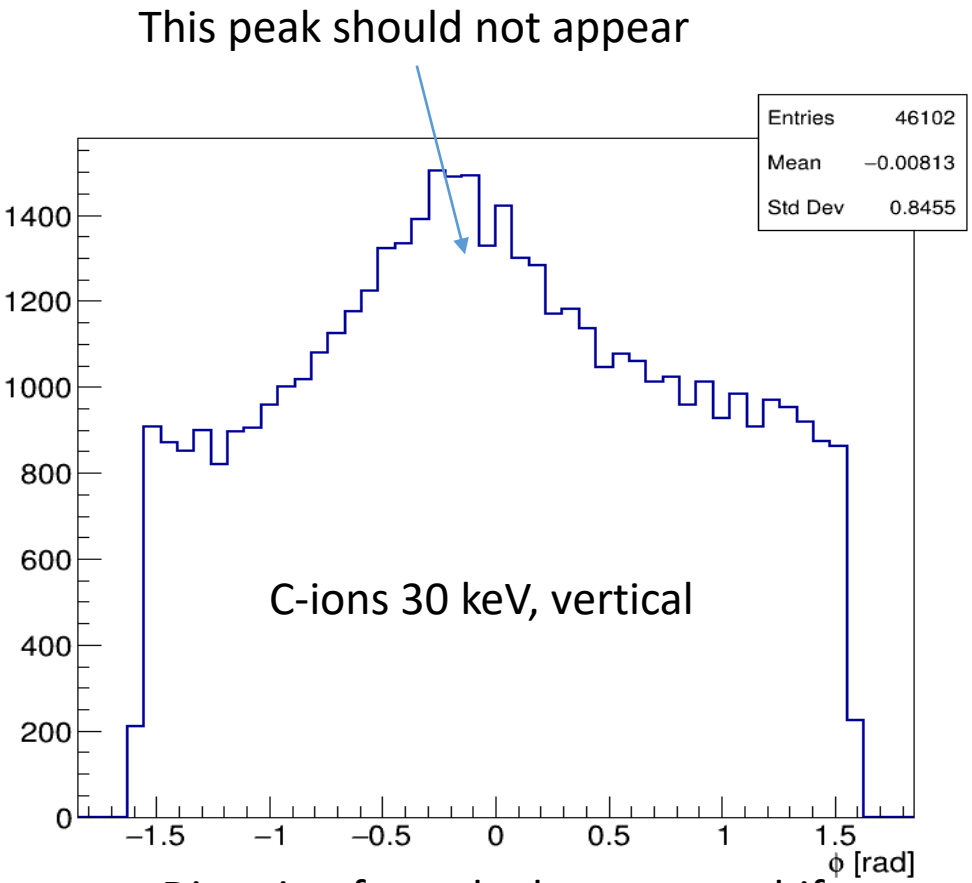
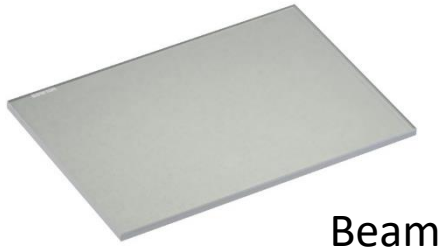
Status of the microscope

Andrey Alexandrov

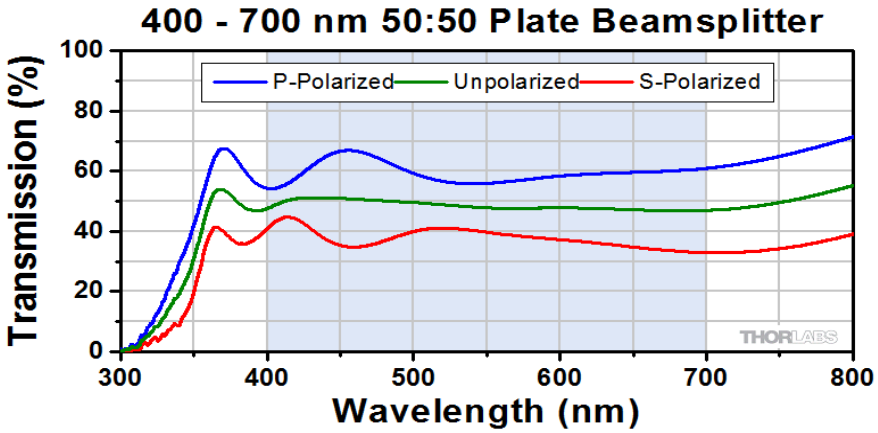
Motivation

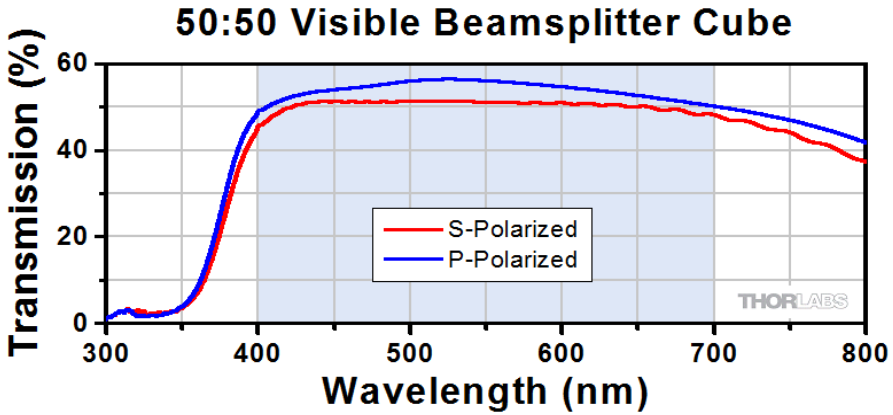

This peak should not appear
Entries 46102
Mean −0.00813
Std Dev 0.8455
1400
1200
1000
800
600
400
200
0
−1.5
−1
−0.5
0
0.5
1
1.5
ϕ [rad]
C-ions 30 keV, vertical
Direction from the barycentre shift

