光與奈米
_從光學顯微到顯“奈”鏡

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Institute of Biophotonics,

National Yang-Ming University, Taipei, Taiwan

http://www.microscopy.tw

The International Year of Light

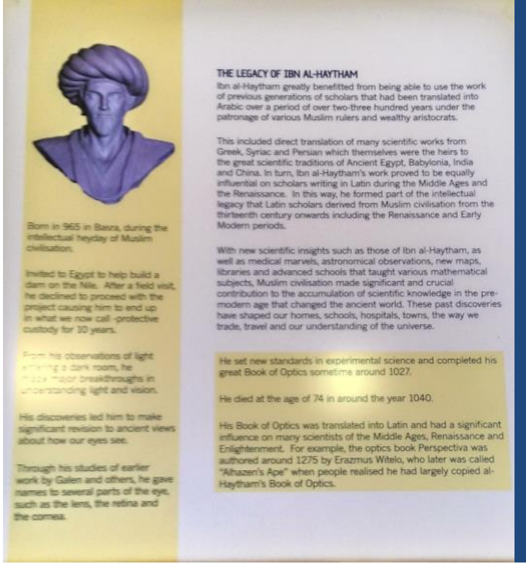
and Light-based Technologies

2015

INTERNATIONAL
YEAR OF LIGHT
2015

Health **Communications** **Economy** **Environment** **Social**

Ibn al-Haytham



http://en.wikipedia.org/wiki/Alhazen

http://global.britannica.com/EBchecked/topic/738111/Ibn-al-Haytham

《墨經》光學八條

- 原文：二臨鑑而立，景到，多而若少，說在寡區。
- 譯文：二人，臨鏡而站，影子相反，若大若小。原因在於鏡面彎曲。
- 原文：鑑位，景一小而易，一大而正，說在中之外內。
- 譯文：鏡子立起，影子小則是鏡位斜，影子大則是鏡位正中，是所謂以鏡位正中為準，分內外的原理。
- 原文：鑑團景一。
- 譯文：無論鏡子大小，影只有一個。
- 原文：景不徙，說在改為。
- 譯文：影子不移，是所謂沒改變的結果。
- 原文：住景二，說在重。
- 譯文：一止而二影，是所謂重複用鏡的結果。
- 原文：景到，在午有端與景長，說在端。
- 譯文：影子顛倒，在光線相交下，焦點與影子造成，是所謂焦點的原理。
- 原文：景迎日，說在摶。
- 譯文：影子在人與太陽之間，是所謂反照的結果。

Declared on 20 Dec 2013

President of the 68th Session of the UN General Assembly formally adopts the resolution for the IYL 2015

"Let there be a Year of Light"

Argentina, Australia, Azerbaijan, Bosnia and Herzegovina, Chile, China, Colombia, Cuba, Dominican Republic, Ecuador, France, Ghana, Guinea, Haiti, Honduras, Israel, Italy, Japan, Mauritius, Mexico, Montenegro, Morocco, Nepal, New Zealand, Nicaragua, Palau, Republic of Korea, Russian Federation, Somalia, Spain, Sri Lanka, Tunisia, Turkey, Ukraine and United States of America

What the UN wants

To promote light technologies for improved quality of life in developed and developing world

To reduce light pollution and energy waste

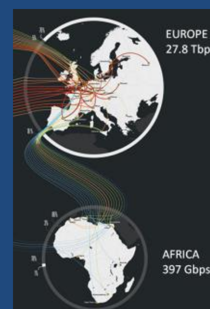
To promote women's empowerment in science

