rev. 10/16 For research use only

# Caspase-2 Fluorometric Substrate, VDVAD-AFC

**CATALOG #**: 1071-200 200 assays (1 x 1 ml)

1071-1000 1000 assays (5 x 1 ml)

**STORAGE:** Store at -20° C, protected from light. Stable for 6 months

MOL. WEIGHT: 770.7

**SEQUENCE:** Ac-Val-Asp-Val-Ala-Asp-AFC

(AFC, 7-amino-4-trifluoromethyl coumarin)

**PURITY:** >98% by HPLC analysis

### DESCRIPTION:

Ready-to-use fluorometric substrate for caspases that recognize the amino acid sequence VDVAD. Caspase activity can be quantified by fluorescent detection of free AFC after cleaved from the peptide substrate VDVAD-AFC at Ex/Em = 400/505 nm using a fluorometer or a fluorescence plate reader. Alternatively, a shift in fluorescence from blue to green upon cleavage can be visualized using a hand-held long-UV lamp. The ready-to-use caspase substrate provides an economic alternative for researchers who perform large amount of caspase assays. Cell Lysis Buffer (Cat.# 1067-100), 2X Reaction Buffer (Cat.# 1068-20), DTT (Cat.# 1201-1), and other reagents used for caspase activity assays are also available separately.

#### ASSAY PROTOCOL:

- Induce apoptosis in cells by desired method. Concurrently incubate a control culture without induction.
- 2. Count cells and pellet 1-5 x  $10^6$  cells or use 50-200  $\mu g$  cell lysates if protein concentration has been measured.
- 3. Resuspend cells in 50 µl of chilled Cell Lysis Buffer (Cat.# 1067-100).
- 4. Incubate cells on ice for 10 minutes.
- 5. Add 50 µl of 2X Reaction Buffer (Cat.# 1068-20, -80) containing 10 mM DTT (Cat.# 1201-1) to each sample.
- Add 5 μl of the 1 mM VDVAD-AFC (50 μM final conc.) into each tube individually and incubate at 37° C for 1-2 hour.
- 7. Read samples in a fluorometer equipped with a 400-nm excitation filter and 505-nm emission filter. For a plate-reading set-up, transfer the samples to a 96-well plate. You may perform the entire assay directly in a 96-well plate.

Fold-increase in VDVAD-dependent caspase activity can be determined by comparing these results with the level of the uninduced control.

## **RELATED PRODUCTS:**

Apoptosis Detection Kits & Reagents

- Annexin V Kits & Bulk Reagents
- Caspase Assay Kits & Reagents
- Mitochondrial Apoptosis Kits & Reagents
- Nuclear Apoptosis Kits & Reagents
- · Apoptosis Inducers and Set
- Apoptosis siRNA Vectors

# Cell Fractionation System

- Mitochondria/Cytosol Fractionation Kit
- Nuclear/Cytosol Fractionation Kit
- Membrane Protein Extraction Kit
- Cytosol/Particulate Rapid Separation Kit
- Mammalian Cell Extraction Kit
- FractionPREP Fractionation System

#### Cell Proliferation & Senescence

- Quick Cell Proliferation Assay Kit
- Senescence Detection Kit
- High Throughput Apoptosis/Cell Viability Assay Kits
- LDH-Cytotoxicity Assay Kit
- Bioluminescence Cytotoxicity Assay Kit
- Live/Dead Cell Staining Kit

# Cell Damage & Repair

- HDAC Fluorometric & Colorimetric Assays & Drug Discovery Kits
- HAT Colorimetric Assay Kit & Reagents
- DNA Damage Quantification Kit
- Glutathione & Nitric Oxide Fluorometric & Colorimetric Assay Kits

#### Signal Transduction

- cAMP & cGMP Assay Kits
- Akt & JNK Activity Assay Kits
- Beta-Secretase Activity Assay Kit

#### Adipocyte & Lipid Transfer

- Recombinant Adiponectin, Survivin, & Leptin
- CETP Activity Assay & Drug Discovery Kits
- PLTP Activity Assay & Drug Discovery Kits
- Total Cholesterol Quantification Kit

# Molecular Biology & Reporter Assays

- siRNA Vectors
- Cloning Insert Quick Screening Kit
- Mitochondrial & Genomic DNA Isolation Kits
- 5 Minutes DNA Ligation Kit
- 20 Minutes Gel Staining/Destaining Kit
- β -Galactosidase Staining Kit & Luciferase Reporter Assay Kit

Growth Factors and Cytokines

Monoclonal and Polyclonal Antibodies