

Newsletter

Of the

New York Microscopical Society

1 Prospect Village Plaza (66F Mt. Prospect Avenue) Clifton, New Jersey 07013-1918 GPS: Latitude 40.8648N, Longitude 74.1540W



April 2013

N.Y.M.S. (973) 470-8733

Volume 7 (27) Number 4

11th Annual Microscope Day at John Jay

Thursday, April 25th, 2013" New" Building, 524 W. 59th St New York, NY 10019, (Fifth floor, Room 5.67) Presented by New York Microscopical Society and John Jay College

Please join us for this event, which begins at 11:00 AM and concludes at 4:00 PM. It is an informal event where speakers will give short presentations with ample time in-between to interact with other attendees and speakers. This year's speakers and their presentation titles (exact times and order of speakers is subject to change):

•11:00 <u>Dr. Lawrence Kobilinsky</u>, Chair, Department of Sciences, John Jay College – *Opening Remarks*•11:15 <u>Stan Petrash</u> – Research Scientist, Henkel Corporation - *Advanced Microscopic Imaging: How People, Microscopes and Digital Cameras "See" Small Things*

•12:00 <u>Gerard Petillo</u> and <u>Anna Tverdovsky</u>, Forensic Firearm Consultants – Using a Microscope to Determine if a Bullet was Fired from a Gun, An Overview of Forensic Firearm Identification •1:00 Lunch

•2:00 <u>Nick Petraco</u>, John Jay College and the CUNY Grad Center – *Microscopical Examination of Household Dust*

•3:00 <u>Peter Diaczuk</u>, John Jay College and the CUNY Grad Center – *Primers and Gunshot Residues* •3:30 <u>Julie Cohen</u>, John Jay College – Yes, Virginia, it is a Science: Using Confocal Microscopy to Compare Toolmarks on Bullets

•4:00 Peter Diaczuk, President, New York Microscopical Society - Closing remarks

Exhibitors: Several firms dealing with microscopes and microscope accessories have graciously agreed to set up exhibits of their products for the day.

Refreshments to be served.

This event is free and open to all those interested in microscopy Photo ID necessary for entry into building

Human liver fluke (Clonorchis sinensis), from the Eric Gravé Image Archive at NYMS (see page 3)



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Save a Tree: Get The Extended Newsletter: By Email Only

Board of Managers (updated)

| Diaczuk, Peter, pedicopete@earthlink.net; (212) 237-885 | 96, Expy June 2013, | President |
|---|-----------------------|---------------------------|
| Scott, John, nyconsnfdn@aol.com; Expy June | 2015,Vice P | resident, Program Chair |
| Pollinger, Mel, pollingmel@optonline.net; (201) 791-9826 | 6, Expy June 2014,Tre | asurer, Editor, Librarian |
| Klaus, Angela, Ph.D., klausang@shu.edu; Expy | / June 2015,Se | cretary, Education Chair |
| O'Leary, Don, dkoleary@verizon.net; (201) 368-8849, | Expy June 2013,Cur | ator, Facilities Manager |
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| Perlowitz, Seymour, perlowitzs@hotmail.com; | Expy June 2013, | Board member |
| Reffner, John Jr., jrr1lp@gmail.com; (cell): (215) 527-18 | 382, Expy June 2014, | Board member |
| Scal, Roland, Ph.D., rscal@qcc.cuny.edu; (718) 631-607 | 1,ExpyJune 2013, | Board member |

Dues and Addresses

Please remember to mail in your Dues to: Mary McCann, Membership Chair **McCann Imaging 161 Claflin Street Belmont, MA 02478**

<u>Junior</u> (under age 18) \$10 Annually <u>Regular</u> \$30 <u>Student</u> (age 18 or above) \$20 Annually <u>Supporting</u> \$60 Annually <u>Corporate</u> (includes one advertisement in NYMS News) \$175 Annually <u>Life</u> \$300 (payable within the year) To avoid missing notices: Notify Mary McCann and Mel Pollinger if you have changed your address, phone or email.

Awards Given by the New York <u>Microscopical Society</u>

The New York microscopical Society takes great pleasure in recognizing and rewarding individuals who have contributed to either the activities of the society or to furthering microscopy. These awards are described in our website and in a pdf file for our email newsletter recipients. All members are eligible to nominate individuals for these various awards, and are encouraged to do so. John A. Reffner, Awards Committee Chairperson

Awards Committee Chair: John A. Reffner Members Jan Hinsch Don O'Leary Mel Pollinger

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New York Microscopical

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NYMS Lapel Pins



Mel Pollinger, Editor 18-04 Hillery St. Fair Lawn, NJ 07410-5207



The Mission of the New York Microscopical Society is the promotion of

theoretical and applied microscopy and the promotion of of education and interest in all phases of microscopy.

Alternate Meeting Notifications

Please note that due to time constraints in publishing, some meeting notices may be available by calling Mel Pollinger at 201-791-9826, or by visiting the NYMS website, or emailing: pollingmel@optonline.net *************

Dues for 2013 are due!

Buy and Read a Good Book on Microscopy.

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Clonorchis sinensis, the Chinese liver fluke, is a human <u>liver fluke</u> in the class <u>Trematoda</u>, Phylum <u>Platyhelminthes</u>. This <u>parasite</u> lives in the liver of humans, and is found mainly in the common <u>bile duct</u> and <u>gall bladder</u>, feeding on <u>bile</u>. These animals, which are believed to be the third most prevalent worm parasite in the world, are endemic to <u>Japan</u>, <u>China</u>, <u>Taiwan</u>, and <u>Southeast</u> <u>Asia</u>, currently infecting an estimated 30,000,000 humans. *Excerpted from Wikipedia*

A Message From NYMS Honorary Member Johan S. Ploem

Dear Mel Pollinger,

I have been mentioned on the Dutch media as a possible candidate for a Nobel Price.

Some members of the N.Y.M.S may be interested in some details on the development of multi-wavelenghs epi-fluorescence microscopy.

