APRIL 2018 # 02

the Spectroscopist

Spectra, Spectra, Everywhere

Three top scientists weigh in on how spectroscopy is permeating every aspect of our lives – and every corner of the globe.

10 – 17

EXPAND
YOUR RANGE
EXTEND YOUR RESOURCES

Avio 500 ICP-OES

The New Avio 500 ICP-OES - High throughput with low cost of ownership

The Avio® 500 ICP-OES combines the productivity you need with the high-quality performance and faster return on investment your work demands. With high sensitivity and superior resolution, your lab can accomplish more, even when dealing with the most difficult samples. And with the lowest argon consumption of any ICP, simultaneous background correction, and high throughput enabled by Dual View technology, it all comes together to expand the range of what you can accomplish. High throughput. Low cost of ownership. Superior performance. It's everything you want in an ICP-OES system.

For more information, visit perkinelmer.com/avio500

PerkinElmer®
For the Better

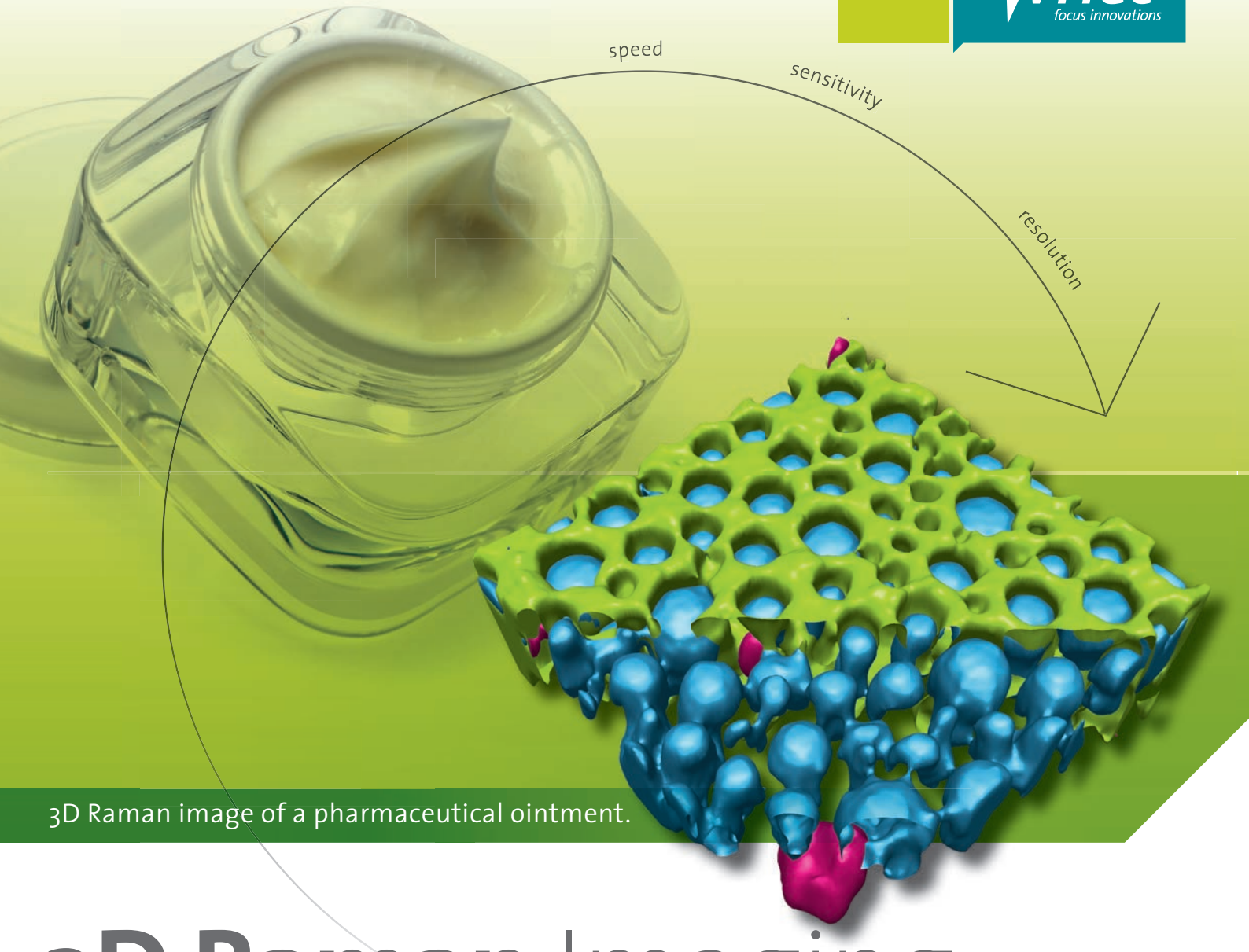
Contents

08

18

26

WITec
focus innovations



3D Raman image of a pharmaceutical ointment.

3D Raman Imaging

Turn ideas into **discoveries**

Let your discoveries lead the scientific future. Like no other system, WITec's confocal 3D Raman microscopes allow for cutting-edge chemical imaging and correlative microscopy with AFM, SNOM, SEM or Profilometry. Discuss your ideas with us at **info@witec.de.**

Raman · AFM · SNOM · RISE **www.witec.de**

MADE IN **GERMANY**

The ~~Two~~ Three Sides of Spectroscopic Investigation

We need to recognize the important symbiotic relationship between theory, application, and instrumentation in analytical science.

The rather peculiar title of this editorial attempts to provide a framework that I have found convenient for considering scientific developments. The first part of the concept involves the classical view of scientific inquiry, that theory guides application. That is, a theoretical framework is developed from which applications spring. Put more colloquially by the baseball catcher Yogi Berra, "If you don't know where you are going, you're apt to end up somewhere else". Abundant examples of this approach can be found. For example, nearly 40 years elapsed between the first description of mass spectrometric principles and their initial application to chemical characterization. Similarly, NMR and Mössbauer spectroscopy were described theoretically long before their application. In the area of physics, the principles of laser operation were established long before a workable system was designed; the use of lasers in chemistry took even more time. This classical view of scientific inquiry can be represented as:

Theory → Guides → Application

Excellent examples of how analytical spectrochemistry has benefited from fundamental findings in atomic physics were provided by the Dutch physicist C.Th.J. Alkemade, then at the University of Utrecht. He stated the philosophy clearly: "Analytical systems can be improved more straightforwardly and more universally if one first understands the basic mechanisms involved and has knowledge of the relevant physical parameters and constants."

The converse side of scientific inquiry is that applications drive theory. In many situations, a sufficiently important application requires that the pertinent theory be developed so an applied problem can be better solved. An appropriate example lies in the field of atomic spectroscopy, which stems largely from the original work of R.W. Bunsen. Long before the Bohr model of the atom was formulated, Bunsen and his colleague Kirchhoff showed that a chemical flame and spectroscope could be used for the determination of alkali metals. This general approach can be embodied as:

Application → Drives → Theory

The processes involved in these two "reactions" are mutually supportive and essential for scientific progress. No matter whether an investigation begins with an important application or a new theory, the opposite partner will benefit. For example, in NMR spectrometry, theoretical developments led and workable instrumentation followed. But when it became evident that NMR could solve important problems in chemistry, medicine, and other fields, the theory was refined further, to encompass the use of multiple pulse sequences, relaxation times, and other parameters to elicit additional information. In turn, those multidimensional parameters enabled even more complex problems to be tackled, and so on.

In both cases, whether theory guides applications or applications drive theory, the NMR example suggests that instrumentation provides a catalyst; it serves as an enabler for theory to solve important problems or for problems to be investigated more fully and lead to improved theories. Instrumentation therefore serves as the third "side" of spectroscopic inquiry. The theory of multi-pulse sequences in NMR was of little use until it could be embodied in scientific instrumentation. Similarly, applications of NMR instruments to real problems suggested the importance of other parameters to characterize. Thus:

Application ↔ Instrumentation ↔ Theory

It is critical that this symbiosis not be overlooked by spectroscopists, other scientists, scientific journals, and especially funding agencies. It is worrisome that such a strong emphasis seems to exist right now in the funding of focused applications of existing theory or instrumentation. Funding mainly application-oriented research and neglecting either instrumentation development or fundamental underpinnings limits future gains needlessly. Instead, it seems prudent to dedicate an ongoing effort to both theoretical work and instrumentation development, even when the immediate application of a new instrument or theory isn't immediately evident. Witness, for example, the development of scanning tunneling microscopy or, as mentioned above, NMR.

Hopefully, the foregoing comments provide convincing evidence that there are not two but really three areas of scientific inquiry: theory, application, and instrumentation. Each is capable of existing and being important on its own, but all benefit from symbiosis. The transfer of information between theory and applications will be driven not only by instrumentation but also by individuals who seek to recognize the importance of instrument and method development and who attempt to understand the fundamentals of instrumentation and the measurement process in greater detail. I am certain you will find that the articles in this issue of The Spectrocopist provide good examples of this symbiosis at work.

