DRAQ7<sup>™</sup> in Image-Based Screening

Far-Red Fluorescent Live-Cell Impermeant DNA Dye

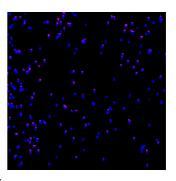


# DR7.APPNOTE: IMAGE SCREEN 002 230323

## 1. DEAD/DAMAGED CELL IDENTIFICATION

### BACKGROUND

In most cell-based assays for high content screening (HCS, target-based or phenotypic) fluorescence-based imaging is preferred. Several parameters can be studied simultaneously: a disease-related readout such as protein translocation can be combined with unrelated features that may reflect toxicity of the compounds (or siRNAs) being tested, whether desired (for example, with an anti-cancer agent) or not. In such live-cell end-point HCS assays it can help to estimate cell death as a measure of toxicity. In more sophisticated temporal- and dose-response assays a real-time measure of cell viability may be advantageous to gain a fuller understanding of cellular sensitivity to a compound. In principle, a simple means of achieving this would be the addition of fluorescent viability dye, typically a cell membrane-impermeant DNA probe that labels the nuclei of membrane-compromised cells.



### WHAT IS THE PROBLEM?

A number of cell membrane-impermeant DNA dyes are candidates to report the presence of dead/damaged cells in HCS assays. In fluorescence-based assays it eases assay design if the viability dye is spectrally separated from most reagents likely to be used in HCS (allowing it to be chosen as a default component) and would include GFP, the spectrum of other fluorescent proteins available to tag biologically-relevant proteins and the cell permeant functional and organelle-specific probes. In this context, propidium iodide (PI) is not ideal due to its broad emission spectrum. Likewise, TOTO-3 and TOPRO-3 have orange/red emission that can occlude the valuable rhodamine-based dyes, depending on filter settings. Meanwhile, DAPI, needs equipment that is UV-enabled and due to spectral overlap with GFP and fluorescein-based probes would require duplicate scanning of samples.

Long-term or real-time monitoring has become of greater interest with the understanding of idiosyncratic toxicity. None of the viability dyes described above have been validated for long-term monitoring of cell health.

Typically these agents need to be prepared fresh from hard compound and cannot be stored long-term in a ready-to-use aqueous format, and are prone to photobleaching.

#### HOW DOES DRAQ7<sup>™</sup> HELP?

As a far-red DNA-binding viability probe DRAQ7<sup>™</sup> immediately avoids spectral overlap with visible-range and UVexcited probes allowing it to be used as a default viability probe. It has been validated for long-term, real-time use and exhibits extremely low photobleaching. DRAQ7<sup>™</sup> only enters cell with compromised membranes and, due to its high dsDNA specificity, reports the nuclear texture of dead and dying cells.

DRAQ7<sup>™</sup> consolidates observation of treatment effect (e.g. protein translocation) and toxicity measurement, cell-by-cell, without duplicating cell culture and reagent costs to run a separate homogeneous ATP-based method.

DRAQ7<sup>™</sup> is stored in a ready-to-use aqueous format with a very long shelf-life.

#### **DRAQ7™** Product Features:

- far-red fluorescing cell impermeant dsDNA probe
- rapid, clear labelling of only the permeabilized cells
- compatible with UV-/violet-excited, fluorescein and rhodamine dyes
- unifies observation of target biology and toxicity
- DMSO-free; long-term assay compatible



For a full price list and further information see www.biostatus.com or contact us at: BioStatus Limited

56a Charnwood Road, Shepshed, Leicestershire LE12 9NP United Kingdom T +44 1509 558 163 | F +44 1509 651 061 | E enquiry@biostatus.com | W www.biostatus.com