To promote education amongst young people

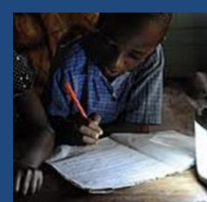
To promote sustainable development

Focus on development

Internet use by population
75% in Europe
16% in Africa

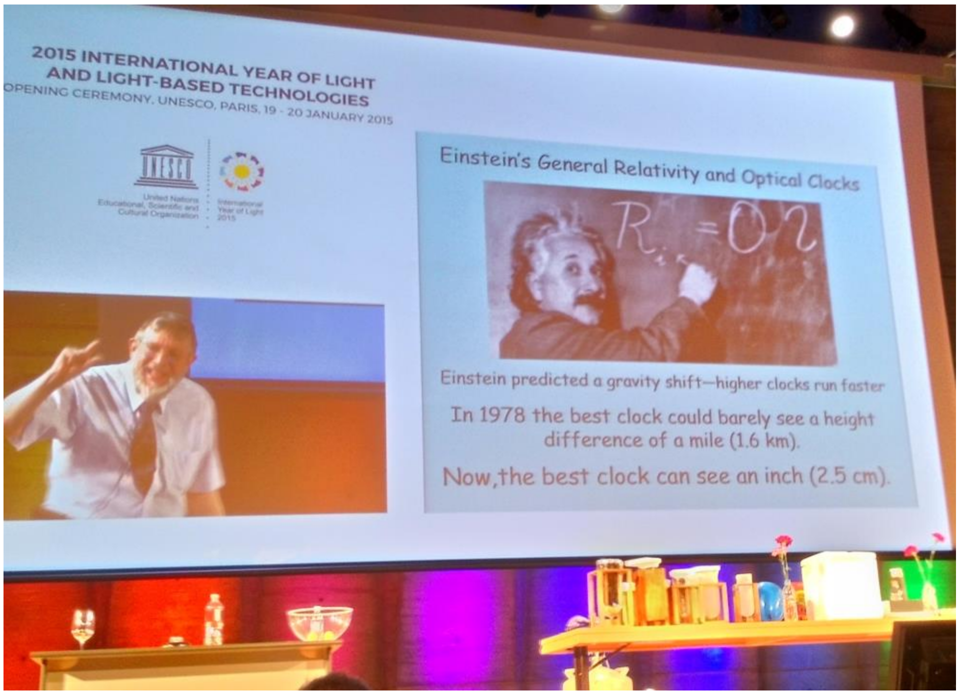


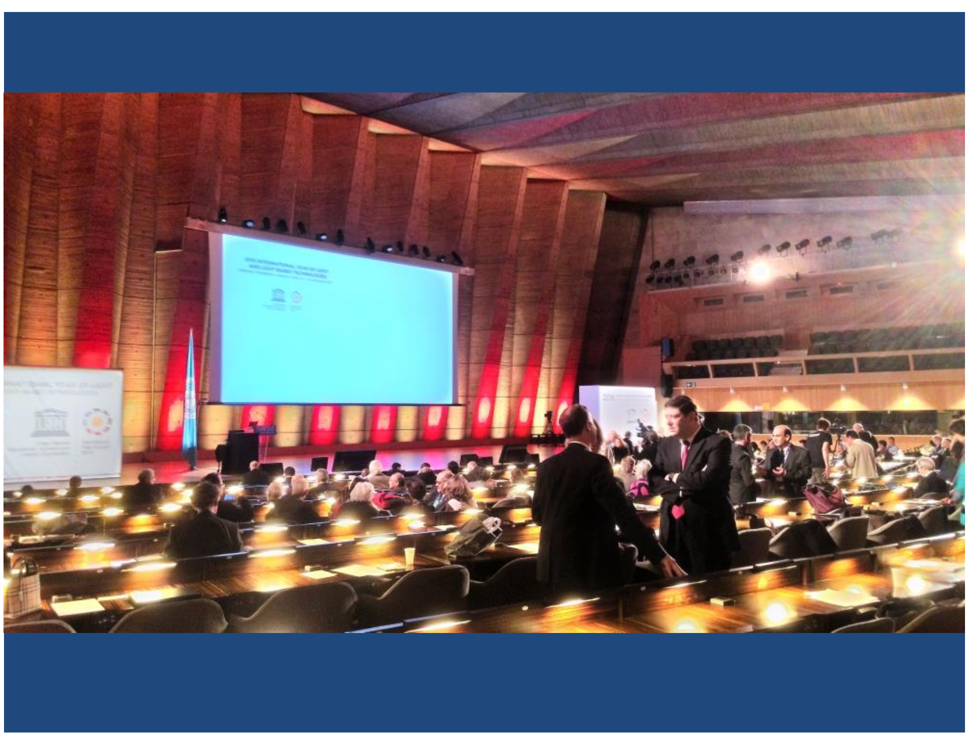
Study after sunset is not possible in many developing countries



UNESCO

United Nations
Educational, Scientific and
Cultural Organization


2015 INTERNATIONAL YEAR OF LIGHT
AND LIGHT-BASED TECHNOLOGIES
OPENING CEREMONY, UNESCO, PARIS, 19 - 20 JANUARY 2015
Einstein's General Relativity and Optical Clocks
Einstein predicted a gravity shift—higher clocks run faster
In 1978 the best clock could barely see a height difference of a mile (1.6 km).
Now, the best clock can see an inch (2.5 cm).



2015 Taiwan Lantern Festival
_ an event claiming 8 million visits

光與奈米
_從光學顯微到顯“奈”鏡

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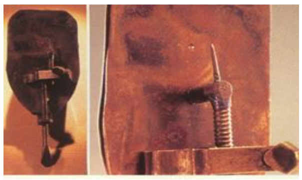
Outline

- Some history
- Current and coming developments
  - Aiming for nanoscopy
- The challenges
- Developments in my labs

Starting in the 16th century from a magnifier

Leeuwenhoek's single-lens microscope



Pacini Compound Microscope (circa 1845)

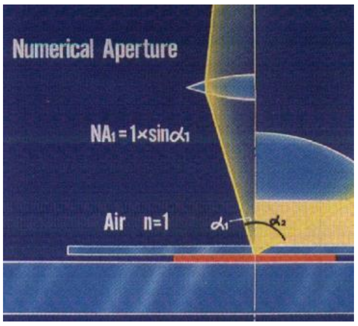
The 19th century saw dramatic progress in the development of the microscope, thanks to the contributions of such great minds as Carl Zeiss, who devoted significant effort to the manufacture of microscopes, Ernst Abbe, who carried out a theoretical study of optical principles, and Otto Schott, who conducted research on optical glass.

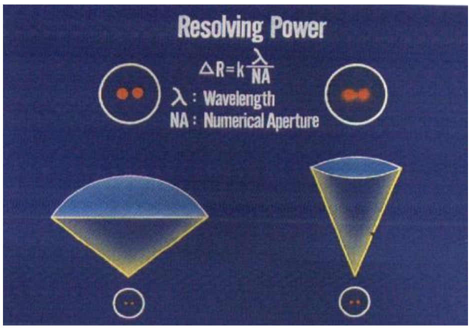
Carl Friedrich Zeiss (1816-1888)

Ernst Abbe (1840-1905)

Otto Schott (1851-1935)

Diffraction Limits





One of the Microscopy Challenges: Elucidating Molecular Dynamics in Biology without Destroying the Molecules

Diffract Limit: **A barrier originated from the Uncertainty Principle**

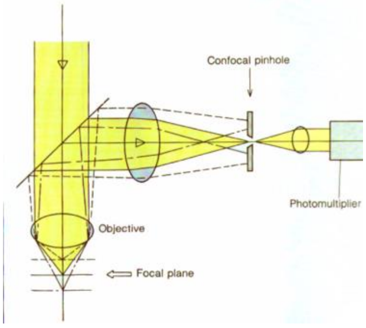
Ernst Abbe memorial at Jena, Germany:

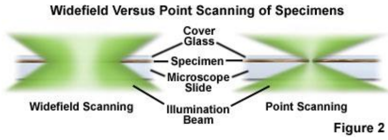
$$\Delta x \Delta p > \frac{\hbar}{2}$$
$$\Delta E \Delta t > \frac{\hbar}{2}$$

