CHENOMX
NMR Mixture Analysis

nmr suite 12.04



Chenomx NMR Suite 12.0

Copyright © 2001-2026 Chenomx Inc.

Table of Contents

1. Notices and Trademarks 1
    1.1. Technical Support 1
    1.2. Disclaimer of Warranties and Liabilities 1
    1.3. Trademarks 2
2. Introduction 3
    2.1. Running Chenomx NMR Suite 3
    2.2. Software Updates 3
    2.3. Tutorial and Sample Files 4
3. Handling Samples and Spectra 5
    3.1. Sample Preparation 5
        Preparing Filter Tubes for Sample Filtration 5
        Filtering the Sample through Microcentrifuge Filter Tube 5
        Internal Standard Solution 5
    3.2. Spectral Data Acquisition 6
        Pulse Sequences 6
        Required NMR Parameters 6
    3.3. Spectral Data Processing 7
        Data Processed with Other Software 7
4. Processor 9
    4.1. Overview 9
    4.2. Information Tools 10
        Spectrum Details (Sidebar) 10
        Processing History (Sidebar) 10
        Calibration Tool 11
    4.3. Processing Steps 11
        First Step: Line Broadening 11
        Second Step: Phase Correction 12
        Third Step: Baseline Correction 13
        Fourth Step: Shim Correction 14
        Fifth Step: Calibrate CSI 15
        Sixth Step: Send to Profiler 17
    4.4. Optional Steps 17
        Region Deletion 17
        Reverse Spectrum 18
        Define Custom CSI 18
        Calibrate pH 19
        Export as JCAMP-DX 20
5. Profiler 21
    5.1. Overview 21
    5.2. Profiling Tools 22
        Compound Table 22
        Cluster Navigator 23
        Enforce Transform Windows 24
        Scale Concentrations 24
        pH Tool 25
        Colors/Stars/Pins/Remember Selection 26
    5.3. Manual Profiling 27
        First Step: Identify Compounds 28
        Second Step: Determine Compound Concentrations 30
        Third Step: Profile Overlapped Regions in a Spectrum 30
    5.4. Advanced controls 31

Minimal transform windows 31
Manual locking of concentrations 31
Manual locking of peak clusters positions 32
Custom calibration of peak cluster centers ranges for automated procedures 33
5.5. Automatic Profiling using a semi-automated approach 33
First Step: Cluster positioning using Compound Snapper 34
Second Step: Concentration optimization using Fit Concentrations 35
5.6. COMPLETE Autofit: fully automated profiling approach 38
How to use COMPLETE AUTOFIT 38
Tips and Limitations of COMPLETE Autofit 45
Note about Legacy Autofit 45
5.7. Importing and Exporting Data 46
Import Profile 46
Export Profiled Compounds 47
Export Compound Table 48
Batch Export 49
Send to Processor 50
6. Library Manager 51
6.1. Overview 51
6.2. Library Tools 51
Compound Table 52
Quick Searches 52
Add Compounds 53
Export Compounds 54
Send to Compound Builder 54
Remove Compounds 55
Update Compounds 56
6.3. Compound Set Tools 56
Compound Sets (Sidebar) 57
Automatic Compound Sets 57
Compound Sets 58
Smart Compound Sets 59
Rename Compound Sets 60
Remove Compound Sets 61
6.4. Reference Card Editor 61
Required Details 61
Optional Details 62
6.5. Information Tools 62
Reference Card Panel (Sidebar) 62
7. Batching and Binning (PRO Only) 63
7.1. Batching 63
Batch Import 63
Batch Process 63
Batch Edit 65
Batch Fit 65
Batch Operations on Selected Compounds or Clusters 65
Batch Spectral Alignment 66
7.2. Spectral Binning 66
8. Compound Builder (PRO Only) 69
8.1. Overview 69
8.2. Information Tools 70
Compound Details 70

Information Panel 71
Cluster Navigator 71
8.3. Build a Compound 72
First Step: Sample Preparation and NMR Acquisition 72
Second Step: Process your Spectrum in Processor 73
Third Step: Import new CNX file into Compound Builder. 73
Fourth Step: Add Peaks, Define Clusters and Optimize Shapes 73
Fifth Step: Set Transform Windows 76
8.4. Optional Steps 77
Compound Fit Style 77
Import Simulation 78
Generate Cluster for Region 79
New Compound 80
Manage Cluster IDs 80
pH Sensitivities 81
9. Spin Simulator 83
9.1. Overview 83
9.2. Simulating Tools 84
New Simulation 84
Spin Systems 84
Spin Definitions 85
Spin Navigator 87
J-Modifiers 87
9.3. Information Tools 89
Spin Definition (Sidebar) 89
Simulation Details 90
10. The Basics 91
10.1. File Management 91
Open Files 91
Importing Spectra 91
Saving Files 92
Closing Files 92
Undo and Redo 93
10.2. Preferences 93
Change Columns 95
Display Options 95
10.3. Sidebar 96
Legend 96
Files 97
Reference Cards 98
Quick Searches 99
Spectrum Details 100
Processing History 101
Compound Sets 101
Simulation 102
10.4. Spectrum Graph 102
Spectrum Thumbnail 103
Select Region 103
Spectrum Overlays 104
Zoom Tools 106
Saving and recalling view settings 107
Export Spectrum Image 108
Copy Spectrum Image to Clipboard 108

Chapter 1. Notices and Trademarks

Developed by Chenomx Inc.

© 2001-2026 Chenomx Inc. No copying or reproduction allowed without express written permission.

1.1 Technical Support

If you have any questions or concerns, please contact Chenomx Technical Support at:

E-mail: <support@chenomx.com>

When you contact Chenomx Technical Support, you may be asked to provide an application log. The application log contains data that can help us track down issues that you may have experienced while using Chenomx NMR Suite.

Menu Location	Help > Save Application Log...
Icon	

To save an application log file

1. Open the Save Application Log dialog.
2. Locate the folder in which you would like to save the application log file.
3. In the File Name field, type a name for the application log file. If you do not specify a different name, the file is saved as `application-YYYY-MM-DD.log`.
4. Click **Save**.

Tips and Tricks

- In the unlikely event that Chenomx NMR Suite encounters an unrecoverable error and crashes, you will be asked to save an application log the next time you run the software. We strongly recommend that you do so, and send the resulting file to Chenomx Technical Support. The information contained in the log file will help us to improve future versions of Chenomx NMR Suite.

Chenomx is located at:

Legal Address:
Chenomx Inc.
1700 Enbridge Centre 10175-101 Street
Edmonton, AB T5J 0H3
Canada

Mailing Address:
Chenomx Inc
PO Box 86, Edmonton Main
Edmonton AB T5J 2G9
Canada

1.2 Disclaimer of Warranties and Liabilities

The software described in this document is furnished under a License Agreement. The software may be used or copied only in accordance with the terms of such agreement. No part of this publication may be reproduced, stored in a retrieval system, or transmitted in

any form or any means electronic or mechanical including photocopying and recording, except as permitted in the License Agreement.

The information contained in this document is subject to change without notice. Chenomx Inc. assumes no responsibility for any errors, omissions, or inaccuracies whatsoever. Chenomx Inc. shall not be held liable for any damages, including special or consequential damages, arising out of the use of the information contained in this document and the software described herein, even if Chenomx Inc. has been advised in advance of the possibility of such damages. Chenomx Inc. makes no warranties with regard to this material including, but not limited to, the implied warranties of merchantability and fitness for a particular purpose.

1.3 Trademarks

Chenomx NMR Suite is a trademark of Chenomx Inc.

Microsoft, Windows, Windows XP, Windows Vista, Windows 7, Windows 8, Windows 10, Windows 11 are either registered trademarks or trademarks of Microsoft Corporation in the United States and/or other countries.

Mac, Mac OS, and OS X are trademarks of Apple Computer, Inc., registered in the U.S. and other countries.

Linux is a registered trademark of Linus Torvalds.

Other brand and product names are trademarks or registered trademarks of their representative holders and should be noted as such.

Chapter 2. Introduction

Welcome to Chenomx NMR Suite! Before you begin analyzing spectra with Chenomx NMR Suite, you need to install and activate the software.

2.1 Running Chenomx NMR Suite

The first time you run Chenomx NMR Suite software, you will see a dialog asking you whether you wish to evaluate or activate the software. If you have not purchased a license for the software, click on **Evaluate** to continue using the software without activating it.

While evaluating the software, you can open and view files, but you will not be able to save your work in Profiler or Compound Builder. Also, certain features such as "Spectral Binning" and "Batch Fit" will be unavailable.

If you have purchased a license, follow these steps to Activate:

1. Open any module of Chenomx NMR Suite. A dialog will appear asking whether you wish to evaluate or activate the software. Click on **Activate** to begin the activation process. The Chenomx License Activation dialog will appear.
2. Send the Activation Key listed in the dialog to Chenomx at `<sales@chenomx.com>`. Your activation key is unique to your computer. If you are installing Chenomx NMR Suite on multiple computers, you'll need provide activation keys for each one.
3. When you receive a license file (`.cnxlic`) from Chenomx, save to any folder on your computer.
4. Open any module. A dialog will appear again asking whether you wish to evaluate or activate the software. Click on **Activate** to return to the Chenomx License Activation dialog.
5. Click the **Install License File...** button.
6. Browse and select your License File, and **Open**. If you would like to allow all users of your computer to use Chenomx NMR Suite, check the box marked **License extends to other users on this computer** (you may need to run the software with administrative privileges to select this option).
7. Click **Activate** and your software will be activated.

You can view your activation details at any time by clicking **Software Activation...** in the **Help** menu in any module.

2.2 Software Updates

Chenomx NMR Suite will periodically check for both MINOR and MAJOR updates and notify you if any are available.

1. MINOR updates are recommended and do not require a new software license. When you see a MINOR update notification, **Download It Now** and install. If you want to dismiss the update notification until later, click on **Remind Me Later**, or **Skip this Update**
2. MAJOR updates may provide new features and compounds. If you install a MAJOR update, you will need to obtain a new license from Chenomx before you can use it.

Subscription Users receive MAJOR updates for free during through the subscription term whereas Perpetual Users must purchase MAJOR updates.

Update checks happen automatically, and update notifications can appear at any time while the software is running. You can disable this function from the software's **Preferences** dialog by clearing the **Check for Updates Automatically** option in the **General** tab.

You can also use the **Check Now** button to force the software to check for available updates, and immediately display a notification.

2.3 Tutorial and Sample Files

The Chenomx NMR Suite simple introduction tutorial is available as a PDF document, and it comes with a small collection of sample files that you'll use as you work through the document.

Menu Location	Help > Tutorial
Icon	

To get started with the tutorial

1. Open any module in Chenomx NMR Suite.
2. In the **Help** menu, click **Tutorial...**
3. You will be prompted to select a Save Location for your Tutorial Files. Click **OK**.
4. The tutorial PDF document and the sample files will be copied into the chosen directory. Open the PDF document to begin.

Note: if you are unable to open the PDF document, you may need to install a free PDF reader such as Adobe Reader® before proceeding.

You can also view sample files directly, without going through the tutorial.

Menu Location	Help > Open Sample File
Icon	

To open a sample file

1. Open either Processor, Profiler, Compound Builder, or Spin Simulator.
2. In the **Help** menu, click **Open Sample File**

Chapter 3. Handling Samples and Spectra

Accurate analysis of your NMR spectra with Chenomx NMR Suite requires preparing good samples and acquiring high-quality spectra.

3.1 Sample Preparation

Preparing Filter Tubes for Sample Filtration

The "Amicon Ultra-0.5 Centrifugal FilterTube" contains trace amounts of Glycerol. If this material interferes with analysis, rinse the device with Milli-Q water before use. Centrifuge the filter tube for 5 minutes at 13,000 rpm or 13,793 rcf. Remove the water from the filtrate receiver (bottom half of the microcentrifuge filter tube) and discard. Repeat steps 2-4 four times to ensure removal of the glycerol preservative.

Caution: Do not allow the membrane in filter devices to dry out once wet. If you are not using the device immediately after rinsing, leave fluid on the membrane until the device is used.

Filtering the Sample through Microcentrifuge Filter Tube

Most types of samples like biofluids, tissue culture extracts and cell lysates (except urine) have molecules larger than 500kDa. In those cases we recommend filtering macromolecular components by using filter tubes before acquiring spectra from the samples. Pipette the sample into the sample reservoir (upper part) of the prewashed microcentrifuge filter tube.

- The reservoir holds a maximum of 500 µL. Because of this limitation, most samples will require two filters for efficient collection of filtered samples. Centrifuge the sample for 30-60 minutes at 13,000 rpm or 13,793 rcf.
- Some samples may need more time in the centrifuge. Centrifuge until you get the maximum amount of sample possible accumulated in the filtrate receiver. Record the time in your laboratory notebook.
- The final filtrate should ideally be clear of any coloration. In most cases, coloration is the result of a filter breakdown, in which case the filtrate should be re-filtered through a new filter.

Transfer the filtered sample into a labeled Eppendorf tube using a pipette.

Internal Standard Solution

Your samples must contain an internal standard for accurate quantification using Chenomx NMR Suite. The internal standard is used as an internal reference to calibrate the concentration and chemical shifts of the analytes.

The internal standard solution recommended for use with Chenomx NMR Suite is prepared by the following procedure:

1. To the desired volume of 99.9% D_2O, add 5 mM (DSS-d6) 3-(Trimethylsilyl)-1-propanesulfonic acid-d6 sodium salt solution as a chemical shape indicator (CSI) and 0.1% w/v sodium azide (NaN_3) to inhibit bacterial growth. D2O is used to help with the lock.

Remember that DSS is hygroscopic in the solid form. For accurate concentrations from a Chenomx NMR Suite analysis, you must determine the final DSS concentration accurately.

2. For optional pH measurement in Processor, add one or more of 100 mM imidazole, 100 mM creatinine and 20 mM DFTMP.
3. Adjust the resulting solution to about pH 6.5.

Add the internal standard as a 1 in 10 (10% v/v) spike to each of your samples. For example, for a 700 µL NMR analysis preparation, add 70 µL of Chenomx ISTD to 630 µL of acquired sample or an ISTD volume equivalent to 10% of the total sample volume to each of filtered samples. Vortex the sample for 30 seconds for uniform mixing. Transfer the sample containing Chenomx ISTD into a NMR tube. It is recommended to add 185 µL and 560 µL into 3mm and 5 mm NMR tubes, respectively.

Tips and Tricks

- To adjust the pH of the sample solutions, you can use a buffered internal standard solution.
- You can purchase the internal standard solution directly from Chenomx.
- DSS is recommended for your CSI, but 3-trimethylsilylpropionate (TSP or TMSP) or formate are also acceptable. Please note: TSP or formate, as their chemical shifts are sensitive to pH.
- For accurate sample analysis with Chenomx NMR Suite, the pH of your samples should be between 4 and 9, and ideally close to 7 for manual analysis.
- For using "Spectral Binning" feature, it is recommended to adjust the pH of all of the samples at a specific number to have an accurate analysis
- If you choose not to add any pH indicators to your samples, you still need to supply a measured pH value for proper analysis with Chenomx NMR Suite (see "Calibrate pH"). More accurate pH values for your samples allow you to make better use of pH-sensitive signatures to profile spectra of the samples.

3.2 Spectral Data Acquisition

For the best results from your analysis with Chenomx NMR Suite, you must acquire high-quality spectra. Pay particular attention to proper shimming, phase correction and baseline correction to produce spectra that you can accurately analyze and interpret using Chenomx NMR Suite.

Pulse Sequences

Using Chenomx NMR Suite, you can quantitatively analyze spectra produced by NOESY pulse sequences (noesypr1d on Bruker instruments) with t_{mix} of 100 ms. . You can use other pulse sequences for identification purposes, but absolute quantification based on alternate pulse sequences may be unreliable.

We do not recommand using gradient-based pulse sequences because they sometimes distort intense peaks at their base.

Required NMR Parameters

For the best Chenomx NMR Suite analysis results, use the following parameters for NMR data collection:

- Solvent = 90% H_2O/10% D_2O
- pH = 7
- Temperature = 25°C
- Acquisition time = 4 s
- Relaxation delay = 1 s
- Spectral width = 12 ppm
- Ghost (dummy) scans = 8

Tips and Tricks

- Chenomx NMR Suite can automatically detect DSS or TSP signals in most spectra. However, automatic detection relies on the water peak being centered in the spectrum. For best results, ensure that the water peak is centered in your spectra.

3.3 Spectral Data Processing

Chenomx NMR Suite can import spectral data from a number of common sources. These include:

- Agilent `fid` files and `phasefiles`
- Bruker `fid` and `1r` files
- JEOL `.jdf` files
- JCAMP-DX `.jdx`, `.jcm` and `.dx` files
- NMRPipe `.fid` and `.ft2` files
- Mestrelab `.mnova` files (uncompressed format only)

Before you analyze a spectrum with Chenomx NMR Suite, you must properly process your raw spectral data using either the Processor module or one of the alternate software packages described in the next section. Note that the following steps can be partially or completely automated when using the Processor module.

- Zero-fill your data, using the smallest power of 2 that is at least twice as large as the number of points acquired in the original spectrum (e.g., for 16000 points in the original spectrum, use zero-filling of 32K points).

  To do this in Processor, select **Automatic** in the import dialog under zero-filling.

- Do not apply any transformations like sine bell or Gaussian functions to your spectra.
- For the best results, you must properly phase and baseline correct all of your spectra.
- For the best results, your spectra should have CSI linewidths of no more than 1.5 Hz. Use an appropriate combination of shim correction and line broadening to adjust the linewidths.

Data Processed with Other Software

If you prefer to process your spectra using a third-party software package, you need to ensure that the Fourier-transformed (frequency domain) data from your processed spectra is available for Chenomx NMR Suite to import. The processed data from the following software packages are supported:

- VNMR and VNMRj (Agilent `phasefile` files)
- XWIN-NMR (Bruker `1r` files)
- Delta (JEOL `.jdf` files)
- NMRPipe (`.ft2` files)
- ACD/Labs software package. Export to the JCAMP-DX format (`.jdx` files).
- Mestrelab `.mnova` files (uncompressed format only)

Spectra processed by most of these software packages requires no additional preparation before they can be imported. However, if you have spectra that were processed in older versions of VNMR or VNMRJ and you would like to retain your processing when you import them into Chenomx NMR Suite, you will need to copy each spectrum's phasefile into its fid folder.

To manually copy an Agilent phasefile into the fid folder

1. Open the spectrum in VNMR and process the spectrum.
2. Change experiments or quit VNMR, making note of the experiment number in which you have opened the spectrum (e.g., `exp2`).
3. In a Solaris terminal window, copy the `datdir` folder into the `mydata.fid` folder where `mydata.fid` is your Agilent spectrum. `mydata.fid` is the folder that you need to access to open the spectrum using Chenomx NMR Suite.

   Use the following commands:

   ```
   cd ~/vnmrsys/data/mydata.fid
   cp -r ~/vnmrsys/expn/datdir .
   ```

   where *`expn`* is the experiment number noted previously, and *`mydata.fid`* is the fid folder you want to access with Chenomx NMR Suite. In this example, *`expn`* would be `exp2`.

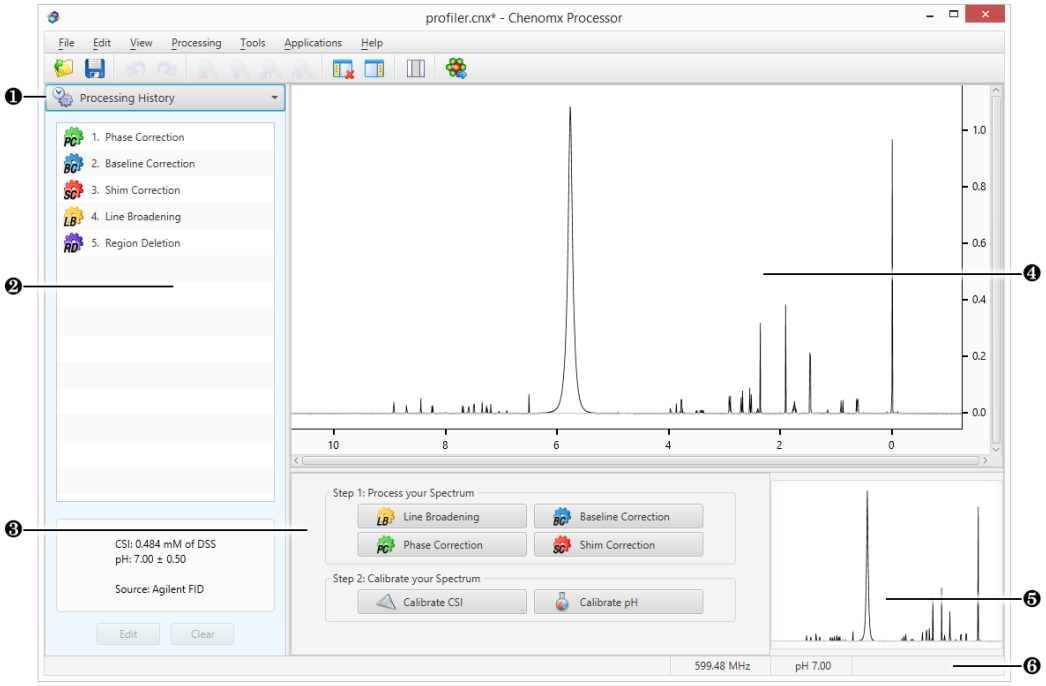
Note that it is possible to automate this process using VNMR if this is something you expect to do often. Consult your VNMR documentation for further information on automating tasks.

Chapter 4. Processor

The Processor module lets you import spectral data from many third-party vendors and software packages (including Agilent, Bruker, JEOL, JCAMP-DX, Mnova, and NMRPipe), perform standard processing and calibration operations and convert your spectra to profile ready Chenomx NMR Suite format (`.cnx`)

For a hands-on introduction to Processor, see "Tutorial and Sample Files" .

4.1 Overview



❶ Sidebar View Dropdown Menu. Lets you change the currently visible Sidebar view.

❷ Processing History. Displays a list of every processing layer that you have applied to the current spectrum, and lets you remove any processing layer that you have applied using the Processor module. Also displays information relating to the selected processing layer.

❸ Processor Dashboard. Contains quick-access buttons for the most commonly used processing features and calibration tools.

❹ Spectrum Graph. Displays the spectrum as well as guidelines used during pH determination, CSI adjustments and other processing steps.

❺ Spectrum Thumbnail. Displays a thumbnail image of the current spectrum. You can also use the thumbnail to navigate the spectrum at various zoom settings.

❻ Status Bar. Displays the current cursor position in the units specified by the **View > X Scale** and **View > Y Scale** settings. Also displays information about recent user actions.

4.2 Information Tools

Spectrum Details (Sidebar)

Menu Location	View > Sidebars > Left Sidebar > Reference Card
Icon	

Menu Location	View > Sidebars > Right Sidebar > Reference Card
Icon	

The Spectrum Details Sidebar view displays a summary of the calibration parameters for the current spectrum. It also shows the spectrum's acquisition details, including spectral width, magnet frequency, pulse sequence, and others. At the bottom of the view, there's also space for additional comments.

Tips and Tricks

- You can edit a spectrum's calibration parameters using Processor's calibration tools.
- A spectrum's acquisition details cannot be edited.
- You can edit a spectrum's comments at any time. You must save the spectrum to retain your changes.

Processing History (Sidebar)

Processing spectra in Processor does not alter your original spectral data. Every processing effect that you add to a spectrum acts as a layer modifying the display of your original data. All of these effects are completely reversible, and are tracked in the Processing History Sidebar view.

While processing a spectrum, you may want to review, edit or reverse one or more of the processing layers that you have already applied. The Processing History maintains a list of every processing layer that you have applied to the current spectrum using Processor. You can review this list to find out the parameters used in each layer, like phase angles or shim correction parameters, step back through past layers to reverse their effects, or edit a layer.

When you import a processed spectrum, Processor imports phase correction, baseline correction and other processing that you may have applied in other processing software. The imported processing effects appear in the Processing History as a layer called **External Processing**. The External Processing layer is not reversible, nor can you remove or edit it.

Menu Location	View > Sidebars > Left Sidebar > Processing History
Icon	

Menu Location	View > Sidebars > Right Sidebar > Processing History
Icon	

How do I review and remove processing layers?

1. Switch to the Processing History view in the Sidebar.
2. Click the name of a processing layer to review its associated parameters; they appear in the Details box below the list.
3. Click the **Clear** button to remove the processing layer that is currently selected.

   Removing a processing layer will also remove all subsequent processing layers.

How do I edit the most recent processing layer?

1. Switch to the Processing History view in the Sidebar.
2. Click the **Edit** button to edit the processing layer that is currently selected.
3. When you have finished your changes, click the **Accept** button.

   Editing a processing layer will cause all subsequent processing layers to be removed.

Tips and Tricks

- The Processing History only tracks processing effects, including phase correction, baseline correction, shim correction, water deletion and line broadening. Changes to spectrum metadata, such as the spectrum details or CSI and pH settings, do not appear in this panel.
- If you have just accidentally removed or edited a layer in the Processing History, you can use the **Undo** button to restore it.

Related Topics

- "Importing Spectra"
- "Processing Steps"
- "Batch Import"

Calibration Tool

It is possible to calibrate the frequency axis manually by right-clicking anywhere in the spectrum and selecting "Set new frequency at this location" in the contextual menu that appears. This will bring up an interface will to specify a resonance frequency at the clicked location.

4.3 Processing Steps

Processor applies each processing step as a layer, altering the shape of your spectrum, but leaving the original spectral data untouched. The Processing History records each layer along with its associated parameters, like phase angles, baseline breakpoints and linewidth adjustments. You can review the layers, edit them, and even remove them if necessary, restoring your spectrum to an earlier state.

When you import a processed spectrum, all processing that you have performed in other software appears in the Processing History as a layer called External Processing; the details of the External Processing layer indicate the format from which the processed data was imported. The External Processing layer is not reversible, and cannot be removed. If you need to work with the spectrum as it appeared before processing in other software, import the raw data (`fid`, `.jdf`, `.fid`) directly.

The following steps reflect best practices for using Chenomx NMR Suite. Please follow these steps in sequence for best results.

First Step: Line Broadening

Line broadening is a mathematical operation that multiplies your fid by an exponential function before the fid is Fourier transformed. Line broadening effectively increases the linewidth in your spectrum while averaging out instrument noise.

You may not need to apply line broadening to every spectrum. If you choose to use line broadening, apply it after phase correction and baseline correction.

While importing a spectrum, you may choose to have Processor automatically apply line broadening of 0 to 1000 Hz. After importing a spectrum, you can apply line broadening manually. In either case, a Line Broadening layer appears in the Processing History; you can remove the layer if necessary.

Menu Location	Processing > Line Broadening...
Icon	

How do I apply line broadening to my spectrum?

1. Switch to Line Broadening mode.
2. Enter a value between 0 and 1000 Hz.
3. Click the **Accept** button.

Tips and Tricks

- If you intend to apply shim correction to your spectrum, you can add line broadening at the same time. See "Fourth Step: Shim Correction" for details.
- To apply line broadening to more than one spectrum at a time, use the "Batch Process" tool.

Related Topics

- "Batch Import"
- "Batch Process"

Second Step: Phase Correction

Phase correction lets you correct phase shifts that may have occurred during data acquisition. Most spectra require some degree of phase correction. Phase should be adjusted before applying baseline correction or shim correction.

You can recognize phase shifts in a spectrum as an asymmetric baseline on either side of peaks and peak clusters in the spectrum. In some cases, you may see whole peaks inverted, pointing down from the baseline instead of up, while extreme phase shifts may also add a periodic oscillation or "rolling" to the baseline.

While importing a spectrum, you may choose to have Processor automatically apply phase correction. After importing a spectrum, you can apply automatic or manual phase correction. In either case, a Phase Correction layer appears in the Processing History; you can remove the layer if necessary.

Menu Location	Processing > Phase Correction...
Icon	

How do I apply phase correction to my spectrum?

1. Switch to Phase Correction mode.
2. *Automatic:* Click the **Auto** button to have Processor automatically calculate phase angles. Use the **Auto Zero-order** to only optimize the zero-order phase (the first-oder phase will be set to 0. Review the spectrum after automatic phase correction to ensure that the results are acceptable. If necessary, you can use the manual controls to adjust the phase angles before accepting them.
3. *Manual:* Click and drag the **Zero-order Phase** and **First-order Phase** sliders, or type angles in the boxes beside each slider. Use the **Fine Tune** checkbox to adjust the spectrum more gradually while moving the sliders.
4. Click the **Accept** button to apply your changes, or click the **Cancel** button to discard them.

Tips and Tricks

- Properly phase all of your spectra, as good phase correction is essential to accurately analyzing your data using Chenomx NMR Suite. Your spectrum is properly phased when its baseline is a smooth curve (or line) across the entire width of the spectrum.

- A good strategy for manual phasing is to pick a peak on either side of your spectrum, adjust the zero-order phase until the baseline on either side of the peak lines up, activate the **Use pivot Point** checkbox, move the red cursor on the location of that peak, and adjust the first-order phase. The use of a pivot point automatically adjusts the zero-phase upon modifcation of the first-order phase so that the resulting phase at the pivot point location stays constant.
- Only phase your spectrum once. If you do not like the results of phase correction, undo it or remove it using the Processing History, then try again.
- Always phase your spectrum before applying baseline correction or any other processing layers.
- Select one of the phase angle controls, and use the arrow keys to make fine adjustments to the phase angle. The arrow keys adjust in increments of 0.15° normally, and in 0.01° increments when the **Fine Tune** checkbox is selected.
- In spectra of aqueous samples, the water peak may be distorted relative to the rest of the spectrum; do not use the water peak to determine phase correction. Instead, try to obtain smooth baseline curves to either side of the water peak.
- During a phase correction session, you can clear your current adjustments by clicking the **Reset** button. After you have accepted your changes, you can clear previous adjustments through the Processing History panel.
- To apply phase correction to more than one spectrum at a time, use the "Batch Process" tool.

