

# Super-resolution and looking deep in a volume

*a melting pot for research groups and small enterprize*

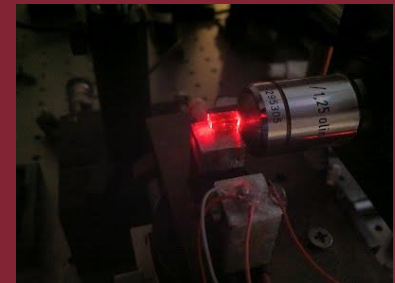
**Eugenio DelRe**

Physics Department, University of Rome “La Sapienza”, Italy



**SAPIENZA**  
UNIVERSITÀ DI ROMA

Frascati – 13th of October 2015



### Group

Giuseppe Di Domenico, PhD Student  
Single-photon sources – F005

Fabrizio Di Mei, PhD Student  
Scale-free optics and giant response – F008

Davide Pietrangeli, PhD Student  
Solitons in gratings - F008  
Digital Materials -F008  
Rogue Waves – F008

Salvatore Silvestri, PhD Student  
Super-resolved imaging – IIT

Piergiorgio Caramazza, Laurea Degree Student  
Mariano Flammini, Laurea Degree Student  
Mario Ferraro, Laurea Degree Student

### Projects

Prin2012 – Rogue Waves  
FIRB PHOCOS – Spatial Solitons  
ISC – Structured Illumination Imaging

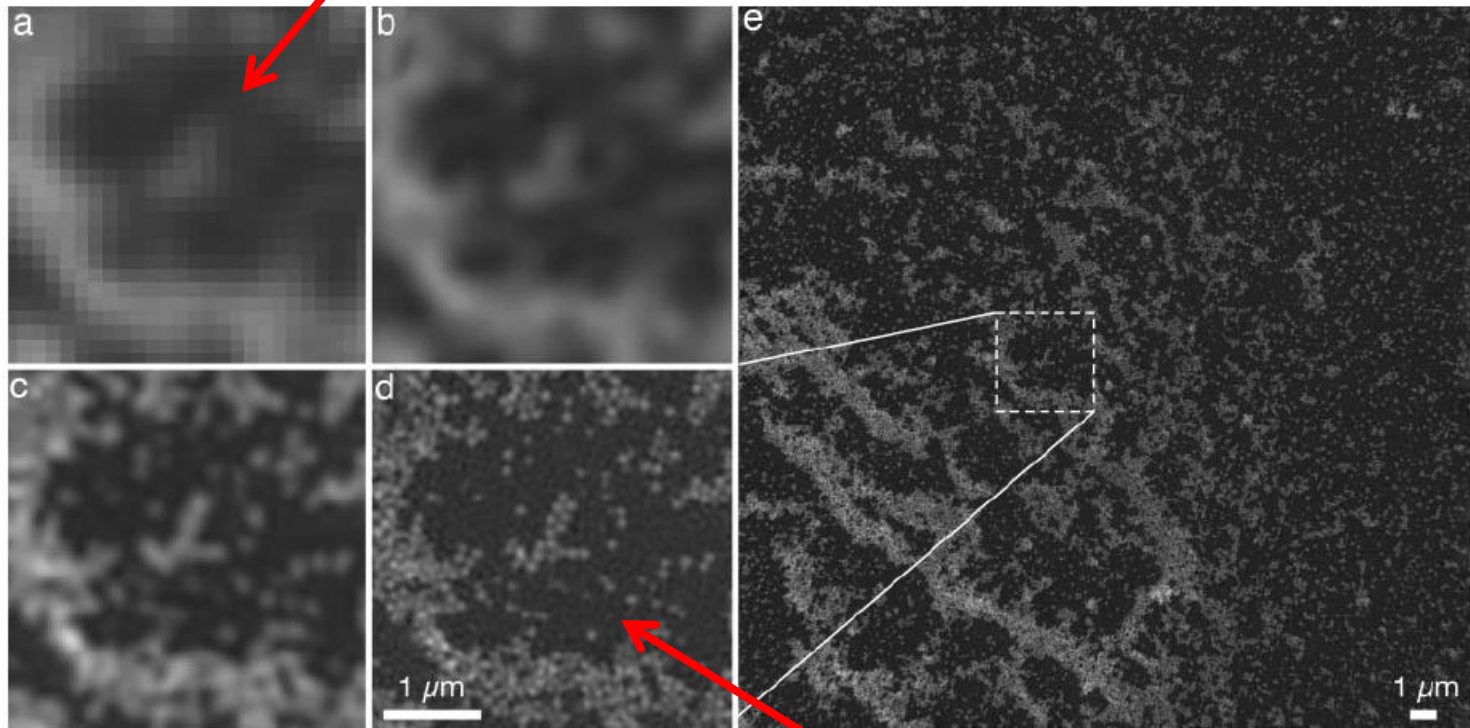


**Fundamental limits to far-field  
(at a distance) imaging using light**



# The Shackles of Diffraction

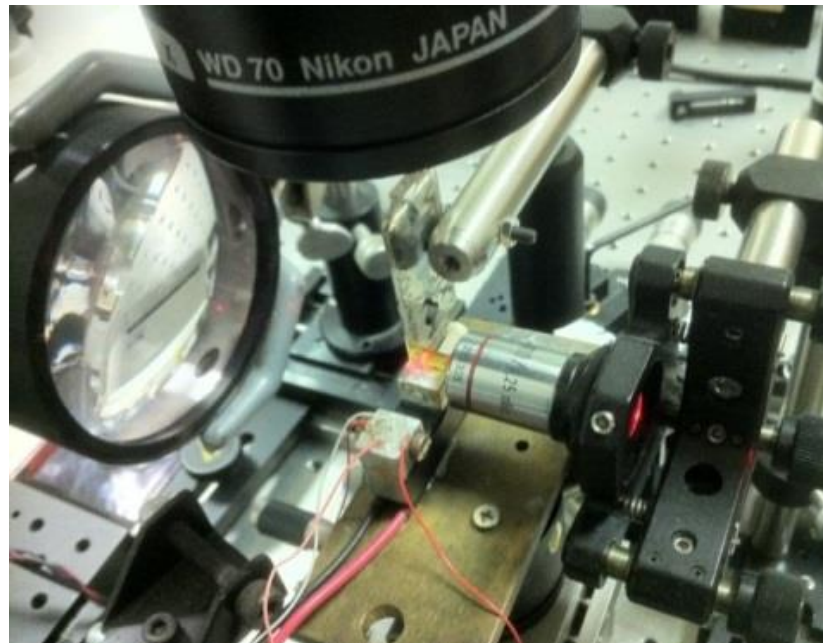
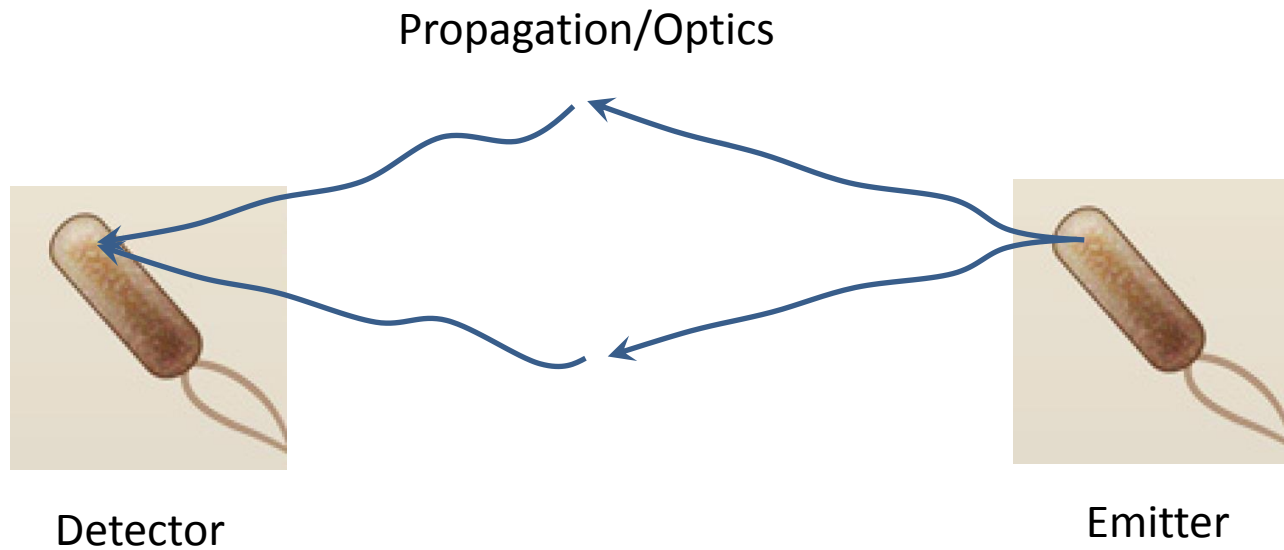
Diffraction-limited (details below  $\sim 200$  nm are **lost**)



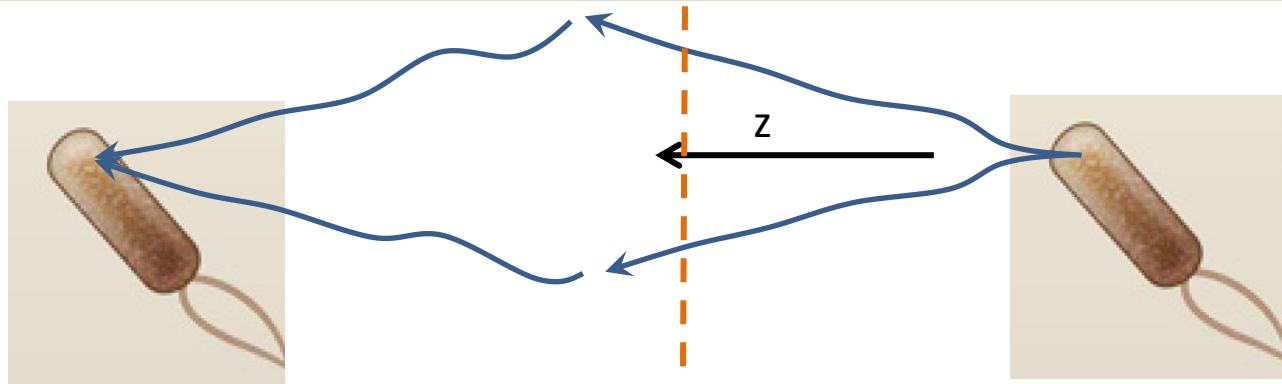
**Fig. 6.** A field of 50-nm fluorescent beads, imaged by conventional microscopy (a), conventional microscopy plus filtering (b), linear structured illumination (c), and saturated structured illumination using illumination pulses with  $5.3 \text{ mJ/cm}^2$  energy density, taking into account three harmonic orders in the processing (d). Because no scanning is necessary, a wide field can be imaged simultaneously.

Super-resolution to below 50 nm (through  
**nonlinear microscopy**)

