

Introduction to Mass Analyzers

Mass Analyzer

- Separate ions within a selected range of mass-to-charge (m/z) ratios
- The most important part of the MS because of the role it plays in the MS's accuracy and mass range
- Controlled by magnetic fields, electric fields or measuring the time it takes an ion to travel a fixed distance
- Performance Characteristics
 - 1) Mass Range
 - 2) Mass Accuracy
 - 3) Mass Resolution (Resolving power)
 - 4) Acquisition Speed
 - 5) Tandem MS Capability

Types of Mass Analyzers

Magnetic Sector

- double focusing sector

Quadrupole

- Single quadrupole mass filter
- Triple quadrupole (QQQ)

Beam-Type Mass Analyzers

Ion trap (IT)

- 3D quadrupole ion trap (QIT)
- 2D linear ion trap (LIT)

Ion-Trapping Mass Analyzers

Ion cyclotron resonance (ICR)

Orbitrap

- LIT – OT (LTQ-OT velos, 기초과학공동기기원)
- Q - OT (Q-Exactive)
- Q - OT - LIT (OT Fusion Tribrid)

Time-of-flight (TOF)

- TOF/TOF (MALDI-TOF-TOF-MS, 기초과학공동기기원)
- Q-TOF (Triple TOF, Nicem)

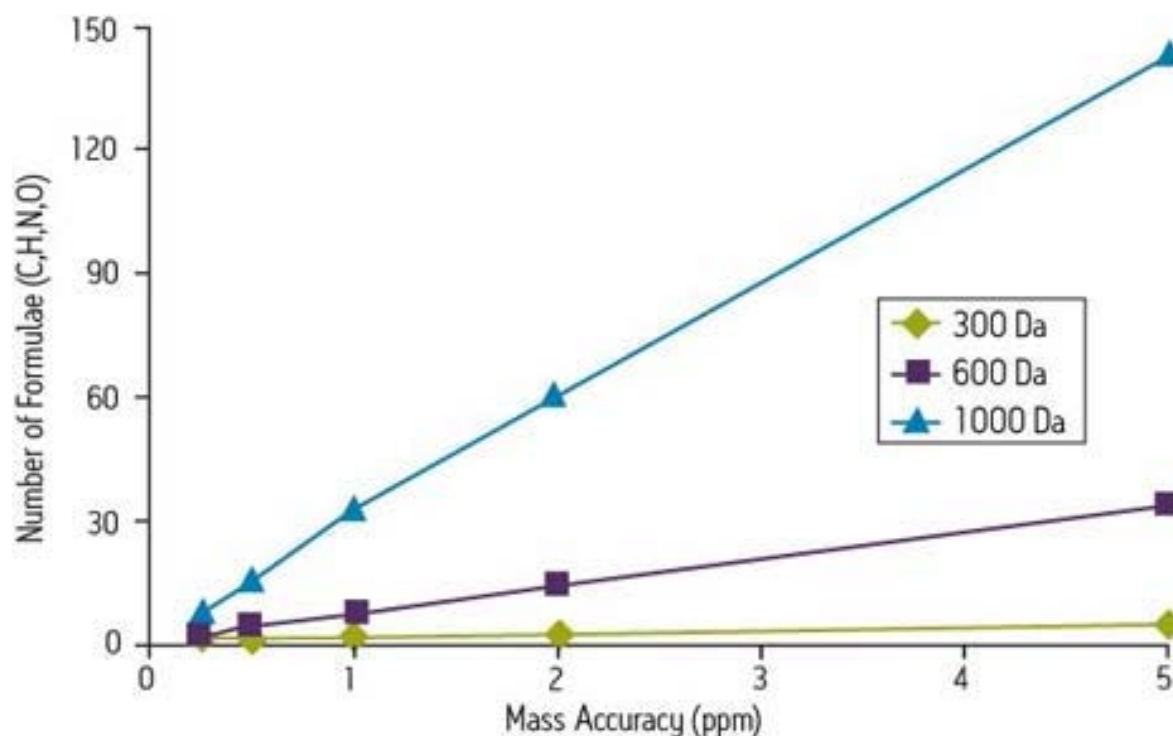
Mass Accuracy

- The difference between measured accurate mass and calculated exact mass.
- Expressed as an absolute unit (u or amu) or as a relative unit (ppm).

$$\text{ppm} = \left(\frac{m_{\text{exp}} - m_{\text{calc}}}{m_{\text{exp}}} \right) \times 10^6$$

How much accuracy do we need, or can realistically achieve, and what are the compromises?

“ For C, H, O, N compositions (C_{0-100} , H_{3-74} , O_{0-4} and N_{0-4}) a nominal mass-to-charge response at 118 needs only an error not exceeding 34 ppm to be unambiguous, where a m/z response at 750 requires precision better than 0.018 ppm to eliminate “all extraneous possibilities.” “



The effect of increasing mass accuracy for unambiguous identification of compounds

<http://www.waters.com>

High Mass Accuracy

Type	Mass Accuracy
FT-ICR-MS	0.1 - 1 ppm
Orbitrap	0.5 - 1 ppm
Magnetic Sector	1 - 2 ppm
TOF-MS	3 - 5 ppm
Q-TOF	3 - 5 ppm
Triple Quad	3 - 5 ppm
Linear IonTrap	50-200 ppm (10 ppm in Ultra-Zoom)

- Instruments must be calibrated to obtain high mass accuracy.
- External calibration: mass spectrum of calibration compounds is acquired in a different experiment than the analyte compound. (Instrument drift can introduce errors.)
- Internal calibration: mass spectrum of calibration compounds are recorded at the same time as analyte compound. (Observing both calibrant and analyte ions in the same experiment can be a significant challenge.)