Third Step: Baseline Correction

Baseline correction removes distortions in the baseline of the spectrum. You can recognize distorted baselines as a net curvature or slant of the regions of your spectrum that contain no signal ("noise" regions). Common forms of baseline distortions include "smiles" (outer regions of the spectrum turned up), "frowns" (outer regions of the spectrum turned down) and "wings" (left and right edges of the spectrum turned up or down, with the middle region flat). If you notice a periodic oscillation or "rolling" of the baseline, check the phase correction of your spectrum before proceeding with baseline correction.

Most spectra require some degree of baseline correction, particularly if you need accurate quantification in Profiler. Apply baseline correction after phase correction, but before shim correction.

While importing a spectrum, you may choose to have Processor automatically apply either simple linear baseline correction (drift correction) or a cubic spline baseline correction based on automatically-determined breakpoints. After importing a spectrum, you can apply automatic or manual baseline correction. In either case, a Baseline Correction layer appears in the Processing History panel; you can remove this layer if necessary.

For more direct control over the baseline correction applied to your spectrum, you can specify breakpoints manually.

Menu Location	Processing > Baseline Correction...
Icon	

How do I apply baseline correction to my spectrum?

1. Switch to Baseline Correction mode.
2. Select the type of baseline to be performed from the Automatic BaseLine Correction pull-down menu. Four different types of baseline are available:
   - *Linear (Drift):* Simple drift correction by passing a baseline through the edges of the spectrum.

- *Chenomx Spline:* Cubic-spline-based baseline adjustment.
- *Whitaker Spline:* Whitaker filter followed by cubic interpolation of equally-spaced points.
- *BXR Spline:* BXR algorithm followed by cubic interpolation of equally-spaced points.

3. You can control the number of automatically generated breakpoints, and in the case of the Whitaker and BXR splines, the smoothness of the produced baseline by clicking on the engineer wheel icon next to the pull-down menu.
4. *Last Applied Baseline:* Loads the settings and the breakpoint locations from the last applied baseline. The baseline type and settings pulldown menus will be updated to show the parameters associated with that baseline. A new baseline calculated with those settings but for the currently loaded spectrum will be shown.
5. *From File:* Loads the baseline from the processing history of a cnx file, if applicable. A baseline operation must be present in the processing history. If multiple baseline operations are contained in the chosen file (note that technically there should never be more than one baseline correction applied), only the first instance is considered. The baseline type and settings pulldown menus will be updated to show the parameters associated with that baseline. A new baseline calculated with those settings but for the currently loaded spectrum will be shown.
6. *Manual:* Click on the spectrum to set breakpoints for a cubic spline fit. Click and drag existing breakpoints (including the endpoints) to move them. Hold down **Control** or **Command** to place breakpoints anywhere in the spectrum, instead of directly on the spectrum line itself. Hold down **Shift** while clicking on an existing breakpoint to delete it. Hold down **Shift** while delimiting a box inside the spectrum to delete multiple breakpoints at once.
7. Click the **Accept** button to apply your changes, or click the **Cancel** button to discard them.

Tips and Tricks

- Only apply baseline correction after you are satisfied with the phase correction in your spectrum.
- When setting breakpoints, be aware of the amount of curvature in the area you are trying to fit. You need to set more breakpoints to fit regions with very strong curvature, such as the areas near the water peak, than you would for a relatively flat region.
- To apply baseline correction to more than one spectrum at a time, use the "Batch Process" tool.

Fourth Step: Shim Correction

Shim correction is a method of reconstructing an ideal spectrum by removing lineshape distortions based on the shape of a reference peak. To successfully apply shim correction to a spectrum, you need to find a single, isolated peak, well-separated from the signals in the rest of the spectrum. If you have added DSS or TSP to your sample, the methyl signal from either of these is an ideal choice.

Shim correction is a linear process, using direct and inverse Fourier transforms, so quantitative relationships among compounds in the spectrum remain intact. Applying shim

correction to a spectrum assumes that all lineshapes in the spectrum are affected by systematic distortions in the same way.

Many spectra can benefit from the use of shim correction, but it is not required. It is particularly useful in reducing the effects of varying shimming techniques, or when you need accurate quantification from regions with strong overlap of signals.

You need to apply shim correction manually after you have imported your spectrum. After you have applied shim correction, a Shim Correction layer appears in the Processing History panel; you can remove this layer if necessary.

Menu Location	Processing > Shim Correction...
Icon	

How do I apply shim correction to my spectrum?

1. Switch to Shim Correction mode.
2. Zoom in to the highlighted peak in the Spectrum Graph. If it is not properly fit to the peak that you are targeting for shim correction, adjust its fit as necessary.
3. If you are using DSS or TSP as your target peak, select the **Use DSS/TSP Satellites** check box. Otherwise, clear the check box.
4. Enter the desired target linewidth.
5. Click the **Accept** button to apply your changes, or click the **Cancel** button to discard them.

Tips and Tricks

- Only apply shim correction after you are satisfied with the phase correction and baseline correction in your spectrum.
- The target linewidth text box initially contains the native linewidth of your spectrum, based on your CSI definition.
- Choosing a target linewidth larger than the native linewidth in your spectrum is equivalent to applying line broadening. Choosing a value smaller than the native linewidth may provide some resolution enhancement, at the expense of a reduced signal-to-noise ratio and possible spectral artifacts.
- To apply shim correction to more than one spectrum at a time, use the "Batch Process" tool.

Fifth Step: Calibrate CSI

Once you've finished processing your spectrum, you should calibrate it so that it can be analyzed in Profiler or used to create compound signatures and simulations in Compound Builder and Spin Simulator.

The chemical shape indicator (CSI) settings form the basis for all fits performed in Profiler and Compound Builder. The accuracy of these settings also have a direct impact on the accuracy of profiled spectra (`.cnx`), exported data (`.txt`), compound files (`.xcpd`) and spin simulations (`.xss`). Calibrate CSI mode lets you view, define and change these settings for the current spectrum.

When you import a spectrum, Processor attempts to automatically locate the CSI, and determine its width and height. You can change these automatically determined parameters using Calibrate CSI mode if necessary.

Menu Location	Tools > Calibrate CSI...
Icon	

How do I modify the CSI settings?

1. Switch to Calibrate CSI mode.
2. Type the concentration of the CSI in your sample in the **Concentration (mM)** text entry box under **Calibration Peak**.
3. Select the CSI present in your sample using the drop-down box under **Chemical Shape Indicator (CSI)**. Chenomx NMR Suite includes CSI definitions for DSS, TSP, formate, fumarate and maleate. A custom CSI can also be defined.
4. *Automatic:* Click the **Calibrate Automatically** button.
5. *Manual:*
   - Click and drag the calibration peak to match it with the spectrum line (black).
   - Adjust the width by dragging the width control (red triangles to either side of the peak) or typing a number directly into the **Width (Hz)** text entry box.
   - Use the up and down arrow keys to adjust peak height, and the left and right arrow keys to adjust peak position.

How do I handle samples that have no CSI?

1. Switch to Calibrate CSI mode.
2. Using the drop-down box under **Chemical Shape Indicator (CSI)**, select **None**.
3. Click and drag the horizontal axis to set a chemical shift reference.
4. Set an appropriate line width for the spectrum by typing a value in Hz under **Calibration Peak**.

Tips and Tricks

- Calibrate the CSI only after you have finished applying all necessary processing layers. If you change a spectrum's processing, you will need to recalibrate its CSI to ensure accurate quantification and generally good fits.
- Processor can only automatically calibrate the CSI if its signal has a chemical shift less than 0 or greater than 10. This includes all of the supported CSIs by default, and some Custom CSIs, as long as they are single peaks.
- If Processor cannot calibrate the CSI automatically when you use DSS or TSP, check the phase correction and baseline correction of your spectrum and try again. If Processor still cannot calibrate the CSI, you need to use the manual tools to locate it instead.
- If you have selected DSS as the CSI, then the indicated center position always reads 0 ppm. If you use any other CSIs, the center position varies based on the current pH value for the spectrum.
- If you would like to adjust the height or width of the calibration peak while keeping its total area constant, check the **Preserve Peak Area** box.
- When you select None as the CSI, the red calibration peak in the Spectrum Graph disappears. You can only adjust the chemical shift and width settings as described above.
- To make adjustments to the CSI in more than one spectrum at a time, use the "Batch Process" tool.

Related Topics

- "Calibrate pH"

Sixth Step: Send to Profiler

Menu Location	File > Send to Profiler
Icon	

When you finish processing a spectrum in Processor, your next task is often to start profiling the spectrum in Profiler. **Send to Profiler** closes the current spectrum in Processor, and re-opens it in Profiler.

Tips and Tricks

- Any changes that you have made to the spectrum in Processor are retained when you send the spectrum to Profiler, and are visible if you bring the spectrum back into Processor. However, you still need to save the spectrum, either in Processor or Profiler, to permanently save those changes to the spectrum file.

4.4 Optional Steps

Region Deletion

Menu Location	Processing > Region Deletion...
Icon	

In many spectra of biofluids or cell extracts, the solvent peak (typically water) may be very large. In some cases, it is so large that seeing any other peaks while completely zoomed out is difficult. Region deletion is a simple processing layer that masks data points within a specified frequency range. It is designed to prevent the height of the solvent peak from dominating the vertical range of the spectrum display.

Using region deletion to remove the solvent peak is optional. Ensure that no peaks of interest occur close to the solvent peak before you apply region deletion, as such peaks may be masked by the region deletion layer.

While importing a spectrum from an aqueous sample, you may choose to have Processor automatically apply region deletion to the water peak. A Region Deletion layer appears in the Processing History panel; you can remove this layer if necessary.

How do I apply region deletion to my spectrum?

1. Switch to Region Deletion mode.
2. Enter the desired frequency range.

   Or

   Interact with the highlighted region in the graph to define the frequency range. You can drag the region by clicking inside of it, and expand or contract it by clicking on either edge of the region.
3. Click the **Accept** button to apply your changes, or click the **Cancel** button to discard them.

Tips and Tricks

- Region deletion works best if it is applied after phase correction.
- Region deletion is not only useful for removing the solvent peak from your spectrum; it can be used to remove other peaks and clusters as well.
- To apply region deletion to more than one spectrum at a time, use the "Batch Process" tool.

Reverse Spectrum

Menu Location	Processing > Reverse Spectrum
Icon	

Reverse spectrum reverses the order of the data points in your spectrum in the frequency domain, effectively flipping the spectrum horizontally.

You only need to use reverse spectrum with certain types of spectra from Bruker spectrometers. If you have trouble locating the CSI peak (when you have a CSI in the sample), or even making any sense of your spectrum after importing it, you may need to reverse the spectrum.

While importing a spectrum, you may choose to have Processor reverse the spectrum automatically. After importing a spectrum, you can reverse the spectrum manually. In either case, a Reverse Spectrum layer appears in the Processing History panel; you can remove this layer if necessary.

Tips and Tricks

- If you notice that a spectrum from a particular spectrometer must be reversed, it is likely that all spectra from that instrument, especially those acquired with the same parameters, need to be reversed.
- If it is necessary, apply Reverse Spectrum first, as other processing layers may be difficult to apply correctly when the spectrum is backwards.
- To apply Reverse Spectrum to more than one spectrum at a time, use the "Batch Process" tool.

Define Custom CSI

If you have spectra that were acquired with a CSI other than the five supported by Chenomx NMR Suite (DSS, TSP, formate, fumarate and maleate), you can define a custom CSI for use with Chenomx NMR Suite.

How do I define the custom CSI?

Menu Location	Tools > Define Custom CSI...
Icon	

1. Open a spectrum of a sample that contains both your custom CSI and a Chenomx-supported CSI (DSS, TSP, formate, fumarate and maleate). This spectrum should already be calibrated using its Chenomx-supported CSI.
2. Open the Define Custom CSI dialog.
3. Adjust the custom CSI peak (red) to match the appropriate peak in the calibrated spectrum.
4. Enter a name for your custom CSI.
5. Click Accept.

Tips and Tricks

- You may only have one custom CSI defined at a time.
- The spectrum that you use to define the custom CSI should also contain a Chenomx-supported CSI (DSS, TSP, formate, fumarate and maleate). This reference spectrum must be calibrated using the Chenomx CSI before you use it to define the custom CSI.
- Once you have defined the custom CSI, you may not delete it. You can only edit its parameters.

Calibrate pH

The signal position and shape of compounds in aqueous solution can be very sensitive to pH. The accuracy of the pH that you set for a spectrum will have an impact on the quality and accuracy of fits that you perform on that spectrum in Profiler, Compound Builder or Spin Simulator. Calibrate pH mode lets you calculate or set the pH for the current spectrum.

If your sample contains any combination of creatinine, imidazole or DFTMP, Processor may be able to calculate the spectrum pH. If your sample does not contain these compounds, or if you prefer to use an externally measured pH, you may enter a pH value manually.

While importing a spectrum, Processor attempts to calculate the pH of the sample automatically using the frequencies of the imidazole, creatinine and DFTMP peaks. You can refine or change this calculated pH in Calibrate pH mode. If Processor cannot calculate the pH of your sample, you need to set it manually.

Whether you use automatic or manual methods to set the spectrum pH, you can also set an uncertainty for the pH value. The pH uncertainty helps to determine the size of the transform windows for pH-sensitive compound signatures in Profiler.

The predicted frequencies of the imidazole, creatinine and DFTMP peaks, based on the pH value that you currently have set, appear in the Spectrum Graph as a pair of blue (imidazole) and purple lines (histidine) at low frequencies (high ppm value)s, a pair of green lines(creatinine) at mid frequency, and a single orange line (DFTMP) at high frequency. You can use these lines during manual pH determination by aligning them with the corresponding peaks for creatinine, imidazole, histidine and DFTMP, when these compounds are present.

Menu Location	Tools > Calibrate pH...
Icon	

How do I set the spectrum pH?

1. Switch to Calibrate pH mode.
2. Set the desired pH using any one of the following methods:
   - Click the **Find Automatically** button.
   - Click and drag the pH slider bar to set the determined frequencies of imidazole, creatinine and DFTMP as close as possible to the actual frequencies of the peaks in the spectrum (if known). The resulting pH appears in the text box beside the slider bar.
   - Select the pH slider control, and use the arrow keys to make fine adjustments to the pH (with the **Fine Tune** checkbox selected).
   - Type the desired numeric pH into the text entry box to the right of the slider bar.

Tips and Tricks

- Setting an accurate spectrum pH helps Profiler choose more accurate starting positions and transform windows for clusters.
- Once you have made coarse adjustments to the pH, you can zoom in to particular indicator lines to make finer adjustments. Select the **Fine Tune** check box to let you make more detailed adjustments over a smaller pH range.
- When aligning the displayed lines to peaks in your spectrum, it is not necessary to have the lines match the peak positions precisely. A common situation is to arrive at two pH values separated by 0.01, one of which places the indicator lines on one side of the peak, while the other places them on the other side of the peak. In such cases, you may use either value.

- If you have selected a CSI other than DSS, then changing the pH changes the position of the horizontal axis, since the frequencies of the TSP and formate peaks are pH-sensitive.
- To make adjustments to the pH in more than one spectrum at a time, use the "Batch Process" tool.

Export as JCAMP-DX

Menu Location	File > Export > JCAMP-DX...
Icon	

Processor has the ability to export your spectrum into the JCAMP-DX file format, which can be imported into most NMR spectral processing software packages.

The effect of processing layers that you have applied to your spectrum will be retained in the exported JCAMP-DX file, but other analysis data such as compound fits and concentration will not.

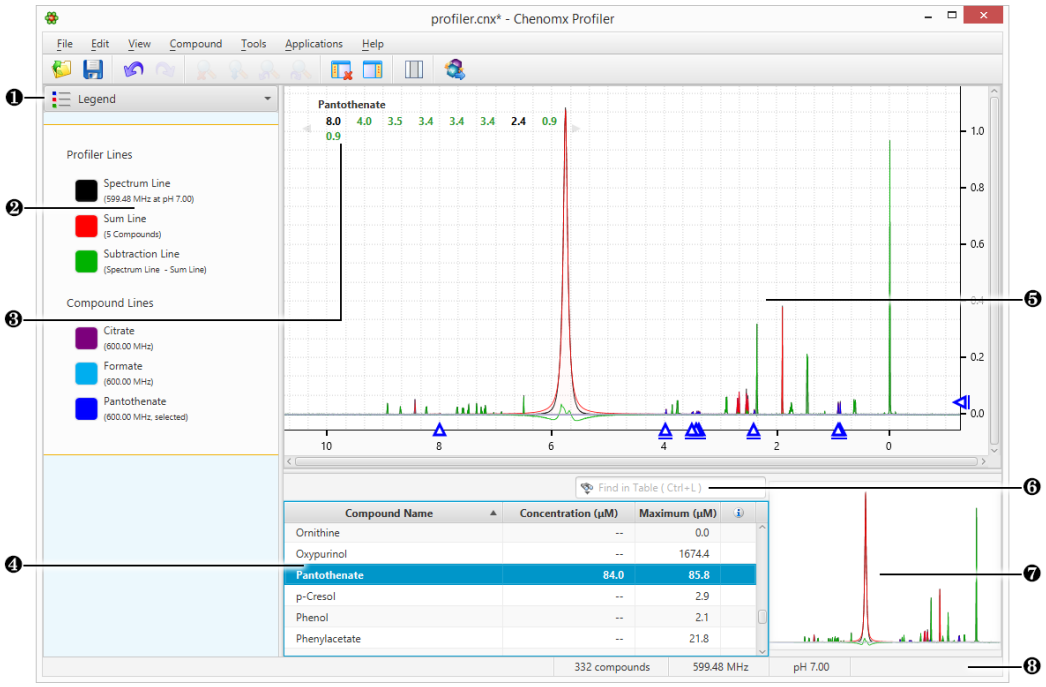
Several spectra can also be exported all at once using **Batch Export JCAMP** from the Tools menu.

Chapter 5. Profiler

The Profiler module helps you to identify and quantify compounds in a sample spectrum, as well as apply spectral analysis techniques like spectral binning (PRO Only). You can fit any available compound to the current spectrum, and narrow down the compounds you're interested in by using compound sets and filters.

For a hands-on introduction to Profiler, see "Tutorial and Sample Files".

5.1 Overview



❶ Sidebar View Dropdown Menu. Lets you change the currently visible Sidebar view.
❷ Legend. A sidebar view letting you customize the lines appearing in the Spectrum View.
❸ Cluster Navigator. Lets you navigate among the clusters of the currently selected compound. You can enable or disable the Cluster Navigator using the **View** menu.
❹ Compound Table. Displays compounds, their calculated concentrations, and their profiling status. You can access signatures of these compounds in the Spectrum Graph by selecting them. As well, you can view additional compound information in the Reference Panel by selecting a compound in the Compound Table and switching the sidebar view to the Reference Card Panel. You can change the default concentration units displayed in the Compound Table using the **View** menu.
❺ Spectrum Graph. Displays the spectrum as well as the sum line and subtraction line. You can modify the display by focusing in on a particular area or displaying and modifying compound signatures.
❻ Quick Search. Lets you filter the Compound Table to focus on a particular compound or group of compounds.
❼ Spectrum Thumbnail. Displays a thumbnail image of the current spectrum. You can also use the thumbnail to navigate the Spectrum Graph at various zoom settings.

- Status Bar. Displays the current cursor position in the units specified by the **View > X Scale** and **View > Y Scale** settings. Also displays information about recent user actions.

Compound Signatures with a *peak-based* fit style, such as those found in the Chenomx Reference Library, are fit to your spectrum by adjusting the compound height and making adjustments (sometimes called 'transforms') to the locations of their clusters. The result is a 'fit', or 'profiled' spectrum.

Peak-based compound signatures profile a spectrum by measuring compound concentrations by the height of each compound signature within the peak heights of your spectrum. When the height of a compound signature is adjusted, the corresponding heights of all peaks and clusters to that compound are modified.

The frequencies (i.e. positions) of the clusters in a compound signature may need to be adjusted during fitting due to slight mismatches in solution pH, ion concentrations, and other matrix effects. These potential errors are captured by a region around each cluster known as its 'transform window'. Profiler displays the transform window for each cluster as two triangles on the spectrum baseline when the cluster is selected. If the pH value for your spectrum has been calibrated correctly, you should be able to move each cluster to obtain a good fit without needing to move outside of the limits of the cluster's transform window.

5.2 Profiling Tools

Profiler provides you with a variety of tools to identify and quantify compounds in your spectra. The Profiler selects compounds from your Library, sets concentrations, determines peak locations, and finds and using extended information about compounds that you would like to profile.

Compound Table

The Compound Table summarizes the results of your profiling work in the active spectrum and lists the compounds that are available to profile. The Compound Table lists both Potential (Maximum) Concentration in the right hand column as well as Measured/Profiled Concentration in the left hand column. You must select at least one compound set in the Compound Sets view to see any compounds in the Compound Table.

The currently selected compound in the Compound Table is the active compound in the Spectrum Graph, where you can adjust its shape, position, and concentration. Select a compound without a concentration in the Compound Table, a preview line for the compound appears in the Spectrum Graph, scaled vertically to suit your current zoom level. You can click on the preview line to begin profiling that compound; its concentration will be set to match the height of the preview line.

Filter the current contents of the Compound Table using Quick Searches for compound names, comments, compound IDs, regions of the spectrum, or specific locations in the spectrum.

Select more than one compound from the Compound Table by holding down **Ctrl** (**Cmd** in Mac OS X) and clicking additional compounds. If you would like to select a range of compounds, click on the first compound and then hold down **Ctrl-Shift** (**Cmd-Shift** in Mac OS X) and click on the last compound. Apply colors, pins, or stars to multiple compounds simultaneously.

Available units of concentration include millimoles per litre (mM), micromoles per litre (µM) and milligrams per decilitre (mg/dL).The maximum allowed concentration for any compound is 5000 mM (5 M). When you select mg/dL as the concentration unit, the maximum

value varies per compound, since the units incorporate the molecular weight of the compound. However, the maximum value is always equivalent to 5000 mM.Concentrations in mg/dL are only available for compounds whose molecular weight has been specified in the compound's Reference Card , and the accuracy of such concentrations depends on the accuracy of the molecular weight provided. The molecular weight has already been correctly specified for all compounds in the default Chenomx Reference Library.

Tips and Tricks

- After typing in the Quick Search box, press the down arrow key to navigate to the Compound Table.
- Set custom display colors for compounds, pin compounds so that they are visible even when they are not selected and toggle stars on compounds to generally mark them for future reference.
- Hover the mouse cursor over some parts of the Compound Table, tooltips appear containing more details. For example, hovering over the optional matched clusters column for a compound shows you which clusters are matched.
- Set compound concentrations directly in the Compound Table, by double-clicking on a compound's concentration cell and typing in a value.
- Right-click on a compound's row in the table. Operations accessible through the context menu include scaling concentrations, automatically fitting the compound to the spectrum, and clearing fits, custom colors, stars or pins.

  There are many columns available for display in the Compound Table. Display any number of the available columns, (at least one visible) and drag column headers in any order. Sort the Compound Table on any column by clicking its column header. Click the header again to reverse the sort order.
- Copy the contents of a cell to the Clipboard by right-clicking on the cell and select **Copy [cell name] to Clipboard**. Paste the contents as text to any other program.
- Compounds modified from their default state (including fits, custom colors, pins and stars) will appear in the Profiled Compounds set, and appear in bold in the Compound Table.
- Press **Delete** with one or more compounds selected in the Compound Table to clear all fits, custom colors, pins and stars for the selected compounds.
- To make more space for the Compound Table, you can hide the Spectrum Thumbnail by clicking **View > Show Spectrum Thumbnail**.

Related Topics

- "Quick Searches"

Cluster Navigator

The Cluster Navigator enables fast navigation among clusters of an active compound. The Navigator appears in the top left corner of the Spectrum Graph as an ordered list of the approximate positions of all the clusters, in ppm. Click on a cluster position and navigate to an optimal view of the cluster.

Tips and Tricks

Menu Location	View > Show Cluster Navigator

- Use the Q and E keys to display the next or previous cluster.

- Clusters that appear green in the Cluster Navigator are 'matched', meaning they closely match the spectrum line as determined by comparing the sum line (red) and the spectrum line (black) near each cluster.
- Click on a cluster and drag along the Cluster Navigator to display multiple clusters in the Spectrum Graph.

Related Topics

- "Compound Table"

Enforce Transform Windows

Transform windows appear alongside compound signatures to guide profiling by restricting the frequency range that you can adjust each cluster. All Chenomx Reference Compounds have had transform windows set to allow the flexibility to fit most common biofluids. You can choose to have the transform windows enforced, preventing you from moving any cluster outside its transform window, or not enforced, allowing you to move any cluster beyond its transform window.

Edit transform windows

Menu Location	Edit > Enforce Transform Windows

1. In the Edit menu, check **Enforce Transform Windows** to lock.
2. On the Edit menu, clear **Enforce Transform Windows** to allow clusters beyond transform windows.

Tips and Tricks

- View the size of the transform that has been applied to any cluster by hovering the mouse cursor over it. The transform size appears in the top right corner of the Spectrum Graph, and visually as a thick blue line connecting the starting position of the selected cluster and its current position.
- When you move a cluster outside its transform window, its transform adjustment triangle turns red. The **Transform** column in the Compound Table will also indicate whether the compound has any clusters that have been moved beyond their transform windows
- Clusters that are outside their transform window appear in the Transforms column of the Compound Table as **Outside Windows**. Note that you may need to turn on the Transforms column in the Compound Table.
- When you move a cluster outside its transform window, the adjustment triangle on the horizontal axis of the graph turns red. Also, when you hover over the cluster of its triangle, the visual indicator for the adjustment is red, as is the text in the top right corner of the Spectrum Graph describing the extent of the adjustment.
- Transform windows for pH sensitive compound signatures (including those found in the Chenomx Reference Library) are determined in part based on the pH uncertainty for the current spectrum. If you find that you frequently need to override the transform windows for a spectrum, user Processor to check the accuracy of its pH calibration, and consider increasing its pH uncertainty value.

Scale Concentrations

Scale Concentrations lets you modify the concentrations of profiled compounds in the current spectrum by a common multiplication factor. You can use this to compensate for

dilution effects, errors in CSI concentration or other systematic influences on compound concentrations.

Menu Location	Compound > Scale Concentration...
Icon	

How to Scale Concentrations of one or more compounds by a common factor

1. Select one or more compounds in the Compound Table, and in the Compound menu, click **Scale Concentrations...**, or go to Edit menu, click **Select All**, and then in the Compound menu, click **Scale Concentrations...**.
2. Enter the multiplication factor. To increase concentrations, use a factor greater than one. To decrease them, use a factor less than one.
3. Click OK.

Tips and Tricks

- You cannot enter a multiplication factor that would result in a concentration higher than 5000 mM for any compound.
- If you set the multiplication factor to zero, all of the profiled compounds are set to 0 mM.
- Scale Concentrations does not affect compounds with unknown concentrations (--).
- If you want to scale concentrations for a specific compound or for all of the profiled compounds in a group of `.cnx` files, use the Batch Edit feature.

pH Tool

Profiler can propose the pH of the sample by analysing the position of the fit peaks clusters that are pH sensitive and report an optimized pH value that would best explain the location of each.

The pH analysis is carried on based on 3 different selection modes:

1. the cluster(s) that are specifically selected for a given compound
2. the set of compounds selected in the compound table
3. if no clusters or compounds are selected, then it analyzes the entire list of profiled compounds.

Compounds with no concentration and clusters with 0 net intensity or without pH-defined chemical shifts are ignored.

The results of the analysis of each clusters/compounds are presented in a scrollable table, with compounds in the first column, cluster ID in the second, best pH value in the third, the valid compound pH range in the fourth, and the pH sensitivity of the cluster in the last column. The sensitivity is reported as the change in ppm around ± 0.1 unit of pH around the global optimum pH that is proposed.

Only the clusters with a sensitivity of ≥ 0.001 ppm are used in the calculations and reported in the list.

The reported optimized pH is the value that minimizes the weighted amount of transforms that would need to be applied to the listed clusters to explain their current position. The listed sensitivity values are used as weights.

It is possible to change the pH of the spectrum directly in the interface. Upon pH change, the clusters transforms are automatically recalculated to preserve the initial positions of the clusters.

Colors/Stars/Pins/Remember Selection

When working with multiple compounds, it can become difficult to distinguish which compounds are contributing which parts to an overlapped region. Set custom colors, 'pin' a compound to ensure it remains visible, or 'star' a compound to label it for later analysis. 'Remember' compounds applies your compound selection from one spectrum across the remaining spectra in your project, which can be useful when using a feature such as 'Fit Automatically'.

Menu Location	Compound > Assign Color...
Icon	

How do I set a custom color for one or more compounds?

1. Select one or more compounds in the Compound Table.
2. In the Compound menu, click **Assign Color...** and select your desired color. Alternatively, if your Compound Table has a Color Column open, click in the Custom Color column () to access the color chooser.
3. Click one of the preconfigured colors in the color chooser, or click **More...** to choose from a larger selection of colors.

