

A TINY TOOLBOX

Stephen W. Carmichael,¹ Mayo Clinic

A recent article by Matthias Rief, Filipp Oesterhelt, Berthold Heymann, and Hermann Gaub² concluded with this sentence: "Single molecule force spectroscopy by AFM has proven to be a powerful addition to the nanoscopic piconewton toolbox." Everything about that conclusion is tiny. Clearly, the atomic force microscope (AFM) has given us a tool to examine structure at or near the atomic level. Earlier work from Gaub's laboratory, reviewed in this column³, demonstrated that the AFM could directly measure the binding force between single molecules of biotin and avidin. This established that the AFM could be used as a tool to measure forces, not just observe structure. Their most recent experiments has added to this tiny toolbox.

Strands of dextran, a long polymer with an average molecular weight of 500,000, were glued to a gold substrate with epoxy-alkanethiols. The glucose units of the dextran were activated with a carboxymethyl group and reacted with streptavidin. It was determined that several streptavidin molecules were chemically bound to each dextran filament. The strands extended into a physiologic buffer so that they formed a "polymer brush." An AFM cantilever with biotin bound to its tip was carefully dipped into this brush until a binding event was registered. Using this "fly fishing mode," Rief *et al.* were able to snag single strands. In cases where multiple strands were bound, they would slowly pull back the tip, rupturing bonds until just one strand was left attached. With a single strand suspended between the tip and the gold substrate, they could manipulate the strand up to the point where the streptavidin-biotin "handle" broke. The characteristics of the tension changes at low forces were entirely consistent with a single dextran filament being stretched.

Molecular dynamics calculations showed that the elasticity at low forces is due to a twisting of C5-C6 bond that results in an elongation of the filament. At forces less than one nanoNewton, this elongation is proportional to the force and gives rise to a segment elasticity of 750 picoNewtons/Ångstrom. Experimental results and theoretical calculations were in agreement.

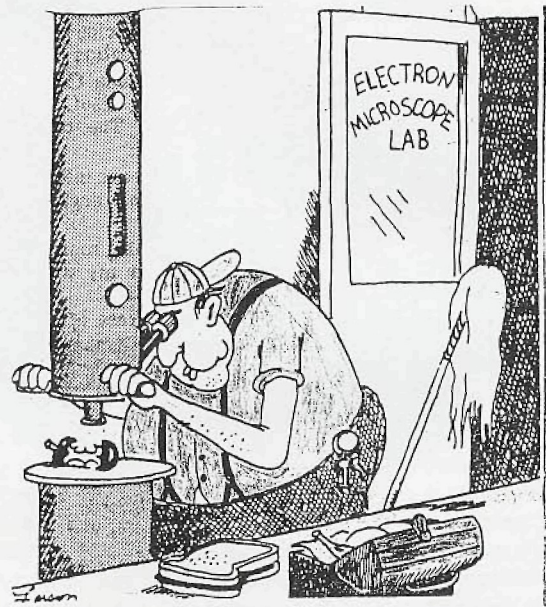
Measurements at higher forces were complicated by the fact that the streptavidin-biotin bond was not strong enough. A tighter bond, holding above one nanoNewton, was created by adsorbing the carboxymethylated dextran filaments (in the absence of streptavidin) onto an AFM tip that had

been made hydrophobic by silanization. At higher tension forces the elasticity showed a discontinuity which molecular dynamics simulations attributed to a snap of the C5-C6 bond angle into a new, stiffer conformation.

Individual filaments could be manipulated repeatedly, up to the rupture limit of its connection to the AFM tip. This experiment showed that the snap of the bond angle was reversible when the force was reduced again.

Rief *et al.* pointed out that this technique allows for the controlled manipulation of individual molecules can reveal details of the molecular basis of the mechanical properties of polymers that could not be obtained otherwise. The application of this tool to biologically significant properties of polymers remains to be performed, but clearly this is an important addition to a tiny toolbox!