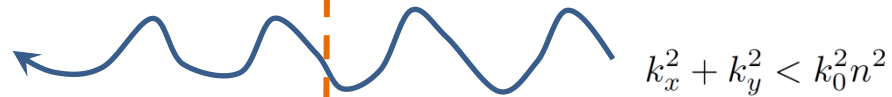
# Imaging using light



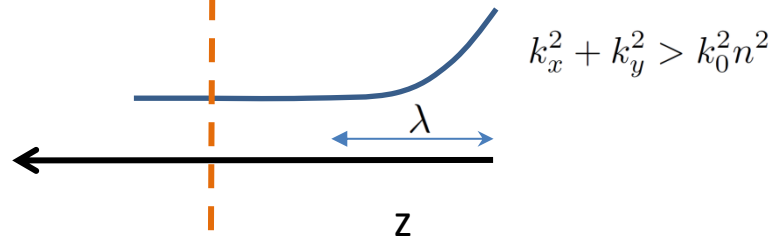
# Imaging using light



Propagating waves

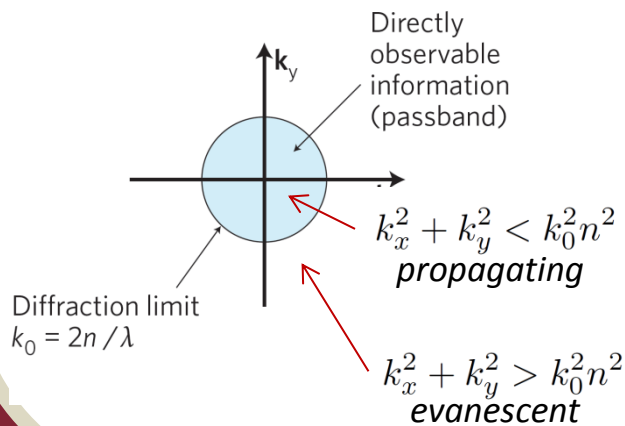


Evanescent waves

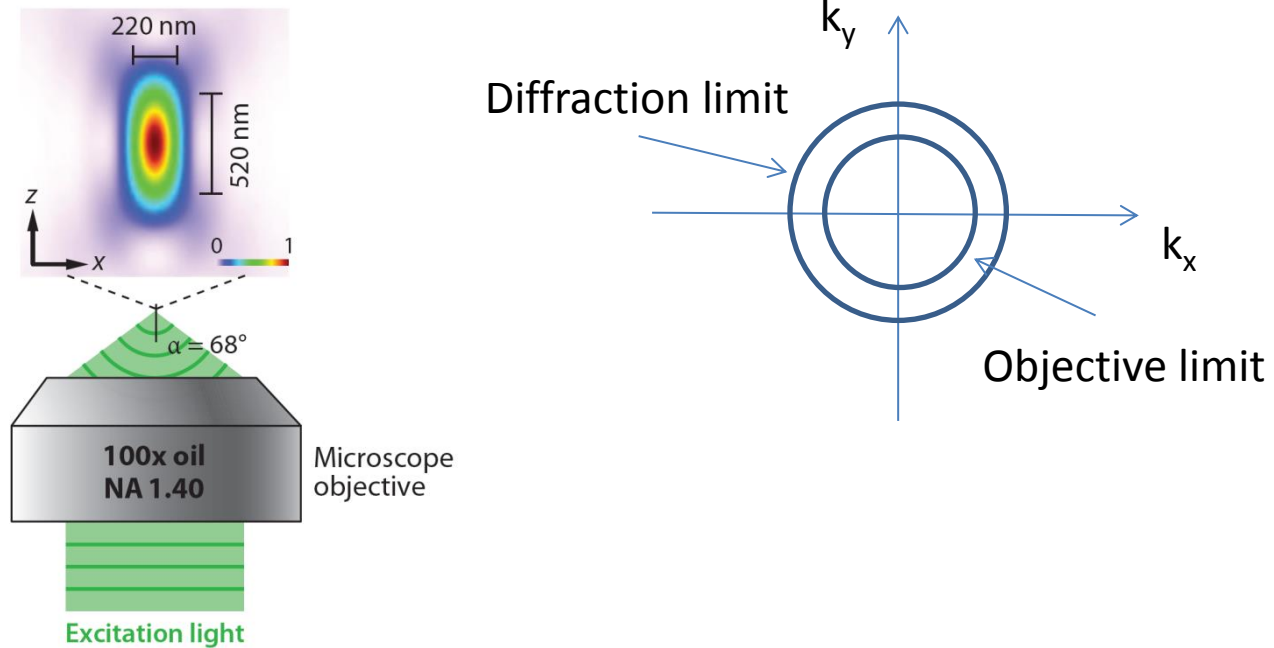


Far-field

Near-field



# Imaging using light



B. Huang, M. Bates, and X. Zhuang, *Annu. Rev. Biochem.* 2009. 78:993–1016 (2009)



Numerical Aperture:  $NA = n \sin \theta$

Collected light

Emitted light

Light emitter

objects

$E(r)$

images

$D(r)$

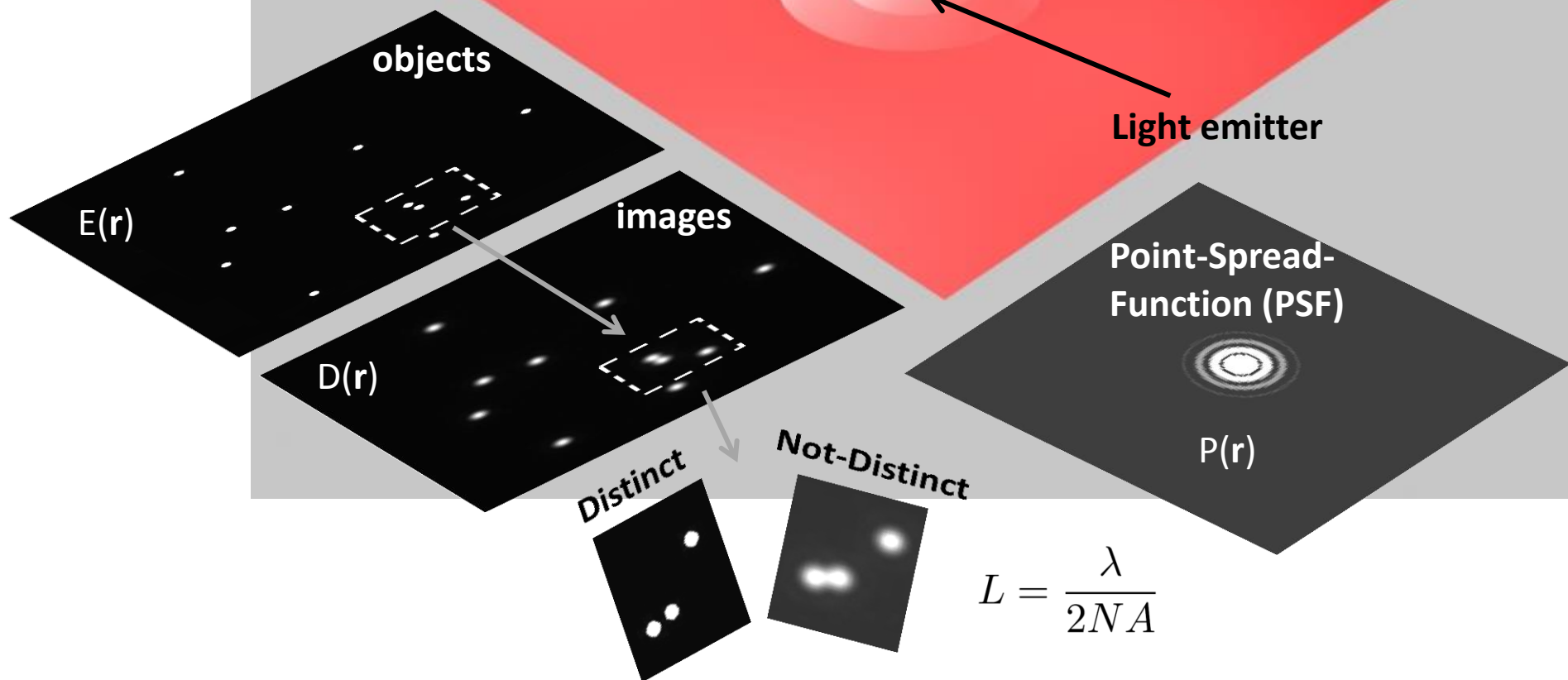
Point-Spread-Function (PSF)

$P(r)$

Distinct

Not-Distinct

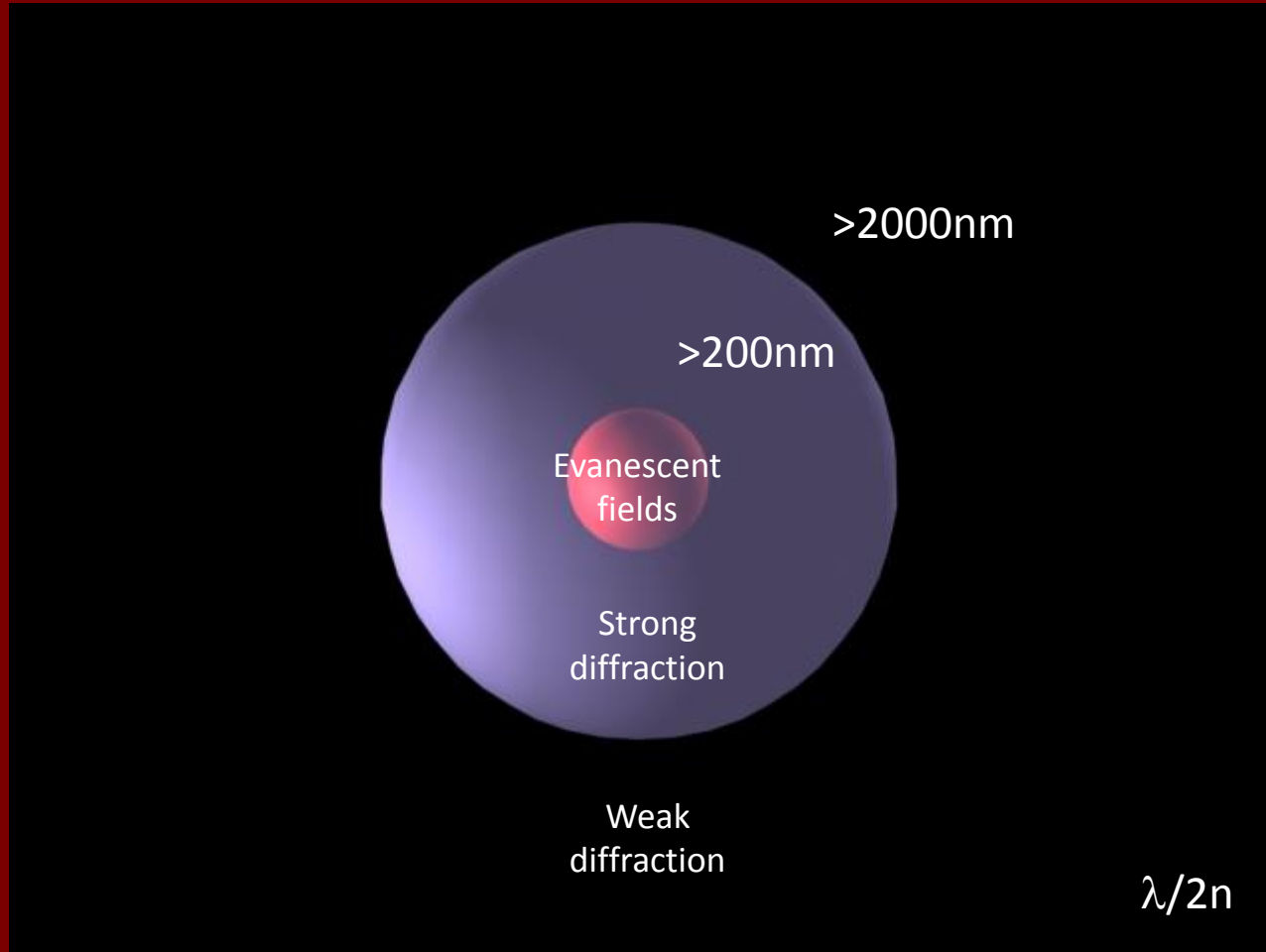
$$L = \frac{\lambda}{2NA}$$





# Our “vision” of the world

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# Science slips beyond conventional optics...

Biology  
Mechanics  
Electronics

Evanescent  
fields

Strong  
diffraction

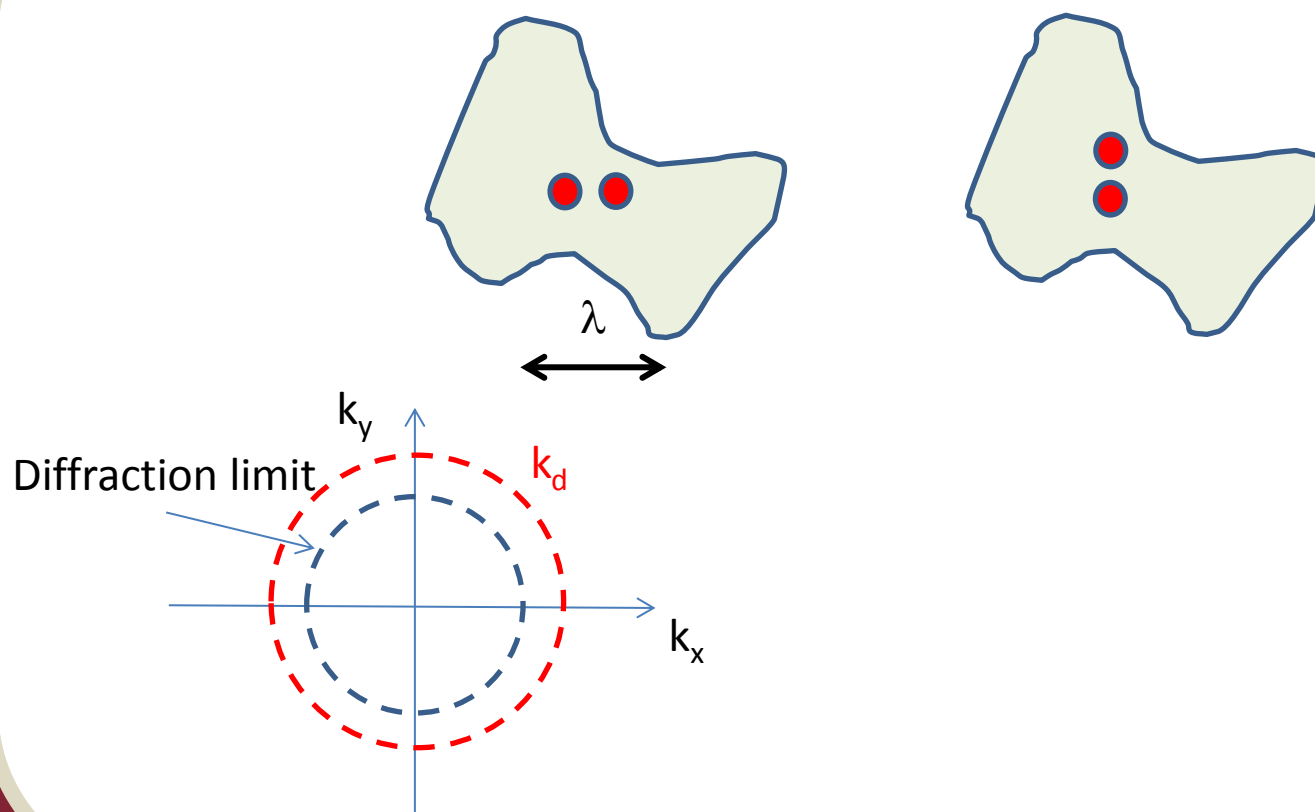
Weak  
diffraction

$$\lambda/2n$$

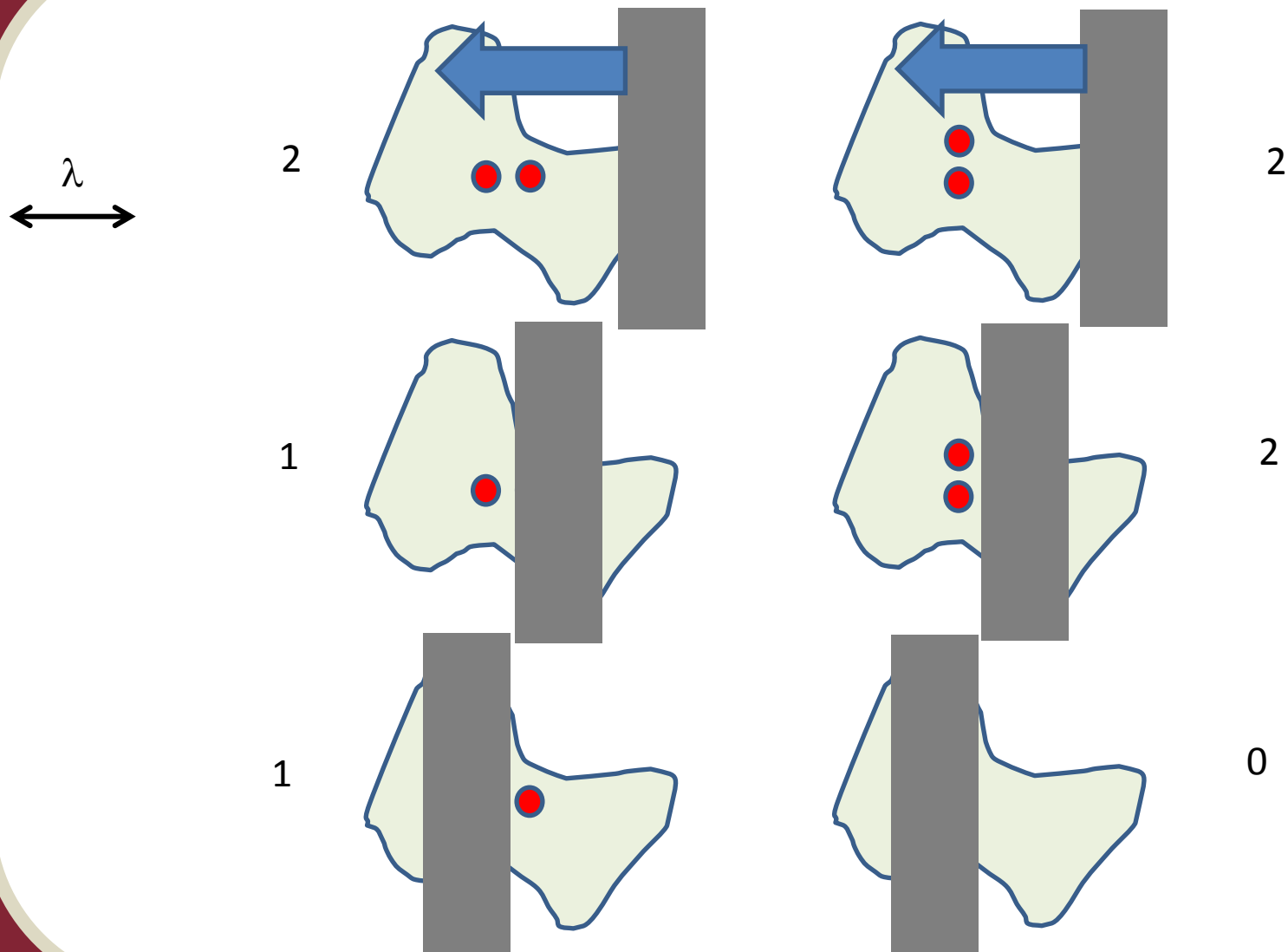
**Seeing things below the  
diffraction/aperture limit at a distance**



# Combining imaging with prior knowledge



# Combining imaging with prior knowledge



# Combining imaging with «prior» knowledge – Super-resolution!

But how?

This is a *multiphoton*, i.e., **NONLINEAR**, process!

Not a single image, but a cumulative effect of many images.

One photon –  $\lambda$

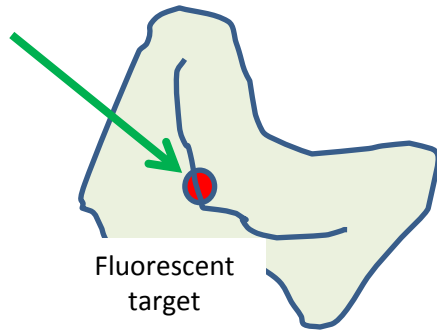
Two photon –  $\lambda/2$

....

