Alkaline phosphatase Conjugation Kit - Lightning-Link® ab102850

Overview

**Product name**
Alkaline phosphatase Conjugation Kit - Lightning-Link®

**Product overview**
Alkaline Phosphatase Conjugation Kit / Alkaline Phosphatase Labeling Kit ab102850 uses a simple and quick process for alkaline phosphatase 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 Alkaline Phosphatase using this kit:
- add modifier to antibody and incubate for 3 hrs
- add quencher and incubate for 30 mins
The alkaline phosphatase 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 HRP.

Notes

This product is manufactured by Expedeon, an Abcam company, and was previously called Lightning-Link® Alkaline Phosphatase Labeling Kit. 702-0005 is the same as the 100 µg size. 702-0010 is the same as the 3 x 100 µg size. 702-0030 is the same as the 3 x 10 µg size. 702-0015 is the same as the 1 mg size.

**Amount and volume of antibody for conjugation to Alkaline Phosphatase**

<table>
<thead>
<tr>
<th>Kit size</th>
<th>Recommended maximum amount of antibody</th>
<th>Maximum antibody volume</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 x 10 µg</td>
<td>3 x 10 µg</td>
<td>3 x 10 µL</td>
</tr>
<tr>
<td>100 µg</td>
<td>100 µg</td>
<td>100 µL</td>
</tr>
<tr>
<td>3 x 100 µg</td>
<td>3 x 100 µg</td>
<td>3 x 100 µL</td>
</tr>
<tr>
<td>1 mg</td>
<td>1 mg</td>
<td>1 mL</td>
</tr>
</tbody>
</table>

1 Ideal antibody concentration is 1mg/ml. 0.5 - 1 mg/ml can be used if the maximum antibody volume is not exceeded. Antibodies > 5mg/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
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
Thiomersal
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.

Tested applications Suitable for: Conjugation

Properties

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

<table>
<thead>
<tr>
<th>Components</th>
<th>1 mg</th>
<th>100 µg</th>
<th>3 x 10 µg</th>
<th>3 x 100 µg</th>
<th>30 µg</th>
</tr>
</thead>
<tbody>
<tr>
<td>AP-Mix</td>
<td>1 x 1mg</td>
<td>1 x 100µg</td>
<td>3 x 10µg</td>
<td>3 x 100µg</td>
<td>3 x 10µg</td>
</tr>
<tr>
<td>Modifier reagent</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
</tr>
<tr>
<td>Quencher reagent</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
<td>1 vial</td>
</tr>
</tbody>
</table>

Applications Our Abpromise guarantee covers the use of ab102850 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conjugation</td>
<td>Use at an assay dependent dilution.</td>
<td></td>
</tr>
</tbody>
</table>
This illustration demonstrates a general procedure and will slightly vary dependent on the conjugate used.

Thomas FC et al. used ab102850 as part of examining the toxicity of p17 protein assemblies. They used the kit to conjugate Alkaline phosphatase to anti-bovine Hp antibody for use in ELISA.

Boxplot showing Hp concentration (μg/ml) in two SCC categories of composite milk samples. * indicates an extreme value (values greater than 3 interquartile range (IQR) away from 25th or 75th percentile); IQR = 3rd quartile - 1st quartile (represented by the height of the box). ° indicates an outlier value (values greater than 1.5 interquartile range (IQR) away from 25th or 75th percentile); IQR = 3rd quartile - 1st quartile (represented by the height of the box).

Grabias B et al. used ab102850 as part of developing a rapid detection of Plasmodium falciparum infection in mosquitoes. They used the kit to conjugate Alkaline phosphatase to anti-Plasmodium falciparum circumsporozoite protein antibody for use in ECL slot blot assay.

Evaluation of whether conjugation of primary mAb 2A10 with alkaline phosphatase (1°-AP) enhanced sensitivity for the detection of Pfocyst prepared from mosquito lysates when compared to the use of AP-conjugated secondary antibody (2°-AP). Detection limits were compared for each protocol by fitting the band intensities of serially diluted oocysts to a Michaelis-Menten regression curve and
establishing a cutoff intensity threshold of mean + 2 SD from unfed mosquito specimens run on the same blot. Labeled primary antibody displayed overall higher band intensities across the range of oocyst dilutions examined and achieved lower limits of detection than the typical sandwich antibody format (0.009 oocyst versus 0.02 oocyst, respectively). The removal of an additional antibody incubation step also contributed to an overall shorter assay time in the newly developed slot blot protocol.

Charlemroj R et al. used ab102850 to run a sandwich ELISA and compare its sensitivity with a microsphere immunoassay based on Luminex using the same set of antibodies.

Thaitrong N et al. used ab102850 to run a microfluidic sandwich ELISA for Acidovorax citrulli (Ac), watermelon silver mottle virus (WSMoV), and melon yellow spot virus (MYSV) screening. Nine different conditions for each disease panel were tested on the microfluidic platform using combinations of three concentrations of capture Ab (11E5, 2D6, and 5E7) and three concentrations of detection Ab (MPC-AP, MYSV6-AP).

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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