SANTA CRUZ BIOTECHNOLOGY, INC.

4E-BP1/2/3 (H-40): sc-98602

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

The multisubunit eukaryotic translation initiation factor (eIF) 4F recruits 40S ribosomal subunits to the 5' end of mRNA. The eIF4F subunit eIF4E interacts directly with the mRNA 5' cap structure. Assembly of the eIF4F complex is inhibited by a family of repressor polypeptides, the eIF4E-binding proteins (4E-BPs). 4E-BP1 (also known as PHAS-1) normally binds eIF4E, inhibiting cap-dependent translation. Hyper-phosphorylation of 4E-BP1 disrupts this binding, activating cap-dependent translation. The PI 3-kinase/Akt pathway and the FRAP/mTOR kinase regulate 4E-BP1. 4E-BP1 is phosphorylated *in vivo* on multiple residues and phosphorylation by FRAP/mTOR on Threonine 37 and Threonine 46 of human 4E-BP1 may prime it for subsequent phosphorylation at sites including Serine 65 and Threonine 70. The corresponding rat residues include Threonine 36, Threonine 45, Serine 64 and Threonine 69. *In vitro,* 4E-BP1 is also phosphorylated by ataxia telangiectasia (Atm) at human Serine 112 (rat Serine 111) in response to an increase in Insulin levels.

REFERENCES

1. Pause, A., et al. 1994. Insulin-dependent stimulation of protein synthesis by phosphorylation of a regulator of 5'-cap function. Nature 371: 762-767.
2. Fadden, P., et al. 1997. Identification of phosphorylation sites in the translational regulator, PHAS-I, that are controlled by Insulin and Rapamycin in rat adipocytes. J. Biol. Chem. 272: 10240-10247.
3. Brunn, G.J., et al. 1997. Phosphorylation of the translational repressor PHAS-I by the mammalian target of Rapamycin. Science 277: 99-101.
4. Gingras, A.C., et al. 1998. 4E-BP1, a repressor of mRNA translation, is phosphorylated and inactivated by the Akt (PKB) signaling pathway. Genes Dev. 12: 502-513.
5. Gingras, A.C., et al. 1999. Regulation of 4E-BP1 phosphorylation: a novel two-step mechanism. Genes Dev. 13: 1422-1437.
6. Yang, D.Q., et al. 2000. Participation of Atm in Insulin signalling through phosphorylation of eIF-4E-binding protein 1. Nat. Cell Biol. 2: 893-898.

SOURCE

4E-BP1/2/3 (H-40) is a rabbit polyclonal antibody raised against amino acids 29-68 mapping within an internal region of 4E-BP2 of human origin.

PRODUCT

Each vial contains 200 µg 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.

PROTOCOLS

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

APPLICATIONS

4E-BP1/2/3 (H-40) is recommended for detection of 4E-BP1, 4E-BP2, 4E-BP3 of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg 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).

4E-BP1/2/3 (H-40) is also recommended for detection of 4E-BP1, 4E-BP2, 4E-BP3 in additional species, including canine, bovine and porcine.

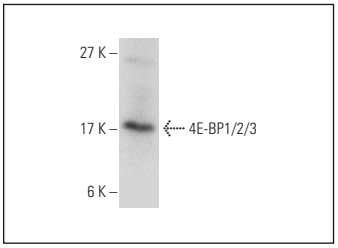
Molecular Weight of 4E-BP1/2/3: 21/16/12 kDa.

Positive Controls: HeLa whole cell lysate: sc-2200.

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.

DATA



4E-BP1/2/3 (H-40): sc-98602. Western blot analysis of 4E-BP1/2/3 expression in HeLa whole cell lysate.

RESEARCH USE

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