

DATA SHEET

eFISHPML/RARA Dual Color Dual Fusion Probe

Catalog No.**FP073-10X- 100µl-10 test****FP073-20X- 100µl-10 test**

Doc No: 932-FP073 Rev: C

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 system is 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 eFISH PML/RARA Dual color dual fusion probe is currently available for Research use only. eFISH PML/RARA Dual color dual fusion probe is designed to be used for the detection of the translocation (15;17)(q24;q21) in formalin-fixed, paraffin embedded tissue or cells by fluorescence in situ hybridization (FISH).

BioGenex eFISH PML/RARA Dual color dual fusion probe comes in hybridization buffer contains orange-labeled polynucleotides (Orange: excitation at 547 nm and emission at 572 nm, similar to rhodamine), which target the PML gene in 15q24.1, and green-labeled polynucleotides (Green: excitation at 503 nm and emission at 528 nm, similar to FITC), which target the RARA gene in 17q21.

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 fluorescent 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 an 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, translocation in tissue sections or within individual cells.

Storage and Handling

The BioGenex eFISH PML/RARA dual color dual fusion 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.

Protocol

Please refer to the 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. These systems were also used for the confirmation of appropriateness of the BioGenex eFISH PML/RARA dual color dual fusion probe.

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

The use of BioGenex eFISH PML/RARAdual color dual fusion probe in interphase and normal chromosome where there is no t(15;17)(q24;q21) translocation produces two green and two orange signals separately. Where as in regions of no t(15;17)(q24;q21) translocation, the signals appear as two green/orange fusion signals in addition to a separate single green and orange signal.

We recommend the use of atleast one control in which the t(8,21) status is known to judge the specificity of the signals.

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

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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