Technical Data Sheet

Antibody Diluent

#62713, 62714, 62715

Instructions

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 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.

Properties

Antibody Diluent used in the dissolving of primary and secondary enzymes and antibodies to required working concentrations.

Presentation

Antibody Diluent is supplied as a ready to use solution in the following volumes: 50, 125 or 500 ml. The solution has a clear colorless appearance.

Application

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

Use in IHC

Use to dilute primary polyclonal or monoclonal antibody to reduce non-specific binding of the primary antibody. The diluent can also be used to dilute secondary, enzyme labelled or fluorochrome-labelled secondary antibody.

In the case of overdiluting antibodies to reduce the background staining, consider the use of a higher concentration (2-4 times) when using Antibody Diluent, in order to improve the sensitivity or antigen detection.

Use in Other Applications

Antibody Diluent 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

Use as antibody diluent. For Immuno-PCR and protein arrays, the diluent can also be used as a washing buffer to rinse the reagents between immunostaining stages.

Stability and Storage

Stable for 2 years when stored unopened at +4°C. Every lot is issued with a certificate indicating the expiration date.

After opening, however, store at +4°C in the refrigerator and be sure to use within 6 months.