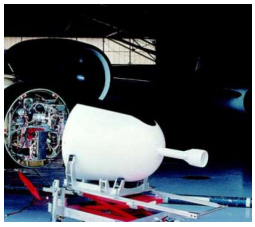
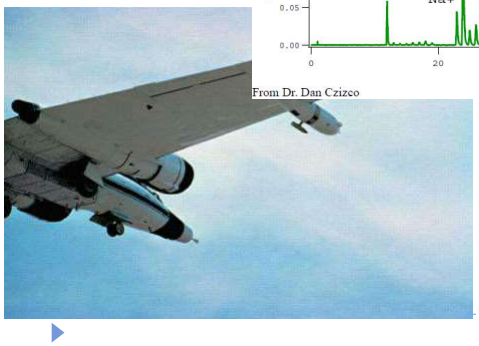
Interpretation - Basics

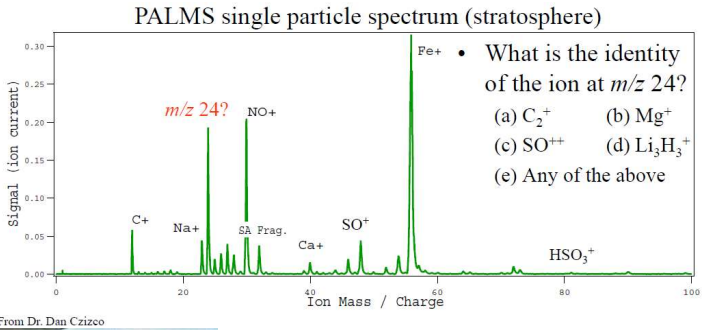
- What is the mass spectrum and what is it's resolution?
- How do we determine the resolution?
- How do we characterize an error with which we determine the mass?
- Standard interpretation procedure

- **Literature - Fred W. McLafferty, František Tureček: Interpretation of mass spectra**

Mass spectrum





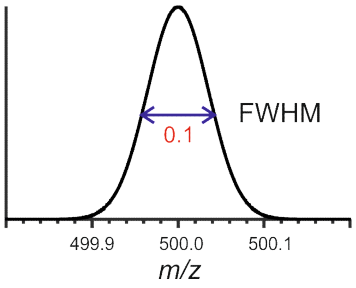


From Dr. Dan Czizco

Mass spectrum =
Recording of ion intensities as a function of their mass-to-charge ratios

This spectrum – a low resolution spectrum – *m/z* gives a nominal mass

Resolution

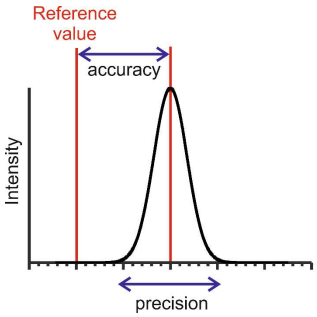


Peak width ($\Delta m_{50\%}$, FWHM)
- Full width at half maximum

Mass resolution ($m/\Delta m_{50\%}$)

Mass = 500
FWHM = 0,1
Resolution = 500/0,1 = 5000

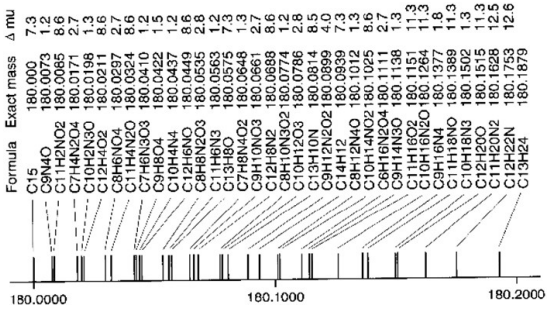
Precision/Accuracy



Real mass = 500
Measured mass = 500,002
Difference = 0,002
Error = 10^6x0,002/500 = 4 ppm

Low vs. high resolution

Exact masses of ions with nominal mass 180 D (only C, H, N, O)



Q:
What resolution do you need to determine elemental composition of ions with m/z 180?

Hint:
minimal Δm_{min} = ?
R > 180/ Δm_{min}

Interpretation – first steps

- I have a sample, what am I going to do?

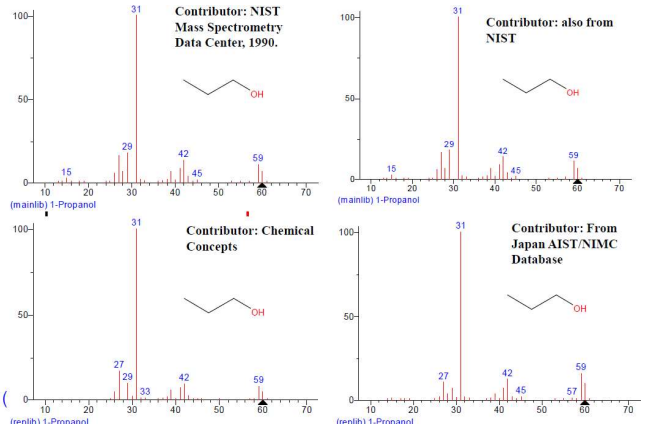
1. I will measure an MS spectrum and compare it to a database
2. I will measure a high-resolution spectrum →
   → elemental composition
3. I will identify the compound according to "Standard interpretation procedure"

- EI database
  - NIST's Chemistry WebBook – 6000 molecule, open access (http://webbook.nist.gov/chemistry)
  - NIST off-line, Wiley – order of magnitude more molecules (~10^5)

- Protein databases, e.g., MASCOT (http://www.matrixscience.com)

Reproducibility and noise

1. Signal to noise ratio depends on the instrument
2. Spectra are affected by impurities (e.g., air, solvents, etc.)
3. Calibration!



Standard interpretation procedure for EI spectra

1. **Known information** (other spectra, history of the sample), clear requirements for the MS measurement, control the *m/z* assignment (calibration)
2. **Elemental composition** – isotopic pattern (for all peaks in the spectrum)
3. **Molecular ion** (largest mass in the spectrum, odd number of electrons, logic neutral losses). Comparison with spectra obtained with CI or other soft-ionization method
4. **Important ions**: odd number of electrons, largest abundance, high mass, largest abundance in a group of the peaks
5. **Appearance of the spectrum**: stability of molecular ion, labile bonds
6. **Possible sub-structures**
   1. Important series of ions with low masses
   2. Important neutral losses from $M^{+\cdot}$ (fragment with high masses)
   3. Characteristic ions
7. **Suggest molecular structure**

   Comparison with a reference spectrum, with spectra of similar compounds, check with fragmentation mechanisms expected for the suggested molecular ion

→ Always step by step!