### abcam

#### Product datasheet

## Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker ab5432

★★★★★ 1 Abreviews 19 References 9 Images

#### Overview

Product name Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker

**Description** Mouse monoclonal [4.3E8.D10] to ATPB - Mitochondrial Marker

Host species Mouse

**Specificity** Detects the beta subunit of ATP synthase (ATPB) from mouse rat and human samples. This

antibody is useful as a mitochondrial marker.

Tested applications Suitable for: WB, ICC/IF, IP

Species reactivity Reacts with: Mouse, Rat, Human

**Immunogen** Tissue, cells or virus corresponding to ATPB. Intact rat mitochondria.

Positive control ICC/IF: 3T3, HEK293T, A431, rat neuronal glial cells, HeLa; IP: rat neuronal/glial cells, THP-1

cells; WB: human testis tissue lysate

**General notes**The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

#### **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 or -

80°C. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.05% Sodium azide

Constituents: 99% PBS, 0.1% BSA

Purity Affinity purified

Clonality Monoclonal

Clone number 4.3E8.D10

**Isotype** IgG1

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#### **Applications**

#### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab5432 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	<b>★</b> ★ ★ ★ <b>(1)</b>	Use a concentration of 1 - 2 µg/ml. Detects a band of approximately 57 kDa (predicted molecular weight: 57 kDa).
ICC/IF		1/100 - 1/1000.
IP		Use a concentration of 2 - 5 $\mu$ g/ml. By immunoprecipatation, this antibody detects an 50 kDa protein representing ATP synthase (ATPB) from solubilized rat brain mitochondria.

#### **Target**

#### **Function**

Mitochondrial membrane ATP synthase (F(1)F(0) ATP synthase or Complex V) produces ATP from ADP in the presence of a proton gradient across the membrane which is generated by electron transport complexes of the respiratory chain. F-type ATPases consist of two structural domains, F(1) - containing the extramembraneous catalytic core, and F(0) - containing the membrane proton channel, linked together by a central stalk and a peripheral stalk. During catalysis, ATP synthesis in the catalytic domain of F(1) is coupled via a rotary mechanism of the central stalk subunits to proton translocation. Subunits alpha and beta form the catalytic core in F(1). Rotation of the central stalk against the surrounding alpha(3)beta(3) subunits leads to hydrolysis of ATP in three separate catalytic sites on the beta subunits.

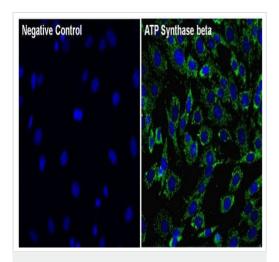
#### Sequence similarities

Cellular localization

#### Belongs to the ATPase alpha/beta chains family.

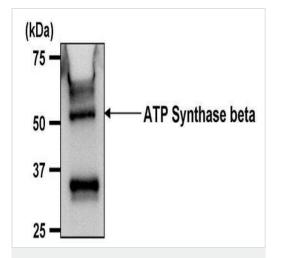
Mitochondrion. Mitochondrion inner membrane. Peripheral membrane protein.

#### **Images**



Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

Immunocytochemistry analysis of ATPB using ab5432 at 5µg/mL concentration shows staining in 4% paraformaldehyde-fixed 3T3 Cells. Secondary was Goat anti-Mouse IgG (H+L) Superclonal Secondary Antibody, Alexa Fluor® 488 conjugate at 1/1000 dilution. ATPB (green), and nuclei with Hoechst 33342 dye (blue) is shown. Negative control has no primary antibody



Immunoprecipitation - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

ATPB was immunoprecipitated from THP-1 whole cell lysate with 5  $\mu L$  ab5432.

Lane 1: ab5432 IP in THP-1 whole cell lysate, with HRP-conjugated goat anti-mouse IgG secondary

**Detection: Chemiluminescence** 



Western blot - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432) at 1  $\mu$ g/ml + Human testis tissue lysate - total protein (ab30257) at 20  $\mu$ g

#### Secondary

Goat Anti-Mouse IgG H&L (HRP) preadsorbed (ab97040) at 1/5000 dilution

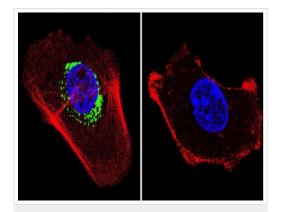
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 57 kDa Observed band size: 57 kDa

# ATP Synthase bela DAPI Merge

Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

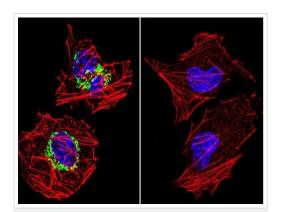


Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

#### Exposure time: 16 minutes

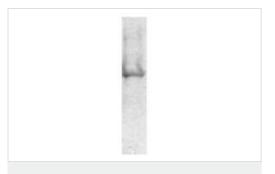
Immunocytochemistry/Immunofluorescent analysis of ATPB (red) in HEK293T cells. Cells fixed with 4% formaldehyde were permeabilized and blocked with 1X PBS containing 5% BSA and 0.3% Triton X-100 for 1 hour at room temperature. Cells were probed with ab5432 at 1:100 overnight at 4°C in 1X PBS containing 1% BSA and 0.3% Triton X-100, washed with 1X PBS, and incubated with a fluorophore-conjugated goat anti-mouse IgG secondary antibody at a dilution of 1:200 for 1 hour at room temperature. Nuclei (blue) were stained with DAPI. Images were taken on a Leica DM1000 microscope at 40X magnification.

Immunofluorescent analysis of ATPB in A431 cells. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with a ATPB monoclonal antibody (ab5432) at a dilution of 1:200 overnight at 4 C and incubated with a DyLight-488 conjugated secondary antibody. ATPB staining (green) F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Images were taken at 60X magnification.

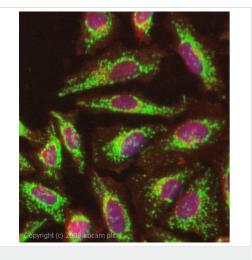


Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432)

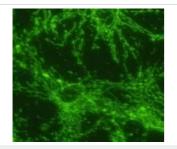
Immunofluorescent analysis of ATPB in HeLa cells. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with a ATPB monoclonal antibody (ab5432) at a dilution of 1:200 overnight at 4 C and incubated with a DyLight-488 conjugated secondary antibody. ATPB staining (green) F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Images were taken at 60X magnification.



Immunoprecipitation - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432) Immunoprecipatation of rat neuronal/glial cell extract using ab5432.



Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432) ICC/IF image of ab5432 stained HeLa cells. The cells were 4% PFA fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab5432, 1 $\mu$ g/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-mouse lgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 $\mu$ M.



Immunocytochemistry/ Immunofluorescence - Anti-ATPB antibody [4.3E8.D10] - Mitochondrial Marker (ab5432) Immunocytochemistry/Immunofluorescence analysis of rat neuronal/glial cell culture using ab5432.

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