abcam

ab75973

Anti-Ferritin heavy chain antibody [EPR3004Y]

Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) is a rabbit monoclonal antibody detecting Ferritin Heavy Chain in **Western Blot, Flow Cytometry (Intra), ICC/IF**. Suitable for **Human, Mouse, Rat**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 110 publications
- Trusted since 2009

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR3004Y
Purification technique	Affinity purification Protein A
Specificity	We recommend following our protocol when testing ICC and recommend using lower dilution in samples having low expression level of Ferritin.
Concentration	0.068 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Mouse
Dilution info	1/1000 - 1/2000
Notes	-

Species	Rat
Dilution info	1/1000 - 1/2000
Notes	-

Species	Human
Dilution info	1/1000 - 1/2000
Notes	-

IHC-P

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	We recommend following our protocol when testing ICC and recommend using lower dilution in samples having low expression level of Ferritin.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/50
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information FTH1

Function	Stores iron in a soluble, non-toxic, readily available form. Important for iron homeostasis. Has ferroxidase activity (PubMed:9003196). Iron is taken up in the ferrous form and deposited as ferric hydroxides after oxidation (PubMed:9003196). Also plays a role in delivery of iron to cells (By similarity). Mediates iron uptake in capsule cells of the developing kidney (By similarity). Delivery to lysosomes is mediated by the cargo receptor NCOA4 for autophagic degradation and release of iron (PubMed:24695223, PubMed:26436293).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Product Specifications
Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) was developed by Abcam using patented rabbit monoclonal antibody technology and is validated for use in Flow Cyt (Intra), ICC/IF, WB in human, mouse, rat samples.
Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) specifically detects Ferritin Heavy Chain (UniProt ID: P02794; Molecular weight: 21kDa) and is sold in 100 µL and 1 mL selling sizes.

Quality and Validation
Abcam's high quality manufacturing and validation processes ensure Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) has high sensitivity and specificity alongside high lot-to-lot consistency and reproducibility.
The specificity of Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) has been confirmed by testing in knockout samples.
Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) has been cited over 113 times in peer reviewed journals and is trusted by the scientific community.
Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) has 7 independent reviews from customers.

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

What are the advantages of a recombinant monoclonal antibody?
This 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 batch production

For more information, read more on recombinant antibodies.

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

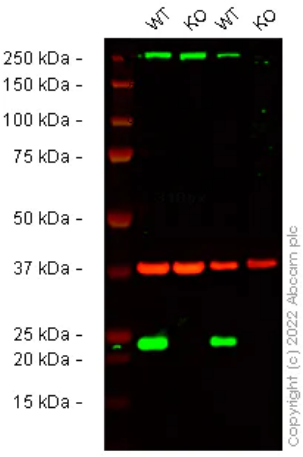
We do not recommend this combination. It is not covered by our product promise.

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.

9 product images



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

False colour image of Western blot: Anti-Ferritin heavy chain antibody [EPR3004Y] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab75973 was shown to bind specifically to Ferritin heavy chain. A band was observed at 24 kDa in wild-type MCF7 cell lysates with no signal observed at this size in FTH1 knockout cell line ab269493 (knockout cell lysate ab269655). To generate this image, wild-type and FTH1 knockout MCF7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.
All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/1000 dilution

Lane 1:
Wild-type MCF7 cell lysate at 20 µg

Lane 2:
FTH1 knockout MCF7 cell lysate at 20 µg

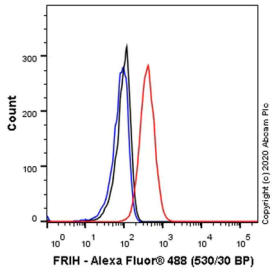
Lane 3:
Wild-type HEK-293T ab255553 at 20 µg

Lane 4:
FTH1 knockout HEK-293T ab260185 cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 21 kDa

Observed band size: 24 kDa

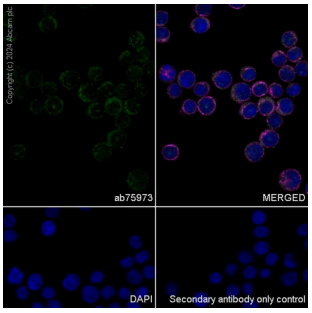


Flow Cytometry (Intracellular) - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Ferritin heavy chain Flow Cytometry (Intracellular) staining using rabbit Anti-Ferritin heavy chain antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized SH-SY5Y (Human neuroblastoma epithelial cell) cells labelling Ferritin heavy chain with ab75973 at 1/50 dilution (1µg) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype

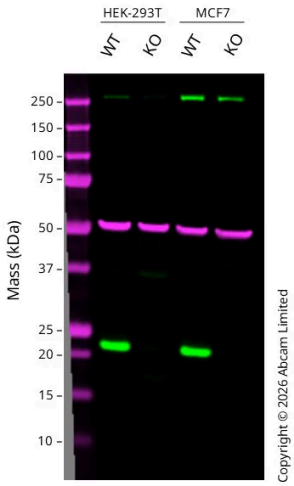
control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Immunofluorescence staining of 4% paraformaldehyde fixed, 0.1% Triton X-100 permeabilized Jurkat cells (human T cell leukemia T lymphocyte from peripheral blood) with ab75973 at a working dilution of 1/100. The secondary antibody, ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed, was used at a dilution of 1/1000. The counterstain, ab195889 anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used at a dilution of 1/200.

