Improving Mass Accuracy on a Unit Resolution Quadrupole Mass Spectrometer

Maria Cristina A. Dancel[1], Ming Gu[2] and David H. Powell[1]

[1]Department of Chemistry, University of Florida, Gainesville, FL; [2]Cerno Bioscience, Danbury, CT

OVERVIEW

- Our main goal is to achieve parts per million (ppm) mass accuracy on a unit resolution quadrupole mass spectrometer *via* internal calibration.

- At least three ions were used to generate calibration curves, with Excel™ and with MassWorks™. The latter software also provided calibrated ion profiles for elemental composition determination.

- Less than 10 ppm mass errors for various known and synthetic compounds were achieved on the quadrupole MS, while the shorter list of possible elemental compositions made chemical formula determination easier.

INTRODUCTION

The accurate mass of a molecular ion or its adduct is often used as proof of chemical composition. By convention, instruments with high mass resolving power were used to collect this information.[1] In 1996, Tyler *et al.* showed that accurate mass measurements could be obtained from a low-resolution instrument as long as the peak of interest did not have interfering peaks that would distort its profile.[2]

Advances in computer technology have allowed higher digitization rates for defining ion peak profiles and centroiding through faster data transfer rates and greater storage capacities. Together with internal calibration, molecular ion masses may be obtained with great accuracy without utilizing a high-resolution instrument. In this study, masses within this limit are reported for synthetic compounds using a commercial software that converts ion peaks to a more Gaussian profile.

METHODOLOGY

Solutions of several proprietary synthetic compounds were introduced *via* a direct insertion probe (DIP) or a gas chromatograph (GC) equipped with a Rtx-5ms column. The sample underwent chemical ionization, with methane as reagent gas, and focused into a DSQ quadrupole mass spectrometer (Thermo Scientific). Perfluorotributylamine (PFTBA) was also introduced into the ion source during MS analysis. Each mass spectrum was collected in "profile mode".
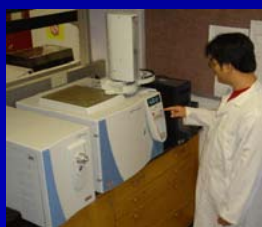
The calibration curve was created in an Excel™ spreadsheet using a minimum of three internal mass ions between *m/z* 69 and 652 (in Figure 1), and was used to calculate the accurate masses for various samples. The results were compared with those obtained with MassWorks™ post acquisition software package where each ion peak profile is evaluated and its centroid mathematically determined. This information was later compared with theoretical isotope patterns, using the Calibrated Line Isotope Profile Search (CLIPS) feature, in order to validate proposed chemical compositions.[3]

Selected References:

1. Webb, K.; Bristow, T.; Sargent, M.; Stein, B. Methodology for Accurate Mass Measurement of Small Molecules: Best Practice Guide; LGC Limited, 2004.
2. Tyler, A.N.; Clayton, E.; Green, B. N. *Anal Chem* **1996**, 68, 3561-3569.
3. Ming Gu; Yongdong Wang; Xian-guo Zhao; Zhe-ming Gu; *Rapid Comm. Mass Spec.* **2006** 20, 764-770.

RESULTS and DISCUSSION

Parameter Development:

Acquired calibration gas spectra showed mass-to-charge deviations even after several attempts to tune and calibrate the DSQ MS (Figure 1). This was attributed to differences in scan parameters during lens optimization and actual data collection. Decreasing the scan rate to 250 scans/s was found to reduce the *m/z* error. Internal calibration remained essential at this optimum rate to correct mass shifts due to sample-inherent interfering ions.
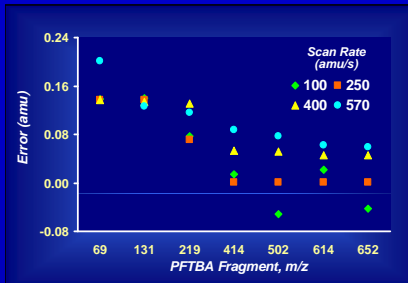

Figure 1. Errors obtained from the average spectrum of a 7-minute acquisition decrease as the scan rates changed from 570 to 100 amu/s.

Analysis of Fatty Acids:

After esterification and GC-MS analysis in electron ionization (EI) mode, a fatty acid (FA) exhibits a molecular ion that is less intense than its fragment ions. To avoid derivatization in this study, a DIP was used to introduce a FA into the ion source, while CI conditions were used to generate the molecular ion and methane gas adducts. Results for stearic acid are shown below.
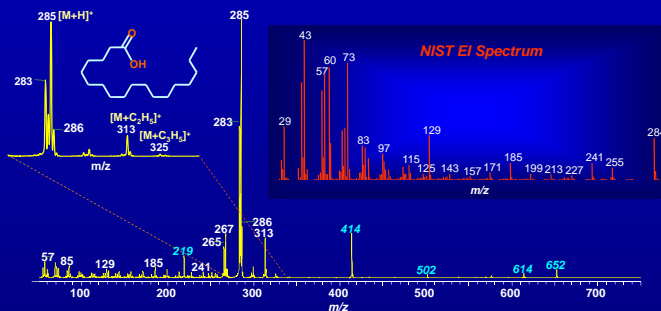

Figure 2. The DIP-CI spectrum of stearic acid (MW = 284) and of the IS PFTBA. The internal standard ions below m/z 219 are present but unlabeled due to FA fragments; these ions are less intense in relative abundance compared to the NIST EI spectrum (inset).

