Cell Navigator® Fluorimetric Lipid Droplet Assay Kit *Blue Fluorescence*

Catalog number: 22731
Unit size: 200 Tests

Component	Storage	Amount (Cat No. 22731)
Component A: Droplite™ Blue	Freeze (< -15 °C), Minimize light exposure	1 Vial
Component B: Staining Buffer	Freeze (< -15 °C), Minimize light exposure	1 Bottle (20 mL)

OVERVIEW

Droplite™ Blue is a lipophilic stain that can specifically stain lipid droplets/adiposomes present in the cells.

Lipid droplets, also known as lipid bodies or adiposomes, are essential cellular organelles responsible for the storage and hydrolysis of neutral lipids. These lipid-rich structures serve as key reservoirs for fatty acids and cholesterol and play a crucial role in biological processes such as energy production, cellular signaling, and membrane maintenance. Impaired cytoplasmic lipid droplet accumulation is associated with various diseases and pathological conditions including metabolic disorders, liver dysfunction, and cardiovascular diseases.

AAT Bioquest's Cell Navigator® Fluorimetric Lipid Droplet Assay Kit provides a powerful and reliable method for accurately studying lipid droplet accumulation within the cells. This kit features Droplite™ Blue, a lipophilic stain that exhibits bright blue fluorescence in a lipid-rich environment while giving minimal fluorescence in aqueous media. Droplite™ Blue is an ideal vital stain for visualizing intracellular lipid droplets using fluorescence microscopy, flow cytometry, or fluorescence microplate readers. The blue fluorescence can be easily detected using standard DAPI filter sets, offering a convenient and efficient tool for lipid droplet analysis.

AT A GLANCE

Protocol Summary

1. Prepare cells with test compounds.
2. Add Droplite™ Blue working solution.
3. Incubate at room temperature or 37°C for 10 to 30 minutes.
4. Read fluorescence intensity with a fluorescence microscope using a DAPI filter set.

Important Note

This protocol is our recommended guideline for live cells, but it can be adjusted to meet your specific requirements. Since Droplite™ Blue exhibits minimal fluorescence in aqueous media, removing the growth medium and staining solution after staining is optional. Stained cells can be fixed with 3–4% formaldehyde. Alternatively, prefixed cells (fixed with 3–4% formaldehyde) can also be stained using the Droplite™ Blue staining solution.

KEY PARAMETERS

Fluorescence microplate reader

Cutoff	420 nm
Emission	450 nm
Excitation	350 nm
Recommended plate	Black wall/clear bottom
Instrument specification(s)	Bottom read mode

Fluorescence microscope

Emission	DAPI filter set
Excitation	DAPI filter set
Recommended plate	Black wall/clear bottom

CELL PREPARATION

For guidelines on cell sample preparation, please visit:

https://www.aatbio.com/resources/guides/cell-sample-preparation.html

PREPARATION OF WORKING SOLUTION

1. To prepare the Droplite™ Blue working solution, dilute 5 µL of the Droplite™ Blue (Component A) in 1 mL of Staining Buffer (Component B).

   Note: 50 µL of Droplite™ Blue (Component A) is enough for one 96-well plate. Protect the solution from light. The optimal concentration of Droplite™ Blue may vary depending on the application. Adjust staining conditions based on the cell type and the permeability of cells or tissues to the probe.

SAMPLE EXPERIMENTAL PROTOCOL

For adherent cells:

1. Grow cells either in a 96-well black wall/clear bottom plate (100 µL/well/96-well) or on cover-slips inside a petri dish filled with the appropriate culture medium.
2. Gently aspirate the culture medium and add equal volume (such as 100 µL/well/96-well plate) of the Droplite™ Blue staining solution.
3. Incubate the cells in a 37°C, 5% CO_2 incubator for 10 - 30 minutes.
4. Remove Droplite™ Blue working solution (Optional).
5. Measure fluorescence at Ex/Em = 350/450 nm with a microplate reader or observe the cells using a fluorescence microscope equipped with a DAPI filter set.

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For suspension cells:

1. Centrifuge the cells at 1000 rpm for 5 minutes to get 1 - 5 × 10^5 cells per tube.
2. Resuspend cells in 500 µL of Droplite™ Blue working solution.
3. Incubate at room temperature or 37°C for 10 to 30 min, protected from light.
4. Centrifuge to remove the Droplite™ Blue working solution, and resuspend cells in 500 µL of pre-warmed medium or buffer of your choice to get 1 - 5 × 10^5 cells per tube (Optional).
5. Monitor the fluorescence increase using a fluorescence microscope equipped with a DAPI filter set.

EXAMPLE DATA ANALYSIS AND FIGURES

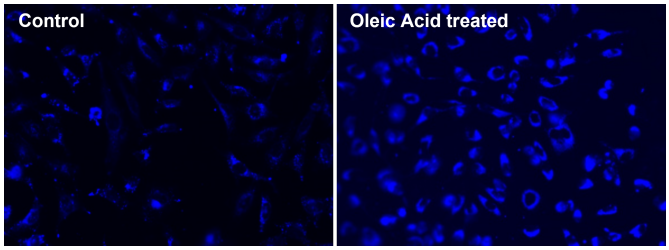

Figure 1. Fluorescence images of intracellular lipid droplets in control (Left) and Oleic Acid treated HeLa cells (Right) using Cell Navigator® Lipid Droplets Fluorescence Assay Kit. HeLa cells were incubated with 300 uM of Oleic Acid for 24 hours to induce intracellular lipid droplets formation. After washing with PBS, the cells were labeled with 1X Droplite™ Blue. Images were acquired with fluorescence microscope using DAPI filter set.

DISCLAIMER

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