Instructions for use

Product name Monosan Antibody Diluent

Intended Use Antibody Diluent is developed for dilution of primary antibodies. Antibodies diluted with Antibody Diluent are primarily used in immunohistochemistry with formalin-fixed paraffin-embedded tissue sections, but also with frozen, HOPE-fixed, and cytological samples as well as in immunoblot procedures.

Applications IHC-P, IHC-Fr, IF

Summary and explanation Antibody diluents used in immunohistochemistry should protect the antibody from microbial contamination and stabilize the antibody chemically. Antibody Diluent reduces non-specific binding of antibodies to tissue sections and is therefore extremely useful in receiving background-free staining results.

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Principle of method

Immunohistochemical staining procedures often start with incubation of a blocking solution to reduce unspecific binding of primary antibody to tissue sections. This step can be omitted if the antibody used is diluted in Antibody Diluent. Antibody Diluent minimises unspecific binding of the primary antibody to the tissue section, reduces surface tension of the antibody solution and improves spreading the reagent on the slide, increases microbial and chemical stability of the antibody, reduces adhesion of antibody to the surface of the vial, and minimises the danger of antibody degradation by proteolytic enzymes.

Reagents provided

100 ml Antibody Diluent (ready-to-use)

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Storage and handling

The solution should be stored at 2-8°C without further dilution. Do not freeze it. Under these conditions the solution is stable up to the expiry date indicated on the label. Do not use product after the expiry date. If stored at room temperature the solution is stable for at least 10 month from the date of delivery. A positive and a negative control have to be carried out in parallel to the test material. If you observe unusual staining or other deviations from the expected results which could possibly be caused by this reagent, please contact our technical support.

Reagent preparation

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Procedure

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Expected results

During the reaction of the substrate with horse radish peroxidase or alkaline phosphatase in the presence of a chromogen, a coloured precipitate is formed at the location of the bound primary antibody. This reaction only takes place if the target antigen is existent in the tissue. The chromogen used determines the colour of the precipitate. The analysis is carried out using a light microscope.

Trouble shooting

If you observe unusual staining or other deviations from the expected results please read these instructions carefully, contact our technical support. Also refer to the instructions of the detection systems containing Peroxide Block for guidance on general troubleshooting

Quality control

We recommend carrying out a positive and a negative control with every staining run. The positive control permits the validation of appropriate processing of the sample. If the negative control has a positive result, this points to unspecific staining. Please refer to the instructions of the detection system for guidance on general quality control procedures.

Performance

Studies have been conducted to evaluate the performance of the kit reagents. The product has been found to be suitable for the intended use

Limitations of procedure

Immunohistochemistry is a complex technique involving both histological and immunological detection methods. It requires a highly trained histotechnologist. Tissue processing and handling prior to immunostaining, for example variations in fixation and embedding or the inherent nature of the tissue can cause inconsistent results (Nadji and Morales, 1983). Inadequate counterstaining and mounting can influence the interpretation of the results. Sanbio guarantees that the product will meet all requirements described from its shipping date until its expiry date, as long as the product is correctly stored and utilized. No additional guarantees can be given. Under no circumstances shall Sanbio be liable for any damages arising out of the use of the reagent provided.

Precautions

Use through qualified personnel only. Wear protective clothing to avoid contact of reagents and specimens with eye, skin and mucous membranes. If reagents or specimens come in contact with sensitive area, wash with large amounts of water. Microbial contamination of the reagent must be avoided, since otherwise non-specific staining may occur. ProClin300 and sodium azide (NaN3) are used for stabilisation. Reaction of sodium azide with lead or copper in drainage pipes can result in the formation of highly explosive metallic azides. Discard the antibody solution in a large volume of running water to avoid formation of deposits. A material safety data sheet (MSDS) for the pure substances is available upon request.

References

1. Elias JM Immunohistopathology – A practical Approach to Diagnosis ASCP Pr
2 Nadji M and Morales AR Ann N.Y. Acad Sci 420:134-139, 1983
3. -

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