Technical Data Sheet

Section Block

#62710, 62711, 62712

Introduction

All antibody preparations have some potential to produce a non-specific reaction in the assay; it originates from:

- Non-specific antibodies that are present in some proportion in any polyclonal antibody preparation, including ones that are affinity-purified
- Low specificity antibodies among specific ones in polyclonal
- Fragments of fallen apart IgGs in stored preparations, including monoclonal
- Separate heavy and light chains of specific antibodies, which are produced by most hybridomas

All these are capable of non-specifically binding to molecules on tissue sections, blots, fixed cells and other objects for immune detection. In the case of retrieved formalin sections, the risk of non-specific reaction is increased, since the proteins comprising the tissue sections are denatured during HIER. This makes many domains accessible that are charged. They are also capable of binding the test immunoglobulins in a non-specific manner.

The standard means of blocking non-specific binding of specific antibody preparation is as follows: Add irrelevant protein, other serum, casein, etc. However, in most cases, many who have tried this found that increasing concentration of such blocking agent leads to a great reduction of specific reaction as well. This is a result of the lack of blocking molecules binding to access sites on section and thus sterically blocking the access to specific antibodies to epitopes of interest.

All of our buffers at Electron Microscopy Sciences are developed for immune assays and contain, instead, short (0.6-2 kD) peptides that are capable of block effectively to non-specific reactions. These do not affect the specific binding of antibody.

Advantages

Section Block represents a new class of blocking solutions based on chemically modified and fragmented ultra-pure casein. Some of the advantages are:

- Reduces unwanted binding of primary antibody and conjugates to charged surface of the slide and tissue section
- Greatly reduces non-specific binding while preserving the specific reaction by saturating potential non-specific sites for protein-protein interactions
- In contrast to BSA-based, IgG-, casein- or serum-based blocking solutions, there is no interaction of specific antibody and blocking protein itself
- It is in a class of its own and is not comparable to any other commercially available or homemade blocking solutions

Applications

Electron Microscopy Sciences recommends this product for research and diagnostic pathology, especially for HIER retrieved sections and polyclonal antibodies.

Use in IHC

1. Apply to cryostat, or depaffinaized formalin-fixed, or processed in Retriever (or other epitope recovery device) tissue section quantity sufficient to cover the tissue (approximately 200 ml). Alternatively, incubate by submerging the sections into the buffer. Use Retriever Slide Chamber (Catalog #62705-01) to block many slides at once with minimal use of Block buffer.

2. Incubate 30 minutes at room temperature.
3. Rinse once with PBS or other suitable buffer, and proceed with immunostaining.

Note: For best results use Altibody Diluent (Catalog #62713) to prepare the solutions of primary and secondary antibodies.

Use in Other Applications

Section Block can also be used to prevent non-specific binding of reagents and to improve sensitivity in the following:

- Immuno-PRC
- Western Blotting
- Protein Arrays
- Immunofluorescent staining of tissue sections and fixed cells
- Flow cytometry on fixed and permeabilized cells

Block in similar manner for 30 minutes at room temperature.

Stability and Storage

- The preparation is stable for 2 years when stored unopened at +4°C
- Every lot is issued with an expiration date
- Once opened, use within 6 months and store at +4°C in the refrigerator