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Datasheet for W09-K01-MU1

Human MAPK1 (ERK2) Knockout A549 Cell Lysate

Overview

Description:	Human MAPK1 (ERK2) Knockout A549 Lysate - W09-K01-MU1
Item No.:	W09-K01-MU1
Size:	100 µg
Applications:	SDS-PAGE, WB
Origin:	Human

Product Details

Background:	ERK2 antibodies detect the ERK2 isoform. Mitogen activated protein kinase 1, also known as MAPK1, ERK, or ERK2, is an integral component of the MAP kinase cascade that regulates cell growth and differentiation. ERK1 and ERK2 are activated by MEK1 and MEK2 in the B-raf signaling pathway resulting in its translocation to the nucleus where it phosphorylates nuclear targets. Human ERK1 and ERK2 are 84% identical in sequence and share common functionality in cells.
Synonyms:	rabbit anti-ERK2 antibody, MAPK1, ERK 2, ERK-2, P42MAPK, PRKM1, PRKM2, MAPK 2, MAP kinase 2, Mitogen-activated protein kinase 2, p42-MAPK, MAP kinase isoform p42, Extracellular signal-regulated kinase 2, ERT1, MAP kinase 1, Mitogen-activated protein kinase 1
Species of Origin:	Human
Clone ID:	Clone 15

Target Details

Gene Name:	MAPK1

Purity/Specificity:	MAPK1 (ERK2) knockout A549 cells were grown in Dulbecco's medium supplemented with 10% fetal bovine serum. Cells were washed with PBS and then incubated on ice in modified RIPA buffer to lyse the cells. Protein integrity was ensured using a cocktail of protease inhibitors with broad specificity for the inhibition of aspartic, cysteine, and serine proteases as well as aminopeptidases (0.1 mM AEBSF HCl, 0.08 µM Aprotinin, 5 µM Bestatin, 1.5 µM E-64, 2 µM Leupeptin Hemisulfate, 1 µM Pepstatin A). Phosphatase inhibitors 1 mM NaF and 1 mM Na3VO4 were also added. Cell debris was removed by centrifugation. Protein concentration was determined by BCA using a commercially available kit. Protein concentration was adjusted to 2 mg/ml with modified 1X RIPA buffer. MAPK1 (ERK2) knockout A549 Clone 15 contains knockout deletions on all three copies of the MAPK1 (ERK2) gene in A549 cells. Each copy contains the same 104bp deletion induced by CRISPR/Cas9. The deletion occurs in exon 2 and disrupts the sequence between amino acids 59 to 94. These mutations induce a frame-shift and result in early stop codons. Validated by Sanger sequencing and Western blot.
Relevant Links:	• UniProtKB - P28482

Application Details

Tested Applications:	SDS-PAGE, WB
Application Note:	Human MAPK1 (ERK2) Knockout A549 Cell Lysate has been tested by SDS-PAGE and western blot and is suitable for use in Western blot, ELISA, Immunoprecipitation and ChIP. No detection of expected band at ~44kDa is observed in MAPK1 (ERK2) knockout A549 when compared with unmodified A549 cell lysates by Western blot.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ChIP:	User Optimized
ELISA:	User Optimized
IP:	User Optimized
WB:	User Optimized

Cell Line Data

Cell Line:	Human A549 (lung epithelial carcinoma)
Lysate Fractionation:	Whole Cell Lysate
Lysate Stimulation:	Not Stimulated
Culture Type:	Tissue Culture
Induction:	None (Control)

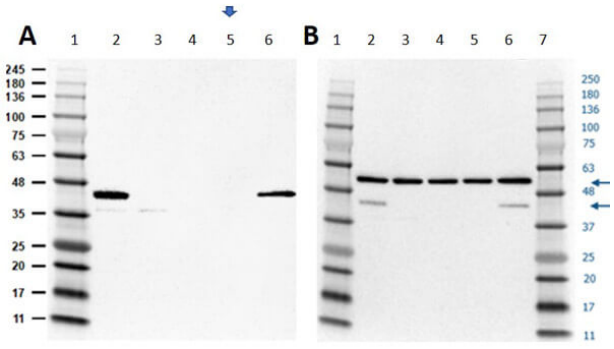
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	2.0 mg/mL by BCA assay
Buffer:	1X RIPA Buffer with HALT Protease and Phosphatase Inhibitors
Preservative:	None
Stabilizer:	None

Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -70° C or COLDER. For extended storage, aliquot contents to minimize freeze/thaw cycles.
Expiration:	Expiration date is one (1) year from date of receipt.

Images



Western Blot
Western Blot of Human MAPK1 (ERK2) Knockout A549 Cell Lysate. Lane 1: Opal Prestained MW Marker (p/n MB-210-0500). Lane 2: A549 WCL Parental (p/n W09-001-372). Lane 3: A549 MAPK1 KO Clone 4. Lane 4: A549 MAPK1 KO Clone 10. Lane 5: A549 MAPK1 KO Clone 15. Lane 6: HeLa WCL Parental (p/n W09-000-364) Load: 10µg lysate/lane. Primary Antibody [Blot A] Anti-MAPK1(ERK2) (p/n 600-401-GP1) ~44kDa; [Blot B] stripped and re-probed with Anti-Tubulin (p/n 600-401-880) ~50kDa; at 1µg/mL overnight at 2 -8°C. Secondary Antibody: Goat Anti-Rabbit IgG HRP (p/n 611-103-122) at 1:30,000 for 1hr at RT. Block: BlockOut Buffer (p/n MB-073) for 1hr at RT. No detection of expected band at ~44kDa is observed in MAPK1 (ERK2) knockout A549 when compared with unmodified A549 cell lysates by Western blot.

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