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George R. Harrison Spectroscopy Laboratory

Massachusetts Institute of Technology

Research Report

Quantitative phase microscopy and spring constants of cholesterol helical ribbons

B. Khaykovich, N. Kozlova, W. Choi, C. Hossain, A. Lomakin, Y. Sung, R. R. Dasari, M. S. Feld and G. B. Benedek

Self-assembly of helical ribbons in complex fluids is an interesting phenomenon that poses fundamental questions about the molecular structure, elastic properties and kinetic evolution of these objects. In particular, solutions that contain cholesterol, non-ionic surfactants and lipids spontaneously form helical ribbons with characteristic pitch angles of 11 and 54° (see Fig. 1). It appears that the range of spring constants of our helices is such that they are suitable for measuring forces acting between nano-scale biological objects such as antigen-antibody and enzyme-substrate interactions. In order to measure the forces, we need to know the spring constant of individual ribbons. We believe that the spring constants can be determined entirely by measuring the external dimensions of the helices. The dependence of the diameter of the ribbons on their thickness provides a crucial link

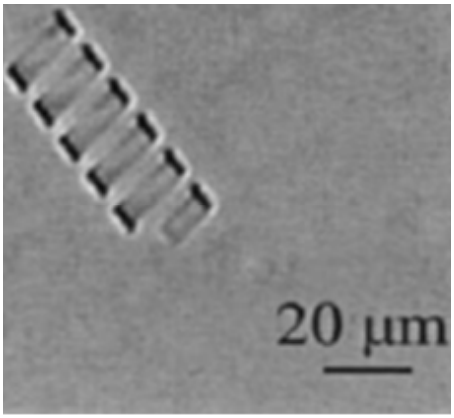


Fig. 1. Phase contrast image of a helical ribbon.

Quantitatve phase *cont. on p. 2*

OPPORTUNITY for Independent diagnostic spectroscopy projects

Kamran Badizadegan

The MIT Laser Biomedical Research Center (LBRC) announces the availability of spectroscopic instruments for projects of outside investigators aimed at developing novel diagnostic applications and demonstrating their feasibility in clinical translational studies. Two instruments will be made available for this purpose: (1) a "mini-FastEEM", capable of collecting diffuse white light spectral reflectance and 340 nm-excited fluorescence; and (2) a Raman spectroscopy instrument capable of collecting Raman spectra from biological tissues over the fingerprint range (400-1960 cm^{-1}). Both instruments collect spectra in ~1 s by means of specially designed optical fiber probes that sample 1 mm^2 regions of tissue. Specially designed display and spectral analysis software are built in. These instruments are fully described at http://web.mit.edu/spectroscopy/research/biomedicaloptics.html.

Purpose and Scope of the Program

The purposes of the LBRC outside user diagnostic instrumentation program are:

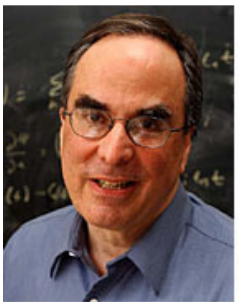
(1) to foster clinical translational research in optical spectroscopy; and

(2) to familiarize young/new investigators with the methods of diagnostic spectroscopy.

The program particularly favors focused clinical projects with well-defined endpoints. Potential investigators with medical credentials are specially encouraged to participate. The LBRC will provide instruments, optical fiber probes, training, instrumentation support and troubleshooting, and support for data analysis as necessary. The research may be performed at the applicant's institution, at MIT, or a combination of the two.

OPPORTUNITY, *cont. on p. 6*

APS Schawlow Prize to Robert Field



Robert Field, the Haslam and Dewey Professor of Chemistry and the Harrison Spectroscopy Laboratory's Associate Director for Scientific Coordination, has been chosen to receive the 2009 Arthur L. Schawlow Prize in Laser Science. This prize, endowed by the NEC Corporation and awarded by the American Physical Society, recognizes Field's "pioneering development and application of multiple resonance laser spectroscopy and effective Hamiltonian models that reveal fundamental mechanisms of chemical bond breaking, electronic rearrangement, intramolecular vibrational redistribution, and unimolecular isomerization."