Mass Resolution (Resolving Power)

$$R = m / \Delta m$$

- The ability of an instrument to perform such a separation of neighboring peaks.
- The ratio of the mass of interest, m , to the difference in mass, Δm , as defined by the width of a peak at a specific fraction of the peak height.
- The terms 'resolution' and 'resolving power' are basically interchangeable.
- There are two major ways **how to define Δm** .
 - 1) 10% valley definition (= 5% peak width definition if symmetrical peak)
 - 2) **FWHM** (Full Width of the peak at Half its Maximum height) definition

Mass Resolution (Resolving Power)

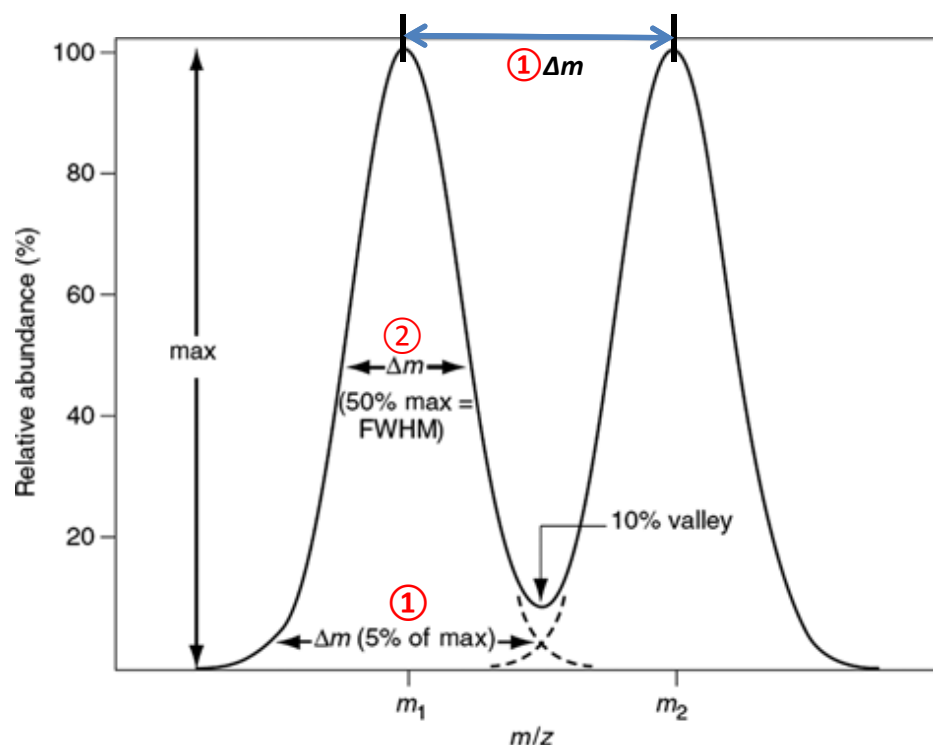
- There are two major ways **how to define Δm** .

1) 10% valley definition (= 5% peak width definition if symmetrical isolated peak)

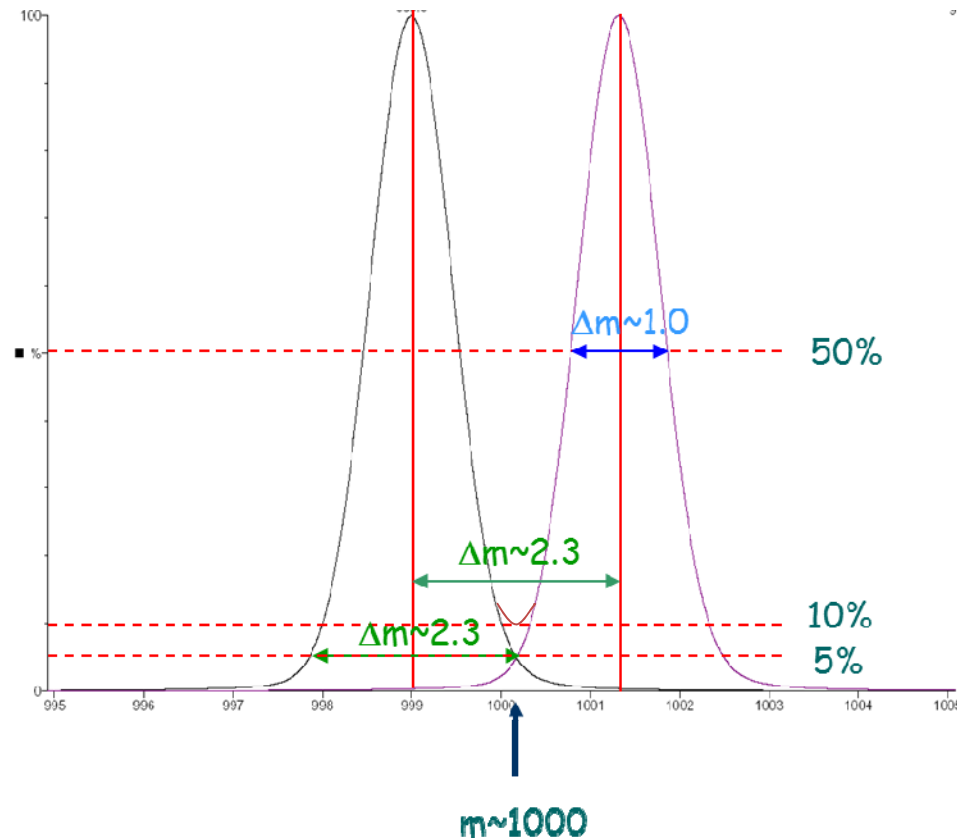
- Useful only for instruments giving Gaussian peaks.
- Magnetic sector instruments

2) FWHM (Full Width of the peak at Half its Maximum height) definition

- Preferentially used for peak with wider tailing (Asymmetric)
- Quadrupole MS, FT-ICR MS, Orbitrap MS and ToF MS instruments



10% valley conditions are fulfilled if the peak width at 5% relative height equals the mass difference of the corresponding ions. (5% + 5% = 10%)



❖ The numerical value arrived at using the FWHM definition is always larger than the value one would determine using the 10% valley definition.

