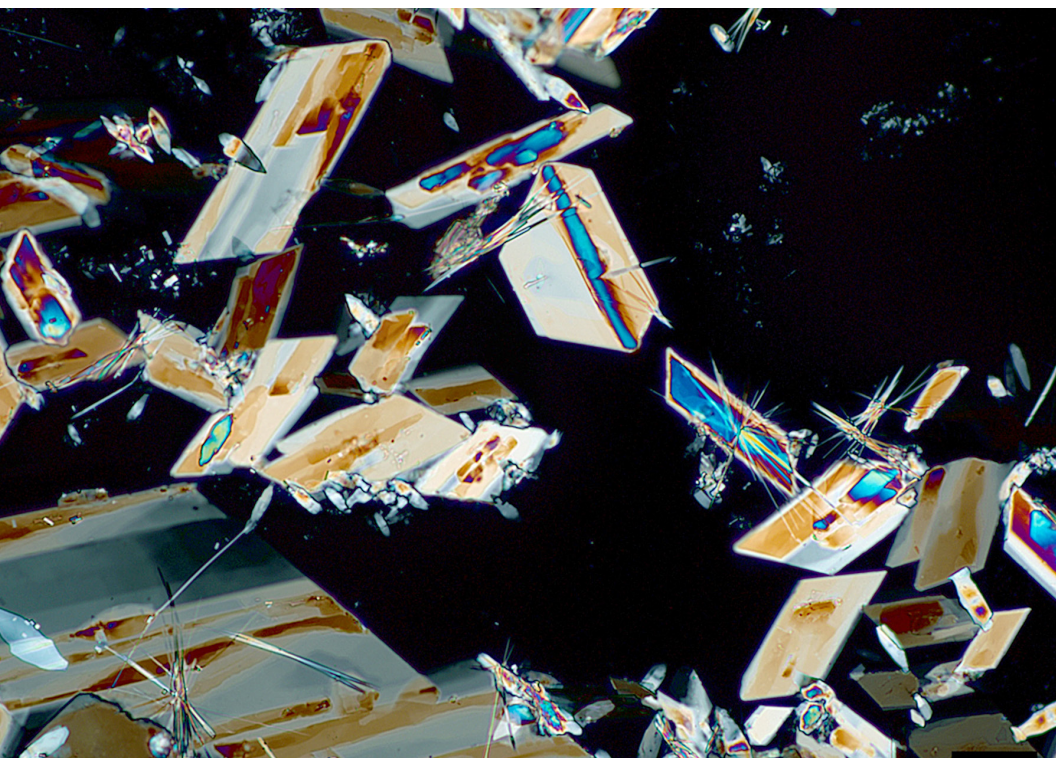


# INTER/MICRO 2017

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**An International Microscopy Conference**

June 12–16, 2017 • Chicago



*Sponsored and hosted by*  
**McCrone Research Institute**

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# Welcome to Inter/Micro 2017

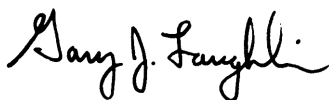
Microscopy is defined as the application of any magnified-image process that enables the visualization of objects that are otherwise invisible to the unaided human eye. Individuals using the instruments capable of producing enlarged images of tiny objects are called microscopists. Those individuals, properly trained, are able to extend their observation and interpretation of large (macroscopic) objects down to submicrometer-size objects that would be difficult, if not impossible, to identify by any other means. McCrone Research Institute and Inter/Micro are dedicated to the advancement of applied microscopy. This includes all light and electron microscopes, microspectroscopes, microprobes, automatic image analyzers, and other microscopes based on X-rays, sound, protons, etc.

Inter/Micro presentations from some of the world's leading amateur and professional microscopists will cover new techniques for improving contrast, increasing resolution, and obtaining and recording more characterization data. Attendees will learn how new, and not-so-new, techniques and instruments are used everyday to solve important problems.

This year marks the 69th anniversary of the Inter/Micro conference, which was introduced by Dr. Walter C. McCrone in 1948 and is now held annually at the McCrone institute in Chicago. Inter/Micro gives all of us the opportunity to get up-to-date on new instruments, new techniques, and new applications of microscopy and microanalysis.

We encourage all speakers to submit research papers based on their Inter/Micro presentations for publication in *The Microscope*, the official journal of this conference. Papers will be peer reviewed and published in the order they are received.

Thank you for attending and participating at Inter/Micro 2017.



Gary J. Laughlin  
Chairman, Inter/Micro

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**ON THE COVER:** Well-formed, birefringent crystals resulting from a microcrystal test for the drug *l*-pseudoephedrine using a dilituric acid reagent; crossed polars. From *A Modern Compendium of Microcrystal Tests for Illicit Drugs and Diverted Pharmaceuticals*, McCrone Research Institute, Chicago.

*Monday, June 12*

## **Techniques and Instrumentation**

*8:00 a.m.–5:00 p.m. Registration and packet pickup, McCrone Front Desk*

*9:00 a.m.–12:10 p.m. Morning Session, McCrone Lecture Room*

*Chair: Andrew A. Havics — pH2, LLC*

### **The Forensic Analysis of 3-D Printer Dust Particles**

Kelly Brinsko Beckert — Microtrace, LLC

### **Raman Has Never Been So Sweet**

Brendan Nytes — Microtrace, LLC

### **Fulgurites and Forensic Science: A Novel Application of Forensic Geology**

Christopher S. Palenik — Microtrace, LLC

*Morning Break*

### **Validating a Raman Microspectrometer in an ISO-Accredited Forensic Laboratory**

Andrew M. Bowen — U.S. Postal Inspection Service

### **Recent Advances in Raman Imaging Microscopy**

Alexander Rzhevskii — Thermo Fisher Scientific

### **Contrast in Reflected Light Microscopy**

Andrew A. Havics — pH2, LLC

*12:10–2:00 p.m. Lunch Break, McCrone Garden*

2:00–5:00 p.m. Afternoon Session, McCrone Lecture Room

Chair: John A. Reffner — John Jay College, CUNY

**Observations on Temperature Variations in Liquid Mounts  
During Light Microscopical Investigations**

Jan Burmeister

**A History of Ammonium Nitrate Disasters; Why is Ammonium  
Nitrate in My Car's Airbag Inflator?**

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**Takata Airbag Death No. 10 — Investigation to Determine  
Projectile Source**

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**Topics in Nonwoven Structures and Fibers**

Walter J. Rantanen — SGS IPS Testing

*Afternoon Break*

**A Forensic Microscopy Approach to Identifying Particulate  
Matter Observed in a Sterile Ophthalmic Solution on Stability**

Mary Lee Ciolkowski — Pharma Technical Services, Bausch & Lomb, a division of Valeant Pharmaceuticals

**“EXCELIBR”: An Excel Spreadsheet for Solving the Optical  
Orientation of Uniaxial and Biaxial Crystals**

C.J. Steven — Geological Sciences, University of Idaho

**An Excel(ent) Guide for Fiber Identification Using Polarized  
Light Microscopy**

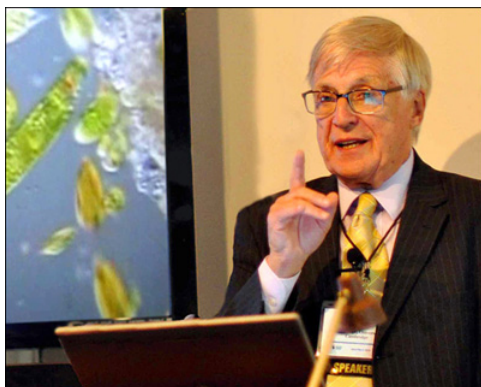
John A. Reffner — John Jay College CUNY

*See Monday presentation abstracts on page 14.*

*Monday, June 12*  
**An Evening with Brian**  
**“100 Talks in 60 Minutes”**

*5:30–7:00 p.m. Mediterranean cuisine dinner, McCrone Garden, \$30. (You may pay for the dinner at the front desk if you did not pre-register.)*

*7:00–8:00 p.m. An Evening with Brian presentation with **Brian J. Ford**, McCrone Lecture Room, free*



This evening's presentation will be the 100th that Brian has given at Inter/Micro since his first in 1969, and it will look back at the previous 99 presentations. The subjects range from color vision in dogs, and a detached leg found in a reservoir, to pioneering papers on the role of microorganisms in pollution control, and food production — not to mention, the world's worst microscopy. Some of these talks gave rise to TV programs and books, but whether they can all fit into the space of an hour remains to be seen.