Presentation of the award will be in October 2009 in San Jose, CA at the APS Division of Laser Science/Optical Society of America Annual meeting.

Also in this issue

Quantitative phase, *cont. from p. 1*

between the elasticity and the external dimensions of the helices.

The thickness of the ribbons is less than 200 nm, which is below the diffraction-limit axial resolution of optical microscopes. However, the tomographic phase microscope recently developed at the MIT Spectroscopy Laboratory [2] is capable of determining the thickness with nanometer accuracy. The microscope uses interference between a sample beam and a reference beam to measure two-dimensional distributions of phase delays induced by a specimen in the sample beam. Since the microscope requires a coherent light source, the phase images show fixed pattern background noise due to diffraction from dust particles in the beam path and from the specimen itself, see Fig. 2(a). We can remove the effects of diffraction by properly synthesizing phase images taken at different angles of illumination. Only the images of the specimen in the focal plane constructively interfere among the phase images taken at different angles. As shown in Fig. 2(b), the part of the ribbon in the focal plane is now clearly visible with high signal-to-noise ratio.

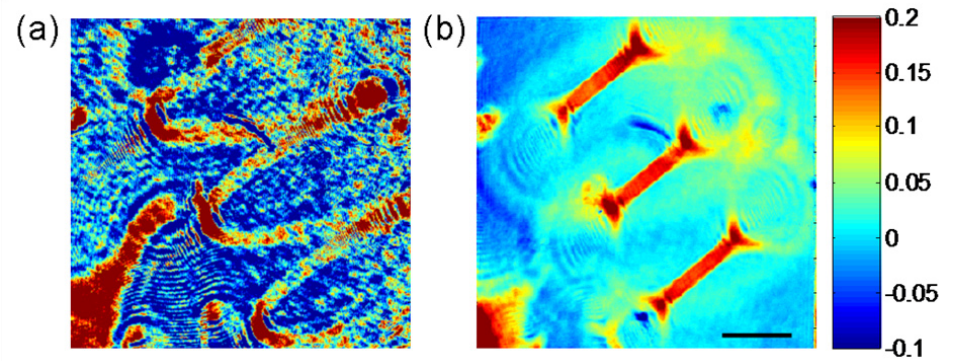


Fig. 2. Quantitative phase images of a helical ribbon (a) taken at zero degrees illumination (b) after synthesizing phase images taken at different angles. Scale bar, 20 microns. Color bar indicates phase in radians at 633 nm.

The difference in phase across the field of view is due to the difference of refractive index between the ribbon and the medium. The thickness t is deduced from the phase by means of the expression $t = (\Delta\phi/2\pi)(\lambda/\Delta n)$, where $\Delta\phi$ is the phase difference between the ribbon and the background, $\lambda = 633$ nm is the wavelength of light, and Δn is the difference in refractive index between the ribbon and the medium. For the ribbon of Fig. 2(b), the thickness is approximately 112 nm while the diameter of the ribbon is about 75 μm (as determined by conventional phase-contrast microscopy). We have measured helical ribbons with diameters ranging from 15 to 140 μm. First results are consistent with the expected diameter-vs.-thickness relation [1].

These dramatic results demonstrate the power of tomographic phase microscopy in measuring biological structures well below the optical diffraction region, and illustrate its application to the problem of self assembly of helical cholesterol ribbons.

References

[1] B Smith, YV Zastavker, GB Benedek. Tension-induced straightening transition of self-assembled helical ribbons, Phys. Rev. Lett. **87** (2001) 278101.

[2] W Choi, C Fang-Yen, K Badizadegan, S Oh, N Lue, RR Dasari, et al. Tomographic phase microscopy, Nature Methods. **4** (2007) 717-719.

THE SPECTROGRAPH

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The Spectroscopy Laboratory houses two laser research resource facilities. The MIT Laser Research Facility provides shared facilities for core researchers to carry out basic laser research in the physical sciences. The MIT Laser Biomedical Research Center, a National Institutes of Health Biomedical Research Technology Center, is a resource center for laser biomedical studies. The LBRC supports core and collaborative research in technological research and development. In addition, it provides advanced laser instrumentation, along with technical and scientific support, free of charge to university, industrial, and medical researchers for publishable research projects. Call or write for further information or to receive our mailings.

