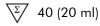


Nuclear Red Solution

REF CS-0003-20



For use in chromogenic in situ hybridization (CISH) procedures

> For research use only. Not for use in diagnostic procedures.

Intended use

The Nuclear Red Solution (CS3) is intended to be used for counterstaining steps in chromogenic in situ hybridization (CISH) applications on formalinfixed, paraffin-embedded specimens. The Nuclear Red Solution is intended to be used in combination with Digoxigenin-labeled Zyto Fast Probes and the Zyto Fast PLUS CISH Implementation Kit AP-NBT/BCIP (T-1061-40).

Clinical relevance

This product is for research use only and not for diagnostic procedures.

Test principle

The chromogenic in situ hybridization (CISH) technique allows the detection and visualization of specific nucleic acid sequences in cell preparations. Hapten-labeled nucleotide fragments, so called CISH probes, and their complementary target sequences in the preparations are co-denatured and subsequently allowed to anneal during hybridization. Afterwards, unspecific and unbound probe fragments are removed by stringency washing steps. Duplex formation of the labeled probe can be visualized using primary (unmarked) antibodies, which are detected by secondary polymerized enzyme-conjugated antibodies. The enzymatic reaction with chromogenic substrates leads to the formation of colored precipitates. After counterstaining the nucleus with a nuclear dye, hybridized probe fragments are visualized by light microscopy.

Reagents provided

The Nuclear Red Solution is available in one size:

CS-0003-20: 20 ml (40 reactions of 0.5 ml each)

Materials required but not provided 5.

- Zyto Fast Probe
- Zyto Fast PLUS CISH Implementation Kit AP-NBT/BCIP (T-1061-40)

The Nuclear Red Solution is intended to be used in CISH procedures using ZytoVision probes and kits. For information on materials required for CISH procedures, please refer to the instructions for use of the respective ZytoVision probe and implementation kit.

Storage and handling 6.

Store at 2-8°C. Return to storage conditions immediately after use. Do not use reagents beyond expiration date indicated on the label. The device is stable until expiration date indicated on the label when handled accordingly.

7. Warnings and precautions

- Read the instructions for use prior to use!
- Do not use the reagents after the expiry date has been reached!
- Do not reuse reagents!
- This product contains substances (in low concentrations and volumes) that are harmful to health and potentially infectious. Avoid any direct contact with the reagents. Take appropriate protective measures (use disposable gloves, protective glasses, and lab garments)!
- If reagents come into contact with skin, rinse skin immediately with copious quantities of water!
- A material safety data sheet is available on our homepage (www.zytovision.com)!
- Avoid any cross-contamination and micro-bacterial contamination of the reagents

Hazard and precautionary statements for CS3:

This mixture is not classified as hazardous in accordance with Regulation (EC) No. 1272/2008.

8. Limitations

- For research use only.
- For professional use only
- Specimen staining, especially signal intensity and background staining, is dependent on the handling and processing of the specimen prior to staining. Improper fixation, freezing, thawing, washing, drying, heating, sectioning, or contamination with other specimens or fluids may produce artefacts or false results. Inconsistent results may result from variations in fixation and embedding methods, as well as from inherent irregularities within
- The performance was validated using the procedures described in the instruction for use of the respective ZytoVision probe and implementation kit. Modifications to these procedures might alter the performance and have to be validated by the user.

Interfering substances

Refer to the instructions for use of the respective Zyto Fast CISH Probe and implementation kit.

10. Preparation of specimens

Refer to the instructions for use of the respective Zyto Fast CISH Probe and implementation kit.

11. Preparatory treatment of the device

The product is ready to use. No reconstitution, mixing, or dilution is required.

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12. Assay procedure

Follow the procedure as described in the instructions for use of the respective Zyto Fast PLUS CISH Implementation Kit. Bring to room temperature ($18-25^{\circ}$ C) before use.

13. Interpretation of results

Refer to the instructions for use of the respective Zyto Fast CISH Probe.

14. Recommended quality control procedures

Refer to the instructions for use of the respective Zyto Fast CISH Probe.

15. Performance characteristics

Refer to the instructions for use of the respective Zyto Fast CISH Probe.

16. Disposal

The disposal of reagents must be carried out in accordance with local regulations.

17. Troubleshooting

Any deviation from the operating instructions can lead to inferior staining results or to no staining at all. Please refer to the instructions for use of the respective ZytoVision probe and kit for further information.

18. Literature

- Isola J, Tanner M (2004) Methods Mol Med 97: 133-44.
- Speel EJ, et al. (1994) J Histochem Cytochem 42: 1299-307.
- Tsukamoto T, et al. (1991) Int J Dev Biol 35: 25-32.
- Wilkinson DG: In Situ Hybridization, A Practical Approach, Oxford University Press (1992) ISBN 0 19 963327 4.

Our experts are available to answer your questions. Please contact <u>helptech@zytovision.com</u>



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