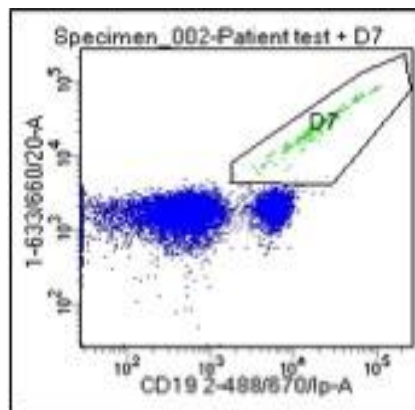




1. IDENTIFYING MEMBRANE-COMPROMISED CELLS IN CELL HEALTH ASSAYS

BACKGROUND

One of the biggest costs in drug discovery and development has been failure of drug candidates late in the process (or worse post-launch) due to unexpected or “idiosyncratic” toxicity, not to mention the risk to patient health. To reduce this risk compound libraries are exposed to a battery of mandatory tests, however these have been performed when the new chemical entity (NCE) is often well advanced. Recently, new *in vitro* toxicity assays have been developed that use physiologically-relevant cells and measure their health under different doses of a compound using a few simple parameters, e.g. total cell count, mitochondrial membrane potential, calcium efflux and plasma membrane integrity. Changes in these are detectable with fluorescent functional probes and an assay based on these parameters would be highly amenable to high-throughput flow cytometry.



WHAT IS THE PROBLEM?

For the measurement of plasma membrane integrity traditional viability dyes (DAPI, PI, 7-AAD) have proved to be incompatible despite the assay’s simplicity. They are spectrally incompatible, have high non-specific binding that drifts upwards with time or poor discrimination of intact, intermediate and leaky/dead cells. These characteristics make them unsuitable. More recent alternatives include TOTO-3 and TO-PRO-3 but these are costly to run in the volumes associated with non-adherent cells such as leukocytes. The chemistry of all these dyes means that they are supplied in hard compound form and require DMSO to get them into solution and cannot be stored in aqueous solution for extended periods.

HOW DOES DRAQ7™ HELP?

As a far-red DNA-binding viability dye DRAQ7™ immediately alleviates the problems of spectral overlap with functional probes such as TMRM, Fluo-4 and Hoechst 33342.

DRAQ7™ rapidly stains cells with compromised membranes giving an excellent separation between intact and damaged/dead cells. It is cost-effective and reliable in such assays.

Uniquely, DRAQ7™ is compatible with long-term, real-time cell health assays meaning that cells can be sampled serially to determine temporal deterioration of cell health. This also means that it can be added to cells for end-point analysis without affecting the protocol.

DRAQ7™ has excellent chemical- and photo-stability. Accordingly, DRAQ7™ is supplied in a ready-to-use aqueous format with a very long shelf-life making it amenable to high-throughput applications where reagents are pipetted from bulk quantities on robotic platforms, often with limited environmental control.

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