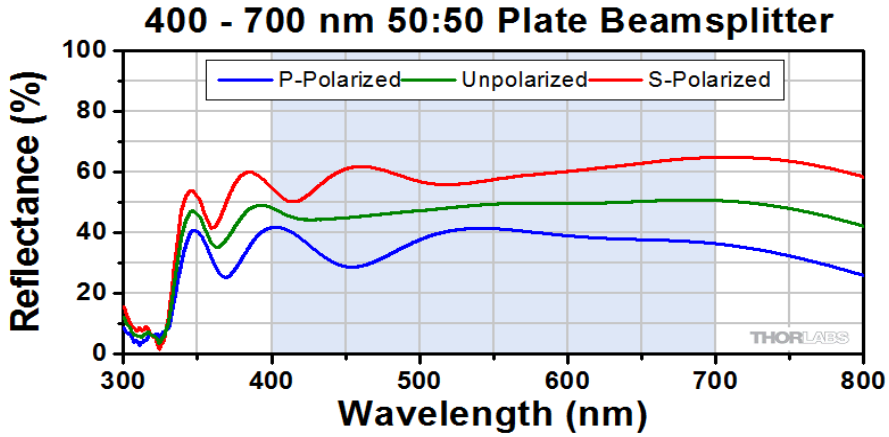


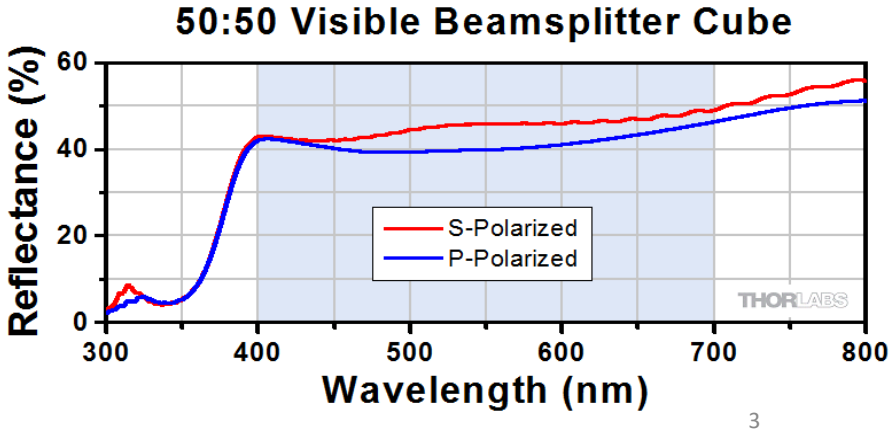
Beam-splitting plate
(Thorlabs BSW10R)

Beam-splitting cube
(Thorlabs BS013)



