



Mestrelab Research

chemistry software solutions

Mnova Training – Basics

For Mnova v14.2.0

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Main Topics

- Installation and Activation of Mnova NMR
- Opening and processing 1D ^1H NMR
- Multiplet analysis for 1D ^1H NMR
- Opening and processing 1D ^{13}C NMR
- Peak picking for 1D ^{13}C NMR
- Opening and analyzing LC-MS
- Reporting and publishing results
- Saving the results



Specifics for <xxxxxx> University (To be completed by instructor)

- The instructions for downloading, installing and activating Mnova:
 - <Link to instruction page>

- The Mnova licenses that <XXXX> University has:
 - Mnova Suite (NMR, NMRPredict & MS), unlimited
 - Etc.

- The sample data used in this tutorial are located at:
 - <Link to data folder>.

Installation and Activation of Mnova, and General Setup*

**You will need to have Mnova Suite (NMR, NMRPredict Desktop and MS) licenses for this tutorial.
For the Advanced tutorial, you will also need Mnova DB and Wiley DB licenses.*

INSTALLATION

Install and activate Mnova in General

- Download and install Mnova from www.mestrelab.com. Choose **File > Help > License Manager** to open the License Manager dialog.
- Activate Mnova using your purchased license files, or apply for 45 day free trial licenses (Click **Get/Install Licenses**)
- Make sure that there are green checkmarks for NMR and other plugins that are supposed to be activated
- For managing campus/site/ concurrent licenses, see <http://www.mestrelab.com/mlicserver>

The Host ID for this computer

Location of the license file

Host ID: W2MFH-7CMP31S2-P5L7T-38REL3JE

Licenses

	State	Plug-in	Issued By	Licensed To	Type	Issued Date	Days to Expi	Update Days	V
35	✔	Mnova Verify	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N
36	✔	Mnova qNMR	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N
37	✔	NMR	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N
38	✔	NMRpredict Desktop	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N
39	✔	PhysChem Properties	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N
40	⚙	Plate Processing	Mestrelab Research S.L.	Dr Chen Peng,...	single	Fri Sep 14 2018	681	681	N

Mnova plugin names

License issued date

License expiring date

Service Licenses

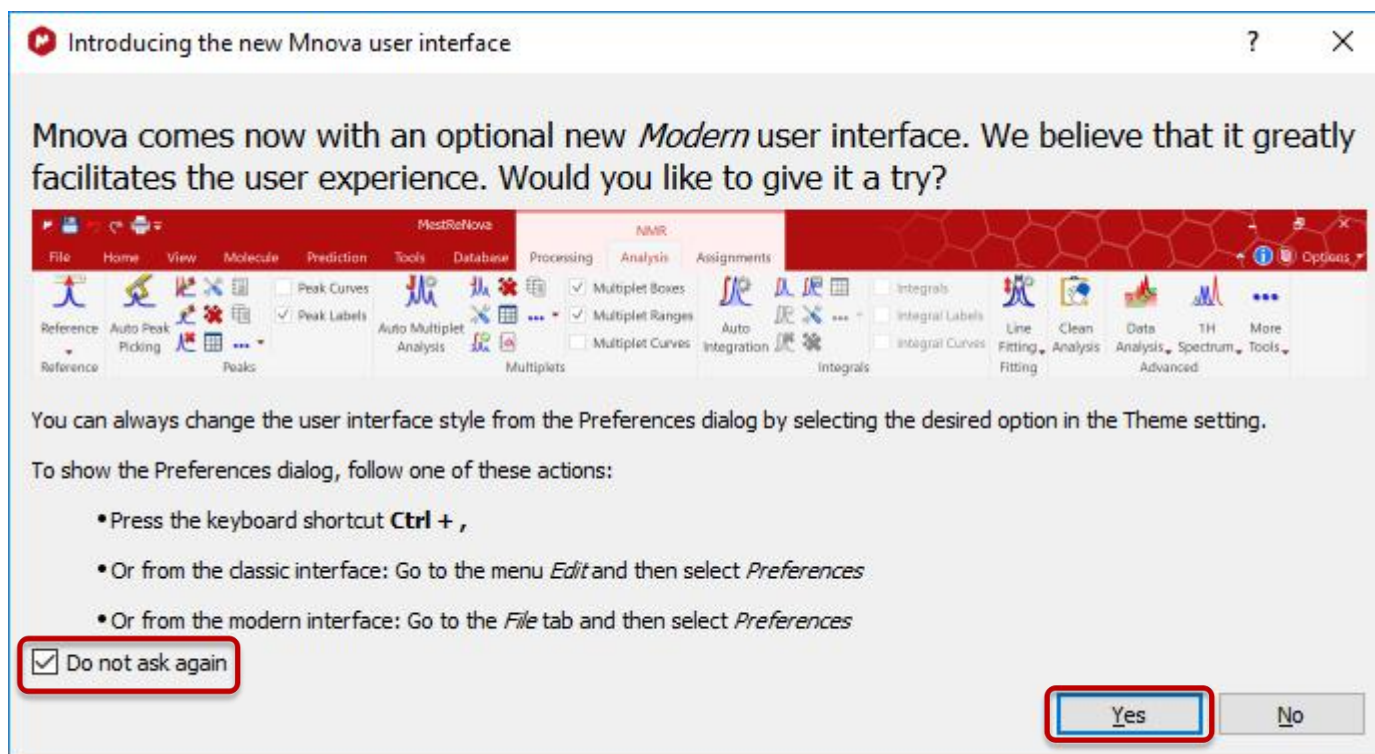
State	Name	Username	Id	Issued Date	Expiry Date	Operations	Tenant Id	Asset Id

Support... Error Summary

Use the New Graphical User Interface (GUI)

PREFERENCES

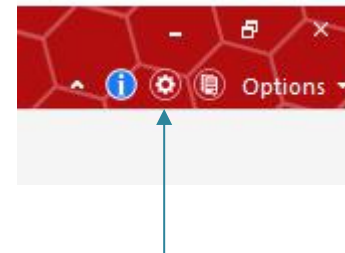
The first time you start Mnova, it asks if you want to use the “Modern” GUI. Choose it and check “Do not ask again”. Restart Mnova, you will enjoy the modular ribbon GUI introduced since version 12.



Note: This instruction is based on the Modern GUI. You can always switch back to the “Classic” GUI from “File/Preferences”.

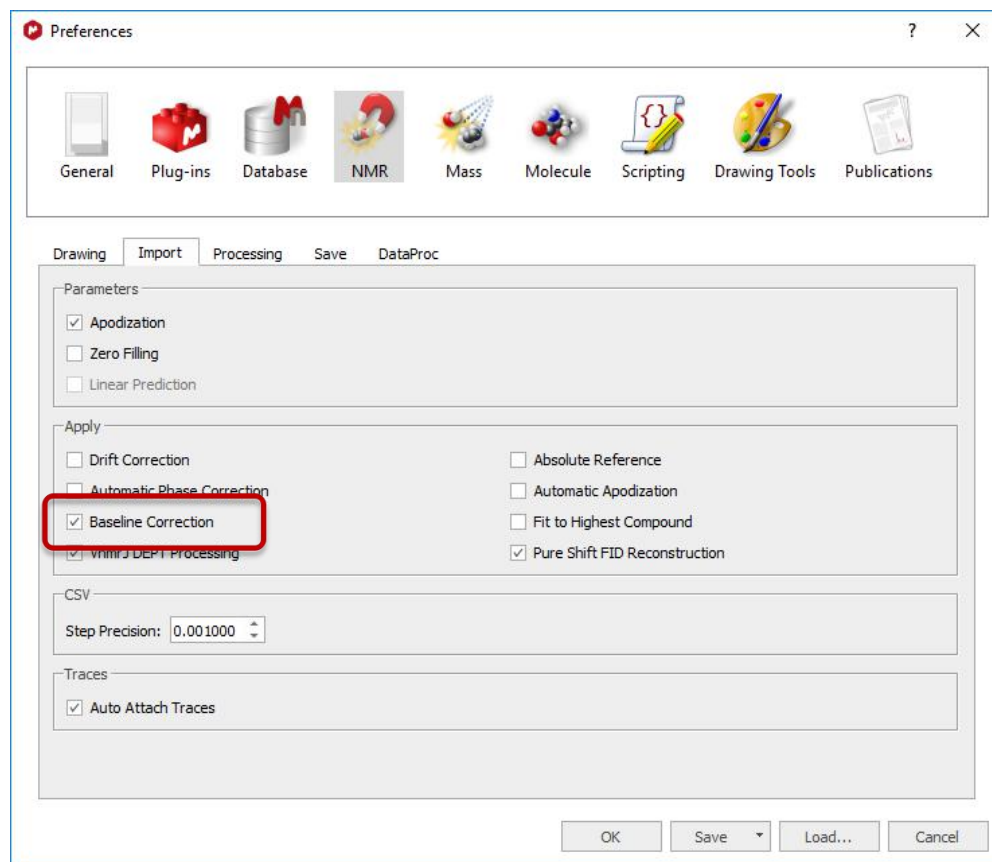
Turn on Auto Baseline Correction

Choose File/Preferences. In the NMR> Import Tab, check Baseline Correction so that baseline is automatically done when you open an NMR spectrum.



Note: Automatic Baseline Correction use the default algorithm of "Bernstein Polynomial with order of 3", or the one that you used previously. This option may make manual phasing of 2D NMR sluggish. In that case, turn baseline off from Processing .

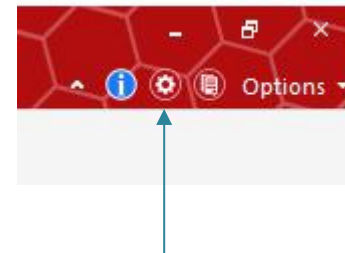
Tip: There are many options and settings that you may change in the Preferences Dialog, especially the resolution of image copying and image exporting in the Drawing Tab.



PREFERENCES

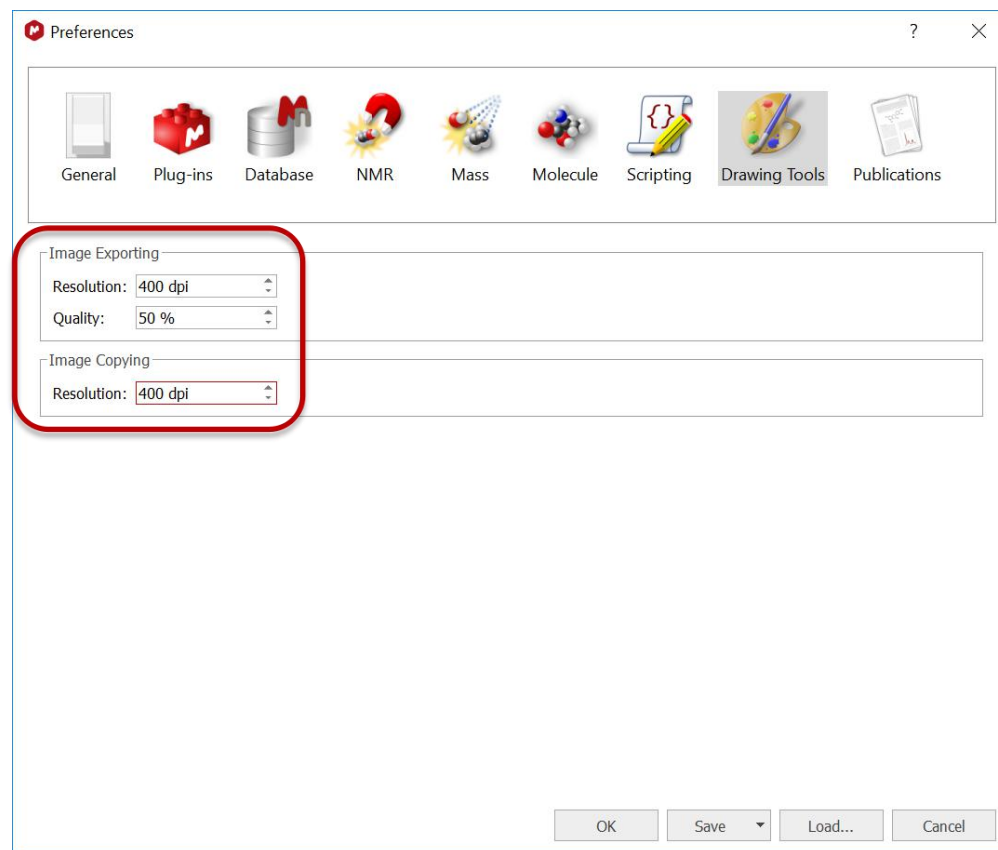
Setup the resolution for publishing spectra

Choose File/Preferences. In the Drawing Tools tab, change the resolutions for Image Exporting and Image Copying to numbers similar to something shown below.



The resolution for Image Exporting is used when you choose File > Save As and save the selected objects in Mnova as a graphical image file.

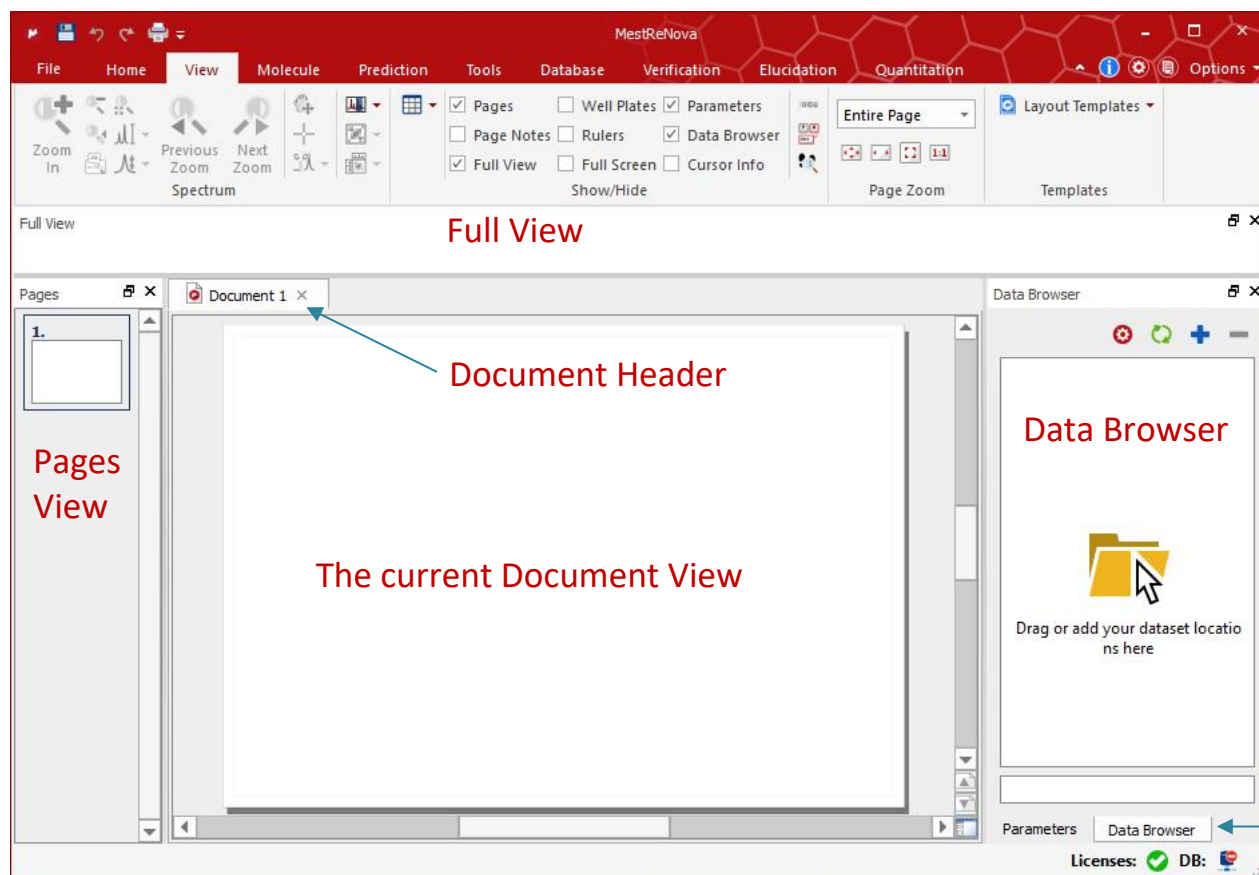
The resolution for Image Copying is used when you copy selected objects in Mnova and paste them to another application.



Setup the Workspace

SETUP

- In the View Ribbon, check the Pages, Full View, Parameters, and Data Browser Views
- Dock and arrange them as shown below



Click here to minimize the ribbon

Document Header

The current Document View

Pages View

Data Browser

Drag or add your dataset locations here

Click here to switch the panels or tables

Setup Data Browser

SETUP

- Click “+” in the Data Browser, navigate to the directory where the sample NMR data are located and click OK to add it.
- Click the Settings button to turn on the display of the meta data, date and time, and enable sorting
- Make sure you see the data files similar to those shown below

The Data Browser window displays a tree view of folders under 'Training Dataset'. The table below shows the files listed in the browser:

Name	Experiment	Comm	Format	Modification date
Training Dataset				2019-11-03T20:18:18
1D and 2D peak assignment				2019-11-03T19:48:53
Brucine 1D and 2D NMR				2019-10-20T19:39:40
Estradiol H1 and HSQC assignment				2019-10-23T22:16:33
Ibuprofen LC-MS and 1D and 2D NMR for assignment				2019-10-21T00:13:20
Strychnine 1D and 2D NMR for assignment				2019-10-31T02:13:33
10	1D-H-zg30	Stryc...	Bruker T...	2019-10-30T19:15:12
11	1D-C-zgpg30		Bruker T...	2019-10-30T19:15:12
12	2D-HH-COSY-co...		Bruker T...	2019-10-30T19:15:13
13	2D-CH-HSQC-E...		Bruker T...	2019-10-30T19:15:13
14	2D-CH-HMBC-h...		Bruker T...	2019-10-30T19:15:14
16	2D-NH-HMBC-h...		Bruker T...	2019-10-30T19:15:14
19	2D-CC-INADEQU...		Bruker T...	2019-10-30T19:15:15
Strychnine.mol				2012-07-16T23:51:54
saved processed.mnova			MestRe...	2019-10-31T02:13:33
1H NMR phase and baseline correction	1D-H-zg		Bruker T...	2019-07-09T18:51:21
Database Search				2019-07-09T18:51:22
MS				2019-07-09T18:51:23
Reaction monitoring				2019-11-03T19:38:53
Results				2019-07-09T18:51:27

The 'Add location' dialog box shows the following fields:

- Path: and Templates/Training for chemists/Training courses by Chen/Training Data Sets
- Label: Training Data Sets

Buttons: OK, Cancel

The 'Data Browser Settings' dialog box shows the following settings:

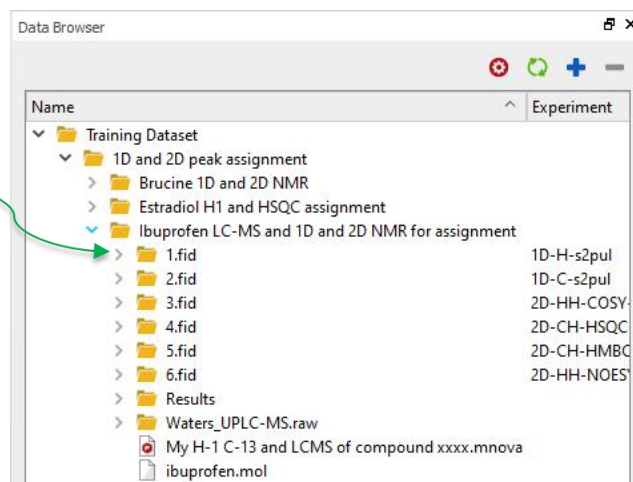
- View:
 - Enable sorting
 - Show date and size
 - Show file meta data
- File Formats:

Format
<input checked="" type="checkbox"/> AB SCIEX Analyst (*.wiff)
<input checked="" type="checkbox"/> AB SCIEX Data Explorer (*.dat *.t2d)
<input checked="" type="checkbox"/> Advion expression CMS (*.datx)

Buttons: Check All, Uncheck All

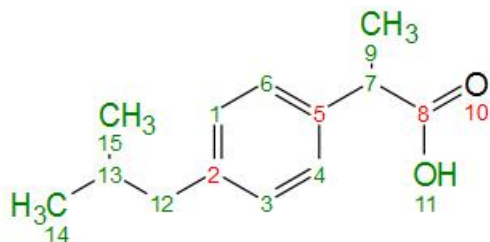
1D ^1H NMR Spectrum Processing, Analysis, and Reporting

Sample data



PROCEDURE

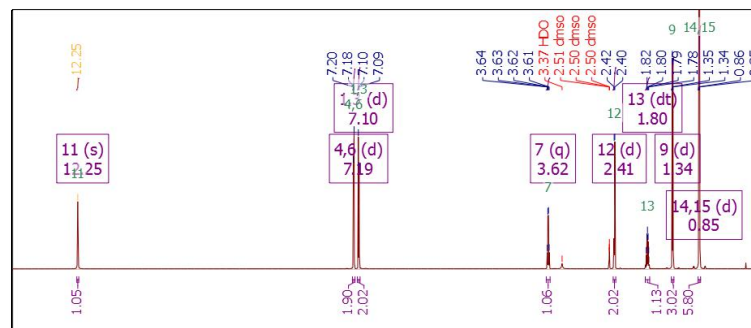
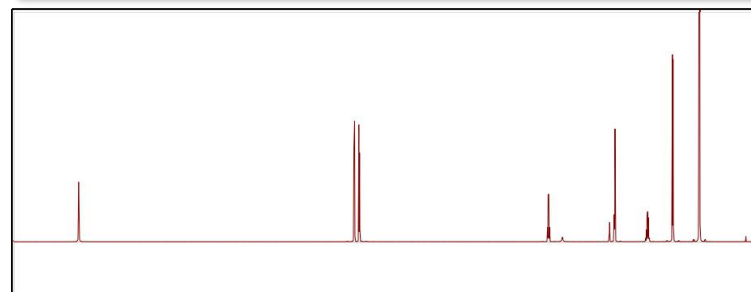
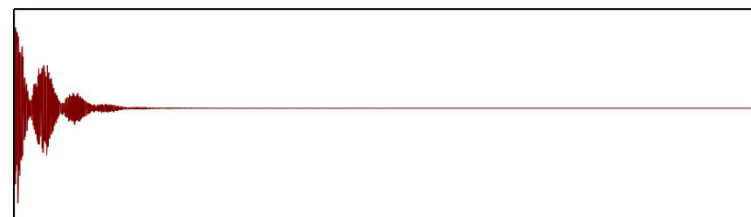
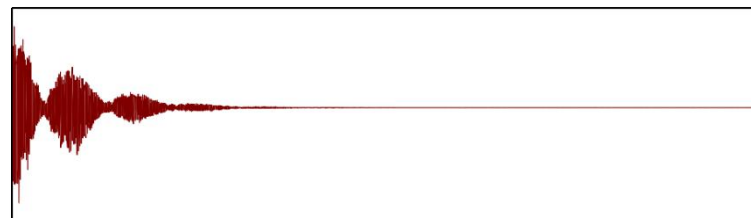
- Open the raw data
- Pre-process the FID: drift correct, apodize, zero fill, linear predict, etc.
- Fourier transform
- Phase correct and baseline correct
- Chemical shift reference
- Peak-pick, integrate, multiplet analysis
- Structure verification and peak assignment
- Report and publish



^1H NMR (600 MHz, DMSO- d_6) δ 12.25 (s, 1H), 7.19 (d, $J = 7.8$ Hz, 2H), 7.10 (d, $J = 7.9$ Hz, 2H), 3.62 (q, $J = 7.1$ Hz, 1H), 2.41 (d, $J = 7.2$ Hz, 2H), 1.80 (dt, $J = 13.5, 6.8$ Hz, 1H), 1.34 (d, $J = 7.1$ Hz, 3H), 0.85 (d, $J = 6.7$ Hz, 6H).

Note: Most of these steps are done automatically by Mnova. However, you retain full control at all times

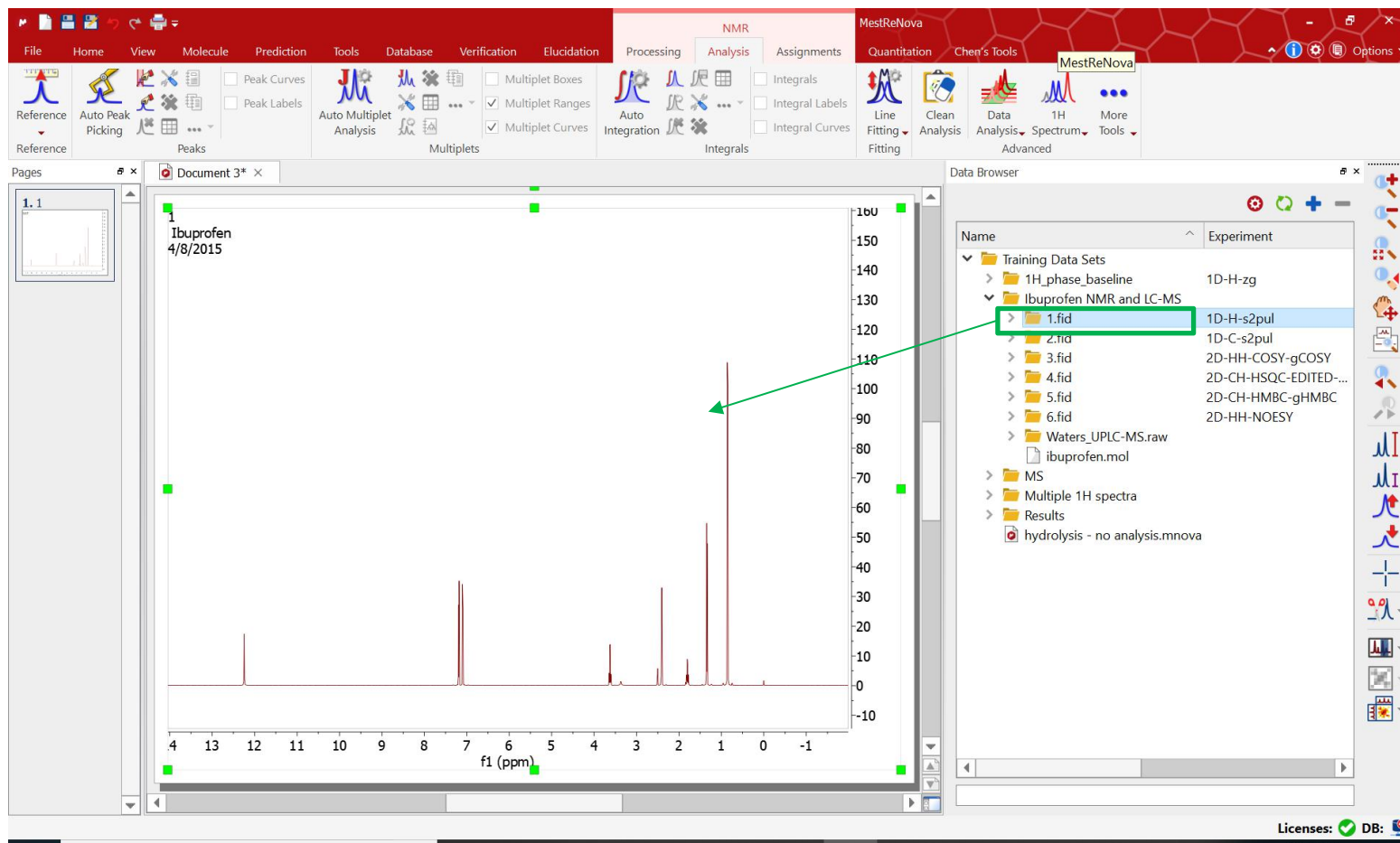
^1H processing and analysis: general procedure



Open a H-1 spectrum

PROCESSING

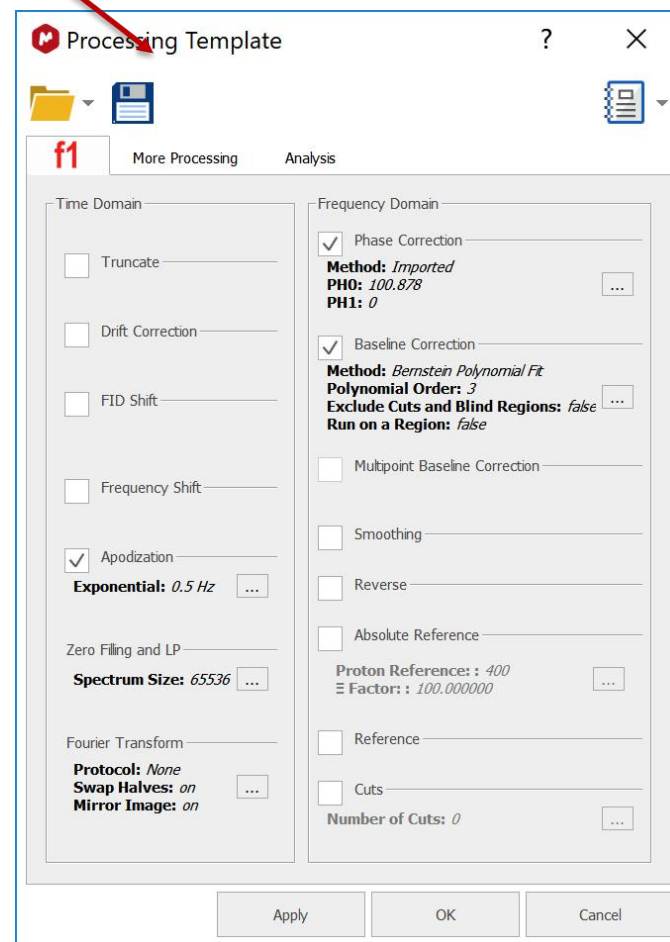
- In Data Browser, expand the folders Training Datasets > Ibuprofen NMR and LC-MS, and drag the “1.fid” folder (1D H-1 spectrum) to the main window.
- Notice the H-1 spectrum is automatically processed and displayed.



PROCESSING

- In most cases, Mnova processes the spectrum automatically using the parameters from the instrument. The spectrum should be well-processed if the original processing parameters were well set. The Processing Tab is for you to re-process the spectrum when needed.
- Choose Processing > Processing Template to verify the processing parameters. Make sure they look the same as displayed on the right.
- Click OK or Apply to re-process the spectrum.

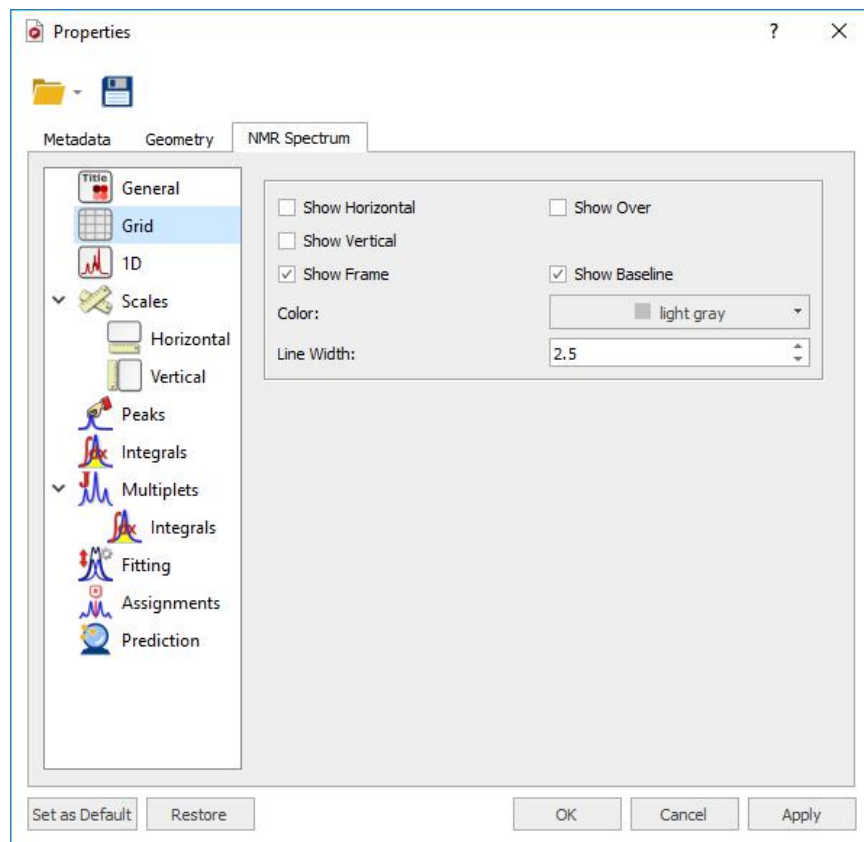
Verify the processing parameters



Change the Display Properties

DISPLAY

- Right click on the spectrum* and choose Properties to open the Properties Dialog, view the properties that can be changed.
- In the Grid Category, uncheck Show Horizontal, and Show Vertical, check Show Baseline
- Click Apply, and then Set as Default to apply the settings to 1D spectra opened in the future

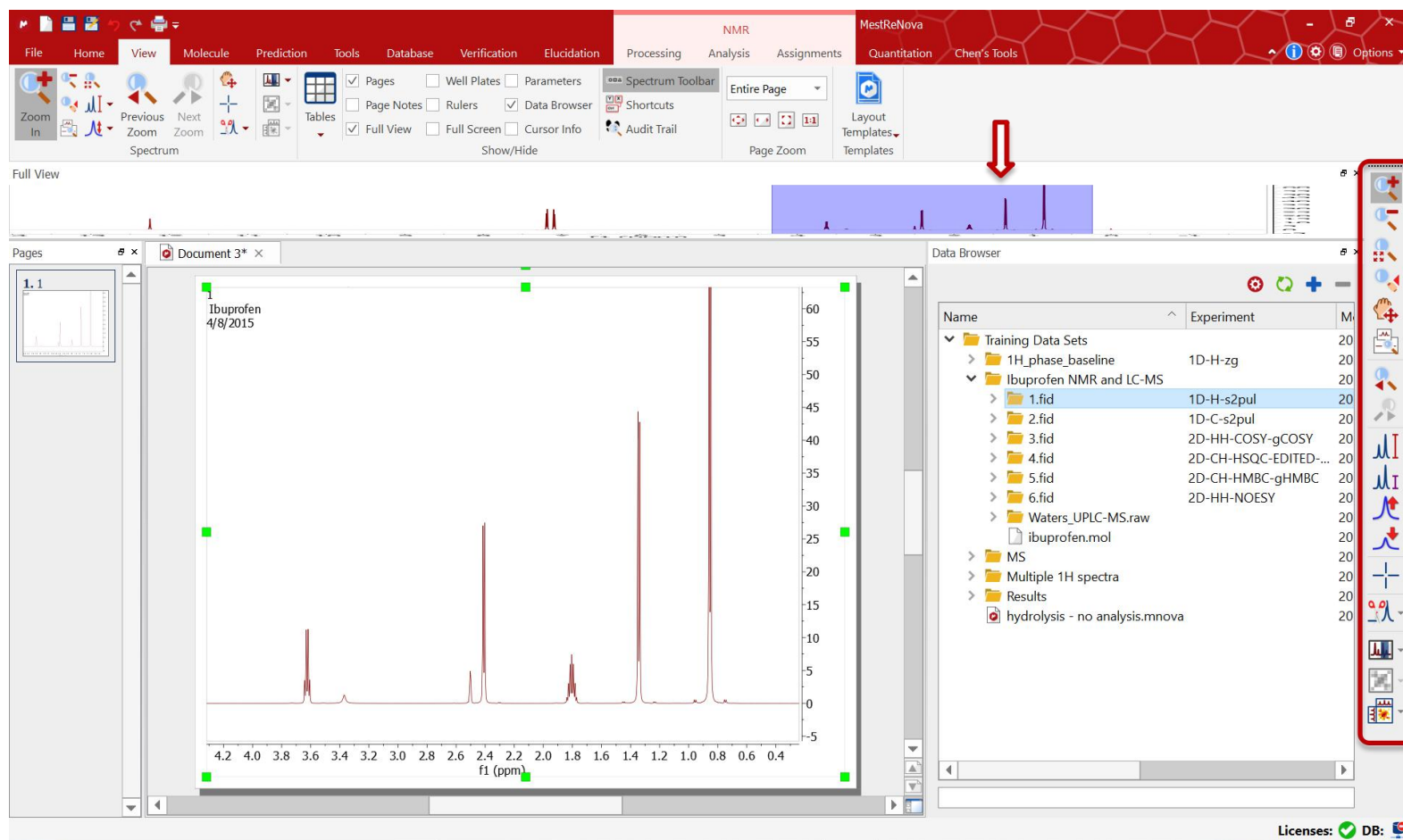


**Starting from Version 14, double-clicking changes the display to full-spectrum if you are in the default pointer mode.*