Menu Location	Compound > Pin to Graph
Icon	

How do I pin one or more compounds?

- Click the pin for the compound in the Pinned column () of the Compound Table.

  Or

  Click the pin next to the compound's line in the Legend ().

  Or

  Select one or more compounds in the Compound Table and, in the Compound menu, click **Pin to Graph**.

Menu Location	Compound > Add Star
Icon	

How do I star one or more compounds?

1. Click the grey star for the compound in the Starred column () of the Compound Table.

   Or

   Select one or more compounds and, in the Compound menu, click **Add Star**.

Menu Location	Tools > Remember Selection
Icon	

How to 'Remember' compounds from my current spectrum to work with in other spectra

1. Select one or more compounds.
2. In the Edit menu, click **Remember Selection**.

Menu Location	Tools > Restore Selection
Icon	

How do I re-apply selected compounds to a new spectrum?

1. Open the spectrum that you want to select your compounds in.
2. In the Edit menu, click **Restore Selection**.

Tips and Tricks

- You can use this feature to quickly select a number of known compounds in the Compound Table to be fit automatically.

Menu Location	Compound > Clear > Color
Icon	

How do I clear a color/pin/star from one or more compounds?

1. Select one or more compounds in the Compound Table.
2. In the Compound menu, click **Clear > Color/Pin/Star**

Menu Location	Compound > Clear > Pin
Icon	

Tips and Tricks

- Set or clear custom colors for compounds visible in the Legend by clicking on the colored box beside the compound in the Legend.
- Setting a custom color/pin/star for a compound adds it to the Profiled Compounds set, even if you have not fit the compound.
- Custom colors/pins/stars are stored with the spectrum when you save it as a `.cnx` file. The colors you select are still associated with the compounds when you next open the file, until you clear them. If you want to set the color of a specific compound in a group of `.cnx` files, use the Batch Edit feature.
- You can export custom colors/pins/stars to a file using Batch Export.

Menu Location	Compound > Clear > Star
Icon	

Related Topics

- "Legend"

5.3 Manual Profiling

Profiling a spectrum (i.e "targeted profiling") can be a subtle and potentially complex task depending on timeframes, research goals, or other requirements. Maintaining a consistent profiling strategy will provide more consistent and more easily-interpreted results.

For more details on specific profiling techniques and how to apply them to your spectra, please see "Tutorial and Sample Files" .

Basic Compound Fitting

1. Select a peak-based compound in the Compound Table and select a cluster from the Cluster Navigator to begin. Chenomx recommends selecting an isolated peak for initial profiling.
2. Click and drag the cluster to adjust the fit. You can also click and drag the blue triangle on the vertical axis of the graph to change only the compound concentration, or drag a blue triangle on the horizontal axis to change only a cluster's frequency.
3. The concentration of the compound will appear in the 'Concentration' column in the Compound Table.

Region-based Compound Fitting

1. Select a region-based compound in the Compound Table. Note that there are no region-based compounds in the default Chenomx Reference Library--you will need to create your own and add it to your Compound Library.
2. Select a cluster in the Cluster Navigator, click and drag the cluster to adjust the fit. Notice how changing the height of the compound has no effect on which part of the spectrum is fit.
3. Concentrations cannot be calculated for region-based compounds. Instead, the otherwise unfit spectrum areas near each cluster will be summed up and displayed in the Area column of the Compound Table. This value is proportional to the true concentration of the compound.

Targeted Profiling

1. Open the spectrum in Profiler
2. Select either the default, or a custom compound set that corresponds to your sample.
3. Identify and profile isolated and prominent signals in the spectrum first(see "First Step: Identify Compounds"). Easily identifiable compounds include CSI compounds like DSS or TSP and pH indicators like imidazole, creatinine or DFTMP.
4. Systematically review the remaining signals in the spectrum; identify and profile as needed.

   Use information and external links from the Reference Panel to help establish the presence of particular compounds in the samples that you are analyzing.

First Step: Identify Compounds

Match the compound signatures from the Chenomx Library with patterns in your sample spectrum. There are several ways that you can do this.

Identify specific compounds

1. Select at least one compound set in the Compound Sets view (see "Compound Sets (Sidebar)").
2. Select a compound in the Compound Table that you believe is present in the sample(see "Compound Table").
3. Review each cluster in the Cluster Navigator and compare the shape of the preview line (light blue) to the spectrum line (black) and decide whether it is reasonable that the preview line could be contributing to the spectrum line in the displayed region.
4. If the shape appears reasonable, click the preview line, and adjust the resulting shape by determining appropriate heights and cluster locations for the compound. If the shape does not appear reasonable...
5. Select another compound in the Compound Table.

Find possible compounds contributing to a specific peak

1. Select a compound set.
2. Right-click on the peak in the experiment spectra and select **Search for Compounds Near x.xx ppm**.
3. Select a listed compounds in the Compound Table to display the preview line and review each cluster in the Cluster Navigator.
4. Compare the shape of the preview line (light blue) and the spectrum line (black) and decide whether or not it is reasonable that the preview line could be contributing to the spectrum line in the displayed region.
5. If the shape appears reasonable, click the preview line, and adjust the resulting shape by determining appropriate heights and cluster locations for the compound. If all clusters appear reasonably well-matched, apply an automatic fit, then adjust the fit for the compound as needed. If the shape does not appear reasonable...
6. Select a different suggested compound from the Compound Table.
7. Finish with the filtered list and clear the frequency filter to continue your analysis with the full compound list (see "Quick Searches").

8. If no compounds in the list of potential candidates provides a reasonable shape near the peak that you are investigating, consider the possibility that the compound responsible for the peaks is either not in the selected compound library, or not in your currently selected compound set.

To identify all compounds contributing to a particular region

1. Select a compound set in the Compound Set Panel.
2. Select a region in the Spectrum Graph (see "Select Region").
3. Search the Compound Table for compounds in the selected region (see "Quick Searches") and select a listed compound.
4. Compare the shape of the preview line (light blue) and the spectrum line (black) and decide whether or not it is reasonable that the preview line could be contributing to the spectrum line in the displayed region.
5. Compare the shape of the preview line (light blue) and the spectrum line (black) and decide whether or not it is reasonable that the preview line could be contributing to the spectrum line in the displayed region. If all clusters appear reasonably well-matched, apply an automatic fit, then adjust the fit for the compound as needed. If the shape does not appear reasonable...
6. Evaluate the remainder of the compounds in the Compound Table.
7. Finish with the filtered list and clear the frequency filter to continue your analysis with the full compound list (see "Quick Searches").

Tips and Tricks

- Profiler can suggest a fit for a selected compound by clicking **Compound > Fit Automatically**, or by pressing the space bar.
- It is also possible to create your own compounds that use a *region-based* fit style, which automatically fit all of the spectrum area in whatever region they are located in. For more information about fit styles, see "Compound Fit Style" .
- Double-click on a peak-based compound's concentration in the Compound Table and type a value directly to set the concentration. Type a number from 0 to 5000 mM (or equivalent concentrations in µM or mg/dL), or type "**--**" to set the concentration to unknown.
- When you have selected a compound in the Compound Table, you can click its shape in the Spectrum Graph, then use the arrow keys to adjust the compound's concentration or the cluster's position. Hold down **Shift** to allow finer adjustments.
- To clear all of the fits for the current spectrum, click **Edit > Select All** and then click **Compound > Clear > Fit.**
- To clear fits only for selected compounds, select the compounds and click **Compound > Clear > Fit.** Multiple compounds can be selected by holding down **Shift** or **Ctrl** (**Cmd** in Mac OS X).
- All compound concentrations and areas start out as unknown (--).
- If a compound's concentration is unknown (--) and you drag it up and then back down to the baseline, the concentration is set to 0.00 mM. To return the concentration to unknown, you must clear it.
- You can export a file describing the transform status of all compounds by clicking **Tools > Batch Export**.

Second Step: Determine Compound Concentrations

If you have automatically fit a compound, you may need to refine the suggested concentration. Note that it is not possible to determine (or adjust) the concentrations of compounds that make use of the region-based **fit style**. For those compounds, you must perform your analysis using compound Areas instead. See "Compound Fit Style" for more details. For more details on techniques for determining optimal profiles for compounds in experimental spectra, see "Tutorial and Sample Files" .

Determine the concentration of a compound

1. Ensure the subtraction line (green) is visible (see "Legend").
2. Set a concentration for the compound so that you can see the clusters clearly and use the Cluster Navigator to locate a cluster that overlaps clusters as little as possible and approximate the compound concentration.
3. Click and drag the height control to adjust the height of the compound until the subtraction line under the cluster approximates a normal spectrum. You may need to move the cluster from side to side somewhat to properly adjust the subtraction line.
4. If any of the clusters for the current compound overlap with compounds that you have already profiled, you may need to re-adjust those compounds as well.
5. Compound concentrations are shown in the Compound Table (see "Compound Table").

Third Step: Profile Overlapped Regions in a Spectrum

For all but the simplest spectra, you will need to deal with compounds whose signatures overlap. A consistent strategy for profiling overlapped regions will produce more consistent results.

For more details on this and other profiling strategies, see "Tutorial and Sample Files" .

To profile compounds in an overlapped region of the spectrum

1. Identify likely compounds contributing to an overlapped region (see "First Step: Identify Compounds").
2. Evaluate each compound for the presence of additional, non-overlapped peaks elsewhere in the spectrum (see "Cluster Navigator").
    a. If they are present:
        i. Look for clearly visible peaks in the overlapped region. Such clusters occur mostly at the edges of the overlap region, but remember to look throughout the region, as some peaks stand out nicely between the peaks of another cluster.
        ii. Use these partially-exposed peaks to establish the optimal frequency for the cluster.
    b. If they are not present:
        i. Look for clusters of peaks in the overlapped region with some clearly visible peaks.
        ii. Use these partially-exposed peaks to establish the optimal frequency for the cluster.

3. Once you have approximately profiled most of the groups in the overlap region, fine-tune the heights and frequencies of each cluster in the region for an optimal shape. Use the subtraction line to guide your modifications, as a well-matched cluster will leave a subtraction line that resembles a normal spectrum with the effects of the cluster removed.
4. In overlap regions, profile multi-peak clusters before single-peak clusters. If urea is present, you may want to profile it last, to minimize its interference with other compounds in the spectrum.

5.4 Advanced controls

New functionalities were introduced in version 12.0, providing the user with much finer control and customization of key features, specifically with the capability of locking clusters and concentration values in place, as well as the possiblity of providing custom calibrations for cluster center limits.

Details are provided in the sections below.

Minimal transform windows

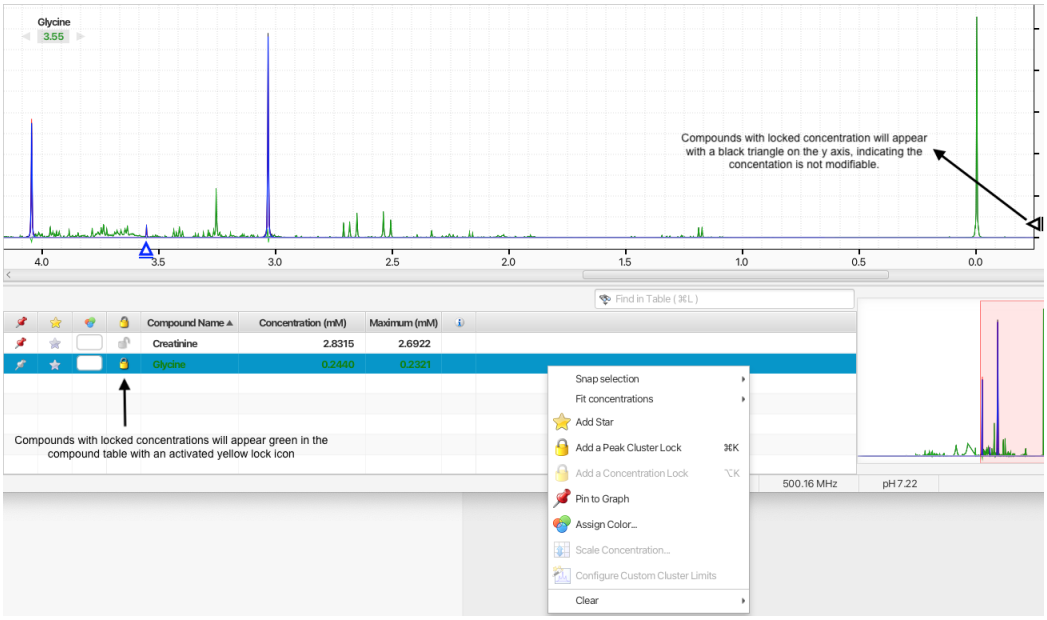
By default, the minimal transform windows on each side of peak cluster is set to 0.02 ppm, even if the pH equations predict a smaller value. It was put in place to account for possible matrix effects with some biological samples, among other things.

You can change this value in the software Preferences under the General tab. Modifying this value affects all clusters and has consequences on the capacity of the software to automatically position clusters. Contrary to manual peak positioning where going outside the calibrated transform window is allowed, calibrated transform windows are strictly respected during automated procedures, unless instructed to do so with a external custom calibration file (see below).

Manual locking of concentrations

Compound concentrations can be locked so that their values cannot be changed by automated procedures or accidently by manual manipulations. Compounds will locked concentrations will appear green in Profiler's compound table.

You can activate the display of the lock statusus of compounds by right-clicking in one of the table column and selecting "Concentration locks". A column with a lock icon will appear. Clicking the lock icon will toggle the lock status on/off.



Alternatively, you can choose to add or clear concentration locks on one or more compounds at once by first selecting the compounds in the compoudn table and then right-clicking on one of the compound's associated rows in the and choosing the appropirate otion. You can also directly add or clear concentration locks using keyboard shutcuts.

You can add or clear concentration locks to a whole series of spectra automatically via **Tools Menu > Batch Operations > Batch Edit**. (Pro version only)

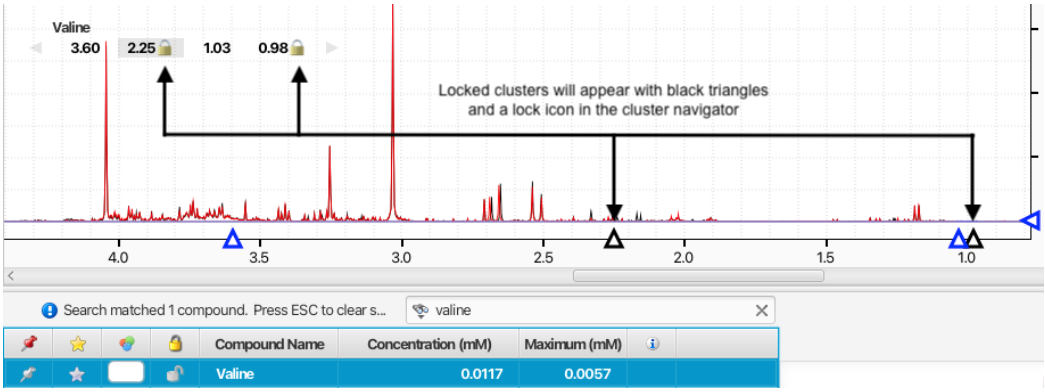
Compound with locked concentrations will appear with a black triangle on the y axis. The triangle will not be movable until the concentration is unlocked.

Manual locking of peak clusters positions

The positions of peak clusters can be locked in place so that their positions will not be modified by automated procedures or accidently by manual manipulations. Locked clusters will appear with a lock icon in the compound Cluster Navigator when the compound is selected.

You can toogle on and off the lock status of a cluster by holding on the command (Macintosh) or alt key (Windows and Linux) while double-clicking on its displayed location in ppm in the Cluster Navigator. Alternatively, you can select a compound and one or more of its clusters from the triangles displayed on the ppm axis, and right-clicking on one of them, or directly via the associated keyboard shutcuts.

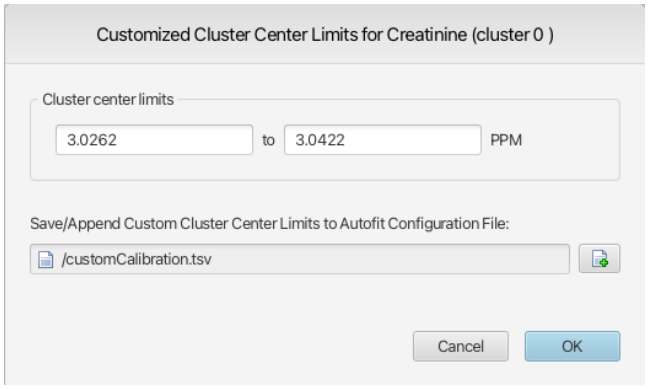
Locked clusters will appear with a black triangle on the ppm axis and they will not be movable until they are unlocked.



Custom calibration of peak cluster centers ranges for automated procedures

For the batch snapping, Legacy Autofit and the COMPLETE Autofit procedures, you can optionally overwrite the automatically calibrated peak cluster transform windows with your own set of custom values. They must be provided to the autopmatic routines via a tab-seperated value (tsv) file.

To define custom cluster limits values for a given cluster, activate the cluster from its associated triangle on the x axis, right-click and select **Configure Custom Cluster Limits**.



Provide custom values for the lower and upper cluster center limits, as well as a new or existing file to which the cluster information will be saved. By default, the values presented in the interface are the calibrated limits of the cluster center for the selected cluster in the currently opened spectrum if no spectral region has been selected prior to calling the function. On the other hand, if a region was selected, the limits of the selected spectral region will be shown. The default selected file will always be the last one used, allowing for fast additions to a growing file.

5.5 Automatic Profiling using a semi-automated approach

Manual Profiling can involve a lot of effort in setting compound cluster positions and compound concentrations. Alternatively, you can use a semi-automated approach, or, with the introduction of version 10.0, a fully automated approach.

With the semi-automated approach, the first step consists of positioning compounds clusters on the spectra, followed by an automated concentration optimization based on those positions. Several rounds may be required.

First have Profiler fit cluster postions using the "First Step: Cluster positioning using Compound Snapper" tool and/or compound concentrations using the "Second Step: Concentration optimization using Fit Concentrations" for either the currently selected compound, a selection of compounds, or all compounds in the compound table.

Tips and Tricks

- Use the Remember Selections feature to the mark compounds that can be reliably Autofit in your project. You can then restore the selection when working on subsequent spectra, allowing you to quickly autofit those compounds.

First Step: Cluster positioning using Compound Snapper

This feature allows for automatic positioning of peak clusters on the spectrum. It will essentially position peak clusters where their shape best match the experimental spectrum, **independent of intensity**.

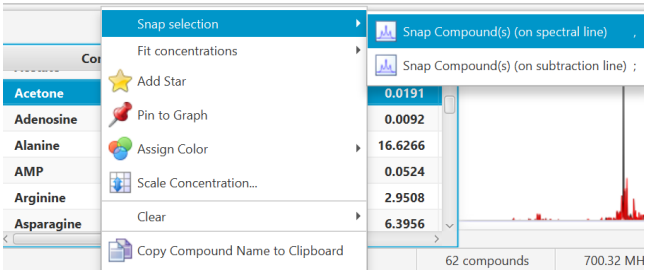
Compound Snapper focuses on optimizing peak cluster locations, individually or in group, depending on the user's selected cluster(s) or compound(s). Compound Snapper will not attempt to set or modify the concentration of compounds. This functionality automatically positions peak clusters on the spectrum at the location that **best matches their global shape**. If two or more clusters overlap within their allowed transform window, all possible combinations are analyzed.

As such, this feature is powerful, and will find the proper cluster combination, as sophisticated as it may be, that matches, according to shape, a relevant spectral region.

A snapping operation can either be performed on the original spectral line, or on the subtraction line, i.e. by considering the "left-over" signal from everything that has been fit already. It can also be done on a compound selected in the compound table and whose concentration has not been set already.

How to use Compound Snapper

- Select one or several clusters from an individual compound, a single or several compounds at once from the compound table, right-click in your selection to bring a contextual menu, or choose 'Snap Selected Compound(s)' from the Compound Menu (main toolbar).



You can also use the keyboard shortcut comma (,) to snap clusters using the experimental spectrum line, or semi-colon (;) to snap clusters using the residual line instead (the residual line is calculated using everything that is already fit except for the clusters/compounds being snapped).

- To snap all clusters resonating in a particular spectral region at once, first select the region with the Select Region tool (main tool bar), right-click and select "Find and Snap Peak Clusters in this Region".

Second Step: Concentration optimization using Fit Concentrations

Once peak clusters have been positioned, compound concentrations can be fit and optimized using constrained penalized linear least squares, according to:

$$\min_{l \le x \le u} \|W(Ax - b)\|_2$$

Where...

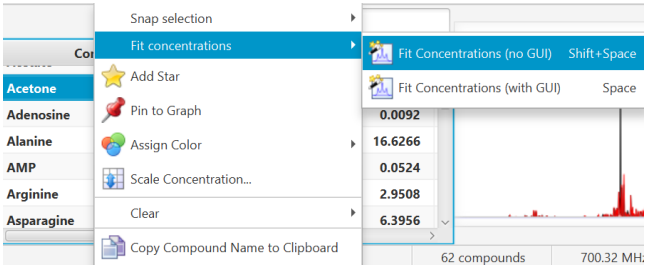
1. A is the model matrix made up the unscaled individual signatures of the compounds being fit. The current locations of the peak clusters are taken into account when the A matrix is calculated.
2. x is a vector made up all of the compound concentrations to be optimized. Each component of the vector, xi, is bounded between li (lower bound) and ui (upper bound). Non-negative restraints are imposed (li ≥ 0) since concentrations cannot be negative.
3. b is the spectral subtraction line, i.e. the experimental spectrum minus the contribution of all compounds part of the current profile that are not part of the optimization process. In other words, if a compound has been fit and his not part of the fitting process, its contribution is subtracted from the experimental spectrum before the process starts.
4. W is a weight vector, which is used to modulate the relative importance of each spectral point. Optionally, the algorithm can increase the weight of the points that are overfit though an iterative penalization process.

   IMPORTANT NOTE: The optimization process will not modify the location of the peak clusters.

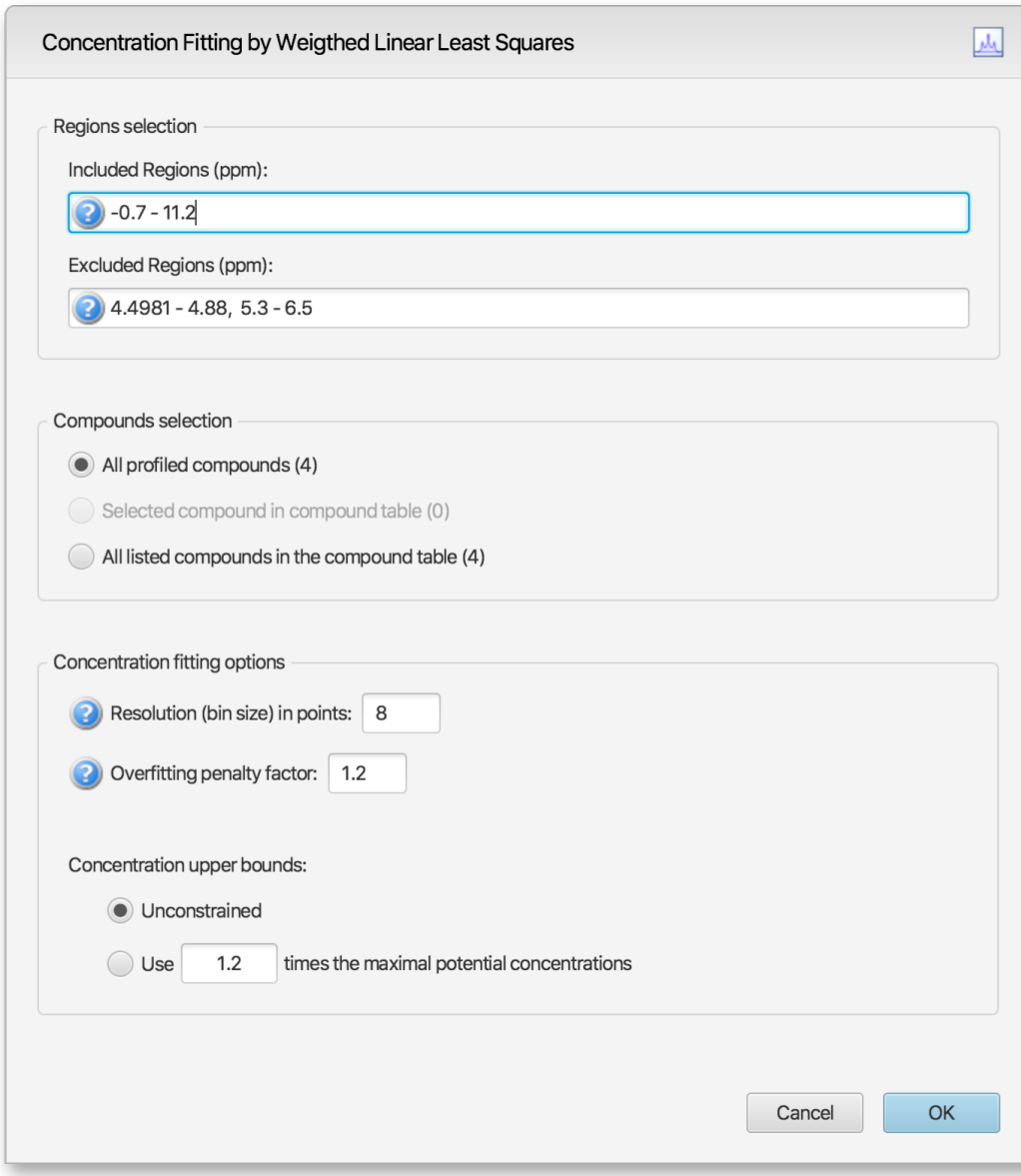
How to Fit Automatically

- Right-click/control-click selected compounds in Profiler's compound table, or on a selected spectral area.

  Concentration Fitting can be performed without a GUI (graphical user interface) by using the Shift+Space bar keyboard shortcut, in which case it uses the default options and values stored in Profiler's preferences.



- Alternatively, temporary settings can be entered via GUI (graphical user interface) using the space bar keyboard shortcut. The interface is composed of three main sections: Regions Selection, Compounds Selection, and Fitting options. The role of each section is described below. The application Preferences panel can be used to control and set all the default values appearing in the interface.



Regions Selection

- Comma-separated list of spectral regions (in ppm units) to be included or excluded in the fit. Each component of the lists must include a lower and upper bound in ppm, separated by a dash. For instance, “0.8-2.3, 3.0-4.2” is a valid list.
- The Included Regions input field displays the limits of the currently selected spectral region (as selected with the Select Region Tool), or of the selected clusters for an individual compound. If no region is currently selected, the Included Regions input field defaults to the entire spectrum.
- The Excluded Regions input field will automatically include the region associated with the water peak. It is recommended that any peaks around the water region be excluded due to attenuation caused by water saturation. Similarly, the region associated with urea (5.4-6.0 ppm) should also be excluded if urea is present.

Excluded regions have prevalence over any included regions, i.e. any spectral point falling within any of the listed excluded regions will automatically be ignored during the fit.

Compound Selections

- Users can optimize either all of the members of the current profile, all of the currently selected compounds in the compound table, or all of the currently listed compounds in the compound table. Note that compounds that do not have a single peak cluster within the participating spectral regions will not be considered for the fit. Their concentrations will not be changed.

Concentration Fitting Options

This group of options control the behavior of the optimization engine.

- Resolution (bin size): The spectrum can be fit using either its full representation, or a binned representation of it. This parameter controls the number of spectral points per bin. Set this parameter to 1 for full resolution. However, it is highly recommended to use a binned representation if the peak cluster positions have not been optimized. Fitting at full resolution will require more memory and CPU time.
- Overfitting penalty factor: The amount of overfitting can be limited by increasing the weights of the spectral points where overfitting occurs. This will effectively increase the importance of the spectral points where this occurs relative to the other ones, adding a penalty to pay for predicting points over the spectrum line. For a given spectral point, the square of the difference between the experimentally observed spectrum intensity and the predicted/calculated one will be multiplied by the factor specified in this field if the difference is negative. The weights are adjusted iteratively and up to 5 cycles of optimizations are performed if the specified penalty factor is >1.0.
- Upper constraints: Non-negative constrained are used during the optimization process so that concentration values remain ≥ 0. Two different settings are offered to control their upper bounds. They can be set to be unlimited via the Unconstrained option, or limited according to their predetermined maximal potential concentration. The maximal potential concentration of a given compound is the highest concentration for which the compound signature would not exceed the experimental spectrum. Because of possible discrepancies between the compound library signatures and the experimental spectrum, mainly due to pH conditions, matrix effects, etc, the potential concentrations determined by Profiler may not be optimal and should only be regarded as guidelines. To give potential concentrations more wiggle room, potential concentrations can be pre-multiplied by a specified constant. The derived set of scaled potential concentration values will then be used as upper bounds.

IMPORTANT NOTE: The baseline is not modeled from the residuals at any stage. It is therefore crucial that the baseline be properly corrected prior to concentration fitting.

Related Topics

- “Calibrate pH”
- “Compound Table”

5.6 COMPLETE Autofit: fully automated profiling approach

Version 10 of Chenomx NMR Suite introduces a new sophisticated, flexible and easy to use tool for automatic profiling of processed spectra.

Typically, when automatically fitting a spectrum, hundreds of variables (cluster positions and compounds concentrations) require optimization. Because compound concentrations are derived from existing peak cluster locations, and because peak clusters are allowed to be positioned within certain ranges ("transform windows"), several rounds of optimization are necessary to achieve a local or global minimum, i.e. a solution that reconstructs the experimental spectrum as best as possible.