1. The author gratefully acknowledges Matthias Rief for reviewing this article.
2. Rief, M., F. Oesterhelt, B. Heymann, and H.E. Gaub, Single molecule force spectroscopy on polysaccharides by atomic force microscopy, *Science* 275: 1295-1297, 1997.
3. Carmichael, S.W., Microscopy Isn't Just For Microscopists, Anymore. *Microscopy Today* 94-5, 28, 1994.



Front Page Image

GREEN CONE OPSIN EXPRESSION IN GOLDFISH RETINA DETECTED WITH NONRETROACTIVE *IN SITU* HYBRIDIZATION

Riboprobes to the goldfish green opsin visual pigment were hybridized to a tangential retinal cryosection and detected with an alkaline phosphatase color substrate (NBT/BCIP). The triangular purple profiles represent labeled green cones. The round profiles are another cone subtype, which expresses a different (ultraviolet) opsin gene.

For further information, review the article *Nonreactive In Situ Hybridization on Cryosections* on page 16 of this issue.

MICROSCOPY TODAY

A monthly newsletter dedicated to the unique interests in microscopy and mailed to some 8,000 microscopists worldwide - at no cost in the U.S. and with a modest international charge.

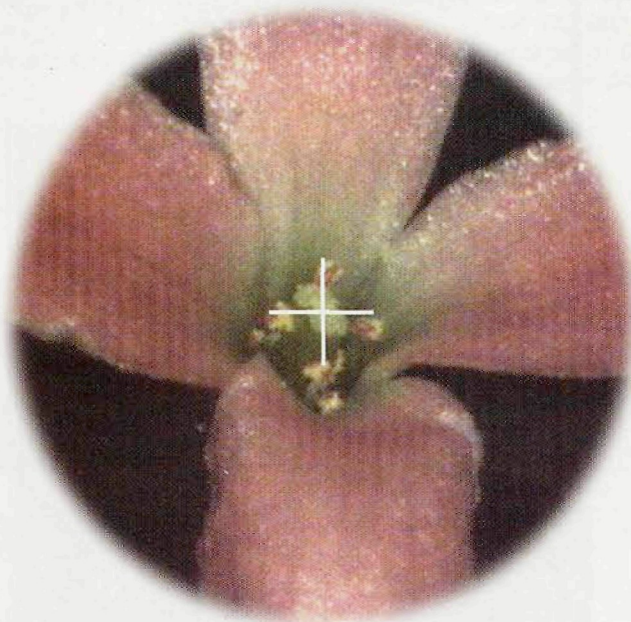
PO Box 620122, Middleton, WI 53562 - Tel.: (608)836-1970 - Fax: (608)836-1969 - eMail: MicroToday@aol.com

<http://www.microscopy-today.com>

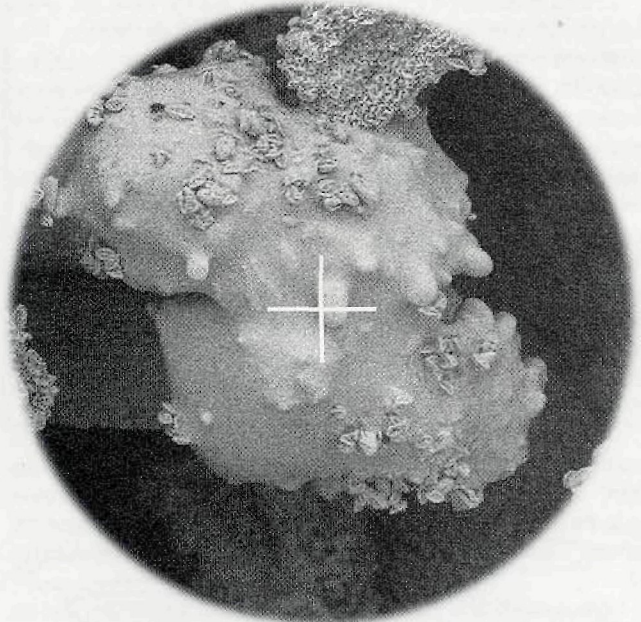
Copyright in 1997 by Today Enterprises. All rights reserved.

Don Grimes, Editor

Topcon Introduces Opti-SEM.TM A Whole New Way Of Looking At An Old Problem.