Navigate in the H-1 Spectrum
















VISUALIZATION

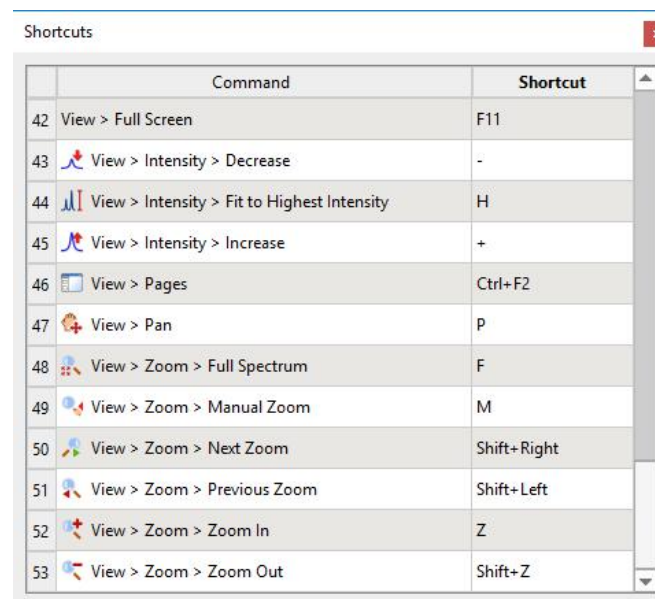
- Use the Spectrum Toolbar to zoom in/out, pan, and change the Y scale (see next slide for details)
- Use the Full View to move to different zoom in area (click or drag)



Spectrum visualization tools

- The Spectrum Toolbar is visible only after you open a spectrum.
- Learn some short-cut keys by choosing View > Shortcuts

	Zoom in/Zoom out (or press Z) *
	Zoom out**
	Full spectrum (or press F)
	Manual Zoom in to defined ppm range
	Pan spectrum (or press P) ***
	Expansion – click&drag to draw an inset (or press E)
	Previous Zoom level
	Next Zoom level
	Fit to Highest Intensity (or press H)
	Fit to highest compound peak
	Increase Intensity (or rotate mouse wheel)
	Decrease Intensity (or rotate mouse wheel)
	Crosshair Cursor (or press C) for measuring <i>J</i> -couplings
	Cut (or press X) to hide parts of the spectrum
	Edit Blind regions



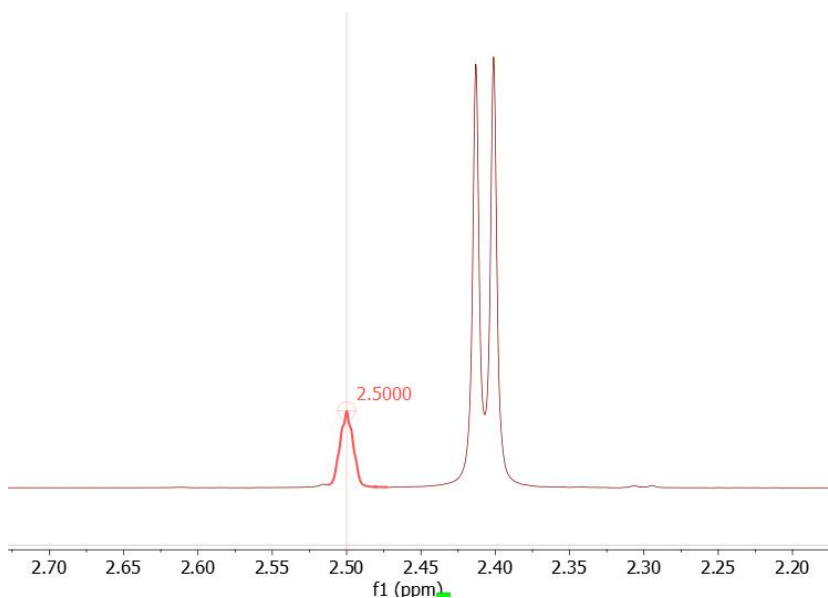
	Command	Shortcut
42	View > Full Screen	F11
43	View > Intensity > Decrease	-
44	View > Intensity > Fit to Highest Intensity	H
45	View > Intensity > Increase	+
46	View > Pages	Ctrl+F2
47	View > Pan	P
48	View > Zoom > Full Spectrum	F
49	View > Zoom > Manual Zoom	M
50	View > Zoom > Next Zoom	Shift+Right
51	View > Zoom > Previous Zoom	Shift+Left
52	View > Zoom > Zoom In	Z
53	View > Zoom > Zoom Out	Shift+Z

* Press **Z** several times to toggle between horizontal/vertical/box zoom

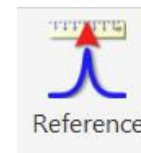
** Press **P** several times to toggle between free/horizontal/vertical panning

ANALYSIS

- This spectrum uses DMSO-d6 as the solvent. We can reference the chemical shifts by setting its middle peak to 2.5 ppm.
- Zoom to the DMSO peak at around 2.5 ppm. Choose Analysis > Reference, and click on the top of the middle peak.
- Set it to 2.5 ppm either manually or from the Solvent List.



Chemical Shift Referencing



Reference along f1

Old Shift: 2.5021 ppm Auto Tuning
New Shift: 2.5000 ppm Range Width: 0.1000 ppm
 Annotation: DMSO-d6


Solvent List

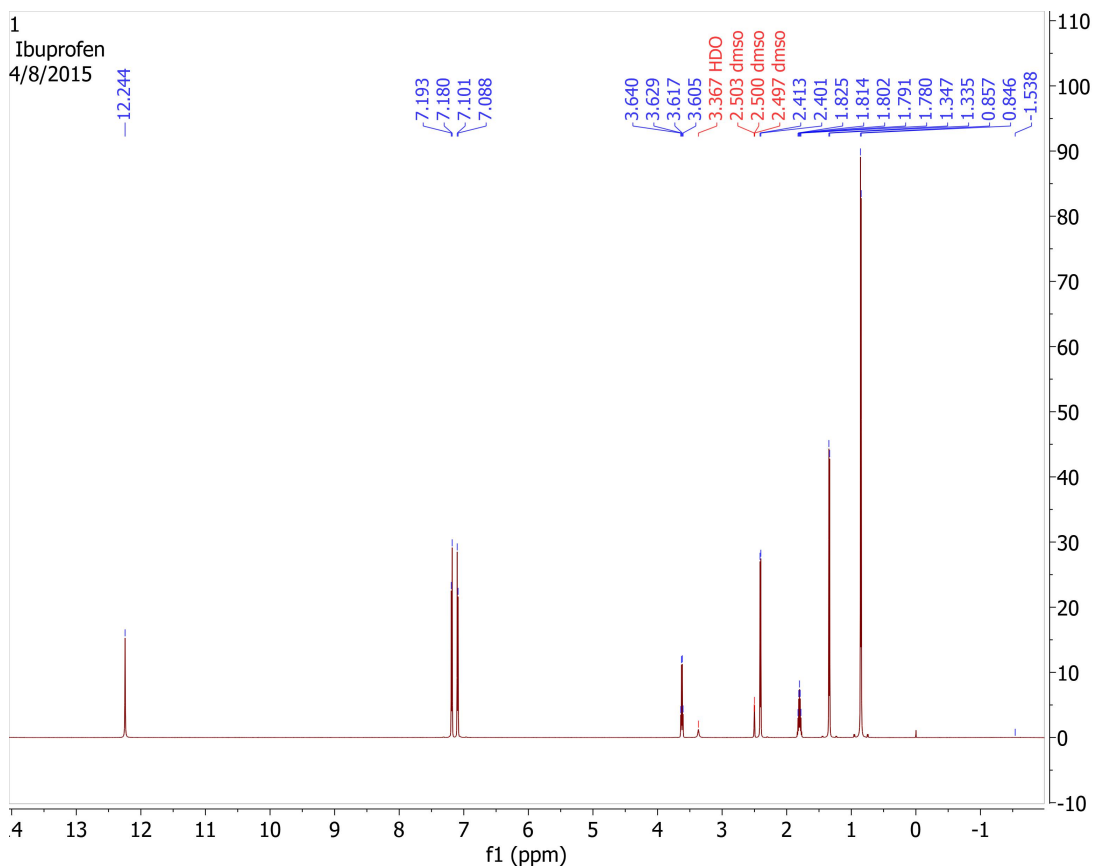
Name	Shift (ppm)	Multiplicity	J (Hz)
Deuterium Oxide	4.790	1	
Dimethyl Sulfoxide-d6	2.500	5	1
	3.330	1	
Ethanol-d6	5.290	1	

Restore Defaults Add... Edit... Delete

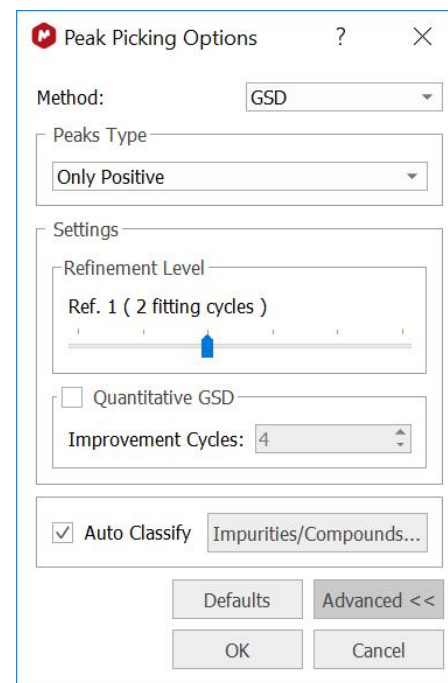
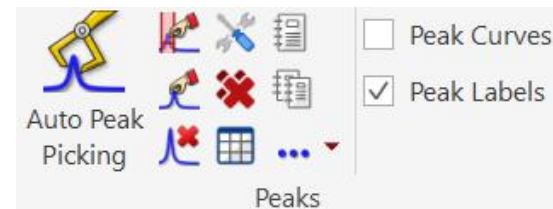
OK Cancel Solvents <<

ANALYSIS

- Click the Peaks > Options  to verify the peak picking options. Default settings are used here as shown to the right.
- Click the Auto Peak Picking tool to pick all the peaks
- Using other peak picking tools to display/delete/add/change peaks as needed.




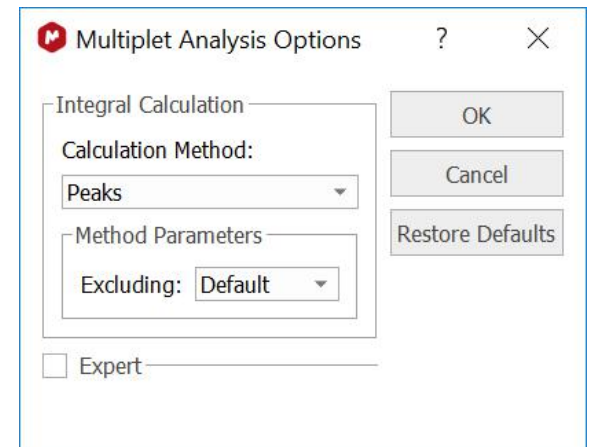
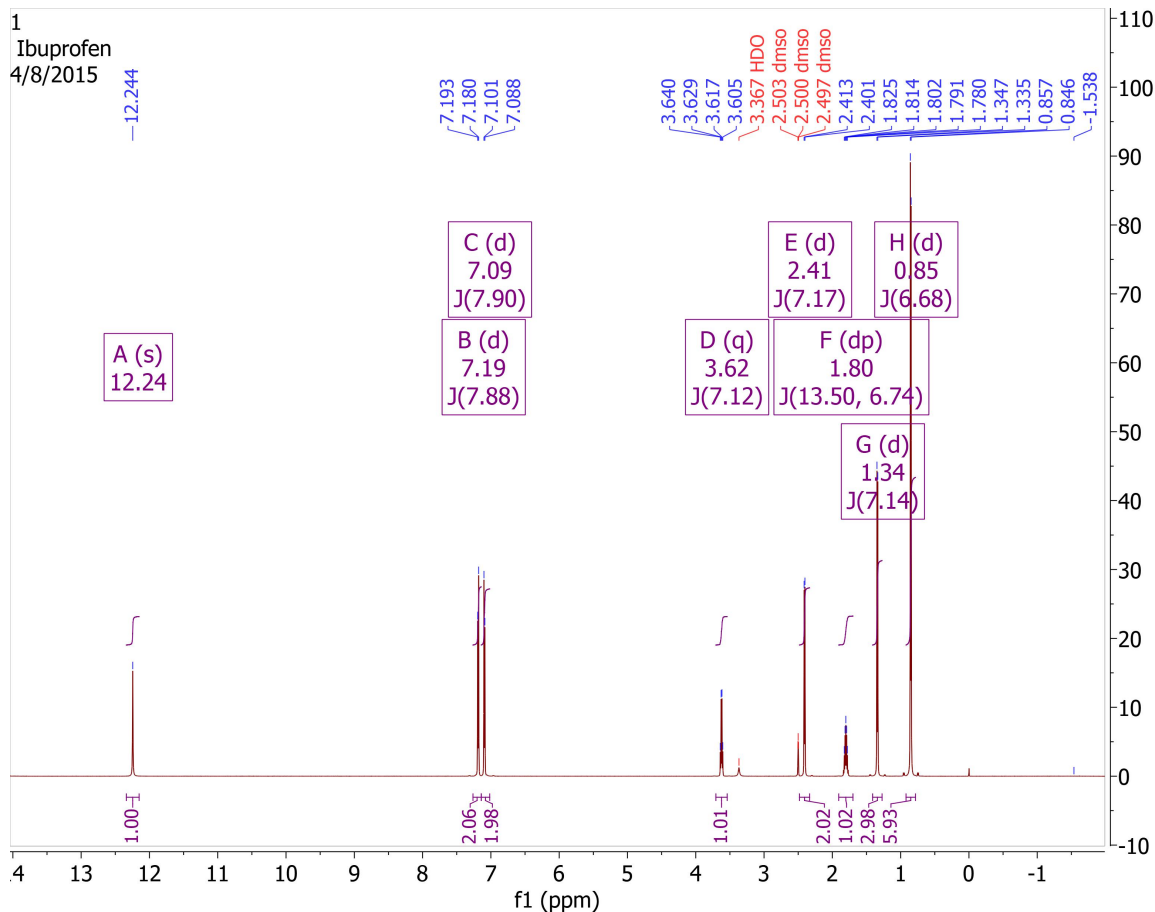
Peak picking



Multiplet analysis

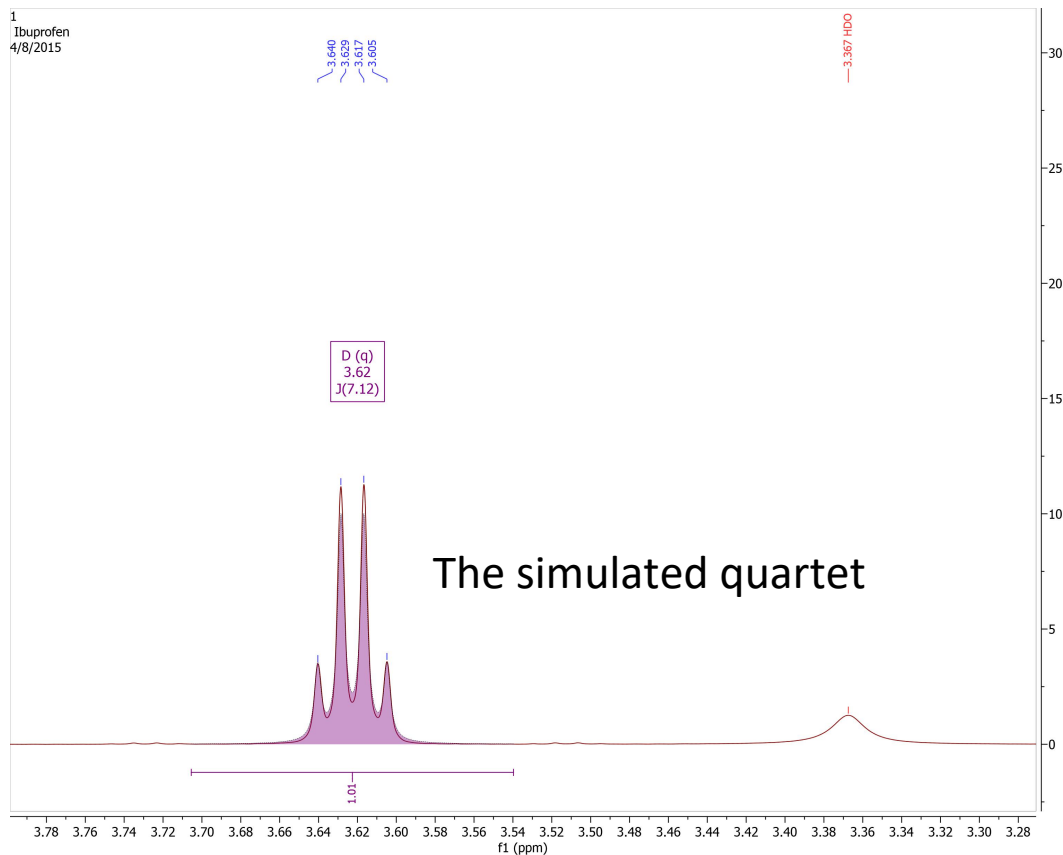
ANALYSIS

- Click the Multiplets > Options  to verify the multiplet analysis options. Default settings are used here as shown to the right.
- Click the Auto Multiplet Analysis tool to do the multiplet analysis based on the picked peaks



ANALYSIS

- Double click on a multiplet label to open the Multiplet Manager.
- Use the tools there to verify and change multiplet analysis results if needed.



Multiplet Manager

Multiplet Manager

3.62 (1H, q, J=7.1 Hz)

Name: D Class: q

δ: 3.623 ppm Middle

J-List: 7.14, 7.12, 7.12 Discard Peaks

Color: Purple

Total Nuclides = 18

Nuclides: 1 Auto

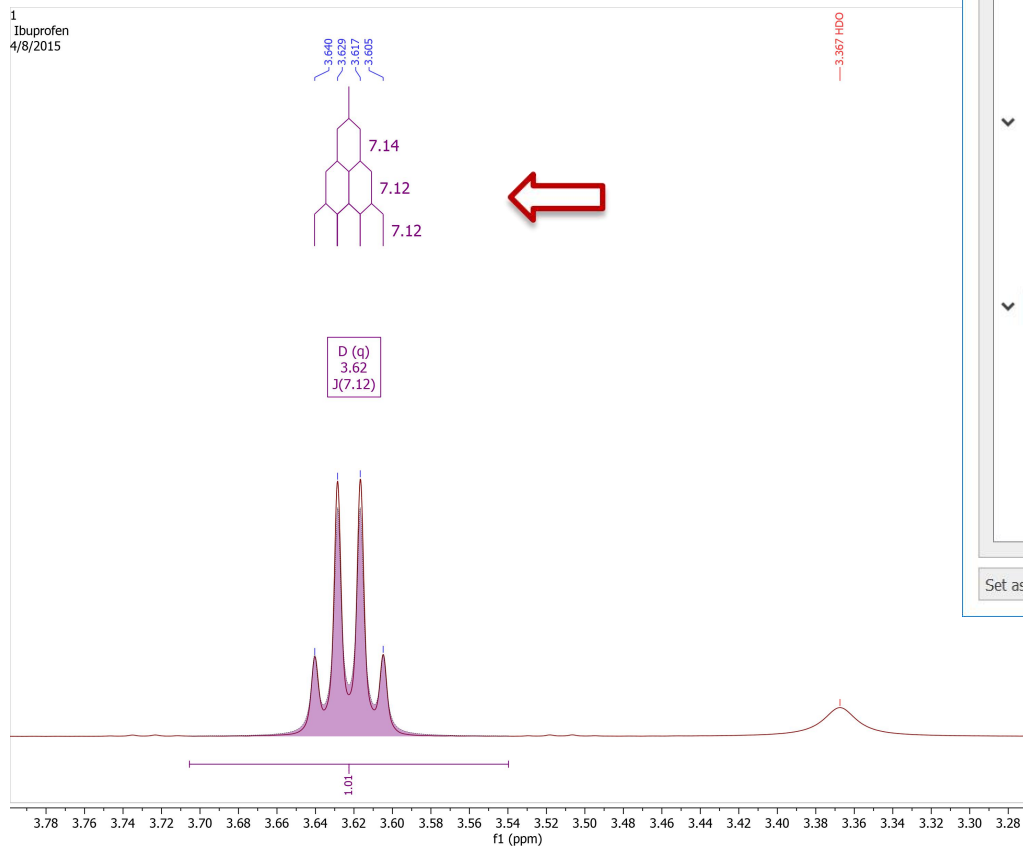
Integral: 1.01

Absolute: 687.448

From: 3.705 To: 3.540

ANALYSIS

- Double click on the spectrum to open the Properties dialog.
- Choose Multiplets, and check J's Tree to display the J-coupling tree for visual verification of the multiplet analysis results.



Multiplet Manager

Properties

Metadata Geometry **NMR Spectrum**

General
Grid
1D
Scales
Horizontal
Vertical
Peaks
Integrals
Multiplets
Integrals
Fitting
Assignments
Prediction

Multiplets Labels

Font: MS Shell Dlg 2

Line Width: 2.5

Label: Name (Category) / Shift / J's

Shift Decimals: 2

Js Decimals: 2

Margin: 55 %

Follow Peak Visibility Rules Show Label Box

J's Tree

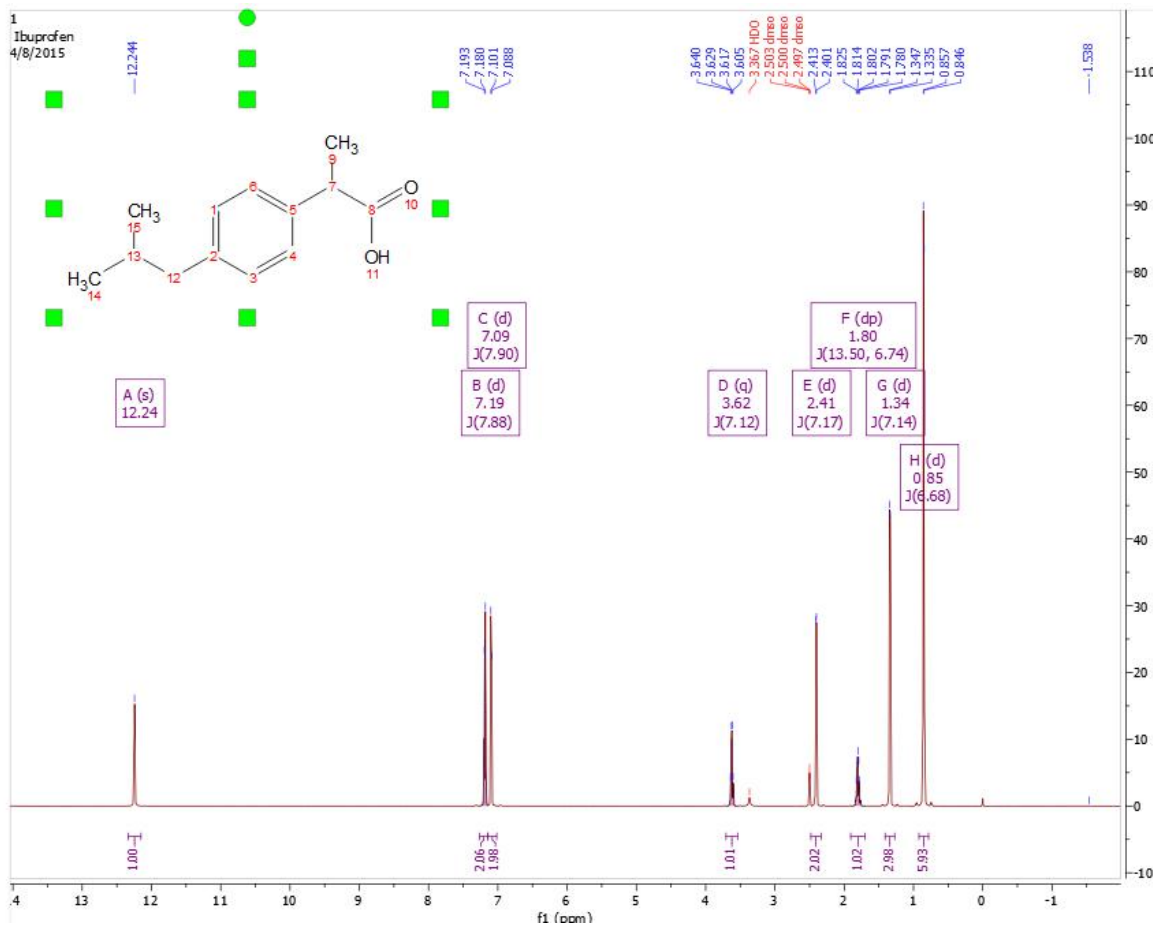
Show Label

Set as Default Restore OK Cancel Apply

Verify the number of Hs

ANALYSIS

- Open the Ibuprofen.mol file from the Data Browser.
- Note the number of protons from multiplet analysis vs. that from the structure



Multiplet Manager

3.62 (1H, q, J=7.1 Hz)

Name: D Class: q

δ: 3.623 ppm Middle

J-List: 7.14, 7.12, 7.12 Discard Peaks

Color: Purple

Total Nuclides = 18 (18 in molecule) ←

Nuclides: 1 Auto

Integral: 1.01

Absolute: 687.448

From: 3.705 To: 3.540

PUBLISHING

- Use the Multiplet Table tool to display the Multiplets Table.
- Click Setup Report to change the reporting format
- Click Report to report the multiplets texts

Multiplets

Report Multiplets Copy Multiplets Setup Report Delete

¹H NMR (DMSO-*d*₆, 600 MHz) δ 12.24 (1H, s), 7.19 (2H, d, *J*=7.9 Hz), 7.09 (2H, d, *J*=7.9 Hz), 3.62 (1H, q, *J*=7.1 Hz), 2.41 (2H, d, *J*=7.2 Hz), 1.80 (1H, dp, *J*=13.5, 6.7 Hz), 1.34 (3H, d, *J*=7.1 Hz), 0.85 (6H, d, *J*=6.7 Hz)

	arr	Shift	Range	H's	ntegra	Class	J's
1	H (d)	0.85	0.92 .. 0.78	6	5.93	d	6.68
2	G (d)	1.34	1.41 .. 1.27	3	2.98	d	7.14
3	F (dp)	1.80	1.91 .. 1.70	1	1.02	dp	6.74, 6.74, 6.75, 6.7...
4	E (d)	2.41	2.48 .. 2.33	2	2.02	d	7.17
5	D (q)	3.62	3.71 .. 3.54	1	1.01	q	7.12, 7.12, 7.14
6	C (d)	7.09	7.14 .. 7.02	2	1.98	d	7.90
7	B (d)	7.19	7.26 .. 7.14	2	2.06	d	7.88
8	A (s)	12.24	12.34 .. 12.15	1	1.00	s	

Report the multiplets

Auto Multiplet Analysis

Multiplets

- Multiplet Boxes
- Multiplet Ranges
- Multiplet Curves

Multiplet R... ? X

J. Nat. Prod.
Angew. Chem.
J. Am. Chem. Soc.
J. Med. Chem.
J. Nat. Prod.
Japanese Patent
Organometallics
Polyhedron
RSC
Tetrahedron
Tetrahedron Letters

Use Extended Solvent Names

Report Assignments

Shift Number of Decimals: 2

Js Number of Decimals: 1

Fill Style: Transparent

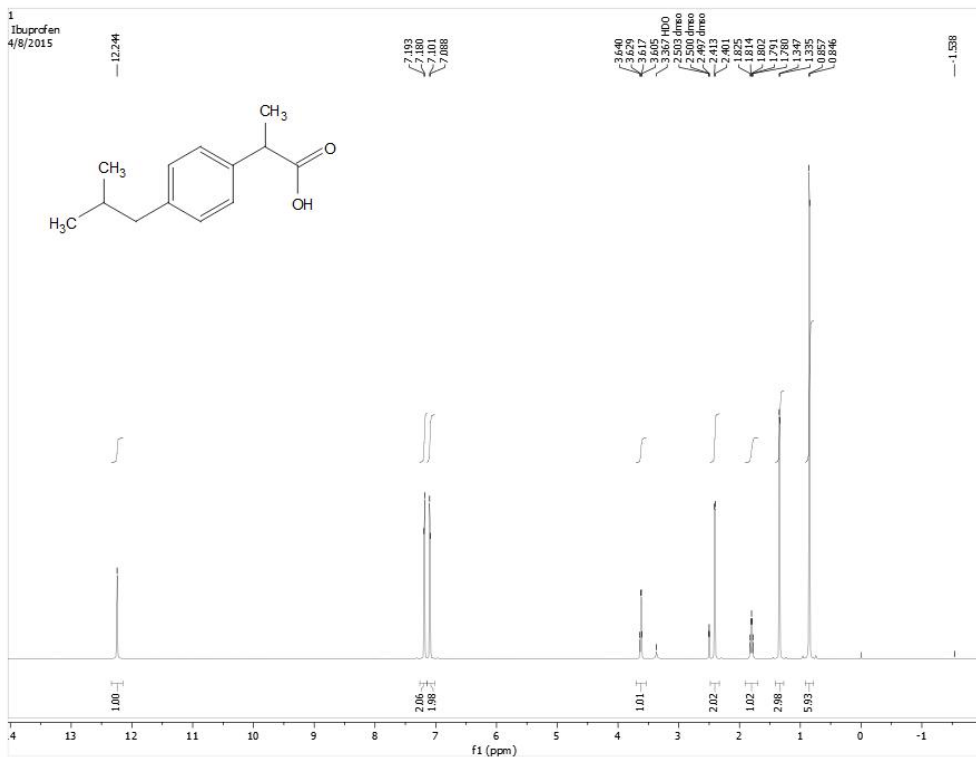
OK Cancel

¹H NMR (DMSO-*d*₆, 600 MHz) δ 12.24 (1H, s), 7.19 (2H, d, *J*=7.9 Hz), 7.09 (2H, d, *J*=7.9 Hz), 3.62 (1H, q, *J*=7.1 Hz), 2.41 (2H, d, *J*=7.2 Hz), 1.80 (1H, dp, *J*=13.5, 6.7 Hz), 1.34 (3H, d, *J*=7.1 Hz), 0.85 (6H, d, *J*=6.7 Hz)

PUBLISHING

- To publish the spectrum on a black and white journal, double click the spectrum to open the Properties Dialog, and set the 1D properties to as shown on the right.
- Choose other properties to display, such as the peak labels, multiplet labels, integrals, etc.
- Copy the spectrum and structure objects and paste them to other documents, such as MicroSoft Word or PPT.

Publishing a spectrum



Properties

Metadata Geometry **NMR Spectrum**

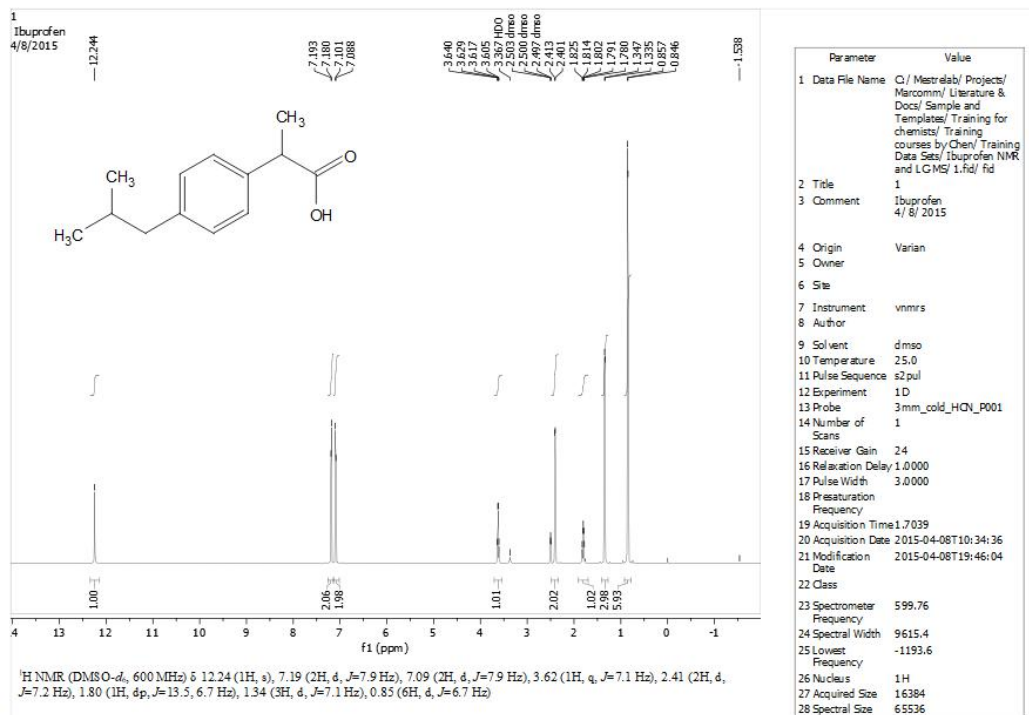
General
Grid
1D
Scales
Horizontal
Vertical
Peaks
Integrals
Multiplets
Integrals
Fitting
Assignments
Prediction

Style: Line
Color: black
Line Width: 1.0

Set as Default Restore OK Cancel Apply

PUBLISHING

- Check View > Parameters Table to display the Parameters Table, and report the parameters on the spectrum. Manually resize the text box to similar to as shown below.
- Report the multiplets and resize the box to as shown below.



Display the parameters

Parameters

Report Copy Setup Customize

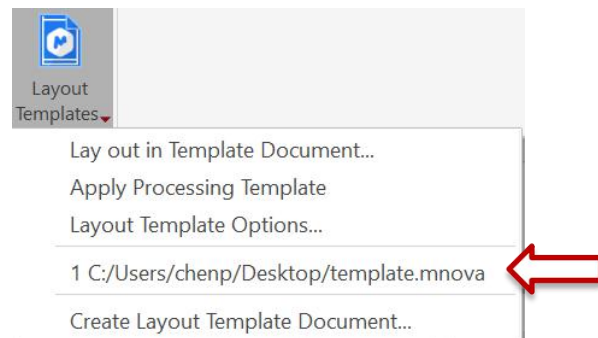
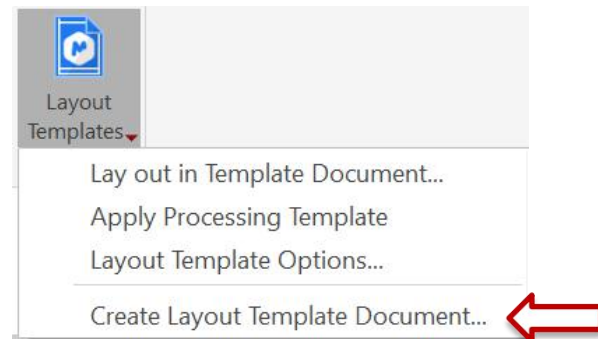
	Parameter	Value
1	Data File Name	C:/Mestrelab/Projects/Marcomm/Liter...
2	Title	1
3	Comment	Ibuprofen 4/8/2015
4	Origin	Varian
5	Owner	
6	Site	
7	Instrument	vnmrs
8	Author	
9	Solvent	dmsol
10	Temperature	25.0
11	Pulse Sequence	s2pul
12	Experiment	1D
13	Probe	3mm_cold_HCN_P001
14	Number of Scans	1
15	Receiver Gain	24
16	Relaxation Delay	1.0000
17	Pulse Width	3.0000

PUBLISHING

- Click on View > Layout Template and choose Create Layout Template to save a layout template. You can edit it.
- Choose File > New and open the H-1 spectrum again, and choose View > Layout Template > [Saved Template Name] to apply it.



