Wavelength (nm)

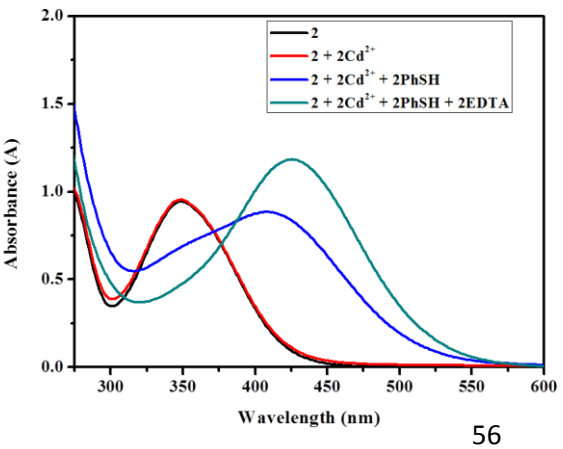

2
2 + 2Co2+
2 + 2Co2+ + 2PhSH
Absorbance (A)
Wavelength (nm)


2
2 + 2Ni+2
2 + 2Ni+2 + 2PhSH
Absorbance (A)
Wavelength (nm)


2
2 + 2Cu2+
2 + 2Cu2+ + 2PhSH
2 + 2Cu2+ + 2EDTA
Absorbance (A)
Wavelength (nm)


2
2 + 2Zn+2
2 + 2Zn+2 + 2PhSH
Absorbance (A)
Wavelength (nm)


2
2 + 2Ag+
2 + 2Ag+ + 2PhSH
2 + 2Ag+ + 2EDTA
after 15 min
Absorbace (A)
Wavelength (nm)


2
2 + 2Cd2+
2 + 2Cd2+ + 2PhSH
2 + 2Cd2+ + 2PhSH + 2EDTA
Absorbance (A)
Wavelength (nm)

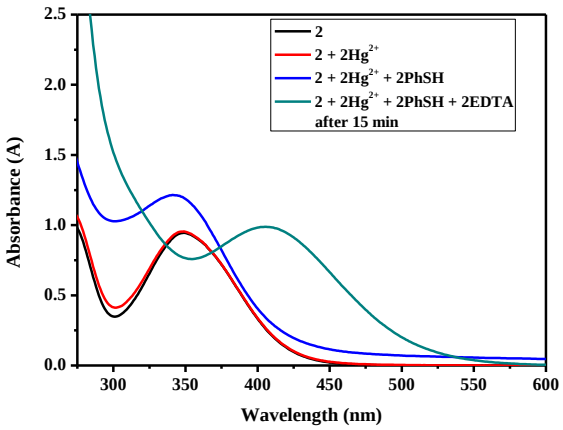


Figure 46. Reversibility of probe **2** upon successive addition of PhSH and *tert*-butyl hydroperoxides.

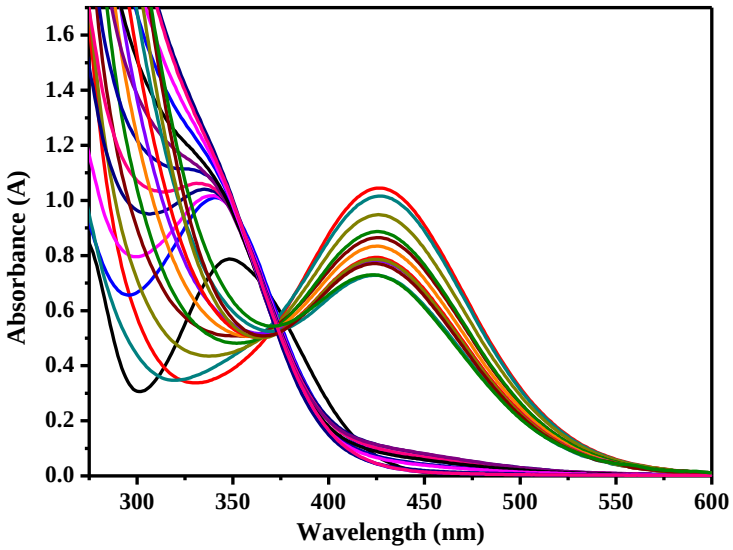
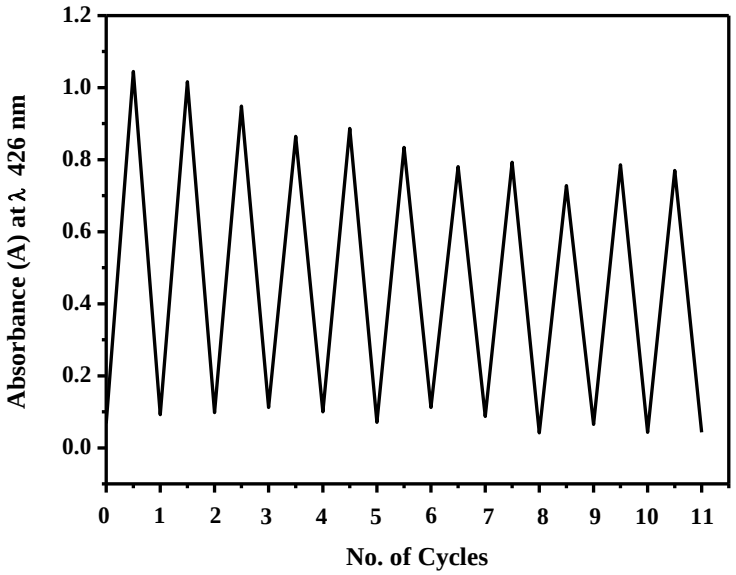


