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# Rapid, Automated Determination of Elemental Compositions of Ions in Mass Spectra Obtained with an Open-Air Ion Source

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## Introduction

The inexpensive autosampler in Figure 1 passes 76 cotton swab, wipe samples supported by a 3-foot long, aluminum bar through the ionizing beam of a DART/TOFMS. The bar rests on two N-scale model railroad flat cars and is pulled by fish line wrapped around a wheel on the shaft of a 7-rpm motor. The bar is also the core element of the field sample carrier in Figure 2 that simplifies sample collection and provides the swab heads nearly ready for analysis. The 1.8 mL glass vials that protect the swab heads and the linear cell array that contains the vials can be removed in about 5 min after the wipe samples are collected. An analysis time of 6s/sample and elimination of extraction, cleanup, and component separation provide high throughput. Dispersive events, Superfund sites, and clandestine drug labs could be characterized, their remediation monitored in real-time, and their cleanup documented rapidly with high spatial resolution.

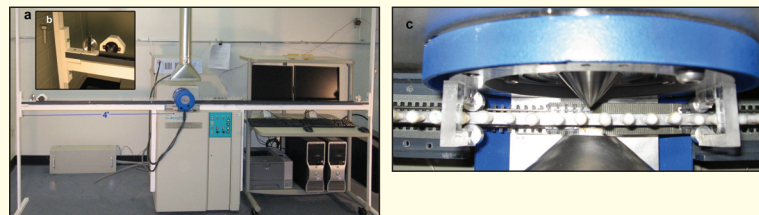


Figure 1. (a) the autosampler; (b) a magnified view of a weight, a vertical adjustment, a pulley, and the motor; and (c) a close-up of the ion source showing two plexiglass and aluminum post, alignment devices that fix the bar location along the ionizing beam axis and prevent rotation of the bar about its axis.

The trade off is that composite mass spectra are often obtained. For dispersed chemicals, rapid identification of one or more analytes is essential for assessing risks to people. Software to deconvolute composite mass spectra was developed to address this need. Precursor and product ions are correlated based on their exact masses and relative isotopic abundances (RIAs).

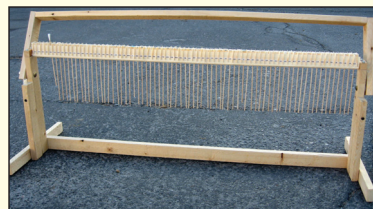


Figure 2. The field sample carrier after collection of four wipe samples. The vial has been removed for the fifth cotton swab, which has been pushed upward just prior to collecting the wipe sample. A wire cutter is used to remove most of the cotton swab stick after a wipe sample is collected.

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## Low, Medium, and High CID Voltages

Grange and Sovocool,<sup>1,2</sup> Suzuki, et al.,<sup>3</sup> and Kaufmann<sup>4</sup> have correlated precursor ions with product ions and their corresponding neutral losses to determine the correct composition for higher-mass precursor ions. To deconvolute composite mass spectra using ion correlation, precursor ions must be classified as such, and product ions must be produced by in-source CID when using a single stage TOFMS.

For N-butylbenzenesulfonamide, the low CID voltage (-15 V) for the top mass spectrum in Figure 3a provided the protonated molecule, an ammoniated adduct ion, a protonated dimer ion, and an ammoniated dimer ion. With the moderate CID voltage (-40 V) for Figure 3a, the precursor ion remained, while the fragile dimeric ions fragmented and were no longer seen. At the highest CID voltage (-70 V) in Figure 3a, the product ions were abundant.

No dimeric ions were seen for the three-component mixture in Figure 3b, but three precursor ions were evident at the two lowest CID voltages. The product ions seen in Figure 3b for the highest CID voltage were produced from all three analytes.

## Multi-Task Software

As illustrated in Figure 4, three computer programs provide output lists of (1) the most abundant ions, precursor ions, adduct and dimer ions, (2) exact masses (and relative isotopic abundances, when ion abundances exceed thresholds) for correlated ions, and (3) unique precursor ion:product ion correlations when multiple precursor ions are found.

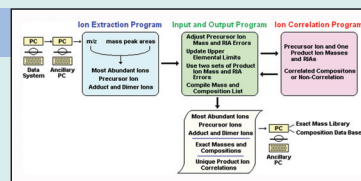


Figure 4. A diagram of the three programs showing their interactions. Importing text files of m/z ratios and mass peak areas, from the data system, yields an output that includes exact masses and corresponding compositions. Comparison of measured exact masses to accurate masses of precursor ions in a library, or comparison of molecular compositions to those in a database, such as SciFinder®, often provides tentative identifications.

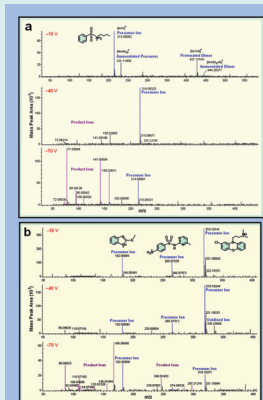


Figure 3. Full scan mass spectra acquired at low (-15 V), moderate (-40 V), and high (-70 V) CID voltages for (a) a single analyte (N-butylbenzenesulfonamide) and (b) a mixture of three compounds (2-(methylthio)benzothiazole, sulfamerazine, and chlorpromazine).

