

PSF

Microscope info:

Image		2026-06-15_63xResolution-1				
image's creation	date					
	method used					
Actual image depth		16				
Microscope type		WideField				
Objective	NA	1.4				
	im. refractive index	1.518				
Channel(s)	Wavelengths		Saturation	sampling (X,Y,Z)		
	Ex. (nm)	Em. (nm)		Nyquist (μm)	Found (μm)	Nyquist/found ratio
Channel 0		410.0	none	0.073x0.073x0.22	0.103x0.103x0.201	1.4, 1.4, 0.9
Channel 1		450.0	none	0.08x0.08x0.242		1.3, 1.3, 0.8
Channel 2		525.0	none	0.094x0.094x0.282		1.1, 1.1, 0.7
Channel 3		570.0	none	0.102x0.102x0.306		1.0, 1.0, 0.7

Warnings:

(No saturated pixels detected). The highlighted undersampled channels may alter the result interpretation. (A subresolution bead is used for all channels).

Resolution table:

Channel	Sig/Backgnd ratio	Dimension	Measured FWHM (μm)	theory (μm)	Fit Goodness	Mes./theory ratio
Channel 0 (em. 410.0nm)	4.0	X	0.434	0.149	0.98	2.91
		Y	0.533	0.149	0.97	3.57
		Z	0.906	0.562	0.99	1.61
Channel 1 (em. 450.0nm)	1.2	X	1.485	0.164	0.42	9.06
		Y	0.648	0.164	0.84	3.95
		Z	1.156	0.617	0.1	1.87
Channel 2 (em. 525.0nm)	4.9	X	0.589	0.191	0.96	3.08
		Y	0.654	0.191	0.96	3.42
		Z	1.187	0.72	0.99	1.65
Channel 3 (em. 570.0nm)	5.3	X	0.321	0.208	0.99	1.55
		Y	0.321	0.208	0.99	1.55
		Z	1.549	0.781	0.94	1.98

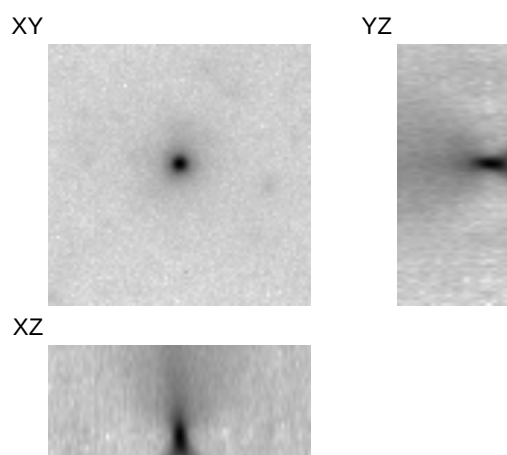
Green: within specifications, red: outside specifications (ie. XY ratios above 1.5 or Z ratio above 2.0)

Lateral asymmetry ratios:

Channel	Ratio
Channel 0 (em. 410.0nm)	0.81
Channel 1 (em. 450.0nm)	0.44
Channel 2 (em. 525.0nm)	0.9
Channel 3 (em. 570.0nm)	1.0

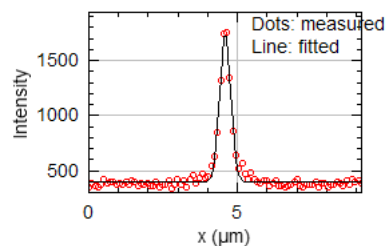
Detailed channel detection info:

Channel #0

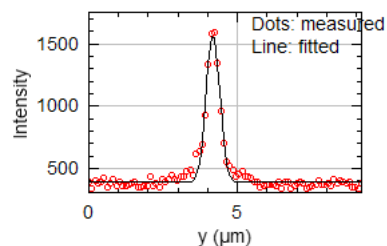


Channel 0 (em. 410.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
4.0	0.81	X	0.434	0.98
		Y	0.533	0.97
		Z	0.906	0.99

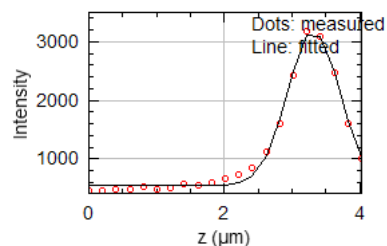
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
Sum of residuals squared: 124281.972
Standard deviation: 37.36879
R²: 0.97827
Parameters:
a = 392.68861
b = 1771.16789
c = 4.59378
d = 0.18441



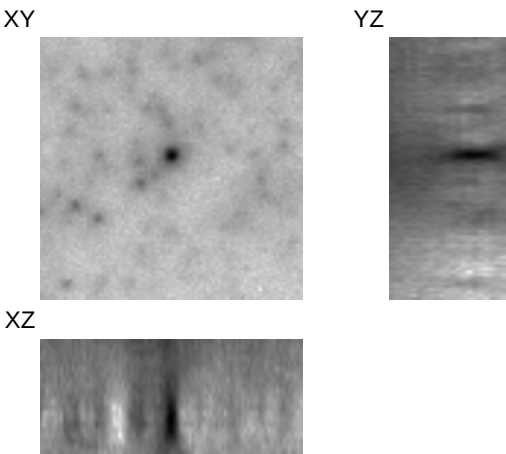
Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
Sum of residuals squared: 155221.313
Standard deviation: 41.76194
R²: 0.97014
Parameters:
a = 394.55477
b = 1585.84526
c = 4.17827
d = 0.22638



