abcam

Product datasheet

DyLight® 488 Conjugation Kit (Fast) - Lightning-Link® ab201799

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Overview

Product name

DyLight® 488 Conjugation Kit (Fast) - Lightning-Link®

Product overview

DyLight[®] 488 Conjugation Kit / DyLight[®] 488 Labeling Kit (ab201799) uses a simple and quick process for DyLight[®] 488 labeling / conjugation of antibodies. It can also be used to conjugate other proteins or peptides. Learn about our **antibody labeling kits and their advantages**.

To conjugate an antibody to DyLight® 488 using this kit:

- add modifier to antibody and incubate for 15 mins
- add quencher and incubate for 5 mins

The DyLight[®] 488 conjugated antibody can be used immediately in WB, ELISA, IHC etc. No further purification is required and 100% of the antibody is recovered for use.

Learn about buffer compatibility below; for incompatible buffers and low antibody concentrations, use our rapid **antibody purification and concentration kits**. Use the **FAQ** to learn more about the technology, or about conjugating other proteins and peptides to DyLight[®] 488.

Custom size conjugation kits up to 100 mg are available on demand. Please contact us to discuss your requirements.

This product is manufactured by Expedeon, an Abcam company, and was previously called Lightning-Link $^{\circledR}$ Rapid DyLight $^{\circledR}$ 488 Labeling Kit. 322-0015 is the same as the 1 mg size. 322-0030 is the same as the 3 x 10 ug size. 322-0010 is the same as the 3 x 100 ug size. 322-0005 is the same as the 100 ug size.

Amount and volume of antibody for conjugation to Dylight ® 488

Kit size	Recommended amount of antibody ¹	Maximum amount of antibody	Maximum antibody volume ²	
3 x 10 µg	3 x 10 µg	3 x 20 μg	3 x 10 µL	
100 μg	1 x 100 μg	1 x 200 µg	1 x 100 μL	
3 x 100 µg	3 x 100 μg	3 x 200 µg	3 x 100 µL	

Notes

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1 mg	1 x 1 mg	1 x 2 mg	1 x 1 mL

¹ Using the maximum amount of antibody may result in less labelling per antibody.

Buffer Requirements for Conjugation

Buffer should be pH 6.5-8.5.

Compatible buffer constituents

If a concentration is shown, then the constituent should be no more than the concentration shown. If several constituents are close to the limit of acceptable concentration, then this can inhibit conjugation.

50mM / 0.6% Tris ¹	0.1% BSA ²	50% glycerol
0.1% sodium azide	PBS	Potassium phosphate
Sodium chloride	HEPES	Sucrose
Sodium citrate	EDTA	Trehalose

¹ Tris buffered saline is almost always ≤ 50 mM / 0.6%

Incompatible buffer constituents

Thiomerosal	Proclin	Glycine
Arginine	Glutathione	DTT

If a constituent of the buffer containing your antibody or protein is not listed above, please check the **FAQ** or **contact us**.

Only purified antibodies are suitable for use, ie. where other proteins, peptides, or amino acids are not present: antibodies in ascites fluid, serum or hybridoma culture media are incompatible.

Storing and handling conjugation kits

Lyophilized Lightning-Link® components are hygroscopic.

Kits are intentionally shipped at ambient temperature with silica gel to avoid exposure to moisture. Upon receipt, store the kit frozen and protect from moisture. Before opening the outer container, allow the lyophilized components to reach room temperature to minimize condensation.

DyLight® is a trademark of Thermo Fisher Scientific Inc. and its subsidiaries.

Properties

Storage instructions

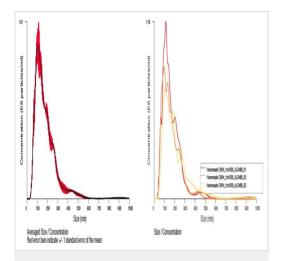
Store at -20°C. Please refer to protocols.