Gary M. Hieftje
Guest Editor

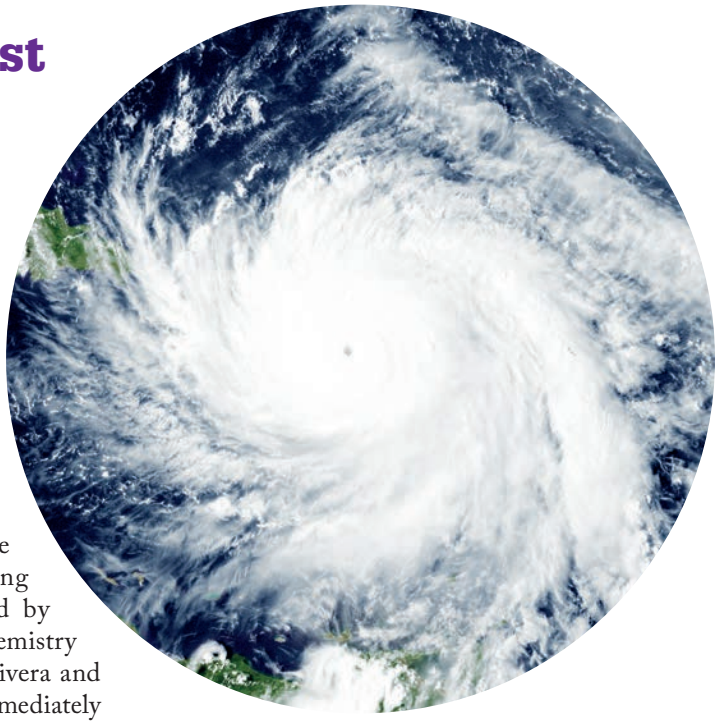
Upfront

Reporting on research, personalities, policies and partnerships that are shaping analytical science.

We welcome information on interesting collaborations or research that has really caught your eye, in a good or bad way. Email: charlotte.barker@texerepublishing.com

Spectroscopist SOS

Professional societies team up to help scientists affected by last year's devastating hurricane in Puerto Rico



When Ellen Miseo, a past President of The Society for Applied Spectroscopy (SAS), read an article in the society's newsletter detailing the disruption being faced by University of Puerto Rico chemistry professor Sam Hernandez-Rivera and his lab earlier this year, she immediately wanted to help. While aid efforts were gradually rebuilding infrastructure, there were no resources to restore laboratories, and water exposure and power outages had rendered much of their equipment inoperable. Hundreds of graduate and undergraduate students had their education put on hold.

Miseo discussed the article with spectroscopy consultant and member of the Coblentz Society governing board Frederick Haibach, and together they hatched a plan to help. "Between us we have an enormous number of contacts in the instrument area, and we thought there must be some way we can get instrument companies to donate and get things to happen," says Miseo.

The pair have already convinced instrument companies to donate money and instruments, and even fly experts out to the island to help scientists get their equipment up and running again. But there's still a long road ahead, says Miseo. "It's going to take over a year to get equipment up and running, labs back in shape, and get students and professors back to being productive."

In the meantime, SAS and the Coblentz Society are offering grants to enable Puerto Rican students affected by the hurricane to carry out experimental work in mainland USA and so complete their degrees. "Students simply fill out a short questionnaire about their research, what was interrupted, what is needed to complete it, and a sign-off from their adviser," says Miseo.

Make a donation at: https://bit.ly/2pWcht7

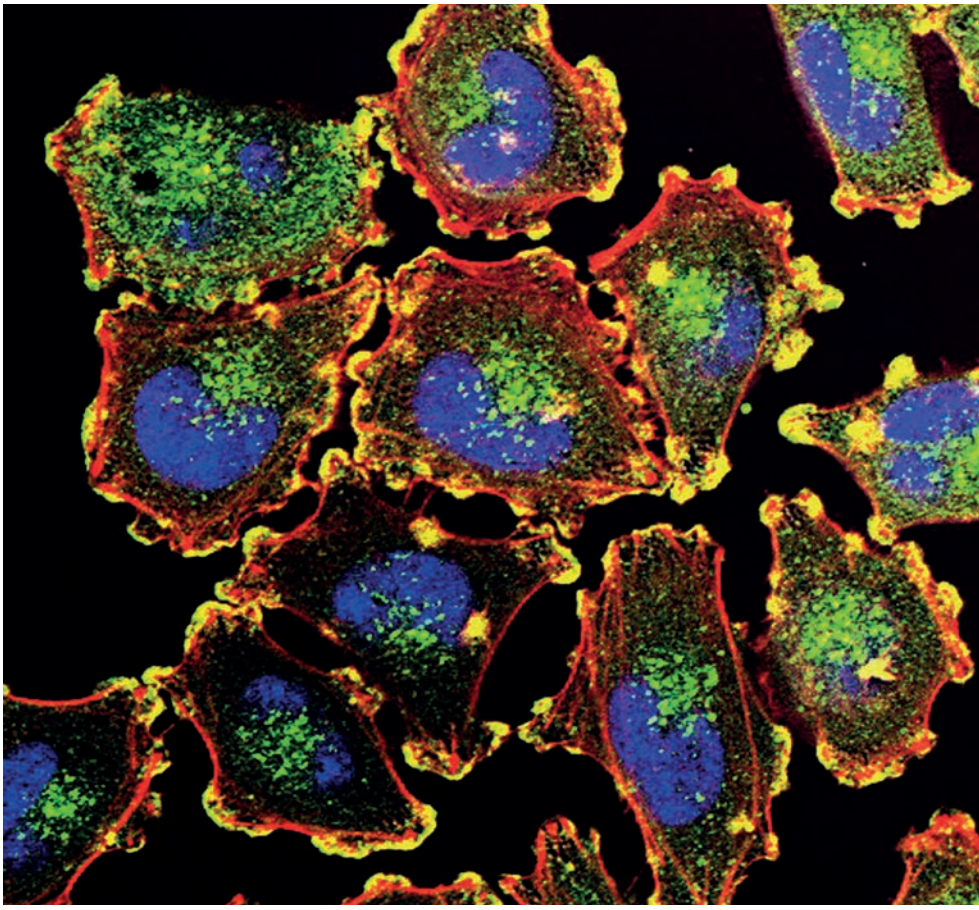
Volunteer your facilities, time or expertise at: https://bit.ly/2pX783f

Scientists and students who would like to request assistance from the fund can contact the societies at: https://bit.ly/2H4o3sY

Screen Test for Spec

How ATR-FTIR spectroscopy could lead to less invasive cancer prescreening

A team from Georgia State University are using Fourier transform infrared (FTIR) spectroscopy in attenuated total reflection (ATR) mode to develop a less invasive prescreening test for melanoma and lymphoma. Here, we talk to Regents' Professor of Physics Unil Perera about his hopes for the method.



What inspired you to develop a prescreen method?

The earlier we detect cancer, the better. Some screening methods, such as colonoscopies, are the gold standard, but are expensive and uncomfortable – to the point where patients avoid being tested. I wanted to devise a simple screening test that would be less invasive, less uncomfortable, and less costly – as well as capable of screening for more than one disease. One day, I want doctors to be able to screen patients annually; if they detect the markers for cancer, then the patient can be referred for further tests.

How far along is the work?

We have used ATR-FTIR to analyze biochemical changes in the blood serum of mouse models. We have shown in our previous work that we can identify colitis and diabetes – more recently, we were able to detect markers of lymphoma and melanoma and discriminate between healthy mice and those with tumors (1). Potentially, the technique could be applied to many different diseases – but it has yet to be proven on human samples.

What's your ultimate aim?

We want to come up with a portable device/detector that sits in a doctor's office. A patient gives blood, and the doctor checks for indications of a particular cancer. Our goal, ultimately, is that the doctor just needs to put in a drop of blood and get an answer – but that's obviously some years away. Our priority right now is to prove that this works in humans.

What keeps you motivated?

I'm happy to make some contribution to human health – even a minor one. After our cancer paper was published (1), I got an email from one of our faculty members, saying that as a survivor of cancer, she was happy to see that I was doing something on early detection. That was really special.

Reference

1. *H Ghimire et al., "ATR-FTIR spectral discrimination between normal and tumorous mouse models of lymphoma and melanoma from serum samples", Sci Rep, 7, 16993 (2017).*

Brain Wave

SERS and SORS could help diagnosticians access your mind...

In 2013, Barack Obama announced the US BRAIN Initiative, a 10-year research endeavor to understand how the brain works. For Bhavya Sharma, Assistant Professor of Chemistry at the University of Tennessee Knoxville, USA, this was a springboard for her own brain imaging research.

"I had learned that, in general, brain imaging techniques provide high spatial resolution but little to no chemical information. Perhaps more importantly for the owner of the brain, techniques that provide chemical information often require holes to be drilled in the skull or portions of the skull to be removed to access brain tissue," she says. Sharma saw Raman spectroscopy as a way to get the information researchers need without physically penetrating the skull. And she knew it could have a big impact. "I believed that if I could make this work, we would be able to develop Raman-based methods for neurological disease diagnosis, particularly for diseases that are difficult to diagnose in the early stages, such as Parkinson's disease," she says.