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
Sum of residuals squared: 113086.607
Standard deviation: 75.19528
R²: 0.99308
Parameters:
a = 541.67917
b = 3183.77642
c = 3.31775
d = 0.38456

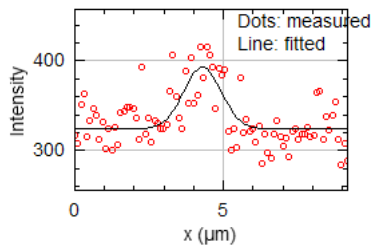


Channel #1

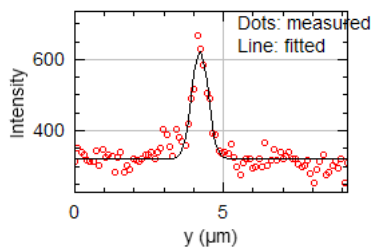


Channel 1 (em. 450.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
1.2	0.44	X	1.485	0.42
		Y	0.648	0.84
		Z	1.156	0.1

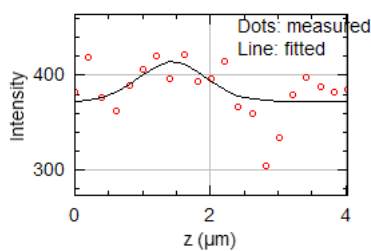
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
Sum of residuals squared: 47758.9122
Standard deviation: 23.16499
 R^2 : 0.44837
Parameters:
 $a = 324.20551$
 $b = 392.90404$
 $c = 4.29006$
 $d = 0.63061$



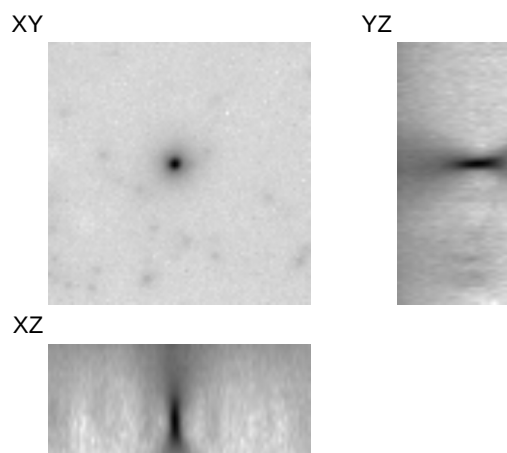
Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
Sum of residuals squared: 68461.1122
Standard deviation: 27.73492
 R^2 : 0.85020
Parameters:
 $a = 320.70011$
 $b = 623.75854$
 $c = 4.21179$
 $d = 0.27517$



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
Sum of residuals squared: 11913.2579
Standard deviation: 24.40621
 R^2 : 0.27184
Parameters:
 $a = 372.51292$
 $b = 414.31161$
 $c = 1.44270$
 $d = 0.49099$

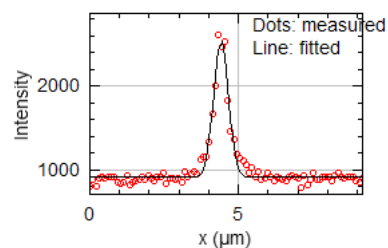


Channel #2

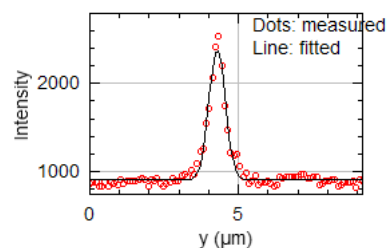


Channel 2 (em. 525.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
4.9	0.9	X	0.589	0.96
		Y	0.654	0.96
		Z	1.187	0.99

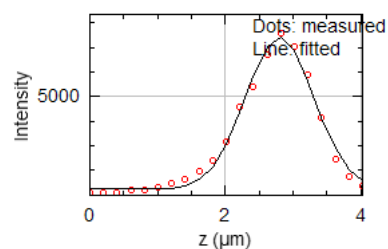
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)/(2*d*d))$
Sum of residuals squared: 442832.524
Standard deviation: 70.53826
R²: 0.95878
Parameters:
a = 909.41369
b = 2536.95517
c = 4.42045
d = 0.25029



