the Analytical Scientist

The Application Book 2022

Recent application notes across a range of exciting topics

Please click the circles to navigate

Micromilling of Uniform Nanoparticles for Space Applications

A NASA lab's ability to develop optimized ceramic nanoparticulate materials for demanding research projects have been enhanced by FRITSCH micromills

By Curtis W. Hill and Lee Allen

The challenge of developing new materials and processes demands laboratory equipment with advanced capabilities. The ability to produce uniform nanoparticles is critical for development of advanced ultracapacitors for energy storage, thermoelectric devices with high figure-of-merit, and materials for NASA's Nuclear Thermal Propulsion system. We have been working to develop much smaller and more uniform particles with Fritsch's Pulverisette Line of micromills. These are capable of producing ultrafine grinding results, down into the nanometer particle size range.

Our laboratory at NASA Marshall Space Flight Center (Huntsville, Ala.) develops materials and processes for NASA's exploration missions and the International Space Station. This involves developing and optimizing materials prop-erties for very demanding applications in energy storage, power generation, and other advanced application areas.

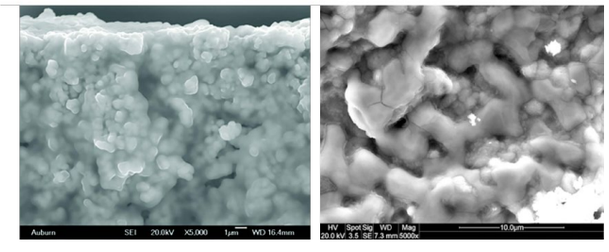
The challenge of developing new materials and processes demands laboratory equipment with advanced capabilities. For instance, the ability to produce uniform nanoparticles is critical for our development of advanced ultracapacitors for energy storage, thermoelectric devices with high figure-of-merit, and materials for NASA's Nuclear Thermal Propulsion system.

However, ceramic powder as-received from suppliers typically has a fairly wide range of particle sizes and is not consistent enough for our high-performance materials research. We have investigated and tested various milling machines and processes, including ball mills and vibratory mills; and although these techniques help reduce D50 particle size as well as improve particle size distribution, the resulting powders are still of insufficient quality for our demanding research.

We have been working with Fritsch's Pulversette line of micromills for the past couple of years to develop much smaller and more uniform particles – these micromills are capable of ultrafine grinding results down into the nanometer particle size range (Figure 1). The laboratory-size mills use smaller, very hard media to achieve extraordinary milling energy.

We use stainless-steel milling bowls lined with zirconia, although mills are available in several other materials and capacities depending upon the materials to be milled. The micromill uses rotation speeds of up to 1,100 rpm and an acceleration force of 95 g for a resulting energy application roughly 150 percent greater than that of classic planetary mills. This extraordinary milling energy results in more economical and efficient milling of particles, providing us with considerably finer grinding results in shorter times.

Figure 1



View the full application note online

LINKS

Videos
Product, Application and Support Videos

Article
Rock, Paper, Scissors: Ways that Milling and Sample Prep Affect our Everyday Lives

Sponsored by FRITSCH

Produced by the Analytical Scientist

Quick Comparisons: ChromaTOF® Sync

Speed up and enhance data comparisons. Strip the busywork away so you can reach better conclusions, faster, with *ChromaTOF* Sync.

Considered by many the national liquor of China, Baijiu is a clear distilled spirit with a long history. It is one of the world's most consumed spirits, and its price can vary dramatically, from the equivalent of a can of beer to thousands of dollars for a single bottle. This variation largely comes from how each baijiu distillery uses its own qu, or fermentation starter. The different qu help create different aroma profiles, which vary by region and in value.

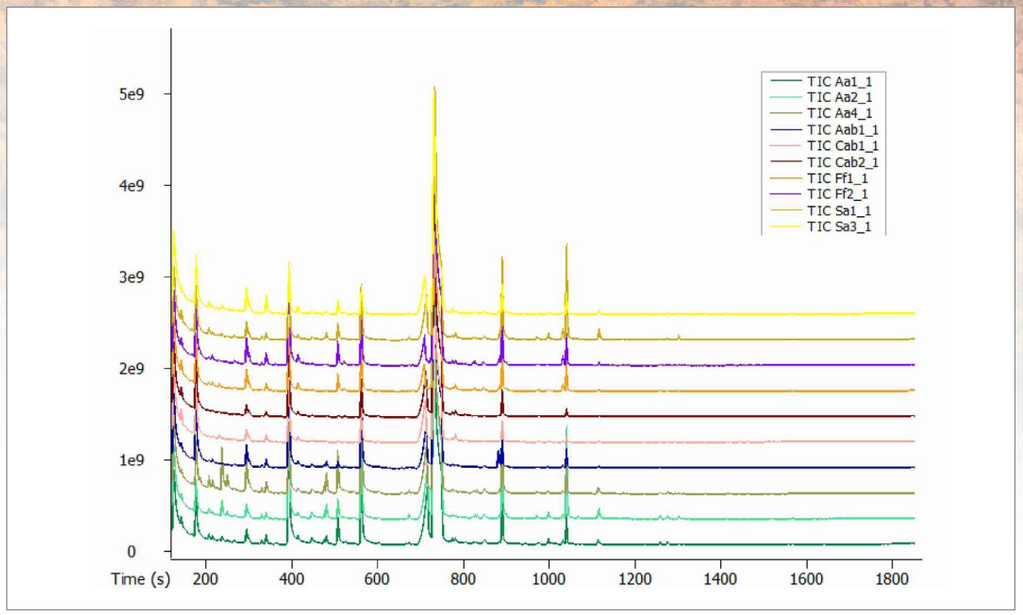
Being able to accurately determine the chemical composition of baijiu samples allows for more confidence in quality and process monitoring.

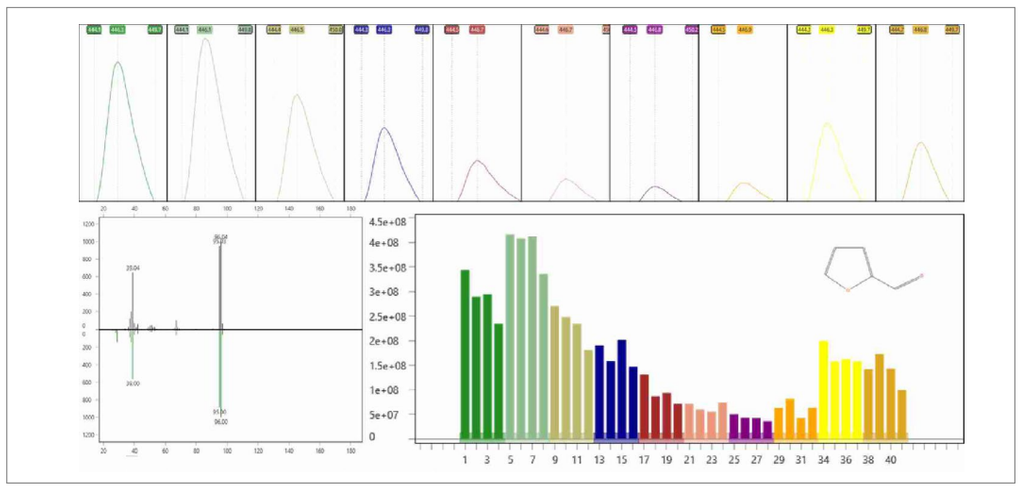
Different baijiu types do tend to be very similar. When 10 baijiu samples were analyzed on a Pegasus® BT, 10 variations of basically the same chromatogram were generated.

