

Peptidylglycine Alpha Amidating Monooxygenase (PAM) Active Protein | PAM active protein

Active Peptidylglycine Alpha Amidating Monooxygenase (PAM)

CATALOG # AAA161894	HOST 293F cell	APPLICATIONS Activity Assay, Cell Culture
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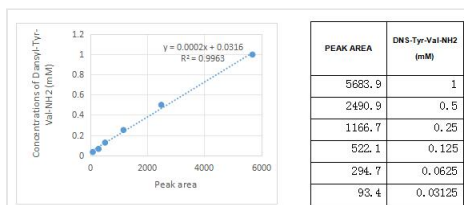
PRODUCT SYNONYMS

Peptidylglycine Alpha Amidating Monooxygenase (PAM); Active Peptidylglycine Alpha Amidating Monooxygenase (PAM); Peptidylglycine Alpha Amidating Monooxygenase; PAM active protein (eukaryotic; 293F cell); Human; PAL; PHM; Peptidylamidoglycolate lyase; Peptidyl-Alpha-hydroxyglycine Alpha-amidating Lyase; Peptidylglycine Alpha-Hydroxylating Monooxygenase4; PAM active protein

PRODUCT SPECIFICATIONS

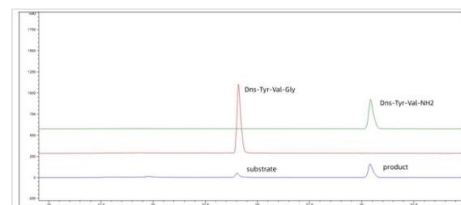
Host	293F cell
Purity/Purification	Greater than 90% by SDS-PAGE
Form/Format	Freeze-dried powder; PBS, pH7.4, containing 0.01% SKL, 1mM DTT, 5% Trehalose and Proclin300.
Sequence	Phe31~Val8 17
Applicable Applications for PAM active protein	Activity Assay, Cell Culture
Species	Human
Expression System	Eukaryotic expression
Tag	N-terminal His Tag
Endotoxin Level	<1.0EU per 1ug (determined by the LAL method)
Isoelectric Point	6.1
Research Area	Enzyme & Kinase
Preparation and Storage	Store at 2-8 degree C for one month, -80 degree C for 12 months

IMAGES



Bioactivity

Figure 3. The standard curve of Dns-Tyr-Val-NH2



Bioactivity

Figure 2. The reaction product compared with standard Dns-Tyr-Val-Gly and Dns-Tyr-Val-NH2.

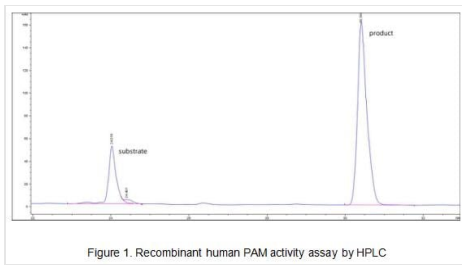


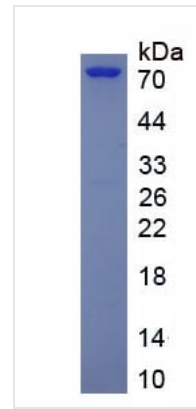
Figure 1. Recombinant human PAM activity assay by HPLC

Bioactivity

Peptidyl-glycine alpha-amidating monooxygenase (PAM) is an enzyme that is required for the biosynthesis of many signaling peptides. It has two enzymatically active domains with catalytic activities-peptidylglycine alpha-hydroxylating monooxygenase (PHM) and peptidyl-alpha-hydroxyglycine alpha-amidating lyase (PAL). These catalytic domains work sequentially to catalyze neuroendocrine peptides to active alpha-amidated products. A typical activity assay was using Dns-Tyr-Val-Gly as substrate, thus the activity of recombinant human PAM was measured by its ability to hydrolyze Dns-Tyr-Val-Gly to Dns-Tyr-Val-NH₂. The reaction was performed in 100 mM MES/KOH pH 6.0, 30 mM KI, 30 mM KCl, 1 uM cupric sulfate, 100 ug/ml catalase (APC418Hu05), 1% (v/v) ethanol, 0.001% (v/v) Triton X-100 and 10 mM ascorbate. 250 ul 0.7 mM substrate of Dns-Tyr-Val-Gly was added with 250 ul various concentrations of recombinant human PAM (1 ug/ml, 5 ug/ml, 10 ug/ml and 20 ug/ml). Incubated at 37 degree C for 30min, the reaction was stopped by addition 6% (v/v) TCA. The product and substrate was detected by RP-HPLC with UV-detection at 280 nm, the analyses were performed at 25 degree C employing a Agilent ZORBAX Poroshell SB C18 column (9.4x250 mm, 5 um), the flow rate was 1 ml/min. The mobile phase consisted of 100 mM sodium acetate (pH 6.5) and 30 min linear gradient of 10-90% acetonitrile. At 30-35 min, the mobile phase consisted of 90% acetonitrile and 10% sodium acetate. The result was shown in Figure 1. As the Figure 1 shows, the substrate have been hydrolyzed to Dns-Tyr-Val-NH₂ after incubating with recombinant human PAM. The retention time of Dns-Tyr-Val-Gly and Dns-Tyr-Val-NH₂ is 24.088 and 30.421 respectively (Figure 2). The specific activity of recombinant human PAM is >6600 pmol/min/ug.

SDS-PAGE

Figure. SDS-PAGE



ADDITIONAL INFORMATION

Product Categories/Family for PAM active protein	Active proteins
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NCBI AND UNIPROT INFORMATION

NCBI GI #	21070984
NCBI GeneID	5066
NCBI Accession #	NP_000910.2; NP_620121.1; NP_620176.1; NP_620177.1
NCBI GenBank Nucleotide #	NM_000919.3
UniProt Accession #	P19021; O43211; O95080; Q16252; Q16253; Q54A45; Q86U53; Q8WVC7; Q9UCG0; A6NMR0; A8K293
Molecular Weight	106,276 Da
NCBI Official Full Name	peptidyl-glycine alpha-amidating monooxygenase isoform a
NCBI Official Synonym Full Names	peptidylglycine alpha-amidating monooxygenase
NCBI Official Symbol	PAM
NCBI Official Synonym Symbols	PAL; PHM
NCBI Protein Information	peptidyl-glycine alpha-amidating monooxygenase
UniProt Protein Name	Peptidyl-glycine alpha-amidating monooxygenase
UniProt Gene Name	PAM
UniProt Synonym Gene Names	PAM; PHM; PAL

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Small volumes of product may become entrapped in the seal of the vial during shipment and storage; if necessary, briefly centrifuge the vial on a tabletop centrifuge to dislodge any liquid in the cap.

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Online datasheet: <https://www.aaabiotech.com/293f-active-protein/peptidylglycine-alpha-amidating-monooxygenase-pam/161894>