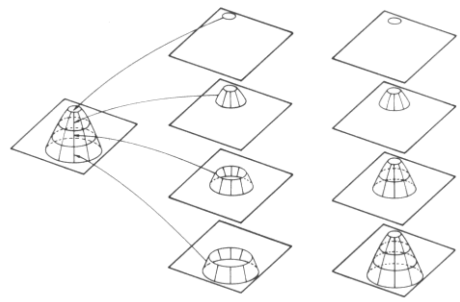
Diffraction Limit: $\Delta R \sim \lambda / NA$

Difficult for life science to be alive at $\lambda < 450$ nm

Confocal Microscopy







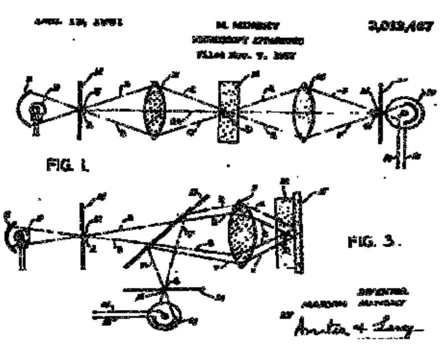
M. Minsky, US Patent 3013467, Dec. 19, 1961

T. Wilson and C.J.R. Sheppard, Theory and Practice of Scanning Optical Microscopy, (Academic, London, 1984)

Peter Török, P. Varga, Z. Laczik and G. R. Booker, Electromagnetic diffraction of light focused through a planar interface between materials of mismatched refractive indices:
an integral representation, J. Opt. Soc. Am. A, 12, 325(1995)

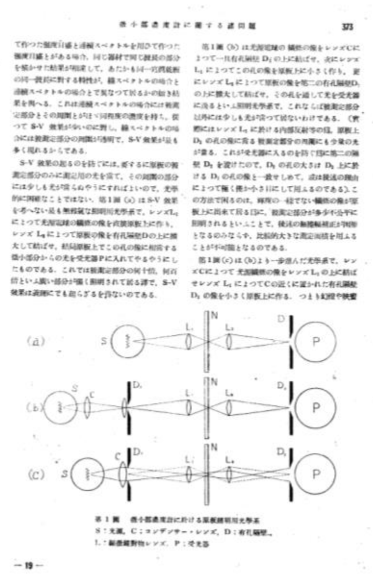
Confocal Microscopy

Marvin Minsky 1957

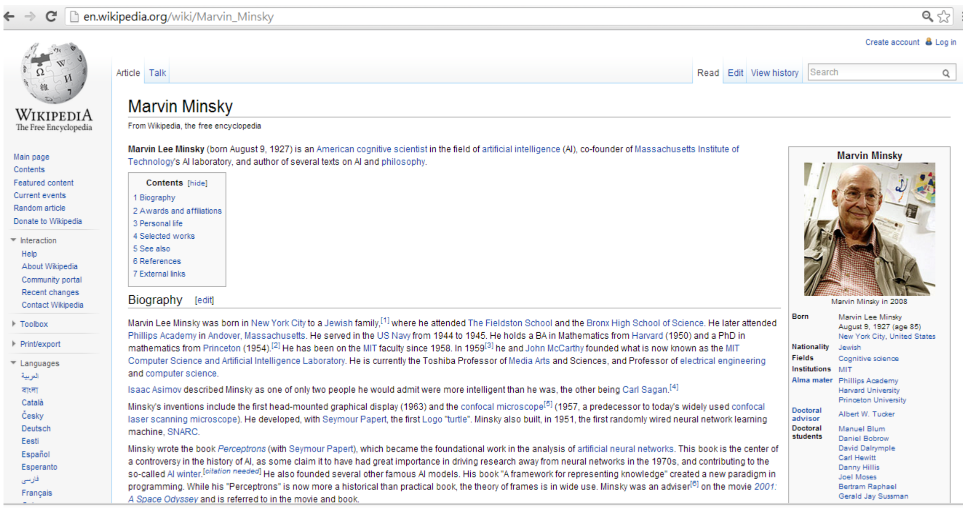


Z Koana 1942

Journal of the Illuminating Engineering Institute of Japan Vol. 26(1942) No. 8



Marvin Minsky; He is still alive !


en.wikipedia.org/wiki/Marvin_Minsky

Marvin Minsky

From Wikipedia, the free encyclopedia

Marvin Lee Minsky (born August 9, 1927) is an American cognitive scientist in the field of artificial intelligence (AI), co-founder of Massachusetts Institute of Technology's AI laboratory, and author of several texts on AI and philosophy.

Contents [hide]
1 Biography
2 Awards and affiliations
3 Personal life
4 Selected works
5 See also
6 References
7 External links

Biography [edit]

Marvin Lee Minsky was born in New York City to a Jewish family,[1] where he attended The Fieldston School and the Bronx High School of Science. He later attended Phillips Academy in Andover, Massachusetts. He served in the US Navy from 1944 to 1945. He holds a BA in Mathematics from Harvard (1950) and a PhD in mathematics from Princeton (1954).[2] He has been on the MIT faculty since 1958. In 1959[3] he and John McCarthy founded what is now known as the MIT Computer Science and Artificial Intelligence Laboratory. He is currently the Toshiba Professor of Media Arts and Sciences, and Professor of electrical engineering and computer science.

Isaac Asimov described Minsky as one of only two people he would admit were more intelligent than he was, the other being Carl Sagan.[4]

Minsky's inventions include the first head-mounted graphical display (1963) and the confocal microscope[5] (1957, a predecessor to today's widely used confocal laser scanning microscope). He developed, with Seymour Papert, the first Logo "turtle". Minsky also built, in 1951, the first randomly wired neural network learning machine, SNARC.

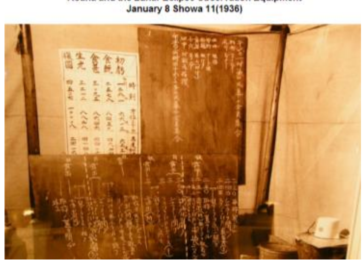
Minsky wrote the book *Perceptrons* (with Seymour Papert), which became the foundational work in the analysis of artificial neural networks. This book is the center of a controversy in the history of AI, as some claim it to have had great importance in driving research away from neural networks in the 1970s, and contributing to the so-called AI winter.[citation needed] He also founded several other famous AI models. His book "A framework for representing knowledge" created a new paradigm in programming. While his "Perceptrons" is now more a historical than practical book, the theory of frames is in wide use. Minsky was an adviser[6] on the movie *2001: A Space Odyssey* and is referred to in the movie and book.