*When he is not speaking at Inter/Micro, Professor Ford travels the world as a keynote speaker and presents his work on television and radio. Ford is a leading authority on the microscope and a best-selling author, whose research is widely quoted in journals and encyclopedias. He is the author of the Critical Focus column, published quarterly in The Microscope journal. Ford was named an Honorary Fellow of the Royal Microscopical Society this year and has served as a fellow of the Open University, fellow and president of past students at Cardiff University, visiting professor at Leicester University, and an associate of Caius College, University of Cambridge, U.K. He has given his Evening with Brian presentations at Inter/Micro for more than 30 years.*

*Tuesday, June 13*

## **Environmental and Industrial Microscopy**

*8:00 a.m.–5:00 p.m. Registration and packet pickup, McCrone Front Desk*

*9:00 a.m.–12:10 p.m. Morning Session, McCrone Lecture Room*

*Chair: Mickey Gunter — Geological Sciences, University of Idaho, Moscow*

### **Application of Rietveld Refinement to Forensic Samples**

Joseph Insana — Microtrace, LLC

### **Does it Fluoresce? Explorations in a Spectral World**

Charles Mazel — NIGHTSEA

### **How Can We Measure “Shape”?**

John C. Russ — North Carolina State University, Materials Science Department

*Morning Break*

### **Microscopical Analyses of Construction Products Derived from Industrial Waste**

Arthur Struss — USG Corp. (retired)

### **Microscopy of Asbestos-Cement Pipe**

James R. Millette — Millette Technical Consulting

### **ISO 22262: The International Standard for Determination of Asbestos in Bulk Materials**

Eric J. Chatfield — Chatfield Technical Consulting Limited

### **Examples of Expert vs. Expert Civil Litigation Errors in Dealing with the Purported Asbestos Content of Talc**

Mickey Gunter — Geological Sciences, University of Idaho, Moscow

*12:10–2:00 p.m. Lunch Break, McCrone Garden*

*2:00–5:00 p.m. Afternoon Session, McCrone Lecture Room*

*Chair: Sebastian B. Sparenga — McCrone Research Institute*

**Look-Alikes Type 1: Objects Within an Order of Magnitude of Size**

Andrew A. Havics — pH2, LLC

**Microscopy in Polymer Product Development**

John R. Reffner — Analytical Sciences, The Dow Chemical Company

**Thermal Recombination of Clay Mineral Components of Ancient Mayan Ceramic Ware as a Means of Differentiating Indigenous Versus Tradeware Using X-ray Diffraction Analysis**

Wayne C. Isphording — Tulane University, Department of Continuing Education

*Afternoon Break*

**What Is This? The Peculiar Tale of a Food Contaminant**

Jason C. Beckert — Microtrace, LLC

**Microbe Power in a Prehistoric Pizza**

Brian J. Ford — Caius College, University of Cambridge, U.K.

**A Microscopical Trip Down Memory Lane: 40+ Years of McCrone Christmas Card Photomicrography**

Sebastian B. Sparenga — McCrone Research Institute

*See Tuesday presentation abstracts on page 23.*



## *Tuesday, June 13* **Exhibitor Booth**

*9:00 a.m.–5:00 p.m. Tuesday, June 13 and Wednesday, June 14,  
McCrone Classroom*

**L**earn about the latest fluorescence microscopy innovations and products from our exhibitor, NIGHTSEA.

## **SMSI Silent Auction**

*12:10–5:00 p.m. Tuesday, June 13 and 9:00 a.m.–3:50 p.m.  
Wednesday, June 14, McCrone Classroom*

**B**id on microscopy equipment and other related items of interest at the annual silent auction benefitting the State Microscopical Society of Illinois (SMSI). Winners will be announced Wednesday afternoon after the speaker presentations.

## **Reggie's Rock Club Rooftop Dinner**

*5:30–8:30 p.m. Reggie's Rock Club, 2105 S. State Street, \$45 (free for  
ASTEE members)*

**U**nwind on a pleasant summer evening with fellow Inter/Micro attendees and sponsors for refreshments and dinner on Reggie's rooftop patio, located just a few blocks away from the McCrone institute. Transportation to Reggie's from McCrone will be provided by Reggie's colorful bus. The rooftop dinner is sponsored by Cargille and the American Society of Trace Evidence Examiners (ASTEE).

*Wednesday, June 14*  
**Chemical and Forensic Microscopy**

*8:00 a.m.–5:00 p.m. Registration and packet pickup, McCrone Front Desk*

*9:00 a.m.–12:10 p.m. Morning Session, McCrone Lecture Room*

*Chair: Skip Palenik — Microtrace, LLC*

**Torture Test: Microscopic Changes in Markings Made by a Tavor Rifle**

Peter Diaczuk — Pennsylvania State University

**Investigation of Malaysia Airlines Flight MH17**

Peter Zoon — Netherlands Forensic Institute

*Morning Break*

**Microcrystal Tests for the Detection of Butylone, Methylone, and Ethylone**

Shan Mei Jones — University of Illinois at Chicago Graduate Program, in association with McCrone Research Institute

**The Effect of Ultraviolet Radiation on the Microspectrophotometry (MSP) of Dyed Fibers — Phase 1**

Meggan B. King — McCrone Research Institute

**A Forensic Study of Known Toner Nanoparticles**

Katie M. White — Microtrace, LLC\*

**Some Lesser Known Microchemists and a Look at Some of Their Work**

Skip J. Palenik — Microtrace, LLC

*12:10–2:00 p.m. Lunch Break, McCrone Garden*

*2:00–5:10 p.m. Afternoon Session, McCrone Lecture Room*

*Chair: JenaMarie Baldaino — FBI Laboratory, Counterterrorism and Forensic Science Research Unit, Visiting Scientist Program*

**Possible Degradation Mechanisms of Antemortem Hair Roots Containing Induced PMRB-Like Features**

Barbara L. Fallon — Federal Bureau of Investigation Laboratory, Counterterrorism and Forensic Science Research Unit

**The Trotter Collection: Microscopical Analysis of Human Scalp Hairs for 21st Century Research Questions**

Sandra Koch — Pennsylvania State University, Department of Anthropology

**Forensic Drug Identification by GC/MS and PLM**

Andrew M. Bowen — U.S. Postal Inspection Service

**Imaginary Microorganisms and Impossible Worlds**

Brian J. Ford — Caius College, University of Cambridge, U.K.

