AutoVectis
HRMS Software

A New Window into Complex Organic Matter Analysis with FTMS: Demonstrating the Benefits of Asymmetric Apodization

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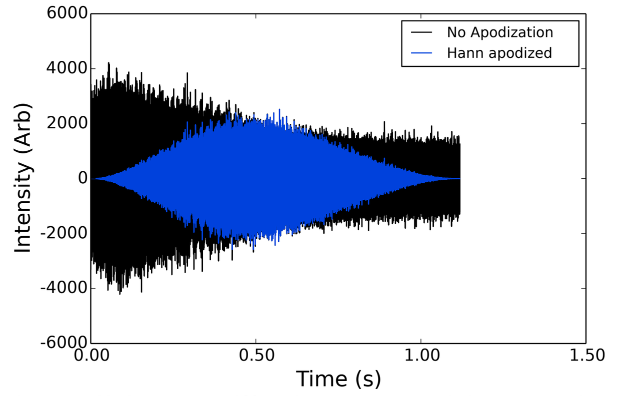
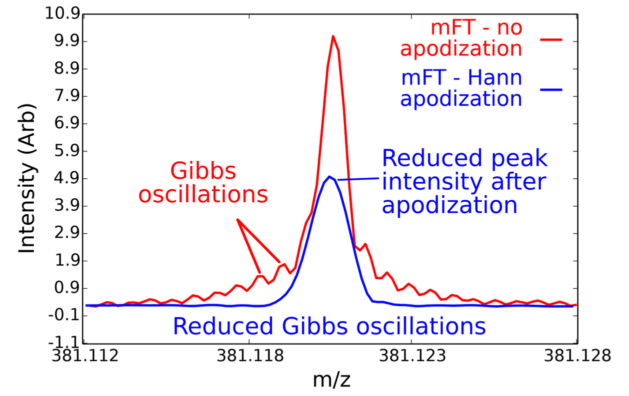
Introduction

The analysis of complex organic matter – e.g. humic and fulvic acids, natural organic matter, petroleomics, biofuels, and direct infusion metabolomics - remains challenging, even for high-resolution FTMS. One often overlooked but critical parameter, in processing FTMS data, is the selection of the apodization function. Many users rely on default settings, unaware of the significant benefit that different apodization functions, particularly asymmetric windows, can have on their results. By optimizing apodization settings, it is easy to enhance signal-to-noise ratio (S/N), detect more peaks, and assign more with more confidence. Here, we show the benefits of asymmetric apodization functions, providing a framework for optimizing FTMS data processing, maximising spectral information retrieval and improving molecular assignments in complex samples.

Gibbs oscillations

If an FTMS transient is Fourier transformed "raw", then the peaks in the resulting mass spectrum will have artefact peaks, known as Gibbs oscillations, that will make interpretation much more complex.





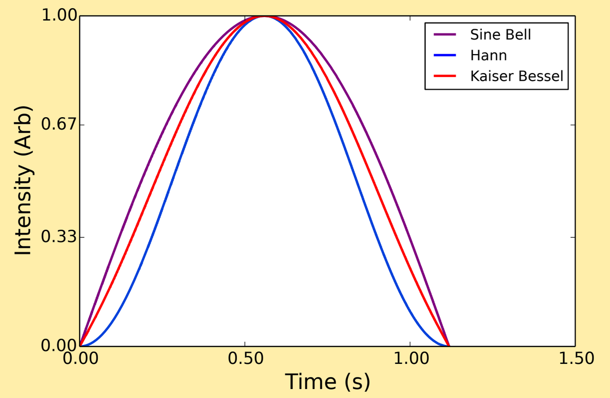
The Gibbs oscillations are (destuctively interfering) artefacts that result from the transient being finite and having sharp ends. It is computationally expensive and difficult to remove Gibbs peaks by filtering peak lists.

