WHITEPAPER

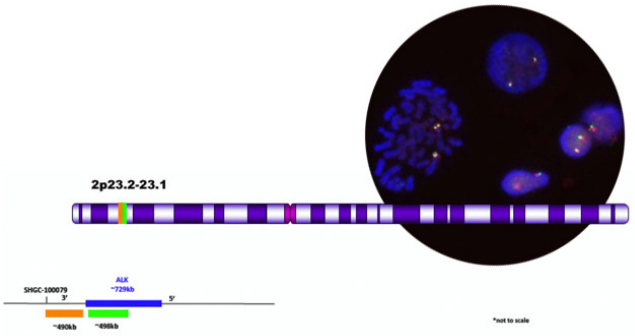
Biocare Basics: Break-Apart Probes

BIOCARE MEDICAL

Biocare Basics: Break-Apart Probes

Fluorescence in situ hybridization (FISH) is a method of detecting disease states and genetic anomalies through the creation of single-stranded DNA fragments designed to complementarily bind to sequences of interest in a cell sample.[3] These constructed DNA "probes" are labeled with fluorochrome tags.[3] The fluorochromes generate fluorescent light when excited with a specialized microscope, allowing the binding site to be visualized.[3] The presence of the fluorescent signal shows that the probe successfully hybridized to the target sequence, which indicates that the sequence is present.[3]

One type of genetic anomaly that FISH is used to detect is a translocation mutation. A translocation mutation occurs when a piece of one chromosome breaks away and attaches to another chromosome.[2] This can result in a gene fusion mutation as the translocation fuses together two genes that were previously separated.[2] Translocation mutations have been identified as a causative factor in various disease states and disorders such as leukemia, breast cancer, muscular dystrophy, and down syndrome.[2]

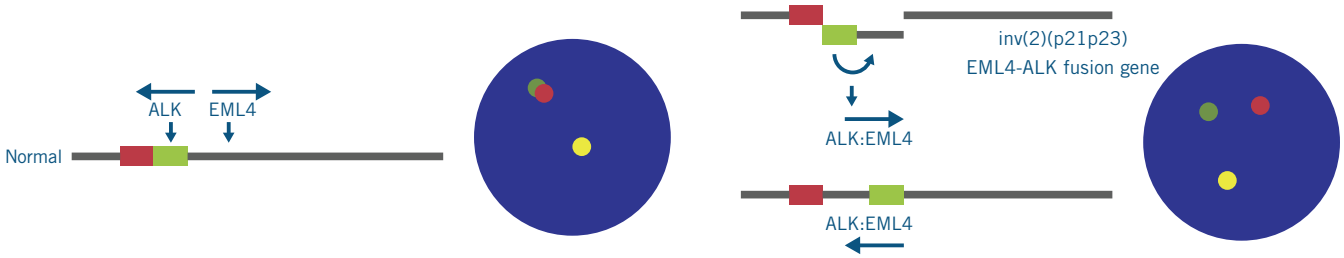


ALK (2p23.2) Break Apart FISH Probe Orange/Green

Some genes have been associated with multiple different translocation mutations that have been associated with cancer. In this case, instead of using specific dual fusion probes to test for each possible translocation, a break-apart probe is used to test whether the gene of interest has translocated in general.[2]

A break-apart probe uses two differently colored probes to bind sequences on either side of a known gene of interest. A separate control probe in a third color binds to another sequence to confirm that hybridization has been successful. If the gene has remained in place, the two colors (in this case, red and green) will appear close together.[3] If there has been a translocation, the two probes will "break apart," and so the red and green colors will appear separate from each other.[3]

Break-apart probes are less specific than dual-fusion probes and provide less information since they do not define the exact place to which the gene of interest has translocated.[3] However, if the procedure design is appropriate, they can give clear, easy-to-interpret results since the separation of two signals is readily recognizable under a microscope.[3] Even so, what constitutes a "normal" signal must be carefully defined since normal signals may occasionally appear slightly separated, depending on the localization and DNA structure.[3] To prevent false-positive results, a signal might be scored positive only at a certain distance.[3]



For more information, please visit our website at biocare.net or call 1-800-799-9499.

1. Donát Alpár; Judit Hermesz; László Pótó; Renáta László; László Kereskai; Pál Jáksó; Gábor Pajor; László Pajor; Béla Kajtár (2008). Automated FISH analysis using dual-fusion and break-apart probes on paraffin-embedded tissue sections. , 73A(7), 651–657. doi:10.1002/cyto.a.20557

2. University of Wisconsin Cytogenetic Services and Molecular Genetics. (2022). Fluorescent In Situ Hybridization (FISH). Wisconsin State Laboratory of Hygiene at the University of Wisconsin-Madison. Retrieved April 19, 2022, from http://www.slh.wisc.edu/clinical/cytogenetics/fish/

3. Ventura, Roland A.; Martin-Subero, Jose I.; Jones, Margaret; McParland, Joanna; Gesk, Stefan; Mason, David Y.; Siebert, Reiner (2006). FISH Analysis for the Detection of Lymphoma-Associated Chromosomal Abnormalities in Routine Paraffin-Embedded Tissue. The Journal of Molecular Diagnostics, 8(2), 141–151. doi:10.2353/jmoldx.2006.050083