Datasheet for ABIN686482

anti-MEK1 antibody (AA 2-150)

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Overview

Quantity:	100 µL
Target:	MEK1 (MAP2K1)
Binding Specificity:	AA 2-150
Reactivity:	Human
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This MEK1 antibody is un-conjugated
Application:	ELISA, Immunohistochemistry (Paraffin-embedded Sections) (IHC (p)), Immunofluorescence (Cultured Cells) (IF (cc)), Immunofluorescence (Paraffin-embedded Sections) (IF (p)), Flow Cytometry (FACS), Immunohistochemistry (Frozen Sections) (IHC (fro))

Product Details

Immunogen:	KLH conjugated synthetic peptide derived from human MEK1
Isotype:	IgG
Cross-Reactivity:	Human
Predicted Reactivity:	Mouse,Rat,Dog,Pig,Chicken,Rabbit
Purification:	Purified by Protein A.

Target Details

Target:	MEK1 (MAP2K1)

Target Details

Alternative Name:	MEK1 (MAP2K1 Products)
Background:	Synonyms: CFC3, MEK1, MKK1, MAPKK1, PRKMK1, Dual specificity mitogen-activated protein kinase kinase 1, MAP kinase kinase 1, MAPKK 1, ERK activator kinase 1, MAPK/ERK kinase 1, MEK 1, MAP2K1 Background: Dual specificity protein kinase which acts as an essential component of the MAP kinase signal transduction pathway. Binding of extracellular ligands such as growth factors, cytokines and hormones to their cell-surface receptors activates RAS and this initiates RAF1 activation. RAF1 then further activates the dual-specificity protein kinases MAP2K1/MEK1 and MAP2K2/MEK2. Both MAP2K1/MEK1 and MAP2K2/MEK2 function specifically in the MAPK/ERK cascade, and catalyze the concomitant phosphorylation of a threonine and a tyrosine residue in a Thr-Glu-Tyr sequence located in the extracellular signal-regulated kinases MAPK3/ERK1 and MAPK1/ERK2, leading to their activation and further transduction of the signal within the MAPK/ERK cascade. Depending on the cellular context, this pathway mediates diverse biological functions such as cell growth, adhesion, survival and differentiation, predominantly through the regulation of transcription, metabolism and cytoskeletal rearrangements. One target of the MAPK/ERK cascade is peroxisome proliferator-activated receptor gamma (PPARG), a nuclear receptor that promotes differentiation and apoptosis. MAP2K1/MEK1 has been shown to export PPARG from the nucleus. The MAPK/ERK cascade is also involved in the regulation of endosomal dynamics, including lysosome processing and endosome cycling through the perinuclear recycling compartment (PNRC), as well as in the fragmentation of the Golgi apparatus during mitosis.
Gene ID:	5604
UniProt:	Q02750
Pathways:	MAPK Signaling, RTK Signaling, Interferon-gamma Pathway, Fc-epsilon Receptor Signaling Pathway, Neurotrophin Signaling Pathway, Activation of Innate immune Response, Toll-Like Receptors Cascades, Autophagy, Signaling of Hepatocyte Growth Factor Receptor, BCR Signaling

Application Details

Application Notes:	ELISA 1:500-1000 FCM 1:20-100 IHC-P 1:200-400 IHC-F 1:100-500 IF(IHC-P) 1:50-200