A comprehensive WEBSITE has been created for the National Dutch Science Museum to illustrate the reason for the incorporation of my early prototype fluorescence epiilluminators in the permanent collection of that museum, as well as the exposition of the

very first Leitz-Leica epi-illumination fluorescence microscope with a Ploemopak fluorescence epi-illuminator in the VISEUM science Museum in Wetzlar. Germany. Website:

www.ploem-fluorescence-microscopy.com

With kind regards Johan Ploem, (J.S. Ploem)

Dear Dr. Ploem: I was elated to hear about your possible candidacy for the Nobel Prize. I will be publishing your information in the April 2013 NYMS Newsletter. I have already forwarded this email to our Board of Managers. Our membership will be elated when they read of your contributions to optical science and are also reminded that you were the recipient of the Abbe Award in 1998 and an Honorary member of the New York Microscopical Society.

I have attached a copy of the March 2013 extended email newsletter. If you would like to continue receiving it by email, please let me know. I did not have your email address until now.

Sincerely, Mel

Inter/Micro 2013

Inter/Micro is an internationally recognized conference that attracts microscopists from all areas of light and electron microscopy. Research presentations during the first three days cover techniques and instrumentation, environmental and industrial microscopy, and forensic and chemical microscopy. The final two days will be a hands on microscopy workshop,

Call for Papers

July 15-19, 2013 - Inter/Micro: 64th Annual Applied Microscopy Conference, Chicago, IL, USA Titles & Abstracts due by April 15, 2013

Upcoming conferences

July 15-19, 2013 Inter/Micro: 64th Annual Applied Microscopy Conference, Chicago, IL, USA

Hosted by: McCrone Research Institute Contact: Julie Antia e-mail: <u>intermicro@mcri.org</u> julie@mcri.org www.mcri.org Phone: 312-842-7100 Fax: 312-842-1078

NYMS Sand collection needs to be catalogued - see pg. 4





Quote of the Day Sent to me by Jeff Glover

"I love science, and it pains me to think that so many are terrified of the subject or feel that choosing science means you cannot also choose compassion, or the arts, or be awed by nature. Science is not meant to cure us of mystery, but to reinvent and reinvigorate it." — Robert M. Sapolsky

Got News? Send it to The Editor. If you have images and/or article related to microscopy, or a letter to the editor, please send it to me. It could be an interesting book, mystery photo, website or anything else you believe may be of interest to your fellow NYMS members, don't be shy, send it to the Editor.

Visitors Always Welcome to NYMS

Although most of our lecture meetings, workshops and classes are held in the NYMS Clifton facility on the last Sunday of the month, the building may be opened for special purposes at other times, by appointment only. For such an appointment, please contact Mel Pollinger by phone at (201) 791-9826, M-F noon to 9:30pm, or by email at pollingmel@optonline.net.

From The Editor... if you have email: Getting the newsletter by email means you can receive an <u>extended pdf version</u> that cannot be sent by "snail mail." Even if you only continue your USPS delivery of the newsletter, NYMS needs your email address for reporting priority events and special news. Being able to contact you quickly by email means better communication between you & NYMS== Mel

Dues for 2013 are due!

Need to use a Microscope?

The various microscopes that are presently set up on the main floor of the New York Microscopical Society building in Clifton, N.J. are there for the use of its members.

Microscope Cleaning Kit

A complete set of tools and accessories to keep your microscope in optimum operating condition. The kit is put together by our Curator/Educational Chairman and available directly from NYMS for only \$35.00 plus shipping & handling, or may be purchased at a meeting. Call or email Mel Pollinger or Don O'Leary for details (see page two for contact numbers).

Also: Slide boxes 100 capacity, used: \$5.00 while they last

Sand Collection Obtained: NYMS recently obtained a collection of 225+ small bottles of sand from around the world. Bottles are labeled with locations. Besides the bottles, there are numerous other containers of sands obtained as part of the collection, these are also labeled with locations or type of sand. All collected globally up to the 1980's. Help is needed to catalog the collection. (See page 3) For additional information please contact Mel Pollinger at pollingmel@optonline.net or (201) 791-9826 up to 9pm weekdays.

Errata from March 2013

Navicula lyon should be Navicula lytra (last page bottom of extended newletter) Gyosigma should be Gyrosigma (Front page bottom)

Answer to Mystery Photo for March 2013



Bug's eye. Squashbug head showing compound eye and antennae. Bug's eye guessed correctly by Jay Holmes. Photo by Mel Pollinger

Mystery Photo for April 2013



Want to take a guess? Send it to me by email or call me: pollingmel@optonline.net, (201) 791-9826

Additional Historical NYMS Supplements Email Newsletter recipients will also be getting copies of NYMS Newsletter pdf back-Issues from 2007. Copies of older newsletters will be sent as I convert them.

Got something you want to sell, trade or publish in the Newsletter and/or on the website? Write, call or send an email message to: 201-791-9826 or <u>pollingmel@optonline.net</u> (images ok) or Mel Pollinger, Editor NYMS Newsletter 18-04 Hillery Street Fair Lawn, NJ 07410





Supporting Member

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NYMS Newsletter Extended Section, April 2013

Directions to NYMS Headquarters

One Prospect Village Plaza (66F Mount Prospect Avenue) Clifton, NJ 07013

GPS: Intersection of Colfax & Mt. Prospect: Latitude 40.8656 N, Longitude 74.1531W, GPS: Our building: Latitude 40.8648 N, Longitude 74.1540 W From George Washington Bridge:

In This Section: •Micro-Garden at the Hinsches part2 •Jan Hinsch Sand Talk •Johan Ploem part 1 •Johan Ploem part 2 •Microscope Day at John Jay •Georgia Microscopical Society •Dr. Cymmek talk in NYC •Sandfest •Membership Application •Items for Sale by NYMS Last page images

Take Interstate Route 80 west to Exit 57A, Route 19 South. Take Route 19 to Broad Street and continue two lights to Van Houten Avenue. Turn Left. Go to second light, Mount Prospect Avenue and turn left. Building 66F is on the left side , one and a half blocks from Van Houton.