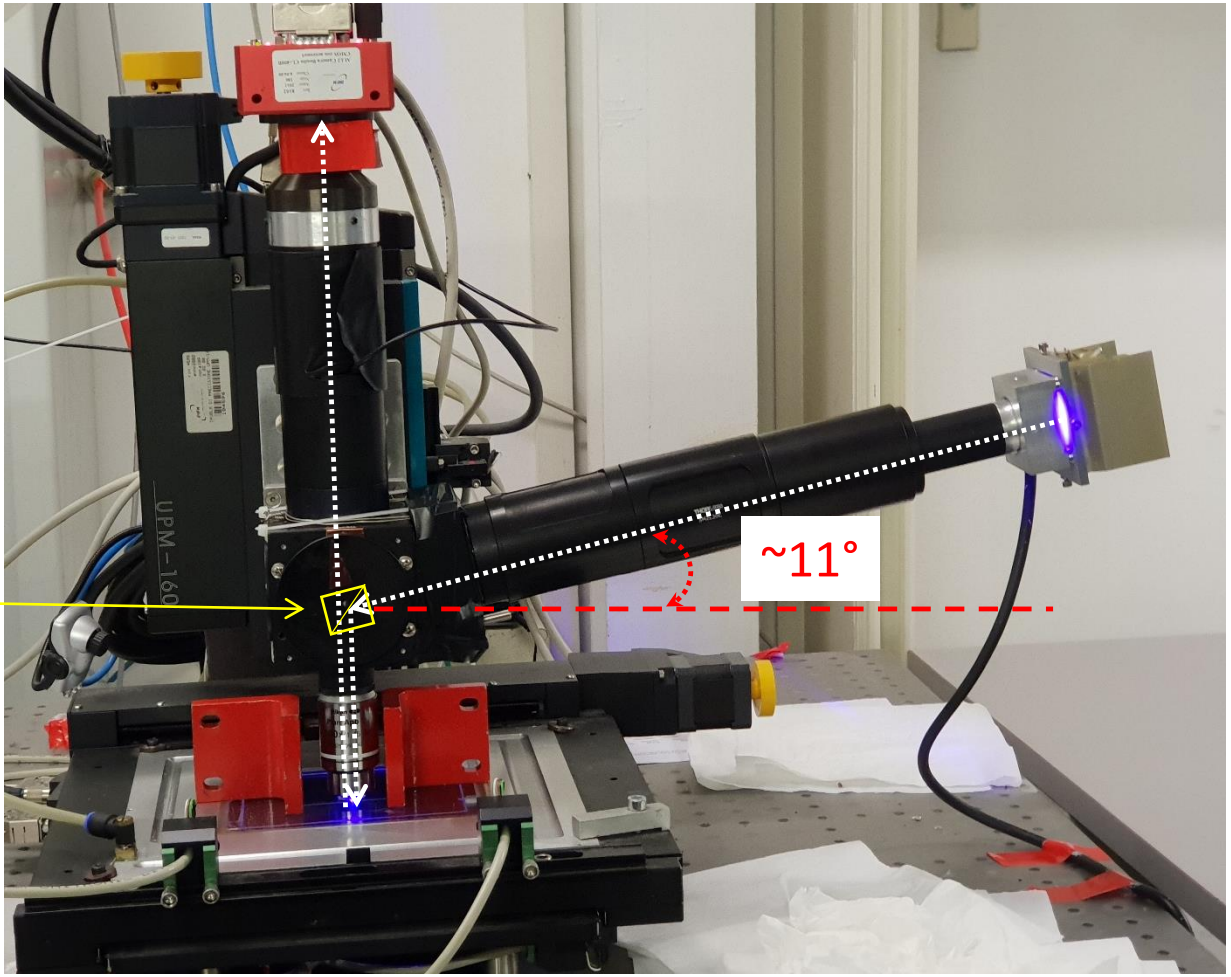




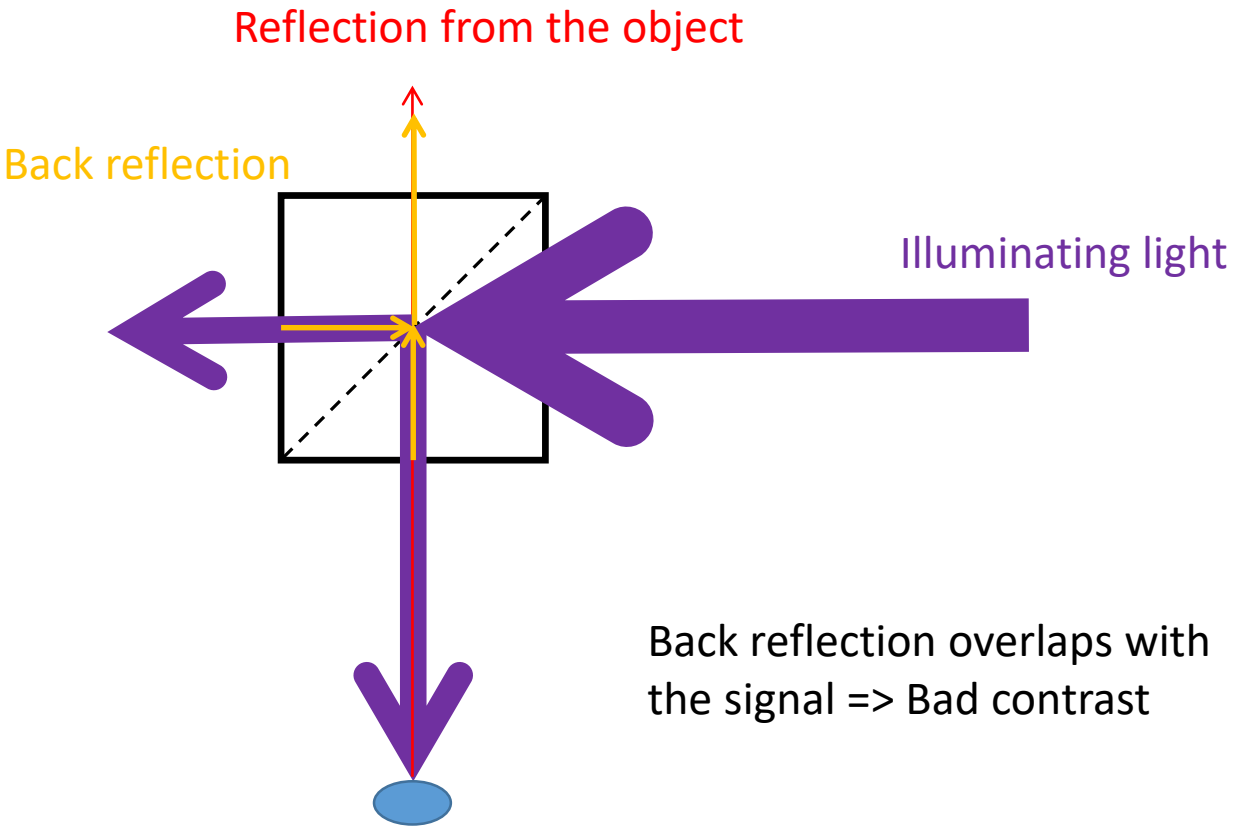


Upgraded microscope

Beam-splitting cube
(Thorlabs BS013)