Table 1: CLIPS results for stearic acid

Formula	Monoisotopic Mass (Da)	Mass Error (ppm)	Spectral Accuracy (%)
$C_{16}H_{35}O$	267.2660	-10.4	95.6
$C_{16}H_{35}O_2$	283.2617	-7.1	74.0
$C_{18}H_{36}O_2$	284.2554	-3.1	72.5
$C_{18}H_{37}O_2$	284.2790	-1.2	92.0
$C_{20}H_{41}O_2$	313.3085	-6.9	98.8

- CLIPS parameters:
  - Rings & double bonds = -100 to 100
  - Number of C atoms = 1 to 100
  - Number of H atoms = 0 to 100
  - Number of O atoms = 0 to 100
  - Mass tolerance = 15 ppm
- The [M+H]+, M+ and [M+H]+ ion profiles interfered with each other, affecting their spectral accuracies; in contrast to [M+C₂H₅]+ as shown in Figure 2.
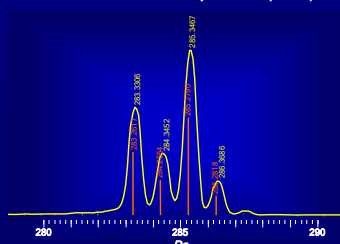

Figure 3. MassWorks™ alignment of the raw and calibrated DIP-CI-MS spectra for stearic acid. On the x-axis, dalton (Da) unit is used since the mass labels do not account for the loss of an electron ($m_{e^-} = 0.00055 u$).

Analysis of Synthetic Compounds:

The accurate mass of the M+ or [M+H]+ ion is preferred by many journals as proof of chemical composition for new synthetic compounds. Fragment ions, in EI and CI modes, are then used to support structure assignment based on NMR. In the methane CI mode, [M+C₂H₅]+ and [M+C₃H₅]+ CI adducts help confirm the assignment of the [M+H]+ ion as shown in Figures 2 & 4. The accurate masses for several compounds, obtained in this study with Excel™ and with MassWorks™, are summarized in Table 2.
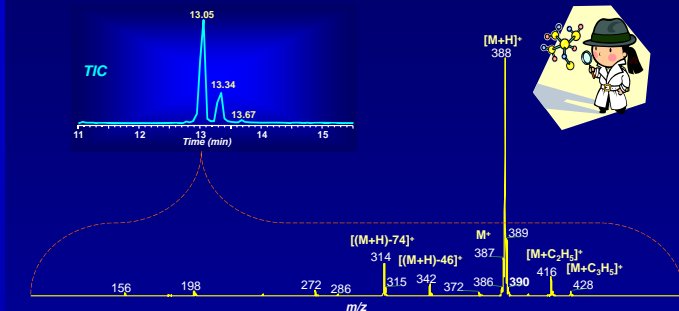

Figure 4. GC-CI-MS analysis showed a proprietary sample had at least three isomers based on their identical CI mass spectra. Neutral losses of 46 and 74 amu were attributed to HOC₂H₆ and HCOOC₂H₆, respectively.

Table 2. Accurate masses of synthetic compounds

Molecular Formula	Theoretical m/z	Method 1[a]		Method 2[b]	
		Measured m/z	Error (ppm)	Measured m/z	Error (ppm)
$C_{14}H_{20}NO_5$	282.1336	282.1347	3.9	282.1348	4.1
$C_{16}H_{22}NO_4$	292.1543	292.1566	7.6	292.1559	5.2
$C_{16}H_{24}NO_4$	294.1700	294.1724	8.3	294.1719	6.3
$C_{23}H_{34}NO_4$	388.2482	388.2454	-7.3	388.2469	-3.6
$C_{23}H_{38}NO_4$	392.2796	392.2813	4.4	392.2799	0.8
$C_{19}H_{15}NOF_3$	330.1120	330.1105	-4.7	330.1133	6.3
$C_{26}H_{36}N$	362.2842	362.2841	-0.3	362.2850	2.0
$C_{25}H_{24}NF_3$	396.1934	396.1916	-4.5	396.1933	0.3
$C_{26}H_{27}NF_3$	410.2091	410.2116	6.4	410.2078	-3.1
$C_{24}H_{22}N_2F_5$	433.1698	433.1714	3.7	433.1687	-2.6

[a] Performed manually using Excel™.
[b] Calculated using MassWorks™ (mass of e⁻ was subtracted).

CONCLUSIONS

- The GC and the DIP temperature-gradient runs minimized interfering ions and complemented the unit resolution of the quadrupole.

- Mass errors less than 10 ppm were consistently achieved with the quadrupole MS for more than 300 compounds (10 was reported here).

- Manual data processing with Excel™ was effective in achieving ppm-level mass accuracy, but operator-dependent and time consuming.

- MassWorks™ shortened the post-acquisition processing time and provided mass errors for several sample characteristic ions.

- The CLIPS feature reduced the list of elemental compositions, making chemical formula selection easier.