Technical Brief

Catalogue No. MCH100109

Muse™ MultiCaspase Assay

Rapid Detection of Cellular Caspase Activity

Assay Features

- Detection of pan-caspase activity in individual cells
- Quick determination of live, caspase-active, caspase-inactive/dead cells and dead cells
- No-wash, mix-and-read format, rapid assay
- Simplified acquisition and analysis
- Minimal number of cells required
- Validated with both adherent and suspension cells
- Accurate and precise

Rapid, Sensitive Detection of Caspase Activity

Caspases (cysteinyl-directed aspartate-specific proteases) propagate programmed cell death (apoptosis) in response to proapoptotic signals. Several caspases also have roles in inflammation, mediating immunity, cell fate specification, cell survival, cell cycle regulation, cell proliferation, and cell migration. While some caspases act to initiate intracellular signaling, effector ("executioner") caspases, such as caspase-3/7, act further downstream and direct cellular breakdown through cleavage of structural proteins.

Assay Principle

The Muse™ MultiCaspase Assay detects the activity of caspases 1, 3, 4, 5, 6, 7, 8 and 9. The assay simultaneously determines the percentage and concentration of cells with caspase activity, in combination with a dead cell dye. Included in the kit are:

(1) A fluorogenic, derivatized VAD-peptide that can detect the activity of multiple caspases

(2) A cell death impermeant DNA binding dye, 7-AAD that provides information on membrane integrity.

The VAD-peptide is derivatized with a fluorescent group and a fluoromethylketone irreversible caspase inhibitor moiety, generating a Fluorescent-Labeled Inhibitor of Caspases (FLICA). The peptide is membrane-permeable and non-cytotoxic. It binds to activated caspases with resulting fluorescent signal proportional to the number of active caspases in the cell which increases signal in the caspase activity axis.

The dead cell marker, 7-AAD, is excluded from live (healthy) and caspase positive cells, but enters and stains membrane compromised later stage apoptotic and dead cells that show increased fluorescence in the viability axis.

Four populations of cells can be distinguished in the assay:

1. Live cells: caspase-3/7(-) and 7-AAD(-)
2. Pan-caspase-active cells: caspase(+) and 7-AAD(-)
3. Late caspase active/Dead cells: caspase(+) and 7-AAD(+)
4. Dead cells: caspase(-) and 7-AAD(+)

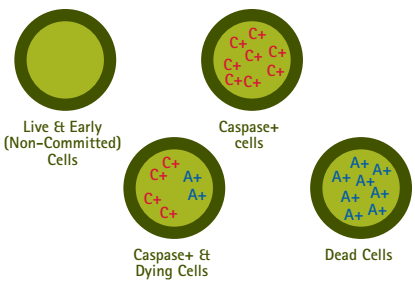

Figure 1.

Principle of the Muse™ MultiCaspase Assay. C represents Caspase peptide substrate while A is the dead cell dye.

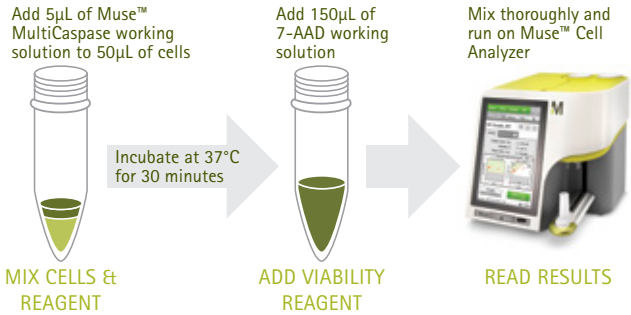

Figure 2.

The Muse™ MultiCaspase Assay uses a simple mix-and-read protocol, enabling easy determination of cells exhibiting caspase activity.

Touchscreen Interface Greatly Simplifies Acquisition and Analysis of Caspase Activity Data

Muse™ MultiCaspase software module guides the user through set-up, acquisition and analysis in a few simple steps.

- Intuitive touchscreen which guides users quickly to results.
- Results include count and percentage of populations automatically displayed after acquisition.
- Easy export of raw data to Excel® format enable archiving of results and additional analysis.

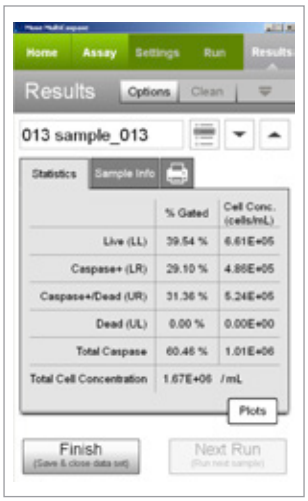

Figure 3. Results obtained using Jurkat cells induced to apoptosis with 1 µM staurosporine, stained with Muse™ MultiCaspase Assay and data acquired on the Muse™ Cell Analyzer.

Versatile and Accurate

The Muse™ MultiCaspase assay is versatile and works with both adherent and suspension cells, providing information on the relative percentages of live, caspase+, late caspase+ and dead cells. Figure 5 demonstrates that the Muse™ MultiCaspase Assay provides accurate percentages of caspase-active cell populations compared to data obtained using comparative platforms.

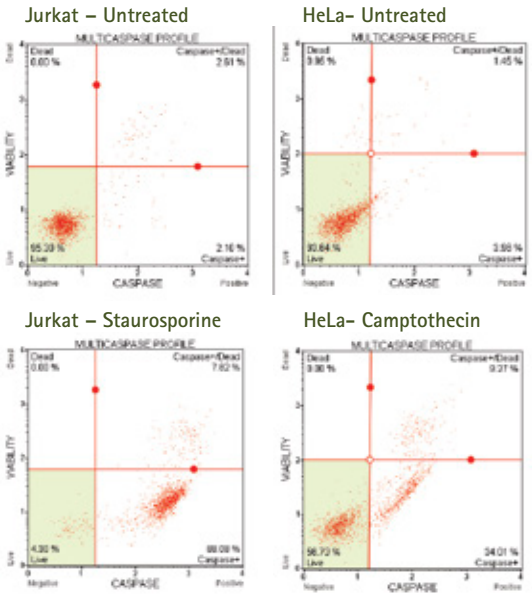

Figure 4. Impact of apoptosis-inducing compounds on HeLa cells and Jurkat cells analyzed using the Muse™ MultiCaspase Assay.

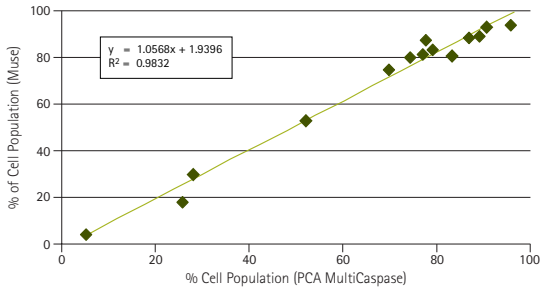

Figure 5. Correlation of caspase-active cell percentages determined using the Muse™ Cell Analyzer with percentages obtained from other flow cytometry platform (guava® PCA) (x-axis). Jurkat cells and HeLa cells were treated with apoptosis inducers and analyzed using the Muse™ MultiCaspase Assay.

Ordering Information

Muse™ Caspase-3/7 Assay	MCH100108	Muse™ Cell Cycle Kit	MCH100106
Muse™ MultiCaspase Assay	MCH100109	Muse™ Count & Viability Kit	MCH100102
Muse™ MitoPotential Assay	MCH100110	Muse™ Annexin V & Dead Cell Kit	MCH100105
Muse™ System Check Kit	MCH100101		

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