In an initial study, Sharma and her team used animal skulls filled with agarose gels to mimic brain tissue, combining surface-enhanced Raman spectroscopy (SERS) and spatially-offset Raman spectroscopy (SORS; combined name: SESORS) to look inside the skulls (1).

"SERS utilizes noble metal nanoparticles to enhance the inherently weak Raman scattered light through an electric field generated at the surface of the nanoparticles, allowing us to collect Raman spectra of analytes down to the nanomolar range)," she says. "In SORS, the laser is incident on the surface of a multilayer sample, with the Raman light scattered from the top layer traveling back along the same trajectory as the incident laser. Some of the photons travel into the lower layer and migrate laterally before being scattered out. The distance between the incident laser and where the light is scattered from the lower layer is the spatial offset." By collecting the Raman scattered light at the spatially-offset location while going through the top layer, the team gained information about the lower layer (see Figure 1).

They were able to measure the Raman spectra of three different neurotransmitters through skull bones across a concentration range of 100 micromolar (μM) to 500 millimolar (mM). But they're keen to take it to the next level. "The measurements we performed were with skulls that are two mm thick, but in humans, the skull is 3-14 mm thick," she says. "Our challenge now is to lower our limits of detection and increase our depth of penetration – but at this point we're already within one mm of measuring the thinnest part of a human skull."

Sharma believes that SESORS holds

great promise for use in non-invasive measurement of neurochemicals in the brain. "We are moving forward with new instrument designs, and have moved onto measuring skulls with greater thicknesses," she says. "Our preliminary results are promising – so stay tuned… We expect to have a lot of new and exciting results to share in the future!"

Reference

1. *AS Moody et al., "Surface-enhanced spatially offset raman spectroscopy detection of neurochemicals through the skull", Anal Chem, (2017). DOI: 10.1021/acs.analchem.7b00985*

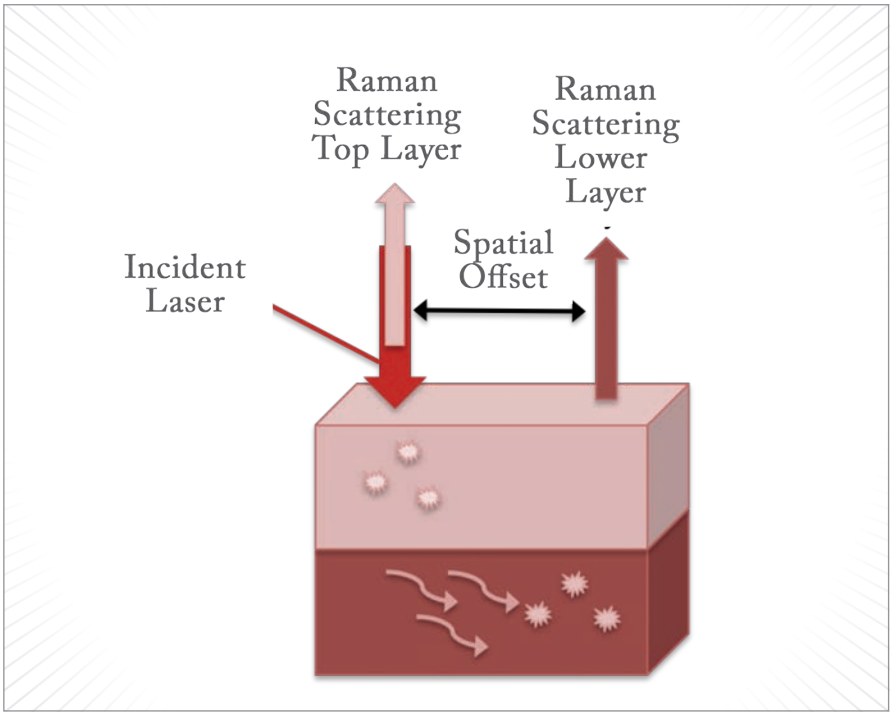
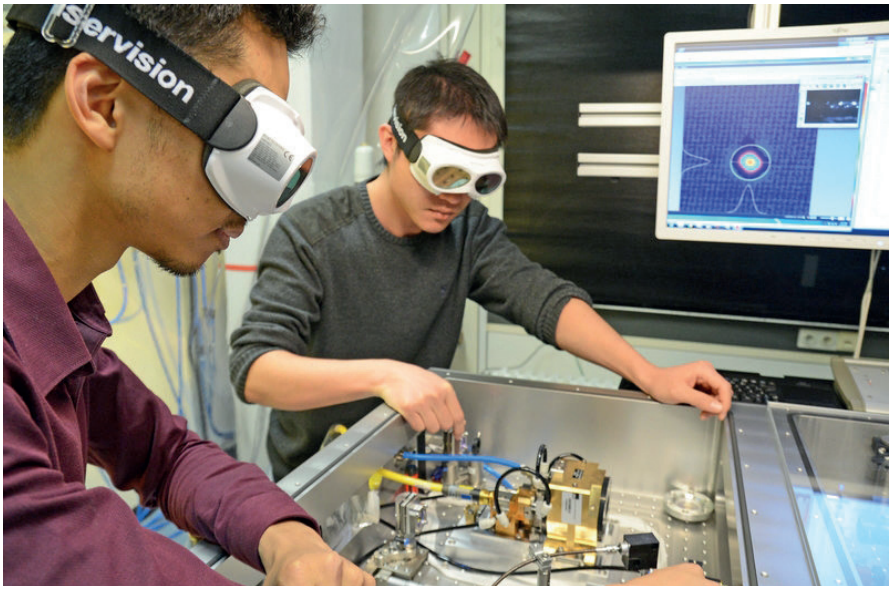


Figure 1. SORS schematic.

Better Source Material

A new mid-IR source could have powerful potential



Credit: Thorsten Naser

What?
A new broadband light source that can produce infrared light in the 4.5–20 µm region, based on a powerful disk-based oscillator.

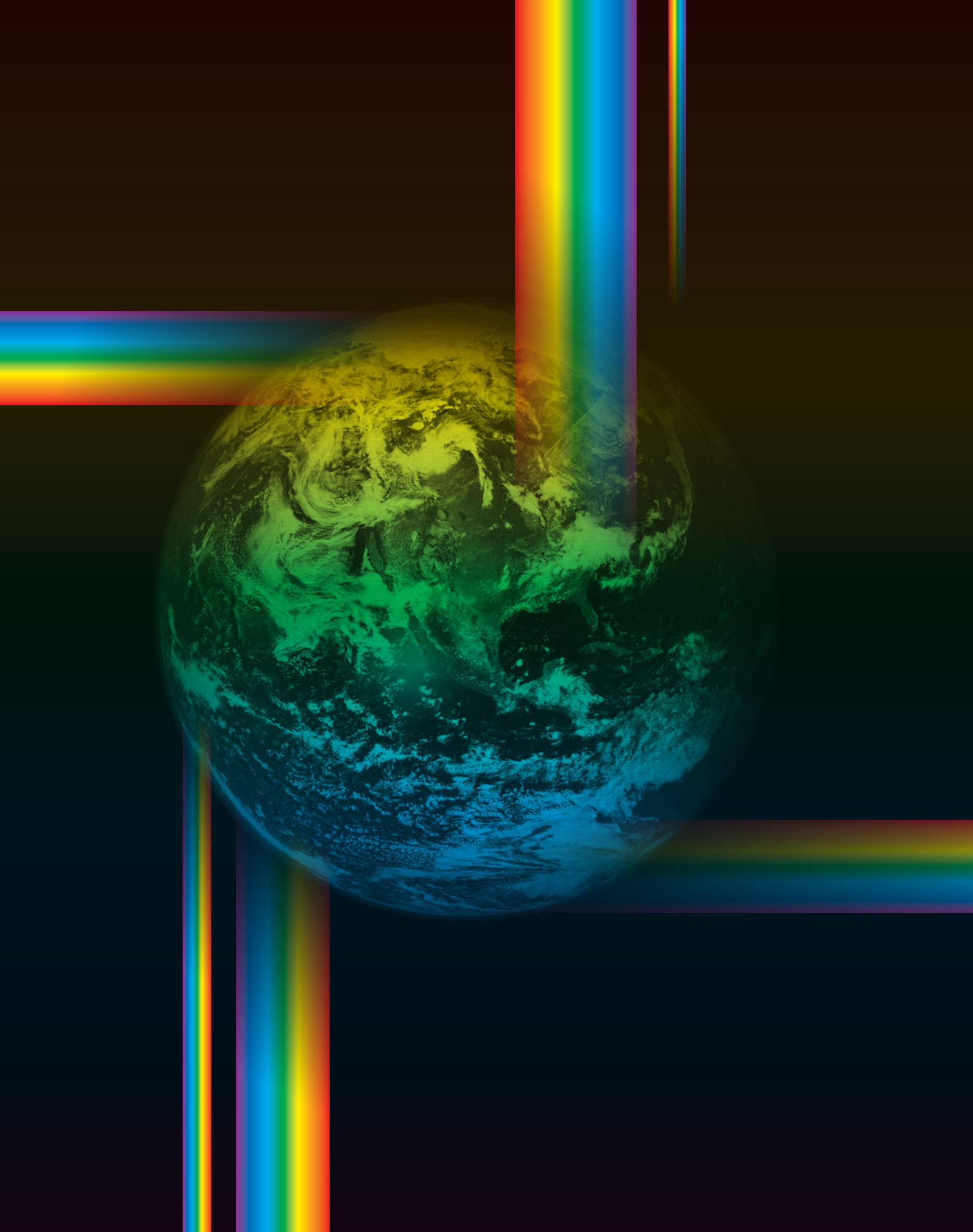
How?
The thin disk is pumped by a thulium-doped fiber laser emitting at 1908 nm, "resulting in a much smaller quantum defect and up-conversion process compared to the traditional Ho/Tm co-doped system" (1). The result? Optical-to-optical efficiency of 22 percent.