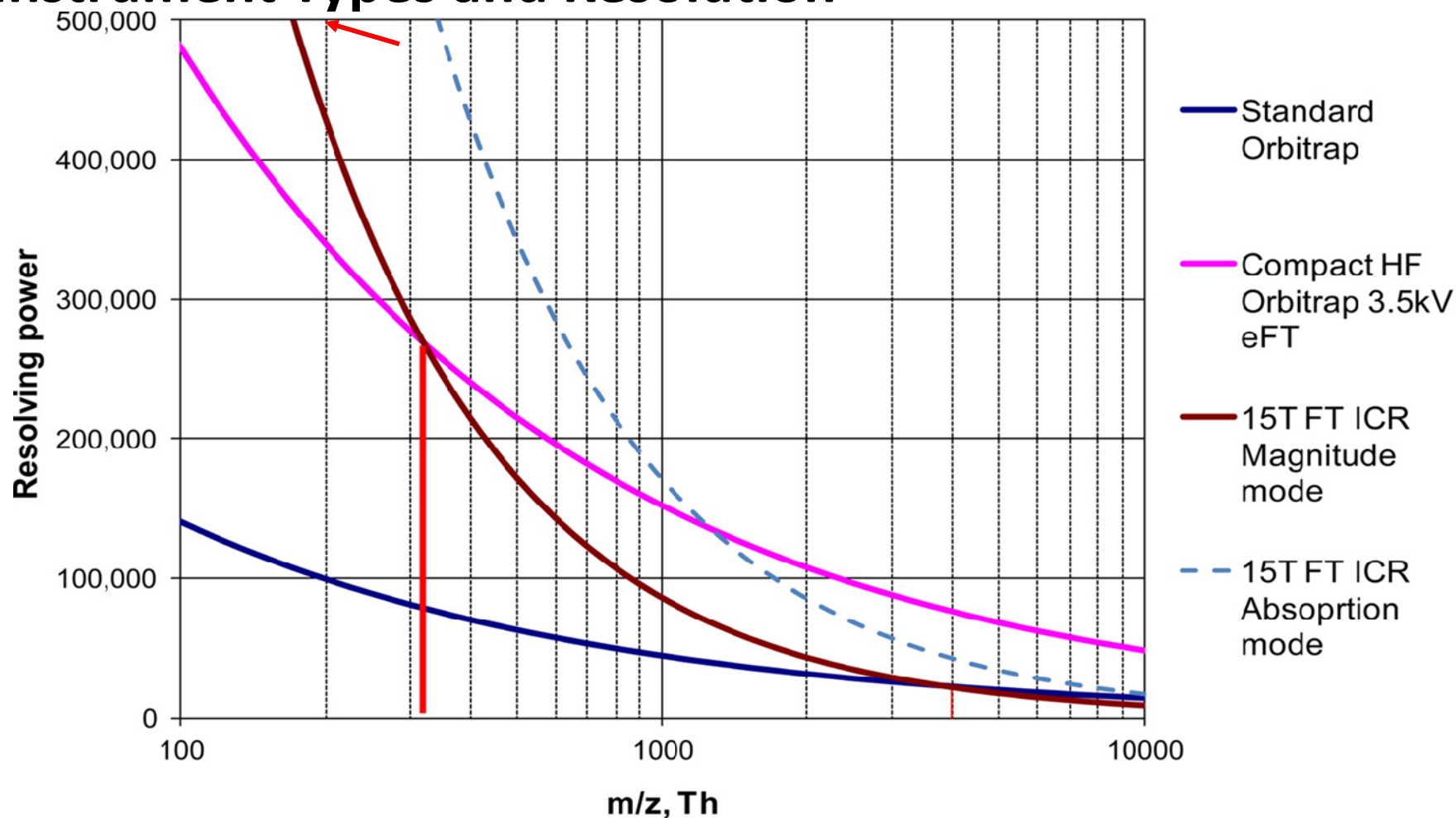
$$R(10\% \text{ valley or } 5\% \text{ peak width}) = 1000/2.3 = \mathbf{430}$$

$$R(\text{FWHM}) = 1000/1.0 = \mathbf{1000}$$

Instrument Types and Resolution

- 1) Transmission quadrupole, linear ion trap: Since Δm is fixed (**unit mass resolution**), the resolution increases linearly with mass ($R \uparrow = m \uparrow / \text{fixed } \Delta m$).
- 2) TOF and double-focusing mass spectrometers operate at **constant resolving power**.
- 3) FT-ICR MS: the resolution **decreases linearly with mass**. When you express resolving power or resolution, you specify the m/z value where you calculate **R**.
Ex) $R = 100,000$ at m/z 400 becomes $R = 25,000$ at m/z 1600.
- 4) Orbitrap MS: the resolution **decreases with square root of mass**.
Ex) $R = 100,000$ at m/z 400 becomes $R = 50,000$ at m/z 1600.