Create a layout template

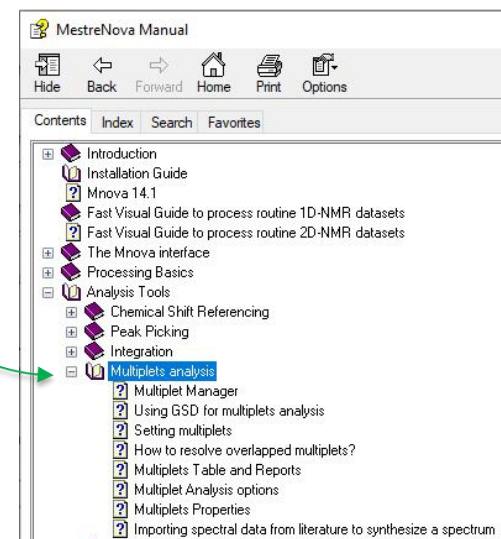


Tip: Mnova uses all pages in the document to create the layout template. So if you have multiple pages, make sure you delete the unwanted ones before creating the layout template.

PROCESSING

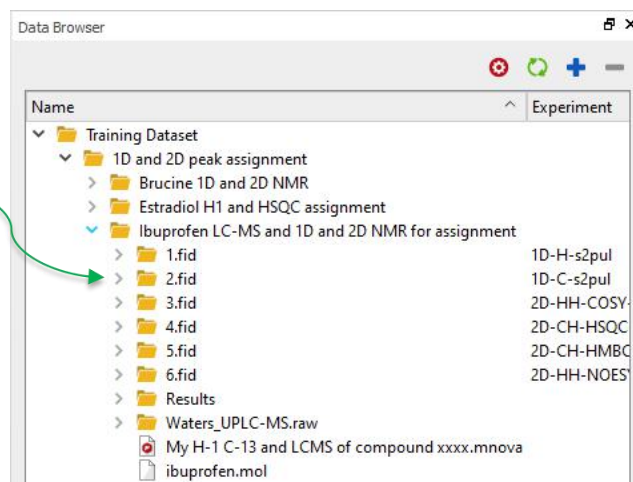
More about H-1 processing

- There are several other 1D H-1 NMR spectra in the tutorial datasets that you can use for practice.
- When the spectrum is more crowded and has more artefacts or impurity peaks, you can also use the manual multiplet analysis tools to have more control
- There are many other ways to correct the multiplet analysis results, such as splitting and assigning individual lines to different multiplets. See File > Help > Contents > Analysis Tools > Multiplet analysis for more details.
- If you start the auto multiplet analysis without any peaks, it will do a peak picking automatically
- Integration is always done automatically during multiplet analysis. If you do manual integration before multiplet analysis, the integration regions will be used for multiplet analysis, and the integration values will be retained as the integrals of the resulting multiplets.



1D ^{13}C NMR Spectrum Processing, Analysis, and Reporting

Sample data



Open a C-13 spectrum

PROCESSING

➤ In Data Browser, open the C-13 spectrum of Ibuprofen.

The screenshot displays the Mestrelab Research software interface. The top menu bar includes File, Home, View, Molecule, Prediction, Tools, Database, Verification, Elucidation, Processing, Analysis, Assignments, Quantitation, and Chen's Tools. The ribbon below the menu bar contains various tool icons for Reference, Auto Peak Picking, Peaks, Auto Multiplet Analysis, Multiplets, Auto Integration, Integrals, Line Fitting, Clean Analysis, Data Analysis, Spectrum, and More Tools.

The main window is divided into several sections. On the left, there are two small preview windows labeled '1. (1) 1' and '2. 2'. The central area shows a large plot of the C-13 NMR spectrum for Ibuprofen, with the x-axis labeled 'f1 (ppm)' ranging from 230 to -10 and the y-axis ranging from -2000 to 26000. The spectrum shows several sharp peaks, with a prominent one at approximately 20 ppm. A green arrow points from the '2.fid' file in the Data Browser to this peak in the spectrum.

On the right side, the 'Data Browser' panel is open, showing a tree view of files and folders. The 'Ibuprofen NMR and LC-MS' folder is expanded, and the '2.fid' file is highlighted with a green box. The table below shows the following data:

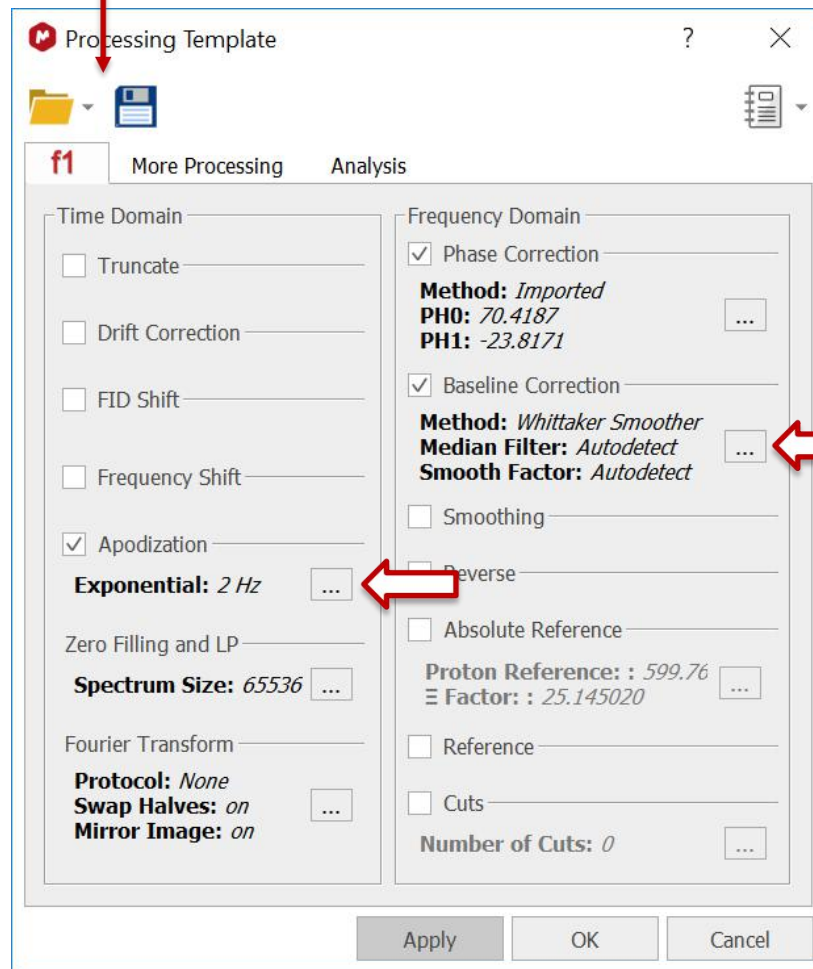
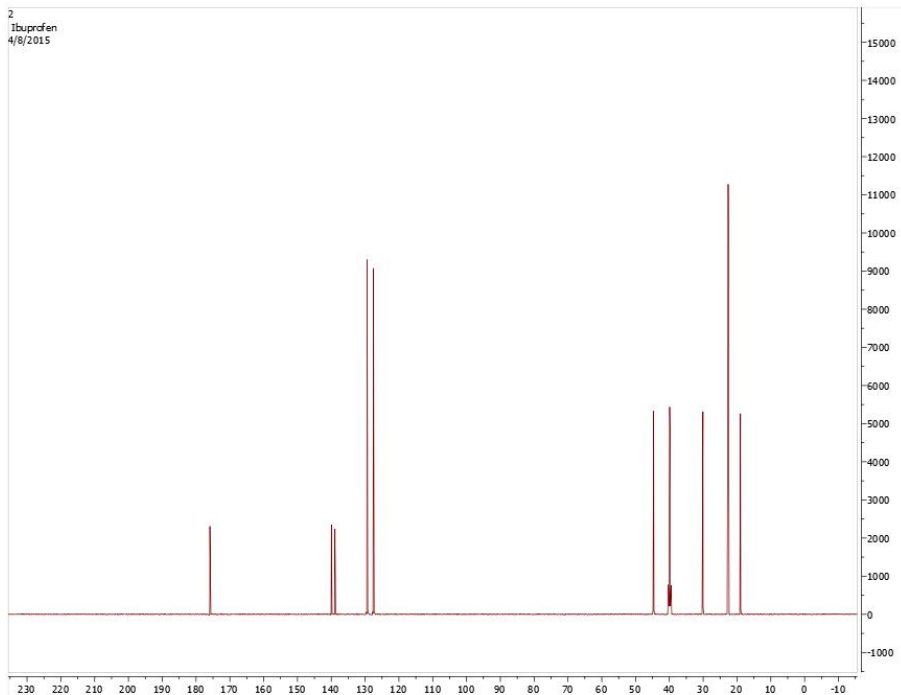
Name	Experiment	M
Training Data Sets		20
1H_phase_baseline	1D-H-zg	20
Ibuprofen NMR and LC-MS		20
1.fid	1D-H-s2pul	20
2.fid	D-C-s2pul	20
3.fid	2D-HH-COSY-gCOSY	20
4.fid	2D-CH-HSQC-EDITED...	20
5.fid	2D-CH-HMBC-gHMBC	20
6.fid	2D-HH-NOESY	20
Waters_UPLC-MS.raw		20
ibuprofen.mol		20
MS		20
Multiple 1H spectra		20
Results		20
hydrolysis - no analysis.mnova		20

At the bottom of the interface, there are tabs for 'Multiplet Manager', 'Multiplets', 'Data Browser', and 'Peaks'. The 'Data Browser' tab is currently active. The status bar at the bottom right shows 'Licenses: DB'.

PROCESSING

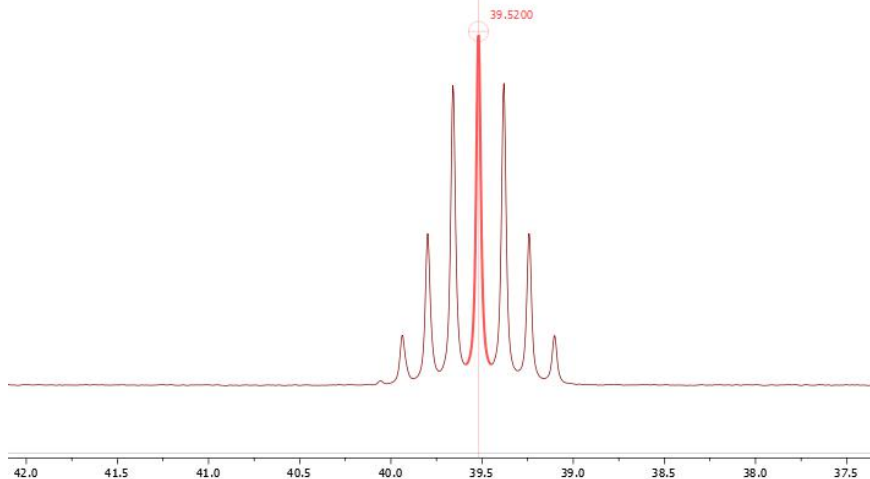
- Choose Processing > Processing Template, and set the parameters similar to the ones shown to the right.
- Click OK or Apply to re-process the spectrum.

Verify the processing parameters

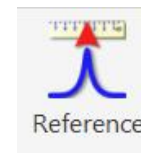


ANALYSIS

- This spectrum uses DMSO-d6 as the solvent. We can reference the chemical shifts by setting its middle peak to 39.52 ppm.
- Zoom to the DMSO peak at around 39 ppm. Choose Analysis > Reference, and click on the top of the middle peak.
- Set it to 39.52 ppm either manually or from the Solvent List.



Chemical Shift Referencing



Reference along f1

Old Shift: 39.9239 ppm Auto Tuning
 New Shift: 39.5200 ppm Range Width: 0.1000 ppm

Annotation DMSO-d6


Solvent List

Name	Shift (ppm)	Multiplicity
Cyclohexane-d12	26.430	5
Dimethyl Sulfoxide-d6	39.520	7
Ethanol-d6	56.960	5
	17.310	7

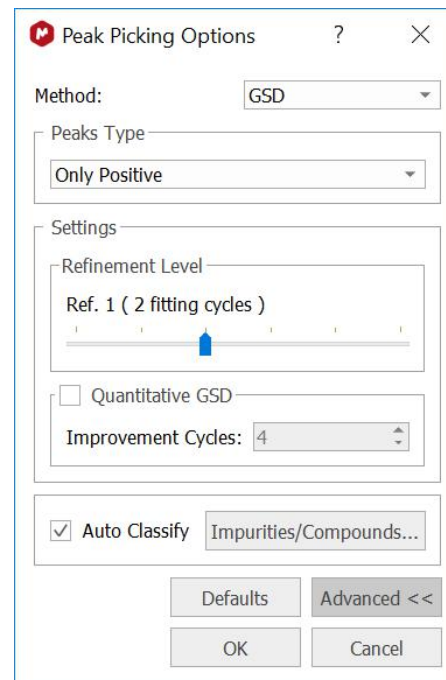
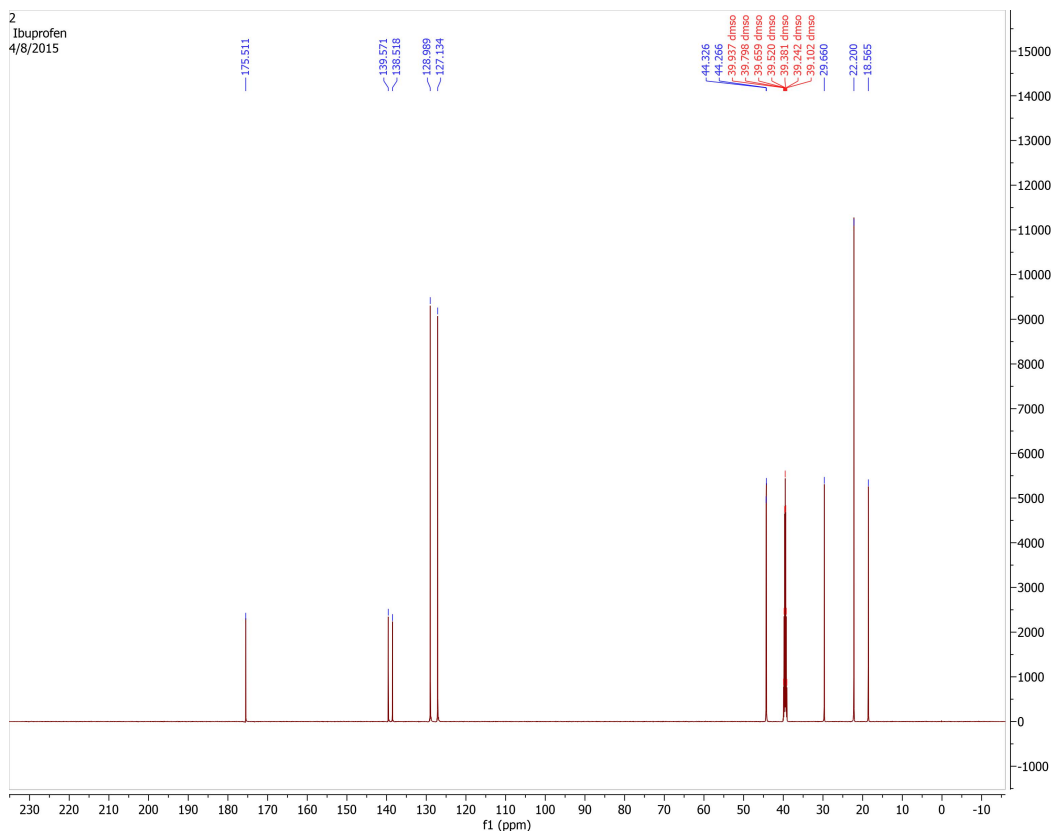
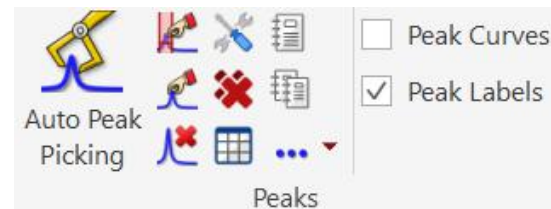
Restore Defaults Add... Edit... Delete

OK Cancel Solvents <<

ANALYSIS

- Click the Peaks > Options  to verify the peak picking options. Default settings are used here as shown to the right.
- Click the Auto Peak Picking tool to pick all the peaks
- Using other peak picking tools to display/delete/add/change peaks as needed.

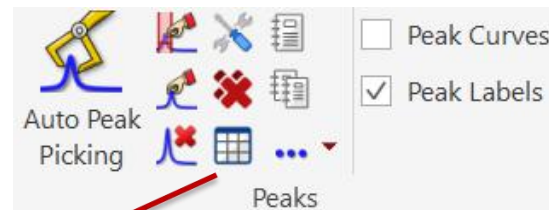
Peak picking



PUBLISHING

- Use the Peak Table tool to display the Peaks Table.
- Click Setup Report to change the reporting format
- Click Report to report the multiplets texts

Report the C-13 peaks



Peaks

Report Peaks Copy Peaks Setup Report Delete Select Peaks

Sync From Spec Filter Sync To Spec Set Flags Set Compound

^{13}C NMR (151 MHz, dmso) δ 175.5, 139.6, 138.5, 129.0, 127.1, 44.3, 44.3, 39.9, 39.8, 39.7, 39.5, 39.4, 39.2, 39.1, 29.7, 22.2, 18.6.

	ppm	intensity	Width	Area	Type	Flags	ty/Corr	notation
1	175.51	2394.8	2.38	1646...	Compound	None		
2	139.57	2468.5	2.76	1701...	Compound	None		
3	139.37	33.1	2.81	254.30	Artifact	Weak		
4	138.69	22.5	2.73	162.48	Artifact	Weak		
5	138.52	2264.7	2.79	1575...	Compound	None		
6	129.18	36.8	2.85	279.84	Artifact	Weak		
7	129.15	53.3	2.99	445.71	Artifact	Weak		

Setup Peak R... ?

J. Am. Chem. Soc.

Ascending order shifts

Ascending order of Js

Only report compound peaks

Report ^{13}C assignments

Report ^{13}C multiplicity

Use Extended Solvent Names

Number of decimals: 1

Fill style : Transparent

2D

Report as points

Report f1

Report f2

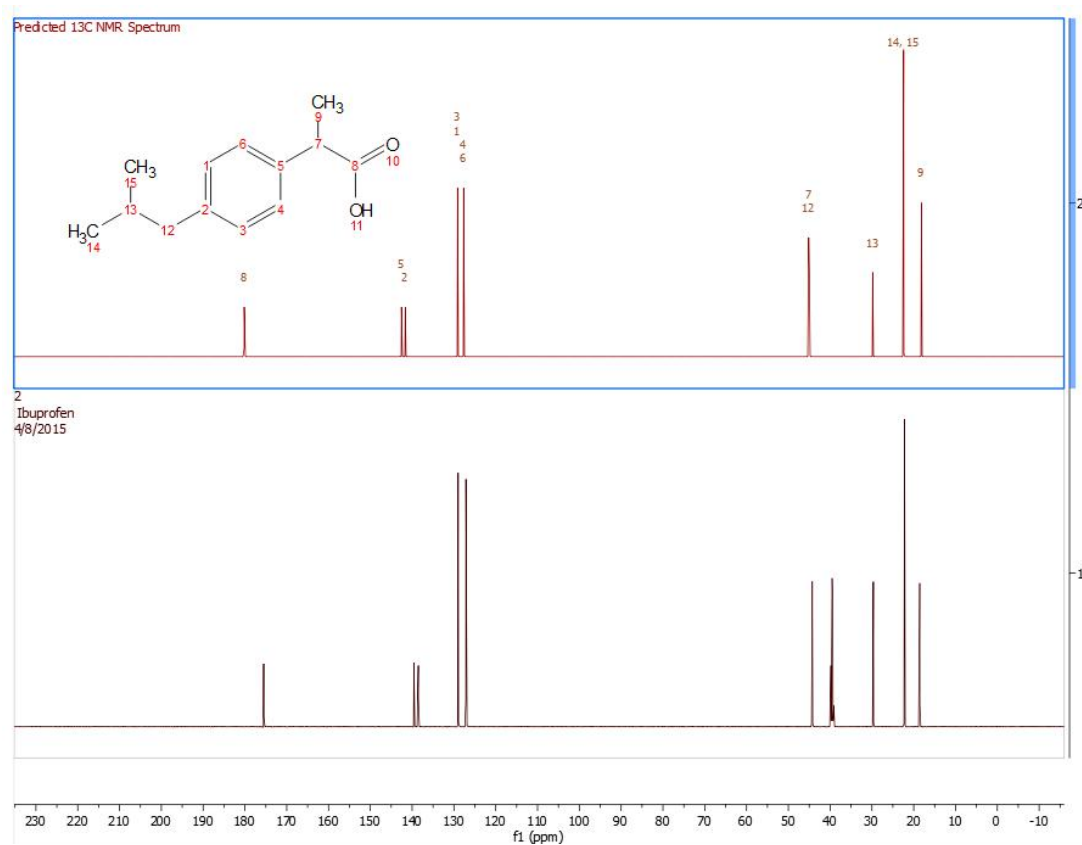
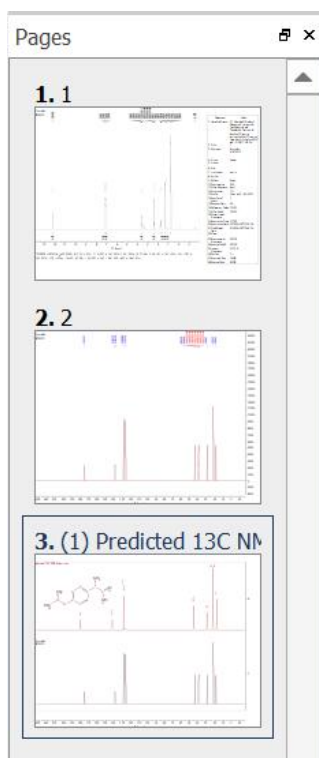
OK Cancel

^{13}C NMR (151 MHz, DMSO- d_6) δ 175.5, 139.6, 138.5, 129.0, 127.1, 44.3, 44.3, 29.7, 22.2, 18.6.

Verify the structure by predict and compare

PREDICTION

- Make a copy of the C-13 spectrum (Ctrl-C and Ctrl-V in the Pages View).
- Open the Ibuprofen.mol to bring in the structure to the C-13 spectrum.
- Choose Predict > Predict Compare.



Tip: if you want to delete the predicted C-13 spectrum from the stack, choose Stacked > Stacked Items Table, and use the Delete tool in the Table to delete the predicted C-13 spectrum.

PROCESSING

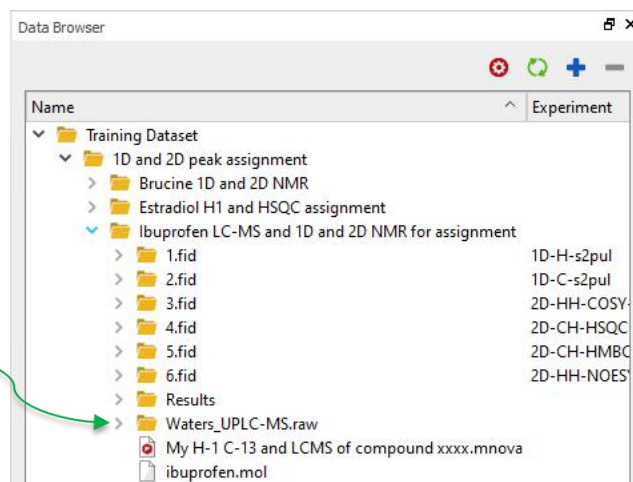
More about C-13 processing and analysis

- There are several other 1D C-13 NMR spectra in the tutorial datasets that you can use for practice.
- When the spectrum is more crowded and has more artefacts or impurity peaks, you can also use the manual peak picking tools to have more control.
- You can also do multiplet analysis for the C-13 peaks, especially when there are F-C couplings, and report the results from the Multiplet Table.



LC/MS Processing, Analysis, and Reporting

Sample data



Open the LC-MS data

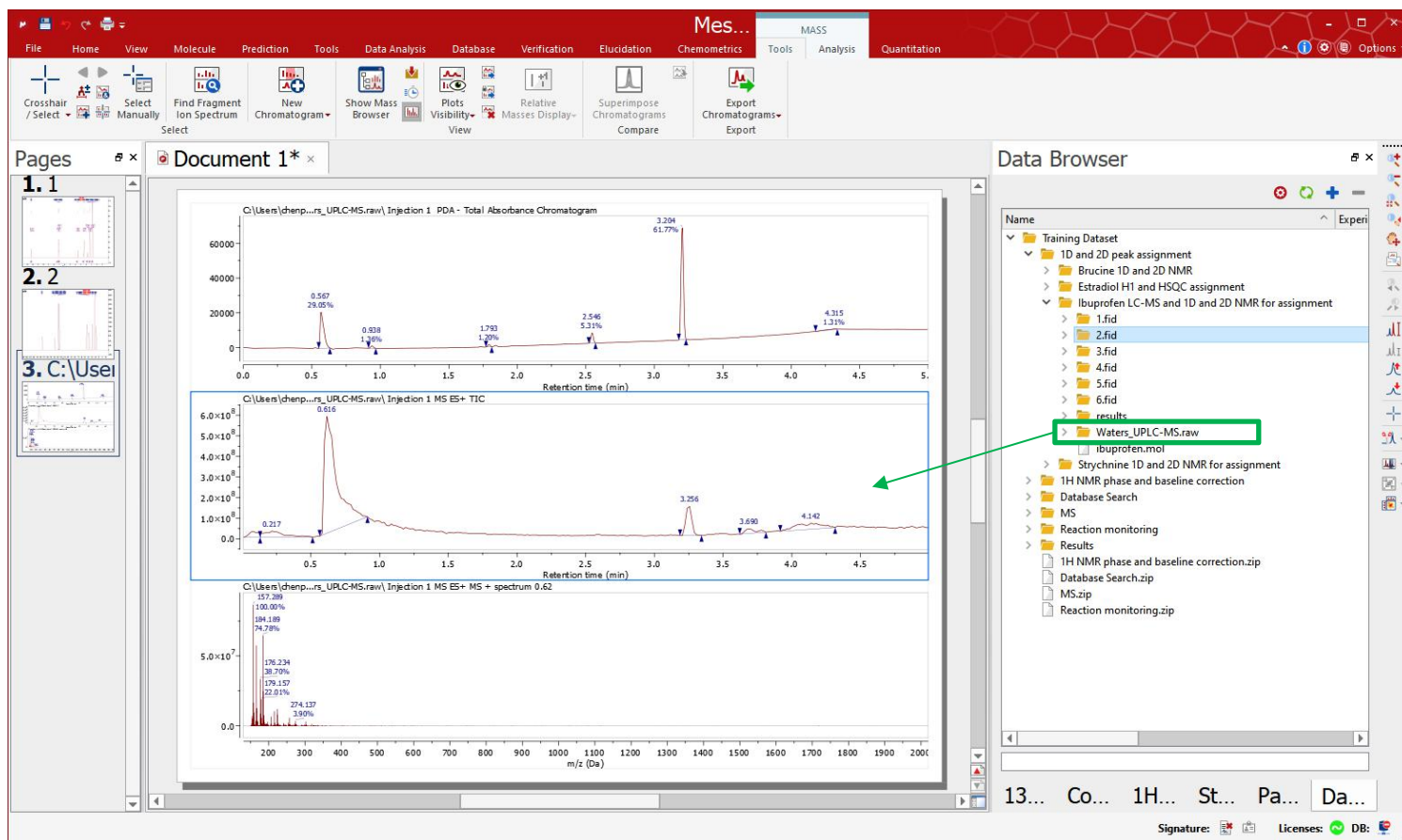
LC-MS

- In Data Browser, open the LC-MS data Ibuprofen (low resolution data acquired on Waters).
- The PDA, TIC and the mass spec at the highest TIC peak are displayed.

H-1

C-13

MS



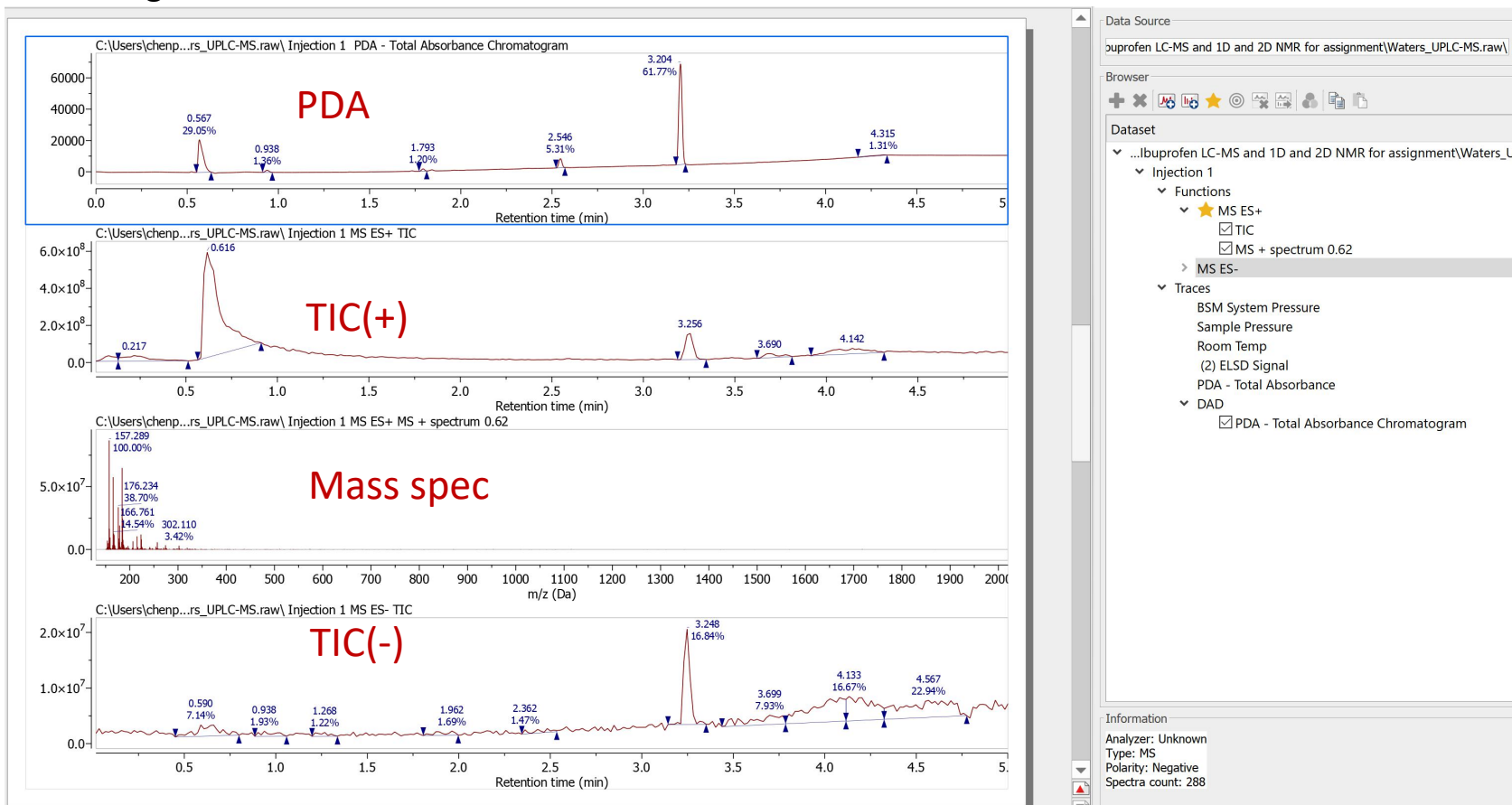
Display chromatograms

VISUALIZATION

- The Mass Browser is automatically displayed.*
- Open the negative polarization TIC by double clicking on “MS ES-”
- Right on the PDA and choose Hide Plot to hide PDA.



Show Mass
Browser

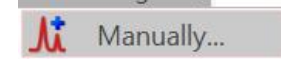
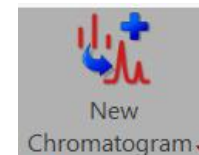


*If the Mass Browser is closed, you can open it using the Show Mass Browser tool in the MASS Tools ribbon.

Verify the elemental composition

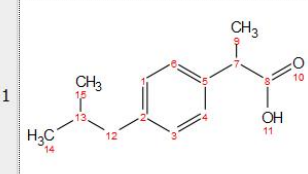
ANALYSIS

- Open the Ibuprofen.mol file from the Data Browser.
- Choose Molecule > Compound Table to find its monoisotopic mass: 206.13
- Highlight the TIC(+), click Mass > New Chromatogram > Manually, and enter a value of 207.13 +/- 0.25 Da to display the new chromatogram (also called Extracted Ion Chrom., EIC)



Compounds

Report Add Delete Setup Graphical Props PhysChem In Columns

Molecule	Properties
	Molecular Formula: C ₁₃ H ₁₈ O ₂ Average Mass: 206.28 Monoisotopic Mass: 206.13 Name: ibuprofen.cdx Label: ibuprofen Color: <input type="checkbox"/> None Assignments: <input type="checkbox"/> None

New chromatogram

Range

From: 207.13 m/z

To: 207.1300 m/z

Tolerance: 0.250 Da

OK Cancel

New chromatogram

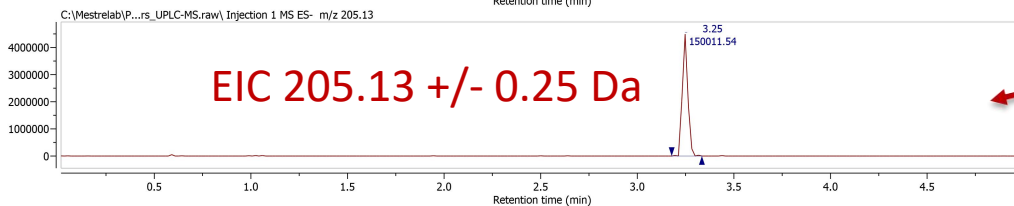
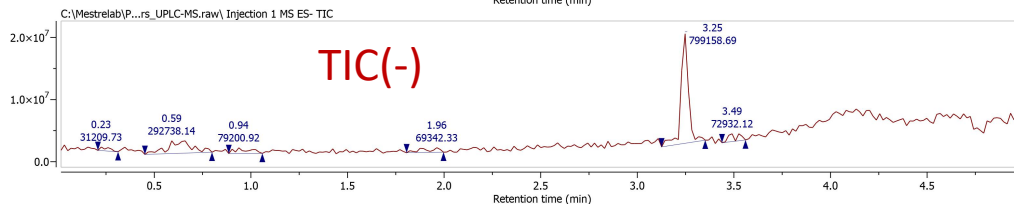
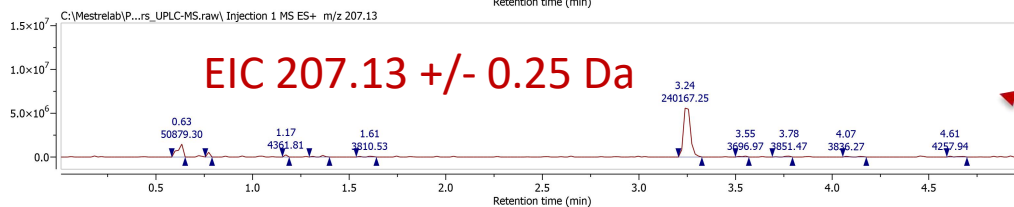
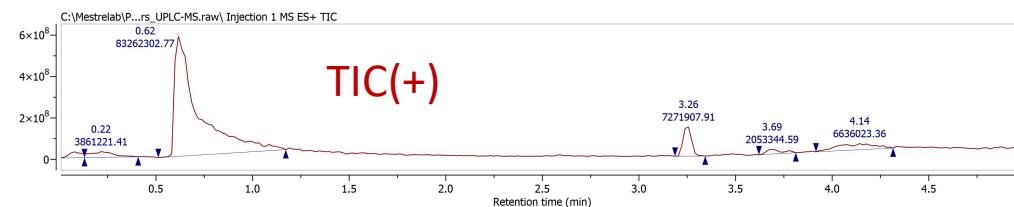
Range

From: 205.1300 m/z

To: 205.1700 m/z

Tolerance: 0.250 Da

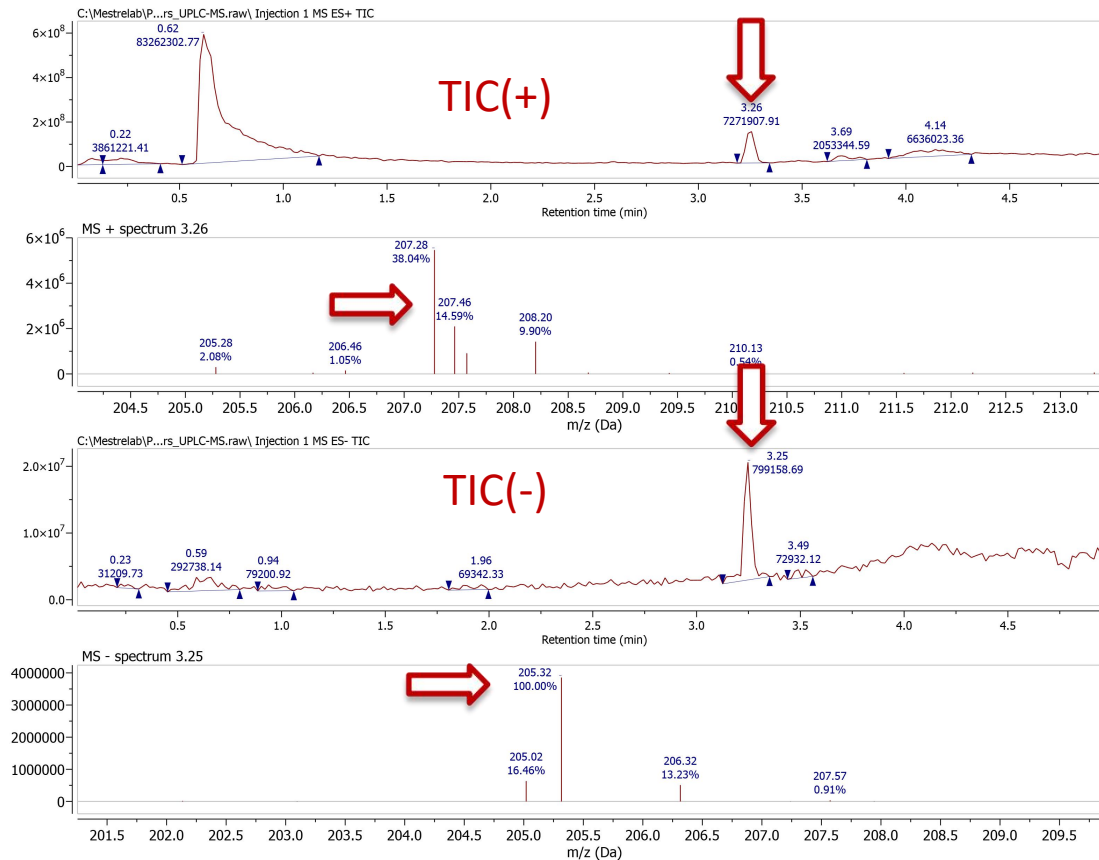
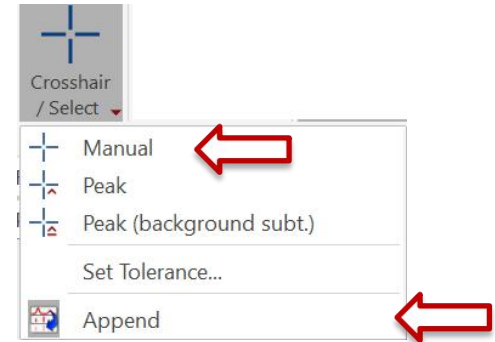
OK Cancel



Find the molecule ion peaks

ANALYSIS

- Click the Crosshair tool, and click on the peak around 3.25 min in both TIC(+) and TIC(-)
- Zoom into the mass spec to find the mol. Ion peaks at around 207.13 and 205.13 Da, respectively.