Rather than processing this data one sample at a time, the data were analyzed with *ChromaTOF* Sync, a new statistical analysis tool. Sync was able to quickly produce a composite sample set peak table, synchronizing the chromatograms for a quick visual overview of the differences. This could then be taken a step further, revealing individual analyte trends throughout the sample set, highlighting statistically significant differences and confirming the correlation of region and aroma profile. Sync is able to pull out single features and display side-by-side profiles, mass spectral comparisons, and bar charts of that same feature throughout all of the data provided, such as this closer look at variation of furfural (CAS: 98-01-1, RI: 833), compared at m/z 95.

A straight-forward and effective approach to challenges – the incredibly complex baijiu samples allows laboratories to explore and interpret their data in the most efficient ways: time-efficient, labor-efficient, and ultimately, cost-efficient.

View the full application note online





LINKS

Pegasus BT GC-TOFMS
Benchtop Gas Chromatography Time-of-Flight Mass Spectrometer

Sponsored by LECO EMPOWERING RESULTS

Produced by the Analytical Scientist

Distinguishing Oxidative Impurities from Ionizable Lipids Used in LNP Formulations Using EAD

Achieving structural elucidation and distinction of oxidative isomer impurities of lipids with electron-activated dissociation (EAD) and the ZenoTOF 7600 system

By Adam Crowe, Nikita Jain, Rehan Higgins, Robert Proos, Matthew Stone, and Kerstin Pohl

In this application note, the comprehensive characterization of impurities from the ionizable lipid (6Z,9Z,28Z,31Z)-heptatriaconta-6,9,28,31-tetraene-19-yl-(dimethylamino) butanoate, commonly known as DLin-MC3-DMA (MC3), is presented. Deep structural elucidation, including the localization of different oxidation products and saturation of double bonds in MC3, was achieved using EAD.

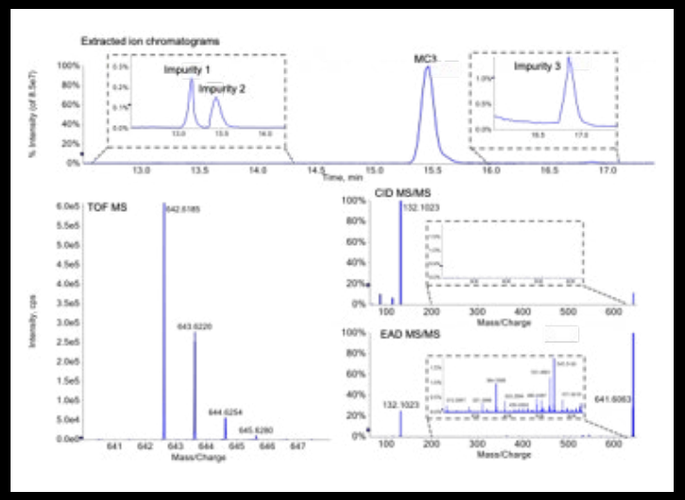


Figure 1. Structural confirmation of MC3 by LC-MS/MS using EAD

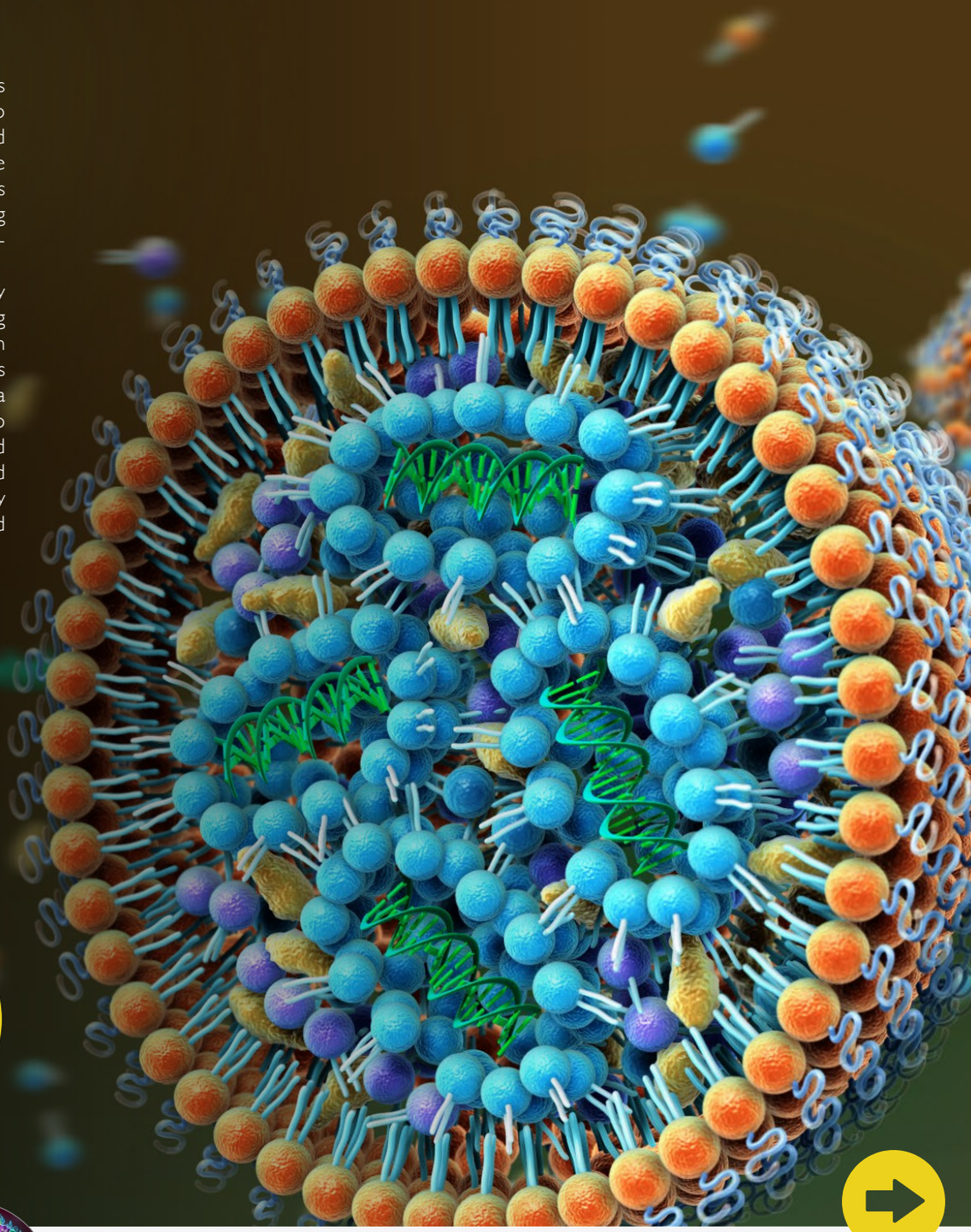
Lipid nanoparticles (LNPs) that are comprised of ionizable lipids are used to deliver oligonucleotides to work as therapeutics or to stimulate the immune system, as seen in the initial mRNA-based COVID-19 vaccines. To ensure product quality, detailed and sensitive characterization of the ionizable lipid and its related impurities is necessary. However, obtaining the level of detail needed is challenging with current liquid chromatography-mass spectrometry (LC-MS)-based methodologies.

