West-Ez Blocking Buffer, 1% Casein

W3720

Storage

Store at 2-8°C.

Contents

- Product Manual
- West-Ez Blocking Buffer, 1% Casein
  contains 1% (w/v) biotin free casein in 25mM Tris,
  150mM NaCl, pH 7.4 with Kathon Antimicrobial Agent

Introduction

West-Ez Blocking Buffer, 1% Casein is a preformulated biotin free casein solution for blocking excess binding sites in ELISA, Western blotting, immunohistochemistry and other immunochemical applications.

Tips

1. Use West-Ez blocking Buffer, 1% Casein as supplied for initial testing. However, other concentrations may be beneficial for specific systems.
2. West-Ez Blocking Buffer, 1% Casein diluted to 0.05% protein and containing 0.05% Tween-20 detergent may be used as a diluent for antibodies to improve signal-to-noise ratios.
3. Because no blocking reagent is optimal for all systems, empirical testing is essential to determine the appropiate blocking buffer for each system. Determining the proper blocking buffer can increase sensitivity and prevent nonspecific signal caused by cross-reactivity between the antibody and the blocking reagent.

Procedure

1. Add the blocking solution to the ELISA well, blotting membrane or immuno-histochemical slide.
2. Incubate 30 minutes to 2 hours at room temperature or 37°C.
3. Continue with standard protocol for the specific application.

Related Products

Product Name	Cat No
Albumin, (IgG, Fattu Acid and Protease Free)	A0100
AffiSelect Cox IV Loading Control Antbody	A0017
AffiSelect GAPDH Loading Control Antibody, 100ug	A0039
AffiSelect Beta-Actin Loading Control Antibody, 100ug	A0042
AffiSelect a-Tubulin Loading Control Antibody, 100ug	A0050
NP-40 Lysis Buffer (2X)	N1200
Affiselect Total Protein Extraction Solution	A0710
RIPA Cell Lysis Buffer (1X) with EDTA	R4100
Xert Protease Inhibitor Cocktail Solution (100X)	P3100
Xpert Prestained Protein Marker (6.5-240 kDa)	P8502
Xpert 2 Prestained Protein Marker (10-240 kDa)	P8503

Troubleshooting Guide

Problem	Possible Cause	Solution
High Background	Incubation tray is contaminated with HRP	Use a clean incubation tray after every step of the procedure
	Use too much primary antibody	Reduce primary antibody concentration to 0.2-1ug/ml
	Insufficient washing	Use a minimum of 20 ml of 1X Wash Buffer for each wash
		Use a clean incubation tray after every step of the procedure
		Add an additional wash cycle for a total of four 5 minute washes
	Overexposed film	Decrease exposure time
	Omitted the brief pre-wash	Wash membrane in 1X Wash Buffer briefly before beginning the protocol
Weak signal	Used insufficient quantities of antigen or primary antibody	Strip and re-probe blot using a higher concentration of antibodies
		Load higher concentrations of sample onto the gel
	Used too much primary antibody	Reduce primary antibody concentration to 0.2-1 µg/ml
	Used too much sample	Reduce amount of sample on the gel
	Inefficient protein transfer	Optimize transfer conditions
Spots within the protein bands	Inefficient protein transfer	Optimize transfer procedure
	Unevenly hydrated membrane	Hydrate membrane according to manufacturer's instructions
	Bubble between X-ray film and membrane	Remove all bubbles before exposing blot to film
Speckling	Over-heating during electrophoresis or transfer	Control temperature during electrophoresis and transfer

GenDEPOT PO Box 454 Barker, Tx 77413 tech@gendepot.com