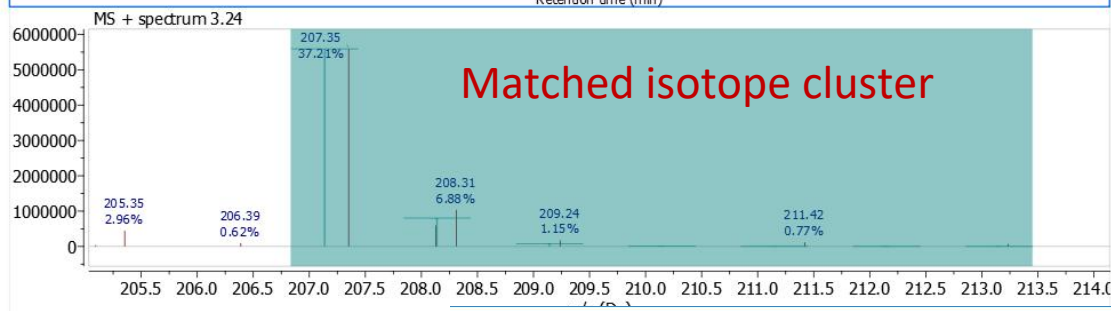
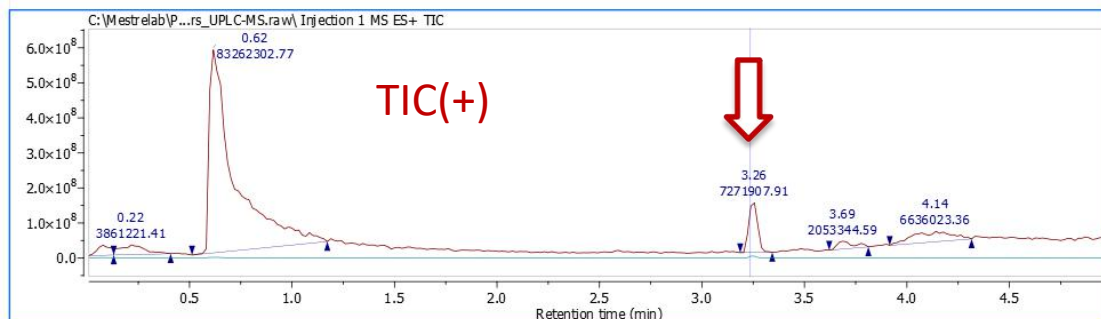


Tip: Use the Mass Browser to hide or delete unwanted plots. Right-click on a plot and choose Move up/Move Down etc. to re-order of the plots

Use Mol Match to verify the elemental composition

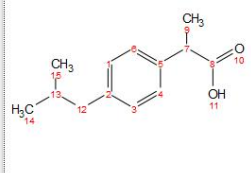
ANALYSIS

- Open the Ibuprofen.mol. Click Molecule Match.
- The Molecule Match Table shows the matching results.
- Click on the structure in the table to display the mol match results on the spectrum



Molecule Match

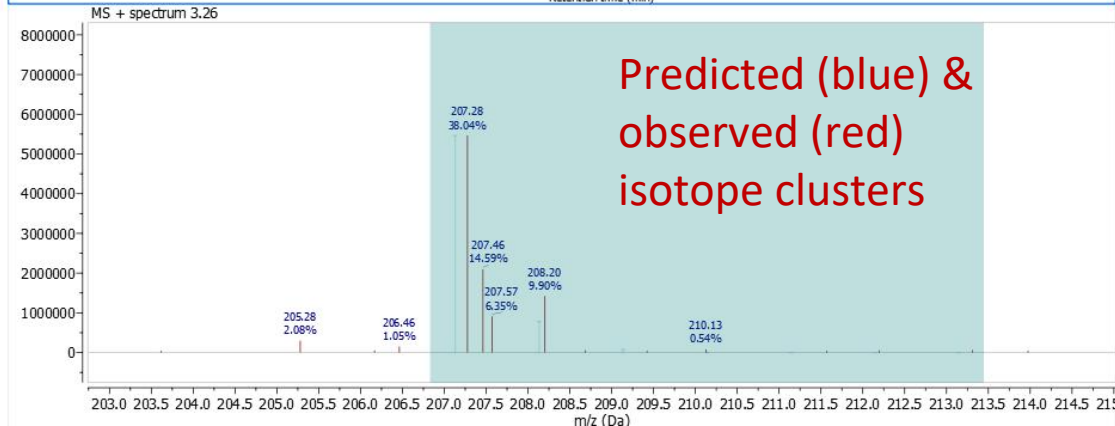
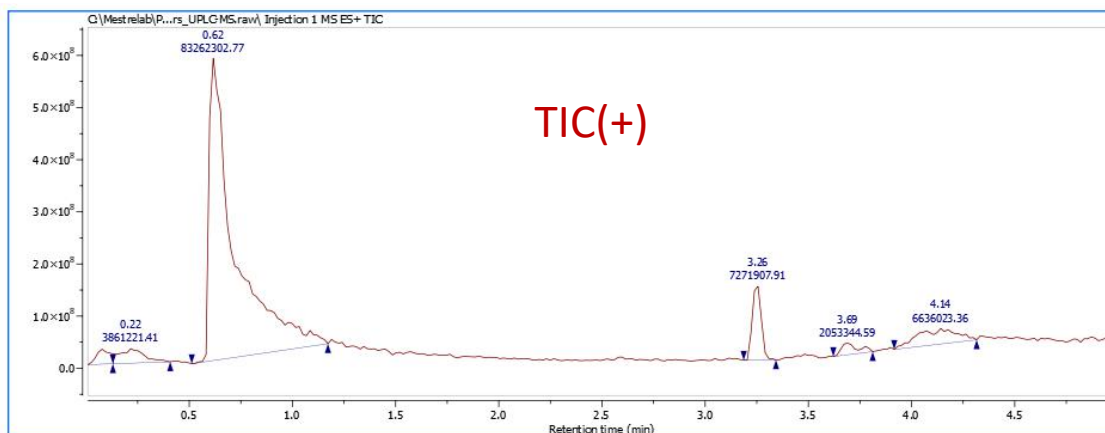
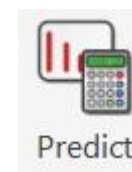
Report Molecule Match View Settings Setup

	Molecule	Formula	Molecular Weig	Match	Match Score	Similarity	MS Purity	RT	Scan	Purity	Matcd	Adduct/Loss
1		C ₁₃ H ₁₈ O ₂	206.131	✓	0.948	0.948	0.051	3.24	187	100.00%		H+ / -

Predict and verify the molecule ion peaks

ANALYSIS

- Click the Predict tool, and choose the MF C13H18O2 and press “+” to use it for prediction
- In the Mass Prediction List, highlight the first row. The predicted molecule ion and isotope peaks are displayed on top of the experiment peak for comparison.



Molecular Formula ? X

Molecular Formula: [] +

Recent

- C13H18O2
- C27H33NO2Cl
- C19H38N2O2

Compounds

	Formula	Weight
1	C ₁₃ H ₁₈ O ₂	206.1307

Mass Prediction X

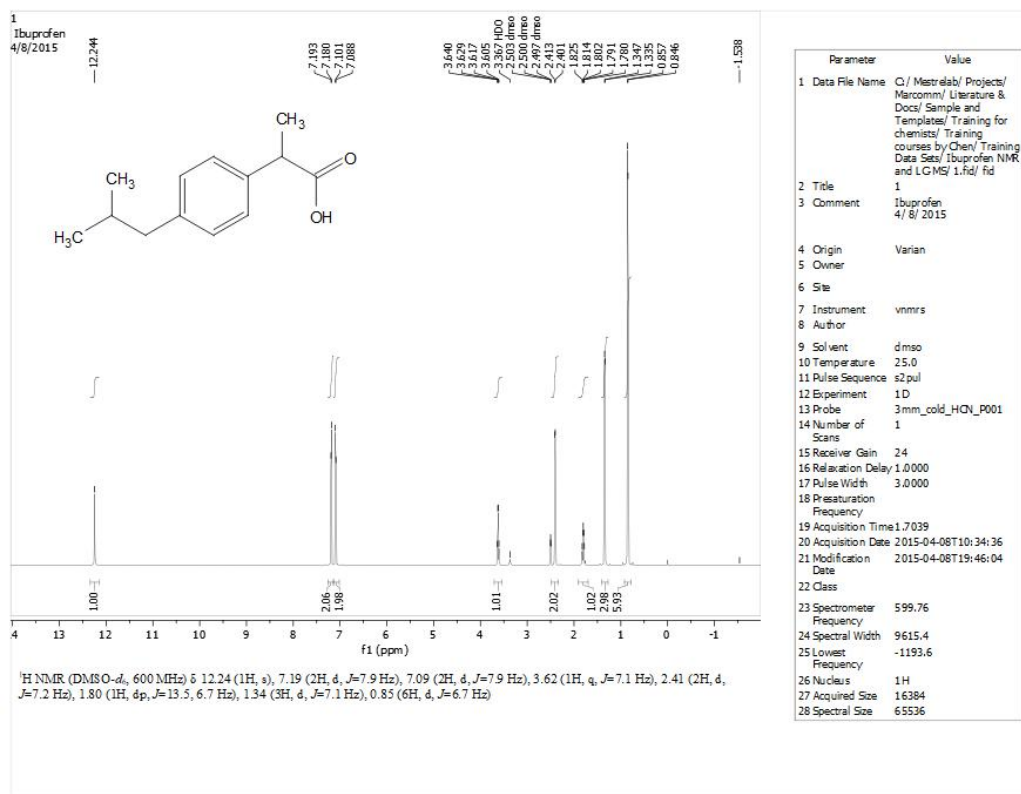
Report Copy Highlight Export Delete Clear Setup

	Formula	Adduct / Loss	rg e Si	m/z	Status
1	C ₁₃ H ₁₈ O ₂	H ⁺ / —	+1	207.13796	✓
2	C ₁₃ H ₁₈ O ₂	Na ⁺ / —	+1	229.11990	✓
3	C ₁₃ H ₁₈ O ₂	K ⁺ / —	+1	245.09384	✓
4	C ₁₃ H ₁₈ O ₂	CH ₃ OH ⁺ / —	+1	239.16417	✓
5	C ₁₃ H ₁₈ O ₂	NH ₄ ⁺ / —	+1	224.16451	✓
6	C ₁₃ H ₁₈ O ₂	— / H ⁺	-1	205.12340	✓
7	C ₁₃ H ₁₈ O ₂	Cl ⁻ / —	-1	241.10008	✓
8	C ₁₃ H ₁₈ O ₂	— / 2 (H ₂ OH ⁺)	-2	84.04750	✓
9	C ₁₃ H ₁₈ O ₂	Br ⁻ / —	-1	285.04957	✓

SAVING RESULTS

- Choose File > Export to PDF to save a PDF report of the page.
- Chose File > Save as to save all the results to a .mnova file.
- In the Advanced Tutorial we will learn to save the results to a database.
- Now can close the document or continue to add other spectra to it.

Save the results



Help information

- Use the Help Facility of Mnova: Help > Contents
- Visit www.mestrelab.com for manuals, tutorials, videos and publications
- Email support@mestrelab.com for technical questions

The screenshot displays the MestReNova software interface. On the left is a red sidebar menu with options: New, Close, Recent, Save, Save As..., Export to PDF..., Save To, Open..., Open Directory..., Open From, Print..., Page Setup..., Help (highlighted with a red arrow), Preferences..., Advanced Plug-ins..., and Exit. The main window is titled 'MestReNova' and has tabs for 'Help' and 'About'. The 'Help' tab is active, showing a 'MestReNova Manual' window. The manual's table of contents is visible, with 'Using GSD for multiplets analysis' selected. A red banner at the top of the manual window reads 'Using GSD for multiplets analysis'. Below the banner, the text states: 'Exploiting the power of GSD for an improved Multiplet Analysis'. It explains that Mnova uses Global Spectral Deconvolution (GSD) for peak picking and multiplet analysis, and that multiplet analysis benefits from automatic analysis with enhanced peak picking capabilities. A graph at the bottom shows a proton spectrum with a red peak at 3.46 ppm, labeled as a triplet.



Mestrelab Research

chemistry software solutions

Mnova Training– Advanced

For Mnova v14.2.0

Oct. 2020

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Mestrelab Research SL

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858.736.4563

Main Topics

- Opening and processing 2D NMR
- Assigning peaks to atoms
- Reporting assignment results
- Creating a database to save the data
- Searching Wiley C & H databases
- Analyzing arrayed spectra for reaction monitoring



Note: This tutorial covers the NMR, NMRPredict, MS, DB plugins of Mnova, and Wiley DB2

Specifics for <xxxxxx> University (To be completed by instructor)

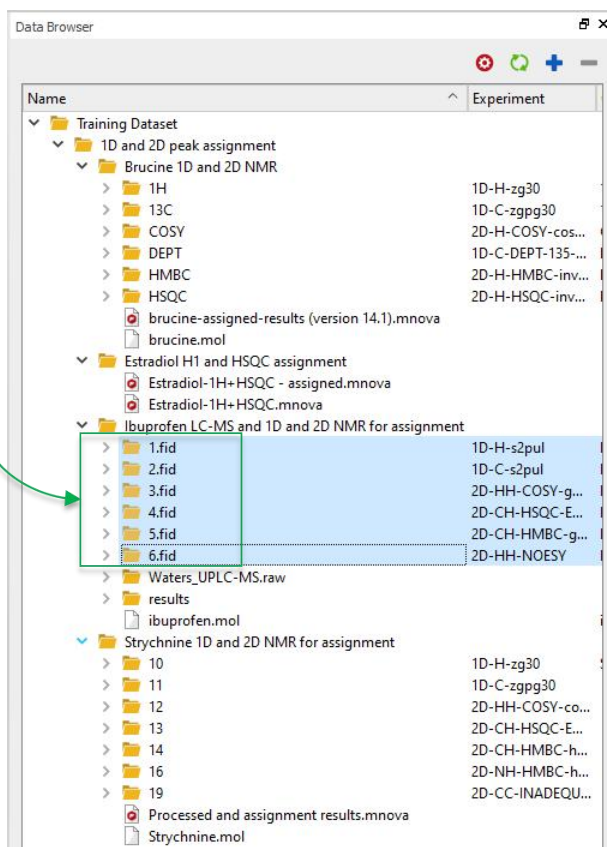
- The instructions for downloading, installing and activating Mnova:
 - <Link to instruction page>

- The Mnova licenses that <XXXX> University has:
 - Mnova Suite (NMR, NMRPredict & MS), unlimited
 - Etc.

- The sample data used in this tutorial are located at:
 - <Link to data folder>.

Processing 1D & 2D NMR Spectra Together

Sample data



Tip: There are a total of 4 datasets with 1D and 2D NMR spectra for practicing spectral processing, peak assignment (and structure elucidation, which is not covered in this tutorial). The Ibuprofen one used here is the simplest one compared to the others (Brucine, Strychnine and Estradiol).

Open a 1D and 2D spectra of Ibuprofen

PROCESSING

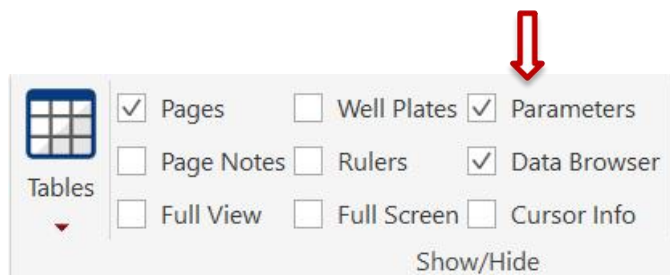
- Choose File > New to open a new blank document.
- In Data Browser, choose the 1D H/C, 2D HSQC, HMBC, COSY, and NOESY spectra and drag all of them to the main window.
- Re-process and analyze the H-1 and C-13 spectra according to the Basic Tutorial.

The screenshot displays the MestReNova software interface. The main window shows a 2D NMR spectrum for Ibuprofen, with the x-axis labeled 'f2 (ppm)' ranging from 14 to -1 and the y-axis labeled 'f1 (ppm)' ranging from -2 to -14. The Data Browser on the right side of the interface shows a tree view of files. A green box highlights a selection of files: 1.fid (1D-H-s2pul), 2.fid (1D-C-s2pul), 3.fid (2D-HH-COSY-gCOSY), 4.fid (2D-CH-HSQC-EDITED-...), 5.fid (2D-CH-HMBC-gHMBC), and 6.fid (2D-HH-NOESY). A green arrow points from this selection to the main spectrum window. The software interface includes a menu bar (File, Home, View, Molecule, Prediction, Tools, Database, Verification, Elucidation, Processing, Analysis, Assignments, Quantitation, Chem's Tools) and a toolbar with various analysis tools.

PARAMETERS

Which is which?

- Check View > Parameters Table to display the Parameters Table
- The Experiment and Pulse Sequence parameters usually indicate the type of NMR data



The 'Parameters' dialog box displays a table with the following data:

	Parameter	Value
7	Instrument	vnmrs
8	Author	
9	Solvent	dmsd
10	Temperature	25.0
11	Pulse Sequence	gHSQCAD
12	Experiment	HSQC-EDITED
13	Probe	3mm_cold_HCN_P001
14	Number of Scans	8
15	Receiver Gain	44
16	Relaxation Delay	1.0000

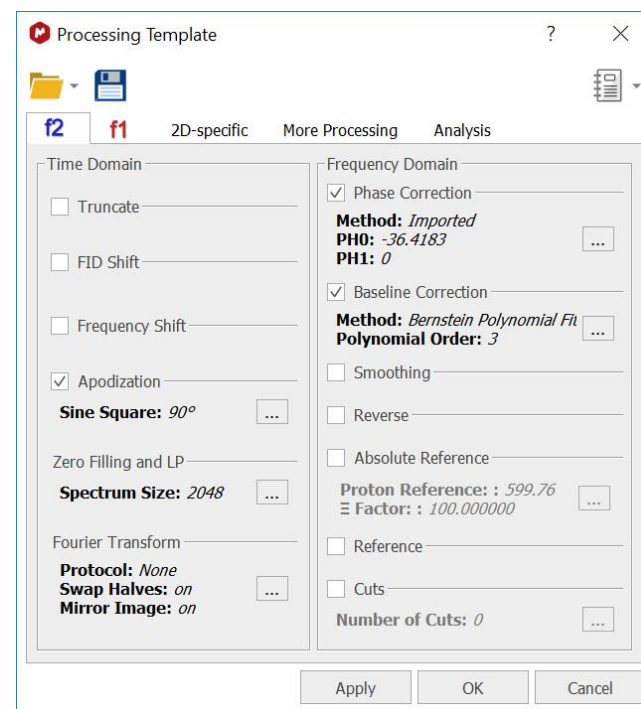
Tip: You can display the Experiment as part of the spectrum title. Double click on the spectrum and setup the Title in the Properties Dialog.

Rules of thumb for 2D processing

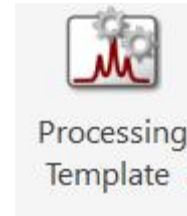
PROCESSING

When a 2D is opened in Mnova, it is automatically processed starting from the raw data (*ser* or *fid*) with the parameters from the instrument. If the results are not satisfactory, you may need to adjust the following parameters to improve the results:

- Apodization: To improve the line shape, resolution and S/N ratio
 - COSY: Use Sine Square 0 for F2 & F1
 - Others: Use Sine Square 90 F2 & F1
- Zero Fill and LP (Linear Prediction): To improve resolution
 - F2: at least double of the original datapoints, 2048 or 4096
 - F1: At least double of the original datapoints, 1024 or 2048
 - F1: Do LP if original datapoints \leq 128 (optional)
- Phase Correction: To improve line shape
 - Use Imported or Automated (Regions) first
 - Do manual correction if needed.
- Baseline Correction: To reduce noise
 - Use Bernstein Polynomial Fit on either dimension
- Other 2D-Specific parameters (optional):
 - COSY, NOESY: Use Symmetrize with caution
 - HSQC, HMBC: Use Reduce T1 Noise Reduction when needed.



Rules of thumb for 2D processing



The 2D processing parameters that you may want to adjust and their recommended values*

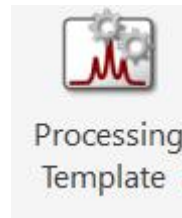
	Apodization	Zero-fill & Linear Prediction	Phase Correction	Baseline Correction	Others
COSY	Sine square 0 on both F2 and F1	At least double to 2K or 4K on F2; or 1K or 2K on F1. Do linear prediction on F1 if # of increments < 128	Use imported parameters first. Use auto or manual phasing if needed on both F1 and F2	Use Bernstein Polynomial Fitting (3 rd order) for F2 and F1	Do COSY-style symmetrization with caution
HSQC	Sine square 90 on both F2 and F1	Same as above	Same as above	Same as above	Use Reduce T1 Noise to reduce T1 noises with caution
HMBC	Same as above	Same as above	Same as above	Same as above	Same as above
NOESY/R OESY	Same as above	Same as above	Same as above	Same as above	Do COSY-style symmetrization with caution

**These are recommended starting points for conventional 2D NMR processing. You can try many of other combinations as appropriate. Once you are satisfied with the results, you can save all the parameters as a processing template, and apply it to similar spectra later.*

Re-process HSQC spectrum

PROCESSING

- Reprocess the HSQC spectrum as shown below.
- Note the apodization functions for F2 and F1
- Note the forward linear prediction for F1 applied here



Processing Template

f2 f1 2D-specific More Processing Analysis

Time Domain

Truncate

FID Shift

Frequency Shift

Apodization

Sine Square: 90° ...

Zero Filling and LP

Spectrum Size: 2048 ...

Fourier Transform

Protocol: None
Swap Halves: on ...
Mirror Image: on

Frequency Domain

Phase Correction

Method: Imported
PH0: -36.4183
PH1: 0 ...

Baseline Correction

Method: Bernstein Polynomial Fit
Polynomial Order: 3 ...

Smoothing

Reverse

Absolute Reference

Proton Reference: : 599.76 ...
Factor: : 100.000000

Reference

Cuts

Number of Cuts: 0 ...

Apply OK Cancel

Processing Template

f1 2D-specific More Processing Analysis

Time Domain

Truncate

Frequency Shift

Apodization

Sine Square: 90° ...

Zero Filling and LP

Spectrum Size: 1024 ...
Forward LP: [128, 256], 112, 15 ...

Fourier Transform

Protocol: Echo-Antiecho
Swap Halves: on ...
Mirror Image: on

Frequency Domain

Phase Correction

Method: Imported
PH0: 0
PH1: 0 ...

Baseline Correction

Method: Bernstein Polynomial Fit
Polynomial Order: 3 ...

Smoothing

Reverse

Absolute Reference

Proton Reference: : 599.76 ...
Factor: : 25.145020

Reference

Cuts

Number of Cuts: 0 ...

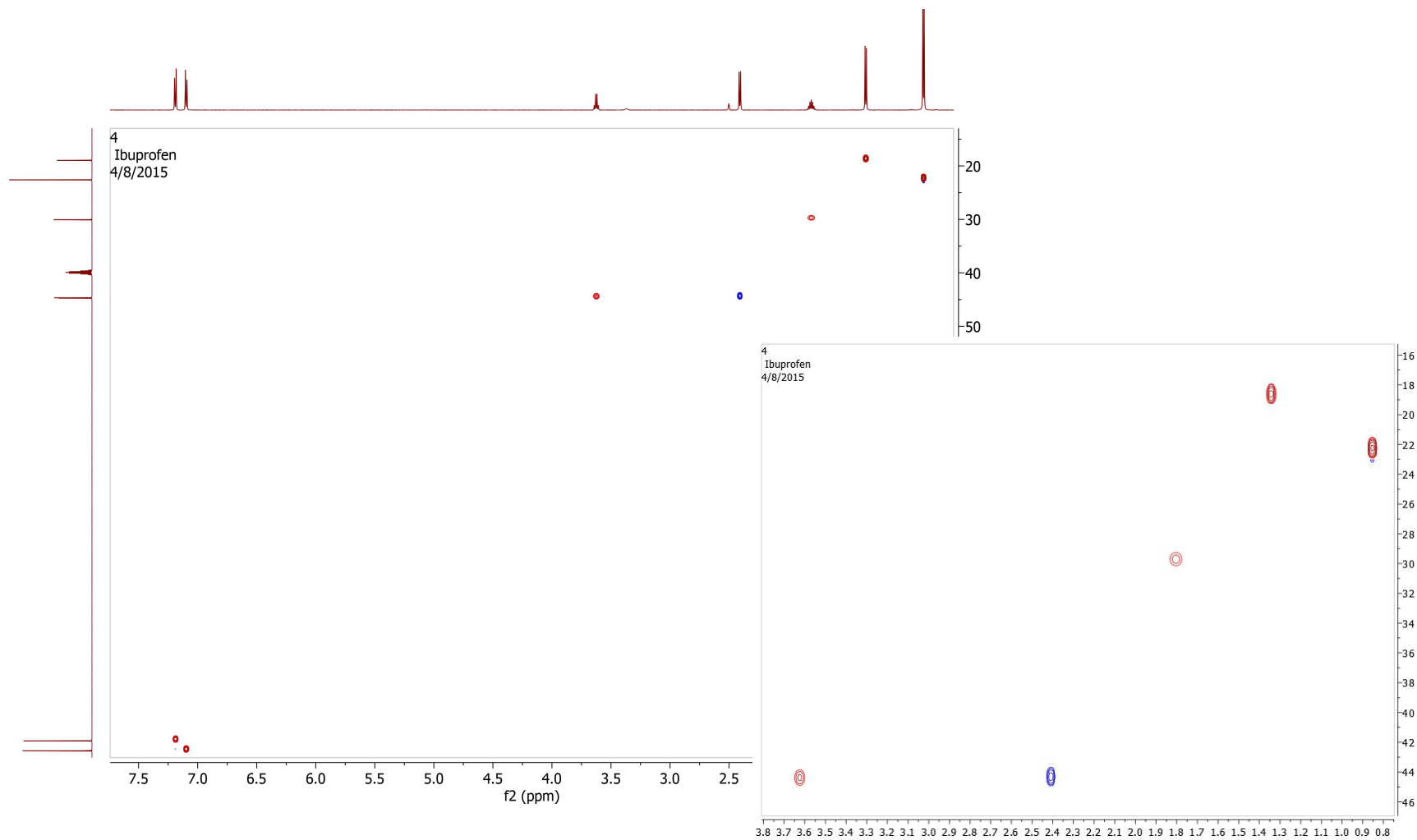
Apply OK Cancel

Note: The Fourier Transform method is automatically set and normally you don't need to change it.

Re-process HSQC spectrum

PROCESSING

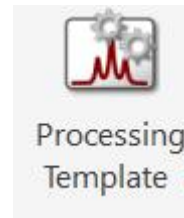
- The re-processed HSQC spectrum shows better line shape, and higher resolution on the F1 dimension



Re-process HMBC spectrum

PROCESSING

- Reprocess the HMBC spectrum as shown below.
- Note the apodization functions for F2 and F1
- Note the forward linear prediction for F1 applied here



Processing Template

f2 f1 2D-specific More Processing Analysis

Time Domain

- Truncate
- FID Shift
- Frequency Shift
- Apodization
- Sine Square: 90°**
- Zero Filling and LP
- Spectrum Size: 2048**
- Fourier Transform
- Protocol: None**
- Swap Halves: on**
- Mirror Image: on**

Frequency Domain

- Phase Correction
- Method: Magnitude**
- Baseline Correction
- Method: Bernstein Polynomial Fit**
- Polynomial Order: 3**
- Smoothing
- Reverse
- Absolute Reference
- Proton Reference: : 599.76**
- Factor: : 100.000000**
- Reference
- Cuts
- Number of Cuts: 0**

Apply OK Cancel

Processing Template

f1 2D-specific More Processing Analysis

Time Domain

- Truncate
- Frequency Shift
- Apodization
- Sine Square: 90°**
- Zero Filling and LP
- Spectrum Size: 1024**
- Forward LP: [128, 256], 112, 15**
- Fourier Transform
- Protocol: Echo-Antiecho**
- Swap Halves: on**
- Mirror Image: on**

Frequency Domain

- Phase Correction
- Method: Imported**
- PH0: 0**
- PH1: 0**
- Baseline Correction
- Method: Bernstein Polynomial Fit**
- Polynomial Order: 3**
- Smoothing
- Reverse
- Absolute Reference
- Proton Reference: : 599.76**
- Factor: : 25.145020**
- Reference
- Cuts
- Number of Cuts: 0**

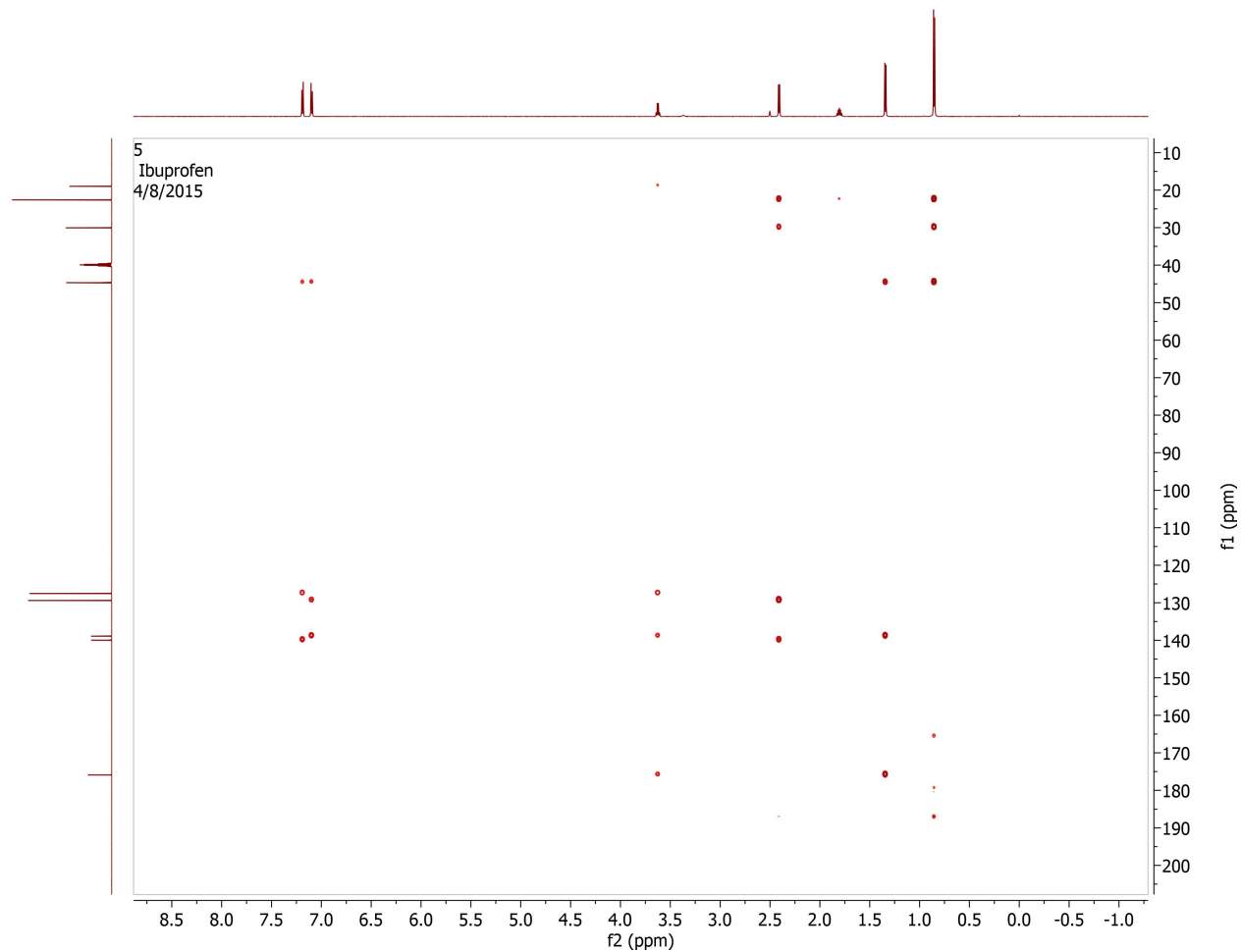
Apply OK Cancel

Note: The Fourier Transform method is automatically set and normally you don't need to change it.

