EZ-Western Stripping Buffer

Cat. No. DG-WSB500

FOR RESEARCH USE ONLY.

NOT FOR USE IN DIAGNOSTIC PROCEDURES.

▪ Product Description

The Stripping Buffer is used in Western Blot experiments to remove primary and secondary antibodies from the transfer membrane (NC, PVDF). It is especially useful in experiments with limited samples when detecting one or more proteins with the same molecular weight. This allows you to confirm proteins without performing multiple electrophoresis and blotting experiments, saving both sample and time.

EZ-Western Stripping Buffer uses the principle of affinity chromatography to effectively separate the antigen-antibody complex while minimizing the denaturation and loss of the membrane-bound protein (antigen).

▪ Contents

Component	Volume
EZ-Western Stripping Buffer	500 ㎖

▪ Storage and Stability

Store at 2 ~ 8°C and it is stable for 1 year from the date of manufacture.

▪ General Protocol

- Before stripping, store the membrane in TBS-T or PBS-T at 4°C.
- If the membrane dries out, the antigen may denature, making it unusable for the experiment.

1. Take the EZ-Western Stripping Buffer from the refrigerator and allow it to reach room temperature before use.
2. Add enough EZ-Western Stripping Buffer to fully submerge the membrane and incubate at room temperature for 15 ~ 30 minutes.
   - For an 8 x 10 cm membrane, use 20 ㎖ to 25 ㎖ of buffer.
   - For antibodies with strong binding, shake the membrane or incubate at 37°C.
   - PVDF membrane makes the membrane look transparent when you put the stripping buffer in it.

3. After incubation, wash the membrane with TBS-T or PBS-T.
   - ➢ Use 50 ~ 100 ㎖ of buffer and shake for about 5 minutes (repeat 2~3 times).
   - ➢ For PVDF membranes, continue washing until the membrane becomes opaque again.
4. Check if the antibody has been removed.
   1) Confirm HRP-labeled reagent removal:
      ① After stripping, treat the membrane with ECL substrate, and expose it to film or an imaging system.
      ② If no signal is detected after 5 minutes of exposure, the HRP-labeled reagent has been removed.
   2) Confirm primary antibody removal:
      ① After treating the membrane with HRP-labeled secondary antibody, detect using the ECL substrate.
      ② If no signal is detected after 5 minutes of exposure, the primary antibody has been removed from the antigen.
5. If a signal is detected after antibody removal, add more EZ-Western Blot Stripping Buffer and incubate for an additional 5 ~ 15 minutes.
   - ➢ Shaking the membrane can improve efficiency.
6. After stripping, proceed with the standard immuno-blotting steps.
   - ➢ Begin from the blocking step.

▪ Notice

1. In the case of certain antigen/antibody, the binding effect is strong, so you can increase the incubation temperature or increase the time.
2. Membrane can repeat strip and re-probe processes, but it can be less sensitive than the initial reaction because antigens are also partially lost during strip process.
   In this case, the sensitive ECL substrate should be used, or the exposure time should be extended.

▪ Related Product

I. EZ-Western series

: Western blot detection kit (ECL solution) and others

Product	Catalog No.	Size
EZ-Western (Nano~mid picogram)	DG-W100	100 ㎖ (A: 50 ㎖ + B: 50 ㎖)
	DG-W250	250 ㎖ (A: 125 ㎖ + B: 125 ㎖)
	DG-W500	500 ㎖ (A: 250 ㎖ + B: 250 ㎖)
EZ-Western Lumi Pico (Low picogram)	DG-WP100	100 ㎖ (A: 50 ㎖ + B: 50 ㎖)
	DG-WP250	250 ㎖ (A: 125 ㎖ + B: 125 ㎖)
	DG-WP500	500 ㎖ (A: 250 ㎖ + B: 250 ㎖)
EZ-Western Lumi Pico Alpha (Fast Detection)	DG-WPAL120	120 ㎖ (A: 60 ㎖ + B: 60 ㎖)
	DG-WPAL250	250 ㎖ (A: 125 ㎖ + B: 125 ㎖)
EZ-Western Lumi La (Mid femtogram, Long duration)	DG-WD100	100 ㎖ (A: 50 ㎖ + B: 50 ㎖)
	DG-WD200	200 ㎖ (A: 100 ㎖ + B: 100 ㎖)
EZ-Western Lumi Femto (Low femtogram)	DG-WF100	100 ㎖ (A: 50 ㎖ + B: 50 ㎖)
	DG-WF200	200 ㎖ (A: 100 ㎖ + B: 100 ㎖)
EZ-Gel Staining Solution	DG-GS1000	1000 ㎖ (Without De-stain)
EZ-BCA Protein Quantification Kit	DG-BCA500	Reagent A: 500 ㎖ Reagent B: 25 ㎖ Standard Sol.: 1 ㎖ X 10
EZ-Bradford Assay Dye Reagent	DG-BRA500	Bradford Reagent: 500 ㎖ Standard Sol.: 1 ㎖ X 10

II. Protein marker

Catalog No.	Product	Qty.
DG-PMC245	3-Color Regular Range Protein Marker, 10-245kDa	250㎕ x 2
DG-PMP245	3-Color Broad Range Protein Marker PLUS, 5-245kDa	250㎕ x 2