MMP-2 proenzyme (human fibroblasts)

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ALX-200-419-C005	5µg

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Handling & Storage

Use/Stability	As indicated on product label or CoA when stored as recommended.
Handling	Avoid freeze/thaw cycles.
Long Term Storage	-80°C
Shipping	Dry Ice

Regulatory Status

RUO - Research Use Only

Product Details

Alternative Name	Matrix metalloproteinase 2, Gelatinase A, 72 kDa Type IV collagenase
Application Notes	Immunogen for antibody generation, control in immunoassays and for characterizing interactions with MMP inhibitors.
Formulation	Liquid. In 50mM TRIS-HCl, pH 7.0, containing 200mM NaCl, 5mM $CaCl_2$, 1µM $ZnCl_2$, 0.05% BRIJ 35 and 0.05% sodium azide.
MW	~72kDa
Purity	≥90% (SDS-PAGE, Western blot)
Purity Detail	No other MMP contaminants are detectable.
Source	Isolated from human rheumatoid synovial fibroblasts. Requires activation.
Specific Activity	≥850mU/mg protein (Y. Masui, et al.; Biochem. Med. **17**, 215 (1977)). One unit is defined as the amount of enzyme that hydrolyzes 1µmol Dnp-Pro-Gln-Gly-Ile-Ala-Gly-Gln-D-Arg-OH per min. at 37°C, pH 7.0.

Technical Info / Product Notes

Activity: Specific activity can be assayed with the synthetic substrate N-(2,4)-dinitrophenyl-Pro-Gln-Gly-Ile-Ala-Gly-Gln-D-Arg (Dnp-peptide) (Masui et al.). Substrate concentration should be 0.5mg/ml in 50mM TRIS-HCl, pH 7.0, 200mM NaCl, 5mM $CaCl_2$, 1µM $ZnCl_2$, 0.05% sodium azide, 0.05% BRIJ35, containing 0.05mg/ml albumine. One unit MMP catalyzes the hydrolysis of 1µmol Dnp-peptide/min. at 37°C and pH 7.0. Alternatively the fluorogenic substrate (7-Methoxycoumarin-4-yl) acetyl-Pro-Leu-Gly-Leu-N-β-Dnp-L-(α,β-diaminopropionyl)Ala-Arg-NH_2 (Knight et al. 1992) can be used. Hydrolysis of the Gly-Leu bond separates the highly fluorescent (7-Methoxycoumarin-4-yl)acetyl group from the 2,4-dinitrophenyl resulting in an increase of fluorogenic intensity. The K_m value for the gelatinase A is $7.0\times10^5 M^{-1} s^{-1}$. Substrate should be kept as a 9.15mM stock solution in DMSO (10mg/ml). In the assay the substrate concentration should be ~25µM. The assay can be performed in a 96-well microtiter plate (100/200µl per well) suitable for fluorogenic measurements (Ex 328 nm; Em 393 nm).

Activation: Requires activation by 2mM (final concentration) APMA or 1mM mersalyl acid for 60-120 min. at 37°C. We do not recommend to use trypsin for activation! Do not dilute enzyme for activation!

Inhibitors: Activated enzyme is inhibited by tissue inhibitors of matrix metalloproteinase-2 (TIMP-2) and by chelators of divalent cations like EDTA or o-phenanthroline.

UniProt ID

P08253

Last modified: May 29, 2024

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