Confocal image showing positive staining in Jurkat cells (shown in green). The counterstain was observed in magenta. Nuclear DNA was labeled with DAPI (shown in blue). Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Ferritin heavy chain Western blot staining using rabbit Anti-Ferritin heavy chain antibody

Western blot: Anti-Ferritin heavy chain antibody [EPR3004Y] ab75973 staining at 1/1000 dilution, shown in green; Mouse anti alpha Tubulin ab7291 loading control staining at 1/20000 dilution, shown in magenta. A band was observed at 21 kDa in Wild-type HEK-293T cell lysates with no signal observed at this size in FTH1 knockout HEK-293T cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3% Milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit 800CW and Goat anti-Mouse 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/1000 dilution

Lane 1:
Wild-type HEK-293T cell lysates at 20 µg

Lane 2:
Western blot - Human FTH1 knockout HEK-293T cell line (ab266581) at 20 µg

Lane 3:
Wild-type MCF7 cell lysates at 20 µg

Lane 4:
Western blot - Human FTH1 knockout MCF7 cell line (ab269493) at 20 µg

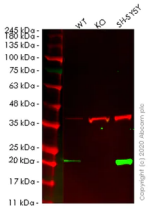
Secondary

All lanes:
Goat anti-Rabbit 800CW & Goat anti-Mouse 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 21 kDa

Observed band size: 21 kDa



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Ferritin heavy chain Western blot staining using rabbit Anti-Ferritin heavy chain antibody

Lanes 1-3: Merged signal (red and green). Green - ab75973 observed at 21 kDa. Red - loading control ab8245 observed at 36 kDa.
ab75973 Anti-Ferritin heavy chain antibody [EPR3004Y] was shown to specifically react with Ferritin heavy chain in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266581 (knockout cell lysate ab256924) was used. Wild-type and Ferritin knockout samples were subjected to SDS-PAGE. ab75973 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2. 5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/1000 dilution

Lane 1:
Wild-type HEK293T cell lysate at 20 µg

Lane 2:
Western blot - Human FTH1 knockout HEK-293T cell lysate (ab256924) at 20 µg

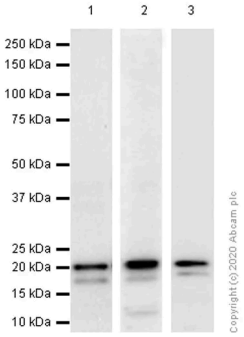
Lane 3:
SH-SY5Y cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 21 kDa

Observed band size: 21 kDa, 36 kDa



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Observed MW: 21 kDA
Blocking/diluting buffer and concentration: 5% NFDM/TBST

All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/2000 dilution

Lane 1:
Jurkat (Human T cell leukemia T lymphocyte) whole cell lysates at 20 µg

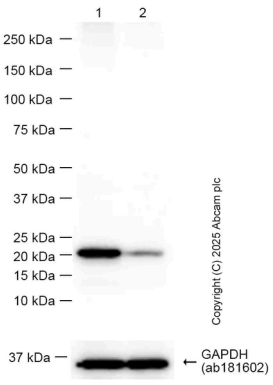
Lane 2:
HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysates at 20 µg

Lane 3:
C6 (Rat glial tumor glial cell) whole cell lysates at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/20000 dilution

Predicted band size: 21 kDa



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

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

All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/1000 dilution

Lane 1:
HeLa (Human cervix adenocarcinoma epithelial cell) + si -scramble whole cell lysate at 20 µg

Lane 2:

HeLa (Human cervix adenocarcinoma epithelial cell) + si - Ferritin Heavy Chain whole cell lysate at 20 µg

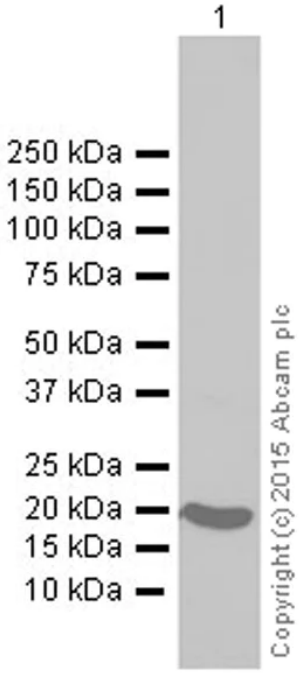
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 21 kDa

Observed band size: 21 kDa

Exposure time: 3s



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST
All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/10000 dilution

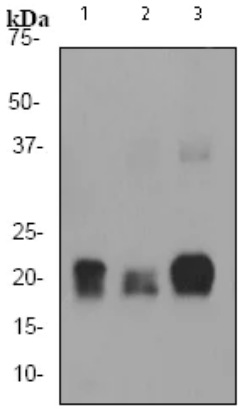
All lanes:
Raw264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 10 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 21 kDa

Observed band size: 21 kDa



Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973)

All lanes:
Western blot - Anti-Ferritin heavy chain antibody [EPR3004Y] (ab75973) at 1/2000 dilution

Lane 1:
Jurkat (Human T cell leukemia cell line from peripheral blood) cell lysate at 10 µg

Lane 2:
SH-SY5Y (Human neuroblastoma cell line from bone marrow) cell lysate at 10 µg

Lane 3:
Human brain lysates at 10 µg

Secondary

All lanes:
HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 21 kDa

Observed band size: 19 kDa, 21 kDa

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