From Lincoln Tunnel:

Follow exit road to NJ route three west. Continue to Bloomfield Avenue exit. Turn right to Circle and go three quarters to Allwood Road West. Mount Prospect Avenue is a few blocks on the right (a small street) Turn right and go to first light (Van Houton) continue. Building 66F is on the left side , one and a half blocks from Van Houton.

From North:

Take Garden state Parkway South to Route 46 Clifton Exit. On 46 Make second exit to Van Houton Ave. Continue to third light Mount Prospect Avenue and turn left. Building 66F is on the left side , one and a half blocks from Van Houten.

From Route 46 coming from west:

Take Broad Street Exit in Clifton and follow Directions above from GW Bridge.

From route 46 coming from East: Take Paulson Avenue Exit in Clifton and follow to Second light, Clifton Ave turn right. Go to next light, Colfax, turn left, go three blocks and turn right on Mount Prospect Ave.. Building 66F is half block on right.

Public transportation from NY:

Take NJ Transit train from Penn Station to Secaucus Transfer Station. Change trains to Bergen Line to Clifton (call NJ Transit for schedules). From Clifton Station cross under tracks to first street and go left one block to Mount

Prospect Street, turn right and Building 66F is one half block on Right.

If you plan to come by bus or train, please copy the links below into your browser:

http://www.njtransit.com/sf/sf_servlet.srv?hdnPageAction=TripPlannerItineraryTo http://www.njtransit.com/sf/sf_servlet.srv?hdnPageAction=BusSchedulesP2PTo http://www.njtransit.com/sf/sf_servlet.srv?hdnPageAction=TrainTo

The Garden Under The Microscope

by Jan & Wiebke Hinsch

Part 2

Insect Eggs, Pollen and Spores

Jan & Wiebke Hinsch gave a talk at our holiday party in December: "The Garden under the Microscope." We will show you an excerpt of their pictures.

Jan and Wiebke look at their garden from different perspectives. Wiebke is a master gardener and her garden reflects her love for plants, design, and environmental responsibility. She taught Jan to look at the garden as an endless source of miraculous things with which to feed his microscope. This talk therefore is a synthesis of Jan's and Wiebke's passions



insect Eggs

Insect eggs are often hidden on the underside of leaves. These are 1 $\frac{1}{2}$ mm in diameter with a perforation ring where the larva can hatch.



Stinkbug Eggs

insect Eggs



Stinkbug Eggs

insect Eggs



These are empty shells of a predatory stinkbug that is beneficial to the garden.

Pollen and Spores

Flowering plants have pollen, lower plants like ferns and mosses produce spores for propagation. Fern spores grow on the underside of the leaf, protected by an indusium. When they are ripe, the indusium opens and the sporangies are exposed. They each have a spring-like spine that at the right time propels the spores into the open.



Stained pollen grains

Spores



Underside of fern





Fern sporangies

Spores



Fern indusium (stained)

Spores



Fern Spores & Sporangies (stained)

Jan Hinsch's Sand Presentation for March 2013

Coinciding with NYMS obtaining two collections of sand samples from worldwide locations, all neatly packaged and labelled, Jan opened the door to a new chapter in the micro-materials available at NYMS for study, and, in some cases, where sufficient material is available, for sampling by our sand collecting members.

Jan's presentation included photomicrographs of sand using polarizedlight, Rheinberg illumination and modified dark-field illumination. He also discussed and presented images and information regarding optical phenomena and devices such as 2V, Refractive index and the use of the Spindle Stage. MP

Sand images by Jan Hinsch



Shell sand from Cancun, Mexico

Quartz sand with Inclusions from Amelia Island, Florida



SECTION

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DOCUMENT DRAWER == CLICK TO OPEN/CLOSE ==

Bas Ploem



Professor Ploem is renowned for his research in fundamental light microscopy, especially his leadership in fluorescence microscopy. He developed the first four-wavelength vertical fluorescence illuminator for excitation with a choice of narrow-band ultraviolet, violet, blue or green illumination. This epi-illumination instrument was first marketed for general fluorescence microscopy by Leica under the trade name "Ploem-opak." The Ploem four wavelength epi-fluorescence illumination systems quickly became a standard for all major optical microscope manufacturers. Today, it is the dominant fluorescence microscopy technology. It is widely used in medicine, biology, and industry. This development significantly contributed to progress in cellular immunology and chromosome genetics. Later he developed, together with Leica, an improved optical design for reflected light microscopy for use in the biological and medical sciences. He proposed the name "Reflection-Contrast Microscopy" for this optical system. This microscope system produces images with very high definition and is successfully applied to thin

http://www.cyto.purdue.edu/cdroms/cyto10a/cytometryhistory/individualhistories/ploem.html

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sections in Immuno-cytochemistry applications. Professor Ploem's collaboration with Leica also lead to a new instrument for correlative microscopy of the same specimen with fluorescence microscopy and scanning electron microscopy (1977). In this instrument, a fluorescence microscope system was built into the vacuum chamber of a scanning electron microscope, permitting simultaneous observation of the same specimen With LM and SEM. The early development of computer-operated microscopes was also advanced by Professor Ploem's significant contributions. For automated image analysis of cervical specimens, a new fully-automated computer-operated microscope, the AUTOPLAN, was developed by Leica in collaboration with Professor Ploem. By combining automated microscopes with image analysis software developments, Ploem and his team catalyzed development of several commercial systems for automated cytology. With his coworkers at Leiden University, one of the first systems for automated cervical cytology screening (LEYTAS) was developed in collaboration with Leica and the Institute for Mathematical Morphology (Fontainebleau, France). In a collaboration with Zeiss, a laser scanning fluorescence microscope was tested as early as 1980.



Bas Ploem and Hans Tanke in front of the Leytas system, developed by Bas for automated cervical screening. The next picture is the prototype of the first epi-illumination fluorescence microscope that he developed in Amsterdam in the sixties (publication in 1967; Leitz Mitteilungen). The two others are prototypes of the epi-systems, with the dichroic mirror shifted in the pathway.

