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Product datasheet

PerCP/Cy5.5® Conjugation Kit - Lightning-Link® ab102911

★★★★★ 1 Abreviews 28 References 画像数 6

医薬用外劇物

製品の概要

製品名

製品の概要

PerCP/Cy5.5® Conjugation Kit - Lightning-Link®

PerCP/Cy5.5[®] Conjugation Kit / PerCP/Cy5.5[®] Labeling Kit ab102911 uses a simple and quick process for PerCP/Cy5.5 labeling / conjugation of antibodies. It can also be used to conjugate other proteins or peptides. Learn about our <u>antibody labeling kits and their advantages</u>.

To conjugate an antibody to PerCP/Cy5.5® using this kit:

- add modifier to antibody and incubate for 3 hrs
- add quencher and incubate for 30 mins

The PerCP/Cy5.5[®] 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 PerCP/Cy5.5[®].

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[®] PerCP/Cy5.5 Labeling Kit. 763-0015 is the same as the 1 mg size. 763-0010 is the same as the 3 x 100 ug size. 763-0030 is the same as the 3 x 10 ug size. 763-0005 is the same as the 100 μ g size.

Amount and volume of antibody for conjugation to PerCP/Cy5.5®

Kit size	Recommended maximum amount of antibody	Maximum antibody volume ¹
3 x 10 µg	3 x 10 μg	3 x 10 µL
100 µg	1 x 100 µg	1 x 100 μL
3 x 100 µg	3 x 100 µg	3 x 100 μL
1 mg	1 x 1 mg	1 x 1 mL

特記事項

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	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.

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.

製品の特性

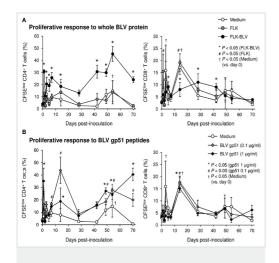
保存方法

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

内容	1 mg	100 μg	3 x 10 μg	3 x 100 μg
Modifier reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl
ab274153 - PerCP/Cy5.5 mix	1 x 1mg	1 x 100µg	3 x 10µg	3 x 100µg
ab274133 - Quencher reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl

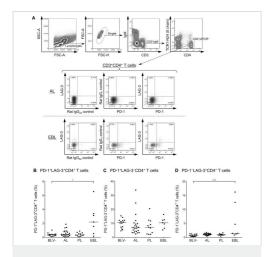
画像

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



Flow Cytometry - PerCP/Cy5.5 Conjugation Kit Lightning-Link (ab102911)

Image from Okagawa, Tomohiro, et al., Front Immunol., 8:650, doi: 10.3389/fimmu.2017.00650. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/



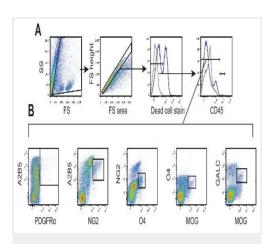
Flow Cytometry - PerCP/Cy5.5 Conjugation Kit - Lightning-Link

Image from Okagawa, Tomohiro, et al., Vet Res., 49(1):50, doi: 10.1186/s13567-018-0543-9. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Okagawa, Tomohiro, et al used PerCP/Cy5.5® Conjugation Kit -Lightning-Link® (ab102911) as part of examining the effect on proliferation of bovine leukemia virus (BLV)-specific T cells of the administration of Boch5D2. They used the kit to conjugate PerCP/Cy5.5® to anti-CD8 antibody, clone CC63, for use in flow cytometry. T-cell proliferation specific for BLV antigen stimulation. Carboxyfluorescein diacetate succinimidyl ester (CFSE)-labeled peripheral blood mononuclear cells were cultured in triplicate with fetal lamb kidney (FLK)-BLV antigen, control FLK antigen (A), or gp51 peptides (0.1 and 1 µg/ml) (B) for 6 days. The percentage of CFSElow cells in CD4+ and CD8+γδTCR- T cells was measured by flow cytometry. CFSElow cells represent cells proliferated during cultivation. Each dot represents the mean of three independent experiments. Significant differences were determined by Dunnett's multiple-comparison test across the time points. *,#,†P < 0.05 versus 0 dpi in each stimulation.

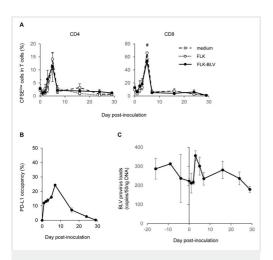
Okagawa, Tomohiro, et al used PerCP/Cy5. $5^{\$}$ Conjugation Kit - Lightning-Link $^{\$}$ (ab102911) as part of examining PD-1 and LAG-3 expression. They used the kit to conjugate PerCP/Cy5. $5^{\$}$ to monoclonal anti-CD3 antibody, clone MM1A, for use in flow cytometry.

