

PSF4x

Microscope info:

Image		2026-06-15 4xResolution				
image's creation	date	2026-06-17 12:51:57				
	method used	from file creation date				
Actual image depth		16				
Microscope type		WideField				
Objective	NA	0.3				
	im. refractive index	1.0				
Channel(s)		Wavelengths		Saturation	sampling (X,Y,Z)	
		Ex. (nm)	Em. (nm)		Nyquist (μm)	Nyquist/found ratio
Channel 0			410.0	12.9%	0.342x0.342x4.451	1.9, 1.9, 0.4
Channel 1			450.0	9.5%	0.375x0.375x4.885	1.7, 1.7, 0.4
Channel 2			525.0	none	0.438x0.438x5.699	1.5, 1.5, 0.4
Channel 3			570.0	none	0.475x0.475x6.187	1.4, 1.4, 0.3

Warnings:

Saturation in highlighted channels may affect result. 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)	3.9	X	5.631	0.697	0.83	8.08
		Y	0.107	0.697	0.01	0.15
		Z	0.984	8.063	0.06	0.12
Channel 1 (em. 450.0nm)	4.1	X	4.078	0.765	0.77	5.33
		Y	0.325	0.765	0.01	0.42
		Z	8.437	8.85	0.01	0.95
Channel 2 (em. 525.0nm)	4.7	X	4.117	0.893	0.62	4.61
		Y	0.877	0.893	0.01	0.98
		Z	41.565	10.325	-0.01	4.03
Channel 3 (em. 570.0nm)	4.5	X	4.439	0.969	0.4	4.58
		Y	0.983	0.969	0.01	1.01
		Z	3.263	11.21	0.06	0.29

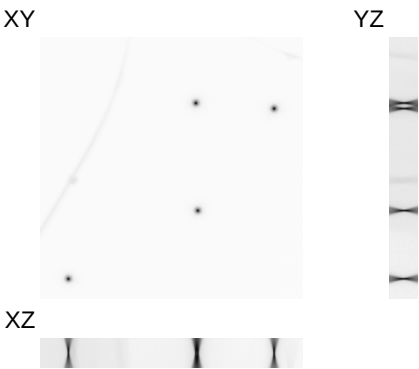
Green: within specifications, red: outside specifications (ie. XY ratios above 1.5 or Z ratio above 2.0)

Lateral asymmetry ratios:

Channel	Ratio
Channel 0 (em. 410.0nm)	0.02
Channel 1 (em. 450.0nm)	0.08
Channel 2 (em. 525.0nm)	0.21
Channel 3 (em. 570.0nm)	0.22

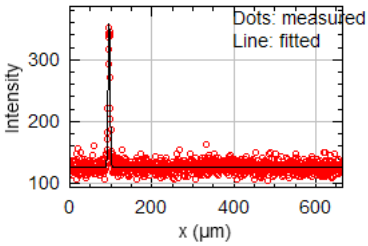
Detailed channel detection info:

Channel #0

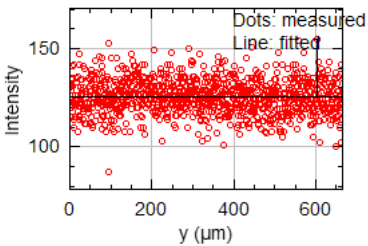


Channel 0 (em. 410.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
3.9	0.02	X	5.631	0.83
		Y	0.107	0.01
		Z	0.984	0.06

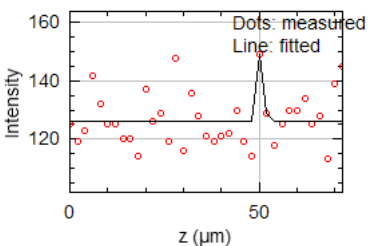
X profile & fitting parameters:
 Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
 Sum of residuals squared: 72950.0466
 Standard deviation: 8.44452
 R^2 : 0.82720
 Parameters:
 $a = 126.26002$
 $b = 359.17369$
 $c = 95.28414$
 $d = 2.39117$



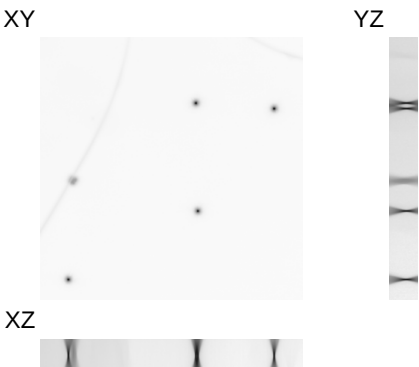
Y profile & fitting parameters:
 Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
 Sum of residuals squared: 76825.7498
 Standard deviation: 8.66594
 R^2 : 0.01119
 Parameters:
 $a = 125.50049$
 $b = 213152.315$
 $c = 604.04156$
 $d = 0.045392$



Z profile & fitting parameters:
 Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
 Sum of residuals squared: 2623.60000
 Standard deviation: 8.53685
 R^2 : 0.16274
 Parameters:
 $a = 126.20000$
 $b = 279.73539$
 $c = 50.78617$
 $d = 0.41790$

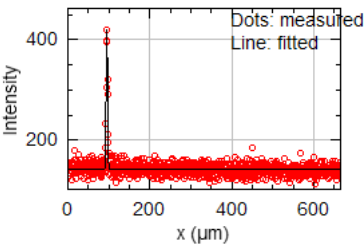


Channel #1

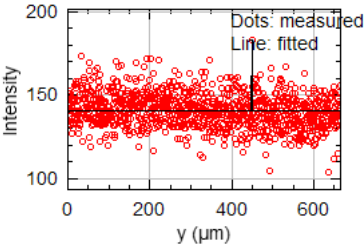


Channel 1 (em. 450.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
4.1	0.08	X	4.078	0.77
		Y	0.325	0.01
		Z	8.437	0.01

