

PSF

Microscope info:

Image		2026-06-16_40xResolution				
image's creation	date	unknown				
	method used	original file info & metadata could not be found				
Actual image depth		16				
Microscope type		WideField				
Objective	NA	1.3				
	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		405.0	none	0.078x0.078x0.276	0.01x0.01x1.0	0.1, 0.1, 3.6
Channel 1		450.0	none	0.087x0.087x0.306		0.1, 0.1, 3.3
Channel 2		525.0	none	0.101x0.101x0.358		0.1, 0.1, 2.8
Channel 3		570.0	none	0.11x0.11x0.388		0.1, 0.1, 2.6

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/Background ratio	Dimension	Measured FWHM (μm)	theory (μm)	Fit Goodness	Mes./theory ratio
Channel 0 (em. 405.0nm)	1.6	X	78.401	0.159	0.65	493.44
		Y	168.179	0.159	0.78	1058.5
		Z	14071.521	0.644	0.42	21853.84
Channel 1 (em. 450.0nm)	1.4	X	0.044	0.177	0.9	0.25
		Y	2.95	0.177	0.91	16.71
		Z	0.545	0.715	-0.02	0.76
Channel 2 (em. 525.0nm)	1.2	X	0.07	0.206	0.56	0.34
		Y	2.772	0.206	0.88	13.46
		Z	22.324	0.835	0.1	26.75
Channel 3 (em. 570.0nm)	1.6	X	6.529	0.224	0.77	29.2
		Y	69.424	0.224	0.71	310.46
		Z	21.23	0.906	0.54	23.43

The annulus drawn around the bead to compute the background mean intensity is bigger than the image (channel 3). Green: within specifications, red: outside specifications (ie. XY ratios above 1.5 or Z ratio above 2.0)

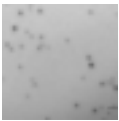
Lateral asymmetry ratios:

Channel	Ratio
Channel 0 (em. 405.0nm)	0.47
Channel 1 (em. 450.0nm)	0.01
Channel 2 (em. 525.0nm)	0.03
Channel 3 (em. 570.0nm)	0.09

Detailed channel detection info:

Channel #0

XY



YZ

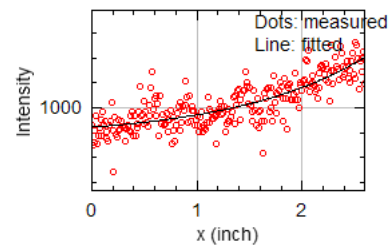


XZ

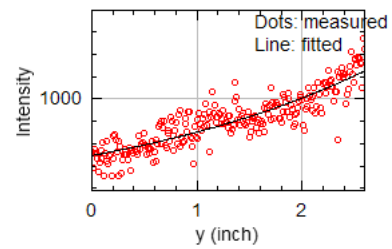


Channel 0 (em. 405.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
1.6	0.47	X	78.401	0.65
		Y	168.179	0.78
		Z	14071.521	0.42

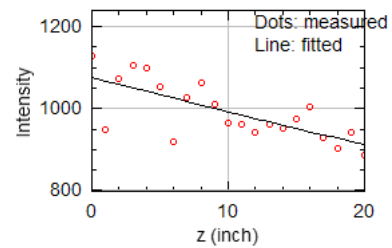
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)/(2*d*d))$
Sum of residuals squared: 801892.251
Standard deviation: 56.74902
R^2: 0.65770
Parameters:
a = 882.10004
b = 1.25855E154
c = 882.27757
d = 33.29374



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)/(2*d*d))$
Sum of residuals squared: 833662.109
Standard deviation: 57.86226
R^2: 0.78151
Parameters:
a = 510.68448
b = 1.34078E154
c = 1888.02394
d = 71.41891

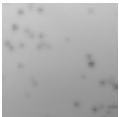


Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)/(2*d*d))$
Sum of residuals squared: 46432.2881
Standard deviation: 48.18313
R^2: 0.52936
Parameters:
a = -874.92460
b = 1.05173E154
c = -157437.519
d = 5975.62470



Channel #1

XY



YZ

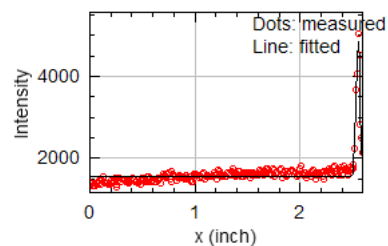


XZ

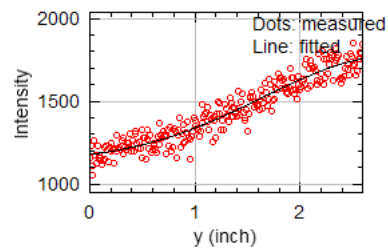


Channel 1 (em. 450.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
1.4	0.01	X	0.044	0.9
		Y	2.95	0.91
		Z	0.545	-0.02

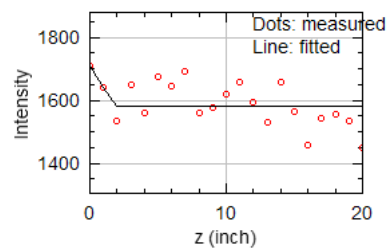
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)/(2*d*d))$
Sum of residuals squared: 3521100.75
Standard deviation: 118.91580
R^2: 0.90489
Parameters:
a = 1567.73742
b = 4863.02568
c = 2.55161
d = 0.018598



