



CyTRAK Orange™ in Plate-Based Cytometry

Orange Fluorescent Live-Cell Permeant DNA and Cytoplasmic Dye

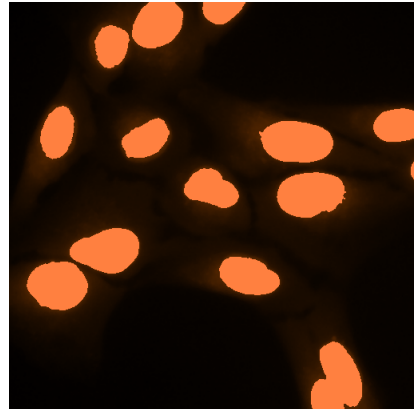


CO5.APPNOTE: PLATE CYTOMETRY 002 040814

1. NUCLEATED CELL LABELLING & NUCL:CYTO SEGMENTATION

BACKGROUND

Microplate- and slide-based cytometers (e.g. TTP Labtech's Acumen eX³, Nexcelom's Celigo S) utilising laser scanning combine the morphological (textural) capability of fluorescence-based imaging with the cell-by-cell multi-parameter analysis of a cytometer. These instruments are particularly valuable for adherent cell types that are not readily compatible with flow cytometry. However, diluted whole peripheral blood samples have been analysed with such instruments for complex white and red cell analysis.



WHAT IS THE PROBLEM?

As with fluorescence microscopy individual cells can be interrogated and counted, with the possibility to segment the cells to their nuclear and cytoplasmic compartments. However, such identification requires addition of a fluorescent DNA counterstain. The ideal DNA counterstain to segment nuclei should meet all of the following criteria: show discrete nuclear staining; be spectrally separated from commonly used chromophores; work in live or fixed cells; report DNA content; be cross-platform compatible for upstream assay development.

Whole blood analysis on slide-based cytometer requires a simple means of differentiating the nucleated and enucleated cells. This could be easily achieved with a DNA counterstain with the characteristics above.

HOW DOES CyTRAK Orange™ HELP?

Orange/red fluorescing cell-permeant DNA-binding probe CyTRAK Orange™ discretely segments nucleus from cytoplasm in microplate-/slide-based cytometry in live or fixed cells, for detailed morphometric analyses. It has almost universal instrument compatibility due to its blue/green optimal excitation. With higher gain settings it is possible to achieve differential counterstaining of both nucleus and cytoplasm with this single probe. It can be combined with UV/violet-excited, FITC/GFP-like and red-excited chromophores (since CyTRAK Orange™ is not) for multi-colour experiments, and contributing to cross-platform compatibility.

CyTRAK Orange™ is cell permeant. It can be added at the end of a staining procedure for live or fixed cell assays. Likewise, CyTRAK Orange™ is compatible with complex samples such as blood / bone marrow to label the nucleated cells, without needing RBC lysis. This allows these to be easily differentiated from RBCs.

CyTRAK Orange™ Product Features:

- ❖ Orange-fluorescing cell-permeant dsDNA probe
- ❖ rapidly and clearly labels all nucleated cells (live or fixed)
- ❖ single-channel dual compartment (nucl:cyto) segmentation
- ❖ compatible with Horizon BV / BUV, FITC/GFP & red-excited dyes
- ❖ water-soluble; ready-to-use from the fridge



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

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