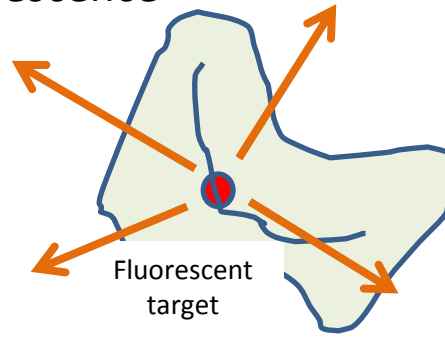


# Imaging through fluorescence

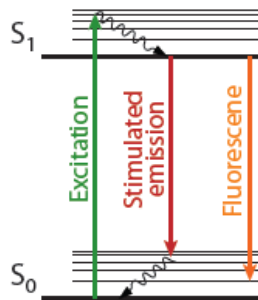
Excitation



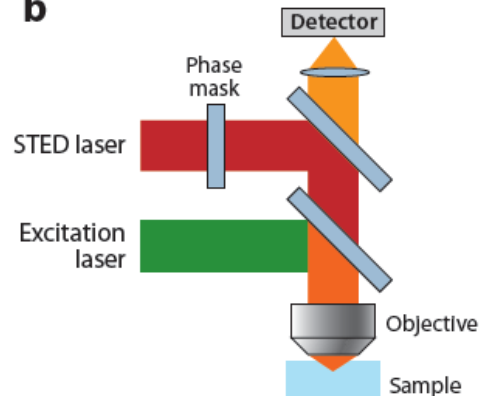
Fluorescence



**a**

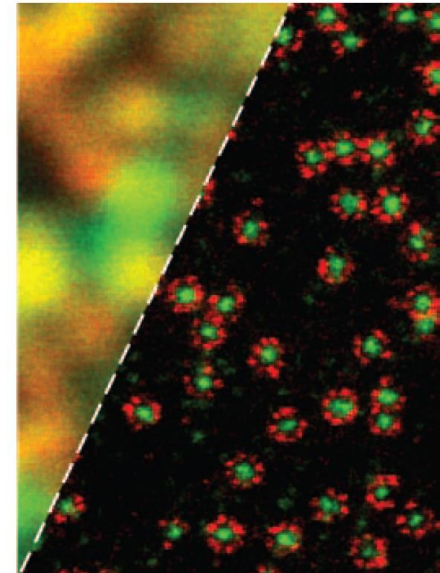


**b**



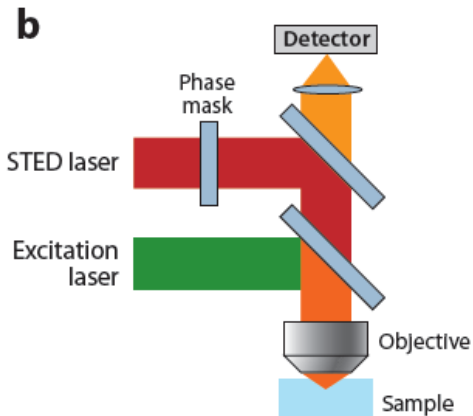
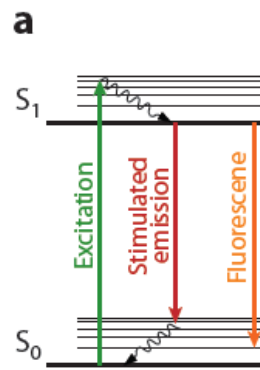
Confocal

STED

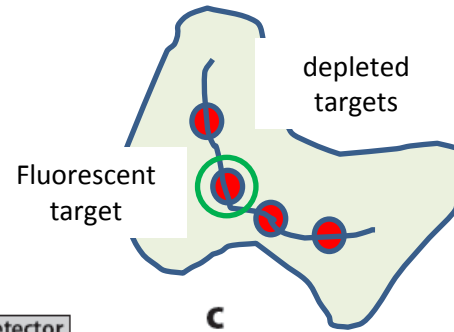


# Super-resolution imaging

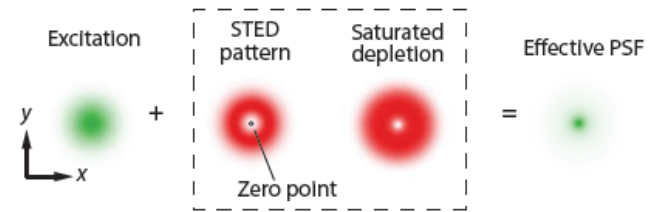
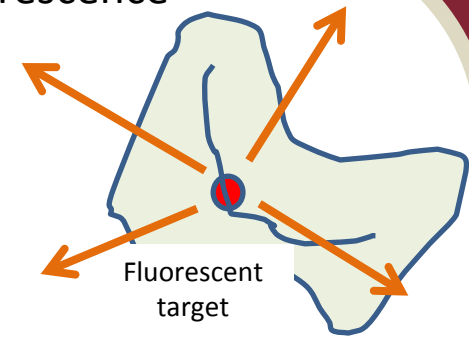
## STED



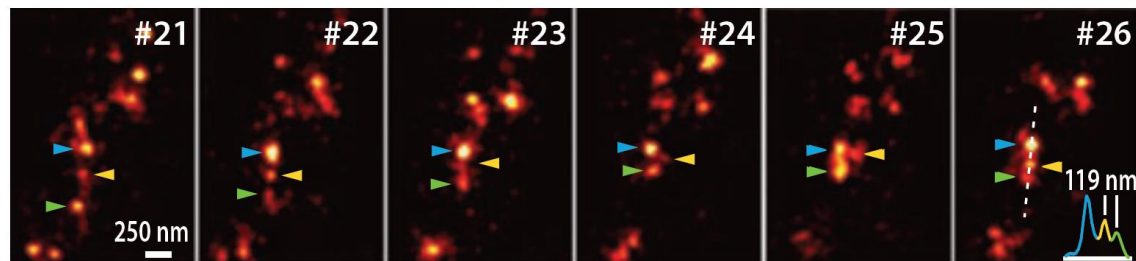
## Excitation



## Fluorescence



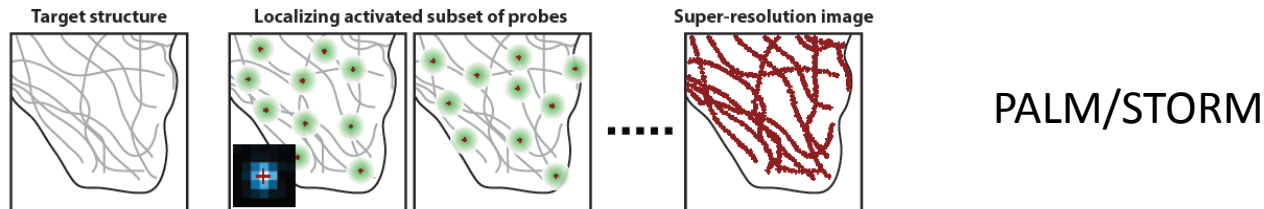
## Stimulated emission depletion microscopy



## Snap-shots of synaptic vesicles



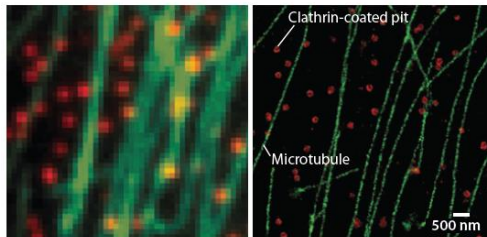
# Super-resolution imaging



Stochastic optical reconstruction microscopy

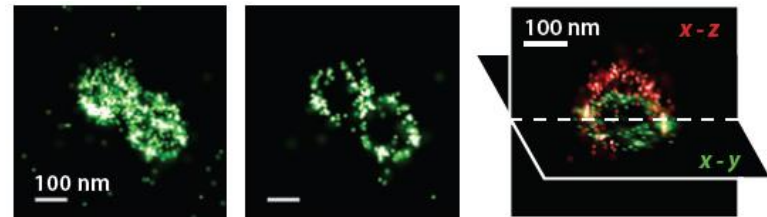
Photoactivated localization microscopy

STORM



microtubules

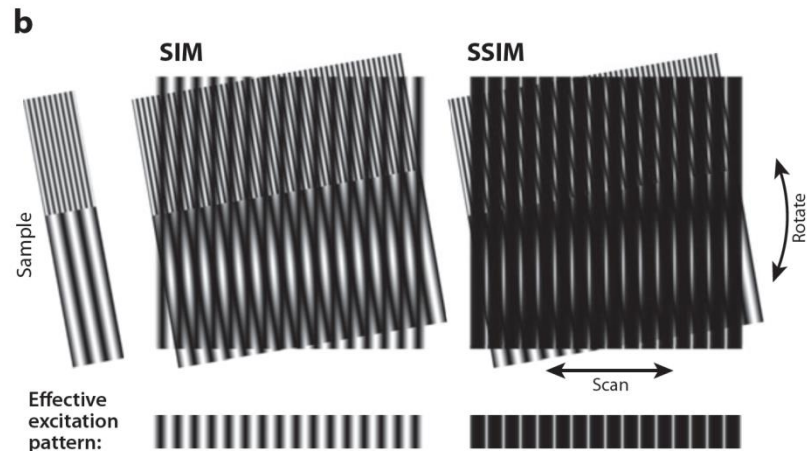
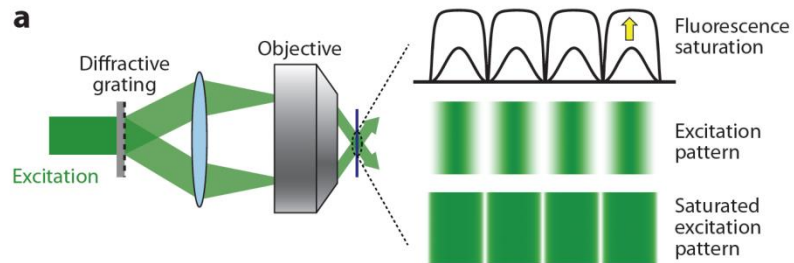
3D STORM



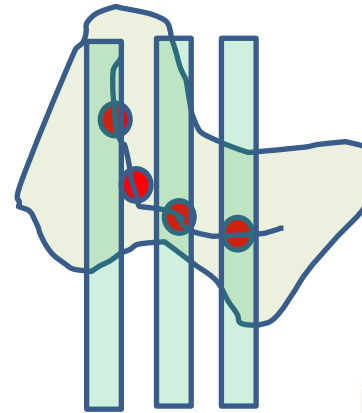
pits

# Super-resolution imaging

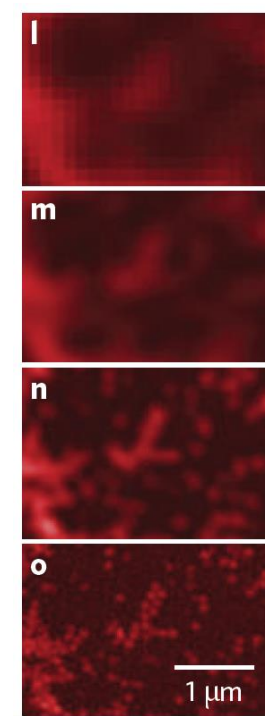
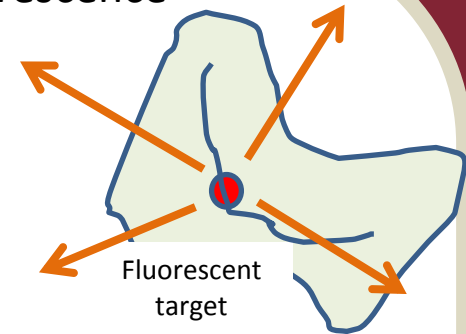
## SIM – Structured Illumination microscopy



