abcam

Product datasheet

APC Conjugation Kit - Lightning-Link® ab201807

107 References 11 图像

概述

产品名称

产品概述

APC偶联试剂盒 - Lightning-Link®

APC Conjugation Kit / APC Labeling Kit (ab201807) uses a simple and quick process for APC 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 APC using this kit:

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

The APC 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 <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 APC.

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[®] APC Labeling Kit. 705-0015 is the same as the 1 mg size. 705-0010 is the same as the 3 x 100 ug size. 705-0030 is the same as the 3 x 10 ug size. 705-0005 is the same as the 100 μ g size.

Amount and volume of antibody for conjugation to APC

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

¹ Ideal antibody concentration is 1mg/ml. 0.5 - 1 mg/ml can be used if the maximum antibody

说明

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volume is not exceeded. Antibodies > 1 mg/ml or < 0.5 mg/ml should be diluted /concentrated.

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.

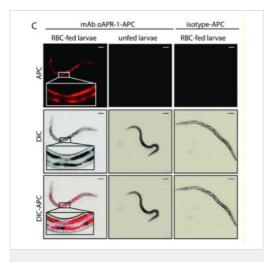
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存放说明

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

组 件	100 µg	1 mg	3 x 10 μg	3 x 100 μg
ab274126 - APC Conjugation Mix	1 x 100μg	1 x 1mg	3 x 10μg	3 x 100µg
Modifier reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl
Quencher reagent	1 x 200µl	1 x 200µl	1 x 200µl	1 x 200µl

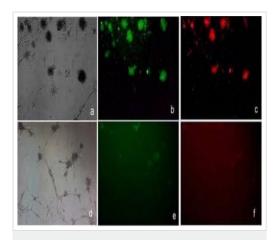
² BSA can also interfere with the use of the conjugated antibody in tissue staining.



APC Conjugation Kit - Lightning-Link

Image from Bouchery, Tiffany, et al., PLoS Pathog., 14(3): e1006931; doi: 10.1371/journal.ppat.1006931. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Bouchery, Tiffany, et al used APC Conjugation Kit - Lightning-Link[®] (ab201807) as part of examining methods for arresting hookworm development. They used the kit to conjugate APC to anti-Na-APR-1 antibody for use with live hookworms in fluorescence microscopy. Infective L3 stage hookworms were fed in vitro for 24 hr with RBC and treated with 10 µg of 11F3-APC monoclonal antibody against Na-APR-1 or with an isotype matched-RELM-APC control antibody. Larvae were allowed to empty their digestive contents for 2 hours in fresh DMEM before internal labelling was evaluated by confocal microscopy. Data representative of 50 larvae cultured in 3 independent experiments.



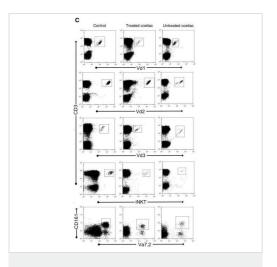
Fluorescence Microscopy - APC Conjugation Kit;-

Lightning-Link(ab201807)

Image from Haque, Nasreen S., Akaash Tuteja, and Niloufar Haque., PloS one 14.7 (2019): e0218944. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Haque, Nasreen S., Akaash Tuteja, and Niloufar Haque used APC Conjugation Kit - Lightning-Link[®] (ab201807) as part of examining as part of examining embryoid body formation. They used the kit to conjugate APC to anti-CCR8 antibody for use in immunofluorescence.

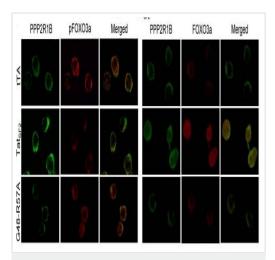
Anti CCL1 inhibits the expression of CCR8 and FoxP3 in mouse mesenchymal stem cells (mMSCs). Untreated (5a,-c) and anti-CCL1 treated (5d,-,f) cells were subjected to immunoflourescence with antibodies against FoxP3 (green;5b and e) or CCR8 (red;5c and f).



Dunne, Margaret R., et al used APC Conjugation Kit - Lightning-Link $^{(\!0)}$ (ab201807) as part of examining coeliac disease. They used the kit to conjugate APC to Vd3 antibody for use in flow cytometry. Dotplots show flow cytometry data for representative control, treated and untreated coeliac donors.

Flow Cytometry - APC Conjugation Kit;- Lightning-Link(ab201807)

Image from Dunne, Margaret R., et al., PLoS One, 8(10): e76008, doi: 10.1371/journal.pone.0076008. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

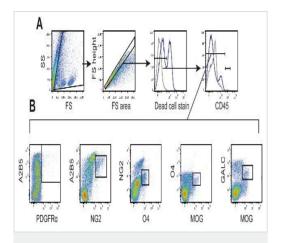


Fluorescence Microscopy - APC Conjugation Kit-Lightning-Link(ab201807)

Image from Kim, Nayoung, et al., PLoS Pathog., 6(9): e1001103. doi: 10.1371/journal.ppat.1001103. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

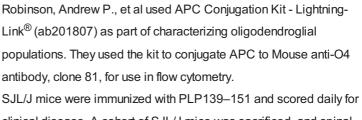
Kim, Nayoung, et al used APC Conjugation Kit - Lightning-Link[®] (ab201807) as part of examining apoptosis in HIV-1-infected CD4+ primary T cells. They used the kit to conjugate APC to anti-pFOXO3a antibody for use in confocal microscopy.

