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DATA SHEET

eFISHD13S319/13q34/CEN 12 Triple Color Probe

Catalog No.
FP078-10X-100µl- 10 test
FP078-20X -200µl- 20 test

Doc No: 932-FP078 Rev: D
Date of Release: 10-Aug-2020

Material Provided: One vial of eFISH probe in hybridization buffer (RTU).

Recommended detection system (Not supplied):
Either of the following detection systemsis recommended depending on the automation/manual platform used:

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

Intended Use:
The BioGenex eFISHD13S319/13q34/CEN 12Triple Color Probe is currently available for Research use only.eFISHD13S319/13q34/CEN 12Triple Color Probe is designed to be usedfor the detection of the human D13S319 regionas well as human 13q34 specific sequences andchromosome 12 alpha-satellites in formalin-fixed,paraffin-embedded tissue or cells by fluorescencein situ hybridization (FISH).

eFISHD13S319/13q34/CEN 12 TripleColor Probe is provided in hybridization buffer. Theprobe contains orange-labeled polynucleotides (Orange: excitation at 547 nm and emission at572 nm, similar to rhodamine), which target theD13S319 region, blue-labeled polynucleotides (Blue: excitation at 418 nm and emission at467 nm, similar to DEAC), which targetchromosome 13q34 specific sequences, andgreen-labeled polynucleotides (Green:excitation at 503 nm and emission at 528 nm,similar to FITC), which target alpha-satellitesequencesof the centromere of chromosome 12.

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/anti-fade 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 translocationin tissue sections or within individual cells.

Storage and Handling
The BioGenex eFISH D13S319/13q34/CEN 12Triple Color Probemust 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.

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Further processing, such as washing and counter-staining, can becompleted according to the user's needs. For a particularly user-friendlyperformance, we recommend the use of a BioGenexeFISH kit.

Disclaimer: The above information is provided for reference only. Each end-user is responsible for developingand 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
The BioGenex eFISH D13S319/13q34/CEN 12Triple Color Probe is a mixture of an orange fluorochrome direct labeled eFISH D13S319 probe specific for the D13S319 locus at 13q14.2, a blue fluorochrome direct labeled eFISH 13q34 probe specific for the chromosomal region 13q34 and a green fluorochrome direct labeled CEN 12 probe specific for the alpha satellite centromeric region of chromosome 12 (D12Z3).The BioGenex eFISH 13q34 probe is specific for the LAMP1 (lysosome-associated membrane protein 1) gene region in 13q34. Due to cross-hybridizations of chromosome 13 al pha satellites to other centromeric regions, probes specific for 13q34 are frequently used for chromosome 13 copy number detection.

Using the BioGenex eFISH D13S319/13q34/CEN 12 Triple Color Probe in a normal interphase nucleus, two orange, two green, and two blue signals are expected. In a cell with deletions affecting the D13S319 locus, a reduced number oforange signals will be observed. Deletions affecting only parts of the D13S319 locus might result in a normal signal patternwith orange signals of reduced size. In a cell with trisomy or polysomy 12, three or more copies of the green signal will be observed, respectively.

Care should be taken not to evaluate overlapping cells, in order to avoidfalse results, e.g. an amplification of genes. Due to decondensedchromatin, single FISH signals can appear as small signal clusters. Thus,two or three signals of the same size, separated by a distance equal to orless 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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