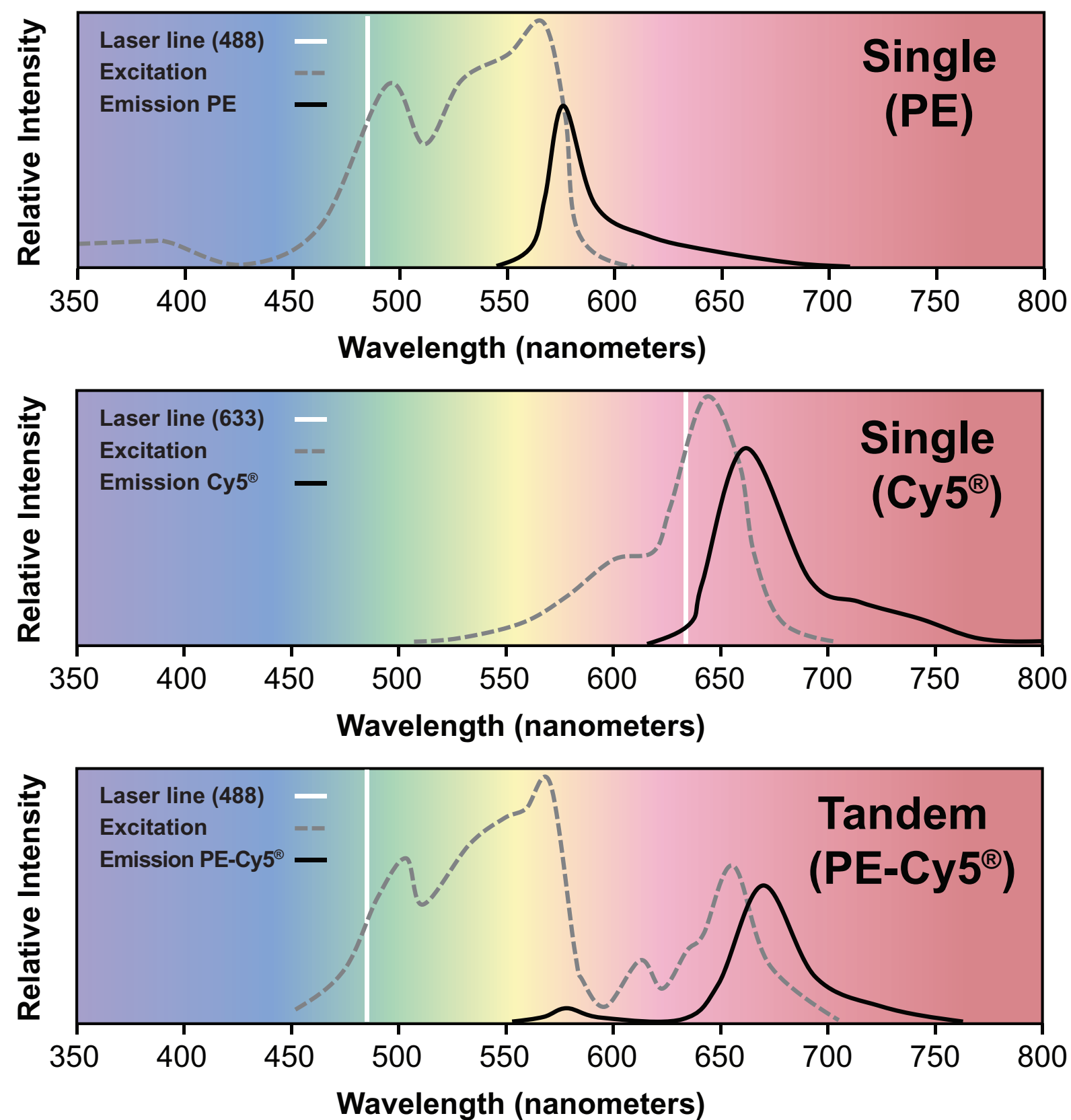


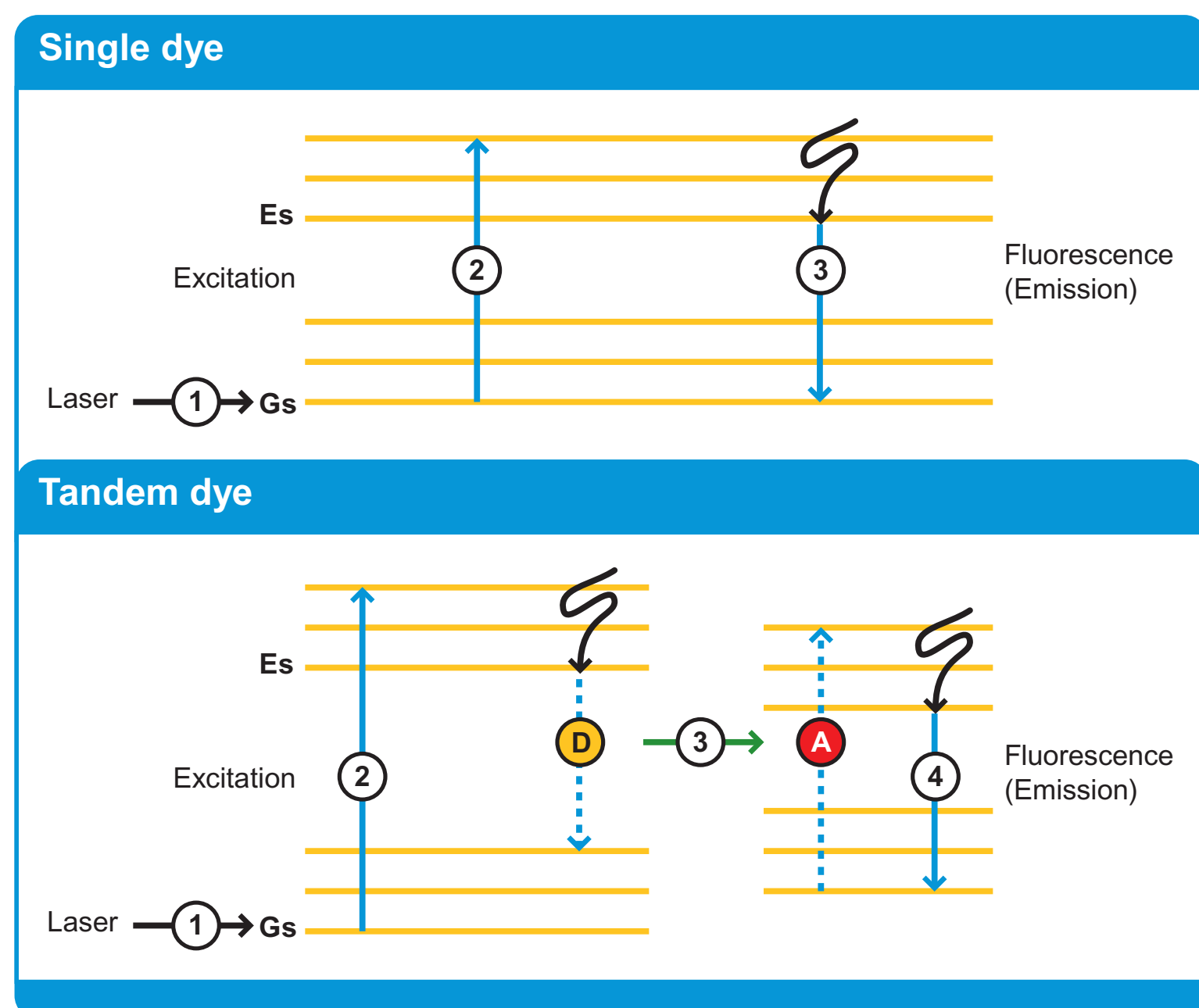
A Guide to Fluorochromes

Excitation and Emission Spectral Profiles



Fluorochromes have unique and characteristic spectra for absorption (excitation) and emission. A single dye is excited at a particular wavelength and emits a photon at a higher wavelength. A tandem dye consists of a donor and acceptor fluorochrome molecule, placed in close proximity, allowing for energy transfer between the two. The tandem dye is excited at the excitation wavelength of the donor molecule and emits a photon at the emission wavelength of the acceptor molecule.