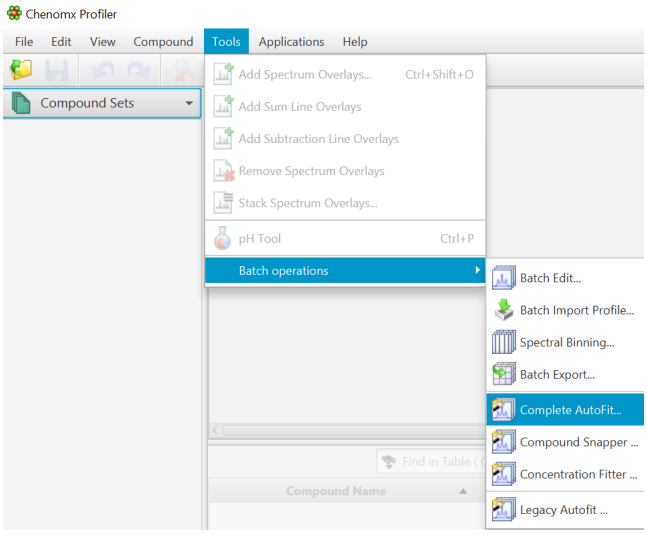
Moreover, different mathematical solutions may recreate the experimental spectrum with similar degrees of fitness/fidelity. For this reason, the software can explore those solutions by repeating the fitting process for each individual input file as many times as the user chooses, and output a solution for each pass.

It is important to keep in mind that automatic profiling is purely a mathematical process that simply relies on a provided compound library. It does have not have the prior chemical knowledge of a human operator. COMPLETE Autofit should not be used blindly and fit results should be reviewed manually.

How to use COMPLETE AUTOFIT

Steps to set up COMPLETE AUTOFIT

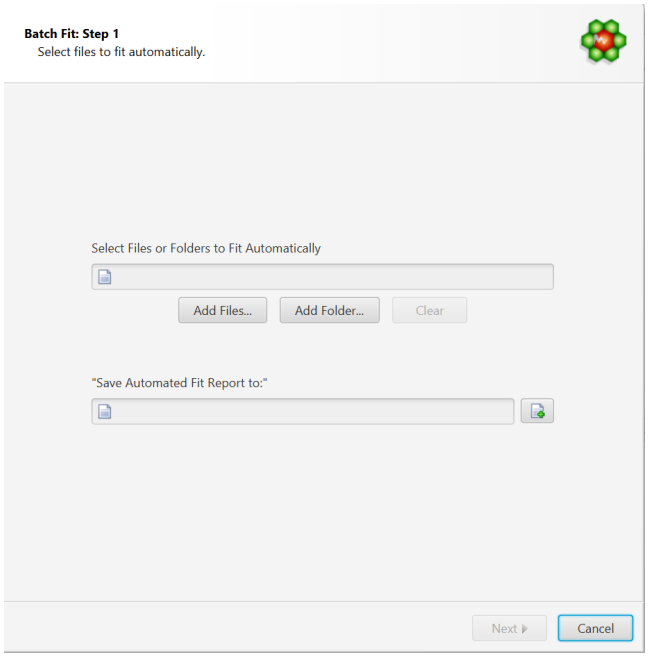
1. Open your processed CNX file within the Profiler Module.
2. Go to Tools -> Batch Operations -> "COMPLETE Autofit"



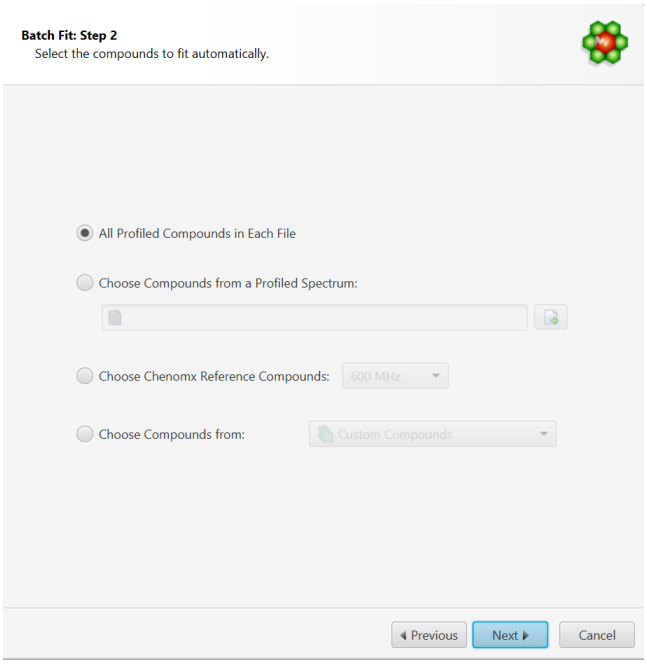
3. **Select Files to Fit Automatically**

   - Select Files or Folders to Fit Automatically: COMPLETE Autofit will do a complete profile of one, or several CNX files. Select your unprofiled CNX files here.
   - Save Automated Fit Report to: A fit report in the form of a tabulated separated value (tsv) file will be generated, with each input file and the sum of residuals

associated with each fit attempt. Specify the destination and name of the report file here.



4. Select the Compounds to Fit Automatically. You can either:



- Choose to fit all the compounds already profiled in each file. In this case, compounds must be already present in their profile and all of the compounds will be included in the fit operation.
- Choose compounds from and already profiled spectrum. Note that it is possible to select only a subset of those in the interface's next step.
- Choose compounds from a Chenomx library

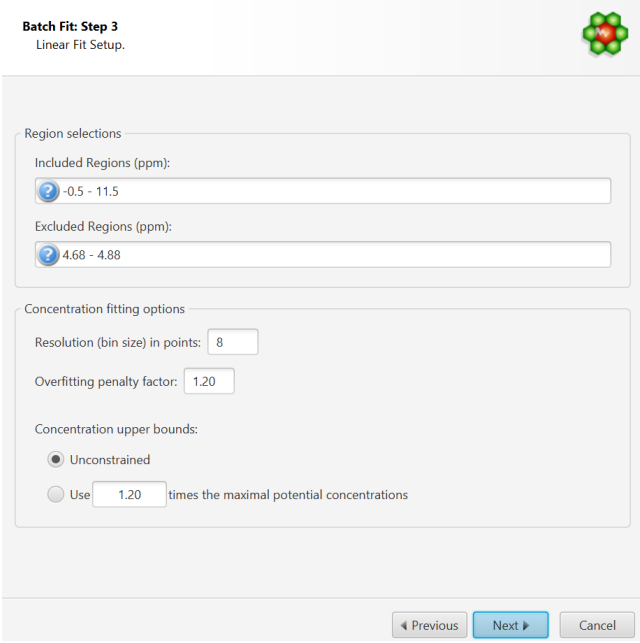
- Choose compounds from a customized compound list defined in Library Manager.

IMPORTANT NOTE: DO NOT ATTEMPT TO FIT THE ENTIRE CHENOMX LIBRARY. THIS WILL RESULT IN OVERFITTING AND AN EXTREMELY LONG PROCESS. TARGET THE COMPOUNDS THAT SHOULD BE VISIBLE AND PRESENT FOR THE TYPE OF SAMPLES YOU HAVE.

5. Compound Selection Refinement

- • Depending on your selection in the previous step, you may be presented with an option to further refine your compound selection. You can move compounds across list boxes to specify whether or not you want to include them in the fit.
- • The checkbox option “Skip compounds that are already fit” is self-explanatory. It allows you to ignore compounds that are already fit. Their contribution will be calculated and taken in consideration to fit the reminder of the compounds, but their current state (cluster positions, concentrations) will not be modified.

6. Concentration Fitting Setup



This interface is composed of two main sections: Regions Selection, and Concentration Fitting options. The role of each section is described below. The application Preferences panel can be used to control and set all the default values appearing in the interface.

a. Region Selection: Comma-separated list of spectral regions (in ppm units) to be included or excluded in the fit. Each component of the lists must include a lower and upper bound in ppm, separated by a dash. For instance, “0.8-2.3, 3.0-4.2” is a valid list.

- The Included Regions input field displays the limits of the currently selected spectral region (as selected with the Select Region Tool). If no region is currently selected, the Included Regions input field defaults to the entire spectrum.

- The Excluded Regions input field will automatically include the region associated with the water peak. It is recommended that any peaks around the water region be excluded due to attenuation caused by water saturation. Similarly, the region associated with urea (5.4-6.0 ppm) should also be excluded if urea is present.

  Excluded regions have prevalence over any included regions, i.e. any spectral point falling within any of the listed excluded regions will automatically be ignored during the fit.

b. Concentration Fitting Options: This group of options control the behavior of the optimization engine.

- Resolution(bin) size:

  The spectrum can be fit using either its full representation, or a binned representation. This parameter controls the number of spectral points per bin. Set this parameter to 1 for full resolution. However, it is highly recommended to use a binned representation if the peak cluster positions have not been optimized. Fitting at full resolution will require more memory and CPU time.

- Overfitting Penalty Factor:

  The amount of overfitting can be limited by increasing the weights of the spectral points where overfitting occurs. This will effectively increase the importance of the spectral points where this occurs relative to the other ones, adding a penalty to pay for predicting points over the spectrum line. For a given spectral point, the square of the difference between the experimentally observed spectrum intensity and the predicted/calculated one will be multiplied by the factor specified in this field if the difference is negative. The weights are adjusted iteratively and up to 5 cycles of optimizations are performed if the specified penalty factor is >1.

- Concentration Upper Bounds and Penalty Factor:

  Non-negative constraints are used during the optimization process so that concentration values remain ≥ 0. Two different settings are offered to control their upper bounds. They can be set to be unlimited via the Unconstrained option, or limited according to their predetermined maximal potential concentration. The maximal potential concentration of a given compound is the highest concentration for which the compound signature would not exceed the experimental spectrum.

  Because of possible discrepancies between the compound library signatures and the experimental spectrum, mainly due to pH conditions, matrix effects, etc, the potential concentrations determined by Profiler may not be optimal and should only be regarded as guidelines. To give potential concentrations more wiggle room, potential concentrations can be pre-multiplied by a specified constant. The derived set of scaled potential concentration values will then be used as upper bounds.

- NOTE: It very important to note that the baseline is not modeled from the residuals at any stage. It is therefore crucial that the baseline be properly corrected prior to concentration fitting.

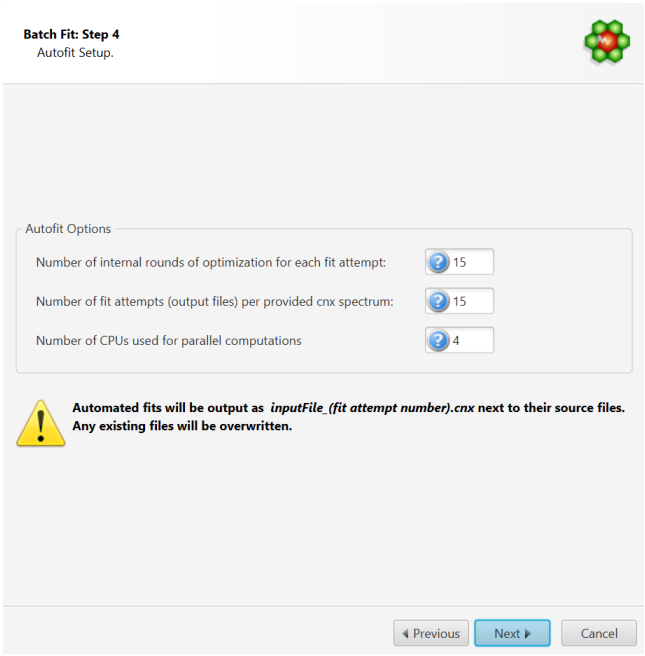
7. Autofit Setup

   This panel controls the overall behaviour of automated fitting:

a. The number of internal iterative rounds is related to the number of times, for each fit attempt of a given spectrum, the algorithm will position clusters and adjust concentrations. It is directly equivalent to the situation encountered with manual fitting, where an operator usually has to do several rounds of going through the list of compounds to be fit and adjust their peak cluster positions and height (concentration).

b. The number of fit attempts control how many output files will be generated for each input spectrum cnx file. By analogy, it is related to how many people would be trying to fit the spectrum if there were a team of operators. Each one could potentially come up with a different solution that produces more or less the same fit quality in terms of sum of squares of the residual line.

c. The number of CPUs control how many CPUs can be used for parallel computations. The number presented in the interface takes into account the maximum number of CPUs present on the computer system, and the number of CPUs that may already be used by another COMPLETE Autofit job.



8. Compound Library Calibration Method

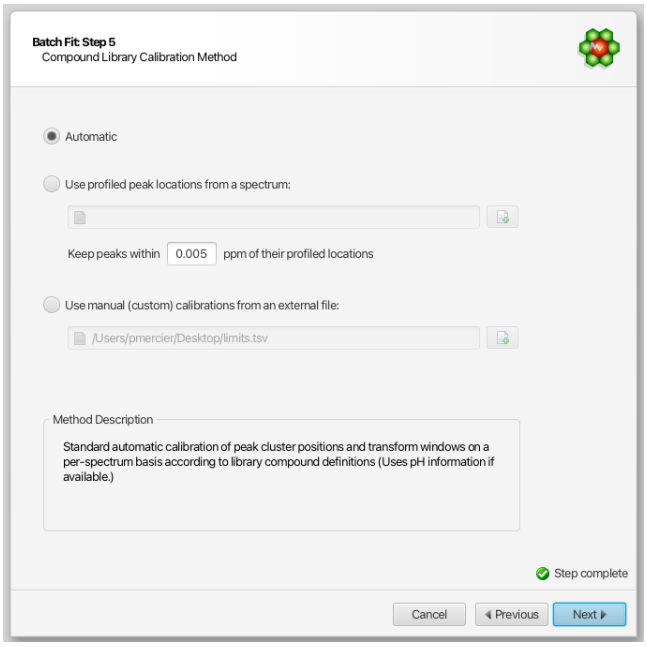
The compound library is automatically calibrated on a per-spectrum basis. The starting peak cluster positions, as well as their range of allowed values (transform windows), are determined based on pH information in each spectrum. Optionally, you can overwrite the positional restraints with your own set of values.

This can be particularly useful in situations where peak multiplets appear outside of their calibrated transform windows, due to matrix effects or other causes. Although Profiler allows for manually positionning multiplets outside of their transform windows, the AUTOFIT procedure strictly enforces them.

A mechanism was put in place to overwrite (post-calibrate) the automatically determined values. You can force the automated procedure to position multiplets around their profiled locations in an external spectrum, within a give range (in ppm units). This method is compound-based, i.e. all clusters of a given compound will be restrained.

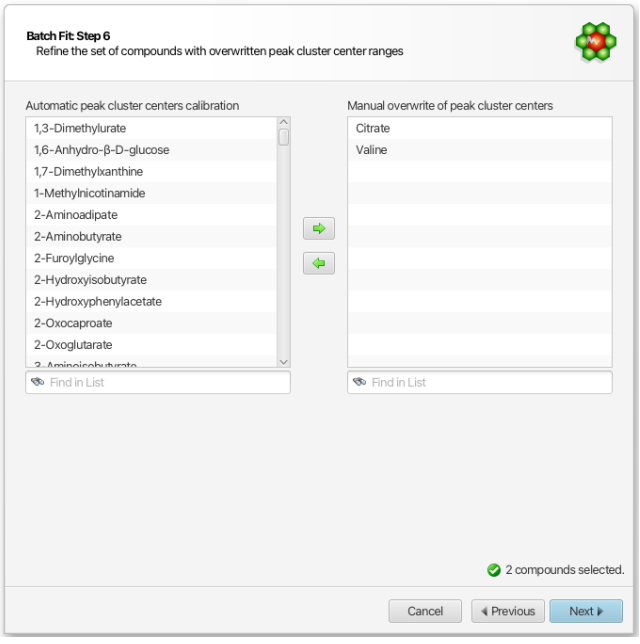
Alternatively, you can opt for a more advanced and flexible cluster-based approach, via an external tab-separated file prepared within Profiler. Refer to the section "Custom calibration of peak cluster centers ranges for automated procedures" for the preparation of such a file.

You can further refine your compound selection in the following next step.



9. Compound Selection for Custom Compound Library Calibration

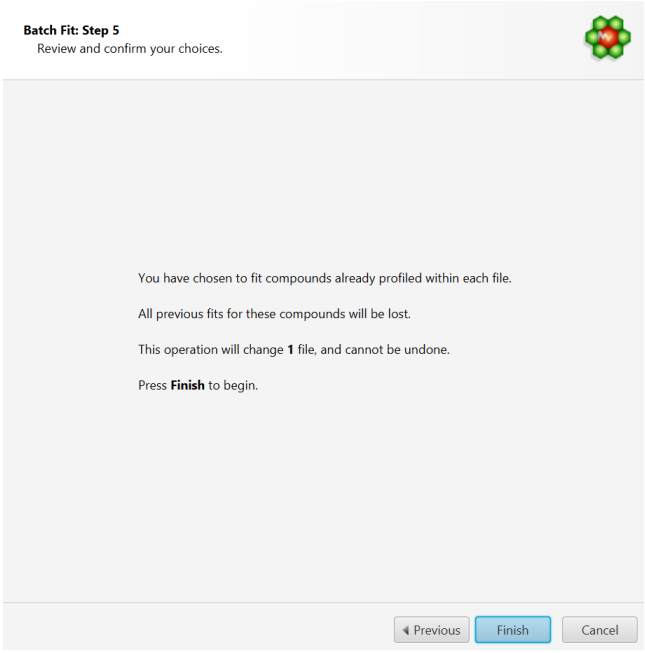
   If you have choosen to post-calibrate (overwrite) the automatic compound calibration with your own set of values, you can further refine which compounds it will apply to from the all of the compounds available from your selected source in the previous step:



At this step, you should exclude compounds known to be particularly very sensitive to pH (exhibit large chemical shift variations with pH). For instance, it is highly advisable to exclude any of the histidine-based compounds if you are operating around pH 7.0, and simply let the automated procedure proceed with the default calibration for those.

Important: If you choose a profiled cnx file as your source of calibration, all the clusters of the selected compounds in this step will be considered, whereas if you choose an external tab-seperated value file, specific clusters can be included or omitted.

10. Review and Finish



- COMPLETE Autofit will generate multiple profiled CNX files for each input spectrum file that will be saved in the same location as the input source. It will not however modify the original source files themselves.
- A fit report will be produced, showing the residuals chi-square value for each of the produced cnx file. For a given input file, the output solution with the lowest chi square (lowest fit error) is automatically saved with the label "_best" appended at the end of its filename.
- A text file (.txt) will also be generated for each exported CNX file with the chi square value achieved for each internal round of iteration. Inspection of those text files can reveal whether or not the number of internal rounds that were specified is enough to achieve a minimum. If the best solution (lowest chi square) is generally obtained only at the last internal iterations, it is a sign that maybe a higher number of iterations is needed.
- It is up to the user to choose the output solution that produced the best fit, if the decision parameter is other than the sum of residuals. For statistical analysis (PCA, PLS-DA, etc.), it may be more relevant to work with concentrations averaged over the series of exported fits rather than single values.

Tips and Limitations of COMPLETE Autofit

- Not all spectra are suitable for automated fitting. Unfiltered spectra showing presence of larger molecules though humps will likely fail. The software does not model the baseline on the fly.
- The raw fid must be correctly processed and the baseline must be adjusted properly. The software does not model the baseline on the fly.
- Set the pH for each sample, either in Processor or Profiler. pH information is used to calculate the boundaries for each peak cluster. The software will not fit peak clusters outside of their calculated transform windows.
- If spectral regions contain intense unknown peaks, exclude those regions from the fit in the provided interface.
- The time required to fit a spectrum is directly dependant on the number of spectral points, and the total numbers of peak clusters and compounds. Avoid under and over fitting, i.e. work with compound subsets that represent what you should expect to find in the type of samples you are analysing.

Note about Legacy Autofit

Version 10 also reintroduces a form of automated fitting found in version 8.6 and earlier of the software, as “Legacy Autofit”. Legacy Autofit may be preferable to older users that want to maintain controls for a project initiated using older versions of Chenomx NMR Suite. For users starting a project using Chenomx NMR Suite Versions 9 or later, consider using more advanced Autofit techniques such as Compound Snapper / Fit Automatically or the COMPLETE Autofit Tool.

The difference between Legacy Autofit and COMPLETE Autofit is explained below.

1. Legacy Autofit first takes the raw spectrum and subtracts the contribution of all previously fit compounds that are not selected for the autofit.
2. Starting from the spectrum obtained in step #1, it scores the compounds selected for autofit to establish the order in which they will be fit.
3. Starting from the best potential candidate, it tries to simultaneously establish the position of the clusters for that metabolite and the maximum intensity (and therefore concentration) of that compound on the spectrum. (If it fails to establish where some clusters are located, they are left at their default/untransformed locations.)
4. The contribution of the potential candidate is then subtracted from the spectrum to yield another spectrum which serves as starting point for the processing of the second candidate compound, using the same procedure.
5. This cycle is repeated until the list of selected compounds for autofitting has been exhausted.
6. One can clearly see that the risk of errors increases at every step, and that eventually not all compounds will succeed in being autofitted. Moreover, as individual compounds are added at their maximal concentrations, the final result does not reflect a global optimization of all compounds, but reflects a successive series of individual optimization. (In order words, after adding compound #x, the algorithm does not reevaluate compound #x-1, #x-2, #x-3, etc.)

The main difference between COMPLETE Autofit and the Legacy Autofit approach relies on the fact that the new algorithm is based on a true “global” optimization process. The

compound concentrations are obtained altogether (globally) using penalized least squares, rather than through successive fit rounds of individual compounds. The algorithm used for positioning peak clusters has also been greatly improved. Be aware though that there is a computational speed cost to those improvements.

5.7 Importing and Exporting Data

Export data from your profiled spectra for further analysis, bin the spectra for analysis by more traditional techniques, or create images of your spectra for use in publications and presentations.

Import Profile

Spectral profiling can be accelerated by importing, partially or entirely, profiled data of spectra that have already been fit. This is particularly efficient when dealing with large series of similar spectra.

How to import a profile from another spectrum

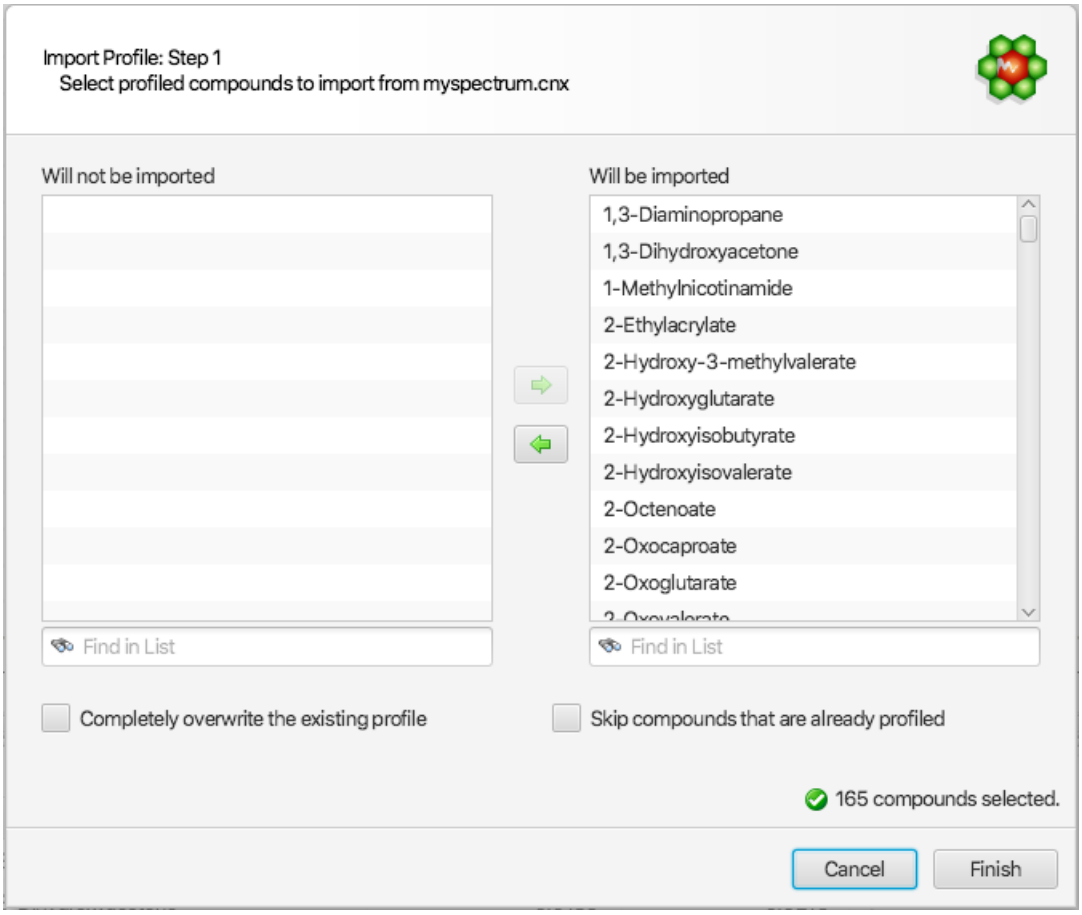
Menu Location	File > Import Profile...
Icon	

There are 4 ways profiled data can be imported:

1. Select 'Import Profile' from the File menu.
2. Select 'Batch Import Profile' from the Batch Operations in the Tools menu.
3. Drag and Drop a cnx file directly in the compound table. The file can also be chosen from Files panel that can be displayed in the left or right vertical sidebar in Profiler
4. Select and drag compounds from the compound table of one opened instance of Profiler to another.

Except for the last option, changes are automatically applied. Any dragged compounds from another profile will automatically overwrite what may exist. The operation is cancellable by selecting Undo Import Profile from the Edit menu.

For the other first 3 options, the following interface will be presented to allow for more options:



The right listbox will automatically be populated with all of the compounds that are available from the source cnx spectrum, and automatically chosen as the default set of compounds to be imported. Compounds that should not be imported can be selected and drag to the "Will not be imported" listbox on the left side of the interface. All compounds chosen for import will merge with the existing profile, and the profile of pre-existing compounds will be automatically replaced by those imported, unless the "Skip compounds that are already profiled" option is selected (see below).

Two additional options are available to control the behaviour of the import operation:

1. Completely overwrite the existing profile: This option will first completely wipe the current existing profile before the selected compounds are imported.
2. Skip compounds that are already profile: If selected, this option will ignore the import of compounds that are pre-existing

Tips and Tricks

- When importing a profile, fully or partially, the imported data completely replaces what currently exists. That applies to peak cluster transforms, compound concentrations, and compound annotations (colors, etc).

Export Profiled Compounds

Exporting profiled compounds creates a compound pack (`.pack`) containing the set of actual compound signatures used to profile a project, including compounds that have been

annotated with stars, pins, or colors. Exchange compound packs with other users of Chenomx NMR Suite, and import new packs via Library Manager. This is one way to be certain that collaborators are profiling spectra for a project using exactly the same collection of compound signatures.

Menu Location	File > Export > Profiled Compounds...
Icon	

How to export a compound pack from the current spectrum

1. Open the Export Profiled Compounds dialog.
2. Enter a name for the exported compound pack.
3. Click OK.
4. In the resulting file chooser, locate the folder and create a name for Compound Pack.
5. Click **Save**.

Tips and Tricks

- To export all of the compounds in a compound set, regardless of whether they have been fit in a particular spectrum, use **Export as Compound Pack** in Library Manager.

Related Topics

- "Add Compounds"
- "Export Compounds"

Export Compound Table

Export Compound Table creates a copy of the current compound table in a format which can be easily imported into any modern spreadsheet program.

Menu Location	File > Export > Compound Table...
Icon	

How to export current compound table from a spectrum

1. Click File > Export > Compound Table.
2. Choose a save location and file format. You can export as comma-separated text (.csv), as tab-separated text (.tsv), or as an excel spreadsheet (.xlsx).
3. Click Save.

Tips and Tricks

- If there are blank cells, there will be corresponding blank values in the exported data (this can happen if the table contains compounds that are missing from your compound library or if you export columns that can have empty cells, like "author", "pulse sequence", or "compound comment")
- The "Status" column will never be exported, even if it is visible in the table.
- The Starred and Pinned columns will export as "Yes" or "No" for each cell, and the Color column will export as a hex RGB value, such as 0000FF for each cell. Cells that have no color assigned will all export as the "default" color for that cell, i.e. the color that the cells compound appears when displayed on the graph.
- Add/remove columns, reorder columns, or click on the header of any column to change the table sort order. The columns you chose to appear in your Compound Table will be exported, and the order of the rows and columns in the exported file will be in the same order as your Compound Table.

Batch Export

Batch Export creates a text file containing all of the relevant details from profiled spectra, in formats that you can easily open in software like Microsoft Excel or Umetrics SIMCA-P.