Johan S. Ploem is Professor Emeritus at Leiden University, the Netherlands. He is a graduate of Utrecht University, the Netherlands, receiving an MD in 1972. He worked as Intern in the Broussais Hospital (Paris, France) with Professor Pasteur Valery-Radot. In 1963, Dr. Ploem was elected a Fulbright Fellow for Study at the Harvard University School of Public Health, receiving a Master of Public Health degree Cum Laude in 1954. He obtained a Ph.D. degree in 1967 from the University of Amsterdam, the Netherlands. Professor Ploem has served as visiting lecturer or professor at various universities: Dundee. Scotland; University of Florida, USA; Monash University, Melbourne, Australia; University of Beijing, China; and at the Free University of Brussels, Belgium. In 1980, he was appointed to a professorship in the Department of Cytochemistry and Cytometry at Leiden University. He retired from that position in 1992.

Numerous honors and awards have been bestowed on Professor Ploem. In 1976 he was elected to the honorary Fellowship of the Royal Microscopical Society, Oxford England. In 1977 he received a Fellowship to the Papanicolaou Cancer Research Institute in Miami, Florida, and in 1979 a Fellowship to the Institute for Cell Analysis at the University of Miami, Florida. In 1982, he was the co-recipient of the C. E. Alken Foundation Award, Switzerland. In 1993, he was elected as the first Honorary Member of the International Society for Analytical Cytology. In 1993, Professor Ploem held the Erica Wachtel Medal Lecture at the British Society for Clinical Cytology meeting. In 1994, the European Society for Analytical Cellular Pathology established a Conference Keynote "Ploem" Lecture for invited scientists at its future general meetings. The International Society of Analytical

http://www.cyto.purdue.edu/cdroms/cyto10a/cytometryhistory/individualhistories/ploem.html

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Cytology invited Professor Ploem to present its inaugural "Robert Hooke" lecture. In 1995, he was invited by the Royal Microscopical Society to give the inaugural CYTO lecture. Professor Ploem has presented more than 200 invited lectures at Symposia and conferences outside of the Netherlands. He authored or co-authored more than 250 scientific publications. Professor Ploem's memberships include the Council of the Dutch Society for Clinical Cytology; the Royal Microscopical Society; the Council of the International Society for Analytical Cytology; the Board of the National Foundation for Scientific Research (Belgium); the Royal Society of Medicine, England; the International Council on Automated and Quantitative Cytology; the Research Section "Krebsfrueherkennung" in Cytology and Hematology of the Bundesministerium fuer Forschung und Technology, Germany; the "Cell Board Subcommittee" of the Medical Research Council; and the Standing Steering Committee on Biomedical Image Processing of the IEEE Computer Society, USA. He is Emeritus Editor of the Journal of Analytical Cellular Pathology.

This curriculum vitae was published at the occasion of Johan Sebastian Ploem receiving the Ernst Abbe Medal and Award from the New York Microscopical Society in the USA, November 17, 1998.

- Copyright
- <u>Reference</u>
- Acknowledgements
- <u>Disc Series</u>
- <u>Corrections</u>
- Purdue University Cytometry Laboratories | 1203 West State Street, West Lafayette, IN 47907
- For more information visit <u>www.cyto.purdue.edu</u> or email <u>cdrominfo@flowcyt.cyto.purdue.edu</u>



Johan Sebastiaan Ploem

From Wikipedia, the free encyclopedia

Website:

www.ploem-fluorescence-microscopy.com

Johan Sebastiaan Ploem (born 25 August 1927, Sawahlunto) is a Dutch microscopist and digital artist, who made significant contribution to the field of fluorescence microscopy.

DUTCH NATIONAL MUSEUM FOR THE HISTORY OF

SCIENCE AND MEDICINE (Boerhaave Museum, Leiden the Netherlands). Ploem's prototype fluorescence epi-illuminators and microscopes form a part of the permanent exposition of the Dutch National Museum of Science and Medicine (Boerhaave Museum, Leiden, the Netherlands) A comprehensive report has been made for this museum presenting published reviews of Ploem's contributions to fluorescence microscopy, citations and web links to his inventions and developments. **This report is published in a website:**

www.ploem-fluorescence-microscopy.com

Contents

- 1 Personal life
- 2 Career
- 3 Work on fluorescence microscopy
- 4 Digital Painting

Personal life

Ploem was born on Sumatra, then part of the Dutch East Indies, where his Dutch father was employed as a coal-mining engineer. At the age of two he returned with his parents to the Netherlands where he remained for the rest of his youth in Heerlen, a town in the south of the Netherlands. He started painting as a small boy and was educated in drawing and painting in Maastricht but after finishing high school opted to study medicine instead of art. During Ploem's entire career painting has remained a factor in his life, even during extensive scientific work in medicine, medical research and advanced microscopy.

Career

Ploem received his education at the University of Utrecht in the Netherlands, Harvard University and the University of Amsterdam. He has since then been employed by a number of academic institutions, including the University of Miami, the University of Amsterdam, and the University of Leiden, where he served as a professor at the Faculty of Medicine. He also cooperated with industry, in particular in the branch of optics and concentrated on research in image analysis, participating in a project aiming to automate cancer cell recognition.

Work on fluorescence microscopy

Around 1962 Ploem started work in collaboration with Schott on the development of dichroic beam splitters for

Johan Sebastiaan Ploem

| Born | August 25, 1927 Sumatra, Indonesia |
|-------------------|--|
| Nationality | Dutch |
| Fields | Advanced Microscopy; Molecular Diagnostics; Digital Painting |
| Institutions | University of Miami; University of Amsterdam; University of Leiden |
| Notable awards | Fellow of the Papanicolaou Cancer Research Institute (1977) C. E. Alken Foundation award |

Notes

Member of the Society of Analytical Cytology, the Dutch Society of Cytology, the International Academy of Cytology and the Royal Microscopical Society, for which he served as president in 1986.

Johan Sebastiaan Ploem - Wikipedia, the free encyclopedia

reflection of blue and green light for fluorescence microscopy using "epi-illumination" ((illumination and detection from one side of the sample)). At the time of his first communication [1965] and publication on epi-illumination with narrow-band blue and green light, he was not aware of the development of a dichroic beamsplitter for UV excitation with incident light by Brumberg and Krylova. Neither was the Leitz company, from which he obtained an "Opak" epi-illuminator with a neutral beamsplitter. This illuminator had to be modified to contain a slider in the incident light path containing four dichroic beamsplitters, for respectively UV, violet, blue and green excitation light. This device, developed at the University of Amsterdam, permitted the easy exchange of different dichroic beamsplitters in the incident light path. The wavelength of the excitation light could thus be easily and rapidly changed.