Marvin Minsky	
Marvin Minsky in 2008	
Born	Marvin Lee Minsky August 9, 1927 (age 85) New York City, United States
Nationality	Jewish
Fields	Cognitive science
Institutions	MIT
Alma mater	Phillips Academy Harvard University Princeton University
Doctoral advisor	Albert W. Tucker
Doctoral students	Manuel Blum Daniel Bobrow David Dalrymple Carl Hewitt Danny Hillis Joel Moses Bertram Raphael Gerald Jay Sussman

http://en.wikipedia.org/wiki/Marvin_Minsky

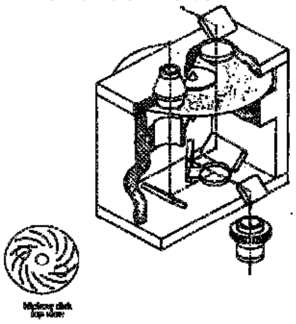
Koana, Zuen

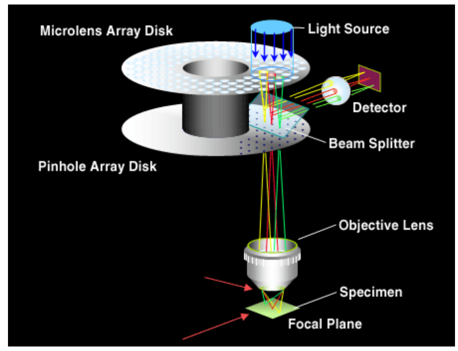

Koana and the Lunar Eclipse Observation Equipment
January 8 Showa 11(1936)



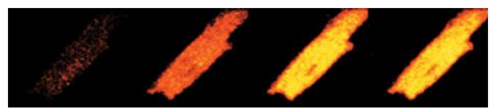
http://museum.c.u-tokyo.ac.jp/2009.html#koana
http://akiroom.com/redbook-e/koana/koana10.html

Petrán 1968, Video Rate Confocal



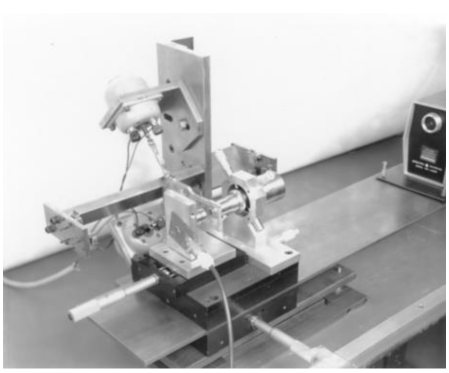


Post-electrostimulation changes in Ca^{2+} in mouse ventricular cardiac muscle cell labeled with Fluo-3 , images taken at 4 ms intervals



16ms 20ms 24ms 28ms

Oxford microscope, 1975

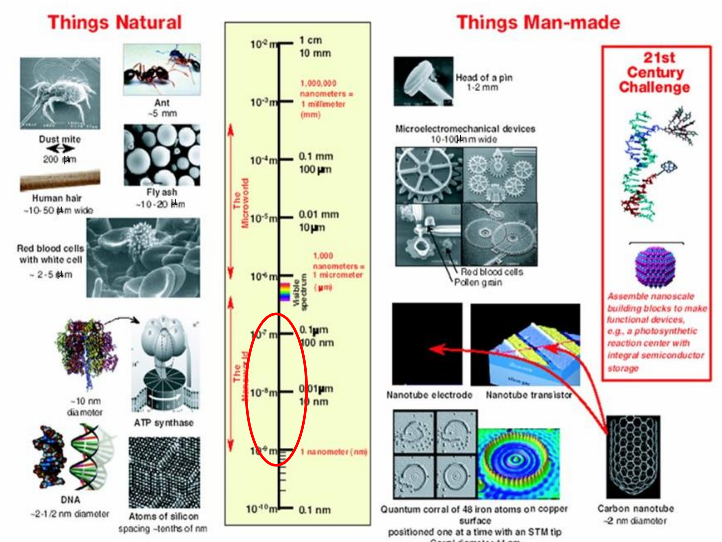


Amar Choudhury, Colin Sheppard, Pete Hale and Rudi Kompfner
Oxford, Summer 1976

The "Discovery" of Nano-World

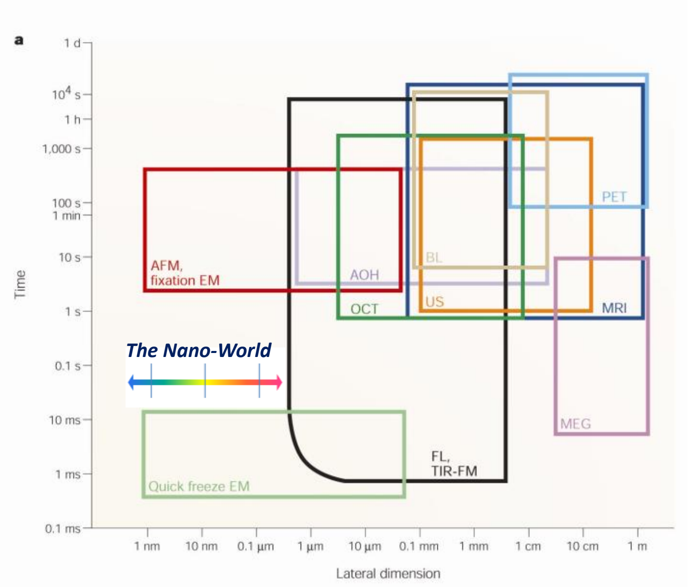
- Introduction and current status
- The less charted territory (challenges)
- The working mechanisms and building blocks
  - Are there "coherent forces" in this range, such as Cooper pairs, plasmonics/polaritons/excitons, or nano-quarsiparticles, ..etc.?
  - Could carbon nanotube, nano-diamonds, or graphene (all carbon) provide the new Legos?
- The tools
  - New concepts and novel technologies
  - Switching of molecules, supercontinuum light sources, CMOS camera, high speed electronics, data center grade computation (modeling high energy physics)
- Prospects and Future Investment, the combination of
  - Super-resolution microscopy, the essence is "switching".
  - High throughput and high content imaging, data center class analysis

