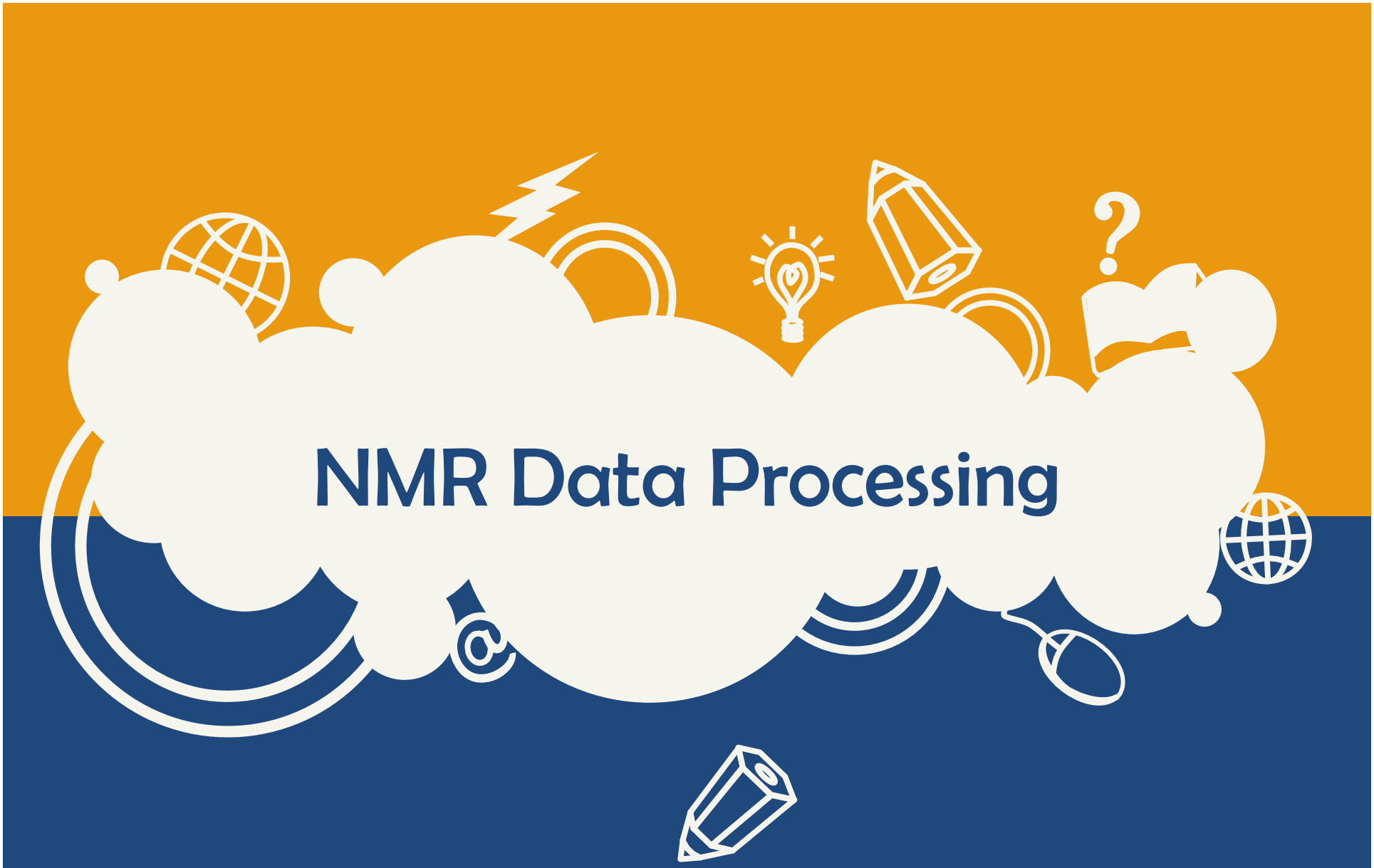
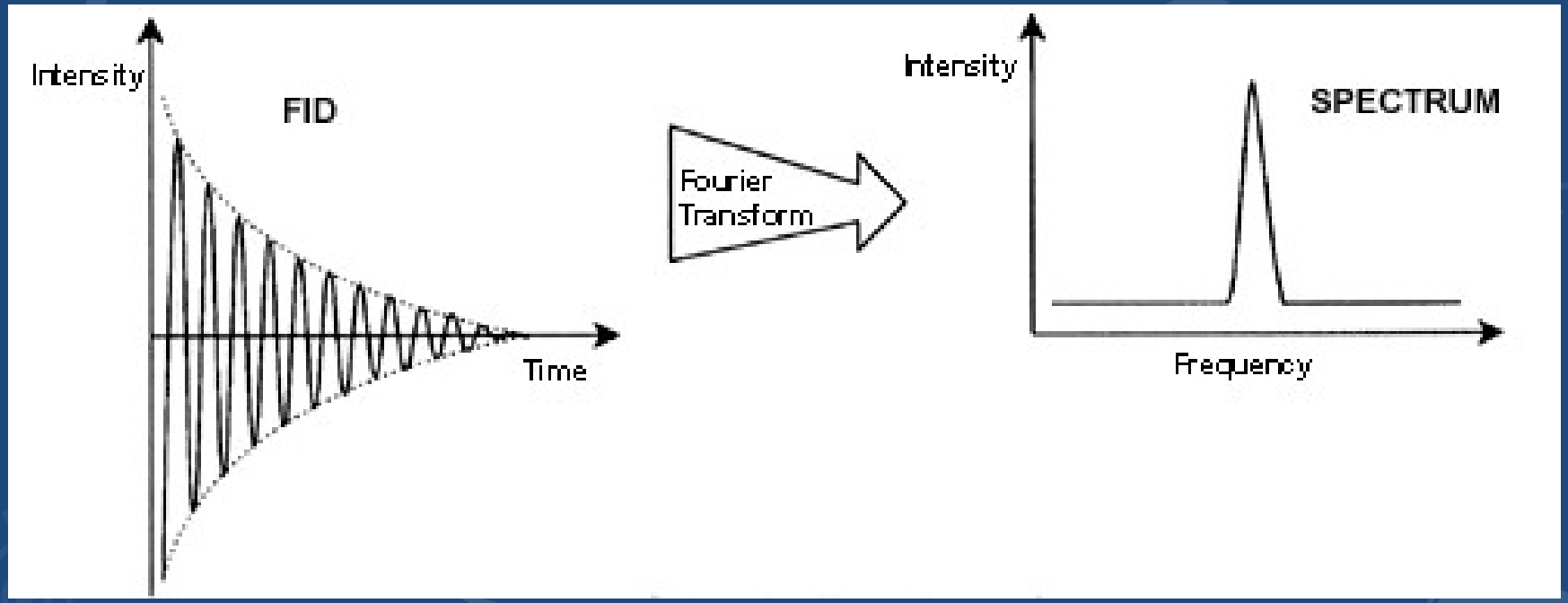


NMR Data Processing

KAIST 화학과 NMR center // 신재선

NMR spectrum



http://www2.chem.uic.edu/nmr/downloads/bruker/en-US/html/Avance%20Beginners%20Guide/en-US/18014398879323275.html

NMR processing?

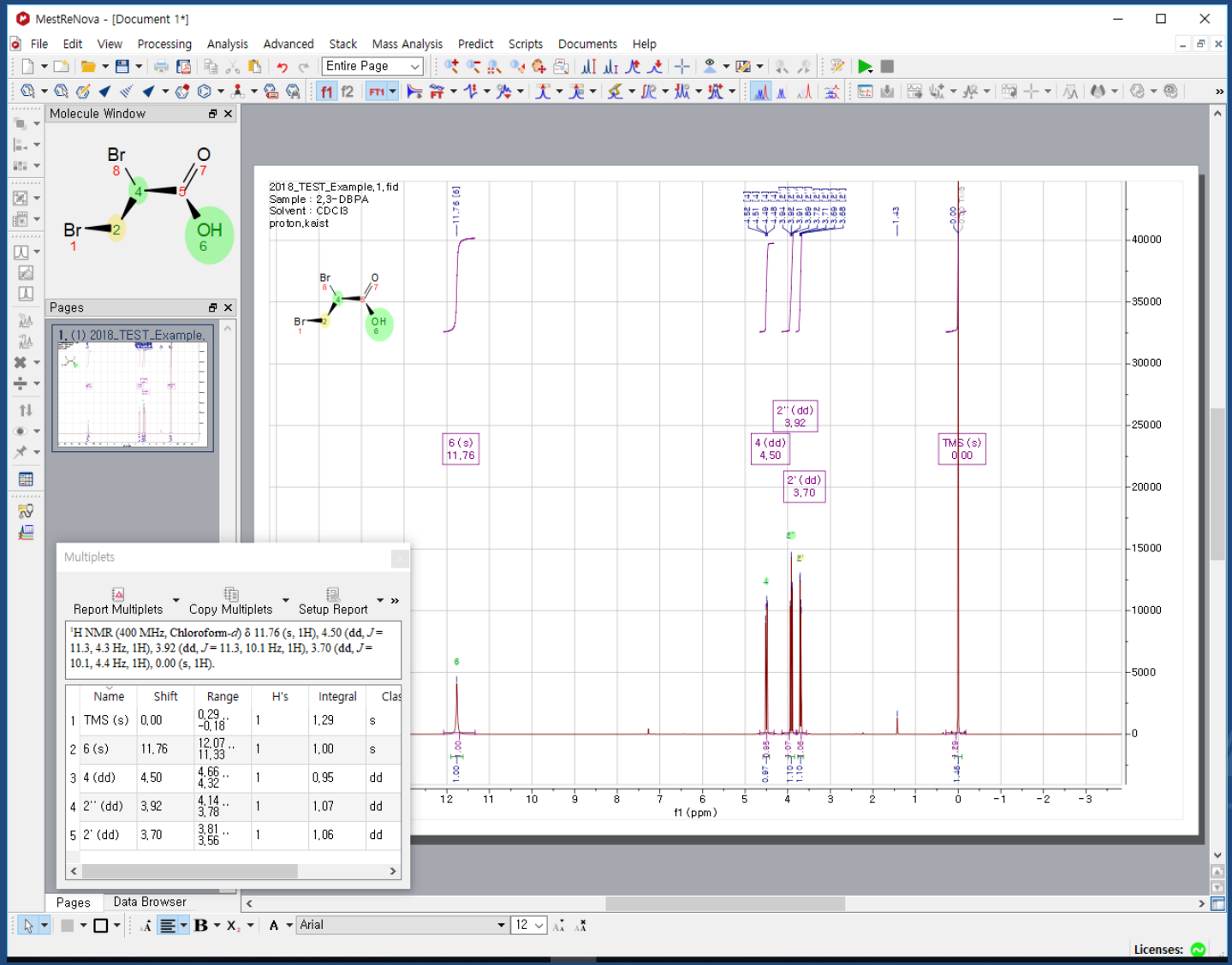
http://www.stockvault.net/photo/104139/toothless-lew

http://www.prodigitalsoftware.com/Astronomy_Tools_For_Full_Version.html

Software

- NMR manufacture
  - Topspin / VNMRJ / Delta
- Free Software
  - ACD NMR processor, SpinWorks 4, iNMR
  - Sparky, CCPN-NMR, NMRViewJ
- Commercial Software
  - Nuts, MestReNova

MestReNova

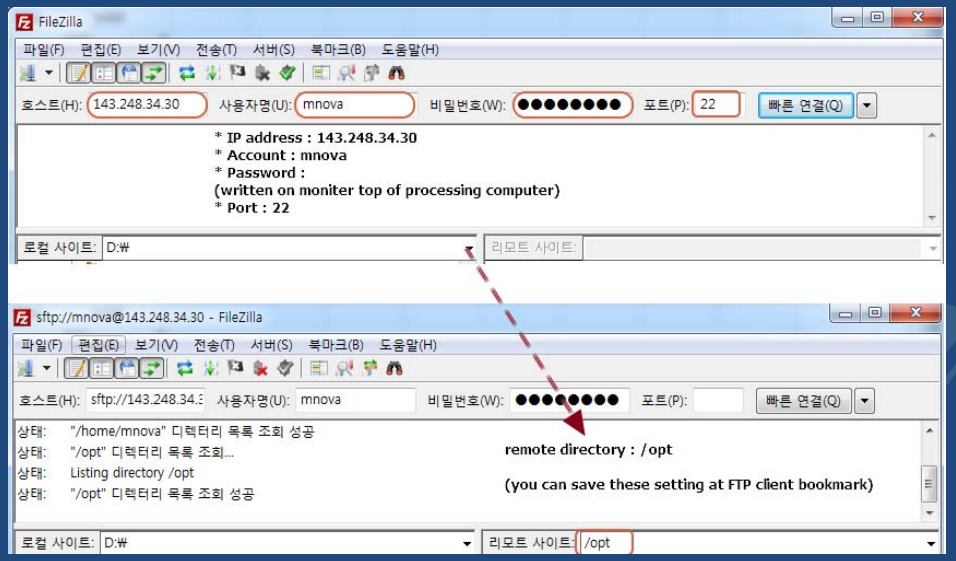

MestReNova - [Document 1*]
File Edit View Processing Analysis Advanced Stack Mass Analysis Predict Scripts Documents Help
Molecule Window
Pages
Multiplets
Report Multiplets
Copy Multiplets
Setup Report
Name Shift Range H's Integral
1 TMS (s) 0,00 0,29 .. -0,18 1 1,29 s
2 6 (s) 11,76 12,07 .. 11,33 1 1,00 s
3 4 (dd) 4,50 4,66 .. 4,32 1 0,95 dd
4 2'' (dd) 3,92 4,14 .. 3,78 1 1,07 dd
5 2' (dd) 3,70 3,81 .. 3,56 1 1,06 dd
Pages
Data Browser
Arial
Licenses:

MestReNova

- Version 14.0.1
- Campus license
  - Actually unlimited users can use in KAIST campus
  - 90 days : out of KAIST campus

