Supporting Information for:

Selenorhodamine Photosensitizers for Photodynamic Therapy of P-glycoprotein-expressing Cells

Jacqueline E. Hill, Michelle K. Linder, Kellie S. Davies, Geri A. Sawada, Janet Morgan, Tymish Y. Ohulchanskyy, and Michael R. Detty*

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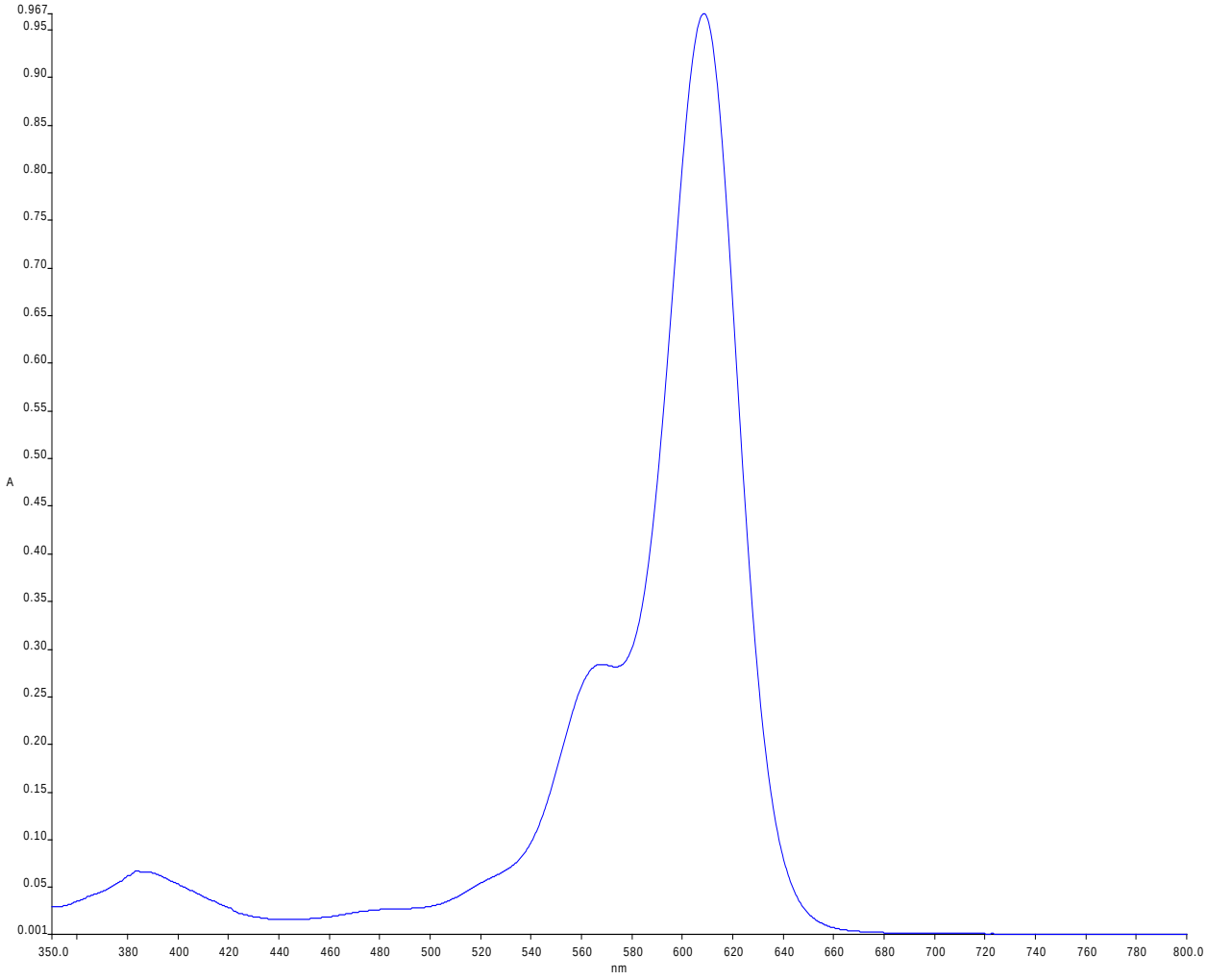
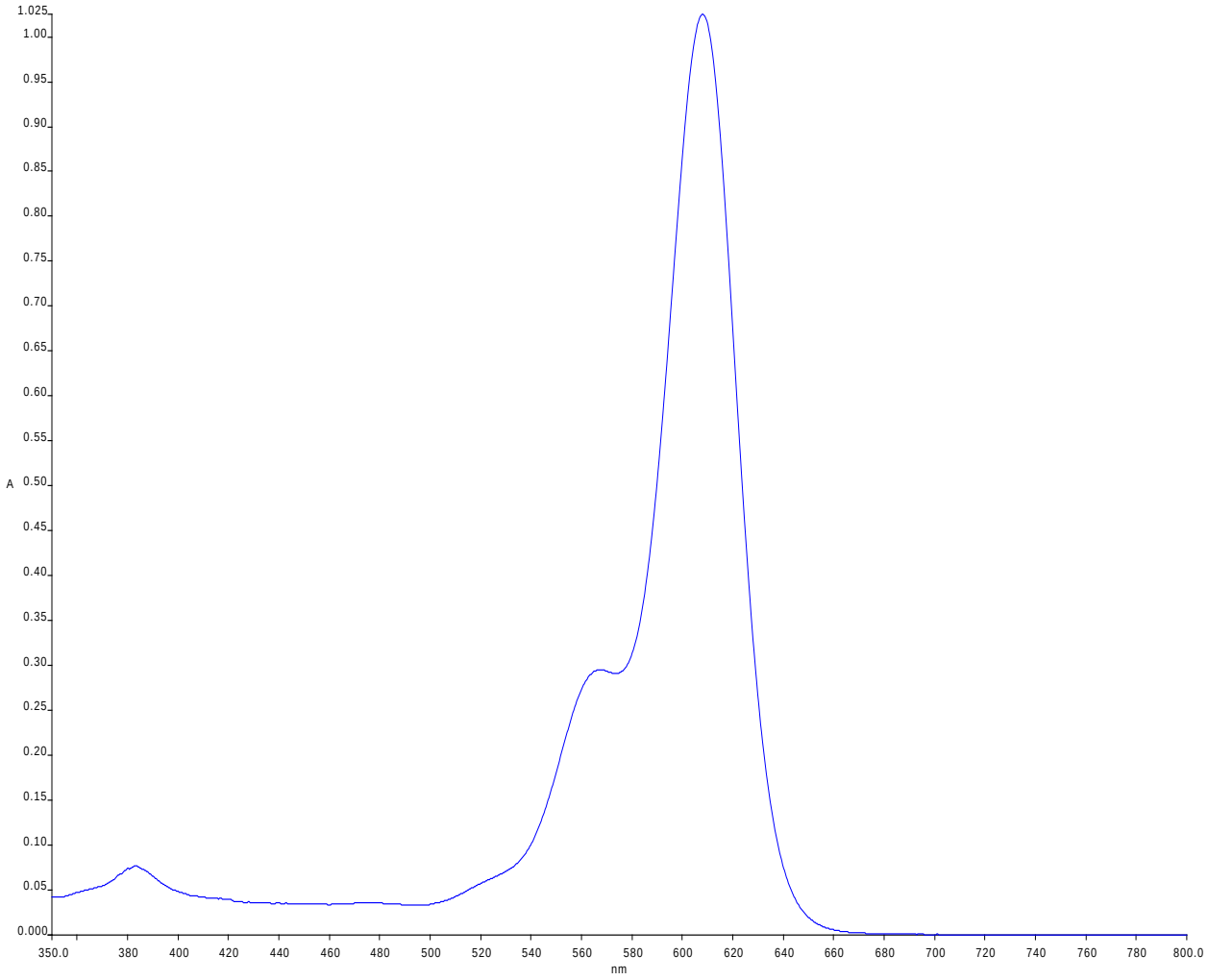
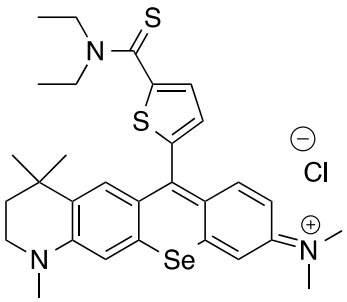


