Accurate Mass Measurement in a Service Environment

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Summary

- Robust method for accurate mass measurement is required by the National Service.
- Traditional method uses 2-stage process on two different spectrometers.
- New instrumentation provides a 1-stage process with full-scan accurate mass.
- Target is to use new instrumentation to improve efficiency whilst maintaining robustness and ease-of-use of traditional method.

Introduction & History of the 2-stage process

- The NMSSC provides an accurate mass service to the UK university community.
- Traditionally, the NMSSC has performed these analyses by peak-matching experiments using sector instruments with a resolution of 38,000.
- In the last 10 years, electrospray has become the technique of choice due to ease and speed of sample introduction, increasing performance over traditional high-vacuum sources.
- Peak-matching requires a characterisation, (low-resolution) spectrum to establish the presence of the expected ion.

Step 1: Characterisation Scan

- Low-resolution characterisation scans were obtained using a Fisons Quattro II (EUCI) with Robohopper autosampler or Waters 2Q4000 with Advion NanoMate (ESI).

Step 2: Peak-Matching

- Accurate mass measurements were obtained using the Finnigan MAT 95Q/ISL mass spectrometer fitted with an electrospray source and operated with the standard operating procedures. 
- Xcalibur v1.4 SR1 with high resolution mass spectrometer, programs v1.4 software was used to produce lists of elemental formulae.
- Typical Source Settings used:
  - Accelerating voltage: 5kV.
  - Capillary temp.: 250°C.
  - Spray voltage: +10kV.
- Samples were loop injected into a stream of water / methanol (1:1) flowing at 30 to 60L/min.
- Polyethyleneimine (PEI) is introduced to the system via the 'Sheath Liquid' port on the ESI source, typically at a flow rate of 10L/min.

Chemicals

PEI (low-molecular weight), ammonium acetate (NH₄OAc), deuterium phosphate (DPA) and caffeine were purchased from Aldrich (Dorset, UK). HPLC grade dichloromethane (DCM), methanol (MeOH) and water were purchased from Fisher Scientific (Loughborough, UK). MEFSA was purchased from Research Plus, Inc. (New Jersey, USA). Ultramark 1621 was purchased from Alfa Aesar (Massachusetts, USA). Samples were obtained as part of the routine service of the NMSSC.

Accuracy of measurements

- Sector instruments have been proven to be highly accurate. 
- The Obritor is a relatively new analyser, which has been reported to have good mass accuracy and dynamic range.
- Data analysis has been performed on the measurements made using both instruments.
- Peak-matching showed a characteristic distribution about 0.
- One-pass accurate mass showed a wider distribution of measurements from the calculated mass.
- SMS mass error is 0.15Da (peak-matching) and 0.45 Da (one-pass accurate mass), see Figure 4.

Examples of analyses facilitated by the new method:

Example 1 – Polyphosphates / polyphosphonates

- Ambient ionisation (API) enhances analysis in negative and positive modes.
- A previous method required NaF to be run concurrently with the sample as a reference compound, negating the DEA effect and rendering this molecule useless.
- New method allows effective addition of DEA to observer and measure [M-H]:
  
  \[ M - nH \]

Example 2 - Characterisation of contaminant

- Using the standard electrospray source with loop-injection or LC autosampler, polarity-switching can be performed in one pass.

Discussion

- Both old and new methods have proved to be robust and effective.

Advantages of new method:

- Improved laboratory efficiency, throughput and cost effectiveness.
- Speed of analysis. 
- Facilitate accurate mass measurement over full-scan range.
- Increased confidence for characterisation due to:
  - Accurate masses of all isotopes.
  - Elemental identification (e.g. 95%) 
- Improved efficiency (ca. 30%) and cost-effectiveness.
- Faster sample turnaround.
- Improved analytical quality and extended capability.

Advantages of old method:

- A broad range of ionisation modes is required for samples unsuited to electrospray.
- Characterisation spectrum is provided, the high speed of the traditional method still makes it useful.
- Both old and new methods will continue to be used for the foreseeable future.

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References