ERBB2 Control Slide Set

REF E-4007-2 2 slides

For the detection of ERBB2 gene amplification by chromogenic *in situ* hybridization (CISH)

For research use only.

Not for use in diagnostic procedures.

1. Intended use

The ERBB2 Control Slide Set is intended to be used as a positive control for the detection of ERBB2 gene amplification by chromogenic *in situ* hybridization (CISH). The ERBB2 Control Slide Set is intended to be used in combination with the Zyto*Dot* CISH ERBB2 Probes and the Zyto*Dot* CISH Implementation Kits (Prod. No. C-3018-40; C-3044-10/-40).

2. Clinical relevance

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

3. Test principle

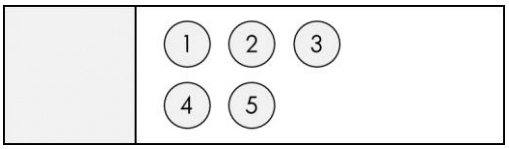
The chromogenic *in situ* hybridization (CISH) technique allows the detection and visualization of specific nucleic acid sequences in cell preparations. Hapten-labeled nucleotide fragments, so called CISH probes, and their complementary target sequences in the preparations are co-denatured and subsequently 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.

4. Reagents provided

The ERBB2 Control Slide consists of four different mammalian cell lines, affected by different levels of ERBB2 gene amplification, and one tissue (myocardial muscle):

- Fixation in 10% neutrally buffered formalin for 24 h at pH 7.0
- Embedding in red-colored paraffin
- Microtome sections of 5 µm thickness
- Mounting on positively charged microscope slides
- Pretreatment for 30 min at 58°C

Description of the slide:
Positioning of the samples on the slide:



1 No ERBB2 amplification, 1-2 gene copies per nucleus
2 Low level ERBB2 amplification, 3-6 gene copies per nucleus
3 High level ERBB2 amplification, large cluster per nucleus
4 No ERBB2 amplification, 1-2 gene copies per nucleus
5 No ERBB2 signals

The ERBB2 Control Slide Set is available in one size:

- E-4007-2: 2 slides

5. Materials required but not provided

- Zyto*Dot* CISH ERBB2 Probe
- Zyto*Dot* CISH Implementation Kits (Prod. No. C-3018-40; C-3044-10/-40).

The ERBB2 Control Slide Set is intended to be used in ISH 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.

6. Storage and handling

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 slide after the expiry date has been reached!
- Do not reuse slides!
- Although the fixation process renders the slides non-infectious, the user is advised to observe the same safety precautions as for handling/disposing potentially infectious agents!
- 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!

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 specimen.
- 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.

9. Interfering substances

Refer to the instructions for use of the respective Zyto*Dot* CISH ERBB2 Probe and implementation kit.

10. Preparation of specimens

Refer to the instructions for use of the respective Zyto*Dot* CISH ERBB2 Probe and implementation kit.

11. Preparatory treatment of the device

1. Remove the label from the ERBB2 Control Slide and label the slide with a pencil.
2. For on slide control mount tissue sample of interest onto the slide.
3. Fix the specimens at 60°C for a minimum of 2 h up to 16 h.

12. Assay procedure

Follow the procedure as described in the instructions for use of the respective Zyto*Dot* CISH Implementation Kit.

13. Interpretation of results

Using the appropriate Zyto*Dot* CISH ERBB2 Probe and implementation kit, hybridization signals on the different sample spots of the ERBB2 Control Slide can be obtained.

Positive staining: In interphases of normal cells or cells without an amplification involving the ERBB2 gene region (sample spots 1; 4), two distinct dot-shaped signals appear (see Fig. 1).

In cells with an amplification of the ERBB2 gene region (sample spots 2; 3), an increased number of the signal or signal clusters will be observed (see Fig. 1).

Negative staining: In cells without the ERBB2 gene region, no signal will appear (sample spot 5).

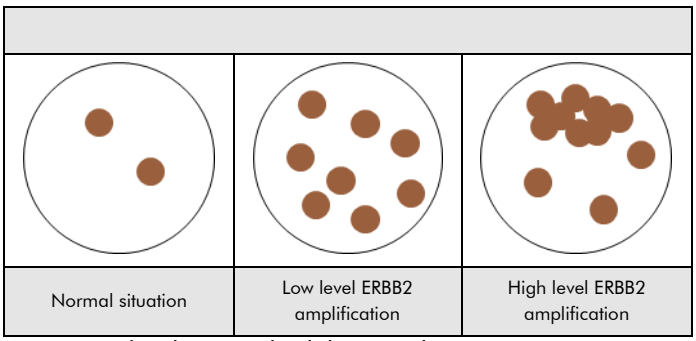


Fig. 1: Expected results in normal and aberrant nuclei

14. Recommended quality control procedures

Refer to the instructions for use of the respective Zyto*Dot* CISH ERBB2 Probe. If the cells on the ERBB2 Control Slide fail to show a positive or negative staining, respectively, results of test samples should be considered as being invalid.

15. Performance characteristics

Refer to the instructions for use of the respective Zyto*Dot* ERBB2 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

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Our experts are available to answer your questions.
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