

DATA SHEET

eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe

Catalog No.**FP096-10X - 100µl- 10 test****FP096-20X - 200µl- 20 test**

Doc No: 932-FP096 Rev: A

Date of Release: 30-Nov-2016

Material Provided: One vial of [eFISH probe](#) in hybridization buffer (RTU).**Recommended detection system (Not supplied):**

Either of the following detection systems is recommended depending on the automation/manual platform used:

eFISH Kit	Cat #	Description
eFISH Histo	DF-500-20X	Automation
eFISH Cyto	DF-510-20X	Automation

Intended Use:

The BioGenex eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe is currently available for Research use only. eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe is designed to be used for the detection of human chromosome 13q12 specific sequences as well as chromosome 18 alpha satellites and chromosome 21q22 specific sequences in formalin-fixed, paraffin-embedded tissue or cells by fluorescence in situ hybridization (FISH).

The BioGenex eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe Probe in hybridization buffer. The probe contains green-labeled polynucleotides (Green: excitation at 503 nm and emission at 528 nm, similar to FITC), which target chromosome 13 specific sequences, blue-labeled polynucleotides (Blue: excitation at 418 nm and emission at 467 nm, similar to DEAC) which target alphasatellite- sequences of the centromere of chromosome 18, and orange-labeled poly-nucleotides (Orange: excitation at 547 nm and emission at 572 nm, similar to rhodamine), which target chromosome 21 specific sequences.

Summary and Explanation

Fluorescence *in situ* hybridization (FISH) is a robust technique of cytogenetic used for the detection of chromosomal aberrations, presence or absence of specific DNA sequence in native context. In this technique florescent probes bind to the target sequence of DNA in chromosome. High specificity and sensitivity coupled rapid and an accurate result has proven role of FISH in both research and diagnosis of solid tumor and hematological malignancies. As technique of cancer cytogenetics, FISH, can be used to identify genetic

aberrations viz., deletions, amplification and translocation in tissue sections or within individual cells. FISH is also used for use in genetic counseling, medicine, and species identification. FISH can also be used to detect and localize specific RNA targets in cells, circulating tumor cells, and tissue samples^{1,2,3,4,5}.

In FISH procedure, fixed tissue sections are pretreated to expose target DNA or mRNA sequences. An appropriately labeled probe is hybridized to the exposed target DNA or mRNA sequences in the cells. Subsequent stringent washing steps remove any probe that is non-specifically bound to the tissue section. Subsequently slides are mounted using DAPI/antifade and can be visualized under fluorescence microscope using appropriate filter set.

Principles of the Procedure

In Situ hybridization (ISH) allows the detection and localization of definitive nucleic acid sequences directly within a cell or tissue. High specificity is ensured through the action of annealing of fluorescence probe nucleic acid sequence to complementary target nucleic acid sequence. ISH techniques can be used to identify genetic aberrations like deletions, amplification, and translocation in tissue sections or within individual cells.

Storage and Handling

The BioGenex eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe must be stored at 2-8°C protected from light and is stable through the expiry date printed on the label.

Specimen Collection and Slide Preparation

Tissues fixed in 10% (v/v) formalin are suitable for use prior to paraffin embedding and sectioning.

FISH Staining procedure

- (a) The BioGenex eFISH probes are supplied in hybridization buffer and used without further dilution.
- (b) Protocol:

Please refer to the eFISH probe specific instruction/protocol for automated or semi-automated [FISH processing platform](#) (Xmatrx®-Infinity, Xmatrx®-Nano and Xmatrx®-mini).

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 BioGenex [eFISH kit](#).

Disclaimer: The above information is provided for reference only. Each end-user is responsible for developing and validating optimal testing conditions for use with this product.

Troubleshooting

Contact BioGenex Technical Service Department at **1-800-421-4149** or your local **distributor** to report unusual staining.

Expected Results

With the use of BioGenex eFISH SPEC 13/CEN 18/SPEC 21 Triple Color probe, the hybridization signals of labeled chromosome 13 specific sequences appear green; the hybridization signals of labeled alpha-satellite-sequences of the centromere of chromosome 18 appear blue, and the hybridization signals of labeled chromosome 21 specific sequences appear orange. In interphases of normal cells or cells without aberrations of chromosomes 13, 18, and 21, two chromosome 13, two chromosome 18, and two chromosome 21 signals appear. In cells with an aneuploidy of one of the chromosomes mentioned above, 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 chromosome 13, 18, and 21 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.

Limitations of the Procedure

Correct treatment of tissues prior to and during fixation, embedding, and sectioning is important for obtaining optimal results. Inconsistent results may be due to variations in tissue processing, as well as inherent variations in tissue. The results from *in situ* hybridization must be correlated with other laboratory findings.

Bibliography

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2. Rudkin, G. T. and Stollar, B. D. (1977). *Nature* 265,472-473.
3. Hougaard, D. M., Hansen, H. and Larsson, L. I. (1997). *Histochem. Cell Biol.* 108,335 -344.
4. Bauman, J. G., Wiegant, J., Borst, P. and van Duijn, P. (1980). *Exp. Cell Res.* 128,485 -490.
5. O'Connor, C. (2008). *Nature Education* 1(1):171.