

# Innova Biosciences A-Z Guide to Fluorochromes

[Lightning-Link® fluorescent antibody and protein labeling kits](#) allow you to covalently label any antibody in under 20 minutes with only 30 seconds hands-on time.

We currently have over 35 labels in [our range](#) of fluorescent dyes. This guide provides a general overview of each fluorochrome in our range including their properties, excitation and emission wavelengths and common applications.

## Allophycocyanin (APC)

Allophycocyanin is a large 105kDa fluorescent phycobiliprotein commonly used in FACS analysis. It absorbs at 650nm and emits at 662nm.

Most commonly APC is used for [ELISA](#), [flow cytometry](#), microarrays and other applications that require high sensitivity but not photostability.

## AMCA

[AMCA](#) (aminomethylcoumarin acetate) is a blue fluorescent dye commonly used in immunofluorescence microscopy and flow cytometry. It has a maximal excitation wavelength of 346nm and a maximal emission wavelength of 442nm.

AMCA has good resistance to photobleaching and has quite a large Stoke's shift.

AMCA is most commonly used for multiple labeling due to the minimal fluorescence overlap with longer wavelength-emitting fluorophores. Furthermore the blue fluorescence of AMCA, when labeled on its own, is not easily seen by the human eye, thus AMCA is popular for [multi-colour flow cytometry](#) and multi labeling for immunofluorescence microscopy.

## Atto dyes

Atto dyes are more photostable when compared to fluorescein and Cy-based dyes. In addition they exhibit long signal lifetimes and have longer excitation wavelengths for reduced background.

[Atto390](#) - is one of a new generation of fluorescent labels with a coumarin structure. It has a strong absorption at 390nm, a high fluorescence at 479nm (extinction coefficient  $2.4 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[Atto488](#) - has been optimised for excitation with an argon laser. It has a strong absorption at 501nm, high fluorescence at 523nm (extinction coefficient  $9.0 \times 10^4$ cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[Atto565](#) - has a strong absorption at 563nm, high fluorescence at 592nm (extinction coefficient  $1.2 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and quantum yield.

[Atto633](#) - has a strong absorption at 629nm, high fluorescence at 657nm (extinction coefficient  $1.3 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield. Atto633 can be used as a substitute dye for DY-633, LightCycler Red 640, Alexa Fluor® 633, and Cy 5.

[Atto700](#) - has a strong absorption peak at 690nm, with an extinction coefficient of  $1.2 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>, and a maximum emission peak at 725nm.

## B-Phycoerythrin (BPE)

BPE is a fluorescent protein from the phycobiliprotein family from the cyanobacteria and eukaryotic algae. It has a strong absorption peak at about 545 nm and has an emission value of 572 nm.

[B-Phycoerythrin](#) is often thought to be less "sticky" than [R-Phycoerythrin](#) therefore BPE conjugates tend to produce less background signal from non-specific binding in certain applications.

Like RPE, BPE is commonly used for [flow cytometry](#) and [immunofluorescence](#).

## Cyanine (Cy) Dyes

Cyanine dyes are bright and stable fluorescent dyes commonly used for flow cytometry.

[Cy3](#) - has a strong absorption peak at 550nm and a secondary absorption peak at 520nm. The maximum emission peak is at 570nm. Cy3 can be used as an alternative to Alexa Fluor® 555, DY-550, Rhodamine Red-X and TAMRA [5(6)-Carboxytetramethylrhodamine].

[Cy5](#) - has a strong absorption peak at 643nm and a secondary absorption peak at 605nm. The maximum emission peak is at 667nm. Cy5 can be used as an alternative to Alexa Fluor® 647, and the phycobiliprotein Allophycocyanin.

[Cy5.5](#) - has a strong absorption peak at 675nm and an emission peak at 694nm. It can be used as an alternative to Alexa Fluor® 680, WellRed DP-3A, DY-676 and LightCycler Red.

## DyLights®

DyLight® dyes are extremely bright and are a close alternative to Alexa Fluor® dyes. DyLight® dyes are highly water soluble meaning that a high dye-protein ratio can be achieved without precipitation of the conjugates. Furthermore DyLight® dyes retain their fluorescence over a pH range of 4-9.

Primary applications of DyLight® conjugates include [immunohistochemistry](#), [western blotting](#), [ELISA](#) or immunofluorescence microscopy.

See below for an overview of each of the DyLights® we currently offer:

[DyLight® 350](#) - is a derivative of AMCA with a vibrant blue fluorescence that is brighter than AMCA and its Alexa equivalent Alexa Fluor® 350. It has a strong absorption at 353nm, high fluorescence at 432nm (extinction coefficient  $1.5 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 405](#) - provides blue fluorescence. It has a strong absorption at 400nm, high fluorescence at 420nm (extinction coefficient  $3.0 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 488](#) - provides green fluorescence. It has a strong absorption at 493nm, high fluorescence at 518nm (extinction coefficient  $7.0 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 550](#) - emits vibrant orange-to-red fluorescence. It has a strong absorption at 562nm, high fluorescence at 576nm (extinction coefficient  $1.5 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 594](#) - generates red fluorescence. It has a strong absorption at 593nm, high fluorescence at 618nm (extinction coefficient  $8.0 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 650](#) - generates far red fluorescence. It has a strong absorption at 652nm, high fluorescence at 672nm (extinction coefficient  $2.5 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 680](#) - provides red fluorescence. It has a strong absorption at 682nm, high fluorescence at 715nm (extinction coefficient  $1.4 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 755](#) - provides far red fluorescence. It has a strong absorption at 754nm, high fluorescence at 776nm (extinction coefficient  $2.2 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[DyLight® 800](#) - provides near infrared fluorescence. It has a strong absorption at 770nm, high fluorescence at 794nm (extinction coefficient  $2.7 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

## FluoProbes®

FluoProbes® are a series of fluorescent dyes developed by Interchim to label biomolecules in advanced fluorescent detection techniques. They offer high fluorescence intensity, excellent photostability and are appropriate for multiple labeling as there is no overlap between dyes allowing for multiplex staining.

