Determination of collision cross sections of proteins using frequency domain linewidths and time domain decay profile fitting of MS1 data from an Orbitrap mass spectrometer

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Introduction and Goals

- Collisional cross sections (CCS) are typically measured by dedicated Ion mobility (IM) mass spectrometers.
- Previous work has shown CCS can be calculated using decay rates of the time-domain transient signal from an Orbitrap™ mass spectrometer.[1]
- Calculation of CCS from FWHM of frequency-domain signals has been demonstrated using FT-ICR.[2]
- Here CCS values are determined for multi-charge state and individual isolated charge state transients using both transient decay fitting and FWHM methods.

Methods and Instrumentation

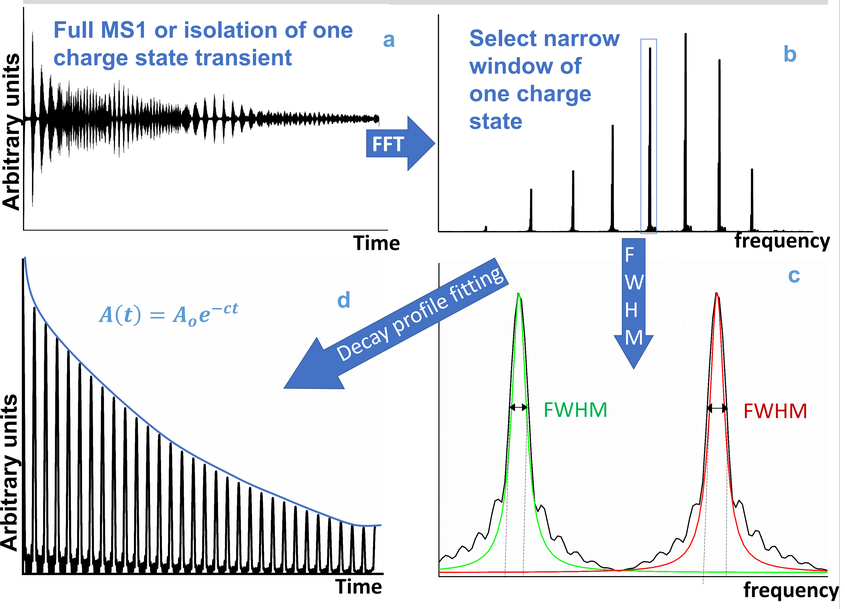

Figure 1. (A) Transients were collected on an Orbitrap™ mass spectrometer (B) A FFT was performed in Matlab, and a selection of a narrow range of data points was chosen for (C) Lorentzian FWHM fitting (OrginLab) or using (D) decay profile fitting method.[1] CCS values for cytochrome C, myoglobin, and ubiquitin were evaluated. Either the decay constant or FWHM can be used in Eqn 1 and 2.

$$\sigma = \frac{FWHM\ (or\ c)}{f_z L N} \quad \textit{Equation 1}$$

$$N = FWHM\ (or\ c)/(f_z * \sigma) \quad \textit{Equation 2}$$

σ, f_z, L represent the CCS, frequency coefficient, and path length, respectively.

Results and Discussion

- Transients for an individual charge state were used to calculate CCS using both methods with close agreement to CCS values based on IM from the literature.
- Transients containing ions of multiple charge states were then used to calculate CCS and were also in close agreement with CCS values from IM measurements.
- CCS calculated using single charge states or multiple charge states closely agree for both transient decay fitting and FWHM methods, suggesting that both methods are viable for CCS measurements of proteins.
- FWHM method could overcome resolution limitations of the decay profile method if peak fitting of data that is not isotopically baseline resolved is accomplished. However, the FWHM method is still currently out-performed by the decay profile fitting method.

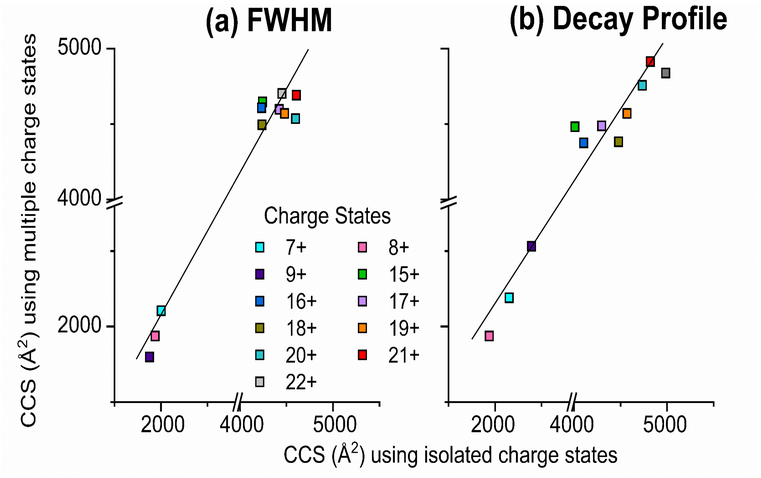

Figure 2. Comparison of CCS values for myoglobin calculated using individual isolated charge states vs multiple charge states for (a) the FWHM method and (b) the decay profile fitting method

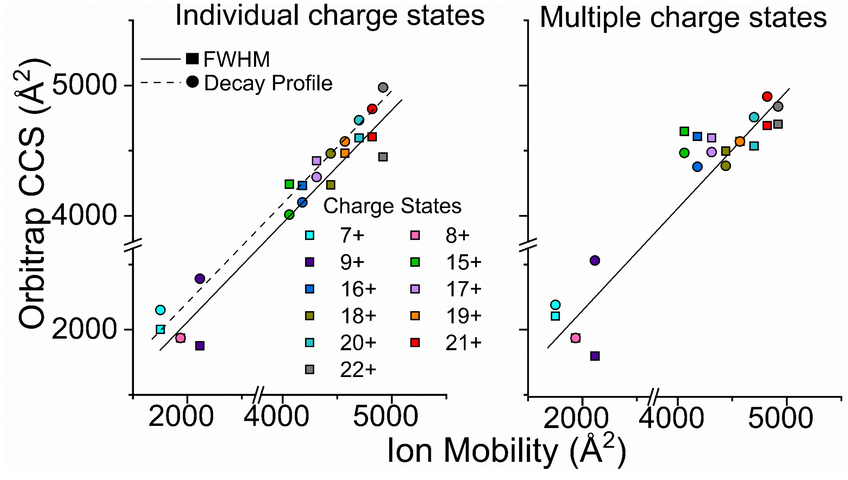

Figure 3. CCS values obtained by the FWHM and decay profile fitting methods (Orbitrap CCS) compared to CCS values from ion mobility.[3]

Conclusions

- CCS of multiple charge states from MS1 transients were determined with decay profile fitting and the FWHM methods with accuracy comparable to CCS obtained by isolating individual charge states or CCS from ion mobility.
- FWHM method measures CCS with accuracy approaching but not yet surpassing the accuracy of the decay profile fitting method.

References and Acknowledgements

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