Nano-Science & Nano-Technology



http://www.terasemjournals.org/GNJournal/GN0301/ck3lg.jpg

Imagining imaging's future



AFM: atomic-force microscopy

AOH: all-optical histology

BL: bioluminescence

FL: fluorescence microscopy at visible wavelengths

MEG: magnetoencephalography

MRI: functional magnetic resonance imaging

OCT: optical coherence tomography

PET: positron emission Tomography

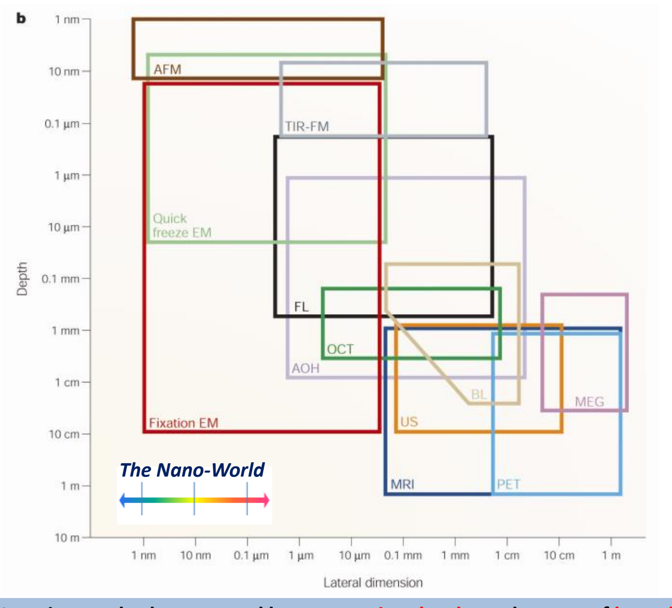
TIR-FM: total internal reflection fluorescence microscopy

US: ultrasound.

Imaging methods compared by their timescales and ranges of lateral dimensions

Roger Tsiem, Imagining imaging's future, Nature Reviews of Molecular Cell Biology 2003 Sep, ss16 – ss21

Imagining imaging's future



AFM: atomic-force microscopy

AOH: all-optical histology

BL: bioluminescence

FL: fluorescence microscopy at visible wavelengths

MEG: magnetoencephalography

MRI: functional magnetic resonance imaging

OCT: optical coherence tomography

PET: positron emission Tomography

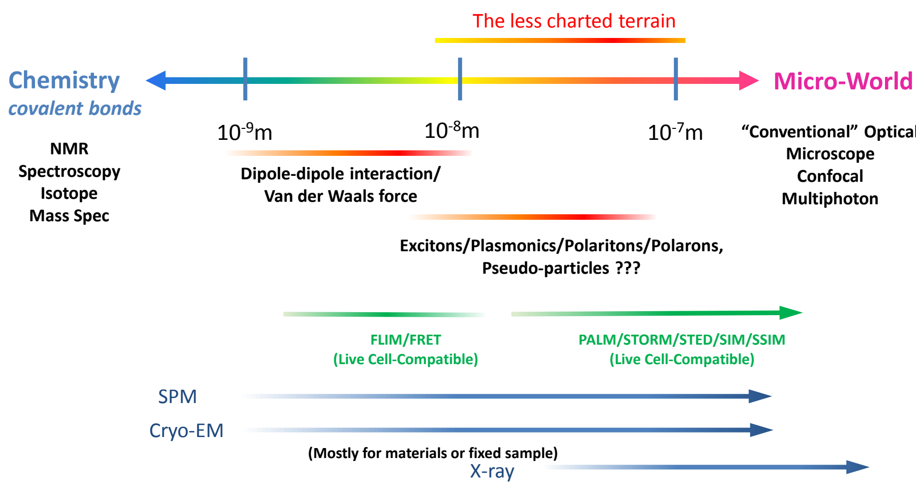
TIR-FM: total internal reflection fluorescence microscopy

US: ultrasound.

Imaging methods compared by penetration depths and ranges of lateral dimensions

Rogen Tsien, Imagining imaging's future, Nature Reviews of Molecular Cell Biology 2003 Sep, ss16 –

The Nano-World _tools and considerations



The Nobel Prize in Chemistry 2014
Eric Betzig, Stefan W. Hell, William E. Moerner

Share this: 1.4K

The Nobel Prize in Chemistry 2014

Photo: A. Mahmoud
Eric Betzig
Prize share: 1/3

Photo: A. Mahmoud
Stefan W. Hell
Prize share: 1/3

Photo: A. Mahmoud
William E. Moerner
Prize share: 1/3

The Nobel Prize in Chemistry 2014 was awarded jointly to Eric Betzig, Stefan W. Hell and William E. Moerner *"for the development of super-resolved fluorescence microscopy"*.

