NomelRT™ Western Blot Stripping Buffer

RUO

Research Use Only

REF

21112



INTRODUCTION

Western blotting is widely used to detect and compare proteins in complex mixtures, and che miluminescence has largely replaced chromogenic substrates as the most convenient and sensitive method of detection. Nitrocellulose and PVDF membranes probed by Western blotting procedures and detected by chemiluminescent or other non- precipitating substrates can be stripped and re-probed using NomelRT* Western Blot Stripping Buffer. One advantage of chemiluminescence is the ability to strip and reprobe the protein mixture on the membrane. Traditional stripping methods use conditions that are effective for only low-affinity antibody-a ntigen interactions or are so harsh that they tend to adversely alter the antigen for subsequen t immunoprobing. No stench smelling and without addition of 2-mercaptoethanol or its analogs. Incubate the blot in NomelRT* Western Blot stripping reagent at roomtemperature.

KIT CONTENTS

Label	Contain
NomelRT™ Western Blot Stripping Buffer	500 ml

STORAGE AND STABILITY

APPLICATIONS

Western Blot

ADDITIONAL REQUIRED EQUIPMENT

- Shakers
- Shaking incubator(optional)

PROTOCOL

After initial probing, be sure to keep membrane wet in TBST buffer. DO NOT DRY!

- 1. Warm the bottle of NomelRT™ Western Blot Stripping Buffer to room temperature.
- Pour 15~30 ml stripping reagent to a clean container and put the blot in the container. Mak e sure that the blot is fully submerged with the stripping buffer.
- 3. Incubate the blot in stripping reagent at room temperature for 10 ~ 30 minutes with agitati on. 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 30 minutes of
- stripping and may require an incubation temperature of 37 °C .

 4. Wash for 5 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 exposur e to a film.

ORDERING INFORMATION

Product Name	Amount	Cat. No.
GangNam-STAIN™ Prestained Protein Ladder	250 ul	24052
WEST-lott™ PVDF Transfer membrane (0.45 um, 0.22 um)	300 x 3000 mm	ITM-P3031 / ITM-P3032
Ponceau S solution	1L	31192
WEST-ZOL plus Western Blot Detection System	200 ml	16024

References

- Kaufmann, S.H., et al. (1987). The erasable Western blot. Anal Biochem 161: 89-95.
- Kaufmann, S.H. and Kellner, U. (1998). Erasure of Western blots after autoradiographic or chemiluminescent detection. In Immun ochemical Protocols. Ed. Pound, J.D. Humana Press, Totowa, NJ., p. 223-35.

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