Figure 47. Reversible behaviour of probe **2** for 11 cycles



Number of Cycles	Absorbance (A) at 426 nm	
	Upon Addition of PhSH	Upon Addition of *t*-BuOOH
1	1.045	0.068
2	1.016	0.092
3	0.949	0.098
4	0.865	0.112
5	0.887	0.100
6	0.834	0.071
7	0.781	0.112
8	0.793	0.087
9	0.728	0.042
10	0.786	0.065
11	0.770	0.043

Figure 48. Behaviour of compound **2** in only methanol medium

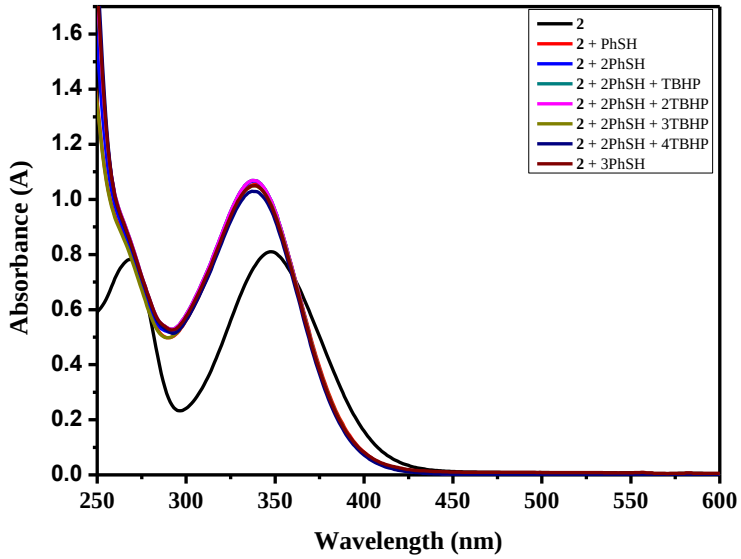


Figure 49. UV-Vis absorbance spectra of selenenylsulfide **2a**.

