

# p-4E-BP1 (Ser 65)-R: sc-18091-R

## 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. Hyperphosphorylation 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.

## CHROMOSOMAL LOCATION

Genetic locus: EIF4EBP1 (human) mapping to 8p11.23; Eif4ebp1 (mouse) mapping to 8 A2.

## SOURCE

p-4E-BP1 (Ser 65)-R is a rabbit polyclonal antibody raised against a short amino acid sequence containing phosphorylated Ser 65 of 4E-BP1 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.

Blocking peptide available for competition studies, sc-18091 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## 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

p-4E-BP1 (Ser 65)-R is recommended for detection of Ser 64 phosphorylated 4E-BP1 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).

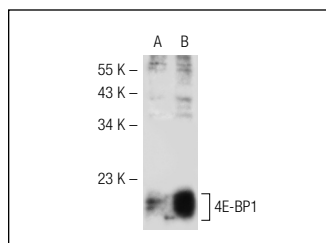
p-4E-BP1 (Ser 65)-R is also recommended for detection of correspondingly phosphorylated Ser on 4E-BP1 in additional species, including porcine.

Suitable for use as control antibody for 4E-BP1 siRNA (h): sc-29594, 4E-BP1 siRNA (m): sc-29595, 4E-BP1 shRNA Plasmid (h): sc-29594-SH, 4E-BP1 shRNA Plasmid (m): sc-29595-SH, 4E-BP1 shRNA (h) Lentiviral Particles: sc-29594-V and 4E-BP1 shRNA (m) Lentiviral Particles: sc-29595-V.

Molecular Weight of p-4E-BP1: 21 kDa.

Positive Controls: 4E-BP1 (h): 293T Lysate: sc-116590 or HeLa whole cell lysate: sc- 2200.

## DATA



p-4E-BP1 (Ser 65)-R: sc-18091-R. Western blot analysis of 4E-BP1 phosphorylation in non-transfected: sc-117752 (A) and human 4E-BP1 transfected: sc-116590 (B) 293T whole cell lysates.

## SELECT PRODUCT CITATIONS

1. Guan, L., et al. 2007. Protein kinase C-mediated down-regulation of Cyclin D1 involves activation of the translational repressor 4E-BP1 via a phosphoinositide 3-kinase/Akt-independent, protein phosphatase 2A-dependent mechanism in intestinal epithelial cells. *J. Biol. Chem.* 282: 14213-14225.
2. Balgi, A.D., et al. 2009. Screen for chemical modulators of autophagy reveals novel therapeutic inhibitors of mTORC1 signaling. *PLoS ONE* 4: e7124.
3. Boukhettala, N., et al. 2010. Effects of essential amino acids or glutamine deprivation on intestinal permeability and protein synthesis in HCT-8 cells: involvement of GCN2 and mTOR pathways. *Amino Acids*. E-published.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog for detailed protocols and support products.