Calibration of z-localization for SML

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The fluorescent spots in each image frame of a calibration experiment are fitted with the following Gaussian function:

$$G(x,y) = I_0 \exp\left[-2\frac{(x-x_0)^2}{w_x^2} - 2\frac{(y-y_0)^2}{w_y^2}\right] + b \tag{1}$$

where I_0 is the peak height, b is the background, (x_0, y_0) is the center position of the peak, and w_x and w_y are the widths of the spot PSF (point spread function) in the x and y directions. The z position of the particle can be extracted from solving the following equation

$$w_{x,y}(z) = w_0 \left[1 + \left(\frac{z-c}{d}\right)^2 + A\left(\frac{z-c}{d}\right)^3 + B\left(\frac{z-c}{d}\right)^4\right]^{1/2} \tag{2}$$

where the parameters w_0, A, B, c, and d are determined from the experimental calibration curves of $w_{x,y}(z)$. Now suppose that these parameters have corresponding errorbars: δw_0, δA, δB, δc, and δd. We estimate the errorbars of $w_{x,y}(z)$ as follows.

We decompose Eq.(2) as

$$w_{x,y}(z) = w_0 f^{1/2}, \qquad f = 1 + \xi^2 + A\xi^3 + B\xi^4, \qquad \xi = \frac{z-c}{d} \tag{3}$$

The error estimate of ξ is

$$\xi = \frac{z-c}{d} = \frac{z}{d} - \frac{c}{d} \tag{4}$$

$$\begin{aligned}(\delta\xi)^2 &= \left(\frac{z}{d}\right)^2\left(\frac{\delta d}{d}\right)^2 + \left(\frac{c}{d}\right)^2\left[\left(\frac{\delta c}{c}\right)^2 + \left(\frac{\delta d}{d}\right)^2\right] \\ &= \left[\left(\frac{z}{d}\right)^2 + \left(\frac{c}{d}\right)^2\right]\left(\frac{\delta d}{d}\right)^2 + \left(\frac{\delta c}{d}\right)^2\end{aligned} \tag{5}$$

The error estimate of f is

$$\begin{aligned}(\delta f)^2 &= (2\xi\delta\xi)^2 + (\xi^3\delta A)^2 + (3A\xi^2\delta\xi)^2 + (\xi^4\delta B)^2 + (4B\xi^3\delta\xi)^2 \\ &= [(2\xi)^2 + (3A\xi^2)^2 + (4B\xi^3)](\delta\xi)^2 + (\xi^3\delta A)^2 + (\xi^4\delta B)^2\end{aligned} \tag{6}$$

Finally we have

$$(\delta w_{x,y})^2 = w_0^2 f\left[\left(\frac{\delta w_0}{w_0}\right)^2 + \frac{1}{4}\left(\frac{\delta f}{f}\right)^2\right] = f(\delta w_0)^2 + \frac{w_0^2}{4}\frac{(\delta f)^2}{f} \tag{7}$$