Optical Image



SEM Image

The Built-In Targeting Microscope Lets You Actually Find What You Want To See.
Think What That Means For Productivity.

With magnification, sometimes more *is* less. Locating a SEM target on a large, complex surface can waste a lot of time. Every time.

An Old Astronomer's Trick.

Astronomers use a small, broad field telescope to aim the main telescope. How easy. How productive. So Opti-SEM borrowed the idea. Its built-in 5x to 40x optical microscope makes targeting just as easy for you.

In fact, everything is easy with Opti-SEM. The Windows GUI interface is so familiar, the operator needs little training. And image averaging assures a clean, crisp image every time. No matter whether you output to digital, video or Polaroid.

Topcon's #1 Target—Affordable Flexibility.

Opti-SEM's stand-alone, industry-standard computer helps keep you at the state-of-the-art for less. Hardware and software upgrades are local and quick. Now when technology

advances, you can afford to move with it.

Better yet, there's an Opti-SEM 300 to meet your particular needs. Including a low-vacuum version. So set your sights on the best value in SEMs today. Topcon. Call 1-800-538-6850 for complete information.



Our Focus Is Clear...To Provide The Best Value
In SEMs Today.

✪ Microscopy & Microanalysis '97 - WOW

Attendance at the recent conference in Cleveland included 1,701 scientific registrants and 820 exhibitor personnel - with 135 exhibiting companies and organizations. The conference may have been the largest ever for this meeting.

Comments from both exhibitors and attendees were very positive. The Sunday evening reception at the Rock and Roll Hall of Fame was extremely enjoyable. Thanks, in particular, to the local arrangements committee.

✪ Digital Instruments Announces Merger Plans with Zygo Corporation

Digital Instruments recently announced the signing of a letter of intent to merge with Zygo Corporation, a leading supplier of optical measurement systems for industrial applications.

Digital Instruments is a world leader in scanning probe/atomic force microscopy (SPM/AFM). This technology allows scientists to see and measure topographical features on the nanometer scale and below. Digital Instruments has sold more than 2,200 NanoScope® SPM Systems and has won numerous awards for its innovation and engineering achievements.

Zygo manufactures non-contact precision surface measuring instruments, precision surfaces, optical components and coatings for industrial manufacturing and research.

Under the terms of the agreement, Digital Instruments will continue to manufacture and market products from their Santa Barbara, CA location.

For further information, contact Monte Heaton, Digital Instruments Director of Marketing, at (805)967-1400, www.di.com

✪ Electron, X-Ray and Ion Probe Spectroscopies - A Primer

Mark your calendars for an all day seminar Friday November 14, 1997 on "Electron, X-Ray and Ion Probe Spectroscopies - A Primer" to be held at Argonne National Laboratory, Argonne, IL. The seminar will be co-sponsored by the Electronic Materials Programming Committee of the Chicago Regional Chapter of ASM International and the Materials Science Division of Argonne National Laboratory.

Speakers will include Professor Vinayak Dravid, Northwestern University and Dr. Nestor Zaluzec, Argonne National Laboratory. Elemental spectroscopic techniques, from surface to volume will be presented. The program will include energy dispersive x-ray (EDX), Auger, x-ray photoelectron spectroscopy (XPS) and secondary ion mass spectroscopy (SIMS).

Complete information will be included in the October issue of *Microscopy Today*. The registration fee is \$30 and includes lunch. Pre-registration is MANDATORY. For registration information contact Anita Brandes at (847)205-2525 or by eMail: G10809@email.mot.com

✪ Luminescence Microscopy and Spectroscopy Society (SLMS)

Members of this Society are involved in cathodoluminescence (CL) and UV-excited fluorescence microscopy and many are involved in the earth sciences, material sciences and ceramics - plus applications in forensics, archaeology, and other fields. The society has been in existence for some 10 years. There is a semi-annual newsletter edited by Professor Kopp of the University of Tennessee. Dues are only \$10.00 for full members and \$5.00 for students.