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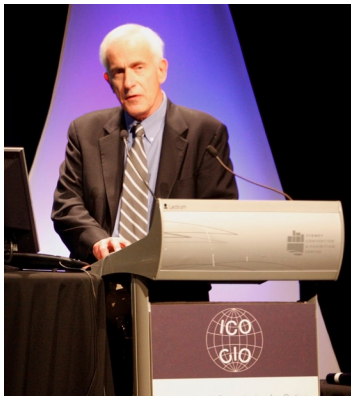
OSA Meggers Award to Michael Feld



Michael Feld delivering his plenary lecture on the occasion of receiving the OSA's William F. Meggers Award in Sidney, Australia, July 7, 2008

At ICO-21, the International Commission for Optics Congress, held last July in Sidney, Australia, Michael Feld, Director of the Harrison Spectroscopy Laboratory and MIT Professor of Physics, received the William F. Meggers Award of the Optical Society of America. This award honors Feld for his "major contributions to the foundations of laser spectroscopy, and for pioneering developments in the application of spectroscopy to biomedicine."

> ... a wonderful tool for exploring the frontiers of physics, chemistry, biology, and medicine.

Feld's plenary lecture, "Spectral interferometry in biology and medicine: Past, present and future," reviewed the recent successes of his group in developing tomographic phase microscopy to create refractive index maps of cells, with application to studying dynamical processes such as the development of malaria (see MIT Tech Talk, September 10, 2008, p. 5).

"For me," Feld noted, "the Meggers Award has special meaning, because its first recipient, George R. Harrison, founded the laboratory that I have directed since 1975. Spectroscopy is a wonderful tool for exploring the frontiers of physics, chemistry, biology, and medicine. It will continue to be a major tool for a wide range of important endeavors, from improving human health to understanding the secrets of the universe."

Research Report

Light scattering study of single live cells

Wonshik Choi, Chung-Chieh Yu, Christopher Fang-Yen, Kamran Badizadegan, Ramachandra R. Dasari, and Michael S. Feld

Light scattering has been widely used to develop new optical diagnostic tools. A commonly employed strategy is to measure the scattered light from tissue as a function of angle and infer tissue properties from the measurements. The virtue of this approach is its ability to identify pre-cancerous ("dysplastic") regions of tissue, often invisible, without the need for high resolution 3D imaging. Because a wide area of tissue can be sampled in a short time, the technique is highly suitable for clinical applications. A case in point is colposcopy, a widely used technique in which a low power microscope scans the cervix to search for dysplasia. In this technique a 3-5% acetic acid solution is applied to the cervix. Dysplastic sites tend to scatter more light than do normal regions of tissue, and so suspect regions of tissue exhibit whitening, called "aceto-whitening." Hence, the increase in light scattering due to application of acetic acid is an indication of dysplasia and can serve to guide biopsy.

However, to obtain an objective diagnosis from light scattering it is important to understand how the cellular structures contribute to the light scattering distribution. One can then develop proper logistics to quantify the morphological structures of the tissue to be used for diagnosis. To study the specific connection between the cellular structures and the light scattering distribution, we need to measure both the 3D maps of refractive index and the light scattering angular distributions from single live cells. Conventional light scattering techniques have limitations in performing these tasks for two reasons. First, the sensitivity is not high enough to record the angular scattering from individual cells. Light scattering from hundreds of thousands of cells must be sampled and averaged; as a result only a statistical comparison is available. Second, up to now there has been no experimental measurement of the 3D refractive index map of live cells. Thus, the analysis has had to rely on numerical modeling of overly simplistic pictures of cells. For example, a cell is often depicted as a spherical nucleus surrounded by a larger spherical region of cytoplasm. This rather artificial cell model may limit the validity of connecting light scattering to cellular structures.

