

Dye: Cyanine 3 (Cy3™)

The Cyanine fluorescent dye family are produced by GE Healthcare. This family of fluorophores are a popular choice for labelling a variety of fluorescence based applications. They also offer bright and intense colours with narrow and spectrally distinct emission bands and are available in different chemistries allowing labelling via amine or thiol groups. The Cy fluorophore family provide high sensitivity and are very photostable and exhibit low non-specific binding.

The Cy3 dye is an orange fluorescing cyanine that produces an intense signal. The Cy3 fluorescent dye is usually synthesized with reactive groups on either one or both nitrogen side chains so that the dye can be chemically linked to either nucleic acids or protein molecules.

Applications

The Cy3 fluorophore is often used for multicolour detection in a range of applications. This fluorescent dye is also often used in fluorescent labelling applications including western blots and in applications involving proteomics such as 2-D gel electrophoresis.

Visualization

Applications using the Cy3 fluorescent dye can be easily visualized using the Syngene Dyversity and G:BOX iChemi range of cooled camera image capture systems. The Cy3 dye has an excitation peak of 550nm and an emission peak of 570nm.

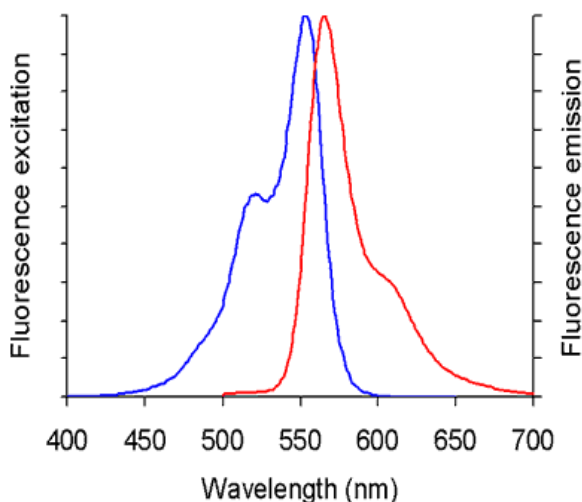


Figure 1 – Excitation (blue line) and emission (red line) spectra of Cy3 dye

System	Lighting	Filter
Dyversity	Epi green from RGB module Cy3	FiltLP Cy3
G:BOX iChemi range of cooled cameras	Epi green from RGB module CY3	FiltLP Cy3

Table 1 - Recommended lighting and filter selection for visualizing the Cy3 dye using Syngene image capture systems

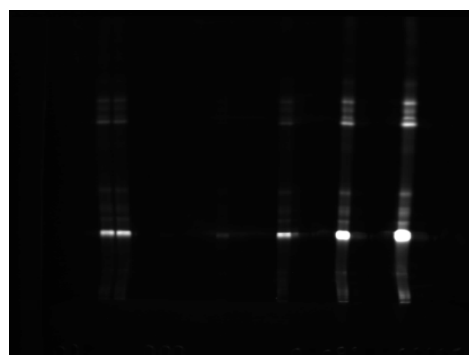


Figure 2 – Polyacrylamide Cy3 gel visualized using G:BOX XT image capture system

Two protein samples Sigma carbonic anhydrase and Sigma bovine serum albumin (BSA) were labelled with Cy3 dye and a dilution series of 1, 10, 100, 500 and 1000ng of protein and dye were loaded on to a polyacrylamide gel. Bands were visualized by exposure to a Cy3 filter. The image was captured using GeneSnap software (Syngene) for an exposure time of 17 seconds using the G:BOX XT Syngene image capture system.

For the ideal image using the Cy3 dye the image should have good resolution, high sensitivity enabling the detection of low intensity bands and a low background improving band visibility as illustrated in the image above.

Syngene reserves the right to amend or change specifications without prior notice. This Application note supersedes all earlier versions.

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UK tel: +44 (0)1223 727123
Email: sales@syngene.com

USA tel: 800 686 4407/301 662 2863
Email: ussales@syngene.com