The instrumentation used includes the familiar SEMs and EMPAs and a significant number of the members are using relatively simple and inexpensive cold cathode based electron beam systems which are small enough in size that they can be mounted directly on the stage of a conventional monocular or binocular transmitted light microscope. The CL of the mineral samples can then be seen directly, in real time, with a minimum of sample preparation and the information revealed on impurity distributions, etc. is otherwise very difficult or impossible to obtain with other existing techniques.

For further information on the SLMS, visit their [www site](http://www.zephyr.rice.edu/SLMS/SLMS.html):

✪ The **Microscopy-Tutor™** is a 90-minute, interactive CD-ROM (for both PC and Macintosh) that teaches the principles and practices of transmitted light microscopy. Developed at the University of Washington, this program is based on an accurate, three-dimensional model of a microscope that allows the user to view and manipulate components as they would on a real instrument. Microscopy-Tutor will be a valuable tool for anyone who uses a microscope or teaches light microscopy.

Microscopy-Tutor is unique for many reasons. The solid model of the microscope is more tangible and accurate than textbook illustrations, and it can show views of the microscope that are difficult to visualize on a real microscope or with photographs. In addition the model allows users to "take apart" the microscope, something that could not be done in a class. Finally, the program actually feels like using a microscope. Manipulations of the microscope are shown concurrently with the effect they produce on the image viewed through the oculars.

It is available on a 30-day free trial basis. If one is not completely satisfied, he has but to write "cancel" on the invoice and return it with no further obligation.

It is priced at \$195.00 and may be ordered by calling Lippincott-Raven Publications at (800)777-2295

✪ AN APOLOGY!

In our last issue, in a "last call" for the Microscopy and Microanalysis '97 Conference, I suggested that "Cleveland might not be the place you would select for your (next) honeymoon". A surprising number of readers took issue with this thought!

This conference happened to be the seventeenth MSA Conference that I have attended and I can honestly say that I enjoyed Cleveland at least as much as I have any other American city. The following is a quote, which I find accurate, from one of the letters that I received:

Cleveland is a vibrant city with an attractive downtown and numerous historical, sports, and cultural attractions. It has an abundant night life, nationally respected medical and educational institutions, and many beautiful beaches along the shores of Lake Erie. In fact, Cleveland is not only a wonderful place to spend your (next) honeymoon, it is also a great place to live and raise a family.

I hereby humbly and sincerely apologize to all in Cleveland!

--- Don Grimes, Editor

BTW: My plan is to take our (next) honeymoon during the ICEM XIV/International Congress on Electron Microscopy in Cancun, Mexico. See you there?

✪ ICEM XIV/International Congress on Electron Microscopy, Cancun Mexico, August 31 - September 4, 1998

The Mexican Society of Microscopy, on behalf of the other Latin American Societies, is proud to host the ICEM-14 in Cancun, a beautiful place on the Caribbean which offers the possibility of enjoying the facilities of modern Mexico amid the land of the ancient Mayan Culture.

ICEM-14 will be the last meeting of its kind in our century. Therefore, we will review the achievements reached so far and glitter into the future of the technique. We also aim to join the newest imaging techniques being developed in TEM and SEM. In particular, optical microscopies are expected to play a major role in this conference.

The program will be very complete and stimulating, with a wide participation of microscopists from all over the world. The Mexican Committee is working on a program with strong academic content and a high level equipment exhibition. Also, a social program with the traditional warmth and human affection of the Latin American people is being planned. All of this will make ICEM-14 one of the best ever.

--- Miguel José Yacamán, Chairman

Note: For information on this upcoming conference, check out their [www site](http://icem.inin.mx):

1998 MICROSCOPY SOCIETY OF AMERICA AWARDS

All MSA Members are reminded that nominations are currently being solicited for the 1998 MSA Awards. The Awards include:

- MSA Distinguished Scientist (Biological Sciences)**
- MSA Distinguished Scientist (Physical Sciences)**
- MSA Burton Medal**
- MSA Outstanding Technologist (Biological Sciences)**
- MSA Outstanding Technologist (Physical Sciences)**
- MSA Morton D. Maser Distinguished Service Award**

Distinguished Scientist Awards: These Awards honor preminent scientists from both the Biological and Physical disciplines who have an internationally recognized record of outstanding achievements in the field of microscopy and microanalysis.