Excitation



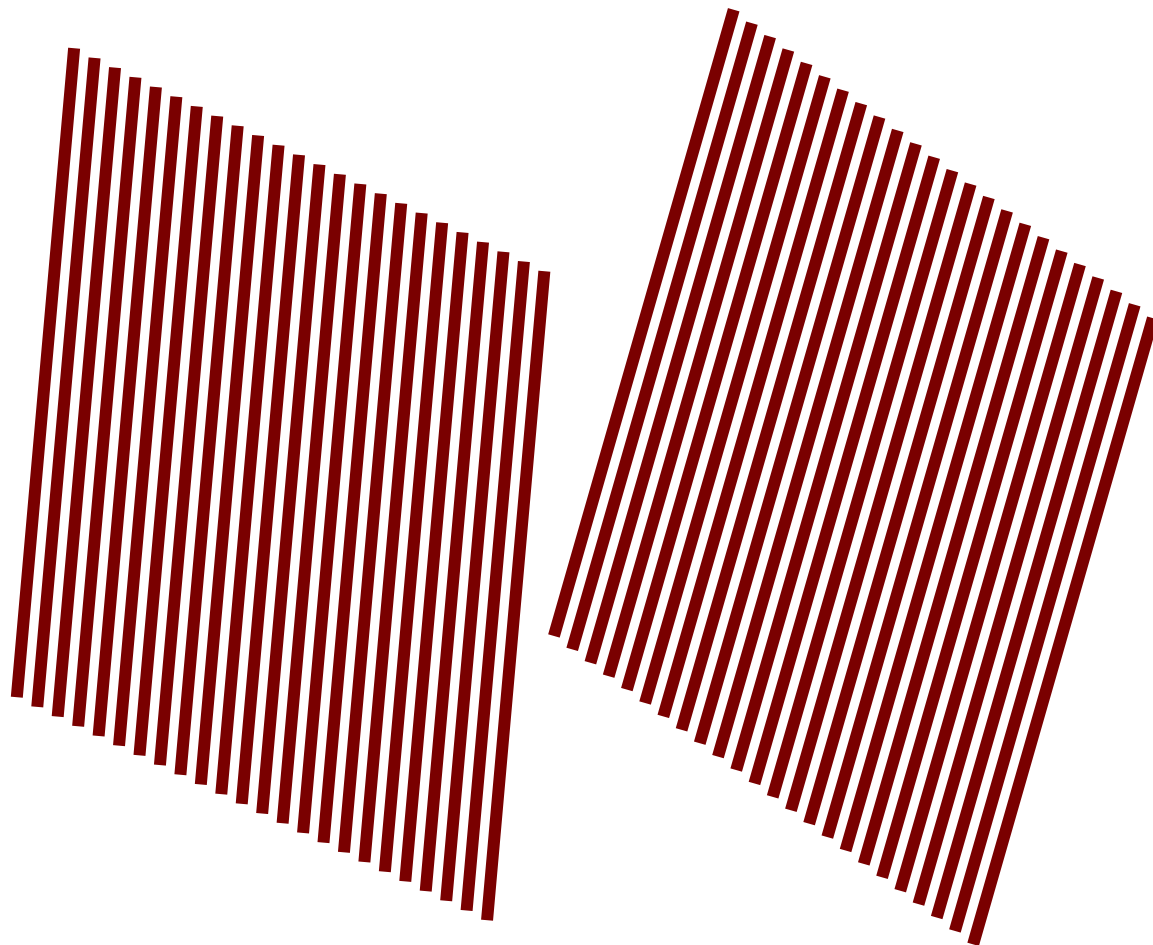
Fluorescence



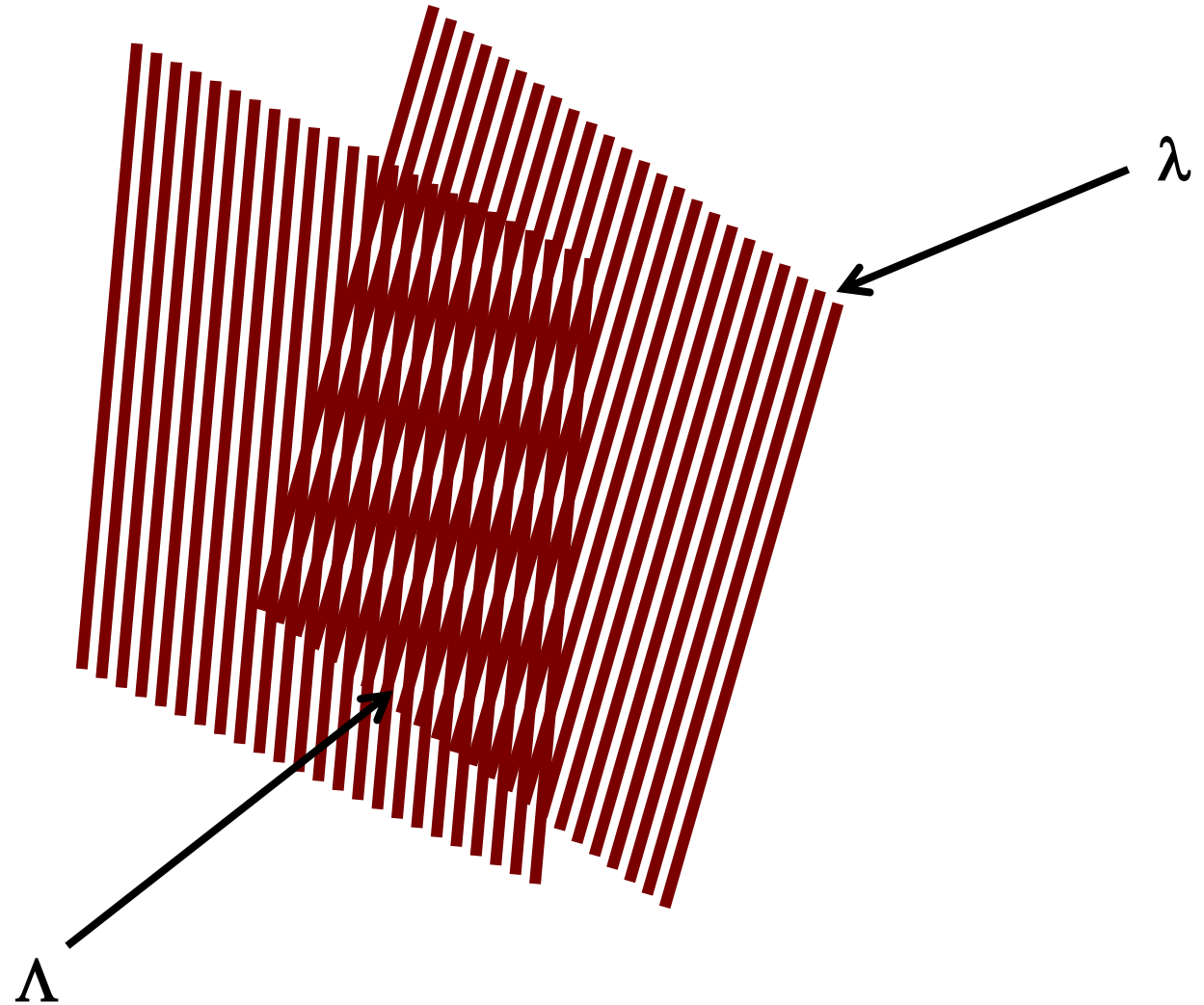
<sup>3</sup> B. Huang, M. Bates, and X. Zhuang, Annu. Rev. Biochem. 2009. 78:993–1016 (2009)



Increasing resolution by *superimposing* known images on *unknown* objects



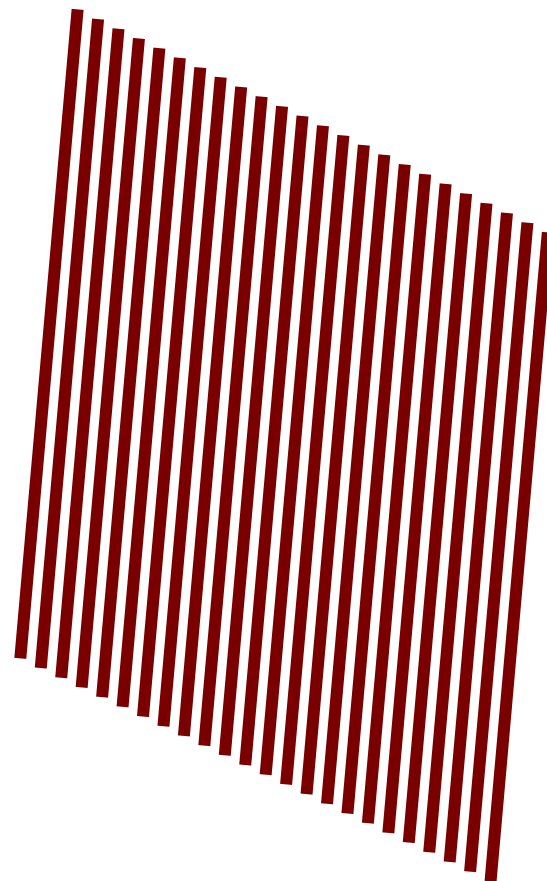
Digital Age revival of an age-long secret...

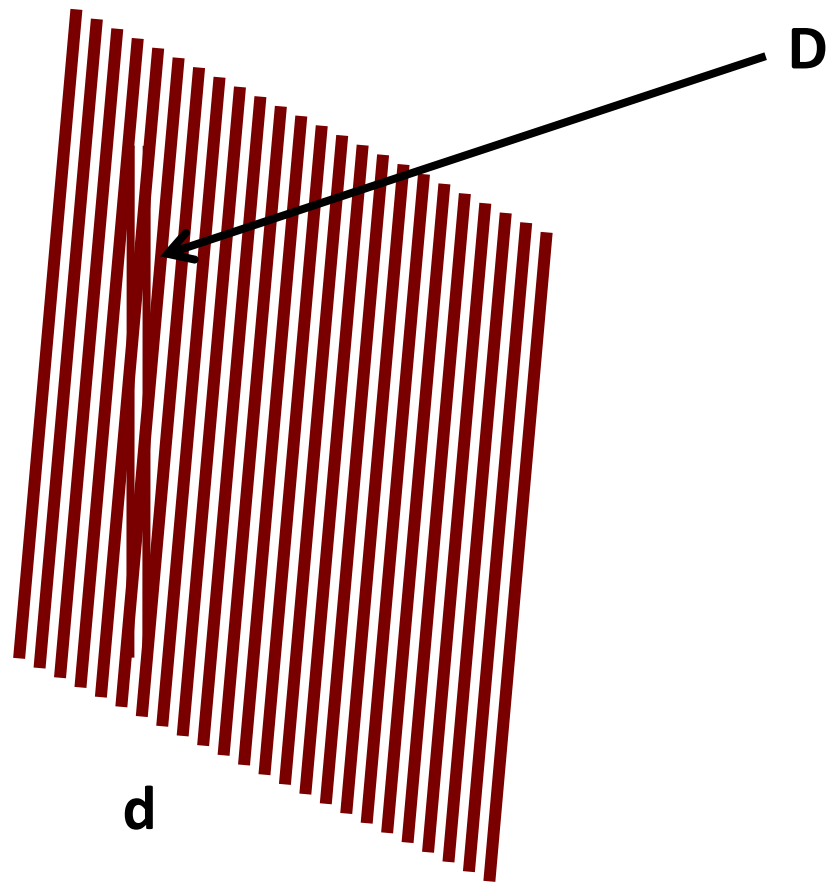


The moiré pattern



**d**

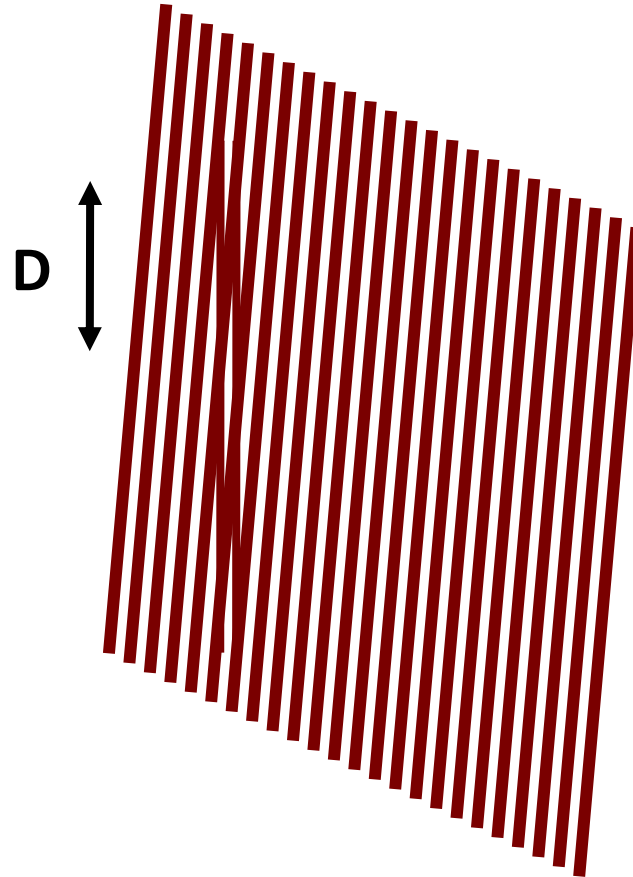






$d < L$

We DO NOT see the two lines

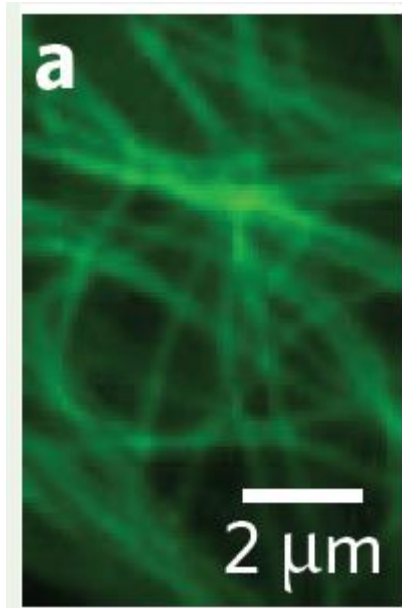


$D > L$

We DO “see” the two lines



Is resolution really increased?



TIRF image of a living  
S2 cell expressing  
GFP-tubulin.

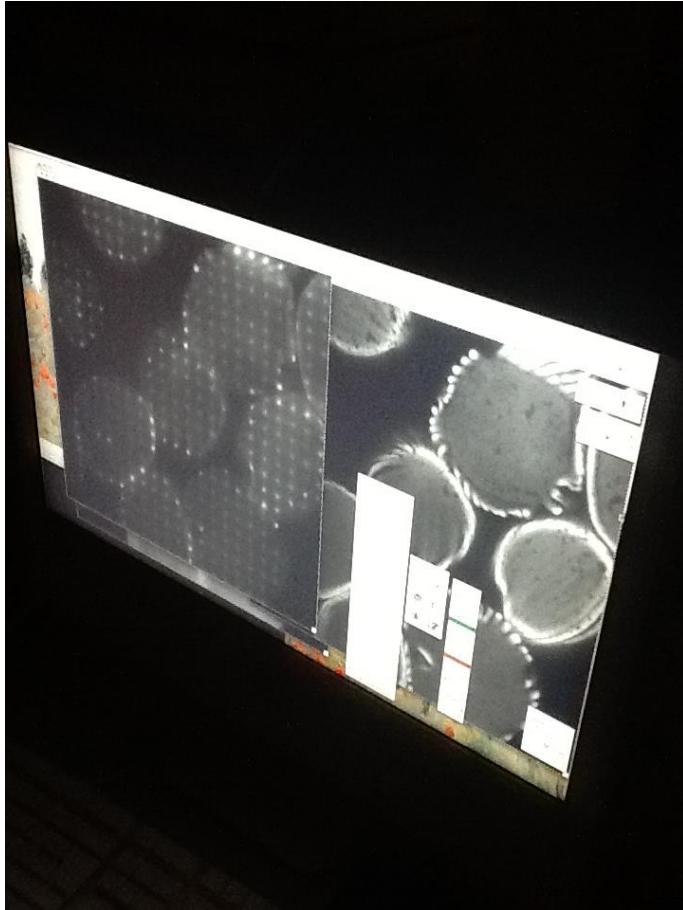


2D SIM

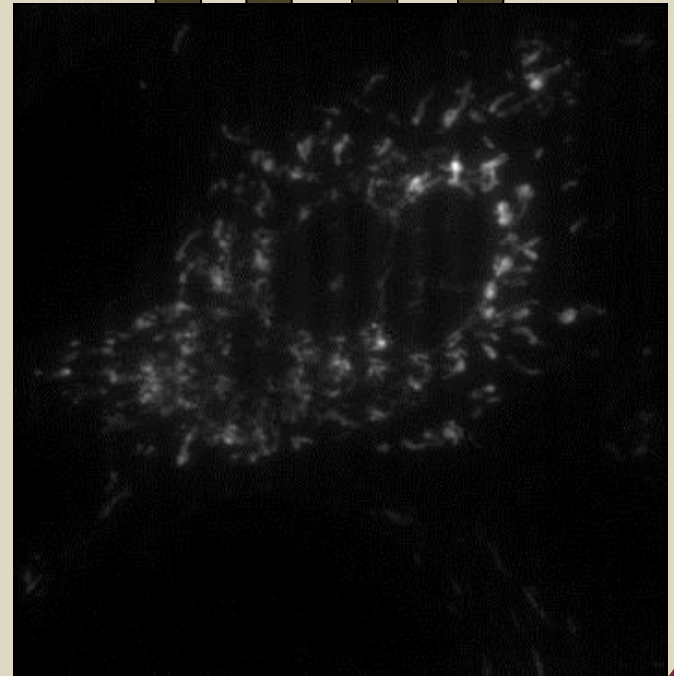
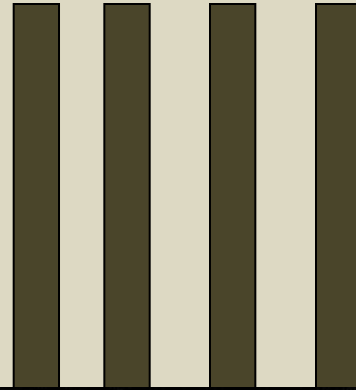
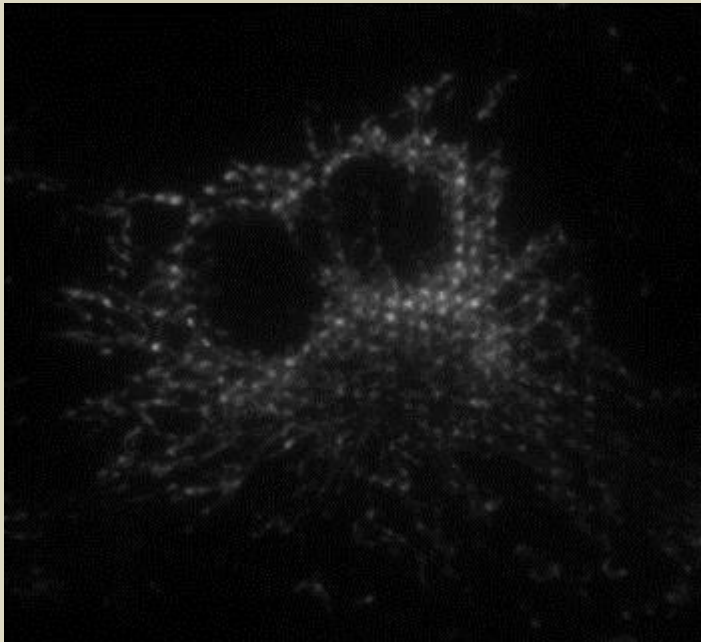
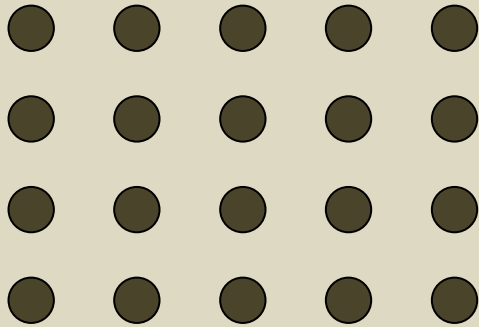