In this application note, we will explore how EAD can efficiently provide complete characterization of different naturally occurring lipids in a single LC-MS run. The applicability of this novel fragmentation mode for the detailed characterization of lipids used for LNPs was tested using MC3 and its related impurities as a model. Within a single experiment, the exact locations of oxygen incorporation of two isomeric species and the double bond reduction of another related impurity were pinpointed using the unique fragment ions produced by EAD. This information can be used to determine drug efficacy and safety from formulated LNPs. Additionally, it can be used to aid rational design of new synthetic lipids.

Key features of lipid impurity characterization with EAD:

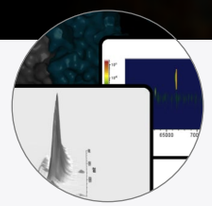
- Confident detection of low-level impurities with the ZenoTOF 7600 system and more than 5 orders of dynamic range
- Information-rich MS/MS spectra for definitive and distinct structural elucidation of related and isomeric singly charged lipid species with EAD
- Detection and identification of lipid impurities at levels below 0.01 percent relative abundance of the main peak by greatly enhanced fragment detection with the Zeno trap.

View the full application note online

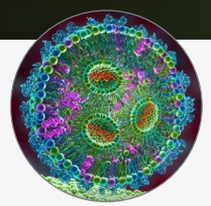


LINKS

On-demand webinar
Handle with care: Ensuring LNP lipid quality for better genetic medicines



Resource Center
ZenoTOF 7600 system biopharma resource center



Overview
mRNA-LNP workflow overview

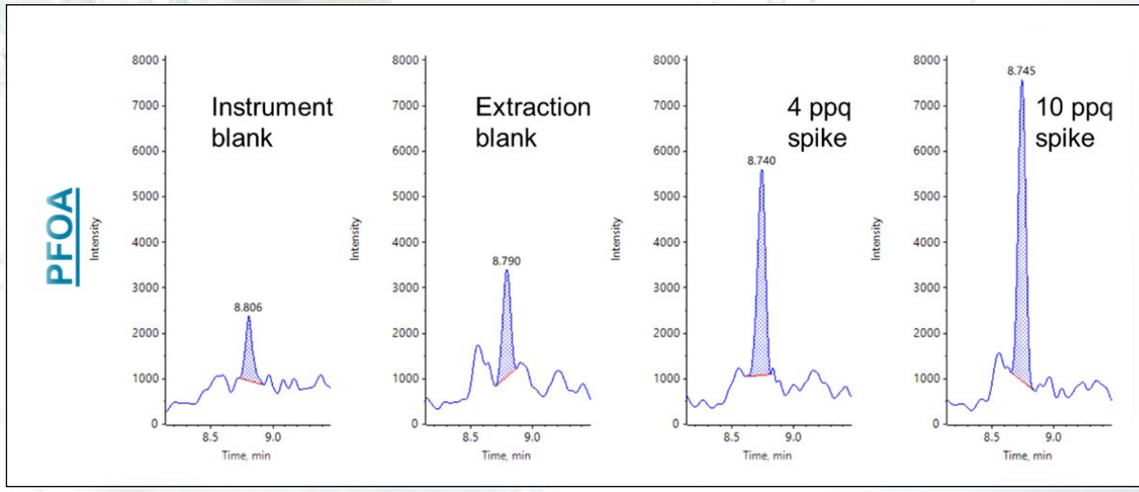
Sponsored by SCIEX The Power of Precision

Produced by the Analytical Scientist

Achieving Low Parts-Per-Quadrillion Detection Limits for PFAS Analysis in Drinking Water

A collaborative exploration of instrument sensitivity and sample preparation for ultra-trace analysis of PFAS on the SCIEX 7500 system

By Thep Phomsopha, Simon Roberts, Craig M. Butt, Karl Oetjen, Matt Noestheden, Sam Lodge, and Andrew N. Patterson



View the full application note online

A collaborative project between SCIEX and Eurofins Environment Testing is described to meet the 2022 US EPA drinking water health advisory levels (HALs) for 4 PFAS compounds using the SCIEX 7500 system. Sample preparation and instrumental methods were developed to achieve low parts-per-quadrillion (ppq) HALs – which is pg/L equivalent – for perfluorooctanoic acid (PFOA, 4 ppq), perfluorooctane sulfonic acid (PFOS, 20 ppq), perfluorobutane sulfonic acid (PFBS, 2 ppb) and hexafluoropropylene oxide dimer acid (HFPO-DA, or GenX, 10 ppt).

PFAS are widely detected contaminants in drinking water and the recent 2022 US EPA drinking water health advisory levels (HALs) set low parts-per-quadrillion (ppq, or pg/L) HALs for PFOA and PFOS. HALs for PFAS are not enforceable regulatory limits but indicate the levels below which adverse health effects are not anticipated, when considering lifetime exposure.

The extremely low HALs require an unprecedented level of cleanliness and instrumental sensitivity. As such, extensive measures were performed to minimize PFAS contamination. The extensive contamination mitigation steps during the sample preparation and instrumental analysis resulted in blank levels that were below the 4 ppq spike level, allowing for accurate and unbiased quantification. Extraction blanks did not show any detectable PFOS and only minor traces of PFBS (0.5-1.2 ppq). PFOA was consistently detected in the extraction blanks at an average of 1.9 ppq (range: 0.5-2.7 ppq), which represented 35 percent of the calculated 4 ppq spike concentration.

The method extraction spikes (n=2) showed excellent recovery and precision at 4 ppq for PFOA, PFBS and HFPO-DA, and at 20 ppq for PFOS. Specifically, the mean extraction recoveries at the 4 ppq spike were: 79.3 percent for HFPO-DA (%CV = 6.2 percent), 139 percent for PFBS (%CV = 19.5 percent) and 138 percent for PFOA (%CV = 0.8 percent). The mean recovery for PFOS at the 20 ppq spike level was 113 percent (%CV = 0.9 percent). These results indicate that the method can achieve the low ppq EPA drinking water health advisory levels for PFOA and PFOS and is several orders of magnitude lower than the levels for HFPO-DA and PFBS.

LINKS

SCIEX 7500 system

PFAS applications and resources

Sponsored by SCIEX The Power of Precision

Produced by the Analytical Scientist

Enhancing Protein Quantification with Microflow Chromatography and Zeno SWATH Data-Independent Acquisition

Exploring the impact of the Zeno trap innovation on quantitative proteomics using microflow LC-MS/MS with SWATH data-independent acquisition workflow

By Christie Hunter and Alexandra Antonoplis

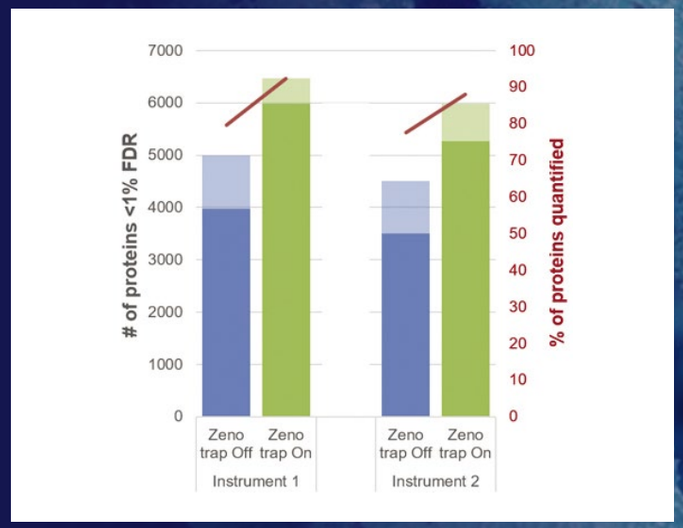
Data independent acquisition (DIA) continues to grow as a key workflow in the field of quantitative proteomics. Here, the improvements in proteins identified and quantified using microflow SWATH DIA coupled with the Zeno trap technology on SCIEX Zeno 7600 system is described. Large improvements in the numbers of proteins quantified were observed when the Zeno trap was activated, especially at the lower sample loadings.

