abcam

ab14748

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) is a mouse monoclonal antibody detecting ATP5A in **Western Blot, Flow Cytometry, IHC-P, ICC/IF**. Suitable for **Cow, Drosophila melanogaster, Human, Mouse, Rat**.

- Clone 15H4C4 is the most cited clone to ATP5A
- Over 550 publications
- Trusted since 2004

Lab Essentials

Key facts

Isotype	IgG2b
Host species	Mouse
Storage buffer	pH: 7.5 Preservative: 0.02% Sodium azide Constituents: HEPES buffered saline
Form	Liquid
Clonality	Monoclonal
Clone number	15H4C4
Purity	IgG fraction
Light chain type	kappa
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →
Purification notes	Near homogeneity as judged by SDS-PAGE. The antibody was produced in vitro using hybridomas grown in serum-free medium, and then purified by biochemical fractionation.

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1 µg/mL
Notes	Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Expected

Species	Rat
Dilution info	-
Notes	-

Species	Cow
Dilution info	-
Notes	-

Species	Mouse
Dilution info	1 µg/mL
Notes	Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Species	Drosophila melanogaster
Dilution info	1-10 µg/mL
Notes	Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Predicted

Species	Pig
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1-10 µg/mL
Notes	-

Expected

Species	Rat
Dilution info	-
Notes	-

Species	Cow
Dilution info	-
Notes	-

Species	Mouse
Dilution info	1/1 - 1/10
Notes	-

Species	Drosophila melanogaster
Dilution info	1-10 µg/mL
Notes	-

Predicted

Species	Pig
Dilution info	-
Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	1 µg/mL

Notes	ab170192 - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	-
Notes	-

Species	Rat
Dilution info	-
Notes	-

Species	Cow
Dilution info	-
Notes	-

Species	Drosophila melanogaster
Dilution info	1 µg/mL
Notes	ab170192 - Mouse monoclonal IgG2b, is suitable for use as an isotype control with this antibody.

Predicted

Species	Pig
Dilution info	-
Notes	-

WB

Tested

Species	Mouse
Dilution info	1 µg/mL
Notes	-

Species	Rat

Dilution info	1 µg/mL
Notes	-

Species	Human
Dilution info	1 µg/mL
Notes	-

Species	Cow
Dilution info	1 µg/mL
Notes	-

Expected

Species	Drosophila melanogaster
Dilution info	1 µg/mL
Notes	-

Predicted

Species	Pig
Dilution info	-
Notes	-

Target data

See full target information ATP5F1A

Function

Subunit alpha, of the mitochondrial membrane ATP synthase complex (F(1)F(0) ATP synthase or Complex V) that 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 (Probable). ATP synthase complex consist of a soluble F(1) head domain - the catalytic core - and a membrane F(1) domain - the membrane proton channel (PubMed:37244256). These two domains are linked by a central stalk rotating inside the F(1) region and a stationary peripheral stalk (PubMed:37244256). 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 (Probable). In vivo, can only synthesize ATP although its ATP hydrolase activity can be activated artificially in vitro (By similarity). With the catalytic subunit beta (ATP5F1B), forms the catalytic core in the F(1) domain (PubMed:37244256). Subunit alpha does not bear the catalytic high-affinity ATP-binding sites (Probable). Binds the bacterial siderophore enterobactin and can

promote mitochondrial accumulation of enterobactin-derived iron ions (PubMed:30146159).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C

Notes

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) is a mouse monoclonal antibody and is validated for use in Flow Cyt, ICC/IF, IHC-P, WB in human, mouse, rat samples.

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) has been cited over 553 times in peer reviewed journals and is trusted by the scientific community.

Abcam's high quality validation processes ensure Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) has high sensitivity and specificity.

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) has 25 independent reviews from customers.

Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) specifically detects ATP5A (UniProt ID: P25705; Molecular weight: 55kDa) and is sold in 100 µg selling sizes.

Antibody clone 15H4C4 is also available pre-conjugated to FITC for your convenience (ab119688).

ATP5A (ATP Synthase F1 Subunit Alpha) is an essential protein for ATP production in mitochondria, forming part of the ATP synthase complex. This complex is crucial for oxidative phosphorylation, the process by which cells generate energy. Mutations in the ATP5A gene are linked to mitochondrial disorders, including combined oxidative phosphorylation deficiency 22 (COXPD22), which manifests as severe metabolic and neurological issues, emphasizing the protein's critical role in energy metabolism.