Burton Medal: The Burton Medal was initiated to honor the distinguished contributions to the field of microscopy and microanalysis of a scientist who is not older than 35 years of age in the January of the award year.

Outstanding Technologist Awards: These Awards honor technologists from both the Biological and Physical Sciences who have made significant contributions such as the development of new techniques which have contributed to the advancement of microscopy and microanalysis.

Morton D. Maser Distinguished Service Award: This Award was initiated to recognize outstanding volunteer service to the Society as exemplified by Mort Maser, who served the Society for many years with great dedication. This award is made to honor an MSA member who has provided significant volunteer service to the Society over a period of years.

The Distinguished Scientist, Burton Medal, and Outstanding Technologist Awards Nominations should include:

- 1) a letter from the primary MSA nominator describing the research accomplishments of the candidate with particular emphasis on the unique technical achievements in the Physical or Biological Sciences; and
- 2) supplemental letters of support from other members of the scientific community.

The Morton D. Maser Distinguished Service Award Nomination should include:

- 1) a letter from the primary MSA nominator describing the basis for the nomination; and
- 2) supplemental letters of support from other members of MSA.

The Deadline for receipt of Awards Nomination Packages is December 31, 1997. Please contact the MSA Awards Committee [Gracie Burke (mgburke@pitt.edu), Stan Erlandsen (stan@lenti.med.umn.edu), Bev Maleef (Beverly_E_Maleeff@sbphrd.com), Jim Bentley (bentleyj@ornl.gov), Bill Gunning (wgunning@magnum.mco.edu)] or the MSA Business Office for additional information.

COMING EVENTS

- ✓ Sept 22/25 '97: **International Conference on Texture and Anisotropy of Polycrystals** (TU Clausthal, Germany). Robert Schwarzer: Fax: (+49)5323-722340, eMail: schwarzer@tu-clausthal.de
- ✓ Sept 25 '97: **Computers in Microscopy** (RMS) Cambridge, UK. Rebecca Morden: eMail: info@rms.org.uk
- ✓ Oct 5/8 '97: **Multinational Congress on Electron Microscopy**. Portoroz, Slovenia. <http://www2.ijs.si/~k5www/MCEM97/index.html>
- ✓ Oct 6/10 '97: **Scanning Electron Microscopy & X-ray Microanalysis**. (SUNY, New Palz, NY. Dr. A.V. Patsis: (914)255-0757, Fax: (914)255-0978
- ✓ Oct 8/16 '97: **Optical Microscopy and Imaging in the Biomedical Sciences** (Marine Biological Lab) Woods Hole, MA. Carol Hamel: (508)289-7401.
- ✓ Oct 20/24 '97: **44th AVS National Symposium**. San Jose, CA. Ms. Marion Churchill: AVS, 120 Wall St., 32nd Floor, New York NY 10005
- ✓ **Optimizing Light Microscopy** (1 day short course)
Nov 3 '97: New York City
Nov 5 '97: Springfield, MA
Nov 7 '97: Boston, MA
Barbara Foster (MME): (413)746-6931, mme@map.com
- ✓ Nov 14 '97: **Electron, X-Ray and Ion Probe Spectroscopies - A Primer** (ASM & Argonne National Lab) Argonne, IL Anita Brandes: (847)205-2525, eMail: G10809@email.mot.com
- ✓ Nov. 17/20 '97: **Eastern Analytical Symposium**. Somerset, NJ. (302)738-6218, Fax: (302)738-5275.
- ✓ Dec 1/5 '97: **MRS Fall Meeting** Boston, MA. MRS: (412)367-3004, Fax: (412)367-4373
- ✓ Dec 13/17 '97: **37th Annual Cell Biology Annual Meeting** Washington, DC: (301)530-7153, Fax: (301)530-7139
- ✓ Jan. 7/10 '98: **Emerging Electron Microscopy Technologies Workshop** (Ted Pella, Inc.) U.C. Berkeley. Kathy Strangenberg: (916)243-2200, Fax: (916)243-3761, tedpel@aol.com
- ✓ March 1/6 '98: **PITTCON '98** New Orleans, LA, (800)825-3221, (412)825-3220, <http://www.pittcon.org>
- ✓ April 19/24 '98: **7th Biennial Frontiers of Electron Microscopy in Materials Science** (FEMMS98) Irsee, Germany: <http://femms98.llnl.gov>, email: weking@llnl.gov
- ✓ May 9/12 '98: **SCANNING '98: (FAMS)** Baltimore, MD. Mary K. Sullivan: (201)818-0086, Fax: (201)818-0086, fams@holonet.net.Internet
- ✓ **Practical Aspects Series of Short Courses at Univ of Maryland**
May 18/22 '98 Scanning Electron Microscopy - Session I
May 19/22 '98 Image Analysis
May 25/29 '98 Scanning Electron Microscopy - Session II
May 26/29 '98 X-ray Microanalysis
Tim Mangel: (301)405-6898, eMail: mangel@zool.umd.edu
- ✓ July 7/9 '98: **MICRO98** (Royal Microscopical Society) London, UK, +44 (0) 1865 248768, Fax: +44 (0) 1865 791237, info@rms.org.uk
- ✓ Aug 31 - Sept 4 '98: **ICEM XIV/International Congress on Electron Microscopy**. Cancun, Mexico. (525)553-4507, Fax: (525)553-4500, email: icem@icem.inin.mx