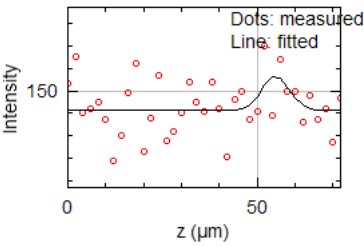
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
Sum of residuals squared: 108456.684
Standard deviation: 10.29652
 R^2 : 0.77094
Parameters:
a = 142.05539
b = 421.38497
c = 94.73442
d = 1.73161



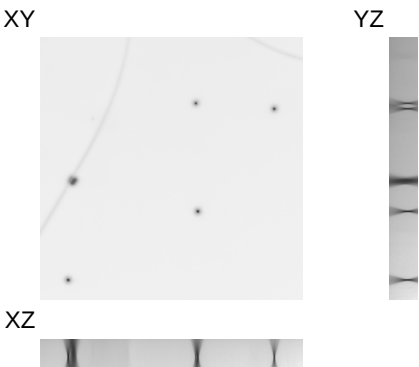
Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
Sum of residuals squared: 105497.205
Standard deviation: 10.15506
 R^2 : 0.01663
Parameters:
a = 140.93151
b = 379.45901
c = 448.75690
d = 0.13782



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
Sum of residuals squared: 4655.65522
Standard deviation: 11.37206
 R^2 : 0.11309
Parameters:
a = 141.44630
b = 156.46578
c = 54.81719
d = 3.58297

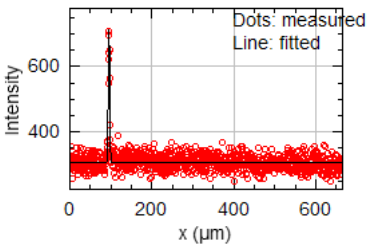


Channel #2

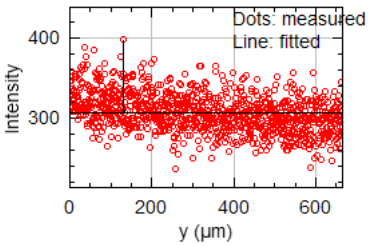


Channel 2 (em. 525.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
4.7	0.21	X	4.117	0.62
		Y	0.877	0.01
		Z	41.565	-0.01

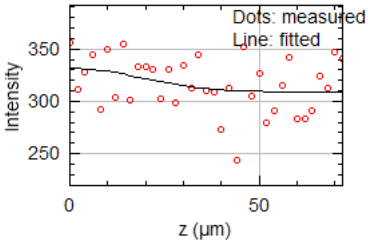
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
Sum of residuals squared: 473584.123
Standard deviation: 21.51596
 R^2 : 0.62182
Parameters:
 $a = 307.21899$
 $b = 713.23951$
 $c = 95.03934$
 $d = 1.74852$



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
Sum of residuals squared: 559163.946
Standard deviation: 23.37931
 R^2 : 0.01840
Parameters:
 $a = 307.52465$
 $b = 410.56217$
 $c = 131.10318$
 $d = 0.37224$



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
Sum of residuals squared: 22051.0909
Standard deviation: 24.74935
 R^2 : 0.09942
Parameters:
 $a = 308.94156$
 $b = 331.14411$
 $c = 1.29621$
 $d = 17.65102$

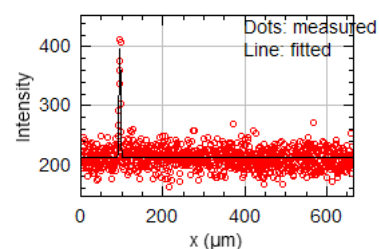


Channel #3

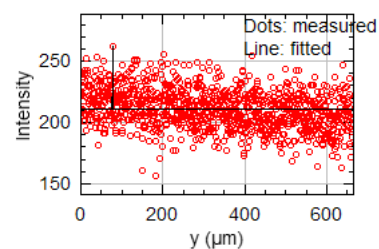


Channel 3 (em. 570.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
4.5	0.22	X	4.439	0.4
		Y	0.983	0.01
		Z	3.263	0.06

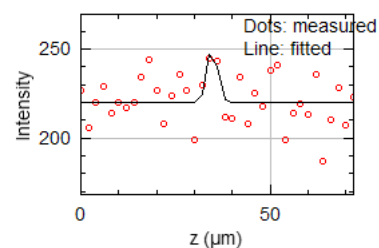
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \cdot \exp(-(x-c)/(2 \cdot d))$
Sum of residuals squared: 265791.434
Standard deviation: 16.11880
R²: 0.40059
Parameters:
a = 211.90014
b = 398.72233
c = 94.64073
d = 1.88528



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \cdot \exp(-(y-c)/(2 \cdot d))$
Sum of residuals squared: 268457.143
Standard deviation: 16.19943
R²: 0.01164
Parameters:
a = 211.12614
b = 263.16236
c = 77.28559
d = 0.41756



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \cdot \exp(-(z-c)/(2 \cdot d))$
Sum of residuals squared: 5670.99386
Standard deviation: 12.55100
R²: 0.16222
Parameters:
a = 219.97113
b = 251.08531
c = 34.68684
d = 1.38581



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 1:04 PM
data	result folder	Z:\CTIU\QC\2026-06-15\Thunder\Processed\PSF4x\
	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_4xResolution	2026-06-17 12:51:57	Ch.0,1 saturated	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:

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

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