## For Research Use Only Blotting Accelerator

Catalog Number: PR20039



Description

This ready-to-use solution is specifically used for membrane blocking and antibody dilution in Western blot experiments and designed to significantly shorten the blotting workflow to around 40 minutes or less. It contains special organic compounds, which can significantly shorten the reaction time of experiments and thus improve experimental efficiency.

This product contains inert proteins of different molecular weight ranges, which can effectively reduce the background and protect the activity of the antibody. Antibody working solution diluted with this product can usually be stored at 4°C for more than 3 months.

This product contains pathology grade preservatives and is compatible with HRP, so it can dilute primary antibodies (including phosphorylated primary antibodies), HRP-labeled primary antibodies, or secondary antibodies. It is also possible to dilute fluorescently labeled primary and secondary antibodies. Exclusion: This product contains BSA and cannot be used to dilute anti-BSA primary antibodies. This product is a semi-transparent colloid. Please do not use it if there are insoluble or lumpy aggregates.

**Package** 

Storage

**Notes** 

100 mL/500 mL

Store at 2-8°C for a year.

- 1. If you use this product for the first time, you may need to optimize the antibody dilution ratio. It is recommended to set the concentration of secondary antibody (generally 0.05-0.1 g/mL) first, and optimize the concentration of primary antibody after setting the reaction time of primary antibody and secondary antibody.
- 2. If there are individual unsatisfactory results, you can adjust the concentration of the primary antibody, secondary antibody, or washing time, and optimize the imaging or exposure time appropriately.
- 3. The diluted primary antibody or secondary antibody can be stored at 4°C for more than 3 months. However, the length of storage time varies among different primary or secondary antibodies, so please make your judgment on the specific stability period.
- 4. This product contains the preservative ProClin 300. Please wear a lab coat and disposable gloves.
- 5. This product is only for professional laboratory scientific research use, not for medical, food, or pharmaceutical use.

1. Do I need to use this product for the entire duration of the blotting protocol?

A: No, it is not necessary. Just follow the recommended conditions for the steps using this product, and the steps not using this product should be done in the normal way.

2. Is there any requirement for the manufacturer of primary antibody or secondary antibody to use this product?

 $\hbox{A: There is no requirement.}\\$ 

3. When using this product, what steps can be set to stop and how should they be set?

A: The optional stopping points for using this product are:

(1) Blocking (optional).

Soak the membrane directly in PBST/TBST wash solution or in this product at 4°C as a stopping point (30 minutes to 24 hours). When restarting the experiment, move the membrane to room temperature to equilibrate the temperature for subsequent experiments.

(2) After the primary antibody.

After the primary antibody reaction, directly soak the membrane in PBST/TBST washing solution and let it stand at room temperature as a stopping point (30 minutes to 4 hours is appropriate). When restarting the experiment, the membrane should be washed 3 times with washing solution for 1 minute each time. Secondary antibody can be added after washing.

(3) After the secondary antibody.

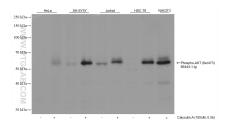
After the secondary antibody reaction, soak the membrane directly in PBST/TBST wash solution and let it stand at room temperature as a stopping point (30 minutes to 4 hours is preferred). When restarting the experiment, the membrane should be washed 3 times with the washing solution for 1 minute each time. After washing, the substrate can be added for imaging.

Caution

Before using this product, please equilibrate to room temperature (25°C) or warm up to 37°C to ensure effectiveness. There is no significant effect on experimental results between incubation at room temperature (25°C) or at 37°C. However, if the room temperature is lower than 25°C, it is recommended to warm up the solution.

**FAQ** 

## Validation Data

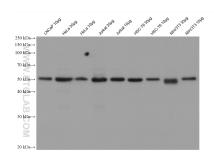


Various samples were loaded at 30 ug per well and Various samples were loaded at different volumes subjected to Western blot, unblocked. Primary antibody 66444-1-Ig (Phospho-AKT (Ser473) Mouse per well and subjected to Western blot, unblocked. Primary antibody 15282-1-AP (HSPD1 Rabbit Monoclonal Antibody) was diluted with PR20039 at Polyclonal Antibody) was diluted with PR20039 at 1:10,000 and incubated at room temperature for 15 minutes. The secondary antibody RGAM001 was 1:50,000 and incubated at room temperature for 18 minutes. The secondary antibody RGAR001 was diluted with PR20039 at 1:20,000 and incubated at... diluted with PR20039 at 1:20,000 and incubated at...

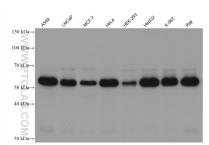
75 kDa –

room temperature for 8 minutes.

100 kDa

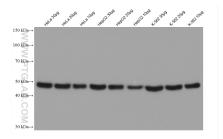


Various samples were loaded at different volumes per well and subjected to Western blot, unblocked. Primary antibody 66031-1-Ig (Alpha Tubulin Mouse Monoclonal Antibody) was diluted with PR20039 at 1:20,000 and incubated at room temperature for 3 minutes. The secondary antibody RGAM001 was diluted with PR20039 at 1:20,000 and incubated at... room temperature for 3 minutes.

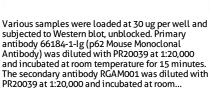


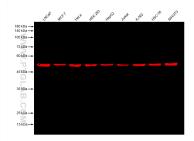
room temperature for 10 minutes.

Various samples were loaded at 30 ug per well and subjected to Western blot, unblocked. Primary antibody 66210-1-1g (JNK Mouse Monoclonal Antibody) was diluted with PR20039 at 1:10,000 and incubated at room temperature for 15 minutes. The secondary antibody RGAM006 was diluted with PR20039 at 1:10.000 and incubated at room... temperature for 10 minutes. temperature for 10 minutes.

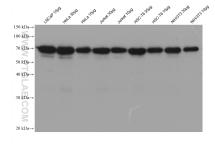


Various samples were loaded at different volumes per well and subjected to Western blot, unblocked. Primary antibody 80713-1-RR (Beta Tubulin Rabbit Recombinant Antibody) was diluted with PR20039 at 1:10,000 and incubated at room temperature for 18 minutes. The secondary antibody RGAR001 was diluted with PR20039 at 1:20,000 and incubated at... room temperature for 8 minutes.





Various samples were loaded at 30 ug per well and subjected to Western blot, unblocked. Primary antibody 80713-1-RR (Beta Tubulin Rabbit Recombinant Antibody) was diluted with PR20039 at 1:20,000 and incubated at room temperature for 15 minutes. The secondary antibody RGAR006 was diluted with PR20039 at 1:10,000 and incubated at... room temperature for 10 minutes.



Various samples were loaded at different volumes per well and subjected to Western blot, unblocked. Primary antibody HRP-66095 (HRP-conjugated Lamin B1 Mouse Monoclonal antibody) was diluted with PR20039 at 1:5,000 and incubated at room temperature for 3 minutes without a secondary antibody involved.