

Specifications and Use

- Sequence** ♦ Mca-Lys-Pro-Leu-Gly-Leu-Dpa-Ala-Arg-NH₂ (Neumann, U. *et al.*, 2004, Anal. Biochem. **328**:166 - 173).
♦ Mca: (7-Methoxycoumarin-4-yl)acetyl, Dpa: N-3-(2, 4-Dinitrophenyl)-L-2,3-diaminopropionyl.
- Molecular Mass** ♦ 1221.34 Da.
- Purity** ♦ > 95% based on high performance liquid chromatography.
- Peptide Content** ♦ 78.9%.
- Quantity** ♦ 1 mg. After dilution, it is sufficient for 600 assays using the recommended conditions.
- Recommended Assay Conditions** ♦ A fluorescence plate reader with excitation at 320 nm and emission at 405 nm is recommended for the measurement of the enzymatic activity. The substrate can be used at the final concentration of 10 μM in a total of 100 μL reaction mixture.

- Applications**
- ♦ The peptide substrate contains a highly fluorescent 7-methoxycoumarin group that is efficiently quenched by resonance energy transfer to the 2,4-dinitrophenyl group. It can be used to measure the activities of peptidases that are capable of cleaving an amide bond between the fluorescent group and the quencher group, causing an increase in fluorescence.
 - ♦ It is an excellent substrate for MMP-1 (collagenase 1), MMP-2 (gelatinase A), MMP-7 (matrilysin), MMP-8 (collagenase 2), MMP-9 (gelatinase B), MMP-11 (stromelysin 3), MMP-12 (macrophage elastase), MMP-13 (collagenase 3), MMP-14 (MT1-MMP), and MMP-16 (MT3-MMP). The cleavage site is the peptide bond between Gly and Leu.
 - ♦ It is also an excellent substrate for active Cathepsins D and E and ADAM10 and ADAM17/TACE.

- Formulation** ♦ A stock solution at 10 mg/mL or 6.46 mM in dimethyl sulfoxide (DMSO).

- Shipping** ♦ The substrate is shipped with cold packs. Upon receiving, store it immediately at the temperature recommended below.

- Storage**
- ♦ Samples are stable for up to twelve months from date of receipt at -20° C to -70° C.
 - ♦ The substrate can be aliquoted and stored at -20° C to -70° C in a manual defrost freezer for six months.
 - ♦ **Avoid repeated freeze-thaw cycles. Minimize exposure to light.**

Comparison of ES010 to ES001 or ES003 using recombinant human enzymes from R&D Systems

Enzyme	Substrates	Fold
MMP-1	ES010/ES001	6.0
MMP-2	ES010/ES001	1.0
MMP-7	ES010/ES001	1.6
MMP-8	ES010/ES001	1.9
MMP-9	ES010/ES001	1.0
MMP-11	ES010/ES001	2.2
MMP-12	ES010/ES001	2.2
MMP-13	ES010/ES001	1.5

Enzyme	Substrates	Fold
MMP-14	ES010/ES001	1.6
MMP-16	ES010/ES001	1.8
ADAM10	ES010/ES003	3.8
TACE	ES010/ES003	2.8
Cathepsin D	ES010/ES001	4.2
Cathepsin E	ES010/ES001	1.6