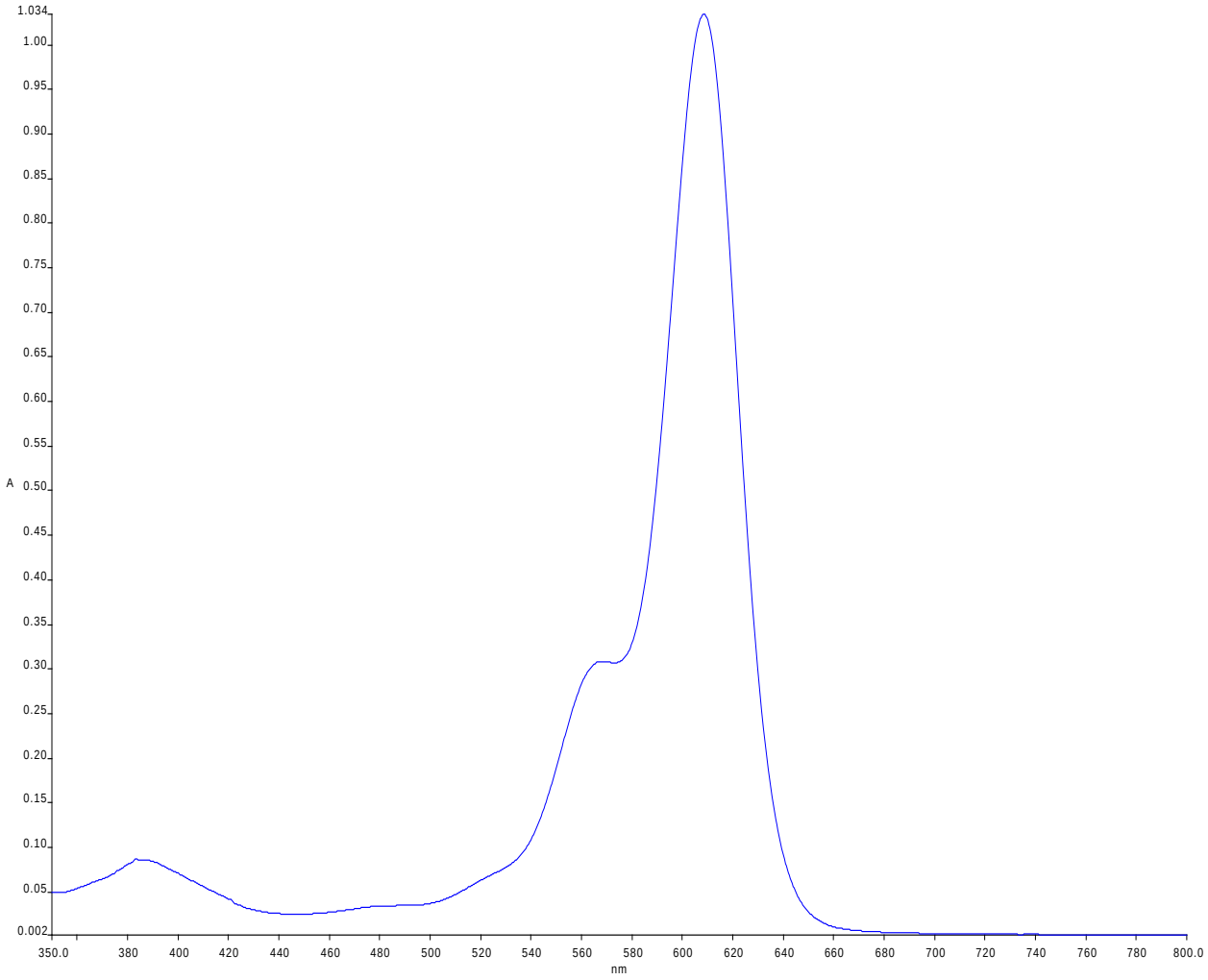
Figure S1a. Electronic absorption spectrum of **9-Se** in MeOH.

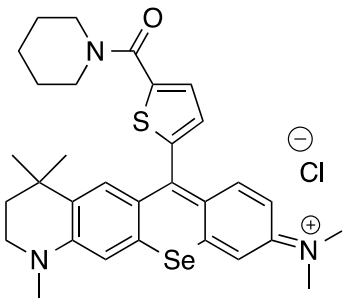




10-Se
I-JH-066
608.23 nm 1.0246

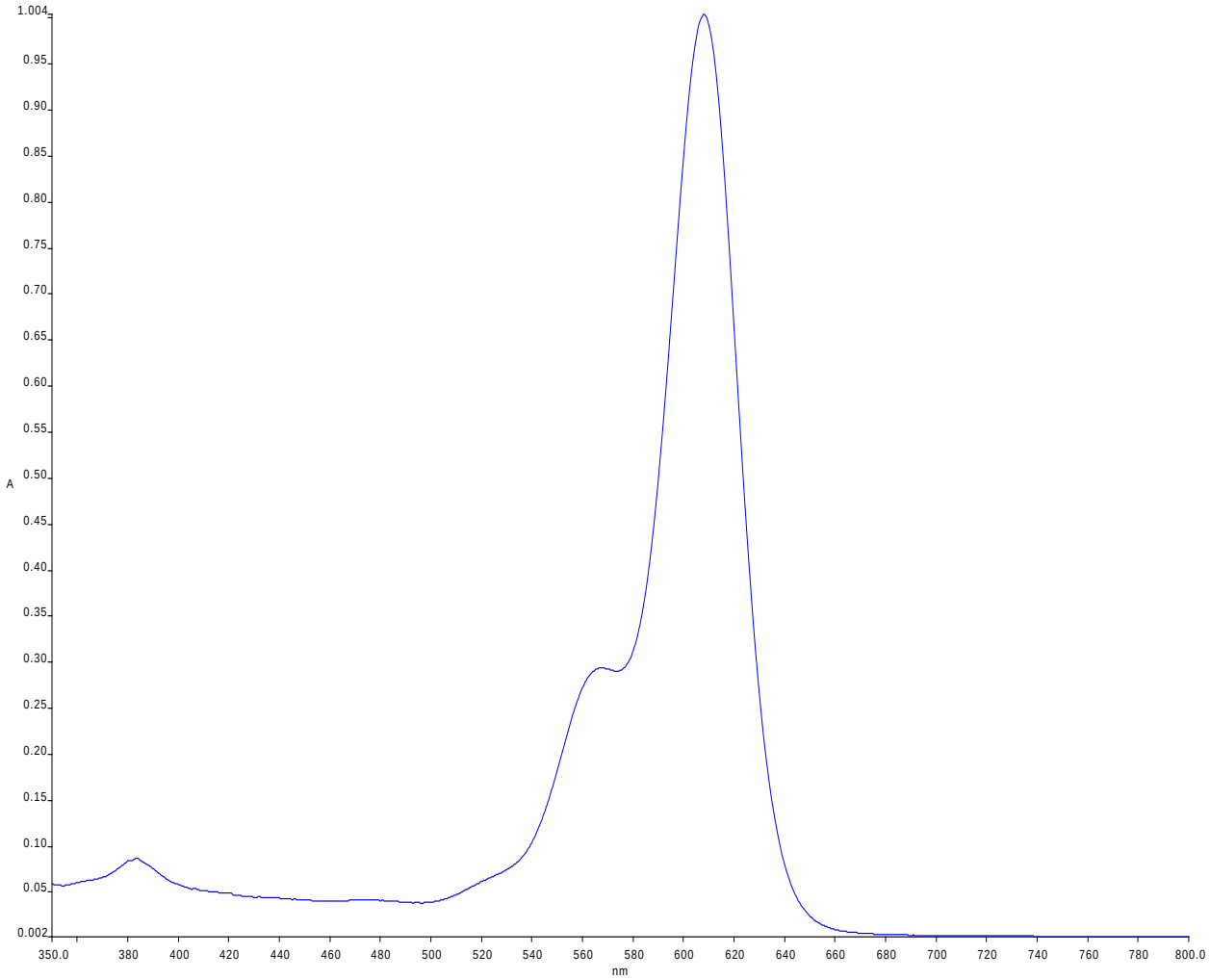
Figure S1b. Electronic absorption spectrum of **10-Se** in MeOH.

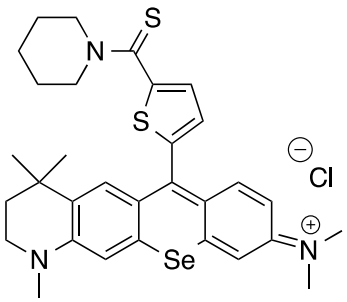




11-Se
I-JH-174
608.23 nm 1.0342

Figure S1c. Electronic absorption spectrum of **11-Se** in MeOH.





12-Se

I-JH-072

608.23 nm 1.0036

Figure S1d. Electronic absorption spectrum of **12-Se** in MeOH.

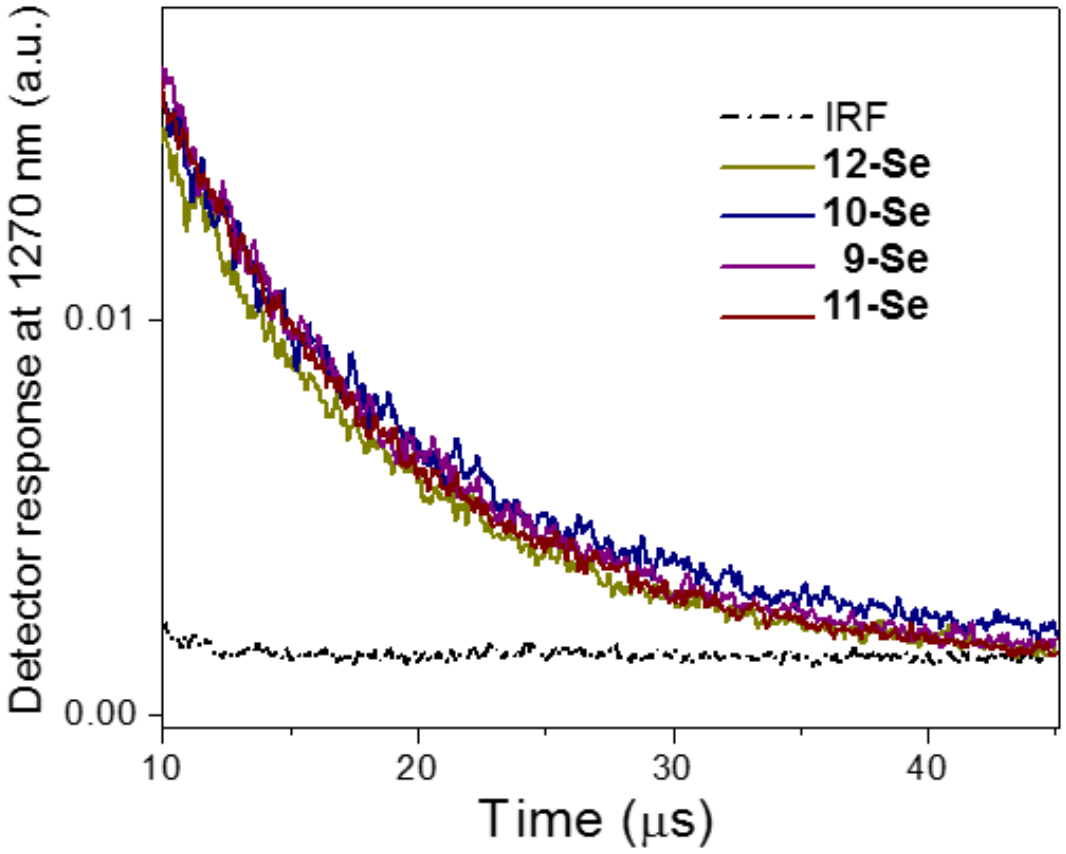


Figure S2. Decays of luminescence from $^1O_2$ sensitized by compounds **15b-18b** used for determination of $\Phi(^1O_2)$. Signal obtained from air-saturated MeOH in the cuvette was used as the instrument response function (IRF).