*Afternoon Break*

**Using SEM-EDS for Quantitative Forensic Glass Comparisons: Some Things to Think About**

Thomas A. Kubic — John Jay College and The Graduate Center, CUNY; Thomas A. Kubic and Associates

**Morphology and Microanalysis of Aluminum Powders**

JenaMarie Baldaino — FBI Laboratory, Counterterrorism and Forensic Science Research Unit, Visiting Scientist Program

*See Wednesday presentation abstracts on page 32.*

*Wednesday, June 14*  
**State Microscopical Society of Illinois**  
**2017 Awards Dinner and Live Auction**

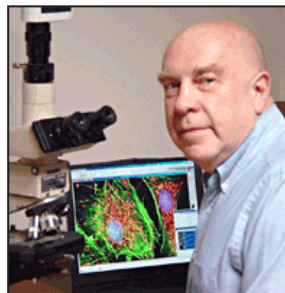
**Dr. John C. Russ • 2017 SMSI August Köhler Award Recipient**

*Presented at Café Bionda,  
1924 S. State Street, Chicago; \$70*

*6:30–7:30 p.m. Social hour and live auction  
hosted by Brian J. Ford*

*7:30–8:30 p.m. Dinner*

*8:30–9:30 p.m. Award announcement and  
presentation*



Join Inter/Micro and the State Microscopical Society of Illinois as they honor Dr. John C. Russ with the 2017 August Köhler Award. Dr. Russ is a world-leading expert in digital image processing and analysis for a variety of disciplines. He is an emeritus professor in the Department of Materials Science and Technology, College of Engineering at North Carolina State University in Raleigh, NC. During his 50-year career, he has focused on imaging for technical, scientific, and forensic applications, and on studying the microstructure and surface topography of metals and ceramics.

Dr. Russ is the author of many books, including *The Image Processing Handbook*, *Image Analysis of Food Microstructures*, and *Forensic Uses of Digital Imaging*. He currently teaches imaging workshops worldwide, consults with companies, and provides expert testimony in criminal and civil cases. In 2006, the New York Microscopical Society honored Dr. Russ with the Ernst Abbe Memorial Award for his contributions to the field of microscopy as a developer of computer-assisted microscopy and image analysis.

*Thursday–Friday, June 15–16*  
**Workshop: Image Processing and  
Measurement**

*9:00 a.m.–5:00 p.m., McCrone Classroom and Laboratory; \$500*

**I**mages are a primary source of information in many fields of science, and especially in microscopy. This two-day workshop, taught Dr. John C. Russ, will emphasize the tools, methods, and workflow used to extract relevant and accurate information from digitized images through the step-by-step application and comparison of algorithms. A variety of images, many of typical specimens such as polished metal surfaces, biological thin sections, microscopic particles on slides, and SEM and TEM images will be studied in the hands-on exercises. Several different public domain and commercial software programs will be used to process and measure images. Students will also learn image-correction techniques such as adjustments for color, brightness, contrast, illumination, and noise reduction. Special attention will be given to comparing different algorithms and explaining their operation. Participants will receive a copy of *The Image Processing Cookbook* for use as a text for the workshop. This publication explains and illustrates the image processing and some of the measurement, and covers the operations using ImageJ, Matlab, ImageProPlus, Photoshop, etc. that will be covered in the workshop.

***Dr. John C. Russ** is a world-leading expert in digital image processing and analysis for a variety of disciplines. He is an emeritus professor in the Department of Materials Science and Technology, College of Engineering at North Carolina State University in Raleigh, NC. During his 50-year career, he has focused on imaging for technical, scientific, and forensic applications, and on studying the microstructure and surface topography of metals and ceramics. Dr. Russ is the author of many books, including *The Image Processing Handbook*, *Image Analysis of Food Microstructures*, and *Forensic Uses of Digital Imaging*. He currently teaches imaging workshops worldwide, consults with companies, and provides expert testimony in criminal and civil cases.*

## PRESENTATION ABSTRACTS

*Monday, June 12*

### **Techniques and Instrumentation**

#### **The Forensic Analysis of 3-D Printer Dust Particles**

Kelly Brinsko Beckert and Christopher S. Palenik — Microtrace, LLC

3-D printers are becoming increasingly efficient and economical and thus more widely accessible to the public. Previous research has documented the release of dust particles during the printing process, however, little is known about their morphology and other characteristic features. This study was undertaken as part of a federal research cooperative agreement (NIJ Award No. 2015-DN-BX-K033) to characterize these particles so that they may be collected, recognized, and analyzed appropriately. Samples were collected from a variety of 3-D printers, representing both consumer- and commercial-grade models. 3-D printers use thermoplastic filaments, typically polylactic acid (PLA) or acrylonitrile butadiene styrene (ABS), although others may be used, including nylon, polyvinyl acetate, and polyurethane. Cotton or polyester-flocked swabs were used to collect dust from various surfaces within the printer chamber and from surrounding areas up to 10 feet away. Particles produced from ABS filaments are most easily recognized based on color and rounded morphology via light microscopy. Fourier transform-infrared (FT-IR) spectra of the particles confirmed the identification of the ABS polymer. Pigments and the ABS polymer matrix were also identified using Raman microspectroscopy. Dust from PLA printers consistently contained finer, submicron-sized particles (relative to background levels) that could be observed by field emission scanning electron microscopy (FEM); however, the size of the particles precluded their specific identification as PLA. This presentation will detail the collection procedures employed to find, isolate, identify, and compare 3-D printer dust particles and include a discussion of their potential applications and limitations as forensic evidence.

## **Raman Has Never Been So Sweet**

Brendan Nytes — Microtrace, LLC

Of the myriad different substances that we encounter in our laboratory, carbohydrates are one of the more common materials observed. They may be present as part of a food product as a starch, in a textile as a cotton fiber, or in pharmaceuticals as a microcrystalline cellulose excipient, etc. These materials are typically identified by their microscopic morphology and optical properties. Sugar, another carbohydrate that is often observed, can also be identified by these characteristics. In some instances, the optical properties may not be readily apparent, and a specific identification of the type of sugar may require a different analytical technique. Well established identification methods, e.g., chromatography, can be time consuming and require sample preparation. Raman spectroscopy has shown promise as a means by which to differentiate carbohydrates, including sugars. The advantage to Raman is that it can be conducted in situ with little or no sample preparation. This talk explores the analysis of various carbohydrates, including sugars and sugar products, by Raman microspectroscopy.

## **Fulgurites and Forensic Science: A Novel Application of Forensic Geology**

Christopher S. Palenik — Microtrace, LLC

The term fulgurite, which derives from the Latin fulgur (meaning “thunderbolt”), was originally intended to refer to amorphous silica produced by lightning strikes. Over time, this term has been more broadly applied throughout the literature to include amorphous silica, or related compositions, produced as a result of high temperature and/or high pressure events that can include anthropogenic activities. The identification and characterization of a suspected fulgurite through polarized light microscopy, electron microscopy, and Raman microspectroscopy can produce a wealth of information about its identity and its pressure/temperature history. The combination of analytical methods can be used to place constraints upon the conditions of fulgurite formation, distinguish between natural and an-

thropogenic origins, and potentially provide some insight into the relative timing of fulgurite formation. This information can be used in a forensic context to provide information about the start of fires occurring under certain circumstances.

### **Validating a Raman Microspectrometer in an ISO-Accredited Forensic Laboratory**

Andrew M. Bowen — U.S. Postal Inspection Service

Raman microspectroscopy has a number of advantages over more traditional instrumentation used in forensic laboratories, including its excellent spatial resolution and confocal capabilities. These make it possible to definitively identify numerous particulate components in complex mixtures, non-destructively, with minimal sample quantity. This presentation will discuss the recent validation of a two-laser Raman microspectrometer in an ISO 17025-accredited forensic laboratory. The validation design and results obtained will be described, together with lessons learned during the process. Practical advice for scientists who plan on conducting future validation studies will be shared, along with issues of potential concern during general use of Raman microspectroscopy in a forensic laboratory.

