



THE POWER OF

GUAVA® EASYCYTE™ CASPASE 3/7 & CASPASE 8 ASSAY KITS:

For rapid, sensitive detection of caspase 3, 7 or 8 enzyme activity in live cells

Guava Caspase Assays

- Specific detection of active Caspase 3/7 or Caspase 8 molecules on a per cell basis
- Optimized for 96-well plate formats
- Works with both adherent and suspension cells
- FLICA reagents are non-cytotoxic and cell permeant, thus eliminating cell lysis and permeabilization procedures
- Staining and data acquisition completed in ≤ 3 hours.

Now you can easily discriminate between apoptotic and non-apoptotic cells based on the simultaneous interrogation of caspase enzyme activity and cell membrane permeability within a single assay. Together with the Guava EasyCyte™ instrument platform, Guava Technologies' new fluorescent-based assays for Caspase 3/7 and Caspase 8 activity offer a powerful single cell analysis approach to assessing apoptotic status in a convenient microplate format at your benchtop. The Guava EasyCyte system's integrated ExpressPlus™ software automates data capture and analysis, providing both a live and dead cell count, plus the relative frequency of cells in the various stages of apoptosis.

Guava Caspase Assays employ a FLICA (Fluorescent Labeled Inhibitor of Caspases) reagent that specifically identifies active Caspase 3 and 7 molecules or Caspase 8 molecules. Each FLICA is comprised of three subunits: a caspase-specific amino acid inhibitor containing a recognition sequence for Caspase 3/7 (DEVD) or Caspase 8 (LETD); a fluoromethylketone moiety (FMK) that forms a covalent linkage with the active enzyme; and a carboxyfluorescein (FAM) reporter. The nuclear DNA stain, propidium iodide (PI), is also included in the assay to simultaneously evaluate membrane integrity and cell viability.

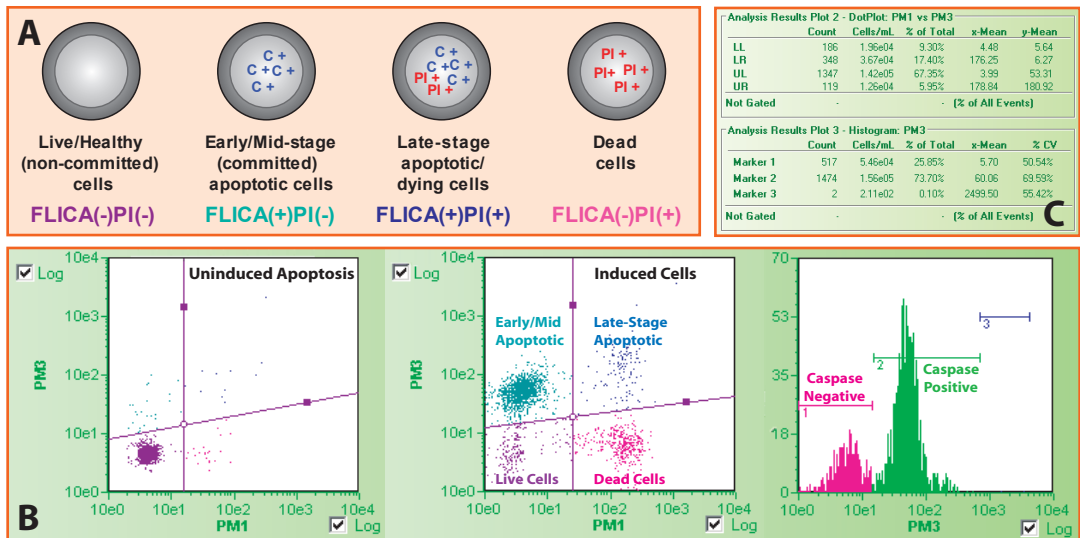


FIGURE 1: Up to four subpopulations of cells can be detected with the Caspase Assay (Fig. 1A). The Guava EasyCyte software permits the visualization and analysis of each of these populations in various formats including dot plots with both gating capability and flexible quadrant markers and a three marker histogram (Fig. 1B) and statistical tables (Fig. 1C). Data can also be exported for printing and further analysis.

SUPERIOR ASSAY PERFORMANCE

Guava Caspase Assays are ideally suited for applications in drug discovery and development (Fig. 2). They are sensitive enough to detect even naturally occurring apoptosis at 2 to 6% of the overall population, while still delivering robust (Fig. 3), reproducible (Fig. 4), and specific detection of Caspase 3/7 or 8 enzyme activity (Fig. 5).