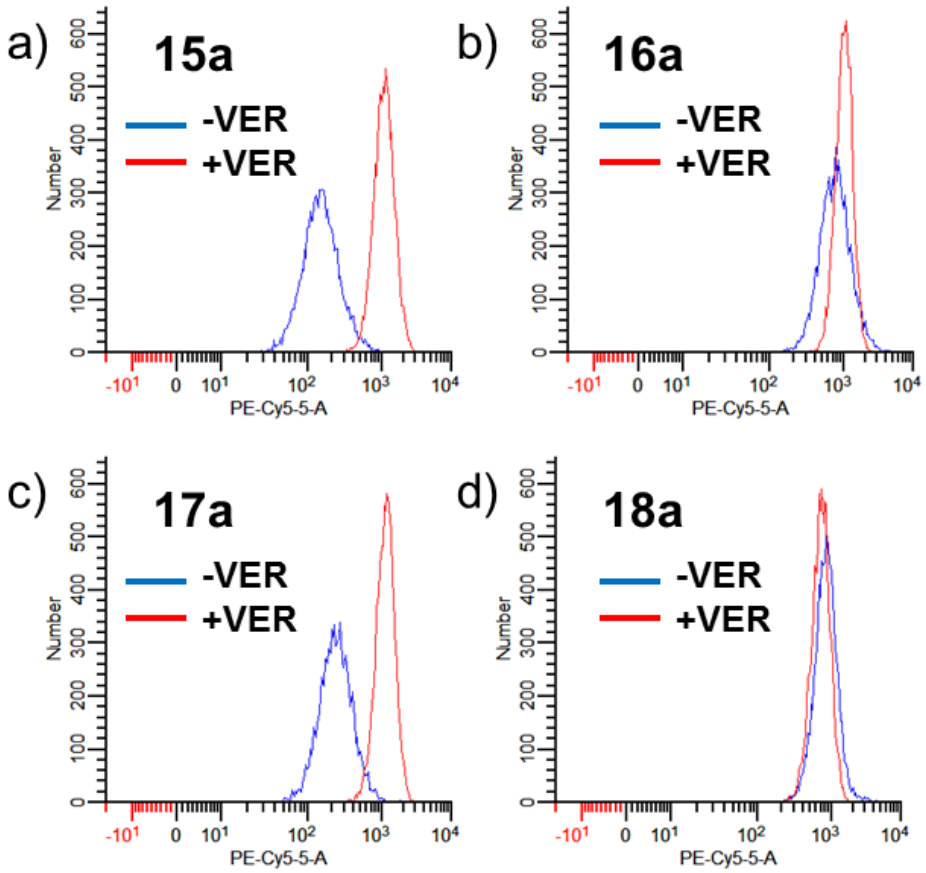


Figure S3. Flow cytometry data for Colo-26 cells incubated 1 h with 2×10^{-7} M **15a-18a** and with or without 1×10^{-4} M verapamil (VER). The histograms show the shift in fluorescence from dye alone (blue) and dye plus verapamil (red).

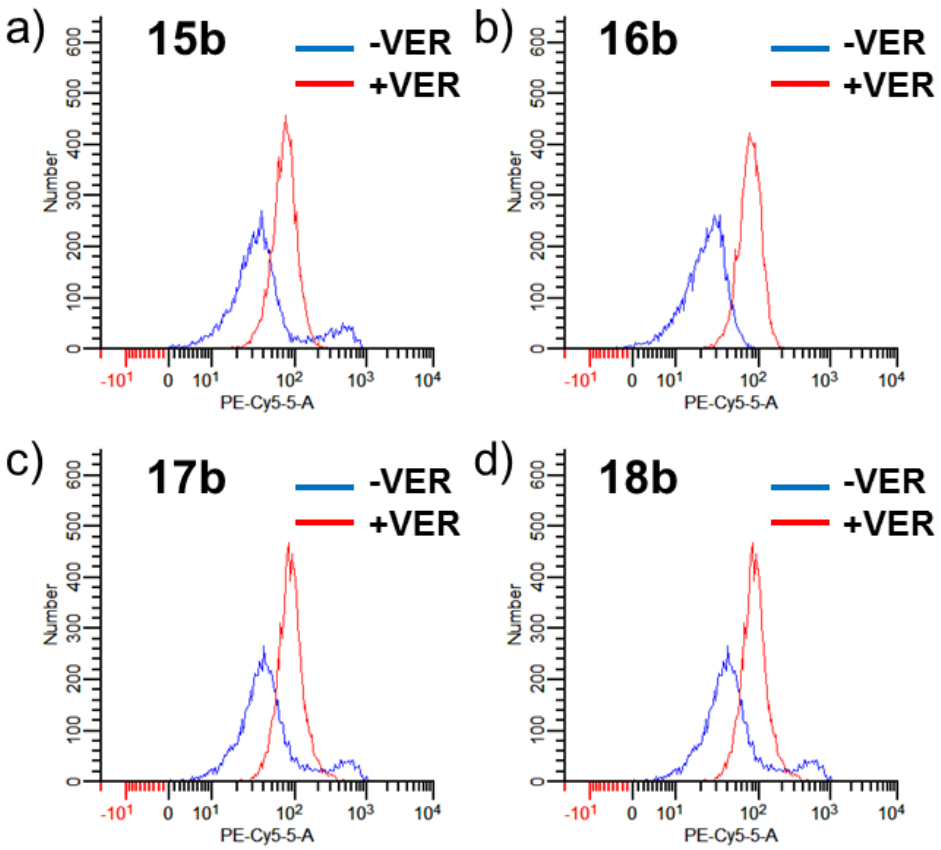


Figure S4. Flow cytometry data for Colo-26 cells incubated 1 h with 2×10^{-7} M **15b-18b** and with or without 1×10^{-4} M verapamil (VER). The histograms show the shift in fluorescence from dye alone (blue) and dye plus verapamil (red).

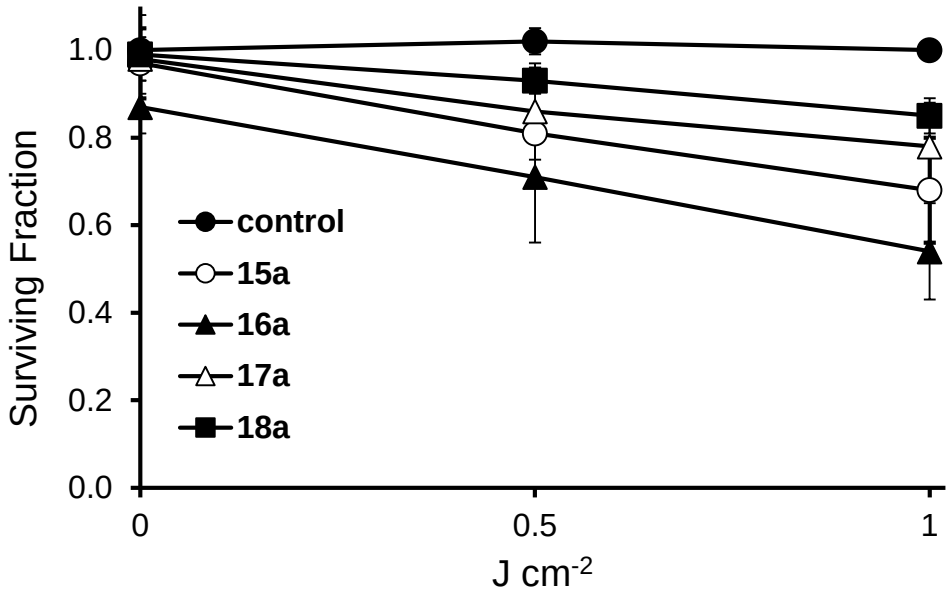


Figure S5. Phototoxicity of 0.5 μM **15a-18a** toward Colo-26 cells with 0.5 J cm^{-2} or 1.0 J cm^{-2} of 350-700-nm light. Error bars are ± SD.