Want a custom formulation?
This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	ATP5A also known as ATP synthase F1 subunit alpha is a protein important for cellular energy production. As part of the ATP synthase complex it plays a

	mechanical role in synthesizing ATP from ADP and inorganic phosphate. The ATP5A protein has a molecular weight of approximately 55 kDa and is widely expressed in the inner mitochondrial membrane across different cell types. Its central function lies in its ability to harness the energy of the proton gradient generated by the electron transport chain to catalyze ATP synthesis.
Biological function summary	ATP5A is essential in cellular respiration serving as a catalytic core of the F1 component of ATP synthase. As part of the multi-subunit enzyme complex ATP synthase is responsible for ATP production the primary energy currency in cells. The ATP5A subunit works in conjunction with other subunits of the enzyme oligomer to facilitate the conversion of energy released during oxidative phosphorylation into a usable form. The protein's efficiency in this biological role underpins its importance in sustaining cellular energy homeostasis.
Pathways	ATP5A plays a pivotal role in oxidative phosphorylation and the electron transport chain integral components of cellular respiration. The oxidative phosphorylation pathway depends on this protein to manage the synthesis of ATP molecules while the electron transport chain creates the proton gradient necessary for ATP production. ATP5A is functionally connected to other proteins in these pathways such as ATP5B and cytochrome c oxidase working in a coordinated manner to ensure efficient energy transfer and maintenance.
Associated diseases and disorders	ATP5A is implicative in mitochondrial disorders and neurodegenerative diseases such as Leigh syndrome and Parkinson's disease. These conditions often arise from deficits in ATP production where ineffective ATP synthase activity can contribute to cellular energy failures. In the context of Parkinson’s disease for instance ATP5A interactions with other proteins like Parkin can contribute to mitochondrial dysfunction an important pathological feature of the disorder. Through such associations alterations in ATP5A activity can significantly impact disease progression and symptomatology.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

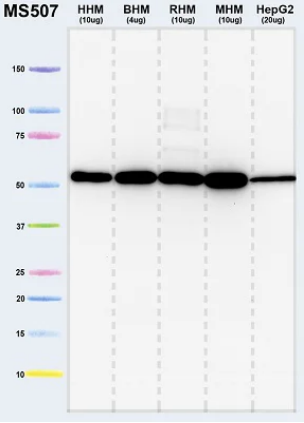
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

11 product images



Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

All lanes:
Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) at 1 µg/mL

Lane 1:
Isolated mitochondria from human heart at 10 µg

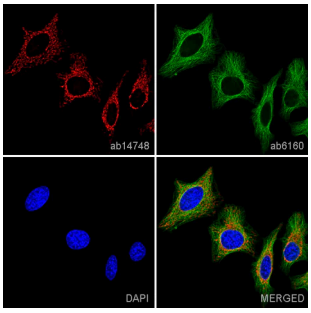
Lane 2:
Isolated mitochondria from bovine heart at 4 µg

Lane 3:
Isolated mitochondria from rat heart at 10 µg

Lane 4:
Isolated mitochondria from mouse heart at 10 µg

Lane 5:
HepG2 (Human liver hepatocellular carcinoma cell line) lysate at 20 µg

Predicted band size: 60 kDa



Immunocytochemistry/ Immunofluorescence - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

ATP5A Immunocytochemistry/ Immunofluorescence staining using mouse Anti-ATP5A antibody

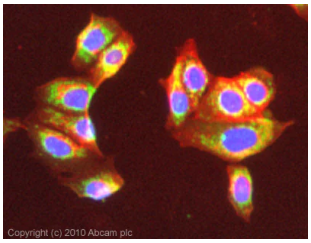
ICC/IF image of ab14748 stained HeLa (Human epithelial cell line from cervix adenocarcinoma) cells.
The cells were fixed in 100% methanol (5 minutes), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1 hour. The cells were then incubated overnight at +4°C with ab14748 at 1 µg/ml (shown in red) and ab6160 (Rat monoclonal to Tubulin) at 1 µg/ml (shown in green).

This was followed by an incubation at room temperature for 1 hour with ab150119, Goat Anti-Mouse IgG H&L (Alexa Fluor® 647) preadsorbed, at 0.5 µg/ml (shown in red) and ab150165, Goat Anti-Rat IgG H&L (Alexa Fluor® 488) preadsorbed, at 0.5 µg/ml (shown in green).