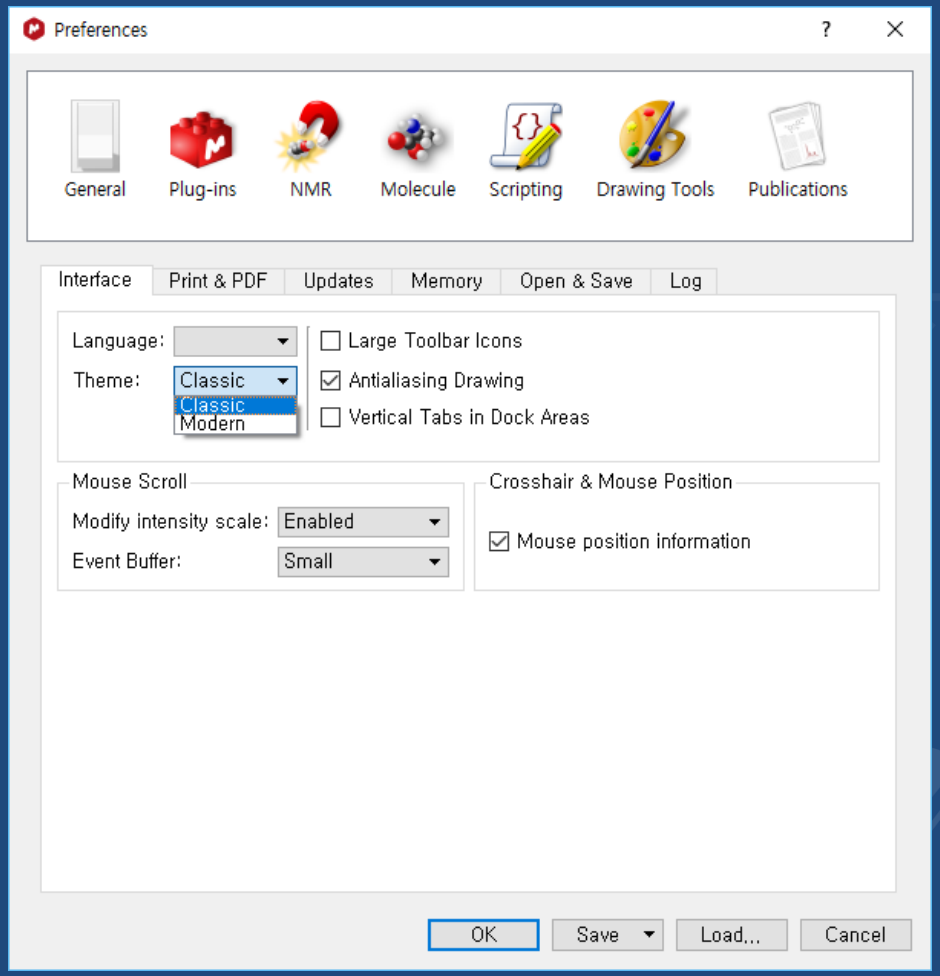
Installation

- Download Mnova
  - http://mestrelab.com/download/mnova/
- Or ...
  - Install Filezilla (FTP client)
  - Download License



Interface

- Modern vs classic

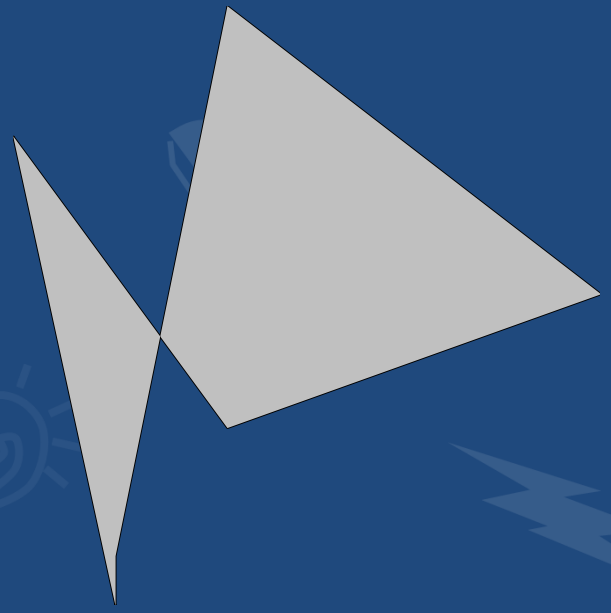


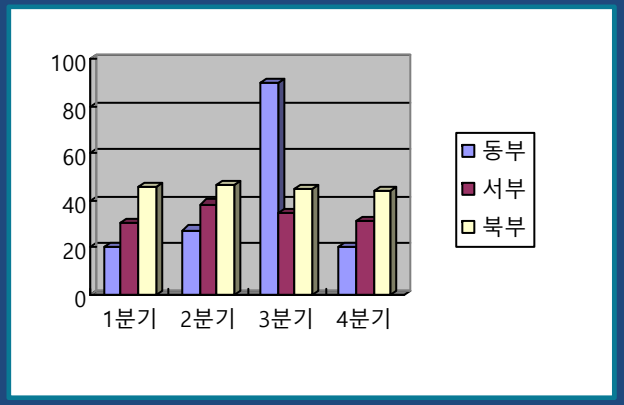
How to use?

- 1D processing
  - qNMR, water suppression
- 2D processing
  - NUS, assignment
- Multiple Spectrum Analysis
  - Reaction Monitoring // Kinetics
  - DOSY
  - T1/T2 relaxation
- Etc...

Home

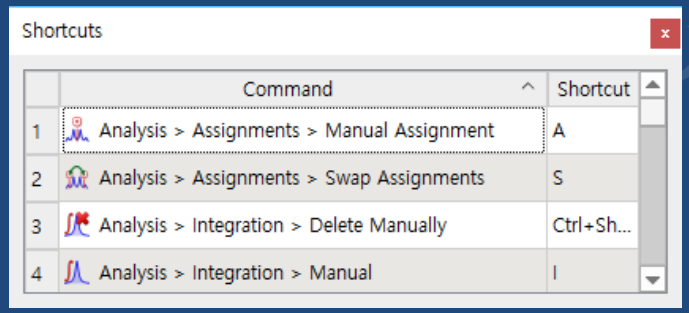
- Drawing
- Insert Object

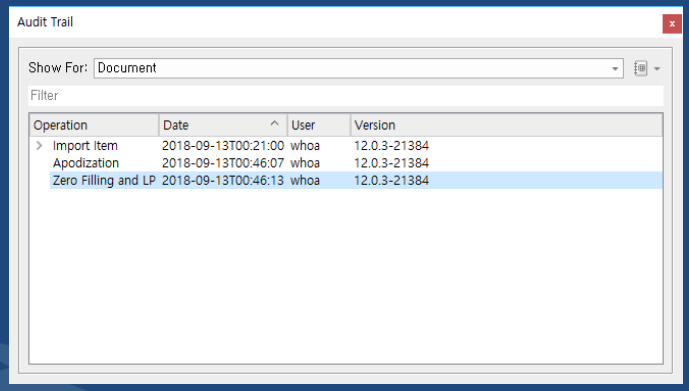


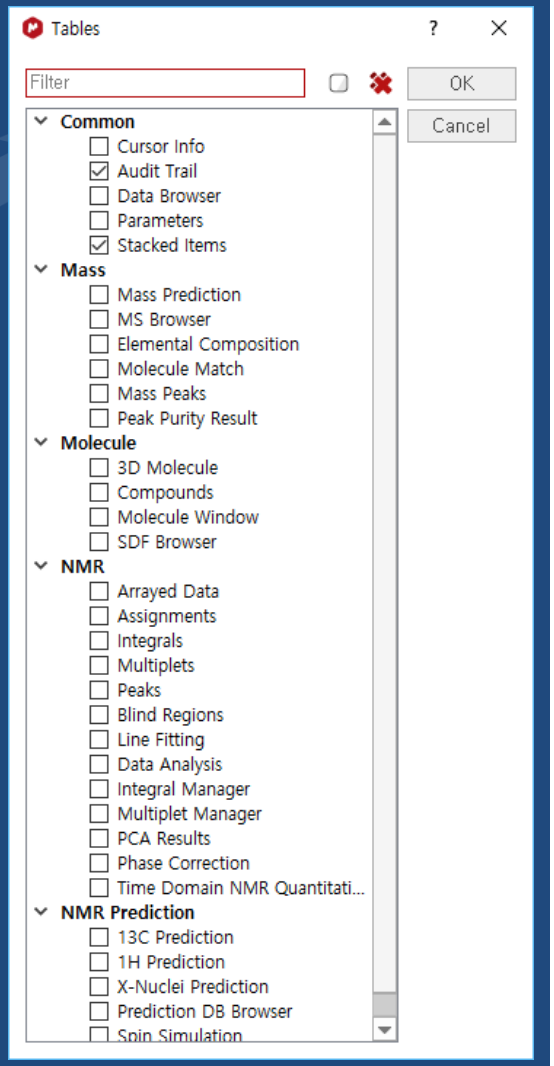


View

- Spectrum Toolbar
- Shortcuts
- Audit Trail
- Tables

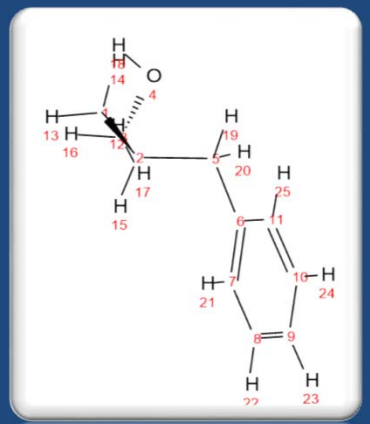


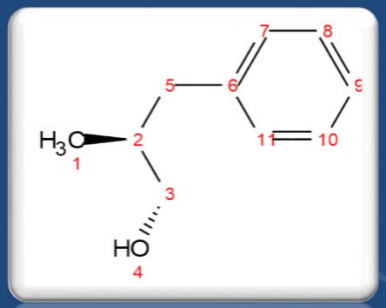


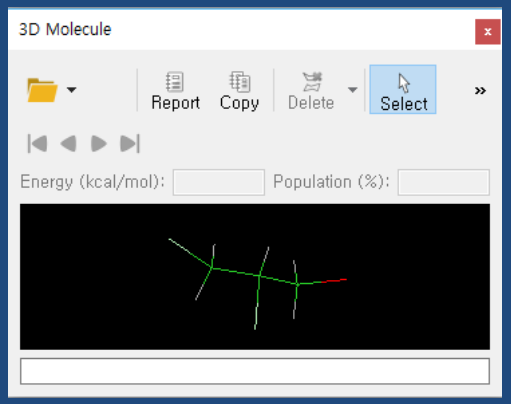


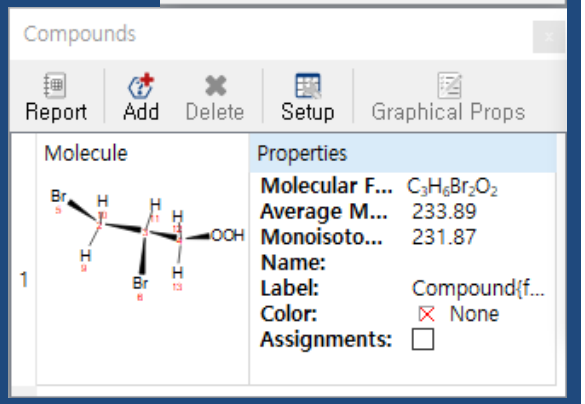
Molecule

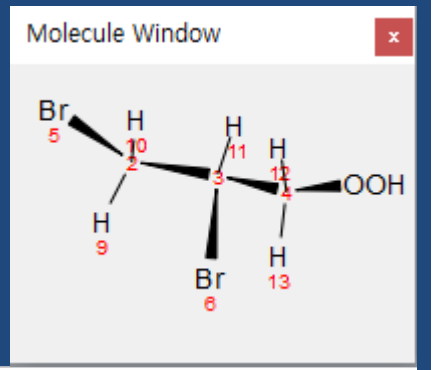
- Draw
  - 2D molecule
  - 3D molecule with energy minimization
  - View → Tables











1D processing

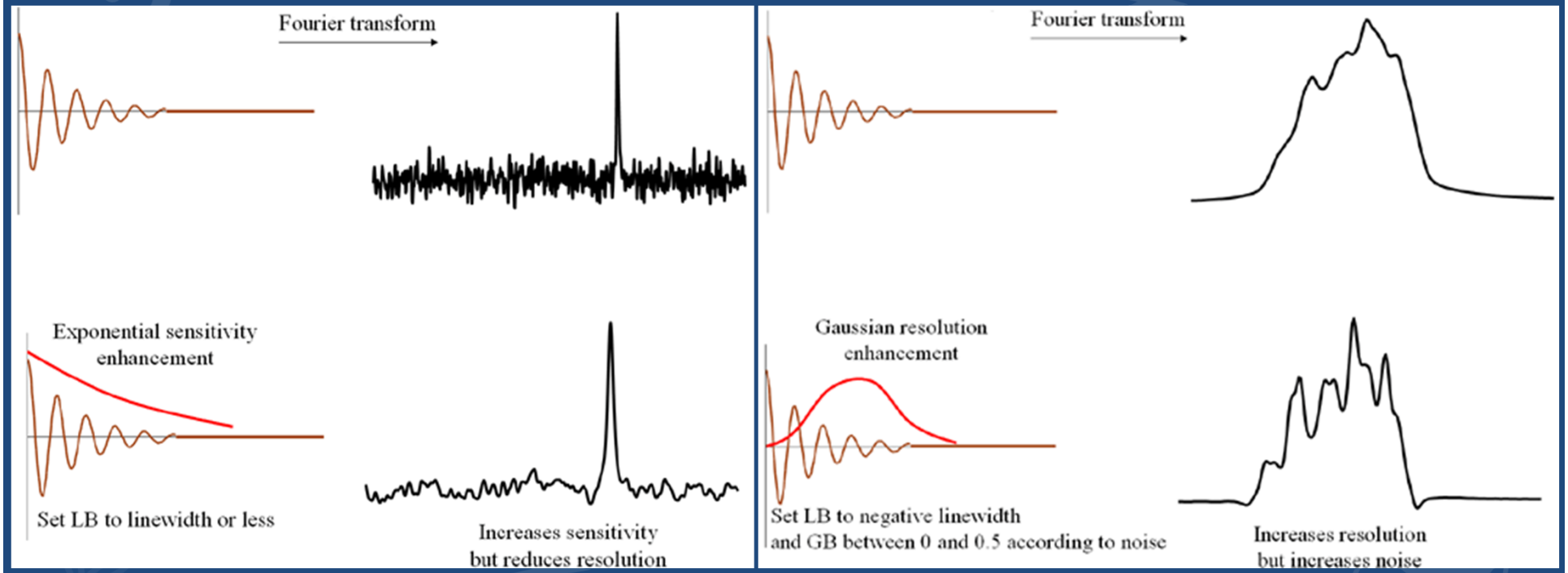
- Apodization
- Zero filling
- Phase correction
- Baseline correction
- Referencing
- Peak picking
- Integration
- Multiplet Analysis

Open files (Ctrl + O)

- Shortcut key : Ctrl+O
- Data type
  - Agilent / Varian : fid
  - Bruker
    - 1D : fid
    - nD : ser
  - Jeol : .jdf
- Or just drag-N-drop folder to Mnova

