Flow Post-its
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Are All Fluorochromes Equal?

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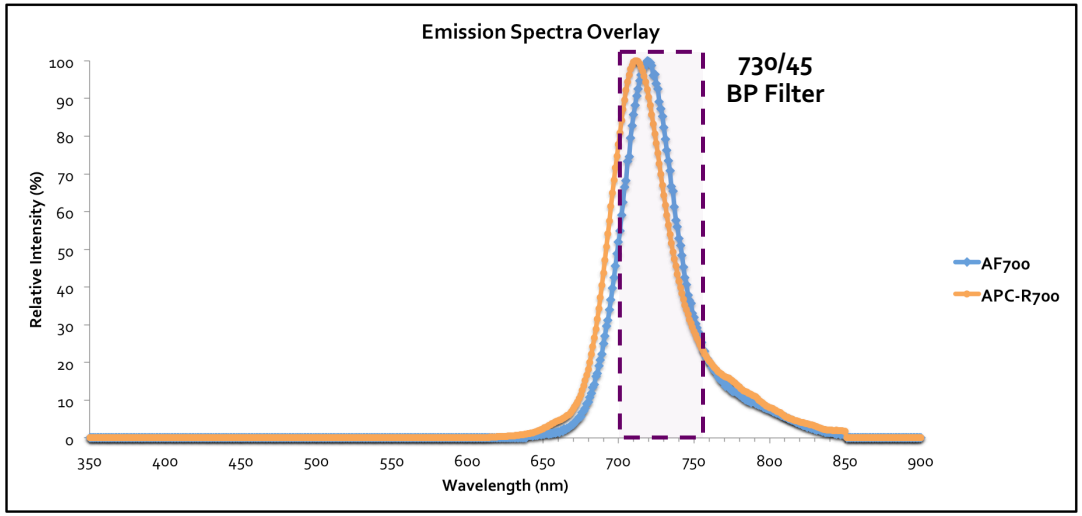
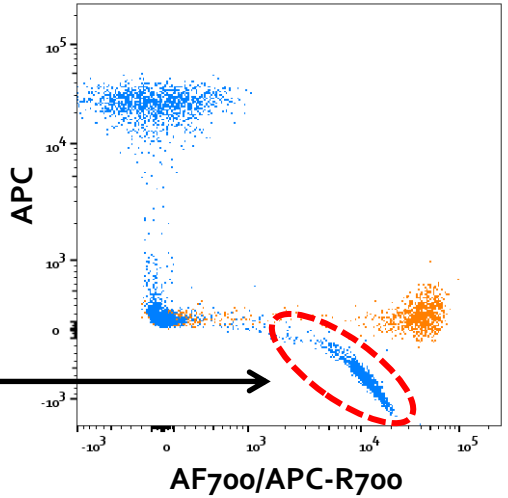
In order to accurately analyze multicolor flow cytometry data, **spectral overlap** must be corrected with compensation or unmixing. When running the single stain controls, it is essential that the **fluorochromes match** those in your experimental samples. Failure to follow this best practice will result in incorrect data.

Case Study: AF700 & APC-R700…One and the same?

- Two color experiment was planned with APC and AF700. Compensation was needed and **APC-R700** was run as a single color in replacement of **AF700**.
- Emission of both fluorochromes show an almost identical spectra.
- Both fluorochromes are excited by the **640nm** laser line.
- 730/45 filter captures the peak of emission for both fluorochromes.
- Single color APC-R700 was brighter than the AF700 in experimental sample.



No!



Sample	Secondary Stain Index
Full Stained Sample (AF700 + APC)	-3.49
Single Stained APC-R700	0.27

Conclusion

Even if fluorochromes have a similar emission spectra, they should not be treated the same. Fluor swapping can and will result in over or under compensation.