Jurkat T cells expressing tTA alone, TatSF2+tTA, or TatSF2G48-R57A +tTA were stained with antibodies against PPP2R1B (first and forth columns of panels, green), pFOXO3a (second column, red), and FOXO3a (forth column, red).

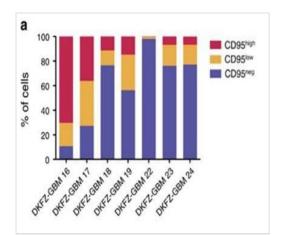


Flow Cytometry - APC Conjugation Kit;- Lightning-

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/



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.



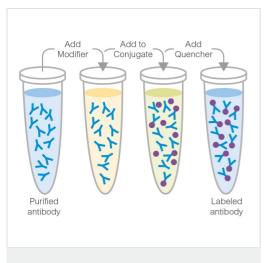
Flow Cytometry - APC Conjugation Kit- Lightning-

Link

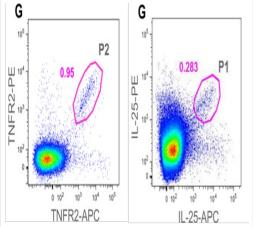
Image from Drachsler, Moritz, et al., Cell death & disease 7.4 (2016): e2209-e2209. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Drachsler, Moritz, et al used APC Conjugation Kit - Lightning-Link[®] (ab201807) as part of examining expression of CD95. They used the kit to conjugate APC to anti-human CD95 for use in flow cytometry.

The graph shows the CD95 expression in seven patient tumor samples.



APC conjugation kit

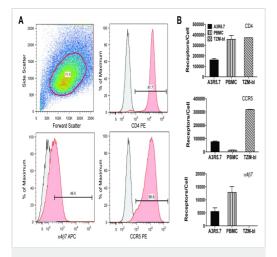


memory B cells from TNFR2 and IL-25 immunised mice.

Starkie DO et al. used ab201807 to identify antigen-specific mouse

APC Conjugation Kit - Lightning-Link® labeling IL-25 and TNFR2 extracellular domain for Flow cytometry

Image from Starkie DO et al., PLoS One, 11(3):e0152282. Fig 3 and 2.; doi: 10.1371/journal.pone.0152282. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/



APC Conjugation Kit - Lightning-Link® labeling antihuman 4ß7 antibody for Flow cytometry

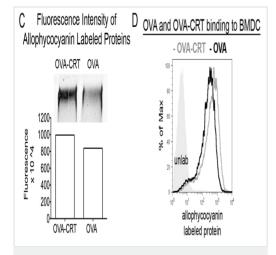
Image from McLinden RJ et al., PLoS One, 8(11):e77756. Fig 2.; doi: 10.1371/journal.pone.0077756. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

McLinden RJ et al. used ab201807 as part of examining HIV-1 neutralizing antibodies.

They used the kit to conjugate APC to anti-human $4\beta7$ antibody for use in flow cytometry.

A. Flow cytometric analysis of CD4, CCR5 and $\alpha4\beta7$ expression in the A3R5.7 cell line. 0.5 x 106 cells were singly stained for 30 minutes with fluorochrome-conjugated antibodies as shown followed by fixation in 2% paraformaldehyde. Data are representative of at least two independent experiments. Isotype controls are shown in grey. Nearly all cells were positive for CD4 and CCR5 while approximately half were positive for $\alpha4\beta7$.

B. Comparison of cell surface CD4, CCR5 and $\alpha4\beta7$ receptor densities in various cell targets. 0.5 x 106 cells were stained with fluorochrome-conjugated antibodies and compared to defined populations of similarly stained Quantum Simply Cellular beads. PBMC were stimulated with CD3.8 bi-specific antibody in the presence of 50U/mL rhlL-2. Assuming monovalent antibody-to-surface receptor binding, the Antibody Binding Capacity (ABC) calculated is equivalent to receptors/cell. Data represents the mean of two separate experiments. TZM-bl cells express high levels of CD4 and CCR5 but are negative for $\alpha4\beta7$ while A3R5.7 cells possess CCR5 and $\alpha4\beta7$ densities more similar to PBMC. CD4 expression on TZM-bl was beyond assay range.



APC Conjugation Kit - Lightning-Link® labeling OVA and OVA-CRT for in-gel Fluorescence and Flow cytometry

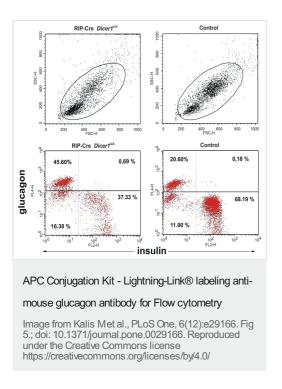
Image from Del Cid N et al., PLoS One, 7(7):e41727. Fig 2.; doi: 10.1371/journal.pone.0041727. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Del Cid N et al. used ab201807 as part of examining antigen cross-presentation.

They used the kit to conjugate APC to ovalbumin (OVA) and ovalbumin-calreticulin fusion protein (OVA-CRT) for use in in-gel fluorescence and flow cytometry.

OVA-CRT and OVA were labeled with allophycocyanin. (C)
Labeling intensity was determined by fluorescence imaging of the proteins after separation by SDS-PAGE (inset). Fluorescence intensity was quantified for the indicated proteins. (D) Binding of fluorescent proteins to BMDC was assessed by flow cytometry.

BMDC were incubated with labeled proteins on ice before being analyzed by flow cytometry. BMDC not incubated with proteins are depicted as a grey filled.



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