#2331 Store at –20°C

Phospho-eEF2 (Thr56) Antibody

Cell Signaling TECHNOLOGY®

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rev. 12/21/15

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

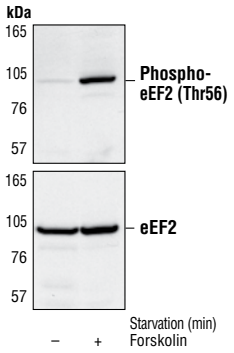
Entrez-Gene ID # 1938
Swiss-Prot Acc. # P13639

Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W Endogenous	H, M, R, Mk, C, Hm	95 kDa	Rabbit**

Background: Eukaryotic elongation factor 2 (eEF2) catalyzes the translocation of peptidyl-tRNA from the A site to the P site on the ribosome. It has been shown that phosphorylation of eEF2 at threonine 56 by eEF2 kinase inhibits its activity (1–4). eEF2 kinase is normally dependent on Ca^{2+} ions and calmodulin (5,6). eEF2 kinase can also be activated by PKA in response to elevated cAMP levels (7–9), which are generally increased in stress- or starvation-related conditions. A variety of treatments known to raise intracellular Ca^{2+} or cAMP levels have been shown to result in increased phosphorylation of eEF2, and thus to inhibit peptide-chain elongation. The inactive phosphorylated eEF2 can be converted to its active nonphosphorylated form by a protein phosphatase, most likely a form of protein phosphatase-2A (PP-2A). Insulin, which activates protein synthesis in a wide range of cell types, induces rapid dephosphorylation of eEF2 through mTOR signaling and may involve modulation of the activity of the PP-2A or the eEF2 kinase or both (10).

Specificity/Sensitivity: Phospho-eEF2 (Thr56) Antibody detects endogenous levels of eEF2 only when phosphorylated at Thr56. It does not recognize eEF2 phosphorylated at other sites.

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Thr56 of human eEF2. Antibodies are purified by protein A and peptide affinity chromatography.



Western blot analysis of extracts from C6 cells, untreated or forskolin-treated (10 µM for 60 minutes), using Phospho-eEF2 (Thr56) Antibody (upper) or eEF2 Antibody #2332 (lower).

Background References:

(1) Nairn, A.C. and Palfrey, H.C. (1987) *J. Biol. Chem.* 262, 17299–17303.
(2) Ryazanov, A.G. et al. (1988) *Nature* 334, 170–173.
(3) Carlberg, U. et al. (1990) *Eur. J. Biochem.* 191, 639–645.
(4) Redpath, N.T. et al. (1993) *Eur. J. Biochem.* 213, 689–699.
(5) Nairn, A.C. et al. (1985) *Proc. Natl. Acad. Sci. USA* 82, 7939–7943.
(6) Palfrey, H.C. et al. (1987) *J. Biol. Chem.* 262, 9785–9792.
(7) Redpath, N.T. and Proud, C.G. (1993) *Biochem. J.* 293, 31–34.
(8) Diggle, T. et al. (1998) *Biochem. J.* 336, 525–529.
(9) Hovland, R. et al. (1999) *FEBS Lett.* 444, 97–101.
(10) Proud, C. (2000) *Translational Control of Gene Expression. Cold Spring Harbor Laboratory Press, NY,* 719–739.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western Blotting	1:1000

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **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.