

Very Cool Clathrin

Stephen W. Carmichael¹
Mayo Clinic
carmichael.stephen@mayo.edu

Clathrin-coated vesicles are the shuttle containers within cells. The vesicles carry lipids and proteins between membrane-bound compartments. Clathrin forms a cage-like structure around the membrane-bound vesicle that is pinched off from the plasma membrane (in endocytosis) or a membranous component of the cytoplasm. Clathrin recruits cargo that is within a vesicle through intermediary proteins known as adaptors that help select membrane-anchored protein and form an interface between the clathrin cage and the membrane bilayer. Whereas many earlier studies have elucidated much of the structure of clathrin, a recent electron cryo-microscopy study by Alexander Fotin, Yifan Cheng, Pitor Sliz, Nikolous Grigorieff, Stephen Harrison, Thomas Kirchhausen, and Thomas Walz provides information on a sub-nanometer scale.² In the same issue of *Nature*, the same laboratory group described the structure of an auxilin-bound clathrin coat and the implications for the mechanism of uncoating,³ but this article will not be discussed in this column.

Clathrin-coated vesicles vary in a dynamic, as well as a static manner. The dynamic requirement is involved when a piece of membrane is pinching off to form a vesicle. The static phase relates to the various sizes of clathrin-coated vesicles. The clathrin molecule is a trimer of three long subunits that radiate symmetrically from a central hub. Each subunit has characteristic straight regions and bends that I will refer to by homology to the lower limb: a thigh (Fotin *et al.* call it the “proximal segment”), knee, leg (“distal segment”), ankle, hindfoot (“linker”), and forefoot (“terminal domain”). These trimers can assemble *in vitro* into small, medium, or larger cages, the latter two being the most common. Fotin *et al.* called the medium-sized structures “hexagonal barrels” and the larger ones “soc-

cer balls.” They reconstituted coats from purified clathrin and adaptor complexes *in vitro* that provided relatively homogeneous specimens with predominately barrels, suitable for electron cryomicroscopy. Images were initially reconstructed at 2.1 nanometers (nm), but by averaging many images, creating density maps, and other manipulations, they were able to achieve a remarkable resolution of 0.79 nm!

The interactions of the trimers were studied in detail. Fotin *et al.* wanted to describe how an assembling cage adapts to cargoes of different sizes and shapes and how the assembly of such an elaborate lattice can be modulated by interactions with regulatory factors. To summarize a complicated description, there are two basic mechanisms at work. One is that individual trimers are flexible, specifically at the knee, ankle, and hindfoot. The hub is relatively inflexible. The second mechanism involves the fitting together of neighboring trimers. They can be arranged at sharp angles to each other, creating a small ball, they may even be arranged in a flat extended array, or various-sized spherical structures in between. They apparently can transition from one configuration to another to fit dynamic requirements during endocytosis.

Clathrin plays several important functions in shuttling cargo within cells. This elegant study by Fotin *et al.* goes a long way in explaining how this molecule can be so versatile in order to play different roles. ■

References

- 1 The author gratefully acknowledges Dr. Stephen Harrison for reviewing this article.
- 2 Fotin A., Y. Cheng, P. Sliz, N. Grigorieff, S.C. Harrison, T. Kirchhausen, and T. Walz, Molecular model for a complete clathrin lattice from electron cryomicroscopy, *Nature* 432:573-579, 2004.
- 3 Fotin A., Y. Cheng, N. Grigorieff, T. Walz, S.C. Harrison, and T. Kirchhausen, Structure of an auxilin-bound clathrin coat and its implications for the mechanism of uncoating, *Nature* 432:649-53, 2004.