Starting with text files from the data system of the exact masses and mass peak areas from the three mass spectra acquired at low, moderate, and high CID voltages, the Ion Extraction Program (IEP) uses the abundance and exact mass difference tests listed in Table 1 to classify ions as precursor, adduct, dimeric, or product ions.

To ensure only abundant ions are saved and to keep outputs simple and uncluttered, the abundance threshold is increased stepwise until no more than 25 ions are saved when a single precursor ion is found, or no more than 50 ions when multiple precursor ions are found.

Table 1: Precursor Ion Tests. Includes criteria for mass peak area, exact mass, and adduct/dimeric ion tests.

Table 1. Precursor, Adduct, Dimeric, and Product Ion Tests.

Table 2: Lists output by the IEP. Product ions are listed in order of decreasing abundance. Includes columns for Most Abundant Ions, Precursor Ions, and Adducted Precursor Ions.

Table 2. Lists output by the IEP. Product ions are listed in order of decreasing abundance.

Table 3: Possible compositions for the precursor ion from metoprolol and for three product ion-neutral loss pairs. Includes Precursor Ion, Correlations, and Re-correlations.

Table 3. Possible compositions for the precursor ion from metoprolol and for three product ion-neutral loss pairs

RIAs are calculated when certain thresholds are met. For 10,000 or more counts for the monoisotopic ion and at least 500 counts for the +1 and +2 isotopic peaks, RIAs are calculated. If the monoisotopic ion abundance exceeds 100,000 counts, 900 is added to the RIA to serve as a flag that a 15% RIA error limit should be used, rather than a 20% error limit.

Table 2 shows the output of the IEP. The Ion Correlation Program (ICP) tests a precursor ion and a single product ion for correlation (see Figure 5). The possible compositions are determined for each mass and RIAs, if available, and for the mass difference of the corresponding neutral loss within the mass and RIA error limits. If a product ion composition and a neutral loss composition sum to provide a possible precursor ion composition and if a precursor ion composition can yield possible product ion and neutral loss compositions, there is correlation.

The Input and Output Program (IOP) inputs the exact masses and RIAs of a precursor ion and each possible product ion in succession to the ICP starting with the most abundant product ion. For Table 2, the first correlation test was between m/z 319 and m/z 167. Correlations found are saved in memory.

Figure 5. Ion correlation for the m/z 319 precursor ion with the 246 product ion and its corresponding neutral loss, but not with the 167 ion. Rings and double bonds are in parentheses.

Upper elemental limits are determined from the possible precursor ion compositions that remain after each ion correlation. If more restrictive elemental limits were found for correlations after the first one, the cycle is repeated starting with the first product ion using the tightest upper elemental limits found.

In Table 3, four compositions were initially possible for the precursor ion, three compositions after correlation with the m/z 116 ion, two compositions after correlation with the m/z 191 ion, and only one composition after correlation with the m/z 226 ion. During the second cycle through the product ions (re-correlations), only one composition was found for the m/z 116 and 191 ions and for their corresponding neutral losses.

Table 3: Possible compositions for the precursor ion from metoprolol and for three product ion-neutral loss pairs. Includes columns for Precursor Ion, Correlations, and Re-correlations.

Table 3. Possible compositions for the precursor ion from metoprolol and for three product ion-neutral loss pairs

The IOP varies exact mass and RIA error limits to limit the number of precursor ion, product ion, and neutral loss compositions, while not overlooking low-abundance product ions. For precursor ions, exact mass and RIA error limits are relaxed as listed in Table 4 until at least one precursor ion is found. For the product ions, two sets of error limits are used to minimize the number of compositions found for abundant ions that provide exact masses accurate to within 2 mDa, and for low-abundance ions, a 4-mDa mass error limit is used.

Table 4: Exact Mass and RIA Error Limits for the Precursor and Product Ions. Includes columns for Precursor Ion and Product Ions.

Table 4. Exact Mass and RIA Error Limits for the Precursor and Product Ions

Finally, the IOP outputs Tables 2 and 5. Table 5 is a list of the ion correlations. For composite mass spectra, the IOP also provides in Table 5 a list of unique product ion correlations with each precursor ion. Because the three analytes had different sets of heteroatoms, deconvolution was very successful for the three-component mixture produced in Table 5.

Additional details of the software are provided in an article under peer review by the journal Rapid Communications in Mass Spectrometry.

Table 5: IOP Output of the Correlated Ion and Neutral Loss Compositions. Includes columns for Exact Masses & Prec. Counts, Lower Elemental Limits, Upper Elemental Limits, and Neutral Losses.

Table 5. IOP Output of the Correlated Ion and Neutral Loss Compositions.

## References

- 1. Grange AH, Winnik W, Ferguson P, Sovocool GW. Rapid Commun. Mass Spectrom. 2005; 19: 2699.
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