Who?
The source is the result of a long-term collaboration between laser technology company Trumpf, and Ference Krausz and Oleg Pronin from the Max-Planck Institute of Quantum Optics, as well as researchers from Ludwig-Maximilians University, Munich.

What next?
The team believes that this development has great potential within a wide variety of spectroscopic techniques. "Time resolved pump-probe spectroscopy, scattering near-field optical microscopy and potentially dual-comb spectroscopy will benefit from the improved coverage and increased power," the research paper states (1). In particular, they believe the new source "holds promise for better specificity for biomedical applications."

Reference

1. *1J Zhang et al., "Multi-mW, few-cycle mid-infrared continuum spanning from 500 to 2250 cm-1", Light Sci Appl, 7, 17180 (2018).*

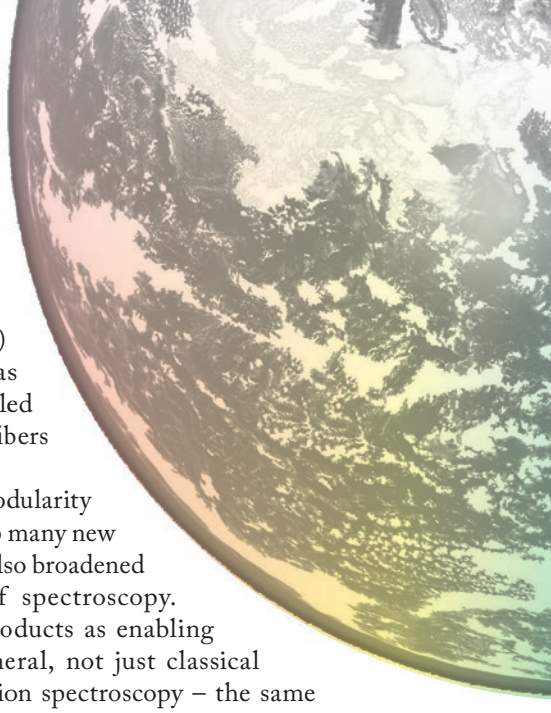


THE **RISE** OF ALL-PERVASIVE **SPECTRA**

Technological advances are allowing optical sensors and mass spectrometers to permeate every aspect of our lives, every corner of our globe – and beyond. What makes spectroscopy so ubiquitous and where are more recent trends taking us? Here, three scientists offer insight across the analytical spectrum.

A SPECTROMETER ON EVERY WRIST

By Mike Morris



In 1992, we launched Ocean Optics to sell what we described as the world's first miniature spectrometer. It featured a very low-cost linear array detector that was originally manufactured for barcode scanners. We discovered this US$20 charge-coupled device (CCD) array detector significantly outperformed $1000 "scientific" photo-diode array detectors. The reason, we came to understand, is that the quality of microelectronics improves with quantity. High-volume manufacturing can use more expensive coaters and other capital equipment. Higher volumes allow for fine tuning and optimization of the process and implementation of more sophisticated quality control systems. The other breakthroughs we needed came from low-cost, high-performance optical fibers, and ever improving and lower-cost computers. These three components formed the basis for our fiber optic, PC-controlled spectrometer.

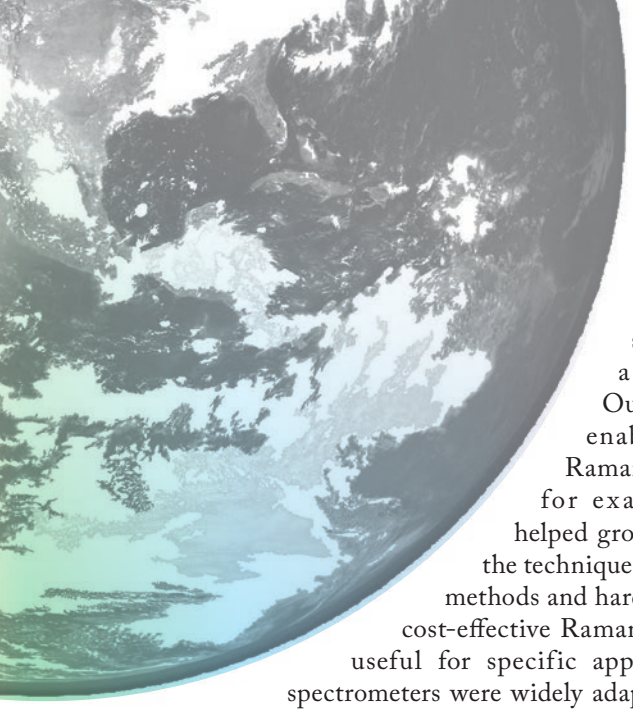
The other breakthrough was a combination of marketing and engineering, two realms that have more common ground than you might think – both are experimental, involve solving problems, and treat the cost of a product as a design variable. We found that we could lower the cost of applying our spectrometer to new applications by making the system highly modular. Our goal was to make the creation of a new "product" or application possible without any additional engineering expense. Therefore, the components of a spectrophotometer (light source, sample compartment, spectrometer, readout and interpretation) were made and sold as individual items, coupled together with optical fibers to create systems.

The lower cost and modularity of our system opened up many new application areas, and also broadened our understanding of spectroscopy. We thought of our products as enabling optical sensing in general, not just classical absorbance and reflection spectroscopy – the same device could measure transmission/absorbance, reflection, elastic and inelastic scattering, interference, polarity, emission, and time-based changes. The affordability of the systems lowered the financial risk of trying out new types of measurements, and also helped in the adoption of new applications in the marketplace. Arguably, our marketing success broadened the awareness and appreciation of spectroscopic measurements and boosted the industry as a whole.

> *"Marketing and engineering share common ground – both are experimental, involve solving problems, and treat the cost of a product as a design variable."*

One of our great successes was in education markets. I went through four years of undergraduate education with only a single day's exposure to a Spec 20 in general chemistry lab. It even had a small sign pasted to it that said, "Do not touch the dials!" Today, many students at all levels (including high school) are using our spectrometers for experiments that include acquiring full spectra from their samples as .xls files, and learning how to manipulate and process data in a spreadsheet – valuable real-life skills. It is exciting to note that spectrometers identical to those that students are using have flown on space missions, been to the depths of the oceans and been used in field studies in all kinds of environments. In many institutions, the chemistry department shares spectrometers with the biology and physics departments, made possible by acquiring a few extra accessories.

We also helped develop many



new optical sensing approaches. Our spectrometer enabled low-cost Raman spectroscopy, for example, which helped grow awareness of the technique and progress in methods and hardware to obtain cost-effective Raman data that was useful for specific applications. Our spectrometers were widely adapted for plasma chemistry applications, especially in electronics and optical vacuum process environments. We also had systems being used to look at absorbance of gases coming from volcanoes, smoke stacks, fence line monitors, automobile exhausts, marine sediments, exhaled breath and atmospheric pollutants. Our fiber optic reflection probe found great use in diffuse reflection of tissue for detecting cancers and other pathologies, and in backscattering measurements used to characterize particle size distribution in slurries, colloids and biological materials. Our spectrometer and a special connector brought spectroscopy to microscopes and telescopes. The spectrometers served as detectors for fiber optic chemical sensors including pH and O_2, and even special sensors for monitoring the breakdown of solid rocket fuel in aging intercontinental ballistic missiles.

The other important attributes of the systems we developed at Ocean Optics were small size and a low power requirement, which allowed portable spectroscopy for samples that can't be brought back to the lab. We pioneered using laptop and notebook PCs and battery-operated light sources to make measurements in situ. More recently, our spectrometers have been added to autonomous vehicles and drones to look at spectral reflection of crops, natural vegetation, minerals and soils, algae in natural waters, and so on. The spectrometers add fine spectral information to coarse hyperspectral imagers and can be deployed for ground truth and sea truth measurements as well. Applications abound in agriculture for managing crops, finding invasive species in waterways, detecting fertilization, water stress, and ideal time to harvest.

The photonics industry is driven by advances in materials science and manufacturing technology. Detector development has been driven by the demand for higher-resolution and lower-cost cameras in cellphones and surveillance devices. The need for higher bandwidth communications has driven the development of better, faster lasers and detectors, higher-performance optical fibers, narrow wavelength filters for wavelength division multiplexing and a host of components for use in the near infrared (NIR) range. Better design tools, including optical modelling CAD software, have enabled improvements in all aspects of optical systems.