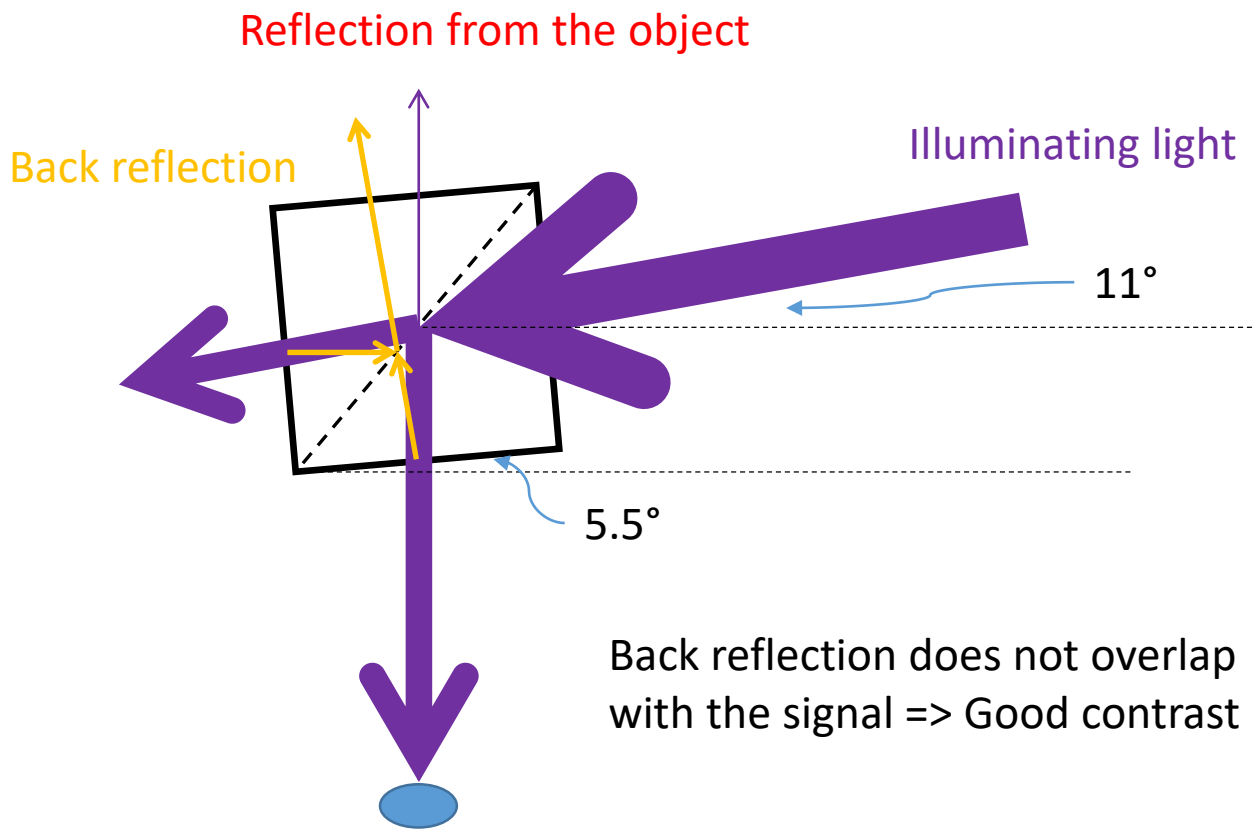


Direct BS mirror replacement

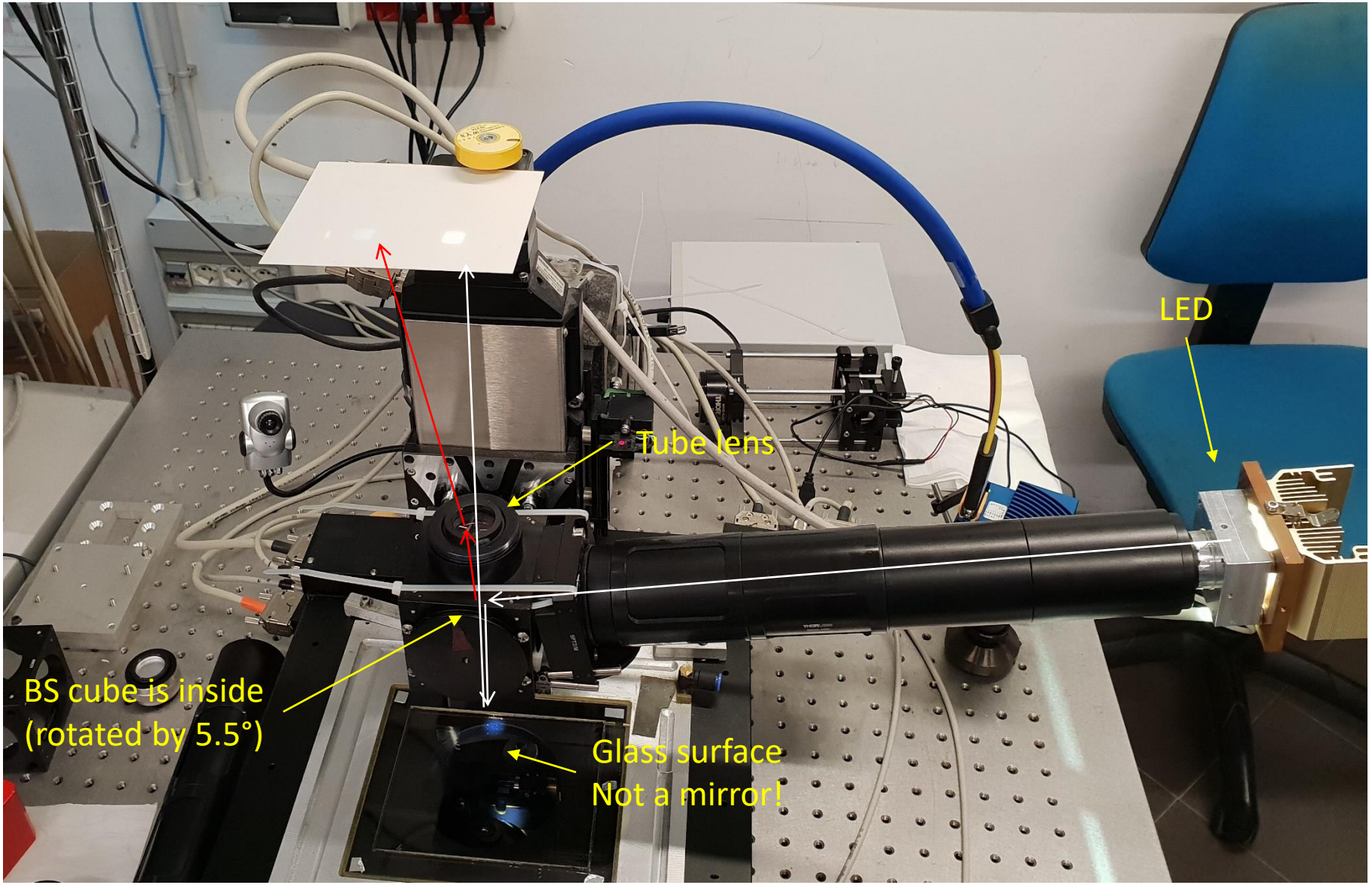


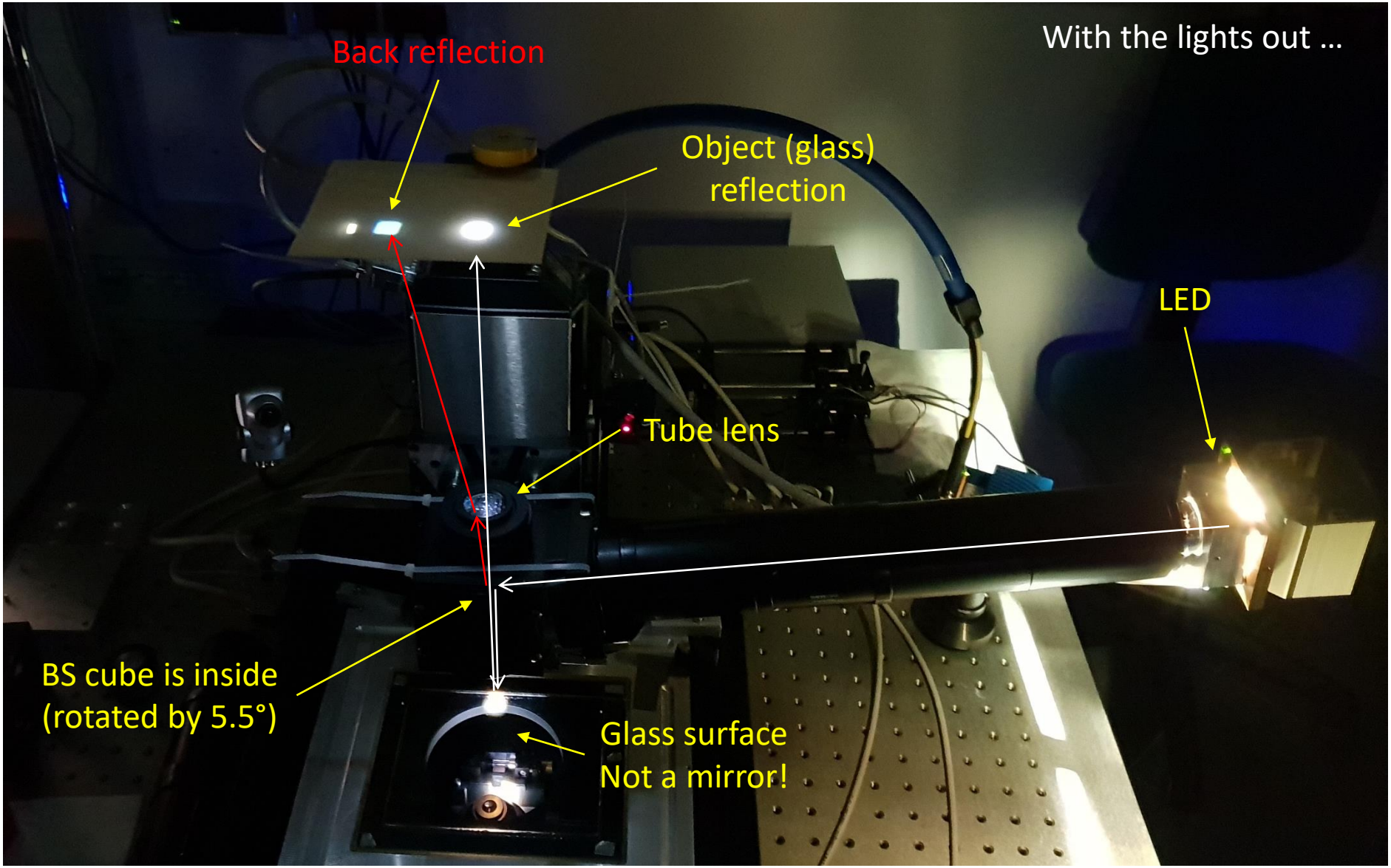
Back reflection overlaps with the signal => Bad contrast

After BS cube rotation

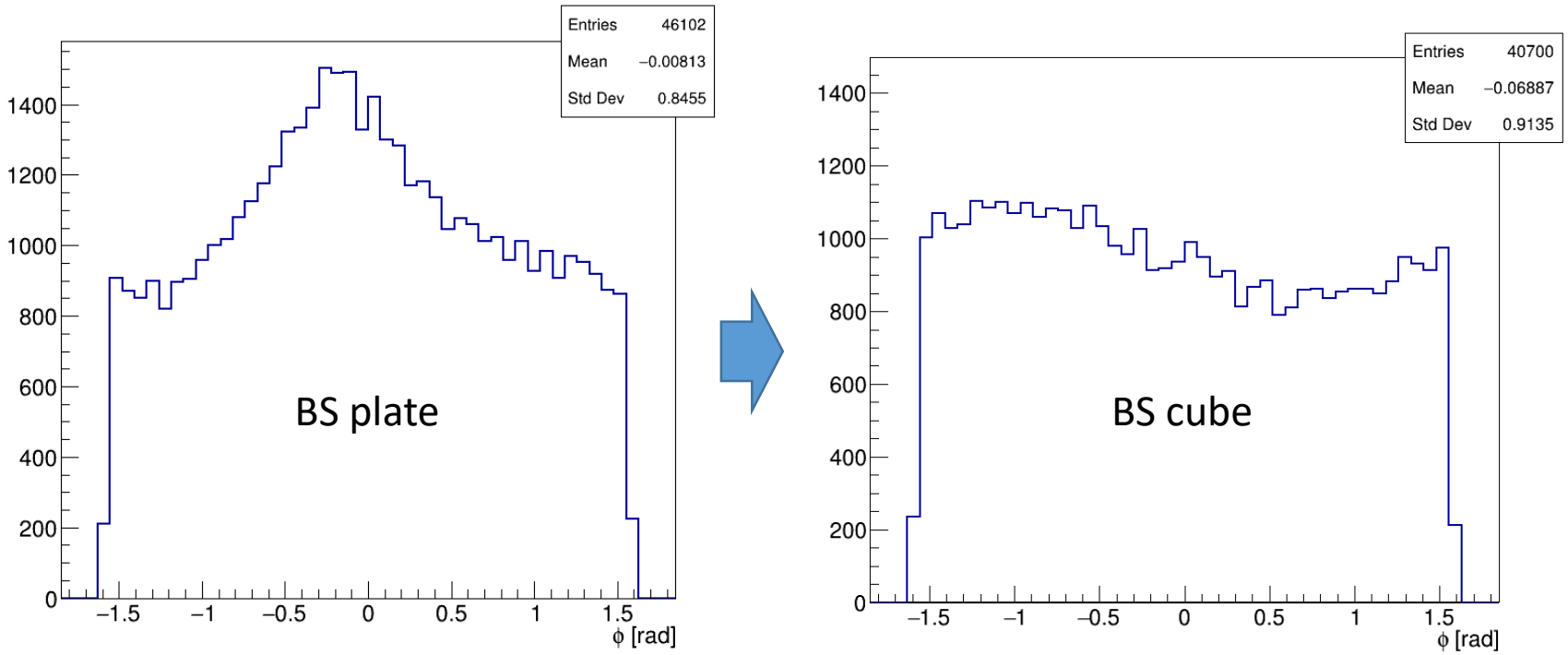


Back reflection does not overlap with the signal => Good contrast


LED
Tube lens
BS cube is inside
(rotated by 5.5°)
Glass surface
Not a mirror!


With the lights out …
Back reflection
Object (glass) reflection
LED
Tube lens
BS cube is inside (rotated by 5.5°)
Glass surface Not a mirror!