As the field of quantitative proteomics continues to evolve, larger biological cohorts are being studied, often using precious samples obtained from biobanks or other difficult-to-obtain sources. This creates two workflow requirements: the need to acquire quantitative data on the digested samples faster and the need to use smaller amounts of samples. For these types of studies, data-independent acquisition (DIA) continues to grow as the workflow of choice for reproducible quantitative analysis of large numbers of proteins from a proteomic sample.

The Zeno trap is a key innovation from SCIEX on the ZenoTOF 7600 system, increasing the instrument's duty cycle to >90 percent across the entire mass range. Thus, it provides large gains in MS/MS sensitivity and enables significant improvements in protein identification workflows, targeted peptide quantification workflows and the SWATH DIA workflow.

Before, the combination of fast microflow chromatography and SWATH DIA enabled the quantification of large numbers of proteins from complex proteomics samples at very high rates, up to 100 samples per day. Now, coupling SWATH DIA with Zeno MS/MS and microflow LC has the potential to enhance a core quantitative proteomics workflow even further.

Here, the use of microflow chromatography in combination with Zeno SWATH DIA was optimized and evaluated compared to traditional SWATH DIA to characterize the impact of the Zeno MS/MS sensitivity gains. Four different gradient lengths (5, 10, 20 and 45 minutes) were tested to cover a range of application needs. Large improvements in the numbers of proteins quantified were observed when Zeno trap was activated, especially at the lower sample loadings. For instance, Zeno SWATH DIA data processed with DIA-NN software permitted up to ~6,100 proteins to be identified using a 45-min microflow LC gradient, with ~95 percent of those proteins quantified at <20 percent CV.

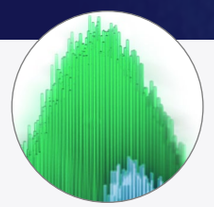


Large improvements in proteins quantified with Zeno SWATH DIA with microflow LC. The numbers of proteins identified at <1% FDR (transparent) and quantified at <20% CV (solid) using SWATH DIA were compared with and without the Zeno trap activated for a 45-min gradient and at 400 ng of sample load. Approximately 50% more proteins were quantified using Zeno SWATH DIA on both instruments. In addition, ~90% of identified proteins were quantified at <20% CV (red line).

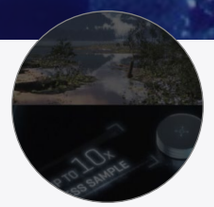
View the full application note online

LINKS

Zeno Trap and EAD



Zeno SWATH DIA



The Pursuit of Ideation and Innovation

Sponsored by SCIEX The Power of Precision

Produced by the Analytical Scientist

Multiresidue Pesticide Analysis with the 6475 Triple Quadrupole LC-MS System

Our comprehensive LC-MS-MS method tackles over 500 pesticide analyses in three food matrices

By Linfeng Wu and Aimei Zou

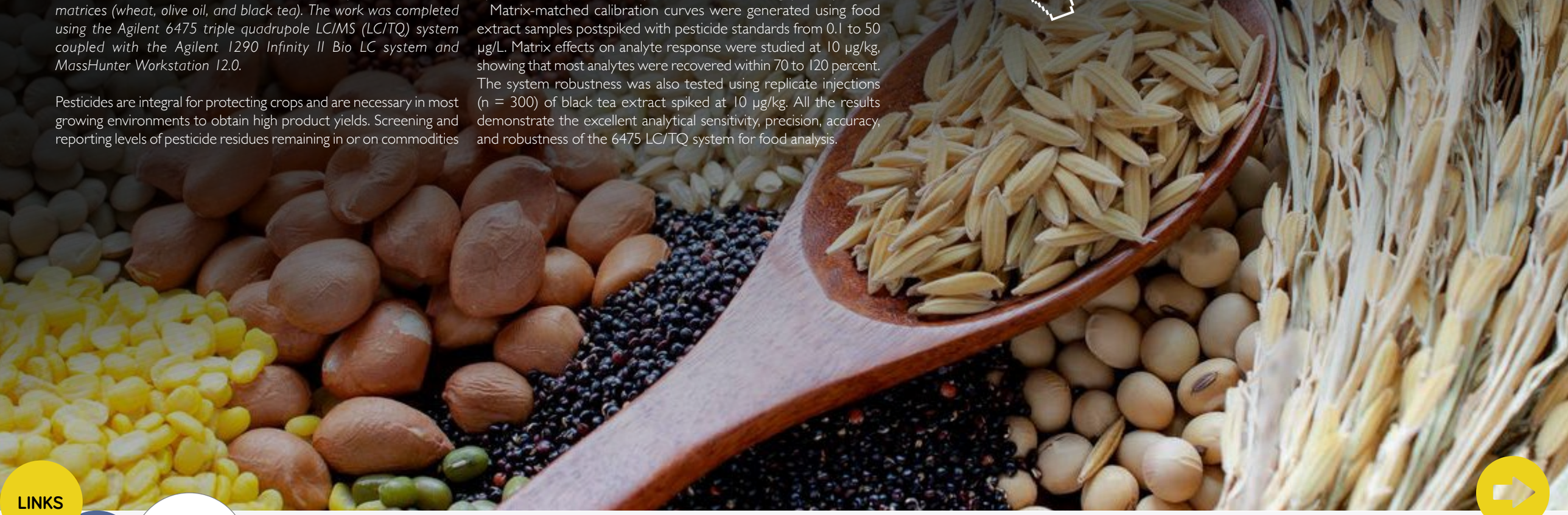
Here, we describe the development and evaluation of a comprehensive LC-MS-MS method for over 500 pesticide analyses in three food matrices (wheat, olive oil, and black tea). The work was completed using the Agilent 6475 triple quadrupole LC/MS (LC/TQ) system coupled with the Agilent 1290 Infinity II Bio LC system and MassHunter Workstation 12.0.

Pesticides are integral for protecting crops and are necessary in most growing environments to obtain high product yields. Screening and reporting levels of pesticide residues remaining in or on commodities are required by many regulatory bodies, e.g., the US-EPA and the European Commission. Major challenges for pesticide analysis in food include many pesticides from various compound classes, diverse complex food matrices, matrix effects, low concentrations of target analytes, and so on.

This application note describes the development and evaluation of an LC/MS/MS method for over 500 pesticides and pesticide metabolites quantitation in three food matrices. These matrices include wheat (high starch content), olive oil (high oil content), and black tea (difficult matrix). The analysis was performed using the Agilent 6475 triple quadrupole LC/MS system (LC/TQ) coupled with Agilent MassHunter Workstation software 12.0. An Agilent 1290 Infinity II Bio LC system and an Agilent Jet Stream (AJS) electrospray ion source were used with the LC/TQ system. The instrument was operated in a dynamic multiple reaction monitoring (dMRM) mode.

