Formulas used:

Lateral ($res^{o}_{x,y}$) and axial (res^{o}_{z}) theoretical resolution values used for single point scanning multiphoton excitation microscopes are calculated as defined in Zipfel, W.R., Willams, R.M. & Webb, W.W. Nonlinear magic: multiphoton microscopy in the biosciences, Nature Biotechnology 21, 1369–1377 (2003):

$$NA \leq 0.7 \quad res^{o}_{x,y} = \frac{0.377 * \lambda_{ex}}{NA} \quad res^{o}_{z} = \frac{0.626 * \lambda_{ex}}{n - \sqrt{n^2 - NA^2}}$$

$$NA > 0.7 \quad res^{o}_{x,y} = \frac{0.383 * \lambda_{ex}}{NA^{0.91}} \quad res^{o}_{z} = \frac{0.626 * \lambda_{ex}}{n - \sqrt{n^2 - NA^2}}$$

NA: numerical aperture, λ_{ex}: excitation 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 $\mathrm{FWHM} = 2d\sqrt{2ln(2)}$

Compliance with the Shannon-Nyquist criterion for k photon excitation uses the following formulas for Shannon-Nyquist distances calculation (and a fixed k value of 2):

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

$$\Delta_{x,y} = \frac{\lambda_{ex}}{4.k.NA} \qquad \Delta_{z} = \frac{\lambda_{ex}}{4.n.\,(1 - cos\,(\alpha))}$$