



## PRODUCT DATA SHEET

**Product:** Ac-VDVAD-pNA (Chromogenic caspase-2, 3 substrate)

**Cat. No.:** AC-027 (25 mg)

**Chemical Name:**

Acetyl-Val-Asp-Val-Ala-Asp-pNA

**Molecular Weight:**

679

**Purity:**

>97% by HPLC

**Description:**

White lyophilized powder. Chromogenic paranitroanilide-peptide substrate for caspase-2 (ICH-1). Release of free pNA is monitored by absorbance at 405 nm ( $\epsilon=9,160 \text{ M}^{-1}\text{cm}^{-1}$ ).

**Introduction:**

Caspase-2 (also known as ICH-1) is a member of the caspase family of cysteine proteases involved in apoptosis. It is a member of the Group II caspases, along with caspases-3 and -7. Group II caspases prefer peptides of the DEXD-type as substrates. However, unlike caspases-3 and -7, Caspase-2 requires a P5 amino acid in the peptide for efficient cleavage. The similar substrate specificities of the Group II caspases suggests that their roles in cells are at least overlapping, if not completely redundant. The requirement for a fifth amino acid in substrates for Caspase-2 means that the DEVD-type, while serving as substrates for caspases-3 and -7, do not work with Caspase-2. For this reason, the Ac-VDVAD-pNA substrate is excellent for studying Caspase-2.

**Specificity:**

Substrate for caspase-2, 3.

**Applications:**

Assay of caspase activity in cell extracts.

**Protocol:**

Soluble in DMSO and aqueous buffers. We recommend preparing a stock solution in DMSO, and diluting into aqueous buffer shortly prior to use.

Suggested procedure only. Each laboratory must determine optimum conditions.

1. Lyse cells in 50 mM Tris-HCl, pH 7.5, 0.3% NP-40, 1.0 mM DTT, at a density of  $2 \times 10^6/\text{mL}$ .

2. Assay 0.01 mL cell lysate in a final volume of 0.1 mL. Assay buffer is cell lysis buffer containing 0.2 mM substrate.

3. Incubate at 37°C for 0-3 hr. Take periodic readings of absorbance at 405 nm.

**Storage and Stability:**

Solid can be stored at room temperature. However, for long term storage, desiccated at 4°C is recommended. Protect from light and moisture. Store stock solutions in DMSO refrigerated or frozen. Stable indefinitely protected from light and moisture. Stock solutions in DMSO can be stored for long periods refrigerated or frozen. Solutions in aqueous buffers should be stored for only short periods of time. Hydrolysis of the substrate will be revealed by the appearance of a yellow color.

**References:**

1. Duan, H. et al. (1996). *J. Biol. Chem.* 271: 16720-24.
2. Faucheu, C. et al. (1995). *EMBO J.* 14: 1914-22.
3. Kamens, J. et al. (1995). *J. Biol. Chem.* 270: 15250-56.
4. Munday, N.A. et al. (1995). *J. Biol. Chem.* 270: 15870-76.
5. Srinivasula, S.M. et al. (1996). *J. Biol. Chem.* 271: 27099-106.
6. Talanian, R.V. et al. (1997). *J. Biol. Chem.* 272: 9677-82.
7. Thornberry, N. et al. (1997). *J. Biol. Chem.* 272(29): 17907-911.

**Limitations:**

For *in vitro* research use only. Not for use in diagnostics or in humans.

**Warranty:**

No warranties, expressed or implied, are made regarding the use of this product. KAMIYA BIOMEDICAL COMPANY is not liable for any damage, personal injury, or economic loss caused by this product.