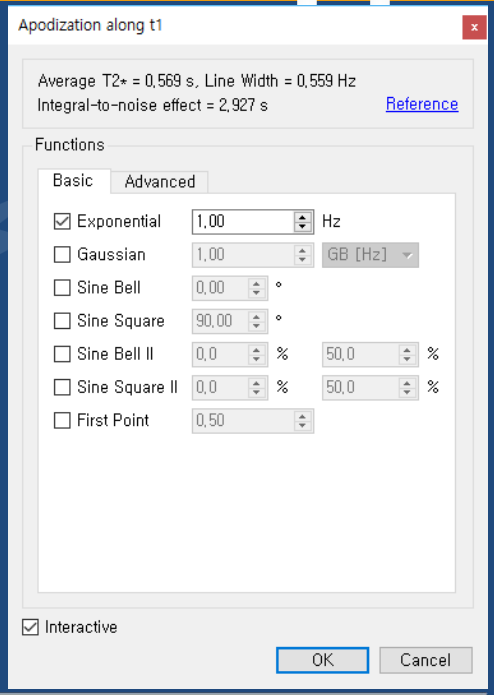
Apodization(W)

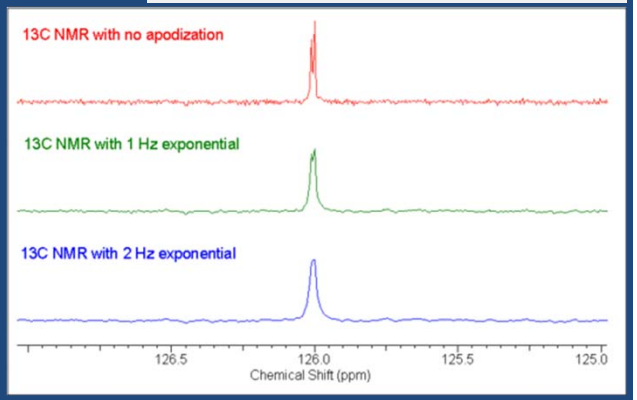
$$d'_k = a_k d_k$$



Apodization(W)

- 1D : optional
  - Proton :
    - Recommended for bad peak shape
    - Exponential // 0 ~ 0.2 Hz
  - Carbon :
    - Recommended for low S/N
    - Exponential // 1 ~ 2 Hz

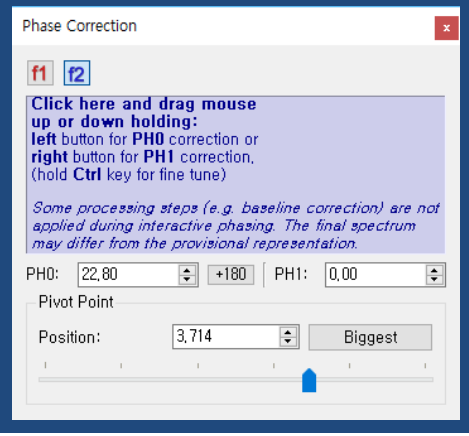




http://blog.acdlabs.com/elucidation/2008/09/how-much-is-too.html

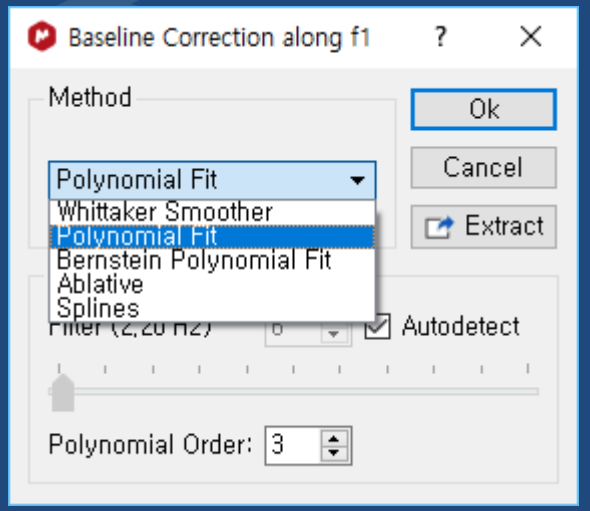
Phase correction(Shift + P)

- Auto phase correction : 1H, 13C
  - Bruker : apk, apk0
  - Agilent : aph or [Autoprocess]
  - Mnova :
- Manual correction
  - Spectrum of Hetero nuclei (11B, 19F, etc ...)
    - NMR tube : Borosilicate with teflon coating
  - Shortcut key (Shift + P)
    - Global correction (0th order)
    - Gradient correction (1st order)



Baseline correction(B)

- Auto baseline correction : 1H, 13C
  - Bruker : abs , absn, absf
  - Varian : bc
  - Mnova
- Manual correction
  - Polynomial Fit
    - Order : < 5 is recommended
    - Filter : Autodetect is recommended



Baseline correction(B)

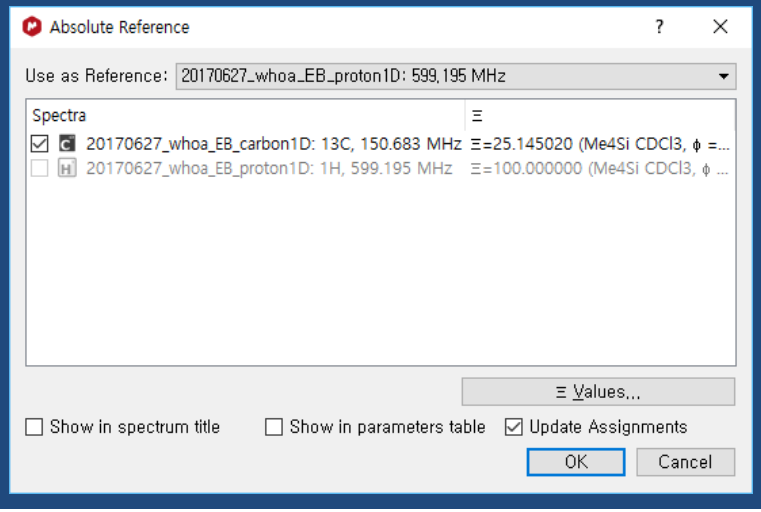
- Manual correction
  - Splines
  - Whittaker smoother
    - Don't use if broad peak is concerned
  - Bernstein Polynomial
  - Ablative
  - Multipoint Baseline Correction
    - Pick Point

Reference (L)

- Reference (L) or Graphic Reference (R)
  - TMS or DSS
  - Solvent peak

Absolute Reference

- IUPAC 2001 (modified 2008)
  - Reference of 1H resonance of TMS for all nuclei
  - http://www.iupac.org/publications/pac/80/1/0059/
- Absolute Reference
  - Bruker : $SR_X = \nu_X - BF1_X$
    - Ex> $\nu^{obs}_{TMS} = SF_H$ = 400.1300082 Mhz
      - BF1 + SR = 400.13 Mhz + 8.24 Hz
    - $\nu_C = \nu^{obs}_{TMS} \times \Xi_C / 100$ = 100.6127705 Mhz
      - 400.1300082 X 25.145020 /100 Mhz
    - $SR_C = \nu_C - BF1_C$ = 1.5 Hz
      - 100.6127705 – 100.6127690 Mhz
    - → SR 1.5
  - Mnova :

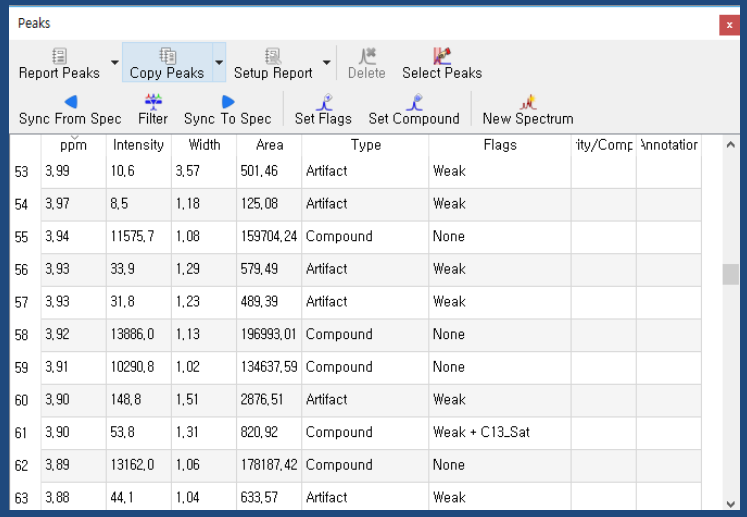


Zoom (Z)

- Zoom
  - Zoom in (Z)
    - Horizontal → Vertical → 2D
  - Zoom out (Shift + Z)
  - Manual Zoom (M)
- Full Screen (F)
- Crosshair (C)
- Back to cursor (ESC)

Peak Picking(K, Ctrl+K)

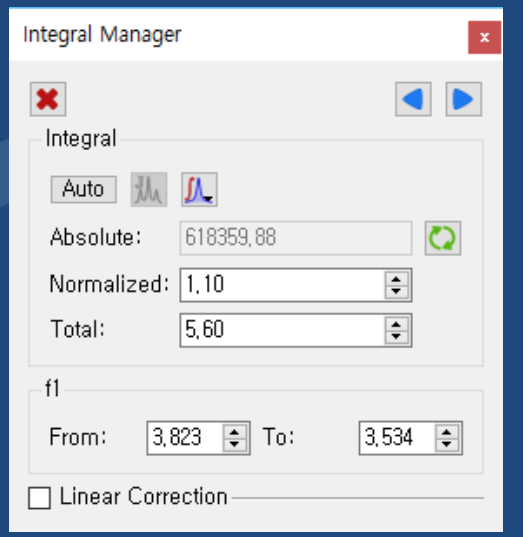
- Automatic Peak Picking
  - Mnova :
- Manual
  - Threshold : K
  - Peak by peak : Ctrl + K
  - Merge
  - Delete : Ctrl + Shift + K
  - Delete all
- Tables → Peaks

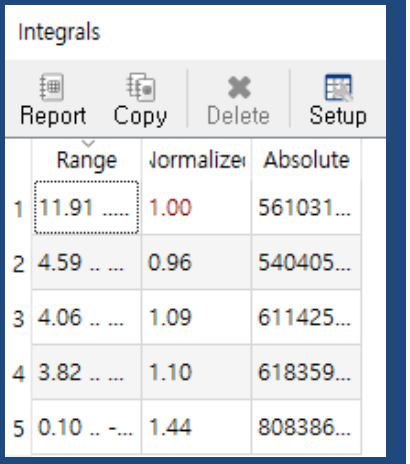


	ppm	Intensity	Width	Area	Type	Flags	ity/Comp	Annotatior
53	3,99	10,6	3,57	501,46	Artifact	Weak		
54	3,97	8,5	1,18	125,08	Artifact	Weak		
55	3,94	11575,7	1,08	159704,24	Compound	None		
56	3,93	33,9	1,29	579,49	Artifact	Weak		
57	3,93	31,8	1,23	489,39	Artifact	Weak		
58	3,92	13886,0	1,13	196993,01	Compound	None		
59	3,91	10290,8	1,02	134637,59	Compound	None		
60	3,90	148,8	1,51	2876,51	Artifact	Weak		
61	3,90	53,8	1,31	820,92	Compound	Weak + C13_Sat		
62	3,89	13162,0	1,06	178187,42	Compound	None		
63	3,88	44,1	1,04	633,57	Artifact	Weak		

Integration (I)

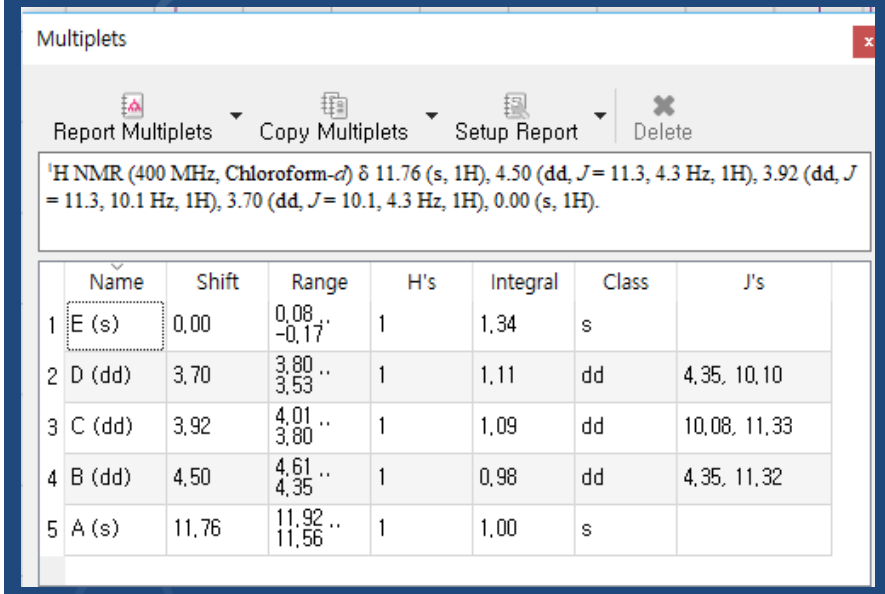
- Integration
  - Bruker : .int
  - Agilent : intmod
  - Mnova : I
- Rule
  - Apply same rule when drag region of peak
  - First integration : 1.00
    - Integral Manager
    - Tables → Integrals

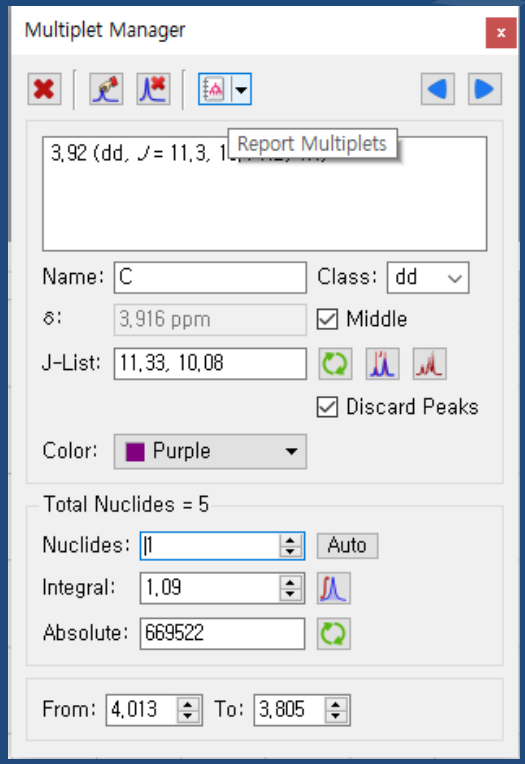


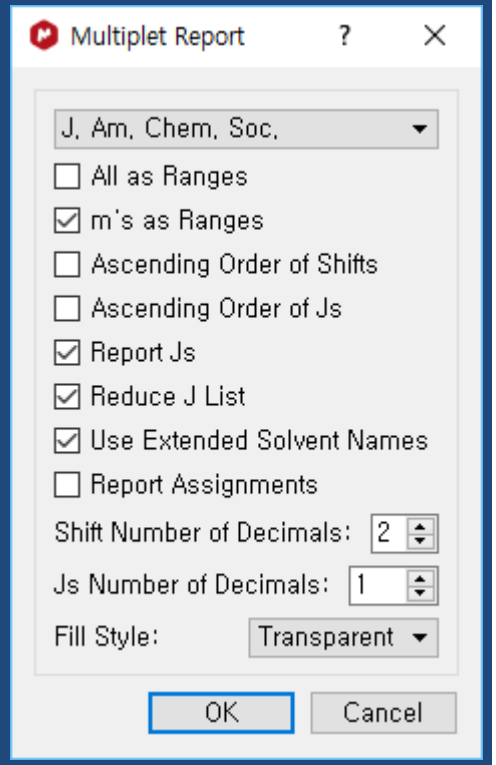


Multiplet(J)