Matrix-matched calibration curves were generated using food extract samples postspiked with pesticide standards from 0.1 to 50 µg/L. Matrix effects on analyte response were studied at 10 µg/kg, showing that most analytes were recovered within 70 to 120 percent. The system robustness was also tested using replicate injections (n = 300) of black tea extract spiked at 10 µg/kg. All the results demonstrate the excellent analytical sensitivity, precision, accuracy, and robustness of the 6475 LC/TQ system for food analysis.

View the full application note online



LINKS

Triple Quadrupole LC/MS
6475 Triple Quadrupole LC/MS

Sponsored by Agilent

Produced by the Analytical Scientist

LC/Q-TOF Analysis and Nontargeted Chemometric Profiling of Meats and Plant-Based Alternatives

Food sensory testing using the Agilent 1290 Infinity II LC and Agilent 6546 LC/Q-TOF over 500 pesticide analyses in three food matrices

By Toh Seok Hwa, Upendra Khurana, Tarun Anumol, and Daniel Cuthbertson

This application note describes a nontargeted profiling method to characterize chemical components of animal and plant-based foods, using a high-resolution accurate mass LC-Q/TOF. Also, various statistical tools are presented that translate accurate mass LC/Q-TOF data into more easily understandable information.

Meat-alternative sources of protein, including plant-based and cell-based foods, are gaining popularity globally due to a combination of consumer interest, regulatory changes, and global food systems. For example, as Singapore aims to achieve 30 percent of its food production levels through self-production by 2030, many established food companies and startups are developing meat-substitute products.

The main drivers for alternative meats and proteins are concerns around environmental sustainability, future food shortages, consumer acceptance, and a demand for more healthy and nutritious foods, among others. Historically, plant-based meat substitute foods have struggled to achieve the same texture and taste as animal meats. However, recent analogs of plant-based meats are significantly more similar in taste, texture, nutrition and composition as traditional meats due to technological advances in production methods.

This application note describes a nontargeted profiling method to characterize chemical components of unknown foods, using a high-resolution accurate mass LC-Q/TOF. Also, various statistical tools are presented that translate accurate mass LC/Q-TOF data into more easily understandable information. Principal component analysis (PCA) of the data can be used to identify compounds, abundance distribution of the compounds in different samples, and how the compounds correspond to target taste profiles. Heat maps and hierarchical clustering of raw ingredients show similar distribution of proteins with target taste profiles.

View the full application note online

LINKS

Quadrupole Time of Flight LC/MS
6546 LC/Q-TOF spectrometry

Sponsored by Agilent

Produced by the Analytical Scientist

Selecting the Optimal Column for Native SEC-MS of Monoclonal Antibodies

How columns affect ionization efficiency

By Andrea Krumm, Tosoh Bioscience GmbH

Three different size exclusion columns were compared for their compatibility with mass spectrometry. The TSKgel UP-SW3000-LS column was superior to other columns due to its low shedding and long-lasting high ionization efficiency.

Characterization of monoclonal antibodies (mAbs) is essential to product safety and efficacy. Size exclusion chromatography (SEC) coupled with mass spectrometry (MS) is increasingly used to identify the accurate molecular mass of mAbs and its impurities. However, traditional SEC generates high particle shedding, which decreases ionization efficiency over time. To avoid shedding for MS and multi-angle light scattering (MALS) applications, Tosoh Bioscience developed TSKgel® UP-SW3000-LS U/HPLC size-exclusion columns. In this application note, the column was coupled with an MS instrument for the analysis of a mAb standard. Data demonstrate that the TSKgel UP-SW3000-LS column surpasses competitive UHPLC columns and a dedicated low shedding column for SEC of proteins in terms of particle shedding observed by MS. Moreover, the column helps maintain ionization efficiency in the electrospray ionization (ESI) source >90 percent compared to the initial injection over >50 injections, thus increasing data quality and reducing ion source cleanings.

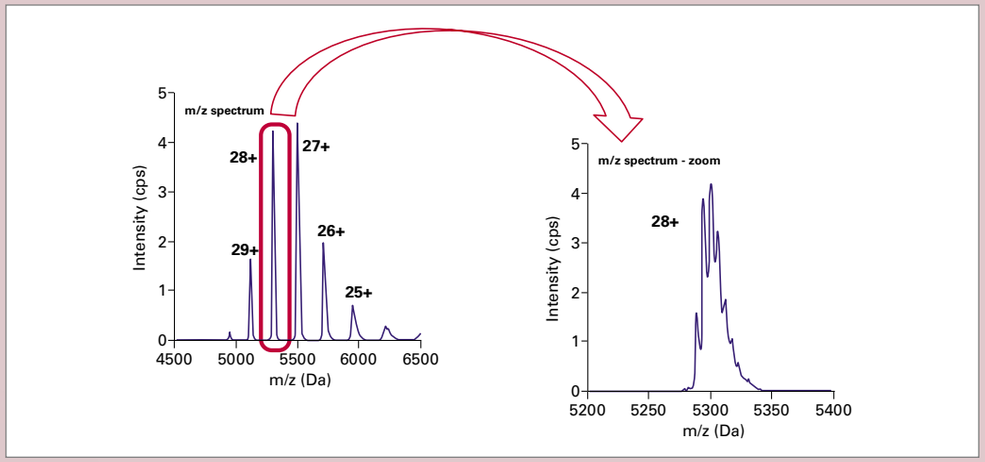


Figure 1. SEC-MS analysis of a mAb

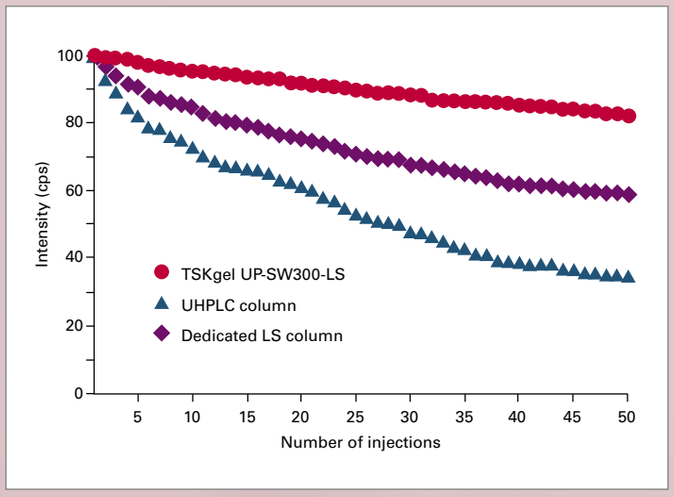
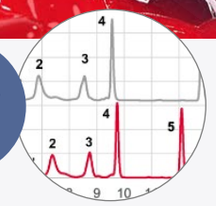


Figure 2. Electrospray ionization efficiency of a mAb

View the full application note online

LINKS



Product
UHPLC size exclusion column for protein analysis by light scattering and mass spectrometry

Webinar
Tosoh Tuesday - SEC analysis of mAbs: advanced detection – advanced columns

Sponsored by TOSOH

Produced by the Analytical Scientist

Monitoring Structural Changes in Polysaccharides Using SEC-MALS

Characterization of linear and branched polysaccharides with SEC-MALS

By Subin Damodaran, Tosoh Bioscience GmbH

The LenS3 MALS detector reveals structural differences in polysaccharides, such as linear conformation versus branching, by providing MW and Rg over the broadest range with superior sensitivity

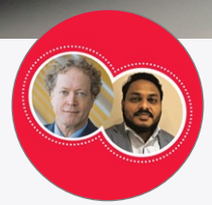
As the most abundant natural biopolymer, polysaccharide's unique chemical and physical properties, as well as their excellent biocompatibility, make them materials of choice in many industries. Due to their wide application range and the complexity of their structure, such polymers need to be examined very thoroughly to fully understand their molecular characteristics. A great variety of conformations and branching behaviours make specific polysaccharides either suitable or problematic for certain applications.