### **Recent Advances in Raman Imaging Microscopy**

Alexander Rzhevskii — Thermo Fisher Scientific

Raman microscopy has become one of the most powerful instrumental techniques for a diverse range of applications in both research and analytical laboratories. In this presentation, I will consider the recent technological advancements in Raman spectral imaging, including EMCCD detectors, fast moving sampling stages, advanced software, etc. These advancements offer additional analytical possibilities in analyzing two- and three-dimensional spatial distributions of materials on a sub-micrometer scale with an information-rich content, while maintaining a reasonable image acquisition time.

Raman imaging microscopy is a valuable technique for hyperchemical characterization of micro- and nanostructures. The essential advantages of Raman spectral imaging will be illustrated with



examples in polymer, pharmaceutical, gemological, semiconductor, ceramic, and biological applications.

## **Contrast in Reflected Light Microscopy**

Andrew A. Havics — pH2, LLC

Detection and resolution of objects in microscopy rely heavily on contrast. In 2011, the author presented a series of contrast techniques almost exclusively for transmitted light microscopy. There are comparably contrast techniques for reflected light microscopy as well. These methods include darkfield (DF), reflected Rheinberg illumination, Rheinberg sandwich-style illumination, polarized light microscopy (PLM), sensitive tint addition, Nomarski Differential Interference Contrast (DIC), staining of specimens, etching (thermal, selective dissolution, interference film, decorative), phase contrast microscopy (PCM), fluorescence, reflection contrast microscopy (RCM), metallization, fringe-based (monochromatic DIC and interference), and color filtration.

## **Observations on Temperature Variations in Liquid Mounts During Light Microscopical Investigations**

Jan Burmeister

The precise measurement of the refractive indices of transparent small particles in an immersion liquid mount, using classical light microscopy and polarized light microscopy techniques, requires exact knowledge of the mount temperature, and that the necessary temperature coefficient calculation be applied to the liquid before stating final measurement results. Commercially available refractive index standards for microscopy are supplied as liquids in bottles with the temperature coefficient printed on the label.

Previous proposals for temperature measurement in a microscope's light path, as published in handbooks and online articles, may be misleading due to the large and thermally inert mass of the mercury-containing thermometer bulbs that are routinely used today.

An experimental setup was devised and duly calibrated using a

miniature NTC thermistor as the temperature sensor and a high-resolution ohmmeter to observe temperature effects in a way that is much closer to reality than in older, obsolete procedures. The results for this particular setup show that only negligible thermal effects are observed in mounts containing three drops of immersion liquid. Extrapolation calculations for situations with less liquid in the mount show that, in such cases, the expectable temperature effects are also small to negligible.

## **A History of Ammonium Nitrate Disasters; Why is Ammonium Nitrate in My Car's Airbag Inflator?**

Richard S. Brown — MVA Scientific Consultants

The storage and transportation of ammonium nitrate has resulted in some of the most horrific and deadly explosions in the last 100 years. The storage and use of ammonium nitrate as an explosive and as a fertilizer remains widespread. Examples of past disasters include but are not limited to:

- April 2, 1916: — “The Great Explosion,” Faversham, Kent, England, 115 dead
- July 26, 1921 — Kriewald, Germany, 19 dead
- Sept. 21, 1921 — BASF Oppau, Germany, 561 dead
- April 29, 1942 — Tessenderlo, Belgium, 189 dead
- April 16, 1947 — Texas City, SS Grandcamp, several hundred dead (all but one member of the fire department died)
- July, 28 1947 — Brest, France, 29 dead
- April 17, 2013 — West Texas, 15 dead

As a result of these “accidents” and subsequent investigations, the processing and storage of ammonium nitrate has changed. How ammonium nitrate reacts in the presence of variable temperature and humidity remains somewhat of a mystery. A summary of past research and incidents serves as a reminder of the explosivity and unpredictability of ammonium nitrate. So why is it in my car's airbag?

## **Takata Airbag Death No. 10 — Investigation to Determine Projectile Source**

Richard S. Brown — MVA Scientific Consultants

In December 2015, the driver of a 2006 Ford Ranger was found dead after colliding with an object in the road. The pickup truck's airbag had deployed during the collision. A subsequent autopsy revealed an apparent gunshot wound in the driver's neck. We were requested by the coroner's office to determine the composition and source of the projectile slug. The suspected source of the slug was the driver's side airbag module located in the steering wheel of the vehicle. Examination of the components of the airbag module revealed that the airbag inflator had ruptured from excessive pressure and that the initiator, inside of the airbag inflator, had fractured into two pieces. One of the pieces penetrated the driver's neck; the other piece was found pressed onto the steering wheel mounting nut. The investigation was supplemented by examining an exemplar airbag inflator using 3-D computer tomography. The types of materials analyzed included labels on the inflator, adhesive, and fracture patterns. Fluorescence microscopy, scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM-EDS), calipers, and a microchemical test for ammonium ions were used during the investigation. The airbag inflator was manufactured by the Takata Corporation and contained ammonium nitrate, which is at the center of the largest recall in automotive history.

## **Topics in Nonwoven Structures and Fibers**

Walter J. Rantanen — SGS IPS Testing

Nonwovens are formed with natural and/or synthetic fibers and produced using different web-forming systems. Various nonwoven fabrics that are produced include wipes, personal care products, toweling, filters, and upholstery backing. The use of light microscopy is beneficial when analyzing nonwoven fabrics due to the complex array of different forming systems and fiber types available. Microscopy can compliment the chemical analysis and some of the physical testing to achieve complete data on a material. Analysis of the fibers by testing methods with the microscope can also deter-

mine qualitative and quantitative information, where other methods lack this ability.

### **A Forensic Microscopy Approach to Identifying Particulate Matter Observed in a Sterile Ophthalmic Solution on Stability**

Mary Lee Ciolkowski — Pharma Technical Services, Bausch & Lomb, a division of Valeant Pharmaceuticals

This presentation will describe a microscopical investigative approach to characterize particles that were observed during stressed stability testing of an ophthalmic solution formulation. Topical ophthalmic solutions should be “essentially free” of visible particulates upon inspection, as specified in the U.S. Pharmacopeia (USP). Ophthalmic solutions are also required to meet subvisible particulate matter requirements per USP Chapters <789/788> at the time of product release and during stability testing to verify manufacturing cleanliness and support shelf life of the finished product. The analytical characterization work needed in cases where USP <788/789> failures occur is considered trace or ultra-trace analysis. For example, the USP <789> limit for particles 10–25  $\mu\text{m}$  is 50 particles/mL. If this limit is exceeded due to a needle-shaped particulate with a density of  $1.5 \text{ g/cm}^3$ , this would correspond to ~8 ppb on a concentration basis for a 5 mL product fill size. Particle isolation and enrichment practices coupled with direct sample analysis using microanalytical techniques are essential to productive problem solving during particulate matter failure investigations, due to the inherent trace concentration levels. A progressive problem-solving approach based on forensic microscopy was utilized to isolate and characterize particulate matter in the ophthalmic formulation and identify its potential source.

### **“EXCELIBR”: An Excel Spreadsheet for Solving the Optical Orientation of Uniaxial and Biaxial Crystals**

C.J. Steven and M.E. Gunter — Geological Sciences, University of Idaho

The polarized light microscope remains the single most useful tool in identifying minerals. Using a spindle stage, the microscopist

can orient a crystal's principle refractive index (RI) vectors with the polarizer. Finding the principle RI vectors is accomplished by using either conoscopic methods, or more simply, using extinction data as inputs in the program EXCALIBR. The use of EXCALIBR has major advantages over conoscopic methods, but it is hindered by interface and compatibility issues. Presented here is a Microsoft Excel spreadsheet, which the authors have named "EXCELIBR," which performs operations similar to EXCALIBR, including solving for the optical orientation of biaxial or uniaxial minerals using extinction data. With Excel as the interface, EXCELIBR is more accessible, familiar, and versatile for the user. This spreadsheet is useful for preliminary screening of a crystal for X-ray studies, optical characterization of minerals, and rapid mineral identification. Included with the crystal orienting calculations are tabs for double variation, RI modeling, and compensator plate calculations.