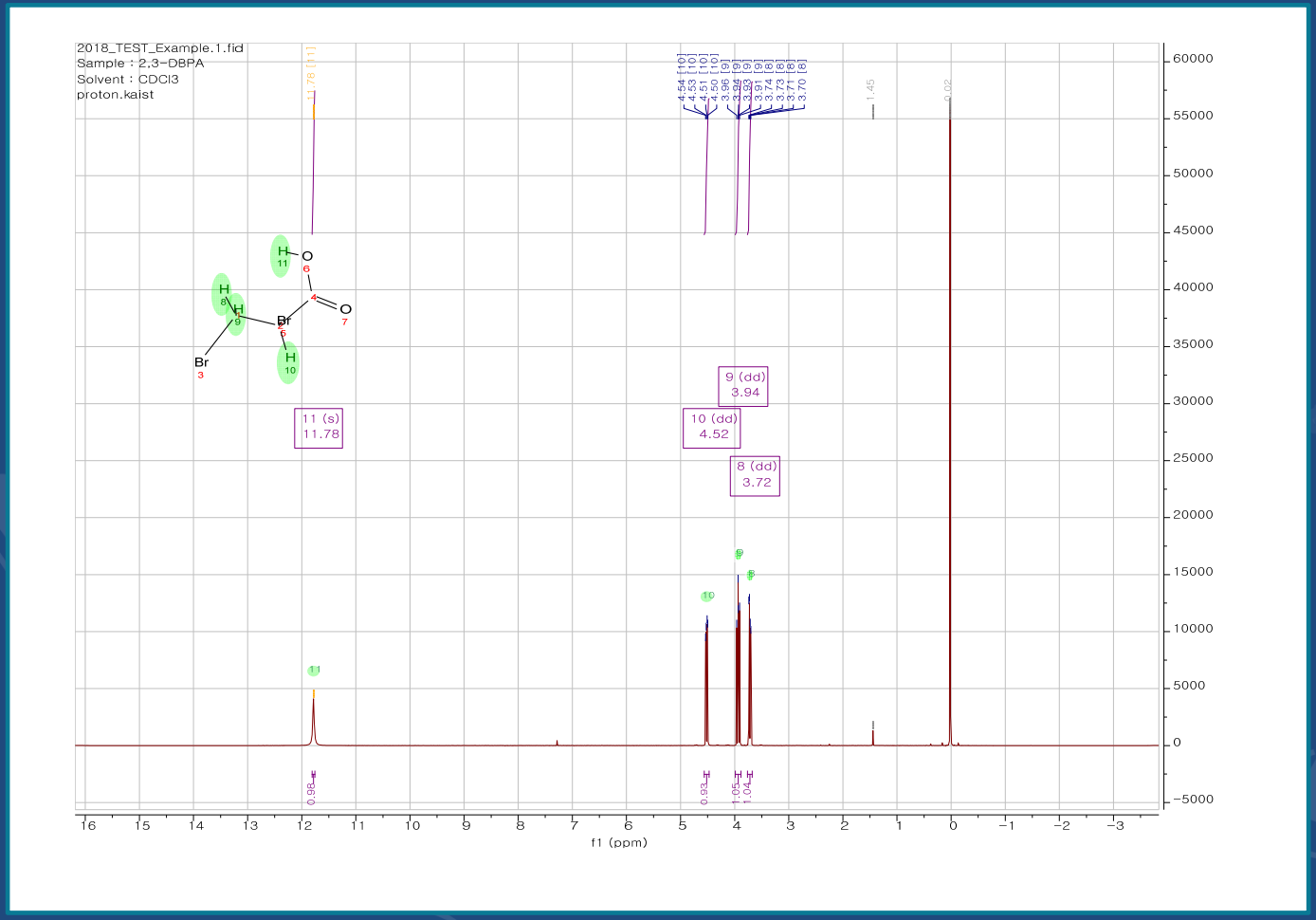
Multiplet analysis (J)



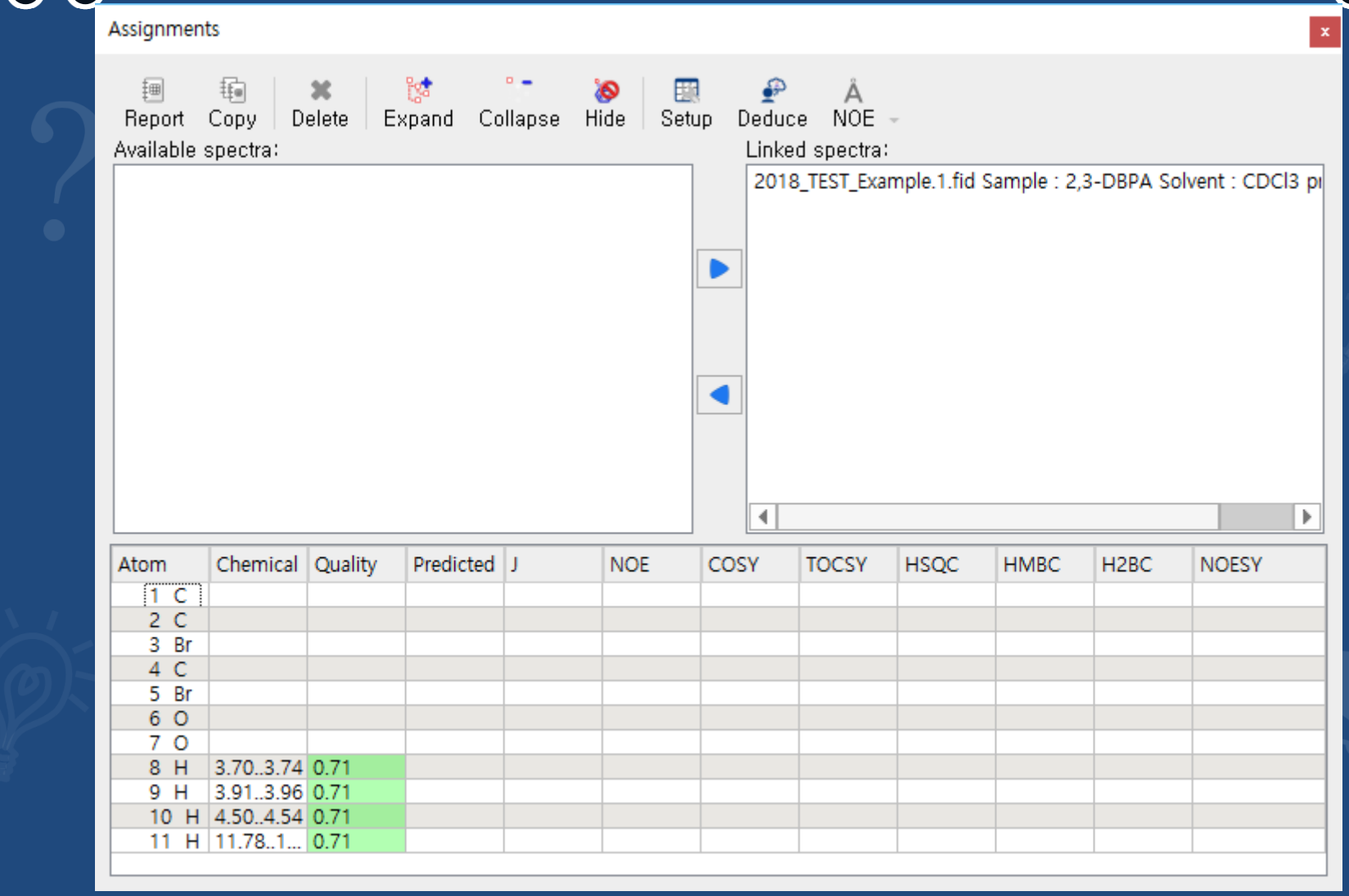




Assignments(A)



Assignments(A)

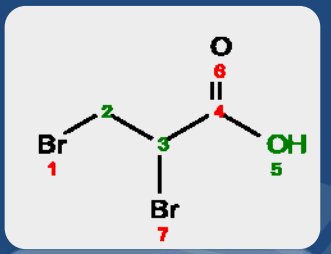


Atom	Chemical	Quality	Predicted	J	NOE	COSY	TOCSY	HSQC	HMBC	H2BC	NOESY
1 C											
2 C											
3 Br											
4 C											
5 Br											
6 O											
7 O											
8 H	3.70..3.74	0.71									
9 H	3.91..3.96	0.71									
10 H	4.50..4.54	0.71									
11 H	11.78..1...	0.71									

qNMR

- Reproducible integration
  - Relaxation delay : 3 ~ 7 X T1
  - Phase correction
  - Baseline correction : Polynomial (oth order)

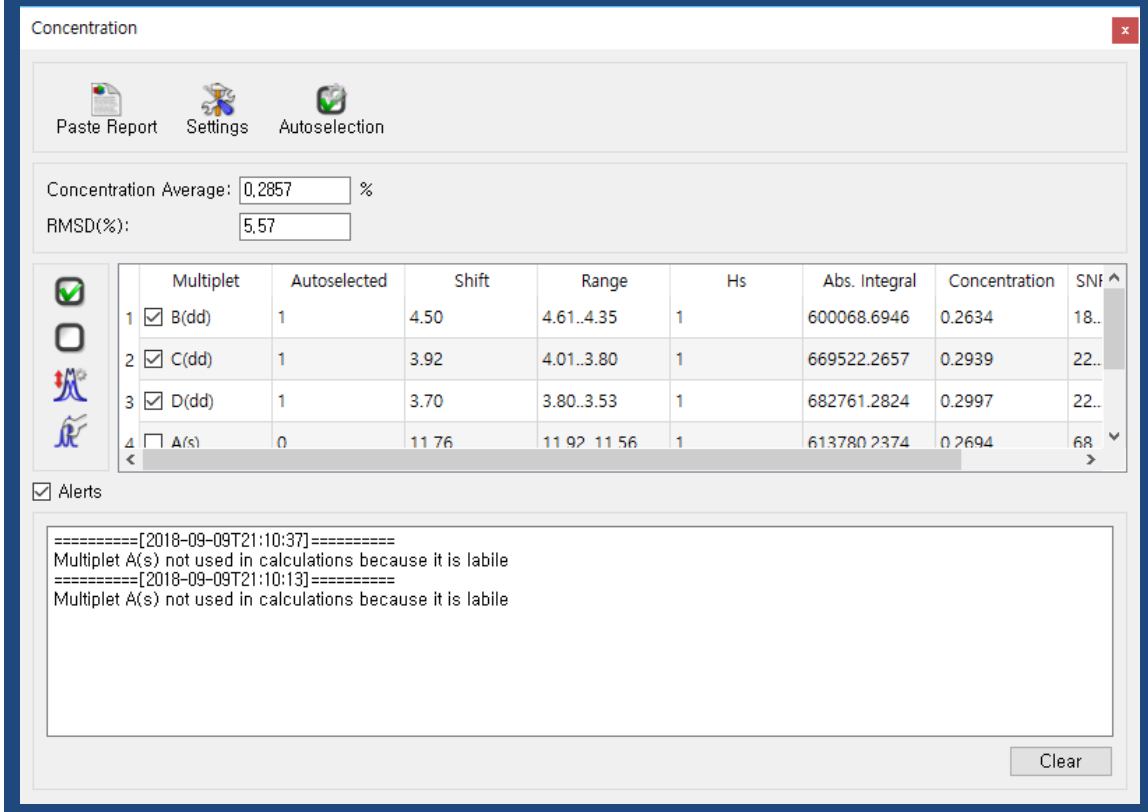
Intensity	5-OH	3-H	2a-H	2b-H	TMS	RMSD(%)
T1(s)	5.37 sec	8.22 sec	1.49 sec	1.36 sec	11.18 sec	
D1=2s	0.68	0.66	0.78	0.79	1.00	7.8971
D1=24s	0.69	0.67	0.71	0.71	1.00	2.7994
D1=60s	0.62	0.64	0.64	0.64	1.00	0.1570

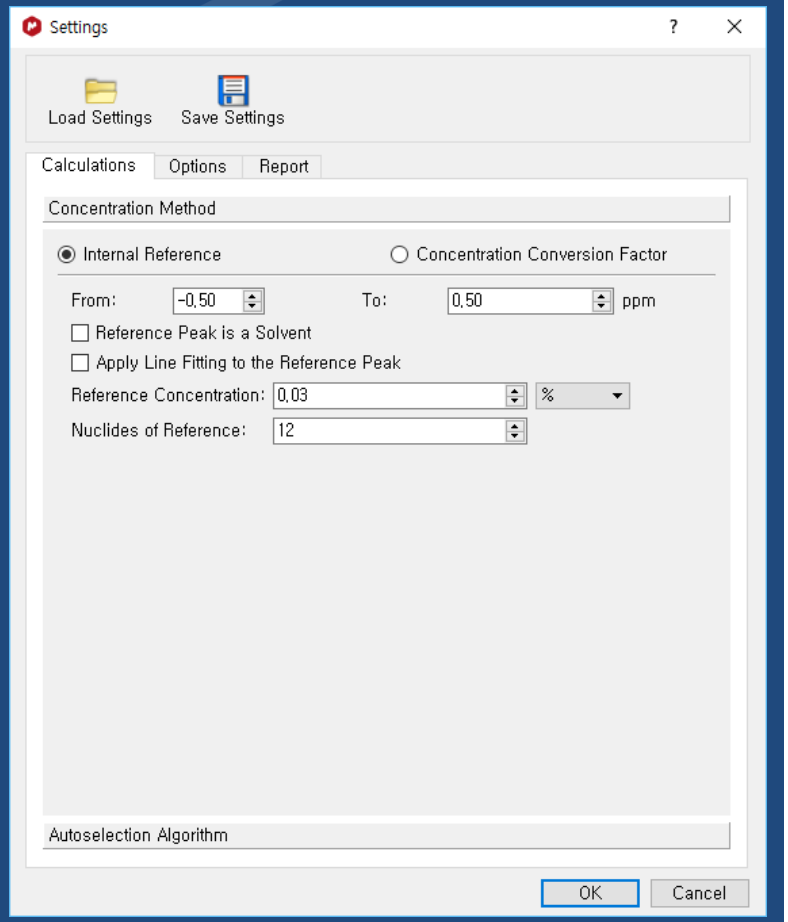


400Mhz, 298K, 90 pulse, 4 s acquisition time, 8 scan

qNMR

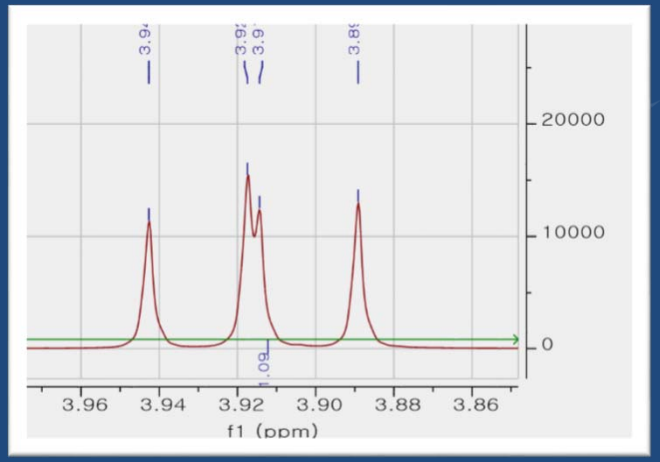
- Advanced → qNMR → Concentration
- Settings