Menu Location	Tools > Batch Export...
Icon	

How to export data from one or more spectra

1. Start the Batch Export wizard.
2. Select the type of data that would you like to export. You can export concentrations in various units, other profiling results like profiled areas, matched clusters and transform summaries, or annotations like stars, pins and custom colors.
3. Choose the source spectra to export, and specify a target folder and filename for the exported text file. You can specify the currently open spectrum, other individual spectrum files or one or more folders containing spectrum files as the source.
4. Choose a save location and file format. You can export as comma-separated text (.csv), as tab-separated text (.tsv), or as an excel spreadsheet (.xlsx).
5. Choose the compounds for which you would like to export data. You can use custom compound sets that you have created in Library Manager to filter the output to specific compounds, or you can export all profiled compounds in all of the spectra.
6. Choose additional data that you would like to include in the exported file. Additional data can include spectrum pH values, Chenomx compound IDs for the exported compounds, or InChI or SMILES strings for the exported compounds.
7. Choose the default layout (files in rows and compounds in columns) or transposed layout (compounds in rows, files in columns) for the output file, depending on how you intend to use the file.

Concentration Confidence Intervals

- When exporting concentrations, the software will also export the statistical standard deviations on concentrations as a second tab in the excel workbook. Concentration confidence intervals (± values) at 90%, 95%, and 99% can be calculated by multiplying the reported standard deviations by 1.645, 1.96, and 2.576, respectively.
- For each compound, the standard deviation of each cluster is calculated individually, by calculating a 'local' sum of difference of squares in the spectral region currently occupied by each cluster. The minimum value found for all clusters is reported. As such, the reported standard deviation should be considered "best-case scenario'.
- Failure to properly correct the spectral baseline will result is larger sum of squares, and therefore larger concentration errors.
- Confidence intervals can be used to assist in the determination of the meaningfulness of reported fit concentrations, and in the decision to include or reject compounds in statistical models.

Tips and Tricks

- Export Areas rather than Concentrations when working with compounds that have a region-based **Fit Style** (see "Compound Details")
- To filter the exported compounds without modifying the source spectra, create a Compound Set or Smart Compound Set in Library Manager, and only export compounds in that Compound Set.

- Use the default layout to export files that you intend to open with statistical analysis packages. Use the transposed layout to export files that you intend to open in a spreadsheet program, or if you prefer the transposed layout.
- If you intend to open the exported file in Microsoft Excel 2003 or earlier, limit the number of compounds to less than 253 for the Default layout, or the number of files to less than 253 for the Transposed layout, since these versions of Excel do not support large numbers of columns.

Related Topics

- "Compound Table"
- "Compound Fit Style"

Send to Processor

Menu Location	File > Send to Processor
Icon	

As you work with a spectrum in Profiler, you may notice that the phase correction, baseline correction or other aspects of the spectrum's lineshapes are not properly adjusted. Similarly, you may import a spectrum directly into Profiler, only to realize that it requires phase correction, baseline correction, or additional pH or CSI calibration. **Send to Processor** closes the current spectrum in Profiler, and opens it in Processor, retaining any changes you have made in Profiler.

Tips and Tricks

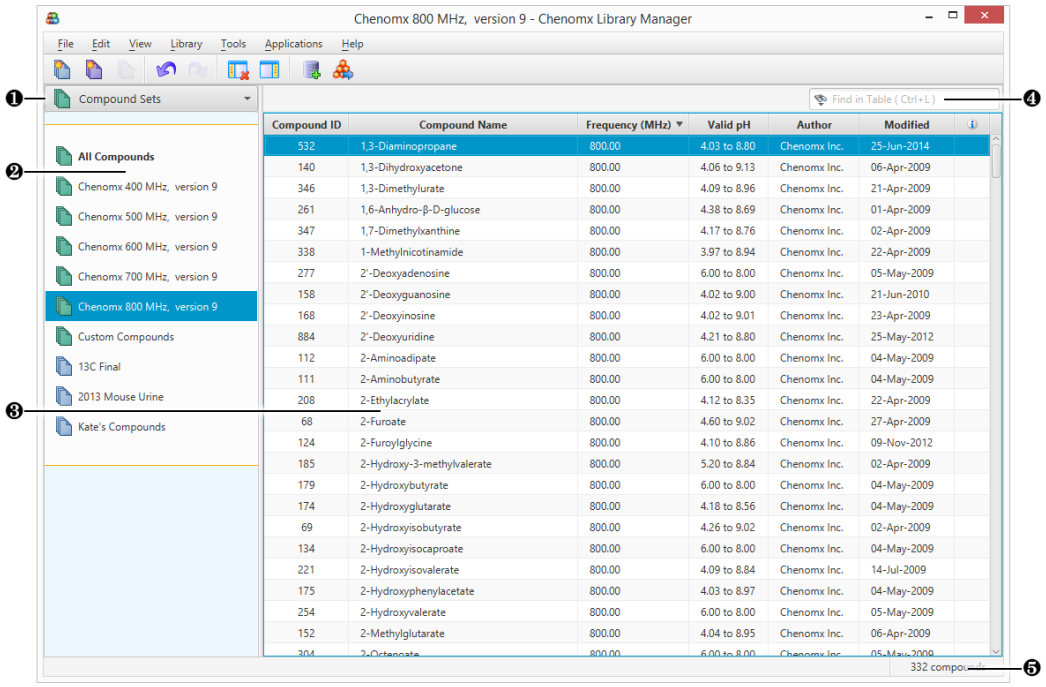
- Any changes that you have made in Profiler are retained when you send the spectrum to Processor, and will be visible if you bring the spectrum back into Profiler. However, you still need to save the spectrum, either in Processor or Profiler, to permanently save those changes to the spectrum file.

Chapter 6. Library Manager

The Library Manager module lets you create and modify compound sets based on the compounds in your library, and it also allows you to add compounds to your library by importing compound packs and `.xcpd` files that you have created with Compound Builder.

For a hands-on introduction to Library Manager, see "Tutorial and Sample Files".

6.1 Overview



❶ Sidebar View Dropdown Menu. Lets you change the currently visible Sidebar view.
❷ Compound Sets. Sidebar view letting you control which compound set appears in the Compound Table.
❸ Compound Table. Displays information about compounds and their status in the library. You can send the signature of a compound to Compound Builder for editing by clicking **File > Send to Compound Builder** or by double-clicking on the compound.
❹ Quick Search. Lets you filter the Compound Table to focus on a particular compound or group of compounds.
❺ Status Bar. Displays information about recent user actions.

6.2 Library Tools

Library Manager lets you browse and manage the contents of your compound library. Your compound library contains the compounds that are available for you to use when profiling a spectrum.

Compound Table

The Compound Table in Library Manager displays compounds in your library that are part of the currently selected Compound Set. When you first open Library Manager, the **All Compounds** set is selected, so the Compound Table displays all of the compounds in your library. When you select a different Compound Set, the Compound Table changes to show the contents of that set.

You can use the Compound Table to help you build new Compound Sets via drag-and-drop and context menus. You can also use text-based Quick Searches to temporarily filter the compounds that are currently visible in the Compound Table.

There are many columns available for display in the Compound Table. You can choose to display any number of them, with a minimum of one visible at any time, and you can drag the column headers to position the columns in any order that you like. You can sort the Compound Table on any column by clicking its column header. Click the header again to reverse the sort order.

Tips and Tricks

- You can add compounds to a Compound Set by dragging them from the Compound Table onto the name of the Compound Set, or by right-clicking on the compound and selecting **Add to New Compound Set** or **Add to**. You cannot add compounds to Smart Compound Sets or to Reference Compound Sets in this manner.
- You can view a compound in the Compound Table in more detail by double-clicking on it or by right-clicking on its entry and selecting **Send to Compound Builder**.

Related Topics

- "Change Columns"
- "Quick Searches"
- "Send to Compound Builder"
- "Remove Compounds"
- "Compound Set Tools"

Quick Searches

The number of compounds appearing in the Compound Table can make it difficult to locate a particular compound. The Quick Search box helps with this by letting you enter search text to temporarily limit the number of compounds appearing in the Compound Table. The search text can by based on compound name, compound IDs, or comments.

How do I limit the compounds visible in the Compound Table?

1. Click the binoculars icon at the left edge of the search box to choose the type of search to use (Compound Name, Compound Comment, Chenomx Compound ID or All). If you do not select something here, the default search type is All.
2. Type your search terms in the Quick Search box at the top left of the Compound Table. The contents of the Compound Table updates to show only compounds matching your current search terms.
3. Click the X at the right edge of the search box or press escape to cancel the search and restore the full list of compounds. Clearing a search also resets the search type to All.

Tips and Tricks

- Use Quick Searches to temporarily limit the number of compounds displayed. Use compound sets to define a more permanent working set of compounds for profiling a particular spectrum.

Related Topics

- "Compound Table"
- "Remove Compounds"
- "Compound Set Tools"

Add Compounds

You can add compounds to your compound library from a variety of sources, including compound files, compound packs, old Chenomx library files and ZIP (compressed) archives. Add Compounds lets you add compounds from all of these sources to your library, which in turn makes the compound signatures available for use in Profiler.

Menu Location	Library > Add Compounds...
Icon	

How do I add Chenomx Reference Compounds to my library?

1. Open the Add Compounds wizard.
2. Select Chenomx Reference Compounds, then click Next.
3. Select the frequencies for which you would like to add Chenomx Reference Compounds.
4. Click Finish to add the selected compounds.

How do I add other compounds to my library?

1. Open the Add Compounds wizard.
2. Select Importable Compounds, then click the Browse button (**...**).
3. Select any combination of compound files (`.xcpd`), compound packs (`.pack`), old Chenomx library files (`.cnxlib`), ZIP files (`.zip`) or folders containing any of these file types, then click Choose Files.
4. Click Next.
5. If you would like the compounds added to a new Compound Set, select the Track Imported Compounds checkbox and choose a name.
6. Click Finish to add the selected compounds.

Tips and Tricks

- When you add compounds, the icon for any compound sets that were created or modified during the addition process will 'pulse' for a couple seconds to indicate that they were affected by the import.

Related Topics

- "Compound Table"
- "Compound Set Tools"

Export Compounds

When you collaborate with other people on a project, you may have different compounds in your library than your collaborator has in theirs. Exporting compounds allows you to exchange compound signatures with other users of Chenomx NMR Suite. You can then be certain that you and your collaborator are always fitting spectra for that project using the same set of compound signatures.

You can export compounds as a single compound pack file (`.pack`), or as collections of individual signature files (`.xcpd`).

Menu Location	Compounds > Export Selected Compounds to > Compound Pack (.pack)...
Icon	

How do I export compounds to a compound pack?

1. Select the compounds or compound set that you would like to export.
2. Open the **Export Compound Pack** dialog.
3. Enter a name for the exported compound pack. The entered name is used to label the associated compound set when the compound pack is added to another library via Library Manager.
4. Click OK.
5. In the resulting file chooser, locate the folder in which you would like to save the compound pack.
6. In the File Name field, type the new name for the compound pack file. The specified name can be different from the name of the compound pack itself, but you may prefer to keep them the same.
7. Click **Save**.

Menu Location	Compounds > Export Selected Compounds to > Compound Files (.xcpd)...
Icon	

How do I export compounds to individual compound files?

1. Select the compounds or compound set that you would like to export.
2. On the Compounds menu, click **Export Selected Compounds to > Compound Files (.xcpd)...**.
3. In the resulting file chooser, select the folder in which you would like to save the compound files.
4. Click **Select**.

Tips and Tricks

- If you only want to export compounds that were actually profiled in a particular spectrum, load the spectrum in Profiler and use **Export Profiled Compounds**.

Related Topics

- "Add Compounds"
- "Compound Set Tools"
- "Export Profiled Compounds"

Send to Compound Builder

Library Manager can let you view the details and lineshape of any compound in your library by sending a copy of that compound to Compound Builder.

Menu Location	File > Send to Compound Builder
Icon	

How do I send a compound to Compound Builder?

1. Select a compound in the Compound Table.
2. Click **Send to Compound Builder**.

   OR

   Double-click on the compound in the Compound Table.

Tips and Tricks

- Changes that you make to compounds that you have sent to Compound Builder will NOT be automatically reflected in your compound library. If you want changes to appear in your library, you need to save them to an `.xcpd` file in Compound Builder, and then re-add that compound file to your library with Library Manager.
- You can send multiple compounds to Compound Builder by selecting them in the Compound Table, right-clicking on your selection, and selecting **Send to Compound Builder**.

Related Topics

- Chapter 8, *Compound Builder (PRO Only)*
- "Compound Table"
- "Quick Searches"
- "Compound Set Tools"

Remove Compounds

If you are certain that you no longer want a compound to be accessible in Chenomx NMR Suite, you can remove it from your library.

Menu Location	Library > Selected Compounds > Remove
Icon	

How do I remove compounds from my library?

1. Select the compound that you would like to remove in the Compund Table.
2. Click **Selected Compounds > Remove**.

Menu Location	Library > Remove Compounds > [Compound Set]
Remove Compounds Icon	

How do I remove all compounds from a compound set?

1. Click **Remove Compounds > [Compound Set]**. This will also remove the Compound Set itself.

Tips and Tricks

- When you have removed compounds from the library, you cannot recover them through Undo. If you still have the original compound files (`.xcpd`) or compound packs (`.pack`) available, you can add them again.

  If you have removed Chenomx Reference Compounds from your library, you can easily recover them by running the Update Compounds or or the Add Compounds feature.

Related Topics

- "Compound Table"
- "Quick Searches"
- "Compound Set Tools"

- "Add Compounds"
- "Update Compounds"

Update Compounds

Whenever you run a new major version of NMR Suite for the first time, you will be prompted to update any Chenomx Reference Compounds that you have installed. If you choose not to update, your reference compounds will remain unchanged. In that case, you may wish to perform this update manually.

Menu Location	Library > Update Compounds...
Icon	

How do I manually update my reference compounds?

1. You can choose to update to the latest version of the Chenomx Reference Library at any time. Click **Update Compounds**.
2. Click **Yes**.

Tips and Tricks

- You can run Update Compounds at any time to restore your Chenomx Reference Compounds to the latest version. This can be useful for restoring missing compounds.
- Update Compounds will only update your library with the latest reference compounds that are available for your version of Chenomx NMR Suite. Newer versions of Chenomx NMR Suite will contain additional reference compounds that cannot be obtained by running this feature.
- It is recommended that you use the same version of the Chenomx Reference Library when profiling spectra that belong to the same study.
- If required, previous versions of the Chenomx Reference Library can be downloaded from our website at http://www.chenomx.com/compounds . A version history is also available.

Related Topics

- "Automatic Compound Sets"

6.3 Compound Set Tools

Compound Sets are groupings of compounds from your compound library that you can use in Profiler. You can create them either by selecting specific compounds or by defining specific rules (Smart Compound Sets). Library Manager includes a number of tools to help you manage your compound sets.

Compound Sets (Sidebar)

Menu Location	View > Sidebars > Left Sidebar > Compound Sets
Icon	

Menu Location	View > Sidebars > Right Sidebar > Compound Sets
Icon	

The Compound Sets view in the Sidebar contains a list of the currently defined compound sets and lets you interact with them by displaying their contents or editing them.

How do I display the contents of a compound set?

1. Open the Compound Sets view in a Sidebar.
2. Select the set in the Compound Sets view. The contents of the set appear in the Compound Table.

Tips and Tricks

- You can select and drag compounds onto a blank part of the Compound Sets view to create a new compound set.
- You can drag compounds onto a Compound Set in the Compound Sets view to add them to the set. You cannot drag compounds onto Smart Compound Sets.
- You can rename compound sets directly in the Compound Sets view. Click on the name of a selected set, type the new name, and press **Enter** to accept the new name.
- Several operations involving compound sets are available by right-clicking on them in the Compound Sets view.

Related Topics

- "Library Tools"
- "Export Compounds"
- "Compound Sets"
- "Smart Compound Sets"
- "Rename Compound Sets"
- "Remove Compound Sets"

Automatic Compound Sets

Some compound sets are created and removed automatically by Chenomx NMR Suite. These sets cannot be edited or deleted, and their contents are automatically updated whenever you add or remove compounds from your library.

For example, a compound set will be created for each available magnet frequency when any of the Chenomx Reference Compounds are added to your compound library. The names of these new compound sets will include their associated magnet frequency, as well as the version numbers of the Chenomx Reference Library that their compounds belong to.

The **Custom Compounds** set is another type of automatic compound set. It contains all of the custom compounds in your library (recall that custom compounds are compounds that you created with Compound Builder.) This compound set will appear automatically the first time you add a custom compound to your library.

Tips and Tricks

- A version history of changes to the reference compounds in the Chenomx Reference Library is available on our website at http://www.chenomx.com/compounds .

Related Topics

- "Add Compounds"
- "Remove Compounds"
- "Update Compounds"

Compound Sets

You may need to create a compound set that does not change as you add new compounds to your library. For example, analysis of an extended dataset may require profiling a fixed set of compounds to help ensure a consistent analysis, or you may want to profile only a specific group compounds. By using a Compound Set, you can include arbitrary compounds on a per-compound basis, hand-picking the members of the set to suit your needs.

How do I create an new, empty Compound Set?

Menu Location	File > New Compound Set...
Icon	

1. Click **New Compound Set**.
2. Enter a name for the new Compound Set.
3. Start adding compounds from your library or other compound sets.

How do I create a new Compound Set containing specific compounds?

1. Select a compound or group of compounds in the Compound Table.
2. Click **New Compound Set**.
3. Enter a name for the new Compound Set.
4. Alternately, drag the selected compounds onto an empty space in the Compound Sets view.
5. Alternatively, right-click on a selected compound or group of compounds in the Compound Table, and click **Add to New Compound Set**.

How do I create a new Compound Set containing all the profiled compounds in a .cnx file?

Menu Location	File > New Compound Set from Profile...
Icon	

1. Click **New Compound Set from Profile...**.
2. Use the **Add Files** or **Add Folder** buttons as many times as desired to select one or more `.cnx` files.
3. Alternately, drag and drop one or more `.cnx` files or folders onto an empty space in the Compound Sets view.
4. Enter a name for the new Compound Set, or keep the name that is already suggested.
5. Click **OK** to create a new Compound Set, which will contain all the compounds that have been profiled in any of your selected `.cnx` files.

How do I add compounds to an existing Compound Set?

Menu Location	Library > Selected Compounds > Add to [Compound Set]
Icon	

1. Select a compound or group of compounds in the Compound Table.
2. Click **Add to [Compound Set]** to add the compounds to an existing set.
3. Alternately, drag the selected compounds onto the name of the Compound Set in the Compound Sets view.

Menu Location	Library > Selected Compounds > Remove
Icon	

How do I remove compounds from a Compound Set?

1. Select the Compound Set from which you would like to remove compounds in the Compound Sets view.
2. Select the compounds that you would like to remove in the Compound Table.
3. Click **Remove**.

Tips and Tricks

- When the contents of any compound set change, usually because you've added or removed compounds from your library, the compound set's icon 'pulses' for a couple seconds.
- When you add new compounds to your library, you can choose to track those compounds by creating a new Compound Set containing only the newly added compounds.

Related Topics

- "Library Tools"
- "Export Compounds"
- "Compound Sets (Sidebar)"
- "Smart Compound Sets"
- "Rename Compound Sets"
- "Remove Compound Sets"

Smart Compound Sets

In Library Manager, you can create Smart Compound Sets based on rules that choose specific compounds from your library. Smart Compound Sets automatically update to reflect changes in your library. For example, if you add new compounds to your library, they immediately appear in any Smart Compound Sets whose rules they match.

Simple rules might be "*all compounds that I can use to profile 800 MHz spectra*" or "*all compounds with a region-based fit style.*" However, you can also use Smart Compound Sets to select "*all compounds useful for profiling 600 MHz spectra, with a valid pH range from 5.54 to 8.39, with a molecular weight less than 300 g/mol and whose name starts with the letter 'a'.*" You can be as specific or as general as you like. You can also merge results from existing Compound Sets, selecting "*compounds that are in Set A, but not in Set D.*"

Smart Compound Sets can help you focus on just the compounds that you are interested in, without having to worry about updating the sets as you add new compounds to your library or update existing compounds.

Menu Location	File > New Smart Compound Set...
Icon	

How do I create a new Smart Compound Set?

1. Open the New Smart Compound Set dialog.
2. Enter a name for the new Smart Compound Set.
3. Enter the desired magnet frequency for the Smart Compound Set.
4. Use the drop-down menus to define any additional search rules that you would like to apply to the Smart Compound Set. Use the + buttons to add new rules, and the - buttons to remove existing rules.

5. Click OK.

How do I edit a Smart Compound Set?

Menu Location	File > Edit Smart Compound Set...
Icon	

1. Select a Smart Compound Set in the Compound Sets view.
2. Click **Edit Smart Compound Set**.
3. Make any desired changes to the name of the set, magnet frequency restriction, or filters.
4. Click OK.

Tips and Tricks

- You cannot manually add compounds to a Smart Compound Set. Only compounds in your library that match the rules that you define in the Smart Compound Set appear in the set. Use **Edit Smart Compound Set** to modify the compounds appearing in a Smart Compound Set.
- Smart Compound Sets update automatically as the contents of your library change. When you add a new compound to your library, it appears in any Smart Compound Set whose rules it matches, even if you created the Smart Compound Set before you added the new compound.
- If the contents of any Smart Compound Set changes, usually because you have added or removed compounds from your library, the set's icon in the Sidebar 'pulses' for a couple seconds.

Related Topics

- "Library Tools"
- "Compound Sets (Sidebar)"
- "Compound Sets"
- "Rename Compound Sets"
- "Remove Compound Sets"

Rename Compound Sets

You can rename Compound Sets or Smart Compound Sets after you have created them.

How do I rename a compound set?

Menu Location	File > Rename Compound Set...
Icon	

1. Select a compound set in the Compound Sets view.
2. Click **Rename Compound Set**.
3. Enter a new name for the set.
4. Press **Enter**.

Tips and Tricks

- Click on the name of a selected compound set, or right-click on the name of a compound set and select **Rename** to rename the set.
- You can rename a Smart Compound Set via the **Edit Smart Compound Set** dialog.

Related Topics

- "Compound Sets (Sidebar)"
- "Compound Sets"
- "Smart Compound Sets"

Remove Compound Sets

You can remove compound sets when you no longer need them.

Menu Location	File > Remove Compound Set...
Icon	

How do I remove a compound set?

1. Select a compound set in the Compound Sets view.
2. Click **Remove Compound Set**.
3. You will be given the option to permanently remove all compounds in this set from your library.

Menu Location	Library > Remove Compounds > [Compound Set]
Icon	

Tips and Tricks

- Right-click on the name of a compound set and select **Remove** to remove the set.
- You can also click **Remove Compounds > [Compound Set]**. This will remove all compounds in this set from your library AND remove the chosen compound set.

Related Topics

- "Compound Sets (Sidebar)"
- "Compound Sets"
- "Smart Compound Sets"

6.4 Reference Card Editor

Menu Location	Tools > Reference Card Editor...
Icon	

When you are working with a compound, Chenomx NMR Suite will often display a relevant reference card in the "Reference Cards". You can use the Reference Card Editor to create your own custom reference cards, which can be applied to your custom compounds or even used to override the default Chenomx reference cards.

Custom reference cards are saved as .xmc files on your file system and can be opened and saved by Reference Card Editor.

A new folder will be created in on your computer for storing and accessing your custom reference cards. This folder is your Custom Reference Cards Location, and it can be configured in "Preferences".

Any custom reference card that you save in the Custom Reference Cards Location will be automatically loaded by Chenomx NMR Suite.

Required Details

There are four required details that need to be filled out before a reference card can be saved.

Required Details

- *Chenomx Compound ID* needs to be the same as the Chenomx Compound ID assigned to your custom compound in Compound Builder. Note that this is not a required

field when creating a new compound in Compound Builder, so you may need to go back to each custom compound and assign it a Chenomx Compound ID before continuing. You should always assign Chenomx Compound IDs that are higher than 100000.

- *Common Name* is the name of the compound that you wish to see as the header in your custom reference card.
- *Formula* is the molecular formula for your custom compound. This field will take atoms and number of atoms that make up the custom compound. (ie. C2O2H4)
- *Weight* is the molecular weight of the custom compound in Daltons. Note this is used in Profiler to calculate mass concentration units.

Optional Details

There are additional optional details that can be added to your custom reference card.

Optional Details

- *IUPAC Name* is the IUPAC (www.iupac.org) name for the custom compound.
- *CAS Registry* is the unique CAS registry number for your custom compound.
- *InChI String* is the IUPAC International Chemical Identifier for your custom compound.
- *SMILES String* is the Simplified Molecular Input Line Entry System string for your custom compound.
- *Structure Image File* is the file that shows the chemical structure of your custom compound. This file can either be a JPEG, PNG, GIF or SVG image type.

6.5 Information Tools

While organizing compound sets, you can access extended information about compounds in your library.

Reference Card Panel (Sidebar)

Menu Location	View > Sidebars > Left Sidebar > Reference Panel
Icon	

The Reference Card Panel Sidebar view displays information specific to the compound that you have selected in the Compound Table (if available). This includes a structure image, molecular formula and weight, alternate names, InChI and SMILES strings, extended descriptive text and links to external reference sites.

Menu Location	View > Sidebars > Right Sidebar > Reference Panel
Icon	

Related Topics

- "Reference Cards"
- "Reference Card Editor"
- "Preferences"

Chapter 7. Batching and Binning (PRO Only)

7.1 Batching

Batch Import

For larger projects, you may want to import groups of spectra into Chenomx spectra (`.cnx`), processing them with a consistent set of parameters to ensure consistent analysis results.

Menu Location	Tools > Batch Operations > Batch Import...
Icon	

How to batch import spectra?

1. Start the Batch Import wizard.
2. Choose the source spectra to import. The source spectra must be in one of the supported file formats ("Spectral Data Processing")
3. If the source spectra include multiple file types, select the type that you would like to import.
4. Choose the CSI and pH calibration options.
5. Choose the processing layers that you want to apply.
6. Specify a target folder for the imported spectra.

Tips and Tricks

- If the source folder contains more than one type of spectrum file, you need to select one type to import in the current session, since different import options may apply to each available file type. You can do additional sessions to import other file types.
- The import settings available through the Batch Import wizard are the same as those available when you import individual spectra ("Importing Spectra").
- Storing the imported spectra next to the source files means that each new spectrum file appears in the same folder as the spectrum file from which it was created. You may find it more useful to have Processor place the imported spectra into an entirely different folder.

Batch Process

The Batch Process tool allows you to add, remove, and change calibration and processing effects on multiple `.cnx` files at the same time.

Menu Location	Tools > Batch Operations > Batch Process
Icon	

How to add/remove/change processing effects for multiple spectra at once:

1. Click on **Batch Process** in the Tools menu.
2. Use the **Add Files** or **Add Folder** buttons as many times as desired to select one or more `.cnx` files. These are the files that you'll be modifying. Click **Next** to move on to the next page.
3. Choose to which calibrations and processing effects you would like to adjust by moving them from the list on the left to the list on the right. You can do this with the arrow buttons or simply by dragging and dropping them with the mouse. Click **Next** to continue.

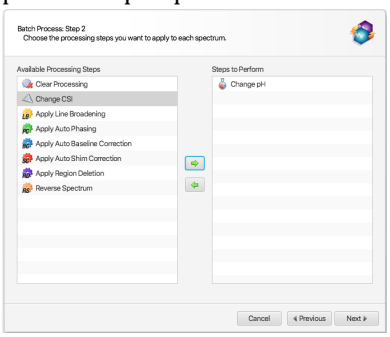
4. You will now see a page of options for each calibration or effect that you have chosen. Adjust the options as you like, clicking **Next** after each.

5. On the final page, you'll see a summary of the changes that you are about to make. Once you are satisfied with them, click **Finish** and your changes will be applied.

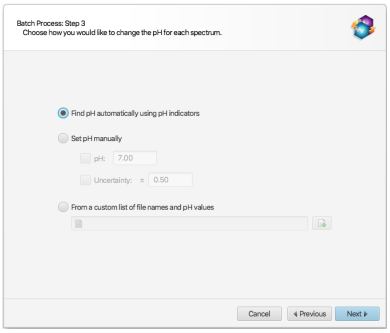
How to change the pH settings for multiple spectra at once:

The pH information of several samples/filenames can be read from an excel workbook and batch applied to a series of spectra.

1. Select Batch Process from the Tools menu. After choosing a set of files, select Change pH as a step to perform in the interface:



2. In the next interface that will appear, choose “From a custom list of file names and pH values”:



The provided excel workbook will be scanned for sample file names and pH information. The program will scan each tab/sheet of the provided excel workbook document (.xlsx) until it finds one that has a single row containing one of the mandatory valid column headers for BOTH the file names and their associated pH values.

The valid column header identifiers for the file names are: “name”, “sample”, or “filename”. The listed file names do not need the “.cnx” extension. If the file names are numerical in nature, and listed without their default “.cnx” extension, for instance: 10, 20,30, the cells must explicitly be formatted as Text inside Excel, not numbers.

The column containing pH information must be labeled “pH”, and nothing else.

Optionally, it is also possible to specify equidistant pH uncertainty ranges (+/-), with one of the following valid column labels: “ΔpH”, “Δ pH”, “+/- pH”, “± pH”, or “pH uncertainty”. If this column is not provided, only the raw pH value of the listed samples will be changed to their listed values.

Once a row with all valid column headers have been found, the rest of the excel sheet will be read from the following row on and the information (filenames and pH values) will be extracted from their corresponding columns.

Tips and Tricks

- The effects and calibrations that you apply with this tool are identical to the ones you can apply manually in Processor. You can read more detailed descriptions of each of them in other sections of this document.
- You can check to see if your changes were applied correctly by opening your any of your newly batch processed spectra in Processor and inspecting its Processing History.

Batch Edit

Batch Edit performs automated profiling operations on one or more compounds in each of a group of Chenomx spectra (.cnx). Remote or add stars, pins, colors, concentration or clsuter locks to compounds, scale compound concentrations, or clear profiled compounds. After selecting an option, choose the compounds that you want to affect. Once completed, you can open the .cnx spectra in Profiler to review the changes that were made.

