Datasheet for ABIN6719663

DyLight™ Multiplex 649/488/800 Trio Western Blot Kit

2 Images

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Overview

Quantity:	1 kit
Reactivity:	Chicken, Mouse, Rabbit
Host:	Goat
Application:	BioImaging (BI), Immunofluorescence (IF), Immunostaining (ISt), Multiplex Assay (MA), Standard (STD), Western Blotting (WB)

Product Details

Purpose:	This kit is suitable for fluorescent western blotting, multiplex analysis, including multicolor imaging, utilizing various commercial gel imaging systems.
Characteristics:	Synonyms: Fluorescent western blotting, Multiplex western blotting, multi-color western blotting, fluorescent labelled antibodies, fluorescent imaging, DyLights, dyLight conjugates Background: The DyLight™ multiplex 649/488/800 Trio fluorescent Western blot kit is suited for simultaneous detection and quantification of specific protein populations in a biological sample. Using a combination of three antibodies selected for minimal cross reactivity, fluorescent detection method enables simultaneous quantitative analysis of multiple proteins within the same sample on the same blot. The DyLight™ multiplex 649/549/800 Trio Western blot kit contain all the necessary components that are optimized for the simultaneous detection of multiple proteins on the same blot using DyLight™-dye labeled secondary antibodies that are visualized in different fluorescence channels (649/549/800). The kit also includes blocking buffer, wash buffer, pre-stained protein standard and an incubation box for convinience and ultimate performance with minimal or no optimization. The fluorescent dyes such as DyLights™ when conjugated to secondary antibodies, offer a variety of benefits over traditional detection methods such as colorimetric and chemiluminescent detection. Multiplex detection using the

Product Details

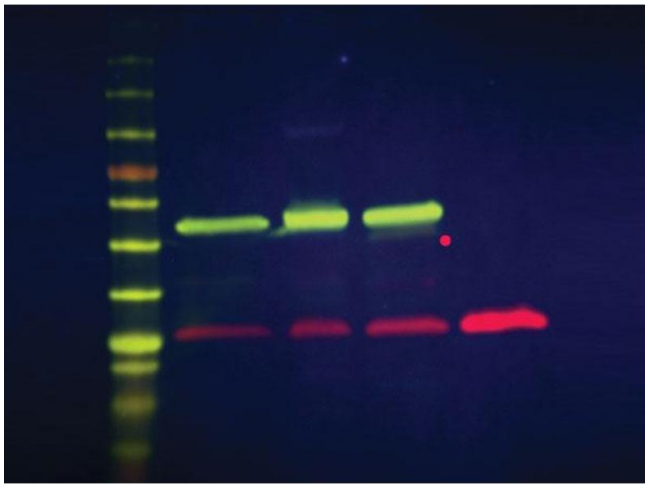
	correct lighting and filter conditions, enables the quantitation of multiple proteins and eliminates the need to strip and reprobe. Other benefits of fluorescent Western blotting include increased sensitivity, excellent signal stability over time as well as precise quantitative analysis with broader dynamic range and high linearity. Due to their exceptional photostability, DyLight™ dye conjugates can be archived and visualized several times without a decrease in signal.
Components:	DyLight™ Multiplex 649/488/800 Trio Western Blot Kit contains Goat anti-mouse IgG (H+L) DyLight™ 649, Goat anti-chicken IgG (H+L) DyLight™ 488, Goat anti-rabbit IgG (H+L) DyLight™ 800, opal pre-stained protein standard, wash buffer (10x TTBS), blocking buffer (2x) and incubation box.

Application Details

Protocol:	Production: The DyLight conjugated secondary antibodies were prepared from monospecific antiserum by immunoaffinity chromatography using Rabbit or Mouse or chicken IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any unwanted reactivities. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Goat Serum, Rabbit or Mouse or chicken IgG and Rabbit or Mouse or chicken Serum. No reaction was observed against Bovine, Goat, Guinea Pig, Hamster, Horse, Human, Rat and Sheep Serum Proteins. These antibodies will react with heavy chains of rabbit or mouse or chicken IgG and with light chains of most rabbit or mouse or chicken immunoglobulins. Blocking buffer is specifically formulated to achieve superior reproducible western blotting images using this system. Wash buffer (10X TTBS) was aseptically filtered through a Millipore 0.22 micron filter into clean, pre-sterilized containers. The product was tested on trypticase soy agar for 24 hours, 48 hours and 72 hours and was found to be negative for bacteria.
Restrictions:	For Research Use only

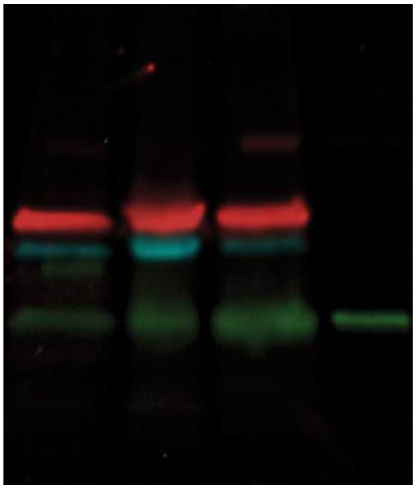
Handling

Storage:	RT,4 °C,-20 °C
Storage Comment:	The secondary antibodies should be stored at 4°C prior to reconstitution. For extended storage aliquot contents and freeze at -20°C. Avoid freeze/thaw cycles. The wash buffer and blocking buffer can be stored at 2-8°C prior to opening. The pre-stained protein Western standards should be stored at 2-8°C. The pre-stained protein standards can also be stored at room temperature for up to 6 months or at -20°C for up to two years.
Expiry Date:	12 months



Immunofluorescence

Image 1. Simultaneous detection of α-tubulin and GFP on a single blot using -labeled secondary antibody conjugates. Simultaneous detection of α-tubulin and GFP on a single blot using -labeled secondary antibody conjugates. Protein lysates from HeLa (lane 1), PC12 (lane 2) and K562 (lane 3) cells and 100ng GFP protein (Lane 4) were run on a gel. The cell lysates were spiked with 25ng, 50ng and 75ng GFP protein. Probing of cell lysates and GFP with mouse anti-α-tubulin and chicken anti-GFP antibodies followed by649 goat anti-mouse IgG (pseudocolored green) and 800 goat anti-chicken IgG (red) conjugates, and then imaged using Syngene G:BOX Imaging System resulted in comparable patterns of detection. Lane 5: Opal Prestained Protein Standard 10-245kDa.



Fluorescene Western

Image 2. Simultaneous detection of three proteins on a single blot using -labeled secondary antibody conjugates. Simultaneous detection of three proteins on a single blot using -labeled secondary antibody conjugates. Protein lysates from HeLa (lane 1), PC12 (lane 2) and K562 (lane 3) cells and 50ng GFP protein (Lane 4) were run on a gel. The cell lysates were spiked with 50ng, 75ng and 150ng GFP protein. Probing of cell lysates and GFP with anti-α-tubulin (mouse), anti-β-actin (rabbit), and anti-GFP (chicken) followed by-649 goat anti-mouse IgG (red),- 800 goat anti-rabbit IgG (pseudocolored aqua) and-488 goat anti-chicken IgG (pseudocolored green) conjugates, and imaged using Syngene G:BOX Imaging System resulted in comparable patterns of detection.