Recently, we have developed an electric field (E-field) based technique that can simultaneously measure angle-resolved light scattering and the refractive index distribution of a single cell (W. Choi et al, Optics Letters 33 1596 (2008)). By exploring the connection between the refractive index structure and the light scattering spectrum, we should be able to elucidate the origin of aceto-whitening at the single cell level.

The following is a summary of our experimental studies. We first used tomographic phase microscopy (TPM), developed by our group to map the refractive index of the cellular sub-structure of live cells (W. Choi et al, Nature Methods 4 717 (2007)). We recorded refractive index tomograms of a HeLa cell in a normal culture medium (Fig. 1a), in a culture medium containing 0.5 % acetic acid (Fig. 1b), and in a normal culture medium after the cell had been subjected to 0.5% acetic acid (Fig. 1c). To connect the structural changes of the cells with the angular scattering, we calculated the forward-directed angular scattering (Fig. 1e) of the HeLa cells from their refractive index tomograms (Figs. 1a-c) using the Born approximation (Fig. 1e). As can be seen in Fig. 1e, the scattering intensity at large angles increased after adding the acetic acid, especially at angles larger than 10 degrees. The Born approximation relates the Fourier transform of the refractive index map to the angular scattering distribution. In this way we connect the spatial frequency of the refractive index structure to the scattering angle. High spatial frequency structure will contribute to large scattering angles, low spatial frequency structure to small angles. The scattering angle of 10 degrees corresponds to the spatial frequency of 1/3 μm^{-1}. Therefore, our analysis verifies that the increase in structures finer than 3μm is responsible for the increase in scattering at large angles by addition of acetic acid.

To support this reasoning, we need to demonstrate by experiment the validity of the Born approximation. To compare with angular scattering distributions from the refractive index tomograms, we measured angular scattering of the same single HeLa cell in Fig. 1a-c under the three different conditions. We measured both the phase and the amplitude of the scattered light at the image plane, constructed the E-field, and took the Fourier transform to obtain the light scattering distribution (Fig. 1d). The results show a close match with the angular scattering calculations from the experimentally measured index map (Fig.1e), which validates use of the Born approximation for calculating angular scattering from the refractive index tomograms.

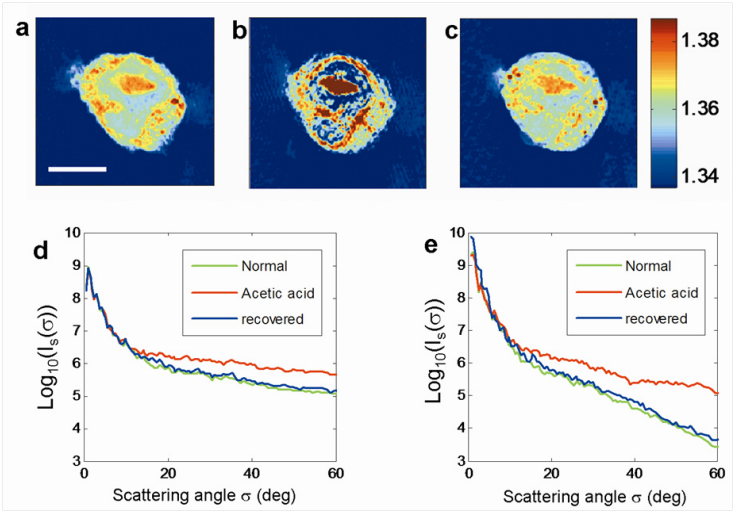


Fig. 1. Angular scattering distributions from a live HeLa cell. Tomographic sections of refractive index tomograms in normal culture medium (a), medium containing 0.5 % acetic acid (b), and in normal culture medium after having been subjected to 0.5% acetic acid (c). (d) Angular scattering distributions measured from the field image under the above three conditions. (e) Angular scattering distributions calculated from the refractive index tomograms (a-c) obtained using TPM. The scale bar in (a) represents 10 μm.

Now that we have demonstrated the capability of our instrument to quantitatively connect the light scattering spectrum with the 3D refractive index map, we will be able to elucidate contributions to the scattering from subcellular structures/organelles. This, in turn, will make it possible to rigorously validate the numerical modeling used in conventional light scattering studies.

