BIOSTATUS

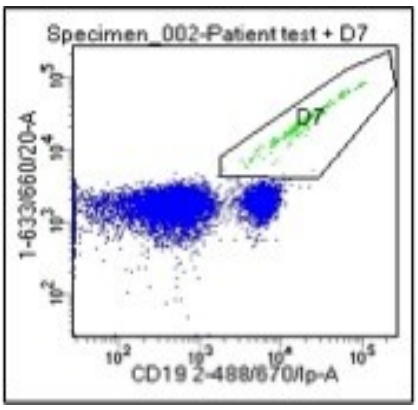
DRAQ7™ in FC-Based Apoptosis Assays

Far-Red Fluorescent Live-Cell Impermeant DNA Dye

1. IDENTIFYING MEMBRANE-PERMEABILIZED CELLS IN APOPTOSIS ASSAYS

BACKGROUND

In apoptosis an ordered series of events leads to destruction of a single cell. The changes that occur can be assayed through DNA fragmentation, caspase activity (using permeant substrates e.g. FLICA™ probes), mitochondrial membrane potential (e.g. using TMRM), cytochrome C release, plasma membrane inversion (using Annexin V) and permeabilization being common choices. Plasma membrane permeabilization is a late hallmark of apoptosis identified with a viability probe. This is combined with other apoptotic features to give temporal context and to identify cells permeabilized by non-apoptotic means (e.g. shear forces).



WHAT IS THE PROBLEM?

To achieve such analysis, traditional viability dyes like propidium iodide (PI) or DAPI have been used. However, each has challenges associated with it.

PI progressively equilibrates with intact cells resulting in these cells drifting right on plots making it difficult to reliably set a gate for positive cells. Importantly, PI's fluorescence completely overlaps with the antibody conjugate R-PE and with TMRM and JC-1, commonly used mitochondrial membrane potential probes.

DAPI is UV-/violet-excited and cannot be combined with UV-excited Hoechst 33342 probe commonly used as an "all-event" marker in apoptosis and cell health assays.

Photo-switching of DAPI and photo-bleaching of PI as well as evidence of semi-permeance make these DNA binding agents incompatible with long-term assays. Typically these agents need to be prepared fresh from hard compound and cannot be stored long-term in a ready-to-use solution.

HOW DOES DRAQ7™ HELP?

As a far-red DNA-binding viability dye DRAQ7™ immediately alleviates the problems of spectral overlap with TMRM (PI) and UV-excited dyes like monochlorbimane (Glu-SH probe) and Hoechst 33342 which be used together with DRAQ7™. For example, DRAQ7™ can be combined with Hoechst 33342, Annexin V-FITC, TMRM a useful and temporally relevant combination of parameters. DRAQ7™ is extremely chemically and photo-stable.

Uniquely, DRAQ7™ is compatible with long-term, real-time cell health assays meaning that cells can be sampled serially to determine the temporal development of the apoptotic events in the assay system.

DRAQ7™ is stored in a ready-to-use aqueous format with a very long shelf-life. It is convenient and easy to use.

DRAQ7™ Product Features:

- far-red fluorescing cell impermeant dsDNA probe
- rapidly and clearly labels only permeabilized cells
- compatible with BUV / BV, FITC & R-PE dyes
- compensation-free dead cell exclusion (via virtual channel)
- water-soluble; ready-to-use from the fridge

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