Re-process HMBC spectrum

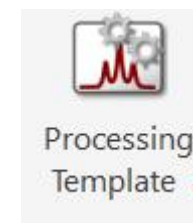
PROCESSING

- The re-processed HSQC spectrum shows better line shape, and higher resolution on the F1 dimension



PROCESSING

Re-process COSY spectrum



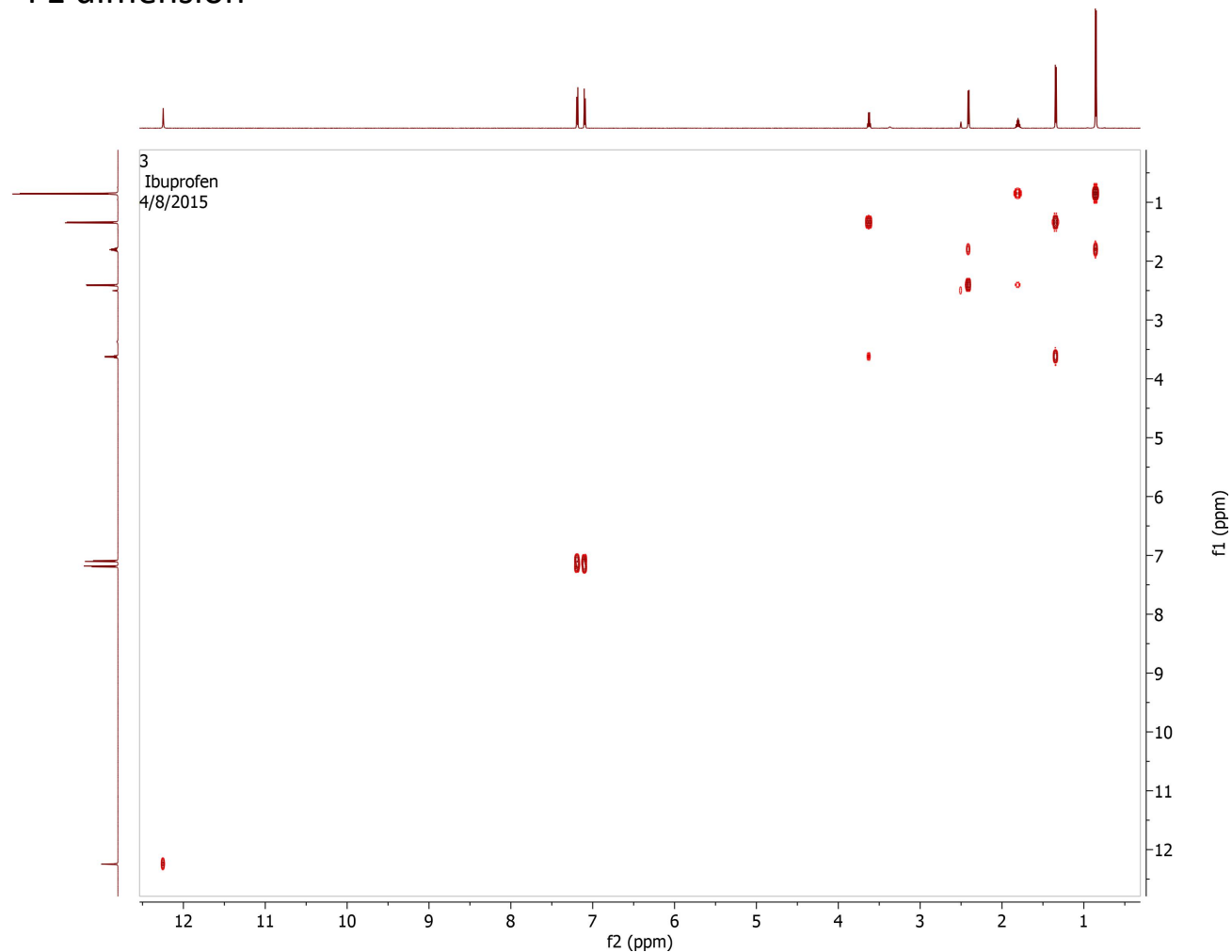
- Reprocess the COSY spectrum as shown below.
- Note the apodization functions for F2 and F1
- Note the forward linear prediction for F1 applied here
- Turn off Symmetrize in the 2D-specific Tab

Note: The Fourier Transform method is automatically set and normally you don't need to change it.

Re-process COSY spectrum

PROCESSING

- The re-processed COSY spectrum shows better line shape, and higher resolution on the F1 dimension



Re-process NOESY spectrum

PROCESSING



- Reprocess the NOESY spectrum as shown below.
- Note the apodization functions for F2 and F1
- Note the forward linear prediction for F1 applied here

The image displays two screenshots of the 'Processing Template' dialog box, illustrating the configuration for re-processing a NOESY spectrum. Red arrows highlight key settings in both windows.

Left Window (f2 tab):

- Time Domain:**
 - Truncate
 - FID Shift
 - Frequency Shift
 - Apodization
 - Sine Square:** 90°
 - Zero Filling and LP:** Spectrum Size: 2048
 - Fourier Transform:** Protocol: None, Swap Halves: on, Mirror Image: on
- Frequency Domain:**
 - Phase Correction (Method: Magnitude)
 - Baseline Correction (Method: Bernstein Polynomial Fit, Polynomial Order: 3)
 - Smoothing
 - Reverse
 - Absolute Reference (Proton Reference: 599.76, Factor: 100.000000)
 - Reference
 - Cuts (Number of Cuts: 0)

Right Window (f1 tab):

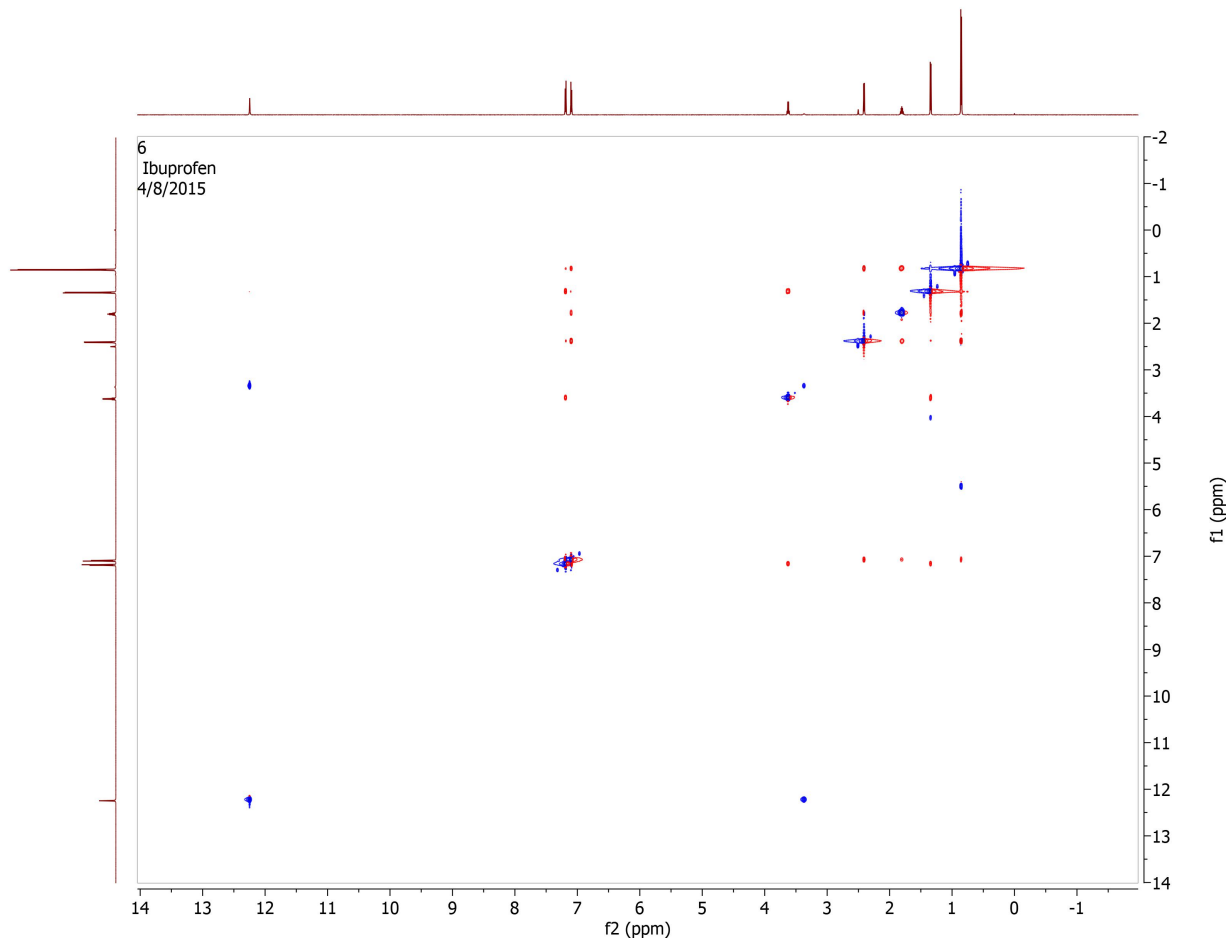
- Time Domain:**
 - Truncate
 - Frequency Shift
 - Apodization
 - Sine Square:** 90°
 - Zero Filling and LP:** Spectrum Size: 1024, Forward LP: [128, 256], 112, 15
 - Fourier Transform:** Protocol: Echo-Antiecho, Swap Halves: on, Mirror Image: on
- Frequency Domain:**
 - Phase Correction (Method: Imported, PH0: 0, PH1: 0)
 - Baseline Correction (Method: Bernstein Polynomial Fit, Polynomial Order: 3)
 - Smoothing
 - Reverse
 - Absolute Reference (Proton Reference: 599.76, Factor: 25.145020)
 - Reference
 - Cuts (Number of Cuts: 0)

Note: The Fourier Transform method is automatically set and normally you don't need to change it.

Re-process NOESY spectrum

PROCESSING

- The re-processed NOESY spectrum shows better line shape, and higher resolution on the F1 dimension, though there still some phase errors



Phase correction for NOESY spectrum

PROCESSING

- Do Manual phase correction for either both dimensions.
- Also applied +180 for PH0 to make the cross peaks negative and diagonal peaks positive



Phase Correction

f1 f2

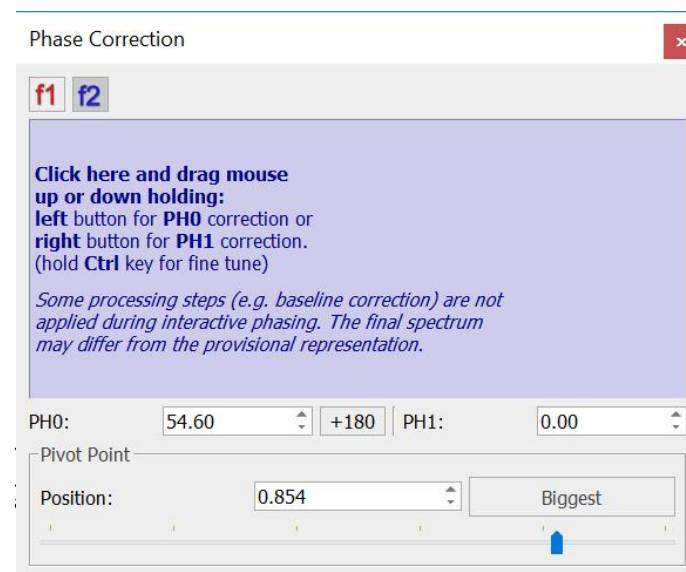
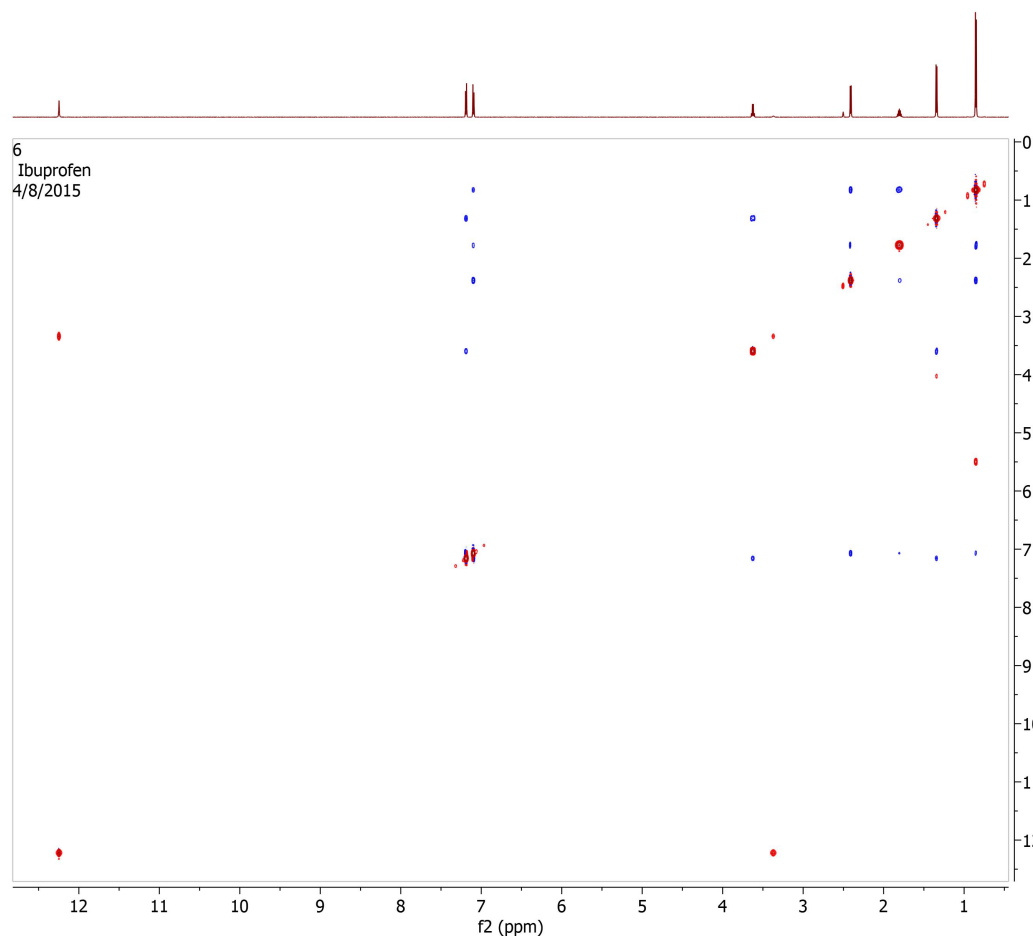
Click here and drag mouse up or down holding: left button for PH0 correction or right button for PH1 correction. (hold Ctrl key for fine tune)

Some processing steps (e.g. baseline correction) are not applied during interactive phasing. The final spectrum may differ from the provisional representation.

PH0: 54.60 +180 PH1: 0.00

Pivot Point

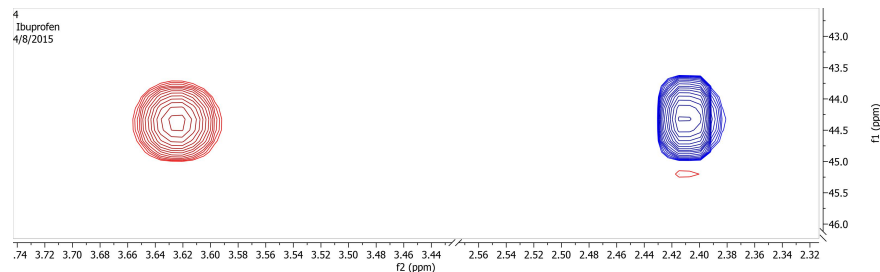
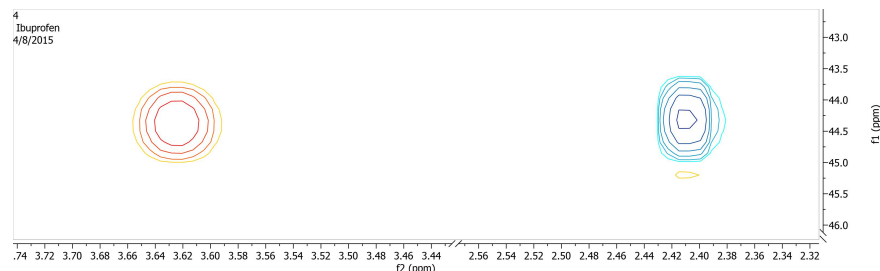
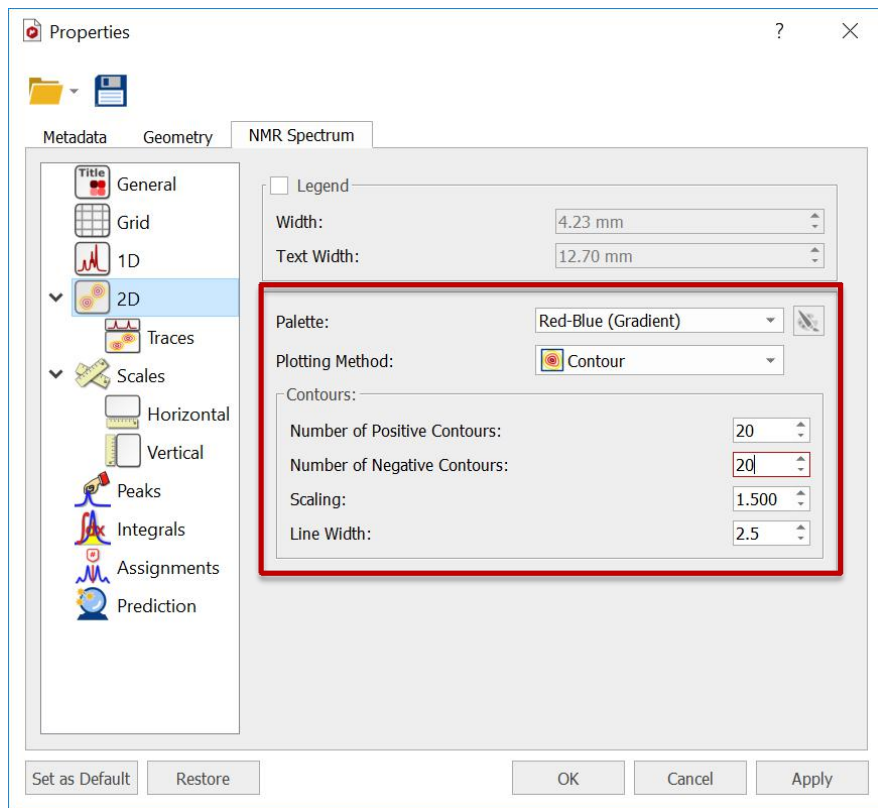
Position: 0.854 Biggest

A screenshot of the "Phase Correction" dialog box in Mestrelab software. It features a title bar with a close button, two tabs labeled "f1" and "f2", and a large blue instruction area. Below the instructions are input fields for "PH0" (54.60), a "+180" button, and "PH1" (0.00). At the bottom, there is a "Pivot Point" section with a "Position" field set to 0.854 and a "Biggest" button, accompanied by a horizontal slider.

Change the Display Properties

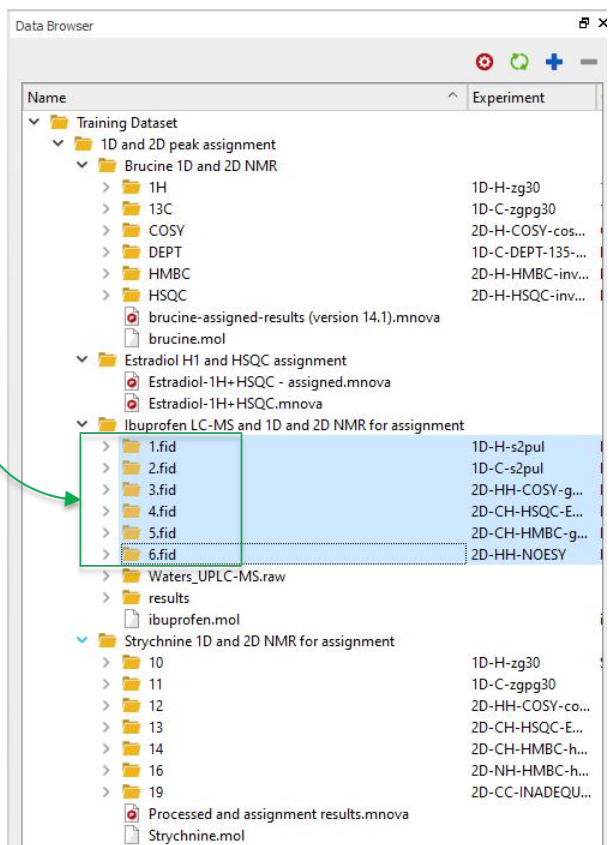
PROCESSING

- Right click on the spectrum to open the Properties Dialog, view the properties that can be changed
- In the 2D Category, adjust the highlighted parameters and click Apply to see the effects
- Click Set as Default to retain the settings for 2D spectra display in the future



Peak Assignment Using 1D & 2D NMR Spectra Together

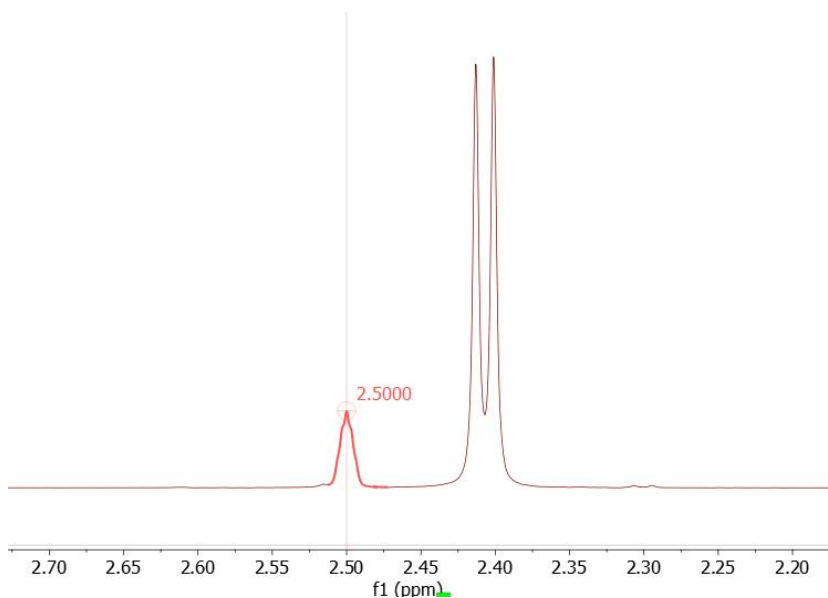
Sample data



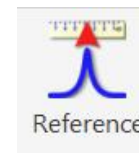
Tip: There are a total of 4 datasets with 1D and 2D NMR spectra for practicing spectral processing, peak assignment (and structure elucidation, which is not covered in this tutorial). The Ibuprofen one here is the simplest one compared to the others (Brucine, Strychnine and Estradiol).

ANALYSIS

- This spectrum uses DMSO-d6 as the solvent. We can reference the chemical shifts by setting its middle peak to 2.5 ppm.
- Zoom to the DMSO peak at around 2.5 ppm. Choose Analysis > Reference, and click on the top of the middle peak.
- Set it to 2.5 ppm either manually or from the Solvent List.



Chemical shift referencing for H-1



Reference along f1

Old Shift: 2.5021 ppm Auto Tuning

New Shift: 2.5000 ppm Range Width: 0.1000 ppm

Annotation: DMSO-d6

Solvent List

Name	Shift (ppm)	Multiplicity	J (Hz)
Deuterium Oxide	4.790	1	
Dimethyl Sulfoxide-d6	2.500	5	1
	3.330	1	
Ethanol-d6	5.290	1	

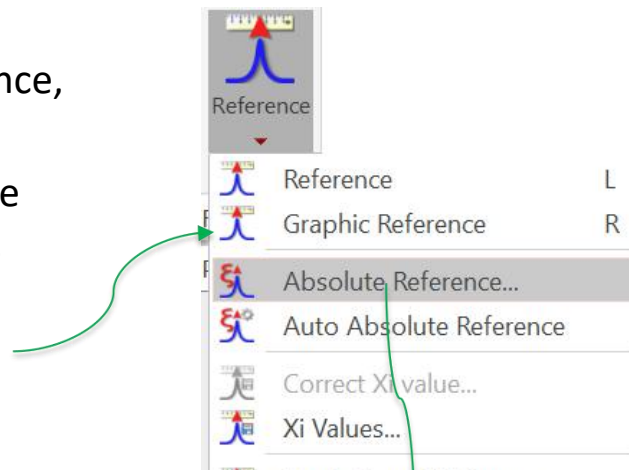
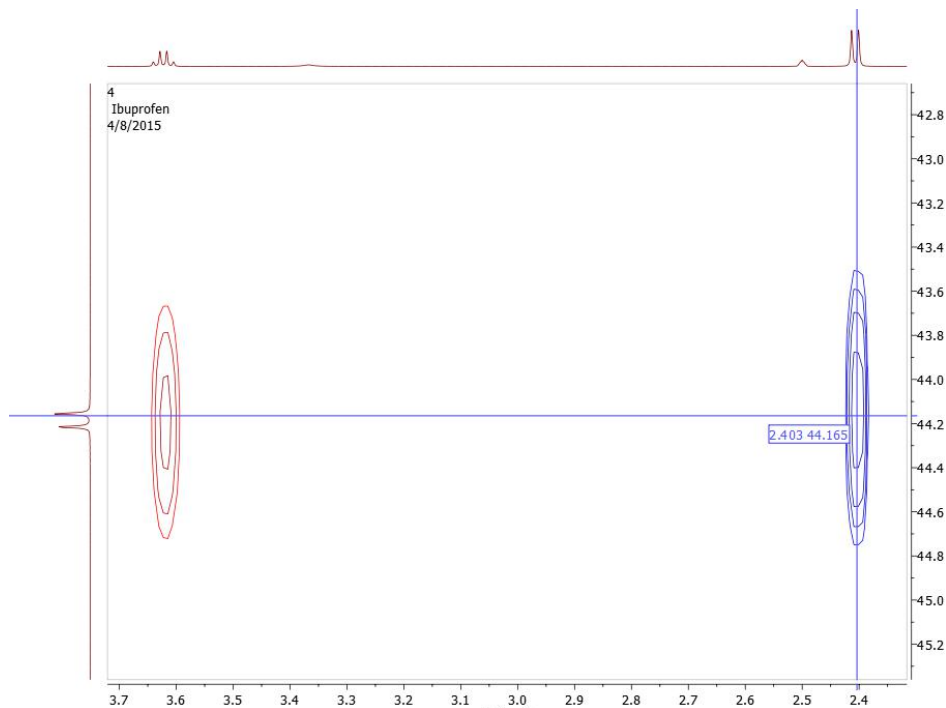
Restore Defaults Add... Edit... Delete

OK Cancel Solvents <<

Chemical shift referencing for other spectra

ANALYSIS

- Choose Analysis > References > Absolute Reference, and click OK to the dialog.
- This applies referencing to all other spectra in the document using the H-1 spectrum as a standard.
- If needed, further align the 1D and 2D spectra manually using the Graphic Reference tool.



Absolute Reference [?] [X]

Use as Reference: 1: 599.763 MHz

Spectra	
<input type="checkbox"/>	1: 1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	2: 13C, 150.826 MHz $\Xi=25.145020$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	2D-COSY: 3
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	2D-HSQC-EDITED: 4
<input checked="" type="checkbox"/>	13C, 150.826 MHz $\Xi=25.145020$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	2D-HMBC: 5
<input checked="" type="checkbox"/>	13C, 150.826 MHz $\Xi=25.145020$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	2D-NOESY: 6
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)
<input checked="" type="checkbox"/>	1H, 599.763 MHz $\Xi=100.000000$ (Me4Si CDCl3, $\phi = 1\%$)

[Xi Values...]

Show in spectrum title Show in parameters table

[OK] [Cancel]

Open the structure for peak assignment

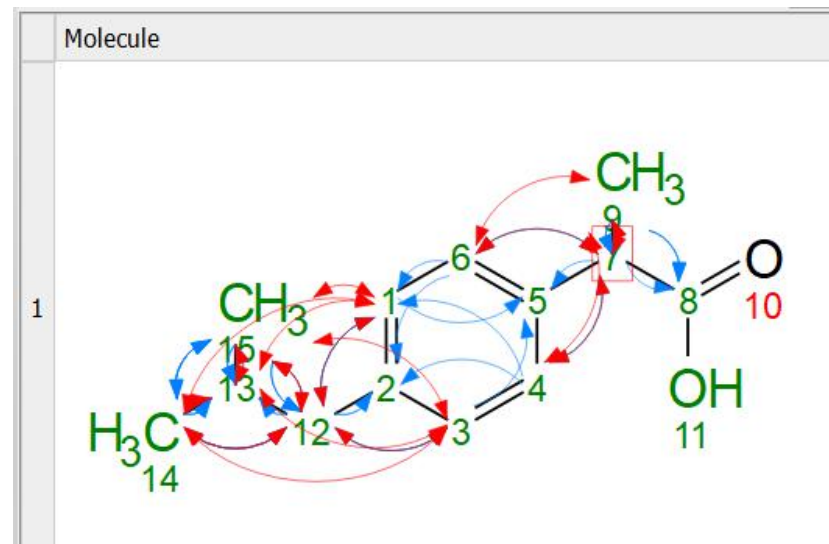
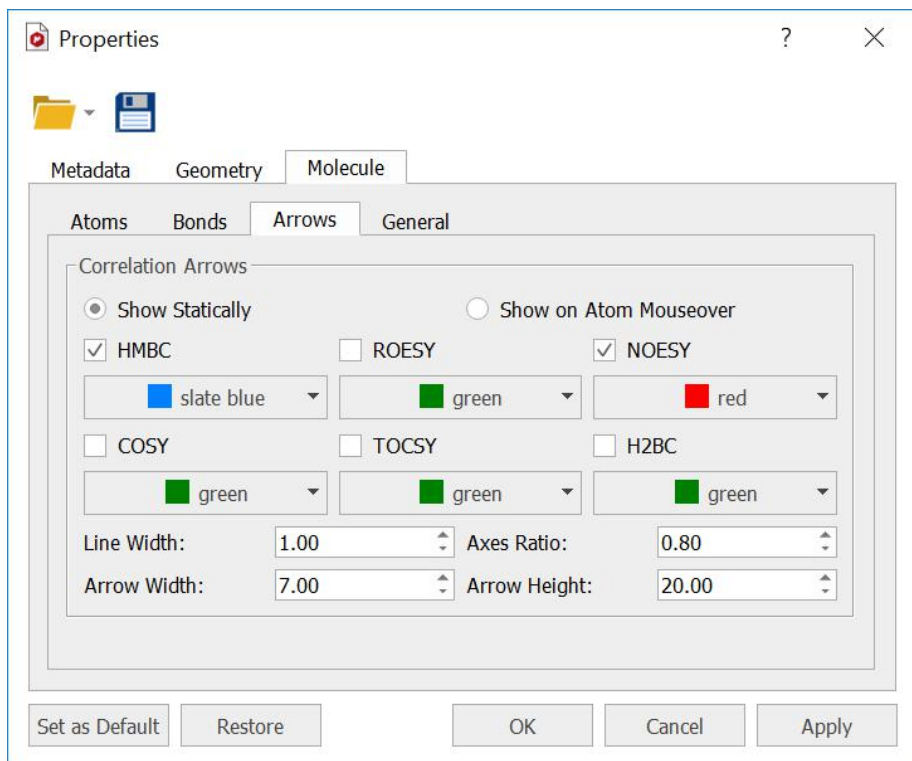
ANALYSIS

- Open the Ibuprofen.mol file from the Data Browser.
- Choose Molecule > Compound Table to show the structure on the side.
- Note: Open the same molecule only once. If needed, use Report on the Compound Table to report the structure to other pages. Do not open the same structure multiple times.
- Display the Assignment Table. Make sure all spectra are “linked” in it.

The screenshot shows the MestReNova software interface. The main window displays the chemical structure of Ibuprofen and its ¹H NMR spectrum. The spectrum shows several peaks with assignments: A (s) at 12.24 ppm, C (d) at 7.09 ppm, B (d) at 7.19 ppm, D (q) at 2.41 ppm, E (d) at 2.41 ppm, G (d) at 1.80 ppm, and H (d) at 1.68 ppm. The chemical structure is shown with atom numbering from 1 to 14. The Assignments panel on the right shows a list of available spectra and a list of linked spectra. The linked spectra list contains four entries: 3 Ibuprofen 4/8/2015, 6 Ibuprofen 4/8/2015, 4 Ibuprofen 4/8/2015, and 5 Ibuprofen 4/8/2015. A green arrow points from the 'Assignment Labels' checkbox in the top toolbar to the 'Linked spectra' list in the Assignments panel.

