abcam

Product datasheet

Rhodamine Conjugation Kit (Fast) - Lightning-Link® ab188286

14 References 5 Images

Overview

Product name

Product overview

Rhodamine Conjugation Kit (Fast) - Lightning-Link®

Rhodamine Conjugation Kit / Rhodamine Labeling Kit ab188286 uses a simple and quick process for rhodamine 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 Rhodamine using this kit:

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

The rhodamine 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.

The excitation and emmision wavelengths for Rhodamine are Ex: 550nm, Em: 570nm.

Learn about buffer compatibility below; for incompatible buffers and low antibody concentrations, use our rapid <u>antibody purification and concentration kits</u>. Use the <u>FAQ</u> to learn more about the technology, or about conjugating other proteins and peptides to Rhodamine.

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[®] Rapid Rhodamine Labeling Kit. 311-0005 is the same as the 100 ug size. 311-0010 is the same as the 3 x 100 ug size. 311-0030 is the same as the 3 x 10 ug size. 311-0015 is the same as the 1 mg size.

Amount and volume of antibody for conjugation to Rhodamine

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	

Notes

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3 x 100 µg	3 x 100 μg	3 x 200 µg	3 x 100 μL
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.

Properties

Storage instructions

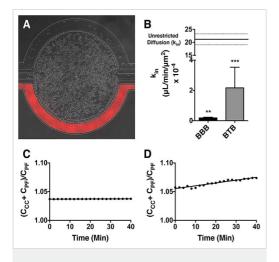
Store at -20°C. Please refer to protocols.

 $^{^2}$ Ideal antibody concentration is 1mg/ml. 0.5 - 1 mg/ml can be used if the maximum antibody volume is not exceeded. Antibodies > 2 mg/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
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
ab273998 - Rhodamine Conjugate	1 x 1mg	1 x 100µg	3 x 10µg	3 x 100µg

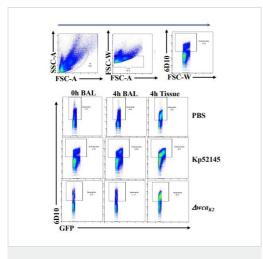
Images



Conjugation - Rhodamine Conjugation Kit (Fast) - Lightning-Link® (ab188286)

Image from Terrell-Hall et al., Oncotarget., 8(48):83734-83744; doi: 10.18632/oncotarget.19634. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/3.0/

Terrell-Hall, Tori B., et al used Rhodamine Conjugation Kit (Fast) -Lightning-Link® (ab188286) as part of examining mechanism of trastuzumab movement. They used the kit to conjugate Rhodamine to trastuzumab for characterization in a novel microfluidic in-vitro. Linear central compartment accumulation of trastuzumab-Rhodamine123 (t-Rho123) in in-vitro blood-brain barrier (BBB) and blood-tumor barrier (BTB) microfluidic chip models. Representative image of model with TRITC labeled t-Rho123 flowing over HUVEC cells in the outer compartment and either astrocytes or JIMT-1 cancer cells in the central compartment (A). Rate of t-Rho123 movement in each model plotted against the unrestricted diffusion k_{in}; ** p<0.0033 significance between BBB model and unrestricted diffusion k_{in}, n=3; *** p<0.0005 significance between BTB model and unrestricted diffusion kin, n=3. All data represent mean ± S.E.M. Each model is significantly different than 0 (p < 0.05) (B). Representative graphs of the rate of accumulation of t-Rho123 in the BBB (C) and BTB (D) microfluidic devices (n≥3).



Flow Cytometry - Rhodamine Conjugation Kit

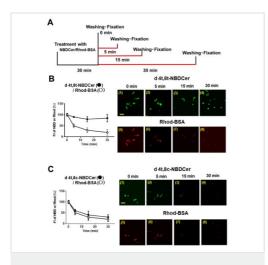
(Fast) - Lightning-Link® (ab188286)

Image from Dumigan, Amy, et al., MBio., 10(6): e02802-19; doi: 10.1128/mBio.02802-19. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Dumigan, Amy, et al used Rhodamine Conjugation Kit (Fast) - Lightning-Link[®] (ab188286) as part of examining innate cell recruitment in K. pneumoniae-infected porcine EVLP model. They used the kit to conjugate Rhodamine to anti-pig granulocyte marker antibody, clone 6D10, for use in flow cytometry.

Gating strategy and representative dot plots for flow cytometric analysis of neutrophil staining using anti-pig granulocyte marker clone 6D10 (B). Dot plots represent 0-h (baseline) and 4-h-postinfection or mock infection BAL samples and 4-h tissue

samples.

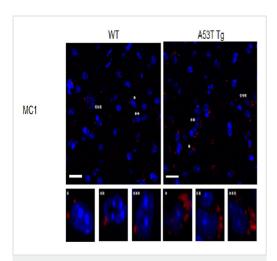


Fluorescence Microscopy - Rhodamine Conjugation
Kit (Fast) Lightning-Link (ab188286)

Image from Usuki, Seigo, et al., Cells, 9(2):517; doi: 10.3390/cells9020517. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Usuki, Seigo, et al used Rhodamine Conjugation Kit (Fast) - Lightning-Link (ab188286) as part of examining the time course of NBD-ceramides (NBD-Cer) and Rhodamine-bovine serum albumin (Rhod-BSA) bound to PC12 cells. They used the kit to conjugate Rhodamine to bovine serum albumin (BSA) for use in Dissociation Time Course for Cell Surface Binding.

(A) Dissociation time course analysis of FI based on binding of 100 nM d NBD-Cer (FI = 5000) and 100 nM Rhod-BSA (FI = 5510), examined using Plexin A1 gene-silencing PC12 cells. (B) Images showing (1 to 4) changes in d4t,8t-NBD-Cer and (5 to 8) changes in Rhod-BSA at the indicated timepoints. The left graph shows a time course plot of FI (%) relative to 0 min. Data are presented as mean \pm SD (n = 3). Scale bar = 100 μ m. (C) Images showing (1 to 4) changes in d4t,8c-NBD-Cer and (5 to 8) changes in Rhod-BSA at the indicated timepoints. The left graph is a time course plot of FI (%) relative to 0 min. Data are presented as mean \pm SD (n = 3). Scale bar = 100 μ m.

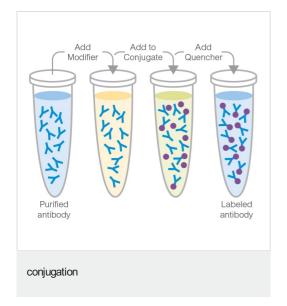


Immunohistochemistry (PFA perfusion fixed frozen sections) - Rhodamine Conjugation Kit (Fast) - Lightning-Link® (ab188286)

Image from Wills, Jonathan, et al., PLoS One, 6(3): e17953; doi: 10.1371/journal.pone.0017953. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Wills, Jonathan, et al used Rhodamine Conjugation Kit (Fast) - Lightning-Link® (ab188286) as part of examining MC1 immunostaining in the striatum of A53T α -Syn mutant mice (A53T Tg) and wild-type mice (WT). They used the kit to conjugate Rhodamine to anti-MC1 antibody for use in immunohistochemistry (PFA perfusion fixed frozen sections).

Sections of striatum of A53T Tg and age-matched WT mice were stained with anti-MC1 antibody to detect p-Tau conformation (red). Nuclei were stained with DAPI (blue). Slides are shown at lowest magnification (upper panels) to highest magnification (lower panels). Asterisks indicate highlighted individual cells shown at higher magnification in lower panels. Scale Bar: 10 μ m.



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