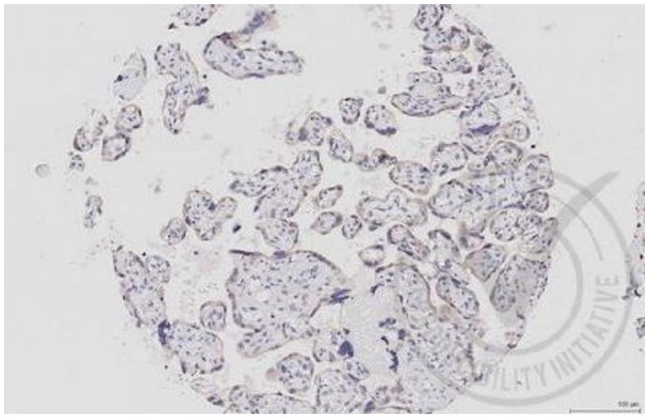
Application Details

	IF(IHC-F) 1:50-200 IF(ICC) 1:50-200
Restrictions:	For Research Use only

Handling

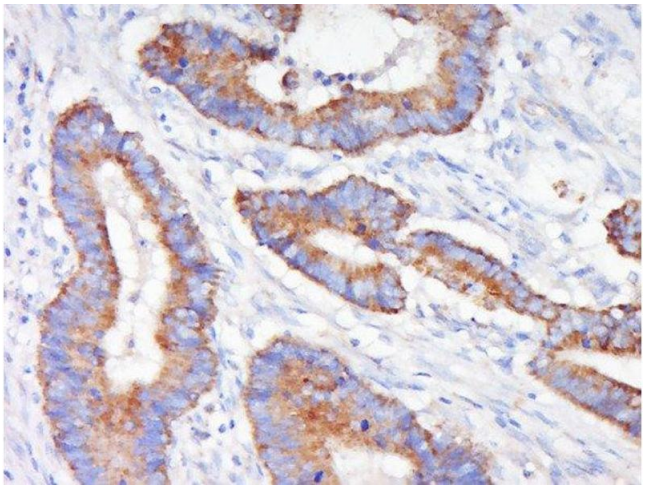
Format:	Liquid
Concentration:	1 μg/μL
Buffer:	0.01M TBS(pH 7.4) with 1 % BSA, 0.02 % Proclin300 and 50 % Glycerol.
Preservative:	ProClin
Precaution of Use:	This product contains ProClin: a POISONOUS AND HAZARDOUS SUBSTANCE, which should be handled by trained staff only.
Storage:	4 °C,-20 °C
Storage Comment:	Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.
Expiry Date:	12 months

Images



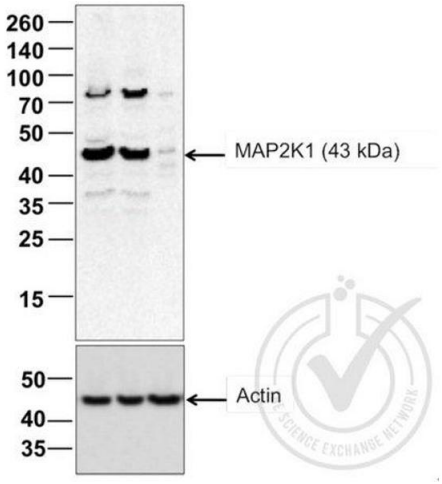
Immunohistochemistry

Image 1. Images provided the Independent Validation Program (badge number 029646)Formalin-fixed and paraffin embedded human placenta labeled with Rabbit Anti-MEK1 Polyclonal Antibody (ABIN686482) at 1:250 overnight at room temperature followed by conjugation to secondary antibody.



Immunohistochemistry (Paraffin-embedded Sections)

Image 2. Paraformaldehyde-fixed, paraffin embedded human colon cancer, Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15min, Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes, Blocking buffer (normal goat serum) at 37°C for 30min, Antibody incubation with MEK1 Polyclonal Antibody, Unconjugated at 1:500 overnight at 4°C, followed by a conjugated secondary for 20 minutes and DAB staining.



Western Blotting

Image 3. Image provided by the Independent Validation Program (badge number 29760). Lane 1: HeLa cell extract, Lane 2: NIH/3T3 cell extracts, Lane 3: c6/36 mosquito cell extract (non-reactivespecies) probed with Rabbit Anti-MEK1 Polyclonal Antibody, Unconjugated at 1:200 overnight at 4˚C. Followed by conjugation to secondary antibody at 1:20000 for 60 min at 26˚C.