Lester Wolfe Workshop in Laser Biomedicine

Molecular tools in diagnostic medicine: The opportunity for biomedical optics

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Recent ground-breaking progress in cell and molecular biology has resulted in tremendous advances in understanding the molecular underpinnings of human diseases. These discoveries have given rise to the new discipline of molecular diagnostics that continues to revolutionize the practice of diagnostic pathology and in vivo imaging. In parallel, the development of targeted therapies against specific molecular markers has generated additional demands for molecular-specific characterization of cells and tissues. This workshop will introduce the participants to the current challenges and opportunities in molecular diagnostics, with special emphasis on the contribution of optical and spectroscopic methods.

State-of-the-art in molecular diagnostics
John Iafrate, Massachusetts General Hospital/ Harvard Medical School

Flip-flopping contrast mechanisms in optical imaging
Samuel Achilefu, Washington University School of Medicine

Molecular pathology with mid-infrared chemical imaging
Rohit Bhargava, Beckman Institute at the University of Illinois

Imaging of molecular assemblies for cancer detection, monitoring and therapy: A plasmonic approach
Konstantin Sokolov, University of Texas at Austin

Tuesday, November 25, 2008, 3:30-6:00 PM
Massachusetts Institute of Technology
Grier Room, 34-401
77 Massachusetts Avenue, Cambridge

Refreshments served at 3:00 PM

Sponsored by the G. R. Harrison Spectroscopy Laboratory, MIT, MGH Wellman Laboratories, the Harvard-MIT Division of Health Sciences and Technology, and the Center for the Integration of Medicine and Innovative Technology (CIMIT)

Seminar on

Modern Optics and Spectroscopy

Fall Semester 2008

September 16	Jelena Mirkovic, MIT **Detecting cervical pre-cancer with "fluorectance" spectroscopy**
October 7	Bruce Weisman, Rice University **Fluorescence spectra of single-walled carbon nanotubes: From discovery to application**
October 14	***2nd Annual Dasari Lecture*** Takeshi Oka, University of Chicago **H_3^+ in the central molecular zone of the Galactic center**
October 21	M. Fatih Yanik, MIT **On-chip femtosecond neurosurgery**
November 4	Allard Mosk, University of Twente, The Netherlands **Opaque lenses: Using disorder to bring laser light to a focus**
November 18	Phillipe Guyot-Sionnest, University of Chicago **Slow electron cooling and spin blockage in colloidal quantum dots**
November 25	Vladimir Petrovic, MIT **Toward pure electronic spectroscopy**
December 2	Marko Loncar, Harvard University **Light-matter interaction in nanophotonic devices**
December 9	Daniel Mittleman, Rice University **Terahertz near-field imaging and spectroscopy**

Tuesdays, 12:00 - 1:00 p.m., Grier Room (34-401)

Refreshments served following the seminar.

Sponsored by the George R. Harrison Spectroscopy Laboratory, Department of Electrical Engineering and Computer Science, and School of Science, MIT

Honors for Millie Dresselhaus

Millie Dresselhaus, active core researcher in the Spectroscopy Laboratory, EECS Professor Emeritus, and Institute Professor, received her PhD from the University of Chicago in 1958. In June that university

Here at a dinner in her honor in the Rotunda of the National Academy of Sciences Building, Millie Dresselhaus, retiring Chair of the AIP Board of Directors and Executive Committee, talks to her successor, geophysicist Lou Lanzerotti. On her right is Marc Brodsky, former AIP Executive Officer, and on her left is Fred Dylla, current AIP Executive Officer.

presented her their esteemed and infrequently awarded Alumnus (Alumna) Medal. Honored as "an internationally known physicist who had done groundbreaking research in condensed matter and worked tirelessly to draw women to science and engineering," she delivered the main address at the University's Alumni Convocation.

In March Millie was celebrated by the American Institute of Physics as she retired from her position as chair of the AIP Board of Directors and its Executive Committee. She is the first woman to have held this position. AIP held a gala dinner in her honor in the rotunda of the National Academy of Sciences building in Washington, DC.