Apodization

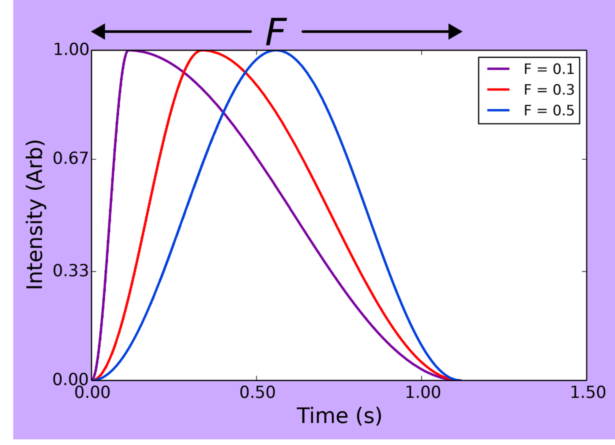
Apodization is much more successful way of removing Gibbs oscillations. A digitized transient is simply a list of numbers - the transient shown above is 2^{22} points long (approx 4M points). An apodization or window function is another list of numbers, with a special shape, the same length as the transient. If we multiply the two lists, we generate the apodized transient. FFT of that produces peaks with significantly reduced Gibbs oscillations - at the cost of peak intensity and resolution.

Window functions

There are many different window functions - here are 3 functions comonly used with Fourier transforms. These result in differences in spectral peak geometries and abilities to remove Gibbs oscillations. Normally, window functions are symmetrical - meaning the apex of the function is half-way along the function.



Asymmetric windows

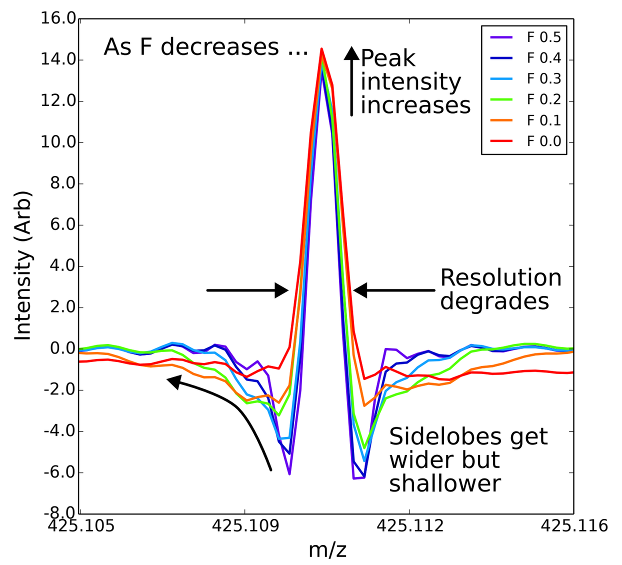


Apodization functions that are asymmetric can be beneficial. In AutoVectis, asymmetric Hann, Sine bell and Kaiser Bessel windows are available. The asymmetry is defined by the *F* value - expressing the position of apex of the window. Each side of the function is a corresponding half window function of that type.

Effect of *F* on peak shape

Changing the asymmetry of the window, by varying *F*, affects the absorption mode peak shapes in the resulting mass spectrum.

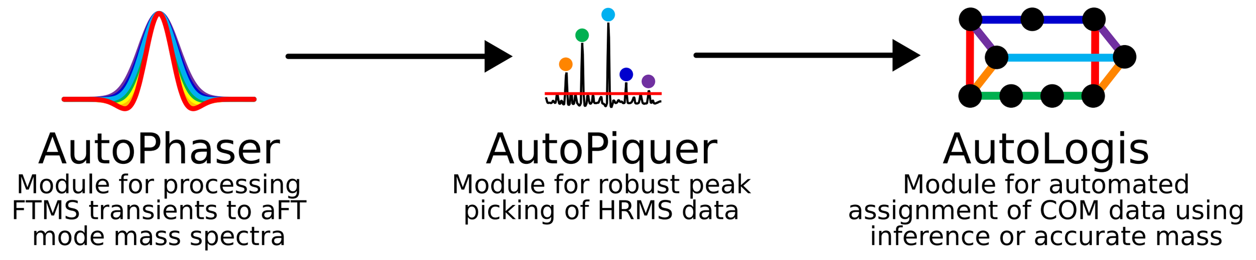
Commonly, transients exhibit damping, meaning that the intensity decreases the longer the transient is recorded - so, lower values of *F* tend to result in increased peak intensities, as the window maximum moves back into the earlier, more intense, early regions of the transient. But, this comes at the cost of reduced resolution.