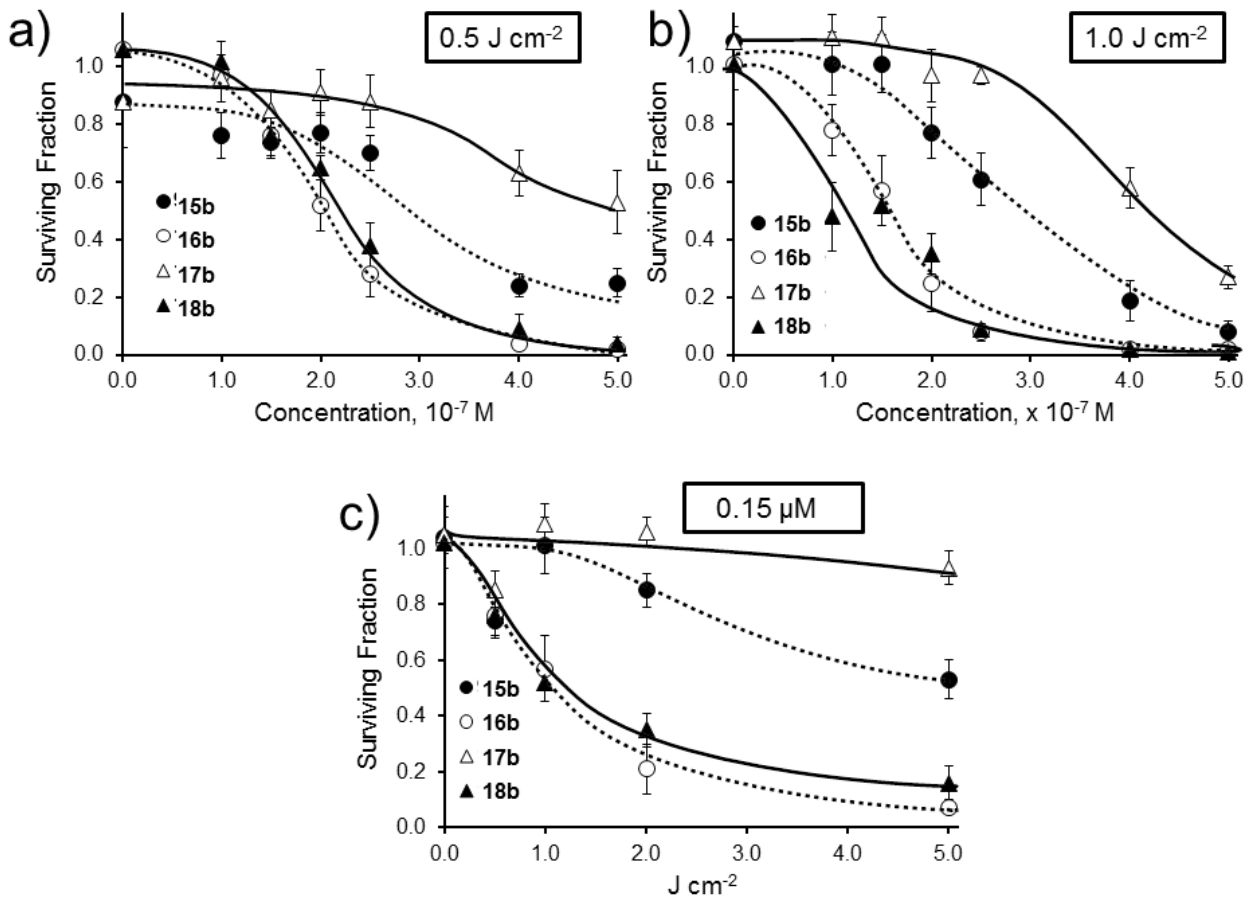


Figure S6. Phototoxicity of **15b-18b** toward Colo-6 cells with irradiation from a tunable dye laser. Irradiation at 611 ± 2 nm for **15b** and **17b** and 613 ± 2 nm for **16b** and **18b** delivered at 3.2 mW cm^{-2} for varying selenorhodamine concentration and a) 0.5 J cm^{-2} of light and b) 1.0 J cm^{-2} of light and c) for 0.15 μM **15b-18b** and varying light doses from 0 to 5 J cm^{-2}. Values of EC_{50} were determined by sigmoidal dose-response (variable slope) analysis. Error bars are ± SD.

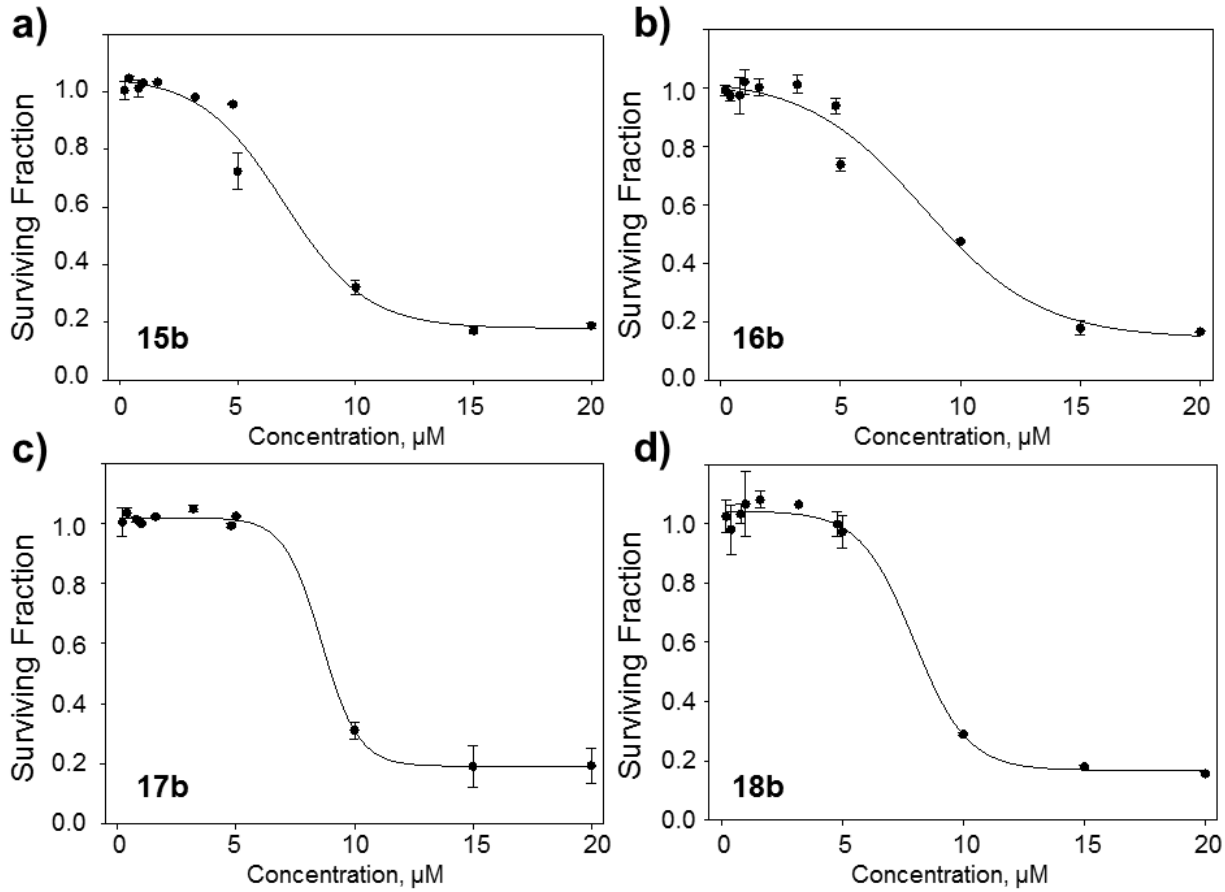


Figure S7. Dark toxicity of **15b-18b** toward Colo-26 cells. Error bars are ± SD. Values of LD_{50} were determined by a sigmoidal dose-response (variable slope) analysis.

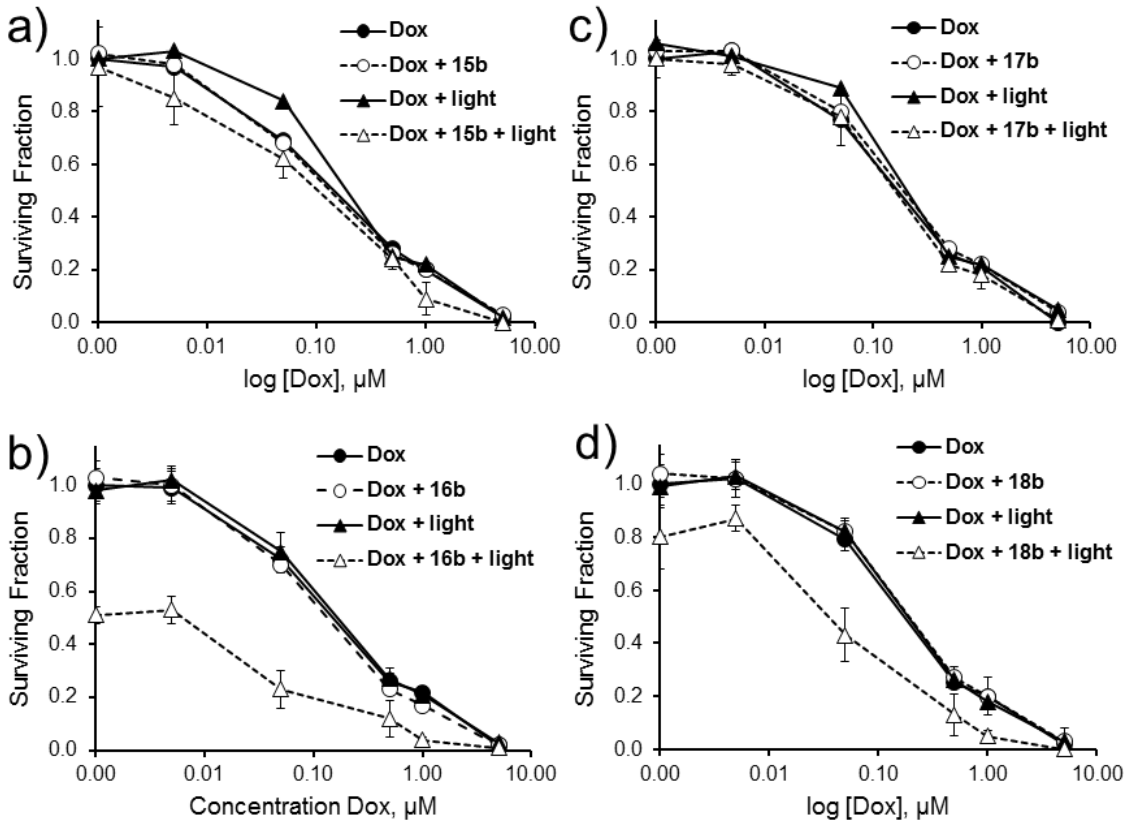


Figure S8. Combination treatment of Colo-26 cells with various concentrations of doxorubicin (Dox) alone or in combination with a) **15b** (0.15 μM), b) **16b** (0.15 μM), c) **17b** (0.15 μM), or d) **18b** (0.15 μM) in the dark or with 1.0 J cm^{-2} of 611-nm (for **16b**and **18b**) or 613-nm light (for **15-b** or **17b**). Values are the mean of six replicates. Error bars are ± SD.

