abcam

Product datasheet

Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] ab207442

Recombinant RabMAb

18 References 6 Images

Overview

Product name Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549]

Description Rabbit monoclonal [EPR19549] to AMPK alpha 1 + AMPK alpha 2

Host species Rabbit

Tested applications Suitable for: WB, IP

Species reactivity Reacts with: Mouse, Rat, Human, Recombinant fragment

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human AMPK alpha 1 and AMPK alpha 2 full length recombinant proteins; MCF7, HepG2,

HeLa, C6, RAW 264.7, PC-12 and NIH/3T3 whole cell lysates; Human skeletal muscle, fetal liver, fetal heart and fetal kidney lysates; Mouse brain and heart lysates; Rat brain, heart and spleen

lysates. IP: MCF7 whole cell lysate.

General notesThis product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**[®] **patents**.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

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ClonalityMonoclonalClone numberEPR19549

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab207442 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/1000. Detects a band of approximately 62 kDa (predicted molecular weight: 62 kDa).
IP		1/40.

Target

Function

Catalytic subunit of AMP-activated protein kinase (AMPK), an energy sensor protein kinase that plays a key role in regulating cellular energy metabolism. In response to reduction of intracellular ATP levels, AMPK activates energy-producing pathways and inhibits energy-consuming processes: inhibits protein, carbohydrate and lipid biosynthesis, as well as cell growth and proliferation. AMPK acts via direct phosphorylation of metabolic enzymes, and by longer-term effects via phosphorylation of transcription regulators. Also acts as a regulator of cellular polarity by remodeling the actin cytoskeleton; probably by indirectly activating myosin. Regulates lipid synthesis by phosphorylating and inactivating lipid metabolic enzymes such as ACACA, ACACB, GYS1, HMGCR and LIPE; regulates fatty acid and cholesterol synthesis by phosphorylating acetyl-CoA carboxylase (ACACA and ACACB) and hormone-sensitive lipase (LIPE) enzymes, respectively. Regulates insulin-signaling and glycolysis by phosphorylating IRS1, PFKFB2 and PFKFB3. AMPK stimulates glucose uptake in muscle by increasing the translocation of the glucose transporter SLC2A4/GLUT4 to the plasma membrane, possibly by mediating phosphorylation of TBC1D4/AS160. Regulates transcription and chromatin structure by phosphorylating transcription regulators involved in energy metabolism such as CRTC2/TORC2, FOXO3, histone H2B, HDAC5, MEF2C, MLXIPL/ChREBP, EP300, HNF4A, p53/TP53, SREBF1, SREBF2 and PPARGC1A. Acts as a key regulator of glucose homeostasis in liver by phosphorylating CRTC2/TORC2, leading to CRTC2/TORC2 sequestration in the cytoplasm. In response to stress, phosphorylates 'Ser-36' of histone H2B (H2BS36ph), leading to promote transcription. Acts as a key regulator of cell growth and proliferation by phosphorylating TSC2, RPTOR and ATG1: in response to nutrient limitation, negatively regulates the mTORC1 complex by phosphorylating RPTOR component of the mTORC1 complex and by phosphorylating and activating TSC2. In response to nutrient limitation, promotes autophagy by phosphorylating and activating ULK1. AMPK also acts as a regulator of circadian rhythm by mediating phosphorylation of CRY1, leading to destabilize it. May regulate the Wnt signaling pathway by phosphorylating CTNNB1, leading to stabilize it. Also has tau-protein kinase activity: in response to amyloid beta A4 protein (APP) exposure, activated by CAMKK2, leading to phosphorylation of MAPT/TAU; however the relevance of such data remains unclear in vivo. Also phosphorylates CFTR, EEF2K, KLC1, NOS3 and SLC12A1.

Sequence similarities

Belongs to the protein kinase superfamily. CAMK Ser/Thr protein kinase family. SNF1 subfamily.

Contains 1 protein kinase domain.

Domain

The AIS (autoinhibitory sequence) region some sequence similarity with the ubiquitin-associated

domains and represses kinase activity.

Post-translational modifications

Ubiquitinated.