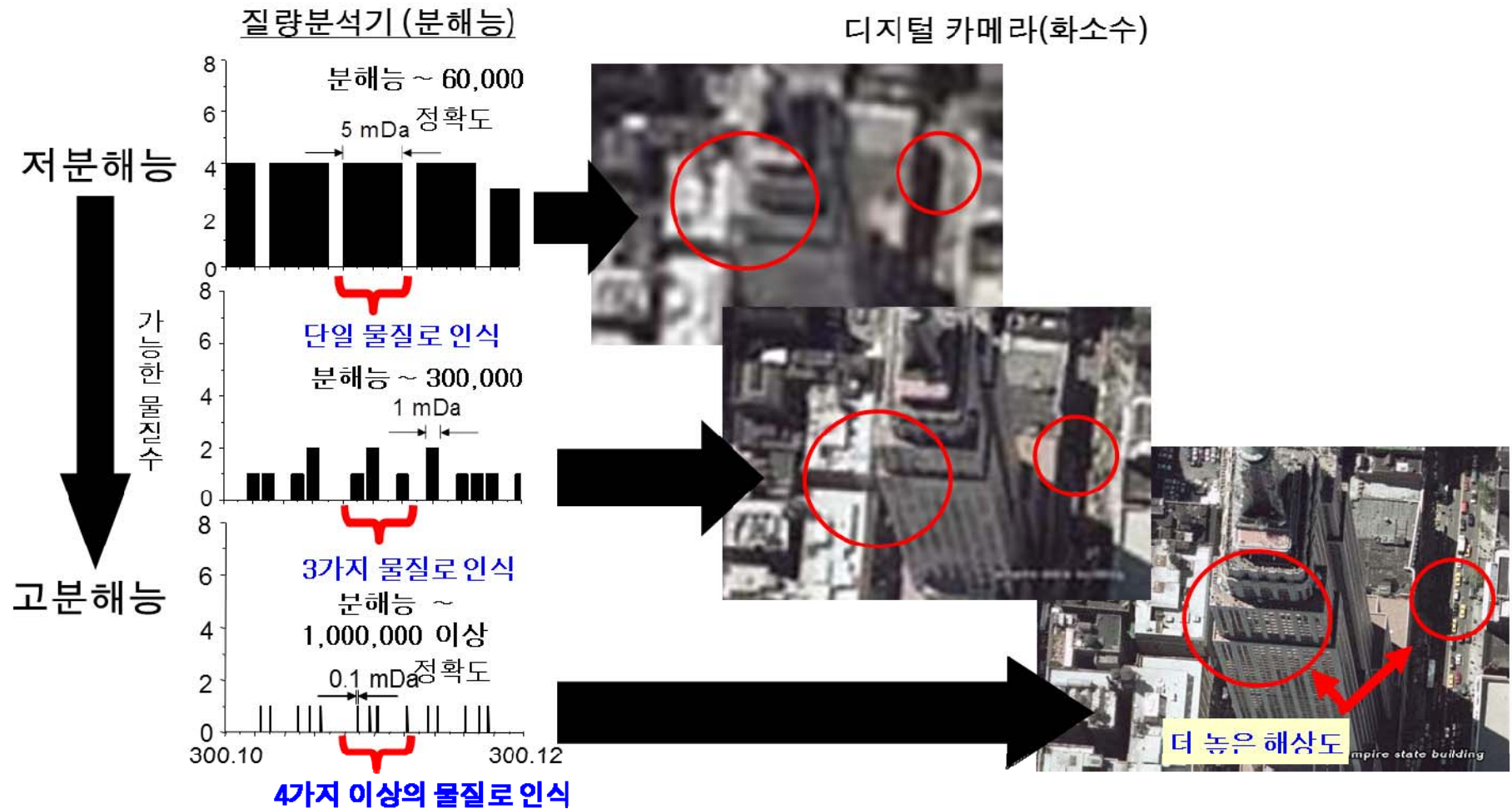
Instrument Types and Resolution



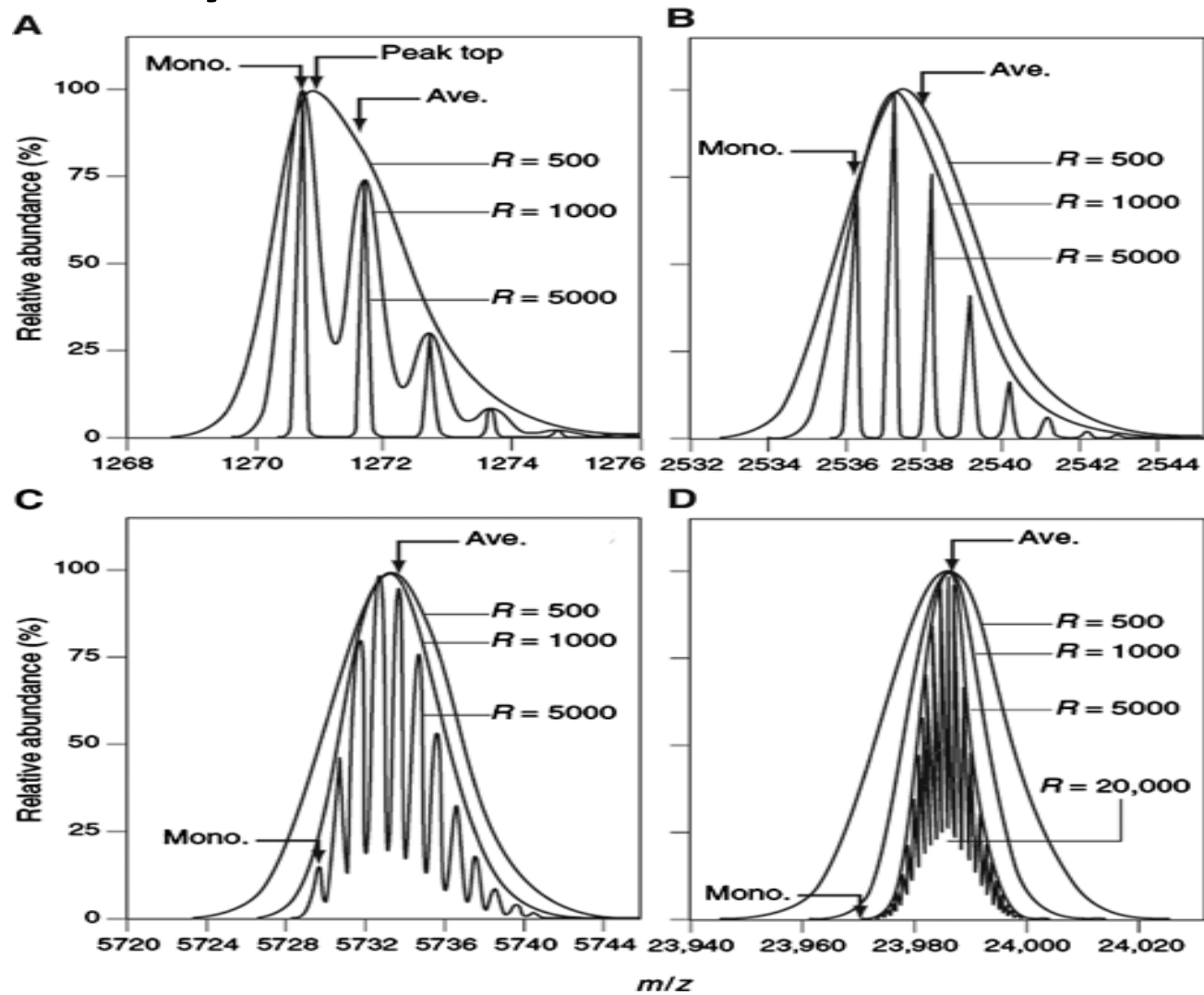
Dependence of resolving power on m/z for the following analyzers (all data are shown for a 0.76 s scan): (i) standard trap (magnitude mode, 3.5 kV on central electrode), (ii) compact high-field trap (eFT, 3.5 kV on central electrode), (iii) FTICR (magnitude mode, 15 T), (iv) FTICR (absorption mode, 15 T).

Roman A. Zubarev; Alexander Makarov; *Anal. Chem.* 2013, 85, 5288-5296.

