



610PR-01

G-Biosciences ♦ 1-800-628-7730 ♦ 1-314-991-6034 ♦ technical@GBiosciences.com

A Geno Technology, Inc. (USA) brand name

Blocking Agents Optimization Trial Packs

(Cat. # 786-946, 786-946P)



think proteins! think G-Biosciences www.GBiosciences.com

INTRODUCTION 3

ITEM(S) SUPPLIED 3

STORAGE CONDITIONS 3

ADDITIONAL ITEMS REQUIRED 3

PREPARATION OF 1X NAP-BLOCKER BEFORE USE 3

IMPORTANT 4

PROCEDURE FOR BLOCKING WESTERN BLOTTING MEMBRANES..... 4

PROCEDURE FOR BLOCKING ELISA PLATE..... 5

PROCEDURE FOR BLOCKING TISSUE FOR IMMUNOHISTOCHEMISTRY 5

RELATED PRODUCTS..... 6

INTRODUCTION

The Blocking Agents Optimization Trial Packs are a set of G-Biosciences four proprietary blocking agents for immunoassays with one being a non-animal protein blocker and another protein free that minimizes the risk of non-specific binding of antibodies during the immunoassay. Each blocking buffer provided in the Trial Pack is suitable for 6 mini blots (7.5cm x 8.5cm size) except NAP-Blocker, which is good for 12 mini blots and are suitable for Western Blot and ELISA experiments.

The Blocking Agent Trial packs are supplied in either TBS or PBS.

ITEM(S) SUPPLIED

Description	Cat. # 786-946 <i>Supplied in TBS</i>	Cat. # 786-946P <i>Supplied in PBS</i>
NAP-Blocker™ [2X]	125ml	125ml
<i>Superior</i> ™ Blocking Buffer	125ml	125ml
<i>Protein-Free</i> Blocking Buffer	125ml	125ml
<i>FirstChoice</i> ™ Blocking Buffer	125ml	125ml

STORAGE CONDITIONS

It is shipped at ambient temp. Upon arrival, store at 4°C. If stored and aseptic techniques are used for handling NAP-Blocker™, it is stable for up to 1 year.

ADDITIONAL ITEMS REQUIRED

- Primary and secondary (labeled) antibodies
- Reagents for immunodetection

NOTE: Gently shake the bottle of NAP-Blocker™ before its use to mix it and follow aseptic techniques for handling the NAP-Blocker and other blocking agents in the pack.

PREPARATION OF 1X NAP-BLOCKER BEFORE USE

Prepare 1X NAP-Blocker™ by mixing 1 part NAP-Blocker™ with 1 part of 1X TBS or PBS (or G-Biosciences' femto-TBST or femto-PBST) for blocking PVDF membranes. For nitrocellulose membranes and ELISA experiments, mix 1 part NAP-Blocker™ with 2 parts 1X TBS or PBS (or G-Biosciences' femto-TBST or femto-PBST).

NOTE: Use TBS for NAP-Blocker™ from 786-946 and PBS for 786-946P

IMPORTANT

- For optimal blocking, do NOT dilute other blocking buffers in the pack except the NAP-Blocker.
- The efficacy of blocking agents varies from application to application, so we recommend empirical testing of blocking buffer and optimization of procedure to increase sensitivity and prevent nonspecific signal and cross-reaction between blocking agent and antibody.
- Use of detergent in blocking buffers is not required for all applications; however, addition of 0.05% Tween[®]-20 often improves blocking efficiency. Use only high quality ultrapure grade Tween[®]-20, we recommend our Proteomic Grade Tween[®]-20 solution (Cat. # DG011, DG012, DG511), which is purified to remove peroxide and carbonyls contaminants that may interfere in some applications.
- 10-fold diluted Blocking Buffer containing 0.05% Tween[®]-20 may be used to dilute antibodies to enhance the sensitivity of the signal.

PROCEDURE FOR BLOCKING WESTERN BLOTTING MEMBRANES

1. Following protein transfer, carefully move the membrane to a suitable size tray.
2. Add enough Blocking Buffer to completely cover the membrane.
3. Incubate for 30-120 minutes at room temperature with agitation.
4. Discard blocking buffer and continue with downstream Western blotting steps.

NOTE: For washing steps, use of femtoTBST™ (Cat. # 786-161) or femtoPBST™ (Cat. # 786-162) will minimize the washing out of immune-complexes and aid in the generation of cleaner backgrounds resulting in a higher signal to noise ratio, a common problem associated with classical TBST or PBST buffer used for washing.

PROCEDURE FOR BLOCKING ELISA PLATE

1. Apply sample to the ELISA plates and incubate for 1-2 hours at room temperature.
2. Apply 300µl of one of the Blocking Buffers to each well. Immediately empty the well by aspiration or inversion. Repeat this step twice more. The incubation is not required, however, the plates may be incubated without any detrimental effects.
3. Continue the downstream ELISA steps.
NOTE: For washing steps, use of femtoTBST™ (Cat. # 786-161) or femtoPBST™ (Cat. # 786-162) will minimize the washing out of immune-complexes and aid in the generation of cleaner backgrounds resulting in a higher signal to noise ratio, a common problem associated with classical TBST or PBST buffer used for washing.
4. For storage of coated plates, invert plates and allow plates to dry completely before sealing in a plastic bag with desiccant.

PROCEDURE FOR BLOCKING TISSUE FOR IMMUNOHISTOCHEMISTRY

1. Incubate tissue in the blocking buffer for 30 minutes at room temperature.
2. Remove the blocking buffer from the tissue.
3. Without rinsing the tissue, continue with immunohistochemistry downstream procedures for detection

RELATED PRODUCTS

Download our Western Blotting and Assay Development Handbooks



<http://info.gbiosciences.com/complete-western-blot-handbook--selection-guide>

<http://info.gbiosciences.com/complete-assay-development-handbook>

For other related products, visit our website at www.GBiosciences.com or contact us.

Last saved: 11/30/2015 IA

This page is left intentionally blank



www.GBiosciences.com