Show 2D correlation as arrows

- Right-click on the structure (or click on Graphical Props in the Compounds Table)
- In the Arrows Tab, turn on the display of HMBC and NOESY correlation as different colors
- The correlations will be displayed when the assignments are added later

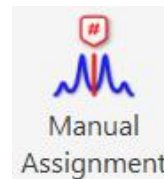


Tip: Click Save as Default button to save the settings

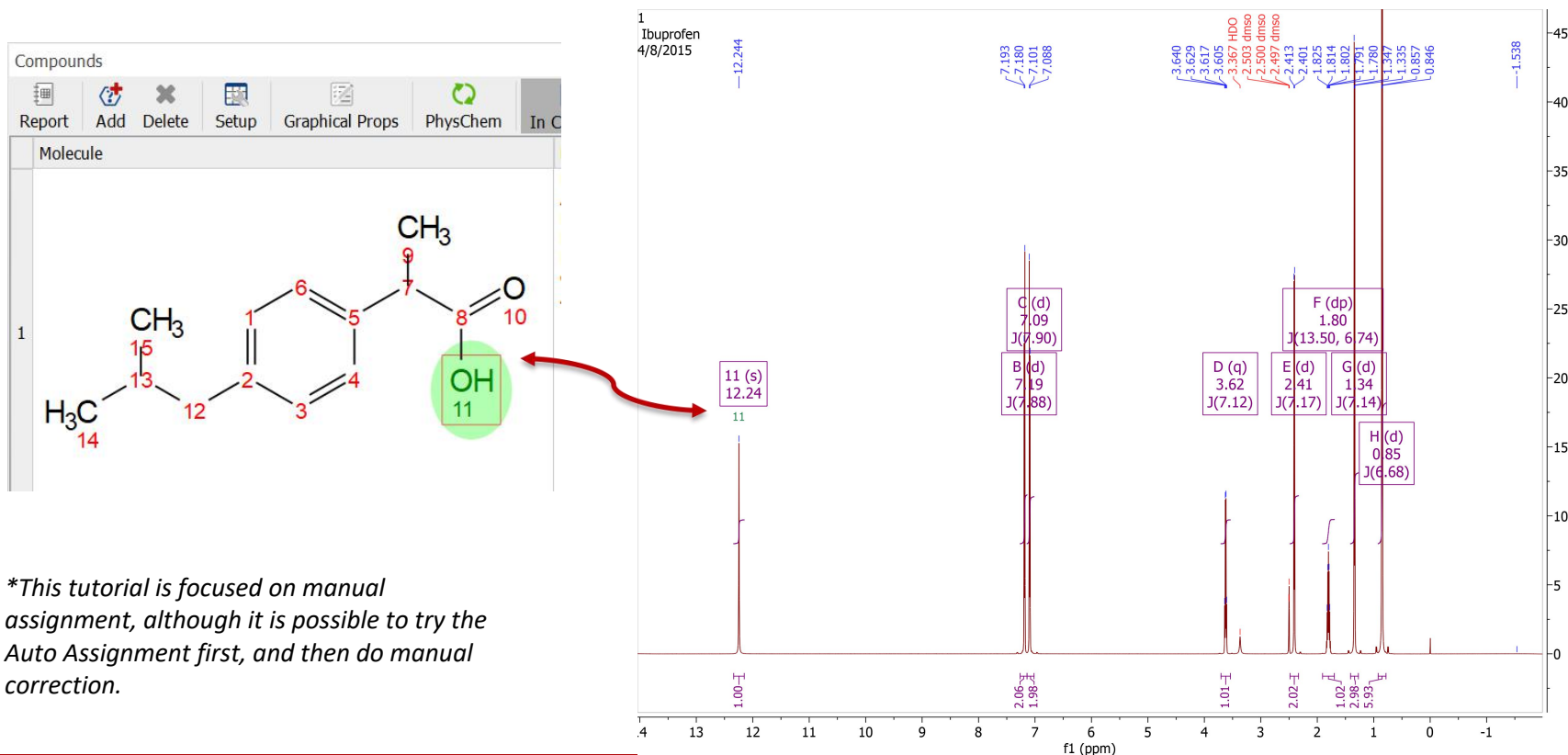
ANALYSIS

- Click A key to switch to Manual Assignment mode
- Click on a multiplet label and assign it to an atom (This is the most common way to assign H-1 peaks)
- Click on a peak and assign it to an atom
- Click and drag on the spectrum, and assign the range to an atom
- View chemical shift assignment grids on the other "linked" spectra

Assign H-1 multiplets

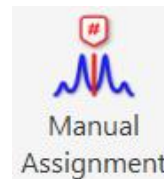


Shortcut = A

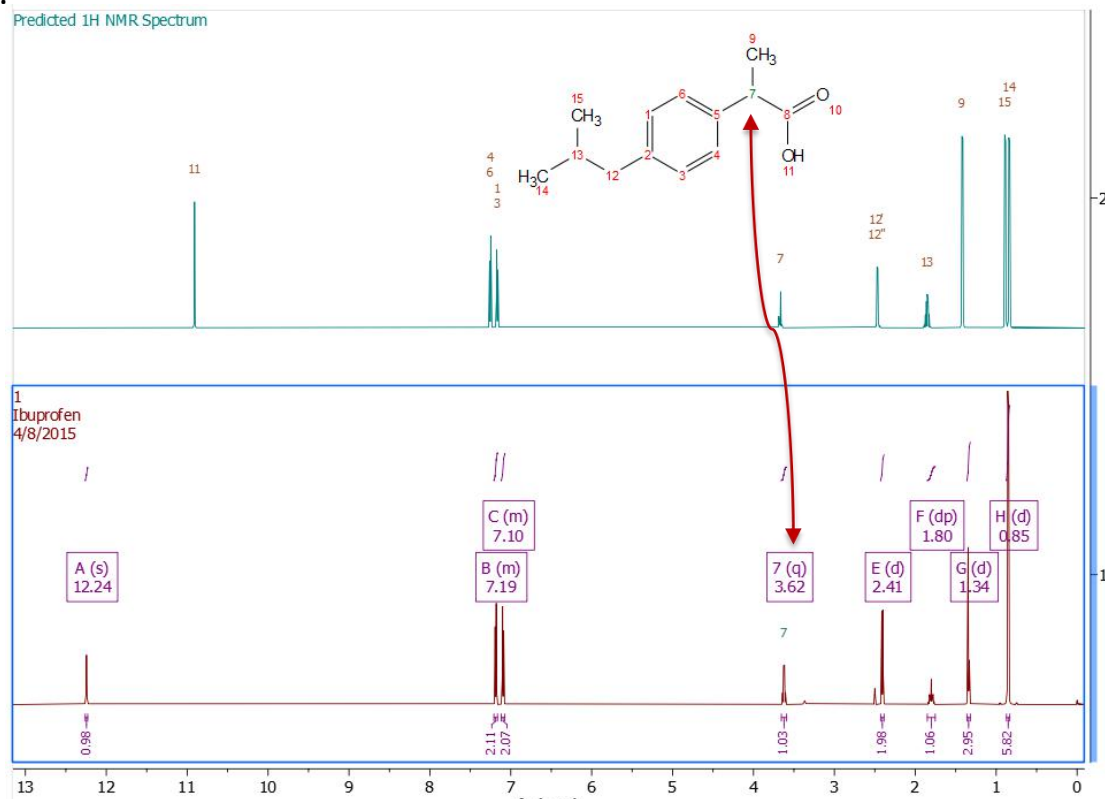


ANALYSIS

- Optionally, you can choose Predict > Predict and Compare to have a predicted H-1 spectrum stacked above the experimental one to guide your assignment of some peaks
- For example, the predicted quartet for H-7 is very close to the quartet observed at around 3.62ppm, so we can assign that observed quartet to H-7, by clicking the atom and then the multiplet label in the Manual Assignment mode.
- You can assign more multiplets in this way.



Shortcut = A



Tip: Use the following short cut keys to accelerate your assignment:

A: Assignment mode

Z: Zoom in mode

S: Swap the assignments of two atoms

<Ctrl+Space>: Switch to Zoom in mode temporarily

<ESC>: Exit from any of the modes above

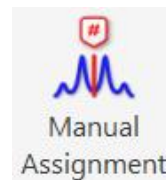
If you need a copy of the structure in a page, click the structure in the Compounds Table and click Report.

To delete the predicted C-13 spectrum, choose Stack > Stacked Items Table, and delete it from the Table.

ANALYSIS

- Make a copy of the C-13 spectrum.
- Choose Predict > Predict Compare to predict the 13C spectrum
- Use the prediction to guide the manual assignment

Assign C-13 peaks

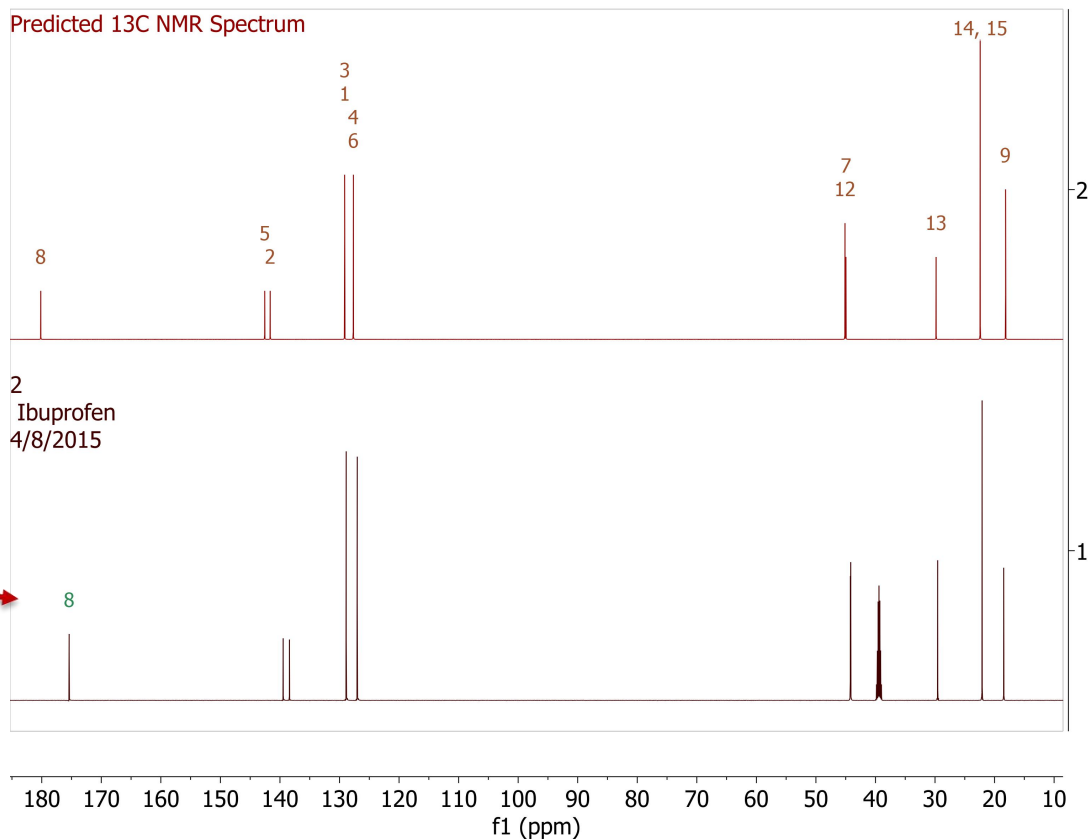
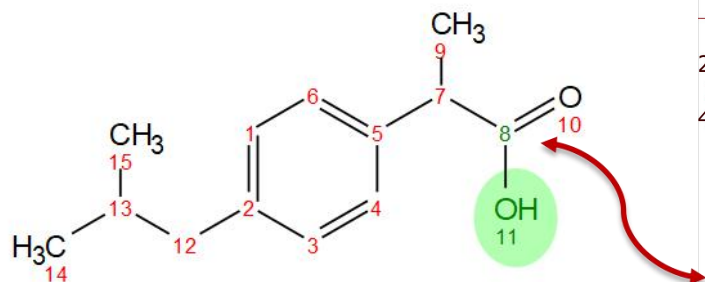


Shortcut = A

Compounds

Report Add Delete Setup Graphical Props PhysChem In

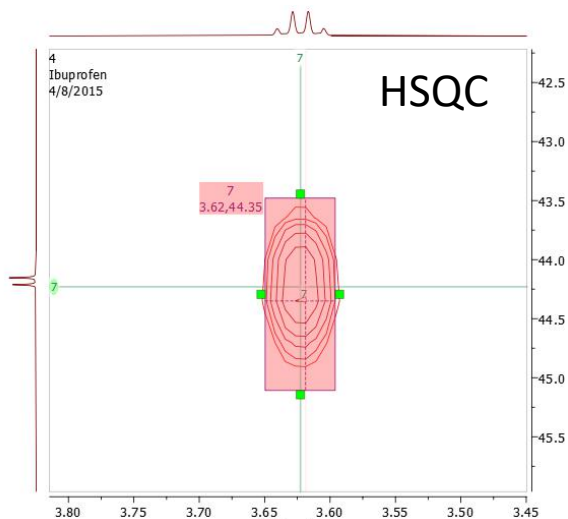
Molecule



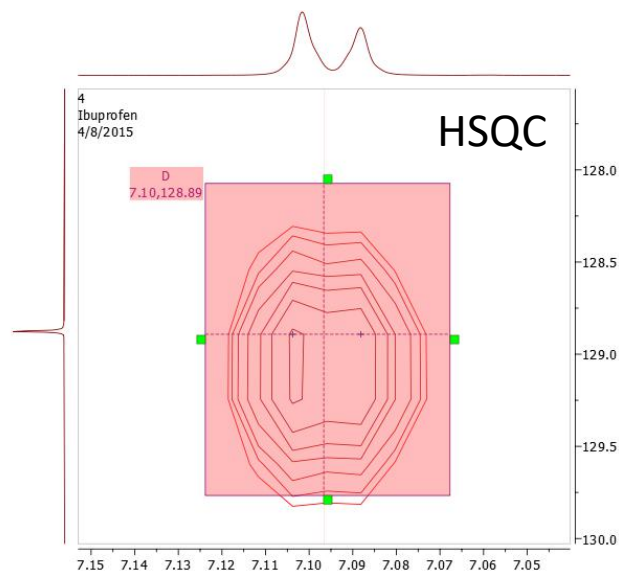
General guidance for assigning 2D spectra

ANALYSIS

- Since Mnova version 14.1, the assignment of a 2D peak is based on a “2D multiplet”, which can be either a single peak or a group of peaks.
- 2D multiplets can be automatically picked using Analysis > Auto Multiplet Analysis, and then manually corrected.
- If you don't pick any multiplets before assigning a 2D peak, it will automatically pick a multiplet around where you clicked in the 2D spectrum during the assignment.
- A 2D multiplet can be manually resized using the green blocks shown around it when you hover the cursor on it.



A HSQC multiplet with a single peak. It is assigned to CH(7)



A HSQC multiplet with two peaks. It is not assigned to any atoms yet

Tip: For better visualization of the assignment results on a 2D spectrum, it is better to suppress the display of the 2D peaks and 2D multiplet labels. Right click on the spectrum and choose Properties, and turn off those options.

ANALYSIS

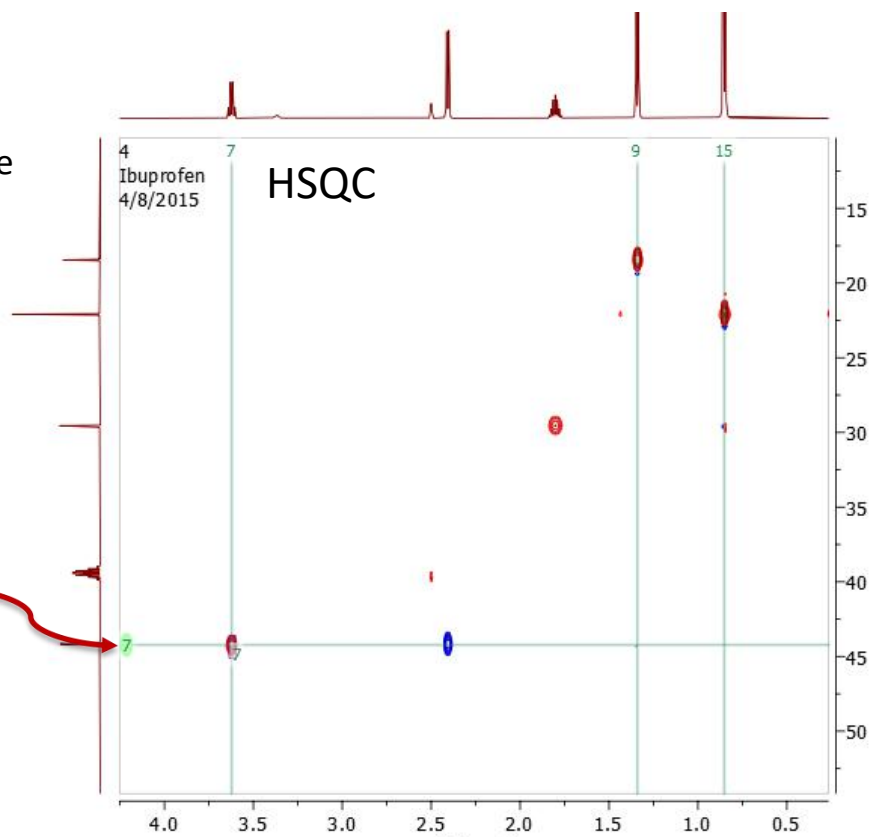
- If you assign a peak from a 1D H-1 or C-13 spectrum, the assignment is displayed for the same nucleus on the 2D spectra automatically
- If you assign a cross peak from a 2D spectrum, the assignments for the corresponding nuclei/dimensions are also displayed on the corresponding 1D and 2D spectra, automatically
- The 1D and 2D assignment results are recorded in the Assignment Table (Assignment > NMR Assignments)
- You can click and drag an assignment line to change the chemical shift.

Assignment lines across the spectra



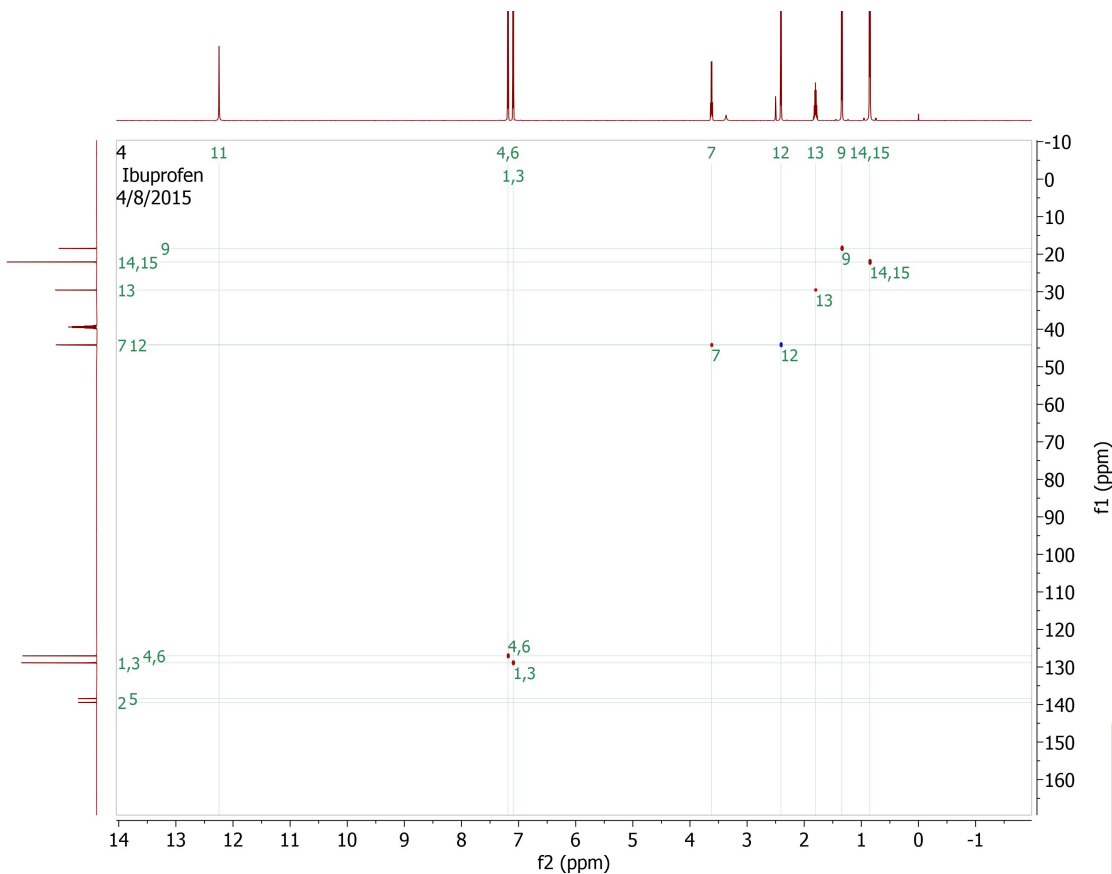
Shortcut = A

Tip: Click and drag the number ("7") to adjust the chemical shift value of this assigned peak in the C-13 dimension. The change will be reflected in all other spectra, as well as in the Assignment Table. You can do this on 1D spectra too.

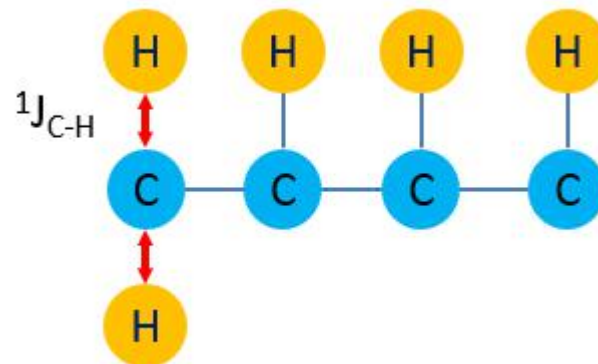


ANALYSIS

- Assign a cross peak to connected H-C atom pair: Click the peak first, then the assigned CH group (or, if H is drawn explicitly, the H first, and C next)
- Hold Alt key to display the Assign Dialog for more choices. Use "original" 1D H or C chemical shifts if available*



Assign HSQC peaks



Assign ? X

Atoms 4,6: ▼ $\delta(1H)$: 1H(f2)=7.186 ppm (multiplet 4,6) ▼

Already assigned (4):7.19 (6):7.19

Replace
 Add
 Keep Original

Assign f1

Atoms 4,6: ▼ $\delta(13C)$: f1=127.02 ppm ▼

Already assigned (4):127.02 (6):127.02

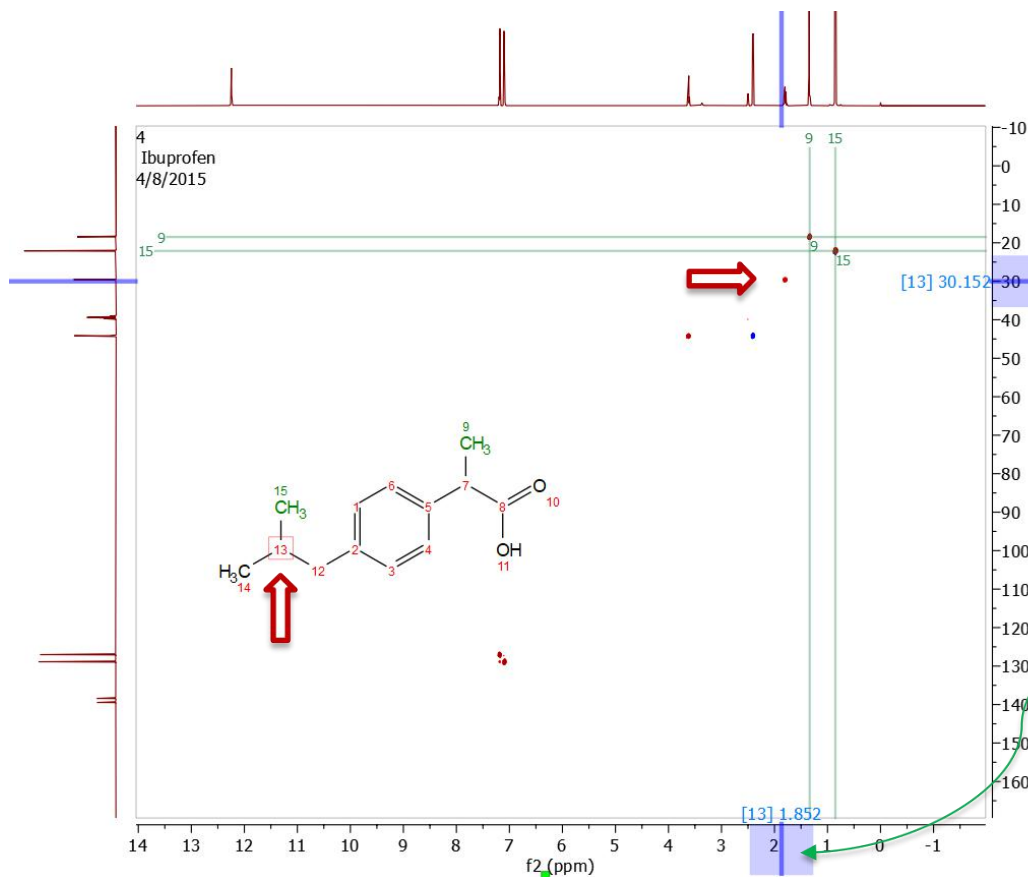
Replace
 Add
 Keep Original

**The assigned chemical shifts can be from 1D or 2D peaks. Since a 1D spectrum typically has higher resolution, it is recommended to use those from 1D. Note you can also fine tune the chemical shift by moving the atom number on an assignment line.*

Use Predict & Highlight to help assigning HSQC peaks

ANALYSIS

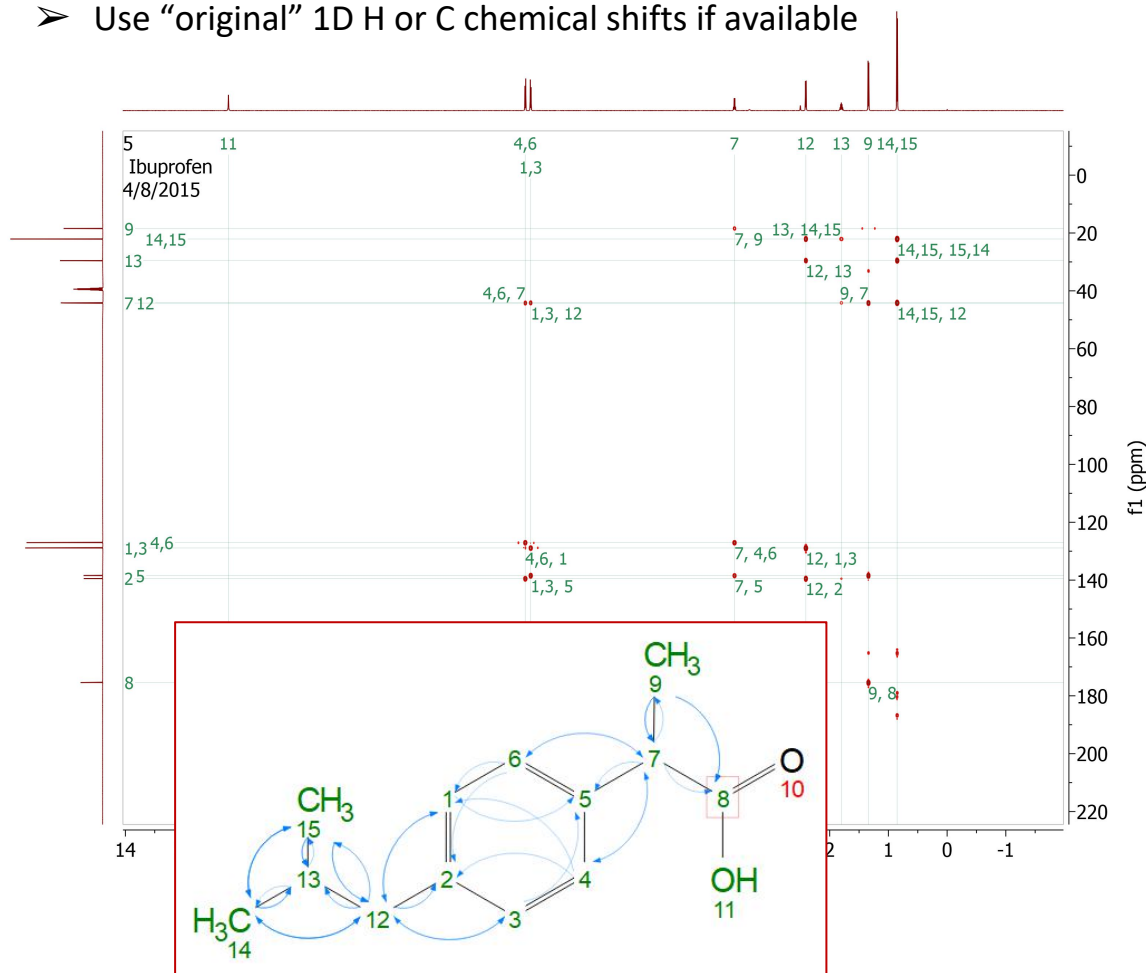
- Optionally, you can choose Predict > Predict and Highlight to display the predicted range of HSQC peaks for CH groups. The ranges can be helpful for assigning HSQC peaks to their corresponding CH groups



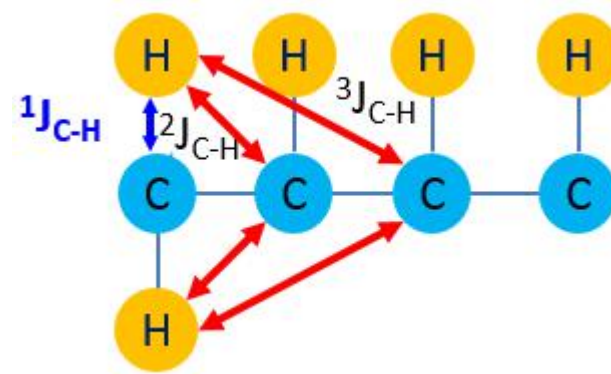
**Click on CH(13), it shows the value and error range of its predicted C-13 and H-1 shifts. This gives you an idea where to look for the cross peak to assign to this CH group.*

ANALYSIS

- Assign a cross peak to connected H-C with 2 or 3 bonds in between: Click the peak first, then the assigned H. Mnova will popup an Assign Dialog for you to choose the assigned C.
- Use "original" 1D H or C chemical shifts if available



Assign HMBC peaks



Assign ? X

Atom 3: f2=7.095 ppm

Already assigned (3): 7.08-7.11

Replace
 Add
 Keep Original

Assign f1

Atom: 12 f1=44.2 ppm

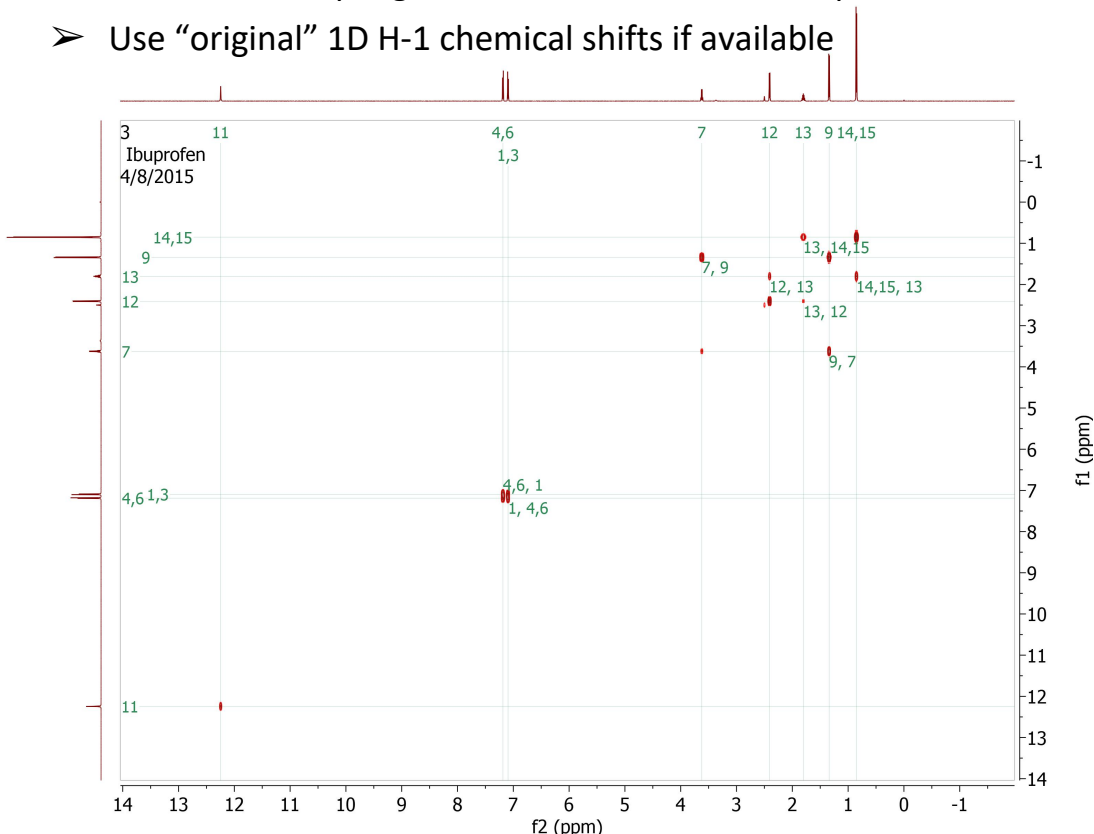
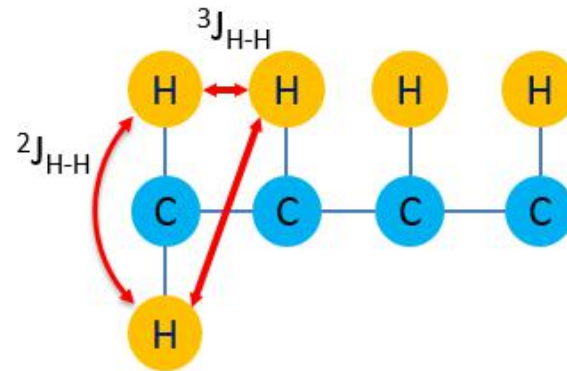
Already assigned (12): 44.20-44.22

Replace
 Add
 Keep Original

ANALYSIS

- Assign a cross peak to a H-H pair with 2 or 3 bonds in between: Click the peak first, then the assigned H on F2. Mnova will popup an Assign Dialog for you to choose the other H.
- Note weak couplings between 4-5 bonds is also possible
- Use "original" 1D H-1 chemical shifts if available

Assign COSY peaks



M Assign ? X

Atoms 4,6: ▼ δ(1H): f2=7.186 ppm ▼

Already assigned (4):7.19 (6):7.19

Replace Add Keep Original

Assign f1

Atom: 1 ▼ δ(1H): f1=7.095 ppm ▼

Already assigned (1):7.09

Replace Add Keep Original

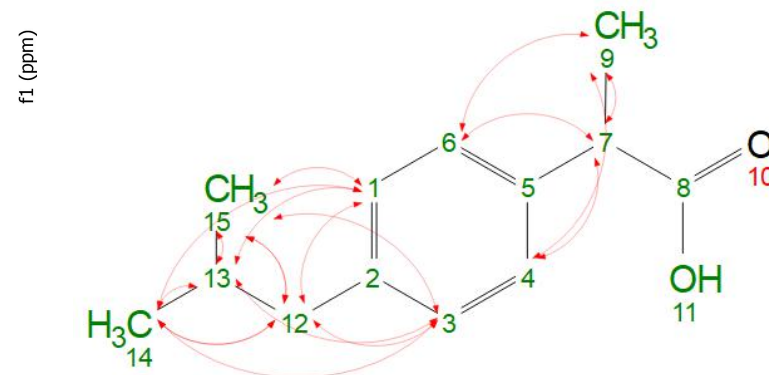
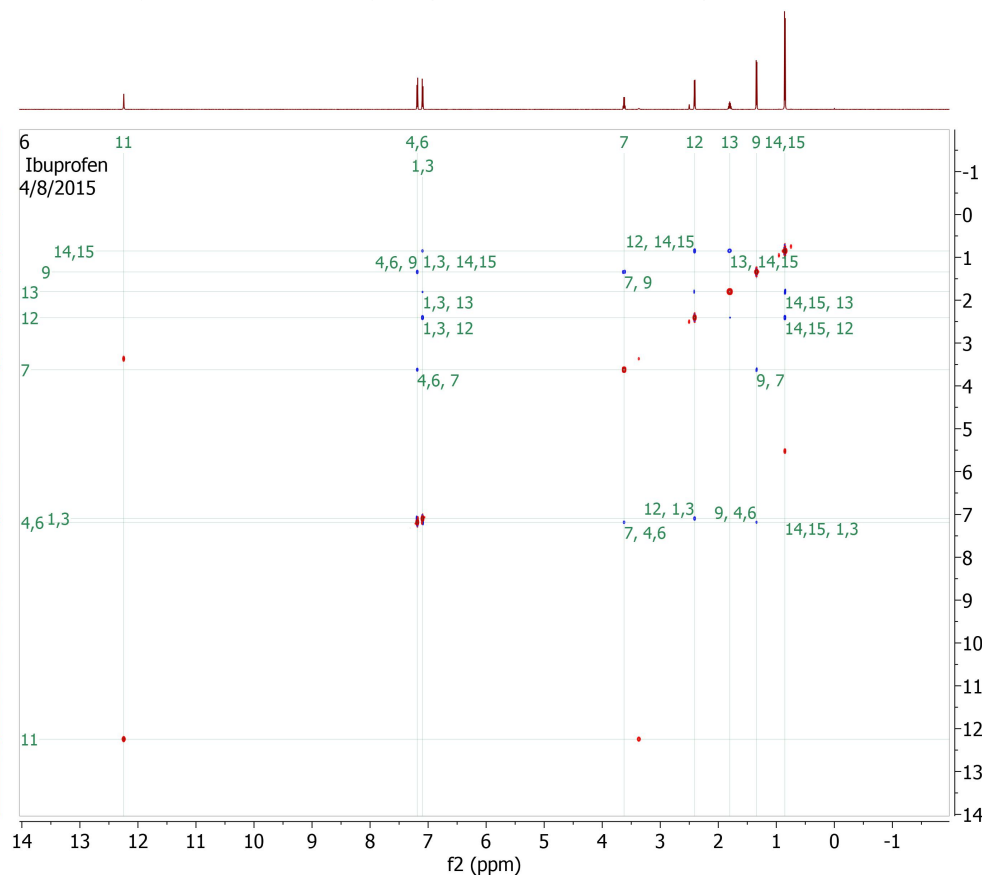
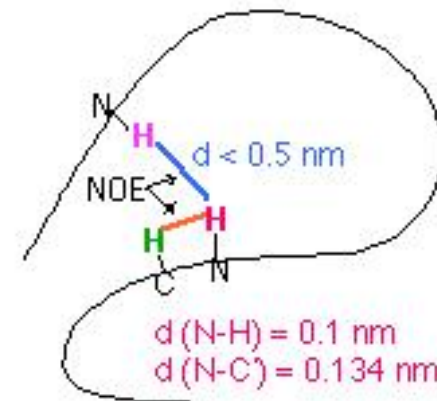
OK Cancel

Tip: Displaying the diagonal line can be helpful to distinguish cross peaks and diagonal ones. To do that, right click and open the Properties Dialog, and check Grids > Show Diagonal.