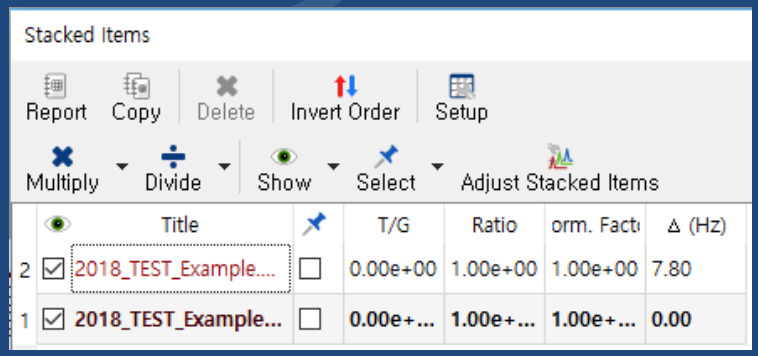
Additionally…

- View
  - Expansion (E)
  - Cut (X)
  - Restore (V)
- Properties



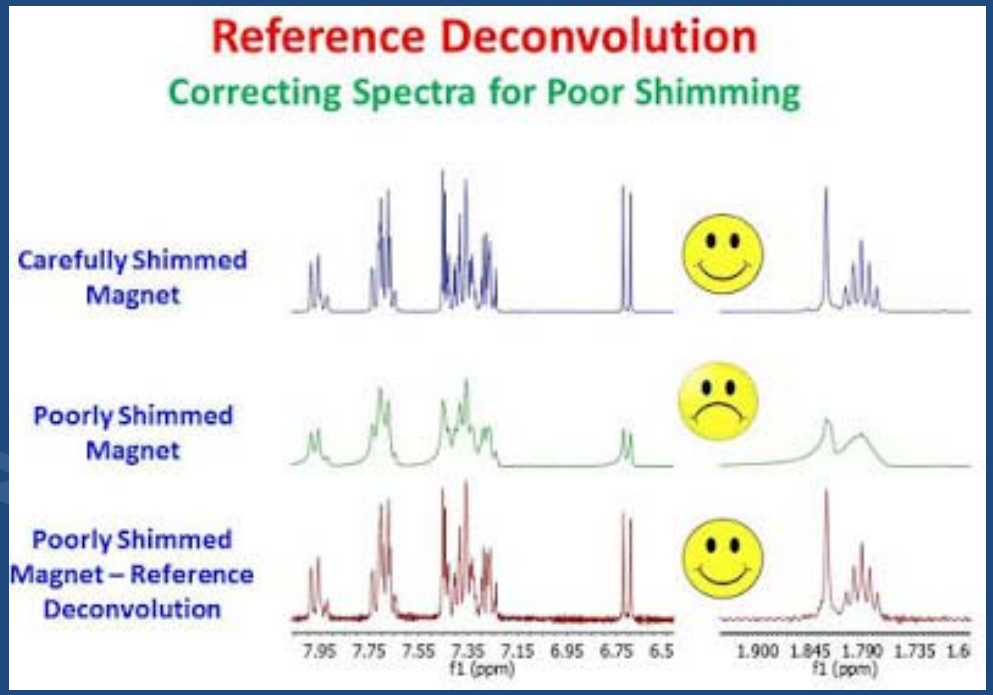
Stack

- Select pages → Menu : Stack -->
  - Stack items
  - Superimpose items
  - Bruker : .md
- Arithmetic
  - 1D NOE experiment : Steady-state NOE



Reference Deconvolution

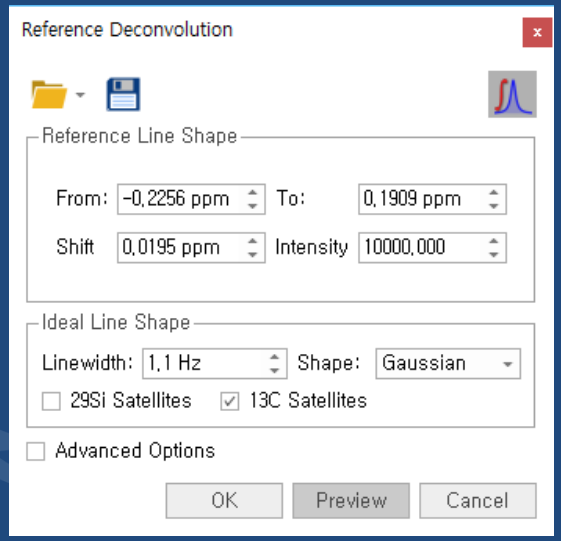
- Improve "spectra of Bad shim"
  - Good : Bad lineshape → ideal lineshape
  - Bad : S/N will be sacrificed

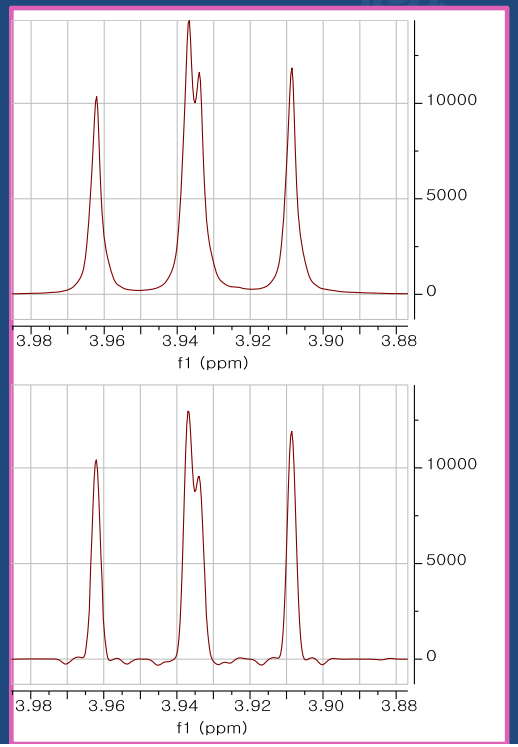


http://u-of-o-nmr-facility.blogspot.com/2017/07/correcting-nmr-spectra-for-poor.html

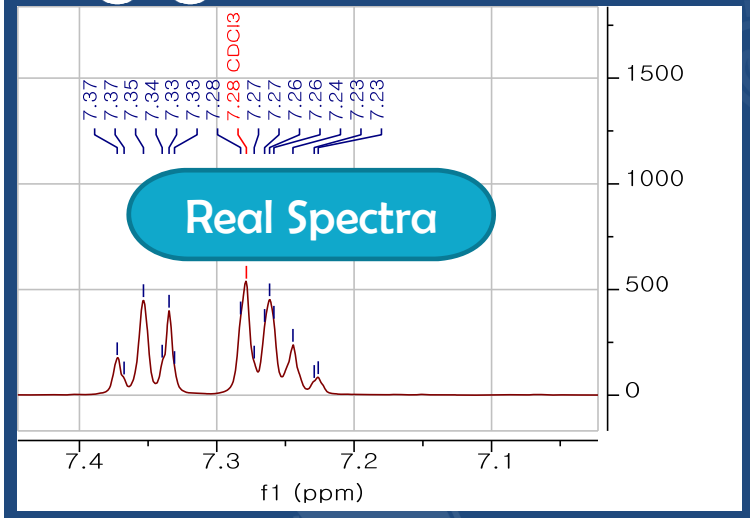
Reference Deconvolution

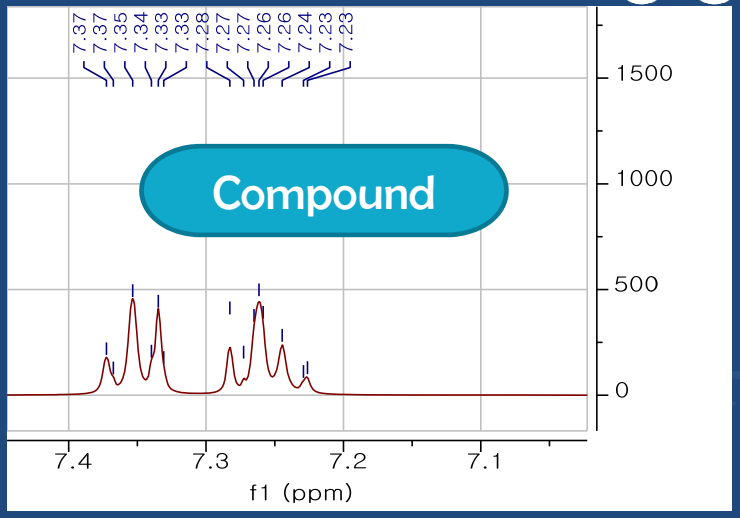
- Bruker : .refdcon (Topspin ver. >=3.5)
- Mnova
  - Integrate “Separated strong singlet peak”
    - Ex> TMS

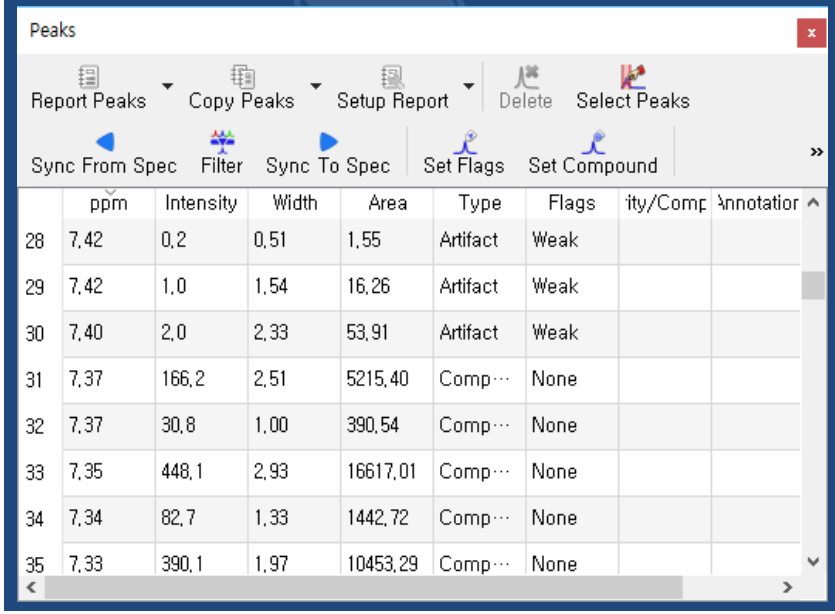




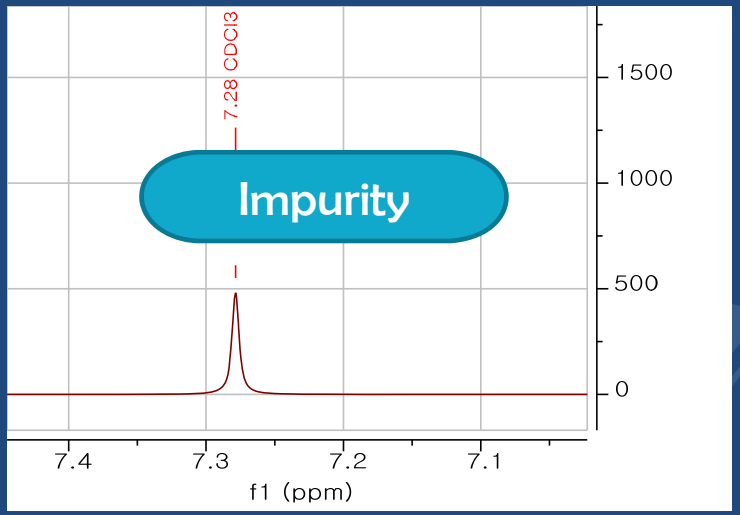
Compound // Impurity





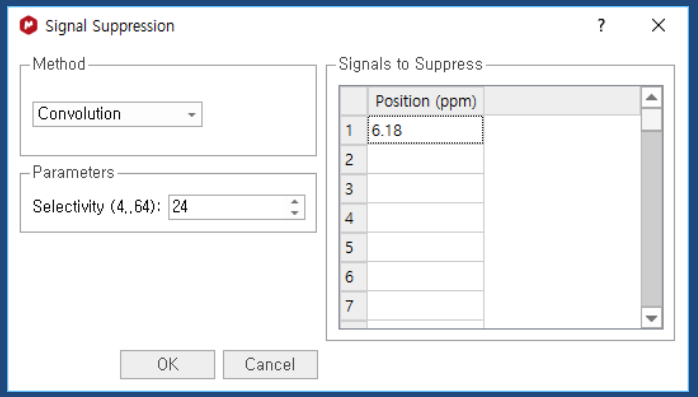


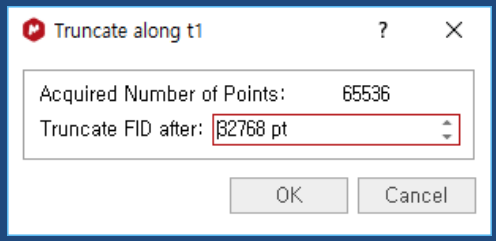
	ppm	Intensity	Width	Area	Type	Flags	ity/Comp	Annotation
28	7,42	0,2	0,51	1,55	Artifact	Weak		
29	7,42	1,0	1,54	16,26	Artifact	Weak		
30	7,40	2,0	2,33	53,91	Artifact	Weak		
31	7,37	166,2	2,51	5215,40	Comp···	None		
32	7,37	30,8	1,00	390,54	Comp···	None		
33	7,35	448,1	2,93	16617,01	Comp···	None		
34	7,34	82,7	1,33	1442,72	Comp···	None		
35	7,33	390,1	1,97	10453,29	Comp···	None		

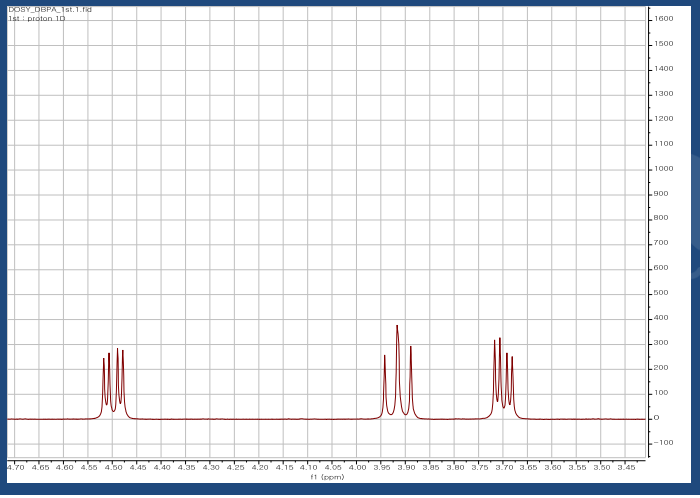


Other functions...