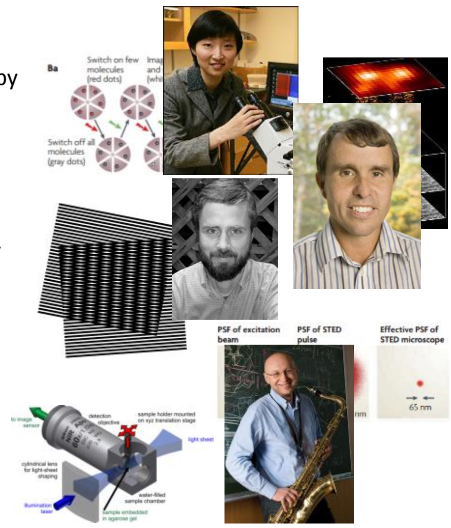
Leading Developments Worldwide

Nano- and Multidimensional-scopy

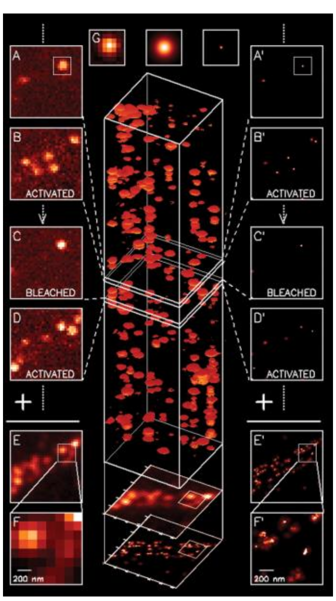
__Beyond the Diffraction Limit

From micro to nano: 200 nm → 100 nm → 30 nm (→ 10 nm)

1. **Single fluorophore Localization microscopy**
   (a) Stochastic optical reconstruction microscopy (STORM), 2006, Xiaowei Zhuang
   (b) Photoactivated localization microscopy (PALM), 2006, by Eric Betzig, Harald Hess, and Samuel Hess.
   (c) SPDM C. Cremer, 2008 or earlier.
2. **Saturated structured illumination microscopy (SSIM),**
   Mats Gustafsson
3. **Stimulated emission depletion (STED),**
   Stefan Hell
4. **Optical Sectioning Microscopy,**
   Ernst Stelzer



Optical Localization Microscopy



1. Single molecule (or fluorophore) activation
2. Single molecule detection
3. Centralizing
4. Image reconstruction

https://www.youtube.com/watch?v=-ioAnvfkJr8

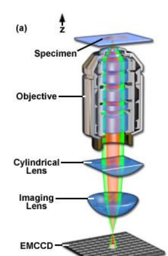
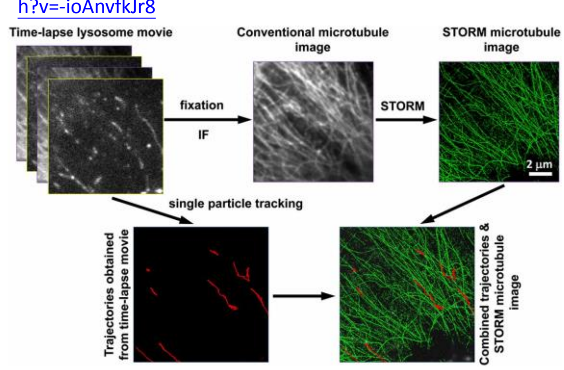


Figure 9

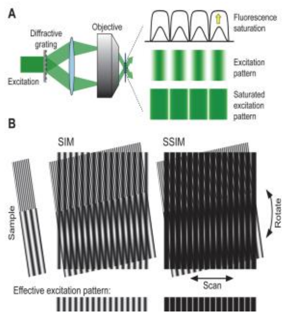


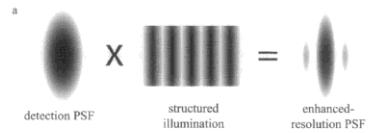
Bálint Š et al. PNAS 2013;110:3375-3380

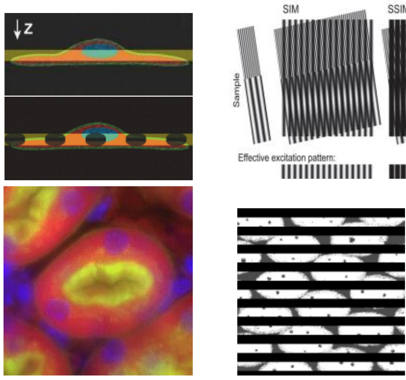
A moiré pattern and structured illumination, SIM and SSIM