² Ideal antibody concentration is 1mg/ml. 0.5 - 1 mg/ml can be used if the maximum antibody volume is not exceeded. Antibodies > 2mg/ml or < 0.5 mg/ml should be diluted /concentrated.

² BSA can also interfere with the use of the conjugated antibody in tissue staining.

Components	1 mg	100 µg	3 x 10 µg	3 x 100 µg
ab274013 - Dylight® 488 Conjugation Mix	1 x 1mg	1 x 100µg	3 x 10µg	3 x 100µg
ab273994 - Modifier reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl
ab273995 - Quencher reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl

Images

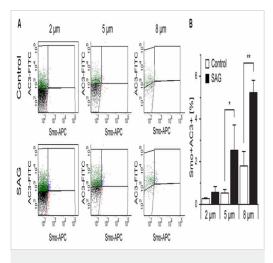


Conjugation - DyLight488 Conjugation Kit (Fast)-Lightning-Link; (ab201799)

Image from Mellema et al., PLoS One, 11(12):e0166045; doi: 10.1371/journal.pone.0166045. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Mellema, M., et al used DyLight[®] 488 Conjugation Kit (Fast) - Lightning-Link[®] (ab201799) as part of examining DyLight[®] 488-conjugated alendronate binding of calcifying nanoparticles (CNP) generated in vitro. They used the kit to conjugate DyLight[®] 488 to alendronate (that selectively and specifically bind to calciumphosphorus surfaces) for use in Nanoparticle Tracking Analysis (NTA[®]).

CNP in the whole urine samples and in vitro generated CNP suspensions were analyzed using the NanoSight LM10HS-48814TS instrument (Malvern Instruments, Worcestershire, UK) with a 488 nm wavelength laser, fluorescent filter (505 nm long pass), and both temperature control and syringe pump modules. A representative histogram shows averaged sizing and relative abundance data (left; SE in red) and individual analyses from the assessment (right) of this representative sample in triplicate. Note that CNP generated in vitro do not acquire the protein/mucus coating known to occur in vivo and the larger forms retain the ability to bind to alendronate.

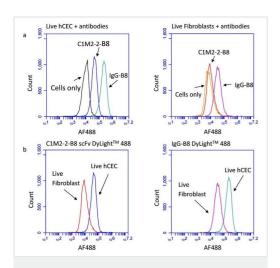


Flow Cytometry - DyLight 488 Conjugation Kit (Fast) Lightning-Link (ab201799)

Image from Monaco, Sara, et al., Front Cell Neurosci., 12:519; doi: 10.3389/fncel.2018.00519. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Monaco, Sara, et al used DyLight[®] 488 Conjugation Kit (Fast) - Lightning-Link[®] (ab201799) as part of examining the co-localization of Smoothened and AC3 in primary cilia. They used the kit to conjugate DyLight[®] 488 to anti-AC3 antibody for use in flow cytometry.

(A) Representative dot plots of untreated (Control) and SAG-treated samples (SAG) from E18 embryos immunostained for AC3-FITC and Smoothened-APC (Smo-APC). (B) Quantification of the percentage of Smo+AC3+ particles. Bar graphs show mean \pm SEM, p-values are calculated with Student's t-test. Statistically significant differences are indicated with asterisks (*p < 0.05, **p < 0.01).



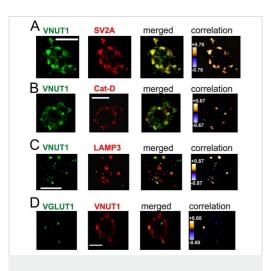
Flow Cytometry - DyLightDyLight® 488 Conjugation

Kit (Fast) - Lightning-Link® (ab201799)

Image from Dorfmueller, Simone, et al., Sci rep., 6:21661; doi: 10.1038/srep21661. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Dorfmueller, Simone, et al used DyLight[®] 488 Conjugation Kit (Fast) - Lightning-Link[®] (ab201799) as part of characterizing antibody-labelled human corneal endothelial cells (hCECs) and stromal fibroblasts. They used the kit to conjugate DyLight[®] 488 to C1M2-2-B8 scFv or lgG-B8 full-size antibody for use in flow cytometry.

