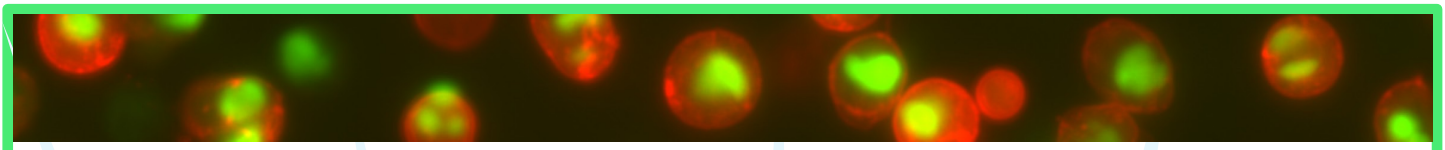

NucView™488 Caspase-3 Detection in Living Cells

NucView™ 488 caspase-3 substrate is the first of a series of novel substrates developed by Biotium to follow caspase activity in intact cells in real-time. We have invented a new fluorogenic enzyme substrate design by attaching an enzyme substrate moiety to a nucleic acid dye. In the case of DEVD-NucView 488 caspase-3 substrate, the NucView™488 DNA dye is attached to the caspase-3 substrate peptide sequence DEVD. Once linked to the substrate peptide, the dye is unable to bind to DNA and remains nonfluorescent. The substrate crosses the plasma membrane to enter the cytoplasm, where it can be cleaved by caspase-3 to release the high-affinity DNA dye, which migrates to the cell nucleus to stain the nucleus with bright green fluorescence (Figure 1). Therefore, the substrate allows both detection of caspase-3 and visualization of nuclear morphology in apoptotic cells (Figure 2) .

Unlike fluorescently-labeled caspase inhibitor assays (FLICA) that use irreversible caspase inhibitors to label active casapses, NucView™488 Caspase-3 Substrate does not interfere with caspase activity, allowing monitoring of caspase activity in real time (Figure 3).

Biotium offers assay kits featuring NucView488 for the measurement of caspase-3 activity in combination with phosphatidylserine translocation (see Figure 4, other side) or mitochondrial membrane potential (see Figure 5, other side) by flow cytometry, microscopy, or fluorescence plate reader.

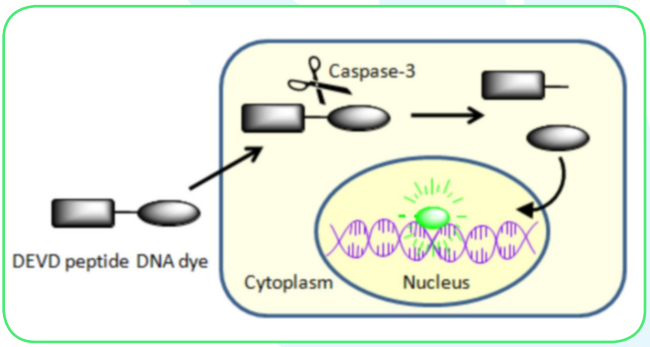

Figure 1. Schematic showing the principle of intracellular caspase-3 detection using NucView™ 488 Caspase-3 Substrate.

Key Features:

- Caspase-3 dependent staining of nuclear DNA in intact cells, allowing real-time monitoring of caspase-3 activity
- For use in adherent or suspension cells
- Rapid, direct staining in cell culture medium with no washing required
- Green fluorescence can be measured by flow cytometry, microscopy or fluorescence plate reader
- Staining is formaldehyde-fixable

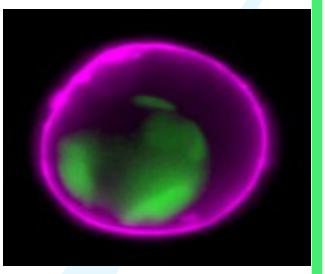

Figure 2. Apoptotic Jurkat cell stained with NucView™ 488 (green) and CF647-Annexin V (magenta). NucView™488 nuclear staining indicates caspase-3 activity and reveals the fragmented nuclear morphology characteristic of apoptotic cells. Annexin staining reveals phosphatidylserine translocation to the extracellular leaflet of the plasma membrane, another marker of apoptosis.

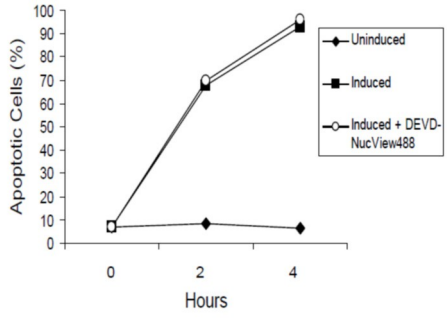

Figure 3. NucView™ 488 Caspase-3 Substrate detection of apoptosis in staurosporine-treated Jurkat cells. The substrate does not interfere wtih apoptosis progression, allowing monitoring of caspase-3 activity in real time. Jurkat cells induced to undergo apoptosis with staurosporine. From: Cen et al. FASEB J. 22, 2243–2252. (2008).