Soon it became clear that excitation with narrow-band blue and green light opened optimal possibilities for the detection of the widely used immunofluorescence labels fluorescein isothiocyanate (FITC) and tetramethylrhodamine isothiocyanate (TRITC). The use of blue and green excitation also minimized autofluorescence of tissue components, an undesired effect encountered with conventional transmitted illumination with UV light. FITC could now be excited with narrow band blue light (using a band interference filter with a half width of 16 nm), close to the excitation maximum at 490 nm (long wavelength blue), with clear observation of the green fluorescence peak emission at 520 nm. Autofluorescence of tissue components was minimized (Fig. 2a, b) resulting in a high image contrast. Excitation of FITC near its excitation maximum enabled such an efficient excitation that even a mercury high-pressure arc lamp, having no strong emission peak in the blue wavelength range, could be used. Furthermore epi-illumination with a green reflecting dichroic mirror enabled for the first time the excitation of Feulgenpararosaniline with the strong mercury emission line at 546 nm (Fig. 3a, b).



Fig. 2a: Tissue cells marked with an immunolabel (FITC) illuminated with wideband UV excitation. Note the tissue structure with blue autofluorescence.

Fig. 2b: Same tissue and same immunostaining with FITC label illuminated with epi-illumination using narrow-band blue (490 nm) light. Note the increased image contrast (Ploem, 1967)

Fig. 3a: Liver tissue. Nuclei stained with Feulgen-pararosanilin for DNA, and visualized with transmitted green light. This stain was known as absorbing stain and not known to be fluorescent. One on the nuclei is illuminated with incident narrowband green light (546 nm) resulting in a red fluorescence emission.

Fig. 3b: Liver tissue. Nuclei stained with Feulgen-pararosaniline for DNA. Epiillumination with narrow band green light (546 nm) and a dichroic beam splitter for reflecting green light. Probably the first example of microscope excitation with green light (Ploem, 1965). Note large image contrast.

In his second publication on the multi-wavelengths epi-illuminator, describing a Leitz prototype with four dichroic beam-splitters, Ploem could acknowledge the contribution of Brumberg and Krylova. The inaccessibility of Russian research in that time period, and the absence of any major industrial development of epi-fluorescence microscopy in Russia or East Germany was the reason that Leitz had not been aware earlier of such a development. The possibility to introduce epi-illumination with UV light, although useful for several applications, had not been a motive for a new technological development at Leitz, since they had already excellent transmitted dark field UV excitation available. The increasing worldwide use of routine immunofluorescence microscopy in medical diagnosis and molecular biology research could, however, profit from the new possibility of epi-illumination using narrow band excitation with blue and

Johan Sebastiaan Ploem - Wikipedia, the free encyclopedia

green light. Since standard high-pressure mercury arc lamps could be used, this seemed a practical proposition. Subsequently Leitz developed a novel multi-wavelength fluorescence epi-illuminator (Leitz PLOEMOPAK) with four rotating dichroic beamsplitters for respectively UV, violet, blue and green light. In successive generations of Leitz illuminators (containing four dichroic beamsplitters) barrier filters and a rotating turret for excitation filters were added. Finally an elegant epi-illuminator was constructed by Kraft containing multiple sets of a combination of an excitation filter, a dichroic beamsplitter and a barrier or emission filter, mounted together in a filter cube, also called filter block (Fig. 4). Since this illuminator permitted the filter cubes to be rapidly turned into the optical light path, multiwavelength illumination of the same section of tissue became a practical proposition. Moreover, the four filter cubes in the illuminator could be exchanged by the user (Fig 1). Different sets of four filter cubes could be assembled, chosen from many filter cubes, containing combinations of excitation, barrier filters and dichroic beamsplitters, developed for different applications. Following suggestions by Ploem, Leitz also produced an inverted microscope with epiillumination. For a review of the Leitz PLOEMOPAK illuminator for multi-wavelength fluorescence microscopy, the reader is referred to a review by Pluta.



Fig. 4: Complete Leitz (Leica) filter cube (block) for fluorescence microscopy containing: excitation filter, dichroic beam splitter and emission (barrier) filter.

The Leitz (Leica) filter cube system was so efficient that now, >45 years later, similar types of filter cubes are still used by most microscope manufacturers for multi-wavelength fluorescence microscopy. This development finally led within Leica to the development of automated multiwavelength fluorescence epi-illuminators accommodating eight filter cubes for various wavelength ranges. When switching between filter cubes, pixel shift on the computer monitor is avoided or stays below the resolution power of a 35 mm film due to a 0-pixel shift technology. This illuminator is now used for fluorescence in situ hybridisation methods (FISH) in the study of chromosomes. Ploem, van der Ploeg and Ploem and Nairn and Ploem further explored the filter combinations that had to be developed for many biomedical applications. This was done in collaboration with Schott and Leitz. Rygaard and Olson developed a novel shortwave pass high transmission interference filter with a very high transmission for blue light and a sharp cut-off towards wavelengths longer than 490 nm. Ploem combined this SP filter with a 1 mm GG 455 filter from Schott, which blocked UV excitation, and suggested the development by Balzers of a similar filter (SP 560 = KP560) for excitation with green light and a filter for excitation with violet light (LP 425 = KP 425). The latter filter was applied in the investigation of neurotransmitters. In Fig. 6a, b the resulting blue fluorescence can be observed. From the optical industry side, early contributions and reviews on these developments were written by Kraft, Walter, Trapp and Herzog.

