Product datasheet **MON-APP165**

Monosan Wash Buffer (20x), 500 ml (for 10 liter)

Reagents

Instructions for use

Product name Monosan Wash Buffer (20x), 500 ml (for 10 liter)

Intended Use The wash Buffer is designed as washing solution for immunohistochemical and immunocytological staining procedures on slides. Wash Buffer is primarily used with formalin-fixed paraffin-embedded tissue sections, but also with frozen, HOPE-fixed, and cytological samples as well as in immunoblot procedures. The Wash Buffer is suitable for manually operated and automated immunohistochemical staining.

Applications IHC-P, IHC-Fr, IF

Summary and explanation Immunohistochemical staining procedures consist of sequential incubation steps with blocking solutions, antibodies and secondary reagents, enzymes and chromogenic substrates carried out on tissue sections. Washing away the applied reagents after each incubation step is critical to receive optimally stained samples. The Wash Buffer is especially designed for effective washing and therefore ensures brilliant staining results.

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Principle of method

The Wash Buffer is a 20fold concentrated phosphate buffer with additives of sodium chloride, detergent, and stabilising substances. For preparation of the working strength solution the buffer concentrate is diluted 1:20 with deionised or distilled water. The resulting solution has a pH of 7.2 (7.0 to 7.4).
The Wash Buffer
• is wetting the tissue sections with detergent and thus reduces surface tension and improves spreading the reagents on the tissue section,
• reduces unspecific binding of reagents on the tissue sample,
• and because of the exact tuned salt concentration effects an excellent preservation of cell morphology.

Reagents provided

500 ml Wash Buffer (20fold concentrated, adequate for 10 litres ready-to-use wash buffer)

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Storage and handling

The solution should be stored at room temperature. It is stable up to the expiry date indicated on the label if undiluted. Do not use product after the expiry date. The diluted working strength solution is stable for about 1 week depending on the ambient temperature. 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

Preparation of the Wash Buffer working strength solution:
• Dilute Wash Buffer concentrate 1:20 with deionised or distilled water and mix thoroughly.
• The pH-value should be at 7.2 (7.0 to 7.4). If necessary adjust pH-value with diluted NaOH or HCl solution.

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, or contact our technical support. Also refer to the instructions of the detection systems for guidance on general troubleshooting.

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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. The Wash Buffer is a 20fold concentrated solution with a mildly acidic pH-value. The correct pHvalue of about 7.0 (+/- 0.2) is achieved after diluting the solution 1:20. Sometimes deionised water has pH-values considerably different from the neutral point (pH 7.0) depending on the preparation method. Experiments have shown that the Wash Buffer can successfully be diluted with deionised or distilled with water in the pH-range of 5.5 up to 9.5. If a detection system with alkaline phosphatase is used please note: larger amounts of wash buffer remaining on the slides can lead to decreasing enzyme activity. 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

Precautions

Use by qualified personnel only. Wear protective clothing to avoid contact of reagent or specimen with eye, skin or mucous membrane. In case of the reagent or specimen coming into contact with a sensitive area, wash the area with large amounts of water. Microbial contamination of the reagent must be avoided, since otherwise non-specific staining may occur. ProClin 300 is used for stabilisation. A material safety data sheet (MSDS) 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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