Advantage of High Resolution

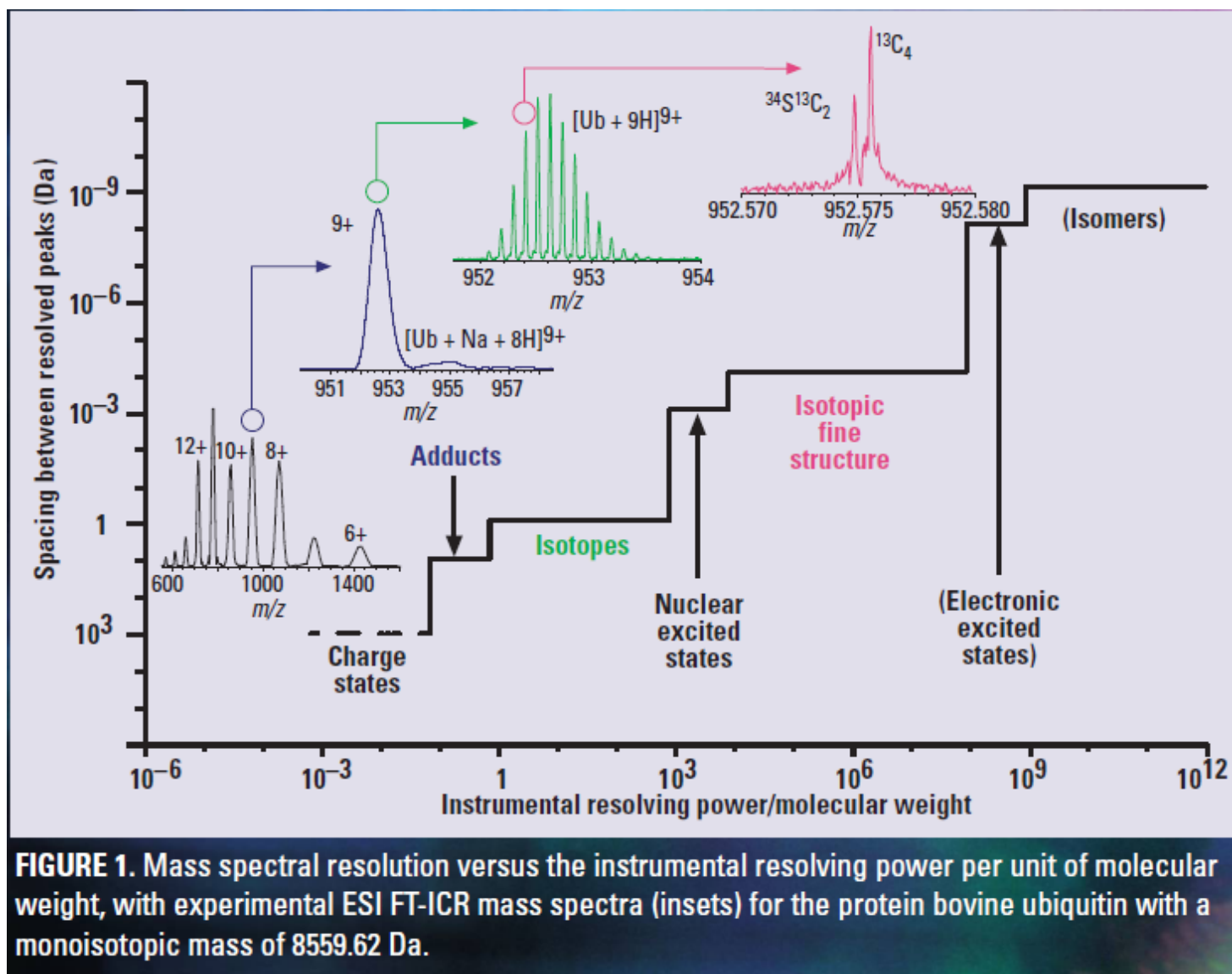


Mass Accuracy and Resolution



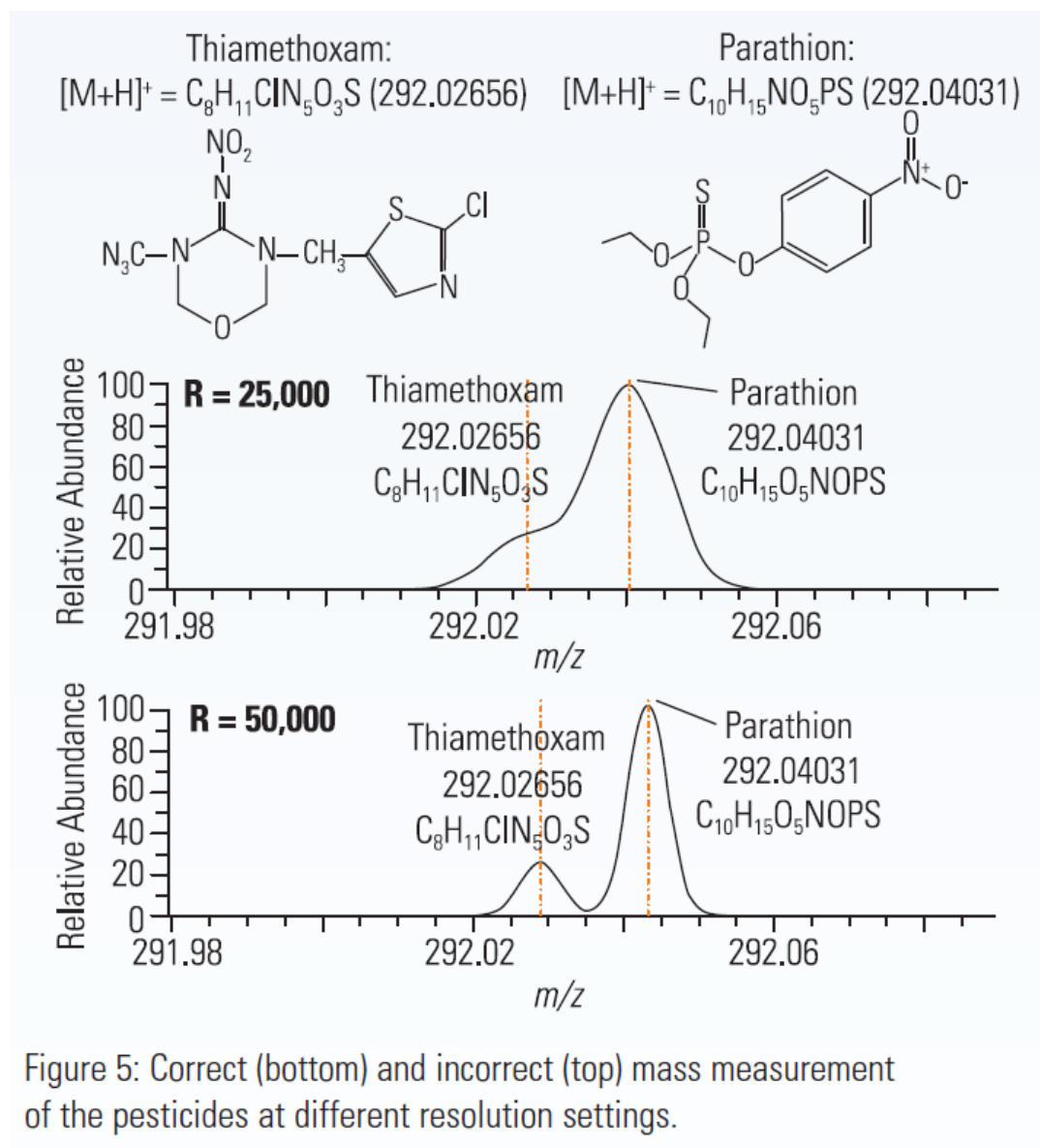
Good mass accuracy can only be obtained from sufficiently sharply and evenly shaped signals.