Optimizing your apodization

Selecting the best apodization and asymmetry (*F* value), during post processing, can make a measurable difference for assignment. Differences in spectral complexity, signal to noise levels and dynamic range, together with factors relating to the specific information you need to extract from the spectrum, will influence the optimum apodization settings to use.

AutoVectis offers unique built in batch processing functions to automate this optimization process, iterating through multiple options. Then, once the processing options are optimized, the rest of the data can be batch processed and automatically assigned with the best settings, using the built-in batch processing workflows.



Videos demonstrating the workflows automating the optimization, processing and assignment of the example data can be found here (follow the QR code) and at:

https://www.youtube.com/@autovectis

Benefit of optimization for COM analysis

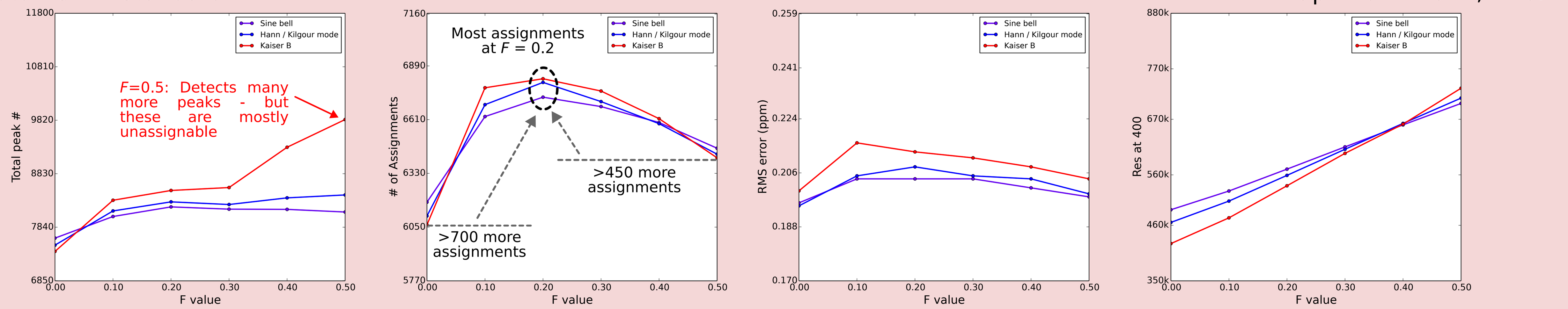
A variety of complex organic matter samples (including Suwannee River Fulvic Acid, SRFA) were analyzed using Orbitrap and Ion Cyclotron Resonance (ICR) FTMS instruments. Data were acquired using standard system acquisition hardware or with an external high-performance data acquisition system (DAQ) (FTMS Booster, Spectroswiss). Data processing was performed in AutoVectis Pro (Vibrat-Ion and Spectroswiss), from raw transient data (Bruker .d, Spectroswiss .h5, MIDAS or ASCII formats) to molecular assignments and graphical outputs, using the built-in COM workflows in AutoVectis. Apodization functions - including Hanning, sine bell, and Kaiser-Bessel windows - were applied across a range of symmetric and asymmetric configurations. The effects on signal intensity, resolving power, peak detection, and assignment statistics were recorded to determine the optima.

