



FISH Pretreatment Buffer

REF / Cat. No.: FA-002-500 500 ml

Instructions for use

Intended use

FISH Pretreatment Buffer is a ready-to-use solution developed for heat induced epitope retrieval (HIER) in formalin-fixed paraffin-embedded tissue sections on slides. This procedure is primarily used in *in situ* hybridization (ISH) assays and in immunohistochemistry.

FISH Pretreatment Buffer is intended for *in vitro* diagnostic use.

Summary and explanations

In situ hybridization is usually performed with tissue sections prepared out of formalin-fixed paraffin-embedded (FFPE) tissue blocks. Cellular structures are very effectively stabilised by formalin fixation which results in optimal morphological preservation of the sample, but on the other hand formalin fixation leads to strong cross-links between proteins impairing the accessibility of the target sequences for DNA probes. Therefore cross-links must be broken up before a probe is applied to the tissue. This is in most cases achieved by a combination of heat induced epitope retrieval (HIER) and proteolysis.

Principle of the method

FISH Pretreatment Buffer is a ready-to-use solution with additives of stabilising substances.

Reagent provided

REF / Cat. No. FA-002-500
500 ml FISH Pretreatment Buffer

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.

Internal (on-slide) and external controls should be carried out in parallel to verify the assay procedure. If you observe deviations from the expected results which could possibly be caused by the reagent, please contact Zytomed Systems' technical support or your local distributor.

Precautions

Use by qualified personnel only. Wear protective clothing to avoid contact of reagent and specimen with eye, skin or mucous membranes. In case of reagent or specimen coming into contact with a sensitive area, wash area with large amounts of water. A material safety data sheet (MSDS) is available upon request.

Reagent preparation

The solution is ready-to-use.

ISH Procedure

FISH Pretreatment Buffer is suitable for heat induced epitope retrieval (HIER) carried out after deparaffinization and rehydration of the tissue sections. Tissue sections used in heat induced epitope retrieval should always be placed on adhesive slides.

1. Pour a sufficient volume of FISH Pretreatment Buffer into a Coplin jar, transfer Coplin jar to a water bath. Please make sure that there is enough volume to cover the tissue sections on the slides completely.
2. Pre-heat water bath to 98° C.
3. When temperature within the Coplin jar is constantly at 98°C, transfer slides into the preheated solution.
4. Incubate for 15 min.
5. After 15 min of incubation, remove slides from the Coplin jar and wash 2 x 2 min in H₂O_{dest} at room temperature.
6. Proceed with *in situ* hybridization assay as usual.

Quality control

We recommend carrying out internal (on-slide) and external controls with every hybridization run to monitor the performance of the reagents used, and for correct interpretation of results.

Troubleshooting

If you observe unusual results please read these instructions carefully, contact Zytomed Systems' technical support or your local distributor. Also refer to the instructions for use of the ZytoPure® FISH Accessory Kit for guidance on general troubleshooting.

Limitations of the procedure

- Interpretation of results should only be done by qualified personnel familiar with the method and probe signal patterns under the supervision of a pathologist. Modifications of the protocol may lead to divergent results. They are the responsibility of the user and are to be validated for correct outcome.
- Positive and negative controls should be performed.
- Unsuitable pre-analytic parameters, like prolonged ischemia, over- or underfixation, insufficient dehydration or embedding of tissue specimens can negatively affect the results.
- Tissue specific problems, like strong autofluorescence, can negatively affect the results.

Performance characteristics

Zytomed Systems has conducted extensive studies to evaluate the performance of the reagent. The product has been found to be suitable for the intended use.

Bibliography









- Lehr HA *et al.* Am J Clin Pathol 115:814-822, 2001
- Vincent-Salomon A *et al.* Histopathology 42:337-347, 2003

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Explanation of the symbols on the product label:

REF	Bestellnummer Catalog Number Reference du catalogue		Verwendbar bis Use By Utiliser jusque		Gebrauchsanweisung beachten Consult Instructions for use Consulter les instructions d'utilisation
LOT	Chargenbezeichnung Batch Code Code du lot		Lagerungstemperatur Temperature Limitation Limites de température	RUO	Nur für Forschungszwecke For Research Use Only Pour la recherche uniquement
IVD	In vitro Diagnostikum In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro		Achtung/Gefahr Warning/Danger Attention/Danger	 Hersteller / Manufacturer / Fabricant Zytomed Systems GmbH Anhalterstraße 16 14163 Berlin, Germany Tel: (+49) 30-804 984 990 www.zytomed-systems.de	
	Achtung/Gefahr Warning/Danger Attention/Danger		Achtung Warning Attention		 Gefahr Danger Danger