Resolution- What Can Be Seen..



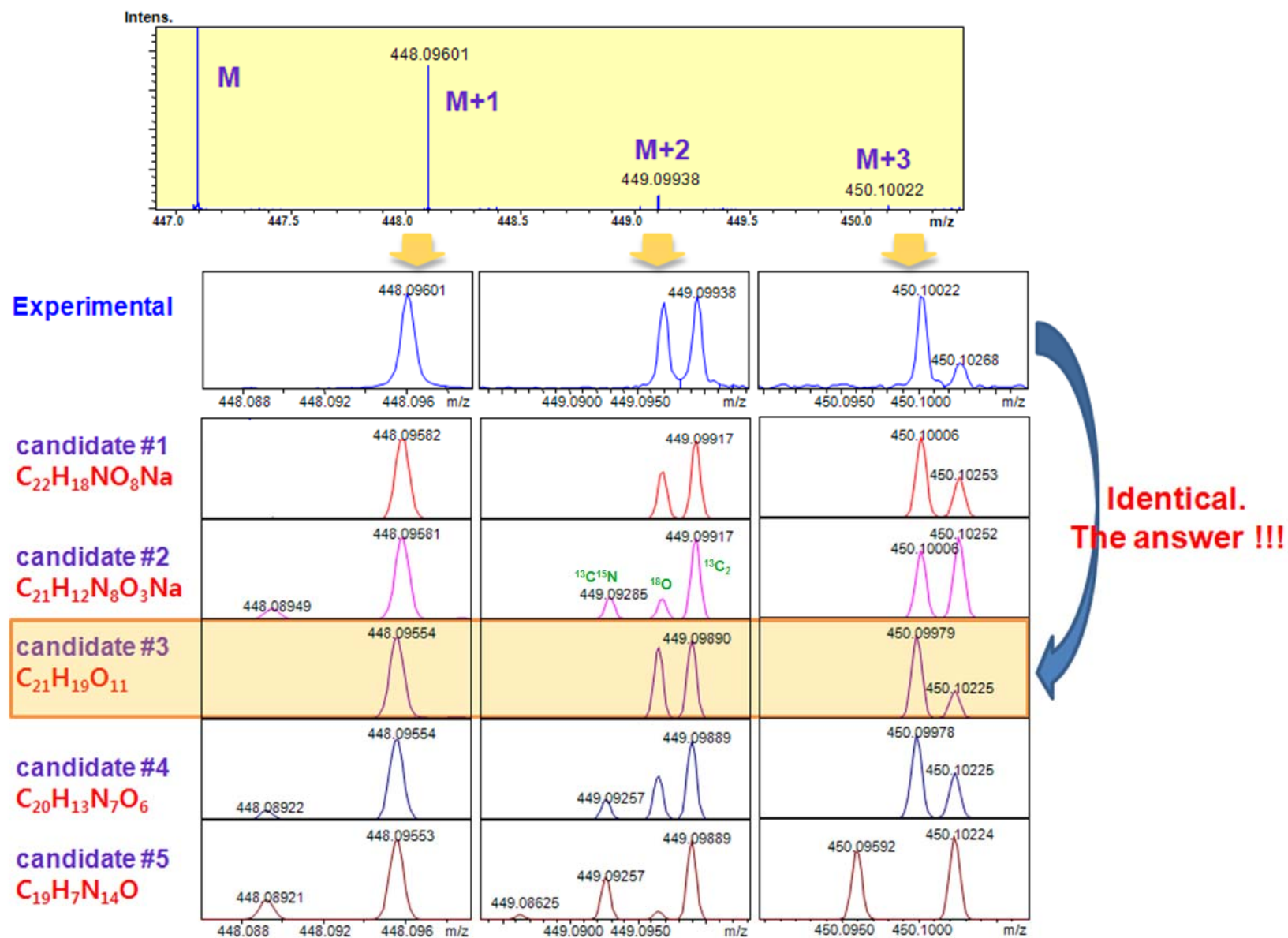
Anal. Chem., 2002, 74 (9), pp 252 A–259 A

