Store at -20°C

#51367

4E-BP1 Control Cell Extracts

100 µl (10 western blots)

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For Research Use Only. Not For Use In Diagnostic Procedures.

Product Includes	Product #	Quantity
4E-BP1 Control Cell Extracts (MCF7 untreated)	64125	100 ul
4E-BP1 Control Cell Extracts (MCF7 + insulin)	87570	100 ul

Background: Translation repressor protein 4E-BP1 (also known as PHAS-1) inhibits cap-dependent translation by binding to the translation initiation factor eIF4E. Hyper-phosphorylation of 4E-BP1 disrupts this interaction and results in activation of cap-dependent translation (1). Both the PI3 kinase/Akt pathway and FRAP/mTOR kinase regulate 4E-BP1 activity (2,3). Multiple 4E-BP1 residues are phosphorylated *in vivo* (4). While phosphorylation by FRAP/mTOR at Thr37 and Thr46 does not prevent the binding of 4E-BP1 to eIF4E, it is thought to prime 4E-BP1 for subsequent phosphorylation at Ser65 and Thr70 (5).

Description: *Nonphosphorylated 4E-BP1 Control Cell Extracts:* Total cell extracts from MCF7 cells, amino acids starved for 1 hour to serve as a negative control. Supplied in SDS Sample Buffer.

Phosphorylated 4E-BP1 Control Cell Extracts: Total cell extracts from MCF7 cells, amino acids starved for 1 hour followed by adding back amino acids for 1 hour and treating with 100 nM insulin for 30 min to serve as a positive control. Supplied in SDS Sample Buffer.

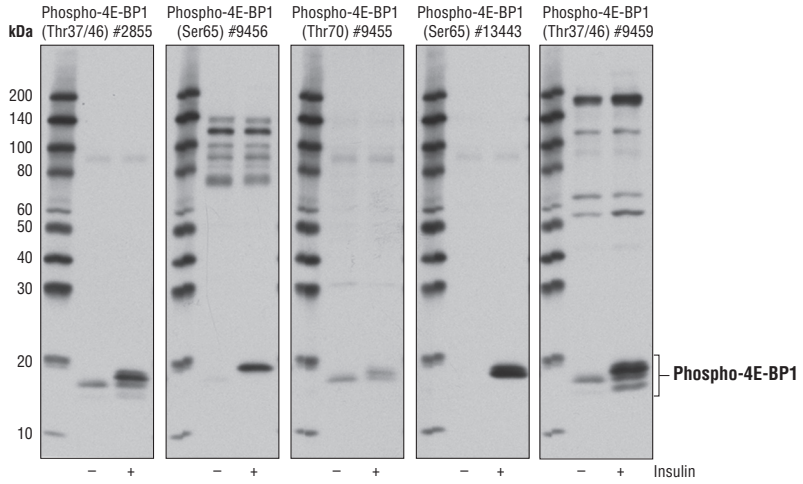
Directions for Use: Boil for 3 minutes prior to use. Load 10 µl of phosphorylated and nonphosphorylated 4E-BP1 Control Cell Extracts per lane.

Background References:

(1) Pause, A. et al. (1994) *Nature* 371, 762-7.
(2) Brunn, G.J. et al. (1997) *Science* 277, 99-101.
(3) Gingras, A.C. et al. (1998) *Genes Dev* 12, 502-13.
(4) Fadden, P. et al. (1997) *J Biol Chem* 272, 10240-7.
(5) Gingras, A.C. et al. (1999) *Genes Dev* 13, 1422-37.

Storage: Store at -20°C. Supplied in SDS Sample Buffer: 62.5 mM Tris- HCl (pH 6.8 at 25°C), 2% w/v SDS, 10% glycerol, 50 mM DTT, 0.01% w/v bromophenol blue or phenol red.

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com



Western blot analysis of 4E-BP1 Control Cell Extracts from MCF7 cells, amino acid starved (1 hr), then either untreated (-) or treated with insulin (100 nM, 30 min; +), using Phospho-4E-BP1 (Thr37/46) (236B4) Rabbit mAb #2855, Phospho-4E-BP1 (Ser65) (174A9) Rabbit mAb #9456, Phospho-4E-BP1 (Thr70) Antibody #9455, Phospho-4E-BP1 (Ser65) (D9G1Q) Rabbit mAb #13443, and Phospho-4E-BP1 (Thr37/46) Antibody #9459.

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Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.