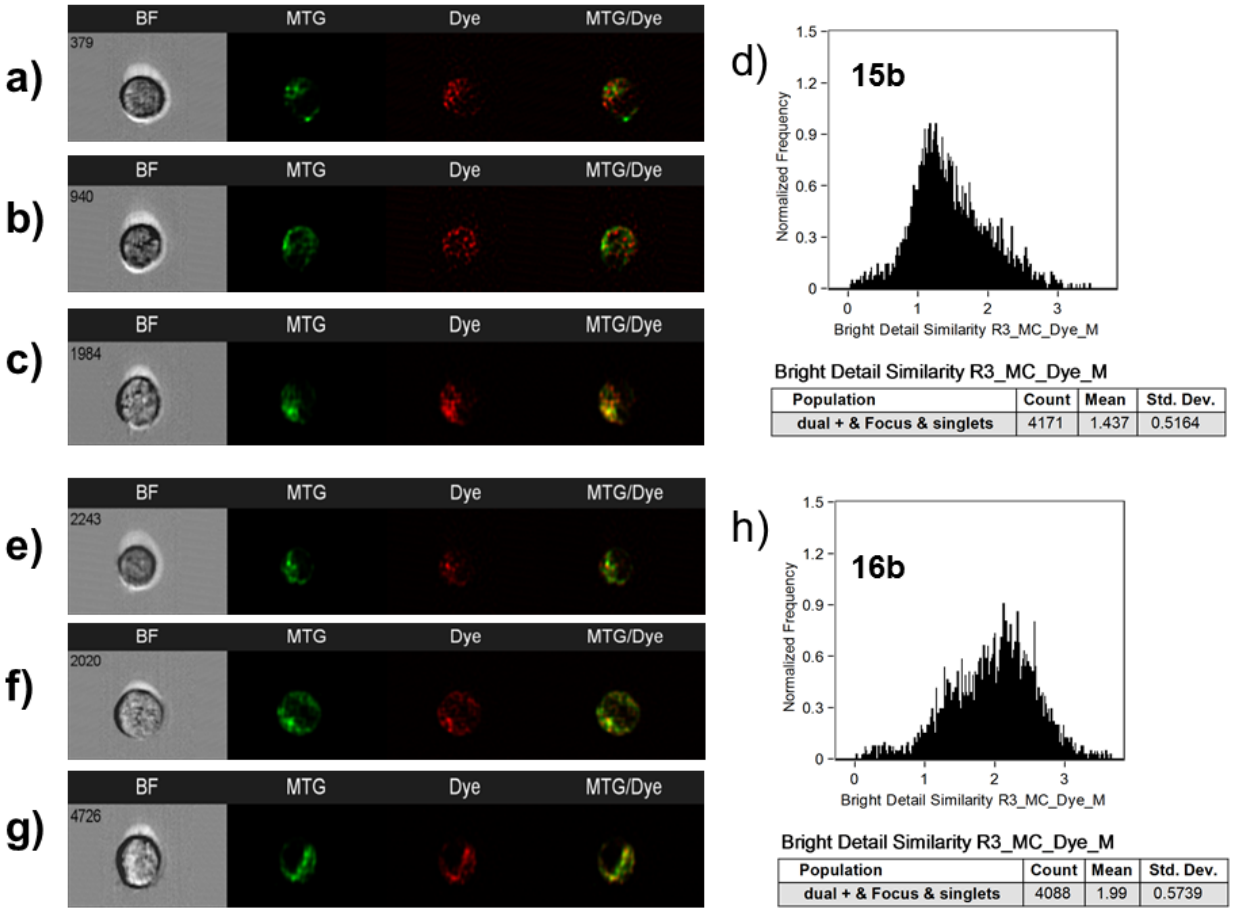
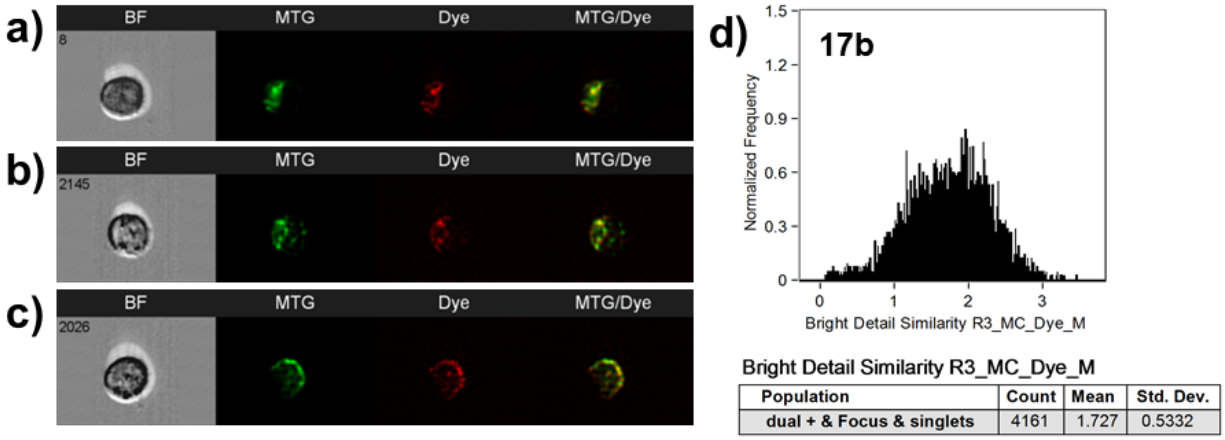


Figure S9. Representative examples of MTG/**15**-stained cells as a bright field image (BF), MTG fluorescence (MTG), **15b** fluorescence (Dye), and a merged image of MTG/**15b** fluorescence (MTG/Dye) for cells with a) low similarity, b) intermediate similarity, and c) high similarity. d) A histogram of the pixel-by-pixel statistical analysis of each cell (n = 4200) analyzed, in which the y-axis is number of cells and the x-axis is the similarity coefficient between MTG and **15b**.

Representative examples of MTG/**16b**-stained cells as a bright field image (BF), MTG fluorescence (MTG), **10-Se** fluorescence (Dye), and a merged image of MTG/**16b** fluorescence (MTG/Dye) for cells with e) low similarity, f) intermediate similarity, and g) high similarity. h) A histogram of the pixel-by-pixel statistical analysis of each cell (n = 4100) analyzed, in which the y-axis is number of cells and the x-axis is the similarity coefficient between MTG and **16b**.



Bright Detail Similarity R3_MC_Dye_M

Population	Count	Mean	Std. Dev.
dual + & Focus & singlets	4161	1.727	0.5332

Figure S10. Representative examples of MTG/**17b**-stained cells as a bright field image (BF), MTG fluorescence (MTG), **17b** fluorescence (Dye), and a merged image of MTG/**17b** fluorescence (MTG/Dye) for cells with a) low similarity, b) intermediate similarity, and c) high similarity. d) A histogram of the pixel-by-pixel statistical analysis of each cell (n = 4200) analyzed, in which the y-axis is number of cells and the x-axis is the similarity coefficient between MTG and **17b**.

I-JH-045

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

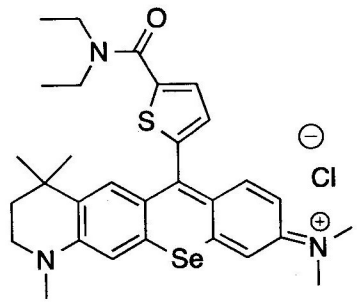
Sample directory:

FidFile: 1-JH-045

Pulse Sequence: PROTON (s2pul)
Solvent: cd3cn
Data collected on: Aug 15 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.9010665 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



9-Se

1-JH-045

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

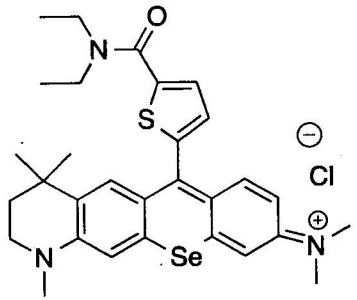
Sample directory:

FidFile: 1-JH-045-2

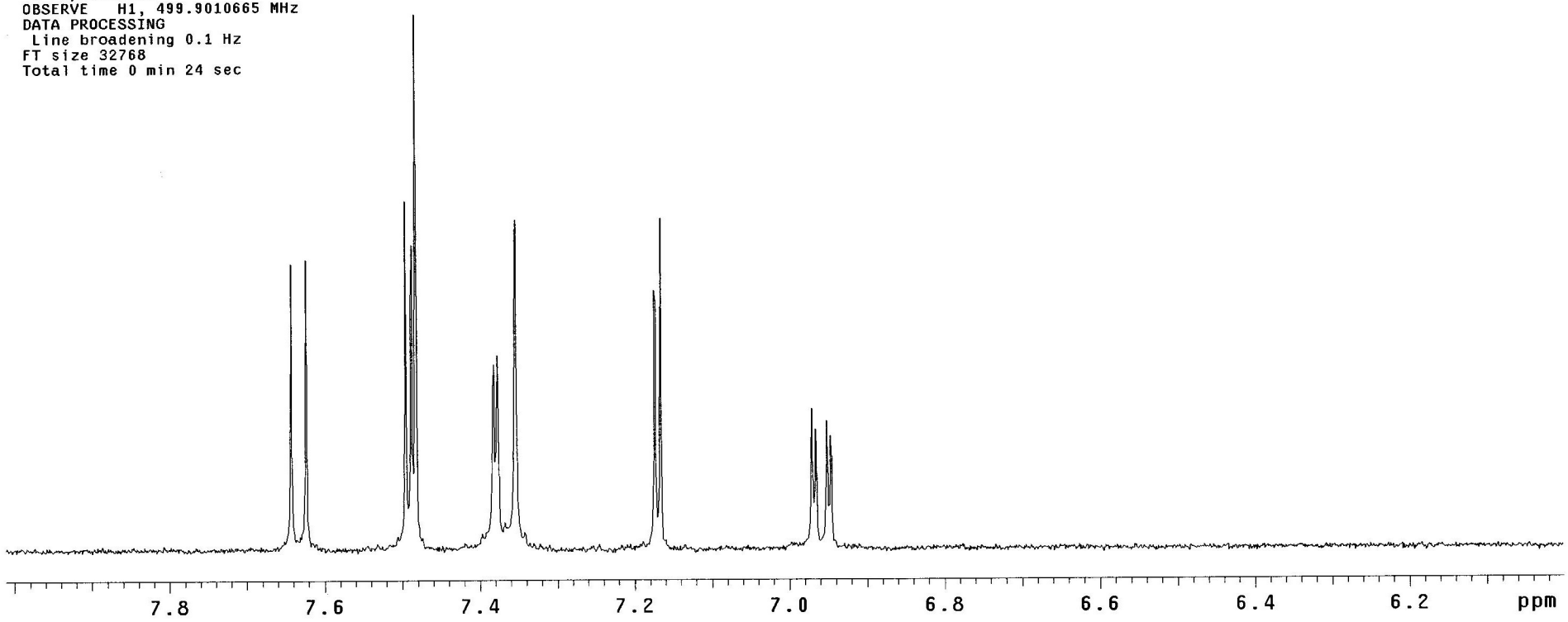
Pulse Sequence: PROTON (s2pul)
Solvent: cd3cn
Data collected on: Aug 16 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.9010665 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