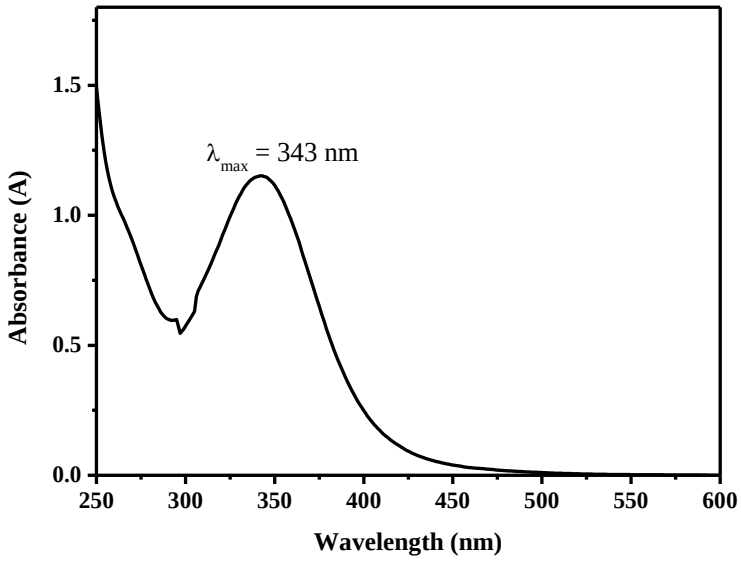
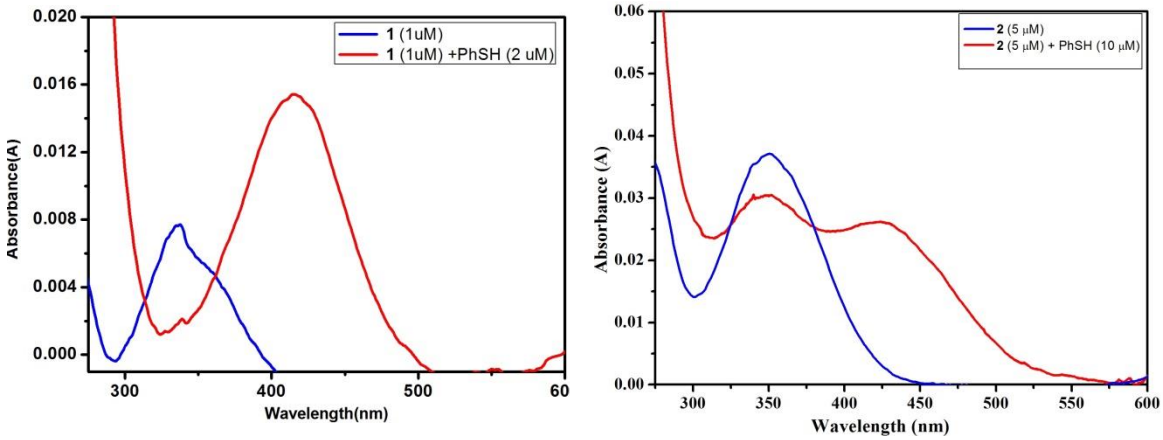


Figure 50. UV-Vis absorbance spectra describing detection limits of **1** and **2**



Crystallographic Details for Compounds 1-4

Data Collection and Structure Solution and Refinement:

Single crystal X-ray diffraction data of compounds **1-4** were collected at 25°C on a Bruker Apex II D8 Venture diffractometer equipped with CMOS detector. Data reduction and integration were performed by SAINT V7.685A12 and absorption corrections and scaling was done using SADABS. All the crystal structures were solved by direct methods using SHELXS 97 and refined by the full matrix least squares method using SHELXL97 present in the program suite WinGX. ORTEP diagrams were generated using ORTEP32. **Table 1** lists the crystallographic and refinement data of **1-4**.

Table 1: Crystallographic and refinement data

DATA	Compound 1	Compound 2	Compound 3	Compound 4
Molecular Formula	$C_{10}H_8N_2O_3S$	$C_{10}H_8N_2O_3Se$	$C_{14}H_{10}N_2O_3Se$	$C_{15}H_{12}N_2O_3Se$
Molecular weight	236.24	283.14	333.20	347.23
Temperature (K)	298	298	298(2)	298(2)
Wavelength (Å)	0.71073	0.71073	0.71073	0.71073
CCDC No.	978032	978031	953727	978033
Solvent system	Dichloromethane	Dichloromethane	Dichloromethane	Dichloromethane
Morphology	Plate	Plate	Block	Plate
Crystal System	Monoclinic	Orthorhombic	Orthorhombic	Orthorhombic
Space Group	$P2_1/c$	$Fdd2$	$Pccn$	$P2_12_12_1$
a (Å)	4.3357(3)	24.513(2)	25.121(3)	6.5803(6)
b (Å)	21.5407(16)	42.263(4)	8.1738(11)	6.6436(6)
c (Å)	11.2435(9)	4.1537(3)	12.8146(17)	32.419(3)
α (^{0})	90	90	90	90
β (^{0})	99.669(2)	90	90	90
γ (^{0})	90	90	90	90
Volume (Å^3)	1035.16(13)	4303.2(7)	2631.3(6)	1417.3(2)
Z/Z'	4/1	16/1	8	4
ρ (g/cm^3)	1.516	1.748	1.682	1.627
μ (mm^{-1})	0.305	3.483	2.862	2.661