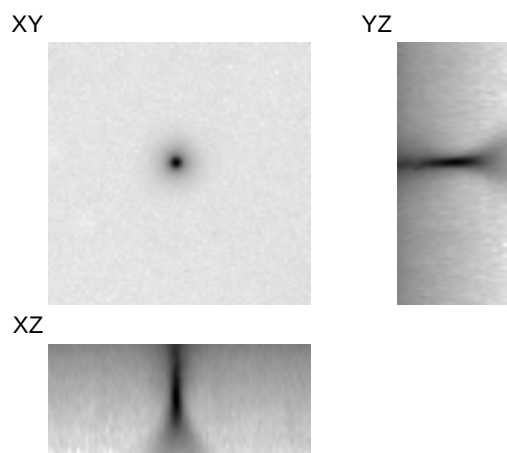
Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)/(2*d*d))$
Sum of residuals squared: 322864.143
Standard deviation: 60.23028
R²: 0.96654
Parameters:
a = 904.47977
b = 2383.29940
c = 4.28548
d = 0.27769



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)/(2*d*d))$
Sum of residuals squared: 1132777.68
Standard deviation: 237.98925
R²: 0.98858
Parameters:
a = 1237.23627
b = 7405.61853
c = 2.80300
d = 0.50427

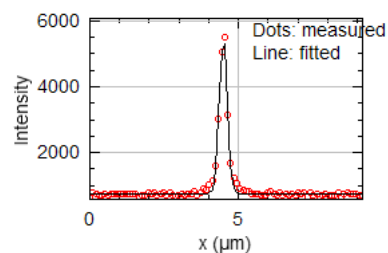


Channel #3

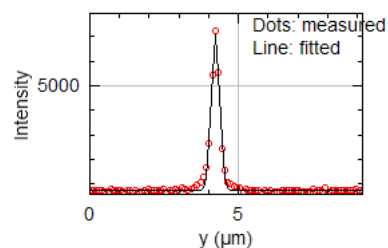


Channel 3 (em. 570.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
5.3	1.0	X	0.321	0.99
		Y	0.321	0.99
		Z	1.549	0.94

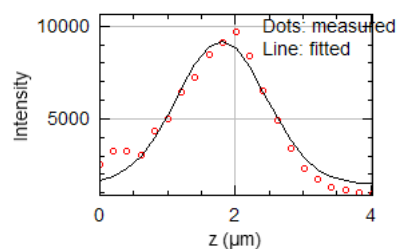
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)^2/(2*d*d))$
Sum of residuals squared: 697776.004
Standard deviation: 88.54479
R^2: 0.98653
Parameters:
a = 757.35434
b = 5556.69689
c = 4.49440
d = 0.13625



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)^2/(2*d*d))$
Sum of residuals squared: 1114428.36
Standard deviation: 111.90025
R^2: 0.98759
Parameters:
a = 769.43036
b = 7092.58093
c = 4.22873
d = 0.13624



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)^2/(2*d*d))$
Sum of residuals squared: 7377760.29
Standard deviation: 607.36152
R^2: 0.95475
Parameters:
a = 1489.19417
b = 9211.09092
c = 1.80051
d = 0.65762



Analysis parameters

Tool & Operator	Tool	PSF Profiler
	Versions	MetroloJ_QC v1.3.1.2, ImageJ v2.16.0/1.54p, Java v1.8.0_452, OS Windows 11
	Operator & date	Hanna, June 17, 2026 12:57 PM
data	result folder	Z:\CTIU\QC\2026-06-15\Thunder\Processed\Processed\PS F\
	Type of saved data	.pdf, .jpg, .xls
	Input data bit depth	16
Dimension order		XY-(C)Z
Discard saturated samples		false
Beads	Bead detection threshold	Legacy
	Center detection method	Legacy Fit Ellipses
	Discard bead if more than one particle are thresholded	false
	Background annulus thickness in μm	1.0
	Background annulus distance to bead edges in μm	0.5
	Multiple beads in image	false
Square Root PSF Image displayed		true
Tolerance	Applied in this report	true
	X & Y FWHM ratios valid if below	1.5
	Z FWHM ratio valid if below	2.0

Analysis log

image name	creation date	saturation	sampling density	status
2026-06-15_63xResolution-1		none	Ch.0,1,2,3 undersampled	analysed

Formulas used:

Lateral ($res_{x,y}^o$) and axial (res_z^o) theoretical resolution values used for widefield microscopes are calculated as defined in Wilhelm, S. Confocal Laser Scanning Microscopy, 2011:

$$res_{x,y}^o = \frac{0.51 \cdot \lambda_{em}}{NA} \quad res_z^o = \frac{1.77 n \cdot \lambda_{em}}{NA^2}$$

NA: numerical aperture, λ_{em} : emission wavelength, n: refractive index of the lens immersion & mounting media.

Axis profiles are fitted using ImageJ Gaussian Curve Fitter and the following formula $y = a + (b - a) * e^{\frac{-(x-c)^2}{2d^2}}$ (Gaussian fitting).

Measured lateral and axial resolution (Full Width at Half Maximum, FWHM) values are derived using $FWHM = 2d\sqrt{2\ln(2)}$

Compliance with the Shannon-Nyquist criterion uses the following formulas for Shannon-Nyquist distances calculation:

$$\alpha = \arcsin\left(\frac{NA}{n}\right)$$

$$\Delta_{x,y} = \frac{\lambda_{em}}{4 \cdot NA} \quad \Delta_z = \frac{\lambda_{em}}{2 \cdot n \cdot (1 - \cos(\alpha))}$$