

Optimizing reference controls for multicolor flow cytometry

Best practices for preventing compensation or unmixing errors



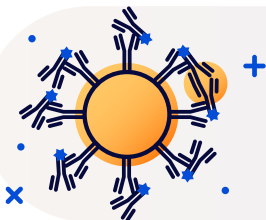
Cell Treatment

Have the reference controls and sample been treated, processed and stained using the same protocol, buffers, fluorochrome and tandem batch?



Rare Cells

Is the number of available cells sufficient to record all reference controls, including rare populations?



Beads

Have beads been tested and validated for the ability to create a compensation or unmixing matrix similar to cells?



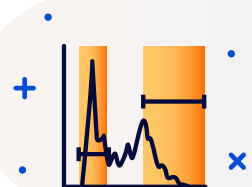
Autofluorescence

Does each reference control have a negative and positive population with the same autofluorescence?



Brightness

Is the positive population of each reference control as bright or brighter than the sample?



Gating

Have you excluded debris and doublets and gated only on a statistically significant number of cells with the highest level of expression?



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