F (000)	488	2240	1328	696
$\theta_{min, max}$	2.64, 24.51	3.32, 27.44	2.62, 24.98	2.51, 28.28
$h_{min,max}$;$k_{min,max}$; $l_{min,max}$	-5, 4; 0, 25; 0, 13	-31, 31; -54, 54; -5, 5	-29, 29; -9, 8; -15, 15	-8, 8; -8, 8; -37, 43
Treatment of hydrogen	Fixed	Fixed	Fixed	Fixed
No. unique/ observed reflections.	1713/1477	2409/ 1981	2310/1919	3504/2862
No. of parameters	145	145	181	191
R_all, R_obs	0.0404, 0.0326	0.0513, 0.0325	0.0797, 0.0694	0.0534, 0.0370
wR_2_all, wR_2_obs	0.0935, 0.0876	0.0594, 0.0555	0.2041, 0.1959	0.0802, 0.0760
$\Delta\rho_{min,max}$(eÅ^{-3})	-0.178, 0.202	-0.272, 0.265	3.764, -0.594	-0.464, 0.690
GooF	1.106	1.065	1.058	1.062

Figure 51. ORTEP diagram for compounds **1**

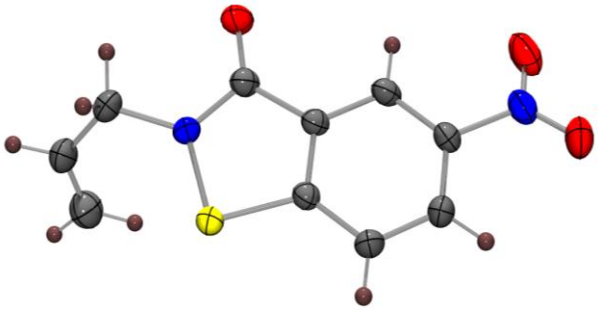


Figure 52. ORTEP diagram for compound **2**

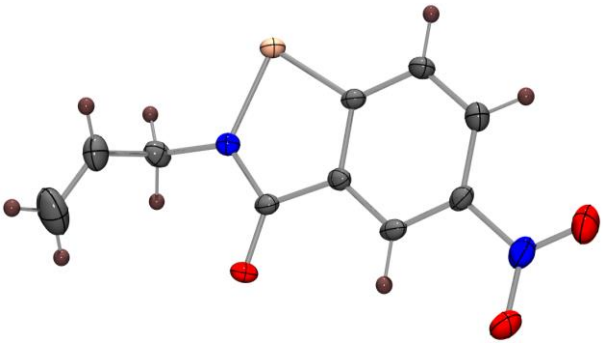


Figure 53. ORTEP diagram for compound **3**

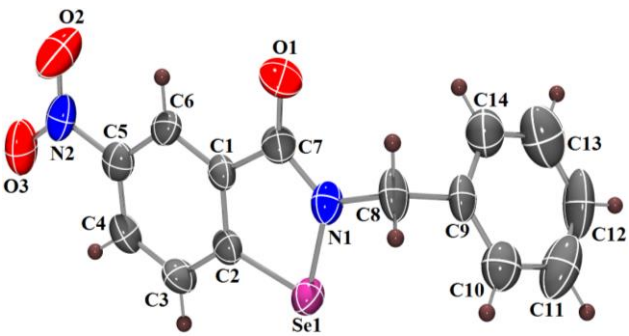
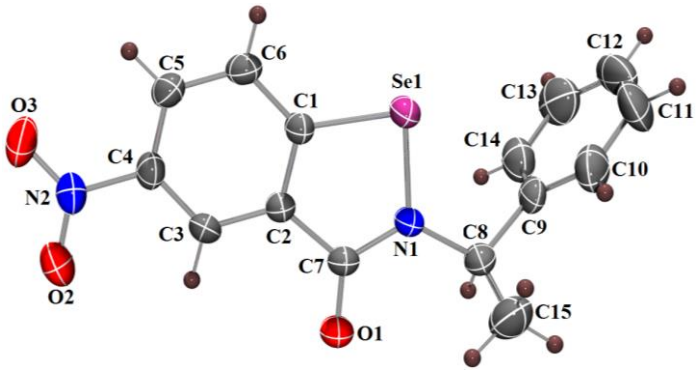


Figure 54. ORTEP diagram for compound **4**



References

1. B. S. Bhakuni, S. J. Balkrishna, A. Kumar, S. Kumar. *Tetrahedron Lett.*, 2012, **53**, 1354-1357.
2. S. J. Balkrishna, B. S. Bhakuni, D. Chopra, S. Kumar. *Org. Lett*., 2010, **12**, 5394-5397
3. S. J. Balkrishna, C. D. Prasad, P. Panini, M. R. Detty, D. Chopra, S. Kumar. *J. Org. Chem.*, 2012, **77**, 9541-9552.