The main classes of filters used in epi-illumination fluorescence microscopy were defined in (1) the primary excitation filters LP (long pass) and SP (short pass) – in the German literature known as KP filter – and (2) the secondary filters such as barrier filters and emission filters. The latter were also described as fluorescence selection filters; these are for instance used to limit the observation to the peak fluorescence at 520 nm of FITC. A recent extensive review on filters for fluorescence microscopy has been given by Reichman. Cormane was the first to demonstrate that narrow band blue light epi-illumination of the fluorescent label FITC gave an optimal contrast in immunofluorescence studies of human skin disease. Transmitted-light excitation with UV light used to cause such a strong auto-fluorescence of elastic fibres in the skin, so that visualization of the fluorescent antibody was severely hindered. The pioneering work of Leitz in epiillumination fluorescence microscopy coincided in the seventies with a worldwide increase in the application of immunofluorescence and other molecular biology methods like FISH in medical diagnosis and research. Hijmans et al. were the first to demonstrate the usefulness of the new Leitz multi-wavelength excitation epi-illuminator for the selective detection of certain classes of immunoglobu lines in cells, using antibodies conjugated with green fluorescent FITC and red fluorescent TRITC. They applied the two-wavelengths excitation method using blue and green light and the selection of the peak fluorescence of FITC by an emission filter at 520 nm (Fig. 7). Brandtzaeg and Klein et al. made similar discoveries in identifying immunologically important cell types, using two-wavelength excitation with the Leitz epi-illuminator. In a staining of blood with "rosette" formation, the two-wavelengths excitation method using UV and green light can demonstrate erythrocytes around a mononuclear cell (Fig. 8).



Fig. 6a: Mesenterium of the rat with small blood vessel surrounded by a blue fluorescent adrenergic (CA) nerve-plexus and yellow fluorescent mast (5-HT) cells. Formaldehyde-induced neurotransmitter fluorescence of CA and 5-HT fluorophores.

Fig. 6b: Same tissue and staining as Fig. 6a: Epi-illumination with narrow-band violet excitation light (LP 3mm GG 400 and SP (KP)425 interference filter), a dichroic beam splitter 495 nm, reflecting violet light and a barrier filter LP460nm. The filter cube permitted for the first time the observation of blue fluorescent adrenergic nerve fibers, distinctly different from yellow fluorescent mast cells (Ploem, 1971).



Fig. 7: Bone marrow cells stained with antikappa TRITC conjugate and an anti-IgG FITC conjugate. Epi-illumination with narrow band green and blue light, resulting in red fluorescence of cells containing TRITC and green fluorescence of cells containing FITC. Some cells contain both FITC and TRITC.



Fig. 8: Human blood cells, "rosette" formation. Erythrocytes stained with the blue fluorescent stain stilbene using epiillumination with UV light. Lymphocytes stained with the orangered stain eosine after excitation with green light (1965).

Digital Painting



Meadow near Sauto, Pyrenees, France

Early days

Bas Ploem started painting as a small boy, making copies of paintings in the house of his parents in Heerlen, a town in the south of the Netherlands. He lived there until he was eighteen. While still at secondary school he used to take the train to the nearby town of Maastricht to attend an evening course in drawing and painting at the "Kunstnijverheidsschool Maastricht", which was later converted into the 'Academie Beeldende Kunsten Maastricht' (Academy of Art, Maastricht). After finishing high school he had the chance to opt for a further education in art, but decided to study medicine instead.

Meeting with the painters Frits and Yves Klein in Paris

During his medical studies at the University of Utrecht in the Netherlands, he had the opportunity to work as an intern in the Hospital Broussais in Paris under the supervision of professeur Pasteur Vallery-Radot, the grandson of Louis Pasteur. Ploem's presence in Paris was important for his knowledge and interest in art since he could regularly visit his cousins in Paris, the painter Frits Klein

and his son Yves. He visited the Kleins when Yves was making his first monochromes.

Analogue paintings

During Ploem's entire career, painting remained a factor in his life, its intensity varying with the workload, first in medicine and later in medical research. On the basis of this lifelong activity as a 'Sunday painter' an exposition of his analogue paintings was organized in 1992 in Pulchri Studio in The Hague on the occasion of his retirement as a

Johan Sebastiaan Ploem - Wikipedia, the free encyclopedia

In the last years of his activities at the faculty of medicine at Leiden University, he concentrated on research in image analysis. He was asked to participate in a European project with the aim of automating cancer cell recognition using computer analysis. It concerned a collaborative project with the German optical company Leitz/Leica Microsystems, and the Institute for Mathematical Morphology in Fontainebleau, France. Together with a team, Professor Jean Serra at this institute had developed an image analysis method, now internationally known as 'Mathematical Morphology' (MM). With his experience as an analogue painter, Ploem quickly saw the possibility of also applying the methods of mathematical morphology to human faces, landscapes, buildings and flowers. Instead of looking at cells, the computer programmed for image analysis, can also look at the optical information of an image scanned into its memory by e.g. a camera. Unfortunately not Mathematical Morphology program was then available for use on a PC. It was only years later, on a visit to the firm Leica Imaging Systems in Cambridge, England, that Ploem accidentally saw a CD with the Mathematical Morphology program that could run in Windows on a PC. He received a copy of this program on loan and immediately started to use it for digital painting experiments.



In the valley of the Eyne, Pyrenees

Mountain flowers as the first topic for digital image analysis

Since Ploem is a nature enthusiast, he started with the application of mathematical morphology programs to the image analysis of meadows covered with mountain flowers. To get inspiration for his art work he frequently made nature walks in the region of the Pyrenees known as the Cerdagne, and specifically in the Eyne valley also known as the 'La Vallée des Fleurs'. These first digital graphics of nature scenes were shown in his exposition at a regional art centre in the Pyrenees (Ossega, June, 1997).

Scientific interest in computer graphics created with mathematical morphology

As he was probably the first person to systematically use mathematical morphology for the creation of digital art, Ploem's work attracted international attention and he was invited as a plenary speaker at an international mathematical conference in Amsterdam in 1998 to explain his new type of digital art. He also received an invitation to show his art work in an exposition at this conference. The organisers of this meeting asked Ploem to write a chapter on his novel technique for digital art in a book (Kluwer, ISBN 0-7923-5133-9) that was published on the occasion of this meeting.

