

Formulas used:

Axial (res_z^0) theoretical resolution value used for widefield microscopes is calculated as defined in Wilhelm, S. Confocal Laser Scanning Microscopy, 2011:

$$res_z^0 = \frac{1,77n*\lambda_{em}}{NA^2}$$

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

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

Measured axial resolution (Full Width at Half Maximum, FWHM) value is derived using $FWHM = 2d\sqrt{2\ln(2)}$

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

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

$$\Delta_z = \frac{\lambda_{em}}{2.n.(1-\cos(\alpha))}$$