Nuclear DNA was labeled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

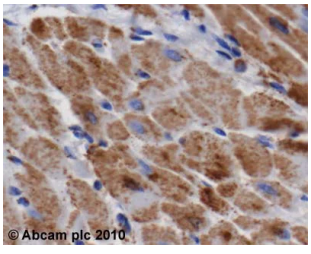
This product also gave a positive signal under the same testing conditions in HeLa cells fixed with 4% formaldehyde (10 minutes).



Immunocytochemistry/ Immunofluorescence - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

ATP5A Immunocytochemistry/ Immunofluorescence staining using mouse Anti-ATP5A antibody

ICC/IF image of ab14748 stained MCF7 (Human breast adenocarcinoma cell line) cells.
The cells were fixed in 4% formaldehyde (10 minutes) and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1 hour to permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab14748, 10 µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-mouse IgG (H+L) used at a 1/1000 dilution for 1 hour. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1 hour. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 µM.

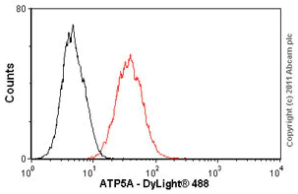


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

ATP5A Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-ATP5A antibody

ab14748 (1 µg/ml) staining ATP5A in human heart (left ventricle), using an automated system (DAKO Autostainer Plus). Using this protocol there is mitochondrial staining of cardiomyocytes. Sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffer citrate pH 6.1 in a DAKO PT link. Slides were peroxidase blocked in 3% H_2O_2 in methanol for 10 minutes. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 minutes and detected with Dako envision flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes.

Slides were counterstained with hematoxylin and coverslipped under DePeX. Please note that, for manual staining, optimization of primary antibody concentration and incubation time is recommended. Signal amplification may be required.



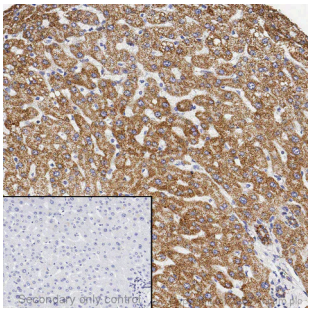
Flow Cytometry - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

ATP5A Flow Cytometry staining using mouse Anti-ATP5A antibody

Overlay histogram showing HepG2 (Human liver hepatocellular carcinoma cell line) cells stained with ab14748 (red line).
The cells were fixed with 4% paraformaldehyde (10 minutes) and then permeabilized with 0.1% PBS-Tween for 20 minutes. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab14748, 1 µg/1x10^6 cells) for 30 minutes at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 minutes at 22°C. Isotype control antibody (black line) was mouse IgG2b [PLPV219] (ab91366, 2 µg/1x10^6 cells) used under the same conditions.

Acquisition of >5,000 events was performed.

This antibody gave a positive signal in HepG2 cells fixed with 80% methanol (5 minutes)/permeabilized in 0.1% PBS-Tween used under the same conditions.

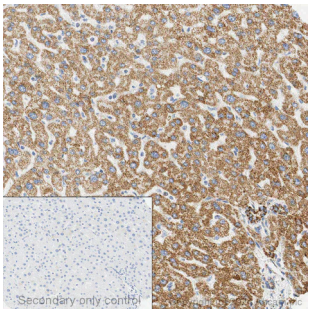


Immunohistochemistry - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

Immunohistochemical analysis of formalin fixed paraffin embedded human liver labelling ATP5A with ab14748 at a concentration of 0.05µg/ml. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with a Bond™ Polymer Refine Detection kit. Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution 2) for 20 mins.

ab14748 anti-ATP5A antibody [15H4C4] was incubated for 15mins at room temperature. Sections were counterstained with Hematoxylin. Image inset shows absence of staining in secondary antibody only control.

Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual)



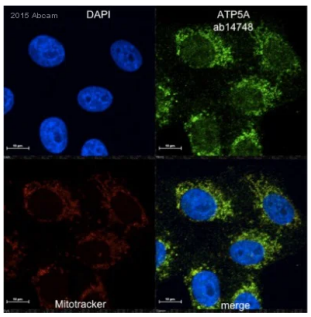
Immunohistochemistry - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

Immunohistochemical analysis of formalin fixed paraffin embedded human liver labelling ATP5A with ab14748 at a concentration of 0.01µg/ml.