Menu Location	Tools > Batch Operations > Batch Edit...
Icon	

Star/pin/color a set of compounds in multiple spectra

1. Select Batch Edit from the Tools menu.
2. Select the spectra.
3. Choose to star, pin, color, scale the concentration of, or clear the profiled data of the compounds of interest.
4. Choose a Chenomx library or a compound set that contains the compounds of interest.
5. Select the compounds and click the right-hand arrow.
6. Click finish to add the changes to the spectra selected.

Related Topics

- "Colors/Stars/Pins/Remember Selection"

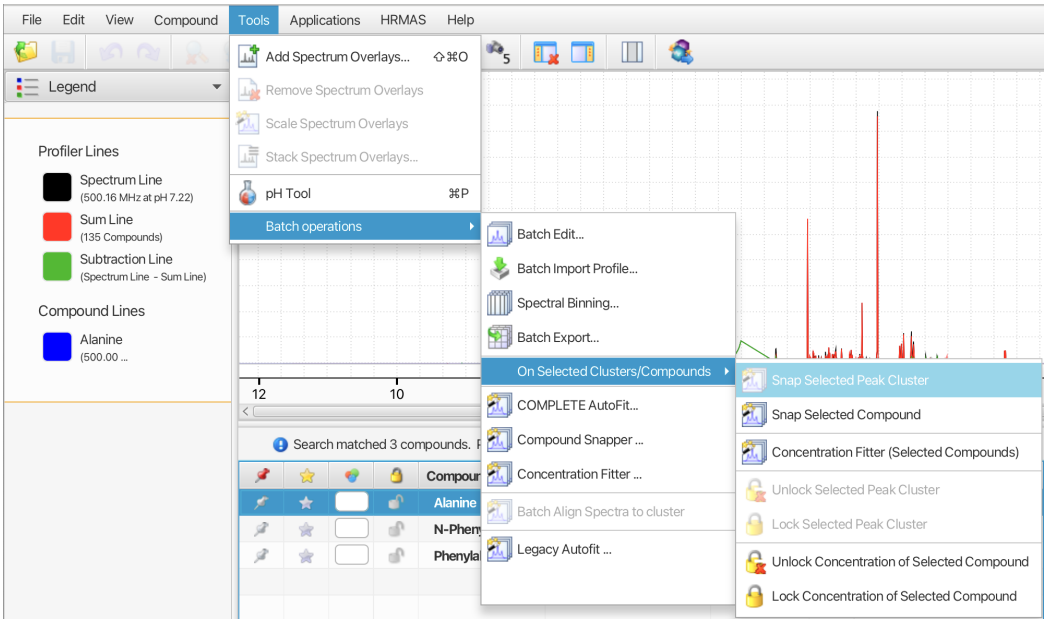
Batch Fit

Batch Fit automatically fits one or more compounds in a group of processed spectra (.cnx). Please refer to Complete Autofit: fully automated profiling approach

Batch Operations on Selected Compounds or Clusters

Most batch operations require the selection of a set of compounds to operate on, which are selected either from a compound library or a profiled spectrum. It is however possible to preselect compounds or clusters to operate and expdite lauch those operations directly from the seleted items.

After selecting a set of compounds from the compound table or clusters from a single selected compound, select **Tools > Batch Operations > On Selected Clusters/Compounds** for a list of potential batch actions.



Batch Spectral Alignment

The software can align a whole series of spectra based on the location of a given peak cluster of a given compound. The location of the cluster choosen to drive the aligment must be know a priori in each spectrum. To specify which cluster should be used to perform the alignment, first select a compound from the compound table and choose a single cluster (out of all of its peak clusters) by clicking on the associated triangle on the ppm axis. Note that at this stage, the selected cluster and compound do no need to be fit. This operation simply tells Profiler which cluster to be used for batch alignment.

Menu Location	Tools > Batch Operations > Batch Align Spectra
Icon	

Align multiple spectra according to the selected cluster

1. Select Batch Operations, Batch Align Spectra from the Tools menu after selecting a single cluster from a single cluster .
2. Select a series of cnx files to align.
3. Click Next.

Spectral alignment will shift the ppm axis in all of the chosen cnx files so that the center of the selected cluster will appear either at a specified location (given in ppm units), or at its "untransformed" locations, i.e. its calibrated location based on pH and in a way that the cluster does not necessitate any transform.

Since spectral alignment shifts the ppm axis in each file, the location of the calibration peak (CSI in Processor) may no longer match its default location. For instance, the methyl peak of DSS may no longer appear at 0 ppm. Any post recalibration of the CSI in Processor will likely shift the CSI to its expected frequency, and nullify the effect of alignment. Therefore, it is highly recommended to make a safety copy of the cnx files prior to using this functionality.

7.2 Spectral Binning

Statistical approaches to analyzing large numbers of spectra often involve data reduction methods like spectral binning to reduce the number of variables that must be considered.

Spectral binning divides spectra into a series of regions, or bins; subsequent analysis involves the integrated area of these bins instead of the raw spectral data. Profiler lets you perform spectral binning on collections of spectra, exporting the results to an excel document (.xlsx) and to tab-delimited text (.tsv) that is easily read by spreadsheet programs and statistical analysis software packages.

Menu Location	Tools > Batch Operations > Spectral Binning...
Icon	

How to bin spectra:

1. Start the Spectral Binning wizard.
2. Choose the source spectra (`.cnx`) for binning, and specify a target folder for the resulting binned data. Specify a series of spectrum files or one or more folders containing spectrum files as the source.

   To repeat a previously saved binning session, specify a binning configuration file (`BinningConfiguration.xml`) to import.

   A binning configuration file is automatically created each time binning results are calculated.
3. Choose a binning method to apply to the specified spectra. Use a defined bin size in ppm or a defined number of bins, or use bins based on the transform windows of the compounds in a Compound Set ("targeted binning").

   If you would like to reuse a predefined set of bins, you can specify a bin definition file (`BinDefinitions.tsv`) to load.
4. Choose regions of the spectrum to exclude from binning, if desired by specifying any regions of the spectrum using start and end values in ppm.
5. Choose a binning target based on the sum line (standard binning) or the subtraction line (residual binning).
6. Choose one of three normalization method:
   a. relative to the total area under each spectrum line.
   b. to units of standardized area (sa). Note that 1 sa is the area under a theoretical DSS methyl peak at 0.50 mM in each spectrum.
   c. None: no normalization are used and area calculted from raw intensities are ouput

Tips and Tricks

- Binning can take a long time, especially on older computers or particularly large datasets. Try a small dataset of five or ten spectra first, before moving on to larger datasets. The time needed for the smaller dataset will help estimate the time needed to complete a binning session.
- If you choose one or more folders as the source, binning includes spectra both directly in the specified folders and in subfolders of the specified folders.
- Binning based on number of bins divides the spectrum into the specified number of bins after excluding the regions that you have defined, if any.
- To do residual binning, each of the source spectra must have been analyzed in Profiler and must contain at least one compound with a defined concentration.
- Excluding regions from a binning dataset can help you to remove the influence of interfering or confounding signals, such as solvent peaks.

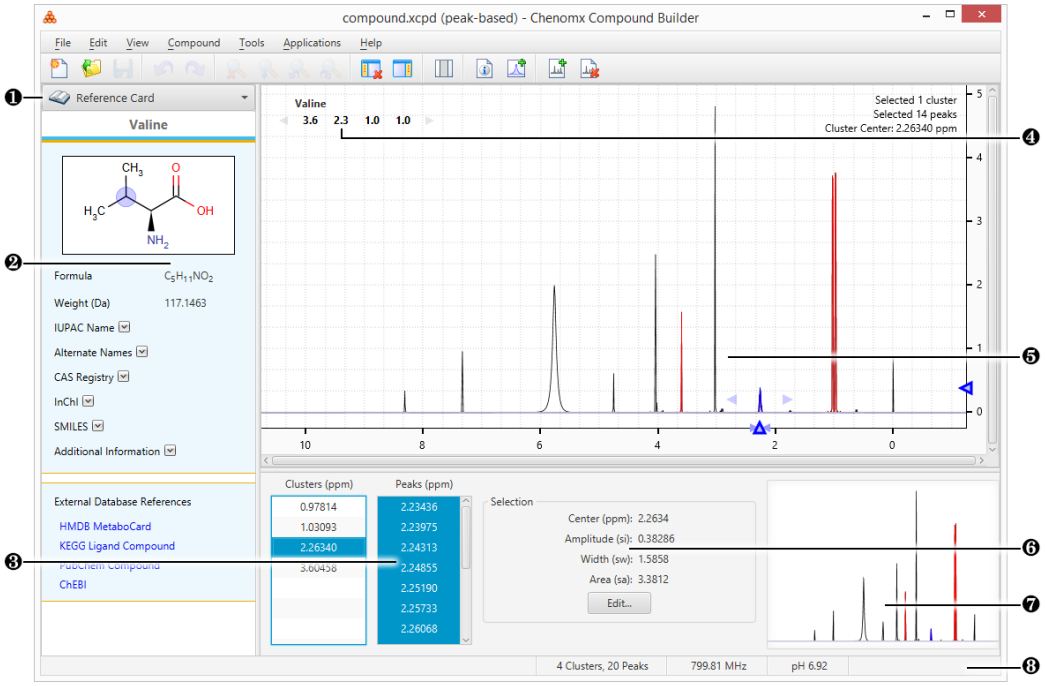
- Normalization based on total area gives the area of a particular bin as a fraction of the total area of all bins, except area in any of the excluded regions that you may have defined.
- Spectral binning is subject to several numerical limits:
    - Each binning session can include a maximum of 5000 spectra.
    - The range that you define when using number of bins or size of bins as a binning method must be at least 1 ppm wide, and can be as large as 1998 ppm (-999 to 999 ppm).
    - You can specify a maximum of 100000 (one hundred thousand) bins or a minimum of 1.
    - You can specify a minimum bin width of 0.00001 ppm and a maximum bin width equal to the width of the range that you have chosen. For example, if you are binning across a range of 2 to 10 ppm, your maximum bin width is 8 ppm.

Chapter 8. Compound Builder (PRO Only)

The Compound Builder module creates compound signatures so that you can identify and quantify your own custom compounds. Add your newly created compound signatures (`.xcpd`) to the compound library using Library Manager. Once in the library, these custom compouds can be used to profile spectra in Profiler.

For a hands-on introduction to Compound Builder, see "Tutorial and Sample Files".

8.1 Overview



❶ Sidebar View Dropdown Menu. Lets you change the currently visible Sidebar view.

❷ Reference Card Panel. Displays additional information for the compound indicated by the Chenomx Compound ID in the Compound Details, including structure, molecular weight, CAS-RN and so on.

❸ Cluster and Peak Lists. Let you edit, select or remove clusters or peaks in the currently open compound. Also lets you define new clusters based on the currently selected peaks or clusters.

❹ Cluster Navigator. Lets you navigate among the clusters of the currently open compound. You can enable or disable the Cluster Navigator using the **View** menu.

❺ Spectrum Graph. Displays the spectrum as well as the sum line and the currently selected cluster(s) or peak(s).

❻ Information Panel. Displays information relating to the currently selected cluster(s) and peak(s).

❼ Spectrum Thumbnail. Displays a thumbnail image of the Spectrum View. The thumbnail can also be used to navigate the spectrum at various zoom settings.

❽ Status Bar. Displays the current cursor position in the units specified by the **View > X Scale** and **View > Y Scale** settings. Also displays information about recent user actions.

8.2 Information Tools

While building compound signatures, you can access extended information about common compounds, view and update details associated with the current compound, and more precisely manage peaks and clusters in complex compounds.

Compound Details

Menu Location	File > Compound Details...
Icon	

Using Compound Details, you can associate information with compound signatures that you build. The information that you enter lets you use the signature with appropriate spectra, display correct information in the Reference Card Panel, calculate accurate concentrations, and manage the contents of your compound library effectively.

Table 8.1. Compound Details

Compound Name	The name of this compound, as you would like it to appear in the Compound Table in Profiler
Compound Author	The author of this compound, as you would like it to appear in the Compound Table in Profiler
Magnet Frequency (MHz)	The frequency of NMR spectra, in megahertz (MHz), that this compound is intended to be used with when profiling.
Fit Style	Specify whether this compound should use a peak-based or region-based fit style when it is profiled in Profiler. For more details, see "Compound Fit Style" .
Sample Concentration (mM)	The concentration of this compound in the sample spectrum that you used as a reference (overlay) when building this compound signature. In millimoles per litre (mM). This value is only used for compounds with the peak-based fit style.
Chenomx Compound ID	Chenomx compound ID associated with this compound
Pulse Sequence	The pulse sequence used to acquire the sample spectrum that you used as a reference (overlay) when building this compound signature
Valid pH	The pH value or range at which you expect this compound signature to be valid
Last Modified	The date and time at which this compound was last modified (and saved).
UUID	The Universally Unique Identifier used to uniquely identify this compound signature in the compound library. Newer and older versions of this compound should maintain the same UUID, but different compounds should always have distinct UUIDs.
Comment	Any text comments associated with this compound signature; you can use this to record any additional information that you consider relevant

Tips and Tricks

- For accurate quantification results, consider verifying the sample concentration that you enter using multiple independent techniques.

- Entering the correct Chenomx Compound ID ensures that information appropriate to the compound appears in the Reference Panel. If your new compound has an ID that isn't recognized by the software, you can use "Reference Card Editor" to create your own reference card.
- Make sure that any new signature that you create has its own distinct UUID. The UUID is used by Chenomx NMR Suite to identify different versions of the same compound, so only different versions of the same compound should share the same UUID.

Related Topics

- "Reference Cards"
- "Build a Compound"
- Chapter 6, *Library Manager*
- "Reference Card Editor"

Information Panel

The Information Panel is always visible at the bottom of the Compound Builder window. It contains current information about the compound signature as you work on it. It lists clusters and peaks that you have created, describes the elements that you have currently selected and the environment on which your compound is based, and lets you access the edit dialog for individual clusters or peaks.

Tips and Tricks

- You can select multiple peaks and clusters in the lists in the Information Panel. Hold down the **Ctrl** or **Cmd** keys while clicking on the desired peaks and clusters. Use **Shift** to select a range of peaks or clusters.
- If you do not have a spectrum overlay, the Information Panel indicates that your compound signature is in the default environment. When you overlay a spectrum, the environment changes to match the overlay.
- The Information Panel displays a warning if your spectrum overlay does not match the information that you provided in the Compound Details. You need to update the Compound Details to match the overlay or choose a new overlay spectrum to remove this warning.

Cluster Navigator

The Cluster Navigator lets you quickly move among the clusters that you have created in the current compound signature. It appears in the top left corner of the Spectrum Graph as an ordered list of the approximate positions of all the clusters of the compound, in ppm. When you click on a cluster position in the navigator, the Spectrum Graph zooms to provide an optimal view of the cluster. You can also use the Cluster Navigator to move sequentially through the list of clusters in either direction.

How do I display the clusters of a compound in the Spectrum View?

Menu Location	View > Show Cluster Navigator

1. Make sure that the Cluster Navigator is visible.
2. Click a cluster position to zoom directly to that cluster. Click the left or right arrow to move sequentially through the list of clusters.

Tips and Tricks

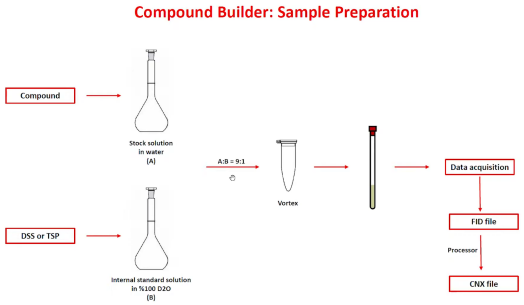
- Clusters that appear green in the Cluster Navigator are pH sensitive, meaning that their heights, positions, and widths all change with spectrum pH.
- Click on a cluster and drag along the Cluster Navigator to zoom to multiple adjacent clusters in the Spectrum Graph.

8.3 Build a Compound

Compound signatures are comprised of clusters, which are in turn comprised of groups of peaks. Compound Builder provides a variety of tools for creating and manipulating these signatures.

First Step: Sample Preparation and NMR Acquisition

To ensure compatibility with both the default Chenomx Reference Library as well as your experiment spectra, acquire your new Compound Signature using careful Sample Preparation and NMR acquisition parameters that will correspond to your Experiment Spectra.



Sample Preparation Steps

1. Prepare a stock solution with a known concentration of your compound
2. Prepare an Internal Standard Solution using either DSS or TSP with 100% D2O
3. Mix Stock Solution with Internal Standard Solution at 9:1 ratio as NMR Instrument requires 10% deuterium to lock.
4. Shake or vortex mixture and adjust pH to 7.0
5. Acquire spectrum by NMR using recommended parameters:
    - Temperature = 25 °C
    - Acquisition time = 4 s
    - Relaxation delay = 1 s
    - Spectral width = 12 ppm

The FID folder acquired from the NMR Instrument can be opened with the Chenomx Processor to create a CNX file. This CNX file can be opened in Compound Builder to create your new Compound Reference Spectrum file (*.XCPD)

Second Step: Process your Spectrum in Processor

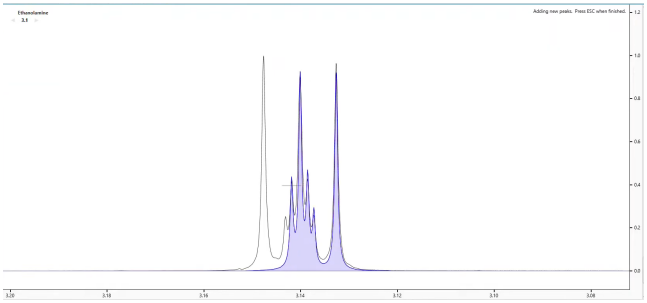
After acquiring your FID file from the NMR Instrument, open and process your Raw Spectrum in the Processor and create a CNX file for compatibility with the Compound Builder. Refer to "Processing Steps" for best practices.

Third Step: Import new CNX file into Compound Builder.

1. Open your processed CNX file reflecting the isolated signals of your new compound.
2. Ensure NMR field strength used to obtain the spectrum matches with the Compound Details Menu.
3. Enter Concentration of compound as it was measured in NMR tube after adding internal standard.
4. Enter Compound Name; pH; Pulse Sequences; UUID as required

Fourth Step: Add Peaks, Define Clusters and Optimize Shapes

Begin Adding Peaks, Defining Clusters and Optimizing Shapes to create your Compound Reference Library Spectrum.



i) Add, Select, and Delete Peaks and Clusters

You can add, select, and delete peaks or clusters by clicking on them in the graph, or in the Information Panel.

Menu Location	Compound > Add Peaks
Icons	

Add peaks to the current signature by clicking on the Compound Spectrum.

1. Switch to Add Peaks mode and the cursor will change to a large crosshair.
2. Click the crosshair within the first Compound Spectrum Cluster to create your first peak. Or... Right-click anywhere in the Spectrum Graph that does not contain a peak and select **Add Peak at x.xx ppm**. The click point defines both the position and the height of the new peak. You are free to change both after you have added the peak.
3. To add additional peaks to the Compound Selected Cluster, hold the **Ctrl** or **Cmd** key while clicking for new peaks.
4. Continue to add peaks until all peaks within the Compound Spectrum have been accounted for. When finished, click **Compound > Stop Adding Peaks**.
5. NOTE: The new peak appears in the peak list in numerical order with the currently displayed peaks, not necessarily at the beginning or end of the list.

Selecting, Deleting Peaks and Clusters

- To Select a Cluster: ensure 'Selection by Cluster' is turned on, then click on the cluster in the Spectrum Graph, or; Click on the Cluster's position in the Information Panel, or; Double Click on the Cluster.
- To Select a Peak: Ensure 'Selection by Cluster' is turned off, then click on the cluster in the Spectrum Graph, or; Click on the Peak's position in the Information Panel.
- To Select Multiple Peaks or Clusters: turn 'Selection by Cluster' on or off, as needed; Hold down the **Ctrl** or **Cmd** key, then click several peaks or clusters, either in the Spectrum Graph, the Information Panel, or both. You can hold down **Shift** instead to select a range of peaks or clusters.
- To Delete Peaks or Clusters: Select the peaks or clusters; Click 'Delete Selected Peaks'.

Additional Notes

- When a cluster is selected, only the peaks associated with that cluster are available to select via the Information Panel.
- When no clusters are selected, no peaks are visible or available to select via the Information Panel.
- If a peak is not visible in the Information Panel, select a different cluster.
- Delete multiple peaks or clusters by selecting them via the Spectrum Graph or the Information Panel.
- Right-click on a selection in the Spectrum Graph or Information Panel and click **Delete Selected Peaks**.

ii) Adjust Peaks

Menu Location	Compound > Delete Selected Peaks
Icon	

In most cases, the initial properties of a peak do not accurately match the spectrum overlay. Modify the height or frequency (center position) of one or more peaks to arrive at a signature that fits the spectrum overlay shape.

Menu Location	Compound > Edit Selected Peaks...
Icon	

Set peak properties to specific values

1. Double-click on a peak or cluster in the Information Panel, Select the peaks or clusters that require adjustment and open the 'Edit Selected Peaks' dialog, or; right-click on a selection in the Spectrum Graph or Information Panel and select **Edit Selected Peaks** NOTE: When you enter specific amplitudes and widths, all of the selected peaks are set to the specified values, regardless of which clusters they are associated with.
2. Enter the desired center (position in ppm), amplitude (height in si) and width (in sw) to apply to all selected peaks or clusters and click OK.

Adjust peak properties

Select the peaks or clusters that you would like to adjust and do any of the following:

- Click and drag the selected peaks or clusters in the Spectrum Graph to adjust their positions and heights. Use the blue triangles on the horizontal and vertical axes for fine control over position and height, respectively.
- Click and drag the blue triangles on either side of the selected peaks and clusters, at approximately half-height, to adjust their widths.

- Use the W and S keys or the up and down arrow keys to adjust the height of the selection up and down; The A and D keys or the left and right arrow keys to adjust left and right; and the Q and E keys to display the next or previous cluster.

Tips and Tricks

- When you adjust the properties for a single peak, you can use mathematical functions of 'ph' instead of constant values to set the center, height and width of the peak. To insert an function expression instead of a single value, right-click in the text box that you would like to change, then click **Expression...**.
- To view or edit an existing expression, click the expression icon (√x) in the text box that you would like to change.

iii) Group Peaks as a Cluster

Especially when building more complex compounds, it may be necessary to group peaks from one or more separate clusters into a single cluster of their own. Grouping peaks into clusters ensures that all of the selected peaks move together when you use the compound signature for profiling, and that they obey the same transform window settings.

How to group peaks as a cluster

Menu Location	Compound > Group Selected Peaks as Cluster
Icon	

1. Right-click on a selection in the Spectrum Graph or Information Panel and select **Group Selected Peaks as Cluster** to group the selected peaks directly.
2. After grouping peaks into a new cluster, you may want to use the Edit Selected Peaks dialog to set them all to a common width.

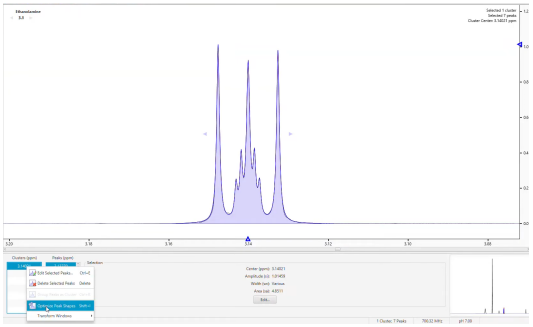
iv) Optimize Selected Peak Shapes

Adding peaks to a signature and adjusting them to approximately model a particular region of the spectrum is fairly simple. However, the task of fine-tuning those peaks to accurately match the lineshape in that region can be tedious. Once you have approximated the shape of a region of the spectrum, use Compound Builder's optimization algorithms to rapidly obtain a more refined match to that region.

Optimize the shape of peaks in a Compound Signature

Menu Location	Tools > Optimize Selected Peak Shapes
Icon	

1. Optimization will generally give better results if your starting shape is reasonably close to the actual shape of the spectrum. In particular, it is important to have the right number of peaks. Simulating the compound in Spin Simulator can often give you a very good starting shape.
2. Select the peaks and clusters (fewer than 50) that you would like to optimize and Click 'Optimize Selected Peak Shapes'.



Fifth Step: Set Transform Windows

Establishing transform windows for your compounds limits the amount that individual clusters can move within the Profiler module and improves the quality of your profiles.

If you set transform windows manually, ensure that they are wide enough to account for the degree of variability in cluster position that you expect to see in your samples. For example, if you routinely analyze spectra with widely varying pH, you will need larger transform windows than if your sample spectra are always close to the same pH.

The most common influences on the position of a cluster in a compound are pH and ion concentrations. To manually set accurate transform windows for a compound, consider acquiring spectra of the pure compound at the outer limits of the conditions you expect to encounter when profiling the compounds in experimental mixtures. This makes it easier to determine maximum extent of these effects on the cluster positions in your compounds.

In the absence of specific information about the behavior of a cluster, you can set a default transform window for the cluster. The default transform window is 0.04 ppm wide, centered on the current cluster center. For a cluster appearing at 1.000 ppm, the default transform window sets lower and upper limits of 0.98 and 1.02 ppm, respectively.

Menu Location	Compound > Transform Window > Edit for Selected Cluster...
Icon	

How to set the transform window for a cluster

1. Select the cluster for which you would like to set a transform window and open the 'Edit Transform Window' Dialog.
2. Select **Limit transforms to**, and enter the new lower and upper limits for the selected cluster. Click OK

How to change the Transform Window for a cluster

1. Select the cluster whose transform window you would like to change and open the 'Edit Transform Window' dialog.
2. Enter the new lower and upper limits for the selected cluster and click OK.

Menu Location	Compound > Transform Window > Set Default on Selected Clusters
Icon	

Menu Location	Compound > Transform Window > Set Default on All Clusters
Icon	

How to set the default Transform Window for cluster(s).

1. Single Cluster: Select the cluster for which you would like to set the transform window and on the Compound Menu, select **Transform Window > Set Default on Selected Cluster**.
2. All Clusters: On the Compound menu, select **Transform Window > Set Default on All Clusters**.

Menu Location	Compound > Transform Window > Clear for Selected Clusters
Icon	

Menu Location	Compound > Transform Window > Clear for All Clusters
Icon	

How to clear the transform window for cluster(s)

- Single Cluster: Select the cluster for which you would like to clear the transform window and on the Compound menu, select **Transform Window > Clear for Selected Clusters**.
- All Clusters: On the Compound menu, select **Transform Window > Clear for All Clusters**.

Menu Location	Tools > pH Sensitivities...
Icon	

How to set pH sensitive Transform Windows

1. On the Tools menu, click **pH Sensitivities** and Click **Load Data Points**.
2. Select the signatures that you would like to use to describe pH sensitive centers and transform windows for the compound.
3. Click **Set Data Points**, then; Click **Optimize Lines**.
4. Review the clusters in the compound. Click **Accept** to apply the automatically generated expressions for cluster centers and transform windows to your compound..

Tips and Tricks

- You should always apply transform windows to any compound that you create. Compounds without transform windows will always appear in the Compound Table in Profiler when performing a Quick Search by region.
- Many of the operations described above for single clusters (setting default transform windows, clearing transform, etc.) can also be performed on multiple selected clusters at once.
- A transform window applies to the center of a cluster, not the positions of its outermost peaks. Consider only cluster centers when you manually set transform windows for your compounds.
- After you have set a transform window for a cluster, if you try to move the cluster beyond either limit of the transform window, the limit moves to accommodate the new position of the cluster. If the transform window was an expression, adjusting it in this manner will replace the expression with a constant.
- You may need to acquire multiple spectra to determine appropriate transform windows for a compound. Remember that some compounds, and some clusters within a compound, are more sensitive than others to pH changes and other matrix effects.
- When you create pH sensitive compounds, expressions (functions of 'pH') for the transform windows are calculated automatically, based on the set of spectra that you select. You can view or edit this expressions by clicking on the expression icon ($\sqrt{x}$) in the text box for the transform.

Related Topics

- "ii) Adjust Peaks"
- "iii) Group Peaks as a Cluster"
- "Cluster Navigator"

8.4 Optional Steps

Compound Fit Style

When creating a compound, adjust **fit style** by using the "Compound Details" dialog. By default all compounds use the *peak-based* fit style, which fit the exact areas and lineshapes of individual compounds while profiling an NMR spectrum. These *peak-based* compounds can be used to quickly and easily deconvolve (break down) overlapping regions in a spectrum and generate accurate profiled concentration values.

Sometimes, however, you may prefer to assign the *region-based* fit style to a newly created compound. Region-based compounds will automatically fit themselves to a spectrum while profiling, ignoring the shape of their own individual peaks and instead fitting the entire region around each of their clusters. This fit style is ideal for compounds whose individual peak shapes are not modelled accurately enough to provide a good fit.

Using the region-based fit style allows you to create new compounds very quickly and without worrying too much about how they are affected by changes to pH, magnet frequency, and other acquisition variables. But it has disadvantages, too.

For example, region-based compounds cannot overlap with other region-based compounds. Also, they are not associated with a sample concentration, so Profiler will never report a concentration value for them. Instead, Profiler calculates the area of a region-based compound by summing up the areas of your spectrum within each of the compound's cluster regions. This total area is proportional to the real underlying concentration of the compound, so it can still be used for various kinds of analysis.