Increasing resolution by *superimposing* known images on *unknown* objects

## Structured Illumination Microscopy



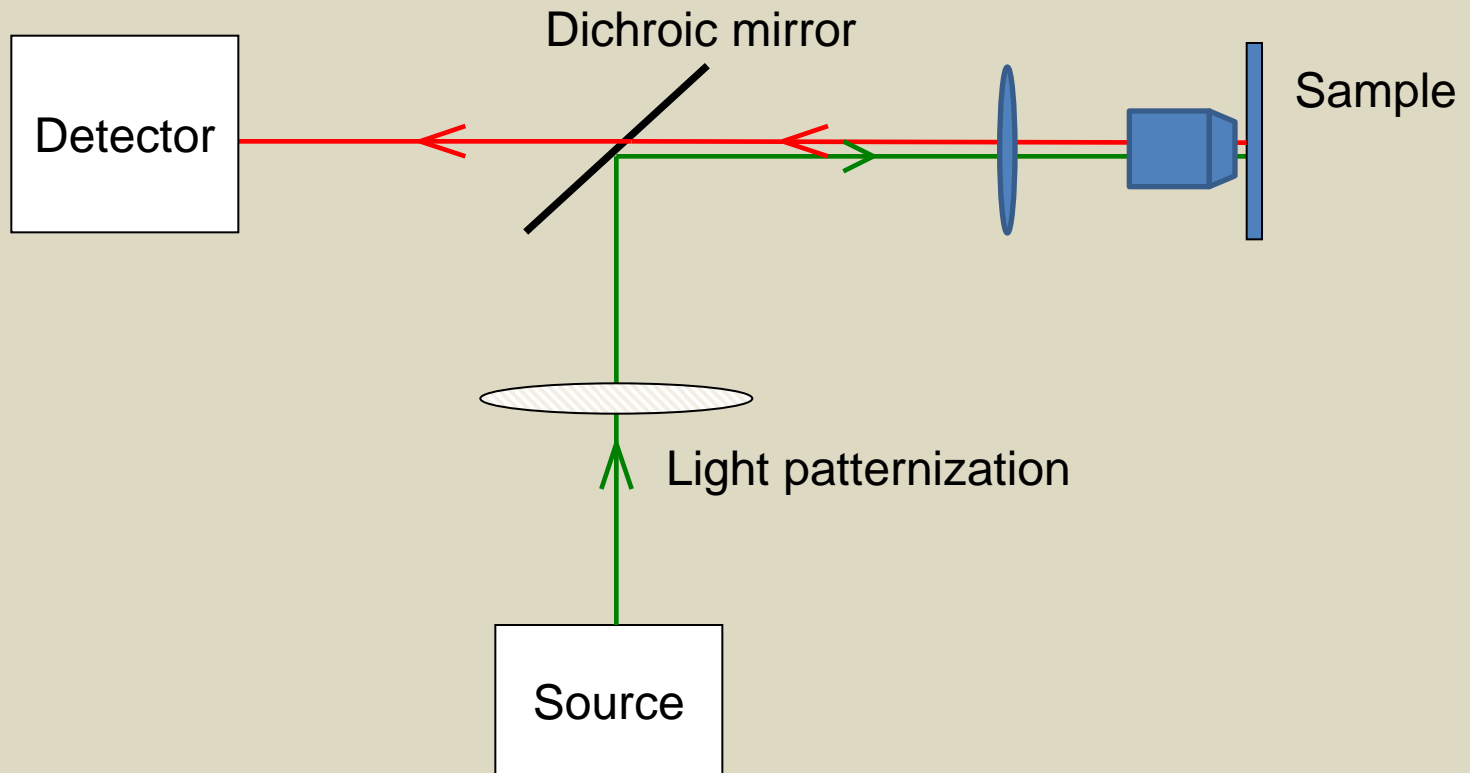
# Patterns



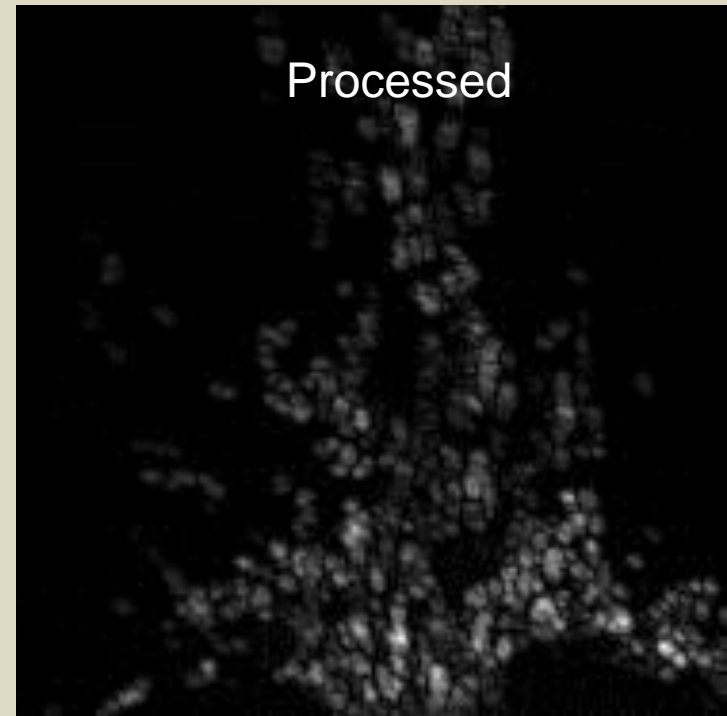
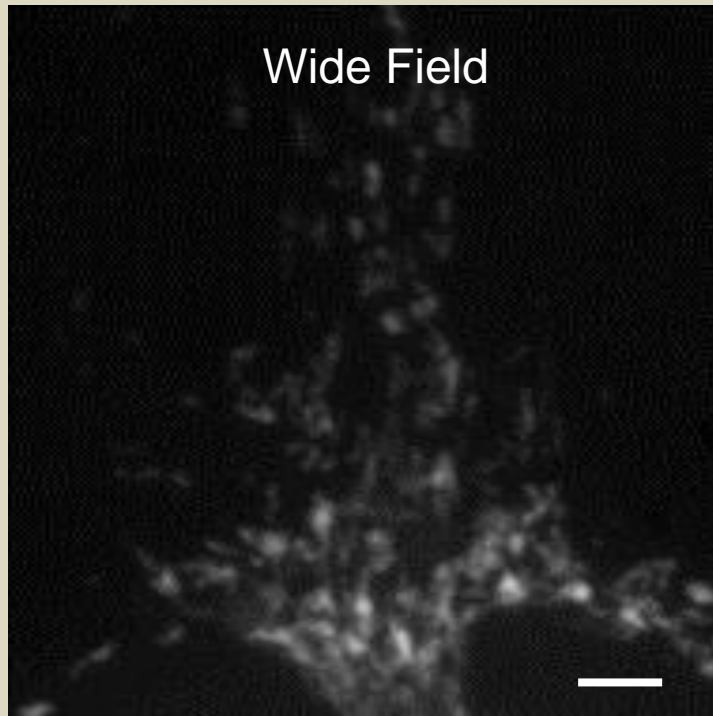
Pattern projected  
onto the sample



## Setup




## Sample results

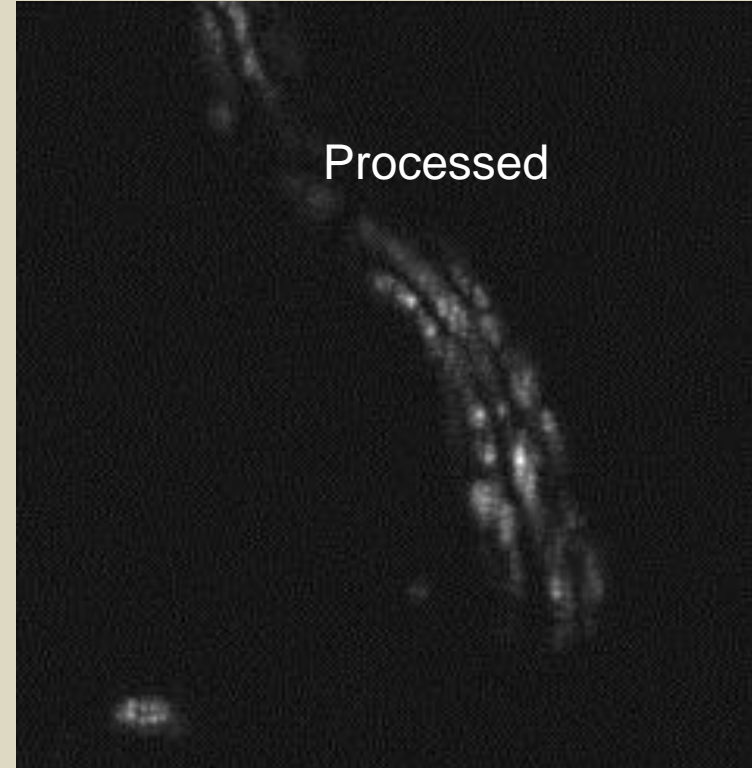
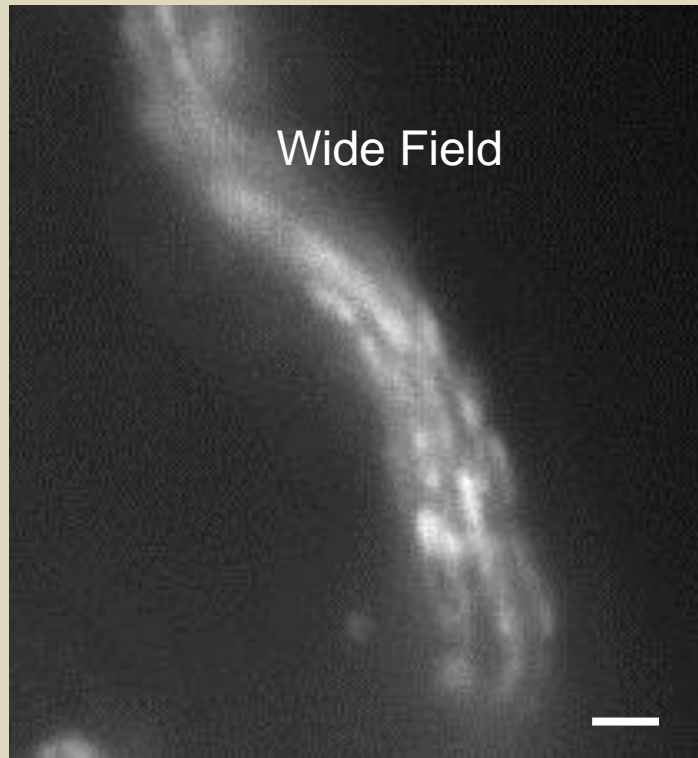


Particular of BPAE cells

4 $\mu$ m



## Sample results



Particular of Human Skin

2 $\mu$ m

## Scale-free optics

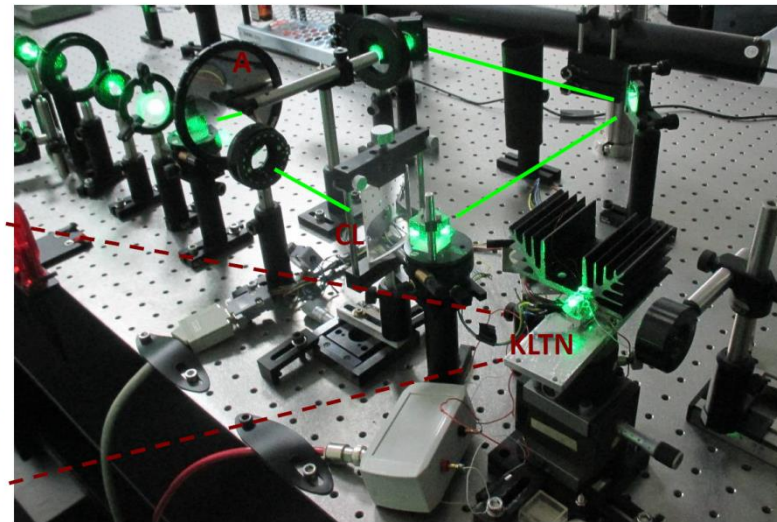
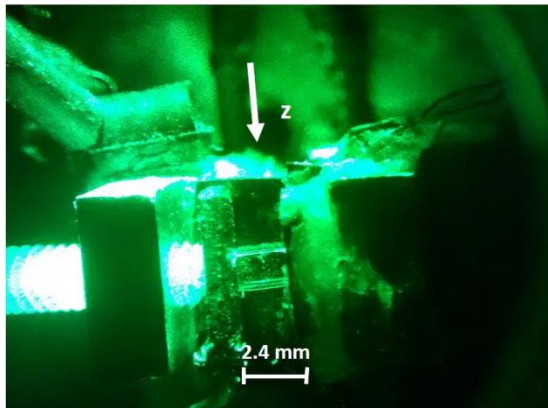
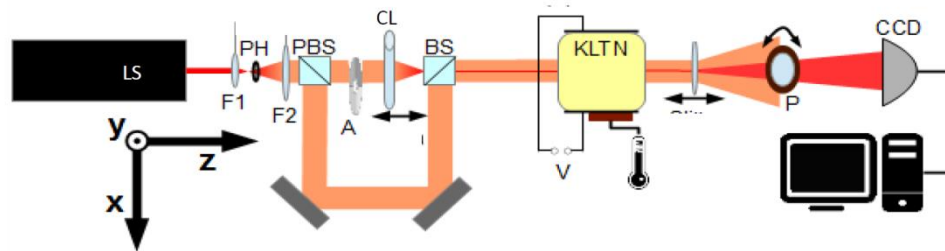