### **An Excel(ent) Guide for Fiber Identification Using Polarized Light Microscopy**

John A. Reffner, Ph.D. and Samuel Kaplan B.S. — John Jay College, CUNY

A Microsoft Excel spreadsheet is a useful tool to guide the analysis of fibers using polarized light microscopy (PLM). The classical PLM method of fiber identification is to measure the principal refractive indices using the Becke line to determine the refractive index of the fiber relative to a surrounding mounting medium. Determining principal refractive indices requires the mounting of fibers in several different refractive index liquids. Synthetic fibers and some natural fibers behave optically as uniaxial crystals. As such, fibers have a continuous gradient of refractive index. The gradient ranges from high to low, corresponding to the principle refractive index values. When fibers are examined using polarized light, the principal refractive index values are aligned parallel and perpendicular to the fiber axis. If a fiber is mounted in a liquid with an index of refraction between the principal values, then at some angle of fiber (and microscope stage) rotation, the Becke line will vanish. The angle between

the fiber axis and the position at which the Becke line disappears is measured. This angle is a function of the fiber composition and refractive index of the liquid. If the refractive index of the liquid and the principal values of the fiber are known, then the angle of rotation can be calculated. Using the equation of an ellipse in spherical coordinates, an Excel spreadsheet was developed that generates tables of data that aid in fiber analysis. This presentation will give examples that demonstrate the utility of this guide for improving the efficiency of fiber identification.

*Tuesday, June 13*

## **Environmental and Industrial Microscopy**

### **Application of Rietveld Refinement to Forensic Samples**

Joseph Insana and Christopher S. Palenik — Microtrace, LLC

Soil is analyzed and examined in a variety of ways for a multitude of reasons. Powder X-ray diffraction is used to identify different mineral phases found within a soil sample. Interpretation of these diffraction patterns in terms of phases present and the amount of each phase can be achieved using a technique known as Rietveld refinement. This is done by fitting a synthetic pattern to the sample pattern through an iterative process. The pattern fitting process as a whole takes into account instrumental variables that affect the pattern, as well as sample properties that include major and minor phases, amorphous contributions, the crystal structure, strain, and texture. When variables are properly constrained, the resulting output can provide an accurate quantitative analysis, which can be evaluated in light of the fit quality compared to the experimental results. By using this approach, Rietveld refinement may provide an alternative approach to forensic soil analysis, providing quantitative results that can be objectively evaluated in terms of quality and uncertainty. This presentation will illustrate the basic principles of Rietveld refinement and its application to a range of synthetic samples, including a blind, unknown sample.

### **Does it Fluoresce? Explorations in a Spectral World**

Charles Mazel — NIGHTSEA

What do bread mold, nylon granules, nematodes, microplastics, bumblebees, epoxy, mouse ears, and electronic components have in common? These are among the variety of things that have been sent to NIGHTSEA to check the capability of our systems. As a purveyor of equipment for viewing and documenting fluorescence, NIGHTSEA encounters a wide variety of opportunities to apply the technique in new applications, overcome challenges in optimizing

a system, and clarify misconceptions on the part of customers.

This talk will cover some of the situations we encounter in working with existing and potential users: testing samples in-house, over-reliance on tables that list only excitation and emission peak wavelengths, neglect of the visible as a source of excitation energy, and more.

Answering the questions “Does it fluoresce?” and “How can I make it fluoresce better?” can involve both empirical and first-principles approaches. These can lead us to off-the-shelf solutions and provide guidance in developing new products and accessories. They can also take us to interesting places, such as a bat cave in New Jersey.

### **How Can We Measure “Shape”?**

John C. Russ — North Carolina State University, Materials Science Department

The identification and classification of objects, both microscopic and macroscopic, is a challenging but important goal in many fields, including research, industrial quality control, and forensics. Color and size are often useful criteria but may vary depending on magnification, illumination, and other factors. Shape is very important for human recognition, but this must be expressed in numeric measurements for computer usage. Several approaches to shape measurement are described and compared. Some of these are complete (able to reproduce the original shape exactly) and some are reductive, extracting a small number of hopefully significant values such as formally dimensionless ratios of dimensions. Some, such as Fourier or wavelet analysis or fractal dimension, depend only on the boundary of the object, while others, such as moments, topology, and cross correlation, use the interior as well. These methods also differ in the amount of computation required and their sensitivity to the quality of the digitized images. But all depend to a considerable degree on the size and quality of the training population used, and on the application of proper statistical analysis methods.



## **Microscopical Analyses of Construction Products Derived from Industrial Waste**

Arthur Struss — USG Corp. (retired)

Companies such as USG Corp. turn air pollutants and waste materials into useful products. Sulfur dioxide emissions from coal-fired power plants are converted to gypsum for wallboard. 100% of the paper used on wallboard is from recycled paper. Fly ash from coal combustion is used in cement. Waste slag from iron and copper smelters is spun into mineral wool for fire-rated ceiling tile and for insulation.

Microscopical analyses are an essential part of utilizing industrial waste in construction products. Flue gas desulfurization gypsum is analyzed microscopically to determine crystal size and morphology and to identify contaminants. Measurement of mineral wool fiber diameter is important, as well as the study of cement porosity and the void structure in wallboard. Examples of these investigations will show how microscopy is used to solve problems related to using industrial waste in construction products.

## **Microscopy of Asbestos-Cement Pipe**

James R. Millette — Millette Technical Consulting

As with most asbestos analysis concerns, microscopy plays an important part in the studies of asbestos-cement (A-C) pipe. There are millions of miles of A-C pipe throughout the world carrying drinking water and wastewater and used as electrical cable conduit. In the U.S., Johns-Manville was a major producer of A-C pipe, and their product name Transite has become a generic term for A-C pipe. CertainTeed was another major U.S. producer in the A-C pipe industry. In 2009, the owner of Eternite, Stephan Schmidheiny, was prosecuted in a criminal trial in Turin, Italy for the asbestos exposures connected with asbestos-cement products. Eternite was a major worldwide producer of A-C pipe with subsidiaries in Europe, South America, and South Africa.

Bulk analysis with polarized light microscopy (PLM) is the usual type of analysis done for A-C pipe. Our analysis of A-C pipe manufactured by CertainTeed and Johns-Manville found them to con-

tain 10–25% chrysotile asbestos and 4–15% crocidolite asbestos by volume. Transmission electron microscopy (TEM) is the mandated instrument in the analysis of drinking water that flows through A-C pipe. The U.S. EPA maximum contaminate level for asbestos in drinking water is 7 million asbestos fibers longer than 10  $\mu\text{m}$ .

Only waters of certain corrosive nature will cause the deterioration of the cement binder in A-C pipe and allow the release of asbestos fibers into the water. Some A-C pipe has been in the ground for decades without any deterioration. However, other A-C pipe systems have deteriorated to the point where shower and sink aerators have been clogged with asbestos fibers released into the water. Scanning electron microscopy (SEM) was very useful in studying the key factors in developing a water stability index that predicts when A-C pipe will be attacked. A transmission electron microscope is typically used to measure the amount of asbestos in the air released from water that has flowed through an A-C pipe into the air through showers and humidifiers. Re-suspension of waterborne asbestos into the air can also occur when water dries on a surface and the fibers are dispersed during a cleaning event. The air analyses follow standard microscope airborne asbestos protocols.