ANALYSIS

Assign NOESY peaks

- Assign a cross peak to spatially approximate H-H pair (~5Å or less): Click the peak first, then the assigned H on F2. Mnova will popup an Assign Dialog for you to choose the other H.
- For small molecules, NOE cross peaks are usually negative in phase. Positive one may be from J-couplings and should be ignored with caution.



Assignments table

REPORT

- Click Assignment > Assignment Table to display the Assignment Table
- The check boxes can be used to turn on/off the display of individual correlations on the structure

Assignments ✕

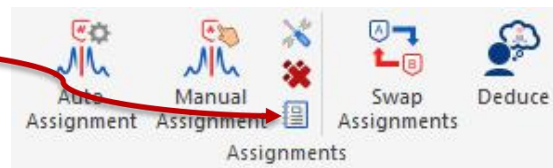
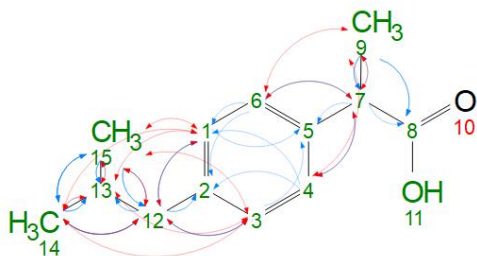
Report Copy Delete Expand Collapse Hide Setup Deduce NOE ▾

Atom	Chemical Shift	Quality	Predicted Shift	J	NOE	COSY	HSQC	HMBC	NOESY
▼ 1 C	128.88		129.12				1	✓ 4, 6, 12	
H	7.09	0.71				✓ 4, 6	1	✓ 5, 12	✓ 12, 13, 14, 15
2 C	139.46		141.62					✓ 4, 6, 12	
▼ 3 C	128.88		129.12				3	✓ 12	
H	7.09	0.71					3	✓ 5, 12	✓ 12, 13, 14, 15
▼ 4 C	127.02		127.66				4	✓ 7	
H	7.19	0.71				✓ 1	4	✓ 1, 2, 7	
5 C	138.41		142.54					✓ 1, 3, 7	
▼ 6 C	127.02		127.66				6	✓ 7	
H	7.19	0.71				✓ 1	6	✓ 1, 2, 7	
▼ 7 C	44.22		44.96				7	✓ 4, 6, 9	
H	3.62	0.71				✓ 9	7	✓ 4, 5, 6, 8, 9	
8 C	175.40		180.15					✓ 7, 9	
▼ 9 C	18.45		18.15				9	✓ 7	
H3	1.34	0.71				✓ 7	9	✓ 7, 8	
10 O									
▼ 11 O									
H	12.24	0.56							
▼ 12 C	44.16		45.12				12	✓ 1, 3, 14, 15	
H2	2.41	0.71				✓ 13	12	✓ 1, 2, 3, 13, 14,...	✓ 1, 3, 14, 15
▼ 13 C	29.55		29.81				13	✓ 12, 14, 15	
H	1.80	0.25				✓ 12, 14, 15	13	✓ 14, 15	✓ 1, 3, 14, 15
▼ 14 C	22.09		22.41				14	✓ 12, 13, 15	
H3	0.85	0.71				✓ 13	14	✓ 12, 13, 15	✓ 1, 3, 12, 13
▼ 15 C	22.09		22.41				15	✓ 12, 13, 14	
H3	0.85	0.71				✓ 13	15	✓ 12, 13, 14	✓ 1, 3, 12, 13

REPORT

- Choose Assignment > Report Assignments.
- Report the assignments on the spectrum or paste the table to another document

Report assignments



Setup Assignments Report ? X

Options

Include 13C and X-Nuclei Assignments

Include 13C Multiplicity

Include 1H Multiplicity

Include Number of protons

Order by Chemical Shift

Report Mean Chemical Shift values

Include Atom Type

Only Copy to Clipboard

Export To File:

Text (TSV) HTML

Decimal Places For 1H:

Decimal Places For 13C and X-Nuclei:

2D Correlations

Format:

n δ(n) Atom(δ)

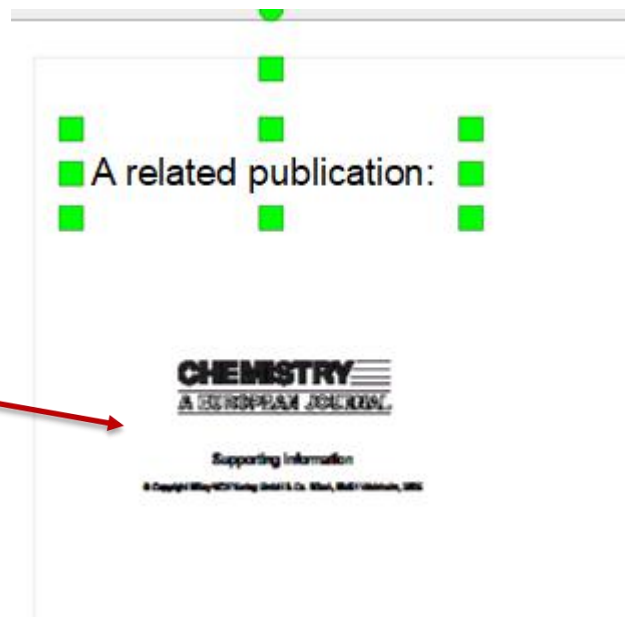
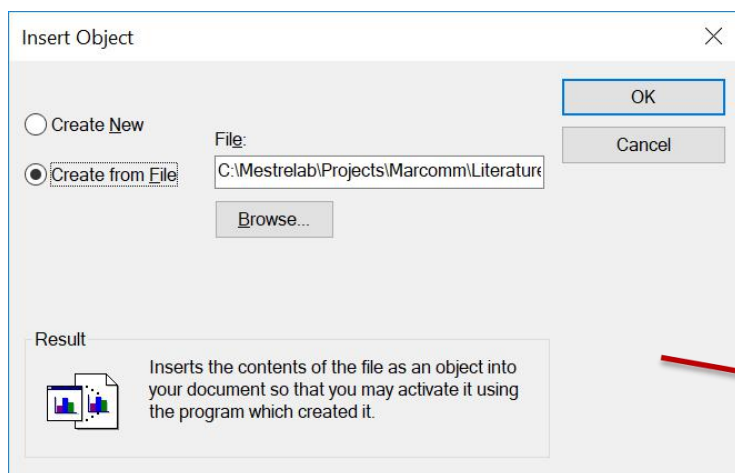
Drop Lines Without Correlation

OK Cancel

No	δ _H (Multiplicity, J, nH)	δ _C (Multiplicity, J)	HSQC-EDITED	HMBC	COSY	NOESY
1	7.09 (d, 7.9 Hz, 1H)	128.9(s)	128.9(1)	44.2(12), 138.4(5)	7.19(4), 7.19(6)	0.85(14), 0.85(15), 1.80(13), 2.41(12)
2	-	139.5(s)	-	-	-	-
3	7.09 (d, 7.9 Hz, 1H)	128.9(s)	128.9(3)	44.2(12), 138.4(5)	-	0.85(14), 0.85(15), 1.80(13), 2.41(12)
4	7.19 (d, 7.9 Hz, 1H)	127.0(s)	127.0(4)	44.2(7), 128.9(1), 139.5(2)	7.09(1)	1.34(9), 3.62(7)
5	-	138.4(s)	-	-	-	-
6	7.19 (d, 7.9 Hz, 1H)	127.0(s)	127.0(6)	44.2(7), 128.9(1), 139.5(2)	7.09(1)	1.34(9), 3.62(7)
7	3.62 (q, 7.1 Hz, 1H)	44.2(s)	44.2(7)	18.5(9), 127.0(4), 127.0(6), 138.4(5), 175.4(8)	1.34(9)	1.34(9), 7.19(4), 7.19(6)
8	-	175.4(s)	-	-	-	-
9	1.34 (d, 7.1 Hz, 3H)	18.5(s)	18.5(9)	44.2(7), 175.4(8)	3.62(7)	3.62(7), 7.19(4), 7.19(6)
11	12.24 (s, 1H)	-	-	-	-	-
12	2.41 (d, 7.2 Hz, 2H)	44.2(s)	44.2(12)	22.1(14), 22.1(15), 29.6(13), 128.9(1), 128.9(3), 139.5(2)	1.80(13)	0.85(14), 0.85(15), 7.09(1), 7.09(3)
13	1.80 (dp, 13.5, 6.7 Hz, 1H)	29.6(s)	29.6(13)	22.1(14), 22.1(15)	0.85(14), 0.85(14), 0.85(15), 7.09(1), 0.85(15), 2.41(12)	7.09(3)
14	0.85 (d, 6.7 Hz, 3H)	22.1(s)	22.1(14)	22.1(15), 29.6(13), 44.2(12)	1.80(13)	1.80(13), 2.41(12), 7.09(1), 7.09(3)
15	0.85 (d, 6.7 Hz, 3H)	22.1(s)	22.1(15)	22.1(14), 29.6(13), 44.2(12)	1.80(13)	1.80(13), 2.41(12), 7.09(1), 7.09(3)

Insert a PDF to the document

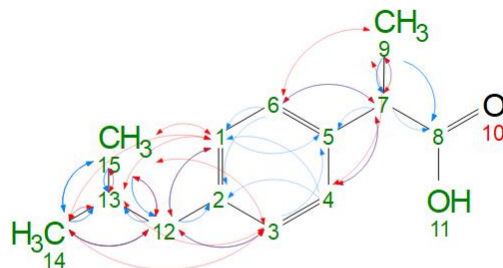
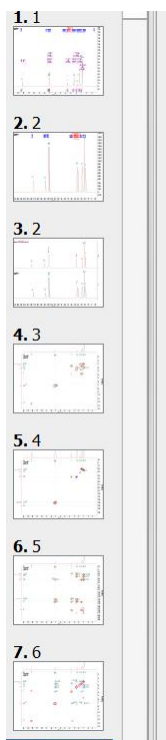
- Choose Home > Insert Object, choose Create from File, and insert a PDF to the document
- A preview logo of the document is displayed.
- Add a text box annotation to it
- You can double click on the preview to open it



Save the results

REPORT

- Choose File > Export to PDF to save a PDF report of the page.
- Chose File > Save as to save all the results to a .mnova file.
- Save all the results to a database (see steps later)
- Now you can close the document or continue to add other spectra to it.



No	δ_H (Multiplicity, J, nH)	δ_C (Multiplicity, J)	H SQC-EDITED	HMBC	COSY	NOE SY
1	7.09 (d, 7.9 Hz, 1H)	128.9(s)	128.9(1)	44.2(12), 138.4(5)	7.19(4), 7.19(6)	0.85(14), 0.85(15), 1.80(13), 2.41(12)
2	-	139.5(s)	-	-	-	-
3	7.09 (d, 7.9 Hz, 1H)	128.9(s)	128.9(3)	44.2(12), 138.4(5)	-	0.85(14), 0.85(15), 1.80(13), 2.41(12)
4	7.19 (d, 7.9 Hz, 1H)	127.0(s)	127.0(4)	44.2(7), 128.9(1), 139.5(2)	7.09(1)	1.34(9), 3.62(7)
5	-	138.4(s)	-	-	-	-
6	7.19 (d, 7.9 Hz, 1H)	127.0(s)	127.0(6)	44.2(7), 128.9(1), 139.5(2)	7.09(1)	1.34(9), 3.62(7)
7	3.62 (q, 7.1 Hz, 1H)	44.2(s)	44.2(7)	18.5(9), 127.0(4), 127.0(6), 138.4(5), 175.4(8)	1.34(9)	1.34(9), 7.19(4), 7.19(6)
8	-	175.4(s)	-	-	-	-
9	1.34 (d, 7.1 Hz, 3H)	18.5(s)	18.5(9)	44.2(7), 175.4(8)	3.62(7)	3.62(7), 7.19(4), 7.19(6)
11	12.24 (s, 1H)	-	-	-	-	-
12	2.41 (dp, 7.2 Hz, 2H)	44.2(s)	44.2(12)	22.1(14), 22.1(15), 29.6(13), 128.9(1), 128.9(3), 139.5(2)	1.80(13)	0.85(14), 0.85(15), 7.09(1), 7.09(3)
13	1.80 (dp, 13.5, 6.7 Hz, 1H)	29.6(s)	29.6(13)	22.1(14), 22.1(15)	0.85(14), 0.85(15), 2.41(12)	0.85(14), 0.85(15), 7.09(1), 7.09(3)
14	0.85 (d, 6.7 Hz, 3H)	22.1(s)	22.1(14)	22.1(15), 29.6(13), 44.2(12)	1.80(13)	1.80(13), 2.41(12), 7.09(1), 7.09(3)
15	0.85 (d, 6.7 Hz, 3H)	22.1(s)	22.1(15)	22.1(14), 29.6(13), 44.2(12)	1.80(13)	1.80(13), 2.41(12), 7.09(1), 7.09(3)

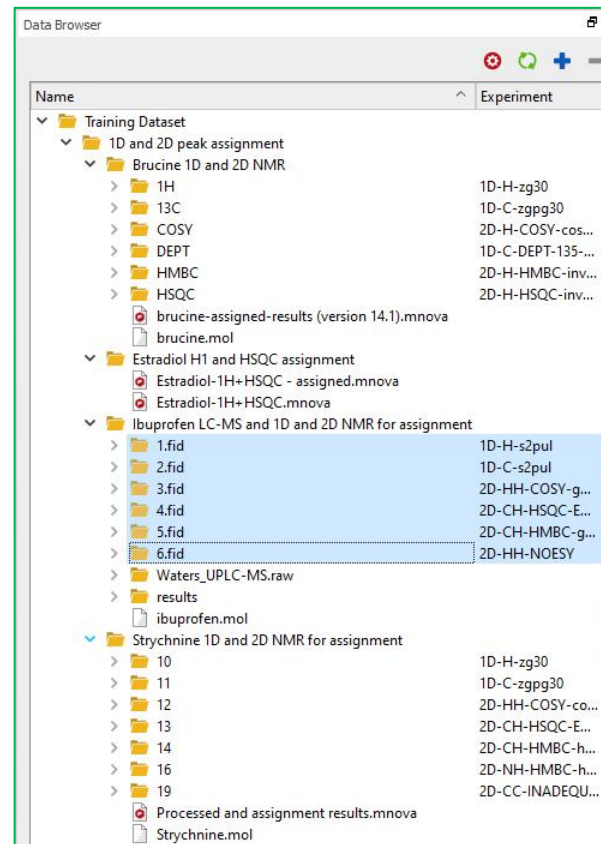
.pdf doc

.mnova doc

Database

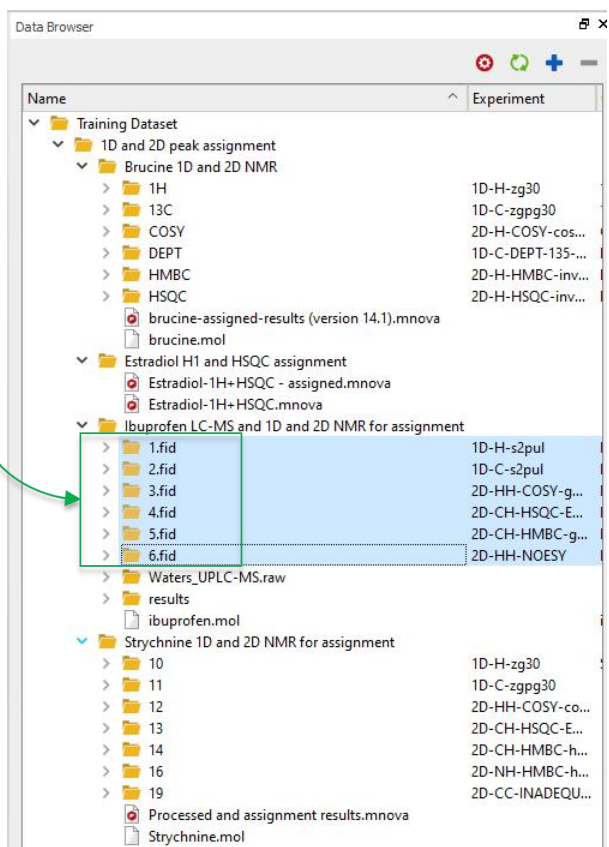
More about peak assignment

- Manual assignment of 1D and 2D peaks is not only an effective way to verify a proposed structure, but also a proven way to learn to understand and analyze 1D and 2D NMR.
- Usually you start with the 1H, 13C and HSQC spectra, if available, with the assistance of the predicted peaks or ranges as a guidance. Then you extend the assignment to the other 2D spectra such as COSY and HMBC. While assigning COSY or HMBC peaks, conflicts with the previous assignments may be discovered and hence corrections can be done. If the conflicts cannot be resolved, it may imply that the structure is wrong.
- There are a total of 4 datasets with 1D and 2D NMR spectra for practicing spectral processing, peak assignment that come with this tutorial: Ibuprofen, Brucine, Strychnine, and Estradiol, along with results of full assignments. You can use them for practice.



Saving Processed Spectra and Analysis Results to a Database

Sample data

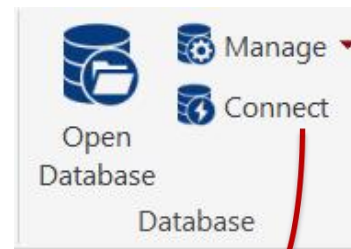


Note: You will need a Mnova DB Client license and install Mnova MyData Server for this session

DATABASE

- Make sure MyData Server is installed and running as a service*
- Connect to MyData Server with the default account setting
- Create and open a new database (Database > Manage > Add)

Install MyData DB Server



Connection

Server: localhost

Port: 5504

User: Test

Password: database

Save Password Show Password

OK Cancel

**If not yet, download the DB MyData Server Installer from <http://mestrelab.com/download/mnova/db/> and install it. You may need to download the latest Java too. No license is needed for MyData DB Server.*

Note: MyData is a personal version of Mnova DB Group or Enterprise Server.

DATABASE

Save data to database



Save to Database ▾

- Choose Database > Save to Database
- Click All in the Select Items dialog
- Click OK to save all items to the new database

Select Items

Select

None

All

	Page	Type	Preview	Description
1	7	Molecule		
2	1	NMR Spectrum		1
3	2	NMR Spectrum		2
4	3	NMR Spectrum		3
5	4	NMR Spectrum		4
6	5	NMR Spectrum		5
7	6	NMR Spectrum		6
8	7	Text	A related publication:	A related publication:
9	7	Text	Chen Peng's assignment results	Chen Peng's assignment results
10	7	Text		Noδ H (Multiplicity...
11	7	OLE Object		

OK Cancel

Browse and download from database

DATABASE

- Choose Database > Browse or Show Record
- Display the contents in Record View (or Table View)
- Choose File > New in Mnova to open a new document
- Right click on the spectrum in the Database Dialog and choose Paste Record to Mnova to download the whole record to Mnova



Database - Record View

File Edit View Configure

Molecule Preview

Button Navigator

mndb://Test@localhost:5504/DB1/1 Molecule Molecule C13H18O2

NMR Preview

Mass Preview

Fields

	Field	Content
1:0:8	Molecular Formula	C13H18O2
1:0:10	Monoisotopic Mass	206.13068
1:1:34	Title	1
1:1:38	Solvent	dmsO
		C:/Mestrelab/Projects/Marcomm/Literature & Docs/Sample and Templates/Training for

1: Ibuprofen
4/8/2015

11 (s) 12.24

1,3 (d) 7.09 [7.90]

4,6 (d) 7.19 [7.88]

7 (e) 3.62 [7.12]

12 (d) 2.41 [7.17]

13 (dp) 1.80 [13.58, 6.74]

9 (d) 1.34 [7.14]

14, 15 (d) 0.65 [6.68]

ft (ppm)

Copy Ctrl+C

Paste Item to Mnova

Paste Record to Mnova

Update 1H Prediction DB

Update 13C Prediction DB

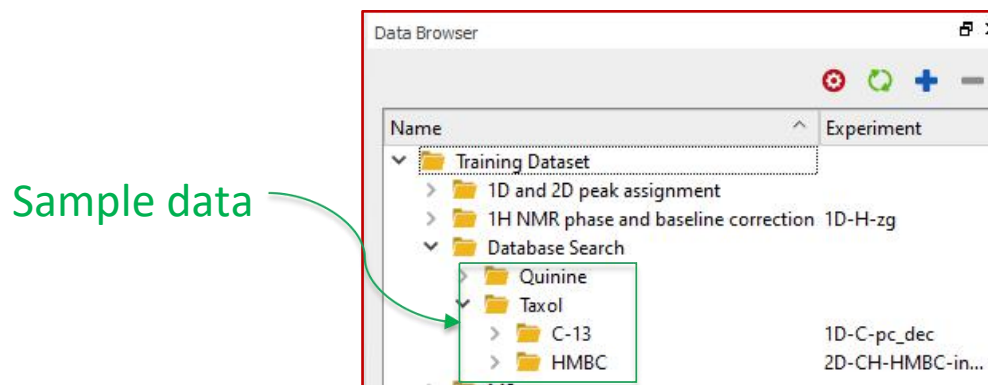
DATABASE

- Search by text
- Search by peaks
- Search by multiplets
- Search by 2D peaks
- Search by structure/substructure

Search your own database



Searching Wiley C-13 and H-1 Databases from Mnova



Note: You will need a subscription to Wiley Databases from Mestrelab.

Search Wiley databases

- Wiley C-13 and H-1 databases are collection of published C13 and H-1 spectra of known organic compounds. They are usually installed on a central server (Mnova DB Enterprise Server) for remote access.
- Make sure you have an account to access the Wiley databases. Contact your library administrators if you don't have it.
- Once connected to the server, the searching methods are the same as with MyData DB Server.

Wiley C-13 database

- 13C NMR Spectra: 268,000
- Structures: 268,000
- Compounds: 228,000
- Replicate Spectra: 40,000
- Collected and reviewed by Wolfgang Robien, with carefully reviewed peak assignments, specific measurement and instrument parameters, where available.
- Using proprietary quality assurance measures developed by the author and Wiley, this is the finest collection of 13C NMR spectra available

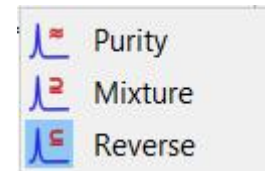
Search Wiley database

- 1H NMR Spectra: 157,000
- Structures: 157,000
- Compounds: 155,000
- Replicate Spectra: 2,000
- Collected and reviewed by Alexander Yarkov, with carefully reviewed peak assignments, and specific measurement and instrument parameters, where available.
- Using proprietary quality assurance measures developed by the author and Wiley, this is the finest and largest collection of 1H NMR spectra available.

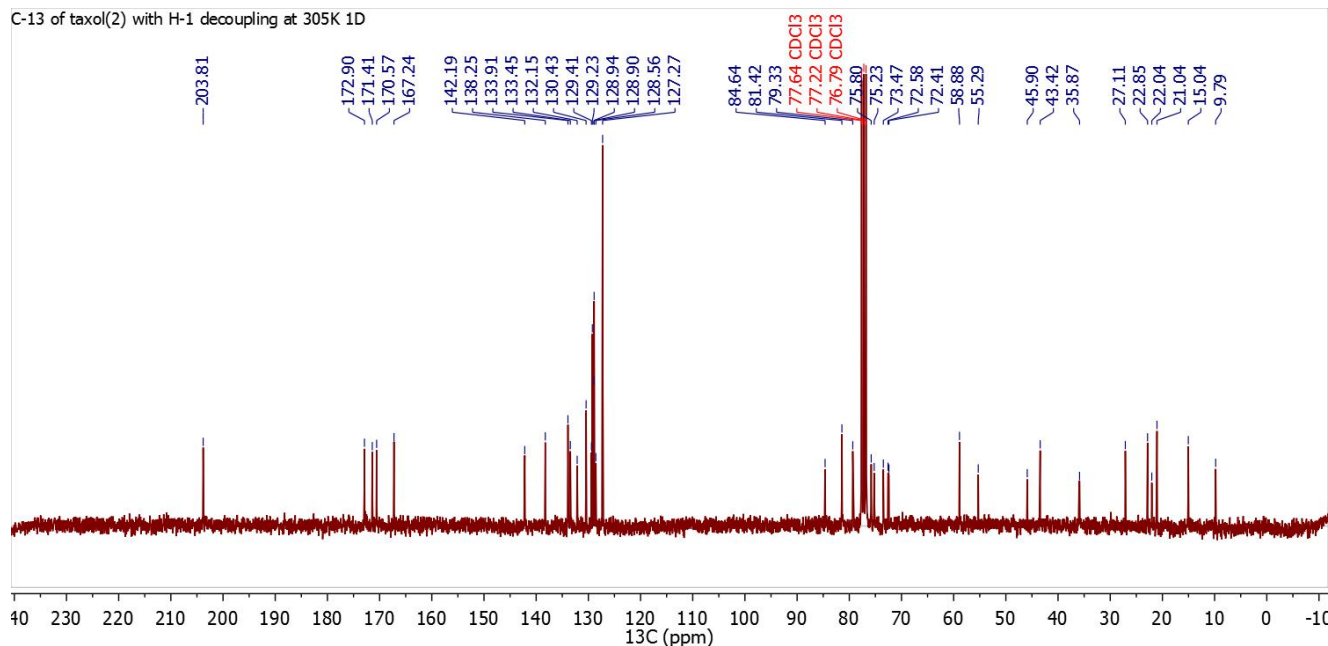
Search a C-13 spectrum

WILEY DB

- Open Training Data Sets > Database Search > Taxol > C-13 > fid
- Do Analysis > Auto Peak Picking, right click and choose peak search
- Peak Search Settings:
 - *Reverse to penalize only unmatched query peaks.*
 - *Tolerance = +/- 1.0 ppm*
- Choose the top hits with score > 800 (out of 1000)
- Taxol has the highest score 948



C-13 of taxol(2) with H-1 decoupling at 305K 1D



C-13 search results

WILEY DB

Hit list, and preview of hits

The screenshot displays the 'Database - Search Results' window with the following components:

- Molecule Preview:** Shows a chemical structure of a complex organic molecule with handwritten red text 'Str.' next to it.
- NMR Preview:** Shows a ¹³C NMR spectrum with handwritten red text '13C' above it.
- Fields Table:**

Field	Content
32923:1:34 Title	TAXOL
32923:1:35 Spectrum Comment	G.N.CHMURNY,B.D.HILTON,S.BROBST,S.A.LO...
32923:1:38 Solvent	CDCL3
32923:1:43 Data File Name	/home/michael/RefDB01/data/WileyCNMR/temp/CNMROC2_001_32922.dx
32923:1:45 Nucleus	13C
32923:1:46 Acquisitio...	
32923:1:55 Spectrome...	500.0
32923:1:57 Spectral W...	125000.0
32923:1:58 Temperature	298.0

Handwritten red text 'Other info' is written next to the 'Nucleus' field.
- Query:** Shows '1D Peaks Query: 39 Peaks'.
- Scores Table:**

Record	Item Type	Item Number	Search Score
1	mndb... NMR S...	1	948
2	mndb... NMR S...	1	948
3	mndb... Molec...	0	923
4	mndb... Molec...	0	923
5	mndb... NMR S...	1	923
6	mndb... NMR S...	1	923
7	mndb... NMR S...	1	897
8	mndb... NMR S...	1	897
9	mndb... Molec...	0	897
10	mndb... NMR S...	1	897
11	mndb... NMR S...	1	897
12	mndb... Molec...	0	897
13	mndb... NMR S...	1	897
14	mndb... NMR S...	1	871
15	mndb... Molec...	0	871
16	mndb... NMR S...	1	871
17	mndb... Molec...	0	871
18	mndb... Molec...	0	871
19	mndb... NMR S...	1	871
20	mndb... NMR S...	1	846
21	mndb... Molec...	0	846
22	mndb... NMR S...	1	846
23	mndb... Molec...	0	846
24	mndb... NMR S...	1	846
25	mndb... NMR S...	1	846
26	mndb... Molec...	0	846

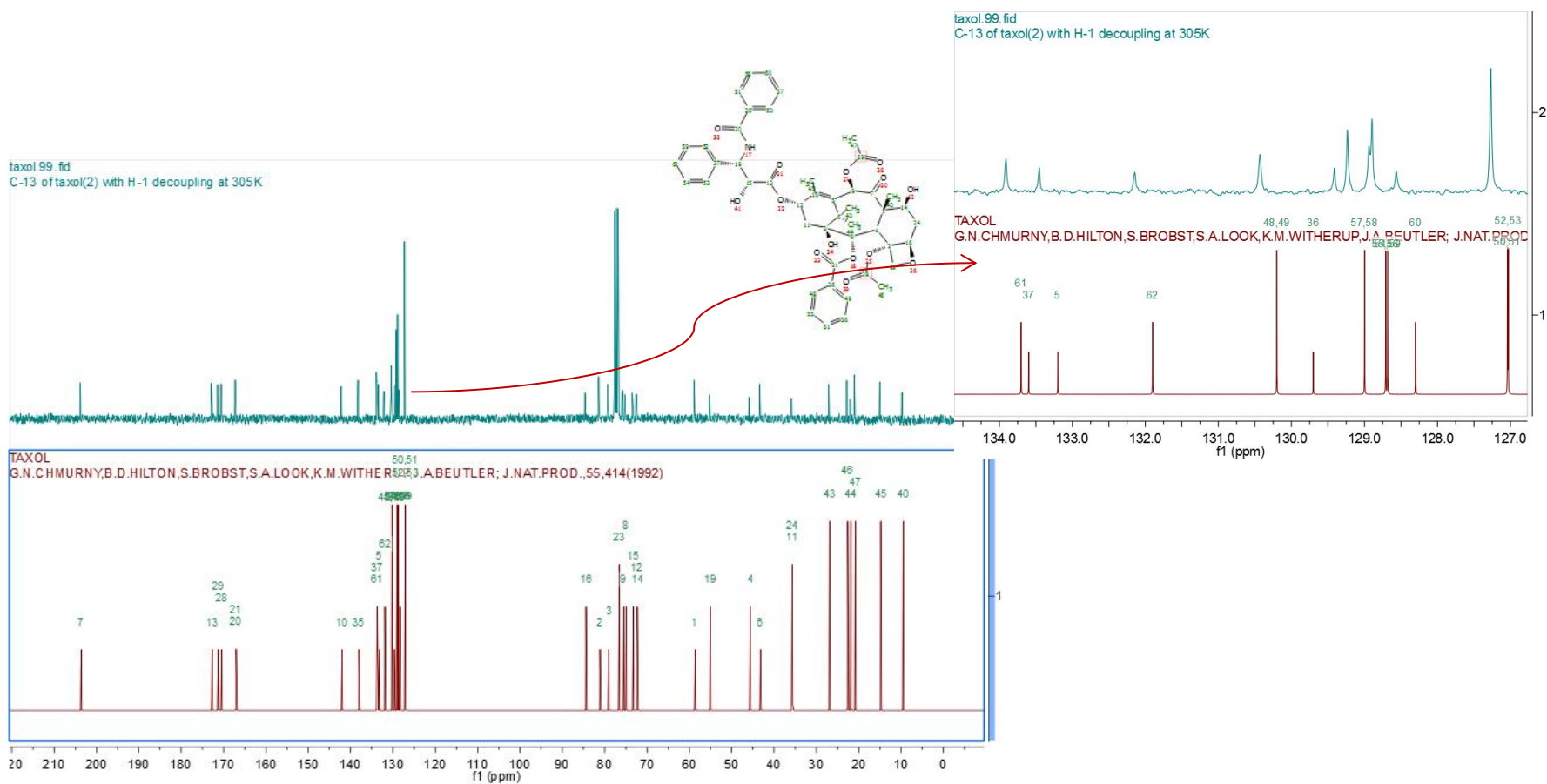
A red arrow points from the 'Hit list' label to the Scores table.

Hit list

Search a C-13 spectrum

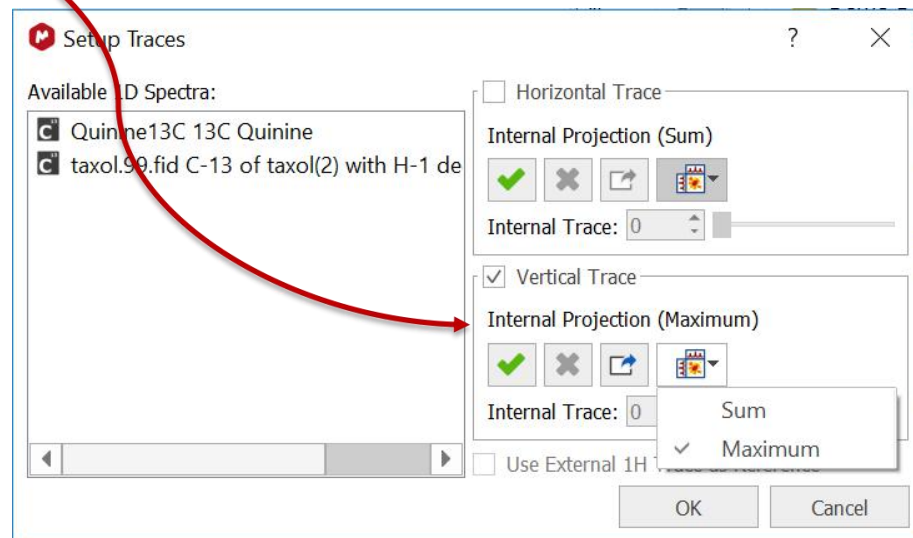
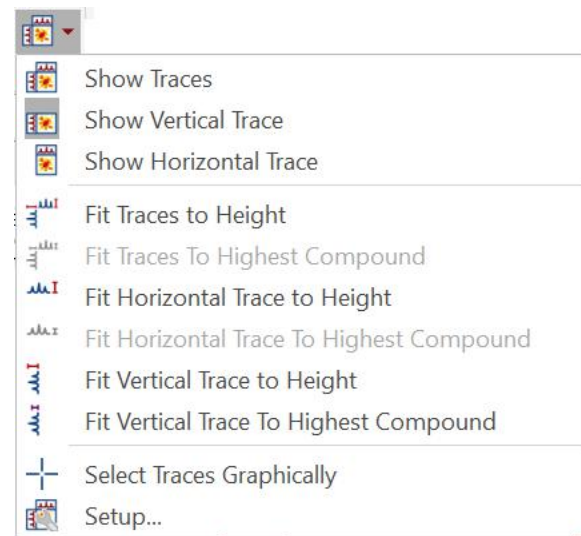
WILEY DB

- Right click on the spectrum, choose Paste Record to Mnova to download the hit spectrum and compare with the experimental one



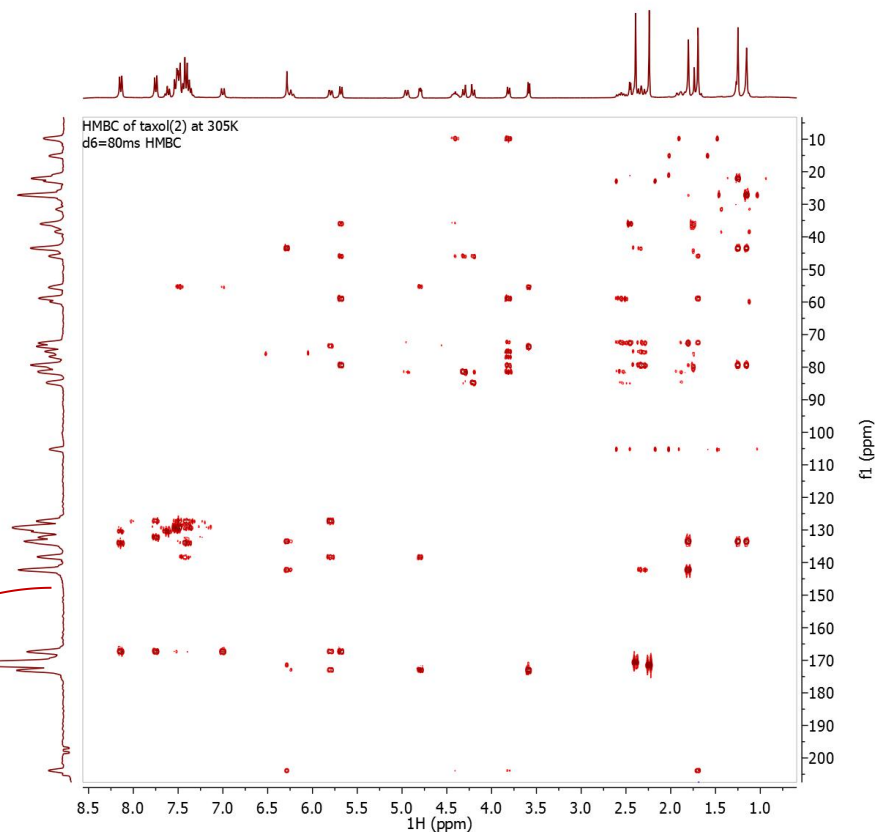
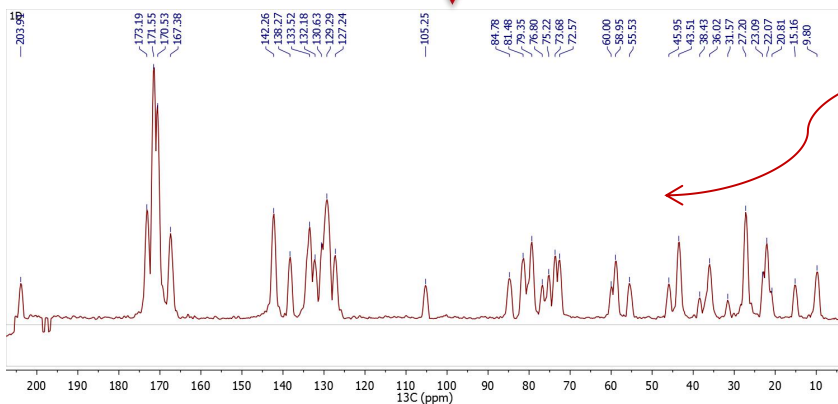
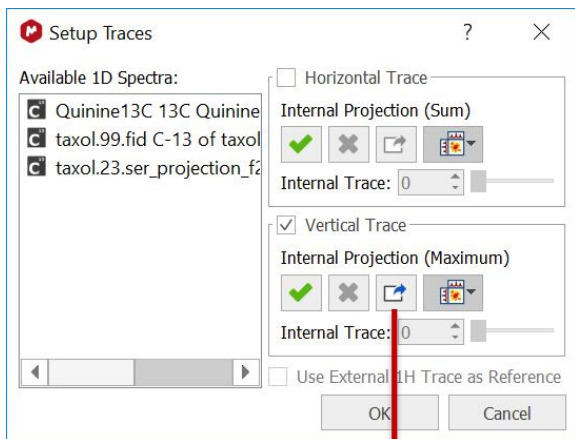
Search C-13 peaks from HMBC

- ❑ Open Training Data Sets > Database Search > Taxol > HMBC > ser
- ❑ Choose Processing > More Processing > Reduce t1 Noise.
- ❑ Choose View > 2D Traces > Setup
- ❑ In the Setup Traces dialog, choose to display Internal Projection (Maximum) as Vertical Trace



Search C-13 peaks from HMBC

- Click Extract in the Setup Trace Dialog to generate a pseudo 1D C13 spectrum in a new page, and do auto peak picking on the pseudo C13 spectrum.