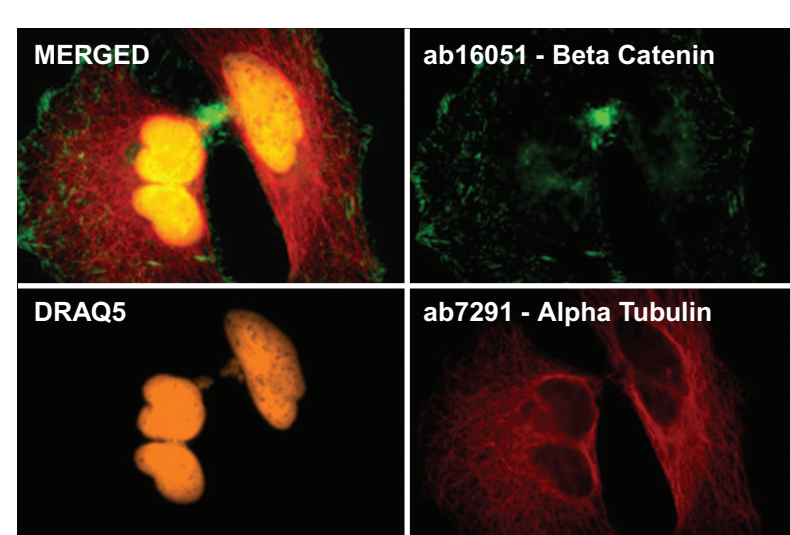
Energy Level Diagrams



For a **single fluorescent dye**, a laser set at the signature excitation wavelength for the dye provides electromagnetic energy to an electron in that molecule (1). The electron moves to an excitation state at the next energy level (Es) (2). Energy is then released in the form of a photon (fluorescence) and the electron moves back down to the lower energy level (Gs) (3). In the case of a **tandem fluorescent dye**, after excitation of the electron by a laser (1) - (2), energy is released by an electron in the donor molecule (D) and absorbed by an electron in the acceptor molecule (A) (3). The electron in the acceptor molecule moves to an excitation state at the next energy level (Es) (4). Similar to a single dye, energy is then released in the form of the photon (fluorescence) and the electron moves back down to the lower energy level (Gs) (4).