INDEX OF ARTICLES

Very Cool Clathrin	3	Fostering LIMS Development Through Open Standards	44
<i>Stephen W. Carmichael, Mayo Clinic</i>		<i>Avrum Goodblatt, U. Penn School of Medicine, Philadelphia</i>	
Metallography for the European Copper Age: Research on the		50th Anniv. Celebrations of Atomic-Resolution Imaging	46
Axe-Blade of the Glacier-Mummy from the Ötztaler Alps in Tyrol .. 8		<i>Thomas F. Kelly and Allan J. Melmed</i>	
<i>Gerhard O. Sperl, Institute for Historical Materials, Leoben, Austria</i>		Attaching Spheres to Cantilevers for Colloidal Probe Force	
Microscopy and Microbes at Plum Island: Protecting		Measurements: A Simplified Technique	48
America's Livestock	16	<i>Yang Gan, University of Newcastle, Callaghan, NSW, Australia</i>	
<i>Thomas G. Burrage, Plum Island Animal Disease Center, NY</i>		Nissl: The Man, The Stain and The Substance	50
Color Metallography	22	<i>John A. Kiernan, The Univ. of Western Ontario London, Canada</i>	
<i>George F. Vander Voort, Buehler Ltd, Lake Bluff, Illinois</i>		Industry News	52
Characterization of Solids from Oilfield Emulsions	28	NetNotes	56
<i>Richard W. Cloud,† Rebecca L. Ramsey,‡ Robert A. Pultz,‡ and</i>		Index of Advertisers	62
<i>Michael K. Poindexter‡, † Nalco Company, Naperville, Illinois; ‡ Nalco</i>			
<i>Energy Services, Sugar Land, Texas</i>			
Automated, Robotic Preparation of Vitrified Samples			
for 2D and 3D Cryo Electron Microscopy	32		
<i>P. M. Frederik¹ and M.H. Storms²; ¹Univ. Maastricht, The Netherlands,</i>			
<i>² FEI Company, Achtseweg Noord Eindhoven, The Netherlands</i>			
Ex-Situ “Auto Lift” Technique for TEM Sample Preparation	40		
<i>Garth “Brian” Cook, Micro Optics of Florida, Davie, Florida</i>			
Writing Nano-Scale Patterns on Insulators Using Variable			
Pressure Electron-Beam Lithography	42		
<i>Floyd Miller and David Frey,* Lehigh University Bethlehem, PA</i>			
<i>and *Carl Zeiss SMT Inc.</i>			
Microscopy for Children	43		
<i>Caroline Schooley, MSA Project MICRO</i>			

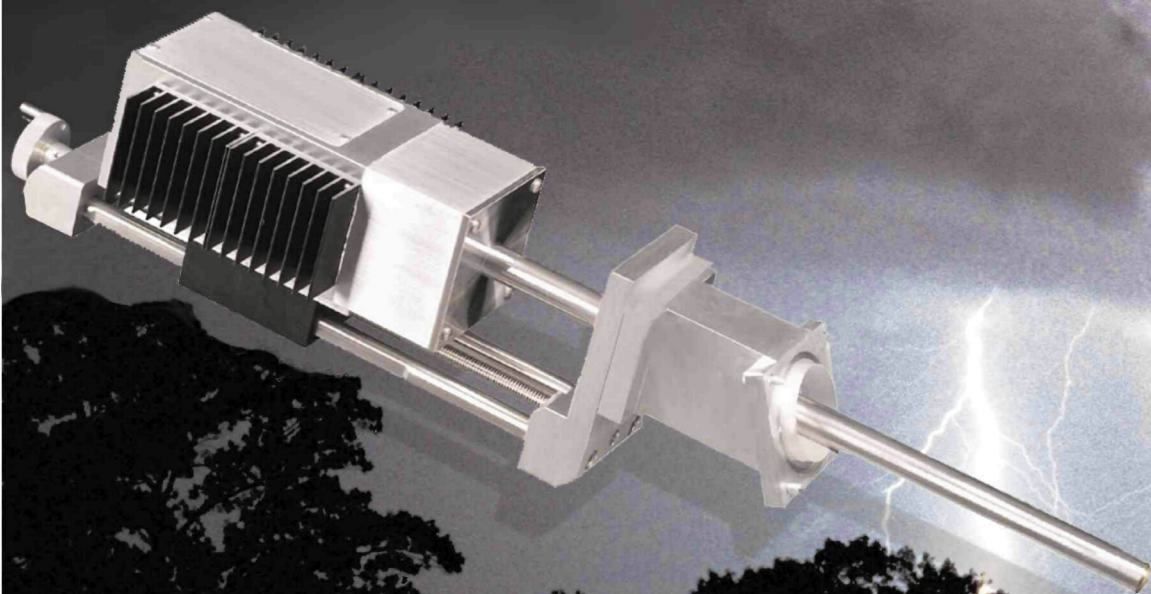
ABOUT THE COVER

From Saunders' "Under the Influence Collection." November's cover is the image of crystals formed from the mix of several rums influencing the crystallization of a vitamin substrate. "Why rum – spending time in Halifax, NS where rum-runners of “yore” smuggled this nectar of the Caribbean past the revenue cutters, this image takes me home. The “hard part” is getting to the last few drops from which to make the crystal. (Zeiss Axiophot, 5/0.15 PlanNeofluar, Transmitted, Cross Pol, 1st order red + a few tricks—no manipulation of the final image. Fuji Realla ASA 100 neg. Final Image 44x65 inch Giclee on canvas.) ...T.H. Saunders