As determined by standard asbestos microscope techniques, significant airborne levels of asbestos can be generated during the installation and repair of A-C pipe, especially when it is cut with a power saw. Airborne releases of asbestos fibers have also been found during situations when pieces of A-C pipe bump or rub against each other.

## **ISO 22262: The International Standard for Determination of Asbestos in Bulk Materials**

Eric J. Chatfield — Chatfield Technical Consulting Limited

The proposal to develop an ISO standard method for determination of asbestos in bulk materials was initially discussed by the ISO/TC 146/SC 3/WG1 Working Group in 1998. ISO 22262 is the method that was developed over 18 years by the working group of international members. ISO 22262 was published in three parts: ISO 22262-1, “Sampling and qualitative determination of asbestos in

commercial bulk materials” in 2012; ISO 22262-2, “Quantitative determination of asbestos by gravimetric and microscopical methods” in 2014; and ISO 22262-3, “Quantitative determination of asbestos by X-ray diffraction method” in 2016.

The purpose of ISO 22262 is to determine the asbestos content of a bulk material with sufficient accuracy to compare analytical results with various definitions of a regulated asbestos-containing material (ACM) used by different jurisdictions. These definitions are “any asbestos,” “greater than 0.1% asbestos,” “greater than 0.5% asbestos,” and “greater than 1% asbestos.” It was recognized that the level of analytical effort required to achieve this objective is variable depending on the regulatory definition, the nature of the material, and the asbestos concentration.

Many commercial building materials, particularly materials, such as sprayed fireproofing, pipe insulation, and asbestos-cement products, contain either a significant proportion of asbestos or no asbestos at all. For these materials, a polarized light microscopy (PLM) examination of the untreated material can often allow confident classification of the asbestos concentration with respect to its regulatory status with only a few minutes of observation. Other types of material present difficulties in both identification and quantification of asbestos. Transmission electron microscopy (TEM) or scanning electron microscopy (SEM) are specified in ISO 22262 as alternative methods for identifying asbestos, when results from PLM examination are ambiguous. When the asbestos concentration is estimated by PLM examination to be lower than approximately 5%, ISO 22262 specifies the use of gravimetric matrix reduction to improve the precision. ISO 22262 provides a tabulation of different materials, with recommended analytical procedures for each type.

### **Examples of Expert vs. Expert Civil Litigation Errors in Dealing with the Purported Asbestos Content of Talc**

Mickey Gunter — Geological Sciences, University of Idaho, Moscow  
Matthew S. Sanchez — RJ Lee Group

Over the past few years, we have served as expert witnesses or as consultants for the defense, in several talc-related civil cases, mainly

dealing with purported asbestos content of talc; our backgrounds are in geology, and especially mineralogy. As such, we have reviewed plaintiff expert reports as well as deposition and trial testimony. We have noted errors that often go uncontested, based on how the litigation evolves. In this presentation (and with follow-up publications) we will show examples taken directly from plaintiff expert reports and testimony that are factually incorrect and can easily be shown to be so based on established geological and mineralogical principals, as well as the peer-reviewed literature. Examples will include 1) incorrect location of deposits, 2) incorrect citations of peer-reviewed literature, 3) misuse of basic geological and mineralogical terminology, and most importantly, 4) misidentification of minerals. To be clear, we will make no challenges to expert opinions, just point out factual errors. As noted above, we have both served as paid expert witnesses for the defense, but they are providing no funds for the preparation of this abstract.

### **Look-Alikes Type 1: Objects Within an Order of Magnitude of Size**

Andrew A. Havics — pH2, LLC

Harry Alden — Alden Identification Service

Microscopists often encounter objects with the microscope that look like other objects. These look-alikes, or microscopic doppelgangers, come in a few types, the first being objects of similar size within an order of magnitude. Some of these look-alikes may be similar objects within a group or something completely outside that grouping. A few examples are fibers that have similar morphology or optical properties of asbestos, fungal spores with similar shape, similar amoebae, similar wood samples, other particles that look like fungal spores, spherical objects of various kinds, crystals with similar growth habits (snow and camphor), tobermite in cement and leaf surfaces, natural and synthetic fibers, clays and ferrous oxides, gunshot residue (GSR) and pyrotechnics, polymer whiskers, and fungal hyphae, etc.

## **Microscopy in Polymer Product Development**

John R. Reffner<sup>1</sup>, Jim Bohling<sup>2</sup>, Paul Brigandi<sup>3</sup>, Casey Wolf<sup>1</sup>, and Jeffrey Cogen<sup>3</sup> — <sup>1</sup>Analytical Sciences, The Dow Chemical Company; <sup>2</sup>Dow Coatings, The Dow Chemical Company; <sup>3</sup>Elastomers, Electrical & Telecommunications, The Dow Chemical Company

This talk will cover two applications of microscopy in industrial research. We have used Cryo-TEM and Cryo-SEM extensively in the development of advanced latex binders for architectural coatings. These binders are typically soft at room temperature, and Cryo-SEM and Cryo-TEM provide unique insights into the structure of the latex and latex-pigment composites. In addition, we have used scanning electron microscopy (SEM), energy-dispersive X-ray spectroscopy (EDS), transmission electron microscopy (TEM), and polarized light microscopy (PLM) for fundamental studies of semi-conductive polymer blends. In the system investigated, control of the morphology and dispersion of the conductive filler (carbon black) was used to produce semi-conductive blends at very low loadings (~1%).

## **Thermal Recombination of Clay Mineral Components of Ancient Mayan Ceramic Ware as a Means of Differentiating Indigenous Versus Tradeware Using X-ray Diffraction Analysis**

Wayne C. Isphording — Tulane University, Department of Continuing Education

Archaeologists have long been aware that valuable information on material exchanges among ancient civilizations can be obtained by identifying the mineral constituents in fragments of ceramic ware. In many cases, the minerals present are useful in determining whether a particular fragment is indigenous to the area where found or whether it represents tradeware from other locations. Even where inscribed design work allows positive source identification, the mineral content can still be used to further confirm this fact and indicate the extent of trade that existed at various ceremonial sites or population centers.

Ceramic materials used by ancient artisans for figurines, urns, pottery, etc. were classically manufactured by firing a mixture of clay and crushed fragments, known as “temper,” to temperatures of

300° C or higher in kilns. Clay components, lacking the addition of temper, were known to shrink and crack during drying or firing, rendering the products unusable. To avoid this, various forms of temper (crushed rock, charcoal, wood ash, etc.) were added to the clay to provide stability and strength. While most temper materials can be identified by polarized light microscopy, clays (which typically comprise the bulk of the objects) lose their identity once they are fired and can no longer be identified. This is unfortunate because the clay mineral component is often distinctive for specific geographic locations. However, a procedure does exist that can “recover” the identity of the original clay. This is done by firing pulverized shards (fragments) of the ceramic ware to temperatures in excess of 1100° C and then quickly quenching the sample. Heating the crushed ceramic powders at high temperature for at least 2 hours allows time for the constituent ions present to recombine and form new, high temperature phases (mullite, enstatite, spinel, cristobalite, etc.). These new phases can be confirmed by X-ray diffraction (XRD) analysis and serve as “fingerprints” for the original clay mineral. Using information from this procedure, it was quickly possible to differentiate indigenous pottery and figurine fragments found in the Guatemalan highlands and further south in Honduras from tradeware manufactured elsewhere in the northern Yucatan Peninsula.