Exposition at universities

When scientists in France became more aware of Ploem as a digital artist, they invited him for a symposium on 'Art et Science' at the University of Caen, France (April, 2001). At the art exposition connected with this symposium, he presented 6 digital graphics that were dominated by chaotic transformations of rock art themes. A similar invitation was made by the University of Basel in Switzerland (April, 2002). His exposition of digital graphics in Basel also showed works which were created with the so-called 'watershed transformation' of Mathematical Morphology, resulting in pictures resembling mountain ranges.

Acceptance of digital art

Generally spoken, digital art still suffers from a lack of acceptance by a wider public. One of the reasons may be that this type of art is sometimes difficult to understand or to explain. For Ploem as one of the early developers of digital graphics using image analysis, it was encouraging that the well known museum 'Fondation Beyeler' in Basel, Switzerland already had a separate curator for digital art in 2002. At the time of his exposition in Basel he was invited for a discussion with this curator about the future of digital art. They discussed the massive interest in digital art while still waiting for the emergence of more significant digital art by young artists who have been growing up with computers. The words 'Digital Art' typed in together in the advanced search made of Google now results in more than 300 million references!

Portraits and architecture

In recent years Ploem has spent time studying more conventional topics in art like portraits and scenes with architecture. The availability of special mathematical morphology software such as the 'Top Hat' transformation made his attempts at creating portraits rather interesting. This famous algorithm was developed by Fernand Meyer. As an assistant of Professor Jean Serra in the earlier mentioned Institute of Mathematical Morphology, he came to the University of Leiden, the Netherlands in order to write programs for the recognition of cancer cells visualized under the microscope. He is now director of this institute in Fontainebleau. The 'Top Hat' algorithm enhances the recognition of structures mainly based on contrast in the image rather than on edge detection. If applied to a human face it makes a novel type of drawing that is different from a drawing made by most artists. If followed by several other MM image transformations, it can produce (digital) portraits characterized by structures that were not directly evident in the original picture.

For the painting of scenes with architectural components Ploem used a program for 3D rendering, that permits structures drawn in 2 dimensions to be visualised in 3D. This enabled him to effectively use perspective in some of his paintings. Whatever the future judgment about the artistic value of Ploem's art form will be, it is clear that he has created novel approaches for the creation of digital graphics on the basis of image analysis, and as such he can be considered as a real pioneer in this field.

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11th Annual Microscope Day at John Jay

Thursday, April 25th, 2013 "New" Building, 524 W. 59th St New York, NY 10019 (Fifth floor, Room 5.67)

Presented by New York Microscopical Society and John Jay College

Please join us for this event, which begins at 11:00 AM and concludes at 4:00 PM. It is an informal event where speakers will give short presentations with ample time in-between to interact with other attendees and speakers.

This year's speakers and their presentation titles (exact times and order of speakers is subject to change):

11:00 Dr. Lawrence Kobilinsky, Chair, Department of Sciences, John Jay College – Opening Remarks

11:15 <u>Stan Petrash</u> – Research Scientist, Henkel Corporation - Advanced Microscopic Imaging: How People, Microscopes and Digital Cameras "See" Small Things

12:00 <u>Gerard Petillo</u> and <u>Anna Tverdovsky</u>, Forensic Firearm Consultants – Using a Microscope to Determine if a Bullet was Fired from a Gun, An Overview of Forensic Firearm Identification **1:00 Lunch**

2:00 <u>Nick Petraco</u>, John Jay College and the CUNY Grad Center – *Microscopical Examination of Household Dust*

3:00 Peter Diaczuk, John Jay College and the CUNY Grad Center - Primers and Gunshot Residues

3:30 Julie Cohen, John Jay College – Yes, Virginia, it is a Science: Using Confocal Microscopy to Compare Toolmarks on Bullets

4:00 Peter Diaczuk, President, New York Microscopical Society - Closing remarks

Exhibitors: Several firms dealing with microscopes and microscope accessories have graciously agreed to set up exhibits of their products for the day.

Refreshments to be served.

This event is free and open to all those interested in microscopy

Photo ID necessary for entry into building



GEORGIA MICROSCOPICAL SOCIETY

April Meeting

The Nikon Small World Exhibit

Saturday April 6th, 2013

Meet by 09:30 am at MVA, Duluth, GA, to go as a group OR meet at Tellus Museum, Cartersville, GA at 10:30 am. MVA group will <u>leave</u> at 09:30 am.



January 19 – April 7, 2013

Photography through the microscope is a technically challenging field, requiring a combination of technical skill and artistic ability. The resulting images are often striking, visually stunning, and sometimes entertaining.

Each year, Nikon holds an international contest to recognize the best photomicrographs, images taken using a microscope,

through its Small World Photomicrography Competition.

The top 20 winning images tour the country in a traveling exhibit – the Nikon Small World Exhibit. Tellus is the only place in Georgia to have the exhibit!

State Science Fair Awards

Walter C. McCrone Award – Julia Abelsky – "Cylindrical Confined Diblock Copolymers and Gold Nanocomposites"

Lucy B. McCrone Award – Jonathan Rong Li – "Facile Solvothermal Synthesis of Silver Nanoparticles and Nanodisks"

Future Meetings

May 22nd - Brian J. Ford, topic to be announced

Any questions, please contact Richard Brown <u>rbrown@mvainc.com</u> or Randy Boltin <u>rboltin@mvainc.com</u>, both at 770-662-8509.

The Georgia Microscopical Society is a Non-profit Organization dedicated to the advancement of all forms of Microscopy. Current Mailing Address is: 3300 Breckinridge Blvd # 400 Duluth, GA 30096-8983 (770) 662-8509

Kirk Czymmek spoke to a joint meeting of the New-York Microscopical Society and the New York Society for Experimental Microscopy on February 6, 2013 at Cornell Weill Medical Center in New York City, about Correlative Microscopy (CM)

Reported by John Scott, Conservator-Analyst, N Y Conservation Fdn

Structure-function relations are always of great and microscopists have long tried for interest. correlatable information by directing different methods to the same specific regions and features of a given sample. Today's CM usually combines data from fluorescence and electron microscopical study of the very same regions within a given specimen. As ever, instrument choice and configuration, and sample preparation are crucial to success, and while changes to samples from preparation and the passage of time complicate the work. mechanical and digital engineering increasingly enable it.