OPPORTUNITY, *cont. from p. 1*

Eligibility

Both new and established investigators may apply. New investigators are specially encouraged to use this opportunity to collect preliminary data for subsequent independent funding. Funded investigators must demonstrate how the use of these instruments will represent a departure from their ongoing work.

Application Guidelines

Applications will be accepted on a rolling basis using guidelines detailed for the LBRC outside projects. Please see http://web.mit.edu/spectroscopy/facilities/guideline.html for detailed information and an application form.

Evaluation of Applications

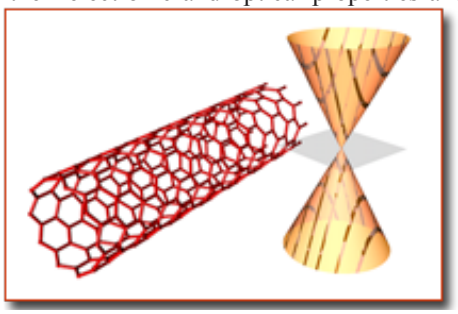
The evaluation committee will be composed of the LBRC core investigators. The committee will review applications and assign a priority rating to each based on the scientific merit and technical feasibility of the proposed research. Initial projects can have a duration of up to 6 months. Continuing applications will be considered, depending on the results of the project and related factors. Awards will be announced on December 1, and projects will start on January 1.

Responsibility of the Awardees

Awardees must submit a written progress report (2 page NIH format) within three months of the completion of their project. In addition, awardees may be invited to present their work at an LBRC Advisory Committee meeting or an NIH site visit. The LBRC must be acknowledged on all publications and presentations of the research in the form of "Supported in part by the MIT Laser Biomedical Research Center, NIH Grant P41-RR02594."

Jing Kong receives Jonathan Allen Junior Faculty Award

MIT's RLE (Research Laboratory of Electronics) has given Assistant Professor Jing Kong a 2008 Jonathan Allen Junior Faculty Award. This award, created to support young faculty, recognizes the potential of her work on controlled synthesis of carbon nanotubes and graphene, and her investigation of their electronic and optical properties and



integration with CMOS circuits. In addition to the honor of the award she receives $50,000 to support her research.

Jing received her B.S. degree in chemistry from Peking University in 1997 and her Ph. D. in chemistry from Stanford University in 2002. She joined the MIT faculty in 2004 as Assistant Professor of Electrical Engineering; she uses the facilities of the Harrison Spectroscopy Laboratory to characterize the helicity of carbon nanotubes.

The award is named for Jonathan Allen, the sixth Director of RLE, who was deeply committed to supporting junior faculty.

Puzzling questions; puzzling answers

Last issue posed two Fermi questions:

1. How many college students are there in the Boston area?

There are a lot of institutions of higher education in and around Boston, more than 10 but probably less than 100. Take the number to be 10 and then compensate for the smallness by assuming that each school has about 10^4 students enrolled. The answer to a Fermi precision is then 10^5 college students in the Boston area.

2. How many photons are there in the Universe?

This question is best answered over (several) drinks in a bar. Remember when answering a Fermi question, you are not supposed to look anything up. Here's one argument. Given the amount of empty space in the Universe, the most numerous photons are probably those associated with the 3-K cosmic background blackbody radiation. So if you multiply the volume of the Universe by the energy density of 3-K radiation and divide by the energy of a 3-K photon, you should get the answer. A random survey of one *Spectrograph* reader led to a value of 10^{87}. (Landau once said "Astrophysicists are often wrong but never uncertain.")

For the next issue

Because spectroscopy is deeply bound up with metrology and the speed of light answer the following two questions:

1. How many minutes are there in a microcentury? Compare this to class and group meeting times.

2. Everyone knows the speed of light in furlongs per fortnight. But what is it in tablespoons per barn per weekend? Send answers to mei_liu@mit.edu.

First Light, *cont. from p. 8*

coloration. It may have been a well-known procedure in its time.

He remarks on how observing colored objects by these colored lights dramatically changes their appearance. But then he goes on from this experiment in perception to something more basic:

"Having placed a paste-board with a circular hole in it between my eye and the flame of the spirits, in order to diminish and circumscribe my object, I examined the constitution of these different lights with a prism…and found that, in the first case when sal ammon[iac], alum, or potash fell into the spirits, all sorts of rays were emitted, but not in equal quantities; the yellow being vastly more copious than all the rest put together, and red more faint than the green and blue."