## **What Is This? The Peculiar Tale of a Food Contaminant**

Jason C. Beckert — Microtrace, LLC

The vast majority of the food contaminants identified at our laboratory can be associated with the product itself, e.g., a clump of ingredients, charred product, etc.; related to the wear of processing equipment, e.g., fine metal particles, rubber, etc.; or they are recognizable objects that are relatively ubiquitous in everyday life, e.g., a staple, an insect, etc. Although each sample is unique, most contaminants can be classified into one of several “usual suspect” categories that are repeatedly encountered. This presentation will discuss a truly unique food contaminant submitted to our laboratory for identification. First impressions were deceiving, and consultation with our client indicated that the sample was even more unusual than

we previously thought. The contaminant was found to be composed of numerous components, and it was clearly assembled by human hands. The rationale behind its construction remains a mystery, and the presenter hopes that the audience will be able to suggest a satisfying answer to this enigmatic case.

### **Microbe Power in a Prehistoric Pizza**

Brian J. Ford — Caius College, University of Cambridge, U.K.

Harnessing the energies of microbes as a means of survival was one of the earliest achievements of prehistoric humans. Circular flat-breads have been produced since ancient times, and the fermented ingredients of the modern pizza are examples of how the traditions of the Stone Age remain with us today. Much of the food we eat is processed, though the role of bacteria and mold in food production is widely ignored. Today, we will review some surprising examples.

### **A Microscopical Trip Down Memory Lane: 40+ Years of McCrone Christmas Card Photomicrography**

Sebastian B. Sparenga — McCrone Research Institute

McCrone Research Institute recently assembled a collection of all the known Christmas cards that were produced for the various McCrone enterprises over the past several decades. It was such an impressive display that it needs to be shared. This talk will review selected Christmas card photomicrographs and provide insight on how some of these magnificent images were produced.

*Wednesday, June 14*

## **Chemical and Forensic Microscopy**

### **Torture Test: Microscopic Changes in Markings Made by a Tavor Rifle**

Peter Diaczuk — Pennsylvania State University

Andrew J. Winter — Centenary University, NJ

The authors were given the opportunity to perform a “torture test” on a new rifle designed by Israeli Weapon Industries (IWI) called the Tavor. We fired just over 2,000 rounds of ammunition through the test rifle in a variety of weather conditions. Comparison microscopy of collected samples of cases and bullets was performed at 200-round intervals to determine whether microscopic changes occurred over time, influencing the ability to determine common origin.

### **Investigation of Malaysia Airlines Flight MH17**

Peter Zoon and Erwin Vermeij — Netherlands Forensic Institute

In the afternoon of July 17, 2014, Malaysia Airlines flight MH17 from Amsterdam to Kuala Lumpur crashed near Hrabove in eastern Ukraine. All 298 passengers and crew were killed. Initial reports hint at a non-accidental cause of the crash. At the time of the crash, an armed conflict between Ukrainian military and armed pro-Russia forces was taking place, which prevented the possibility of investigating at the crash site. Human remains and wreckage parts were eventually returned to the Netherlands.

On July 23, 2014, the disaster victim identification (DVI) process started at the Cpl. Van Oudheusden barracks in Hilversum. A forensic triage was set up within the DVI process to obtain forensic evidence. Mobile CT scanning, X-ray scanning, and handheld X-ray scanning were used in this triage to identify fragments from the wreckage. In December 2014, wreckage parts arrived in the Netherlands and a similar forensic triage was established.

Scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM-EDS) analysis of the fragments in com-



bination with a focused ion beam (FIB) setup to create in situ surface cross-sections were successful in identifying the origin of the fragments. Laser ablation ICPMS analysis yielded quantitative elemental compositions of the recovered fragments and the relationship between the fragments from the human remains and the wreckage parts.

### **Microcrystal Tests for the Detection of Butylone, Methylone, and Ethylone**

Shan Mei Jones — University of Illinois at Chicago Graduate Program, in association with McCrone Research Institute

Synthetic cathinones, commonly known as “bath salts,” have become more abundant on the U.S. drug market. These drugs are classified as stimulants and, therefore, have similar pharmacological effects as amphetamines. While butylone and methylone are both Schedule I drugs in the U.S., ethylone has yet to be scheduled. Microcrystal tests for these three drugs were researched and developed because it is difficult to identify synthetic cathinones by the usual forensic methods. Two common microcrystal test reagents — picric acid and picrolonic acid — form unique rosettes identifiable with each drug. Butylone and methylone form rosettes with picrolonic acid, while ethylone and methylone form rosettes with picric acid. All crystals were then analyzed using polarized light microscopy (PLM) and attenuated total reflectance (ATR) infrared microspectroscopy.

### **The Effect of Ultraviolet Radiation on the Microspectrophotometry (MSP) of Dyed Fibers — Phase 1**

Meggan B. King — McCrone Research Institute

This talk will discuss the initial applied research project design to study changes in dyed man-made fibers that result as an effect of environmental conditions, especially exposure to natural and artificial ultraviolet (UV) radiation over long periods of time. The McCrone Research Institute in Chicago has received funding from the National Institute of Justice (NIJ) under the FY 2016 Research and Development in Forensic Science for Criminal Justice Purposes

(Award No. NIJ-2016-DN-0145) to provide a practical application of microspectrophotometry (MSP) as a tool for understanding how ultraviolet radiation can affect the color and dyes of fiber evidence and improve the discrimination, identification, and individualization of man-made polymer fiber products for the forensic scientist.

### **A Forensic Study of Known Toner Nanoparticles**

Katie M. White and Christopher S. Palenik — Microtrace, LLC

Whether we are aware of them or not, small particles abound in the environments that surround us. Small particles may be engineered for use in manufactured products, be present in dusts generated from man-made industrial processes, or occur naturally in the environment. Some of these particles are just barely visible, while others are so small that they cannot be resolved by the human eye. These subvisible and submicrometer particles (nanoparticles) offer potential as forensic evidence, but they are presently unexploited due to the challenges that their small size present.

One example of subvisible particles is the toner powder used in laser printers and copiers. Presently, most existing research on forensic toner analysis focuses on document examination, i.e., analysis of printed toner, rather than on trace evidence. However, toner is widely used, and these small particles are easily transferred and rarely noticed. Identification of trace amounts of toner, e.g., on hands or clothing or in dust, could be used to provide investigative leads or associate them with a scene and/or victim, particularly if the particles are suggestive of a specific toner.

This presentation will discuss the results from an analytical study of more than 50 different toner samples. This research evaluates microscopic morphologies observed by light microscopy and scanning electron microscopy, and chemical properties determined by Raman spectroscopy, of the known toner samples, providing methods that can be used in the forensic laboratory to identify and classify toner particles. Analytical differences observed within the sample set, the prevalence of background toner particles in different environments, and limitations of this approach will be covered.

## **Some Lesser Known Microchemists and a Look at Some of Their Work**

Skip J. Palenik — Microtrace, LLC

Most delegates at Inter/Micro are familiar with the names Behrens, Emich, Chamot, Feigl, Benedetti-Pichler, and Cheronis, as well as their contributions to classical microchemical analysis. This presenter became acquainted with them from an early age, when he began his practical studies of microchemistry. Over the years, growing familiarity with the publications of these authors widened to include the acquisition of many of the books and articles that they recommended in their works. These new references were gradually acquired and studied, as were the works of new generations of microchemists who followed these pioneers. By the time I began my career with Dr. McCrone, the amount of research reported in this field had diminished and current work in it was published primarily in journals such as the *Microchemical Journal* and *Mikrochemica Acta*. The articles published in these journals, their references, and book reviews provided leads to new researchers and their works. This talk will present some of the results of these researches into the work of other notable microchemists whom this author believes should be brought to the attention of those still employing these useful and delicate procedures.