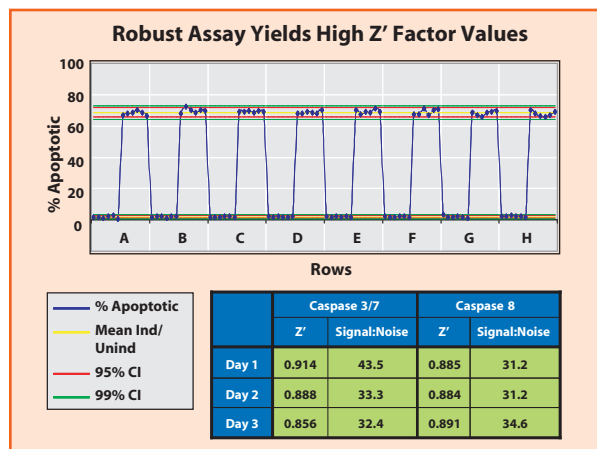


FIGURE 3: Each point on the graph represents the mean value for the percent of apoptotic cells (FLICA+) identified in each well of a 96-well microplate. Jurkat cells were induced with 1 μ M staurosporine for 3 hours prior to performing Caspase assay. Values for induced and uninduced cells remained within the 95% confidence interval. Assay stability and robustness (signal-to-noise ratios > 20:1) over three separate days are shown in the table.

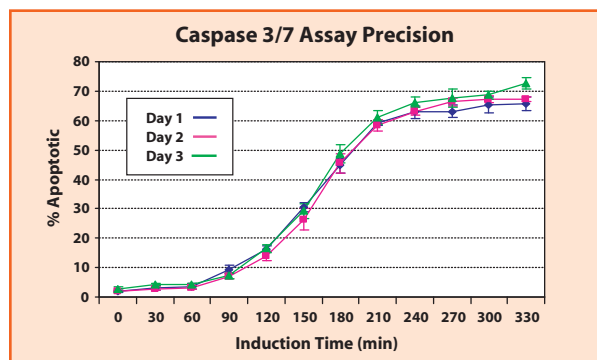


FIGURE 4: Time course results demonstrate the reproducibility of the Caspase 3/7 Assay. Jurkat cells were treated with 1 μ M staurosporine. Similar results were obtained with Caspase 8 Assay (not shown).

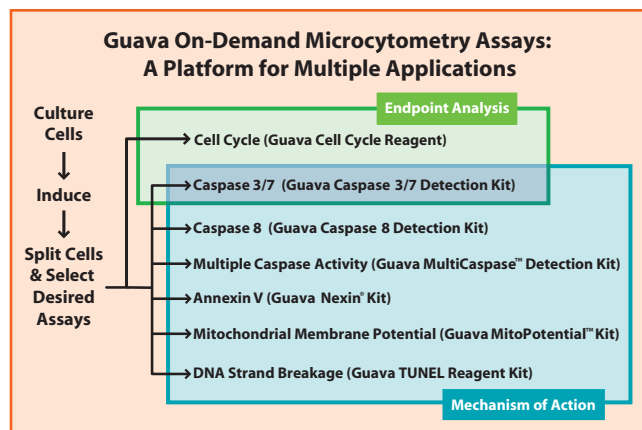


FIGURE 2: Caspase Assays can also be used in conjunction with any Guava apoptosis kit, as well as with the Guava Cell Cycle Assay, for efficacy screening, endpoint analysis, or understanding the mechanism of action.

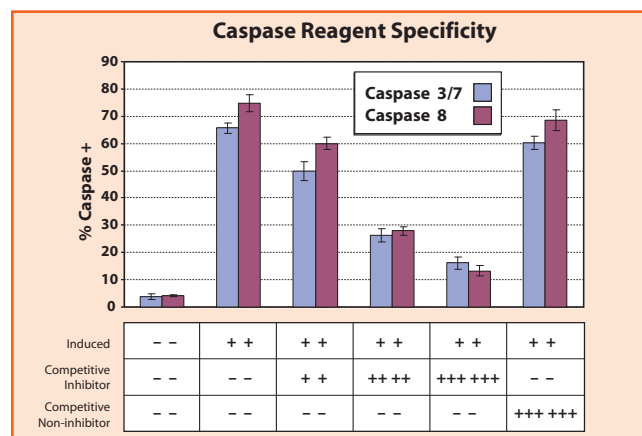


FIGURE 5: Jurkat cells were treated with 1 μ M staurosporine for 3 hours (induced) or DMSO for non-induced. Caspase assays were then performed in the presence of increasing concentrations of competitive inhibitor: + = 10 μ M, ++ = 50 μ M, +++ = 100 μ M. For Caspase 3/7, competitive inhibitor = zDEVD-FMK; Caspase 8, competitive inhibitor = zLETD-FMK. Binding specificity of each Caspase reagent was further confirmed through cross-competition with a non-competitive inhibitor. For Caspase 3/7, non-competitive inhibitor = zLETD-FMK; Caspase 8, non-competitive inhibitor = zDEVD-FMK.

ORDERING INFORMATION

Guava EasyCyte Caspase 3/7 Kit (100 test kit) 4500-0290

Guava EasyCyte Caspase 8 Kit (100 test kit) 4500-0300

For pricing, please request a quotation.



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