Results of optimization

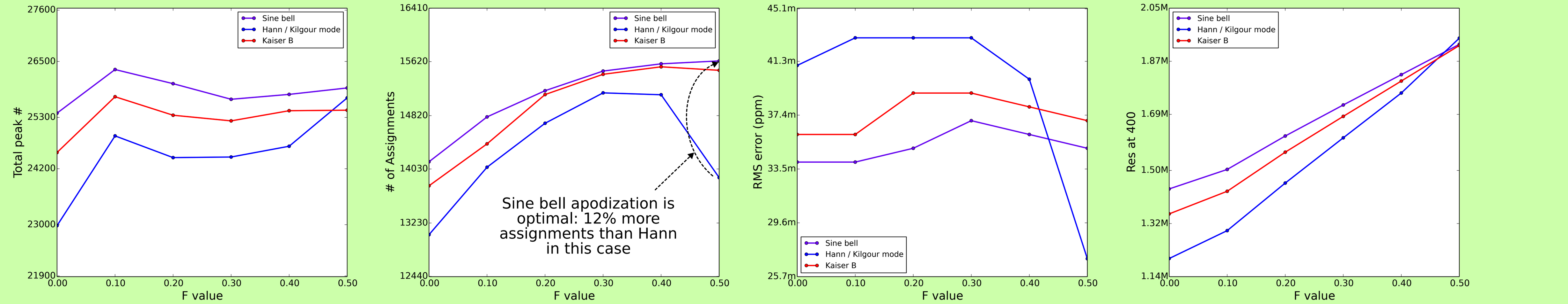
For example, in a 12T Bruker SolariX ICR analysis of an SRFA standard, changing from the normal symmetry to asymmetric apodization (F=0.2) increased molecular assignments by ~10% for both Hanning and Kaiser windows, a significantly greater improvement than the ~0.2% difference between window types. Half-bell apodization (e.g., half-Hanning) provided higher S/N but reduced resolving power, leading to fewer assignments. For the half-bell window, baseline interference patterns would also require computationally expensive and less robust baseline corrections.

Similar assignment benefits are often seen whe processing the data from other FTMS instruments, as shown below. Choosing the right window and apodization asymmetry can get you more assignments from the same data.

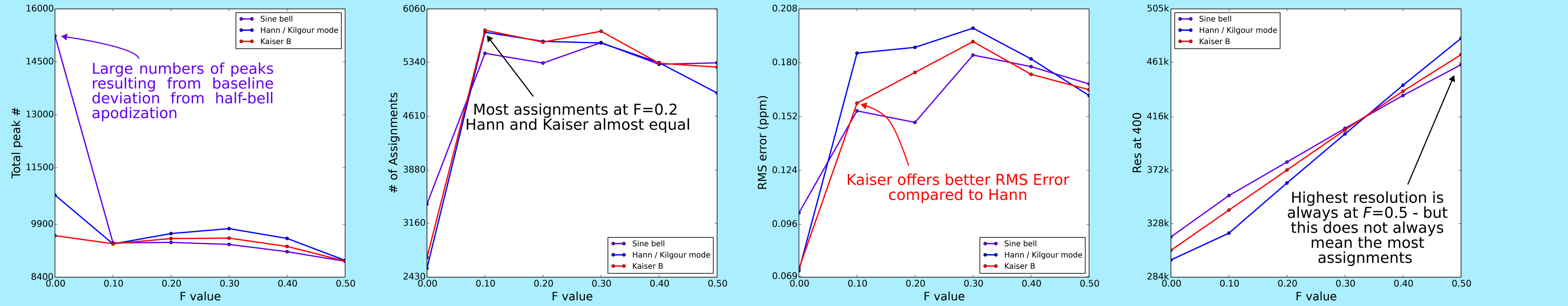
12T data - SRFA Standard — Optimum: Hann, *F*=0.2



21T data - SRFA standard — Optimum: Sine, *F*=0.5



Orbitrap - Booster - Bio-crude sample — Optimum: Kaiser, *F*=0.1



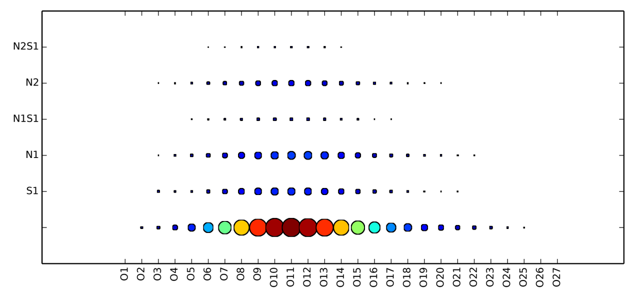
Batch workflows for COM/DOM analysis

Processing of DOM data is complex - and consequently the software workflows are often similarly complicated. AutoVectis has automated processing workflows for the processing and assignment of DOM data - either from FTMS transients or from already processed mass spectra from any instrument (including TOF). It uses graphical interfaces rather than command line access; consequently it is easier to learn and use.

