# abcam

# Product datasheet

# PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® ab102918

★★★★★ 1 Abreviews 106 References 画像数 13

#### 製品の概要

#### 製品名

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PE / R-Phycoerythrin Conjugation Kit - Lightning-Link®

R-PE Conjugation Kit/ R-PE Labeling Kit ab102918 uses a simple and quick process for PE 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 R-PE using this kit:

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

The PE 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 R-PE.

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  $^{\circledR}$  R-PE Labeling Kit. 703-0004 is the same as the 3 mg size. 703-0003 is the same as the 5 x 600 ug size. 703-0010 is the same as the 3 x 60 ug size. 703-0030 is the same as the 3 x 10 ug size. 703-0015 is the same as the 600  $\mu$ g size. 703-0005 is the same as the 60  $\mu$ g size.

#### Amount and volume of antibody for conjugation to R-PE

Kit size	Recommended amount of antibody	Maximum antibody volume <sup>1</sup>		
3 x 10 µg	3 x 10 µg	3 x 10 µL		
60 µg	1 x 60 µg	1 x 60 µL		
3 x 60 µg	3 x 60 µg	3 x 60 µL		
600 µg	1 x 600 µg	1 x 600 μL		

# 特記事項

5 x 600 µg	5 x 600 μg	5 x 600 μL		
3 mg	1 x 3 mg	1 x 3 mL		

The selling size of this product is now based on the amount of antibody that can be conjugated with the kit; the amount of antibody advised that can be used with the kit has also been updated to reflect what will give the best conjugation results. The quantity and formulation of reagents provided have not changed, if you have been previously using the kit successfully with a different amount of antibody, there is no need to change the way that you are using the kit.

# **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 <sup>1</sup>	0.1% BSA	50% glycerol		
0.1% sodium azide	PBS	Potassium phosphate		
Sodium chloride	HEPES	Sucrose		
Sodium citrate	EDTA	Trehalose		

<sup>&</sup>lt;sup>1</sup> 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<sup>®</sup> 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.

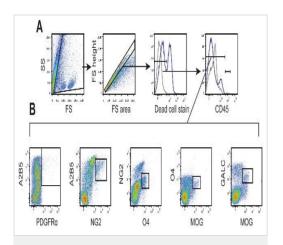
<sup>&</sup>lt;sup>1</sup>Ideal antibody concentration is 1mg/ml. 0.5 - 1 mg/ml can be used if the maximum antibody volume is not exceeded. Antibodies > 1 mg/ml or < 0.5 mg/ml should be diluted /concentrated.

#### 保存方法

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

内容	60 µg	600 µg	5 x 600 μg	3 x 60 µg	3 x 10 µg	1 x 3 mg
Modifier reagent	1 x 200µl	1 x 200µl	1 x 1200µl	1 x 200µl	1 x 200µl	1 x 1200µl
Quencher reagent	1 x 200µl	1 x 200µl	1 x 1200µl	1 x 200µl	1 x 200µl	1 x 1200µl
ab274122 - R-PE mix	1 x 60µg	1 x 600µg	5 x 600µg	3 x 60µg	3 x 10µg	1 x 3mg

#### 画像

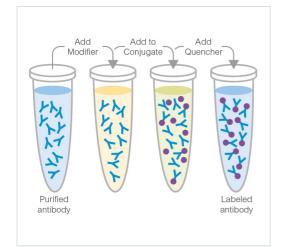


Flow Cytometry - PE / R-Phycoerythrin Conjugation Kit Lightning-Link

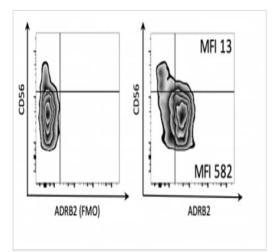
Image from Robinson, Andrew P., et al., PloS one, 9(9):e107649. doi: 10.1371/journal.pone.0107649. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Robinson, Andrew P., et al used PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® (ab102918) as part of characterizing oligodendroglial populations. They used the kit to conjugate PE / R-Phycoerythrin to Mouse monoclonal anti-GALC antibody for use in flow cytometry.

SJL/J mice were immunized with PLP<sub>139-151</sub> 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<sup>-</sup> or CD45<sup>low</sup>. (B) Oligodendroglial cells were defined by double positive staining: A2B5+PDGFRalpha<sup>+</sup> early OPCs, A2B5+NG2+ intermediate OPCs, NG2+O4+ late OPCs, O4+MOG+ pre-myelinating oligodendrocytes, and GALC+MOG+ mature oligodendrocytes.