Dye	Max. excitation wavelength (nm)	Max. emission wavelength (nm)	Excitation laser lines (nm)
Methoxycoumarin	360	410	
DyLight [®] 405	400	420	
Alexa Fluor [®] 405	402	421	
Brilliant Violet 421™	407	421	
HiLyte Fluor™ 405	404	428	
DyLight [®] 350	353	432	
Alexa Fluor [®] 350	346	442	
Aminocoumarin (AMCA)	350	445	
BD Horizon™ V450	404	448	
Pacific Blue™	404	456	360,405,407
EviTag™ quantum dots-Lake Placid Blue	470	490	
AMCyan	457	491	
BD Horizon™ V500	415	500	
Cy2 [®]	489	506	488
Chromo™ 488	488	517	
DyLight [®] 488	493	518	
Alexa Fluor [®] 488	495	519	488
FAM	494	519	
Fluorescein Iso-thiocyanate (FITC)	495	519	488
EviTag™ quantum dots-Adirondack Green	505	520	
Chromo™ 505	505	526	
HiLyte Fluor™ 488	501	527	
Alexa Fluor [®] 514	518	540	
EviTag™ quantum dots-Catskill Green	525	540	
Alexa Fluor [®] 430	434	541	
Pacific Orange™	403	551	
Alexa Fluor [®] 532	532	554	
HEX	535	556	
EviTag™ quantum dots-Hops Yellow	545	560	
Chromo™ 546	545	561	
Cy3 [®]	548	561	488,514
Alexa Fluor [®] 555	555	565	
HiLyte Fluor™ 555	550	566	
5-TAMRA	541	568	
Alexa Fluor [®] 546	556	573	532
DyLight [®] 550	562	576	
Phycoerythrin (PE)	496,566	576	488
Tetramethyl Rhodamine Isothiocyanate (TRITC)	557	576	
EviTag™ quantum dots-Birch Yellow	560	580	
Cy3.5 [®]	576	589	568,543
Rhodamine Red-X	570	590	
PE-Dyomics [®] 590	488	599	
EviTag™ quantum dots-Fort Orange	585	600	
ROX	575	602	
Alexa Fluor [®] 568	578	603	532
Red 613	480,565	613	
Texas Red [®]	595	613	568,543,514
HiLyte Fluor™ 594	593	616	
PE-Texas Red [®]	566	616	
Alexa Fluor [®] 594	590	617	
DyLight [®] 594	593	618	
EviTag™ quantum dots-Maple-Red Orange	600	620	
Alexa Fluor [®] 610	612	628	
Chromo™ 494	494	628	
Alexa Fluor [®] 633	632	647	
SureLight [®] APC	652	657	
DyLight [®] 633	638	658	
Allophycocyanin (APC)	650	660	595,633,635,647
Chromo™ 642	642	660	
Quantum Red	488	660	
SureLight [®] P3	614	662	
Alexa Fluor [®] 647	650	665	595,633,635,647
Cy5 [®]	647	665	633,635
PE-Cy5 [®]	565	666	488
SureLight [®] P1	545	666	
PE-Alexa Fluor [®] 647	567	669	
PE-Dyomics [®] 647	488	672	
DyLight [®] 650	654	673	
HiLyte Fluor™ 647	650	675	
Peridinin Chlorophyll (PerCP)	477	678	488
IRDye [®] 700DX	680	687	
Alexa Fluor [®] 660	663	690	
PE-Cy5.5 [®]	565	693	488
APC-Cy5.5 [®]	650	694	595,633,635,647
Cy5.5 [®]	675	694	647
TruRed	490,675	695	
HiLyte Fluor™ 680	678	699	
Alexa Fluor [®] 680	679	702	
DyLight [®] 680	692	712	
Alexa Fluor [®] 700	702	723	
APC-Cy7 [®]	650	774	595,633,635,647
Alexa Fluor [®] 750	749	775	
Cy7 [®]	753	775	
PE-Dyomics [®] 747	488	776	
DyLight [®] 755	754	776	
HiLyte Fluor™ 750	753	778	
PE-Cy7 [®]	566	778	488
IRDye [®] 800RS	770	786	
DyLight [®] 800	777	794	
IRDye [®] 800CW	778	794	
Alexa Fluor [®] 790	782	805	

Multicolor staining example



DRAQ5™ (ab108410) allows rapid staining of dsDNA/nuclei of LIVE or FIXED cells. It is ideal for use in multi-color analysis and compatible with GFP labels. As well as offering half the scan times of Hoechst or DAPI, DRAQ5™ can be used in a greater variety of applications. The image on the left shows HeLa cells stained with 5µM DRAQ5™ to label nuclear DNA (pseudocolor orange). Pre-absorbed goat polyclonal secondary antibodies to rabbit IgG - H&L were used to visualise beta Catenin (DyLight[®] 488, ab96899, green) and alpha Tubulin (DyLight[®] 594, ab96899, red).

Nucleic Acid Dyes	Max. excitation wavelength (nm)	Max. emission wavelength (nm)	Excitation laser lines (nm)
DAPI	359	461	325,360,405,407
Hoechst 33258	352	461	
Hoechst 33342	350	461	
SYTOX Blue	431	480	
YOYO-1	491	509	
SYTOX Green	504	523	
TOTO-1, TO-PRO-1	509	533	
Mithramycin	450	570	
SYTOX Orange	547	570	
Chromomycin A3	445	575	
CyTRAK Orange™*	457,488,549	615	
Ethidium Bromide	493	620	
Propidium iodide (PI)	305,540	620	325,360,488
DRAQ5™	646	681,697	
DRAQ7™	599,644	678,697	

*CyTRAK Orange™ stains both nucleus and cytoplasm