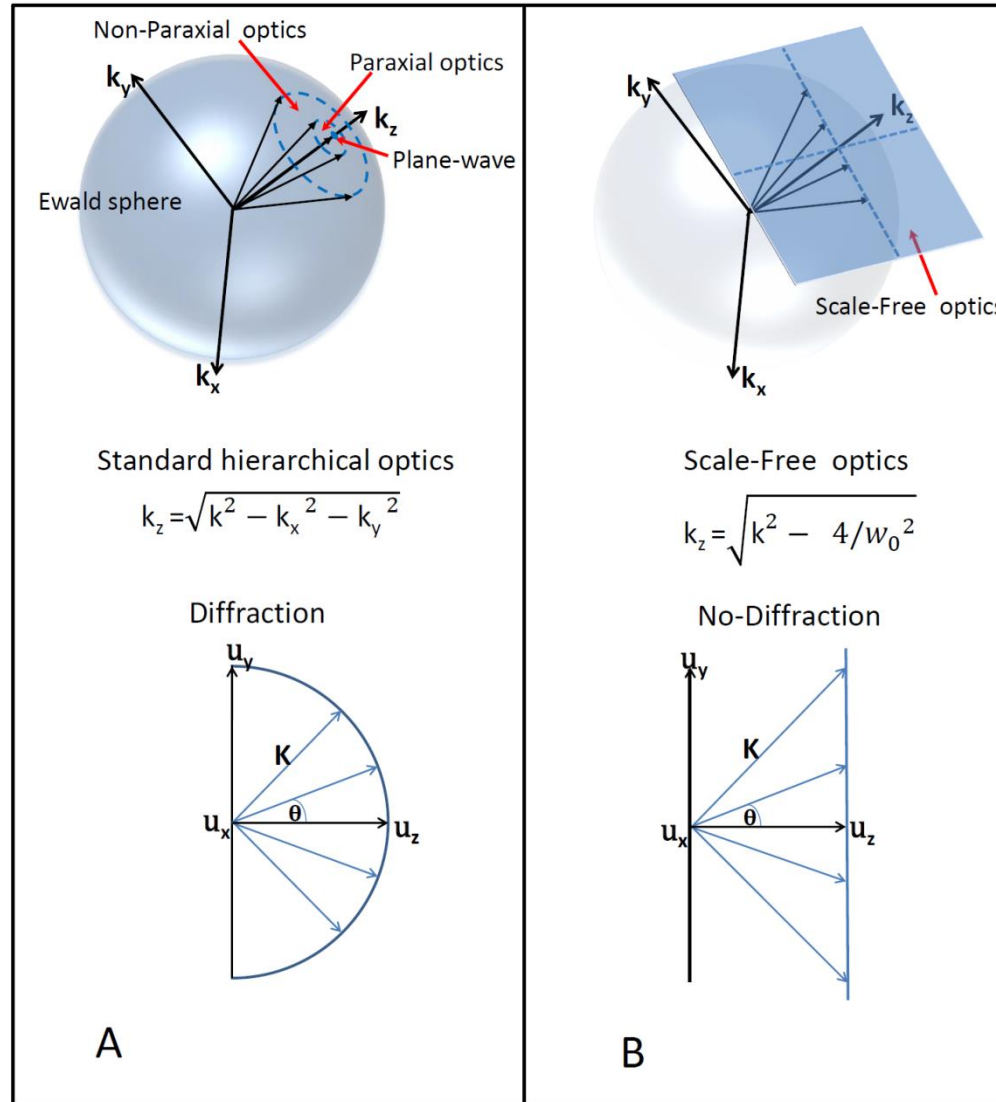
# Scale-free optics and diffractionless waves in nanodisordered ferroelectrics

E. DelRe<sup>1\*</sup>, E. Spinozzi<sup>1</sup>, A. J. Agranat<sup>2</sup> and C. Conti<sup>3</sup>

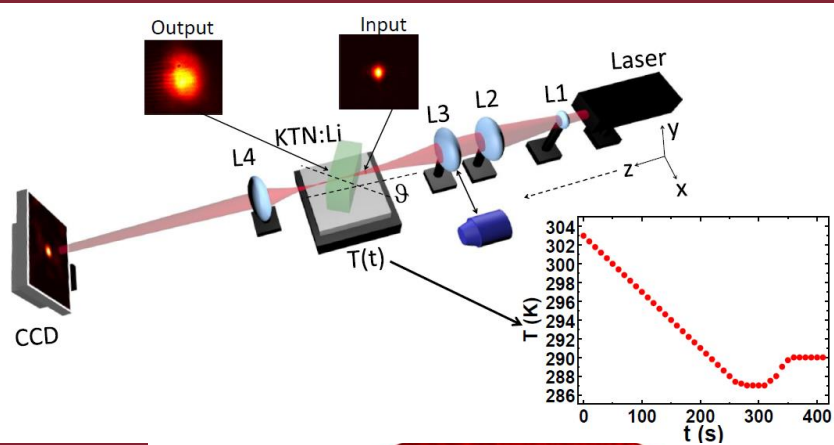
## Experimental setup



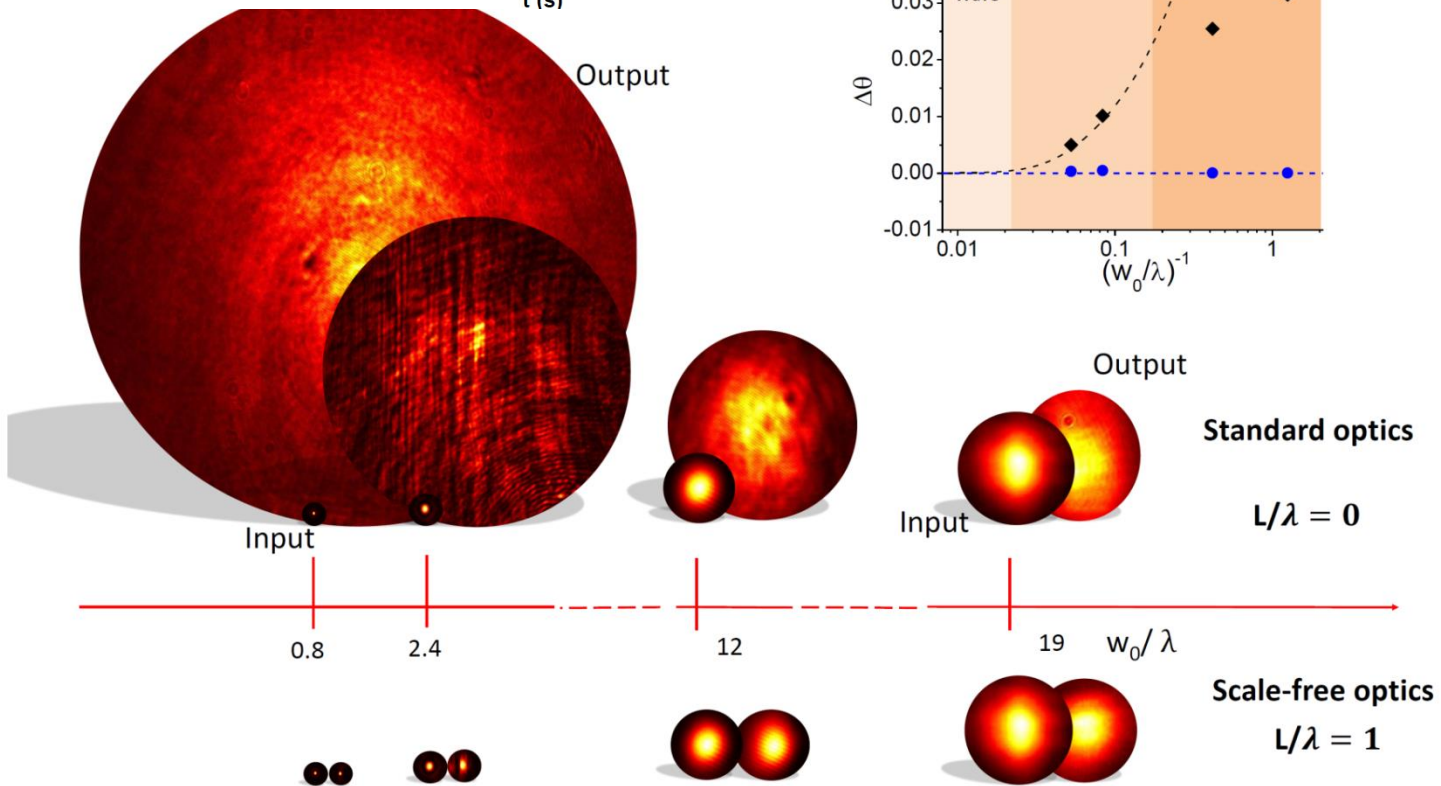
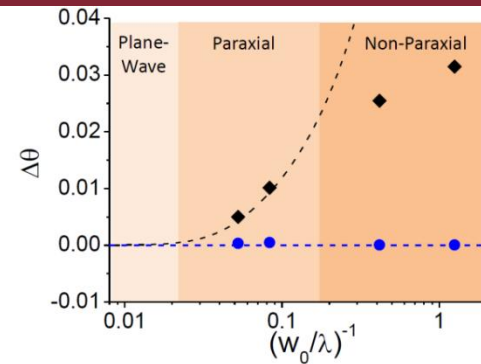
# Comparing standard optics and scale-free optics



$$-\frac{\nabla^2 E}{E} + \left(\frac{L}{\lambda}\right)^2 \left(\frac{\nabla|E|^2}{2|E|^2}\right)^2 = k^2$$



## Scale-invariance: the full picture



PHYSICAL REVIEW A **92**, 013835 (2015)

## Observation of diffraction cancellation for nonparaxial beams in the scale-free-optics regime

F. Di Mei,<sup>1,2</sup> D. Pierangeli,<sup>1</sup> J. Parravicini,<sup>1</sup> C. Conti,<sup>3,1</sup> A. J. Agranat,<sup>4</sup> and E. DelRe<sup>1,3,\*</sup>

## Challenge

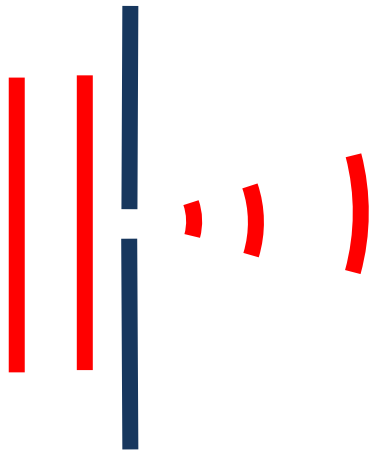
Propagate a **non-paraxial subwavelength-sized beam** in a volume  
along **macroscopic distances** for imaging



# Diffraction

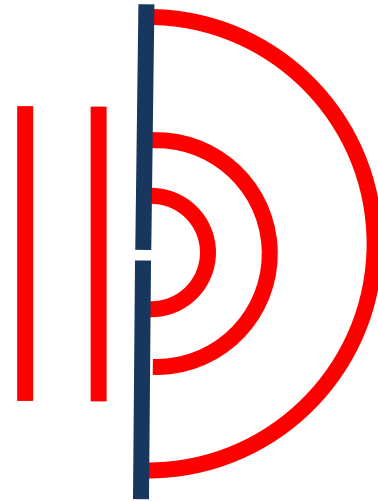
$$d \gg \lambda$$

Small angular aperture



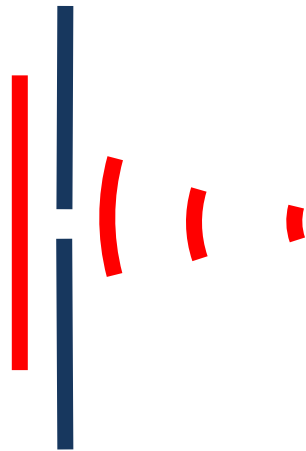
$$d \approx \lambda$$

Large angular aperture



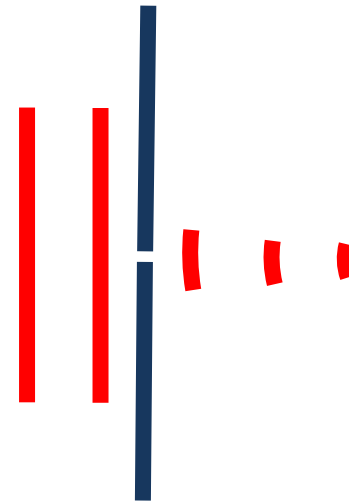
# Diffraction

? angular aperture

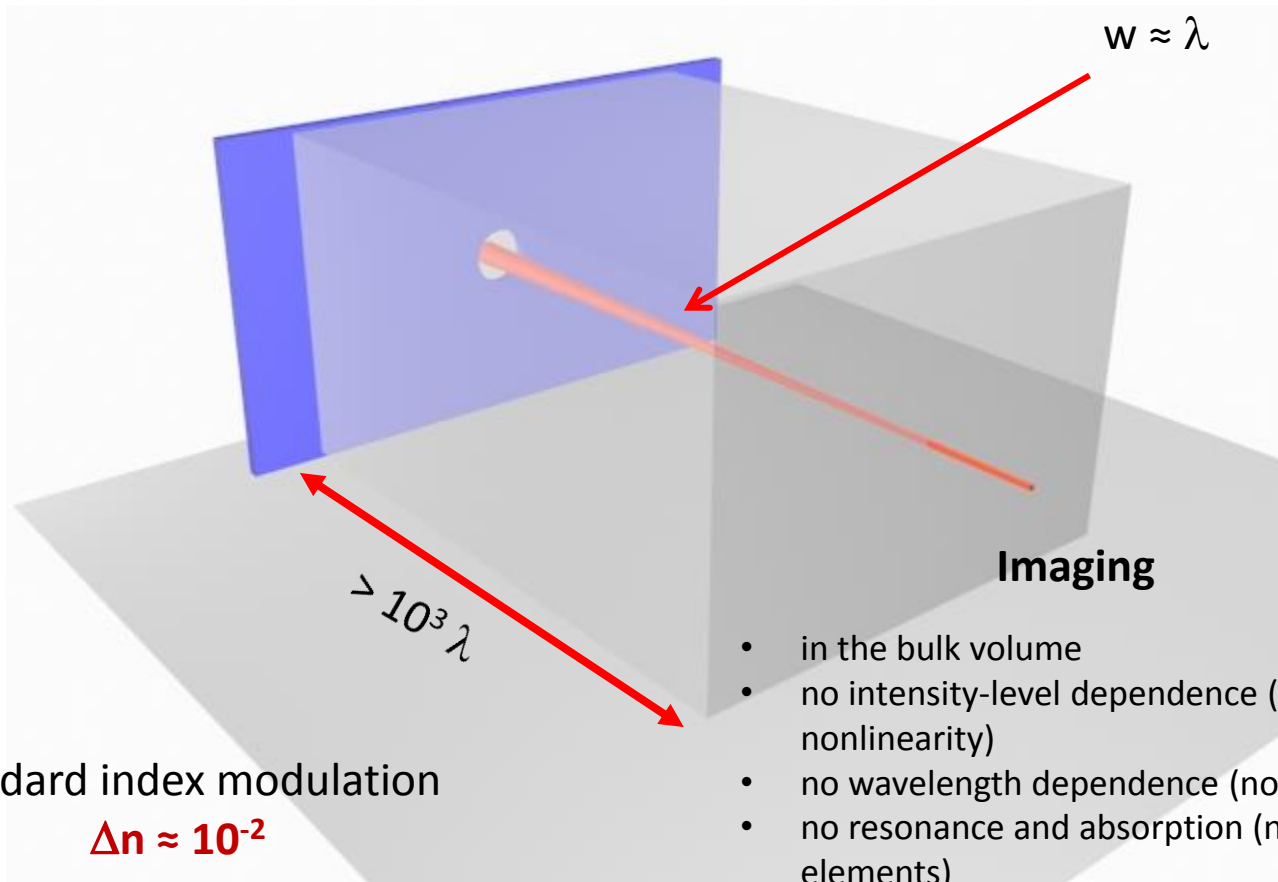


$$d \gg \lambda$$

? angular aperture



$$d \approx \lambda$$



Standard index modulation

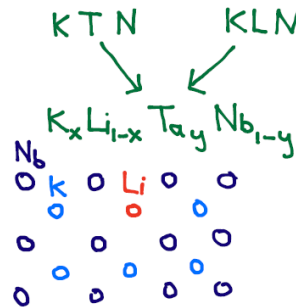
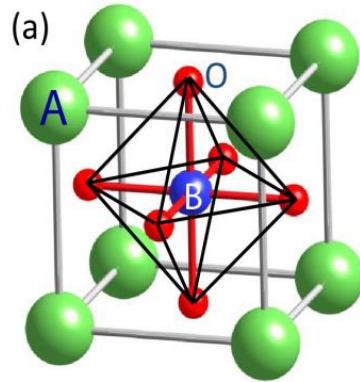
$$\Delta n \approx 10^{-2}$$

**Imaging**

- in the bulk volume
- no intensity-level dependence (no standard nonlinearity)
- no wavelength dependence (no periodicity)
- no resonance and absorption (no conducting elements)
- must work beyond paraxial optics