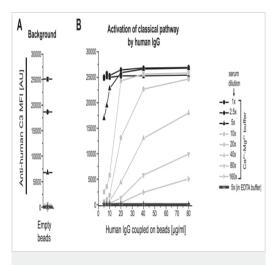
Conjugation - R-Phycoerythrin Conjugation (ab102918)



Bigler MB et al. used ab102918 to assess expression of the epinephrine receptor (ADRB2) on CD3- CD56+ lymphocytes by flow cytometry.

PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® labeling ADRB2 antibody for Flow Cytometry

Image from Bigler MB et al., PloS One, 10(12):e0145635. Fig 3.; doi: 10.1371/journal.pone.0145635. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

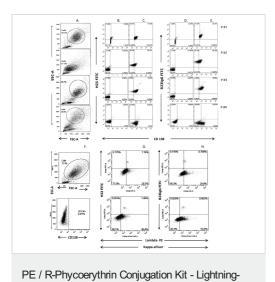


PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® labeling F(ab')2 antibody for Bead array

Image from Ayoglu B et al., PLoS One 9(5):e96403. Fig 2.; doi: 10.1371/journal.pone.0096403. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Ayoglu B et al. used ab102918 to detect complement activation driven C3 deposition on beads.

For measuring the background, classical, lectin & alternative and only alternative pathway activation, a serially diluted serum sample (1:1–1:160) was applied to empty, human lgG. The assay buffer for serum dilution contained either Ca2+-Mg2+, which promotes complement activation, or EDTA, which blocks complement activation. Plot displays for each serum dilution the respective median fluorescence intensity (MFI) value against varying concentrations of human lgG.

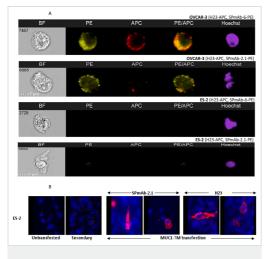


Link® labeling anti-MUC1 SP antibodies FACS

Image from Koyjazin R et al., PLoS One, 9(1):e85400. Fig 3.; doi: 10.1371/journal.pone.0085400. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Kovjazin R et al. used ab102918 to evaluate the cell surface presence of the MUC1 SP domain by FACS.

The cell surface presence of the MUC1 SP domain was evaluated by gated FACS analysis on PC cells in fresh BM aspirates obtained from MM patients (A–E) and normal naive sample (F–H).



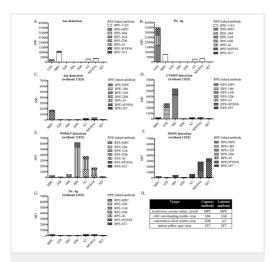
PE / R-Phycoerythrin Conjugation Kit - Lightning-

 $\label{link} \mbox{Link} \mbox{$\mathbb{R}$ labeling SPmAb-6 antibody for Imagestream}$ 

analysis

image from Kovjazin R et al., PLoS One, 9(1):e85400. Fig 2.; doi: 10.1371/journal.pone.0085400. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

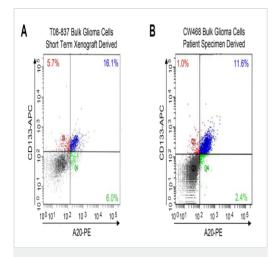
Kovjazin R et al. used ab102918 for detection of MUC1 SP on the membrane of MUC1-positive tumor cells.



PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® labeling a panel of antibodies for Microsphere immunoassay based on Luminex

Image from Charlermroj R et al., PloS One, 8(4):e62344. Fig 2.; doi: 10.1371/journal.pone.0062344. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/ Charlermroj R et al. used ab102918 as part of an experiment studying four different plant pathogens: a fruit blotch bacterium Acidovorax avenae subsp. citrulli (Aac), chilli vein-banding mottle virus (CVbMV, potyvirus), watermelon silver mottle virus (WSMoV, tospovirus serogroup IV) and melon yellow spot virus (MYSV, tospovirus).

(A-G) X-axis is antibody-coated microsphere and y-axis is median fluorescent intensity (MFI) from each RPE-labeled antibody. (H) Summary of selected antibody pair sets for the detection of the four plant pathogens.



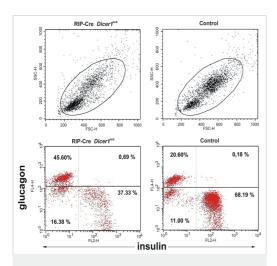
PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® labeling A20 antibody for FACS

Image from Hjelmeland AB et al., PLoS Biol., 8(2):e1000319. Fig 2.; doi: 10.1371/journal.pbio.1000319.Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Hjelmeland AB et al. used ab102918 as part of an experiment studying the mechanisms that cause A20 regulator to have differential effects on tumor growth and cancer cell behaviors.

They used the PE labelling kit to conjugate phycoerythrin to A20 antibody for use in FACS.

FACS plots are shown for T08-837 (A) and CW468 (B).