9-Se



I-JH-045Cl-saltcarbon

Sample Name:

Data Collected on:
roesy.chem.buffalo.edu-mercury300
Archive directory:

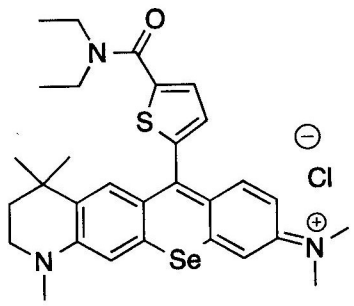
Sample directory:

FidFile: I-JH-045Cl-saltcarbon

Pulse Sequence: CARBON (s2pul)
Solvent: cdcl3
Data collected on: Jul 26 2013

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 0.868 sec
Width 18867.9 Hz
2304 repetitions
OBSERVE C13, 75.4536435 MHz
DECOUPLE H1, 300.0754430 MHz
Power 38 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 32768
Total time 536 hr, 56 min



9-Se

I-JH-045Cl-saltcarbon

Sample Name:

Data Collected on:
roesy.chem.buffalo.edu-mercury300
Archive directory:

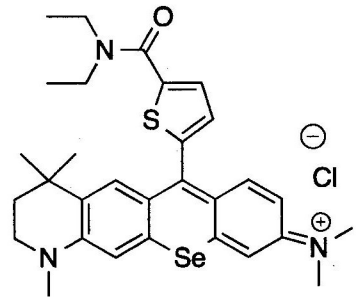
Sample directory:

FidFile: I-JH-045Cl-saltcarbon

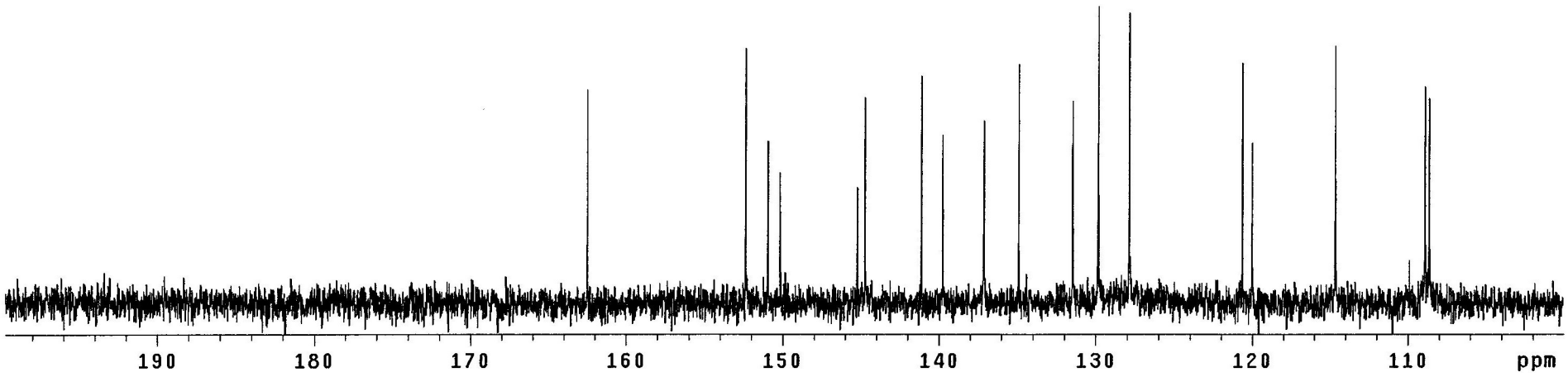
Pulse Sequence: CARBON (s2pul)
Solvent: cdcl3
Data collected on: Jul 26 2013

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 0.868 sec
Width 18867.9 Hz
2304 repetitions
OBSERVE C13, 75.4536435 MHz
DECOUPLE H1, 300.0754430 MHz
Power 38 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 32768
Total time 536 hr, 56 min



9-Se



1-JH-039pure

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

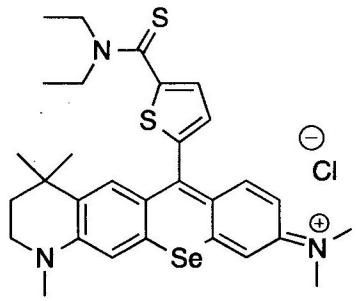
Sample directory:

FidFile: 1-JH-039pure

Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Jul 26 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993732 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



10-Se

1-JH-039pure

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

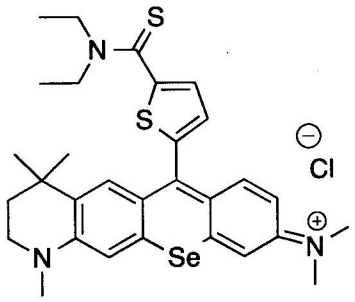
Sample directory:

FidFile: 1-JH-039pure

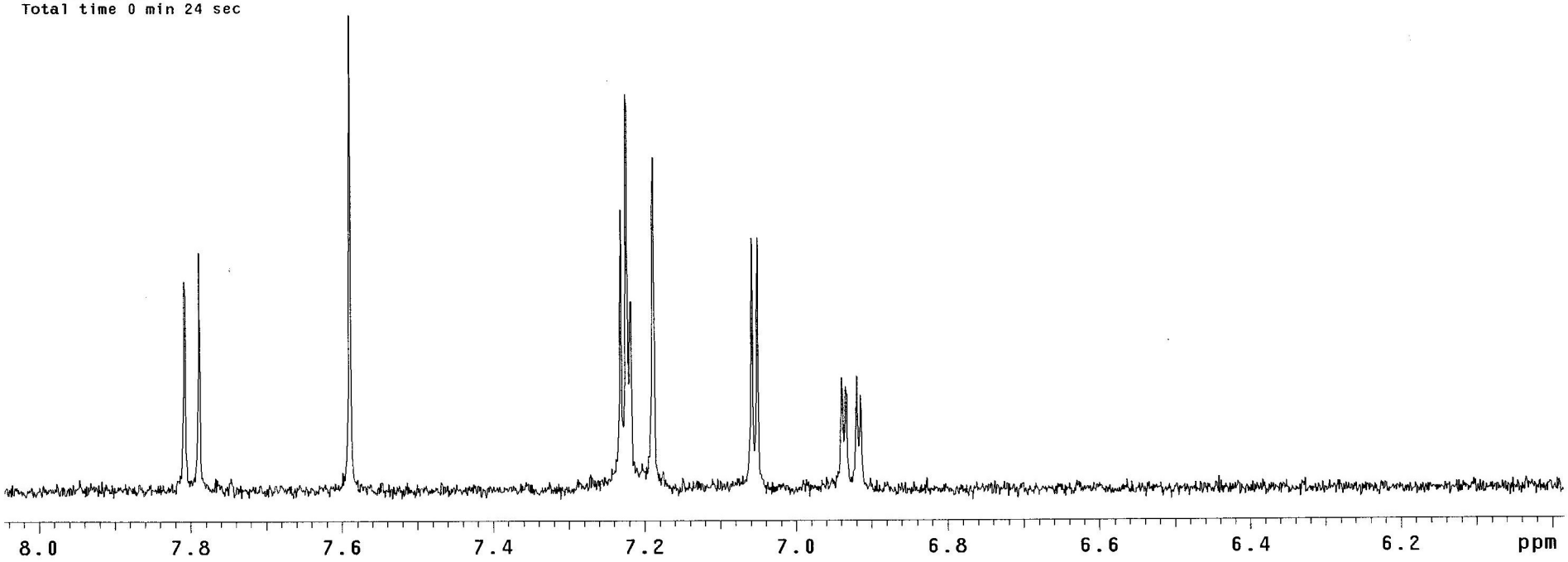
Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Jul 26 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993732 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



10-Se



1-JH-039pure

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

Sample directory:

FidFile: 1-JH-039pure

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Solvent: cd2cl2
Data collected on: Jul 26 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

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Acq. time 2.048 sec
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DATA PROCESSING
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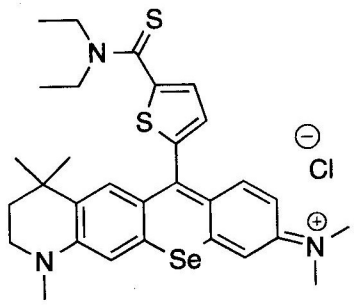
FT size 32768
Total time 0 min 24 sec



10-Se

1-JH-039carbon

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

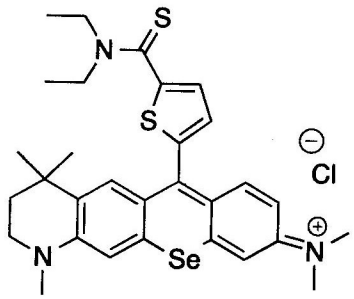
Sample directory:

FidFile: 1-JH-039carbon

Pulse Sequence: CARBON (s2pul)
Solvent: cd2cl2
Data collected on: Aug 5 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 1.042 sec
Width 31446.5 Hz
21760 repetitions
OBSERVE C13, 125.6997917 MHz
DECOUPLE H1, 499.9018501 MHz
Power 36 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.1 Hz
FT size 65536
Total time 569 hr, 43 min



10-Se

1-JH-039carbon

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

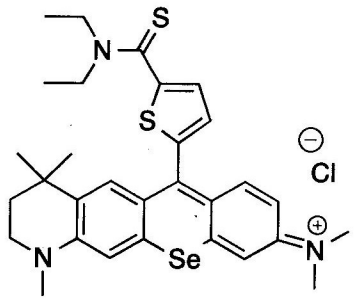
Sample directory:

FidFile: 1-JH-039carbon

Pulse Sequence: CARBON (s2pul)
Solvent: cd2cl2
Data collected on: Aug 5 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 1.042 sec
Width 31446.5 Hz
21760 repetitions
OBSERVE C13, 125.6997917 MHz
DECOUPLE H1, 499.9018501 MHz
Power 36 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.1 Hz
FT size 65536
Total time 569 hr,.43 min



