Z-VAD-FMK

Catalog number: 13300
Unit size: 1 mg

Component	Storage	Amount
Z-VAD-FMK	Freeze (< -15 °C), Minimize light exposure	1 mg

OVERVIEW

Z-VAD-FMK (carbobenzoxy-valyl-alanyl-aspartyl-[O-methyl]- fluoromethylketone) is a cell-permeable, irreversible pan caspase inhibitor that binds to the catalytic site of caspase proteases and can inhibit induction of apoptosis. For inhibition of apoptosis, Z-VAD-FMK should be added at the same time that apoptosis is induced. The peptide is O-methylated in the P1 position on aspartic acid, providing enhanced stability and increased cell permeability. Fluoromethyl ketone (FMK)-derivatized peptides act as effective irreversible inhibitors with no added cytotoxic effects.

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles.

Z-VAD-FMK stock solution
Add appropriate amount of DMSO into Z-VAD-FMK to make 2-5 mM Z-VAD-FMK stock solution.

Note Store the unused Z-VAD-FMK stock solution at -20 °C in single use aliquots.

SAMPLE EXPERIMENTAL PROTOCOL

The following protocol can be used as a guideline and should be optimized according to your needs.

1. Treat your samples as desired to induce apoptosis.
2. Add Z-VAD-FMK along with the treatment of your choice.

   Note We recommend using 10-100 µM of final concentration ofZ-VAD-FMK to inhibit caspases such as 50 µM for caspase-8.

   Note Optimal time and concentration for incubation needs to be determined experimentally.
3. Stain samples as you desired.
4. Monitor the fluorescence intensity with a proper instrument.

EXAMPLE DATA ANALYSIS AND FIGURES

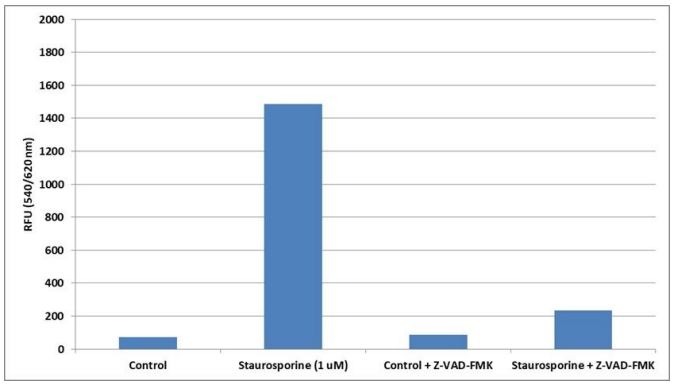

Figure 1. Inhibition of caspase activities in Jurkat cells with pan caspase inhibitor Z-VAD-FMK (Cat.# 13300). Jurkat cells were seeded on the same day at 200,000 cells/90 µL/well in a Costar black wall/clear bottom 96-well plate. The cells were treated with or without 1 µM of staurosporine for 5 hours. The Z-VAD-FMK (50 uM) was added and co-incubated with staurosporine. The caspase-8 activity was detected with Cell Meter™ Caspase 8 Activity Apoptosis Assay Kit (Cat.# 22816). The fluorescence intensity was measured at Ex/Em = 540/620 nm (Cutoff = 610 nm) with Flexstation 3 fluorescence microplate reader (Molecular Devices).

DISCLAIMER

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