PE / R-Phycoerythrin Conjugation Kit - Lightning-Link® labeling anti-mouse insulin antibody for Flow Cytometry

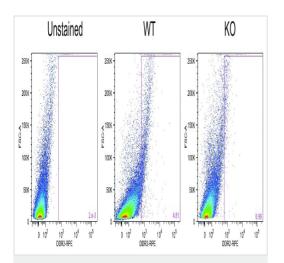
Image from Kalis, Met al., PLoS One, 6(12):e29166. Fig 5.; doi: 10.1371/journal.pone.0029166. Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/

Kalis M et al. used ab102918 as part of examining beta-cells deletion of Dicer1.

They used the kit to conjugate PE to anti-mouse insulin antibody for use in flow cytometry.

Islet cells suspension from 50 islets from 8 weeks old littermates and RIP-Cre Dicer1 $\Delta/\Delta$  mice were stained for insulin and glucagon and analyzed by flow cytometry. Side scatter (SSC) and forward scatter (FSC) analysis of islet cells and the respective gate used to identify live cells. Littermate control islet cells contained significantly more insulin-positive cells (P<0.001) and less glucagon-positive cells (P<0.05) than the RIP-Cre Dicer1 $\Delta/\Delta$  islet cells.

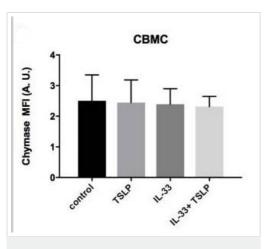
Representative of 3 independent experiments.



PE conjugation kit used to label DDR2 antibody for Flow Cytometry

Bagchi RA et al. BMC Biol. 2016 Mar 17;14:21 Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/ RA Bagchi et al used PE conjugation kit / PE labeling kit ab102918 as part of examining the conversion of fibroblasts to myofibroblasts. They used the PE labeling kit to conjugate phycoerythrin to a DDR2 antibody for use in flow cytometry.

Charts are forward-scatter plots illustrating flow cytometry analysis of cardiac cells from WT and scleraxis KO mice. Left column, unstained cells; center column, stained cells from WT tissue; right column, stained cells from scleraxis KO tissue. Results are representative of assessments from n = 3 independent tissue samples. Purple outline denotes labeled cells, and is derived from unstained plots.



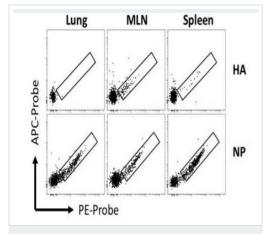
PE conjugation to an anti-chymase antibody with ab102918 PE Conjugation Kit

Image from Rönnberg E et al., Frontiers in immunology., 10, 1361. Fig 4C.; doi: 10.3389/fimmu.2019.01361 Reproduced under the Creative Commons license https://creativecommons.org/licenses/by/4.0/.

Ronnberg E et al. conjugated PE to an anti-chymase antibody with ab102918 PE Conjugation Kit as part of investigating the role of IL-33 in mast cells.

The mean fluorescence intensity of chymase expression in cells was measured by intra-cellular flow cytometry staining and normalized to the respective isotype control.

This data shows that there was no significant change in the expression on chymase in cord blood-derived mast cells (CBMC) when treated with different combinations of IL-33 and TSLP.

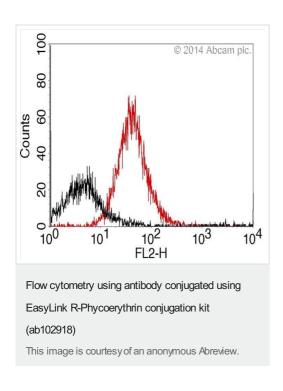


Conjugation of recombinant influenza A H1N1 NP protein with PE or APC fluorochromes using ab102918

Image from Tan HX et al., Frontiers in immunology., 10, 611.. Fig 4A; doi: 10.3389/fimmu.2019.00611
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Tan H-X et al. labeled recombinant influenza A H1N1 NP protein with PE or APC fluorochromes using ab102918 PE / R-Phycoerythrin Conjugation Kit and <u>ab201807</u> APC Conjugation Kit.

Representative HA and NP probe staining in flow cytometry of GC B cells (B220+ lgD- CD38lo GL7+) isolated from lung inducible bronchus-associated tissues (iBALT), mediastinal LN (MLN), and spleen of mice at d35 post-infection with A/Puerto Rico/8/34 (PR8).



Flow cytometry histogram showing integrin beta-3 positive population of platelets from wildtype and knockout mice. Integrin beta-3 antibody was conjugated using EasyLink R-Phycoerythrin conjugation kit (ab102918). Flow cytometry was performed using platelets from wild type and integrin beta-3 knockout mice. Mice that expressed beta-3 (shown in red) had a clear shift in FL-2 fluorescence over beta-3 knockout mice (shown in black).

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