Upgrade result
(more details in Asada's and Valerio's talks)

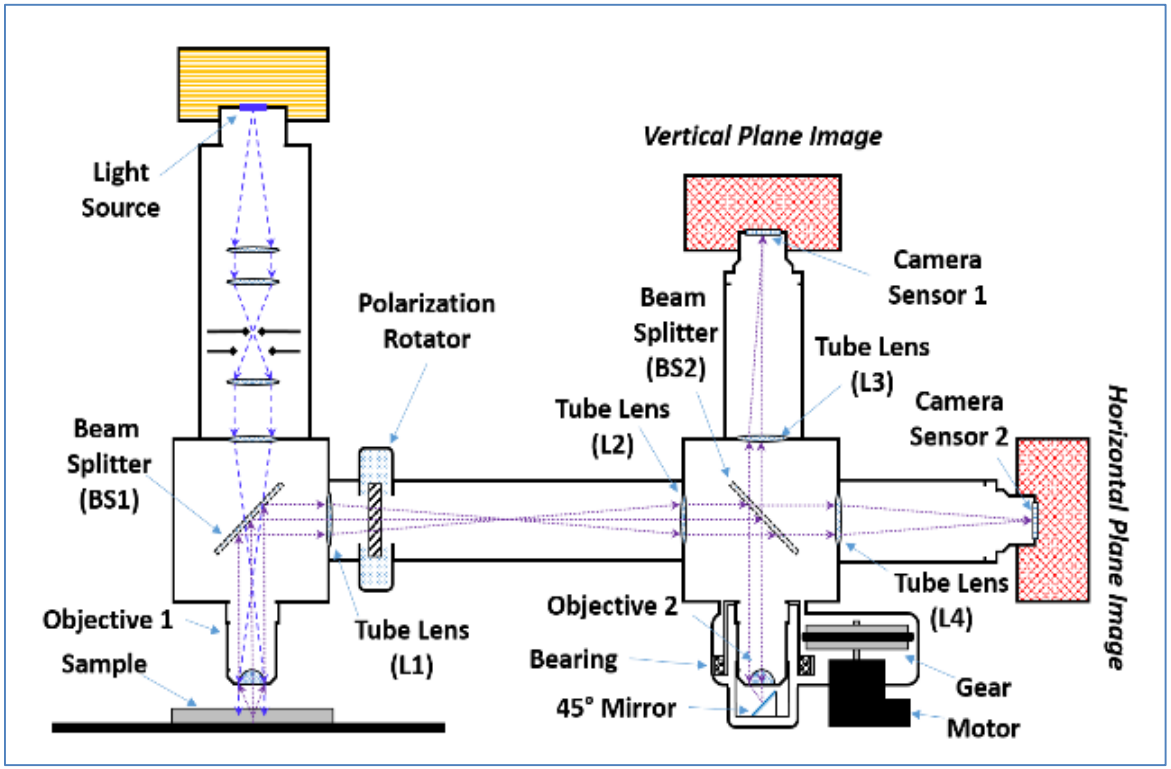


Direction from the barycentre shift

3D super-resolution microscope for color analysis

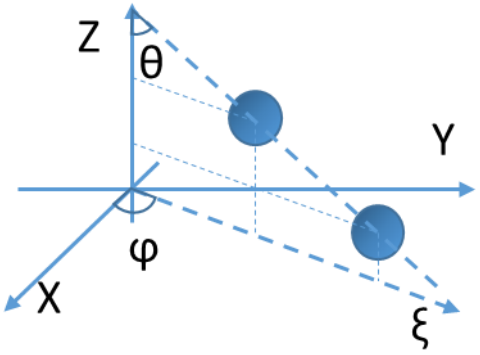
Andrey Alexandrov

3D microscope

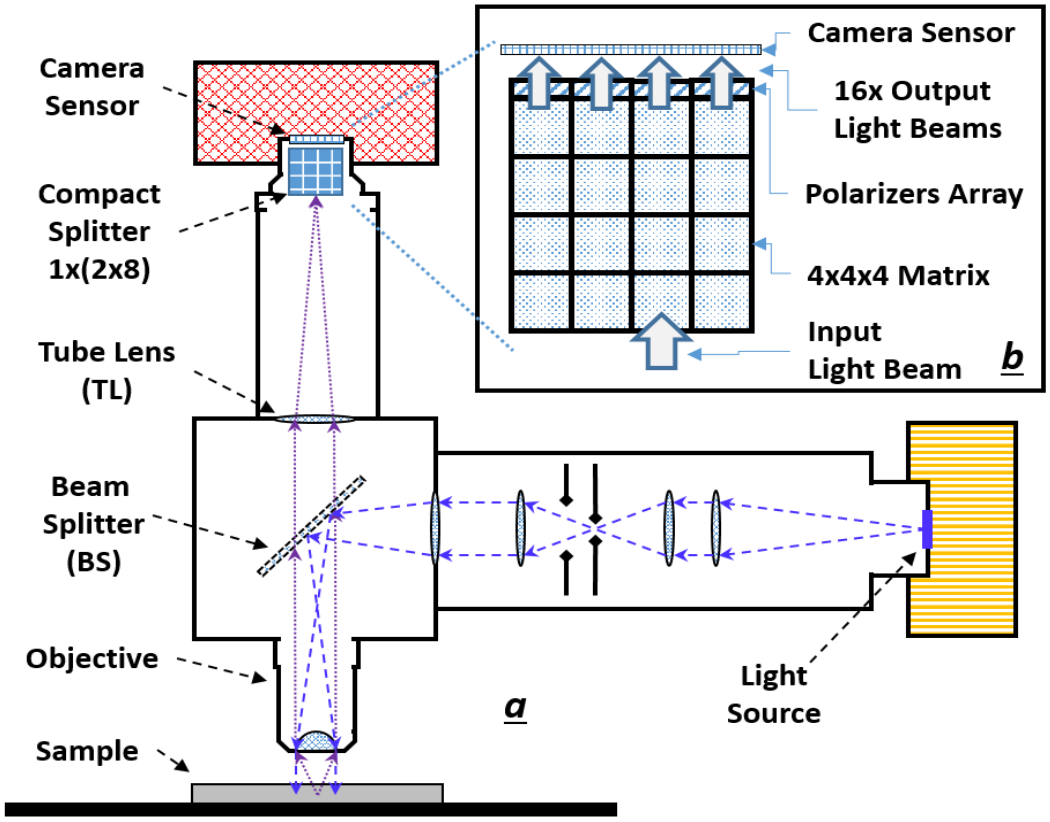


Weak Points:

- LC is not adapted for white light => no color analysis
- Slow LC operation => slow DAQ
- One image per polarization
  - Static position => slow DAQ
  - Sensitive to vibrations => Limited accuracy
- Two-phase measurement => slow DAQ