These photonics advances have benefited spectroscopy. Our miniature spectrometers are now available in the NIR-SWIR (short wave infrared) range; low-cost mid infrared (MIR) array sensors are now becoming available and will soon lead to the introduction of similar low-cost MIR spectrometers. The combination of pulsed high-speed lasers with very fast detectors in spectrometers has made another imaging technique possible – optical coherence tomography (OCT). In fact, the blending of spectroscopy and imaging is gaining traction with unique use of two dimensional camera sensors in a variety of hyperspectral and multispectral systems. New architectures of CCD arrays with two bins per pixel explicitly allow for measurement of fluorescence lifetimes in imaging systems.

"The blending of spectroscopy and imaging is gaining traction with unique use of two dimensional camera sensors in a variety of hyperspectral and multispectral systems."

Advances in non-laser light sources have also been great for spectroscopy. Most significant is the expansion in LEDs, which are easy to switch on/off and can be operated at high speeds. LED sources are steadily reducing in cost, increasing in power and becoming available in an ever-broadening wavelength range (from deep UV to IR). As a result, the size and power requirements of spectroscopy-based systems have been reduced. In addition, narrow-band light allows for a narrow range of spectral measurements, with much less stray light.

Our dream at Ocean Optics was to someday put a spectrometer in every home – to make spectroscopy a common and indispensable tool. The advance that may make

"Optical data in unstructured situations may be just as valuable as more traditional setups."

that possible is the Internet, specifically the Internet of Things (IOT). The Internet provides a remarkable infrastructure for gathering, storing and providing data, analysis and interpretation to millions of users. To date, its capacity has been barely used. However, big data analytics, statistical analyses, AI and deep learning techniques are changing that. These new ways of getting answers are being used to overstep the traditional boundaries of instruments and measurements in rather unique ways.

Today's analytical models for tracking and predicting important phenomena may include data from many different instruments and sensors, as well as from the stock market, social media, remote sensing satellites and government agencies. In this new world, data is only judged by how it contributes to the solution. As such, optical data in unstructured situations may be just as valuable as more traditional setups. Recently, a research study was launched to use data from Apple watches to attempt to diagnose atrial fibrillation. That data being collected include heart rate and activity levels, but could also include other parameters, such as

A grad student wearing one of our spectrometers on his tin foil hat. He is using it to measure SO_2 and other gases in the air column as he walks around a volcano.

oxygen saturation, as well as data from social media, GPS location, air pollution, epidemiology of local rates of heart issues and so on. Now imagine if we swapped the two-wavelength sensor in the watch with a spectrometer; the possibilities are fantastic – not just a spectrometer in every home, but on every wrist!

Mike Morris is President and CEO of Spectrecology and founder of Ocean Optics Inc, Florida, USA.

ALL CHANGE

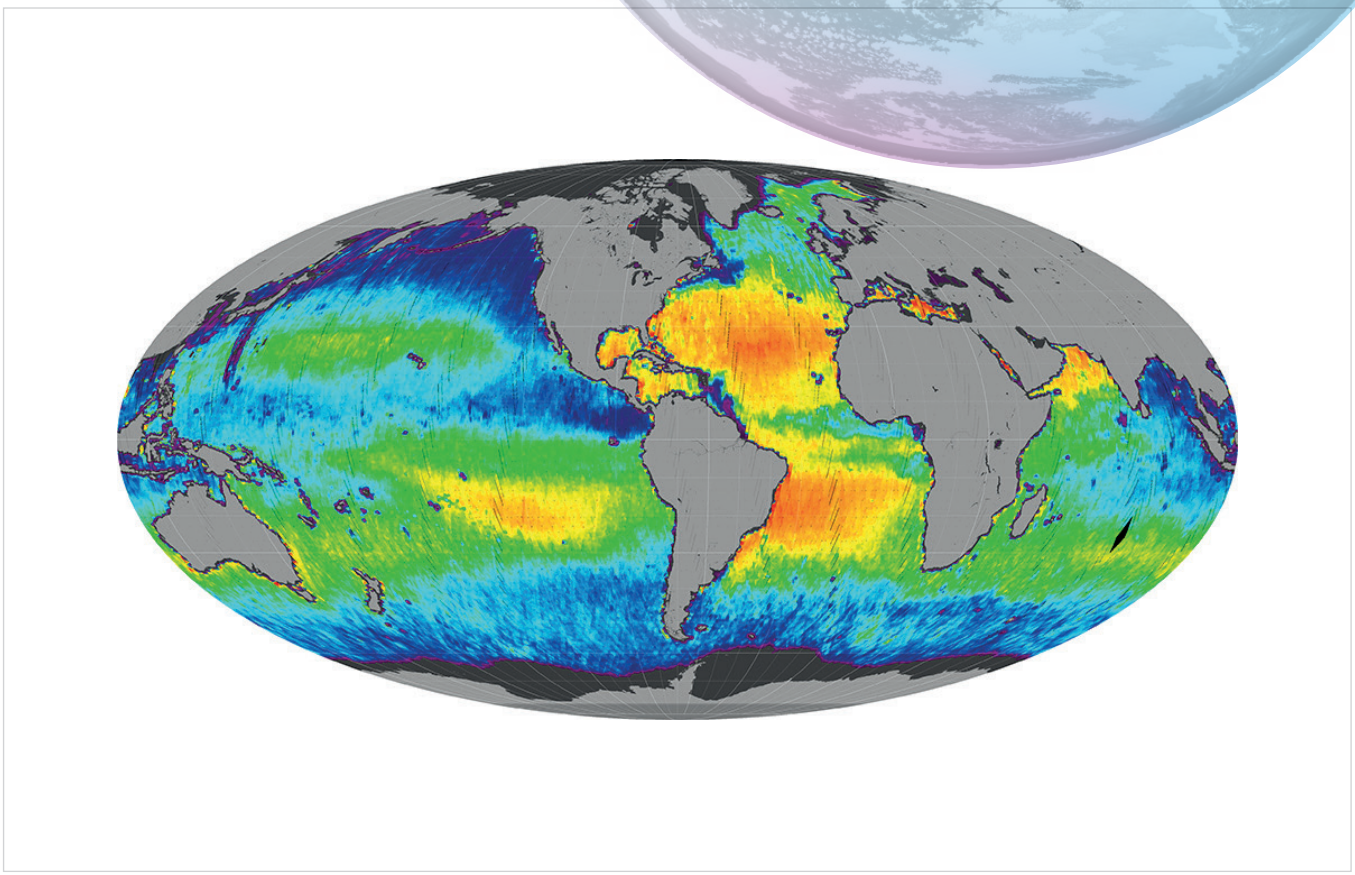
By Benjamin W. Smith

The most noticeable trend in analytical spectrometry is for compact, portable, embeddable, implantable, point-of-need instrumentation. Driven by smaller electronics, reduced power requirements and improved analytical performance, the most obvious example of this trend is the rapid evolution of compact Raman instruments. These handheld instruments are now found in law enforcement vehicles and airport security sites, where they are used for rapid field identification of a wide variety of chemical substances. Another example is how fluorescence imaging is taking on a whole new role in biochemical research. Solid state detectors and light sources (lasers and LEDs) play an important and growing role in all such instrumentation advances.

"It is likely that more compact, efficient ionization sources will eventually replace the rather cumbersome ICP."

Optical spectroscopic methods also lend themselves very well to remote sensing and there will always be new challenges in this realm. Satellite-based sensors now monitor a wide range of atmospheric and terrestrial species and provide essential data for tracking changes on our planet. One of my favorite examples is the NASA Aquarius satellite which, since 2011, has been mapping the salinity of the oceans using microwave emissivity, producing a map of the entire planet roughly once per week

Changes are also happening in atomic spectrometry, where there are a number of specific challenges and opportunities. We only began making reliable quantitative atomic emission measurements about 100 years ago, but the past century has seen a continual, gradual progress towards lower detection limits, greater selectivity and wider dynamic range detection, with all sorts of new approaches. As with molecular methods,



NASA Aquarius satellite salinity data for June 2015.

instrumentation is gradually becoming smaller and less expensive. Flame emission, spark and arc emission, flame and furnace atomic absorption and plasma emission have all played significant roles throughout much of the 20th century. All of these methodologies are now being replaced with ICP-MS.

In retrospect, it is rather obvious that mass spectrometric detection should eventually replace atomic emission, absorption and fluorescence, as ions are easier to detect than photons, and yield important isotopic information. ICP-MS approaches the ultimate limits of detection, is applicable to virtually every element, and exhibits very wide dynamic range. A huge range of analytical problems can still be served with the older techniques but it is clear that ICP-MS is already the method of choice whenever it is available. In addition, laser sampling has made possible increasingly refined, spatially resolved analysis on a wide variety of samples ranging from rocks to frozen tissue sections.

In future, it is likely that more compact, efficient ionization sources will eventually replace the rather cumbersome ICP, and array detectors for ions will eventually make it possible to build compact mass spectrometers with a multiplex advantage.

The purpose of any analysis is to solve a problem, and many problems lend themselves to spectroscopic solutions. For instance, generating new data for healthcare is undoubtedly one of the most important analytical challenges of the near future and spectrometric methods will surely play an important role. It is not hard to imagine a compact mass spectrometer with the power to provide a complete elemental and molecular profile from a small droplet of blood.