Region-based compounds can also used for simple compound identification.

How to change the fit style of a compound

1. Click **Compound Details**.
2. Choose **region-based fits** or **peak-based fits** on the **General** tab.

Tips and Tricks

- You don't have to worry too much about the exact shape of each cluster in your region-based compounds. Just make sure that the peaks are far enough apart to capture each cluster's entire area.
- The "Generate Cluster for Region" and "iv) Optimize Selected Peak Shapes" features provide a quick and easy way to generate and fit cluster shapes for your region-based compounds.

Related Topics

- "Generate Cluster for Region"
- "iv) Optimize Selected Peak Shapes"

Import Simulation

Importing lets you use an existing spin simulation (`.xss`) or legacy compound album (`.cca`) to guide the construction of a new compound signature.

How to import a spin simulation or compound album

Menu Location	File > Import Simulation...
Icon	

1. Open the Import Compound dialog.
2. Locate the folder containing the simulation or album file that you would like to import.
3. Select the simulation or album file and click **Open**.
4. Choose a calibration method.

   If you are calibrating to a spectrum file (`.cnx`), click the Browse button (...) and locate the spectrum file you would like to use.

   The spectrum that you choose is overlaid in the Spectrum View.
5. Click **OK**.

Tips and Tricks

- Calibrate a signature to a real spectrum for accurate results when using the signature for an analysis. This will ensure that the simulation is run at the same magnet frequency as the calibration spectrum when it is imported.
- Calibrating to the Default Environment instead of a spectrum will run the simulation at 800 MHz while importing it. It will have a pH of 7.00, and CSI settings corresponding to DSS at 0.500 mM (height of 1 si, width of 1 sw).
- The magnet frequency and spectrum pH for the current spectrum overlay appear in the status bar at the bottom of the Compound Builder window. If there is no spectrum overlay, the displayed values correspond to the default environment.

Related Topics

- "Spectrum Overlays"
- Chapter 9, *Spin Simulator*
- "Compound Details"

Generate Cluster for Region

You may wish to build a compound signature that will let you verify that the compound is present in a spectrum, without necessarily providing the full range of flexibility that a chemically accurate signature affords. You can quickly create these limited-use signatures using automatically generated clusters. Generated clusters lack the chemical accuracy of signatures that are backed by spin simulations, but provide a quick model of the lineshape of a compound under specific conditions.

How do I generate a cluster to match a region of the spectrum?

Menu Location	Tools > Generate Cluster for Region
Icon	

1. Select the region of the spectrum that you would like to match with a generated cluster.
2. Click **Generate Cluster for Region**.

Tips and Tricks

- Unlike Spin Simulated clusters, generated clusters have no formal theoretical basis. They simply contain a number of Lorentzian peaks that approximate the overall shape of part of the overlaid source spectrum. These generated peaks will *not* let you accurately account for significant changes in the spectrum based on magnet frequency, pH or other systematic variations.
- You should only expect a compound with generated clusters to fit accurately if you are profiling spectra that have been acquired under conditions very similar to the compound's source spectrum. This restriction makes compounds with generated clusters ideal candidates for the *region-based* fit style (see "Compound Fit Style" .)
- Generated clusters are already optimized. Using the Optimize Selected Peak Shapes feature on them will not give any better results.
- When you generate a cluster, Compound Builder will only use up to 100 peaks to match the selected region. If the region you are trying to match cannot be accurately modeled using 100 peaks, try selecting a smaller region, adding more peaks manually, or simulating the compound in Spin Simulator and importing the simulation.

Related Topics

- "Select Region"
- "Import Simulation"
- "i) Add, Select, and Delete Peaks and Clusters"
- "ii) Adjust Peaks"
- "iv) Optimize Selected Peak Shapes"

New Compound

Menu Location	File > New Compound...
Icon	

The **New Compound** feature creates a new compound file that contains the size, position, and shape of the clusters and peaks that make up a single compound's NMR signature. The new compound starts with default "Compound Details" , including a magnet frequency of 800 MHz and a sample concentration of 5 mM.

Tips and Tricks

- When you open or create a compound, it does not have a spectrum overlay by default. Add a spectrum overlay before you begin building the compound's signature.
- You can overlay one spectrum at a time in Compound Builder.
- You need to save the new compound (as an `.xcpd` file) to retain changes that you have made.
- Spectrum overlays are not retained when a compound is saved.
- If the compound that you've created is not always going to be a good fit when it is used to profile real NMR signatures, you might want to switch your compound's **Fit Style** from from *peak-based* to *region-based* (see "Compound Fit Style").

Related Topics

- "Spectrum Overlays"
- "Compound Fit Style"
- "Compound Details"

Manage Cluster IDs

Clusters have ID numbers that are normally not visible to the user. But if you create a new versions of an existing compound or multiple compound signatures for the same compound (for example, if you are going to make a pH sensitive compound) then it is important to make sure that the cluster ID for each cluster is the same in every compound signature.

How do I change the cluster IDs in a signature?

Menu Location	Tools > Manage Cluster IDs...
Icon	

1. On the Tools menu, click **Manage Cluster IDs...**.
2. Click and drag the clusters in the list to change their order.

   Or

   Select a cluster in the list and use the green up and down arrows to change the position of the cluster in the list.
3. Click OK.

Tips and Tricks

- Cluster IDs are used by the pH sensitivities dialog to determine which clusters should be considered as corresponding to the same signal across all of your selected data points. Make sure that the clusters appear in the proper order in each of your data points before applying pH sensitivities.
- A cluster that appears upfield from another cluster at one pH may appear downfield from the same cluster at a different pH. Consider overlaying all of your data point spectra (`.cnx`) together in Processor or Profiler to more easily see the behavior of clusters across the full pH range.
- If you have already used a compound to profile spectra, replacing that compound in your library with a new version that has different cluster IDs will make your existing fits of that compound invalid.

Related Topics

- "pH Sensitivities"

pH Sensitivities

You can make compound signatures that you create in Compound Builder sensitive to the pH of the spectrum that you are profiling. When used in Profiler, the clusters of pH sensitive compounds will automatically adapt their centers and transform windows based on the profiled spectrum's pH, ensuring that they start closer to their correct position and simplifying compound identification in complex regions of the spectrum.

Menu Location	Tools > pH Sensitivities...
Icon	

How do I make a compound signature pH sensitive?

1. Make sure that you have a set of compound signatures (`.xcpd` files) available touse as data points when calculating pH curves. You will create these signatures using Compound Builder. They must satisfy the following criteria:
   - All of the compound signatures must have the same number of clusters, in the same order.
   - Each signature must have its valid pH set to the pH value from the signature's originating sample, via the Compound Details dialog. Use a single value instead of a range.
2. Select a signature to use as a 'master' signature. This signature will become the pH sensitive compound signature, and therefore should be very well fit to its associated spectrum file (`.cnx`). The remaining signatures must have accurate cluster locations, but otherwise do not need to be perfectly fit to their associated spectra.
3. Open the master signature file (`.xcpd`) in Compound Builder, and overlay its associated spectrum file (`.cnx`).
4. On the Tools menu, click **pH Sensitivities...**.
5. Click **Load Data Points...** and choose the your compound signatures.
6. Click **Optimize Lines**.
7. Review the resulting pH curves for each cluster by clicking the various tabs at the top of the pH Sensitivities dialog.
8. Click **Accept** to apply the automatically generated cluster center and transform window expressions to the clusters in your master signature, and set the valid pH

of the master signature to the range defined by the pH values of your individual data points.

Tips and Tricks

- Clusters that appear green in the Cluster Navigator are pH sensitive, meaning that their heights, positions, and widths all change with spectrum pH. If a compound signatures's cluster centers and transforms were set with the pH Sensitivities dialog, all of its clusters should be green.
- If you manually adjust any cluster after applying pH sensitivities to the compound, some or all of its automatically generated pH sensitivity expressions will be replaced with constant values. These clusters are no longer entirely pH sensitive, and will not appear green in the Cluster Navigator.
- If the clusters in your set of signatures appear in different orders in some of the files, use Manage Cluster IDs to rearrange the cluster IDs in the signatures so that they match.
- If the signatures you use as data points are at a different spectrometer frequency than your master signature, you may need to apply an offset to ensure accurate pH curves. Click **Lock to Current Clusters** in the pH Sensitivities dialog to calculate the offset before reviewing and accepting the pH curves.

Related Topics

- "ii) Adjust Peaks"
- "Fifth Step: Set Transform Windows"
- "Manage Cluster IDs"
- "Compound Details"

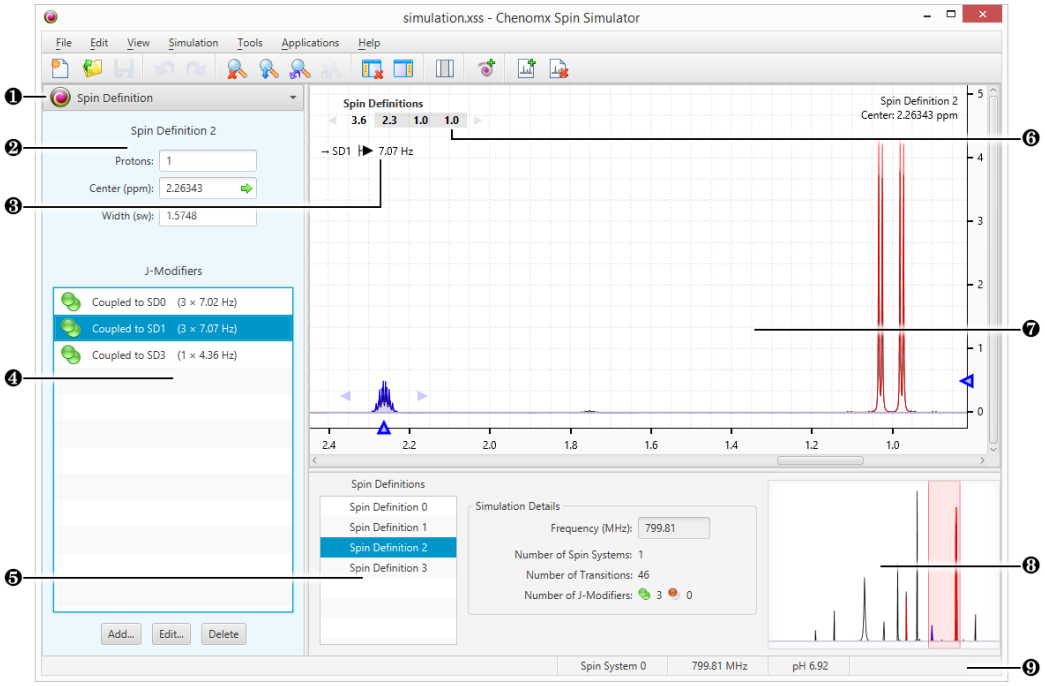
Chapter 9. Spin Simulator

Spin Simulator is a powerful tool for creating simulations of NMR spectra, based on user-defined spin systems and reference spectra. You can use these simulations as starting points for creating your own compound signatures, especially if you need to create signatures at multiple magnet frequencies, or you want to create robust signatures with a strong theoretical backing.

To use Spin Simulator effectively, you'll need a basic understanding of NMR theory, and you should know how to interpret NMR spectra, especially J-coupling. If necessary, Chenomx experts can also provide compound fitting services and help develop specialized compound libraries for you.

For a hands-on introduction to Spin Simulator, see "Tutorial and Sample Files".

9.1 Overview



❶ Sidebar View Dropdown Menu. Lets you change the currently visible Sidebar view.
❷ Spin Definition. Sidebar view that displays the currently selection Spin Definition.
❸ J-Value Editor. Lets you vary the J-value for the currently selected J-modifier.
❹ J-Modifier List. Lets you add, select, edit or remove J-modifiers associated with the currently selected spin definition.
❺ Spin Definition List and Simulation Details. Facilitates selection of spin definitions and spin systems. Also displays information about the current simulation, including the simulation frequency and numbers of spin systems, transitions, and J-modifiers represented in the simulation.
❻ Spin Navigator. Lets you navigate among the spin definitions of the currently open simulation. You can enable or disable the Spin Navigator using the **View** menu.

❼ Spectrum Graph. Displays the spectrum overlay as well as the simulation line and the currently selected spin definitions.
❽ Spectrum Thumbnail. Displays a thumbnail image of the current spectrum. You can also use the thumbnail to navigate the Spectrum View at various zoom settings.
❾ Status Bar. Displays the current cursor position in the units specified by the **View > X Scale** and **View > Y Scale** settings. Also displays information about recent user actions.

9.2 Simulating Tools

Spin Simulator provides you with a variety of tools to help you create accurate spin simulations by adding, manipulating and removing spin definitions and coupling constants, grouping spin definitions into spin systems, and viewing summary details about your current simulation.

Spin simulation is a computationally intensive process, and takes more time as the simulation you are creating becomes more complex. Generally, the amount of time required to recalculate a simulation increases more or less linearly with the number of spin definitions (protons) in the simulation, and exponentially with the number of distinct J-couplings. Spin Simulator monitors the overall complexity of your simulation as you work, and if you attempt to take an action that would result in unusable simulation times, the action is cancelled, and you see a warning dialog indicating that the resulting simulation would be too complex.

New Simulation

Menu Location	File > New Simulation...
Icon	

New Simulation creates a new spin simulation with the default simulation frequency of 800 MHz.

Tips and Tricks

- The new simulation does not have a spectrum overlay by default. Add a spectrum overlay before you continue editing the simulation.
- You can overlay only one spectrum at a time in Spin Simulator.
- You need to save your simulation to an `.xss` file retain the changes that you have made.
- Spectrum overlays are not retained in the saved files.

Related Topics

- "Spectrum Overlays"
- "Simulation Details"

Spin Systems

If you need to model distinct structures in a single simulation, such as different enantiomers in solution, you may want to use spin systems to organize them. A spin system in Spin Simulator is a set of spin definitions that you have defined as only interacting with each other. These spin systems can have their own abundance in solution.

How do I add a new spin system?

Menu Location	Simulation > Add New Spin System...
Icon	

1. Click **Add New Spin System**.
2. If this is the first spin system you have added, a new dropdown list appears above the Spin Definition list, showing the available spin systems. Additional spin systems also appear in this dropdown list.

How do I select a different spin system?

1. In the Spin Definition list, select the spin system to which you would like to switch from the drop-down list.

   Alternately, click on a spin definition belonging to the other spin system in the Spectrum Graph.

How do I delete a spin system?

Menu Location	Simulation > Delete Current Spin System...
Icon	

1. Select a spin system from the drop-down list above the spin definition list.
2. Click **Delete Current Spin System**.

Tips and Tricks

- Deleting a spin system deletes all spin definitions and J-modifiers associated with the spin system as well.

Related Topics

- "Spin Definitions"
- "Spin Navigator"
- "Spin Definition (Sidebar)"
- "Simulation Details"

Spin Definitions

Spin Simulator creates computer models of the interactions among the members of a spin system. Within Spin Simulator, you may consider a member of a spin system as equivalent to a single proton, or a group of protons that are both chemical shift equivalent and magnetic-equivalent. Spin definitons describe the members of spin systems, and are the basic building blocks of the models created by Spin Simulator.

A spin definition consists of three basic pieces of information: The number of protons represented by the spin definition, the center of the spin definition in ppm (equivalent to a chemical shift in an NMR spectrum), and a width in sw (analogous to a linewidth at half-height in an NMR spectrum).

You can organize spin definitions into spin systems, as described elsewhere in this chapter (see "Spin Systems").

How do I add a new spin definition?

Menu Location	Simulation > Add Spin Definitions
Icon	

1. Click the Add Spin Definitions button on the toolbar, or from the menu under Simulation > Add Spin Definitions.
2. Click on the screen the ppm location where you want the new spin definition. A single proton spin definition will be added to the simulation. You can change the number of protons and add J-Modifiers later.
3. Click on the Stop Adding Spin Definitions button on the toolbar (or press ESC) to leave spin definition addition mode.

How do I edit a spin definition?

1. Select the spin definition by clicking on it in the Spectrum Graph, or by selecting it in the Spin Definition list in the Simulation Details panel.
2. Do any of the following:

- Click and drag the blue triangle on the horizontal axis to change the center of the spin definition.
- Click and drag the blue triangle on the vertical axis to change the height of the simulation.
- Click and drag the spin definition directly in the Spectrum View to change its center and the simulation height simultaneously.
- Click and drag the blue triangles on either edge of the spin definition (at around half the height of the tallest peak) to change the width of the spin definition.
- Use the W and S keys or the up and down arrow keys to adjust the height of the whole simulation up and down.
- Use the A and D keys or the left and right arrow keys to adjust the position of the selected spin definitions left and right.
- Use the Q and E keys to display the next or previous spin definition.

How do I reassign a spin definition from one spin system to another?

Menu Location	Simulation > Reassign Selected Spin Definitions...
Icon	

1. Select a spin definition in the Spectrum Graph or by selecting it in the Spin Definition list in the information panel.
2. Click **Reassign Selected Spin Definitions...**.
3. Select either a target spin system, or select a new spin system.
4. All spin definitions coupled to the current selected spin definitions will also be reassigned. Uncheck the check box if you want to uncouple the selected spin definitions and move only the selected spin definitions instead.

How do I delete a spin definition?

Menu Location	Simulation > Delete Selected Spin Definition
Icon	

1. Select a spin definition in the Spectrum Graph or by selecting it in the Spin Definition list in the information panel.
2. Click **Delete Selected Spin Definition**.

Tips and Tricks

- A spin definition can describe no more than ten protons. However, given that a spin definition typically describes chemically and magnetically equivalent protons, a value greater than three (a methyl group) is often not necessary.
- A spin system can contain any number of protons and spin definitions, as long as the total number of transitions is less than 2000. If you need to create a simulation requiring more than 2000 transitions, you will need to use multiple spin systems.

  The total number of transitions in the current simulation appears in the simulation details (see "Simulation Details").
- Height adjustments always affect the whole spin system, not just the currently selected spin definition. If you need to adjust the heights more than is possible in Spin Simulator, finish the simulation, save it and import it in Compound Builder, then make your adjustments there.
- Deleting a spin definition also deletes any J-modifiers that you have associated with that spin definition.

Related Topics

- "Spin Systems"
- "Spin Navigator"
- "J-Modifiers"
- "Spin Definition (Sidebar)"
- "Simulation Details"
- "Import Simulation"

Spin Navigator

The Spin Navigator lets you quickly move among the spin definitions that you have created in the current simulation. It appears in the top left corner of the Spectrum Graph as an ordered list of the approximate positions of all the spin definitions of the simulation, in ppm. When you click on a spin definition's position, the view zooms to provide an optimal view of the spin definition. You can also use the Spin Navigator to move sequentially through the list of spin definitions in either direction.

How do I display the spin definitions of a simulation in the Spectrum Graph?

Menu Location	View > Show Spin Navigator

1. Make sure that the Spin Navigator is visible.
2. Click a spin definition's position to zoom directly to that spin definition. Click the left or right arrow to move sequentially through the list of spin definitions.

Tips and Tricks

- Click on a spin definition and drag along the Spin Navigator to include adjacent spin definitions in the Spectrum Graph.
- Use the Q and E keys to display the next or previous cluster.

Related Topics

- "Spin Systems"
- "Spin Definitions"
- "Spin Definition (Sidebar)"
- "Simulation Details"

J-Modifiers

When you create a new spin definition, it appears on the Spectrum View as a single, unsplit peak. J-modifiers let you split a spin definition into more complex shapes, to fit the shape of real spectra. J-modifiers can be uncoupled, affecting only one spin definition, or coupled, linking the spin definition to another one via its coupling constant. Coupled J-modifiers result in more accurate simulations, as Spin Simulator can use them to calculate higher-order coupling effects, accounting for tenting and other complex behaviour.

How do I add an uncoupled J-modifier to a spin definition?

Menu Location	Compound > Add New J-Modifier...
Icon	

1. Select the spin definition to which you would like to add a J-modifier.
2. Open the **Add New J-Modifier...** dialog.
3. Enter a J-value for the new J-modifier, in Hz.

4. Select Uncoupled Split, and specify a split count for the new J-modifier.
5. Click OK.

How do I add a coupled J-modifier to a spin definition?

1. Select one of the two spin definitions that you would like to link with a coupled J-modifier.
2. Open the **Add New J-Modifier...** dialog.
3. Enter a J-value for the new J-modifier, in Hz.
4. Select Coupled, and select the spin definition to which you would like to link.
5. Click OK.

How do I edit an existing J-modifier?

Menu Location	Compound > Edit Selected J-Modifier...
Icon	

1. Select a spin definition.
2. Select a J-modifier that you would like to edit.
3. Open the **Edit Selected J-Modifier...** dialog.
4. Make your desired changes.
5. Click OK.

How do I couple two spin definitions using splits that I have already defined?

1. Select one of the two spin definitions that you would like to link, and select the uncoupled split that will link it to the other spin definition.
2. Open the **Edit Selected J-Modifier...** dialog.
3. Select Coupled, and select the spin definition to which you would like to link.
4. Select the uncoupled split from the second spin definition that you would like to link to.
5. Click OK.

How do I delete a J-modifier?

Menu Location	Compound > Delete Selected J-Modifier
Icon	

1. Select a spin definition.
2. Select a J-modifier that you would like to delete.
3. Click **Delete Selected J-Modifier**.

How do I uncouple a J-modifier?

Menu Location	Compound > Uncouple
Icon	

1. Select a spin definition.
2. Select a J-modifier that you would like to uncouple.
3. Click **Uncouple**.

Menu Location	Compound > Ungroup Selected Splits
Icon	

Menu Location	Compound > Group Selected Splits
Icon	

How do I ungroup J-modifiers?

1. Select a spin definition.
2. Select an uncoupled split with a split count greater than 1.
3. Click **Ungroup Selected Splits.**.

How do I group several J-modifiers?

1. Select a spin definition.
2. Select several uncoupled splits that you would like to combine. Hold down **Ctrl** or **Cmd** to multi-select J-modifiers.
3. Click **Group Selected Splits.**.

Tips and Tricks

- J-Modifiers can only link two spin definitions that are in the same spin system.
- When coupling two spin definitions, uncoupled splits are only shown if the degree of splitting in the second spin definition makes theoretical sense according to the number of protons in the first spin definition.
- When you delete a coupled J-modifier, you will delete it from both spin definitions to which it applies. If you only want to delete the J-modifier from one spin definition, uncouple it first.
- Uncoupling a coupled J-modifier will result in an uncoupled split on each of the two previously coupled spin definitions, each split having the same J-value as the original coupled J-value.
- Most operations involving J-modifiers are available by right-clicking on a J-modifier in the Simulation view.

Related Topics

- "Spin Systems"
- "Spin Definitions"
- "Spin Definition (Sidebar)"

9.3 Information Tools

While creating spin simulations, you can view and update spin systems, spin definitions and J-modifiers associated with the current compound, and view overall details on the current simulation.

Spin Definition (Sidebar)

The Spin Definition Sidebar view shows the currently selected spin definition which allows you to edit the properties, and create, edit or remove J-modifiers.

Menu Location	View > Sidebars > Left Sidebar > Spin Definition
Icon	

Menu Location	View > Sidebars > Right Sidebar > Spin Definition
Icon	

How do I edit a spin definition and J-modifiers using the Spin Definition view?

1. Select a spin definition by clicking it in the Spectrum Graph, or by selecting it in the Spin Definition list.
2. Click in any of the text fields and enter a new value to edit the number of protons, center or width of the spin definition.
3. Edit J-modifiers by doing any of the following:
   - Click **Add...** to add a new J-modifier to the current spin definition.
   - Click **Edit...** to edit the selected J-modifier in the current spin definition.
   - Click **Delete** to delete the selected J-modifier in the current spin definition.

Tips and Tricks

- Double-click on a J-modifier to change its coupling constant and coupling status, including which spin definition the current one is coupled to, if any.

Related Topics

- "Spin Systems"
- "Spin Definitions"
- "J-Modifiers"

Simulation Details

The bottom center pane in the Spin Simulator window displays the Spin Definition list and simulation details, i.e. information about the current simulation. Simulation details include the simulation frequency, numbers of spin systems and spin definitions (spins), and numbers of transitions present in the current simulation.

Tips and Tricks

- When you overlay a spectrum, the simulation frequency is set to match that of the spectrum overlay.
- When you do not have a spectrum overlay, you can change the simulation frequency in the simulation details to explore the effects of spectrometer frequency on your simulated compound.

Related Topics

- "Spectrum Overlays"
- "Spin Systems"
- "Spin Definitions"

Chapter 10. The Basics

This chapter covers basic software operations including: file operations, navigation, display options and preferences that directly affect your day-to-day use of Chenomx NMR Suite.

10.1 File Management

Open Files

You can open Chenomx NMR Suite spectra files (`.cnx`) in Processor and Profiler, and you can use them as overlays in Processor, Profiler, Compound Builder and Spin Simulator. You can open Chenomx compound files (`.xcpd`) in Compound Builder, and you can open Chenomx spin simulation files (`.xss`) in Spin Simulator, or import them in Compound Builder.

Tips and Tricks

- When you open the next or previous file, the zoom settings, selected compound and selected compound sets, where applicable, are preserved.
- You can also open files using the Files Sidebar Panel (see "Files"). This is especially useful for skimming through sets of spectra.
- If you want to open a Agilent spectrum named `Example.fid`, the raw data is stored in the file named `Example.fid\fid`, while the processed data is stored in the file named `Example.fid\datdir\phasefile`.
- If you want to open a Bruker spectrum named `Example\1`, the raw data is stored in the file named `Example\1\fid`, while the processed data is stored in the file named `Example\1\pdata\1\1r`.

Importing Spectra

When you import a spectrum from a supported format (see "Spectral Data Processing") in Processor or Profiler, you can choose from a number of import options.

Table 10.1. Import NMR Data Options

Sample contains a Chemical Shape Indicator (CSI)	Indicate whether a Chemical Shape Indicator (CSI) is present in the sample. Specify the CSI used, and enter its concentration in millimoles per litre (mM). You can choose DSS, TSP, Formate, or a Custom CSI (see "Define Custom CSI").
Sample contains one or more pH Indicators	Indicate whether any pH indicators are present in the sample. If they are, Processor will attempt to automatically detect the pH of the sample, subject to the specified uncertainty, using signals from imidazole, creatinine and DFTMP. You must have at least one of these pH indicators present in the sample to attempt automatic pH detection. If the pH is not detected, it will be set to 7.00. You can change the pH manually after the spectrum is imported.

Phase Correction	Automatically apply phase adjustments to your spectrum. When importing Agilent raw data (`fid`), you are given two options: **Auto** applies automatically-determined phase angles, while **Import from VNMR** imports the phase angles found in the VNMR `procpar` file.
Automatic Baseline Correction	Automatically apply baseline correction to your spectrum. **Linear (Drift)** applies a simple two-point linear baseline correction, while **Spline** applies a cubic spline baseline correction based on automatically-determined breakpoints.
Automatic Shim Correction	Automatically apply shim correction to your spectrum. You can set a target linewidth in the **Linewidth (Hz)** field.
Reverse Spectrum (Bruker only)	Reverse your spectrum horizontally. Only available when you import Bruker raw spectrum files (`fid`).
Line Broadening (Hz)	Apply an exponential FID weighting (line broadening) function to your data. Enter a positive value in Hz.
Delete Water Region	Automatically remove the water region from the spectrum during the import process.
Zero Fill To (FIDs only)	Enable zero-filling. **Automatic** zero-fills to the nearest power of two that is at least twice as large as the number of points acquired in your spectrum. This is normally the correct choice. You can also choose a specific number of data points, if you prefer. This option is only available when you import raw spectrum data (e.g. `fid` files).
Defaults	Restore all import settings for this type of imported spectrum to their default values.

Saving Files

Tips and Tricks

- Saving compound files in Compound Builder or simulation files in Spin Simulator preserves only the mathematical peak descriptions created during the fitting process. Compound Builder and Spin Simulator do not retain spectrum overlays in the saved files.
- In the event that Chenomx NMR Suite stops running unexpectedly, (due to, for example, a power outage or system crash) the next time you run the software you may be given the option to restore your unsaved data. Restoring your data is not always possible, however, so it is still highly recommended that you save your work frequently.

Closing Files

Tips and Tricks

- You cannot close a file that has unsaved changes without having the option of saving the changes. If there are no unsaved changes, the file is simply closed without further interaction.

Undo and Redo

You can undo recent changes to the currently open file, and redo recently undone changes.

Menu Location	Edit > Undo
Icon	

Tips and Tricks

- You can only redo changes that you have just undone. If you undo a change, then make other changes, you can no longer redo the change that was undone.

Menu Location	Edit > Redo
Icon	

Menu Location	File > Preferences
Icon	

10.2 Preferences

In Preferences, you can specify default folders, default settings for the Spectrum Graph, colors of various graph elements and some module-specific settings.

How do I turn off animated zooming?

1. Switch to the General module in Preferences.
2. Clear the check box labeled **Enable Zoom Animation**.

How do I change colors?

1. Click the swatch box corresponding to the color you would like to change and select
2. Click **OK** to accept the selected color.

How do I set the initial zoom?

1. Select a module tab.
2. Select the **Initial Zoom** check box.
3. In the Initial Zoom fields, enter the default values (in ppm) for the range of the spectrum to display.

How do I specify the default file folder?

1. Select a module tab.
2. Click the **Always use this folder** button.
3. Click the default folder browse button (**...**) and select desired folder.

