

# Advanced Mass Spectrometry

## Introduction

A compound is tentatively identified when its low resolution mass spectrum matches a single library mass spectrum. Similarity of the mass spectra and retention times for the analyte and a purchased standard verify the compound identification. Unfortunately, problems can occur. Most analytes are not found among library mass spectra. Multiple plausible matches often occur. A low level of an analyte in a complex environmental extract that coelutes with another compound can yield a poor quality mass spectrum. Finally, the mass spectrum may contain only a few ions, which makes spectral matches less certain. Ion Composition Elucidation (ICE), a new high resolution mass spectrometric technique developed by ESD scientists, often allows the identification of compounds under these non-ideal circumstances. Quadrupole and ion trap mass spectrometers provide nominal (nearest integer) masses for compounds. Hence,  $N_2^+$ ,  $CO^+$ , and  $C_2H_4^+$  are indistinguishable with a mass of 28. However, the exact masses of these ions are 28.00615, 27.99491, and 28.03130 atomic mass units (amu). High mass resolution mass spectrometers measure such exact masses and easily determine the compositions of these ions, even when all three are present simultaneously. But when the number of possible compositions increases exponentially with an ion's mass and above 150 amu, multiple compositions are possible. In addition, until now, exact masses of ions have been determined using probe introduction or peak matching, techniques which require about 1 min, thereby precluding the measurement of exact masses as compounds elute from a gas chromatograph.

## Instrumentation and Software

Using a Finnigan MAT 900S double focusing mass spectrometer, the data acquisition technique of ICE, Mass Peak Profiling from Selected Ion Recording Data (MPP-SIRD), acquires data from which exact masses are calculated for multiple analytes during a GC/MS run. MPP-SIRD also provides the 100-fold sensitivity enhancement of selected ion recording relative to full scanning and allows resolving powers of up to 20,000 to be used routinely. This provides excellent mass selectivity to discriminate against mass interferences that are commonly seen for complex environmental extracts. The full potential of MPPSIRD for determining ion compositions is realized by measuring not only the exact mass of an ion, but in addition, the exact masses of the mass peak

profiles greater in mass by 1 and 2 amu that arise from atoms of heavier isotopes ( $^{13}C$ ,  $^2H$ ,  $^{15}N$ ,  $^{17}O$ ,  $^{18}O$ ,  $^{33}S$ , and  $^{34}S$ ). Also measured are the abundances of these two profiles relative to the profile of the ion that contains only the lowest isotopes of its elements. The three measured exact masses and two relative abundances are entered into an in-house Profile Generation Model (PGM) to determine the ion compositions that provide calculated values consistent with the measured values. Usually, only one composition is possible for ions with masses less than 600 amu. The 4-fold increase in the mass limit for which ion compositions can be determined provided by ICE greatly enhances the utility of HRMS for identifying compounds.

## Applications

### Site Characterization

A small amount of a tar-like sample from a Superfund site in West Virginia was dissolved in methylene chloride and injected onto the GC/HRMS. ICE was used to determine the compositions of 51 apparent molecular ions, most of which contained NS,  $N_2S$ , or  $N_2S_2$ . Many of the mass spectra contained the  $C_8H_7NS^+$  ion characteristic of alkyl benzothiazoles. These results suggested the bulk of the hazardous waste was still bottom material from a dye or rubber plant. A nearby dye plant was indeed responsible for the waste.

### Compound Identification

An increased incidence of childhood cancer was observed in Toms River, NJ. A municipal well that serviced 50,000 customers located 1 mile from the Reich Farms Superfund site contained several isomeric compounds that were not identified by conventional mass spectrometry. ICE provided the compositions of the molecular ion and 10 fragment ions. Mass spectral interpretation of the fragment ion compositions indicated that the compounds had a tetralin core with attached cyano and alkylcyano groups. With this information in hand, the corporation with an interest in our study provided us a standard from a polymerization process currently used to produce acrylonitrile:styrene polymers that contained three of the five isomers found in the well water. The isomers were 2:1 acrylonitrile:styrene adducts. Region 2 supervised toxicity studies, after the compound identities and their source were known.

