

LinkAble™ HRP Antibody Conjugation Kit

Intended Use

LinkAble HRP Antibody Conjugation Kit offers a simple and efficient procedure for covalent conjugation of antibodies with HRP by targeting primary amine groups (e.g. lysines). Designed for ease of use giving reliable, reproducible results, this LinkAble kit streamlines the conjugation process to create an HRP antibody conjugate quickly and without specialized expertise.

The kit is optimized for single-use. Multi-packs are available providing flexibility to meet varying experimental needs.

Key Benefits

Ease & Efficiency: Streamlined protocols ensure a simple, fast, and reliable conjugation process. The kit requires only 30 seconds of hands-on time and produces conjugates that are ready to use in 2 hours, without any loss of antibody associated with buffer exchange and purification.

Reproducibility: Optimized reagents deliver consistent, high-quality results. The kit is available in 25 µg, 100 µg, and 1 mg antibody input sizes, which have been optimized to perform identically across all three sizes, allowing scaling of conjugation reactions.

Streamlined & Optimized Usability: The ready-to-use format eliminates the need for extra steps in the procedure, minimizes hands-on time, maximizes convenience, and reduces the risk of erroneous calculations. An intuitive, user friendly design includes screw cap vials that can be easily centrifuged to recover the sample.

Technical Hints & Limitations

- Although it is recommended to use a purified antibody in a suitable buffer (e.g. PBS) and free from additives, the LinkAble HRP Antibody Conjugation Kit produces high quality conjugates in the presence of some buffers/additives.

Note: The kit is not compatible with certain components, see table below.

Components	Compatible
Amine-free buffer (MES, PBS, HEPES, bicarb, borate)	Yes
Tris	≤ 100 mM
Sugars (trehalose / sucrose)	Yes
EDTA	Yes
Proclin	Yes
Sodium azide	No
BSA	≤ 0.1%
Glycerol	≤ 10%
Amine containing reagents (glycine, arginine)	No

- If multiple, non-optimal parameters are used in the conjugation reaction (concentration, additives, etc.) poor quality conjugates may result.
- Centrifuge all vials before resuspending to ensure all contents are at the bottom of the vials.
- In each step, gently pipette the sample up and down to mix.
- Purification of the conjugated antibody is not necessary, though a user may choose to do so.

Precautions

Wear appropriate protective gloves, clothing, and eye/face protection. Wash hands thoroughly after handling.

Review SDS information for all kit components, including the borate buffer.

Storage & Stability

12 months from date of receipt. Store kit at $\leq -20^{\circ}\text{C}$ immediately upon receipt.

Materials Provided

Size	10X Buffer	Reagent A*	Reagent B*
25 μg	200 μL	1 vial	1 vial
100 μg	200 μL	1 vial	1 vial
1 mg	200 μL	1 vial	1 vial
3 x 25 μg	200 μL	3 x 1 vial	3 x 1 vial
3 x 100 μg	200 μL	3 x 1 vial	3 x 1 vial
5 x 1 mg	3 x 200 μL	5 x 1 vial	5 x 1 vial

**Reagent A & B are lyophilized.*

Other Materials Required

Antibody to be conjugated. Refer to Technical Hints and Limitations for more information on the antibody requirements.

Explore R&D Systems antibodies, including the Novus portfolio, with conjugation-ready, BSA- and azide-free formats at rndsystems.com/products/antibodies.

Starting Mass & Volume of Antibody

RECOMMENDED

The kit produces high quality conjugates using IgG antibodies with a concentration of 100 $\mu\text{g}/\text{mL}$ or greater. For optimal results, it is recommended to carry out the conjugation reaction with antibody concentration at 1 mg/mL.

Reaction Size	Min (μg)	Optimal (μg)	Max (μg)
25 μg	10	25	100
100 μg	25	100	400
1000 μg	250	1000	4000

General Protocol

1. Add sufficient volume of 10X Reaction Buffer to the antibody so the final Reaction Buffer concentration is 1X.
2. Transfer the antibody solution in 1X Reaction Buffer to Reagent A tube and gently pipette up and down to dissolve.
3. Incubate the tube at room temperature for 60 minutes **in the dark**.
4. Transfer the antibody solution to Reagent B tube and gently pipette up and down to dissolve.
5. Incubate the tube at room temperature for 60 minutes **in the dark**.
6. The conjugated antibody is now ready to use.
Note: No conjugation clean-up is required before the labeled antibody can be used. Do not add sodium azide for long term storage.

Conjugate Storage

The conjugated antibody should be stored at 4°C .

Troubleshooting

The efficiency of the conjugation reaction means there are small quantities of materials in each of the vials. We recommend centrifuging before starting the conjugation to ensure all material is at the bottom of the vial.

For additional troubleshooting assistance or any other questions, contact our technical support team. rndsystems.com/support/contact-us/technical-support