This study demonstrates that structural differences in polymers can be investigated in depth by size exclusion chromatography (SEC) coupled with multi-angle light scattering (MALS) with pullulans and dextrans as examples. In the given example, higher MW dextrans tend to exhibit increased branching on their backbone, leading to the formation of a more compact structure in solution. Ultimately, this results in higher retention volumes and lower Rg values compared to linear pullulans of similar MW. In practice, elucidating structural changes in the low MW and low Rg region requires a light scattering instrument with high sensitivity and capable of detecting very slightly anisotropic scattering, such as the LenS3 MALS detector.

View the full application note online

LINKS

Product Information
EcoSEC Elite
GPC System



Webinar
Tosoh Tuesday: Put your polymers for biomedical applications under the right light

Sponsored by TOSOH

Produced by the Analytical Scientist

Streamlining Forced Degradation Data Management in Drug Development

Accelerating stability testing and supporting project management with decision support software

By Joseph DiMartino and Jesse Harris

Forced degradation studies are an essential task in pharmaceutical development. This work requires consolidating large amounts of analytical and chemical data, which is often spread across multiple teams. This application note describes how decision support software can support forced degradation research.

Forced degradation studies determine the stability of pharmaceutical compounds and are necessary to confirm the efficacy, quality, and purity of drug products. These studies occur during pharmaceutical development, managed by teams that are often spread across multiple locations and use incompatible electronic systems to capture information. Teams typically use general-purpose software such as Microsoft Excel to share data. While these applications are commonplace, they are unsuited for forced degradation research since they lack chemical intelligence, cannot process analytical data, and do not enable collaboration.

Luminata® allows researchers to consolidate process and analytical data in one interface. It also includes several tools that support forced degradation research.

Structural information capture

Scientists will first use tools to predict degradant pathways to plan stress testing conditions. Degradants are typically stored as an SDfile or Word document within a file sharing system. These structures and associated metadata are therefore disconnected from the observed molecules, meaning this data is not easily accessible.

With Luminata, you can link the API with all degradants predicted in different conditions. Theoretical degradants generated by third-party applications (e.g., Lhasa Zenith) are represented in degradation pathways. Luminata stores this predicted information alongside analytical data for each forced degradation study so you can compare experimental and predicted results.

Simplifying data handling

Forced degradation studies require a pre-determined set of tests. Luminata's Forced Degradation Wizard allows you to select from a list of experiments to run, in addition to custom studies needed for a specific project. This simplifies data management, increases consistency, and reduces the risk of error.

Project management support

Once all forced degradant records have been created, they can be viewed in the Dynamic Project Map (Figure 1). Project progress is displayed using a stoplight system, enabling you to see the status of the forced degradation study. This supports project management and avoids duplicative experiments. Metadata, such as ELN and registration numbers, can also be included.

Take the stress out of stress testing

Luminata allows researchers to consolidate scientific data and supports forced degradation studies with specialized tools. The software streamlines data management and enables collaboration, leading to more effective research.

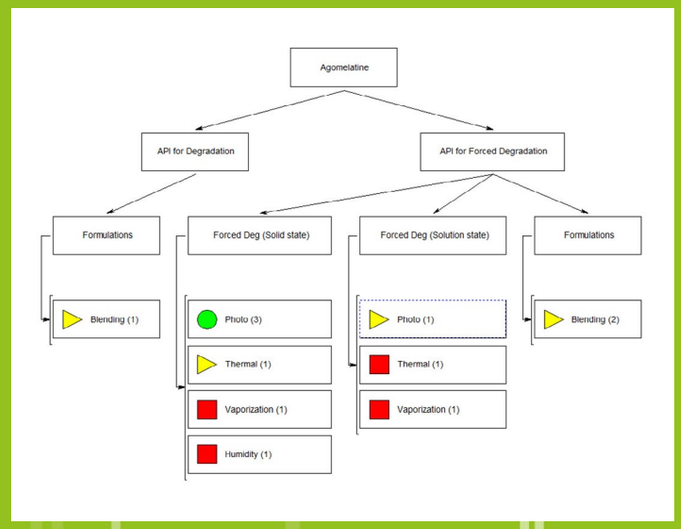
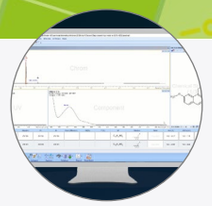


Figure 1: Luminata's forced degradation Dynamic project map

View the full application note online

LINKS

Case Study
Real Results from Bridging Gaps in Data Management



Product Page
Enterprise Decision Support Software for Pharmaceutical & Chemical Product Development

Sponsored by ACD/Labs

Produced by the Analytical Scientist

Eliminating the Logistical Challenges of NMR Data Processing with Browser-Based Software

Implementing Spectrus JS in a natural product researcher's workflow

By Sarah Srokosz and Kristóf Cank

Moving NMR data processing to the web browser provides unique benefits compared to traditional desktop software. A natural products researcher from the University of North Carolina at Greensboro shared his experience of switching to browser-based NMR data processing with Spectrus JS and how it not only streamlined his workflow, but also positively impacted his reported results alongside other lab-wide systems.

Instrument-vendor software is primarily designed for NMR data acquisition, often making processing of that data in the same application challenging. Third-party software has helped overcome these challenges by introducing vendor-neutral applications that are designed with processing workflows in mind. However, these applications still require local installation, putting strain on resources such as computer memory and storage, and limiting when and where NMR data can be processed.

At the University of North Carolina at Greensboro, a natural product researcher was among the first to adopt the first commercial browser-based NMR data processing application, Spectrus JS. In addition to the typical benefits of third-party applications, Spectrus JS is cloud-based and gives users access to a complete set of NMR data processing tools in any web browser. In this application note, the researcher details his experience implementing Spectrus JS and the impacts it had on his work to isolate and characterize bioactive compounds from fungi, plants, and bacteria.

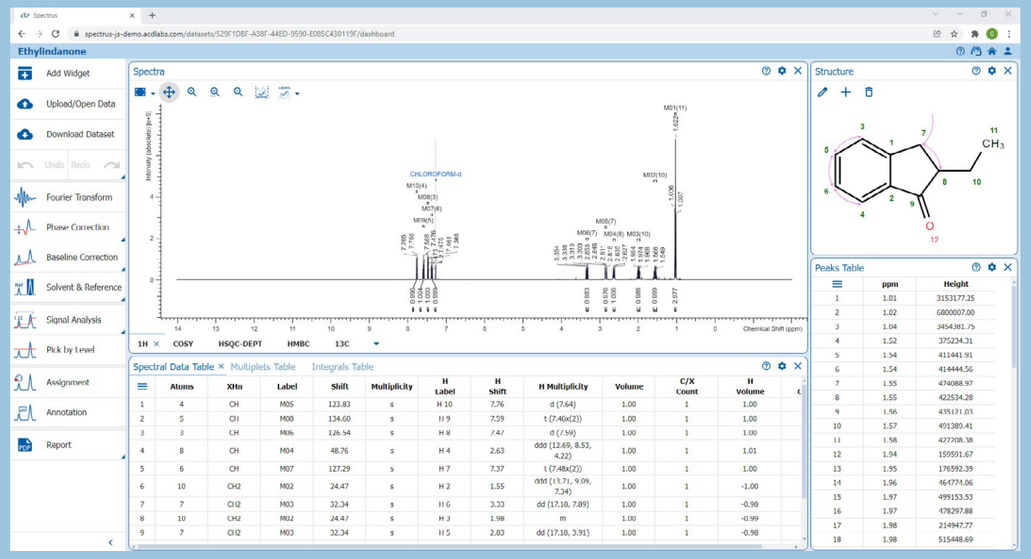
In the switch from the lab's previous NMR data processing software to Spectrus JS, the researcher noted it was important that the software fit into their existing processes and systems without major disruption. The browser-based nature of Spectrus JS alongside its intuitive interface made this transition smooth, even for the researcher's trainee who had significantly less experience processing NMR data.

