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LEADGENE BIOMEDICAL, INC.

Product Information & Manual

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HyLink™ HRP Conjugation Kit

Cat no. LDG0001RC/LDG0002RC/LDG0003RC

Product Overview

Package component

Item	LDG0001RC (10 µg x 2)	LDG0002RC (100 µg x 2)	LDG0003RC (1 mg x 1)
HRP	2 vials	2 vials	1 vial
10X Modifier	1 vial	1 vial	1 vial
Reduction reagent	1 vial	1 vial	1 vial
10X Quencher	1 vial	1 vial	1 vial

Description

Horseradish peroxidase (HRP) is a 44 kDa glycoprotein that can be modified to crosslink with antibodies or other proteins, without compromising the enzymatic activity. HRP conjugates can be widely used in ELISA, western blotting and IHC due to its enzymatic characteristics. Leadgene HyLink™ HRP Conjugation Kit is designed for HRP conjugation of a small quantity (10 µg-1 mg) of antibody or protein. It provides a rapid and easy process with high efficiency to conjugate HRP to antibody or protein. The kit provides all the necessary components and high-activity HRP for conjugation.

Storage and Stability

(1) Stored at -20°C.
(2) Is stable for one year under proper storage conditions.

Procedure

HRP conjugation protocol:

(1) Dissolve antibody in PBS or other buffer that do not contain amine, Tris, NaN3 or glycerol. Add **10X Modifier** to antibody (e.g. 1 µL of 10X HRP modifier for 9 µL of antibody).
(2) Spin down the vial of **HRP** before use.
(3) Make sure all buffers are well dissolved before use. If not, please vortex the vial to make salts dissolved.
(4) Remove cap of the vial of **HRP** and pipette antibody into the vial. Mix gently by pipetting several times.
(5) Cover the cap on the vial and incubate in the dark at room temperature for 3 hours.
(6) After incubating, add 1/20 volume of **Reduction reagent** (e.g. 1 µL of reduction reagent for 20 µL of antibody-HRP mixture) and mix gently by pipetting. Incubate at room temperature for 30 minutes.
(7) Add **10X Quencher** (e.g. 1 µL of 10X Quencher for 9 µL of antibody-HRP mixture) and mix gently by pipetting. The conjugates can be used after 30 minutes.

* For protein conjugation, the amount of protein can be calculated by formula below:

Quantities of protein = quantities of kit (e.g. 10 µg) x (M.W. of target protein) / (150 (M.W. of IgG)

Important notes

(1) Antibody concentrations of 1-4 mg/mL generally give optimal results. Recommended amount and volume of antibody for optimal results.

Kit size	Antibody amount	Reaction volume
2 x 10 µg	10-20 µg	4-20 µL
2 x 100 µg	100-200 µg	40-200 µL
1 x 1 mg	1-2 mg	400-2000 µL

(2) Common non-buffering salts (e.g. sodium chloride) have no effect on conjugation efficiency. Avoid buffer component that contains primary amine (e.g. amino acidor ethanolamine) and thiols (e.g. 2-Mercaptoethanol or DTT).

Components that have on effect or little effect on labeling reaction:

-up to 20 mM Tris

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-up to 10% glycerol

-up to 0.02% sodium azide

Disclaimer

This product is for research use only and is not intended for diagnostic use.

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