


ZytoLight
SPEC MYC/CDKN2A/ERBB2/ZNF217
Quadruple Color Probe

REF Z-2204-200

 20 (0.2 ml)

For the detection of the human genes MYC, CDKN2A, ERBB2, and ZNF217 by fluorescence *in situ* hybridization (FISH)



IVD

In vitro diagnostic medical device

according to EU directive 98/79/EC



Fluorescence-labeled polynucleotide probe for the detection of the human genes MYC, CDKN2A, ERBB2, and ZNF217, ready to use

Product Description

- Content:** ZytoLight SPEC MYC/CDKN2A/ERBB2/ZNF217 Quadruple Color Probe (PL162) in hybridization buffer. The probe contains blue-labeled polynucleotides (ZyBlue: excitation at 418 nm and emission at 467 nm, similar to DEAC), which target the MYC gene in 8q24.21, red-labeled polynucleotides (ZyRed: excitation at 580 nm and emission at 599 nm, similar to Texas Red), which target the CDKN2A gene in 9p21.3, green-labeled polynucleotides (ZyGreen: excitation at 503 nm and emission at 528 nm, similar to FITC), which target the ERBB2 gene in 17q12-q21.1, and gold-labeled polynucleotides (ZyGold: excitation at 532 nm and emission at 553 nm, similar to Rhodamine 6G), which target the ZNF217 gene in 20q13.2.
- Product:** Z-2204-200: 0.2 ml (20 reactions of 10 µl each)
- Specificity:** The ZytoLight SPEC MYC/CDKN2A/ERBB2/ZNF217 Quadruple Color Probe (PL162) is designed to be used for the detection of the human genes MYC, CDKN2A, ERBB2, and ZNF217 in formalin-fixed, paraffin-embedded tissue or cells by fluorescence *in situ* hybridization (FISH).
- Storage/Stability:** The ZytoLight SPEC MYC/CDKN2A/ERBB2/ZNF217 Quadruple Color Probe (PL162) must be stored at 2...8°C protected from light and is stable through the expiry date printed on the label.

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| Use: | This product is designed for <i>in vitro</i> diagnostic use (according to EU directive 98/79/EC). Interpretation of results must be made within the context of the patient's clinical history with respect to further clinical and pathologic data of the patient by a qualified pathologist! |
| Safety Precautions: | <p>Read the operating instructions prior to use!</p> <p>Do not use the reagents after the expiry date has been reached!</p> <p>This product contains substances (in low concentrations and volumes) that are harmful to health. Avoid any direct contact with the reagents. Take appropriate protective measures (use disposable gloves, protective glasses, and lab garments)!</p> <p>If reagents come into contact with skin, rinse skin immediately with copious quantities of water!</p> <p>A material safety data sheet is available on request for the professional user!</p> |

Principle of the Method

The presence of certain nucleic acid sequences in cells or tissue can be detected by *in situ* hybridization using labeled DNA probes. The hybridization results in duplex formation of sequences present in the test object with the labeled DNA probe.

Duplex formation (with sequences of the genes *MYC*, *CDKN2A*, *ERBB2*, and *ZNF217* in the test material) is directly detected by using the tags of fluorescence-labeled polynucleotides.

Instructions

Pretreatment (dewaxing, proteolysis, post-fixation) should be carried out according to the needs of the user.

Denaturation and hybridization of probe:

1. Pipette 10 μ l ZytoLight SPEC MYC/CDKN2A/ERBB2/ZNF217 Quadruple Color Probe (PL162) each onto individual samples

A gentle warming of the probe, as well as using a pipette tip which has been cut off to increase the size of the opening, can make the pipetting process easier. Avoid long exposure of the probe to light.

2. Avoiding trapped bubbles, cover the samples with a coverslip (22 mm x 22 mm). Seal the coverslip, e.g. with a layer of hot glue from an adhesive pistol or with rubber cement

3. Denature the slides at 75°C ($\pm 2^\circ$ C) for 10 min, e.g. on a hot plate

Depending upon the age of the sample and variations in the fixation stage, it may be necessary to optimize the denaturing temperature (73°C-77°C).

4. Transfer the slide to a humidity chamber and hybridize overnight at 37°C (e.g. in a hybridization oven)

It is essential that the tissue/cell samples do not dry out during the hybridization step.

Further processing, such as washing and counter-staining, can be completed according to the user's needs. For a particularly user-friendly performance, we recommend the use of a ZytoLight FISH system by ZytoVision. These systems were also used for the confirmation of appropriateness of the ZytoLight SPEC MYC/CDKN2A/ERBB2/ZNF217 Quadruple Color Probe (PL162).

Results

With the use of appropriate filter sets, the hybridization signals of the labeled MYC gene appear blue, the hybridization signals of the labeled CDKN2A gene appear red, the hybridization signals of the labeled ERBB2 gene appear green, and the hybridization signals of the labeled ZNF217 gene appear golden. In interphases of normal cells or cells without aberrations of chromosomes 8, 9, 17, and 20, two MYC signals, two CDKN2A signals, two ERBB2 signals, and two ZNF217 signals appear. In cells with an amplification of the MYC, ERBB2, and/or ZNF217 gene or a deletion of the CDKN2A gene, a different signal pattern is visible in interphases.

In order to judge the specificity of the signals, every hybridization should be accompanied by controls. We recommend using at least one control sample in which the MYC, CDK2NA, ERBB2, and ZNF217 gene copy number is known.

Care should be taken not to evaluate overlapping cells, in order to avoid false results, e.g. an amplification of genes. Due to decondensed chromatin, single FISH signals can appear as small signal clusters. Thus, two or three signals of the same size, separated by a distance equal to or less than the diameter of one signal, should be counted as one signal.

Our experts are available to answer your questions.

Literature

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