4th Anniversary **Sahara** 4th Anniversary
The Detector That Revolutionized EDS

High performance EDS without liquid nitrogen

- Convenience - especially for multi-user environment
- Cool only when needed - 20 second cooldown
- No tending detector during vacation or plant closings
- No liquid nitrogen related safety hazards



- Excellent light-element sensitivity
- Boron detection
- **AND NOW** premium resolution of 129eV!

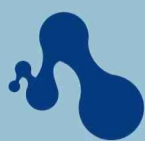
Sahara - proven quality & performance

For more information visit our web site: www.pgt.com/sahara.html

Princeton Gamma-Tech, Inc.
1026 Route 518
Rocky Hill, NJ 08553
Web site: www.pgt.com

Tel: (609) 924-7310
Fax: (609) 924-1729
Toll free: 800-229-7484
e-mail: sales@pgt.com





McCRONE ATLAS

OF MICROSCOPIC PARTICLES

Relieve your particle
identification **HEADACHES!**

The screenshot displays the McCrone Atlas software interface. At the top, there are navigation tabs: OVERVIEW, DATA, PLM, SEM, EDS, FTIR, and RAMAN. The main window is titled "Polarized Light Microscopy" and shows a sample identified as "23: Aspirin". The interface is divided into several sections:

- Top Left:** Time stamps "8:001000, 9:001001, 10:001010".
- Particle Data:** A text box containing the following information: "This sample consists of a ground-up, coated aspirin tablet. The main ingredient is, of course, the active drug, aspirin - see Aspirin (Pure) (AMP:7), but as added excipient here there is considerable corn starch - see Corn Starch (AMP:6). Gum Arabic and/or methyl cellulose may also be present, in addition to the coating itself." Below this text, it says "In the 100X views (Figures P23-1 P23-2 P23-3)".
- Main View:** A large image showing green, star-shaped aspirin particles under polarized light. Below the image are controls for "ZOOM 100%" and "BRIGHTNESS".
- Bottom Left:** Three smaller thumbnail images labeled "P23-8", "P23-9", and "P23-10".
- Bottom Right:** A section titled "Conditions/Comments" with the text: "P23-9 200X, Fully-Crossed Polarizers, Over heated. Recrystallized."

- Particle images and information at your fingertips!
- Spend less time searching and more time finding the answers you need!
- The most comprehensive particle identification resource available!
- Trustworthy results from the leader in microanalysis!

www.mccroneatlas.com

COMING EVENTS

2005

- ✓ **Materials Research Society**
esp: In-Situ Electron Microscopy of Materials, Symposium MM
November 28- December 2, 2005, Boston, MA
info@mrs.org
- ✓ **9th International Symposium on Biomineralization**
December 6-9, 2005. Pucón, Chile
www.cimat.cl/biomin09
- ✓ **American Society for Cell Biology**
December 10-14, 2005, San Francisco, CA
www.ascb.org

2006

- ✓ **TMS**
March 12-16, 2006, San Antonio, Texas
ckobert@tms.org
- ✓ **PITTCON 2006**
March 12-17, 2006, Orlando, Florida
www.pittcon.org
- ✓ **The American Chemical Society**
March 26-30, 2006, Atlanta, Georgia
natlmtgs@acs.org
- ✓ **American Soc. for Biochemistry and Molecular Biology**
April 1-5, 2006, San Francisco, CA
www.asbmb.org
- ✓ **Focus On Microscopy 2006**
April 9-12, 2006, Perth, Australia
www.FocusOnMicroscopy.org
- ✓ **NIST/Microbeam Analysis Society Particle Workshop 2006**
April 24-26, 2006, Gaithersburg, Maryland
www.nist.gov/particle
- ✓ **SCANNING 2006**
April 25-27, 2006, Washington, DC
www.scanning.org
- ✓ **Lehigh Microscopy School**
June 4-16, 2006, Bethlehem, PA
www.lehigh.edu/microscopy
- ✓ **Short Course: AFM and other Scanned Probe Microscopies**
June 12-16, 2006, Raleigh, North Carolina
www.ncsu.edu/aif/afmcourse
- ✓ **Microscopy and Microanalysis 2006**
July 30-August 3, 2006, Chicago, IL
www.msa.microscopy.com
- ✓ **ICEM XVI International Microscopy Congress**
September 3-8, 2006, Sapporo, Japan
www.imc16.jp
- ✓ **Society for Neuroscience**
September 9-14, 2006, Washington, DC
info@sfn.org
- ✓ **American Society for Cell Biology**
December 9-13, 2006, San Diego, CA
www.ascb.org