Benjamin W. Smith is a Scientist at the Department of Chemistry, University of Florida, Florida, USA.

MASS ACTION

By Jessica Prenni

The field of analytical mass spectrometry plays an important role in so many areas. In the academic lab it is used to study and advance our understanding of basic questions in biological and physical sciences. Outside of the research laboratory it plays a key role in pharmaceutical development, forensic science, food analysis, and clinical diagnostics. It enables us to test our water to make sure it is clean, screen for explosives at the airport to keep us safe, and diagnose illnesses quickly and accurately.

As scientists we should all serve a goal – to contribute to science and the betterment of our world. However, the specific aim of each spectroscopist is not the same, nor should it be. We need spectroscopists who are focused on the theory of spectroscopy and the continued advancement of our analytical tools. Fundamental research is critical to the forward momentum of science and technology. We have to understand our world before we can make it better.

We also need spectroscopists who are focused on the application of these tools and the intersection of analytical science with other disciplines.

Instrumentation, such as quadrupoles, ion traps and time-of-flight are relatively mature – we are seeing incremental improvements each year but not the leaps and bounds of the past. In contrast, ion mobility spectrometry-mass spectrometry is an area in which we are just starting to realize the potential – there is great room for advancement and I can't wait to see where this technology will go.

Mass spectrometry is becoming available to the masses – the technology is becoming accessible to almost all scientific areas and is increasingly easy to use. I see this trend continuing with increased integration of mass spectrometry in many sectors. For example, the food industry is a space in which mass spectrometry could have a huge impact in the future. There is desperate need to improve the production efficiency, nutritional quality and safety of our food, and spectrometry can play a role in all of these areas.

The biggest practical hindrance right now is a lack of coordination around informatics – everyone is processing their data in different ways, which makes it very difficult to compare data across laboratories and/or instrument platforms. To solve problems like these, we need to train more analytical scientists, and those new scientists need to be from more diverse backgrounds – diversity drives innovation.

> *"Fundamental research is critical. We have to understand our world before we can make it better."*

Jessica Prenni is Associate Professor and Faculty Advisor of the Proteomics & Metabolomics Facility at Colorado State University, Colorado, USA.

An Elemental Regeneration

Emerging applications in life sciences and environmental analysis are driving renewed interest in the unique properties of ICP-TOFMS – in particular, its ability to detect complete elemental mass spectra from short transient events, such as single nanoparticles.

By Alexander Gundlach-Graham

Prior to moving to Switzerland, I did my PhD at Indiana University in Gary Hieftje's group, where I studied a new form of velocity-based mass spectrometry – distance-of-flight mass spectrometry (DOFMS). My research into DOFMS was formative, and taught me about mass spectrometer design, construction, and operation. I've carried the interest and expertise I gained in velocity-based mass spectrometry to my current work at ETH Zurich, developing analytical methods for the high-throughput analysis of diverse inorganic nanoparticles from environmental samples by ICP-TOFMS.

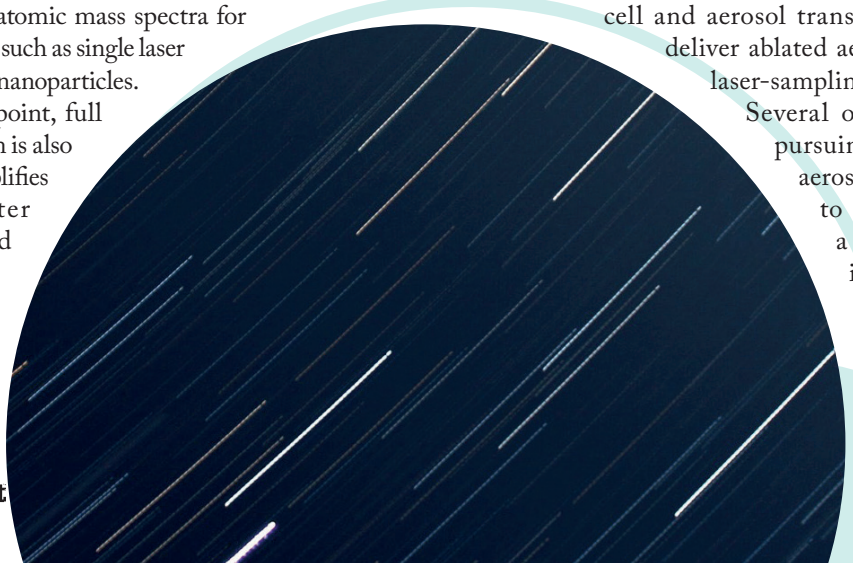
As I began searching for a postdoc position, I was aware of renewed interest in full-spectrum atomic mass spectrometry, largely because of key applications in biomolecule detection and laser-ablation imaging. The potential encouraged me to join the Günther group, who were central to the revival of inductively coupled plasma time-of-flight mass spectrometry (ICP-TOFMS).

ICP-TOFMS rediscovered

ICP-TOFMS is not a new technique; it was pioneered in the early 1990s by the Hieftje group and then commercialized by two instrument companies. However, ICP-TOFMS had limited commercial success because the sensitivity and dynamic range lagged too far behind other scanning-based ICP mass spectrometers. Notably, the initial limitations of ICP-TOFMS were largely the result of the inadequacies of high-speed digitization electronics at the time. Today, high-speed detection electronics have improved (and continue to do so), so ICP-TOFMS has matured into an approach that can compete with other ICP-MS instrument designs in terms of sensitivity, limits of detection, and linear dynamic range. Moreover, it offers unique measurement possibilities through high-speed, full-spectrum ICP-MS analysis.

In my time at ETH Zurich, I've worked predominantly with two ICP-TOFMS instruments: a prototype instrument that was developed through a collaboration between TOFWERK (Thun, Switzerland) and the Günther group, as well as a commercial ICP-TOFMS instrument (icpTOF, TOFWERK). The collaboration between the Günther group and TOFWERK is a great example of how a university-based research group can work in symbiosis with a company in developing a new tool; the academic group provides vision and expertise in compelling and significant applications, while the company provides technical know-how and the environment to develop a user-ready instrument.

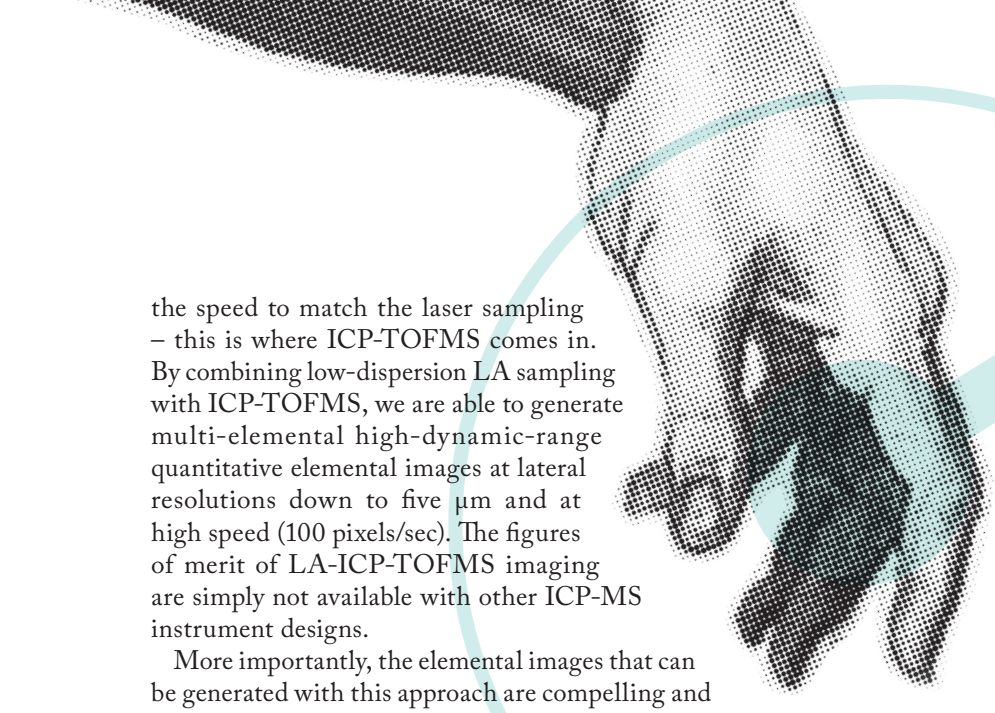
I work mainly on two applications: elemental imaging with laser ablation, and single-nanoparticle detection and quantification. In both of these research areas, ICP-TOFMS provides measurement characteristics that are not available and often fundamentally not possible on other elemental mass spectrometers. The key benefit of ICP-TOFMS is that it provides full spectral coverage and very rapid data acquisition speeds. For laser ablation and single-particle analysis this is a major advantage because it means that we can measure complete atomic mass spectra for discrete transient events, such as single laser ablation events or single nanoparticles. From a practical standpoint, full mass spectrum detection is also beneficial because it simplifies mass spectrometer experimental setup and means that we never miss any unexpected – but pertinent – isotopic signals.



