



## ZytoDot CEN 8 Probe

0.4 ml

For the detection of human alpha-satellites of chromosome 8 by  
chromogenic *in situ* hybridization (CISH)

For in vitro diagnostic use

Product No.: C-3016

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**Digoxigenin labeled polynucleotide probe for the detection of human  
alpha-satellites of chromosome 8 centromeres by CISH,  
ready to use**

**Product description**

**Content:** 0.4 ml (25 reactions of 15  $\mu$ l each) ZytoDot CEN 8 Probe in hybridization buffer. The probe contains labeled polynucleotides (digoxigenylated) which target alpha-satellite-sequences of the centromere of chromosome 8.

**Product No.:** C-3016 (ZytoDot CEN 8 Probe)

**Specificity:** The ZytoDot CEN 8 Probe is designed to be used for the detection of chromosome 8 alpha-satellites in formalin-fixed, paraffin-embedded tissue or cells by *in situ* hybridization via CISH.

**Storage/Stability:** The ZytoDot CEN 8 Probe must be stored at -20°C (short-time storage at 4°C is possible) and is stable through the expiry date printed on the label.

**Use:** This product is designed for in vitro diagnostic use. Interpretation of results must be made within the context of the patient's clinical history with respect to further clinical and pathologic data of patient by a qualified pathologist!

**Safety Precautions:** Read the operating instructions prior to use!

Do not use the reagents after the expiry date has been reached!

This product contains formamide and kathon in low concentrations and volumes. Avoid any direct contact with the reagents. Take appropriate protective measures (use disposable gloves, protective glasses, and lab garments). The following risk and safety phrases apply to: R61 May cause harm to the unborn child. S53 Avoid exposure – obtain special instructions before use. S45 In case of accident or if you feel unwell, seek

medical advice immediately (show the label, where possible)!

If reagents come into contact with skin, rinse skin immediately with copious quantities of water!

A material safety data sheet is available on request for the professional user!

## Principle of the method

The presence of certain nucleic acid sequences in cells or tissues can be detected via *in situ* hybridization, using tagged DNA probes. The hybridization results in duplex formation of specific sequences present in the test object with the tagged DNA probe.

Duplex formation of the digoxigenin-labeled probe (with sequences of chromosome 8 alpha-satellites in the test material) can be visualized using a primary (unmarked) anti-digoxigenin antibody, which is detected by a secondary polymerized enzyme-conjugated antibody. The enzymatic reaction of a chromogenic substrate leads to the formation of a color precipitate that is visualized by light microscopy.

## Instructions

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

Denaturation and hybridization of probe:

**1.** Vortex the ZytoDot CEN 8 Probe and pipette 15  $\mu$ l onto individual sample

*Distribute dropwise on the whole target area to avoid local concentration of probe. Alternatively, add probe to the center of a coverslip and place coverslip upside down on target area.*

**2.** Avoiding trapped bubbles, cover the samples with a coverslip (22 mm x 22 mm) and seal with rubber cement (Fixogum)

**3.** Denature the slides at 94°C ( $\pm$ 2°C) for 5 min, e.g. on a hot plate

**4.** Transfer the slides to a humidity chamber and incubate overnight at 37°C, e.g. in a heating oven

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

Further processing, such as washing, detection, and counter-staining can be completed according to the user's needs. For a particularly user-friendly performance, we recommend the use of a *ZytoDot* CISH system (ZytoVision). These systems were also used for the confirmation of appropriateness of the *ZytoDot* CEN 8 Probe.

## Results

In order to judge the specificity of the hybridization signals, every hybridization should be accompanied by controls. In an interphase nucleus of normal cells or cells without aberrations of chromosome 8 two chromosome 8-specific punctual signals appear which can be clearly distinguished from the background. In cells with an aneuploidy of chromosome 8, a different signal pattern or cluster is visible in interphases.

**Our experts are available to answer your questions.**

## Literature

Waye JS and Willard HF (1987) *Nucleic Acids Res* **15**: 7549-69.

Tsukamoto T, et al. (1991) *Int J Dev Biol* **35**: 25-32.

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*As of: February 15, 2007 (1.0)*