Mouse (monoclonal) Anti-4E-BP1 Unconjugated

PRODUCT ANALYSIS SHEET

Catalog Number:	AHO1382
Lot Number:	See product label
Quantity/Volume:	100 µg/0.2 mL
Clone Number:	554R16
Isotype:	IgG1 κ (mouse)
Form of Antibody:	Purified immunoglobulin in phosphate buffered saline, pH 7.2, with 1% bovine serum albumin.
Preservation:	0.1% sodium azide (Caution: sodium azide is a poisonous and hazardous substance. Handle with care and dispose of properly.)
Purification:	Purified from ascites by affinity chromatography.
Immunogen:	Recombinant human 4E-BP1 protein expressed in *E. coli*.
Specificity:	Eukaryotic initiation factor 4E binding protein 1 (4E-BP1), also known as PHAS, is a ~20 kDa member of a family of eIF4E-binding proteins whose binding affinity to eIF4E is regulated by its phosphorylation. It inhibits cap-dependent translation by binding to eIF4E on the same site that overlaps the binding site for eIF4G, preventing its binding to the latter and eventually leading to an increase in mRNA translation. The phosphorylation of 4E-BP1 is critical in determining cell fate by controlling translation initiation and apoptotic potency. 4E-BP1 is hyperphosphorylated in response to several external stimuli including hormones, growth factors, mitogens, cytokines and G-protein–coupled receptors and in response to stress conditions including nutrient deprivation. The phosphorylation of 4E-BP1 increases in response to activated phosphoinositol 3'-kinase (PI-3K) or its downstream effector Akt/PKB. 4E-BP1 is believed to mediate PI-3K and FRAP/mTOR signaling and is phosphorylated on at least six serine and threonine sites (Thr 37, Thr 46, Ser 65, Thr 70, Ser 83, and Ser 112). The phosphorylation of these sites is believed to occur in an orderly fashion where phosphorylation of threonine 37 and 46 by FRAP/mTOR is a priming step for subsequent phosphorylation of 4E-BP1 at the carboxy-terminal sites.
Species Reactivity:	Human, mouse and rat.
Applications:	This antibody is suitable for use in Western blotting.
Suggested Working Dilutions:	For Western blotting, the recommended concentration is 1 µg/mL. The optimal antibody concentration should be determined for each specific application.
Recommended Positive Control:	Human HEK293 cells, mouse 3T3-L1 cells and rat L6 cells.
Storage:	Store at 2-8ºC. For long term storage, aliquot into small volumes and store at –20ºC. Avoid repeated freeze-thaw cycles to prevent denaturing the antibody.

This product is for research use only. Not for use in diagnostic procedures.

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References:

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Zhou, L., et al. (2005) 4E-binding protein phosphorylation and eukaryotic initiation factor-4E release are required for airway smooth muscle hypertrophy. Am. J. Respir. Cell Mol. Biol. 33(2):195-202.

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Li, S., et al. (2002) Translational control of cell fate: availability of phosphorylation sites on translational repressor 4E-BP1 governs its proapoptotic potency. Mol. Cell Biol. 22(8):2853-2861.

Gingras, A.C., et al. (2001) Hierarchical phosphorylation of the translation inhibitor 4E-BP1. Genes Dev. 15(21):2852-2864.

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Related Products:

Antibodies:

4E-BP1[pT^{46}], Cat. # 44-1170G
mTOR [pS^{2448}], Cat. # 44-1125G
AMPKα1/2 [pT^{172}], Cat. # 44-1150G
eIF4G [pS^{1108}], Cat. # 44-526
p70 S6 kinase [pT^{229}], Cat. # 44-918G
Rib. Protein S6 [pS^{236}], Cat. # 44-921G
c-Fos [pT^{325}], Cat. # 44-281G
c-Jun [pS^{73}], Cat. # 44-292G
mTOR, Cat. # AHO1232
SHP2 [pS^{576}], Cat. # 44-557G
AKT, Cat. # AHO1112
Akt/PKB pan, Cat. # 44-609G
Akt/PKB [pT^{308}], Cat. # 44-602G
Akt/PKB [pS^{473}], Cat. # 44-621G
MEK1 [pSpS$^{218/222}$] /2 [pSpS$^{222/226}$], Cat. # 44-454G
ERK1&2 [pTpY$^{185/187}$], Cat. # 44-680G
NFAT-1 [pS^{54}], Cat. # 44-944

- 20 kDa

1 2 3

Western Blot Analysis

Proteins from cell extracts of human HEK293 cells (lane 1), mouse 3T3-L1 cells (lane 2), and rat L6 cells (lane 3) were resolved by SDS-PAGE and transferred to PVDF. The membranes were incubated with this 4E-BP1 monoclonal antibody (clone 554R16) at a concentration of 1 µg/mL for two hours at room temperature. After washing, the membranes were incubated with a goat F(ab')$_2$ anti-mouse IgG alkaline phosphatase conjugated antibody (Cat. # AMI4405) at a 1:2000 dilution. Bands were detected with CDP-substrate using the WesternStar™ method (Tropix) and Kodak BioMax film.

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