

EZBlock™ Blocking Buffer and Signal Enhancer

03/19

(Store at 4°C)

Cat. No.**M4104-100 100 ml EZBlock™ Blocking Buffer and Signal Enhancer****M4104-500 500 ml EZBlock™ Blocking Buffer and Signal Enhancer**

I. Introduction:

BioVision's EZBlock™ Blocking Buffer and Signal Enhancer is a blocking solution for Western blot analysis. EZBlock™ Blocking Buffer and Signal Enhancer not only provide blocking and primary and secondary antibody hybridization in one step but also enhance the signal developed with HRP (horseradish peroxidase) or AP (alkaline phosphatase) substrates. It, therefore, serves as both blocker and enhancer in Western analysis. With the three-in-one step procedure, EZBlock™ Blocking Buffer and Signal Enhancer is an efficient and cost effective solution for the time consuming and laborious Western procedure.

II. Applications:

- Western blot

III. Content:

EZBlock™ Blocking Buffer and Signal Enhancer

IV. Storage & Handling:

Store Buffer at 4°C. EZBlock™ Blocking Buffer and Signal Enhancer is stable for 1 year.

V. Procedure using EZBlock™ Blocking Buffer and Signal Enhancer in Western blot application:

1. After transferring the Western blot, immerse the PVDF or NC membrane in PBST buffer for 5 minutes.
2. Dilute the primary and secondary antibody with appropriate amounts of EZBlock™ Blocking Buffer and Signal Enhancer.
(Note: For 1: 10,000 dilution of both primary and secondary antibodies, add 2 µl of the primary and secondary antibody to 10 ml of the EZBlock™ Blocking Buffer and Signal Enhancer in two separate tubes. Thoroughly mix the antibody with EZBlock™ Blocking Buffer and Signal Enhancer by inverting the tubes).
3. Pour the primary antibody- EZBlock™ Blocking Buffer and Signal Enhancer into the prepared container first, followed by the addition of the secondary antibody- EZBlock™ Blocking Buffer and Signal Enhancer into the same container.
4. Incubate the membrane immediately in the antibody- EZBlock™ Blocking Buffer and Signal Enhancer at room temperature for 1 -2 hours with gentle agitation.
(Note: The membrane should be immersed in the mixture within 10 minutes after mixing the primary and secondary antibodies for the optimal performance)
5. Wash the membrane with PBST/TBST three times with gentle shaking.
6. Drain excessive wash buffer and proceed immediately to develop the membrane with ECL or colorimetric system.

Notes:

- After mixing the primary and secondary antibodies, the membrane needs to be immediately immersed in the mixture within 10 minutes for obtaining the optimal performance.
- The dilution for the secondary antibody should be at least 1:10,000 or more. Higher level of background noise will be observed as a result with a high concentration of secondary antibody.
- Do not incubate membrane in EZBlock™ Blocking Buffer and Signal Enhancer for over 4 hours to avoid high background. Overnight incubation is especially not recommended.
- The primary/secondary antibodies mixed in EZBlock™ Blocking Buffer and Signal Enhancer maybe reused within 3 days.
- Enhancing effect may trail off along with the increasing storage time or repetitiveness. Keep the mixed solution refrigerated. For critical experiment or strong signal, fresh preparation of antibody- EZBlock™ Blocking Buffer and Signal Enhancer is required.
- When the antibody concentration is too high or if the prolonged incubation takes place, it will cause high background. When excessive background occurs, please try the followings:
 - (a) Reduce/optimize primary and/or secondary antibody concentrations.
 - (b) Use dot-blot test to optimize antibody concentrations.
 - (c) Reduce/optimize incubation time.

VI. RELATED PRODUCTS

EZBlock™ (TBS) Blocking Buffer (2117)	EZBlock™ T20 (PBS) Blocking Buffer (2143)
EZBlock™ T20 (TBS) Blocking Buffer (2140)	Western Blot Substrate Kit (K820)
EZLys™ Bacterial Protein Extraction Reagent (8001)	EZLys™ Yeast Protein Extraction Reagent (8003)
EZLys™ Tissue Protein Extraction Reagent (8002)	EZLys™ lysozyme, human (8005)
Protein Quantitation kit (K810)	BCA Protein Quantitation Kit (K812, K813, K814)
Protein Carbonyl Content Assay Kit (K830)	Protease & Phosphatase inhibitor cocktails (K283, K284)
Protease inhibitor cocktails (K271, K272, K277, K278, K279)	
Ready-to-use IHC/ICC kit (Biotin free), One-Step HRP Polymer anti-Mouse, Rat and Rabbit IgG (H+L) with DAB (K405)	

FOR RESEARCH USE ONLY! Not to be used on humans