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)/(2*d*d))$
Sum of residuals squared: 870583.949
Standard deviation: 59.12970
R^2: 0.91016
Parameters:
a = 1134.82838
b = 1775.74598
c = 2.89671
d = 1.25257

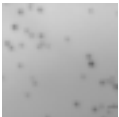


Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)/(2*d*d))$
Sum of residuals squared: 85646.0000
Standard deviation: 65.43928
R^2: 0.17149
Parameters:
a = 1581.99999
b = 2487.81307
c = 0.45941
d = 0.23130



Channel #2

XY



YZ

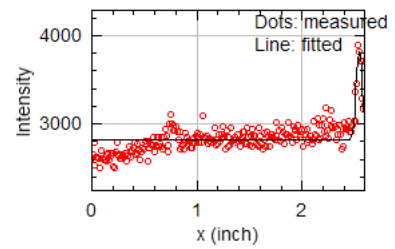


XZ

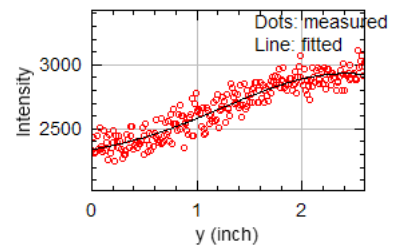


Channel 2 (em. 525.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
1.2	0.03	X	0.07	0.56
		Y	2.772	0.88
		Z	22.324	0.1

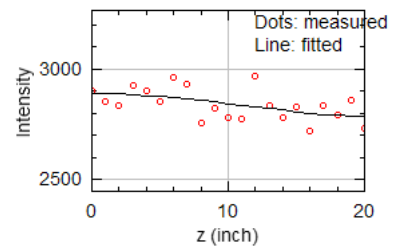
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)/(2*d*d))$
Sum of residuals squared: 3719186.75
Standard deviation: 122.21495
R^2: 0.56303
Parameters:
a = 2817.27314
b = 3814.02529
c = 2.54340
d = 0.029576



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)/(2*d*d))$
Sum of residuals squared: 1443558.09
Standard deviation: 76.14080
R^2: 0.87904
Parameters:
a = 2256.86686
b = 2931.71942
c = 2.42342
d = 1.17699

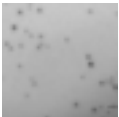


Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)/(2*d*d))$
Sum of residuals squared: 76321.3804
Standard deviation: 61.77434
R^2: 0.26796
Parameters:
a = 2900.82081
b = 2782.86944
c = 21.36325
d = 9.48014



Channel #3

XY



YZ

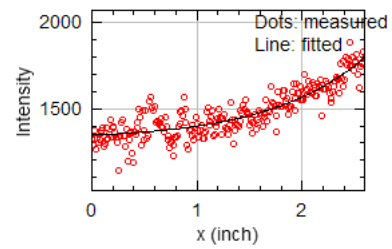


XZ

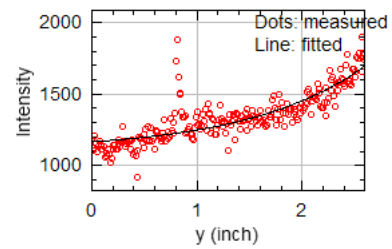


Channel 3 (em. 570.0nm)				
Sig./Backgnd ratio	LAR	Dimension	FWHM	Fit goodness
1.6	0.09	X	6.529	0.77
		Y	69.424	0.71
		Z	21.23	0.54

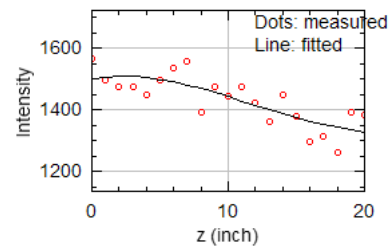
X profile & fitting parameters:
Fit equation $I(x) = a + (b-a) \exp(-(x-c)/(2*d*d))$
Sum of residuals squared: 1100908.73
Standard deviation: 66.49301
R^2: 0.77742
Parameters:
a = 1328.29389
b = 42036.3056
c = 10.88284
d = 2.77279



Y profile & fitting parameters:
Fit equation $I(y) = a + (b-a) \exp(-(y-c)/(2*d*d))$
Sum of residuals squared: 2155151.72
Standard deviation: 93.03348
R^2: 0.71620
Parameters:
a = 1107.87091
b = 1.34078E154
c = 780.94624
d = 29.48151



Z profile & fitting parameters:
Fit equation $I(z) = a + (b-a) \exp(-(z-c)/(2*d*d))$
Sum of residuals squared: 50031.6425
Standard deviation: 50.01582
R^2: 0.62801
Parameters:
a = 1300.31327
b = 1509.05394
c = 2.11319
d = 9.01556



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, July 2, 2026 10:00 AM
data	result folder	Z:\CTIU\QC\2026-06-16\Evident\Processed\PSF\40x\Processed\PSF\
	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
2026-06-16_40xResolution	unknown	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:

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

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