Search C-13 peaks from HMBC

WILEY DB

- Right click on the C-13 spectrum and do a peak search again. Taxol is listed as one of the top hits again

The screenshot displays the Mestrelab Research software interface with the following components:

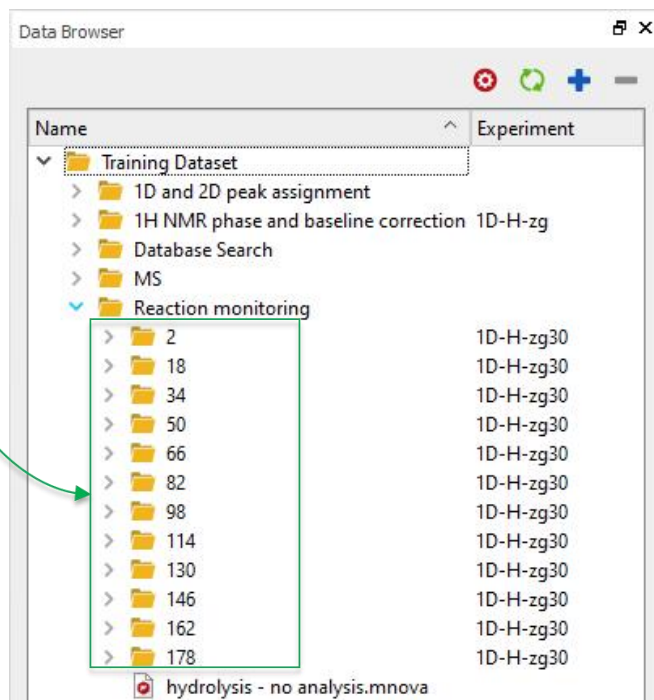
- Molecule Preview:** Shows the chemical structure of Taxol.
- Fields:** A table with the following data:

Field	Content
32923:0:8 Molecular Formula	C47H51NO14
32923:0:10 Monoisotopic Mass	853.330959
32923:1:34 Title	TAXOL
32923:1:35 Spectrum Comment	G.N.CHMURNY,B.D.HILTON,S...
32923:1:38 Solvent	CDCL3
32923:1:43 Data File Name	/home/michael/RefDB01/data/WileyCNMR/temp/CNMROC2_001_32922.dx
- NMR Preview:** A 13C NMR spectrum showing peaks at various chemical shifts.
- Mass Preview:** A mass spectrum showing the molecular ion peak.
- Scores:** A table of search results:

Record	Item Type	Item Number	Search Score
1	mndb...	NMR S...	882
2	mndb...	NMR S...	882
3	mndb...	Molec...	882
4	mndb...	NMR S...	882
5	mndb...	NMR S...	882
6	mndb...	NMR S...	882
7	mndb...	Molec...	852
8	mndb...	NMR S...	852
9	mndb...	Molec...	852
10	mndb...	NMR S...	852
11	mndb...	Molec...	852
12	mndb...	NMR S...	852
13	mndb...	NMR S...	852
14	mndb...	NMR S...	852
15	mndb...	Molec...	852
16	mndb...	NMR S...	852
17	mndb...	NMR S...	852
18	mndb...	Molec...	823
19	mndb...	NMR S...	823
20	mndb...	Molec...	823
- Query:** A 1D Peaks Query showing 34 peaks.

Processing Arrayed Spectra for Reaction Monitoring

Sample data



Stack a few spectra

ARRAYED SPECTRA

- Open the first 3 spectra from the Multiple 1H spectra folder in Data Browser
- The Stacked Ribbon is visible if you highlight multiple spectra in the Pages View

The screenshot displays the MestReNova software interface. The top menu bar includes File, Home, View, Molecule, Prediction, Tools, Database, Verification, Elucidation, STACK, Processing, Analysis, Assignments, Quantitation, and Chen's Tools. The STACK menu is expanded, showing options like Stack Options, Extract Active Item, Adjust Stacked Items, Auto Scale, Multiply Divide, Mode Invert, Show Select, Stacked Items Table, Align Spectra, Reference Alignment, DOSY Transform, and Arrayed Data Table.

The main window shows a 1D ¹H NMR spectrum titled "Multiple 1H spectra.34.fid". The x-axis is labeled "f1 (ppm)" and ranges from 16 to -4. The y-axis ranges from -2400 to 2400. The spectrum shows a sharp peak at approximately 1.2 ppm and a smaller peak at approximately 4.8 ppm.

The Data Browser on the right shows a tree view of the file structure. The "Multiple 1H spectra" folder is expanded, and three subfolders (2, 18, and 34) are highlighted. A red arrow points from the "34" folder to the spectrum in the main window.

The Pages view on the left shows three thumbnails of the spectra, labeled "1. Multiple 1H spec", "2. Multiple 1H spec", and "3. Multiple 1H spec". A red arrow points to the "3. Multiple 1H spec" thumbnail.

ARRAYED SPECTRA

Stack a few spectra

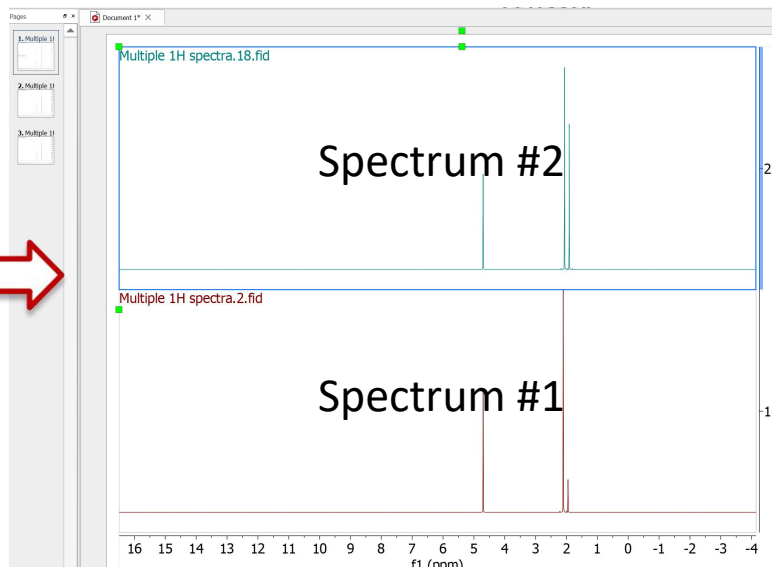
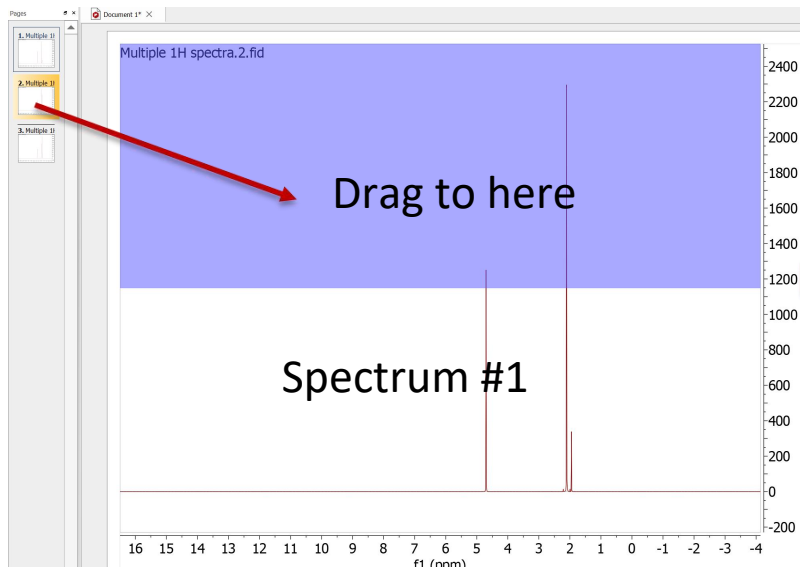
- You can use the Stack Items or Superimpose Items tools to stack or superimpose the highlighted spectra in the Pages View, or:
- Drag the thumbnail of another spectra from the Pages View to the current spectrum to stack them in desired way.
- Continue to drag the 3rd spectrum to the stack. Note you can put the spectrum to the top, middle or bottom, or to replace an existing spectrum in the stack.
- Try the different Stacking Mode, and other tools in the Stacked Ribbon



Stack
Items



Superimpose
Items



Stack many spectra

ARRAYED SPECTRA

- Choose Tools > Loaded Scripts > Directory Spectra Stack, navigate to the directory “Multiple H-1 Spectra” in the training dataset. Click OK to import and stack all of them.

Import Spectra Stack

Data Folder: s by Chen/Training Data Sets/Multiple 1H spectra ...

Order: By Name

File Path Filtering

File Name Masks: fid *.fid *.jdx

Folder Name Masks:

Preview Files

Spectral Data Filtering

Parameter Nucleus =

Chunking

First Spectrum: 1 Number of Chunks: 1

Chunk Size: 1 Step to Next Chunk: 2

Visualization

View: Stacked Decimation Step: 1

Palette: Default

Import Array Values

File: ...

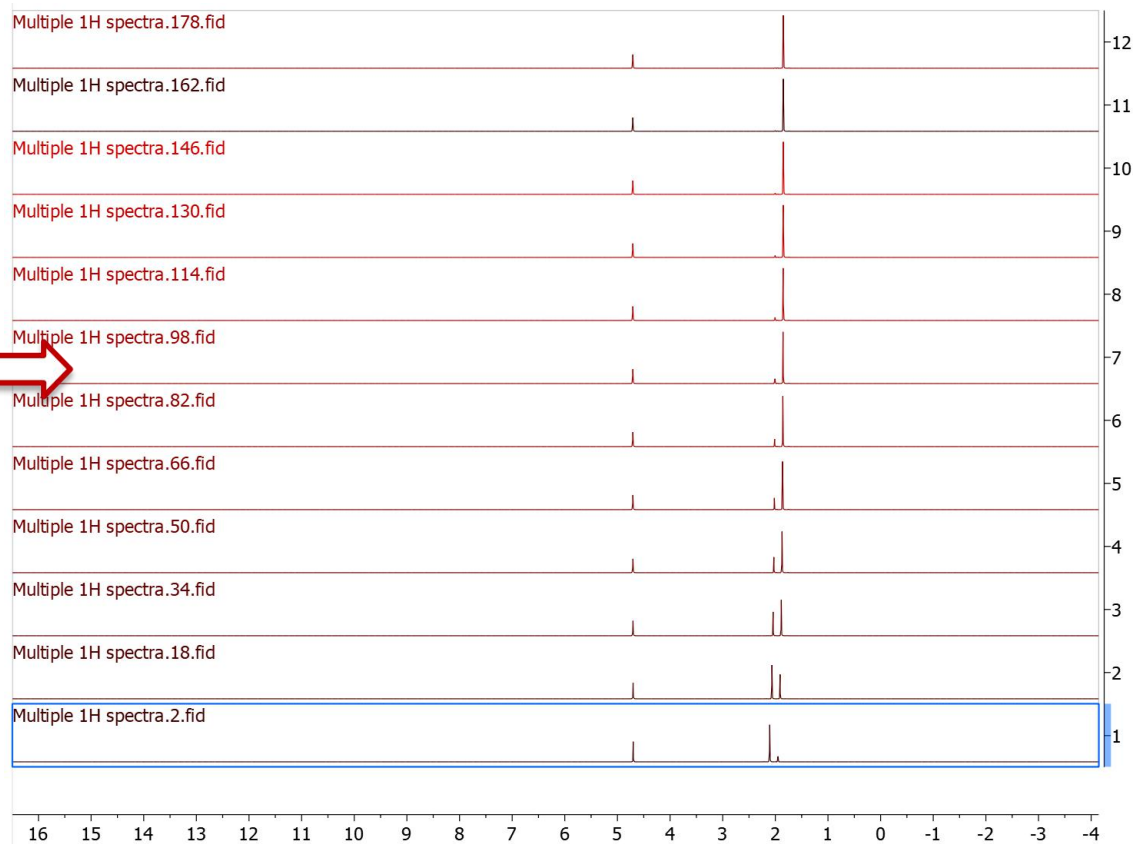
Processing Template

File: ...

Backup

Folder: C:/Users/chenp/AppData/Local/Temp ...

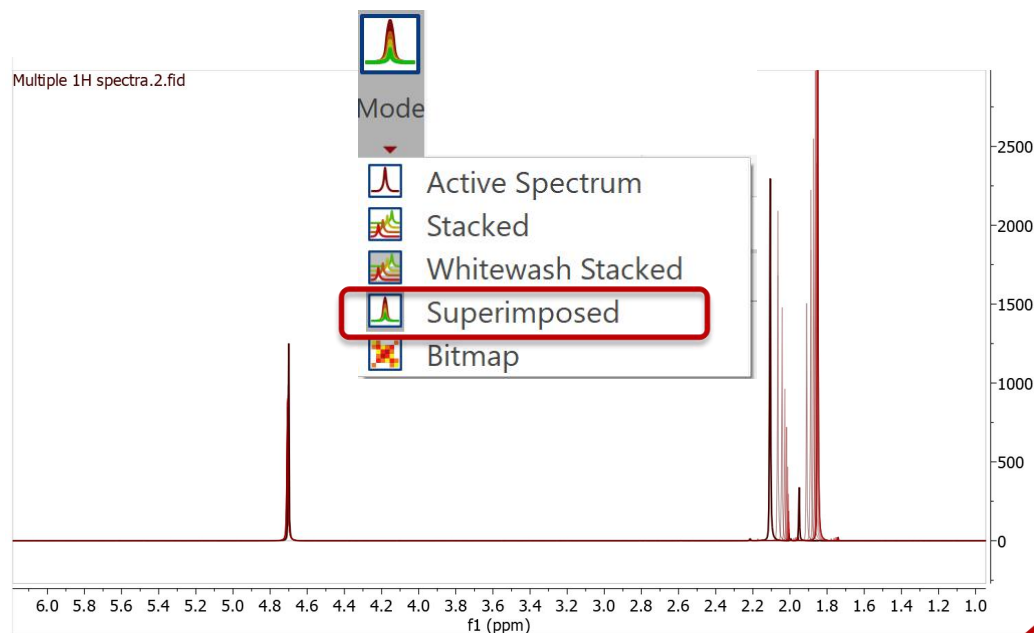
OK Cancel



Stacking mode and Stacked Items Table

ARRAYED SPECTRA

- Choose Stacked > Mode to try different display modes. Choose Superimposed mode to make sure the baseline and phasing is OK for all spectra.
- Choose Stacked > Stacked Items Table to display the Table. You can manipulate the spectra in many ways using the tools on this Table.
- If needed, you can reprocess all or selected spectra



Stacked Items Table

Stacked Items

Report Copy Delete Invert Order Setup

Multiply Divide Show Select Adjust Stacked Items

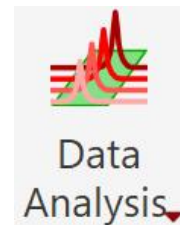
	Eye	Title	T/G	Ratio	Norm. Factor	Δ (L)
12	<input checked="" type="checkbox"/>	Multiple 1H spectra.178.fid	0.00e+00	1.00e+00	1.00e+00	0.00
11	<input checked="" type="checkbox"/>	Multiple 1H spectra.162.fid	0.00e+00	1.00e+00	1.00e+00	0.00
10	<input checked="" type="checkbox"/>	Multiple 1H spectra.146.fid	0.00e+00	1.00e+00	1.00e+00	0.00
9	<input checked="" type="checkbox"/>	Multiple 1H spectra.130.fid	0.00e+00	1.00e+00	1.00e+00	0.00
8	<input checked="" type="checkbox"/>	Multiple 1H spectra.114.fid	0.00e+00	1.00e+00	1.00e+00	0.00
7	<input checked="" type="checkbox"/>	Multiple 1H spectra.98.fid	0.00e+00	1.00e+00	1.00e+00	0.00
6	<input checked="" type="checkbox"/>	Multiple 1H spectra.82.fid	0.00e+00	1.00e+00	1.00e+00	0.00
5	<input checked="" type="checkbox"/>	Multiple 1H spectra.66.fid	0.00e+00	1.00e+00	1.00e+00	0.00
4	<input checked="" type="checkbox"/>	Multiple 1H spectra.50.fid	0.00e+00	1.00e+00	1.00e+00	0.00
3	<input checked="" type="checkbox"/>	Multiple 1H spectra.34.fid	0.00e+00	1.00e+00	1.00e+00	0.00
2	<input checked="" type="checkbox"/>	Multiple 1H spectra.18.fid	0.00e+00	1.00e+00	1.00e+00	0.00
1	<input checked="" type="checkbox"/>	Multiple 1H spectra.2.fid	0.00e+00	1.00e+00	1.00e+00	0.00

Click and drag here to change the order

Check/uncheck these to show/hide spectra

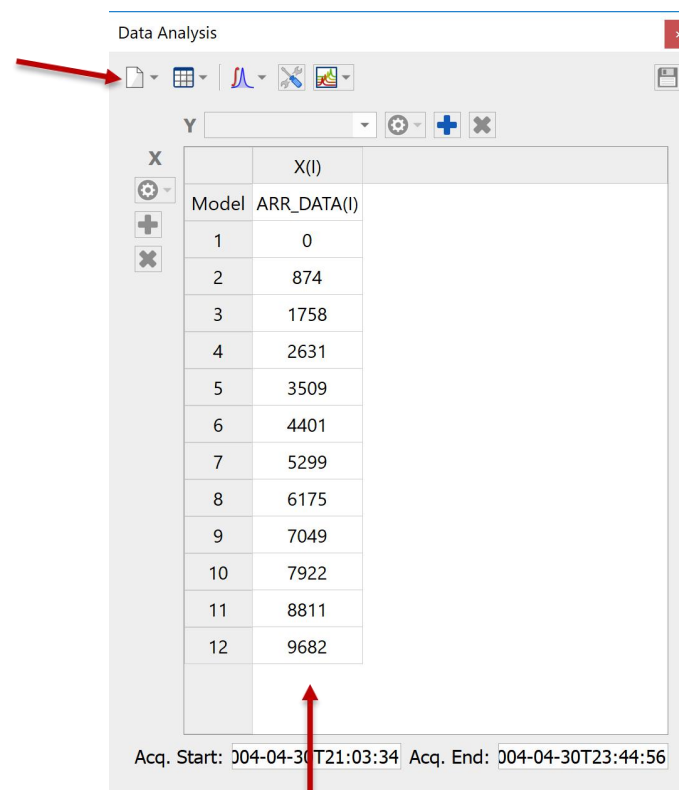
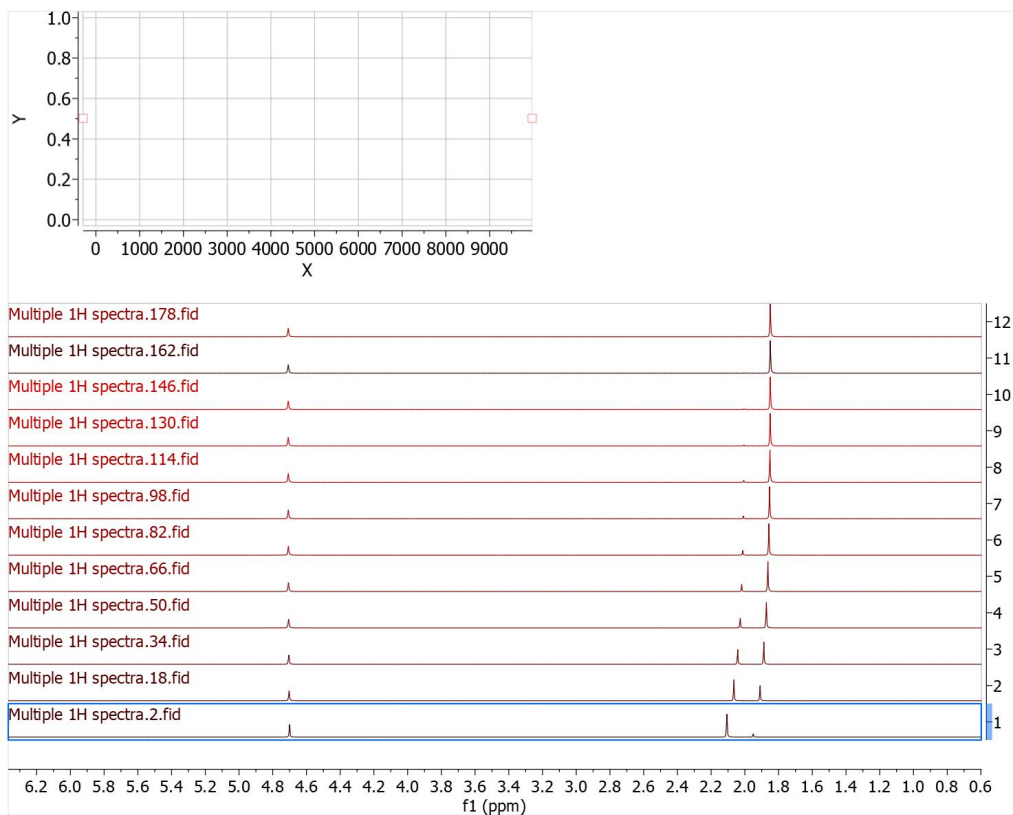
Check these to choose spectra to change

Analyze arrayed spectra



ARRAYED SPECTRA

- Choose Analysis > Data Analysis > Show Table to display the Data Analysis Table.
- Click on the Empty Graph to import the X values (reaction time in this case) and display an empty XY graph.



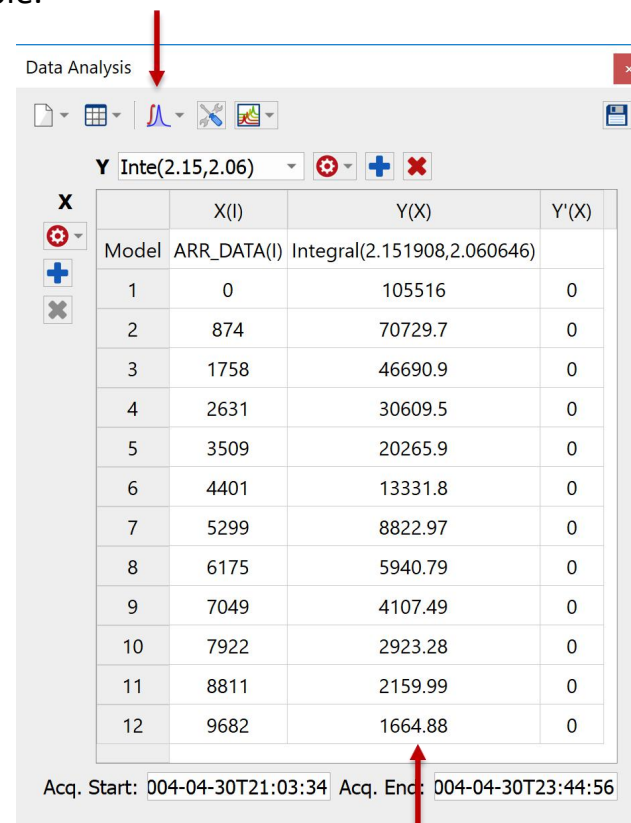
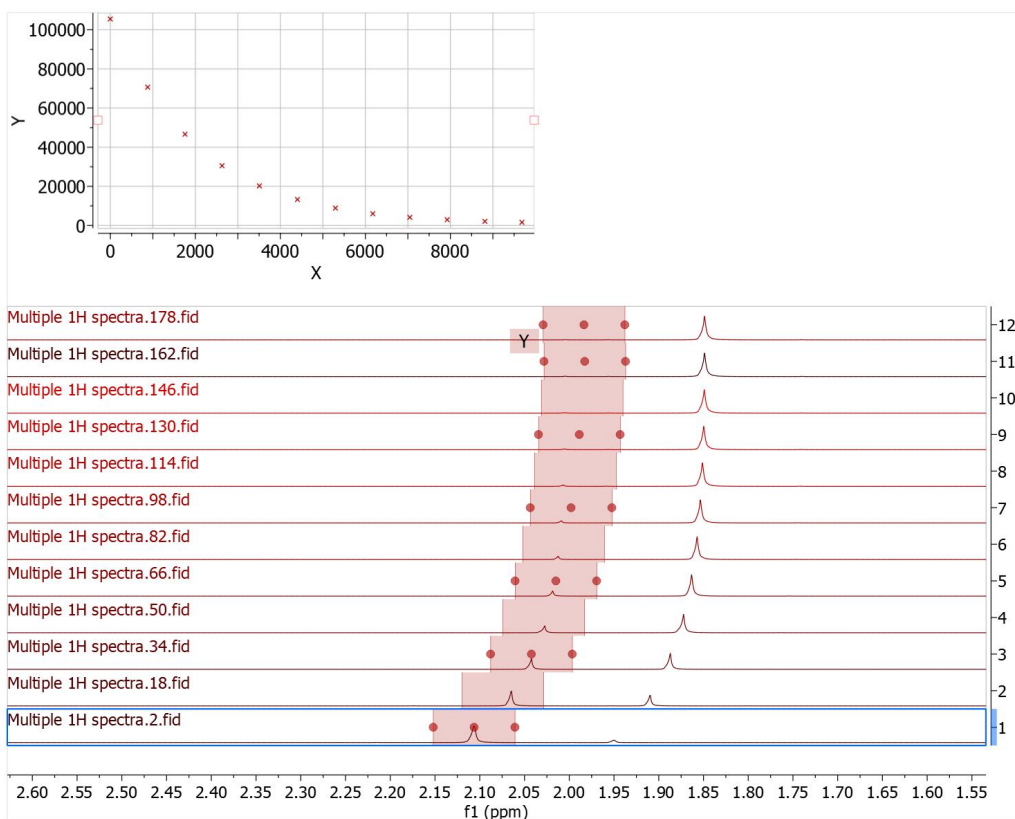
Reaction time

Integrate arrayed spectra



ARRAYED SPECTRA

- Click the Pick Integral tool.* Click and drag on first (bottom) spectrum to define the integration range
- If needed, adjust the handlers to change the integration range **
- The integrals are displayed on the XY graph and in the Data Analysis Table.



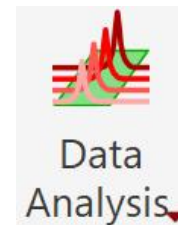
*Use Sum Method (default) for integration unless you are integrating overlapped peaks. Click Options for Integration in the Analysis Ribbon to verify. ** You can increase the # of handlers by using the Edit Model Option Tool.

Integrals

ARRAYED SPECTRA

- Click the Model cell under Y'(X) in Data Analysis.
- Choose the 3rd function, and click Calculate to fit the XY values to a first order reaction (with offset)

Fit XY to a function



Double click here

Data Analysis

Y: **Inte(2.15,2.06)**

	X(I)	Y(X)	Y'(X)
Model	ARR_DATA(I)	Integral(2.151908,2.060646)	
1	0	105516	0
2	874	70729.7	0
3	1758	46690.9	0
4	2631	30609.5	0
5	3509	20265.9	0
6	4401	13331.8	0
7	5299	8822.97	0
8	6175	5940.79	0
9	7049	4107.49	0
10	7922	2923.28	0
11	8811	2159.99	0
12	9682	1664.88	0

Acq. Start: 004-04-30T21:03:34 Acq. End: 004-04-30T23:44:56

Y'-Column Model Function

	Name	Function	Initialization	Report	Description
1	Linear Fit	A+B*x	A= 0, B= 0		Zero Order Reaction Rate
2	Mono-exponential Fit	B*exp(-x*F)			Exponential Decay, First Order Reaction Rate
3	Three Parameter Exponential Fit	B+F*exp(-x*G)			Exponential Decay, First Order Reaction Rate With Offset
4	Inverse Linear Fit	1/(A+B*x)	A= 1, B= 0		Second Order Reaction Rate
5					

Restore Defaults

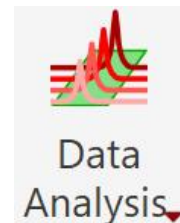
Fitted Parameters

Calculate

B= 312.388, F= 105518, G= 0.000471256
rError = 2.00941e-06, probnotmono = 0.958368

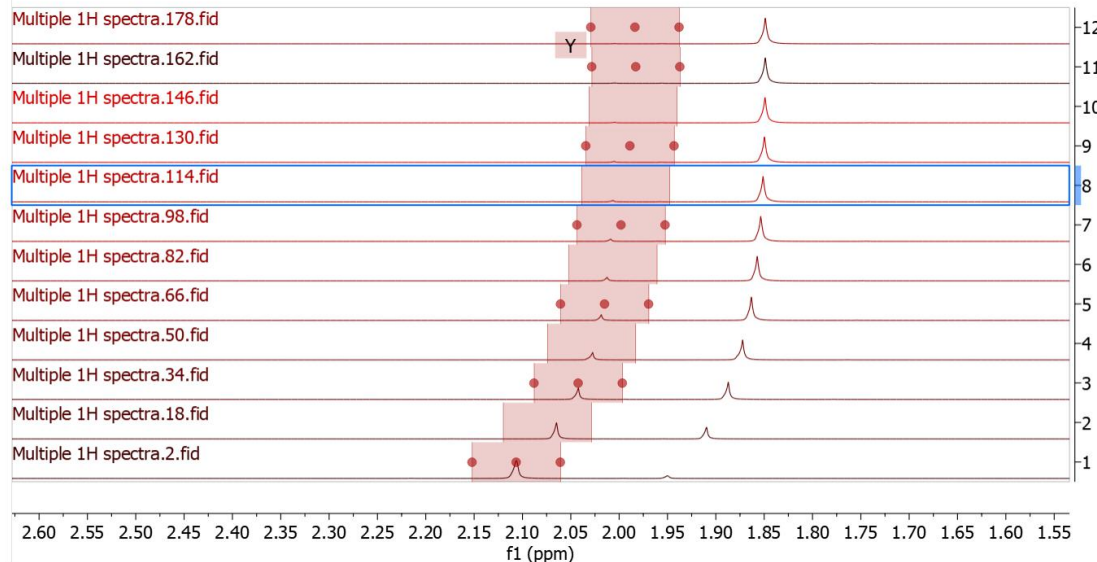
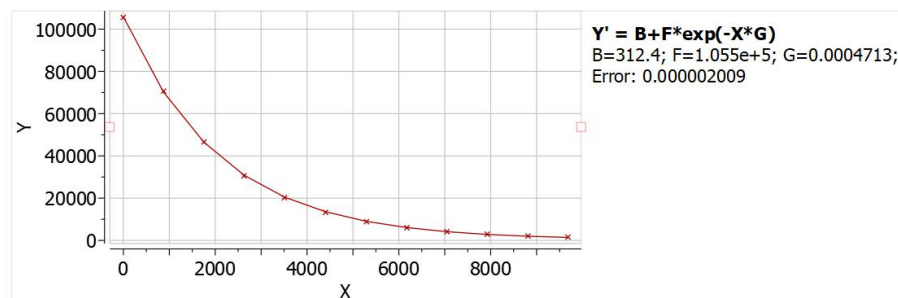
OK Cancel

Report kinetic parameters



ARRAYED SPECTRA

- Click the Report tools from the Data Analysis Panel to report the results next to the XY Graph
- Choose Report to Clipboard and paste the results to another document
- Repeat these steps for the other peaks around 1.91 ppm.



Data Analysis

Y: Inte(2.15,2.06)

X	X(I)	Y(X)	Y'(X)
Model	ARR_DATA(I)	Integral(2.151908,2.060646)	B+F*exp(-x*G) B= 312.388 F= 105518 G= 0.000471256
1	0	105516	105831
2	874	70729.7	70208.4
3	1758	46690.9	46394.2
4	2631	30609.5	30851.6
5	3509	20265.9	20503.6
6	4401	13331.8	13574.2
7	5299	8822.97	8998.29
8	6175	5940.79	6060.56
9	7049	4107.49	4120.01
10	7922	2923.28	2835.76
11	8811	2159.99	1972.11
12	9682	1664.88	1413.35

Acq. Start: 2004-04-30T21:03:34 Acq. End: 2004-04-30T23:04:56

Fitting results

HELP INFORMATION

- Use the Help Facility of Mnova: Help > Contents
- Visit www.mestrelab.com for manuals, tutorials, videos and publications
- Email support@mestrelab.com for technical questions

The screenshot displays the MestReNova software interface. On the left is a red sidebar menu with options: New, Close, Recent, Save, Save As..., Export to PDF..., Save To, Open..., Open Directory..., Open From, Print..., Page Setup..., Help (highlighted with a red arrow), Preferences..., Advanced Plug-ins..., and Exit. The main window is titled 'MestReNova' and has a 'Help' tab selected. The 'Help' menu is open, showing options: Help (Get help using Mnova), License Manager (Get licenses information like u...), Request Licenses (Buy or request evaluation lice...), and Check for Updates (Check if you are using the late...). The 'MestReNova Manual' is open, showing a table of contents with 'Using GSD for multiplets analysis' highlighted. To the right, a red banner reads 'Using GSD for multiplets analysis' with the sub-heading 'Exploiting the power of GSD for an improved Multipl...'. Below this, text explains that Mnova uses Global Spectral Deconvolution (GSD) for peak picking and multiplet analysis, and that multiplet analysis benefits from automatic analysis with enhanced peak picking capabilities. A graph shows a proton NMR spectrum with a triplet peak at 3.40 ppm, with other peaks at 3.21, 3.40, and 3.46 ppm.