- Signal suppression
- Truncate
- Binning
  - S/N ∧, resolution ∨
- Smoothing
- Resolution Booster







2D processing

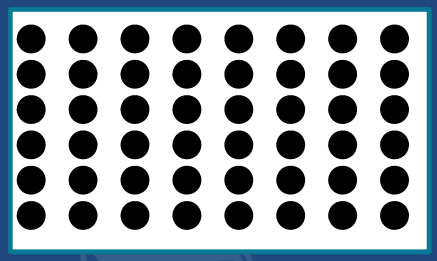
- Apodization
- Zero filling
- Linear Prediction
- Phase correction
- Baseline correction
- Referencing
- Peak picking
- Assignment

In advance...

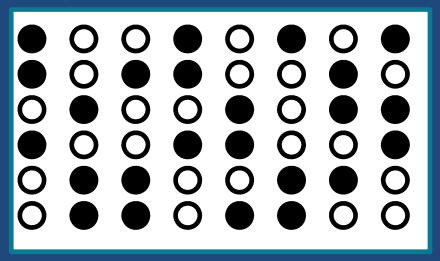
- Acquisition Parameter
  - NS : Number of Scan
  - SW : Spectral Width
  - O1P, O2P : Offset
  - TD : Number of point
    - NUS
- Experiment
  - Homonuclear : COSY, NOESY, ROESY, TOCSY
  - Heteronuclear : HSQC, HMBC, H2BC,

Non-uniform Sampling

- Save Time for 2D acquisition
  - 25 ~ 50%

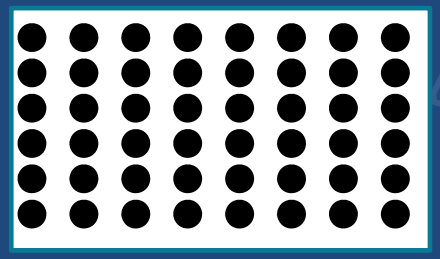


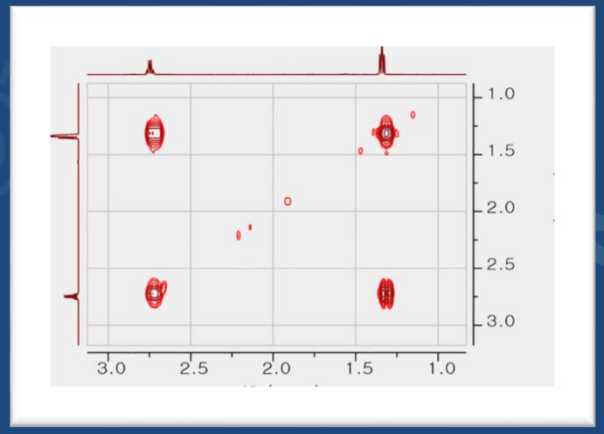
Uniform sampling



Non -Uniform Sampling

NUS processing



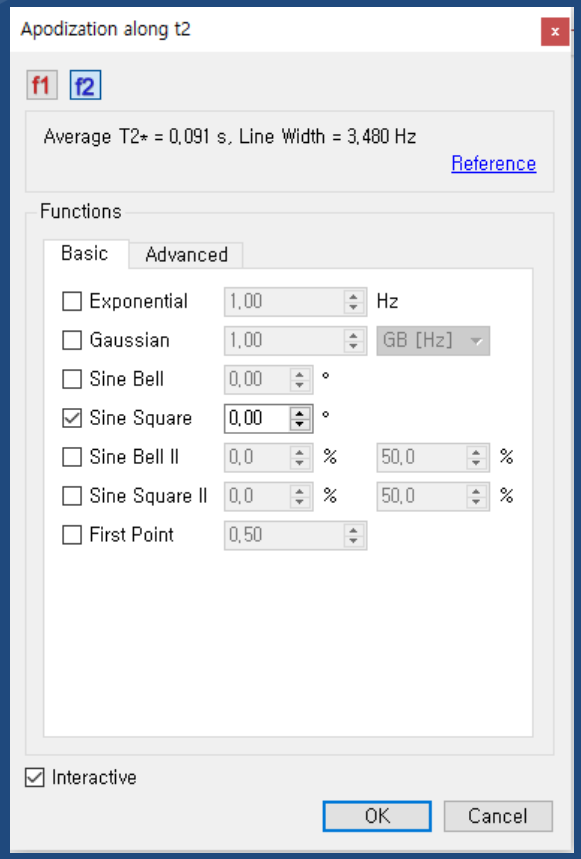


2D spectra processing

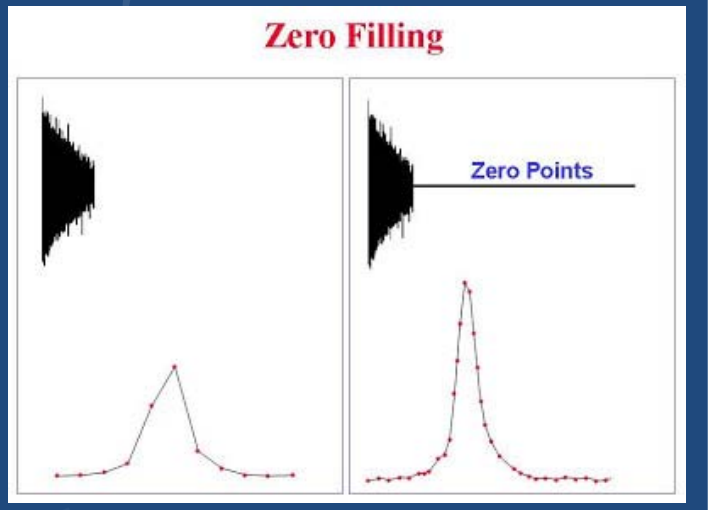
- FT
  - Bruker : xfb
  - Varian : wft2da
- Convolution
  - Bruker : dcon2d
  - Varian : dconi
- Extract 1D
  - Bruker : rser
  - Varian : ds

Apodization (W)

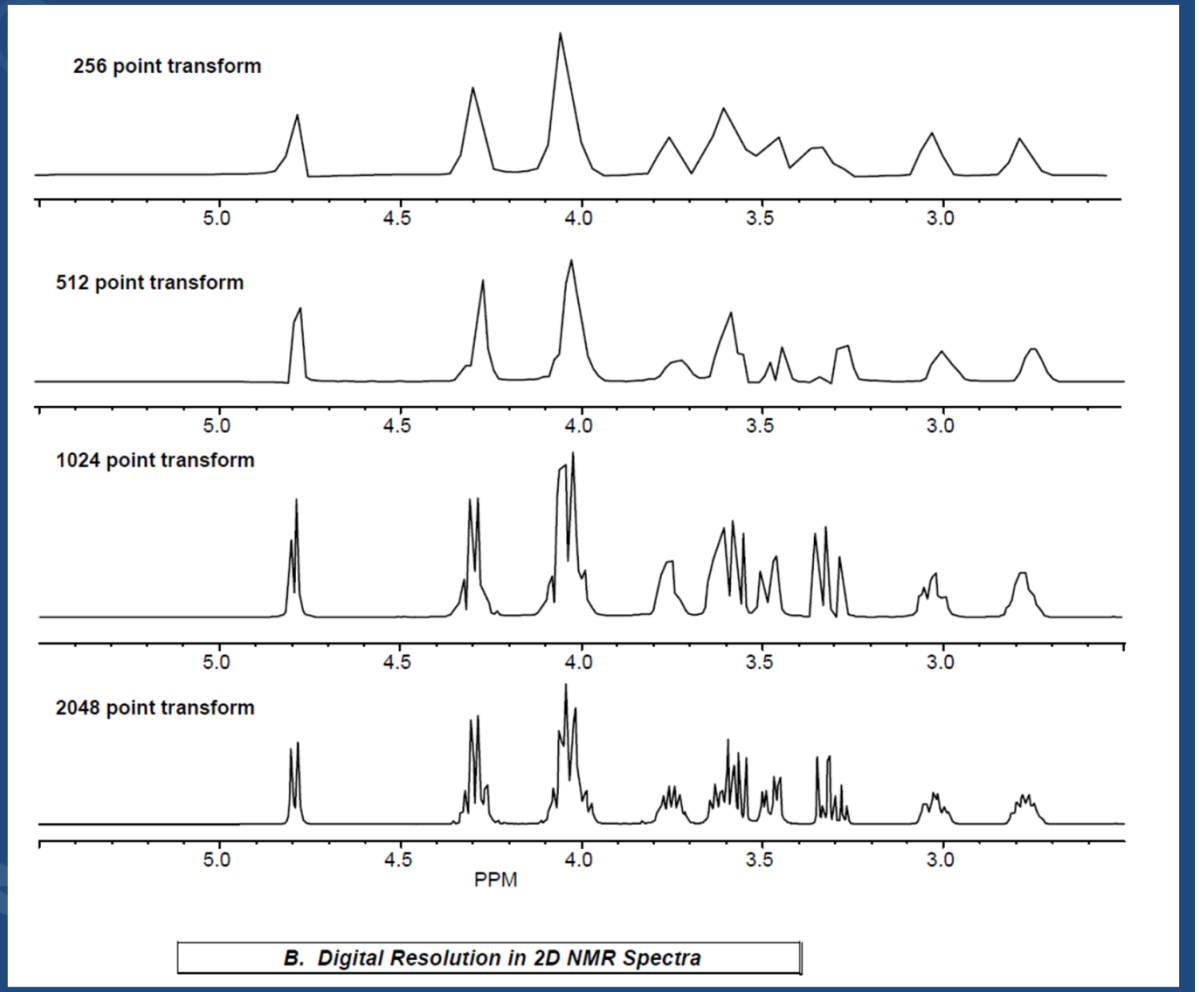
- 2D : strongly recommended
  - Magnitude : SSB 0 deg
    - COSY, HMBC
  - Phase sensitve : SSB 90 deg
    - NOESY, edited HSQC
- Apply both F2 and F1



Zero Filling



http://u-of-o-nmr-facility.blogspot.kr/2007/11/zero-filling.html

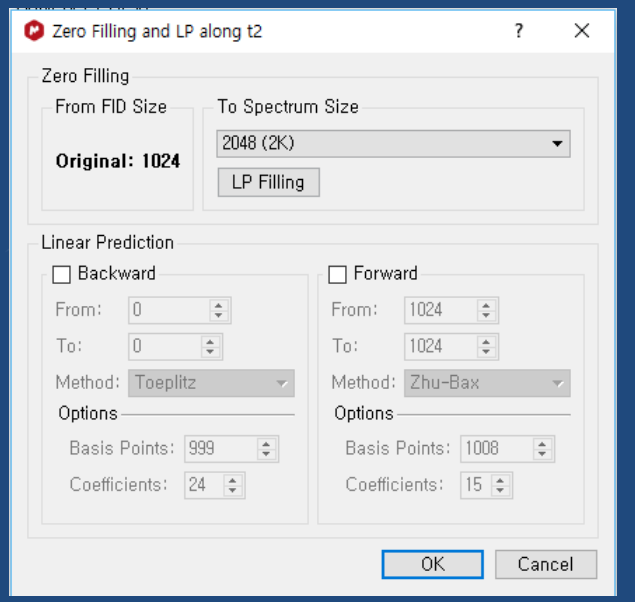


B. Digital Resolution in 2D NMR Spectra

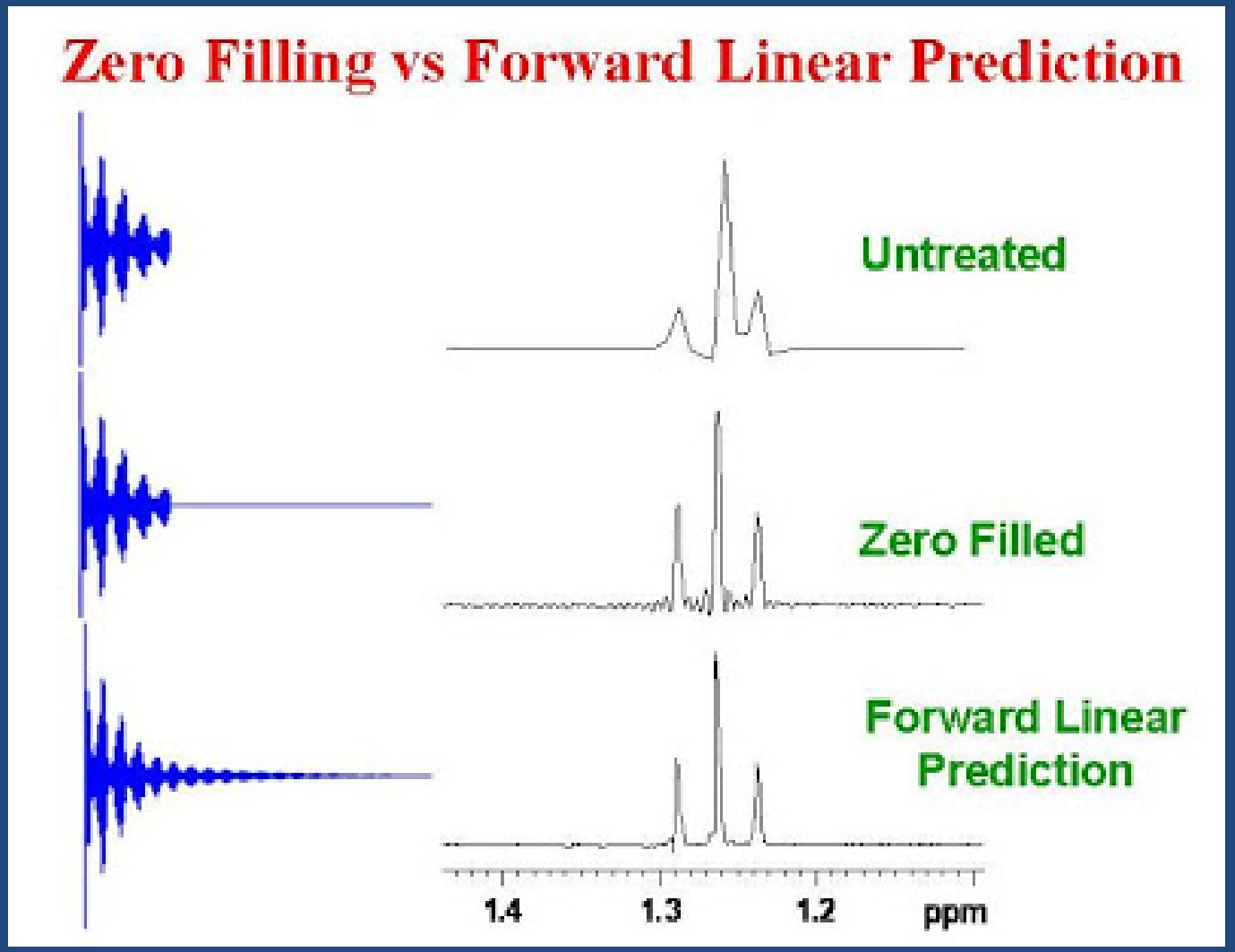
http://nmr.asu.edu/page8/files/zerofill.pdf

