DNAscope (TM): A novel chromogenic in-situ hybridization technology for high-resolution detection of DNA copy number and structural variations

Genomics

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Genomic DNA anomalies such as copy number variations (gene duplication, amplification, deletion) and gene rearrangements are important biomarkers in many disease types. DNA in-situ hybridization (ISH) is the gold standard method to directly visualize these molecular alterations in formalin-fixed paraffin-embedded (FFPE) tumor tissues at single-cell resolution within a histological section. However, currently available fluorescent ISH (FISH) assays provide limited morphological detail due to the use of fluorescent nuclear staining compared to chromogenic staining. Furthermore, FISH techniques rely on expensive fluorescence microscopes, risk loss of fluorescent signal over time and involve tedious imaging at high magnifications (100X). There is thus an unmet need for a sensitive and robust chromogenic DNA-ISH assay that can enable high-resolution detection of genomic DNA targets with the ease of bright-field microscopy.

We present here DNAscope - a novel chromogenic DNA-ISH assay - for detecting and visualizing genomic DNA targets under a standard light microscope. DNAscope is based on the widely used RNAscope® double-Z probe design and signal amplification technology and provides unparalleled sensitivity and specificity with large signal dots readily visualized at 40X magnification and with full morphological context. Furthermore, DNAscope ensures specific DNA detection without interference from RNA due to the use of a novel RNA removal method. Using a duplex chromogenic detection assay in red and blue, we demonstrate highly specific and efficient detection of gene rearrangements (ALK, ROS1, RET and NTRK1), gene amplification (ERBB2, EGFR, MET) and deletion (TP53 and CDKN2A). The DNAscope assay has been carefully optimized for probe signal size and color contrast to enable easy interpretation of signal patterns under conventional light microscopy or digital pathology. Compared to conventional FISH assays, DNAscope probes are standard oligos that are designed in silico to be free of any repetitive sequences and can be rapidly synthesized for any DNA target. In conclusion, the DNAscope assay provides a powerful and convenient alternative to commonly used FISH assays in many research applications.