Expression of PD-1 and LAG-3 on CD4+ T cells in BLV-infected cattle. A Gating strategy and representative dot plots for expression analyses of PD-1 and LAG-3 on lgM–CD3+CD4+ $\gamma\delta$ TCR– T cells from peripheral blood of BLV-infected cattle (AL and EBL). Values in the quadrants indicate percentages of cells. Percentages of PD-1+LAG-3+CD4+ T cells (B), PD-1+LAG-3-CD4+ T cells (C), and PD-1-LAG-3+CD4+ T cells (D) in CD3+CD4+ T-cell population in peripheral blood from BLV-uninfected (BLV – ; n = 15), AL (n = 22), PL (n = 11), and EBL cattle (n = 7). Bars indicate group median percentage. Significant differences between each group were determined using a Kruskal–Wallis test, where P < 0.05 and P < 0.001, indicated by asterisks (* and ****, respectively).



Flow Cytometry - PerCP/Cy5.5 Conjugation Kit-Lightning-Link

Image from Robinson, Andrew P., et al., PloS one, 9(9): e107649. doi: 10.1371/journal.pone.0107649. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/



Flow Cytometry - PE/Cy7® Conjugation Kit - Lightning-Link® (ab102911)

Image from Nishimori, Asami et al. PloS one vol. 12,4 e0174916. 26 Apr. 2017, doi:10.1371/journal.pone.0174916. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Robinson, Andrew P., et al used PerCP/Cy5.5 $^{\circledR}$ Conjugation Kit - Lightning-Link $^{\circledR}$ (ab102911) as part of characterizing oligodendroglial populations. They used the kit to conjugate PerCP/Cy5.5 $^{\circledR}$ to Mouse monoclonal anti-NG2 antibody for use in flow cytometry.

SJL/J mice were immunized with PLP139–151 and scored daily for clinical disease. A cohort of SJL/J mice was sacrificed, and spinal cords were analyzed by flow cytometry (n=5). (A) Cells were distinguished from debris by forward and side scatter then singlet cells were gated. Live cells were gated by dead cell exclusion, and CNS resident cells were identified as CD45– or CD45low. (B) Oligodendroglial cells were defined by double positive staining: A2B5+PDGFRα+ early OPCs, A2B5+NG2+ intermediate OPCs, NG2+O4+ late OPCs, O4+MOG+ pre-myelinating oligodendrocytes, and GALC+MOG+ mature oligodendrocytes.

Nishimori, Asami et al used PerCP/Cy5.5 $^{\circledR}$ Conjugation Kit - Lightning-Link $^{\circledR}$ (ab102911) as part of examining bovine leukemia virus infection. They used the kit to conjugate PerCP/Cy5.5 $^{\circledR}$ to antibovine CD8 antibody for use in flow cytometry.

A BLV-infected cow (#368, Holstein, female, 538 kg, 31 months old) was inoculated with 530 mg (1 mg/kg) of the purified 4G12 intravenously. (A) The proliferation of CD4+ and CD8+ T cells against BLV antigen. Peripheral blood mononuclear cells (PBMCs) isolated from the cow which was inoculated with 4G12 were labeled with carboxyfluorescein diacetate succinimidyl ester (CFSE) and cultured without stimulation (medium) or with the supernatant of FLK or FLK-BLV cells for 6 days. After the cultivation, the proliferation of T cells was immediately analyzed by flow cytometry. A P-value less than 0.05 was considered statistically significant. #, P < 0.05 (FLK-BLV, versus day 0; one-way ANOVA followed by Dunnett's test). (B) Changes in PD-L1 occupancy on circulating IgM+ B cells calculated by the binding of 4G12 to bovine PD-L1. The occupancy was estimated as the percentage of the in vivo PD-L1 binding occurred at the total available binding sites. (C) Changes in BLV provirus loads in the cow inoculated with 4G12; the y-axis shows the number of BLV

copies included in 50-ng DNA extracts of PBMCs. Data are means \pm SEM of at least three replicate experiments.

Add Add to Add Modifier Conjugate Quencher

Labeled

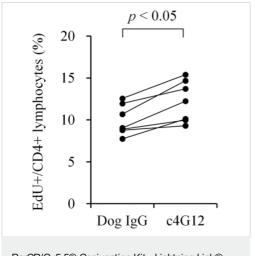
antibody

This illustration demonstrates a general procedure and will slightly vary dependent on the conjugate used.

Conjugation - PerCP/Cy5.5® Conjugation Kit (ab102911)

Purified

antibody



PerCP/Cy5.5® Conjugation Kit - Lightning-Link® labeling anti-canine CD4 antibody for Flow cytometry

Image from Maekawa N et al., Sci rep., 7(1):8951. Fig 2.; doi: 10.1038/s41598-017-09444-2. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Maekawa N et al. used ab102911 as part of examining a canine chimeric monoclonal antibody targeting PD-L1.

They used the kit to conjugate PerCP/Cy5.5[®] to anti-canine CD4 antibody for use in flow cytometry.

To evaluate cell proliferation, nucleotide analogue 5-ethynyl-2'-deoxyuridine (EdU) was added to the medium on day 2, and cells were harvested after incubation for another 2 h. The lymphocyte population was gated by forward scatter and side scatter, and the incorporation of EdU in (c) CD4+ cells was measured by a flow cytometer. Statistical analysis was performed with a Wilcoxon signed rank-sum test.

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