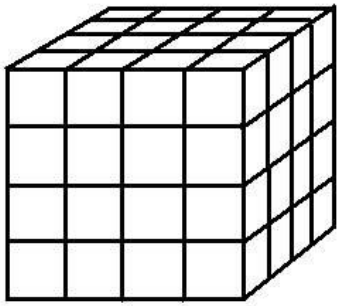


New 3D Super-Resolution Microscope

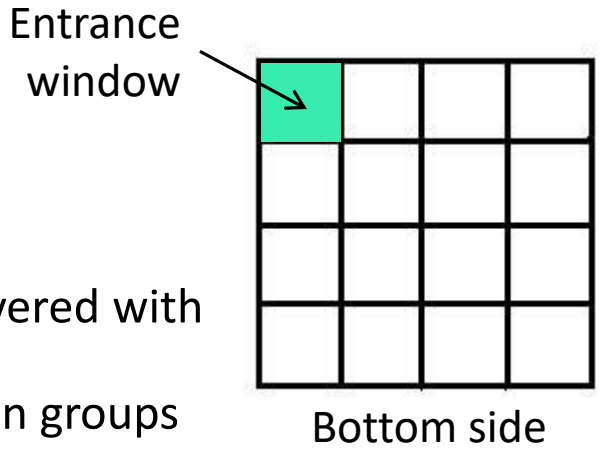


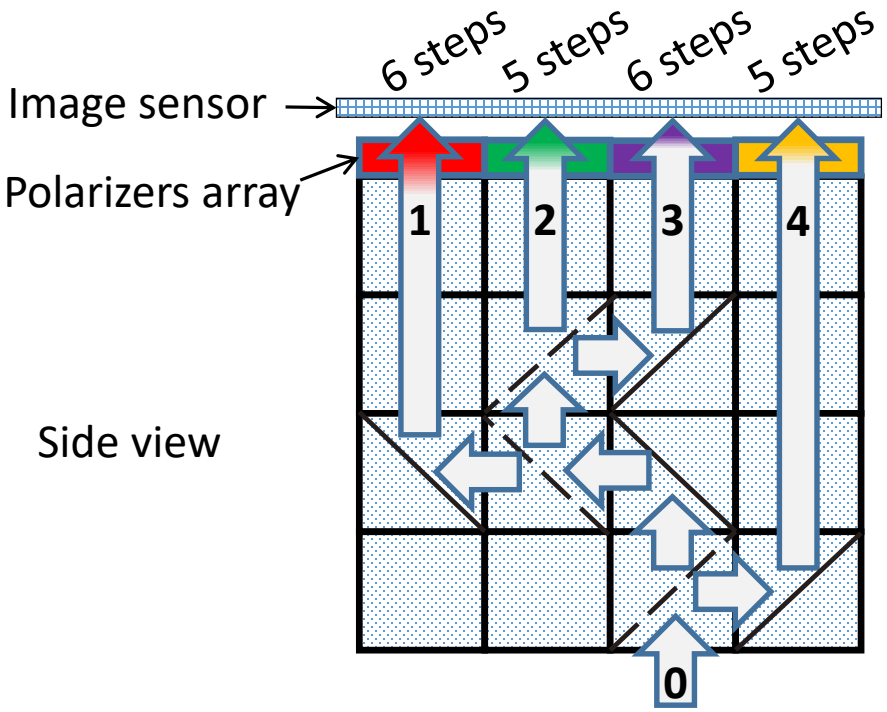
8 polarizations × 2 focal planes with a single shot!

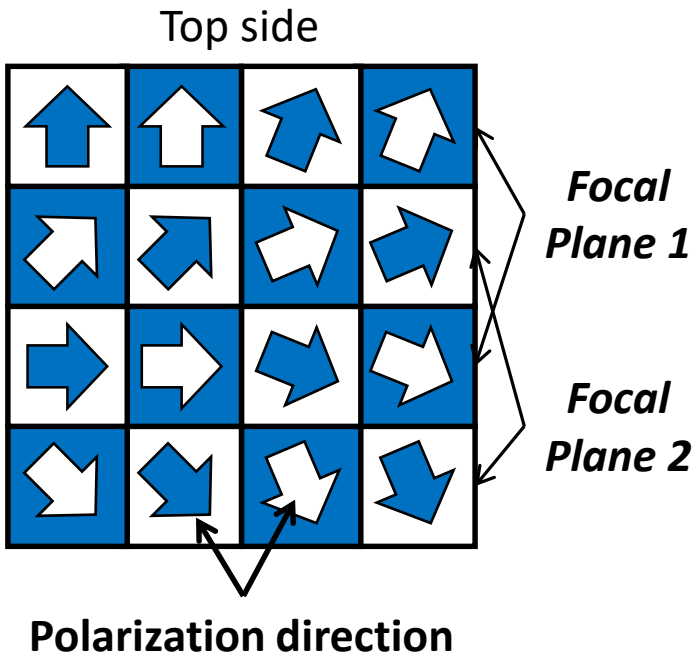
16x compact splitter



- 4x4x4 elements cube
- 5 mm x 5 mm x 5 mm elements
- 1 entrance window
- 16 exit windows in 2 groups (8+8) covered with static polarizers
- 1 step optical path difference between groups