Primary hCECs or stromal fibroblasts were incubated with C1M2-2-B8 scFv or lgG-B8 full-size antibody conjugated to DyLight[®] 488 (4 µg/ml). (a) The histogram for unlabelled hCECs (black) is overlaid with those for scFv and lgG-B8 labelled cells as indicated. Similarly, the histogram for unlabelled stromal fibroblasts (orange) is overlaid with those for scFv and lgG-B8 labelled fibroblasts. (b) The histograms for C1M2-2-B8 scFv labelled hCECs and stromal fibroblasts were shown on the same graph, as for the histograms for lgG-B8 labelled hCECs and stromal fibroblasts. The peak separation between fibroblasts and hCECs were similar in magnitude with either scFv or lgG labelling.

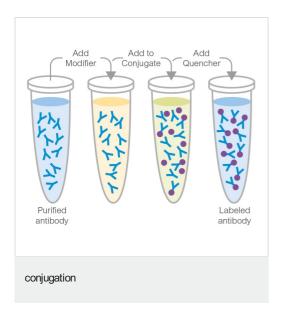


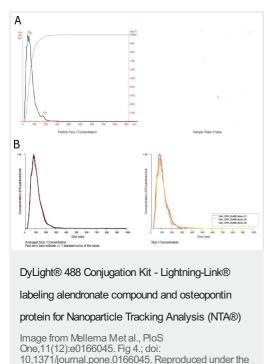
Fluorescence Microscopy - DyLight488 Conjugation

Kit (Fast)- Lightning-Link (ab201799)

Image from Lalo, Ulyana, et al., PLoS Biol., 12(1): e1001747; doi: 10.1371/journal.pbio.1001747. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Lalo, Ulyana, et al used DyLight® 488 Conjugation Kit (Fast) -Lightning-Link[®] (ab201799) as part of examining the colocalization of vesicular ATP transporters with exocytotic organelle markers. They used the kit to conjugate DyLight[®] 488 to various antibodies for use in multiphoton fluorescent microscopy. Living, acutely isolated cortical astrocytes were labeled with antibodies to vesicular transporters (VNUT1 and VGLUT1) and synaptic vesicle (SV2A) and lysosomal (LAMP3 and Cathepsin-D) markers. Antibodies were conjugated to fluorescent dyes DyLight488 (green) and DyLight594 (red) prior to astrocyte labeling. (A-D) The representative two-photon fluorescence images (maximal intensity projections of Z-stack) and results of colocalization analysis, carried out using NIH ImageJ 1.43 software. The correlation between green and red fluorescence (images in the right column) is depicted as a product of the relative differences from the mean (PDM) for each pixel; the pseudocolor PDM images were generated as an output of ImageJ analysis routine. Positive values (bright yellow) are indicative for good co-localization of green and red signals, negative values (blue-violet) indicate segregation, and black color shows the lack of correlation. Note the different extent of scale for PDM values in (A-D). All scale bars in (A-D) are 5 μm.





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Mellema M et al. used ab201799 as part of examining mineraloorganic nanoparticles in feline urine.

They used the kit to conjugate DyLight[®] 488 to alendronate compound and osteopontin protein for use in Nanoparticle Tracking Analysis (NTA[®]).

A: Representative histogram showing size distribution and cumulative percentage (left) of submicron particulate matter in healthy feline urine after labeling with DyLight 488 conjugated alendronate. A representative frame from the captured video analyzed by NTA is shown as well (right). Note the strongly preferential binding to primary CNP that lack an outer layer of protein or mucus.

B: Representative histogram showing averaged sizing and relative abundance data (left; SE in red) and individual analyses from the assessment (right) of this representative sample of healthy feline urine after labeling with DyLight 488 conjugated osteopontin. Note the strongly preferential binding to primary naturally-occurring CNP that lack an outer layer of protein or mucus.

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