Successfully validated (Immunohistochemistry (IHC))

by Immunohistochemistry Core, NYU Langone

Report Number: 029646

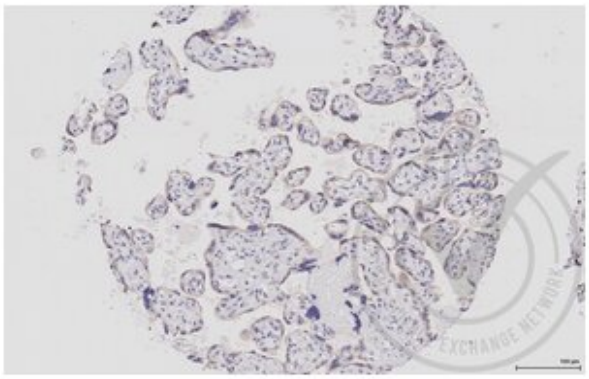
Date: Mar 28 2014

Lot Number:	90915
Method validated:	Immunohistochemistry (IHC)
Positive Control:	Human placenta
Negative Control:	Human breast adipocytes
Notes:	Signal detected in positive control sample and not in negative control sample.
Primary Antibody:	- Antigen: Mitogen-Activated Protein Kinase Kinase 1 (MAP2K1) - Catalog number: ABIN686482 - Supplier: Bioss - Supplier catalog number: bs-1433R - Lot number: 90915
Secondary Antibody:	- Antibody: Biotinylated goat anti-rabbit/anti-mouse (Kit) - Supplier: Ventana Medical Systems - Catalog number: 760-091 - Lot number: D07640BA
Isotype:	- Antibody: Rabbit IgG isotype control - Supplier: Ventana Medical Systems - Catalog number: 790-2014 - Lot number: C11245
Controls:	• Positive control: Human placenta tissue stained with antibody • Negative control: Human breast adipocytes tissue stained with antibody • Isotype control: Human placenta tissue stained with isotype control • Secondary only control: Human placenta tissue stained with secondary antibody only
Protocol:	• Immunohistochemistry was performed on a Ventana NEXes automated platform; instrument manufacturer specific reagents are italicized. • 1. Slides were preheated in convection oven at 60°C for 30 min • 2. Deparaffinization procedure: - 3 changes of Xylene, 5 min each - 3 changes of 100% Ethanol, 3 min each - 3 changes of 95% Ethanol, 3 min each - Rinsed in distilled water, 3 changes • 3. Heat retrieval procedure - Slides retrieved in 10.0 mM Citrate, pH6.0 in a 1000W microwave oven (~100°C) for 15 min. - Slides were allowed to cool (in citrate) for 30 min. - Slides were washed x 3 in Distilled water • 4. NEXes instrument procedure, iView DAB paraffin protocol (*abridged*): - Slide chamber warmed to 37°C • 5. Slides rinsed with *reaction buffer* x3 • 6. *iView Inhibitor (H2O2)* applied and incubated for 4 min

- 7. Slides rinsed with *reaction buffer*
- 8. Antibody Application - Primary antibody diluted 1:250 in PBS (100 microliter applied/slide) - Ventana Isotype control applied neat - Slides Incubated overnight at room temperature (~12 hours ~25°C)
- 9. Slides rinsed with *reaction buffer* x3
- 10. *iView Biotinylated IgG* applied and incubated for 8 min
- 11. Slides rinsed with *reaction buffer*
- 14. *iView Streptavidin-Horseradish Peroxidase* applied and incubated for 8 min
- 15. Slides rinsed with *reaction buffer*
- 16. *iView DAB/H2O2* applied and incubated for 8 min
- 17. Slides rinsed with *reaction buffer*
- 18. *iView Copper* applied and incubated for 4 min
- 19. Slides rinsed with *reaction buffer*
- 20. Slides washed in Dawn Detergent/tap water
- 21. Counterstain Procedure - Hematoxylin (Leica 560 MX) 30 sec - Slides washed in tap water, 1 min - Decolorized (10% Acetic Acid in 70% ethanol), 1 min - Slides washed in tap water, 1 min - Bluing (Austin Clear Ammonia), 1 min - Slides washed in tap water, 1 min
- 22. Dehydration/coverslipping procedure: - 3 changes of 95% Ethanol, 3 min each - 3 changes of 100% Ethanol, 3 min each - 3 changes of Xylene, 5 min each - Mounted with Permount
- 23. Imaging: Leica SCN 400F Whole Slide Scanner with Digital Image Hub and Leica Slidepath software