The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32min with DISCOVERY cell conditioning solution (CC1) 100°C, pH8.5. ab14748 anti-ATP5A was incubated at 37°C for 16min.

Sections were counterstained is with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

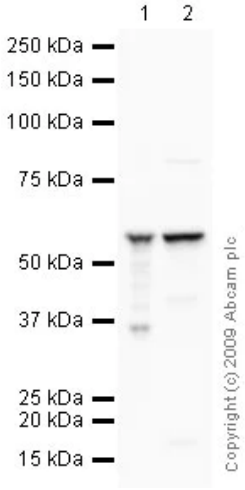
Customers are encouraged to optimise antigen retrieval conditions, antibody concentration, incubation times and temperature for best results in their own IHC assay workflow (automated and manual)



This image is courtesy of an anonymous customer review

Immunocytochemistry/ Immunofluorescence - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

ab14748 staining ATP5A in MDA-MB-231 cells by ICC/IF (Immunocytochemistry/immunofluorescence).
Cells were fixed with formaldehyde, permeabilized with 1% Triton X-100 and blocked with 10% BSA for 1 hour at 21°C. Samples were incubated with primary antibody (1/100 in BSA + 0.02% Tween 20) for 1 hour at 21°C. A DyLight® 550-conjugated goat anti-mouse IgG polyclonal (1/500) was used as the secondary antibody.



Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

All lanes:
Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748) at 1 µg/mL

Lane 1:
Human liver tissue lysate at 10 µg

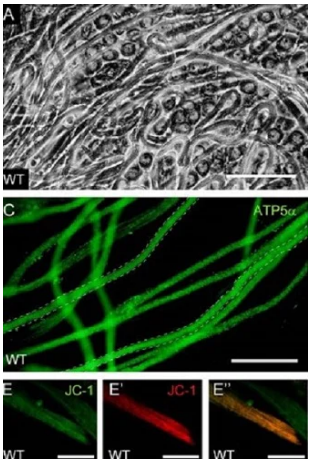
Lane 2:
HepG2 (Human hepatocellular liver carcinoma cell line) whole cell lysate at 10 µg

Secondary

All lanes:
Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Predicted band size: 60 kDa

Observed band size: 36 kDa, 55 kDa



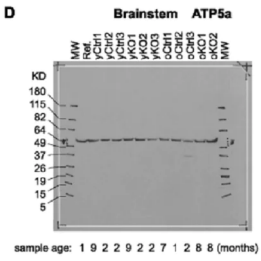
Immunocytochemistry/ Immunofluorescence - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

Testes of *bb8*^{ms} mutants show defects in post-meiotic, elongated spermatids.
(A-B) Spermatids from WT (A) and *bb8*^{ms} (B) testis both have elongated cysts, but there are large spherical vesicles in the mutant (arrows) by phase contrast microscopy. Scale bars: 100 µm.

(C, D) Mitochondria of elongated spermatids stained with ATP5α antibody ab14748 in WT (C) and in *bb8*^{ms} (D) mutants. ATP5α positive staining of the large vesicles in the cysts are indicated by arrow. Scale bars: 50 µm.

(E, F) JC-1 staining positive large vesicles (arrows) are absent from WT (E), but present in *bb8*^{ms} elongated cysts (F). Scale bars: 25 µm.

Vedelek et al PLoS One. 2016 Aug 16;11(8):e0161289. doi: 10.1371/journal.pone.0161289. eCollection 2016. Fig 4. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/



Kayser et al PLoS One. 2016 Jan 29;11(1):e0148219. doi: 10.1371/journal.pone.0148219. eCollection 2016. Fig S2. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

Western blots were probed for HNE-damaged mitochondrial protein from brainstem (A), cerebellum (B) and "rest" brain (R). The sample designation indicates the age group (y for P25-P35, o for P45-P55), the genotype (KO, Ctrl for controls) and a number to distinguish independent samples.
Panel D is the blot from panel A reprobed for the mitochondrial marker ATPase (ATP5a) using ab14748 to demonstrate that extended sample storage did not degrade sample protein in general. Black lines in the MW lanes are magic marker on the film to indicate the positions of the prestained molecular weight standards on the blot.

For full image please see paper.

All lanes:
Western blot - Anti-ATP5A antibody [15H4C4] - Mitochondrial Marker (ab14748)

Predicted band size: 60 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.