High Resolution

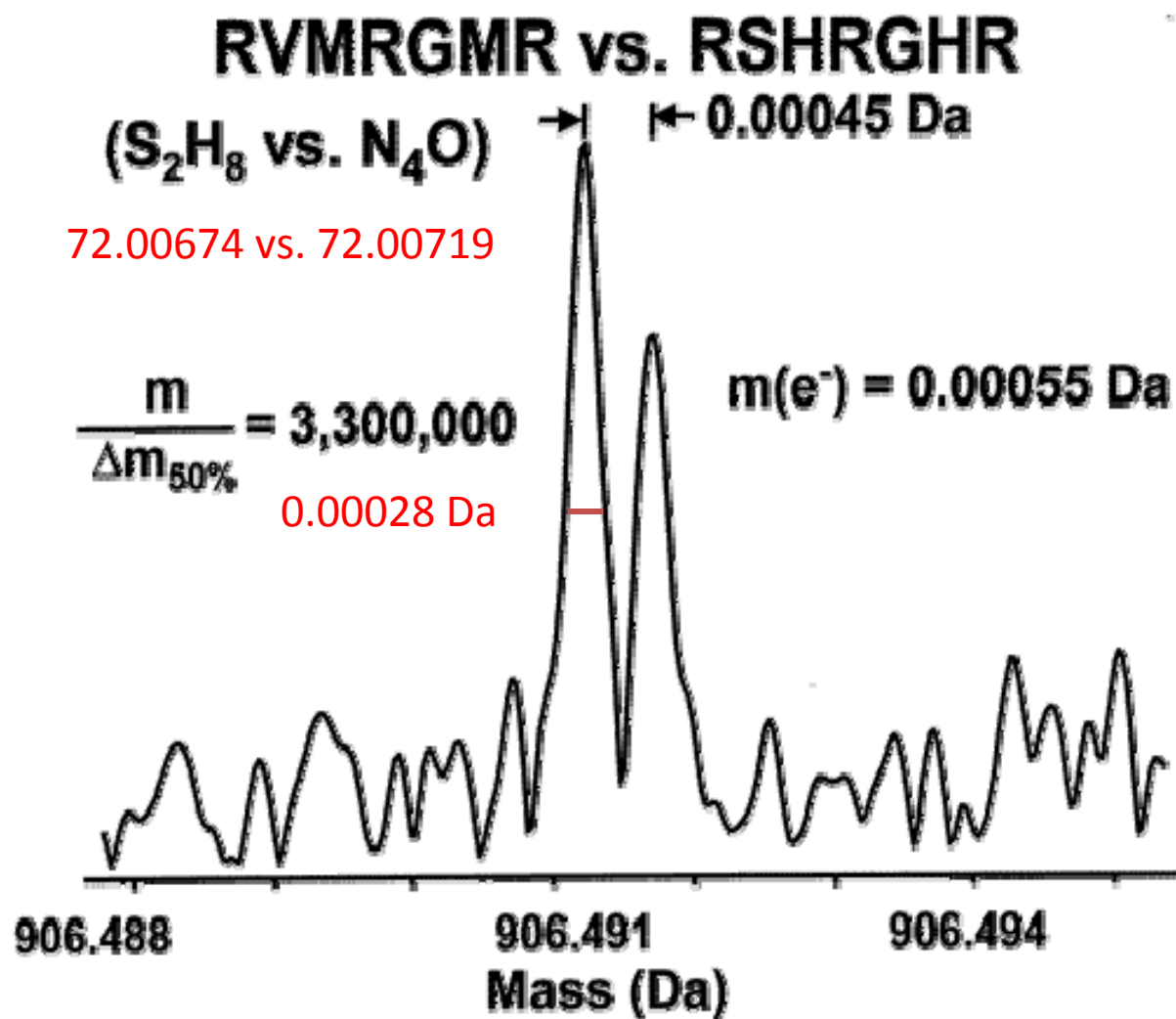


~14 mDa

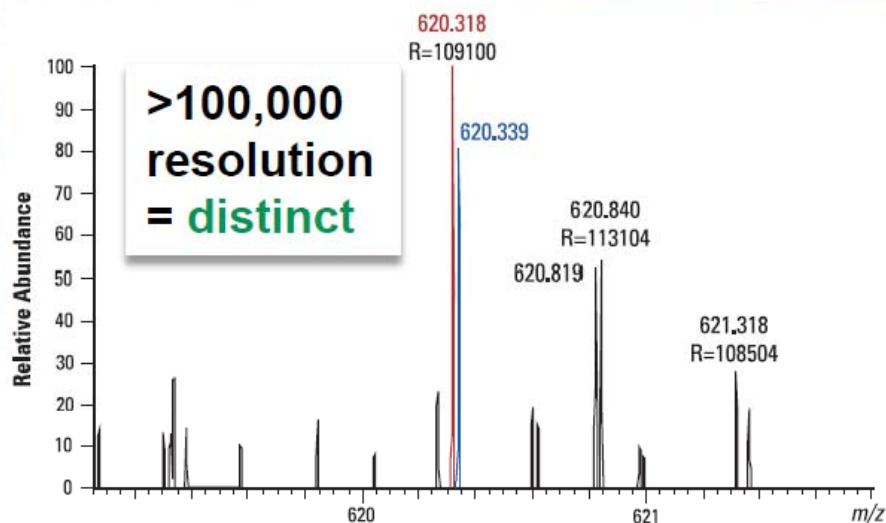
Determination of Molecular Formula



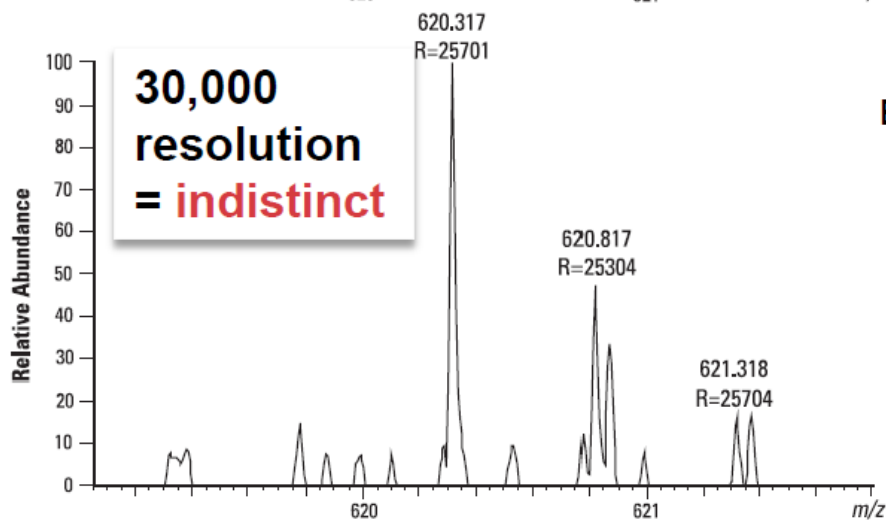
Ultrahigh Resolution



SILAC quantification: Resolution vs Accuracy



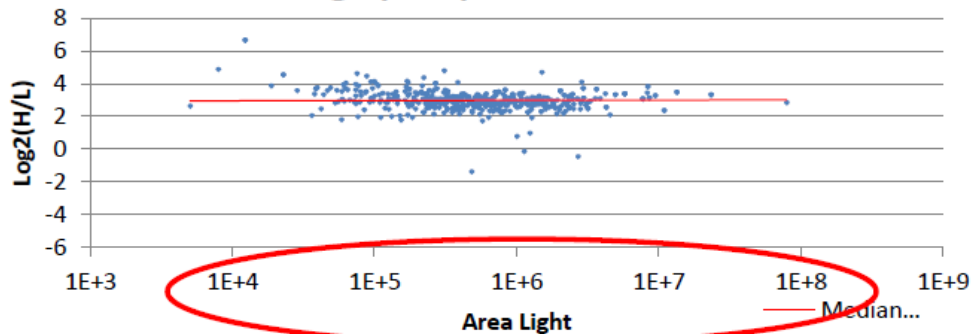
$$Resolution = \frac{M}{\Delta M}$$



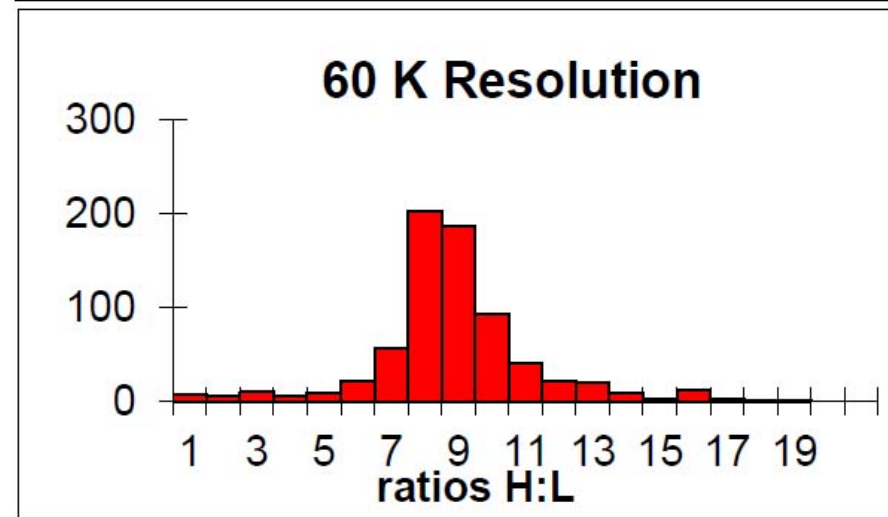
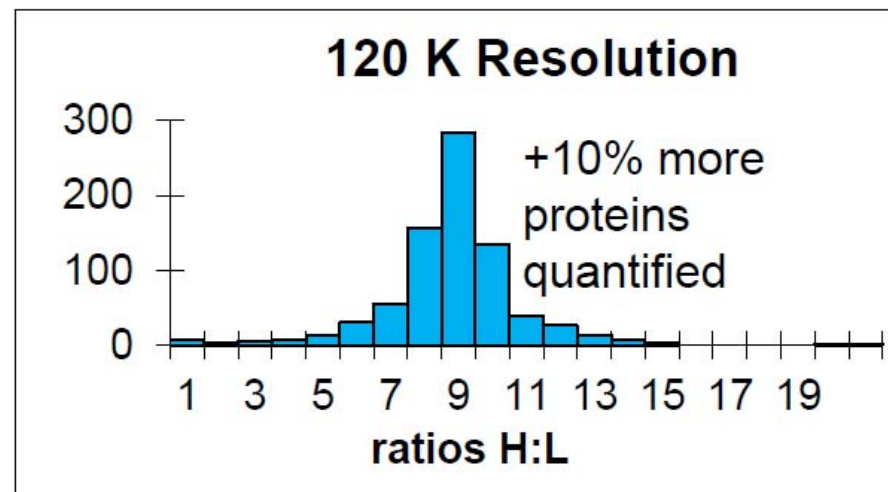