Zero Filling

- Bruker : SI
- Mnova
  - Select dimension : F2
  - Processing → Zero Filling and LP
    - "From FID Size"
      - To Spectrum Size : upper size
        - Ex> original : 512 ~ 972 // To : 1024
      - Check available hard disk usage



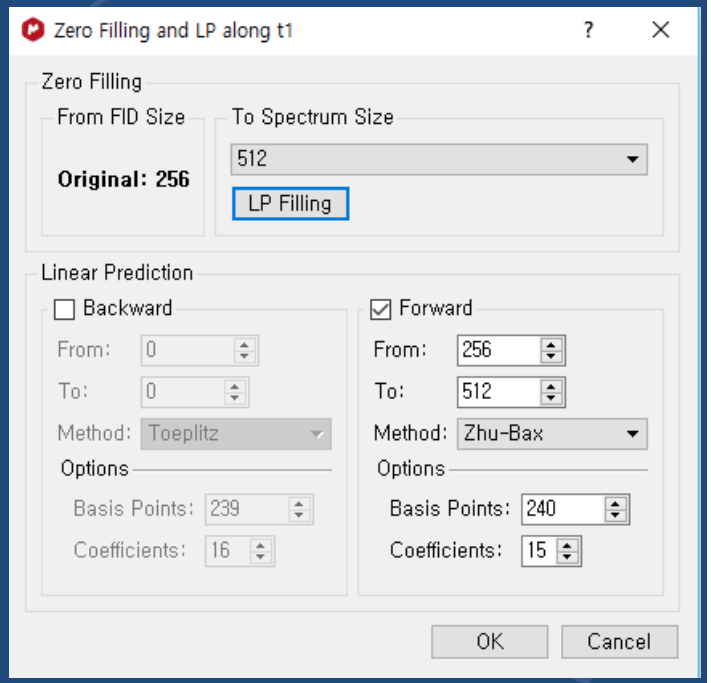
Linear Prediction



http://u-of-o-nmr-facility.blogspot.kr/2008/03/forward-linear-prediction.html

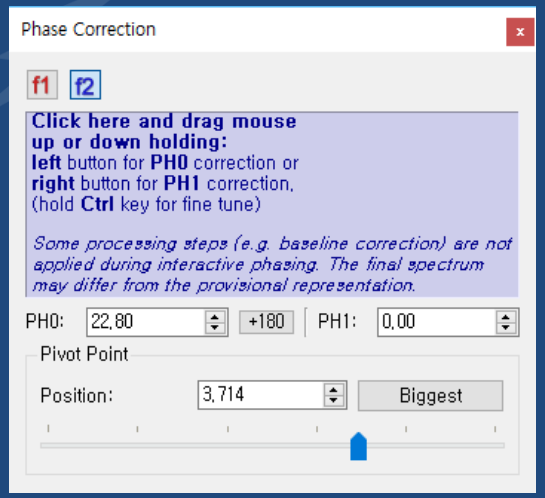
Linear Prediction

- Select dimension : F1
- Processing → Zero Filling and LP
  - "From FID Size"
    - To Spectrum Size : upper size
      - Check available hard disk usage
  - Click : LP Filling
    - "Forward" should be checked



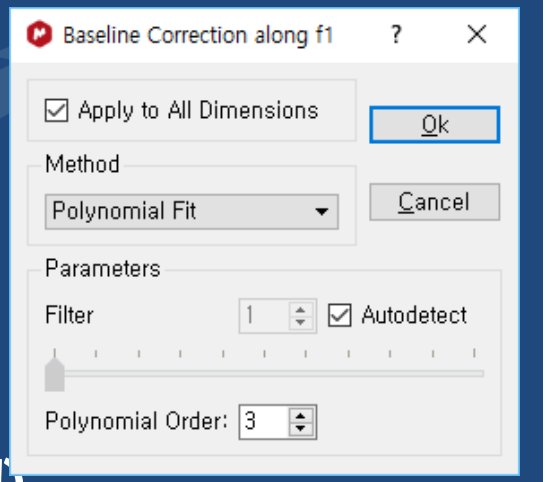
Phase correction(Shift + P)

- For Phase-sensitive spectrum
  - For NOESY, edited HSQC
    - Cf> mag COSY, HMBC
- Auto phase correction
  - Bruker : .ph
  - Mnova :
- Manual correction
  - Shortcut key (Shift + P)
    - Global correction (0^{th} order)
    - Gradient correction (1^{st} order)

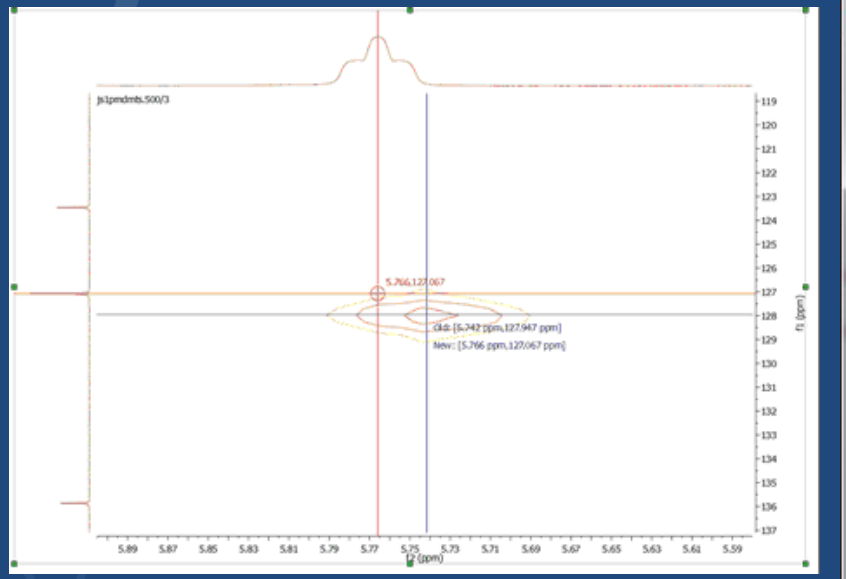


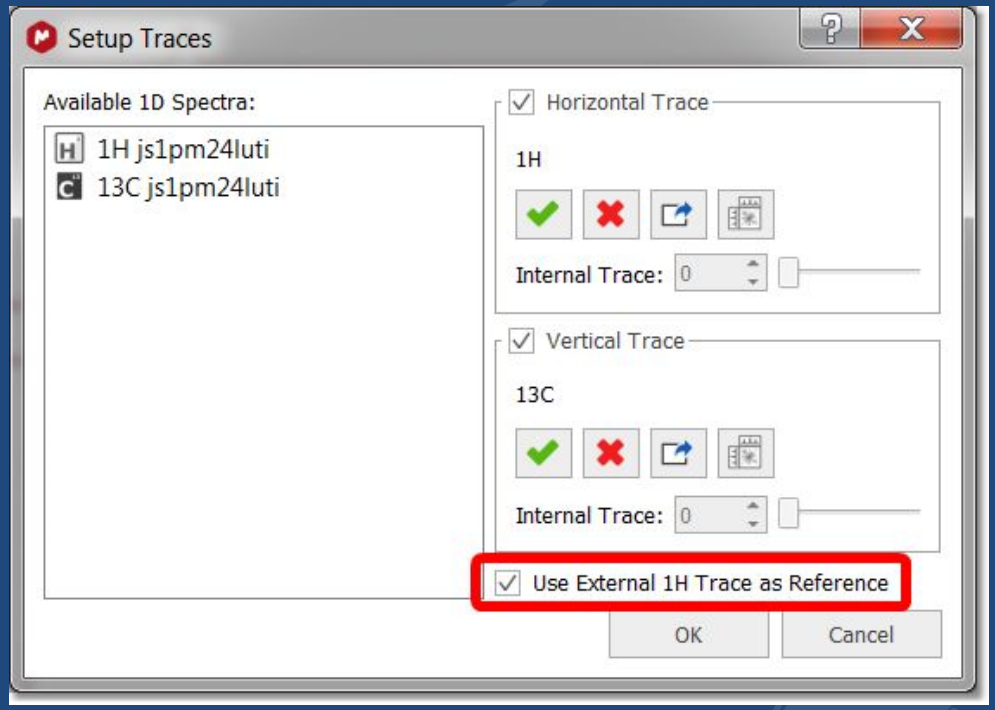
Baseline correction(B)

- Auto baseline correction : 1H, 13C
  - Bruker : abs , absn, absf , abs2
  - Mnova
- Manual correction
  - Polynomial Fit
    - Order : 0 ~ 20 (<5 recommended)
    - Filter : autodetect
  - Check “Apply to All Dimensions”
    - If user want, can apply f2, f1 separately.



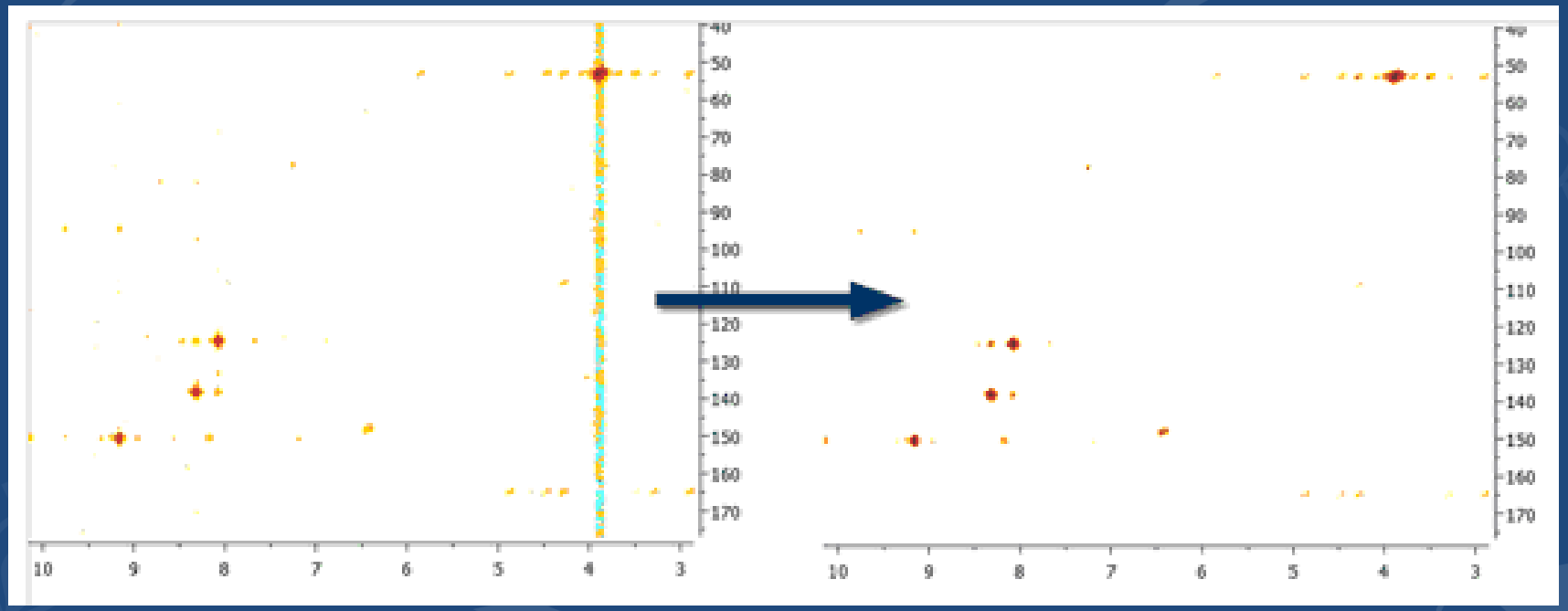
1D external trace



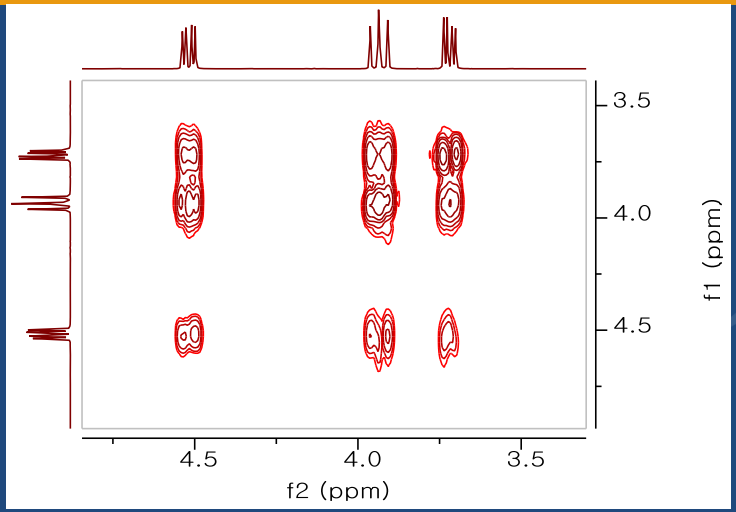


Reduce t1 Noise

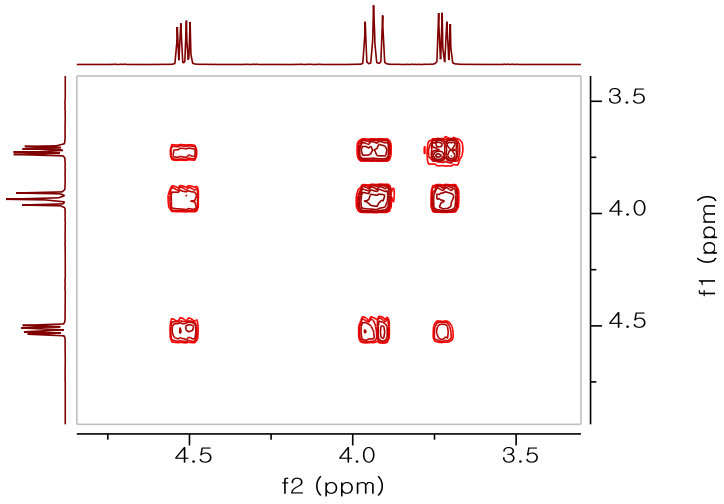
- For heteronuclei spectra(ex>HSQC)