After continuing to use Spectrus JS in his daily research, the researcher noticed the software streamlined his usual workflows by:

- Simplifying data processing for spectra acquired from multiple shared instruments
- Integrating seamlessly with other software and systems in his lab
- Assembling and displaying the information he needed instantaneously

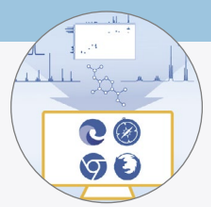
The flexibility of browser-based NMR data processing streams the researcher's workflow by eliminating the unnecessary time- and energy-consuming logistical considerations required by previous software. In addition, the transition to Spectrus JS allowed the researcher to produce more accurate and meaningful results and records without any significant disruptions to his lab's established processes.

View the full application note online



LINKS

Product Page
Browser-Based Analytical Data Processing



Webinar
The Next Generation of NMR Software

Blog
The Freedom of Browser-Based Analytical Data Processing

Sponsored by ACD/Labs

Produced by the Analytical Scientist

Rapid Nitrosamine Analysis

Simple, Rapid Analysis of N-Nitrosodimethylamine (NDMA) Impurity in Ranitidine Products Using SIFT-MS

By Mark J. Perkins and Vaughan S. Langford

Mutagenic N-nitrosamine impurities are found at trace concentrations in certain pharmaceutical products as by-products of synthesis or, less commonly, through migration from packaging materials. They are traditionally analyzed using chromatographic techniques that require significant sample preparation and have relatively low sample throughputs. SIFT-MS simplifies and speeds up analysis of trace volatile N-nitrosamine impurities, with a throughput of 12 samples/hr (three times faster than gas and liquid chromatography methods assuming they utilize fully automated sample preparation) and only 70 minutes to first result (including full calibration set; over twice as fast as chromatographic methods). This application note describes headspace-SIFT-MS analysis of ranitidine products and achieves a limit of quantitation of 2 ng g-1 for NDMA in 500 mg of drug product.

The known or suspected mutagenicity of many N-nitrosamines – in particular, N-nitrosodimethylamine (NDMA) and N-nitrosodiethylamine (NDEA) – means that their presence in any product to which humans are exposed is of concern. Despite the occurrence of these compounds being well-known in water, beverages, and foods, their discovery in sartan medicines in 2018 came as something of a shock to the pharmaceutical industry. Elastomeric sources of N-nitrosamines – as used in various packaging components – are typically very well controlled due to improvements made in the manufacture of elastomers due to historic issues in that industry. Investigations have revealed that most N-nitrosamine issues arise from nitrosating agents used in synthesis – especially when secondary amines are present.

Following substantial industry investigation and consultation, the European Medicines Agency (EMA) and United States Food and Drug Administration (FDA) have issued acceptable intakes. The limits for the more volatile N-nitrosamines are summarized in Table 1. With acceptable intakes at low nanogram levels per day, highly sensitive and selective analytical methods are required. Typically, these are based on gas or liquid chromatography with longer analysis times and more complex sample preparation (e.g., dissolution and centrifugation needed). They are also off-line methods, having a relatively long time to result for the first sample when calibration is considered, and a relatively low sample throughput. In contrast, selected ion flow tube mass spectrometry (SIFT-MS) greatly simplifies sample preparation (drug product is simply weighed into the vial) and eliminates the slow chromatography step, by utilizing soft chemical ionization directly to gas-phase and headspace samples. This approach not only speeds up the analysis, but it can also significantly reduce other sample preparation because no derivatization of the highly polar nitrosamines is required.

This application note demonstrates that headspace-SIFT-MS is well suited to quantitative screening analysis of volatile nitrosamines in drug products because it is highly sensitive and selective. It also provides significant advantages over routine chromatographic analysis through delivery of faster results and higher sample throughputs.

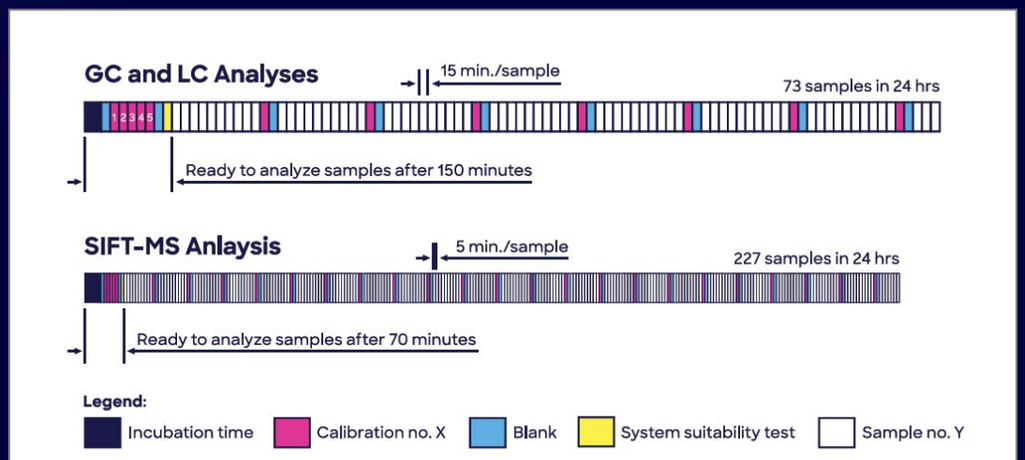


Figure 1. Headspace SIFT-MS enables high-throughput screening of volatile nitrosamines, providing enhanced quality control and quality assurance.

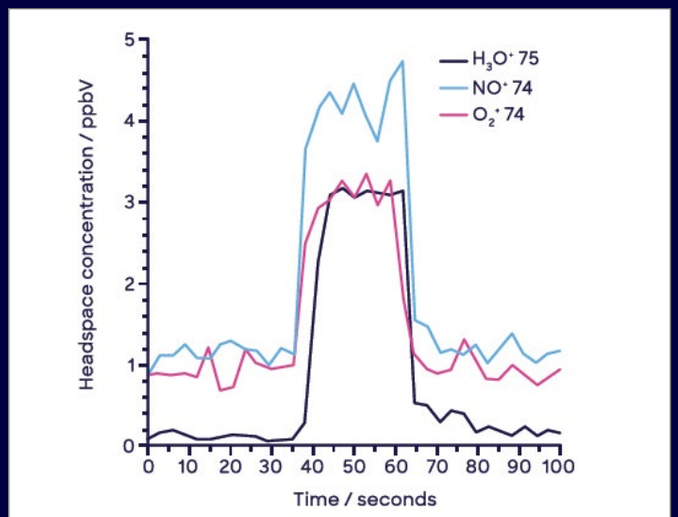


Figure 2. Example headspace injection with synchronous SIFT-MS analysis of NDMA at 2.6-ng spike level (10-mL sample vial).