Four orders of magnitude for 1:10 ratio

B.

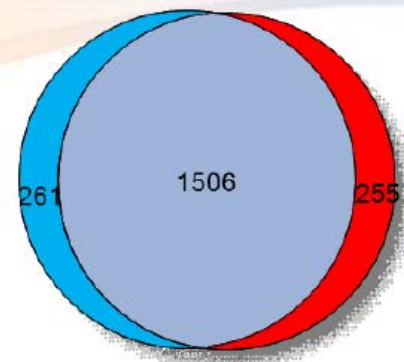
Log₂(H/L) vs. Area L



SILAC Quantification: Resolution vs Accuracy

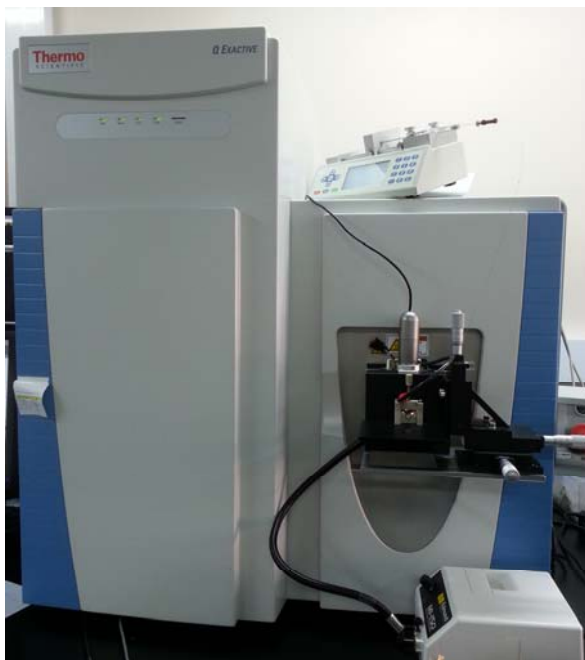


Protein ID results



HeLa cell labeled
with R¹⁰&K⁸, mixed in
H:L=10:1
OT Elite :1x FTMS + 20 x
ddrCID,

Q-Exactive classic



R = 140,000 at m/z 200

Orbitrap Fusion Tribrid



R = 500,000 at m/z 200