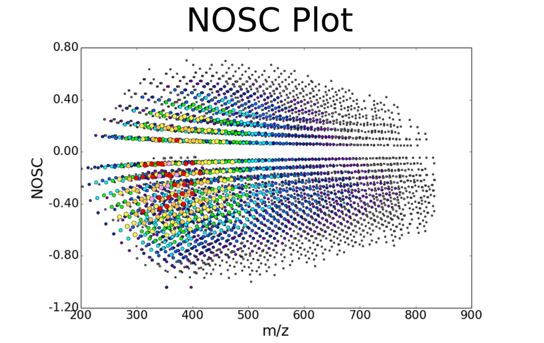
AutoVectis also offers batch processing capabilities that we have used, on batches of many hundreds of files, to automate the processing - freeing up valuable user time for other tasks, as well as reducing the risk of accidental errors creeping into the results. These batch methods are valuable for automating the optimization of processing methods, as demonstrated here, as well as for permitting the robust processing of large batches of data.

Results, from batch processing, can be exported to text, Excel and to SQLite databases, either from individual results files or by combining results of large batches - to make it easy to import into other downstream workflows, such as PCA or other statistical models. But, AutoVectis also offers a wide variety of results graphics for easy data visualization during method development as well as for use in posters, reports and presentations:

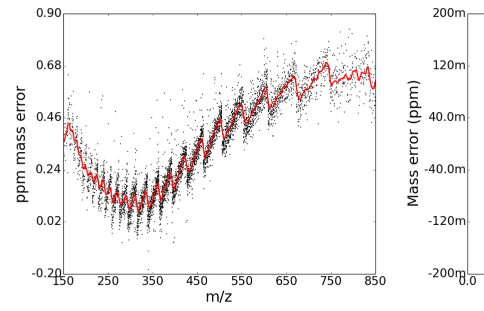
Homologous Class Plot



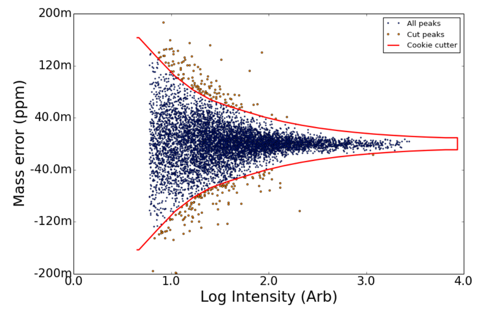
NOSC Plot



Advanced recalibration



Confidence filters



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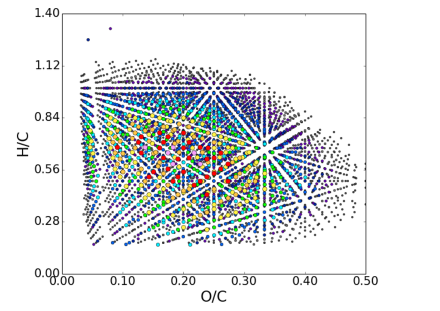

Conclusions

When processing FTMS data, many users are missing out on the benefits that more advanced apodization could bring them. We hope this poster shows why you might want to investigate if your default apodization window is the best for your data and if you might benefit from using an asymmetric apodization. AutoVectis offers the widest range in different apodization options for FTMS processing, including offering asymmetric apodization using Hann (Kilgour mode, in the Bruker software), Sine Bell and Kaiser Bessel functions.

The automated batch processing options in AutoVectis streamline optimizing your apodization - as well as offering automated batch processing for other processing parameters, and for complete end-to-end processing: transient or mass spectrum through to batch reports and databases of assignment results.