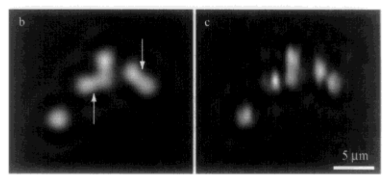
The Late Mats Gustaffson

http://www.bioengineering.ucsf.edu/faculty-mats_gustafsson.vp.html

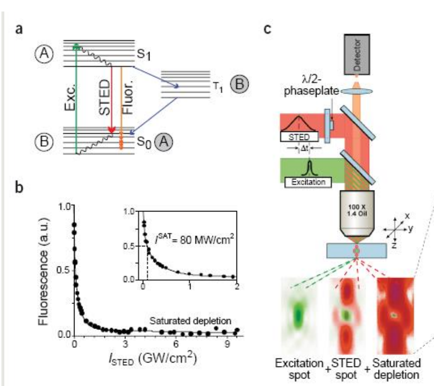




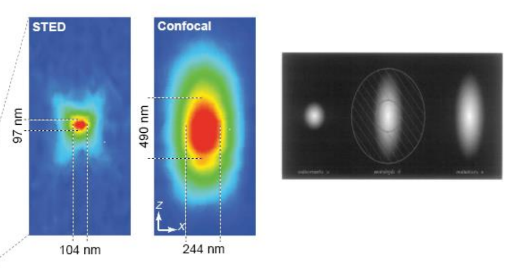


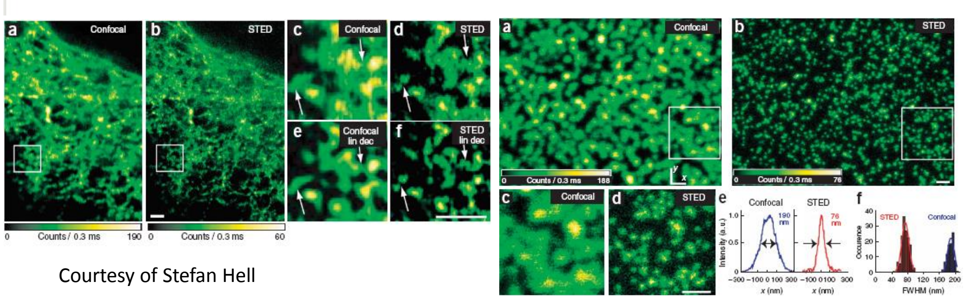


Courtesy of Mats Gustaffson



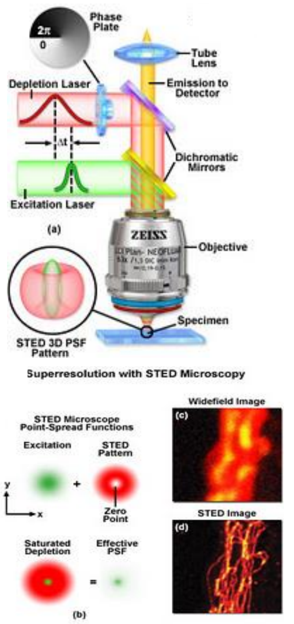
NATURE BIOTECHNOLOGY
V21 # 11 NOVEMBER 2003





Courtesy of Stefan Hell

STED, STimulated Emission Depletion Microscopy



Courtesy of Stefan Hell

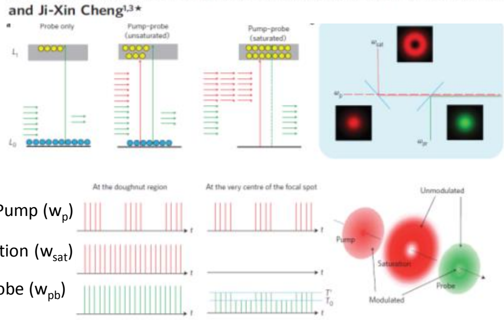
nature photonics

LETTERS

PUBLISHED ONLINE: 28 APRIL 2013 | DOI: 10.1038/NPHOTON.2013.97

Far-field imaging of non-fluorescent species with subdiffraction resolution

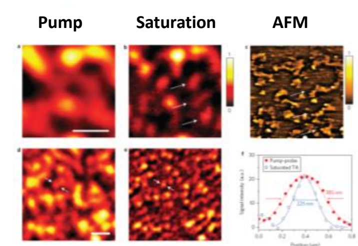
Pu Wang[1†], Mikhail N. Slipchenko[1†], James Mitchell[2], Chen Yang[3], Eric O. Potma[4], Xianfan Xu[2] and Ji-Xin Cheng[1,3]*



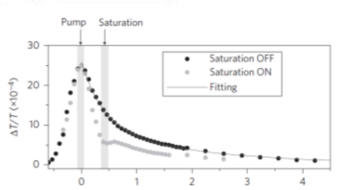
Pump (w_p)

Saturation (w_{sat})

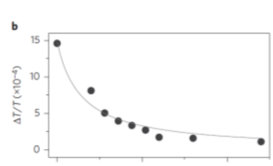
Probe (w_{pb})



Subdiffraction-limited imaging of graphite nanoplatelets

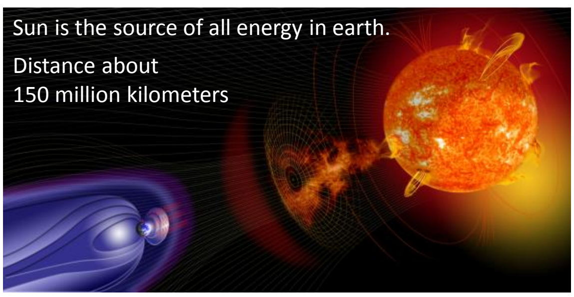


Pump-probe delay (ps)

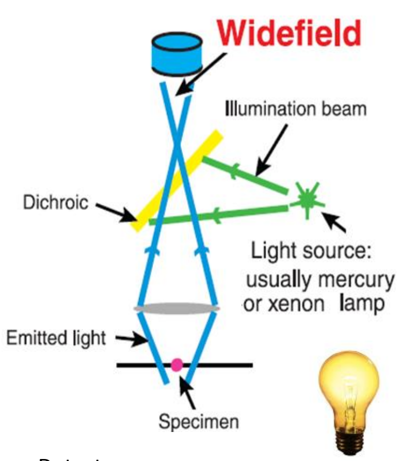


Power density (MW/cm²)

Illuminating the bio-samples properly

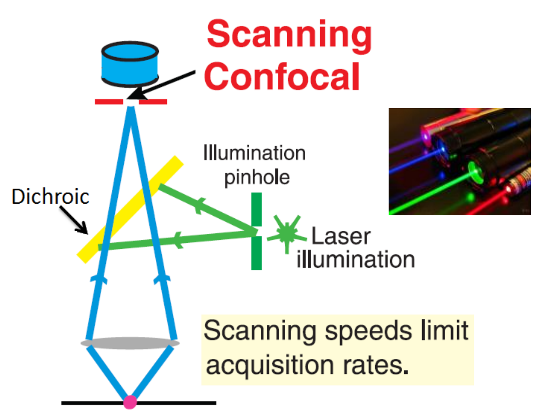


The average intensity at top of the atmosphere 1,360 watts/m^2.
At surface is 1030 watts/m^2
(1.03 X10^{-6}mili-watt/μm^2)
By NASA satellite missions.



Detector- eye or a camera

200 nW incident or
320 W/cm^2 peak irradiance.
Which is comparable to that of Sun's irradiation to our body.

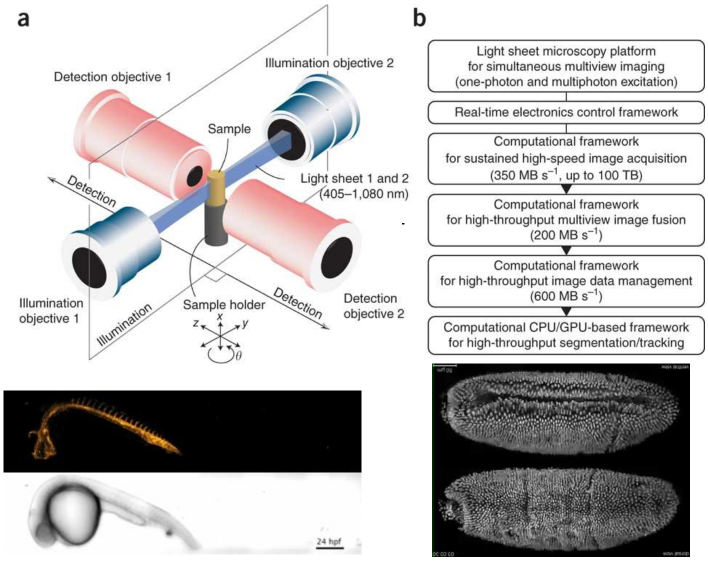


Point-like illumination and point-like detection results in a focal spot
10^5 W/cm^2 for 400 nm ex. 20 X .75 N.A. obj.