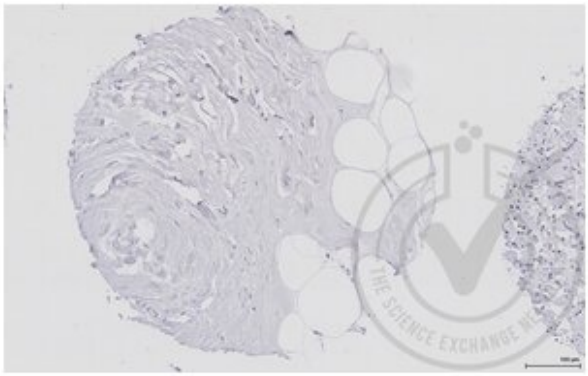
Experimental Notes:

- Deviations from protocol/procedure supplied by manufacturer:
- Step 1: Heated tissue 60°C for 30 minutes; manufacturer heats for 45 minutes.
- Step 2: No ethanol wash was performed during deparaffinization; manufacturer includes 1 wash of 80% ethanol for 3 minutes.
- Step 3.1: Slides were heated for 15 minutes; manufacturer provides a range of 15-20 minutes.
- Step 3.2: Slides were cooled for 30 minutes; manufacturer cools for 20 minutes.
- Step 4: Italicized reagents and incubation time are fixed instrument parameters.
- Step 5: Secondary species-specific serum block not used; manufacturer blocks with 5% normal goat serum for 2 hours.
- Step 8.1: Antibody diluted in PBS at 1:250; manufacture did not recommend diluent or dilution.
- Step 8.2.1: Primary antibody incubated at room temperature overnight; manufacturer incubates overnight 4°C with agitation.



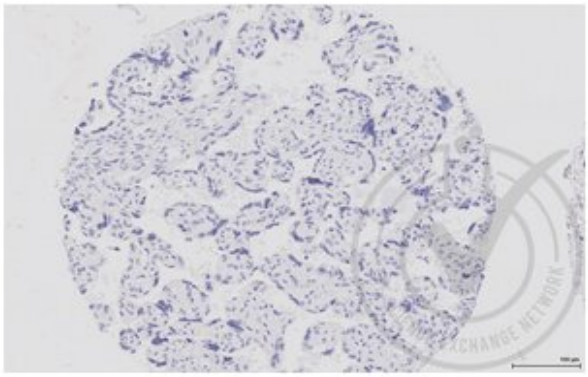
Validation image no. 1 for anti-Mitogen-Activated Protein Kinase Kinase 1 (MAP2K1) (AA 2-150) antibody (ABIN686482)

Figure 1: Human placenta tissue stained with anti-MAP2K1 (brown) and counterstained with hematoxylin.



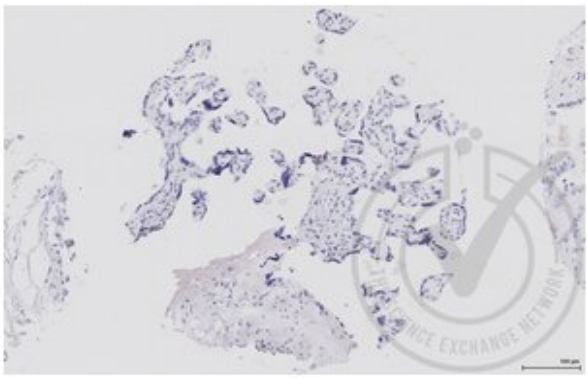
Validation image no. 2 for anti-Mitogen-Activated Protein Kinase Kinase 1 (MAP2K1) (AA 2-150) antibody (ABIN686482)

Figure 2: Human breast adipose tissue stained with anti-MAP2K1 (brown) and counterstained with hematoxylin.



Validation image no. 3 for anti-Mitogen-Activated Protein Kinase Kinase 1 (MAP2K1) (AA 2-150) antibody (ABIN686482)

Figure 3: Human placenta tissue stained with isotype control antibody (brown) and counterstained with hematoxylin.



Validation image no. 4 for anti-Mitogen-Activated Protein Kinase Kinase 1 (MAP2K1) (AA 2-150) antibody (ABIN686482)

Figure 4: Human placenta tissue stained with secondary antibody only (brown) and counterstained with hematoxylin.