

Anti-Erk1 (pT202+pY204) + Erk2 (pT185+pY187) Antibody [PD00-38]

HA724560



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, IF-Cell
Molecular Wt:	Predicted band size: 41/43 kDa
Clone number:	PD00-38

Description: The activation of signal transduction pathways by growth factors, hormones and neurotransmitters is mediated through two closely related MAP kinases, p44 and p42, designated extracellular-signal related kinase 1 (ERK 1) and ERK 2, respectively. ERK proteins are regulated by dual phosphorylation at Tyrosine 204 and 187 and Threonine 177 and 160 residues mapping within a characteristic Thr-Glu-Tyr motif. Phosphorylation at both the Threonine 202 and Tyrosine 204 residues of ERK1 and Threonine 185 and Tyrosine 187 residues of ERK2 is required for full enzymatic activation. The structural consequences of dual-phosphorylation in the ERK2 include active site closure, alignment of key catalytic residues that interact with ATP, and remodeling of the activation loop. In response to activation, MAP kinases phosphorylate downstream components on serine and threonine. Upstream MAP kinase regulators include MAP kinase kinase (MEK), MEK kinase and Raf-1. The ERK family has three additional members: ERK 3, ERK 5 and ERK 6.

Immunogen: Synthetic peptide within Human aa 166-215 / 379.

Positive control: A431 cell lysate, SK-Br-3 cell lysate, human human thyroid carcinoma tissue, MCF-7.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P27361 Human | P28482 Human

Recommended Dilutions:

WB	1:1,000
IHC-P	1:5,000
IF-Cell	1:50

Storage Buffer: 1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). Store at -20℃ long term.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

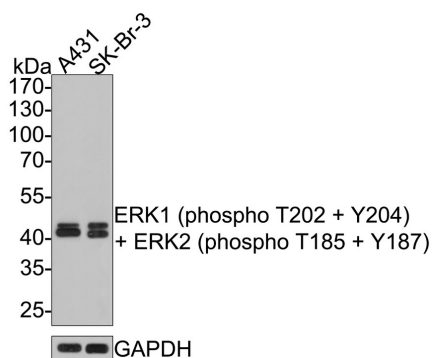


Fig1: Western blot analysis of Erk1 (pT202+pY204) + Erk2 (pT185+pY187) on different lysates with Rabbit anti-Erk1 (pT202+pY204) + Erk2 (pT185+pY187) antibody (HA724560) at 1/1,000 dilution.

Lane 1: A431 cell lysate

Lane 2: SK-Br-3 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 41/43 kDa

Observed band size: 41/43 kDa

Exposure time: 2 minutes;

10% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA724560) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

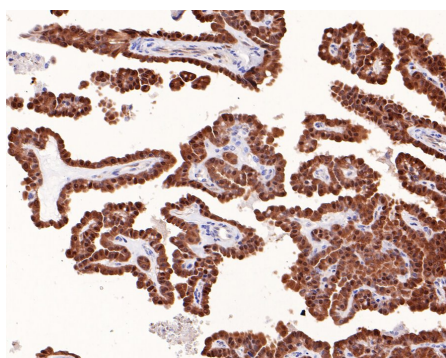


Fig2: Immunohistochemical analysis of paraffin-embedded human thyroid carcinoma tissue with Rabbit anti-Erk1 (pT202+pY204) + Erk2 (pT185+pY187) antibody (HA724560) at 1/5,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA724560) at 1/5,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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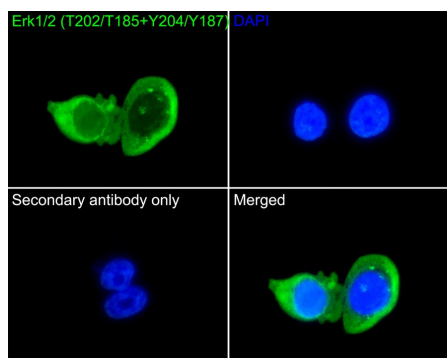


Fig3: Immunocytochemistry analysis of MCF-7 cells labeling Erk1 (pT202+pY204) + Erk2 (pT185+pY187) with Rabbit anti-Erk1 (pT202+pY204) + Erk2 (pT185+pY187) antibody (HA724560) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Erk1 (pT202+pY204) + Erk2 (pT185+pY187) antibody (HA724560) at 1/50 dilution in 2% negative goat serum overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Wang ZH et al. Sunitinib mesylate inhibits proliferation of human colonic stromal fibroblasts in vitro and in vivo. J Zhejiang Univ Sci B 15:701-12 (2014).
2. Assi J et al. Transglutaminase 2 overexpression in tumor stroma identifies invasive ductal carcinomas of breast at high risk of recurrence. PLoS One 8:e74437 (2013).

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