New approach, new measurements

In laser-ablation ICP-MS, a pulsed laser is used to ablate a small quantity of a solid material and the resultant aerosol is transported to the ICP-MS, where the elemental and isotopic composition of the ablated aerosol is measured. Laser ablation is a clean way to sample solid materials and, by controlling the position of the laser, it can be used to measure sample material with lateral and depth resolution. Combined with ICP-MS, laser ablation can provide element and isotope composition at high lateral resolution. Laser ablation is fundamentally a pulsed technique: a laser impinges on a material surface, which causes ablation, and a cloud of sample material is generated – all in a timescale of nanoseconds to hundreds of microseconds. In conventional LA-ICP-MS, sample transport systems are used to "smear" laser-ablated aerosol particles from many laser-ablation events so that timescale of the mass spectrometry analysis matches that of the laser-ablation sampling. Spreading the time profile of LA aerosols leads to more accurate quantification for analysis with scanning-based ICP-MS instruments; however, it also means that information about each LA shot is lost and that lateral and depth resolution is limited by the overlap of signals from temporally (and physically) adjacent laser shots. The high-resolution LA-ICP-TOFMS system we've developed in the Günther lab is fundamentally different to this conventional LA-ICP-MS approach.

In our lab, we use a "pulse-resolved" LA-ICP-TOFMS imaging strategy – the complete transient signal of each LA event is measured without overlap from adjacent LA event signals. This imaging strategy prevents the image blur that arises when ablated aerosols from different locations are mixed in the aerosol transport system. To perform high-speed pulse-resolved LA-based imaging, research in the Günther lab has focused on development of a low-dispersion LA cell and aerosol transport system that can deliver ablated aerosol from individual laser-sampling events within 10 ms. Several other groups are also pursuing low-dispersion LA aerosol transport; however, to provide meaningful and quantitative information about the material sampled at each LA position, we need an ICP-MS instrument that has



the speed to match the laser sampling – this is where ICP-TOFMS comes in. By combining low-dispersion LA sampling with ICP-TOFMS, we are able to generate multi-elemental high-dynamic-range quantitative elemental images at lateral resolutions down to five µm and at high speed (100 pixels/sec). The figures of merit of LA-ICP-TOFMS imaging are simply not available with other ICP-MS instrument designs.

More importantly, the elemental images that can be generated with this approach are compelling and offer access to otherwise unavailable information about trace-, minor-, and major-element spatial distributions across a specimen. In my research, I have focused on the application of LA-ICP-TOFMS imaging for analysis of geological samples, where high-resolution elemental imaging combines with observations about specimen textures to provide insight into rock formation and history. LA-ICP-TOFMS has also been shown to be very promising for elemental bioimaging. By staining tissues with isotope-labeled antibodies and performing LA-ICP-TOFMS imaging experiments, highly multiplexed distributions of biomolecules can be measured at subcellular lateral resolution. This approach is called imaging mass cytometry (commercialized by Fluidigm, USA) and was partly developed in collaboration with our lab. Apart from antibody-labeled elemental imaging, LA-ICP-TOFMS will also have substantial impact in the burgeoning field of elemental bioimaging, where LA-ICP-TOFMS approaches will help researchers hone their understanding of the roles and diagnostic potential of native metals in biological systems.

"Another promising application of ICP-TOFMS is single-particle ICP-MS analysis, in which particle number concentrations of nanoparticles and elemental compositions of single-nanoparticles are measured."

Addressing a (nano)particular problem

Another promising application of ICP-TOFMS is single-particle ICP-MS analysis, in which particle number concentrations of nanoparticles and elemental compositions of single-nanoparticles are measured. The expanding use of inorganic nanoparticles in a variety of consumer goods (everything from lotions to fabrics to food packaging) and industrial processes, makes measurement of individual nanoparticles, especially in complex natural media, an emerging analytical challenge. Nanoparticles are hard to detect – they are small (hundreds to millions of atoms), dilute in terms of total mass concentration, and often present in matrices that contain naturally occurring colloids with similar element compositions.

We have a good understanding of how nanoparticles behave within the products or processes for which they are intended; however, their eventual fate is harder to predict. Nanomaterials make their way into the environment through many routes, whether through disposal into water or solid-waste handling systems that are not designed to deal with nanomaterials, or through incidental release, such as from automotive exhaust. Predicting the impact of nanomaterials and nanoparticles being released into the environment requires the modeling of particle fate and transport, understanding routes of exposure, and performing toxicology experiments. All of these studies need to be supported by analytical measurements. It's important that we better understand the amount and chemical species of inorganic nanoparticles in natural systems, and develop systems for monitoring them.

Currently, there is no single measurement (or even a defined group of measurements) that is used to characterize a broad range of nanoparticles in natural systems. We are working to develop single-particle ICP-TOFMS as a comprehensive measurement system for non-targeted analysis of diverse inorganic nanoparticles. In single-particle ICP-MS, liquid samples with dispersed inorganic nanoparticles at environmentally relevant concentrations (~105 particles/mL) are introduced into the ICP-MS instrument. When

Multi-Element Single-Particle ICP-TOFMS Signal

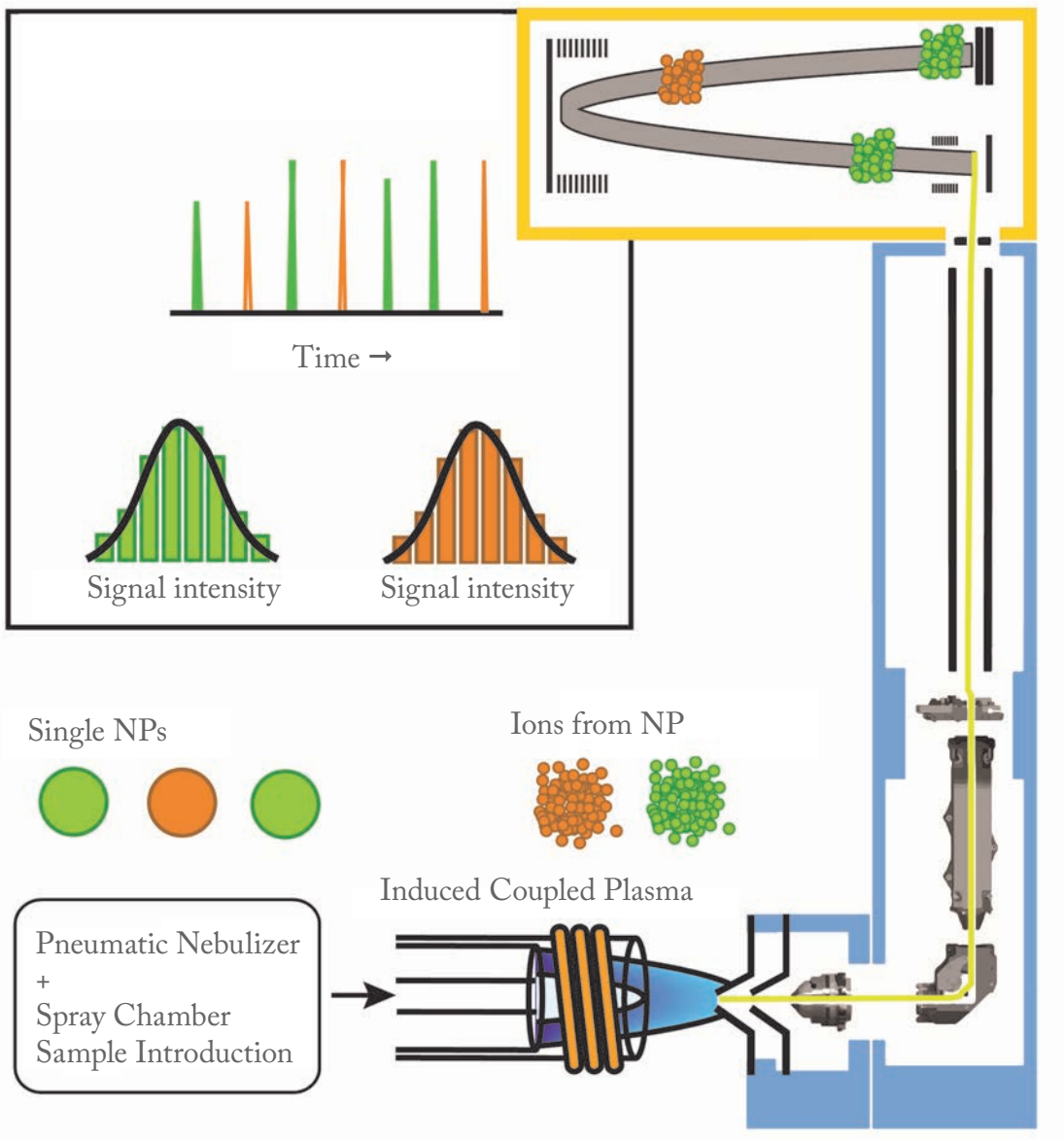


Figure 1. In single-particle ICP-TOFMS (schematic of icpTOF shown here), dispersed NPs are introduced into the ICP, where they are vaporized, atomized, and ionized. Each NP produces a dense cloud of ions that is recorded as a short burst (300–500 µs) of signal by the mass spectrometer. The magnitude of the single-particle signal correlates to particle mass and the frequency of bursts to particle number concentration. With ICP-TOFMS, ions of all m/z are measured simultaneously, so a diverse range of NPs (including multielemental NPs) can be analyzed in a single run.

