Recombinant Human DPPIV/CD26

Catalog Number: 1180-SE

DESCRIPTION

Source	Mouse myeloma cell line, NS0-derived Asn29-Pro766, with a C-terminal Asp-Ile and 6-His tag Accession # Q53TN1
N-terminal Sequence Analysis	Asn29
Predicted Molecular Mass	86 kDa

SPECIFICATIONS

SDS-PAGE	115 kDa, reducing conditions
Activity	Measured by its ability to cleave the fluorogenic peptide substrate, Gly-Pro-7-amido-4-methylcoumarin (GP-AMC). The specific activity is >2,500 pmol/min/µg as measured under the described conditions. See Activity Assay Protocol on www.RnDSystems.com.
Endotoxin Level	<1.0 EU per 1 µg of the protein by the LAL method.
Purity	>95%, by SDS-PAGE under reducing conditions and visualized by silver stain.
Formulation	Supplied as a 0.2 µm filtered solution in MES and NaCl. See Certificate of Analysis for details.

Activity Assay Protocol

Materials

- Assay Buffer: 25 mM Tris, pH 8.0
- Recombinant Human DPPIV/CD26 (rhDPPIV) (Catalog # 1180-SE)
- Substrate: H-Gly-Pro-AMC (Bachem, Catalog # I-1225), 10 mM stock in DMSO
- F16 Black Maxisorp Plate (Nunc, Catalog # 475515)
- Fluorescent Plate Reader (Model: SpectraMax Gemini EM by Molecular Devices) or equivalent

Assay

1. Dilute rhDPPIV to 0.2 ng/µL in Assay Buffer.
2. Dilute Substrate to 20 µM in Assay Buffer.
3. Load into a black plate 50 µL of 0.2 ng/µL rhDPPIV and start the reaction by adding 50 µL of 20 µM Substrate. As a Substrate Blank combine 50 µL of Assay Buffer and 50 µL of Substrate.
4. Read at excitation and emission wavelengths of 380 nm and 460 nm, respectively, in kinetic mode for 5 minutes.
5. Calculate specific activity:

$$\text{Specific Activity (pmol/min/µg)} = \frac{\text{Adjusted } V_{max}\text{* (RFU/min) x Conversion Factor** (pmol/RFU)}}{\text{amount of enzyme (µg)}}$$

*Adjusted for Substrate Blank

**Derived using calibration standard 7-amino, 4-Methyl Coumarin (Sigma, Catalog # A-9891)

Final Assay Conditions

Per Well:

- rhDPPIV: 0.010 µg
- Substrate: 10 µM

PREPARATION AND STORAGE

Shipping

The product is shipped with dry ice or equivalent. Upon receipt, store it immediately at the temperature recommended below.

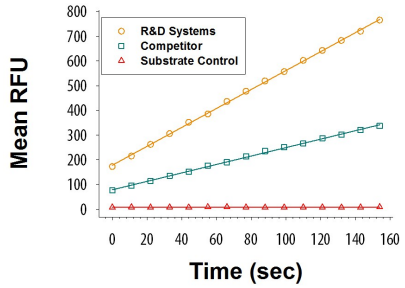
Stability & Storage

Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 6 months from date of receipt, -20 to -70 °C as supplied.
- 3 months, -20 to -70 °C under sterile conditions after opening.

DATA

Enzyme Activity



Recombinant Human DPPIV/CD26 (Catalog # 1180-SE) is measured by its ability to cleave the fluorogenic peptide substrate, Gly-Pro-7-amido-4-methylcoumarin (GP-AMC). The activity (orange) is approximately 2-fold greater than the competitor's DPPIV/CD26 (green).

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BACKGROUND

DPPIV/CD26 (EC 3.4.14.5) is a serine exopeptidase that releases Xaa-Pro dipeptides from the N-terminus of oligo- and polypeptides (1, 2). It is a type II membrane protein consisting of a short cytoplasmic tail, a transmembrane domain, and a long extracellular domain (3-5). The extracellular domain contains glycosylation sites, a cysteine-rich region and the catalytic active site (Ser, Asp and His charge relay system). The amino acid sequence of the mouse DPPIV/CD26 extracellular domain is 84% and 91% identical to the human and rat counterparts, respectively. In the native state, DPPIV/CD26 is present as a noncovalently linked homodimer on the cell surface of a variety of cell types. The soluble form is also detectable in human serum and other body fluids, the levels of which may have clinical significance in patients with cancer, liver and kidney diseases, and depression.

DPPIV/CD26 plays an important role in many biological and pathological processes. It functions as T cell-activating molecule (THAM). It serves as a cofactor for entry of HIV in $CD4^+$ cells (6). It binds adenosine deaminase, the deficiency of which causes severe combined immunodeficiency disease in humans (7). It cleaves chemokines such as stromal-cell-derived factor 1α and macrophage-derived chemokine (8, 9). It degrades peptide hormones such as glucagon (10). It truncates procalcitonin, a marker for systemic bacterial infections with elevated levels detected in patients with thermal injury, sepsis and severe infection, and in children with bacterial meningitis (11).

References:

1. Misumi and Ikehara (1998) in *Handbook of Proteolytic Enzymes*. Barrett, *et al.* (eds) San Diego: Academic Press, p. 378.
2. Ikehara, *et al.* (1994) Methods Enzymol. **244**:215.
3. Marguet, *et al.* (1992) J. Biol. Chem. **267**:2200.
4. Bernard, *et al.* (1994) Biochemistry **33**:15204.
5. Viver, *et al.* (1991) J. Immunol. **147**:447.
6. Callebaut, *et al.* (1993) Science **262**:2045.
7. Kameoka, *et al.* (1993) Science **261**:466.
8. Ohtsuki, *et al.* (1998) FEBS Lett. **431**:236.
9. Proost, *et al.* (1999) J. Biol. Chem. **274**:3988.
10. Hinke, *et al.* (2000) J. Biol. Chem. **275**:3827.
11. Wrenger, *et al.* (2000) FEBS Lett. **466**:155.

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