This technical note describes a change in the cell-by-cell analysis in CytoSpectre 1.2. In versions 1.0 and 1.1, the cell-by-cell analysis was implemented by setting all background pixels to zero prior to spectral estimation. Each cell in turn was analyzed by treating all other cells as background. The drawback of this approach is that in some images, the sharp edge created at the cell boundaries causes ringing artefacts in the spectral estimate, which can lead to distorted results.

A more robust approach for performing cell-by-cell analysis was implemented in version 1.2. Instead of setting all pixels that are not part of the target cell to zero, the image is weighted with a Tukey’s tapered cosine window [1] centered on the target cell. The edges of the window follow a cosine function and thus produce smoothly decreasing intensities at the cell boundaries. This avoids disrupting the spectral estimation. On the other hand, the flat top of Tukey’s window gives equal weight to most pixels within the cell region, as opposed to e.g. a Hann window.

For each cell labeled in the segmentation mask, analyzed in turn, the new procedure is as follows:

1. The bounding box of the cell is computed.
2. The bounding box is extended along its shorter dimension from both ends such that the side lengths become equal. A square box $B$, having the size $N_B \times N_B$, is thus obtained.
3. A 1D Tukey’s window is constructed using the MATLAB function $\text{tukeywin}$. The window length $L$ is $20N_B + 1$ and the parameter $r$ controlling the ratio of cosine-tapered section length to the entire window length is 0.5.
4. A 2D circularly symmetric Tukey’s window evaluated at the grid points of an $N_B \times N_B$ grid is obtained from the 1D window via linear interpolation.
5. The 2D window is placed at the location of $B$ and padded with zeros to produce a window image $W$ having equal dimensions with the input image $I$. If parts of the 2D window extend beyond the image boundaries, these parts are cropped.
6. A weighted input image is obtained as the pixel-wise product $I^\wedge = I \circ W$.

The rest of the analysis proceeds as described in the CytoSpectre paper [2] for version 1.0.