# Material

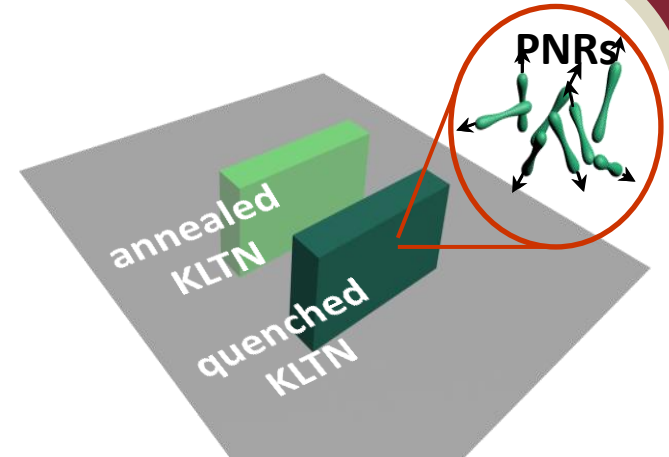
## Dipolar glass forming inside the KTN:Li crystal



Compositional  
disorder

+

deep-in-band Cu impurities → Huge non-linear EO response



E. DelRe et al. Nat. Photon. **5**, 39 (2011)

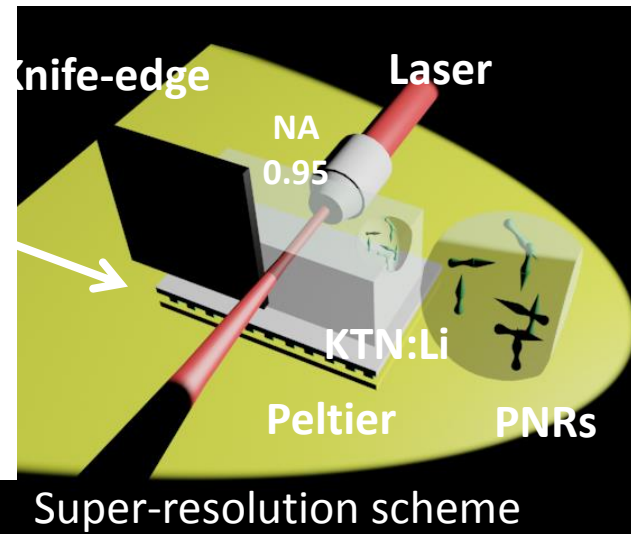
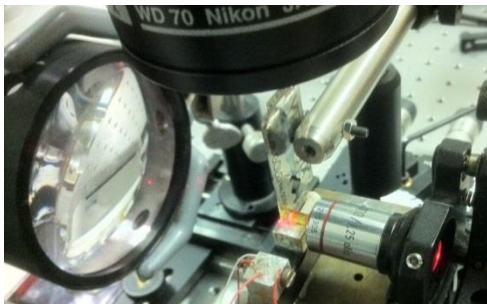
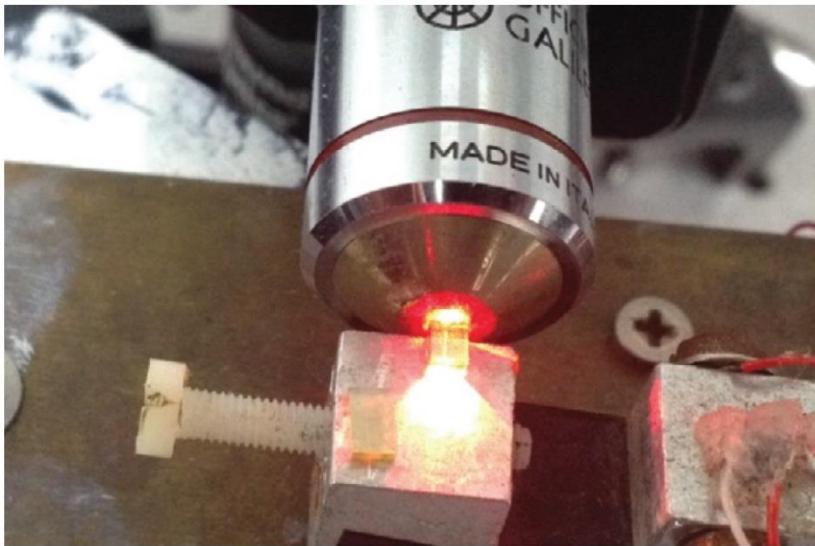
A.A. Bokov, Z. Ye Journal of Material Science 41 31-52 (2006)



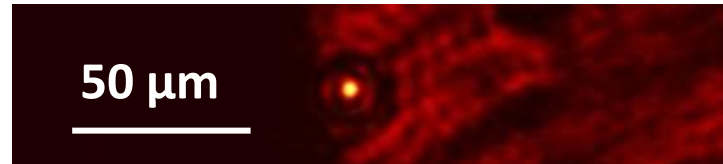


## Subwavelength anti-diffracting beams propagating over more than 1,000 Rayleigh lengths

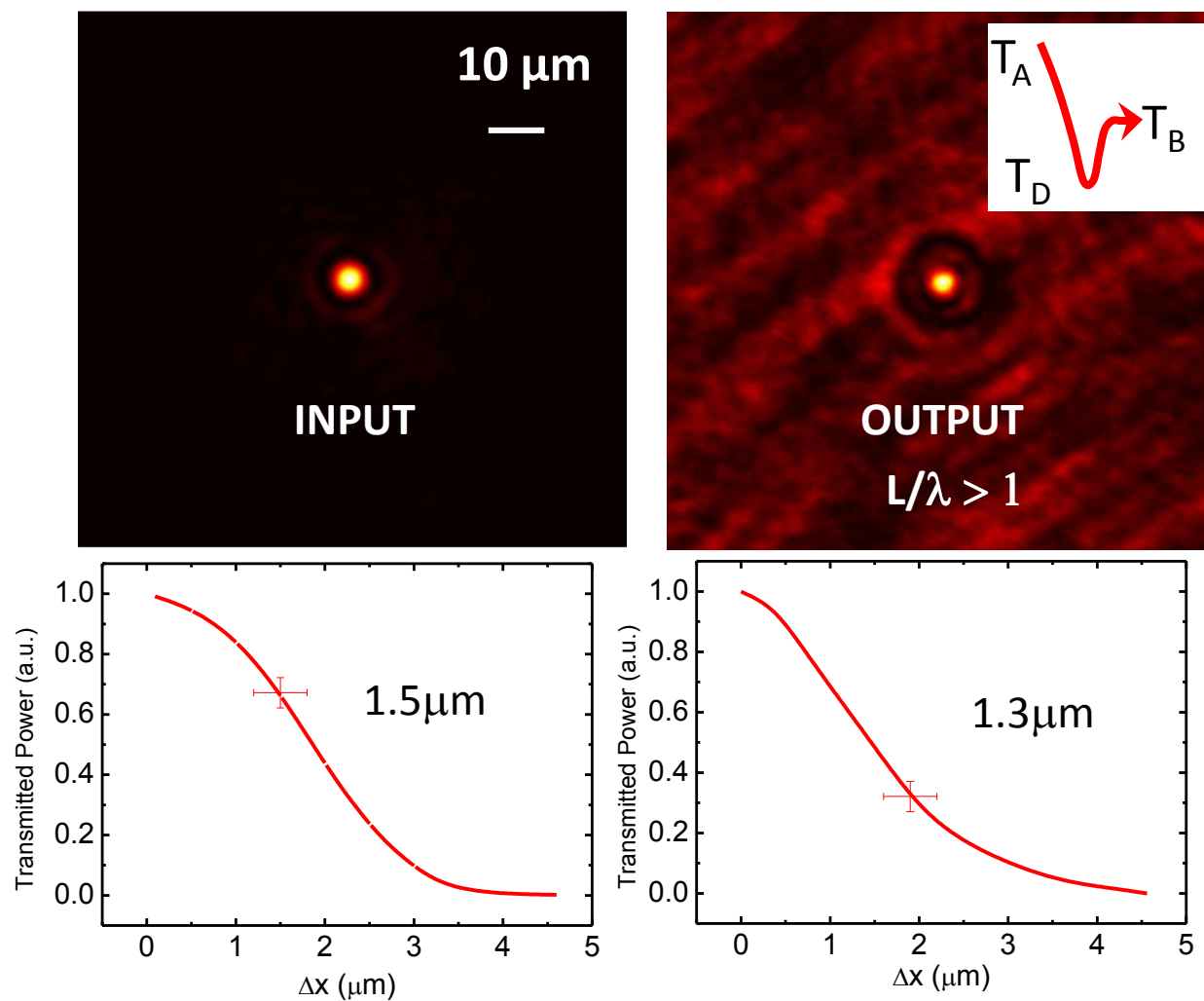
Eugenio DelRe<sup>1\*</sup>, Fabrizio Di Mei<sup>1,2</sup>, Jacopo Parravicini<sup>1</sup>, Gianbattista Parravicini<sup>3</sup>, Aharon J. Agranat<sup>4</sup> and Claudio Conti<sup>1,5</sup>



50  $\mu\text{m}$

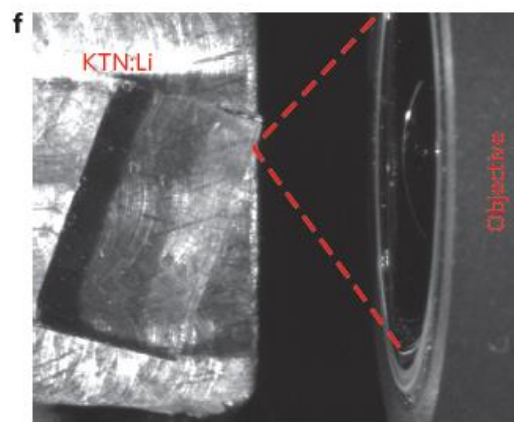
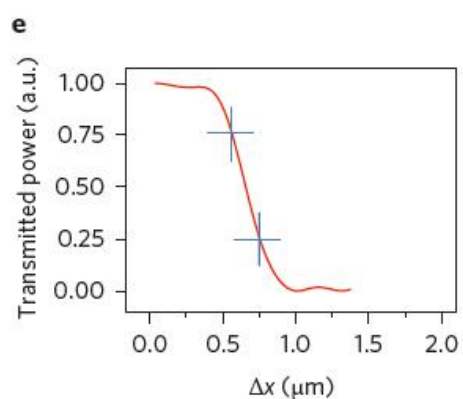
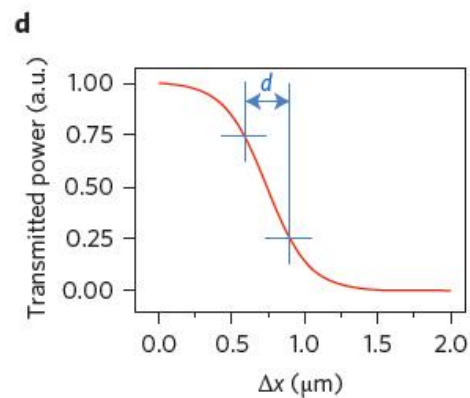
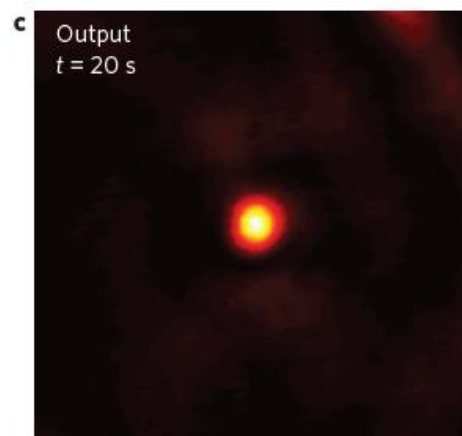
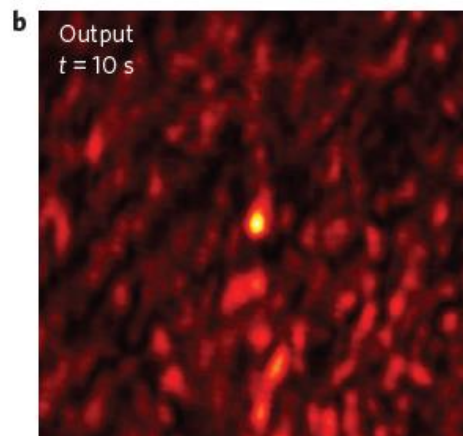
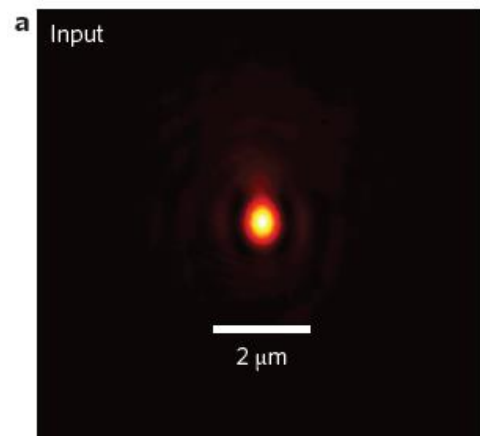


## Massive beam propagation ( $L/\lambda=1.1$ )



# Subwavelength massive beam propagation ( $L/\lambda=1.1$ )

0.28 micrometers...



