Nucleolus Red™ DCS1 *Dead Cell Staining*

Catalog number: 17684
Unit size: 100 Tests

Component	Storage	Amount (Cat No. 17684)
Nucleolus Red™ DCS1	Freeze (< -15 °C), Minimize light exposure	100 Tests

OVERVIEW

Nucleolus Red™ DCS1 is a cell-impermeant fluorescent dye that selectively binds to RNA within the nucleolus, a non-membrane-bound nuclear compartment responsible for ribosomal RNA (rRNA) transcription and processing. Upon binding to RNA, the dye exhibits strong red fluorescence, whereas its interaction with DNA results in only weak fluorescence, providing exceptional specificity for nucleolar RNA detection.

This dye is particularly useful for studying nucleolar morphology and function in dead or permeabilized cells. It enables clear visualization of nucleoli and can be used in conjunction with nuclear counterstains such as DAPI or Nuclear Green™ DCS1 to provide additional structural context. The nucleolus plays a critical role in cellular processes, including ribosome biogenesis, DNA damage response, autophagy, viral pathogenesis, and cellular senescence. Morphological changes in the nucleolus have also been implicated as potential biomarkers for cancer diagnostics. Nucleolus Red™ DCS1 is compatible with fluorescence microscopy, flow cytometry, and high-content imaging assays, making it a valuable tool for researchers investigating nucleolar dynamics and cellular homeostasis in various biological and disease contexts.

AT A GLANCE

Protocol Summary

1. Prepare cells in a growth medium.
2. Incubate the cells with the Nucleolus Red™ DCS1 working solution at 37°C for 15–30 minutes.
3. Observe under a fluorescence microscope using a Cy5 filter set.

KEY PARAMETERS

Fluorescence microscope

Emission	Cy5 Filter Set
Excitation	Cy5 Filter Set
Recommended plate	Black wall/clear bottom

CELL PREPARATION

For Adherent Cells

1. Plate cells overnight in growth medium at 10,000–40,000 cells per well in 90 µL for a 96-well plate or 2,500–10,000 cells per well in 20 µL for a 384-well plate.

For Non-Adherent Cells

1. Centrifuge cells from the culture medium to pellet, then resuspend in fresh culture medium.
2. Seed 50,000–100,000 cells/well in 90 µL for a 96-well poly-D-lysine plate or 10,000–25,000 cells/well in 20 µL for a 384-well poly-Dlysine plate.
3. Before the experiment, centrifuge the plate at 800 rpm for 2 minutes with the brake off.

   Note: The optimal cell density should be determined individually for each cell line.

PREPARATION OF WORKING SOLUTION

Nucleolus Red™ DCS1 Working Solution

1. Add 10 µL of Nucleolus Red™ DCS1 stock solution to 1 mL of HHBS buffer (AAT Bio Cat no. 20011) and mix thoroughly. The working solution remains stable at room temperature for up to 2 hours.

   Note: 100 µL of Nucleolus Red™ DCS1 stock solution is sufficient for one 96-well plate. Any unused stock solution can be aliquoted and stored at ≤ -20°C for several months, provided the tubes are tightly sealed and protected from light. Avoid repeated freeze-thaw cycles to maintain stability.

SAMPLE EXPERIMENTAL PROTOCOL

1. Treat the samples as needed. Then, remove the cell culture medium and wash the cells with DPBS or a buffer of your choice.
2. Fix the cells by treating them with 4% formaldehyde at room temperature for 20 minutes.
3. Discard the formaldehyde and wash the cells twice with DPBS or a buffer of your choice.
4. Add 100 µL per well for a 96-well plate or 50 µL per well for a 384-well plate of Nucleolus Red™ DCS1 working solution to the cell plate. Incubate at 37°C for 15–30 minutes, keeping the plate protected from light.

   Note: The optimal concentration of Nucleolus Red™ DCS1 depends on the specific application. Using a concentration higher than the recommended working solution may be toxic to cells. Staining conditions can be adjusted based on the cell type and its permeability to the probe.

5. Remove the working solution from each well. Wash the cells three times with DPBS or a buffer of your choice. Then, add 100 µL per well for a 96-well plate or 50 µL per well for a 384-well plate to cover the cells.
6. Use a fluorescence microscope with a Cy5 filter set to observe the fluorescence signal in the cells.

EXAMPLE DATA ANALYSIS AND FIGURES

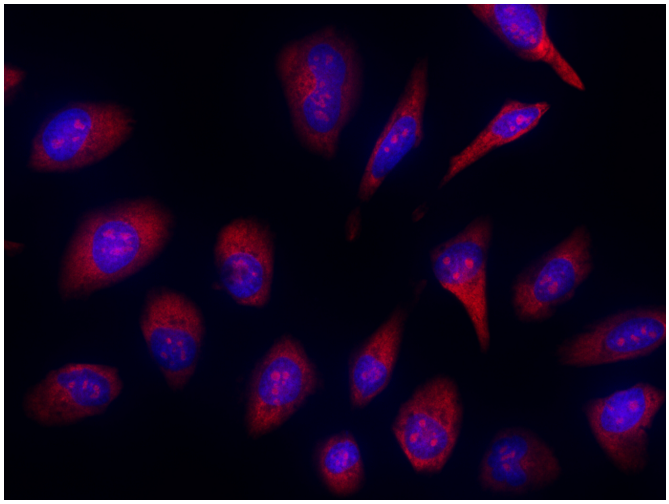

Figure 1. Fluorescence image of Fixed HeLa cells stained with Nucleolus Red™ DCS1 (Cat# 17682) and Nuclear Blue™ DCS1 (Cat# 17548)

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