> "Because the ICP is a robust atomization and ionization source, we can also quantify the amount of measured element(s) in each nanoparticle."

a nanoparticle passes through the ICP, it is vaporized, atomized, and ionized so that a dense cloud of ions is produced and detected as a signal burst by the mass spectrometer. If this signal burst is more intense than dissolved background signal levels, we are able to count the particles. In fact, single particle-ICP-MS can even detect nanoparticles in a matrix that has a higher mass concentration of analyte in the dissolved fraction than the nanoparticulate fraction. Because the ICP is a robust atomization and ionization source, we can also quantify the amount of element(s) in each nanoparticle. With conventional ICP-MS instruments, only a single isotope can be monitored across the short timescale of a single-particle event; TOFMS allows for all elements to be measured within each single-particle event. By using ICP-TOFMS, we can measure both multi-elemental signatures of individual particles and a diverse range of nanoparticle types – in a single analysis.

To illustrate the advantages of ICP-TOFMS for multi-element detection of single nanoparticles, consider our proof-of-principle study, measuring engineered nanoparticles of cerium dioxide (CeO_2) in a soil matrix with cerium-containing natural nanoparticles. In this study (1), we were able to distinguish the two types of cerium particles (engineered and natural), based on conserved multi-element fingerprints from each nanoparticle type. With a machine-learning classification algorithm, we were able to quantitatively count engineered nanoparticles in a matrix with over 100× more natural than engineered cerium nanoparticles. Importantly, without TOFMS, we would have only been able to measure the signal of 140Ce+ for each nanoparticle, and could not have differentiated the Ce-containing natural nanoparticles from the CeO_2 engineered nanoparticles – single-particle

ICP-TOFMS has enabled a real leap forward in measuring inorganic nanoparticles in complex matrices.

ICP-TOFMS impact

I have described just a few of the many applications where fast, multi-element analysis is critical. Other applications primed for ICP-TOFMS include multiplexed isotope tags for biomolecule quantification, and combination with fast separations. I believe that this next generation of commercial ICP-TOFMS instruments will become a mainstay in atomic mass spectrometry. Such instruments provide sufficient sensitivity for most trace-element analysis laboratories and offer measurement capabilities – high-speed full-spectrum detection – to meet the challenges of contemporary elemental analysis. Even more exciting is the potential for more powerful ICP-TOFMS instrumentation in future; higher sensitivity and dynamic range is certainly achievable. Emerging applications – especially in biological sciences and environmental analysis – that simply cannot be tackled with alternative elemental mass spectrometry approaches will no doubt drive further development of ICP-TOFMS. All in all, it is a fun time to be working with ICP-TOFMS – and a thrill to be a part of this renaissance of elemental mass spectrometry.

Alexander Gundlach-Graham is a Research Scientist at ETH Zurich, Switzerland.

Reference

1. *A Praetorius et al, "Single-particle multi-element fingerprinting (spMEF) using inductively-coupled plasma time-of-flight mass spectrometry (ICP-TOFMS) to identify engineered nanoparticles against the elevated natural background in soils", Environ Sci: Nano 4, 307–314 (2017).*

«I want to know immediately what substance it is!»

The Mira DS is the new handheld material identification system from Metrohm. Identify illicit substances and explosives faster and safer than ever!

- **Powerful** – Life saving decision making in the palm of your hand
- **Fast** – get results in seconds
- **Flexible** – be prepared for every treacherous situation
- **Robust** – MIL-STD-810G and IP67 certified

Watch a demo on **metrohm.com/mira-ds**

Metrohm

All Hands on Spec

Sitting Down With... Karen Faulds, Professor, Department of Pure and Applied Chemistry, University of Strathclyde, Glasgow, UK.

Were you always interested in science?
As a small child, I was always asking "Why?" – I wanted to know how things worked, and a simple answer was never good enough! I also wanted to do something practical and work with my hands, to physically create something with meaning. I think that's why I enjoy analytical science – it has a clear end goal, and many applications are instantly useful.

Why focus on SERS particularly?
I fell into SERS during my PhD. I was originally interested in forensic applications, and my PhD (partly sponsored by the Home Office) was on using SERS for the detection of drugs of abuse, with the eventual aim of developing a roadside saliva drug test, using a portable Raman instrument. Although my initial interest was in the forensic application of SERS, during my postdoc I started to look more at bioanalytical applications, in particular DNA detection.

What appealed to you about the technique?
It was a very exciting time when I first started doing SERS. Raman instrumentation was developing rapidly – going from large laboratory instruments, to benchtop, to portable. The SERS technique was starting to develop into a quantitative technique and focus was shifting away from understanding the theory (which is still an important area of course!) into developing it into useful applications. SERS substrates were becoming more reproducible, along with a growing understanding of how to control surface chemistries – giving quantitative analytical results. This opened up lots of really exciting applications for SERS, and I was hooked!

Is instrumentation still advancing?
Absolutely. We now have high-end Raman instruments for fast, high-resolution imaging, but also handheld portable devices. We're starting to reach the stage of "point-of-use" analysis – providing we get the SERS conditions and assays right, as well as creating high-resolution images that can give subcellular biochemical information.

What is the main focus of your work?
We're developing biomarker detection using SERS for different disease states – sepsis, cardiovascular disease and cancer – and bacterial detection for clinical and food safety applications.

What's the advantage of using SERS in those applications?
In short: sensitivity and multiplexing capabilities. Fluorescence can be equally sensitive, but it becomes challenging to differentiate multiple fluorescent components in a mixture because the output is a broad band spectrum; if you have multiple bands present, they start to overlap – particularly when only one excitation wavelength is used. With Raman, because you've got the molecularly specific vibrational fingerprint spectrum, you can start to deconvolute more within a mixture. We're aiming to get more information back per sample – to detect and quantify multiple analytes in one analysis. We want to be able to do one test rather than three or four, and then move these tests into "point-of-use" environments. I look forward to the day (hopefully in the near future!) when we get that test into the clinic, where it can help speed up diagnosis and lead to earlier medical interventions.

You're an advocate of multidisciplinary research – why is that so important?
In our work, clinical input is vital. For example, there is no point in developing a test or trying to understand a disease pathway that is of no help to a clinician either in terms of diagnosis and/or treatment. Early discussion with biomedical scientists and clinicians is vital in directing research towards what will actually make a difference. These problems can't just be solved by one discipline - you can't be an expert in every area, and that's why you work as part of a team. Luckily, I enjoy interacting with people! I have had the privilege to be involved in multidisciplinary teams (chemists, biologists, physicists, clinicians, engineers) for most of my career and much prefer working in a team towards a common goal, rather than working in isolation.

You're also actively involved in educating the next generation...
That's right; I work closely with Duncan Graham at Strathclyde - we run our research groups together and training PhD students is very important. In addition, I am the Strathclyde Director of the Centre for Doctoral Training in optical medal Imaging (OPTIMA), which is a joint venture between the University of Strathclyde and the University of Edinburgh. We're training 60 PhD students over five years and each student has a physical sciences advisor, and a clinical or biomedical supervisor. Students won't just get a thorough grounding in the scientific aspects, but will also cover entrepreneurship and the translation of science – the collaborative environment fosters creativity and sparks ideas for new research areas. It is a continual privilege to get the chance to work with such a fantastic group of students and postdocs.

What's next for you and your lab?
We're about to start a program on bacterial detection in food. We've looked at bacterial detection before, but never in the food production environment – so there is a whole new set of issues. Currently, testing for bacterial contamination in food is done off-site – samples are sent to a central lab, where it can take a week to get results. That's a long time if food has been contaminated. Our aim is to detect bacteria "on the line" to save both time and money. Bacteria multiply very quickly, so if contamination is suspected, everything has to be sterilized and production has to start again – it's a costly process. Faster testing using SERS could also help with traceability – where has that contamination come from, and how has it spread?

SPEX CertiPrep®

CERTIFIED REFERENCE MATERIALS FOR EVERY APPLICATION

INORGANIC

AA | ICP | ICP-MS
LC-ICP-MS | XRF

ORGANIC

GC | GC/MS
HPLC | LC/MS

ACCREDITATION

Accredited by A2LA for
ISO/IEC 17025 & ISO Guide 34

SPEX CERTIPREP

Serving the scientific
community since 1954

Accredited by A2LA for ISO/IEC 17025:2005 and ISO 17034:2016.
Certified by DQS to ISO 9001:2015.

SPEX CertiPrep

US Address:
203 Norcross Avenue • Metuchen, New Jersey 08840
732.549.7144 • crmsales@spexcsp.com • www.spexcertiprep.com

UK Address:
2 Dalston Gardens, Stanmore HA7 1BQ, UK
+44 (0) 208 204 6656 • SPEXEurope@spex.com • www.spexeurope.com