1000 Rayleigh lengths ( $L_z=2.6 \text{ mm}$ )

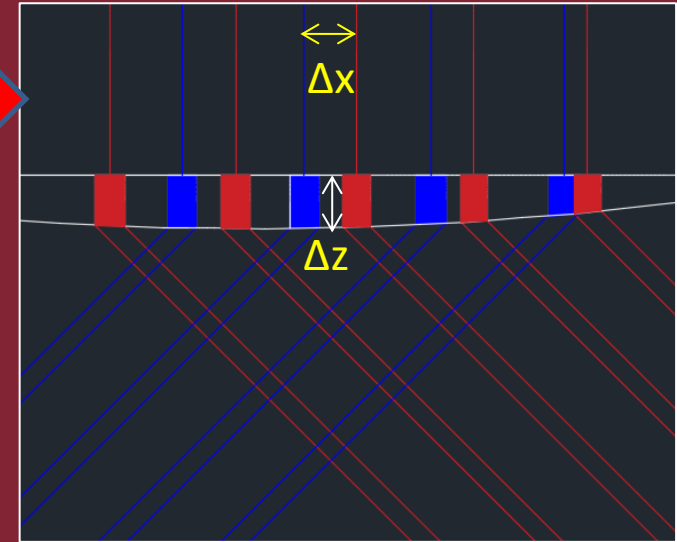
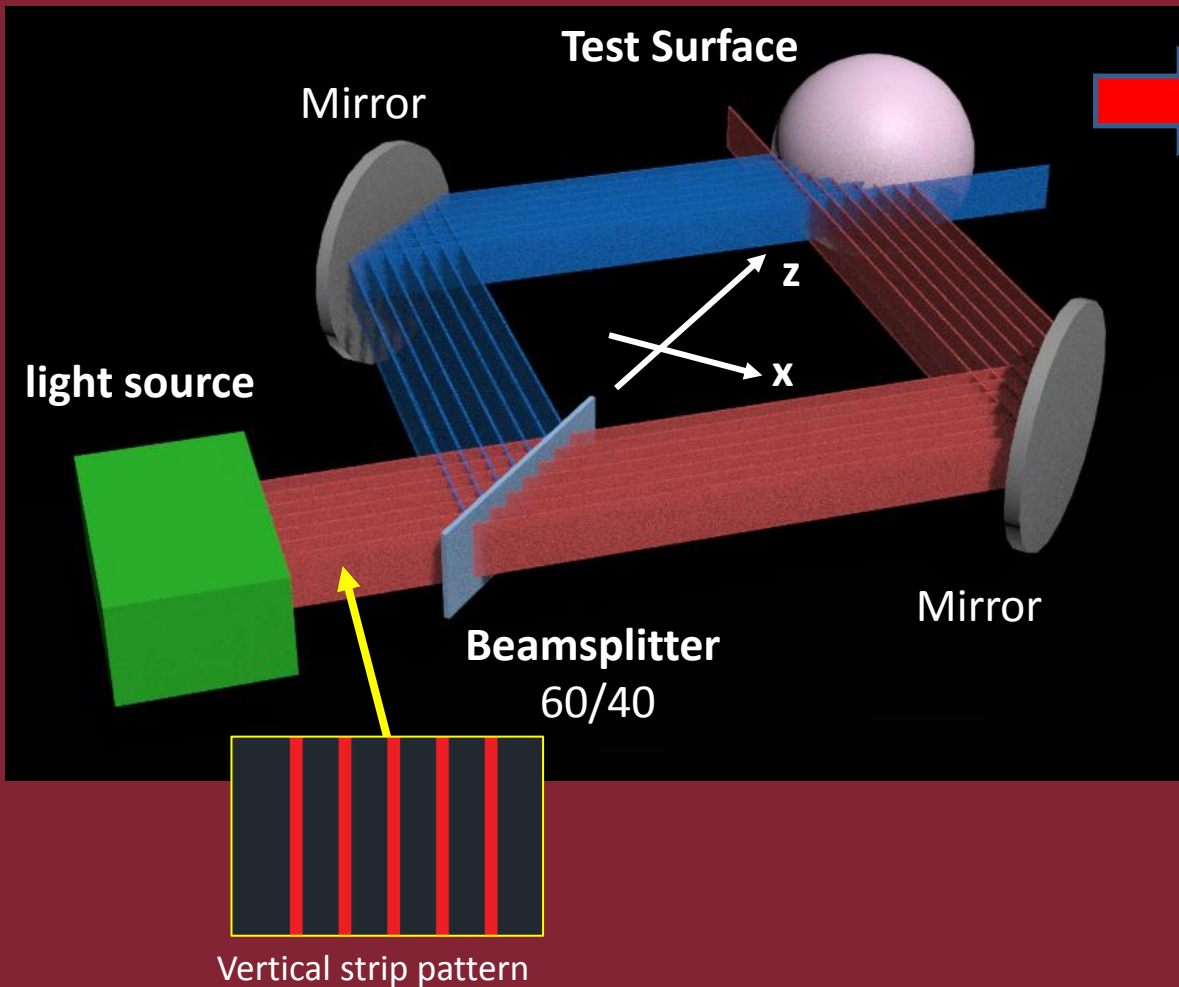


## **Structured illumination 3D scan**



# Idea: structured light with periodic on-off pattern

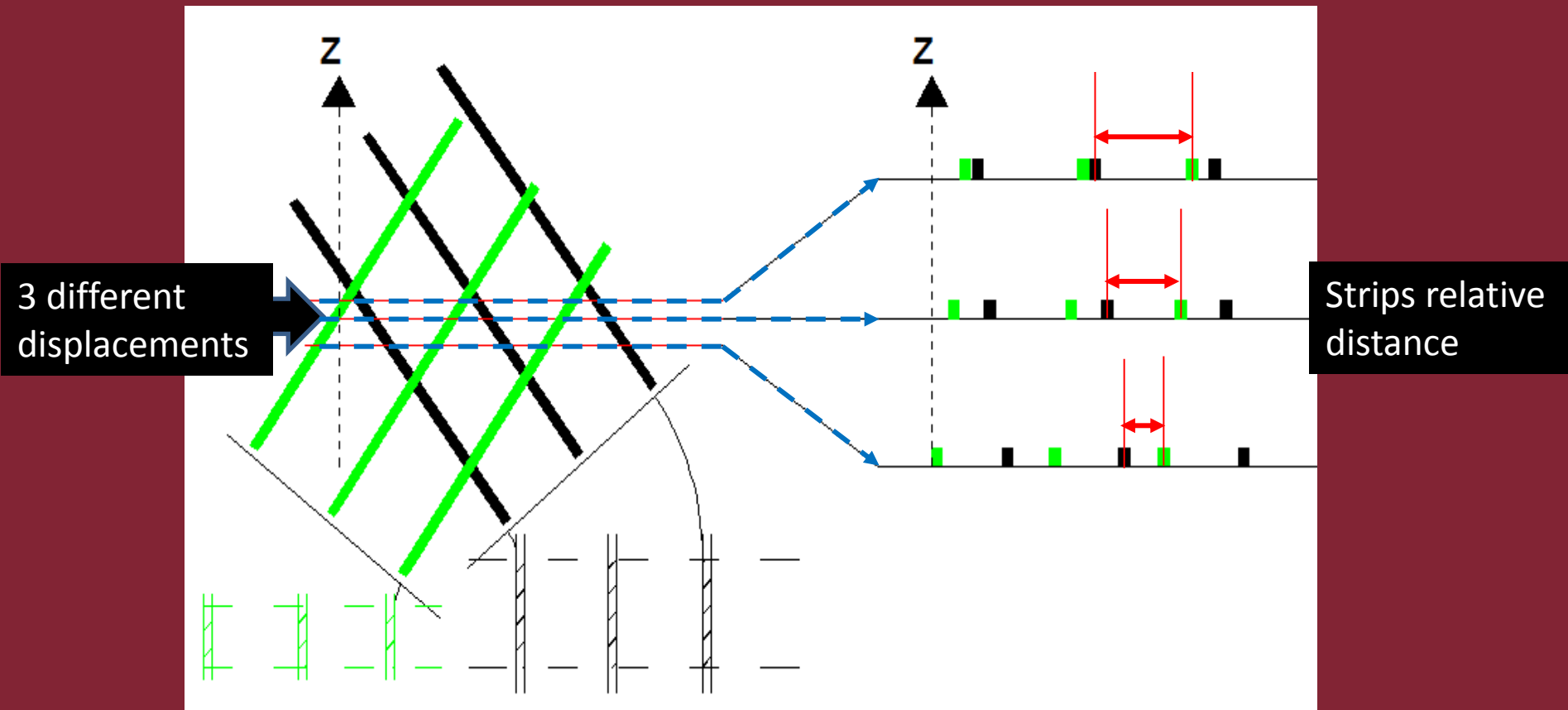
## Experimental setup



Locally, relative distance between strips ( $\Delta x$ ) provides information about relative surface displacement ( $\Delta z$ )



## Strips distance versus surface displacement

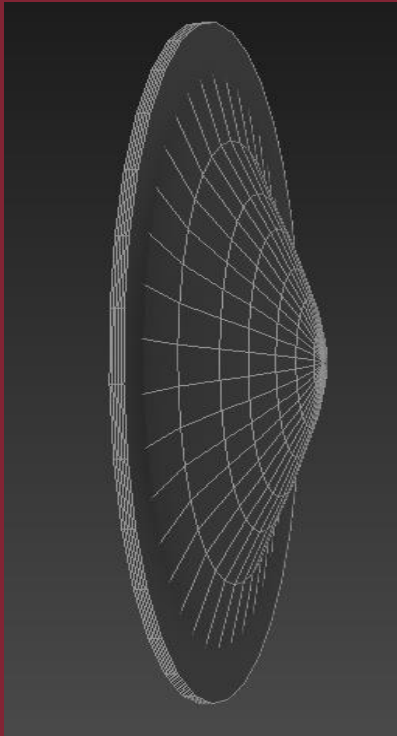


Surface displacement produces relative distance modifications between pattern strips coming from two different directions



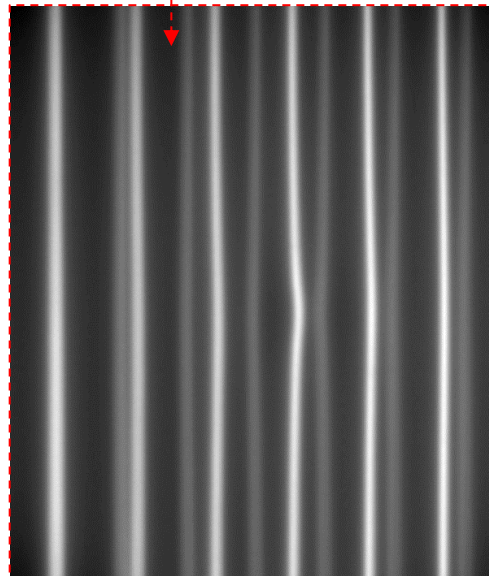
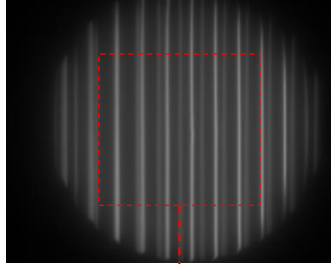


## Test surface and acquisitions



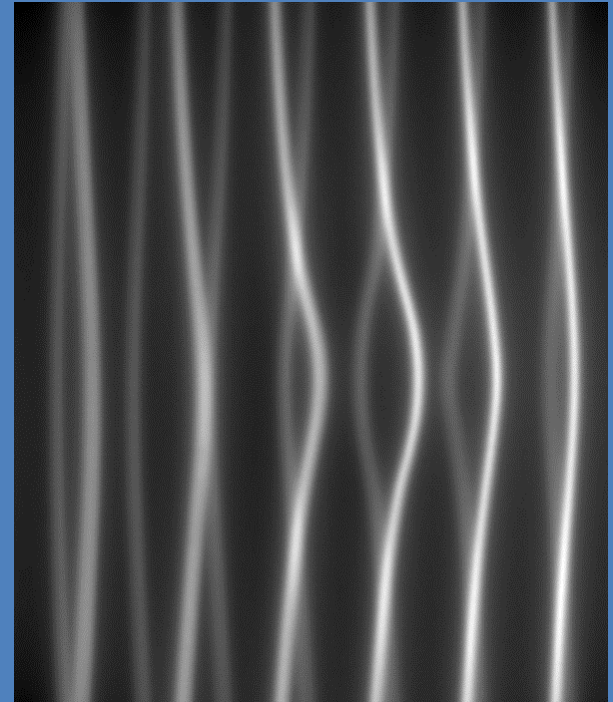
Elastic membrane  
deformed with spherical  
profile in range 0 to 3 mm

Single Frame



Processed portion  
(displacement of 0,3mm)

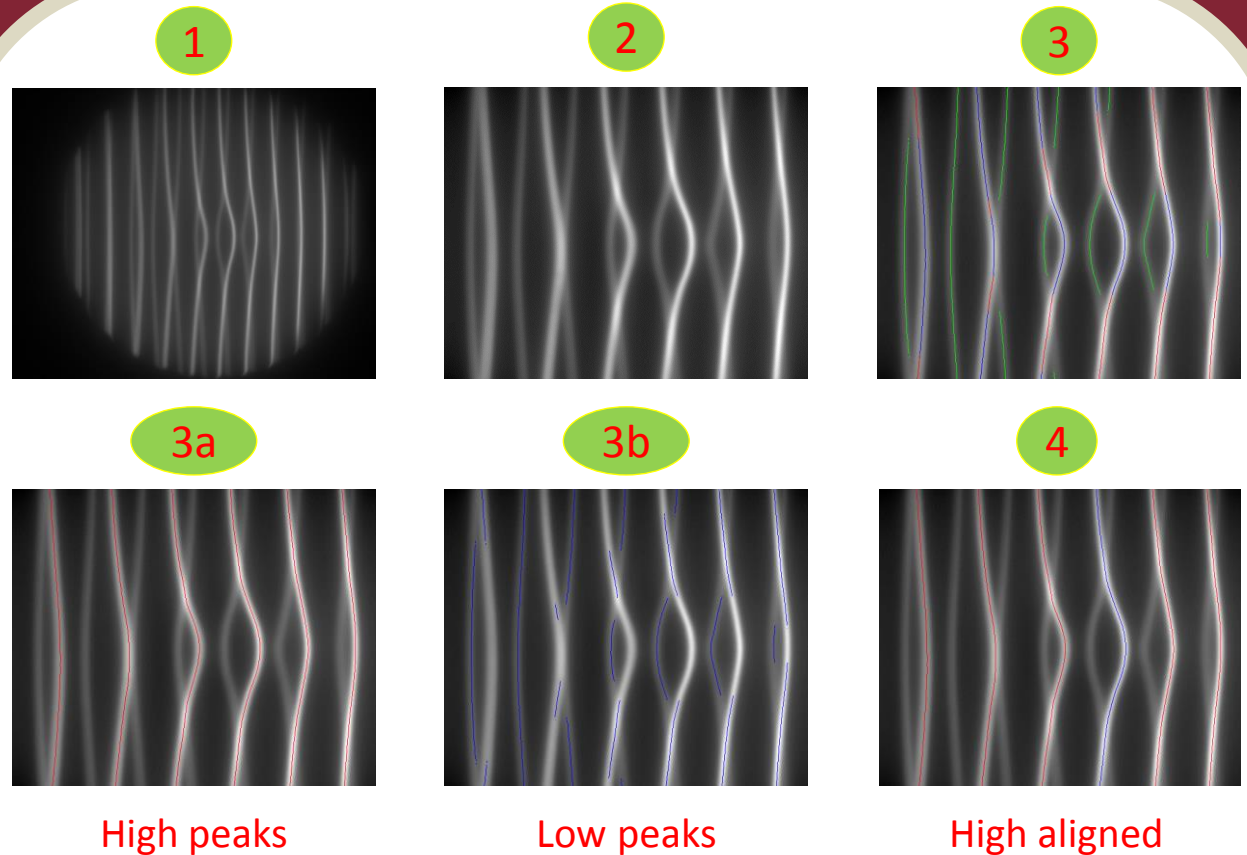
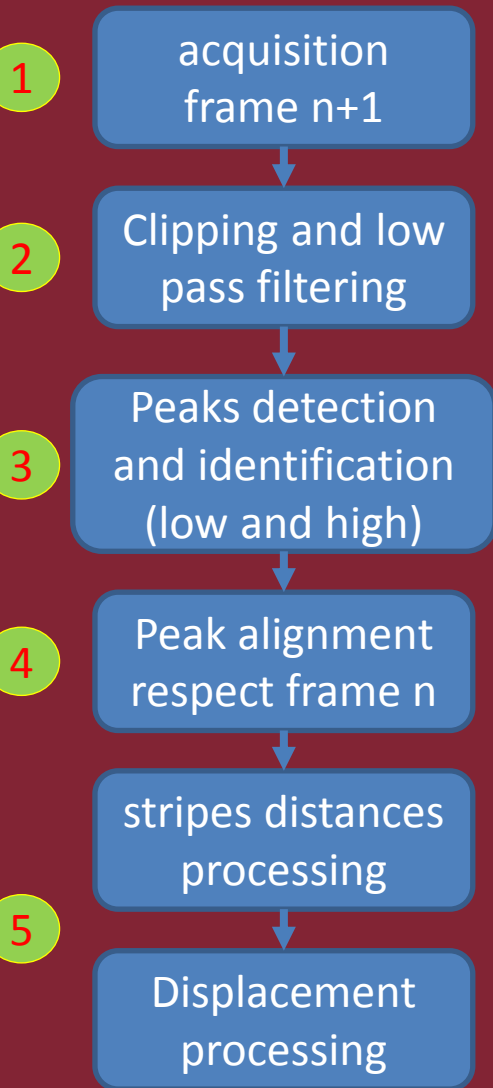
Displacement of 3 mm)



Worst case: intensity  
variations, period variations  
and strips overlap



## Processing algorithm



Processing is performed using parallel algorithms to improve efficiency