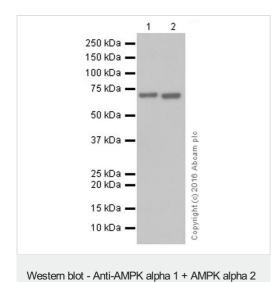
Phosphorylated at Thr-183 by STK11/LKB1 in complex with STE20-related adapter-alpha (STRADA) pseudo kinase and CAB39. Also phosphorylated at Thr-183 by CAMKK2; triggered by a rise in intracellular calcium ions, without detectable changes in the AMP/ATP ratio. CAMKK1 can also phosphorylate Thr-183, but at a much lower level. Dephosphorylated by protein phosphatase 2A and 2C (PP2A and PP2C). Phosphorylated by ULK1 and ULK2; leading to negatively regulate AMPK activity and suggesting the existence of a regulatory feedback loop

between ULK1, ULK2 and AMPK.

Cellular localization

Cytoplasm. Nucleus. In response to stress, recruited by p53/TP53 to specific promoters.

Images



antibody [EPR19549] (ab207442)

All lanes: Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) at 1/2000 dilution

Lane 1: Human AMPK alpha 1 full length recombinant protein Lane 2: Human AMPK alpha 2 full length recombinant protein

Lysates/proteins at 0.01 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

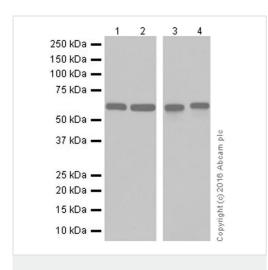
1/100000 dilution

Predicted band size: 62 kDa Observed band size: 62,64 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.

Human AMPK alpha 1 and AMPK alpha 2 full length recombinant proteins contains aa1-559 and aa1-552 with a His-Tag®.



Western blot - Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442)

Lanes 1-2: Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) at 1/10000 dilution

Lanes 3-4: Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) at 1/1000 dilution

Lane 1 : MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lane 2: HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 4: Human skeletal muscle lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 62 kDa **Observed band size:** 62 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442)

All lanes : Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) at 1/1000 dilution

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Lane 1 : Human fetal liver lysate

Lane 2: Human fetal heart lysate

Lane 3: Human fetal kidney lysate

Lysates/proteins at 10 µg per lane.

Secondary

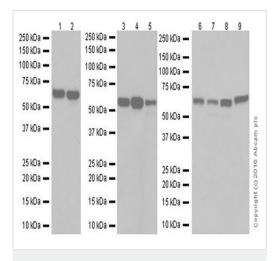
All lanes: Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to

the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 62 kDa **Observed band size:** 62 kDa

Exposure time: 10 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) **All lanes :** Anti-AMPK alpha 1 + AMPK alpha 2 antibody [EPR19549] (ab207442) at 1/1000 dilution

Lane 1: Mouse brain lysate

Lane 2: Mouse heart lysate

Lane 3: Rat brain lysate

Lane 4: Rat heart lysate

Lane 5: Rat spleen lysate

Lane 6: C6 (Rat glial tumor cell line) whole cell lysate

Lane 7: RAW 264.7 (Mouse macrophage cell line transformed

with Abelson murine leukemia virus) whole cell lysate

Lane 8: PC-12 (Rat adrenal gland pheochromocytoma cell line)

whole cell lysate

Lane 9: NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell

lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$ at

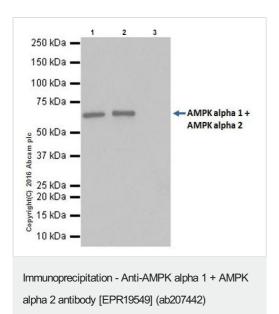
1/100000 dilution

Predicted band size: 62 kDa **Observed band size:** 62 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1/2: 30 seconds; Lane 3-5: 3 minutes Lane 6-

9:5 seconds.



AMPK alpha 1 + AMPK alpha 2 was immunoprecipitated from 0.35 mg of MCF7 (Human breast adenocarcinoma cell line) whole cell lysate with ab207442 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using ab207442 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10000 dilution.

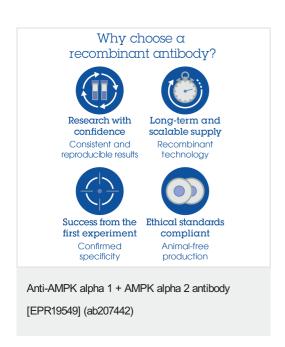
Lane 1: MCF7 whole cell lysate 10ug (Input).

Lane 2: ab207442 IP in MCF7 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab207442 in MCF7 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.



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