



α -mannosidase (Y-80): sc-98847

BACKGROUND

Autophagy is a membrane trafficking mechanism that delivers cytoplasmic cargo to the vacuole/lysosome for degradation and recycling. The proper functioning of eukaryotic organelles is largely dependent on the specific packaging of cargo proteins within transient delivery vesicles. The cytoplasm to vacuole targeting (Cvt) pathway is an autophagy-related trafficking pathway whose cargo proteins, aminopeptidase I and α -mannosidase, are selectively transported from the cytoplasm to the lysosome-like vacuole in yeast. In the budding yeast *Saccharomyces cerevisiae*, α -mannosidase, also known as AMP Deaminase or Ams1, is a marker enzyme of the vacuolar membrane. α -mannosidase has no typical signal sequence, but is located on the inner surface of the vacuolar membrane. The enzyme is synthesized as a polypeptide and is converted to a shorter polypeptide by the vacuolar processing protease, proteinase A. α -mannosidase forms an oligomer in the cytoplasm, however its mechanism of vacuolar delivery has not been established. Vacuolar localization of α -mannosidase is blocked in mutants that are defective in the Cvt and autophagy pathways.

REFERENCES

1. Yoshihisa, T. and Anraku, Y. 1990. A novel pathway of import of α -mannosidase, a marker enzyme of vacuolar membrane, in *Saccharomyces cerevisiae*. *J Biol Chem.* 265: 22418-22425.
2. Hutchins, M.U. and Klionsky, D.J. 2001. Vacuolar localization of oligomeric α -mannosidase requires the cytoplasm to vacuole targeting and autophagy pathway components in *Saccharomyces cerevisiae*. *J Biol Chem.* 276: 20491-20498.
3. Shintani, T., Huang, W.P., Stromhaug, P.E. and Klionsky, D.J. 2002. Mechanism of cargo selection in the cytoplasm to vacuole targeting pathway. *Dev. Cell* 3: 825-837.
4. Huang, W.P. and Klionsky, D.J. 2002. Autophagy in yeast: a review of the molecular machinery. *Cell. Struct. Funct.* 27: 409-420.
5. Thumm, M. 2002. Hitchhikers guide to the vacuole-mechanisms of cargo sequestration in the Cvt and autophagic pathways. *Mol. Cell* 10: 1257-1258.

SOURCE

α -mannosidase (Y-80) is a rabbit polyclonal antibody raised against amino acids 732-810 mapping at the C-terminus of α -mannosidase of *Saccharomyces cerevisiae* origin.

PRODUCT

Each vial contains 200 μ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

STORAGE

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

RESEARCH USE

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

APPLICATIONS

α -mannosidase (Y-80) is recommended for detection of α -mannosidase of *Saccharomyces cerevisiae* origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], 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).

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 A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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.

PROTOCOLS

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