This is, so far as I am aware, the first time that anyone took light from a source other than the sun or an unadulterated flame, ran it through an aperture, and subsequently through a prism to break it into its individual components.

Sal ammoniac is ammonium chloride, NH_4Cl, and it lacks a metal ion that would give color to the flame (an ammonium ion would not do so). Alum is $KAl(SO_4)_2{\cdot}H_2O$, while potash is potassium carbonate, K_2CO_3. None of these would be expected to produce a copious yellow light. Potassium does have yellow lines in its spectrum, but they don't predominate – the classic potassium-colored flame is lilac in color. What Melvill was seeing was certainly the intense sodium D line, undoubtedly due to the contaminating sodium that often occurs in potassium salts. The fact that red was fainter than green or blue is consistent with the flame spectrum of potassium salts, in which green and blue are stronger than the longer wavelength lines.

"In the light of spirits mixed with nitre or sea salt, I could still observe some blue, though excessively weak and diluted; with the latter, the green was equally faint; but, with the former, pretty copious. But, when either of these salts were used, could hardly see any vestige of the red at all…

The proportion in which the bright yellow exceeds the other colours in this light, is still more extraordinary than in the former; insomuch that the hole seen through the prism appears uniformly of this yellow, and as distinctly terminated as through a plain glass."

This would again be pretty much consistent with modern observations. Nitre is potassium nitrate, KNO_3, and Melvill's sample must have had relatively much more sodium in it than his other salts did to make the red seem so dim. Sea salt, made by evaporating sea water, could be expected to have predominantly sodium ions (30%), with manganese next in prevalence (3.7%), followed by calcium (1.2%) and then potassium (1.1%). It's not surprising that green and blue should be weaker in this case, where there is much less potassium and with manganese not producing visible spectral lines, while calcium has red predominant.

Furthermore, Melvill observed that this light had a significant difference from sunlight or the light from a plain alcohol flame:

"Because the hole appears through the prism quite circular and uniform in color; the bright yellow which prevails so much over the other colours, must be of one determined degree of refrangibility; and the transition from it to the fainter colour adjoining, not gradual, but immediate."

The spectrum of sodium has a distinct and well-defined line. This would have been easier to observe if Melvill had used a narrow slit rather than a circular hole (as Wollaston would do a century and a half later, enabling him to see those narrow dark lines in the solar spectrum). Had he done so, he might even have been able to observe that the bright yellow color actually resulted from two closely-spaced lines. But it's important not to judge him by the standards of our own knowledge, gained from standing on the shoulders of giants. Melvill was groping through country utterly unknown to him or to anyone else, and his discovery that the spectrum was discrete, rather than continuous, was a major discovery that

the bright yellow ... must be of one determined degree of refrangibility

could have allowed for a conceptual leap.

Could have, but did not. Melvill's topics go in different directions after this, and within two years he was dead. If he had lived, perhaps he would have revisited this topic and pressed onward and measured the relative positions of lines, establishing the science of spectroscopy a century and a half earlier than it was founded.

But perhaps not. Claudius Ptolemy in the first century built experimental apparatus and performed careful measurements of the refraction of light through interfaces between air, glass, an water, producing results very nearly identical to modern measurements. He came close to discovering Snell's Law, but didn't quite do so. He was distracted by a different mathematical formulation and settled on an incorrect model. Neither he nor his students revisited the phenomenon. Had they done so, they might have noticed that the formula didn't work for small angles (most modern accounts of Ptolemy's work incorrectly state that it was correct at small angles), and the science of optics might have advanced much more rapidly than it did. In the long run, it doesn't matter. Several others discovered the law of refraction (even before Snell), and Wollaston, Herschel, Fox, Fraunhofer, and others eventually rediscovered the basics of spectroscopy unearthed byThomas Melvill and then forgotten.