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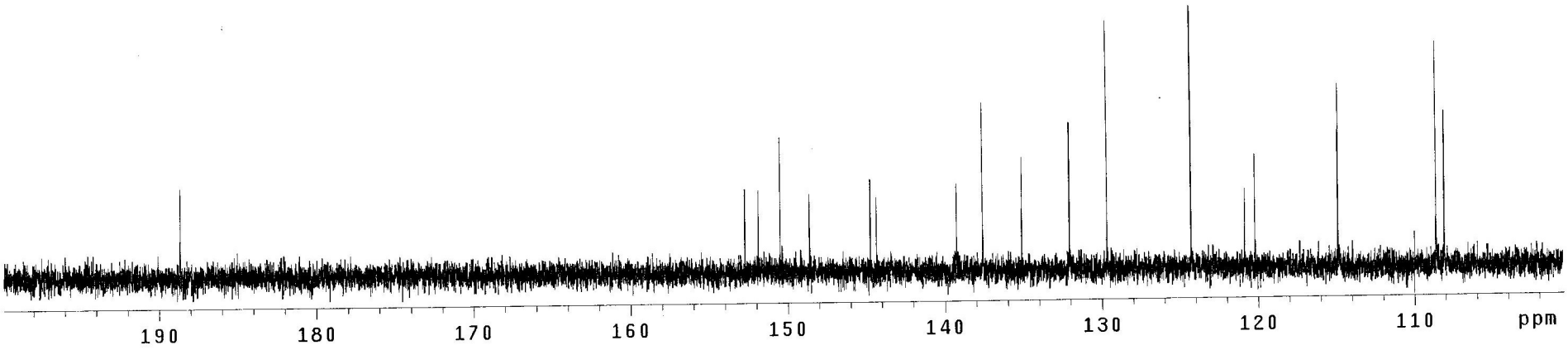

1-JH-041spot2

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

Sample directory:

FidFile: 1-JH-041spot2

Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Aug 6 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

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Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
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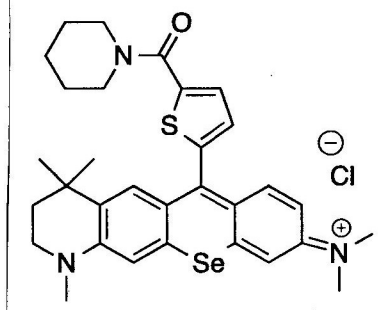
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



11-Se

1-JH-041spot2

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

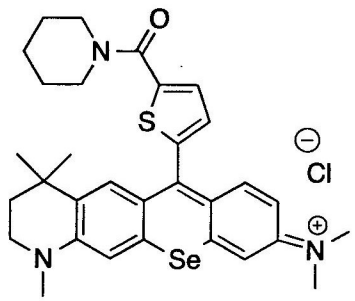
Sample directory:

FidFile: 1-JH-041spot2

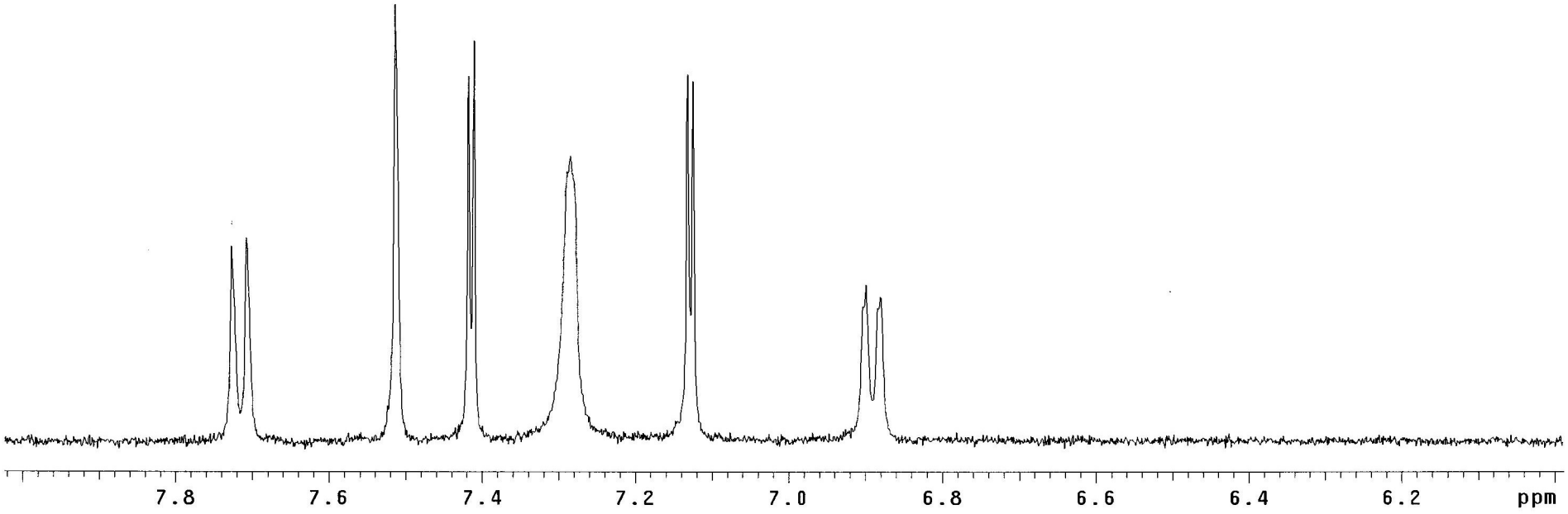
Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Aug 6 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993712 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



11-Se



1-JH-041spot2

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

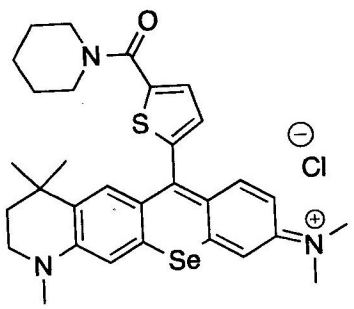
Sample directory:

FidFile: 1-JH-041spot2

Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Aug 6 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993712 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



11-Se

I-JH-041Cl-saltcarbon

Sample Name:

Data Collected on:
roesy.chem.buffalo.edu-mercury300
Archive directory:

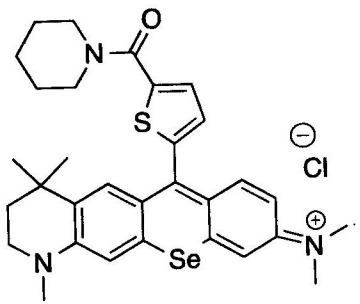
Sample directory:

FidFile: I-JH-041Cl-saltcarbon

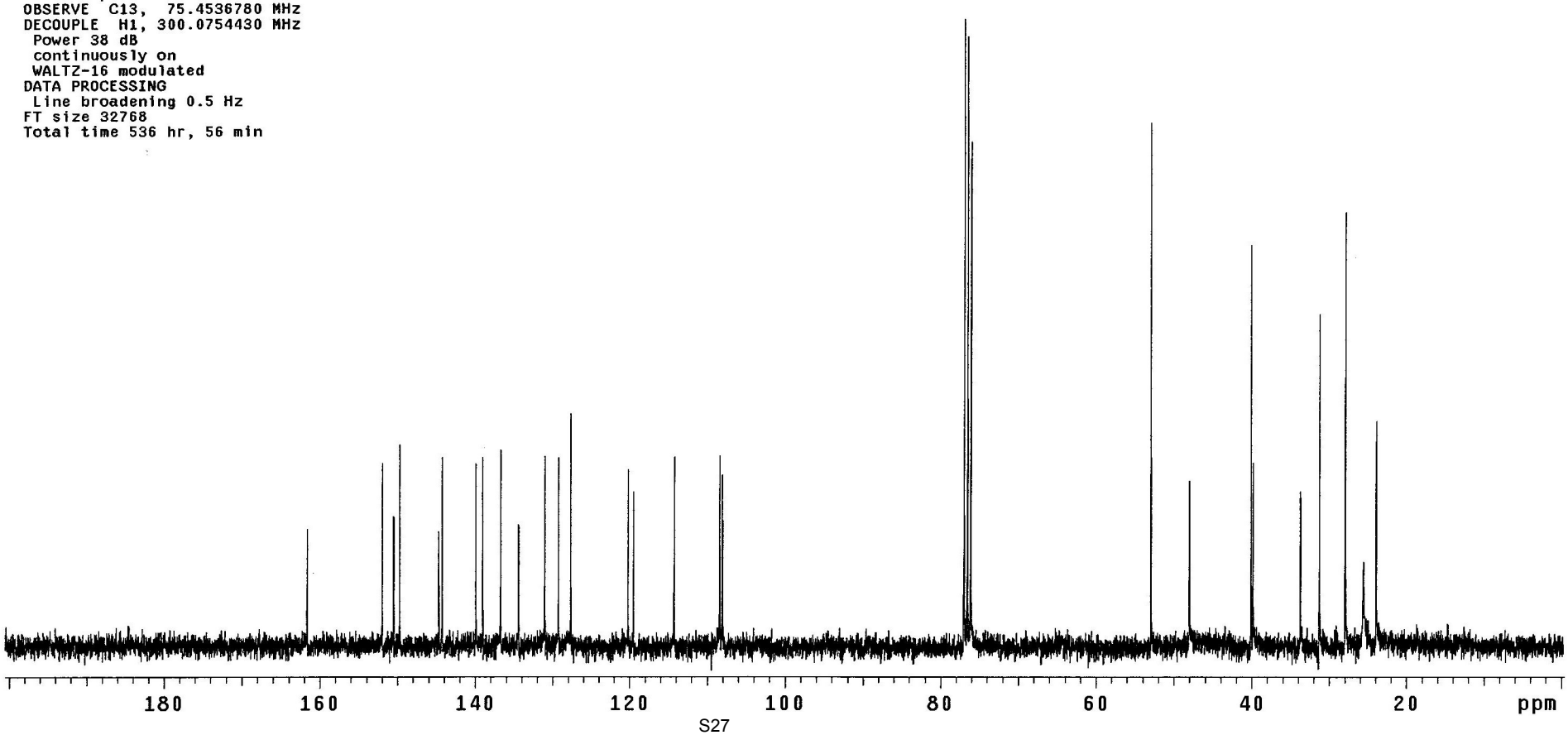
Pulse Sequence: CARBON (s2pul)
Solvent: cdcl3
Data collected on: Jul 30 2013

