SANTA CRUZ BIOTECHNOLOGY, INC.

p-PAM (Ser 949)-R: sc-22263-R



BACKGROUND

Peptidylglycine α -amidating monooxygenase (PAM) catalyzes the two-step formation of bioactive $\alpha\text{-amidated}$ neural and endocrine peptides from their glycine-extended precursors. PAM is a bifunctional protein that contains a peptidyl-glycine α -hydroxylating monooxygenase and a peptidyl- α -hydroxyglycine α -amidating lyase catalytic domains. Tissue-specific alternative splicing and endoproteo-lysis generate both soluble and integral membrane mono- and bifunctional PAM proteins. PAM is highly expressed in ovary, testis, lung, heart septum, anterior pituitary and hypothalamus, and to a lesser extent in liver, ventricle, atrium and neuro-intermediate lobe. The 3'-untranslated region of PAM mRNA has a novel 20-nucleotide cis element, which is able to interact with cellular cytosolic protease-sensitive factor(s). The cytosolic domain of the PAM protein contains multiple signals determining its subcellular localization. PAM interacts with three related cytosolic proteins, designated P-CIPs (PAM cytosolic interactor proteins). P-CIP2 is a protein kinase that phosphorylates PAM at serine 949. Phosphorylation of PAM in the cytosolic domain of PAM plays a critical role in the trafficking of PAM. PAM in rat sciatic nerves is proteolytically processed during the axonal transport of secretion granules.

REFERENCES

- 1. Husten, E.J., et al. 1993. Use of endoproteases to identify catalytic domains, linker regions and functional interactions in soluble peptidylglycine α -amidating monooxygenase. J. Biol. Chem. 268: 9709-9717.
- 2. Yun, H.Y., et al. 1995. Phosphorylation of the cytosolic domain of peptidyl glycine α -amidating monooxygenase. J. Biol. Chem. 270: 30075-30083.
- 3. Takasugi, H., et al. 1996. Distribution and processing of peptidylglycine α -mediating monooxygenase activity in rat dorsal root ganglia and sciatic nerves. Neurochem. Int. 29: 397-403.
- 4. el Meskini, R., et al. 1997. Estrogen regulation of peptidylglycine α -amidating monooxygenase expression in anterior pituitary gland. Endocrinology 138: 379-388.
- 5. Fraboulet, S., et al. 1998. Identification of a novel CIS-element in the 3'untranslated region of mammalian peptidylglycine α -amidating monooxygenase messenger ribonucleic acid. Endocrinology 139: 894-904.
- 6. Girard, B., et al. 1999. Characterization and regulation of peptidylglycine α -amidating monooxygenase (PAM) expression in H9c2 cardiac myoblasts. Cell Tissue Res. 298: 489-497.
- 7. Caldwell, B.D., et al. 1999. The novel kinase peptidylglycine α -amidating monooxygenase cytosolic interactor protein 2 interacts with the cytosolic routing determinants of the peptide processing enzyme peptidyl glycine alpha-amidating monooxygenase. J. Biol. Chem. 274: 34646-34656.

CHROMOSOMAL LOCATION

Genetic locus: PAM (human) mapping to 5q21.1; Pam (mouse) mapping to 14 E2.3.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

SOURCE

p-PAM (Ser 949)-R is a rabbit polyclonal antibody raised against a short amino acid sequence containing Ser 949 phosphorylated PAM of human origin.

PRODUCT

Each vial contains 200 µg lgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-22263 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-PAM (Ser 949)-R is recommended for detection of PAM of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-PAM (Ser 949)-R is also recommended for detection of correspondingly phosphorylated PAM in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for PAM siRNA (h): sc-106802, PAM siRNA (m): sc-155926, PAM shRNA Plasmid (h): sc-106802-SH, PAM shRNA Plasmid (m): sc-155926-SH, PAM shRNA (h) Lentiviral Particles: sc-106802-V and PAM shRNA (m) Lentiviral Particles: sc-155926-V.

Molecular Weight of PAM: 120/94/84/45 kDa.

Positive Controls: MIA PaCa-2 cell lysate: sc-2285.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.