See other side for NucView™ kits and ordering information

Biotium
www.biotium.com

NucView™488 Caspase-3 Assay Kits

NucView™ 488 Caspase-3 Assay Kit for live cells contains NucView™ 488 Caspase-3 substrate in DMSO and caspase-3 inhibitor Ac-DEVD-CHO.

NucView™ 488 Caspase-3 substrate and sulforhodamine 101-annexin V kit includes deep red fluorescent sulforhodamine 101 (Texas Red®-annexin V for dual detection of caspase-3 activity and phosphotadylserine translocation in intact cells (Figure 4).

The NucView™ 488 and MitoView™ 633 Apoptosis Kit includes far red fluorescent MitoView™ 633 mitochondrial membrane potential dye for simultaneous measurement of caspase-3 activity and mitochondrial membrane potential (Figure 5).

NucView™ 488 Caspase-3 substrate is also offered as a 1 mM stock solution in DMSO or PBS. DMSO facilitates NucView™ 488 Caspase-3 staining in some cell types. The substrate is offered in PBS for use in DMSO-sensitive cell types.

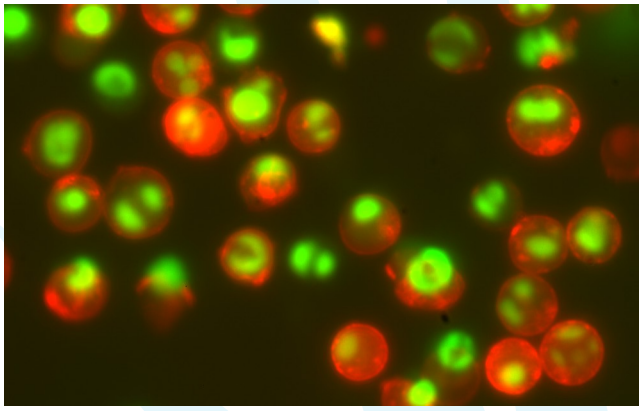

Figure 4. Staurosporine-induced apoptotic Jurkat cells stained with NucView™ 488 Caspase-3 Substrate and Sulforhodamine 101 (Texas Red®) Annexin V Dual Apoptosis Assay Kit. Caspase-3 activity is indicated by green nuclear staining, while exposed phosphatidylserine on the plasma membrane surface is stained red by Texas Red® Annexin V.

Ordering Information

Catalog number	Product description
30029	NucView™ 488 Caspase-3 Assay Kit for Live Cells
30030	NucView™ 488 Caspase-3 Substrate and Sulforhodamine 101 (Texas Red®) Annexin V Dual Apoptosis Assay Kit
30062	NucView™ 488 and MitoView 633 Apoptosis Kit
10403	NucView™ 488 Caspase-3 Enzyme Substrate 1mM in PBS
10402	NucView™ 488 Caspase-3 Enzyme Substrate 1mM in DMSO

For current pricing, please visit www.biotium.com.

Related Products

Biotium offers a variety of apoptosis research tools, including mitochondrial membrane potential dyes, CF™dye-Annexin V conjugates, direct fluorescence TUNEL labeling kits featuring CF™dye dUTP conjugates, cell viability and proliferation assays, and live/dead cell quantitation kits. For more information, please visit our website at www.biotium.com.

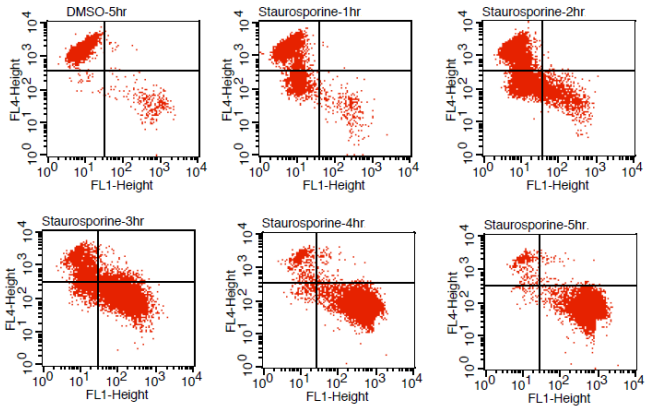

Figure 5. Flow cytometry analysis of caspase-3 acitvity and mitochondrial membrane potential in staurosporine-treated Jurkat cells using NucView™ 488 and MitoView™ 633 Apoptosis Kit. NucView™499 staining is plotted on FL1 (x-axis) and MitoView™633 staining is plotted on FL4 (y-axis). As apoptosis progresses, NucView™488 signal increases while mitochondrial membrane potential measured by MitoView™633 staining decreases.

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