Royal Microscopical Society

1997/8 Programme of Forthcoming Meetings and Courses

1997

- 10 Sept Microlocalization of Chemicals in Biological Systems
- 21/25 Sept Computers in Microscopy Course and Colloquium
- 10/14 Nov Ultrastructural Immunocytochemistry Course
- 12 Nov 25th Scottish Microscopy Group Symposium

1998

- 24 Feb National Cell Sorting Workshop
- 27 Feb Microscopy of Interfaces - Joint EMAG/RMS Meeting
- 4 March Annual Immunocytochemistry Meeting
- 6/8 April Microscopy of Composite Materials - in Materials
- 14/18 April Focus on Multidimensional Microscopy
- 20/24 April Spring School in Electron Microscopy
- June Food Microstructure
- June Immunophenotyping Meeting
- 7/9 July MICRO98 - Microscopy Conference and Exhibition
- 20/24 July Light Microscopy Summer School, Principles & Applications
- 27/31 July Light Microscopy Summer School, Confocal & Stereology
- 28/30 July Bench-top Flow Cytometry Workshop
- September Lectins Workshop
- September Immunocytochemistry Course
- September Flow Cytometry Course
- November Ultrastructural Immunocytochemistry Course

For further details about these or other RMS activities:

Tel.: +44 (0) 1865 248768

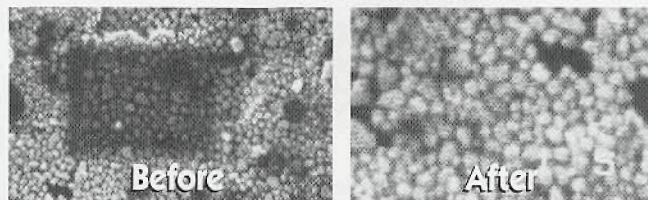
Fax: +44 (0) 1865 791237

eMail: meetings@rms.org.uk

<http://www.rms.org.uk>

Carbon Build-up Stopped!

SEM Laboratory Secret Revealed:



SEM manufacturers won't admit it, but most SEMs are subject to contamination build-up—even dry pumped systems. To stop hydrocarbon condensation, smart SEM users rely on the XEI Scientific SEM-CLEAN™ system.

Result: Outstanding pictures at low kV and high resolution and no oil on EDS X-ray detector windows. The Nitrogen purge of the inexpensive SEM-CLEAN system cleans your electron microscope while you're away.

SEM-CLEAN™ Stops the Oil

XEI
SCIENTIFIC

3124 Wessex Way, Redwood City, CA 94061-1348

650-369-0133 ■ Fax 650-363-1659

<http://www.msa.microscopy.com/SM/XEI/XEIHomePage.html>