# Advanced Mass Spectrometry

---

## Disinfection Byproducts

The mass spectrum of a brominated compound found in chlorine-disinfected well water displayed only a molecular ion and one fragment ion above the chemical noise. ICE determined that the molecular ion composition was  $C_{24}H_{31}O_2Br$ . Replacing the Br atom with an H atom yielded a composition consistent with Quinbolone, an anabolic steroid that enhances growth in cattle. A feed lot was located nearby. Use of ICE led to a plausible hypothesis for the compound's identity, where conventional mass spectrometry could not.

## Risk Assessment

Extracts of malodorous monitoring well water from the Tower Chemical Superfund site near Orlando, FL, contained numerous compounds. A pesticide manufacturer had used DDT as a feed stock to produce two marketable pesticides. ICE determined the molecular ion compositions and provided confirmation of library matches for a group of compounds containing Cl atoms, a group containing S atoms, and a group containing both Cl and S atoms.

The pesticide manufacturer was responsible for the first group, anaerobic bacteria were probably responsible for the second group, and biotransformation may have produced the compounds containing both Cl and S atoms. Estimates of the relative amounts of the compounds present based on peak areas in the total ion chromatograms were also compiled. Region 4 is using this information in its risk assessment of the site.

## Environmental Forensics

ICE provides a listing of the ion compositions of the ions in a mass spectrum and limits a compound's identity to one of a usually small number of isomers. These ion compositions and the retention time provide compelling evidence for tracking a compound upstream (or up a sewer system) to its source. Once the source is known, the compound is more likely to be identified. Even if the compound were not identified, toxicological tests could still be made and remediative steps could be taken, if needed.

## Technology Transfer

Currently, ESD is the only laboratory performing ICE. ESD plans to transfer this technology to other laboratories, especially to the EPA Regional labs that have double focusing mass spectrometers. Most of the MPPSIRD software is written in Lotus 1-2-3 (v. 9.0), while the PGM is written in Quick Basic 4.5.

Consequently, only the data system code for different mass spectrometers must be revised for a successful transfer of ICE to other instruments. ESD anticipates that a week working with an instrument's operator would be sufficient to make ICE operational.

## Availability

ESD's double focusing mass spectrometer, data acquisition and interpretation tools (ICE), and interpretive mass spectroscopists provide a unique resource for investigating environmentally important analytical problems. The EPA's regional personnel are invited and encouraged to take full advantage of this resource through the ESD's Technology Support Center to solve otherwise intractable analytical problems.

## References

<http://www.epa.gov/nerlesd1/chemistry/ice/default.htm> provides access to a dozen articles and a dozen posters that fully describe MPPSIRD, the PGM, and their application to several analytical problems, including those mentioned above.

Also accessible from this URL are "ICE is Nice," a narrated, animated PowerPoint presentation describing the scientific basis for ICE with three applications, and "ICE is Easy," which illustrates stepwise use of ICE with a Finnigan MAT 900 or MAT 95 data system.

## For Further Information

For questions about ICE or its availability to solve analytical problems encountered in your lab, contact: Dr. Andrew Grange, U.S. Environmental Protection Agency, P.O. Box 93478, Las Vegas, NV 89193-3478, Phone: (702) 798-2137, Fax: (702) 798-2142, email: [grange.andrew@epa.gov](mailto:grange.andrew@epa.gov), or Dr. Wayne Sovocool, U.S. Environmental Protection Agency, P.O. Box 93478, Las Vegas, NV 89193-3478, Phone: (702) 798-2212, Fax: (702) 798-2142, email: [sovocool.wayne@epa.gov](mailto:sovocool.wayne@epa.gov).