References

Melvill's work was printed in the relatively obscure Essays and Observations, Physical and Literary, No. IV. It was, fortunately, reprinted in the Journal of the Royal Astronomical Society of Canada, **8**, 231-272 (1914); available online at http://adsabs.harvard.edu/full/1914JRASC...8..231M

Tony Rothman's article on Galois's last hours appeared in American Mathematical Monthly, **89**, 84 (1982). It is available online at http://www.physics.princeton.edu/~trothman/galois.html

On the use of beechwood ashes for coloring glass see the footnote by John G. Hawthorne and Cyril Stanley Smith on pp. 55-6 of *On Divers Arts* by Theophilus, Dover Books 1979/ U. Chicago 1963

Spectral Lines

First light – Thomas Melvill and the beginnings of spectroscopy

Stephen R. Wilk

On the evening of May 30, 1832 Évariste Galois sat up writing out mathematical formulas. He was only 20 years old, and the next morning he was to engage in a duel that he was fated to lose. Fearing that he was to die, legend tells us, he frantically tried to set down all his revolutionary theories on paper before the death he was sure awaited him, scribbling "There is not Time!" on the margin of his paper.

It's a dramatic story, but when Tony Rothman researched the story – he intended to write a play based on he incident – he found that it wasn't true. Galois did die in the duel and did set down some work on the night before, but the frantic attempt to set down the remainder of his work for posterity with woeful asides did not happen. The situation, in fact, is dramatic enough without it, and tragic enough. Galois did make contributions to group theory, abstract algebra, and other areas, and he would probably have gone on to make other contributions had his life not been cut so short. And so he goes to join the ranks of promising scientists who died too early. Such as John Goodricke, the deaf-mute astronomer who, among other things, discovered and measured several variable stars and died of pneumonia (contracted while observing stars on a dark night) at the age of twenty -one. Or Thomas Melvill, who died at the age of 27 in 1753 less than two years after reading his paper "Observations on Light and Color."

The credit for starting the field of spectroscopy is usually given to William Wollaston. In 1807 he observed the dark bands in the solar spectrum, which Fraunhofer later carefully measured. It seems a somewhat inverse method of founding a science more usually associated with light emission. Indeed, the field of flame spectroscopy is held to originate with the work of John Herschel and William Fox Talbot in the late 1820s, and it was some time later that it was noticed that the dark solar bands corresponded to certain flame emission bands. But tucked away in a footnote is the observation that "some work" on flame spectra had been done in the eighteenth century by Melvill.

I have a love of beginnings and had never heard of Melvill. Who was this unsung founder of our science, and what had he actually done?

Thomas Melvill was a Scottish natural philosopher who attended Glasgow University. In 1752 he delivered talks to the Medical Society of Edinburgh on two nights a month apart – January 3 and February 7. In them he addressed a number of topics on the nature of light and color, building upon the work of Newton. He examines and rejects the wave theory of light, agreeing with Newton's corpuscular theory. He then remarks upon the extremely small size and rarefied nature of these particles, which never seem to interact directly. He talks at length about the absorption of light by solid bodies and the attendant heating, observes that the way that light is reflected from drops of water on certain plant leaves indicates that they are not fully in contact with those leaves, and then goes off to discuss the appearance of colored objects under light of different colors. This naturally leads to an explanation of how to generate lights of different colors in the first place. He takes as his baseline "white" the light of an alcohol lamp. He then observes that "Bodies of all the principal colours, viz., red, yellow, green and blue, are very little altered when seen by the light of burning spirits: but, if salts be continually mixed with them during the burning, different changes ensue."

I have not been able to find the earliest references to the use of foreign material to add color to flames. Certainly it was known by the nineteenth century, when packets of powder were sold that would produce different hues from a fireplace flame. You can find direction today on internet sites for how to produce such effects. I have little doubt – but no documentation – that such tricks have been known for a very long time. In the course of tens of thousands of years of throwing things onto fires – for a long time the only artificial source of light and heat – people must have learned of many substances that could be placed on a fire to alter the color. (I note, for instance, that beechwood ashes contain so much manganese that they were used in the Middle Ages to color glass.) Melvill does not say how he came by his knowledge of flame

First Light, *cont. on p. 7*

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