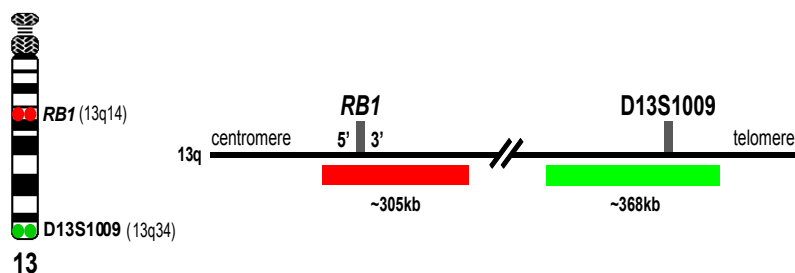


## Intended Use

The *RB1*/D13S1009 DNA-FISH Probe is designed to detect loss of the *RB1* gene on chromosome 13q14 relative to the control marker, D13S1009 on chromosome 13q34, using fluorescence *in situ* hybridization (FISH). The *RB1* gene is a well characterized tumor-suppressor gene and bi-allelic inactivation of the gene due to mutations and/or deletions is causal for the development of Retinoblastoma (RB). Deletion of the *RB1* gene is also common in a wide variety of solid tumors and hematologic malignancies such as chronic lymphocytic leukemia (CLL), multiple myeloma (MM), acute myelocytic leukemia (AML), myelodysplastic syndrome (MDS), and chronic myeloproliferative disorders.<sup>[1-3]</sup> The *RB1* gene is proximal to the D13S25 locus at 13q14, which is often co-deleted with the *RB1* gene in some B-cell hematologic malignancies.

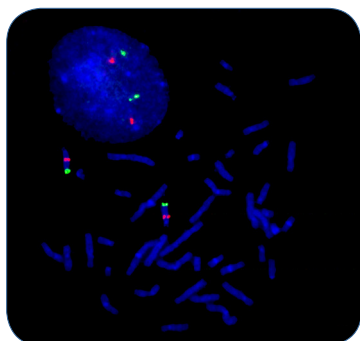


### Schematic of the *RB1*/D13S1009 DNA-FISH Probe:

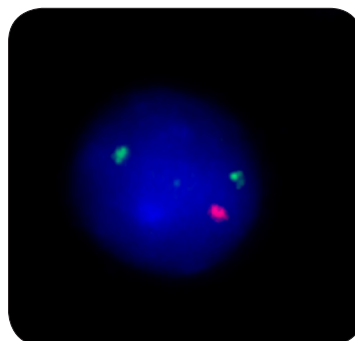
Horizontal red and green bars indicate the regions covered by the probes (approximate to scale, NCBI Build 36.1/Hg18/2006). The *RB1* probe (red) spans the entire gene while the D13S1009 probe (green) hybridizes to the region surrounding the D13S1009 locus marker and serves as a control.

## Signal Interpretation

In normal diploid interphase nuclei and metaphase chromosomes, the probe generates two red and two green signals corresponding to the two normal homologous chromosomes 13 (Figure 1). In cells with deletion of an entire chromosome 13, the number of red (*RB1*) and green (D13S1009) signals will decrease. In cells with interstitial deletion of chromosome 13, in which the *RB1* locus is deleted and the D13S1009 locus is retained, one red (*RB1*) and two green (D13S1009) signals will be observed (Figure 2). Please note that the *RB1* probe size (305 kb) extends beyond the gene, therefore, small internal *RB1* deletions may not result in a detectable loss of signal. If unexpected signal patterns are observed, hybridization to metaphase chromosomes is recommended.



**Figure 1:** Normal diploid metaphase and interphase nucleus (from normal peripheral blood specimen) with 2 red (*RB1*) and 2 green (D13S1009) signals.



**Figure 2:** Interphase nuclei with 1 red (*RB1*) and 1 green (D13S1009) signals.

## References

1. Lee, W.-H., et al., *Science*, 1987. 235: 1394-99.
2. Steigenbauer, S., et al., *Blood*, 1993. 81(8): 2118-24.
3. Dao, D.D., et al., *Leukemia*, 1994. 8(8): 1280-4.

## Fluorescence Microscopy Filter Requirements

Fluorophore	Excitation <sub>max</sub>	Emission <sub>max</sub>
Green	496 nm	520 nm
Red	580 nm	603 nm
DAPI	360 nm	460 nm

Instructions for use are available at [www.cancergeneticsitalia.com](http://www.cancergeneticsitalia.com)