As presented, CM usually involves repeatedly locating and orienting to specific features of a sample, for analysis by different methods such as any number of LM modes, especially photoactivated localization microscopy (PALM), with EM modes and/or with themselves, to reveal distinct characteristics and their relations. Sub-Abbe-limit "high resolution light microscopy," also known as stimulated emission depletion (STED) fluorescence confocal laser microscopy, has been an important tool. The correlative microscopist compares or combines congruent single mode images for richly integrated multimodal information.

Kirk Czymmek has been very active in CM for many years, and his cogent, well paced presentation conveyed clear technical guidance, astute observation and interpretation, and exciting glimpses into quite a few actual and potential applications.

Much CM relates LM information with data from SEM, SEM/EDS, and/or TEM, so method sequencing is key, since sample preparation for one mode can ruin the sample for another. For instance, fluorescent probes quench over time and are lost in in some SEM environments eg with heavy metal coatings, so fluorescence data must be collected first. Hysteresis is another factor, since preparation and analysis processes can distort samples. Czymmek illustrated achievement of repeatability and increasingly prompt close registration of successive images using location mapping and retrieval aids such as traditional diamond scribes, SEM finder grids, mapping

patterns sputtered with finder grids as masks, imbedded microdots, Zeiss' 'Shuttle and Find' sample holder and software system, and more.

Czymmek also described guite a number of other CM enabling technologies and systems: liahtsensitive polymer slide coatings for binding thin sections on slides, Leica's CrvoJane® Tape-Transfer system for cryosectioning, 'en bloc' confocal optical sectioning, green fluorescent protein for gene expression studies, water compatible embedding resins such as LR White for extending gfp fluorescence time en bloc and even in hiah concentrations of alcohol. engineered ascorbate peroxidase genetically encoded for highly resolvable no-light mini-singlet oxygen activated EM labelling of cell structures, and so forth. Czymmek mentioned that CM is not limited to relating LM to EM information, and showed his

relating LM to EM information, and showed his correlation of confocal laser microscopy section images with subsequent LM images of microtomed sections. He Iso illustrated micro computed tomography (mCT) of an immuno-labelled whole flower specimen embedded in LR White resin.

One of Czymmek's ongoing efforts is to develop statistically significant datasets derived from CM analysis of appropriately sized sample sets. Czymmek reports that high resolution light microscopy is becoming "super-resolution microscopy" with direct stochastic optical reconstruction microscopy (dSTORM) enhanced by STED using more precisely targeted immunofluorescent labelling probes such as calcofluor and anti-GFP with Alexa 647 dye, and ever more ultra thin sectioning of Lowicryl resin imbedments.

Clearly CM, and CM for statistically significant datasets, can be extremely labor and time intensive, especially for analyses of many, many closely spaced sections whether en bloc or microtomed. Correlative super-resolution microscopy and creative mechanical and digital engineering are easing the burden, bringing Czymmek's goals for statistically significant results closer. Results like the information-rich images and spellbinding rotatable 3D visualizations Czymmek shared in NYMS' joint meeting with NYSEM make it all worthwhile--and highly effective for experimental microscopy! Czymmek recommends reading a recent introductory article on CM, and for reference, a recent book:

The article:

The power of correlative microscopy: multi-modal, multi-scale, multi-dimensional.<u>Caplan J</u>, <u>Niethammer M</u>, <u>Taylor RM 2nd</u>, <u>Czymmek KJ</u>. <u>Curr Opin Struct Biol.</u> 2011 Oct;21(5):686-93. doi: 10.1016/j.sbi.2011.06.010. Epub 2011 Jul 21.

http://www.ncbi.nlm.nih.gov/pubmed/21782417 the abstract

http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3189301/?report=reader read the article here

The book:

Mueller-Reichert, Thomas, Verkade, Paul. Correlative Light and Electron Microscopy. Elsevier Science & Technology Books, 2012. ISBN-13: 9780124160262, ISBN: 0124160263

~ JS

ISCS SANDFEST 2013







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November 21 through November 24, 2013

Symposium Family Science Night Sand Trading Auctions Annual ISCS Meeting Field Trips Kids' Activities Workshops Banquet Art and Photographs

For More information about SandFest 2013 visit www.iscs.sigmabookstore.com



New York Microscopical Society

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New York Microscopical Society Items For Sale

N.Y.M.S. Microscope Covers

| Item # | Size | Member Price | List Price |
|--------|--------------------------------|--------------|------------|
| MT-003 | Small Microscope or Stereo | \$18.00 | \$20.00 |
| MT-004 | Lab Microscope or Large Stereo | \$23.00 | \$25.00 |
| MT-005 | Large Lab Scope | \$28.00 | \$30.00 |
| MT-009 | Large Lab Scope with Camera | \$31.00 | \$33.00 |
| MT-010 | Universal Scope with Camera | \$36.00 | \$40.00 |
| MT-012 | X-large Scope | \$45.00 | \$50.00 |

N.Y.M.S. Microscopes (see next page for images)

| 185 | Monocular Dissecting Microscope | \$85.00 | \$99.00 |
|---------|---------------------------------------|----------|----------|
| 131 | H.S. Student Microscope | \$190.00 | \$245.00 |
| 131-FLU | H.S. Student Microscope (Fluorescent) | \$200.00 | \$255.00 |
| 125-LED | H.S. Student Microscope (LED) | \$240.00 | \$309.00 |

Other Items

| NYMS Glossary of Microscopical Terms | \$20.00 |
|--------------------------------------|---------|
| NYMS Patch | \$5.00 |
| Microscope Cleaning Kit | \$35.00 |
| NYMS Lapel Pin | \$10.00 |



Model 131: Tungsten Model 131-FLU: Fluorescent





Model 125-LED Cordless

Model 185: 20x



Quartz sand: Polarized-light photomicrograph by Jan Hinsch



Arabo-ascorbic acid recrystallized from hot water and glucomannan, 50x (P1203021) Polarized-light photomicrograph by Mel Pollinger