David J. Stephens, *et al.* *Science* ***300, 82 (2003)***

Light Sheet Microscopy (SPIM)

Photon budget (or statistics) is the key.



Correlative fluorescence and electron microscopy

Confocal STED EM Correlative STED & EM

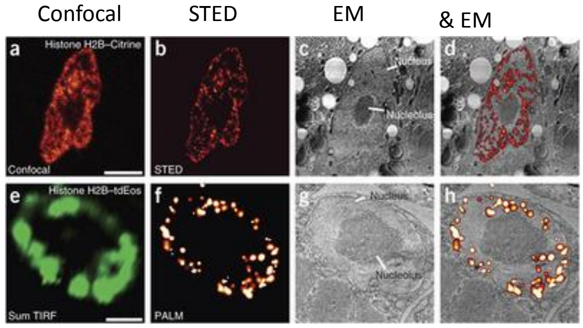
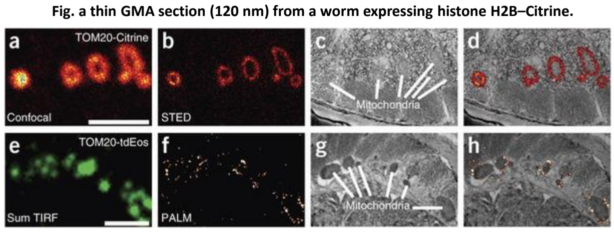


Fig. TOM20 fusion proteins.

Fig. a thin GMA section (120 nm) from a worm expressing histone H2B–Citrine.

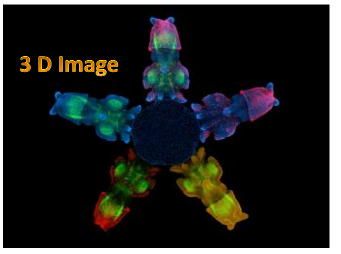


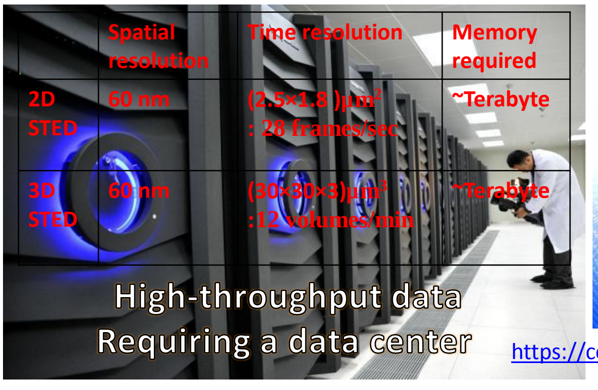
Nature Methods 8, 80–84 (2011)

http://www.nature.com/nmeth/journal/v8/n1/full/nmeth.1537.html

The New Paradigm

Massive data processing





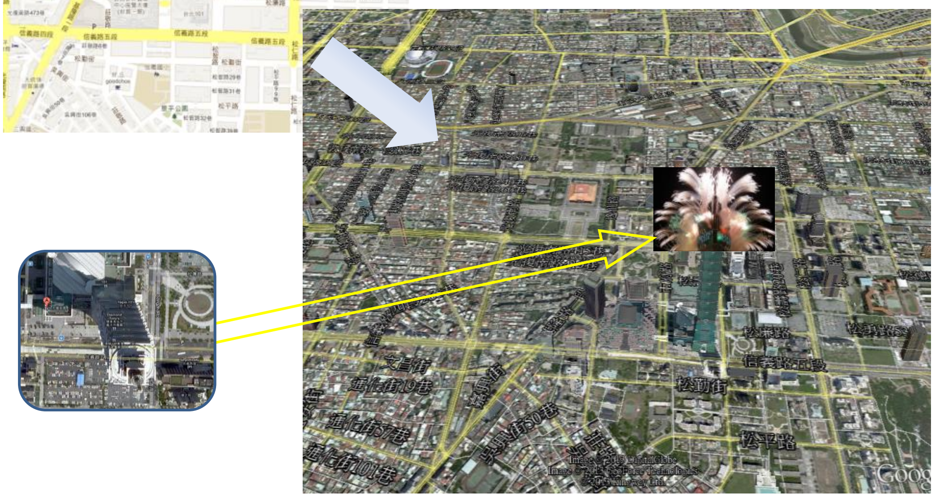
	Spatial resolution	Time resolution	Memory required
2D STED	60 nm	$(2.5\times1.8)\mu m^2$: 28 frames/sec	~Terabyte
3D STED	60 nm	$(30\times30\times3)\mu m^3$:12 volumes/min	~Terabyte

https://code.google.com/p/monashmie/

Summary

1. Optical instruments are most versatile and non-invasive in probing the nano-world.
2. Novel concepts and new tools have enabled unprecedented opportunities. Three-dimensional imaging with an optical resolution as high as ~20 nm in the lateral direction and 40–50 nm in axial dimension has been achieved routinely.
3. As resolution improves beyond diffraction limit, the amount of data explodes accordingly. A new paradigm is reached, data center class processing seems imperative.
4. In addition to better imaging capacity, new fluorescent probes that are brighter, more photostable and switchable fluorophores that have high on-off contrast and fast switching rate are also crucial.

Google Earth-like Visualization



Thank You for Your Attention !