Cat # SL100324 Store at 4~23 ^{0}C

BlotFresh™ Western Blot Stripping Reagent

☐ **Small** **125 ml**

☐ **Large** **500 ml**

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This product is for laboratory research ONLY and not for diagnostic use

Description:

BlotFresh™ Western Blot Stripping Reagent is formulated to be effective for removal of antibodies from Western blots that have been developed with chemiluminescence or radioactive iodine or other isotopes. The membrane can be nitrocellulose or PVDF/nylon. The stripped membrane is OK for re-probing as that of regular western blot.

Reuse of blots offers many advantages:

• Effective use of samples that are available in limited amounts
• Comparison of images obtained with different antibodies in the same blot
• Confirmation of results with the same or different antibodies
• It is simply more economical and less time consuming to reuse blots

Advantages of BlotFresh™ Western Blot Stripping Reagent:

• Strip blot in 5~15 minutes
• Strip antibodies at room temperature (no heating is required)
• No pungent smelling without addition of 2-mercaptoethanol and its analogs.
• Strip solution comes in 1x solution and it is ready to use. Just soak blots in the solution for 5~15 minutes with strong agitation for complete dissociation of antigen-antibody complex
• Blots are reusable within 60 minutes after a short incubation in blocking buffer

Instructions

After initial probing, be sure to keep membrane wet in TBST buffer in fridge. NEVER LET THE BLOT DRY!

1. Pour 15~30 ml stripping reagent to a clean container and put the blot in the container. Make sure that the blot is fully submerged with the stripping buffer.
2. Incubate the blot in stripping reagent at room temperature for 5~15 minutes with strong agitation. Though incubation with the high affinity antibodies need to be optimized, 15 minutes stripping at room temperature is usually sufficient for most of antibodies.

Note: Optimization of both incubation time and temperature is essential for best results. In general, higher affinity antibodies will require at least 15 minutes of stripping and may require an incubation temperature of 37 0C .

3. Wash for 2x5 minutes in TBS-T at room temperature using large volumes (e.g. 100 ml) of wash buffer.

Note: To test the stripping effect, pour ECL reagent on blot followed by 5 minutes exposure to a film.

4. Block blot with TBS-T buffer with 5% defat milk powder for 45 minutes at room temperature.
5. Immunodetection as normal.

References:

1. Kaufmann, S.H., et al. (1987). Anal. Biochem. 161, 89-95.
2. Kaufmann, S.H. and Kellner, U. (1998). Erasure of Western blots after autoradiographic or chemiluminescent detection. In Immunochemical Protocols. Ed. Pound, J.D. Humana Press, Totowa, NJ, 223-235
3. Schrager, J.A., et al. (2002). J. Biol. Chem. 277, 6137-6142
4. Sorci, G., et al. (2003). Mol. Cell. Biol. 23, 4870-4881

Storage: Upon arrival store this product at 4 ^{0}C ~room temperature. For long term, store the product at 4 ^{0}C. Incubation of blots with cold (4 ^{0}C) BlotFresh™ Western Blot Stripping Reagent at room temperature does NOT affect the stripping effect. Product shipped at ambient temperature.

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