FluoProbes are suggested to reduce high background in flow cytometry and in slide microscopy and are also commonly used in applications such as [FRET](#), polarisation anisotropy fluorescence, quenching and life time resolved fluorescence.

[FluoProbes®647H](#) - has a strong absorption at 653nm, high fluorescence at 674nm (extinction coefficient  $2.5 \times 10^5$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[FluoProbes®594](#) - has a strong absorption at 591nm, high fluorescence at 616nm (extinction coefficient  $9.2 \times 10^4$  cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield. FluoProbes®594 can be used as a substitute dye for BODIPY® TR-X, Carboxynaphthofluorescein and Alexa Fluor® 594.

## Fluorescein

[Fluorescein](#) is one of the most popular fluorescent reagents used in biological research because of its water solubility, intense fluorescence and high absorptivity. It has a peak excitation occurring at 494nm and peak emission of 521nm.

Fluorescein conjugates are very popular for flow cytometry and immunofluorescence and are also used for immunohistochemistry.

## PerCP (Peridinin-Chlorophyll-Protein Complex Label)

[PerCP](#) is a small 35kDa phycobiliprotein isolated from red algae. It has a strong absorption peak at 482nm and a secondary absorption peak at 442nm. PerCP has a large Stokes shift (195nm) with its maximum emission peak at 677nm. There is minimal overlap with other fluorochromes making PerCP conjugates especially useful for multi-colour analysis.

PerCP is commonly used in fluorescence-activated cell sorting (FACS) and flow cytometry.

## R-Phycoerythrin (RPE)

[RPE](#) is a fluorescent protein from the phycobiliprotein family, present in red algae and cryptophytes. It has an absorption peak of 488 and 535 nm and it has a strong emission peak at 575nm. RPE is closely related to BPE and these are the most intense fluorescent phycobiliproteins providing an orange fluorescence. RPE conjugates are commonly used for immunofluorescence and flow cytometry and are ideal for multiple labeling with other fluorochromes.

## Rhodamine

[Rhodamine](#) dyes are used mainly in flow cytometry, fluorescence microscopy and ELISA. Rhodamine has an absorption peak of 544nm and maximum emission is obtained when it is excited at 576 nm.

## Tandem dyes

A tandem dye consists of two fluorescent molecules, usually one of a larger size than the other, which are covalently attached to each other and use the emission of one dye and excitation properties of the other. Using Förster resonance energy transfer (FRET), the excitation energy is passed from one fluorophore to the other, emitting photons of light.

Tandem dyes are commonly used in flow cytometry as they are normally manufactured for the standard 488nm laser. They are also favourable for multi-colour fluorescence especially when in conjunction with singular dyes.

[APC/Cy5.5](#) - the APC in this tandem functions as an energy donor for the Cy5.5 and is excited at 633nm. Energy is transferred from the APC to the Cy5.5 via FRET. Energy received from APC is emitted by Cy5.5 in the form of long wavelength light at 694nm.

[APC/Cy7](#) - the APC in this tandem excites at 633nm and transfers its energy to Cy7. The Cy7 emits the energy received from the APC in the form of long wavelength light at 776nm.

[PE/Atto594](#) - the PE in this tandem is excited at 488nm and transfers its energy to the Atto594. The Atto594 emits the energy received at 627nm.

[PE/Cy5 Label](#) - the PE in this tandem is excited at 488nm and is the energy donor for the Cy5. The Cy5 emits the energy received from the PE in the form of long wavelength light at 679nm.

[PE/Cy5.5](#) - the PE is excited at 490nm and functions as an energy donor for the Cy5.5. Energy is transferred from the PE to the Cy5.5 and the Cy5.5 emits the energy received from the PE in the form of long wavelength light at 695nm.

[PE/CY7](#) - the PE is excited at 488nm and functions as an energy donor for the Cy7. Energy is transferred from the PE to the Cy7 and Cy7 emits the energy received from the PE in the form of long wavelength light at 776nm.

[PE/TEXAS RED®](#) - the PE is excited at 488nm and functions as an energy donor for the TEXAS RED®. The TEXAS RED® emits the energy received from the PE in the form of long wavelength light at 615nm.

[PerCP/Cy5.5](#) - the PerCP is excited at 482nm and transfers its energy to the Cy5.5 via energy resonance transfer. The Cy5.5 emits the energy received from the PerCP in the form of long wavelength light at 700nm.

## TEXAS RED®

[TEXAS RED®](#) is a red fluorescent dye commonly used in histology, immunohistochemistry, FACS and multi-colour flow cytometry and fluorescence microscopy. It has a strong absorption at 595nm, with a high fluorescence at 615nm (extinction coefficient 8.0 x10<sup>4</sup> cm<sup>-1</sup>M<sup>-1</sup>) and high quantum yield.

[Click here](#) to see the full list of our fluorescent dyes, their emission and excitation colours and for more information.

Lightning-Link®, is a registered trademark of Innova Biosciences  
DyLight® is a registered trademark of Thermo Fisher Scientific Inc. and its subsidiaries.  
Alexa Fluor® and TEXAS RED® are registered trademarks of Life Technologies Corporation  
FluoProbes® is a registered trademark of Interchim  
Cy and CyDye are trademarks of GE Healthcare