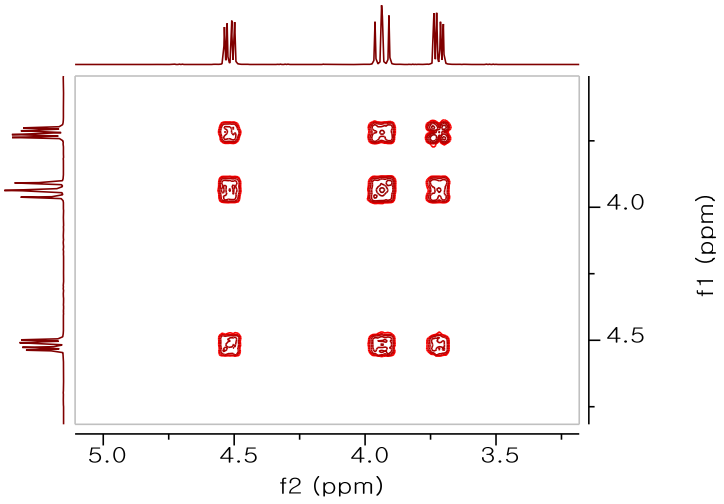
Symmetrization vs Covariance



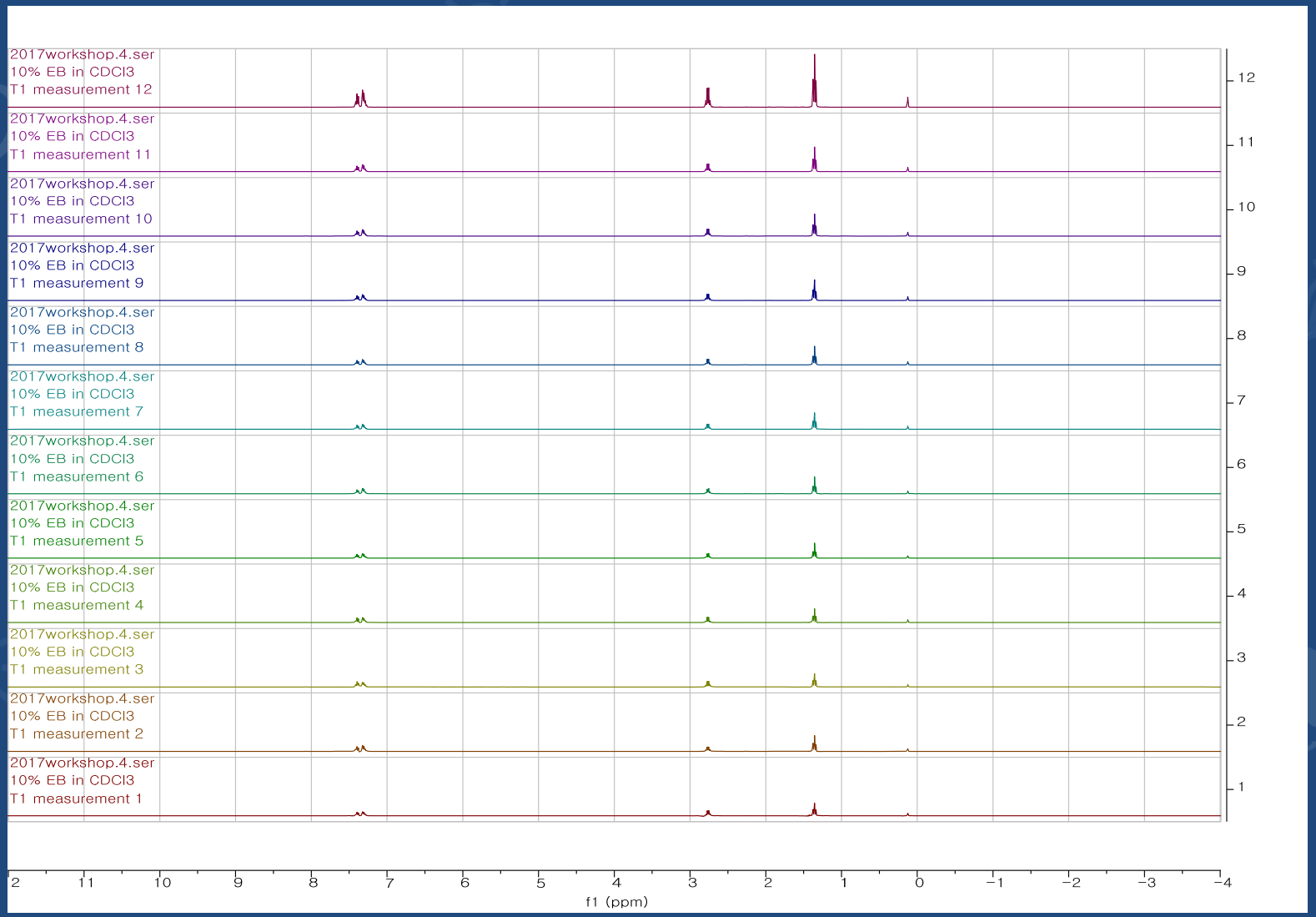
Symmetrization



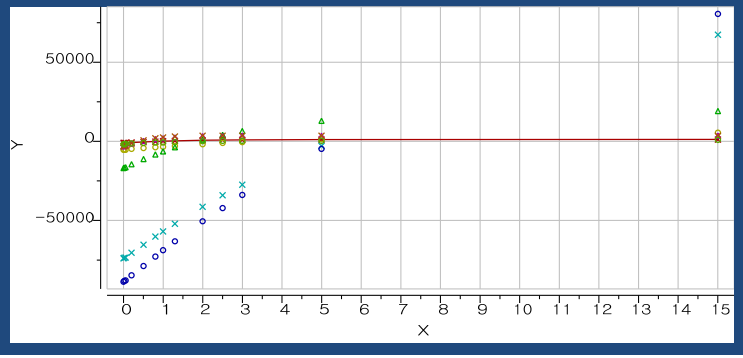
Covariance



T1 relaxation



T1 relaxation



Y'-Column Model Function

Functions

	Name	Function	Initialization	Report	
1	Linear Fit	A+B*x	A= 0, B= 0		Zero Order R
2	Mono-exponential Fit	B*exp(-x*F)			Exponential D
3	Three Parameter Exponential Fit	B+F*exp(-x*G)			Exponential D
4	Inverse Linear Fit	1/(A+B*x)	A= 1, B= 0		Second Orde
5					

Restore Defaults

Fitted Parameters

Calculate

B= 1240,41, F= −2307,86, G= 0,699326
rError = 0,0114327, probnotmono = 0,79731

OK Cancel

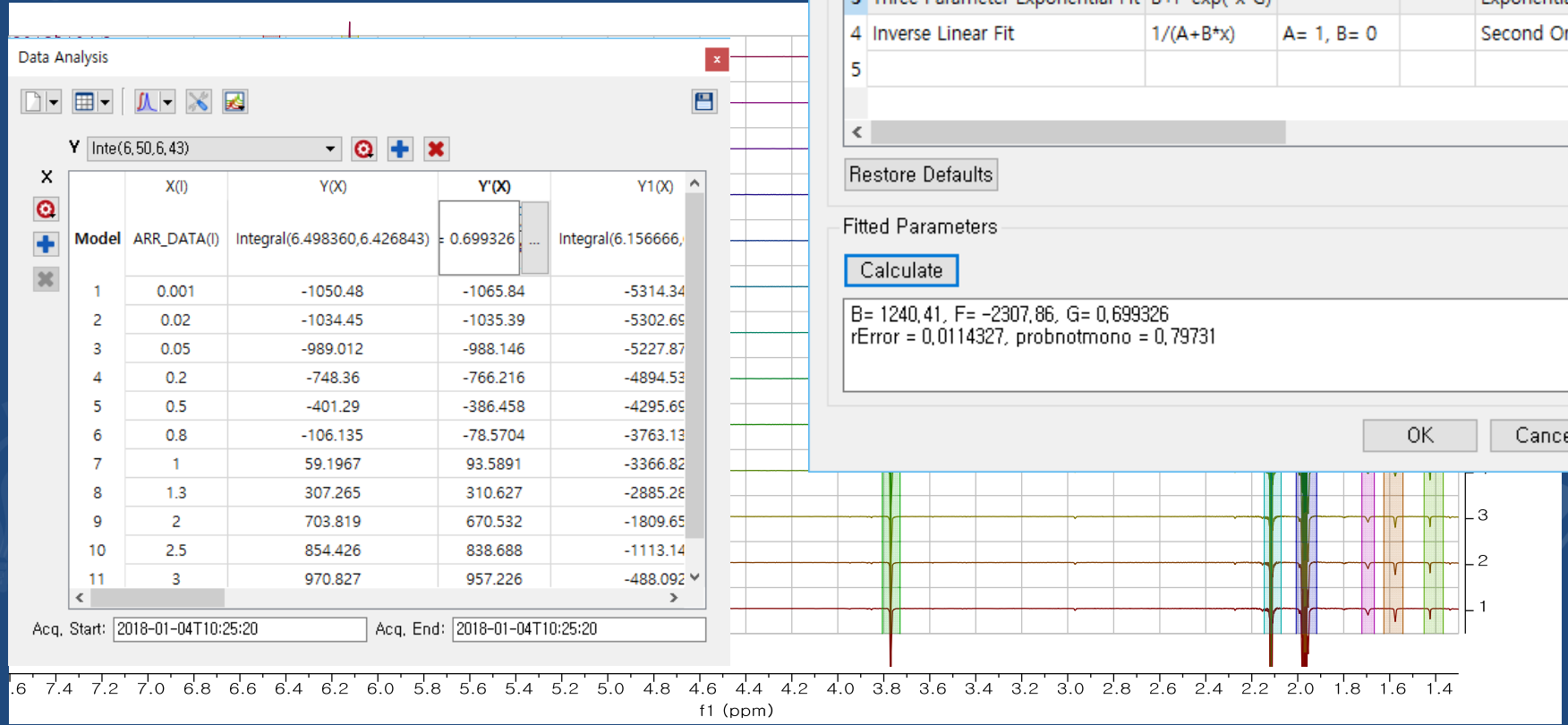
Data Analysis

Y Inte(6,50,6,43)

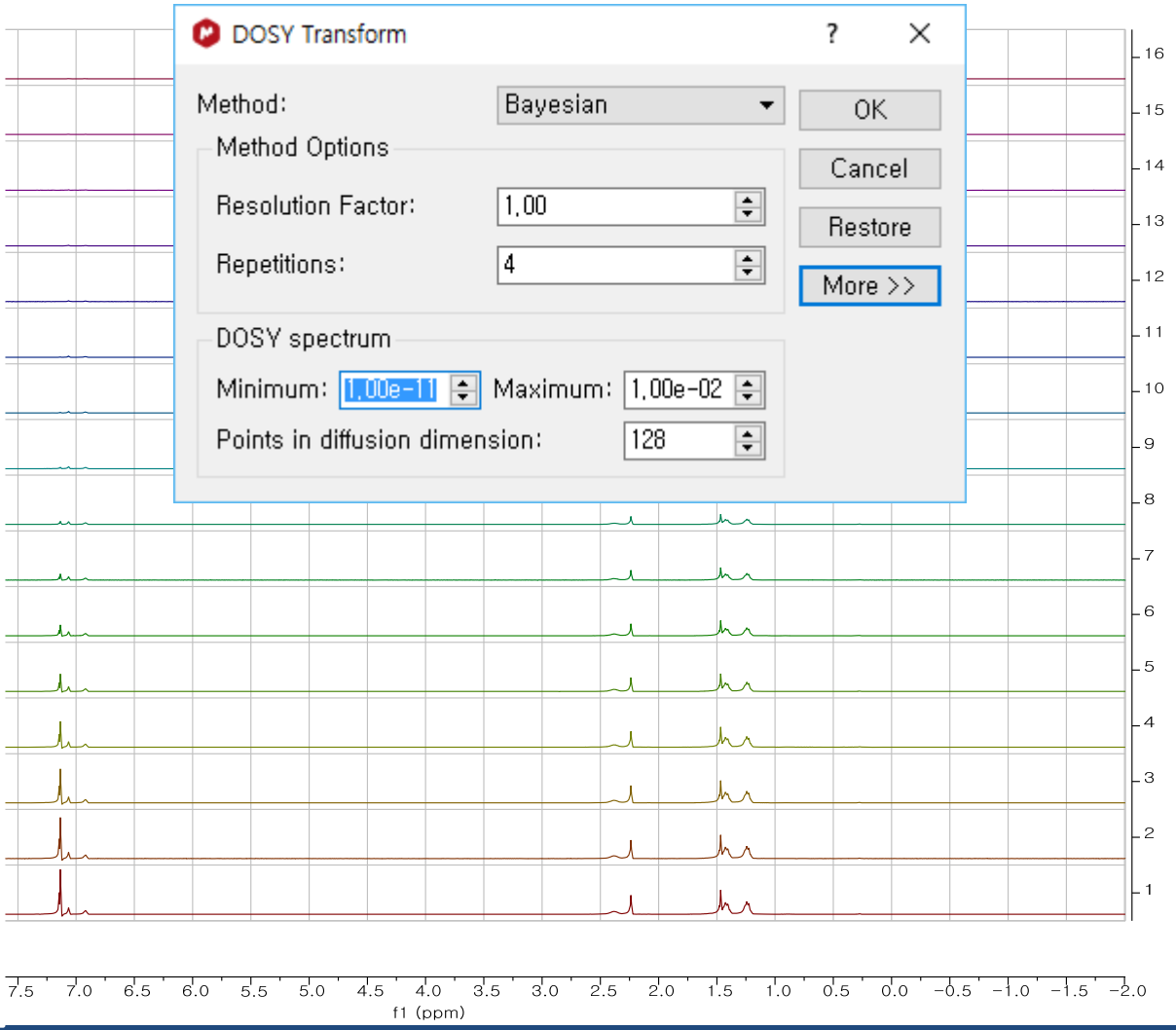
X

Model	X(I) ARR_DATA(I)	Y(X) Integral(6.498360,6.426843)	Y'(X) = 0.699326	Y1(X) Integral(6.156666,
1	0.001	-1050.48	-1065.84	-5314.34
2	0.02	-1034.45	-1035.39	-5302.69
3	0.05	-989.012	-988.146	-5227.87
4	0.2	-748.36	-766.216	-4894.53
5	0.5	-401.29	-386.458	-4295.69
6	0.8	-106.135	-78.5704	-3763.13
7	1	59.1967	93.5891	-3366.82
8	1.3	307.265	310.627	-2885.28
9	2	703.819	670.532	-1809.65
10	2.5	854.426	838.688	-1113.14
11	3	970.827	957.226	-488.092

Acq. Start: 2018-01-04T10:25:20 Acq. End: 2018-01-04T10:25:20



DOSY Transform



DOSY Transform