Nuclear Black™ DCS2 *Cell-Impermeable*

Catalog number: 17692
Unit size: 200 tests

Component	Storage	Amount (Cat No. 17692)
Nuclear Black™ DCS2	Freeze (< -15 °C), Minimize light exposure	200 tests

OVERVIEW

Nuclear Black™ DCS2 is live cell impermeable. It is the first and novel fluorescence suppression reagent optimized for live-cell imaging applications targeting red fluorescence. Some commonly used nuclear stains for live cell applications may lead to uNuclear Black™ DCS1 is live cell impermeable. It is the first and unique fluorescence suppression reagent optimized for live-cell imaging applications.clarity of red fluorescent signals from live, healthy cells in microscopy assays. Its cell-impermeable design ensures that it remains outside intact live cells and does not affect the data from live cells. Nuclear Black™ DCS2 is the first specialized reagent designed to suppress red nuclear fluorescence from dead cells in live-cell imaging experiments.
Nuclear Black™ DCS2 is ideal for live-cell fluorescence microscopy, high-content screening, and assays where signal from dead cells interferes with imaging. It's simple protocol integrates easily into existing workflows to improve imaging results.

AT A GLANCE

1. Prepare cells in growth medium.
2. Stain cells with green nuclear stain for live cells.
3. Incubate cells with Nuclear Black™ DCS2 working solution at 37°C for 30 minutes.
4. Analyze under fluorescence microscope with Cy5 filter set.

CELL PREPARATION

For adherent cells:

Plate cells overnight in growth medium at 10,000 to 40,000 cells/well/100 µL for a 96-well plate or 2,500 to 10,000 cells/well/50 µL for a 384-well plate.

For non-adherent cells:
Centrifuge the cells from the culture medium and suspend the cell pellets in fresh culture medium at 50,000-100,000 cells/well/100 µL for a 96-well poly-D lysine plate or 10,000-25,000 cells/well/50 µL for a 384-well poly-D lysine plate. Centrifuge the plate at 800 rpm for 2 minutes with the brake off before your experiment.

Note: Each cell line should be evaluated individually to determine the optimal cell density.

PREPARATION OF WORKING SOLUTION

Nuclear Black™ DCS2 working solution
Prepare 0.2X to 1X Nuclear Black™ DCS2 working solution in HHBS buffer (AAT Cat# 20011), and mix well. The working solution is stable for 2 hours at room temperature.

Note: The provided stock solution is 100X.

Note: Unused Nuclear Black™ DCS2 stock solution can be aliquoted and stored at ≤ -20 °C for several months if the tubes are sealed tightly. Protect from light and avoid repeated freeze-thaw cycles.

SAMPLE EXPERIMENTAL PROTOCOL

The following protocol can be used as a guideline. You can use a live-cell stain that stains the nuclei of dead cells as an alternative to a live-cell nuclear stain.

1. Prepare the cell samples and treat cells as desired.
2. Optional: Remove the cell culture medium and wash cells with DPBS or buffer of your choice.
3. Treat cells with live-cell nuclear stain such as Nuclear Red™ LCS1 (AAT Cat# 17542).
4. Add 100 µL/well (96-well plate) or 50 µL/well (384-well plate) of Nuclear Black™ DCS2 working solution and incubate for 30 minutes at 37°C in a 5% CO2 incubator, protected from light.
   Note: The optimal concentration of the Nuclear Black™ DCS2 varies depending on the specific applications. Concentration higher than the working solution can be toxic to cells. The staining conditions may be modified according to the particular cell type and the permeability of the cells or tissues to the probe.
5. Observe the fluorescence signal in cells using a fluorescence microscope with a Cy5 filter set.

EXAMPLE DATA ANALYSIS AND FIGURES

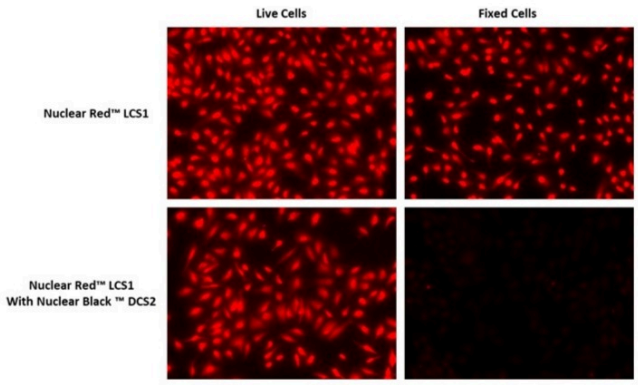

Figure 1. Fluorescence image of live and fixed HeLa cells stained with Nuclear Red™ LCS1 (Cat. #17542) only and stained with Nuclear Red™ LCS1 (Cat. #17542) and Nuclear Black™ DCS2 (Cat. #17692). Nuclear Black™ DCS2 quenches the staining of Nuclear Red™ LCS1 in fixed, but not the live cells.

DISCLAIMER

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