2007

- ✓ **Microscopy and Microanalysis 2007**
August 5-9, 2007, Fort Lauderdale, FL
www.msa.microscopy.com

Please check the "Calendar of Meetings and Courses" in the MSA journal "Microscopy and Microanalysis" for more details and a much larger listing of meetings and courses.

MICROSCOPY TODAY

The objective of this publication is to provide material of interest and value to working microscopists!

The publication is owned by the Microscopy Society of America (MSA) and is produced six times each year in odd months, alternating with MSA's peer-reviewed, scientific journal *Microscopy and Microanalysis*. We greatly appreciate article and material contributions from our readers—"users" as well as manufacturers/suppliers. The only criterion is that the subject matter be of interest to a reasonable number of working microscopists. *Microscopy Today* has authors from many disparate fields in both biological and materials sciences, each field with its own standards. Therefore *MT* does not have a rigid set of style instructions and encourages authors to use their own style, asking only that the writing be clear, informative, and accurate. Length: typical article length is 1,500 to 2,000 words plus images, longer articles will be considered. Short notes are encouraged for our Microscopy 101 section.

MICROSCOPY TODAY

ISSN 1551-9295

Ron Anderson, Editor

randerson20@tampabay.rr.com

José Mascorro, Technical Editor

jmascor@tulane.edu

Phil Oshel, Technical Editor

peoshel@wisc.edu

Thomas E. Phillips, Contributing Editor

PhillipsT@missouri.edu

Dale Anderson, Art Director

microscopytoday@tampabay.rr.com

Regular Mail to:

Microscopy Today, P.O. Box 247, Largo, FL 33779

Courier Mail to:

1001 Starkey Road, Lot #374, Largo, FL 33771

Telephones:

1-(727)507-7101 • Fax: (727)507-7102 • Cell: (727) 631-1022

e-Mail:

microscopytoday@tampabay.rr.com

www Page:

http://www.microscopy-today.com

Colophon: *Microscopy Today* is created using components of Adobe Creative Suite CS2®

Total Circulation: 14,444

Disclaimer: By submitting a manuscript to *Microscopy Today*, the author warrants that the article is original (or that the author has the right to use any material copyrighted by others). The use of trade names, trademarks, etc., does not imply that these names lack protection by relevant laws and regulations. *Microscopy Today*, the Microscopy Society of America, and any other societies stated, cannot be held responsible for opinions, errors, or for any consequences arising from the use of information contained in *Microscopy Today*. The appearance of advertising in *Microscopy Today* does not constitute an endorsement or approval by the Microscopy Society of America of the quality or value of the products advertised or any of the claims, data, conclusions, recommendations, procedures, results or any information found in the advertisements. While the contents of this magazine are believed to be accurate at press time, neither the Microscopy Society of America, the editors, nor the authors can accept legal responsibility for errors or omissions.

© Copyright, 2005, The Microscopy Society of America. All rights reserved.

The Next Step... Innovative Solutions for Nano-Technology



ULTRA FESEM

An extension of our leading SUPRA™ series with new In-column EsB detector for superb high resolution imaging and unmatched compositional data.



CrossBeam®

The ideal workstation for all your Ion milling, TEM sample prep and high resolution SEM imaging requirements.



LIBRA®EFTEM

Unique In-column OMEGA filter combined with Koehler illumination for unrivalled flexibility in imaging and analysis.

Enabling the Nano-Age World®

Carl Zeiss SMT Inc
Nano Technology Systems Division
One Zeiss Drive
Thornwood, New York 10594
USA

Tel. +1 914 / 747 7700
Fax +1 914 / 681 7443
info-usa@smt.zeiss.com
www.smt.zeiss.com/nts