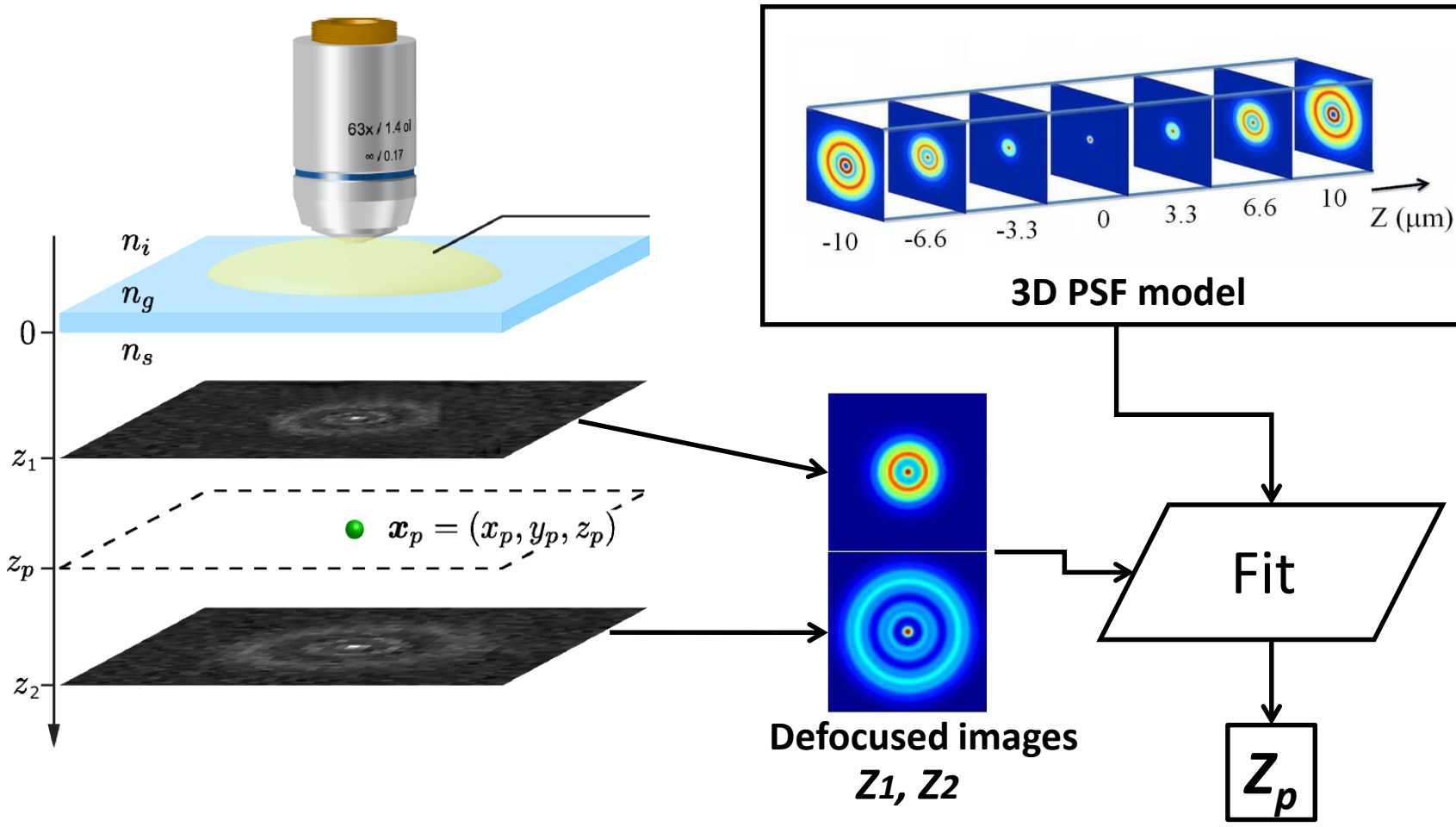




Mikrotron EoSens 25CXP color camera

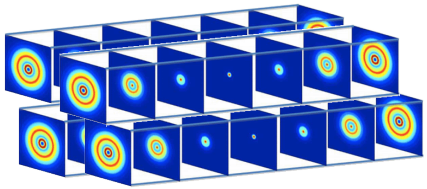
	EoSens® 25CXP+ (monochrome / color)
Resolution	25 Mpix
Active pixels	5,120 x 5,120 px
Interface	CoaXPress® @ 4 x 6.25 Gbit/s
Frame rate (8 bit)	80 fps
Sensor	OnSemi PYTHON 25k
Sensor type	CMOS global shutter
Sensor format	35 mm FF
Active sensor area (H x V)	23.04 x 23.04 mm
Pixel size	4.5 x 4.5 µm
Sensitivity (mono)	5.8 V/lux*s @ 550nm
Color depth	10 / 8 bit
Dynamic range	59 dB
Shutter time (steps)	1 µs
Shutter time range	1 µs – 0.1 s
Max. Jitter	±4 ns
Interface Connector	DIN 1.0 / DIN 2.3
Mount option	F-Mount
Dimensions (W x H x L w/o mount)	80 x 80 x 66 mm
Weight (w/o mount)	540 g
Power consumption	12 W
Power supply	12 - 24 V DC

Z-coordinate reconstruction
Dual-plane method

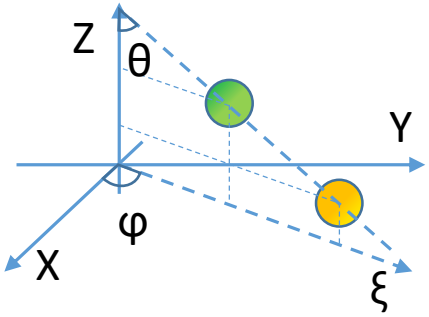


Biophys J. 95(12):6025-43 (2008)

Training

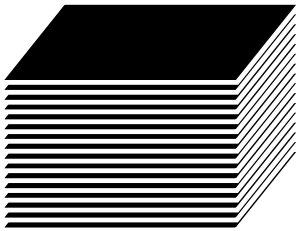


3D PSF collection



3D track model

Image generator

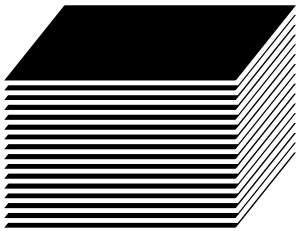


2*Npol Color Images

Neural Network

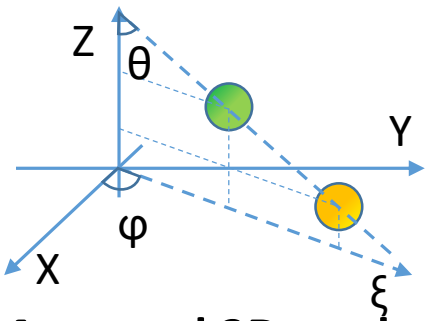
Measurement

DAQ



2*Np Color Images

Neural Network



Measured 3D track

Advantages of the new 3D SR microscope

- ✓Adapted for white light illumination -> color analysis
- ✓Image synchronization by design
- ✓Insensitive to vibrations -> better accuracy
- ✓3D+SR+color reconstruction in motion! -> faster DAQ
- ✓Simple design -> cheaper