## **Possible Degradation Mechanisms of Antemortem Hair Roots Containing Induced PMRB-Like Features**

Barbara L. Fallon, M.S.<sup>1</sup>; Jack Hietpas, Ph.D.<sup>1,2</sup>; and JoAnn Buscaglia, Ph.D.<sup>1</sup> — <sup>1</sup>Federal Bureau of Investigation Laboratory, Counterterrorism and Forensic Science Research Unit; <sup>2</sup>Pennsylvania State University Department of Biochemistry and Molecular Biology

A postmortem root band (PMRB) is a microscopic feature resulting from degradation to the pre-keratinized region of the roots of anagen hairs obtained from cadavers. A PMRB forms due to degradation of the intermacrofibrillar matrix (IMM) (or cell membrane structures) in the cortex resulting in elongated, gas-filled voids<sup>3</sup>. Hypothesized mechanisms for in vitro band formation include am-

monium ions or evolved ammonia gas that may chemically attack the IMM; a change in hair pH that may lead to IMM degradation or collapse; or decomposition gases that may become trapped inside the cortex, thus mechanically disrupting the integrity of the root.

This work investigated these possible mechanisms of the IMM damage in the formation of PMRBs. PMRB-like bands were induced in intact hairs submerged in water, ammonium acetate, and pH 7.8 buffer. Microtomed hair slices exposed to the same solutions displayed mixed results. When observed, ultrastructural features of the induced PMRB-like bands were consistent with true PMRBs. These results may suggest a combined mechanism whereby chemical attack weakens the IMM and facilitates mechanical damage to form PMRBs.

<sup>3</sup>Hietpas, J.; Buscaglia, J.; Richard, A.H.; Shaw, S.; Castillo, H.; and Donfack, J. "Microscopical Characterization of Known Postmortem Root Bands Using Light and Scanning Electron Microscopy," *Forensic Science International*, 267, pp 7–15, 2016.

## **The Trotter Collection: Microscopical Analysis of Human Scalp Hairs for 21st Century Research Questions**

Sandra Koch, Nina G. Jablonski, and Mark D. Shriver — Pennsylvania State University, Department of Anthropology

Mildred Trotter, an anatomist and physical anthropologist at Washington University in St. Louis, conducted much of the early quantitative research on human hair form from the late 1920s to the mid-1950s. Her work greatly influenced our understanding of variation in the cross-sectional shape and diameter of scalp hair in a wide variety of human populations. Hair samples from her original studies, housed at the Smithsonian Institution, were used in this study. Sampling from this collection was undertaken in an effort to update her analyses with high-resolution microscopy and modern image-analysis software. The use of these techniques makes possible more detailed descriptions of morphological diversity in scalp hair forms within and among populations and the addition of novel discriminating morphological characteristics, which can inform human hair research.

For this project, hairs were sampled from 27 population groups

represented in the collection. The hair samples were sectioned and then imaged using oil immersion light microscopy. Maximum and minimum diameters were measured to calculate the degree of ellipticity and for comparison with the hair index and area calculations recorded in Trotter's notebooks and publications. Further analysis was conducted using the particle analysis function within the ImageJ image-processing program to assess the number, distribution, and relative density of melanosomes present within a hair cross section. The goal of this research work is to explore correlations between patterns of microscopical and genetic variation, and provide quantitative data for further research.

## **Forensic Drug Identification by GC/MS and PLM**

Andrew M. Bowen — U.S. Postal Inspection Service

Gas chromatography/mass spectrometry (GC/MS) is often called the workhorse of the forensic drug laboratory. It is well suited for this purpose, providing two independent analytical results: retention time and mass fragmentation pattern. Like all instruments, GC/MS has limitations. Of primary concern for drug chemists are its inability to distinguish between some structural isomers and its inability to determine the salt form of identified compounds. Some laboratories accept these limitations and communicate any uncertainties in their reporting language. Other laboratories use additional instrumentation, commonly Fourier-transform infrared spectroscopy (FT-IR), to pursue more definitive identifications. The primary limitation of FT-IR is that spectra of mixtures can be challenging to interpret. Chemical extractions can sometimes separate the drug of interest from other compounds present, but this is not always possible. Polarized light microscopy (PLM) is capable of distinguishing different salt forms of compounds as well as different positional isomers, even when the controlled substance is a minor component in a mixture. Analysis by PLM takes only minutes and requires a minimal sample. This presentation will share examples from casework and training that illustrate how GC/MS results can be supplemented by PLM to provide additional specificity to the identification of controlled substances.

## **Imaginary Microorganisms and Impossible Worlds**

Brian J. Ford — Caius College, University of Cambridge, U.K.

Reference books contain unrecognizable portrayals of microscopic organisms, and many earlier encyclopedias pay little attention to the accuracy of their accounts. How does this relate to the medieval imaginings of philosophers? And is modern science always better than the scribes from 500 years ago? Just as unicorns were ridiculous inventions, some present-day images are unreal, and they reveal how little science has progressed.

## **Using SEM-EDS for Quantitative Forensic Glass Comparisons: Some Things to Think About**

Thomas A. Kubic — John Jay College and The Graduate Center, CUNY; Thomas A. Kubic and Associates

Alex Comanescu and Tiffany Millet — The Graduate Center, CUNY

Energy-dispersive X-ray spectroscopy (EDS), especially employing a scanning electron microscope (SEM) for an excitation source, is a core elemental characterization technique for micro and ultra-micro size samples. This technique has long been used for adding evidential value to the microscopical forensic examination of transfer evidence. Glass chips are one specific class of transfer evidence for which this technique has been employed. In this talk, we will present the results of follow-up work on 46 glass samples for which visible microspectrophotometric results were previously reported.

As a strategy for comparing glass samples by SEM-EDS, criminalists often normalize the data and compare ratios of peak heights or areas to one common major element such as silicon. Courts today often require data about the frequency of occurrence of similar spectra to assist the trier of facts about the weight of this comparative evidence. One of the goals of this project was to provide insight into this matter.

Being mindful of a caveat by Dr. Peter Zoon of the Netherlands Forensic Institute to be careful about how we generate and use data obtained with computerized black-box software, we evaluated the quantitative data generated from the blue glass samples and a pair of

standards from the National Bureau of Standards (now National Institute of Standards and Technology [NIST]), with commercial software utilizing both their standardless and standard algorithms. In addition, we processed the raw data using NIST's Desktop Spectrum Analyzer II (DTSA II), a software tool for quantifying EDS spectra. We will present the results of these investigations.

## **Morphology and Microanalysis of Aluminum Powders**

JenaMarie Baldaino<sup>1</sup>, Jack Hietpas<sup>1,2</sup>, and JoAnn Buscaglia<sup>3</sup> — <sup>1</sup>FBI Laboratory, Counterterrorism and Forensic Science Research Unit, Visiting Scientist Program; <sup>2</sup>Pennsylvania State Forensic Science Program; <sup>3</sup>FBI Laboratory, Counterterrorism and Forensic Science Research Unit

Online sharing of YouTube videos and instructional manuals on how to construct improvised explosive devices (IEDs) inform amateur bomb makers of the easily accessible household materials that can be used to make aluminum (Al) powder. Previously, we discussed Al powders produced from ball-milling Al foil and extracted from spray paints containing Al flake. The results obtained from scanning electron microscope photomicrographs demonstrate that Al powder manufactured by ball milling could be confidently differentiated from those extracted from an Al flake-containing spray paint. Furthermore, scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM-EDS) analysis of the spray paints containing Al flake provided additional information that could differentiate between brands and among products within brands.

Additional methods to either obtain or extract Al powder include melting Al cans to form an Al ingot, then using a steel file to produce Al powder; grinding Al foil with a coffee grinder; binary exploding targets; and extracting Al powder from pyrotechnics such as sparklers and firecrackers. This presentation will discuss the differences in Al particle surface characteristics and elemental compositions of these additional sources using SEM-EDS.

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