View the full application note online

LINKS

Further Information
SIFT-MS Pharmaceutical Applications and Information

Webinar
Rapid, Simplified Residual Solvent and Volatile Impurity Analysis Using SIFT-MS

Sponsored by Syft

Produced by the Analytical Scientist

Spectral Phenotyping Helps Preserve Our Food Security

Drought impacts analyzed in leaves using spectroscopy

By Lorenzo Cotrozzi, Raquel Peron, Mitchell R. Tuinstra, Michael V. Mickelbart, and John J. Couture

This article examines how NIR spectroscopy can be used to create data models to analyze the stress of drought on maize leaves. Predictive spectral models were created that can be used to estimate the impacts of varying drought conditions on food security.

Advancements in phenotyping techniques capable of rapidly and nondestructively detecting impacts of drought on crops are necessary to meet the 21st-century challenge of food security. Here, we describe the use of hyperspectral reflectance to predict variation in physiological and anatomical leaf traits related with water status under varying water availability in six maize (Zea mays) hybrids that differ in yield stability under drought. We also assessed relationships among traits and collections of traits with yield stability.

Measurements were collected in both greenhouse and field environments, with plants exposed to different levels of water stress or to natural water availability, respectively. Leaf spectral measurements were paired with a number of physiological and anatomical reference measurements, and predictive spectral models were constructed using a partial least-squares regression approach. All traits were relatively well predicted by spectroscopic models, with external validation (i.e. by applying partial least-squares regression coefficients on a dataset distinct from the one used for calibration) goodness-of-fit (R^2) ranging from 0.37 to 0.89 and normalized error ranging from 12 to 21 percent. Correlations between reference and predicted data were statistically similar for both greenhouse and field data.

Our findings highlight the capability of vegetation spectroscopy to rapidly and nondestructively identify a number of foliar functional traits affected by drought that can be used as indicators of plant water status. Although we did not detect trait coordination with yield stability in the hybrids used in this study, expanding the range of functional traits estimated by hyperspectral data can help improve trait-based breeding approaches.

View the full application note online

Sponsored by SVC SPECTRA VISTA CORPORATION

Produced by the Analytical Scientist

High-Throughput Screening of Surface Contaminations

FT-IR for precision mechanics QA/QC wby Infrared Laser Imaging (QCL)

FTIR microscopy is a proven method to locate and identify unknown substances and contaminants based on their molecular vibration. This kind of analysis provides reliable, unambiguous indications of where a contamination is coming from and how to troubleshoot processes.

The quality requirements for precision mechanics are enormous. They are often part of complex assemblies that require many unique components. If one of these components carries a critical impurity, the entire process is in danger.

FTIR microscopy is a proven method to locate and identify unknown substances and contaminants based on their molecular vibration. This kind of analysis provides reliable, unambiguous indications of where a contamination is coming from and how to troubleshoot processes. Many laboratories do not want to miss the well-proven performance of FTIR – especially for unknown substances. The HYPERION II combines modern, IR Laser Imaging with the comprehensive chemical info of full FTIR spectra. The HYPERION II software was designed to give an easy and lightweight user experience when changing IR modes. With a simple "click," you switch back and forth between techniques and analyze your samples with the highest efficiency.

View the full application note online

LINKS

LUMOS II
The FT-IR Microscopy and Imaging Powerhouse

Guide
QCL Microscopy

Sponsored by BRUKER

Produced by the Analytical Scientist

Correlative Raman Imaging of Polymeric Materials

Raman microscopy combined with AFM and SNOM delivers more comprehensive sample characterization

By WITec GmbH

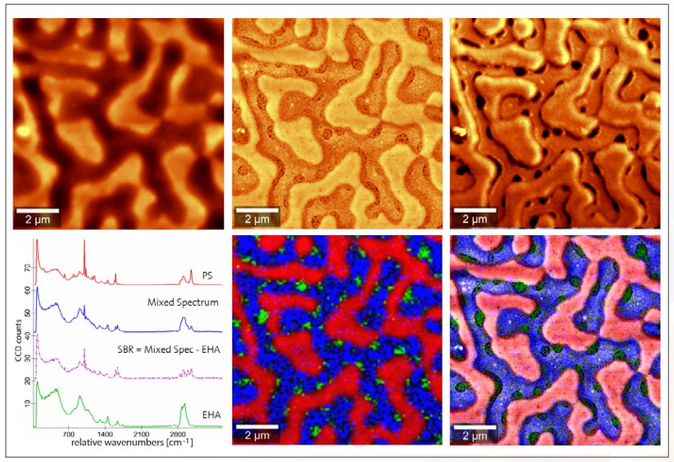
This study shows a series of correlated Raman imaging, atomic force microscopy and scanning near-field optical microscopy measurements that characterize and depth-profile polymer samples. The substances investigated include spin-coated PS-SBR-EHA and PMMA-SBR blends, an orange juice container inner coating, adhesive layers on paper and bioinspired nanofibers.

Polymers have widely varying chemical, mechanical and optical properties. Knowledge of their morphology and composition is crucial to advancing their development and monitoring their production. Raman imaging correlated with atomic force microscopy and scanning near-field optical microscopy can facilitate the investigation of polymer sample properties that are difficult to measure with conventional methods.

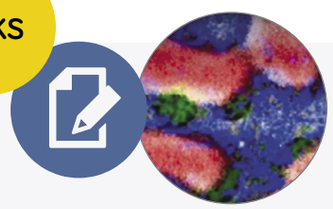
Confocal Raman microscopy is a spectroscopic technique that is able to characterize a sample nondestructively and without staining or other specialized sample preparation, and a due to their confocal beam paths, three-dimensional measurements such a depth profiles can be performed. The chemical sensitivity of high-quality confocal Raman microscopes also allows data acquisition from extremely small sample volumes and concentrations. AFM measurements reveal structural information at the sub-micron level and SNOM can evaluate a specimen optically with resolution below the diffraction limit. Cantilever-based SNOM sensors can be used with the same beam-deflection optics and control electronics as an AFM and are also more robust than traditional SNOM tips. A truly correlative system can use all three methods in concert on the same sample area and the data recorded provides a more thorough understanding than possible with the individual approaches in isolation.

In this study a thin-film mixture of PMMA-SBR is investigated and its components are differentiated with Raman-AFM, a Raman-AFM-SNOM measurement of a PS-SBR-EHA mix is presented, depth-profiles of a polymer coating and an adhesive layer are shown, and the topography and chemical composition of bioinspired nanofibers are characterized with Raman-AFM. The Raman spectra of the constituent substances are displayed along with color-coded images that show their distributions over a sample area in x-y scans or the positions and thicknesses of their layers in scans along the z-axis.

The instrument used for all measurements was a WITec alpha300 RAS correlative Raman-AFM-SNOM microscope. This system fully integrates all three techniques in a modular architecture with a common software environment. The speed, sensitivity and resolution of the alpha300 series, along with its intuitive user interface and configurability, enables researchers to carry out challenging experiments quickly and routinely.



LINKS



WITec Polymers Applications Page

WITec Homepage

Sponsored by OXFORD INSTRUMENTS | WITec

Produced by the Analytical Scientist