Tips and Tricks

- When you click on a color swatch to change a color, the simple color chooser that appears initially lets you choose from a selection of 15 common, well-distinguished colors. Colors in this selection that are already used in the current module's preferences are indicated with a faint white checkmark, while the currently selected color is indicated with a strong white checkmark.
- The low and high frequencies for the initial zoom must be at least 1 ppm apart.
- The **Advanced Preferences** button allows you to specific additional preferences and special preference overrides.

Related Topics

- "Spectrum Graph"

Table 10.2. Preferences Overview

General Preferences	
Application Updates	Lets you check for software updates, and toggle whether the software will check for updates automatically.
Zooming Preferences	Lets you change the color of the zoom box, and toggle the zooming animations used when you change zoom settings in the Spectrum Graph.
Reset Welcome Dialogs	Turns on initial welcome dialogs for all modules, in case you want to see them again after choosing **Don't show this message again**.
Graph Settings	
Show Spectrum Line	Toggles the default display of the spectrum line in the Spectrum Graph in Processor and Profiler, and lets you set its default color.
Show Sum Line	Toggles the default display of the sum line in the Spectrum Graph in Profiler, and lets you set its default color.
Show Compound Line	Toggles the default display of the compound line in the Spectrum Graph in Compound Builder, and lets you set its default color.
Show Simulation Line	Toggles the default display of the simulation line in the Spectrum Graph in Spin Simulator, and lets you set its default color.
Show Subtraction Line	Toggles the default display of the subtraction line in the Spectrum Graph in every module except Library Manager, and lets you set its default color.
Selection Line	Changes the default color of the selection line in the Spectrum Graph in Profiler, Compound Builder and Spin Simulator.
Overlay Lines	Changes the default color of spectrum overlay lines in the Spectrum Graph in Profiler, Processor, Compound Builder and Spin Simulator.
CSI Line	Changes the default color of the CSI line in the Calibrate CSI Spectrum Graph in Processor.
Processing Tools	Changes the default color of the processing tools in Processor, including the baseline correction breakpoints, shim correction peak, adjustment triangles, etc.
Initial Zoom (ppm)	Sets the frequency range of the Spectrum Graph that initially appears when you open a file.
File Preferences	
Display Recent Files	Sets the number of files displayed when you click **File**, then **Open Recent**.
Default File Folder	Sets the default folder for saving and opening files. Options are to use the last used folder, or always use the specified folder.
Library Manager	

Custom Reference Cards Location	Sets the folder where you can save Custom Reference Cards. The software will load these cards to use with your custom compounds, or override default cards for compounds.

Change Columns

In modules that have Compound Tables (Profiler and Library Manager), you can specify which of the available columns are visible in the Compound Table.

Menu Location	View > Change Columns...
Icon	

How do I change the visible columns in the Compound Table?

1. Open the Change Columns dialog.
2. Select the check boxes for the columns you would like to display, and clear the check boxes for the columns you would like to hide.
3. Click OK.

Tips and Tricks

- You can quickly display or hide a particular column by right-clicking in the header row of the Compound Table and clicking the column that you would like to display or hide.

Display Options

You can customize how modules in Chenomx NMR Suite display information by toggling the display of graph elements in the Spectrum Graph, or by changing the units used for display. You can find all of the following options in the **View** menu.

Table 10.3. View Menu Options

Concentration Units	Changes the units that compound concentrations are represented in. Available units are mM, µM and mg/dL.
X Scale	Changes the units represented on the horizontal scale in the Spectrum Graph. Available units are ppm and Hz.
Y Scale	Changes the units represented on the vertical scale in the Spectrum Graph. Available units are si (standardized intensity) and ni (normalized intensity).
Sidebar > Compound Sets	Switches the Sidebar to the Compound Sets view. Available in Profiler and Library Manager.
Sidebar > Legend	Switches the Sidebar to the Legend view. Available in Processor, Profiler, Spin Simulator and Compound Builder.
Sidebar > Processing History	Switches the Sidebar to the Processing History view. Available in Processor.
Sidebar > Reference Card	Switches the Sidebar to the Reference Card Panel view. Available in Profiler, Compound Builder and Library Manager.
Sidebar > Simulation	Switches the Sidebar to the Simulation view. Available in Spin Simulator.
Show Left Sidebar	Toggles the display of the Left Sidebar.

Show Right Sidebar	Toggles the display of the Right Sidebar.
Show Spectrum Thumbnail	Toggles the display of the spectrum thumbnail in modules with a Spectrum View.
Show Cluster Navigator	Toggles the display of the cluster navigator in Profiler and Compound Builder.
Show Spin Navigator	Toggles the display of the spin navigator in Spin Simulator.
Show J-Value Editor	Toggles the display of the J-Value editor in Spin Simulator.
Show Grid Lines	Toggles the display of the grid lines on the Spectra Graph.
Show Stack Slider	Toggles the display of the stack overlay slider in Processor and Profiler.
Change Columns	Specify which of the available columns are visible in modules with a Compound Table.

- You can also change the colors of many elements in the Spectrum Graph by adjusting settings in the Preferences dialog.

Related Topics

- "Preferences"

10.3 Sidebar

Menu Location	View > Show Left Sidebar
Icons	

The Sidebars are used to display frequently-used information, available on both the left and right side of the screen. Several views are available in each Sidebar, including: Legend (all modules except Library Manager), Files (all modules except Library Manager), Reference Card Panel (Profiler, Compound Builder and Library Manager), Compound Sets (Profiler and Library Manager), Simulation (Spin Simulator) and Processing History (Processor).

Menu Location	View > Show Right Sidebar
Icons	

Tips and Tricks

- You can open and close the left and right sidebars independently, to display as much or as little extra information as you like.
- Use the drop-down menu at the top of a Sidebar to choose which view you would like available in that Sidebar.
- A Sidebar must be opened before you can access any particular view in it.
- If you set the contents of one Sidebar to be the same as the other Sidebar, the contents of the Sidebars will simply swap sides.

Related Topics

- "Information Tools" , in Chapter 4: *Processor*
- "Information Tools" , in Chapter 9: *Spin Simulator*
- "Information Tools" in Chapter 8: *Compound Builder (PRO Only)*

Legend

The Legend is an interactive display of the various lines currently appearing in the Spectrum Graph. You can use the Legend to toggle the display of each line, change their colors, or even remove overlays or individual compound lines from the display completely.

Menu Location	View > Sidebars > Left Sidebar > Legend
Icon	

How do I toggle the display of a line?

1. Switch to the Legend view in the Sidebar.
2. Select the check box next to a line to display it, or clear the check box to remove it from the Spectrum Graph.

Menu Location	View > Sidebars > Right Sidebar > Legend
Icon	

How do I change a line color using the Legend?

1. Click the colored box beside an entry in the Legend.
2. Click one of the preconfigured colors in the color chooser, or click **More...** to choose from a larger selection of colors.

How do I remove an overlay using the Legend?

1. Click the X beside an overlay entry in the Legend.
2. Or click **Remove All Overlays** in the dropdown menu for the Overlay Lines section.

How do I automatically set the colors for all the current overlays or pinned compounds?

1. Click **Set All to Different Colors** or **Set All to Default Color** () in the dropdown menu for the Overlay Lines or Compound Lines section.

Tips and Tricks

- Items in the legend are organized into sections. Many of these sections have a dropdown menu in the top right corner. If you click on this dropdown, you can see additional actions that can be performed on all the items in that section.
- The topmost section of the Legend contains the default lines that are always available in the current module.
- You can change the colors of the default lines in a module using the preferences for that module.
- Each of the default lines for a module can be toggled on or off temporarily using the Legend. To change the default state of these lines, use the Preferences for the module that you would like to change.
- Changing the color of a compound line using the Legend in Profiler is equivalent to setting a custom color using the Compound Table. The custom color is saved with the spectrum, and will be used when the spectrum is reopened.

Related Topics

- "Spectrum Overlays"
- "Preferences"
- "Compound Table"
- "Colors/Stars/Pins/Remember Selection"

Files

The Files view is an interactive display of other files appearing in the same folder as your currently open file.

Menu Location	View > Sidebars > Left Sidebar > Files
Icon	

Menu Location	View > Sidebars > Right Sidebar > Files
Icon	

Tips and Tricks

- Click **Next** or **Previous** to open a file adjacent to the currently open file.
- There is a dropdown menu next to the **Next** and **Previous** buttons that allows you to control which aspects of the current display will be preserved whenever you click on either button. For example, you can choose whether to maintain the current zoom level in the newly opened file, or the current selected compounds, or the current spectrum overlays.
- You can open a file in a new window by right-clicking on the entry in the Files view and selecting **Open in New Window...**.
- You can still use the **Next** and **Previous** buttons when the Files view is not open. Simply press **Ctrl-Shift-R** or **Ctrl-Shift-W** to open the next or previous file, respectively, or click the appropriate option in the File menu.

Related Topics

- "Spectrum Overlays"
- "Preferences"
- "Compound Table"
- "Colors/Stars/Pins/Remember Selection"

Reference Cards

Menu Location	View > Sidebars > Left Sidebar > Reference Card
Icon	

The Reference Card Panel Sidebar view displays information about specific compounds including structure image, molecular formula and weight, alternate names and links to external reference sites. Use this information to verify the identity of the compound that you are fitting and better understand the signature and behaviour of the compound in your sample.

Menu Location	View > Sidebars > Right Sidebar > Reference Card
Icon	

The compound represented in the Reference Card Panel in Profiler and Library Manager is the compound that you currently have selected in the Compound Table. In Compound Builder, it is the compound matching the Chenomx Compound ID that you have specified in the Compound Details dialog.

The structure image for all Chenomx reference compounds have been annotated to show the atoms that contribute to each compound's NMR signature. In Profiler and Compound Builder, you can click on an atom in the structure image, and the clusters associated with it will be be selected. Likewise, when you select a cluster, the associated atoms on the structure will become highlighted.

Tips and Tricks

- Some sections in the Reference Card Panel are collapsed by default, including Alternate Names and Alternate CAS Registry. To expand them, click the button next to the section name. Click the button again to collapse the section.
- Each of the External Database References are links to the indicated database. Click a link to open the associated database entry in your system's web browser.

Related Topics

- "Preferences"
- "Profiling Tools"
- "Build a Compound"

- "Compound Details"

Quick Searches

Quick Searches let you temporarily limit the number of compounds appearing in the Compound Table based on search criteria in order to locate a particular compound in large, or numerous Compound Sets. The search criteria can include text searches based on compound name, location, compound IDs, compound comments or all of these. You can also search by frequency, listing all compounds appearing at a specific frequency, or within a frequency range.

How to perform Quick Search

1. Locate the search box at the top right of the Compound Table or press **Ctrl-L** to automatically focus the search field.

   Click the binoculars icon at the left edge of the search box to choose the type of search to use (Compound Name, Compound Comment, Compound ID, Location or All). If you do not specify a search type, the default is All.

2. Type your search terms in the Quick Search box. The contents of the Compound Table update to show all matches to your current entry.

3. Click the X at the right edge of the search box to cancel the search and restore the full list of compounds. Clearing the search field also resets the search type to All.

Search for compounds near a single or multiple frequencies using the Spectrum View

1. Right-click on the spectrum, and click **Search for Compounds Near x.xx ppm** to initiate a single location search, or **Add x.xx ppm to search locations** to append the current location to the existing search location(s).

2. Click the X at the right edge of the search box to cancel the search and restore the full list of compounds.

Search for compounds in a single or multiple frequency ranges using a selected region

1. Use the Select Region tool to select a region of the spectrum.

2. Right-click on the selected region, and click **Search for Compounds in this Region** or **Add Region to search locations**.

3. Click the X at the right edge of the search box to cancel the search and restore the full list of compounds.

Search for compounds containing multiplets with given peak frequencies

1. Right-click on the spectrum, and click **Search for multiplet with a peak at x.xx ppm** to initiate a search, or **Add x.xx ppm to search locations (current multiplet)** to add a component to the current multiplet being searched for, or **Add x.xx ppm to search locations (additional multiplet** to start a new multiplet.

Search For Compounds is an alternate method for setting up a Quick Search based on a frequency or range that you type, rather than based on clicked locations in the Spectrum View.

Tips and Tricks

Menu Location	Tools > Search For Compounds...
Icon	

- Quick Searches temporarily limit the number of compounds displayed. Use Compound Sets created in Library Manager to define a more permanent working set of compounds for profiling a particular type of spectra.

- Reset the Quick Search box and clear all filters at any time by pressing **Esc**.
- Click repeatedly anywhere under the sum line (red) in the Spectrum Graph to cycle through the compounds contributing to the sum line at the frequency that you clicked.
- Right-click anywhere under the sum line (red) in the Spectrum Graph, the resulting context menu shows a list of compounds contributing to the sum line at the frequency that you clicked. Select a compound from the list to make it the active compound.
- In Profiler, you can perform frequency and frequency range searches directly from the Quick Search box. For example, type **[2.89** to find compounds appearing at 2.89 ppm, or type **@3.0 to 7.0** to find compounds appearing between 3.0 and 7.0 ppm.
- In Profiler, you can perform multiplet-based searches directly from the Quick Search box. For example, type **[2.512** to find compounds with a multiplet containing a peak at 2.512 ppm, or type **[2.512 2.542** to find compounds with a multiplet containing both a peak at 2.512 and 2.542 ppm. To append a multiplet to the search, use something like **[2.51; 2.542], [2.654**

  The general syntax is as follow:

  Multiplets must start with a left bracket [and end with a right bracket] (but the last multiplet can remain opened). Multiplets must be separated by a coma, and listed peak frequencies in ppm within each multiplet must be separated by semicolons.

Related Topics

- "Spectrum Graph"
- "Compound Table"

Spectrum Details

Menu Location	View > Sidebars > Left Sidebar > Spectrum Details
Icon	

Menu Location	View > Sidebars > Right Sidebar > Spectrum Details
Icon	

The Spectrum Details Sidebar view displays information about your currently loaded spectrum. This information appears in three sections.

- **Calibration** shows a summary of the calibration parameters for the current spectrum and includes CSI, CSI concentration, and the spectrum's pH. These values can be edited in the Processor's calibration tools.
- **Acquisition** displays details about how your spectrum was originally acquired and imported, including the spectrum's spectral (sweep) width, magnet frequency, pulse sequence, etc. These details cannot be edited.
- **Comments** section is used for additional user comments about the spectrum. If any notes were imported from the original spectrum source, they'll appear in this section. You can replace or expand this text however you wish.

Tips and Tricks

- You can hide any section in the Spectrum Details view by clicking on the section's title bar. Click on the title again to restore the hidden section.
- You can only change the details in the **Calibration** section by using Processor's CSI and pH calibration tools.
- You must save the spectrum to retain any changes that you have made to the **Comments** section.

Related Topics

- Chapter 3, *Handling Samples and Spectra*
- "Importing Spectra"
- ""
- "Calibrate pH"

Processing History

The Processing History view in Processor tracks processing effects that you add to a spectrum, recording details about each effect and letting you edit or reverse them as needed.

Related Topics

- "Processing History (Sidebar)"

Compound Sets

Compound Sets (Sidebar)

Compound sets are groups containing compounds found in your Library. You can use compound sets that you have created in Library Manager to make the task of profiling your spectra easier, by placing compounds that interest you the most at your fingertips.

The Compound Sets view lets you modify the contents of the Compound Table to include any combination of the Compound Sets that you have created in Library Manager. The panel contains a list of every compound set that is currently defined. Contents of the checked compound sets appear in the Compound Table.

How do I select multiple compound sets?

Menu Location	View > Sidebars > Left Sidebar > Compound Sets
Icon	

1. Click the checkbox next to the Compound Sets that you want to add to the Compound Table.

How do I select a single compound set?

1. Click on the name (as opposed to the checkbox) of the Compound Set that you want to select. All other Compound Sets will be deselected.

Menu Location	View > Sidebars > Right Sidebar > Compound Sets
Icon	

Tips and Tricks

- Reference Compound Sets, which contain any Chenomx Reference Compounds in your library, appear automatically in the Compound Sets view. If your library contains any non-Chenomx compounds, including ones that you created yourself, they will appear in a separate compound set called **Custom Compounds**.
- When you filter the Compound Table by frequency or range, only compounds from all the currently active compound sets are considered, since these are the only compounds in the table. You can use this, for example, by creating a compound set containing only compounds that appear in urine samples. Then, when this urine compound set is the only one active, any quick searches that you perform will only return compounds that appear in urine samples as potential hits.
- A compound set called **Profiled Compounds** always appears at the top of the Compound Sets view in Profiler. The Profiled Compounds set includes all compounds in the current spectrum that have modified fits, custom colors, pins or stars.

- **Profiled Compounds** also contains subsets that appear as you start profiling compounds. These sets are automatically updated as you work, and allow you to quickly select profiled compounds that meet specific critia. For example, you can select to view only the compounds that have a defined fit, or only the starred compounds.
- The Profiled Compounds set updates continuously, so if you clear a compound, it immediately disappears from Profiled Compounds. In this case, you can still select the compound from its original compound set.
- If you change the contents of a compound set in Library Manager, your changes immediately appear in Profiler, including modified selection rules for Smart Compound Sets, manual changes to Compound Sets by adding or removing compounds, and created or deleted compound sets.

Related Topics

- "Profiling Tools"
- "Automatic Compound Sets"
- "Compound Sets"
- "Smart Compound Sets"

Simulation

The Simulation view in Spin Simulator lets you select, view and edit elements of the currently open spin simulation, including spin systems, spin definitions and J-modifiers.

Related Topics

- "Spin Definition (Sidebar)"

10.4 Spectrum Graph

The Spectrum Graph displays your NMR spectrum as a graph of intensity vs. frequency. In addition to the simple zoom and scroll that you may be familiar with in other software, Chenomx NMR Suite includes a number of specialized tools for manipulating your view of your spectrum.

- Click and drag directly on the spectrum. A red zoom box appears to show you the region you are about to zoom in on.
- Use the horizontal and vertical scroll bars appearing at the bottom and right sides of the Spectrum Graph.
- Middle-click anywhere in the Spectrum Graph to apply an Auto Zoom.
- Select a region using the "Select Region" tool, and double-click in the region, or right-click in the region and select **Expand Region**. The region expands horizontally to fill the Spectrum Graph.
- Select a region using the "Select Region" tool, and middle-click in the region, or right-click in the region and select **Auto Zoom Region**. The region expands horizontally to fill the Spectrum Graph, and an Auto Zoom is applied.
- Place the mouse cursor anywhere in the Spectrum Graph, and use the mouse scroll wheel to adjust the Vertical Zoom.

- Place the mouse cursor anywhere in the Spectrum Graph, then hold down the **Shift** key and use the scroll wheel on your mouse to scroll horizontally.
- Place the mouse cursor anywhere in the Spectrum Graph, press and hold the **Control** key (**Command** for Mac OS X) and use the mouse scroll wheel to adjust the Horizontal Zoom.
- Press **+** (plus) or **-** (minus) to adjust the Vertical Zoom.
- Press ***** (asterisk) or **/** (slash) to adjust the Horizontal Zoom.

Spectrum Thumbnail

The Spectrum Thumbnail appears in the bottom right corner of modules with a Spectrum View, and displays a miniature version of the Spectrum Graph. You can interact with the Spectrum Thumbnail to change what's shown in the Spectrum Graph.

Menu Location	View > Show Spectrum Thumbnail

How do I toggle the display of the Spectrum Thumbnail?

1. Click **Show Spectrum Thumbnail**.

How do I use the Spectrum Thumbnail?

- Click and drag in the Spectrum Thumbnail. A red zoom box appears to show you the region you are about to zoom in on, and remains to indicate the region currently displayed in the Spectrum Graph.
- Click and drag inside the zoom box to move it. The Spectrum Graph updates to show the new region.
- Click and drag one of the solid borders of the zoom box to move that border, or click and drag one of the corners to move the size and shape of the displayed region.
- Click or double-click anywhere in the Spectrum Thumbnail outside the zoom box to show the entire spectrum in the Spectrum Graph. The results are the same as using the "Show Entire Spectrum" zoom tool.

Select Region

You can use the Select Region tool as an alternate zoom function, to specify areas with which to filter the Compound Table, and to obtain information about the selected region itself, including the region's width and the area under various lines within the region.

Menu Location	View > Zoom > Select Region
Icons	

How do I select a region?

1. Activate Select Region.
2. Click and drag across a region of the Spectrum Graph to select the region.

Tips and Tricks

- You can also activate Select Region by double-clicking anywhere in the Spectrum View.
- Double-click in the selected region to expand the region horizontally to fill the Spectrum Graph.
- Middle-click in the selected region to expand the region horizontally to fill the Spectrum Graph and apply an Auto Zoom.
- Additional information about the selected region appears in the top right corner of the Spectrum Graph. The information displayed always includes the region width

in Hz and ppm. Depending on the module, the display can also include the areas under the compound line, overlay line, simulation line, spectrum line or sum line within the selected region, as well as the percentage of the selected area fit by the sum line (Profiler only).

- In Profiler, you can use the selected region to filter the contents of the Compound Table via Quick Searches. Right-click on the selected region, then click **Filter for Compounds in Selected Region**.
- The selected region remains in the Spectrum Graph until you click on an empty region of the Spectrum Graph, select a new region using the Select Region tool, or expand the current selected region to fill the Spectrum Graph.

Related Topics

- "Compound Table"
- "Quick Searches"

Spectrum Overlays

Overlay Chenomx spectra (`.cnx`) in any module with a Spectrum View. Overlaying a spectrum in Compound Builder or Spin Simulator can aid the compound signature creation process, while overlays in Profiler and Processor can provide useful visual references to other spectra in a dataset.

Menu Locations	Tools > Add Spectrum Overlays Tools > Set Spectrum Overlay
Icon	

How do I overlay a spectrum in Compound Builder and Spin Simulator?

1. Click **Set Spectrum Overlay**.
2. Locate the folder containing the spectra that you would like to overlay, and select the spectra.

How do I overlay multiple spectra in Profiler and Processor?

1. Click **Tools > Add Spectrum Overlays** or drag-and-drop files or directories directly into the Legend panel.
2. Choose whether to **Add Files** or **Add Folder**
3. Locate the files or single folder containing the spectra that you would like to overlay.
4. Select the type of information to overlay: either the experimental line, the sum line or the subtraction line.
5. Select whether to color all overlays as a default grey color, or whether to automatically assign different colors to each overlay using the radio buttons.
6. Select how the overlays should be scaled relative to the main displayed spectra, either using the default setting (based on the intensity of the internal standard), or the total area in specified spectral regions.
7. Click **OK**.

How do I change the scale of the overlayed spectra?

1. You can rescale the overlayed spectra by choosing **Scale spectrum overlays** from the Tools menu or by selecting a spectral region using the Select Region tool, right-clicking in the defined region and choosing **Rescale overlays to this region.**
2. You can also interactively modify the scaling of an individual overlay by clicking on the tiny graph icon next to the overlay line name and moving the slider. The sliders

for all overlay lines can be toogle on/off by hovering the mouse over the "Overlay Lines" label at the top of the overlay lines list and selecting **Hide/Show All Scaling Sliders.**

The sliders are self-adjusting. They will automatically extend their scale once you drag the slider knob to the limit on either end. You can also use the mouse wheel to either shink or grow the scale of the sliders, allowing for very fine or coarse adjustments.

You can also customize the scaling ratio of the overlays by right-clicking on the overlay names and selecting **Set Scaling Ratio**. A window will open showing the current scaling ratio setting in a text field.

How do I remove a single spectrum overlay?

1. In the Legend view of the Sidebar, click the X beside the overlay that you would like to remove.

How do I remove all spectrum overlays in Profiler and Processor?

Menu Locations	Tools > Remove Spectrum Overlays Tools > Remove Spectrum Overlay
Icon	

1. Click **Tools > Remove Spectrum Overlays**.

How do I automatically set the overlay colors?

1. Click the **Automatic Coloring** () icon in the Legend Sidebar panel.

   This will assign a different color to each spectrum overlay. Clicking it again will set all overlays back to the default color.

How do I stack overlaid spectra in Profiler or Processor?

Menu Locations	Tools > Stack Spectrum Overlays...
Icon	

1. Click the **Stack Spectrum Overlays...** menu item in the Tools menu.

   This will open a dialog for you to input the height of all overlays.

2. Or you can use the stack overlay widget on the left hand side of the graph to manually adjust the stack height.

Tips and Tricks

- You can drag cnx files (even directories) from your desktop or the Files panel directly into the Legend panel to automatically overlay their respective spectra.
- Double-click on a listed overlay line to load the corresponding file.
- You can also remove all spectrum overlays by clicking on the X beside the Overlay Lines section in the Legend.
- You can only overlay spectra in the Chenomx NMR Suite format (`.cnx`). If the spectrum you would like to use is in a different, supported format, you need to import and save it in Processor before using it as an overlay.
- In Compound Builder and Spin Simulator, you can only have one spectrum overlay at a time. In Profiler and Processor, you can have up to one hundred spectrum overlays.
- In Compound Builder, if you overlay a spectrum whose frequency does not match the magnet frequency specified in the Compound Details, you are notified of the mismatch, and given the chance to update the Compound Details to match the spectrum overlay.

Related Topics

- "Sidebar"
- Chapter 4, *Processor*
- "Compound Details"

Zoom Tools

Displaying specific ranges can be useful for preparing illustrations or focusing on a particular area of the Spectrum Graph.

Menu Location	View > Zoom > Set Zoom
Icon	

1. Open the Set Zoom dialog.
2. Enter the horizontal (X-Axis) in ppm, and vertical range (Y-Axis) in standard intensity (si) that you would like to display, or select Entire Axis to zoom out to the full spectrum height/width. NOTE: When you open the Set Zoom dialog, the values that appear initially correspond to your current view. If you change the values in only one dimension, your current settings in the other dimension remain intact.
3. Click OK.

Undo/Redo Zoom

Just as Undo and Redo let you recall recent actions, Undo Zoom and Redo Zoom let you recall recent views of the spectrum.

Menu Location	View > Zoom > Undo Zoom
Icon	

Menu Location	View > Zoom > Redo Zoom
Icon	

Increase/Decrease Vertical/Horizontal Zoom

Increase/Decrease Vertical/Horizontal Zoom lets you incrementally adjust the vertical/horizontal scale of the current view, without changing the displayed frequency range.

Menu Location	View > Zoom > Increase Vertical Zoom
Icon	

Menu Location	View > Zoom > Decrease Vertical Zoom
Icon	

Menu Location	View > Zoom > Increase Horizontal Zoom

Icon	

Menu Location	View > Zoom > Decrease Horizontal Zoom
Icon	

Increase/Decrease Horizontal Zoom lets you incrementally adjust the horizontal scale of the current view, without changing the displayed intensity range.

Auto Zoom

Auto Zoom lets you quickly adjust the vertical scale of the current view so that the tallest peak in the currently displayed frequency range fits nicely in the viewing pane.

Menu Location	View > Zoom > Auto Zoom
Icon	

Tips and Tricks

- You can apply Auto Zoom to the Spectrum Graph at any time by clicking anywhere on the spectrum with the middle mouse button.

Show Entire Spectrum

Show Entire Spectrum lets you quickly expand the current view, setting both the vertical and horizontal scales to their largest possible values.

Menu Location	View > Zoom > Show Entire Spectrum
Icon	

Saving and recalling view settings

View settings an easily be saved and recalled at any point in time. The view settings are global (shared across all modules) and are rememered between different sessions with the software. Up to 5 different view settings can be saved. Only the limits on the ppm axis get stored.

To save a view, press and hold the mouse for at least two seconds on one of the five view setting buttons on the toolbar. After two seconds, the icon will change to a flashing camera. You can then release the mouse to save the view setting. A camera shutter sound will be emitted to signify that the view has been saved in the corresponding slot. A text notification will also appear in the module's' status bar at the bottom left of the module window.

To recall a view, simply process with a regular click on one of the five view setting buttons in the module's toolbar or via the View menu.

Menu Location	View > Zoom > Recall view setting
Icon	

Export Spectrum Image

When you prepare publications or presentations based on your work in Chenomx NMR Suite, you may need to include images illustrating the fitting process or particular regions of the spectrum. You can export spectrum images from any module with a Spectrum Graph into a variety of image formats.

Menu Location	File > Export > Spectrum Image...
Icon	

How do I export an image of the current spectrum?

1. Start the Export Spectrum Image wizard.
2. Choose a file format. You can choose a from a variety of vector or raster image formats, including portable network graphics (`.png`), graphics interchange format (`.gif`), scalable vector graphics (`.svg`), or portable document format (`.pdf`).
3. Choose a folder and filename for the exported image.

Tips and Tricks

- Choose **Do not shade area under lines** to ensure that none of the peak shading effects are retained in the exported image. Disabling shading can be useful when you export an image of an area containing many clusters, as shading can create a very heavy look.
- If you are exporting the graph image for display on another computer (in an email or webpage, for example), you should use a raster format like `.png` or `.gif` for the best results.
- If you are exporting the graph image to be embedded as a small figure in a paper document or on a large wall poster, you should use a vector format like `.svg`.

Related Topics

- "Spectrum Graph"
- "Display Options"

Copy Spectrum Image to Clipboard

You can use Copy Spectrum Image to Clipboard to quickly make an image of the current spectrum available to paste directly into another application, without creating an intermediary file.

Menu Location	Edit > Copy Spectrum Image to Clipboard
Icon	

Tips and Tricks

- The resolution and size of the spectrum image copied to the clipboard is the same as the Spectrum Graph on your screen. If adjusting combinations of your module window size and/or screen resolution do not let you produce images of a high enough quality, consider using Export Spectrum Image.

Related Topics

- "Spectrum Graph"
- "Export Spectrum Image"
- "Display Options"