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 0.868 sec
Width 18867.9 Hz
2176 repetitions
OBSERVE C13, 75.4536780 MHz
DECOUPLE H1, 300.0754430 MHz
Power 38 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 32768
Total time 536 hr, 56 min



11-Se



I-JH-041Cl-saltcarbon

Sample Name:

Data Collected on:
roesy.chem.buffalo.edu-mercury300
Archive directory:

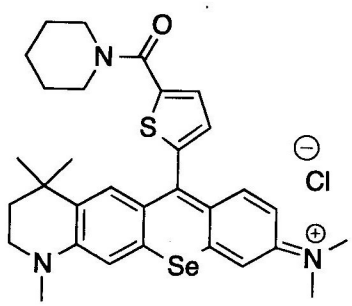
Sample directory:

FidFile: I-JH-041Cl-saltcarbon

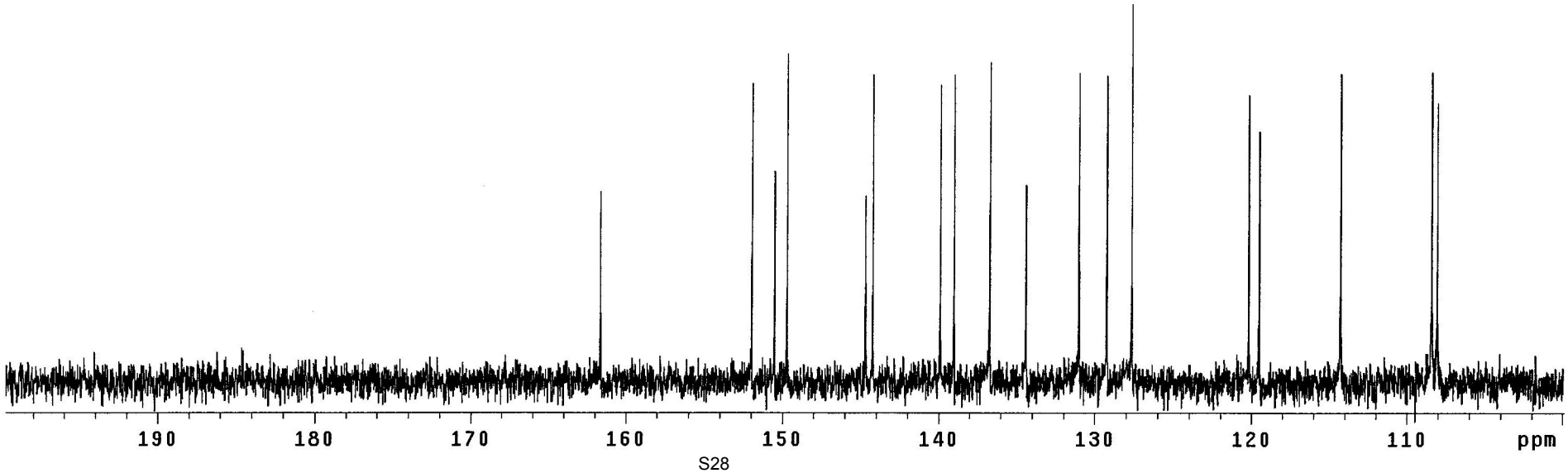
Pulse Sequence: CARBON (s2pul)
Solvent: cdcl3
Data collected on: Jul 30 2013

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 0.868 sec
Width 18867.9 Hz
2176 repetitions
OBSERVE C13, 75.4536780 MHz
DECOUPLE H1, 300.0754430 MHz
Power 38 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 32768
Total time 536 hr, 56 min



11-Se



1-JH-040

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

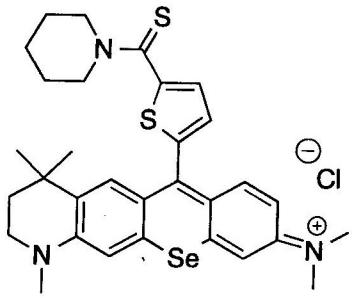
Sample directory:

FidFile: 1-JH-040

Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Jul 30 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993722 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



12-Se

1-JH-040

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

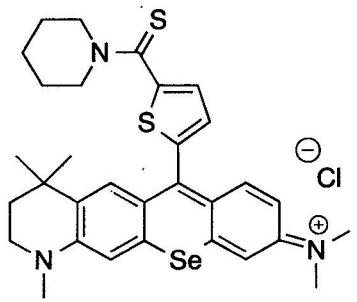
Sample directory:

FidFile: 1-JH-040

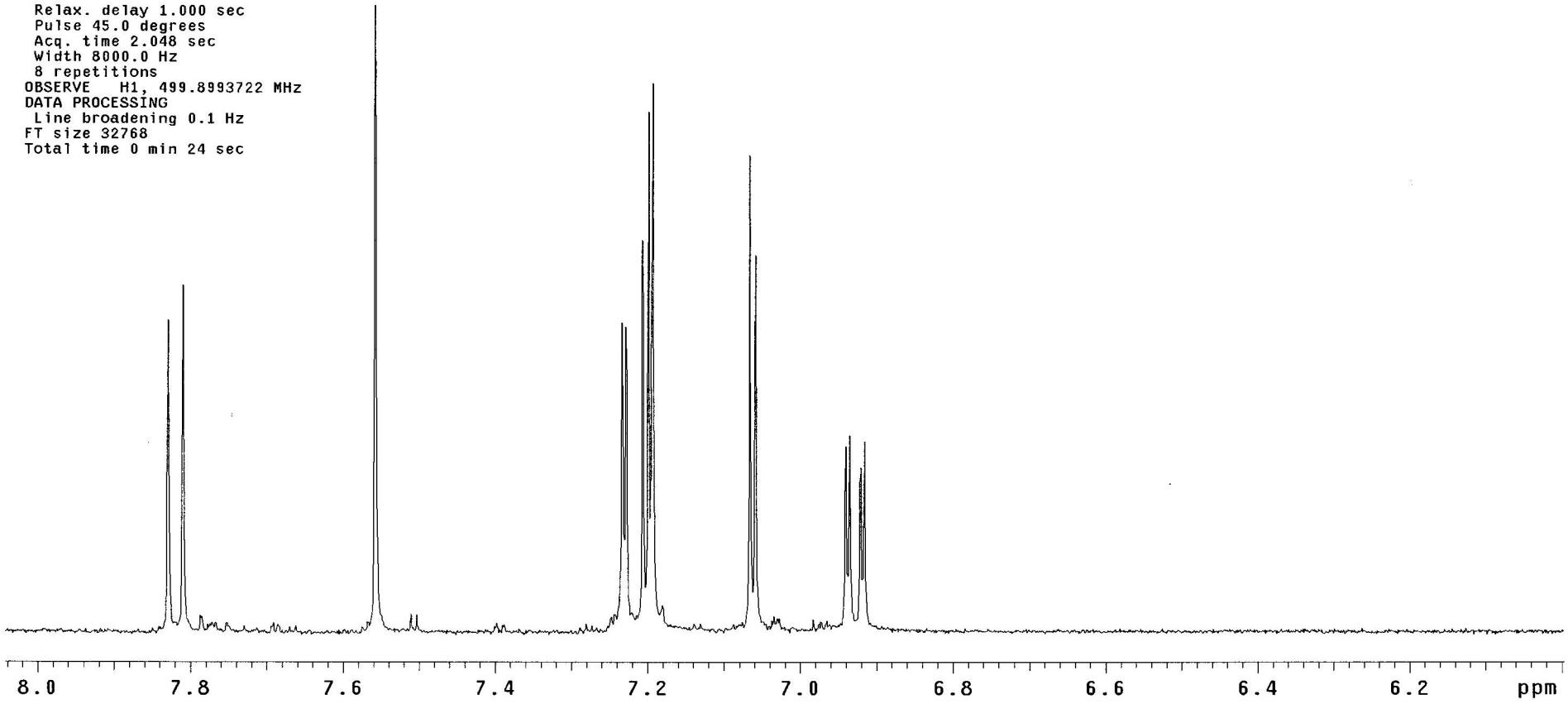
Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Jul 30 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993722 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



12-Se



1-JH-040

Sample Name:

Data Collected on:
chemnmr500.chem.buffalo.edu-inova500
Archive directory:

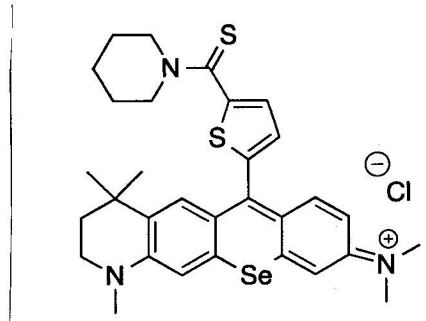
Sample directory:

FidFile: 1-JH-040

Pulse Sequence: PROTON (s2pul)
Solvent: cd2cl2
Data collected on: Jul 30 2012

Temp. 25.0 C / 298.1 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 2.048 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.8993722 MHz
DATA PROCESSING
Line broadening 0.1 Hz
FT size 32768
Total time 0 min 24 sec



12-Se

I-JH-040-carbon

Sample Name:

Data Collected on:
nmr300.chem.buffalo.edu-mercury300
Archive directory:

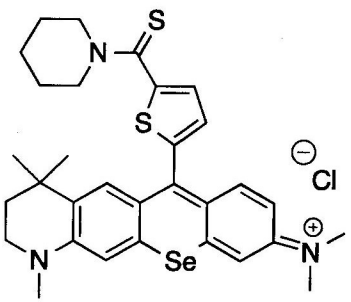
Sample directory:

FidFile: CARBON

Pulse Sequence: CARBON (s2pul)
Solvent: cd2cl2
Data collected on: May 7 2014

Temp. 23.7 C / 296.9 K
Operator: Detty

Relax. delay 1.000 sec
Pulse 45.0 degrees
Acq. time 0.868 sec
Width 18867.9 Hz
24704 repetitions
OBSERVE C13, 75.4537551 MHz
DECOUPLE H1, 300.0760191 MHz
Power 38 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 32768
Total time 536 hr, 56 min



12-Se